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Infectious Diseases of the Gastrointestinal Tract

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Calves

Escherichia coli

Escherichia coli is a normal inhabitant of the gastrointestinal (GI) tract of warm-blooded animals and ubiquitous in the farm environment. Disease caused by *E. coli* in calves may present as enteric or septicemic illness and is an important cause of neonatal mortality in dairy calves. Failure of passive transfer and management practices that allow exposure of neonatal calves to large numbers of *E. coli* are of central importance in the pathogenesis of disease. Because of substantial genotypic and phenotypic variation, it is possible to subgroup *E. coli* into a large number of different serotypes. The commensal *E. coli* are important members of the normal gut microbiota, and only a small fraction of the total *E. coli* population in nature are classified as pathovars or pathotypes. Modern analytical methods have permitted more detailed identification of virulence associated factors and elucidation of specific virulence mechanisms in the classification of pathogenic serotypes of these gram-negative organisms. Broadly speaking, *E. coli* are classified based on several serologic and antigenic parameters, including cell wall or somatic (O) antigens, capsular (K) antigens, pilar or fimbrial (F) antigens, and flagellar (H) antigens. Heretofore, pilus antigens were sometimes classified as K antigens, but recent reference to pilus antigens as F antigens reduces confusion in this area. Among the diarrheagenic pathotypes of *E. coli* the most significant in neonatal calves are the enterotoxigenic *E. coli* (ETEC), which is the most commonly confirmed noncommensal pathotype of *E. coli* in cattle. Enteropathogenic (EPEC), enterohemorrhagic (EHEC), and Shiga toxin–producing *E. coli* (STEC) are pathotypes that are also isolated from diarrheic calves, but their role in neonatal calf diarrhea remains more controversial because they can also be found in healthy individuals. Increasing concern about zoonotic illness and antimicrobial resistance among these pathotypes of *E. coli* is impossible to ignore if one works in the cattle industry, dairy cattle in particular being identified as important reservoirs for zoonotic ETEC, EHEC, and STEC disease.

Septicemia (Septicemic Colibacillosis, Colisepticemia)

Etiology

Colisepticemia in neonatal calves can be considered a disease of poor management. Failure of passive transfer is the primary risk factor for this disease. Colostral transfer of immunoglobulins may be compromised by short dry periods, preparturient leaking of colostrum, assumption that a calf has nursed colostrum just because it is left with the dam for 24 hours, primiparous heifers that have poor-quality colostrum, and many other factors. Provision of an inadequate immunoglobulin mass can be a problem with both commercial colostrum substitutes or replacers as well as dam or farm sourced colostrum. In addition, poor maternity area and poor calf pen hygiene promote exposure of calves to the multitude of strains of *E. coli* capable of causing septicemia, the majority of which are commensal. Filthy conditions; calving areas that are dirty, wet, overcrowded, or overused; and failure to dip navels are additional factors that predispose to this problem. Sanitation and hygiene with respect to collecting, storing, and administering colostrum are important factors in the provision of adequate passive transfer and the prevention of colibacillosis.

Invasive *E. coli* of many subgroups are capable of opportunistic, septicemic infection of neonatal calves. Various reviews suggest an involvement of a multitude of possible *E. coli* types. Variations may be explained by geographic or environmental differences.

Calves with less than 500 mg IgG/dL are very prone to septicemic *E. coli,* and those with 500 to 1000 mg IgG/dL are defined as having partial failure of passive transfer (FPT) and are also at increased risk. Adequate transfer of passive immunoglobulin that ensures at least 1000 IgG mg/dL serum (10 mg/mL serum) or preferably 1600 mg/dL serum is likely to prevent the disease.

Septicemia caused by *E. coli* most commonly occurs from 1 to 14 days of age. The onset of disease tends to occur earlier in this time frame when calves are exposed to high numbers of *E. coli* soon after birth (i.e., in the maternity pen). Poor or nonexistent transfer of passive immunoglobulins to the calf also hastens the onset of disease. Invasive *E. coli* may gain entrance through the navel, intestine, or nasal and oropharyngeal mucous membranes. After invasion and septicemia occur, clinical signs develop rapidly and usually are apparent within 24 hours. Calves with partial FPT or those exposed to less virulent *E. coli* strains may develop more chronic signs of disease over several days.

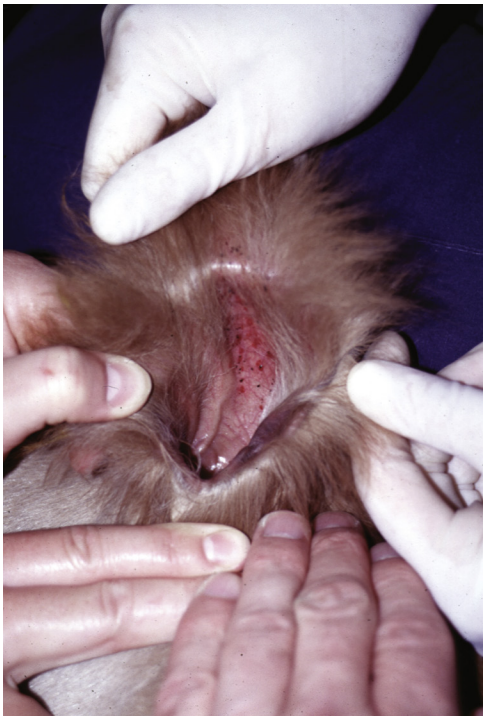
Septicemic calves shed the causative *E. coli* in urine, oral secretions, nasal secretions, and later in the feces, provided they survive long enough to develop diarrhea. Thus, transmission may occur among communally housed calves, crowded calves, or uncleaned maternity stalls because of the heavily infected secretions of sick and septicemic calves. Because septicemic calves can shed large numbers of the organism before clinical signs are evident, contamination of communal pens and common-use feeding devices (e.g., esophageal feeding tubes) and direct contact with the infected calf or its feces or urine may promote spread of infection. Infected calves allowed to remain in the maternity area will amplify the level of environmental contamination, thereby placing other neonates born in that area at risk. Similar amplification may occur in calf housing areas and reinforces the biosecurity need for spatial and temporal separation between occupants, as well as the appropriate and routine disinfection of calf housing.

Clinical Signs

Peracute signs of depression, weakness, tachycardia, and dehydration predominate when highly virulent strains of *E. coli* cause septicemia. Affected calves usually are less than 7 days of age and may be less than 24 hours old. Although often present early on, fever is usually absent by the time obvious clinical signs of fulminant disease occur, when endotoxemia and the resultant poor peripheral perfusion often render the animal normothermic or hypothermic. Exceptions to this rule are calves with peracute disease that collapse when exposed to direct sunlight on hot days; such calves can be markedly hyperthermic. Signs of dehydration are mild to moderate in most cases. The suckle reflex is greatly reduced or absent, and the vasculature of the sclerae is markedly injected. Petechial hemorrhages may be visible on mucous membranes and extremities, particularly the pinnae of the ears (Fig. 6.1). The limbs, mouth, and ears are cool to the touch. Affected calves show progressive weakness and lethargy, often becoming comatose before death. Diarrhea is often seen but may not be apparent in peracute cases.

Evidence of localization of infection in certain tissues may become apparent in cases that survive the acute disease. Hypopyon may be present, as may uveitis, which is evidenced by miotic pupils with increased opacity to the aqueous fluid ("aqueous flare"). Hyperesthesia, paddling, and opisthotonus (Fig. 6.2) are signs suggestive of septic meningitis. Diarrhea may be present in some calves with colisepticemia. Lameness may result from bacterial seeding of joints or growth plates. Signs of omphalophlebitis may be present. Weakness, poor body condition, and recumbency secondary to weakness or joint or bone pain may be present in chronic cases. These localized and less fulminant infections may occur in slightly older calves (>7 days of age). Fever is often present in calves with joint ill or meningitis.

Clinical signs of acute septicemia may be difficult to differentiate from those of acute ETEC infection because dehydration, weakness, and collapse may be common to both. Age at onset can be valuable in the differentiation of ETEC infections because calves are most often within the first 72 to 96 hours of life with this condition and frequently less than 48 hours old. Furthermore, septicemic calves tend to be less dehydrated and have less watery diarrhea than calves



• **Fig. 6.1** Ear of 5-day-old Jersey calf demonstrating petechial hemorrhage associated with *Escherichia coli* septicemia.

• **Fig. 6.2** Seven-day-old Holstein calf with opisthotonus associated with *Escherichia coli* meningitis and septicemia as a result of failure of passive transfer.

with ETEC diarrhea; also, diarrhea tends to develop in the terminal stages of septicemia. Historical data may indicate other neonatal calves have recently shown similar signs or died at less than 2 weeks of age. Other differential diagnoses for acute colisepticemia include hypoxia or trauma during birth, simple hypothermia or hypoglycemia, septicemia caused by *Salmonella* spp., and congenital defects of the central nervous or cardiovascular systems. Polyarthritis caused by *Mycoplasma* spp. is an important differential diagnosis for septic arthritis secondary to colisepticemia but tends to be seen in considerably older calves. Salt poisoning, hypoglycemia, congenital neurologic disorders, traumatic injuries, and intoxications (e.g., lead) should be considered as differential diagnoses for meningitis secondary to colisepticemia. We have also seen several herds in recent years with young calves presenting with neurologic signs indistinguishable from meningitis for which the ultimate diagnosis was ionophore toxicity caused by overdosing before feeding milk replacer. Failure of passive transfer and meningitis were not involved.

Ancillary Data

Calves with peracute *E. coli* septicemia often have elevated packed cell volumes resulting from dehydration and endotoxic shock. The total white blood cell (WBC) count is variable but is frequently low or within normal ranges. Generally, a left shift is observed, and toxic changes (e.g., azurophilic cytoplasm, nuclear hypersegmentation, and Dohle bodies) are often apparent on cytologic examination of blood neutrophils. Plasma fibrinogen concentration is variable. Hypoglycemia is a common finding, and metabolic acidosis, although common, usually is less severe than in calves recumbent as a result of ETEC. In fact, an acid–base and electrolyte determination that does not demonstrate a severe metabolic acidosis in a recumbent, diarrheic, dehydrated calf less than 14 days of age usually portends septicemia. Blood cultures provide the greatest specific diagnostic aid, but results may not be forthcoming in time to help the patient.

Acutely, subacutely, and chronically septicemic calves may have detectable clinical signs of localization of infection that allow a more definitive diagnostic test (e.g., cerebrospinal fluid [CSF] tap for patients showing signs of meningitis or arthrocentesis to confirm septic arthritis) (Fig. 6.3). In chronic cases (calves 2 weeks of age or older), the serum immunoglobulin concentration (and serum total globulin concentration) may be normal or increased as a result of de novo synthesis of antibodies in response to the well-established bacterial infection.

Diagnosis

Whenever clinical signs suggest the diagnosis, the calf's serum immunoglobulin levels should be analyzed. Although adequate levels of IgG do not rule out the disease, calves with IgG of 1600 mg/dL or more based on a single radial immunodiffusion test are highly unlikely to develop septicemic *E. coli* infection. Specific laboratory evaluation of immunoglobulin

• **Fig. 6.3** A 1-week-old calf affected with subacute *Escherichia coli* septicemia. The calf has fever, diarrhea, dehydration, and a septic carpal joint. The calf had inadequate immunoglobulin levels.

levels is preferable to field techniques when confirmation of FPT is essential but may not provide timely results for practitioners. Therefore, even though dehydration may falsely elevate blood protein levels, these field techniques may be useful. Adequate immunoglobulin levels are suggested by serum total protein of 5.5 g/dL or more in calves less than 7 days of age. The use of a Brix refractometer is a simple, inexpensive on-farm tool for the monitoring of neonatal dairy calf immunity levels. A plasma value of 10% or more can be used to classify calves with successful transfer of passive immunity. Other reports have found that using a Brix percentage of 8.5% or less had optimal sensitivity (100%) for detecting failure of adequate immunoglobulin transfer. Previous literature has suggested that serum γ-glutamyl transferase (GGT) activity greater than 200 IU/L in 1 day old calves could be used as a cut-off point for adequacy of passive transfer. However, values for serum GGT can vary widely in healthy and sick calves and measurement of serum GGT should not be used as a gold standard in measuring colostral absorption. The development of visible turbidity in the 18% solution of sodium sulfite turbidity test is a reliable test for adequate passive transfer. A commercial turbidimetric assay (Midlands Bio-Products, Boone, IA) is available for on-farm determination of IgG concentration in serum samples from neonatal calves.

Blood cultures provide definitive diagnosis of bacteremia but usually provide this information too late to be of practical value. When multiple calves are affected, however, blood cultures can help to differentiate *E. coli* septicemia from septicemia caused by other pathogens (e.g., *Salmonella* spp.); this differentiation is relevant for determining the source of infection and initiation of preventive measures. Furthermore, antimicrobial sensitivity testing of blood culture isolates may aid in directing therapy, especially if a common etiologic cause is identified. Clinicians and producers should be aware of the differences between pathotypic strains of *E. coli* (e.g., ETEC) capable of producing severe disease in calves with adequate passive transfer and the everyday, commensal,

and environmental *E. coli* often associated with sepsis caused by FPT. This is an important distinction, lest clients concentrate preventive efforts and management on specific vaccination programs rather than colostrum and neonatal calf management.

Treatment

Treatment of peracute *E. coli* septicemia usually is unsuccessful because of overwhelming bacteremia and endotoxemia in the patient. Signs progress so quickly that most septicemic calves are recumbent and comatose by the time of initial examination. Shock, lactic acidosis, hypoglycemia, and multiple organ failure are common in peracute cases.

If treatment is attempted, correction of endotoxic shock and acid–base and electrolyte abnormalities, effective antimicrobial therapy, and nutritional support are the primary goals. Intravenous (IV) balanced electrolyte solutions should contain dextrose (2.5%–10%), and sodium bicarbonate (20–50 mEq/L) if the plasma bicarbonate concentration is (<10 mEq/L), to address hypoglycemia and metabolic acidosis respectively. Adjustments of the concentration of dextrose and sodium bicarbonate in polyionic fluids can be guided by subsequent serum chemistry results. Maintaining normoglycemia in some peracute and acute septicemic calves can be extremely challenging due to consumption of administered glucose by bacteria. Antimicrobials used to treat neonatal septicemia should be bactericidal and possess a good gram-negative spectrum, such as ceftiofur, trimethoprim-sulfa, or ampicillin. Parenteral administration is necessary to achieve effective blood concentrations. Aminoglycosides such as gentamicin or amikacin can be used alone or in conjunction with the synergistically acting beta-lactam antibiotics (e.g., ceftiofur, penicillin, or ampicillin). Unfortunately, it is not uncommon to find *E. coli* cultured from blood or tissue of calves with septicemia resistant to ceftiofur, ampicillin and gentamicin. The use of potentially nephrotoxic aminoglycosides in a dehydrated patient with prerenal azotemia must be weighed against the potential bactericidal activity of the drugs. Given the present concerns regarding aminoglycoside use in food animals in the United States, use should be limited to situations in which other antibiotics have proven ineffective or in patients for which in vitro susceptibility testing has revealed an absence of approved, labeled alternatives. Furthermore, a minimum 18-month slaughter withdrawal must be enforced for calves that receive aminoglycosides. Use of fluoroquinolones (e.g., enrofloxacin, danofloxacin) and florfenicol in dairy calves is currently not permitted for anything other than respiratory disease under federal law in the United States.

If the previous therapy stabilizes the patient, a transfusion of 1 to 2 L of whole blood from (preferably) a bovine leukemia virus (BLV) and most importantly, bovine viral diarrhea virus (BVDV) negative cow should be performed because FPT is either assumed or confirmed. This translates to a dosage of 20-40 mL of whole blood per kilogram for the calf. Bovine plasma may also be used at the same dosage rate as whole blood and is currently commercially available in the US (Lake Immunogenics, Ontario, NY). Nutritional support ideally entails frequent feedings of small volumes of whole milk or good-quality milk replacer. Partial or total parenteral nutrition (TPN) may be considered for valuable calves, particularly those with concurrent and significant enteritis. Deep, dry bedding; good ventilation; and good nursing care are essential adjuncts to medical treatment.

Specific sites of localized infection also may require specific therapy. As an example, patients manifesting seizures because of meningitis may require diazepam (initial 5 mg dose for a neonatal calf, increasing by 5 mg increments, to effect) to control seizures. Calves with septic joints often require joint lavage. In many cases, especially chronic ones, arthrotomy is necessary to remove fibrin clots and more inspissated material from infected joints.

Chronic cases usually are cachectic; have polyarthritis, bronchopneumonia, and diarrhea; and have an extremely poor prognosis. Although recumbent, weak, dehydrated, and emaciated, these patients tend to have relatively normal acid–base and electrolyte values, so fluid therapy is often of limited value.

Prevention: Colostrum and Management

Sporadic cases of *E. coli* septicemia are unfortunate events, but endemic neonatal calf losses resulting from this disease demand a thorough evaluation of management regarding dry cows, periparturient cows, and newborn calves. There are two basic questions that require answers: (1) Are newborn calves being fed sufficient volumes of high-quality colostrum soon enough after birth? And (2) is the environment likely to harbor large numbers of *E. coli* during the periparturient and neonatal period? In other words, two facets of the dairy operation must be carefully critiqued: colostrum management and the hygiene of the maternity area and neonatal calf pens. A few basic concepts regarding colostrum should be understood:

1. Maternal immunoglobulin is concentrated in the mammary gland of the dry cow via an active transport mechanism during the last few weeks of gestation. Although IgG_1 is the major immunoglobulin transferred, IgG_2, IgM, and IgA are found as well. Resultant colostrum contains IgG at much higher concentrations than maternal serum, and transfer of maternal antibody into colostrum temporarily decreases maternal serum IgG_1 levels. Colostrum should be obtained as close to the time of calving as possible; colostrum collected more than 2 hours after calving shows a significant reduction in total IgG concentration.
2. A minimum of 40 dry days and a maximum of 90 dry days result in the best quality colostrum.
3. Assume dry cows that leak milk before parturition or collection of colostrum have lost the "best" colostrum.
4. Holstein calves must ingest at least 100 g of IgG_1 in the first 12 hours of life for adequate passive transfer

of immunoglobulins. The immunoglobulin concentration in colostrum deemed "acceptable" ranges from 30 to 60 g IgG/L; obviously, if larger volumes of more dilute colostrum are fed, adequate immunoglobulin mass would then be provided. However, many dairy calves simply allowed to nurse dairy breed dams to satiety will not voluntarily ingest an adequate volume of colostrum to meet their required immunoglobulin intake.

5. Certain genetic lines of cattle may be prone to low immunoglobulin levels in colostrum. For example, beef cattle tend to have higher levels than Holsteins. This may reflect genetic selection or merely reflect the dilutional effects of the greater milk volume in dairy cattle.
6. A colostrometer (a hydrometer that measures specific gravity of fluid, thereby indirectly measuring solids and, it is hoped, immunoglobulin concentration) is a common on-farm tool used to assess colostrum quality. Colostrometer readings may be affected by the colostrum temperature; whereas higher temperatures underestimate quality, lower temperatures overestimate quality. Therefore, readings should be made when the colostrum is at room temperature (20° to 25°C). That aside, there is considerable overlap in specific gravity readings among colostrums with low and high immunoglobulin concentration. Previous recommendations state the hydrometer should have a colostrum specific gravity reading of 1.050 or greater at room temperature for adequate immunoglobulin levels. However, given the large number of variables that affect colostrum-specific gravity (e.g., protein concentration, lactation number, cow breed, and temperature), use of the 1.050 cutoff value will misclassify many (up to two thirds) poor-quality colostrums as acceptable.
7. Other measures of colostrum quality and immunoglobulin mass are being increasingly deployed on dairy farms. An alternative to the use of a colostrometer is indirect assessment of IgG concentration in colostrum by Brix refractometry. A Brix reading of 21% to 23% correlates with the target value of 50 g IgG/L, and colostrum with a Brix refractometer value less than 18% should be categorized as being of poor quality. Recent research suggests that Brix-refractometry is the most accurate way of identifying colostrum quality (at least in terms of IgG concentration) on farm. Direct measurement of colostrum IgG concentration is also possible using commercially available immunoassays. One of these cow-side immunoassay kits (Colostrum bovine IgG quick test kit, Midlands Bio-Products, Boone, IA) has been demonstrated to identify poor-quality colostrums (those with IgG concentrations <50 g/L) with 93% specificity; in other words, this test appears to be superior to the hydrometer in accurately identifying poor-quality colostrum.
8. Volume or weight of colostrum at first milking. Weighing the colostrum is a simple method of selecting likely higher quality colostrums. In a large study of Holstein cows, first-milking colostrum weighing <8.5 kg (18.7 lb) was shown to have significantly higher colostral IgG_1 concentration than colostrums weighing >8.5 kg. By discarding (or feeding to older calves) Holstein colostrums that weigh more than 8.5 kg, a producer might increase the percentage of high-quality colostrum being fed to calves. Not surprisingly however, the weight or volume of colostrum is a less sensitive indicator of IgG content than other methods such as refractometry and so should only be relied on when other methods are unavailable.
9. Pooled colostrum from each cow's first milking may not ensure adequate immunoglobulin content because the poor-quality (dilute) colostrums tend to lower the immunoglobulin concentration of the entire pool. The practice of pooling colostrum also may increase *Mycobacterium avium* subspecies *paratuberculosis* (MAP), *Salmonella* Dublin and leukemia virus infection rates.
10. Microbiologic quality and cleanliness of colostrum. An increasing volume of data suggests that bacterial contamination of colostrum is a substantial problem on farm and that levels of bacteria in excess of 100,000 colony-forming units (CFU) per mL indicate a problem. Not only do high bacterial load colostrums represent an infectious disease risk, but there is also evidence that there is an increased risk of FPT when colostrum with such high levels of contamination is fed, likely linked to the binding of free IgG in the gut lumen by bacteria. The rapid proliferation or doubling time of bacteria at body temperature obligates producers to quickly refrigerate or freeze colostrum that is not to be immediately fed. Preservatives such as potassium sorbate can be added so as to increase safe storage time up to at least 96 hours at 4°C. If colostrum is not to be used within a few days, it should be frozen; more effective and rapid freezing, thereby curtailing bacterial growth, can be achieved by storage in 1- to 2-L bags rather than the conventional gallon jugs. Colostrum can become heavily contaminated with bacteria if good milking hygiene is not practiced at the first milking. Clean teats and udders, clean milking equipment, sanitized storage containers, and sanitized feeding equipment are necessary to limit the possibility of colostrum becoming a culture medium for pathogenic bacteria, including *Salmonella* spp. and virulent strains of *E. coli.* McGuirk and Collins have provided goals for bacterial contamination of colostrum:
    - Total bacteria: <100,000 colony forming units (CFU)/ml
    - Fecal coliforms: <10,000 CFU/ml
    - Other gram-negative bacteria: <50,000 CFU/ml

- Streptococci (non-*Streptococcus agalactiae*): <50,000 CFU/ml
- Coagulase-negative *Staphylococcus*: <50,000 CFU/ml

Increasingly, larger farms are investing in the specialized equipment necessary for pasteurization of colostrum for the removal of infectious pathogens. Given the physical and chemical characteristics of colostrum, it is probable that careful quality control over time will be necessary to ensure that the pasteurization equipment is maintaining its efficacy. Initial data suggest that pasteurization, when performed properly, can be an excellent tool, increasing adequacy of passive transfer rates and reducing preweaning morbidity from diarrhea.

11. Springing heifers' colostrum has traditionally been considered lower quality than older cows. However, based on immunoglobulin concentration, colostrum from heifers is comparable with cows beginning their second lactation. In theory, the younger heifers have less immunologic "experience" than cows, so it is possible that the antibody "spectrum" (the number of different antigens to which antibodies are produced) is less in heifers than cows. However, the impact of this theoretical issue on calf health remains unproven. Until contrary data are made available, heifer colostrum should be evaluated and administered on the same basis as cow colostrum.

Given this current summation of colostral quality research, the following recommendations are made for newborn dairy calves:

1. High-quality colostrum cleanly collected from Johne's disease–negative and BLV-negative cows may be stored for use. If not fed within 2 hours of milking, colostrum should be refrigerated in sanitized 1- or 2-L containers until fed; as mentioned earlier, use of larger containers may limit prompt cooling, thereby promoting bacterial overgrowth. Fresh colostrum may be refrigerated for no more than 1 week and frozen for up to 1 year. If frozen, thawing should be performed slowly in warm water. Microwave thawing, if done carefully, can be used to thaw colostrums without overheating and denaturation of colostral antibodies. However, if microwave thawing is used, thawing for short time periods, periodically pouring off the thawed liquid and using a rotating tray may help to limit overheating.
2. Calves should receive 4 L of high-quality first milking colostrum during the first 12 hours of life (3 L is often sufficient for Jerseys, Shorthorns, and Guernseys). The first 2 L should be fed within 1 to 2 hours after birth, and the second 2 L should be fed before 12 hours of life. Many operations have chosen to feed all 4 L by esophageal feeder in a single feeding to larger calves.
3. The exact means of feeding (nipple vs tube) is less important than the timing of colostrum feeding, the volume fed, and the total immunoglobulin mass contained in that volume of colostrum.
4. Passive transfer status can be tested directly by radial immunodiffusion (RID) or indirectly by measurement of serum total protein levels or Brix refractometry per centage. Between 1 and 7 days of age, adequate passive transfer is indicated by a serum IgG concentration >1000 mg/dl and a serum total protein >5.5 g/dL or a Brix refractometry reading of at least 8.4% and preferably closer to 10%. If the serum sodium sulfite turbidity test is used, use of the 1+ endpoint (turbidity in 18% solution) as an indicator of adequate passive transfer status will maximize the percentage of calves correctly classified by this assay.
5. Regular evaluation of passive transfer status of all newborn calves in a herd allows the veterinarian to objectively monitor colostrum management over time. An on-farm testing method that has been scientifically validated (e.g., serum total protein, immunoassay kit, RID, or sodium sulfite turbidity test) should be used. On a herd basis, producers should aim for greater than 75% (i.e., at least 9 of a sample size of 12) of all calves tested between 2 and 7 days of age to demonstrate a total protein of greater than 5.5 g/dL when maternal colostrum is the means of passive transfer. Periodic quality control checks of conventional or Brix refractometers (against distilled water or sugar solution standards for calibration) is advised.
6. Calves born prematurely or from difficult births may have a variety of physical problems (e.g., swollen tongue) and physiologic disturbances (e.g., hypoxia) that may impact their ability to suckle colostrum or absorb immunoglobulins from the gut. Special attention should be paid to these calves to ensure adequate colostrum ingestion, and subsequent testing of serum is recommended to allow early detection and correction of FPT. For valuable calves believed to be at high risk by virtue of dystocia or phenotype (e.g., large in vitro fertilization calves), IV plasma (where available) or whole-blood transfusion from an appropriately screened donor may be considered in the first day of life. Calves fed colostrum of questionable quality, those not receiving their first colostrum feeding until after 12 hours of life, and calves of exceptional value warrant testing for FPT. Calves found to have FPT should receive a 40-mL/kg dose of commercial plasma or whole bovine blood or plasma from their dam or from a BLV- and BVDV-negative cow.
7. Esophageal feeders and bottles used to feed colostrum must not be used for older or ill calves and must be disinfected and dried between uses.
8. Colostral supplements and replacers are frequently used in place of, or in addition to, colostrum. Some of these products may provide IgG concentrations that are reportedly adequate but do not supply IgA, vitamins, maternal leucocytes, growth factors, and so on, that are normally present in colostrum. Furthermore, these products often do not result in serum protein or immunoglobulin levels equal to that of colostrum-fed calves when used per label. Some of these products, especially those marketed as supplements, contain very low immunoglobulin mass. Although colostrum replacers containing

immunoglobulins derived from serum, milk, colostrum, or eggs provide IgG for newborn calves, none appear to be equal or superior to natural colostrum when used as a replacement. Use of such products has recently been implemented in certain herds as a tool to limit transfer of infectious agents to the calf via colostrum, such as MAP, the causative agent of Johne's disease, the bovine leukosis virus, *S.* Dublin, and *Mycoplasma bovis.* The most commonly used colostrum replacers in the United States will not reliably achieve the total protein or total solids cut-off point of 5.5 g/dL in the serum in a 2- to 7-day-old calf when used per label, so expectations will need to be lowered if a testing policy is in place. Adequate total protein and IgG concentrations can often be achieved when additional doses or bags of such replacement products are used, but with this comes an increased financial cost. Recent research suggests that when 200 g of IgG (two packets) of a common colostrum replacement product is administered to newborn calves, antibody titers to common viral pathogens reach comparable levels and persist for at least as long as those seen with approximately 4 L of maternally derived colostrum. It is our opinion that the mixing of supplements or replacement products with maternal colostrum as a theoretical means of boosting neonatal IgG levels should be avoided. Although unproven, we believe there might be an increased risk of acute *Clostridial* spp. diarrhea when these are fed together.

In addition to the possibility of decreased efficiency of IgG absorption, maternity (calving) pen management practices that predispose to *E. coli* septicemia must be corrected. The importance of maternity pen hygiene cannot be overstated because no level of passive immunoglobulin transfer can protect completely against gross filth in the environment, and conversely, even calves with partial or complete FPT may survive when cleanliness is exceptional. Dry cows should not be kept in filthy environments that allow heavy fecal contamination of the coat and udder. Maternity stalls or calving areas should be cleaned, disinfected, and adequately bedded between uses by different cows.

Newborn calves should be removed from the calving area as soon as possible after birth because they will inevitably incur fecal–oral inoculation as they attempt to stand and nurse. Ideally, calves should be moved from the maternity area into individual hutches without being allowed to contact one another. This may not be feasible on larger dairies with limited manpower. In such situations, a small "safe pen" for calves can be constructed adjacent to the maternity pens. A safe area is a sheltered, fenced-in, well-drained, bedded concrete-floor pen located in or near the maternity area. These typically measure approximately 20 ft by 20 ft. Walls should be constructed to prevent contact with cows or bedding from the maternity area. This small area can be cleaned and disinfected daily with relative ease, and fresh bedding can be added. A large gate to facilitate cleaning with a bucket loader should be installed at one end of the safe pen to facilitate efficient (and therefore regular) removal of all bedding before cleaning and disinfection, which should be rigorous and regularly scheduled. This pen becomes the holding area for all newborn calves in the maternity area. Personnel on the dairy are made responsible for moving newborn calves into the safe pen as soon as possible after birth; use of gloves and footbaths will aid in the prevention of pathogen spread via boots or clothing. Subsequently, calves are less likely to become rapidly inoculated with pathogens from the maternity area. The calves are kept here until the calf attendant can provide colostrum and move them to hutches. It is critical that the safe pen be disinfected regularly and not be used as a long-term housing area for calves, or accumulation of pathogens is inevitable. On large dairies, particularly those experiencing high calf morbidity and mortality problems in the first 2 weeks of life, it may be cost-effective to dedicate one employee to the maternity pen whose sole responsibility is the prompt removal of newborn calves and near immediate colostrum administration. Only larger dairies will be able to implement this because 24-hour coverage will be necessary to monitor all calvings.

Enterotoxigenic *Escherichia coli*

Etiology

ETEC produce enterotoxins that cause secretory diarrhea in the host intestine. ETEC are characterized by the presence of both specific adhesins and by the production of specific toxins. The adhesins of relevance in farm animal species are all encoded by fimbrial operons and are referred to as F-4, F-5, F-6, F-17, and F-41. Several types of enterotoxins have been identified, and a single ETEC may be capable of producing one or more enterotoxins. Both heat-labile (LT I, LT II) and heat-stable (STa, STb) enterotoxins have been identified in ETEC infections. In calves, ETEC producing the low-molecular-weight STa cause the majority of neonatal diarrhea problems.

Pathogenic ETEC must be able to attach to the host enterocytes to create disease. Once adhered, the organism releases enterotoxin, which induces the intestinal epithelial cell to secrete a fluid rich in chloride ions. Water and sodium, potassium, and bicarbonate ions follow chloride, creating a massive efflux of electrolyte-rich fluid into the intestinal lumen. Although some of this fluid is reabsorbed in the colon, the efflux of secreted fluid exceeds the colonic capacity for fluid absorption, and watery diarrhea results.

Because enterotoxins are nonimmunogenic, efforts to control ETEC in calves have centered on inducing antibody against fimbrial proteins. The fimbrial adhesins that allow pathogenic ETEC to attach to enterocytes are proteins that initially were categorized with the capsular (K) antigens. Currently, classification of fimbrial adhesins as F antigens helps to avoid confusion, but the literature still occasionally refers to K-99, K-88, and so forth rather than the current designation, F-5 and F-4, respectively. In calves, F-5 (K-99) has received the most attention regarding diagnostics and vaccine production for calves. However, ETEC possessing other fimbrial antigens or multiple fimbrial antigen types including F-6, F-17, and F-41 are capable of causing diarrhea in calves. Current literature suggests that F-4 is not associated

with diarrhea in calves and that F-17 is found most often in diarrheic calves compared with other fimbrial antigen types, although there is some doubt as to the pathogenic role of F-17, and some sources maintain that it requires other fimbrial antigens to be present to be pathogenic. F-5 and F-41 currently have the strongest association with the presence of diarrhea and are currently considered the most common and significant of the ETEC adhesins. Some ETEC possess several types of fimbriae, and both F-41 and F-5 types may be isolated from an individual diarrheic calf. Colostrum possessing passive antibodies against a specific ETEC fimbrial type will protect newborn calves against that specific F type but will not cross-protect against others. Current and old nomenclature alongside toxin type elaborated for several common ETEC are given in Table 6.1.

As is the situation with nonpathotypic *E. coli* septicemia, ensuring prompt feeding of adequate levels of colostrum is extremely important to protect calves against ETEC. However, because of lack of cross-protection against various fimbrial antigens, even calves with excellent passive transfer are at risk for ETEC with F types other than those against which the dam has provided colostral antibodies. Colostrum containing antibodies against specific F types will prevent attachment of homologous ETEC to calf enterocytes by coating the fimbriae binding sites. Therefore, colostral protection is a local effect of IgG in the gut. To be effective, colostrum containing antibodies against ETEC F antigens must be fed as early in life as possible, lest ETEC colonize the gut before colostrum has been consumed. Although one experimental design showed colostral F-5 antibodies to be effective up to 3 hours after experimental oral challenge with ETEC F-5, it is more practical to assume colostrum should "beat" the ETEC to the gut. Other management factors in addition to colostral feeding are also important in the pathogenesis of ETEC diarrhea. Conditions that allow or encourage buildup of ETEC in the dry cows, in the maternity or neonatal calf facilities, or in stored colostrum increase the risk of ETEC diarrhea, as is true with *E. coli* septicemia. Marrow products in colostrum may also decrease the risk of ETEC by binding to toxin receptors or preventing proliferation of pathogenic bacteria.

Affected calves are usually 1 to 7 days of age, with most cases seen in calves less than 5 days of age. For example, calves are most susceptible to F-5 ETEC during the first 48 hours of life and thereafter begin to build resistance to the organism. Concurrent infection with rotavirus may extend the age of susceptibility to ETEC diarrhea to approximately 10 days of age. In older calves, continued exposure to heavy inocula of pathotypic ETEC may result in intestinal colonization and shedding of the organism in normal or diarrheic stools, thereby facilitating new infections in other neonates.

TABLE 6.1 Fimbrial Antigens for ETEC

Designations		
New	**Old**	**Toxin**
F-4	K-88	LT I
F-5	K-99	STa
F-6	987-P	STa
F-41		Sla

Clinical Signs

These signs vary from mild diarrhea with resultant spontaneous recovery to peracute syndromes characterized by diarrhea and dehydration that progress to shock and death within 4 to 12 hours.

Because of the multitude of ETEC types and variability in their pathogenicity, as well as the influence of passive transfer, individual farms may have sporadic or endemic problems resulting from ETEC. Mild disease is common on many farms and seldom is brought to a veterinarian's attention. These calves have loose or watery feces but continue to nurse (Fig. 6.4). Spontaneous recovery or apparent response to the farmer's favorite "calf-scour" treatment (usually an oral antibiotic) is the rule. Owners usually call for veterinary assistance only when peracute cases develop, a high morbidity is apparent, calves fail to respond to over-the-counter (OTC) medications, or death in neonatal calves is experienced.

Peracute cases may produce dehydrated, weak, and comatose calves within hours of the onset of the disease.

• **Fig. 6.4** A 5-day-old calf with mild "calf scours" caused by enterotoxigenic *Escherichia coli* (ETEC). The perineum, tail, and hocks are stained by watery or soupy diarrhea. This type of diarrhea could be caused by enteric pathogens other than ETEC, and clinical signs are not specific for diagnosis.

Historically, these calves usually have nursed normally and appeared healthy until signs develop. Dehydration and weakness are the predominant signs (Fig. 6.5). Mucous membranes are dry, cool, and sticky. The suckle reflex is weak or absent.

Most peracute cases show evidence of voluminous diarrhea (Fig. 6.6), with watery feces coating the tail, perineum, and hind legs. Some calves with peracute disease may not have diarrhea; however, the pooling of fluid in the intestinal lumen creates abdominal distension, and fluid splashing sounds can often be detected by simultaneous auscultation and ballottement of the right lower abdominal quadrant. Mild, transient colic may be noted early in the disease course. Bradycardia and cardiac arrhythmia accompany the systemic signs in some peracute cases and result from hypoglycemia, hyperkalemia, or both. Atrial standstill has been documented in some bradycardiac calves with hyperkalemia. Rectal temperatures usually are normal or subnormal if the calf is recumbent.

Differentiation of *E. coli* septicemia from peracute ETEC infection often is difficult in the field setting because of several commonalities, including overlap in age at presentation alongside peracute onset and profound depression. In prodromal, peracute ETEC cases, the presence of massive fluid in the intestine, as evidenced by abdominal contour, simultaneous auscultation and ballottement, or abdominal ultrasonography, is a key indicator that the characteristic voluminous diarrhea is impending. Furthermore, on resuscitation with IV fluids, ETEC cases typically break with voluminous diarrhea, and provided the concurrent abnormalities in hydration, electrolyte, acid–base, and glucose status are addressed properly with IV fluids, calves with ETEC typically show rapid clinical improvement. In contrast, calves with *E. coli* septicemia show less voluminous diarrhea, and diarrhea typically develops later in the disease course. Also, in contrast to ETEC cases, calves with *E. coli* septicemia typically fail to demonstrate a dramatic clinical response to fluid resuscitation. Neonatal salmonellosis, especially that associated with *Salmonella* Dublin acquired in the immediate postnatal period, is another important differential.

• **Fig. 6.5** Patient with peracute enterotoxigenic *Escherichia coli* diarrhea that is recumbent, extremely dehydrated, and has severe metabolic acidosis.

Acute ETEC cases show obvious watery diarrhea, progressive dehydration, and weakness over 12 to 48 hours. The character and color of the feces vary as well, but feces usually are voluminous, watery, and yellow, white, or green. Such calves may have low-grade fever or normal temperatures and deterioration in the systemic state and suckle response. Continued secretory diarrhea gradually worsens the hydration and electrolyte deficiencies; weight loss is rapidly apparent, especially if fluid intake is decreased by reduced suckling.

Translocation of bacteria from the gut into the systemic circulation is an uncommon event when ETEC is the sole agent involved because these organisms do not invade the deeper layers of the gut wall and they incite minimal intestinal inflammation. Therefore, evidence of localized infection (e.g., hypopyon, arthritis) is uncharacteristic of ETEC infection and more indicative of colisepticemia or septicemia secondary to other enteric diseases, especially salmonellosis.

Clinical Pathology

Peracute infections resulting from ETEC cause severe secretory diarrhea that results in a classic metabolic acidosis

• **Fig. 6.6** Peracute enterotoxigenic *Escherichia coli*) diarrhea with voluminous diarrhea. The calf had a plasma bicarbonate level of 6 mEq/L, a systemic pH of 6.98, and a potassium level of 8.4 mEq/L. After sodium bicarbonate therapy, the calf made a quick recovery.

with low plasma bicarbonate and low venous pH. Hyperkalemia and hypoglycemia also are characteristic. Hyperkalemia results from efflux of K^+ from the intracellular fluid in exchange for excessive hydrogen ions in the extracellular fluid (ECF). Reduced renal perfusion also contributes to retention of K^+ in the ECF. Mild hyponatremia and hypochloremia are inconsistently present. Dehydration is generally greater than 8%, and corresponding elevations in packed cell volume (PCV) and total protein are typical. The WBC count usually is normal, although elevated numbers of WBC may be present because of extreme hemoconcentration, and stress leukograms occasionally are discovered. Leukopenia, left shifts, and toxic cytologic changes in neutrophils are uncommon in ETEC infections, and those findings more likely support *E. coli* septicemia or salmonellosis.

Hypoglycemia is more likely to be present if the interval between feedings is prolonged; this finding is not present in all peracute cases. Blood values for a typical case are shown in Table 6.2. Mild azotemia resulting from prerenal causes (reduced renal perfusion) is common and should be kept in mind when use of potentially nephrotoxic drugs is considered in these patients.

Diagnosis

The diagnosis is suggested by the calf's age, physical signs, and laboratory data. Peracute ETEC may be difficult to differentiate from *E. coli* septicemia and salmonellosis in neonatal calves based on clinical signs alone. Response to appropriate fluid therapy strongly supports ETEC infection, as does confirmation of adequate serum immunoglobulin in the patient.

Definitive diagnosis requires isolation or demonstration of an *E. coli* possessing pathogenic F antigens that allow intestinal attachment in calves having typical clinical signs. When submitting diagnostic samples, the clinician should indicate that ETEC infection is a possibility and should request typing of *E. coli* isolates for F antigens (by immunofluorescence, slide agglutination, or polymerase chain reaction [PCR]) and, if available, for enterotoxin (by PCR or, rarely, by ligated gut loop assays). In fatal cases, ETEC can be cultured from the ileum; a section of ileum should be tied off, placed in a sterile container, and transported on ice packs to the laboratory. Isolation of ETEC from diarrheic feces of older calves (>5 days of age) is generally considered to merely reflect the presence of the pathogen in the calf population on that particular farm. In such cases, fresh specimens of jejunum and ileum should be examined carefully for histologic evidence of attachment of ETEC to enterocytes. These findings suggest participation in enteric disease by ETEC rather than simple intestinal colonization by the organism. Obtaining samples for culture before antibiotic therapy, particularly when oral antibiotics are being given, is an important factor in the diagnostic workup of a potential ETEC outbreak.

Histologic examination of fresh samples of ileum and jejunum of affected calves greatly aids in confirming the diagnosis of ETEC infection. Sections of ileum should be cut into 2- to 3-cm lengths and then split longitudinally and swirled in 10% neutral buffered formalin solution to aid in rapid fixation of the mucosa. Samples for histology should not be tied off because this delays fixation of the mucosa. In classic ETEC infection, a dense population of gram-negative rods is found adherent to the mucosa of the ileum.

Because mixed infections with combinations of ETEC, rotavirus, coronavirus, and *Cryptosporidium* spp. are common, feces or intestinal contents should also be analyzed for viral and protozoal pathogens. Salmonellosis also must be included in the differential diagnosis because many types of *Salmonella* spp. can cause severe diarrhea, dehydration, shock, and acid–base disturbances similar to ETEC. Fever, neutropenia, and a left shift are more commonly observed in *Salmonella* patients. In addition, enterotoxemia resulting from *Clostridium perfringens* must be considered, especially in peracute cases with abdominal distension but no diarrhea. Calves with clostridial enterotoxemia may be weak, dehydrated, or "shocky" but seldom have as dramatic a metabolic acidosis as that found in ETEC infections.

TABLE 6.2 Laboratory Data from a Typical Peracute Enterotoxigenic *Escherichia coli* Infection in a 3-Day-Old Holstein Calf

Variable	Measured Value	Reference Range
Na^+	127	132–150
K^+	8.1	3.9–5.5
Cl^-	104	97–106
HCO_3^-	12	20–30
Total CO_2	10	26–38
Ven pH	7.09	7.35–7.50

Treatment

Appropriate replacement and maintenance fluids constitute the primary therapy for ETEC infection in neonatal calves. Correction of metabolic acidosis and hypoglycemia and reestablishment of normal hydration status are imperative. Calves with peracute signs or those that are recumbent require IV therapy. Calves that can stand but show obvious dehydration, have cool and dry mucous membranes, and have a reduced or absent suckle reflex also should initially be given IV therapy. Calves that are ambulatory and have a good suckle response usually can be treated with oral fluids.

Assessment of concentrations of required electrolytes based on subjective clinical parameters rather than objective laboratory tests is empirical but sometimes necessary in field situations. Therefore, rules of thumb include:

Recumbent calves 12% to 15% dehydrated; base deficit 15 to 20 mEq/L

Weak calves 8% to 12% dehydrated; base deficit 10 to 15 mEq/L

Ambulatory calves 5% to 8% dehydrated; base deficit 5 to 10 mEq/L

These rules of thumb are not absolute, and chronic low-grade bicarbonate loss or increased D-lactate production in the gut may create profound acidosis over a period of days in a calf having only minimal signs of dehydration. A 40-kg calf that is judged 10% dehydrated will need 4 L of fluid simply to address current needs. For all calculations of replacement electrolytes, a 50% ECF will be assumed for neonates. Therefore, a 40-kg calf will be assumed to have 40 x 0.5 = 20 L ECF compartment. If this 40-kg calf has a venous plasma bicarbonate concentration of 10 mEq/L, and 25 to 30 mEq/L is the desired normal level, then 15 to 20 mEq of bicarbonate must be replaced in each liter of ECF. Therefore 20 L x 15 mEq = 300 mEq would be necessary to correct the bicarbonate deficit associated with the metabolic acidosis. Total CO_2 of venous blood also may be used to calculate base deficits in lieu of HCO_3 values.

A great deal of research data as well as strong individual clinical opinion exist as to the most appropriate composition of initial fluid therapy for ETEC infections in calves. An effective solution, first proposed by Dr. R.H. Whitlock, is formulated by adding 150 mEq of $NaHCO_3^-$ to 1 L of 5% glucose. This combination is used for the initial 1 to 3 L of IV therapy, depending on the severity of measured or suspected metabolic acidosis. Glucose corrects hypoglycemia if present, and both bicarbonate and glucose facilitate potassium transport back into cells, thereby lessening the potential cardiotoxicity associated with hyperkalemia. Some reports minimize the importance of hyperkalemia and suggest using IV potassium in the initial fluid. The authors of these papers emphasize that dehydrated calves having severe ETEC-induced secretory diarrhea have a total body K^+ deficit despite having an elevated ECF potassium concentration. Although this may undoubtedly be true, it seems risky to tempt fate by administering K^+-containing solutions as the initial therapy for a patient known to be hyperkalemic. This is especially true for a patient with bradycardia or arrhythmias because death has occasionally occurred when potassium-containing fluids have been given as initial therapy. After plasma K^+ and HCO_3^- levels are quickly improved by the initial 1 to 3 L of 5% dextrose with 150 mEq $NaHCO_3^-$/L, potassium-containing fluids can be safely used. Balanced electrolyte solutions such as lactated Ringer's solution suffice for maintenance fluid needs, but supplemental $NaHCO_3^-$ and dextrose may be required to address continued secretory losses and anorexia. Response to treatment usually is dramatic in calves with secretory ETEC diarrhea. Calves that were initially recumbent usually appear much improved after administration of 2 to 4 L of appropriate IV fluids and usually can stand within 6 hours and begin to nurse within 6 to 24 hours of the initiation of therapy. This type of prompt response strongly suggests a correct diagnosis and tends to rule out septicemia because septicemic calves seldom respond so promptly, if at all. Depending on the setting (field vs clinic), maintenance or intermittent IV fluid therapy may be continued or replaced by oral fluids in those calves that quickly regain a suckle response and are eager to eat.

Antibiotic therapy for peracute ETEC infections remains controversial, with current concerns focused on antimicrobial residues in meat and indiscriminate and unnecessary use of antimicrobials leading to resistance. However, in peracute cases, the overlap of many clinical signs with colisepticemia often prompts the clinician to include antimicrobial treatment in the therapeutic regimen. In a Canadian study, diarrheic calves 5 days of age or younger were found to be at significantly greater risk of bacteremia than older calves; this age range obviously includes calves at risk for ETEC infection. Furthermore, in cases with fever and severe debilitation, the veterinarian is often prompted to consider the possibility of complicating conditions such as bacterial pneumonia. In his thorough reviews of the subject, Constable found published evidence supporting the logic and clinical efficacy of antimicrobial use to all calves that exhibit systemic signs of illness or that have blood or mucosal casts in their stool. Extra-label drug use can be justified because the health and potential survival of the patient is threatened, and there is a paucity of approved, labeled claims for the treatment of neonatal diarrhea. In the United States, however, clinicians must always be mindful of federal law that prohibits extra-label third-generation cephalosporin use and limits the use of fluoroquinolones to specific clinical indications. Because of the challenges in differentiating ETEC, *E. coli* septicemia, and peracute salmonellosis, clinicians are often faced with patients in whom it may be impossible to clinically differentiate true bacteremia from merely lumenally confined enteric bacterial infections when making therapeutic decisions. Parenteral antimicrobials that are bactericidal with a gram-negative spectrum therefore have intuitive appeal for undifferentiated cases. In patients with pure ETEC infections, oral antimicrobial treatment offers the potential benefit of reducing the number of ETEC in the gut, and by reducing the source of enterotoxin, one might also reduce the drive for hypersecretion and ameliorate the metabolic consequences of the enteric infection. Oral amoxicillin trihydrate (10 mg/kg orally every 12 hours) or amoxicillin trihydrate–clavulanate potassium (12.5 mg combined drug/kg orally every 12 hours) for at least 3 days for treatment of such cases is justifiable if the ETEC strain is believed or proven to be susceptible to the antibiotic. Repeated use of these products over the long term is likely to induce resistance and cause further disruption of the intestinal microbiome; therefore, long-term efforts must focus on prevention rather than treatment. Recommended treatments for diarrheic calves with signs of severe systemic illness (e.g., fever and weakness that persist after fluid resuscitation) include ceftiofur, amoxicillin, potentiated sulfonamides (25 mg/kg IV or intramuscularly [IM] every 24 hours) or ampicillin (10 mg/kg IM every 12 hours). Systemic antibiotics usually are continued for 3 to 5 days based on the calf's clinical response, temperature, and character of the feces.

Data from across the world increasingly report that many ETEC cases that result in high calf mortality have limited antibiotic susceptibility, and therefore sensitivity testing or mean inhibitory concentration (MIC) levels should always be determined when the herd history or clinical data suggest high morbidity and mortality from ETEC.

Feces usually remain more watery than normal for 2 to 4 days. If diarrhea persists beyond this time, concurrent infection with other organisms is likely. Other treatments for peracute cases may include flunixin meglumine (1.1–2.2 mg/kg IV every 24 hours) for potential endotoxemia, resolution of fever, and reduction of pain associated with fluid-filled bowel. Repeated dosages of this product carry the risk of renal and GI injury because continued use of flunixin meglumine interferes with vasodilatory prostaglandin synthesis in the gut and kidney.

Milk or milk replacer should be withheld for no more than 24 to 36 hours, during which time a high-quality oral electrolyte energy source may be fed several times (four to six times) daily. Holding ETEC-infected calves off milk or replacer for prolonged times creates weight loss from inadequate energy intake and places calves at risk of starvation. Even though many oral electrolytes are supplemented with dextrose as an energy source, no commercial oral electrolyte solution provides enough energy for maintenance needs, especially for dairy calves in hutches during winter weather. Weight will be lost, and starvation may occur if these electrolyte solutions are fed as the only ration for more than 1 or 2 days. In highly valuable calves undergoing hospitalization, treatment with parenteral nutrition offers an excellent option to at least approximate maintenance calorific needs while the calf is nil per os (NPO). Calves with ETEC are so significantly catabolic that they will still lose weight despite calorific supplementation with IV lipid and amino acids. Careful monitoring of blood glucose for hyperglycemia and strict attention to aseptic technique, as well as catheter and fluid line maintenance, are important when administering parenteral nutrition. Consequently, it is rarely practical outside of a referral hospital.

The alkalinizing potential of oral electrolyte solutions is of great importance, especially when those solutions are used as ongoing therapy for peracute cases after initial IV fluids or when those solutions are used as sole therapy of less severely affected calves having ETEC. Continued HCO_3^- loss accompanying secretory ETEC diarrhea must be anticipated and treated. Therefore, oral electrolyte solutions containing bicarbonate or some other alkalinizing anion are most helpful. The optimal oral electrolyte solutions typically possess 70 to 80 mEq of alkalinizing potential per liter (typically as bicarbonate, propionate or acetate), dextrose, and electrolytes; these should be fed at 4 to 6 L/day. Oral electrolyte solutions that when mixed with water are nearly isotonic are preferred over those that are markedly hypertonic.

Concerns regarding adding oral electrolyte solutions to milk or milk replacers revolve around the alkalinizing solutions' tendency to interfere with abomasal clot formation. Therefore, oral electrolytes are fed during separate feedings at least 30 minutes before or after a milk feeding. Calves do not digest sucrose effectively, and addition of table sugar to "home remedy" electrolyte mixtures will reliably worsen fluid and electrolyte losses in diarrheic stools. After 24 to 36 hours of oral electrolyte treatment, calves may be fed small volumes of milk or milk replacer. Calves that respond rapidly to initial fluid resuscitation can be started back on small volumes of milk or milk replacer at an earlier time. During recuperation, calves should be deeply bedded in dry straw (Fig. 6.7) or similar bedding material and provided shelter from rain and snow. When milk feedings are resumed, feedings are best performed in small volumes frequently. If this is not possible, whole milk or replacer should be divided into at least two to three daily feedings. Supplemental oral electrolyte solutions can be continued if ongoing fluid and electrolyte losses are assumed to result from continued diarrhea, and these solutions should be fed at intervals between whole milk or replacer feedings. Unless the calf is hypoglycemic or acidotic, isotonic electrolyte solutions are preferred because they allow a more normal abomasal transit than do hypertonic solutions.

• **Fig. 6.7** A Holstein calf in deep straw bedding. This depth of bedding is appropriate for recovery and normal "nesting" behavior of calves in winter weather. The lower limbs are not visible from outside the pen. (Courtesy of Dr. Sheila McGuirk.)

Treatment of acute ETEC infections in calves that are ambulatory and still able to suckle may not require IV therapy. Cessation of whole milk or replacer feeding coupled with substitution of oral electrolyte–glucose solutions for 24 to 36 hours may be sufficient. Bicarbonate loss and resulting metabolic acidosis should not be underestimated, however. It is imperative to use highly alkalinizing electrolyte glucose solutions to provide 4 to 6 L of fluids per day. Parenteral antibiotics are indicated if the affected calf is febrile, and oral antibiotics may be administered when the herd medical history indicates involvement of a highly pathogenic ETEC. Milk or replacer should be reinstituted after 24 to 36 hours, and electrolyte feedings can then be used as fluid supplements in the intervals between milk feedings as needed.

Mild ETEC infections seldom require veterinary care. Spontaneous recovery is the rule, and supportive care with oral electrolyte solutions frequently is used by owners in such cases. Use of OTC remedies is widespread among dairy farmers treating mild ETEC infections or nonspecific "calf scours." Although little scientific evidence is found to justify these products, anecdotal testimonials from farmers exist for many oral products and protectants. Oligosaccharides have received increased interest in recent years, the suggested therapeutic rationale being that they provide competitive binding sites within the intestinal lumen for the fimbrial adhesins of ETEC, thereby facilitating intestinal transit and elimination. Although their administration via water has been shown to reduce intestinal coliform counts in experimental ETEC infection, there has yet to be conclusive proof of improved clinical outcomes in challenge studies. OTC calf diarrhea products that contain methscopolamine, atropine, or products that reduce intestinal motility are contraindicated and may cause bloat and ileus if overdosed. Bismuth subsalicylate is palatable and can be used safely in calves. Most recently, an OTC crofelemer extract Neonorm™, a natural product with antisecretory properties, was shown to significantly increase fecal dry matter of neonatal calves with experimentally induced enterotoxigenic *E. coli* diarrhea.

Prevention

Prevention assumes prime importance when a high morbidity rate, significant mortality rate, or both occur on a dairy farm. It is not unusual to encounter 70% to 100% morbidity and mortality when virulent strains of ETEC are present. These strains also tend to be resistant to many antibiotics. The usual situation is that the owner tries multiple OTC products on the first few affected calves and then calls for veterinary assistance to select a "better" antibiotic. One or more calves may die or require intensive therapy before a thorough investigation of the problem ensues.

The veterinarian must avoid the temptation to simply provide or suggest a "newer" or better antibiotic if the problem is to be solved. Feces must be submitted from *more than one* acutely affected calf. If necessary, bull calves should be raised in an identical manner to heifers just to allow them to develop disease and allow early sampling. A qualified diagnostic laboratory must identify the *E. coli* as an ETEC strain with specific attachment antigens and determine antibiotic susceptibility. Diagnostic efforts beyond mere speciation are vitally important.

Management must be meticulously assessed as to the cleanliness of dry cows, colostrum, feeding instruments, maternity areas, and newborn calf facilities. Evidence of successful passive transfer of immunoglobulins must be evaluated in several consecutive calves to rule out *E. coli* septicemia or poor colostral feeding as the major cause of ETEC infection. Culturing of colostrum at milking and from the bucket or bottle immediately before its feeding can be used to assess the cleanliness of colostrum milking procedures, colostrum storage, and feeding instrument hygiene. Readers are directed to the previous section on colisepticemia for more details on assessment of colostrum management. Evaluation of whole milk or milk replacer feeding and the detailed specifics by which diarrheic calves are being treated become extremely important. Mixing errors leading to hyperosmolality of oral feedings (especially electrolyte solutions) or milk replacer can add a further and compromising degree of nutritional diarrhea in some situations. It is remarkable how many extra supplements can be added to the feeding regimen on problem farms such that the end product becomes a contributor to the persistence and severity of the diarrhea. The osmolality of electrolyte solutions should be in the range of 300 to 600 mOs/L, although some solutions at the high end of this range may be problematic if ad libitum water is not available. The ad libitum provision of palatable fresh water is imperative, hyperosmolality of administered oral fluids being compounded by the lack of water by which the calf can compensate. The two biggest challenges to adequate water intake are the health or severity of illness in the calf and the freezing conditions during northern U.S. winter months. Water intake is maximal immediately after milk feeding, so at the very least fresh, warm unfrozen water should be made available shortly after milk or milk replacer intake. Measuring total solids in milk replacer, as fed, is a highly relevant part of a herd investigation as is checking osmolality of any oral rehydration solutions, again as fed. Total solids of milk replacer solutions should never exceed 18%, and it is always reassuring to be able to demonstrate consistency by virtue of a less than 1% variation from feeding to feeding. Milk replacer hygiene is just as relevant as total solids or osmolality; substantial bacterial contamination of milk, colostrum, or milk replacer because of unsanitary equipment, powder storage, or the use of unpasteurized waste milk has detrimental health consequences for all calves but especially those for whom adequacy of passive transfer was compromised. Targets established by Dr. Sheila McGuirk are a total bacterial count of less than 20,000 CFU/mL, a total coliform count of less than 1000 CFU/mL and a total *E. coli* count of less than 100 CFU/mL for pasteurized waste milk; for milk replacer, the set points are less than 10,000 CFU/mL, less than 1000 CFU/mL, and zero, respectively. Samples for testing should be sampled as fed; for example, they should be taken via the nipple in a group pen situation rather than at the time of mixing.

If an ETEC with attachment antigens such as F-5 is identified in the feces of more than one affected calf, more specific preventive measures can be instituted. Management factors including colostral feeding must be emphasized, lest preventive vaccines are looked on by the farmer as a "silver bullet" that obviates any need for management changes. When specific F antigen ETEC are involved, a commercial bacterin containing these F types can be administered to the dry cows 6 weeks and 3 weeks before freshening or at manufacturer's recommended times. Autogenous bacterin manufacturers should be required to show data on endotoxin

levels in bacterins because administration of endotoxin-rich vaccines to adult cattle can cause dramatic production losses or abortion. When vaccines are first used those calves born in the immediate 3-4 weeks following may still not receive adequate specific colostral antibody protection because of insufficient time and may be given commercially available oral monoclonal antibodies against F-5 (K99). This should only work if a commercial product is available and has been confirmed as the attachment factor for the ETEC in question. Monoclonal antibody products must be given immediately after birth before colostrum is fed (Ecolizer, Elanco Animal Health, Greenfield, IN, and First Defense, Immucell, Portland, ME). Valuable calves at risk and born to these same dry cows also may receive oral antibiotics for the first 3 to 5 days of life in an effort to prevent infection with the ETEC identified, and selection of appropriate antibiotics should be based on antibiotic susceptibility testing of the causative organism.

Rarely, a particular serotype of *E. coli* other than the F-5 type is isolated from the small intestine of scouring neonatal calves. If the organism is consistently confirmed as the pathogen (based on samples from multiple affected calves) and commercial dry cow vaccines have not altered the incidence of disease, an autogenous bacterin should be considered. However, the use of autogenous bacterins can only be justified when an absolute diagnosis of a highly pathogenic ETEC has been confirmed by isolates from several affected calves and commercial bacterins fail to stop the disease. Because free endotoxin content may be high in some autogenous vaccines made from gram-negative organisms, the manufacturer should "wash" the preparation to reduce endotoxin content, and data on endotoxin content in the final product should be requested. It is important to resist the temptation to initiate autogenous bacterin production using a nonspecific *E. coli* isolate obtained from one or more calves that merely had colisepticemia as a result of FPT.

Other *Escherichia coli* Diarrhea

Etiology

Although less common than ETEC, other forms of pathotypic *E. coli* have been identified as causes of calf diarrhea. Enteropathogenic strains are defined as those capable of attachment and effacement of intestinal cell microvilli. Attaching and effacing (AEEC) *E. coli* are EPEC that do not produce enterotoxins but may produce cytotoxins of various types. They do not possess *Shigella*-like invasiveness. These organisms have been isolated from calves with diarrhea that have histologic evidence of effacement of microvilli in the cecum, colon, and distal small intestine. Cellular degeneration may ensue if the organisms produce cytotoxins. These histologic changes enable differentiation of AEEC from ETEC that attach to enterocytes but do not cause histologic damage. Because the lesions of EPEC typically involve the large intestine, dysentery and diarrhea may be observed. Malabsorption, maldigestion, and protein loss are characteristic of disease with AEEC or EPEC. Calves from 2 days of age up to 4 months of age may be infected, and other enteric pathogens often are present concurrently. They can also be identified in the feces of healthy calves such that their role is somewhat controversial. For example, a recent large meta-analysis of the epidemiology of *E. coli* in calves demonstrated that EPEC are twice as likely to be found in healthy calves as diarrheic ones. However, cattle are frequently identified as being an important reservoir for these and other pathotypic *E. coli* in humans.

Shiga toxin-producing *E. coli* (STEC) are another type of *E. coli* that produce hemorrhagic colitis and the hemolytic uremic syndrome in humans. These organisms are often more broadly grouped within the enterohemorrhagic *E. coli* (EHEC) and occasionally have been found in sick calves. Some of these strains invade the mucosa to reside in the lamina propria of the large intestine and produce a severe hemorrhagic colitis. Ulcerative colitis with hemorrhage may be present grossly and microscopically in necropsy specimens. STEC are defined by their ability to produce at least one of the Shiga toxins, Stx1 or Stx2, and the role of these pathogens in cattle has not been conclusively elucidated, although fatal cases have been documented. Approximately 75% of human outbreaks with EHEC have been linked to bovine derived products or cattle, highlighting the importance of public health awareness for this zoonotic pathogen to the dairy industry. Enteroaggregative *E. coli* (EAEC), diffusely adherent *E. coli* (DAEC), and enteroinvasive *E. coli* (EIEC) are yet more groups of pathotypic *E. coli* with characteristic pathogenetic and histologic features that can be linked to specific, different toxins; however they are of uncertain relevance in calf diarrhea. EAEC, for example, have the ability to form mucoid biofilms and secret potent cytotoxins linked genetically to a plasmid that encodes both for fimbriae and toxins. This plasmid may be found in the same strains of *E. coli* responsible for calf diarrhea and septicemia in neonatal farm animals.

Clinical Signs

As observed with ETEC diarrhea, dehydration, depression, and weakness are common signs associated with EPEC, AEEC, and STEC (EHEC) infections in calves. Dysentery or fresh blood in the feces, when present, suggest severe colitis and distinguish the disease from ETEC secretory diarrhea. Fever tends to be more common with AEEC and STEC because of mucosal damage and erosive or ulcerative damage to the intestine. Diarrhea is profuse in some calves and intermittent but blood and mucus tinged in others. Tenesmus may be observed as a result of colonic inflammation. Blood loss in the feces may be negligible with some AEEC or severe enough to cause anemia and hypovolemic shock in some with STEC (EHEC). Dysentery or frank blood in the feces always dictates that *Salmonella* spp. be ruled out as a cause of the diarrhea because clinical signs of AEEC and STEC (EHEC) can closely resemble those found in patients with *Salmonella* infection. Affected calves usually are 4 to 28 days of age, and morbidity and mortality vary greatly.

Laboratory Data

Calves affected with EPEC, AEEC, and STEC have maldigestion and malabsorption and may have protein loss

from erosive or ulcerative colonic lesions. Therefore, total protein and the albumin fraction of serum may be low. Anemia may be present because of GI blood loss. Total WBC counts may be normal or low with a left shift. Although shock and lactic acidosis may create a metabolic acidosis in recumbent patients, calves still standing tend not to have a remarkable base deficit because the pathophysiology of their diarrhea is different than the secretory diarrhea of ETEC.

Diagnosis

The emphasis for most veterinary diagnostic laboratories is in the identification of typical ETEC fimbrial antigens when processing diagnostic samples from calf diarrhea cases. The definitive diagnosis of other pathotypic *E. coli* requires more specific analytical methods to demonstrate specific toxins such as Stx1, Stx2, and enterotoxin or to demonstrate specific in vitro adhesion or cytopathic effect. Categorization and typing of these organisms can only be performed by specialized diagnostic laboratories (e.g., Animal Diagnostic Laboratory, Pennsylvania State University, University Park, PA). Because coexisting enteric, bacterial, viral, or protozoan infection is present in most calves with AEEC or STEC, feces should be analyzed for rotavirus, coronavirus, ETEC, *Salmonella* spp., *C. perfringens* type C, and *Cryptosporidium parvum.* If the incidence of diseased calves with diarrhea is found to be high, fecal samples from several acute cases should be evaluated to ensure that the suspected AEEC or STEC is in fact consistently the cause of calf diarrhea on this farm.

Treatment

Therapy is similar to that for ETEC infection except that whole blood transfusions of 1 to 2 L of blood may be necessary in calves with severe dysentery and fecal blood loss. Ceftiofur is the most frequently used parenterally administered antimicrobial for this disease. Broad-spectrum antibiotics such as gentamicin (6.6 mg/kg intramuscular (IM) or IV every 24 hours), amikacin (15 mg/kg SC or IV every 24 hours), or trimethoprim–sulfa combinations (22 mg/kg SC, IM, IV or orally every 12 hours) are also used because of the microvillus or mucosal damage within the intestine, but these represent extra-label drug use in the United States, and aminoglycosides should only be administered after appropriate laboratory diagnostics and understanding of prolonged meat withdrawal. Prognosis is guarded for calves with AEEC or STEC infections unless intensive care is provided. Colonic, cecal, or distal ileal pathology may be so severe as to cause ulceration or perforation of the intestine in some cases. Because of the gross and histologic intestinal pathology, corticosteroids and prostaglandin inhibitors are contraindicated except when used once in conjunction with initial shock therapy because these drugs reduce cytoprotective mechanisms within the bowel.

Because of the maldigestion and malabsorption created by these organisms, oral electrolyte–energy sources may be less useful than in ETEC. These products, however, usually are recommended for at least the first 36 to 48 hours of therapy. Calves continuing to have diarrhea after 48 hours can be returned to milk or replacer feeding but may be candidates for TPN if they are valuable enough to warrant the expense.

Prevention

Because AEEC and STEC do not possess typical F-5 fimbriae, commercial dry cow bacterins and monoclonal antibodies against F-5 are unlikely to prevent future outbreaks. Therefore, management procedures should be examined carefully and corrected when found deficient. If multiple isolates confirm a single AEEC or STEC strain, autogenous bacterins administered twice during the dry period may be considered. Colostral management (hygiene and feeding) and passive transfer of immunoglobulins must be assessed.

If rotavirus, coronavirus, *C. parvum,* or other enteric pathogens are found to be concurrent problems, these should be addressed from a management and preventative standpoint. The frequent association of these pathogens with AEEC and STEC raise concern that these pathogens may be the primary cause of intestinal injury, and the AEEC or STEC in fact may be secondary.

The veterinarian responsible for herd health must consider the public health concerns associated with some AEEC or STEC. Currently, the 0157:H7 strain has caused a great deal of bad publicity for the dairy industry because cattle have been blamed as carriers of this organism that may infect people, causing severe colitis and occasionally hemolytic uremic syndrome. Therefore, sanitation, disinfection, and very careful handling of feces to avoid human infection are indicated.

Rotavirus

Etiology

Rotaviruses are members of the Reoviridae family and are classified further via complicated division into groups (serogroups), serotypes, and subgroups. The rotaviruses cause diarrhea in multiple species, including humans. Although the rotaviruses share certain antigens and cross-infection of species occurs with some strains, in general resistance is specific, and cross-protection against heterologous strains is poor. They are not zoonotic.

Calves usually are infected by group A and less commonly by group B serotypes. Initially identified by Mebus and coworkers, the Nebraska rotavirus isolate was used extensively for study and vaccine production. Other group A serotypes have been identified in the United States and abroad. Exposure to rotaviruses apparently is widespread in the cattle population based on serologic surveys. Older calves and adult cattle serve as carriers of the virus, shedding the virus intermittently in feces. In addition, up to 20% of healthy calves may shed rotavirus. As a rule, rotaviruses coexist with other neonatal enteric pathogens such as ETEC and *C. parvum* in herd calfhood diarrhea outbreaks. Experimental mixed infections of rotavirus with bovine viral diarrhea virus (BVDV) have been shown to result in more severe diarrhea than infection with either of these agents alone, suggesting some synergistic effect in pathogenicity.

Neonatal calves (<14 days of age) are at greatest risk for infection by enteric rotavirus, and most infections occur during the first week of life. Prevalence of infection in neonatal calves born on dairy farms harboring the virus is high, morbidity is high (50% to 100%), and mortality varies greatly. Clinical manifestations of disease and mortality in calves are influenced by several factors, including level of immunity to the virus, magnitude of viral inoculum, viral serotype, concurrent infection of the gastrointestinal tract or other systems, stress, and crowding. Germ-free calves infected by rotavirus have self-limiting diarrhea and rapid recovery. Infected calves in field situations may have inapparent, mild, moderate, or fatal disease. As is true with most enteric pathogens, the younger the patient, the higher the likelihood of severe disease because of losses of water, electrolytes, and body nutrient reserves secondary to diarrhea.

Rotavirus infection is limited to the small intestine and characterized by destruction of villous enterocytes and subsequent replacement of these columnar cells by immature and more cuboidal cells derived from the intestinal crypts. Although these new immature cells are resistant to further viral infection, they are unable to carry out the normal digestive and absorptive tasks necessary for villous enterocytes because of deficient disaccharidase and sodium–potassium ATPase activities. Therefore, rotavirus diarrhea is characterized by maldigestion and malabsorption. To further complicate matters, the intestinal crypt cells continue their normal secretory function, which is no longer balanced by absorptive villous function. Thus, net secretion outweighs absorption and contributes further to diarrhea. Increasing intraluminal osmotic pressure also may draw further water into the bowel as lactose and other undigested nutrients pass through the gut and are fermented in the colon to volatile fatty acids. Bacterial fermentation of undigested lactose creates both D- and L-isomers of lactic acid; in diarrheic calves, absorption of the slowly metabolized D-isomer may result in accumulation of this acid in the systemic circulation, thereby contributing to the development of metabolic acidosis and clinical depression. Water and electrolyte losses of variable severity occur in affected calves.

The level of local passive immunity conferred to calves by colostral intake somewhat determines the risk and relative severity of infection. Colostrum with a high virus-neutralizing antibody titer (≥1:1024) against rotavirus is protective against experimental infection. However, unless colostrum or colostrum–milk combinations with titers this high continue to be fed, this local protection "wears off" within a few days, and the calf becomes susceptible to infection. Colostral rotavirus specific IgA is likely most responsible for protection. Colostrum or colostrum–milk combinations with lower virus neutralizing titers may impart partial protection. Feeding of colostrum having very high levels of IgG_1 antibodies against rotavirus soon after birth may establish high circulating humoral antibodies against rotavirus. Although this humoral protection will not, by itself, protect a calf from infection, a portion of these IgG_1 antibodies is secreted back into the intestine over time and is thought to confer additional local protection against infection.

Data from numerous studies consistently demonstrate that rotavirus is either the most common or second most common infectious agent identified in calf diarrhea investigations.

Clinical Signs

No pathognomonic signs of rotavirus exist in dairy calves that allow differentiation of the disease from ETEC or other enteropathogens. In addition, infections may be subclinical, mild, moderate, or severe based on factors such as inoculum dose and serotype virulence of virus, immunity of the calf, concurrent enteric or other system infections in the calf, and other stressors.

Depression, reduced suckle response, diarrhea, and dehydration comprise the major clinical signs. Fever, hypersalivation, and recumbency may be observed in some patients. Feces usually are watery and yellow in pure rotavirus enteritis. Because mixed infections are common, however, the color, consistency, and composition of the feces vary greatly.

Signs of depression, dehydration, and shock are more likely to occur in the youngest calves (< 5 days of age) and seldom occur in calves more than 2 weeks of age unless D-lactic acid production is high. Recumbent calves usually have profuse watery diarrhea and abdominal distention of the right lower quadrant with fluid-filled small intestine.

Ancillary Data

Laboratory data are not specific enough to aid in the diagnosis of rotavirus enteritis in calves. Severely affected calves will develop a metabolic acidosis with low plasma bicarbonate. Other electrolytes and glucose values tend to be low but vary with the severity and duration of disease.

Diagnosis

Diagnosis requires identification of rotavirus particles or nucleic acid in the feces of acutely infected calves. Feces should be collected within the first 24 hours of illness and diarrhea. Until recently, feces submitted to qualified diagnostic laboratories were examined by electron microscopy (EM) to observe viral particles or subjected to testing using a latex agglutination test or an enzyme-linked immunosorbent assay (ELISA) to detect viral antigen. Fluorescent antibody (FA) stains also are available for tissue analysis from fatal cases. The accuracy of EM and ELISA tests, although high and commonly in agreement in side-by-side studies, is certainly less sensitive than PCR methods, such that the majority of diagnostic laboratories have now moved to PCR as the standard test for the diagnosis of rotavirus infection. Because of the frequency of mixed infections, feces submitted from acute neonatal diarrhea cases should be analyzed for viruses, bacteria, and *C. parvum*. Feces from more than one acute case in the herd must be tested before staking an entire prevention program on one etiologic agent. Affected calves should also be assessed for adequacy of passive transfer of immunoglobulins to rule out FPT.

Treatment

Treatment is nonspecific and generally follows therapy described for ETEC regarding indications and types.

Several differences are noted, however:

1. Because of villous enterocyte pathology, the efficiency of absorption of oral electrolyte and energy sources is likely reduced compared to ETEC infections. Obviously, this comment is relative, not absolute because generally less than 100% of the small intestinal villi are damaged. Therefore, absorption of some proportion of the glucose, electrolytes, and water that comprise the oral fluids will occur, and aggressive oral fluid therapy (4–6 L/day) is still indicated in this disease. Isotonic electrolyte replacements may be preferable unless the calf is hypoglycemic. Electrolyte solutions containing glutamate mixed with yogurt may speed intestinal recovery, although this is not proven in calves.
2. Maldigestion and malabsorption influence the duration of diarrhea and the digestibility of whole milk or milk replacers in patients with viral enteritis. When diarrhea from rotavirus becomes evident, the damage to the intestinal lining has already occurred, and only time and supportive care can allow the intestine to heal. Nutritional support is a critical component of that supportive care, particularly because rotaviral scours may persist for 3 to 7 days. Producers should be counseled that provision of milk or milk replacer is necessary in viral enteritis even though the maldigestion of the milk nutrients may contribute in part to the pathologic process. Denial (for >24 hours) of milk feeding to a calf with viral diarrhea places the calf at significant risk for cachexia and may lower its resistance to opportunistic disease. Death from starvation may occur in such cases, particularly during times of inclement weather (Fig. 6.8). To quote Dr. Chuck Guard, "If a calf scours for a week, and all that the calf is fed is oral electrolyte replacer, then that calf will be well hydrated and will have absolutely perfect blood electrolyte concentrations and acid–base balance on the day it starves to death." Producers should learn to live with the "more-in, more out" rule: The more milk that goes in the front end, the more diarrhea comes out the back end. However, this process is not necessarily harmful because digestion and absorption of some fraction of milk nutrients is likely to occur, and these nutrients are necessary to support the tissue synthesis required to return the intestine to normal.

   Any exacerbation of fluid losses and acidosis that may result from maldigestion of milk nutrients can be offset by aggressive fluid and electrolyte replacement. Ideally, the affected calf should be fed small amounts frequently with the addition of lactase-containing tablets. Because maldigestion is part of the pathophysiology, common sense suggests that dividing milk feeding into smaller but more frequent meals would allow improved digestion and lower the amount of carbohydrate reaching the colon. Excessive amounts of carbohydrates reaching the colon may worsen diarrhea, systemic metabolic status especially D-lactic acidosis, and the clinical status of the calf.
3. Maturation of immature villous replacement cells of crypt origin will allow the intestinal tract to return to normal within several days to 1 week in most patients that recover.

• **Fig. 6.8** A 3-week-old Red and White Holstein calf with chronic diarrhea and emaciation caused by rotavirus and *Cryptosporidium* infection. The calf was normally hydrated and had normal electrolytes but was deteriorating because of malabsorption and maldigestion and cachexia. This is one of the first calves we successfully treated using parenteral nutrition (in 1982).

Intravenous fluid therapy is necessary for recumbent, extremely dehydrated, or "shocky" patients and patients that have lost their suckle reflex. It is best guided by acid–base and electrolyte determinations. If this is not practical or available, however, the most severely affected calves with acute diarrhea should be assumed to have metabolic acidosis, low bicarbonate, high potassium, and low glucose values. Guidelines for fluid therapy are available in the section on treatment of ETEC. Parenteral nutrition may be "lifesaving" in calves with cachexia.

Although there is no need for antibiotic therapy in pure rotaviral enteritis, the likelihood of mixed infections and the pathologic damage to enterocytes that fosters attachment of bacterial pathogens may be reason enough to treat severely affected calves with systemic antibiotics.

Control

Rotavirus is ubiquitous in cattle populations; therefore, management procedures that decrease the magnitude of exposure of neonatal calves to rotavirus must be the focus of preventive efforts. Cleaning maternity pens between calvings, immediately removing the calf from the dam (and thus exposure to feces), placing the calf in an individual hutch that has been cleaned and relocated since removal of the last occupant, and feeding the calf from its own nipple bottle or pail rather than a common feeding device all help reduce spread of viral pathogens. Feces from

a clinically diseased calf may contain hundreds of millions of viral particles per gram and can contaminate inanimate objects and workers' feet, clothing, and hands to be passed to a naïve calf. The use of a safe pen can also be considered (see discussion in section on colisepticemia).

Vaccination of newborn calves or dry cows is somewhat controversial because passive humoral immunoglobulins derived from colostrum probably are not as effective as passive local immunoglobulins derived by continued feeding of colostrum or colostrum–milk combinations that contain high antibody levels against rotavirus. Oral modified-live virus (MLV) vaccination of newborn calves before feeding them colostrum has been practiced. However, it is somewhat cumbersome for most management teams and risks bacterial infection and FPT because colostrum is withheld until several hours after the MLV oral vaccination to prevent inactivation of the vaccine by colostral antibodies. Although neonatal calf vaccination can induce cell-mediated immunity and secretory IgA and IgM against rotavirus of vaccine serotype, efficacy in field studies has been questioned. The utility of PCR assays as the favored diagnostic test for the diagnosis of rotaviral diarrhea is also complicated when MLV vaccines have been administered to neonates. Viral nucleic acid of vaccinal origin can give false-positive results for a length of time that likely exceeds false-positive results given by EM or ELISA. The precise length of time that the vaccinated calf will remain fecal PCR positive is unknown, but this eventuality should be considered probable for a minimum of 3 days after vaccination.

Because colostrum, colostrum–milk combinations, or milk containing virus-neutralizing antibodies with a titer of 1:1024 or greater will protect the gut from infection by local means, feeding such material to calves for the first 14 to 30 days of life usually will prevent rotavirus infection. This also requires that the serotype of rotavirus to which the calves are exposed be the same as that from which the colostral antibodies have been derived. It also requires that management prevent overwhelming exposure of neonatal calves to challenge with this or other combined infections.

Boosting the level of rotavirus antibody in colostrum is a potential means to prevent enteric rotavirus infection if calves are fed adequate to large amounts of colostrum to achieve local protection. If colostrum is only fed for 1 or a few days, the local protective effect will "wear off," and the calf will become susceptible to rotavirus enteritis. Continued feeding of colostrum is ideal but often not practical. Initial postnatal ingestion of very high antibody-containing colostrum may in fact create high enough humoral antibody levels to establish high levels of secretory IgG_1 antibodies in the gut. Boosting the level of colostral antibodies against rotavirus usually is done by vaccinating the dry cow with MLV or killed vaccines containing rotavirus and coronavirus, sometimes in combination with ETEC antigens. Currently, the killed products generally are recommended, and the dry cow should be vaccinated 6 and 3 weeks before freshening (or according to manufacturer's recommendations) and subsequently given booster shots each year, no less than 4 weeks before freshening. No vaccine or antibody can overcome massive viral challenge, and conversely, less concern for passive protection is necessary when management excels at reducing risk for the newborn calf. Given the practical limitations and expense and impracticality of continued colostrum (or colostrum supplement) feeding of calves, the producer should focus on initial colostrum administration to newborns, maternity pen and hutch hygiene, dry cow vaccination, and controlling spread by fomites and personnel. Incidence of rotavirus diarrhea has been decreased on some farms and reported in two clinical trials by mixing some colostrum (10%) with milk or replacer for up to 30 days.

Being a nonenveloped virus, rotavirus is stable in the environment (6 months in fecal matter) and relatively resistant to the effects of some disinfectants. Decontamination of hutches and maternity pens requires thorough physical efforts to remove fecal matter and other organic debris because most disinfectants show reduced, even negligible, activity in their presence. Application of appropriately diluted bleach, a phenolic, or a peroxysulfate disinfectant to a thoroughly cleaned solid surface, with provision of long (> 10 minutes) contact time and subsequent sunlight exposure and drying, will effectively reduce the number of infectious rotavirus particles. Heavily soiled areas, such as the ground beneath calf hutches, may need to be stripped down to the packed surface and exposed to sunlight and dry conditions for several days to weeks (depending on weather conditions) before being considered habitable for the next calf.

Coronavirus

Etiology

Based on seroprevalence studies, the bovine coronavirus (BCoV) responsible for calf diarrhea is quite prevalent in U.S. cattle herds, as is rotavirus. There is much debate among researchers at this point as to whether BCoV isolates obtained from calf and adult diarrhea cases are the same virus or distinct from those that have been incriminated in respiratory disease outbreaks in feedlot and dairy calves. Whether or not there are antigenic or genomic differences in BCoV strains that mediate different organ tropism is similarly unclear. Winter dysentery in adult cattle has been associated with BCoV, and the same strain that causes diarrhea in calves has been used to experimentally create winter dysentery in adult cattle. Therefore, the upper age limit of susceptibility to infection by this agent is apparently longer than traditionally thought.

Although not as common as rotavirus as a cause of viral enteritis in dairy calves, coronavirus has been identified in neonatal calf diarrhea outbreaks, especially in the winter months and with mixed infections. A number of studies indicate that clinical disease associated with BCoV in calves is more severe than rotavirus, with higher mortality rates. Affected calves tend to be slightly older than calves infected with pure ETEC or pure rotavirus. They average 7 to 10

days of age at onset, with some observed as late as 3 weeks of age. The virus causes a severe enterocolitis characterized by villous enterocyte destruction in the small intestine and destruction of both ridges and crypts in the large intestine. Maldigestion, malabsorption, and inflammation all contribute to the pathophysiology of coronavirus diarrhea in calves. The virus is cytolytic, and affected villous enterocytes in the small intestine are replaced by cuboidal cells from the crypts, but the colonic lesions leave denuded mucosa in affected areas of the colon. The severity of this damage helps explain why coronavirus enteritis, unlike rotavirus, may cause some flecks of blood to appear in the stool and can kill calves even in a germ-free isolation facility. Thus, in the natural setting, coronavirus enteritis creates a severe clinical diarrhea and can also be associated with > 50% mortality when combined with other viral, bacterial and *C. parvum* infections.

Clinical Signs

Acute, severe diarrhea, as well as dehydration, reduced appetite or suckle reflex, and progressive depression and weakness are typical, albeit nonspecific, signs of coronavirus infection in calves. Because of the colonic pathology, mucus and flecks of blood may be quite apparent in feces. Coronavirus is also commonly found in the respiratory tract of young calves, and a pneumonia–enteritis complex may occur in these individuals.

Ancillary Data

Coronavirus enterocolitis causes variably severe changes in acid–base and electrolyte status that are also common to *E. coli* and rotavirus. In severe coronavirus infections or mixed infections that include coronavirus, metabolic acidosis and low plasma bicarbonate are the rule. Potassium values vary with the severity and duration of the diarrhea and acidosis. Hemoconcentration secondary to the diarrhea elevates PCV and total protein values. Leukograms are variable. Although nonspecific, the acid–base and electrolyte assessments are of greatest value for individual patient management.

Diagnosis

Submission of feces from calves with acute or peracute diarrhea provides the best diagnostic approach for live patients. Feces collected during the first 24 hours of diarrhea are best. Electron microscopy, ELISA, or PCR may be used to detect virus. As with rotavirus, PCR testing has generally superseded antigen or electron microscopy as the diagnostic test of choice. FA testing of tissue samples obtained from both the small and large intestines is advised for necropsy specimens, principally because one is usually uncertain of the etiologic cause at the time. The spiral colon has, however, been demonstrated to be the best diagnostic sample source at postmortem for specific identification of coronavirus infection. Because of the cytolytic nature of coronavirus, the virus can disappear rapidly from tissue. Therefore, chronically affected calves are not good candidates for etiologic diagnostic sampling.

Treatment

Treatment principles are the same as those previously listed under ETEC and rotavirus treatment. As with rotavirus, oral electrolyte–energy sources may be less efficiently absorbed in coronavirus infections because of enterocyte loss. However, even given these limitations, oral electrolyte–energy sources may contribute to the patient's well-being during the time of intestinal repair. Diarrhea is likely to persist to some degree for 1 week with coronavirus because of the severe enterocolitis. Systemic antibiotics are often indicated to help affected calves cope with secondary bacterial infection of the lung, gut, and other systems.

Control

Every effort should be made to control management factors that predispose calves to infection. These are described in the control of rotavirus. Because coronavirus is an enveloped virus, its persistence in the environment and resistance to disinfectants are considerably lower than those of rotavirus. Dry cows should be vaccinated at 6 and 3 weeks before calving with a killed rotavirus and coronavirus vaccine and boosted each year thereafter at 4 weeks prepartum. Because it is assumed that local antibody is more important than humoral antibody, the feeding of colostrum containing high antibody levels against coronavirus is advantageous, and when possible, prolonged feeding of such colostrum during the first 30 days of life might confer greater protection for problem farms. Active immunization of calves at birth with multivalent products containing coronavirus (usually in combination with *E. coli* and rotavirus) is also quite commonly practiced but should be considered a less reliable and effective method of protecting calves compared with the absorption of high IgG_1 concentration colostrum from immunized dams. Specific antibody products are available and can be administered to newborn calves at birth. One such product contains K-99 antibodies and coronavirus antibodies (First Defense, ImmuCell Corp., Portland, ME) derived from hyperimmune bovine colostrum. In an experimental challenge study with BoCV, dairy calves fed a commercial product containing spray-dried bovine serum showed increased feed intake and higher scores for certain clinical parameters as compared with control calves. The expense of such products is considerable, and discussion of the cost and therapeutic benefit is often warranted before use. Most recently, a modified-live coronavirus vaccine (Bovilis Coronavirus, Merck Animal Health, Madison, NJ) for intranasal administration to calves was shown to decrease the incidence and severity of scours.

Cryptosporidium Infection

Etiology

Cryptosporidiosis is an important cause of diarrhea in neonatal calves that occurs most commonly from 5 to 28 days of age. Cryptosporidiidae are a family of coccidian protozoans grouped with the Sarcocystidae and Eimeriidae families in the suborder Eimeriina. Similar to other coccidia, members of the Cryptosporidiidae family have both

sexual and asexual components to their life cycle but differ from other coccidia in having less host specificity. *Cryptosporidium* spp. are much smaller than *Eimeria* spp. and are therefore difficult to detect in fecal flotation. Laboratory techniques that use acid-fast stains or immunologic techniques greatly aid detection. The true prevalence and pathogenicity of cryptosporidiosis in calves have only recently been appreciated.

At the time of the original description of the parasite in a calf, cryptosporidiosis was thought to be a novel or sporadic infection that most likely affected immunocompromised calves. During the 1980s, it became apparent that the organism was much more prevalent, epidemic to endemic on many farms, and a primary or component cause of neonatal calf diarrhea.

There are currently 30 named species of *Cryptosporidium* in humans and animals, but their taxonomy and nomenclature are confusing. Recent molecular methods rely heavily on single-stranded ribosomal RNA (ssrRNA) sequencing for species identification, but one can still find genotype assignments as a form of taxonomic nomenclature in the literature. Molecular and experimental evidence suggests that humans and cattle are hosts for 14 and 13 of these species, respectively, although there will likely be increases in these numbers in the future (the number of new species identified is growing at about 1 per year). However, not all of these potentially infective species are frequent parasites of clinical importance in either host species. In cattle, the majority of infections are associated with *C. parvum, C. bovis, and C. andersoni*. Zoonotic transmission from cattle to people is most commonly caused by *C. parvum*, and neither *C. bovis* nor *C. andersoni* are considered as major a human health risk as *C. parvum*. *C. parvum* can infect calves, lambs, young pigs, foals, humans, and other species such as suckling rodents. Public health concerns regarding spread of *C. parvum* from animals to people are real and require diligence in the diagnosis and management of this parasite.

C. parvum is an intestinal pathogen of preweaned calves, but both *C. andersoni* and *C. bovis* are more likely to be found in the feces of calves postweaning (highest prevalence is in calves from weanling age to yearlings). However, neither of these two species is typically associated with clinical disease in cattle. *C. andersoni* is located within the peptic glands of the abomasum rather than the intestine, where, at least histologically, it can cause thickening of the abomasal folds. *C. pestis*, *C. ubiquitum*, *C. suis*, *C. hominis*, and *C. ryanae* are all species that have been identified in cattle by ssrRNA methods, but their relevance as bovine pathogens is uncertain.

In cattle, neonates are at greatest risk of infection and disease because age-related resistance seems to be strong; this trend is less evident in humans. Veterinarians, students, technicians, and other individuals involved in handling affected calves, feeding equipment, bedding, or even clothing from in-contact individuals may develop clinical disease if strict hygienic measures are not followed. Immunocompetent hosts usually develop self-limiting diarrhea. However, the organism causes a particularly devastating disease in immunocompromised hosts, in whom persistent infections can occur.

The organism usually infects via the fecal–oral route, but contaminated ground water, improperly treated municipal water, and contaminated feedstuffs can induce infection. The infective dose of *Cryptosporidium* likely varies among individual animals and humans, but the infective dose in a susceptible individual may be less than 100 oocysts. Given that infected calves may shed millions of infective oocysts in each gram of diarrheic stool, there is strong potential on many farms for accumulation of massive infectious challenge. *C. parvum* is the most common species seen in calves up to 1 month of age and the only species typically found in calves younger than 3 weeks of age. Large epidemiologic surveys from diagnostic laboratories in the United States demonstrate that more than one third of submissions from diarrheic calves in the first month of life have detectable cryptosporidial oocysts.

Sporulated oocysts are readily infective to neonatal calves and release sporozoites that infect primarily the small intestinal (but some colonic) enterocytes by infecting the microvillus brush border. A parasitophorous vacuole that resides adherent to the cell but outside the cytoplasm is formed. The life cycle phases of *C. parvum* then result in destruction of cells as the parasitophorous vacuoles break to release merozoites that infect other host cells. The subsequent sexual life cycle phase results in formation of oocysts infective to susceptible hosts. Villous atrophy, villous fusion, and inflammation of intestinal crypts ensue. Autoinfection within the intestine occurs, wherein specialized oocysts are released to infect other enterocytes without exiting the host. Clinical signs of diarrhea reflect a mixed pathophysiology of maldigestion, malabsorption, and osmotic effects with or without secretory and inflammatory factors. The autoinfection process has been hypothesized to account for occasional protracted or relapsing cases that can result in cachexia. The prepatent period for *C. parvum* is 3 to 4 days, although the majority of clinical cases are not detected until at least 7 days of age unless there is massive, immediate postpartum exposure. Damage to the microvilli appears to predispose the calf to combined infections with *E. coli*, viruses, or *Salmonella* spp. Therefore, it is unusual in dairy calves to find only *C. parvum* when investigating endemic calf diarrhea. However, because oocyst shedding typically begins with the onset of clinical signs and persists until several days after diarrhea resolves, fecal testing may tend to reveal this pathogen more consistently than rotavirus or coronavirus, which are shed early in the disease course and for a shorter period of time than *C. parvum*. This reiterates the importance of testing affected calves early in the disease course when investigating the etiology of calf diarrhea. Combinations of enteric pathogens in neonatal calves complicate treatment, worsen the clinical signs and prognosis, tend to result in higher mortality, and predispose to malnutrition.

C. parvum may by itself produce severe diarrhea in immunocompromised calves and those exposed to inclement weather or poor nutrition.

Clinical Signs

Diarrhea, dehydration, and reduced appetite are the major clinical signs and thus do not differentiate *C. parvum* infection from bacterial and viral enteropathogens in neonatal calves. Morbidity tends to be greater than 50% in calves less than 3 weeks of age, but the mortality rate is low unless mixed infections occur or supportive treatment is less than adequate. When *C. parvum* is the only pathogen, diarrhea usually persists for up to 7 days, but most calves do not lose their ability to nurse or their interest in nursing during this time. When mixed infections occur, dehydration, acid–base and electrolyte abnormalities, and dysentery are possible. Malnutrition is a possible sequela to *C. parvum* infections when poor supportive therapy and poor nutritional quality coexist with the rather chronic diarrhea. Malnutrition is quite common in *C. parvum*–infected calves raised outside in hutches during winter weather extremes in northern climates. Because these calves normally have greatly increased caloric needs over calves raised at moderate temperatures, maldigestion, malabsorption, and fluid losses greatly compromise their well-being.

Diagnosis

Microscopic identification of *C. parvum* oocysts has been relied on historically for positive diagnosis but may require a trained microscopist! In most instances, standard flotation on feces from acutely affected calves is performed, but very fresh necropsy tissue samples of the ileum and colon also may be examined after tissue preparation and staining. Acid-fast stains are commonly used to assist in the identification of *C. parvum.* Immunofluorescence, ELISA, and PCR are all believed to be more sensitive and specific than microscopy. The antigen-based techniques probably detect all of the species documented to be infectious in calves, as do the immunofluorescence tests, but do not discriminate among species. PCR methods detect lower levels of infection than antigen-based tests, the former being sensitive to as low as 50 oocysts per gram of feces. Fecal flotation and acid-fast staining probably are only sensitive down to 10^3 to 10^6 oocysts per gram. This latter fact is not too problematic diagnostically because many clinically affected animals are shedding very high numbers ($\geq 10^6$). Because they are of similar size, microscopy cannot distinguish oocysts of *C. parvum* from *C. andersoni* or *C. bovis*. Genetic analysis of bovine or human isolates may be performed to aid in epidemiologic investigations, particularly when zoonotic cases are suspected. Even when *C. parvum* is suspected and confirmed, mixed infections should be considered likely and feces submitted for bacteriologic and virologic evaluation.

Treatment

Treatment is almost always merely supportive and consists of fluids by whatever route indicated by the severity of clinical dehydration. In addition, a high-quality source of nutrients such as whole milk or a quality milk replacer should be continued to be fed if at all possible. Mixed infections with other primary enteropathogens may necessitate a short period of feed withdrawal. If oral electrolyte energy sources are fed during the acute phase of diarrhea, they should not remain the only source of nutrients for more than 24 hours. Thereafter, milk or replacer should be fed at least twice daily and oral electrolyte–energy sources fed between milk feedings to compensate for the fluid losses caused by *C. parvum* diarrhea. During cold or extreme winter weather, hutch-sheltered or neonatal calves left outside should receive milk or high-quality replacer at least three times daily if twice-daily feeding fails to maintain body condition or *C. parvum* diarrhea, maldigestion, and malabsorption interfere with efficient utilization of nutrients.

Antibiotics are not necessary, although they may be indicated in mixed infections that include bacterial pathogens. Many drugs have been tested for efficacy against *Cryptosporidium* spp., but none has been found to be completely effective or economically justifiable. Standard coccidiostats are ineffective with the exception of lasalocid at doses so high as to be toxic to calves. Recent work in Europe and Canada has demonstrated that treatment with halofuginone lactate will reduce oocyst shedding and delay the onset of diarrhea in calves, but similar to many other investigated therapies, it does not significantly impact the incidence or severity of diarrhea in treated calves compared with control animals. Paromomycin, nitazoxanide, azithromycin, and a few other drugs have shown some activity against *C. parvum*, and ongoing research to benefit human patients with AIDS might drive future discoveries in this area. These drugs could potentially be used in valuable calves with cryptosporidiosis. Halofuginone (100 μg/kg) for 7 days is approved in several countries for the preventive treatment of cryptosporidiosis in calves, and although this will decrease oocyst shedding, it may not reduce diarrhea. In a report by Dr. Ollivett, treatment of *C. parvum*–infected calves with nitazoxanide reduced the duration of oocyst shedding and improved fecal consistency. Considerable research effort toward the development of effective vaccines over the past decade has yet to prove successful. Egg yolk antibody products for passive protection of new born calves, as well as recombinant subunit *C. parvum* vaccines for active immunization of dry cows have been closely examined but with inconsistent results. There is no currently available passive or active immunization protocol for this disease.

Control

Because specific treatment is not possible, prevention assumes supreme importance. Unfortunately, many dairy farms fail to effectively control *C. parvum* when environmental contamination becomes extreme. Although diseased calves serve as the primary source of environmental contamination, oocysts are also spread by movement of laborers, equipment, and animals. Given the low dose of oocysts necessary

to cause disease in susceptible animals, the morbidity rate can become unacceptably high. Therefore, control requires a careful, open-minded reexamination of all management practices related to calf rearing, including maternity pen hygiene; colostrum management; cleaning of feeding equipment, hutches, and the ground surrounding hutches; labor allocation; and the order by which laborers feed and handle calves.

First, calf facilities should include individual calf housing rather than grouping. Ideally, newborn calves should be separated from the dam at birth and moved to a cleaned and disinfected calf hutch on new dry ground or concrete. Placing a calf in a hutch on the same ground as that used for the previous occupant will not work because *C. parvum* oocysts can persist for months in such areas, and the ground beneath used calf hutches is often heavily contaminated. Bedding from within and around hutches should be completely removed and disposed of, the ground stripped bare, and the bare ground allowed several days to weeks of sunlight exposure under dry conditions before being considered habitable for another calf. Extremes of temperature can more rapidly inactivate the oocysts, such that infectivity is lost at 40°C and at −22°C. Moving cleaned hutches to new ground or placing them on concrete slabs that can be cleaned, disinfected, and allowed to dry between calves is the best technique. Because *Cryptosporidium* oocysts are highly resistant to the effects of almost all disinfectants, hutches and feeding equipment must be vigorously and thoroughly scrubbed with soap and water and rinsed well with hot water to physically dislodge oocysts. Drying and ultraviolet light are relatively effective against the oocysts; therefore, more hutches should be made available than would be occupied at any given time; a 20% vacancy rate for newly scrubbed hutches will often allow ample time for sun exposure and drying of recently emptied hutches. The role that moisture can play in encouraging oocyst persistence has been further demonstrated by epidemiologic studies that find that a slope of 5% to 10% in the calf housing area will diminish risks of infection, but rainfall will increase shedding rates. A peroxygen-based disinfectant (Virkon-S, Antec International, Sudbury, Suffolk, United Kingdom) has been shown to reduce the infectivity of *Cryptosporidium* oocysts under experimental conditions and is currently used in some veterinary hospitals to disinfect thoroughly cleaned surfaces. Newer generation, peroxygen-based compounds have also shown promise for reduction of oocyst viability.

Calves should have individual feeding implements, and removal of manure from the hutch should be done in such a way that calves are not exposed to manure from neighboring calves or hutches. When doing chores, laborers should move from young calves to older calves and from healthy to sick calves to limit spread of oocysts. It should also be remembered that calves raised in inclement weather, those kept on wet bedding, or calves receiving inadequate nutrition causing failure to gain weight or even loss of weight in the first week of life are more susceptible to having clinical disease after *C. parvum* infection.

Salmonellosis (Calves)

Etiology

Salmonella is a genus of gram-negative, facultative anaerobic, rod-shaped bacteria belonging to the family Enterobacteriaceae. Two species are recognized: *Salmonella enterica*, which is further divided into six subspecies, and *Salmonella bongori*. More than 2600 *Salmonella* serovars, differentiated by their antigenic composition, have been identified to date. Serovar classification is based on O (somatic), H (flagellar), and Vi (capsular) antigens. Most current serogroups are divided by O antigens and listed by capital letters (e.g., A, B, C, D and E). The majority of cattle isolates are *Salmonella* of types B, C, and E, which are non–host specific, or *Salmonella* Dublin (type D), which is host-adapted to cattle. The vast majority of serovars with veterinary and human medical significance belong to *Salmonella enterica* subsp. *enterica* and are typically referred to by their serovar classification for convenience (e.g., *Salmonella enterica* subsp. *enterica* serovar Newport, is conventionally abbreviated to *Salmonella* Newport). Despite the diversity of serovars, relatively few are responsible for a large proportion of clinical infections among cattle and other mammals. Many of these serovars are capable of causing clinical disease in a broad range of host species (e.g., Enteritidis, Typhimurium, and Newport). However, others are almost exclusively associated with a single host species (host-restricted serovars, such as Abortus ovis in sheep) or have a predilection for a particular host species but can also cause disease in other hosts (host-adapted serovars, such as *S.* Dublin in cattle).

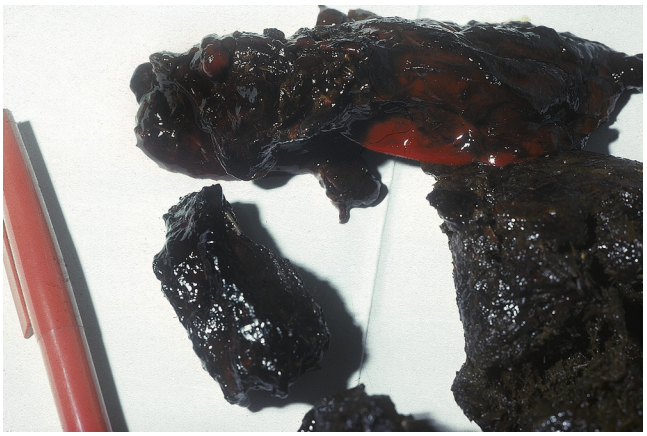
Although many infections remain subclinical, *Salmonella* is an important cause of disease among calves and adult dairy cattle. Salmonellosis is costly for dairy producers because of treatment and labor expenses, mortality, reduced milk yield, and increased cull rates. Salmonellosis as a sporadic cause of diarrhea has been long recognized in cattle, but intensive management systems have contributed to endemic disease in dairy calves, veal calves, and adult dairy cattle. Currently, salmonellosis ranks as one of the two most important bacterial causes of diarrhea in adult dairy cattle (MAP being the other) and has surpassed *E. coli* in this respect in calves on many operations. *Salmonella* spp. also pose a formidable threat to human health, causing approximately 1.2 million illnesses, 20,000 hospitalizations, and 400 deaths annually in the United States alone. Antimicrobial resistance among *Salmonella* isolates magnifies the problem, limiting treatment options and increasing the risk of therapeutic failure. In humans, infection with resistant *Salmonella* strains has been associated with adverse patient outcomes. Such human health relevance continues to heighten and accentuate the scrutiny of antimicrobial use in food-producing animals.

Some broad characterization of the clinical syndromes associated with certain serotypes can be made. With group B infections such as *S.* Typhimurium, as well as with many group E infections, a herd outbreak of diarrhea and

septicemia may occur in adults and calves. Abortion or early embryonic death may occur as a result of acute endotoxemia and shock. However, as the population develops immunity to the agent, clinical signs often dissipate within 1 to 2 months. Infections with group C *Salmonella* are more difficult to characterize because infection and clinical disease may persist in the herd for variable periods of time. Infection may also be perpetuated over the long term by environmental contamination or by group C *Salmonella* cycling through rodents, birds, or insects. Infection with the host-adapted *S.* Dublin (the most common group D isolate from cattle) is characterized by establishment of a higher percentage of carrier animals in the population. When *S.* Dublin is established in a population, some adults experience asymptomatic infection and may serve as shedders (even in milk and colostrum), and pneumonia, septicemia, and acute death become the primary manifestations of disease in calves. Some calves that develop infection and survive will become long-term carriers. Sporadic abortions may also occur with *S.* Dublin.

Calves with acute, chronic, or carrier intestinal infections shed the organism to varying degrees in their feces; this serves as the major source of infection to other naive calves via fecal–oral transmission. Calves with peracute or acute disease often are septicemic and may shed organisms from other secretions such as saliva and urine. Fecal–oral transmission is the norm for types B, C, and E *Salmonella*; infection of the distal small intestine, cecum, and colon ensues. Mucosal injury causes maldigestion, malabsorption, and loss of protein and fluid. A secretory component to the diarrhea also is thought to contribute to further electrolyte and fluid depletion. *S.* Dublin is unique in that respiratory signs may predominate, and transmission may occur via several secretions as well as feces. Adult dairy cattle may be carriers and harbor *S.* Dublin in the intestine or mammary gland. Milk, colostrum, or feces from infected or carrier *S.* Dublin cows can be infective to calves. Clinical epidemics of many *Salmonella* types, including *S.* Dublin, are common in calves in the northeastern United States and other parts of the country. Geographic differences in serogroup prevalence do occur, but widespread transport of calves or adult cows and assembly of herds from distant locations has tended to negate geographic limits for various *Salmonella* serogroups.

Factors that adversely affect the normal enteric flora tend to favor growth of *Salmonella*, which are common, albeit low in number components of the GI flora of carrier or "normal" cattle. Parturition, transport, concurrent disease, anesthesia, and withholding of feed and water are just a few of the stresses that cause intestinal ileus, reduced host immunity, or shifts in enteric bacterial populations that induce proliferation of *Salmonella.* In calves, antibiotics that alter the intestinal flora may also favor the growth of *Salmonella.* When shedding of large numbers of organisms occurs in a carrier animal, naive calves are at increased risk if crowding, poor sanitation, use of common feeding implements, or



• **Fig. 6.9** Blood in the stool from a calf with salmonellosis.

comingling, concurrent diseases, or stress are present. Both humoral and cellular immune mechanisms are involved in resistance to *Salmonella.* Calves persistently infected (PI) with BVDV are at high risk for developing acute salmonellosis following exposure to the organism.

Clinical Signs

Fever and diarrhea are the hallmark signs of clinical salmonellosis in dairy calves. Fever may precede clinical signs of diarrhea but seldom is detected before calves begin to show diarrhea and appear ill. Fresh blood and mucus in the feces (some calves may have blood in feces before diarrhea) also are common with *Salmonella* enteritis (Fig. 6.9). Blood-stained mucus or whole blood clots may be apparent based on the severity of infection and *Salmonella* type. Clinical signs associated with septic physitis, arthritis, meningitis, and pneumonia may be seen in some calves. Sporadic or endemic disease may occur, and although calves from 2 weeks to 2 months are most commonly affected, those of any age may develop the disease. Newborn calves can develop severe disease in the first few days of life when either the maternity area or calf housing facility into which they are born or moved is massively contaminated. This can be a feature of herd outbreaks of clinical salmonellosis when parturient adult and sick fresh cows are shedding excessive numbers of organisms into the environment or when calf housing and common use equipment becomes heavily contaminated due to a large number of clinical cases. Because of the true carrier status and the ability to pass the organism via colostrum and milk from asymptomatic dams, special consideration should be given to *S.* Dublin as a differential when clinical signs suggestive of salmonellosis occur in calves during the first week of life on a farm. Tremendous variation in clinical severity of disease exists based on the virulence and infecting dose of the *Salmonella* serotype in question, and the age, immune status, and existence of concurrent disease in the calf. Type E *Salmonella,* such as *S.* Anatum, tend to cause mild signs of diarrhea and fever with variable morbidity and low mortality, but types B and C are

• **Fig. 6.10** One day's death toll of neonatal calves from a dairy farm suffering high mortality in cattle of all ages during an epidemic caused by a highly virulent *Salmonella* Typhimurium strain.

more likely to cause high morbidity and variable mortality based on strain, exposure dosage of organisms and the immunologic status of the calf. Neonatal calves have a greater risk of death caused by *Salmonella* spp. because of septicemia and fluid losses leading to severe dehydration and electrolyte imbalances (Fig. 6.10).

Peracute septicemia resulting from *Salmonella* types B, C, and D may cause death before diarrhea becomes obvious. These calves rapidly dehydrate into their intestinal tract, have abdominal distension as a result of filling of the small and large intestine and sometimes forestomach. They may die secondary to bacteremia or endotoxemia induced by release of cell wall products of this gram-negative infective agent. Bacteremic calves shed large numbers of *Salmonella* organisms in other bodily secretions and feces and quickly contaminate premises. Acute cases caused by types B, C, and E show classical acute diarrhea, often with fresh blood and mucus in the feces, as well as fever and dehydration. Feces are foul smelling (septic tank odor) and vary in color and consistency, with the most virulent strains causing profuse watery diarrhea with whole blood clots present. The infected calves are frequently bacteremic, and pneumonia, arthritis, physitis, and meningitis may occur.

Acute disease in calves usually is associated with high morbidity and variable mortality that is dictated by the interaction of strain virulence and calf immunology. Fecal contamination of the environment is especially problematic when calves are housed in group housing, raised slatted stalls, or crowded areas, or when born in a stall used both as a maternity pen and sick cow area. Milk and feces from adult cows shedding *Salmonella* and contaminated feeds or feeding devices are all potential sources of *Salmonella*. Chronic infection leads to long standing or intermittent diarrhea, weight loss, hypoproteinemia, and failure to thrive. Some chronically infected calves typically evolve from epidemics of acute salmonellosis in dairy calves and thus enhance the risk of exposure for naive herdmates.

Acute infection associated with *S.* Dublin may be much harder to diagnose because diarrhea may not be the principal sign. Fever, depression, and respiratory signs may be most obvious in acute *S.* Dublin infections in calves. Although diarrhea may be present, it is seldom the predominant clinical sign, which may lead to the erroneous assumption that a calf pneumonia problem exists on the farm. Fever and depression unresponsive to antibiotics may be observed. Abortions may also occur on the premises. Calves infected with *S.* Dublin are typically 4 to 8 weeks of age although it can also be seen in neonatal calves and group-housed calves postweaning. Clinical signs of acute salmonellosis caused by *S.* Dublin in 4-week or older calves are most commonly respiratory in nature combined with depression caused by acute bacteremia, hematogenous pneumonia and septic shock.

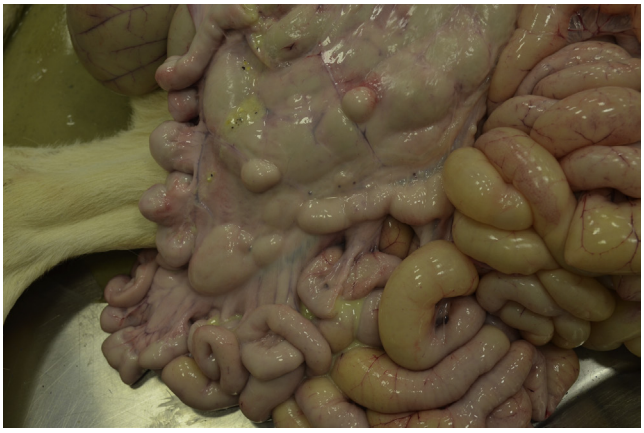
Laboratory Data

Peracute or acute infection with *Salmonella* has variable effects on the patient's leukogram. A degenerative left shift with neutropenia and band neutrophilia is considered typical for severely affected animals, but it is not consistent, and many patients have neutrophilia or normal leukograms. Although blood may be present in the feces, hemoconcentration tends to mask mild anemia resulting from any blood loss. PCV is normal or elevated because of dehydration. Total protein and albumin concentrations are usually normal or low because of protein loss into the gut and malabsorption. Renal function may be compromised by dehydration, reduced renal perfusion, endotoxemia, or nephritis secondary to bacteremia with renal infection. Peracute and acute infections cause inflammatory, secretory, and malabsorption–maldigestion types of diarrhea and result in metabolic acidosis and hyponatremia and hypochloremia. These electrolyte changes are particularly common in instances when the calf loses electrolytes and water in the diarrheic stool but is only allowed access to water for rehydration. Potassium may range from high (peracute) to low (subacute, chronic) depending on severity and duration of diarrhea, subsequent fluid losses, and acid–base status.

The leukogram in *S.* Dublin–infected calves is extremely variable and reflects duration of infection. Acute cases may be neutropenic with a left shift, severely neutropenic, or have normal WBC counts. Subacute or chronic *S.* Dublin infections have a mild to moderate neutrophilia or stress leukogram.

Diagnosis

Regardless of the type or strain of *Salmonella,* isolation of the organism, coupled with history and clinical signs, confirms the diagnosis. Fecal cultures are the standard test necessary to identify serotypes other than *S.* Dublin, but fecal, blood, transtracheal wash, or lung tissue samples may be necessary to identify *S.* Dublin. Fecal samples submitted from suspect calves should be sent to qualified diagnostic laboratories equipped to culture enteric pathogens. When neonatal calves are involved, the laboratory should be forewarned that



• **Fig. 6.11** Marked mesenteric lymphadenopathy at postmortem in a 10-week-old ill-thriven Holstein calf in association with chronic *Salmonella* Dublin infection (see also Video Clip 6.1).

• **Fig. 6.12** Classical fibrinonecrotic or diphtheritic membrane lining the intestine of a calf that died from subacute *Salmonella* Typhimurium enterocolitis.

Salmonella and *E. coli* are suspected. When affected calves vary in age from neonatal to several months old, *Salmonella* is more likely than *E. coli* because the latter tends to more commonly affect only neonates. Sample handling is pivotal in reaching a definitive diagnosis, and practitioners should familiarize themselves with their local diagnostic laboratory requirements and recommendations for maximizing the chances of a positive culture from feces, environmental samples, or postmortem tissues. *Salmonella* spp. are quickly overgrown by many other fecal organisms, and pre-enrichment or the use of specific selective transport media may be indicated for samples obtained in the field.

Calves that die peracutely should be necropsied and cultures obtained from the ileum, cecum, or colon. In addition, the mesenteric lymph nodes (Fig. 6.11 and see Video Clip 6.1), gallbladder, and heart blood should be cultured. Calves that die after respiratory and enteric signs should have lung and intestinal samples cultured for *S.* Dublin.

Although pathologists associate salmonellosis with gross enteric lesions such as diphtheritic membranes in the distal small or large intestines, it must be emphasized that peracute *Salmonella* types B or C and acute *S.* Dublin infections often cause minimal demonstrable gross lesions. This fact has been borne out by observing necropsy specimens from many calf mortality epidemics and dictates bacteriologic investigation rather than empiric gross determination of etiology.

Subacute or chronic cases may have fibrinonecrotic or diphtheritic membranes scattered throughout the large and distal small intestine (Fig. 6.12). Petechial hemorrhages and edematous mesenteric lymph nodes are other gross pathologic findings in some cases.

Treatment

Fluid and antibiotic therapy are the cornerstones of treatment for calves with salmonellosis. Decisions as to route of fluid administration are based on physical signs, severity of the diarrhea, and economic considerations. As with *E. coli* infections, calves that are "shocky," unable to rise, and severely dehydrated and those that have no suckle response should be given IV fluids. Calves that are ambulatory, able to suckle, and only moderately dehydrated usually can be managed with oral and possibly SC fluids. Peracute and acute salmonellosis caused by types B, C, D or E may result in a metabolic acidosis similar to that found in ETEC infections. However, losses of Na^+ and Cl^- tend to be more severe in salmonellosis than those found in ETEC-infected calves. Bicarbonate-rich solutions are indicated in peracute *Salmonella* infections and should be considered when profound depression or shocklike signs accompany peracute diarrhea. After correction of metabolic acidosis, balanced electrolytes may be used IV or oral fluids substituted (see section discussing treatment of ETEC). Oral electrolyte–energy solutions are helpful but limited by the maldigestion, malabsorption, and inflammatory lesions in the patient's intestinal tract. Diarrhea tends to persist longer in *Salmonella* infections than ETEC infections and may become chronic if the intestine is permanently damaged.

Whole blood transfusion occasionally is necessary because of fecal blood losses, and colloids are commonly indicated as a result of severe hypoproteinemia associated with albumin loss from the inflamed intestine. Whole blood (free of BLV and BVDV) is sometimes more economical than plasma for calves having severe hypoproteinemia. Hetastarch is currently a reasonably priced alternative to plasma, but it only augments colloidal pressure, lacking globulins and other homologous proteins that can be more specifically therapeutic in septic or inflammatory conditions.

Severe peracute infections that result in shock may necessitate one-time administration of corticosteroids or flunixin meglumine in conjunction with IV fluids. Continued or repeated use of full dosages of either of these products warrants caution because their side effects on the gastric mucosa and renal vasculature appear to be augmented by volume depletion.

Antibiotic therapy for calves having salmonellosis is somewhat controversial and deserves comment.

Reasons not to use antibiotics include:

1. Fear of creating antibiotic-resistant strains that may present a risk to humans and animals in the future.
2. Although antibiotic therapy may aid clinical recovery, it does not stop fecal shedding or positively affect the duration of fecal shedding.
3. *Salmonellae* are facultative intracellular organisms, and antimicrobial penetration into the infected host cell is often limited even for antimicrobials that show in vitro efficacy against the organism.

Reasons to use antibiotics include:

1. Bacteremia is common with salmonellosis of any type in neonates and is very common with *S.* Dublin.
2. Veterinarians cannot always predict which calves are septicemic and which are only endotoxemic when faced with signs of shock and severe diarrhea.
3. Although intracellular penetration of infected host cells by antibiotics may be limited, adequate blood concentrations of an effective antibiotic may limit spread of infection from the gut to other tissues by acting on the organisms that are free in the blood and ECF.
4. Clinical impressions suggest a shorter course of disease and higher recovery rate when antibiotics are used.
5. Secondary infections (e.g., septic meningitis, arthritis) are possible in severely ill calves with salmonellosis.

Antibiotic therapy is justified for calves with peracute or acute signs that suggest overwhelming infection. Calves having mild signs, that are asymptomatic, and with chronic disease do not appear to benefit from antibiotic therapy. Given the characteristically unpredictable nature of antibiotic susceptibility exhibited by *Salmonella* spp., selection should be based on culture and sensitivity. Currently, some strains are resistant to beta-lactam antibiotics, macrolides, and tetracyclines but are frequently sensitive to aminoglycosides, fluoroquinolones, and trimethoprim–sulfas. Antibiotic therapy should be maintained at least 5 to 7 days for peracute and acute salmonellosis and is more likely to be necessary in type B, C, and D infections than with type E. A decision not to use antibiotics for calves with salmonellosis is easier to enforce when the mortality rate is low or nonexistent. This same decision is impossible to enforce when a high mortality rate occurs because owners will not tolerate such losses and will demand antibiotics or change veterinarians in the hope of saving sick calves. Ultimately, decisions on antibiotic use must be based on humane considerations weighed against the public health concerns related to induction of resistance in an organism with demonstrated zoonotic potential. Wholesale treatment of calves at risk and the use of oral antibiotics such as tetracycline for all calves in a group are contraindicated because these techniques are more likely to be ineffective or lead to antibiotic resistance. Antibiotics should be considered as potential components of the treatment regimen, with aggressive fluid and electrolyte replacement, good nursing care, and maintenance of adequate nutrition as primary considerations. Experience suggests that resolution of a salmonellosis problem on a dairy requires far more critical and influential decisions than antibiotic selection for individual cases.

Control

Although an individual calf sporadically becomes infected with *Salmonella* spp. as a result of stress or FPT, endemic infection is increasingly the rule in dairy and veal operations. Infected calves shed large numbers of organisms into the environment, and contamination is worsened by the fluid characteristic of feces in diarrheic calves. Infection spreads quickly when calves are grouped in confinement or crowded into pens. Fecal contamination of feed, water, or feeding devices is common, and septicemic calves may shed organisms in other body secretions as well as feces. *S.* Dublin–infected calves may shed organisms from body secretions, feces, or the respiratory tract. Inapparent or subclinical infections are common and represent a constant source of environmental contamination.

Cleanliness and disinfection of housing units are extremely important to the control of salmonellosis because a primary determinant of the severity of infection appears to be the magnitude of challenge or infective dose. Many severe outbreaks have resulted from contamination of feed, and this area deserves particular scrutiny in any herd investigation because this route serves as a very efficient means of oral inoculation for new hosts.

Detection of previous infection with *S.* Dublin can be achieved through a well-established ELISA assay currently available through the New York State Diagnostic Laboratory. Both milk samples (group or bulk tank samples for example) and serum can be used in this assay, providing a much more sensitive test than traditional culture methods. In other parts of the world, this assay is being used for surveillance and control of *S.* Dublin on a national level through screening of herds every 3 months. It seems unlikely at this point in time that a national program of this nature will come into effect in the United States, but it is worth remembering that *S.* Dublin tends to be more pathogenic in human patients than many other serovars and that its prevalence is undoubtedly on the increase in this country. One has to be careful when interpreting a positive ELISA result in young calves because of the possibility of passively derived antibody rather than naturally acquired titers. It is probable that passively derived antibody for *S.* Dublin wanes along the same time frame as other colostrally derived antibodies, hence calves might be expected to be negative by early postweaning and certainly before breeding age unless true postnatal exposure has occurred. Detection of carrier animals by culture is very difficult because of inapparent infections, variable patterns of shedding, and the failure of negative cultures to completely rule out a carrier state. In the case of non-Dublin serovars recovered animals continue to shed type B, C, and E organisms for 3 to 6 months or more in some instances. Therefore, control measures based on detection

and elimination of infected animals or carriers are more easily instituted when small groups are infected; they can be impractical and unlikely to succeed in very large free-stall herds where animal movement, comingling of heifers from multiple source farms, and new purchases are the norm.

Vaccination against *Salmonella* spp. is controversial. Because cell-mediated immunity is a major factor in host resistance, killed bacterins that stimulate only humoral immunity give questionable protection. Vaccination of dry cows with specific *Salmonella* bacterins may protect neonatal calves somewhat for the first 2 to 3 weeks of life but probably not thereafter. Autogenous bacterins developed from the specific *Salmonella* serotype involved in an epidemic may be more helpful in this regard. Advances in on-farm efficacy in vaccines for the protection of cattle against *Salmonella* spp. has been challenged by the numerous serotypes encountered on many farms over time and the fact that cross protection between serotypes is notoriously poor, especially with killed products. Killed vaccines administered to neonatal calves have not performed well in research trials, primarily because calves appear to respond poorly to the oligosaccharide side chain antigens that comprise the protective antigens. Both commercial and autogenous bacterins must be used with caution because anaphylactic or endotoxic reactions are possible and are thought to represent an inherited hypersensitivity to endotoxin or other mediators. Aromatic-dependent *S.* Typhimurium and *S.* Dublin strains have been used as MLVs in calves and appear promising because they stimulate both cellular and humoral immune responses. One such product is available commercially in the United States for protection against *S.* Dublin (Enterverne-D, Boehringer Ingelheim Vetmedica, St. Joseph, MO); it is labeled for use in calves older than 2 weeks of age and has enjoyed quite widespread use as a component of *S.* Dublin control efforts on many farms. A word of caution, however, is indicated with the product because it can be associated with life-threatening anaphylactoid reactions, and producers and veterinarians need to discuss this risk and be prepared for this eventuality when administering it to calves preweaning. The use of this vaccine will, of course, interfere with serologic testing efforts to identify individuals previously naturally infected with the organism. This product has also been demonstrated to inspire a serologic increase in antibody titers against *S.* Dublin when administered to dry cows, and this can result in increased antibody levels in calves by passive transfer. The health benefits of this increase in *S.* Dublin antibody titer in either the dam or calves are not known however at this time.

The use of a commercially available subunit vaccine has become quite commonplace in adult cattle within the U.S. dairy industry, but the product (*Salmonella* Newport Bacterial Extract [SRP], Zoetis, Florham Park, NJ) is not labeled for use in calves. Recent research has demonstrated that this product can elicit specific IgG antibody against *S.* Newport in colostrum when administered at dry off and 4 weeks later and that this can be transferred passively to new born calves after ingestion of colostrum. Unfortunately, at this point in time, what is unknown is whether this passive antibody will confer a protective effect for calves against natural challenge to either the vaccinal serotype or other strains in a farm environment. This subunit vaccine utilizes siderophore receptors and porins as antigens from *S.* Newport to stimulate an immune response against iron acquiring mechanisms employed by the bacterium (as well as other serotypes of *Salmonella* and other gram-negative species) during rapid bacterial growth. By "starving" the bacteria of iron, there will hopefully be attenuation and reduction in the severity of any subsequent infection; possibly conferring cross protection against multiple serotypes, even with other gram-negative species. There are philosophical similarities with the use of gram-negative core mutant products such as J5 and Endovac-Bovi as a component of *Salmonella* control in calves in that these products raise antibody against conserved epitopes of the gram-negative bacterial cell wall to hopefully ameliorate the pathogenic effects of endotoxin or lipopolysaccharide (LPS), but similarly do not entirely prevent infection. The use of these gram-negative core mutant products as a means of inspiring passive protection through dry cow immunization and colostral transfer, or by active immunization of preweaned calves also continues to enjoy some acceptance within the dairy industry, but there is relatively little scientific evidence to support either approach. For example, although immunization of calves with commercial J-5 vaccines has been shown to reduce mortality from salmonellosis in an experimental challenge study, in a large field trial, J-5 immunization of calves did not affect survival to 100 days.

Control of epidemic salmonellosis in dairy calves entails the basic principles of infectious disease control. Isolation of active cases, hygiene, disinfection, education of handlers, and perhaps culling or depopulation may be required. Whole-herd epidemics are frightening experiences that lead to tremendous public health concern. It is critical that the producer understands that after an outbreak is well established, control measures may mitigate the severity of the outbreak but often fail to immediately bring resolution. Patience, persistence, and communication are important. The reality for many farms, and commercial heifer rearers in particular, is that salmonellosis has become an endemic issue that requires persistent management efforts to position calves for health rather than disease when inevitable challenge with the pathogen occurs.

Methods of *Salmonella* control in calves include:

- Establish diagnosis via culture and sensitivity or serology (in older calves, >3months old) for *S.* Dublin; conduct investigation of the premises.
  - A. Several affected animals should be cultured in epidemics to confirm a common pathogen, although some clinically ill animals will be culture positive on a single sample.
  - B. Cultures of feces must always be performed.
  - C. Colonic contents, bile, and mesenteric lymph nodes may be cultured from necropsy submissions.

D. If *S.* Dublin infection is suspected, blood, tracheal wash, or lung tissue samples may be cultured in addition to feces. Serology can be used after waning of any colostrally derived antibody by 3 months of age for evidence of *S.* Dublin in older calves also.
E. Carefully examine herd medical records and conduct an inspection of the premises to characterize the spatial and temporal characteristics of the problem. Is there a common sick cow pen or maternity stall, for example?
F. Critique flush water flow patterns and traffic patterns for personnel and vehicles.
G. Trace feedstuffs (including colostrum, whole milk, milk replacer, and water, as well as solid feeds) from their storage, preparation, transport to the animals, and delivery to the individual. Consider the possibility of contaminated feed, feed storage areas and transport equipment, feeding utensils, feed bunks, or buckets as potential sources of oral inoculation of healthy animals. Culture and then disinfect accordingly.
H. It is often useful to culture milk or milk replacer, water, and dry feeds for *Salmonella* spp. at the time of preparation or initial storage, during transport in containers, and after they are placed in the final container and presented to the calf (i.e., in a bucket or nipple feeder). This helps to identify potential sites of contamination and amplification of the organism. Producers often forget that milk and milk replacer are excellent culture media for *Salmonella* spp., and initially small inocula can become tremendous pathogen loads as the organisms replicate in feeds.

- *Isolate* infected animals:
  A. This measure is relative and imperfect because some infected animals may not appear ill. However, calves with fluid feces, fever, dehydration, or other related signs should be isolated because they are shedding billions of organisms into the environment.
  B. *Salmonella* control is often a numbers game, and reduction in pathogen load requires evaluation of all facets of calf handling and colostrum and milk feeding. Carefully scrutinize each and every step of the process of calf handling from parturition (maternity area hygiene) through placement in the hutch or pen. Often the first material ingested by the newborn calf is directly from the maternity environment, immediately after birth, and before consuming colostrum. In an outbreak on a Colorado dairy, a colleague of the authors, Dr. Rob Callan, isolated *Salmonella* Infantis from the nose and mouth of a newborn calf less than 5 minutes after being born. This calf had been pulled several yards across the ground of the maternity pen to a separate area where calves were then fed colostrum. In this situation, *Salmonella* was likely ingested well before consuming colostrum, greatly increasing the risk of infection. Although maternity pen hygiene is essential for minimizing exposure in calves, prompt removal of calves to individual calf hutches can also be pivotal in control. Washable industrial wheelbarrows and wheeled bins make for excellent transfer vehicles for moving calves from the maternity area to designated calf housing. Alternatively, a calf "safe pen" can be considered (see previous section on colisepticemia).
  C. All feeding and cleaning implements for sick calves should be scrubbed with soap and hot water and then disinfected between uses and never used on healthy calves.
  D. Distance can be an effective buffer for reducing pathogen load to calves. When possible, calves should be moved to new, well-protected ground that is well removed and upwind from adult cows and protected from water flow from the main operation.
  E. If labor is spread thin, it may be easiest for the producer to hire additional labor or reallocate personnel such that certain people are solely dedicated to the husbandry of calves. Disinfection and good hygiene take time, and personnel with multiple time demands often fail to fully and persistently implement good sanitation practices when handling calves. Epidemiologic studies from Europe have suggested that a single dedicated employee whose sole responsibility is the timely and hygienic collection and administration of colostrum to neonates is a significant positive factor in the control of *S.* Dublin.
  F. Identify and remove all animals PI with BVDV from the herd.
- Therapy for infected animals:
  A. Fluids to maintain hydration, acid–base balance, and serum electrolyte concentrations.
  B. Additional protection from weather stress.
  C. Maintain adequate nutrition.
  D. Treat with appropriate antibiotics when indicated.
- Physically clean environment, improve hygiene, and disinfect premises.
  A. Separate calves from adult cattle in every way possible (particularly critique flush water flow patterns and maternity pen hygiene because these are common means of spread of infection from adults to calves).
  B. Clear maternity stalls and disinfect between calvings. Consider use of a "safe pen" for calves (see previous section on colisepticemia) and cleanable, plastic wheeled bins or wheelbarrows for transfer of calves out of the maternity area.
  C. Do not house preweaned calves in a group, especially during an outbreak. If group housing is the preferred husbandry system for nursing calves, then endemicity is inevitable; it can still be a consequence when calves are individually housed.
  D. Disinfect with a disinfectant approved to kill *Salmonella* spp. after physically cleaning organic debris from surfaces.
  E. Being an opportunist, *Salmonella* spp. often flourish and cause disease in cattle populations under conditions of suboptimal nutrition or reduced immune function. Poor transition cow management, a high

prevalence of animals PI with BVDV, and alterations in feed intake brought about by temperature extremes or poor bunk management are examples of the "intangibles" that often determine whether *Salmonella* infection becomes problematic on a given operation. With respect to calves, the same stressors of nutrition and climate can contribute to higher morbidity and mortality rates.

- Educate the farm owner and workers regarding public health concerns:
  A. Farm workers, calf handlers, and their families frequently become infected by *Salmonella* spp. during calf epidemics, and workers must be educated on how to minimize this risk.
  B. Insist on handlers wearing separate footwear and coveralls when handling infected calves. Allocate labor to prevent cross-infection from diseased calves to healthy calves. If certain personnel are responsible for all calves, have these individuals handle healthy calves first. Disinfect boots, hands, and implements. Be very careful about personal hygiene.
  C. Do not drink raw milk from adult cows if any signs of enteric disease or abortion have been observed in the herd.
- Recognize that recovered animals will shed intermittently or constantly for some time and thus represent significant risk to uninfected animals and people. Therefore, ongoing hygiene, disinfection, and surveillance are necessary.
- Immunization with modified-live (preferred) or killed vaccines (commercially available or autogenous) should be considered as an adjunct measure to be used only when the aforementioned management changes do not result in satisfactory abatement of the problem. Producers should be reminded that immunity is finite, and increased herd-level immunity brought about by the use of a vaccine is unlikely to succeed over the long term if initiated without meaningful changes in husbandry and hygiene.
- Feeding prebiotics or oligosaccharides may be beneficial.

Clostridium perfringens Type C Enterotoxemia.

Etiology

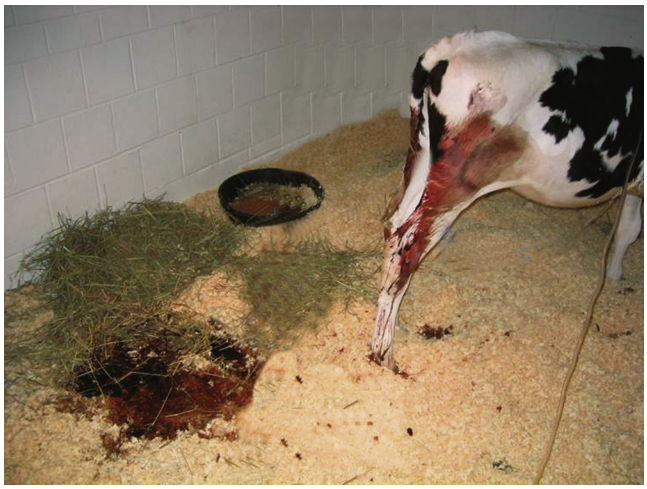
Enterotoxemia caused by *C. perfringens* type C is a commonly fatal disease that occurs in dairy and beef calves. Enteric disease caused by types A, B, and D has been reported in calves but is far less common. Neonates are most commonly affected, although disease losses in older calves (usually ≤3 months of age) can be significant. *C. perfringens* type A is a gram-positive anaerobic bacterium that is part of the normal intestinal flora of vertebrates. Intake of large quantities of soluble carbohydrate or protein is considered a risk factor for the development of type C enterotoxemia; the organism undergoes explosive growth under such conditions, creating a "superinfection" of the enteric lumen and producing exotoxins (termed *major lethal toxins*) that cause the majority of damage to host tissues. The exact reason for clinical disease is often difficult to determine in sporadic cases but usually can be linked to "pushing" calves nutritionally when endemic problems are observed in a herd. Feeding of large volumes of milk or milk replacer, especially in the form of large meals, appears to be a common triggering factor. Heavy grain feeding, foraging on grain crops, sudden access to high-quality forage, or overfeeding after a period of hunger are also considered risks.

Beta toxin (CPB) is the principal major lethal toxin of type C, although variable amounts of alpha toxin (CPA) are produced by this organism too. The contribution of CPA to the pathogenicity of *C. perfringens* type C infections is considered minimal. CPB is a necrotizing toxin that forms membrane pores in susceptible cells such as those of the intestinal epithelium and thereby induces necrosis of enterocytes in the small intestine. This induced intestinal damage allows toxin access to the deeper layers of the gut wall, which creates extensive submucosal necrosis and intraluminal hemorrhage. Alpha toxin is a phospholipase that destroys lecithin within host cell membranes and membranous organelles. Terminally, multisystemic signs of disease can result from absorption of the major lethal toxins and from other gut-origin toxins or organisms gaining entry to the bloodstream via the damaged gut epithelium.

Beta toxin is a protein that is inactivated by exposure to trypsin. Thus, the lethal effects of CPB may be exacerbated in neonates because of either low pancreatic trypsin production or the presence of trypsin inhibitors in colostrum. When calves ingest large volumes of milk or concentrate, the calf's pancreatic enzymes may be sufficiently diluted to prevent inactivation of CPB; alternatively, the organism may proliferate to such a degree that the massive amounts of toxin released simply exceed the limited volume of trypsin available luminally in the gut. Similarly, feedstuffs such as sweet potatoes and soy beans that contain natural trypsin inhibitors may occasionally be associated with clinical disease due to this organism in more mature animals. We have also observed severe *C. perfringens* diarrhea in neonatal calves that were fed a normal feeding of colostrum or milk but that had a large amount of colostral supplements or replacers added to the meal.

Clinical Signs

Signs of enterotoxemia are acute or peracute and consist of colic, abdominal distension, dehydration, depression, and diarrhea. Sudden death or at least such rapid progression of signs that the calf is not observed to be ill for long before death can occur in peracute infections. Colic and abdominal distension usually precede diarrhea, and although the feces are loose, they are never as voluminous or watery as those found in calves with ETEC or salmonellosis. Feces in some enterotoxemia calves contain obvious blood and mucus (Fig. 6.13). Acute cases characterized by abdominal distension and colic may mimic intestinal obstructions unless diarrhea develops to rule out this out. Ballottement of the right lower quadrant reveals increased fluid in the small intestine. Progressive dehydration, depression, abdominal



• **Fig. 6.13** A 5-week-old Holstein with acute and severe hemorrhagic enteritis caused by *Clostridium perfringens* type C. The calf recovered after intensive therapy with intravenously (IV) administered antibiotics (penicillin and ceftiofur), IV fluids, clostridium antitoxin, blood transfusion, flunixin meglumine, gastroprotectants, and transfaunation.

distension, and shock ensue unless intensive therapy is instituted. Neurologic signs are observed occasionally in the terminal stages of fatal cases of type C enterotoxemia. Affected calves usually have been in excellent condition and are often reported to have been vigorous eaters.

Ancillary Data

Blood work seldom is helpful or specific in enterotoxemia patients. Hemoconcentration is a given, but the leukogram and serum chemistry may be normal. In subacute cases, the serum albumin may be low because of intestinal losses and some loss into the peritoneal cavity. Hyperglycemia and glycosuria have been purported to be diagnostic but are more indicative of *C. perfringens* type D in lambs, not enteric clostridiosis in cattle. Any stressful disease may result in hyperglycemia and glycosuria in neonatal ruminants, and these findings are not pathognomonic for enterotoxemia in calves.

Acid–base and electrolyte data are not dramatically abnormal. Enterotoxemia calves *do not* usually have as severe a metabolic acidosis as calves severely affected with acute ETEC or *Salmonella*.

Diagnosis

Other than the physical signs, there are few clues to assist in the diagnosis of enterotoxemia. For fatal cases, necropsy findings often are quoted as diagnostic. However, they seldom are, and necropsy is used primarily to rule out other diseases. In field situations, it may be impossible to obtain meaningful samples and have them reach a diagnostic laboratory in time to be helpful. Recent pathologic description of confirmed cases has detailed that hemorrhagic, coagulative intestinal necrosis is common, especially affecting the jejunum and ileum and that hemorrhagic stool is more common than nonhemorrhagic stool but not invariant. Pathologic lesions can also be found outside the small intestine, with the abomasum, spiral colon, and cecum often demonstrating extensive hemorrhagic necrosis. All dead animals have some *C. perfringens* in their intestines, so the relative numbers, types and toxins present must be assessed to accurately diagnose the type of *C. perfringens* involved and attach significance to the organism. Intestinal enzymes tend to break down alpha and beta toxins within hours of death. Commensal *C. perfringens* type A may proliferate in the gut and invade tissues within a short period after death, especially in warm weather. In fact, postmortem enteric proliferation of *C. perfringens* type A may be so extensive that it masks the presence of type C when luminal contents are cultured. The absolute diagnosis of enterotoxemia caused by type C organisms requires culturing *C. perfringens* from the gut; genotyping by PCR to determine that the isolate is type C; demonstration of gross or histologic lesions; and, if available, testing to identify beta toxin from the intestine of fatal cases (usually by ELISA). In the less common type B and type D enterotoxemias in cattle, absolute diagnosis requires identification of the organism by culture and genotyping in addition to demonstration of the epsilon toxin in the case of type D. Genotypic analysis is usually performed by multiplex PCR (mPCR), although mouse protection testing can also be used.

Calves with acute enterotoxemia may be diagnosed primarily based on clinical signs of colic, abdominal distension due to increased fluid within the small intestine, dehydration, diarrhea, and a rapidly progressive course. Ancillary data, if available, can help rule out other differential diseases. Progressive shock secondary to abomasal perforation with diffuse peritonitis can be ruled out by transabdominal ultrasonography and paracentesis. Acid–base and electrolyte determinations on venous blood and fecal cultures help rule out ETEC and acute salmonellosis because enterotoxemia calves seldom have a profound metabolic acidosis.

Feces or enteric contents may be cultured and assayed for toxins. Toxin identification is laborious and difficult because the toxins are labile and may be rapidly degraded. Proper sampling, storage, and shipment to a qualified laboratory are essential.

Treatment

Supportive treatment requires IV fluids (crystalloids and colloids such as plasma or Hetastarch; 5–10 mL/kg) with appropriate electrolytes and glucose to rehydrate the calf. Ideally, IV potassium or sodium penicillin (44,000 U/kg IV every 6 hours) should be given for the first 24 to 48 hours of therapy but can then be replaced by procaine penicillin (44,000 U/kg IM every 12 hours) if the calf is improved. Calves that are in shock may also be given dexamethasone or flunixin meglumine (0.5–1.1 mg/kg IV) as one-time treatments.

Resolution of clinical signs is gradual and slow. Abdominal distension sometimes takes days to resolve, and diarrhea becomes sporadic rather than voluminous. Recovering calves have variable appetites primarily based on their degree of abdominal distension and hydration status. Recovery in successful cases may require fluid and antibiotic support for up to 7 days. Progressive intestinal ulceration and subsequent

perforation with peritonitis have been observed as an occasional complication in recovering calves. Therefore, repeated use of nonsteroidal and steroidal drugs is contraindicated to avoid further damage to the intestinal tract. Prolonged ileus and failure of abomasal emptying may evolve as a problem in recovering calves. If conservative therapy with IV fluid support and antibiotics fails to resolve this problem and the patient becomes more distended after drinking milk or electrolytes, metoclopramide (0.1–0.25 mg/kg SC every 8 to 12 hours or as a continual infusion) may be helpful to increase abomasal emptying and relieve abdominal distension. Administration of proton pump inhibitors or histamine antagonists may also be considered as described by Ahmed et al in calves with poor abomasal emptying as a means of trying to increase luminal pH and lessen the chances of mucosal ulceration (cimetidine 50–100 mg/kg orally every 8 hours, ranitidine 10–50 mg/kg orally every 8 hours, or preferably, ranitidine 1.5 mg/kg IV every 8 hours). Antitoxins are available commercially, and although they may be of use early in the course of the disease, efficacy of these products is difficult to determine. Blood or plasma transfusions may be needed because of intestinal damage and protein loss.

Control

Presentation of excessive amounts of starch, sugar, or soluble protein into the abomasum or intestine is considered pivotal in the development of enterotoxemia; thus, all potential influences on this critical pathogenic event must be considered when formulating a preventive plan. Evaluation of ration net energy, fiber content and forage length, bunk space, animal hierarchy within a pen, feeding frequency, the rate and magnitude of changes in ration between successive production groups, and feed mixing practices are essential to identify and correct problems with carbohydrate overload or slug feeding in older calves and adults. For pasture-fed animals, turnout onto a new pasture should be very gradual (e.g., day 1, 15 minutes of grazing; day 2, 30 minutes; day 3, 1 hour; day 4, 2 hours, and so on). Prevention of enterotoxemia in nursing calves requires consideration of environmental or management factors that may trigger ingestion of larger than normal volumes of milk or replacer. Decreasing the volume of milk fed per feeding by increasing the frequency of feedings has met with some success. Milk and milk replacer should be fed at or near body temperature to prevent induction of ileus or esophageal dysfunction.

Vaccination with *C. perfringens* toxoids is indicated for herds that have experienced sporadic or endemic enterotoxemia. When successful diagnostic tests confirm a specific type, toxoids obviously should contain that type. When specific types have not been identified, types C and D toxoid usually are suggested because type C is most commonly identified in calf enterotoxemia.

All dry cows and heifers should be vaccinated twice, 2 to 4 weeks apart (or according to manufacturer's recommendations); thereafter yearly boosters should be given 1 month before calving, and calves should be vaccinated with the same vaccine at 8 and 12 weeks of age. Immunization of neonatal calves has been used for enterotoxemia control in problem herds. However, no change in antibody titers to *C. perfringens* toxins has been demonstrated in immunized calves (immunized at ~7 weeks of age) or lambs (immunized twice up to 6 weeks of age) that received colostrum from vaccinated dams.

Clostridium perfringens Enterotoxemia and Abomasitis

Etiology

Sporadic cases of enterotoxemia associated with *C. perfringens* type A have been reported in calves. Abomasitis, abomasal tympany and bloat, and ulceration of the abomasum have also been linked to *C. perfringens* type A. It is uncertain whether the *Clostridium* organism is always the sole cause of this condition, and *C. septicum, C. fallax, Salmonella* spp., and *Sarcina* spp. have also been implicated in the condition in calves and other ruminants.

Abomasitis is a sporadic disorder of neonatal to weanling age calves, lambs, and kids. This disease is characterized by diffuse, hemorrhagic to necrotizing inflammation of the abomasal mucosa, frequently involving the deeper layers of the abomasal wall in severe or chronic cases. Intramural emphysema and edema of the abomasal wall may be present. Abomasal ulceration and perforation may occur in a subset of affected animals.

A variety of putative etiologies for this form of abomasitis existed historically, including primary bacterial or fungal infection, immunosuppression, and pica; trauma from coarse feed or trichobezoars; and vitamin or mineral deficiencies. In 1987, investigators at Kansas State University detected *C. perfringens* types A and E in stomach contents of affected calves and the following year reproduced the disease experimentally by intraruminal inoculation of *C. perfringens* type A in calves. The ability of this organism to produce gas is considered to contribute to the abomasal dilation and intramural emphysema evident in affected animals. More recently, *S.* Typhimurium DT104 was isolated from the abomasal wall of midwestern veal calves with abomasitis. Although authors of earlier case reports associated copper deficiency with abomasitis and abomasal ulcers in beef calves, Roeder and colleagues demonstrated that, in the absence of copper deficiency, abomasitis could occur spontaneously and be induced experimentally. Thus, although copper deficiency may act as a contributory factor for abomasitis and enteric disease of calves, it does not appear to be a requisite factor for either condition. Abomasal stasis and ruminal accumulation of milk have also been proposed as risk factors for the abomasitis/ulceration and the disease is further discussed in Chapter 5.

Diagnostic studies indicate that *C. perfringens* type A is now the most common bacterial isolate in cases of dairy calf clostridial enteritis and that affected calves typically exhibit tympany, hemorrhagic abomasitis, and abomasal ulceration. The presence of the organism as a member of the normal flora of all mammals has clouded its definitive etiologic role, but because inoculation with the organism can reproduce the disease experimentally, much interest has focused on

this organism, particularly in herds that experience repeated cases, or outbreaks, of peracute abomasitis, tympany, and death in nursing calves.

Clinical Signs

Clinical signs include lethargy, abdominal tympany, colic, bruxism, fluid distension of the abomasum, diarrhea, and death. Although the number of case studies concerning abomasitis is few, on review of the available literature, the case fatality rate appears to be very high (75%–100%). Typically, significant signs of tympany and colic precede diarrhea, which is usually low in volume. Appropriate diagnostics are described in the previous section.

Treatment

Treatment of enterotoxemia caused by *C. perfringens* type A is similar to that used for types C or D. Unfortunately, antitoxin for types C and D has unknown efficacy in the treatment of type A cases. For abomasal tympany or abomasitis, IV fluid therapy, plasma therapy, parenteral antibiotic therapy, and antitoxin administration as described in the previous section are warranted in the initial medical management. Orogastric tube passage and decompression may be helpful in some cases; elevation of the calf's forequarters while the tube is placed may be helpful in releasing gas; however, because the predominant site of tympany is the abomasum, not the rumen, the therapeutic benefit of orogastric intubation is often minimal. Oral antibiotics such as penicillin or tetracycline may be helpful in reducing the rate of intraluminal gas production. Decompression of the abomasum via percutaneous ventral abomasocentesis has been described, and intraluminal injection of antibiotics could be performed after decompression. The blind trocharization of the left paralumbar fossa of tympanitic calves in the field is to be avoided. It is understandably tempting to attempt to decompress what can be a massively distended abdomen in this more typical anatomic location on the mistaken belief that it is the rumen that is causing the distension, but we have seen several lacerated abomasums when a 14-gauge needle is placed dorsally in this way—as the thin, distended abomasum deflates, the needle continues to cut into the organ like cheesewire. In cases that become progressively distended or in valuable calves, a laparotomy to drain the abomasal contents and oversew severe abomasal ulcers is often lifesaving. Laxatives appear to be of limited benefit in affected individuals, and large doses of magnesium oxide–hydroxide laxatives are likely contraindicated because they may exacerbate metabolic alkalosis seen in early stages of the disease, induce hypermagnesemia, and simply pull more fluid into the gut lumen.

A large, right-sided tympanic resonance in an ill calf may be a case of abomasal or cecal volvulus, and surgical exploration is indicated if initial medical management does not quickly result in resolution of tympany. Similarly, a left-sided tympanic resonance may reflect left displacement of the abomasum (LDA), and given the apparent high rate of ulceration of the abomasum associated with LDA in calves, surgical exploration is similarly warranted in those cases that do not respond to medical management. Abomasotomy may be indicated for refractory cases of abomasal tympany. Abomasotomy allows for removal of luminal foreign bodies such as hairballs and putrefying milk, both of which may prevent a satisfactory response to medical management.

Prevention

In dairy calves, poor milk hygiene, intermittent feeding of large volumes of milk, and feeding cold milk or milk replacer, often via bucket, have been empirically incriminated as potential contributory factors for abomasal tympany, ulceration, and abomasitis. Anecdotal reports indicate that changing from bucket to bottle feeding, increasing the frequency of milk or milk replacer feeding and decreasing the volume fed at each feeding, as well as maintaining milk or replacer at body temperature until it is fed, may reduce the incidence and severity of this condition. A vaccine (*C. perfringens* type A toxoid, Novartis Animal Health, Larchwood, IA) that induces high antibody titers against alpha toxin, a primary virulence factor of *C. perfringens* type A, is available in the United States for prevention of diseases in cattle caused by this organism. As a dry cow vaccine, this product may increase colostral titers against alpha toxin, but the efficacy of this product in reducing calfhood diseases caused by *C. perfringens* type A is currently undetermined. A small proportion of *C. perfringens* type A strains associated with abomasal tympany and abomasitis in dairy calves are also CPB2 positive (the beta2 toxin encoding gene), so there may be some occasional therapeutic benefit to the administration of antitoxin to *C. perfringens* type C or the prophylactic use of toxoids against type C.

Giardiasis

Giardia duodenalis (also referred to as *G. lamblia* or *G. intestinalis*) is a common protozoal parasite of mammals, and the increased diagnostic sensitivity of PCR over previous immunofluorescence techniques has allowed prevalence investigations in a number of dairying areas worldwide. There are seven major genotypes (called Assemblages) of *G. duodenalis*, three of which have been demonstrated in cattle, specifically assemblages A, B, and E. Some of the specific interest in giardiasis relates to its zoonotic potential, although of the 3 genotypes identified in cattle in the United States, only assemblages A and B are considered zoonotic. Studies in the United States have revealed that the nonzoonotic assemblage E is the most common genotype in dairy calves and calves in cow and calf beef operations. Longitudinal studies have revealed that preweaning calves are more likely to be infected compared with postweaning or breeding-age heifers. The prevalence of infection in adult dairy cows in the United States appears to vary significantly from farm to farm, ranging from 0% to as high as 25%. Prevalence rates in replacement stock vary among studies but are commonly in the 40% to 70% range for preweaning-age calves and in the 25% to 40% range postweaning. A number of studies have revealed a low prevalence of zoonotic assemblage A,

either in isolation, or in mixed infections with assemblage E in dairy calves, so dairy heifers must still be considered a zoonotic risk.

The clinical relevance of *G. duodenalis* is uncertain, prevalence studies in the literature tending to be based on fecal sampling of healthy, nondiarrheic calves or the sampling of diarrheic calves when other likely and well-known pathogens were not investigated extensively. Caution should therefore be taken when the organism is identified in either preweaned or group-housed calves because Koch's postulates have not been fulfilled for this protozoan in cattle, and it is a common finding in healthy calves, especially if modern, highly sensitive molecular techniques are used diagnostically. Fenbendazole at 15 mg/kg for 3 days has been a recommended treatment when practitioners are convinced that giardiasis is a clinical issue, specifically when other etiologic causes of loose stool, with or without blood, have been ruled out by appropriate sampling.

Campylobacteriosis

Much of the interest in *Campylobacter jejuni,* alongside other non-*jejuni* species such as *C. coli* and *C. faecalis,* stems not from their proven role as agents of calf diarrhea but because of the zoonotic risk that meat and dairy products contaminated with these organisms pose to human health. Furthermore, increased attention to antimicrobial resistance patterns among food animal–derived isolates of *Campylobacter* spp. heightens awareness of antimicrobial use in the dairy and beef industry and the potential role that both therapeutic and nontherapeutic drug use may play in increasing the public health threat of these foodborne pathogens. *C. jejuni* and *C. coli* can commonly be found in the feces of healthy calves (especially *C. jejuni*), and several studies have demonstrated that the prevalence of culture-positive fecal samples in dairy calves preweaning is in the range of about 15% of all calves on farm and that this rate is similar when diarrheic calves and normal calves are sampled. There has been an increasing level of tetracycline resistance among *C. jejuni* isolates from dairy animals in the United States in recent years, with some studies reporting up to 50% of strains being resistant. Worldwide, both fluoroquinolone and macrolide resistance are also becoming equally threatening problems.

Clostridium difficile

Rather similarly to campylobacteriosis, the predominant reason for recent interest in *C. difficile* comes not from an established role as a causative pathogen of calf diarrhea but because of the human health implications of the organism. Numerous studies across the world, including the United States, have demonstrated that the feces of dairy and veal calves as well as adult cows can contain toxigenic strains of *C. difficile*, albeit at low prevalence rates. Experimental inoculation of neonatal dairy calves with a highly toxigenic strain of the organism did not cause diarrhea or produce detectable toxin in their feces.

Other Possible Infectious Causes of Calf Diarrhea

With the advent of PCR, reverse transcriptase PCR (RT-PCR), and next-generation sequencing, molecular diagnostic techniques have become increasingly more sensitive and versatile in the past decade, and there has been an increase in the identification of novel viral agents in the feces of neonatal calves with diarrhea. Multiplex PCR assays for example enable screening for an array of different agents simultaneously, and there are several publications from diagnostic laboratories identifying bovine enterovirus, bovine norovirus, bovine torovirus, nebovirus, aichivirus, and bovine astrovirus in fecal samples obtained from dairy calves in the United States, South America, Africa, and the Far East. It is important to point out that many of these agents can also be found in the feces of normal calves and that for each of the viral agents listed, there has as yet been no successful recreation of enteritis experimentally in well-designed studies. In many cases, these agents have been identified along with more traditional viral, bacterial, and protozoal infections as part of a mixed infection in diarrheic calves. Control and prevention methods for viral diarrhea, specifically for what are all contagious, feco-orally spread organisms, would not deviate markedly from those listed earlier in this chapter.

Diarrhea and Emaciation Caused by Milk Replacer Feeding

Etiology

There has been a great deal of change in composition and formulation of modern-day milk replacers compared with early milk replacers produced during the 1950s and 1960s. Similar to adult cow rations, milk replacers may be formulated on a least-cost basis for ingredients, especially those comprising the crude protein fraction because this is the most expensive component. Since diarrhea and emaciation caused by inadequate milk replacer feeding is often initially considered to be an infectious diarrhea it is discussed here.

Milk proteins have been the preferred protein source for milk replacers, and pasteurized skim milk powder (low heat prepared and then spray dried) is ideal. Unfortunately, the price of skim milk has risen to a point where it is no longer economically possible to include it as the total source of protein in most milk replacers. Most milk replacer proteins are now derived from whey protein concentrate, dried whole whey, dried whey products that are byproducts of cheese manufacturing with casein and fat extracted, or spray-dried plasma (often from other species, e.g., porcine).

Other protein sources such as modified soy protein, soy protein concentrate, soy protein isolate, and special processed soy flour also have been used. Special processing of these soy protein sources chemically or by heat is necessary to deter allergic gastroenteritis in calves that ingest them. Such processing reduces antinutritional factors in soy proteins and allows for a soluble product. Moreover, soy proteins seldom are fed as the entire protein source and often comprise less

than 50% of total protein, thus allowing their inclusion and successful use for milk replacer protein sources.

The total protein content of a milk replacer should be a minimum of 20%, with most current minimal recommendations suggesting at least 22%. Milk proteins should make up as much of the protein as possible, and processing of the proteins should not damage the nutrient by subjecting it to high temperatures or other factors.

Fat content of milk replacers is another source of controversy between feed companies and nutritionists. Countless feeding trials have been conducted to show that each company's product is the perfect feed. However, in northern climates, there is no question that a 20% or higher fat content is best based on field observations.

The fiber level in milk replacers roughly correlates with plant origin sources of protein in some instances. With the advent of acceptable soy protein sources, however, fiber levels cannot be the sole means of evaluation. Early milk replacers with soy flour or another soy source added could be judged somewhat by crude fiber because each 0.1% crude fiber suggested 10% of the protein to be of plant origin. Inclusion of modified soy protein, soy protein concentrate, and soy protein isolate, however, does not increase the fiber content significantly. Therefore, crude fiber is not of great correlative value when evaluating current milk replacer protein content.

Yet another controversial aspect of milk replacer feeding involves physiologic "clotting" in the calf abomasum. Milk fed by conventional means causes reflex esophageal groove closure and diversion into the abomasum rather than forestomach. In the abomasum, milk quickly is separated into a casein and milk fat coagulum and a liquid component; whey. Chymosin (rennin) and pepsin in the presence of calcium and hydrochloric acid assist this separation. The whey, which contains lactose, protein, immunoglobulins, and minerals, passes into the duodenum for digestion, but the casein or fat coagulum is digested slowly. For many years, it was thought that milk replacers had to "clot" in the abomasum or else they were inferior and caused diarrhea and poor growth. Because only milk or skim milk feeds have casein and whey components, tests for clotting were most applicable to milk replacers with skim milk as the source of protein. In essence, tests for clotting were designed to detect skim milk-origin whey proteins that had been heat denatured by excessive temperatures during processing or drying and therefore would not clot. Because most current milk replacers have a high composition of whey protein or soy-origin protein, they do not clot, yet they appear to be well digested.

In addition to the composition of milk replacers, practitioners should be familiar with the common errors associated with milk replacer feeding. The amount of milk replacer fed may or may not be enough for maintenance and growth of suckling calves. Similarly, the dilution may be too great or the owner may be skimping because of economic pressures. Replacer should be reconstituted at approximately 12.5% solids (similar to whole milk) and fed at least twice daily for a total of *at least* 10% to 12% body weight. Cold weather extremes and northern winter housing necessitate higher volumes or a third or fourth feeding each day. Some manufacturers do not recommend enough milk replacer to meet maintenance *and* growth requirements under these conditions. Therefore, recommended total amounts may be erroneous.

Another problem with some milk replacers is the high sodium content, which may cause neurologic signs if free water is unavailable for whatever reason! Fresh water consumption being most consistent immediately after milk ingestion, it is imperative that fresh, palatable, unfrozen water is available to all calves immediately after milk feeding. The issue of malnutrition related to inadequate calorific intake of lower quantity or lower quality protein and fat content milk replacers is significantly compounded when calves have illnesses during the first month of life. The calorific needs of calves with, or recovering from, enteritis or pneumonia are multiples of normal maintenance requirements; the lower the fat and protein content, the greater the necessary volume to be ingested will be. This phenomenon is predictable and repeatable during the winter months in the northern United States when endemic scours and respiratory disease problems are often at their worst.

In the past, it was more commonplace for newborn calves to receive colostrum until 3 to 4 days of age and then be switched to replacer or whole milk. This is no longer as frequently practiced, and milk replacer may be fed as early as day 2 of life in most settings. Yet another common feeding error for farmers using milk replacer is not increasing the amount fed as the calf ages. In other words, the calf receives 1 cup of milk replacer in 2 quarts of water twice daily at 4 days of age and is still being fed the same amount at 4 weeks of age. Only through a step-by-step discussion with the owner and by careful observation can the veterinarian detect and correct some calf feeding problems.

High-quality calf starter grains can mask the effect of a poor-quality milk replacer; some authors believe that up to 50% to 75% of calf weight gain before weaning may result from high-quality calf starter intake. Milk replacers containing antibiotics or decoquinate are advertised widely, but their value is difficult to assess because studies have yielded contradictory results.

Calf diarrhea, emaciation, or both can result from errors in milk replacer feeding. The preceding discussion lists some of the common problems in milk replacer composition and feeding. The true "etiology" of milk replacer-related calf mortality varies but includes;

- Poor-quality milk replacer (i.e., one with <22% protein, 20% fat, or a poor-quality or over-processed protein source)
- Feeding at the wrong dilution
- Feeding the wrong amount (usually not enough; see later section on feeding for disease or during cold stress)

Clinical Signs

Calves with malnutrition from poor-quality milk replacer appear thin; have dull hair coats with patchy alopecia; usually have diarrhea that coats their perineum, tail, and hind legs; and are hungry. Affected calves have a normal or subnormal temperature unless an opportunistic infection (e.g., pneumonia)

causes a fever in the terminal stages. Calves have no body fat and are weak. Owners complain about calf death that usually occurs at 3 to 6 weeks of age and attribute death to chronic diarrhea or pneumonia. Calves may die suddenly but often remain hungry and willing to nurse even if recumbent 1 to 2 days before death. All calves in the preweaning group look thin. The owner may report that the calves look good for the first week but then seem to deteriorate. Calves that survive to weaning often do well on high-quality solid feeds and regain condition.

Ancillary Aids

If calves are dying as early as 3 weeks of age, enteric pathogens and parasites must be ruled out by submission of either fecal samples from live animals or feces and gut samples from necropsy samples. It may be necessary to assess blood selenium and vitamin E values from calves that become recumbent. Total protein values may be low because of persistent low protein intake or fecal losses associated with enteritis, a result of poor-quality protein sources. Blood work results are normal unless a stress leukogram exists. Assessment of adequacy of passive transfer is also prudent.

Necropsy confirms malnutrition based on serous atrophy of fat in the epicardial grooves, omentum, and perirenal areas. Gut contents are often fluid, reflecting either poor digestion of nutrients in the cachectic state or opportunistic, secondary enteric infections of the compromised host. Pneumonia may be present as a concurrent condition.

Diagnosis

Diagnosis usually can be made by inspection of the calves coupled with a careful history and evaluation of the milk replacer and feeding procedures. Differential diagnoses include infectious causes of neonatal diarrhea, coccidiosis, and selenium deficiency. Whereas calves that are younger than 3 weeks of age require careful consideration of infectious enteric bacterial, viral, or protozoan pathogens, older calves between 3 weeks of age and weaning require consideration of coccidiosis and salmonellosis. The owner will be adamant that an infectious disease is responsible because so many calves appear to be affected by ill thrift and looser than normal stool.

Improper preparation and mixing are occasional factors that augment malnutrition. Careful reading of the instructions on milk replacers or consultation with a nutritionist affiliated with the manufacturer may reveal that mixing at hot temperatures (104° to 106°F) is required for complete solubilization of fat in the replacer; subsequent cooling to body temperature is necessary for acceptance by calves. In such cases, greasy residue may be detected in the mixing vessel as well as in the bottles or buckets following ingestion by the calves.

Inspection of the whole group of calves is very helpful, especially when the veterinarian routinely observes the calves at monthly visits. The sight of a whole group of malnourished but hungry and bright nursing calves in a barn that usually has well-conditioned calves almost guarantees that the owner has switched to a new (cheaper) milk replacer. Calves "eat until they die" and appear hungry even though they are in poor condition. Necropsy of fatal cases confirms serous atrophy of fat and allows other diseases to be ruled out after submission of appropriate samples.

Prevention

Correction and prevention merely require the feeding of a high-quality milk replacer at proper dilution and in proper quantities. The owner must be convinced that milk replacer is not the place to save pennies. In fact, given the increased costs associated with calf losses in such cases, it can be stated that the most expensive milk replacer a producer can buy is often the cheapest one. Milk replacer is never as good as whole milk for calves; therefore, whenever possible, owners should be encouraged to feed calves whole milk that is at least 22% crude protein and 20% crude fat (dry matter basis) unless there is a problem with Johne's disease, *Salmonella, Mycoplasma,* or leukosis in the herd. Milk discarded because of antibiotic residues is not ideal and should not be fed to group-housed calves. It carries an increased risk for transmission of several contagious infectious diseases unless pasteurization is performed. Many owners need to reassess the costs of feeding milk versus milk replacer because feeding proper quantities of high-quality milk replacer may be nearly as expensive as whole milk and can never be as good a diet. Use of a pasteurizer for feeding waste milk to calves has been shown to be of economic benefit on larger dairies. Pasteurization of waste milk is worthy of consideration for operations that routinely produce calves for sale as replacement stock because milk-borne transmission of infectious agents of concern (e.g., *Mycoplasma* spp.) may be reduced.

Soured colostrum and pickled colostrum can be excellent sources of feed for calves, but their storage problems and an increased potential for spreading pathogens frequently discourage farmers from using them.

If the veterinarian has made a diagnosis of diarrhea and emaciation caused by milk replacer issues and feeding of pasteurized whole milk cannot be done, the following instructions can be followed:

1. Ensure adequate colostral feeding to ensure passive transfer of immunoglobulins during the first 12 hours of life. Quality control for proof of adequacy of passive transfer is advisable; total protein measurement by handheld refractometer to confirm that more than 75% of all calves between 1 and 7 days of age have total protein levels of 5.5 g/dL or higher is recommended. If a Brix refractometer is used, the reading should be greater than 8.5%.
2. Feed colostrum for the first 3 days of the calf's life at 10% to 12% body weight but only if sure the cow is not shedding *M. paratuberculosis* or *M. bovis.* On larger dairies, consider colostrum pasteurization.
3. Begin feeding a high-quality milk replacer on day 4 at 10% body weight:
   - Minimum 22% protein—most or all of milk origin if possible
   - Minimum 20% fat
   - Minimal crude fiber
4. After the first week, quantity can gradually be increased to maintain at least 10% to 12% body weight intake.

5. Be sure dilution factors for milk replacer are correct to mimic the total solids of milk.
6. All calves should have fresh water and a high-quality calf starter available at all times. Feed starter in small amounts initially until the calf begins to eat well enough that the starter does not spoil in the feeder. High-quality hay can be available in small quantities starting at 2 weeks of age.
7. Regularly monitor the preparation and feeding temperature of the milk replacer.

An excellent calf starter, adequate feed intake, and good management may mask the effects of a poor-quality milk replacer. This is why some farms seem to have "starving" calves on a specific milk replacer but others seem to achieve acceptable growth with the same product.

Feeding Dairy Calves Milk Replacer for Cold, Illness, or Stress

Because calves are born with little to no body fat that can be mobilized for energy, nutrition becomes a critical element of health management, welfare, and future performance. Nutritional management of the young dairy calf is very dynamic, with requirements for protein and energy increasing each week so that calves can double their birth weight by 56 to 60 days of age. Accomplishing an optimal rate of gain is made more complex in cold weather, hot weather, or for any calves living in conditions outside of their thermoneutral zone (58° to 72°F). It is also significantly complicated by concurrent disease and convalescence. Although the National Research Council (NRC) provides excellent guidelines for feeding calves under a wide range of environmental temperatures, it does not offer specific guidance for feeding calves under conditions of heat stress, disease, immune challenge, vaccination, or other environmental or management stressors. Most veterinary practitioners and academicians are in agreement that the protein and energy requirements for calves are increased under these conditions, but unfortunately, precise feeding guidelines are not available for each situation.

To provide the calf with the ability to meet increased nutritional demands, whether it is attributed to cold weather, hot weather, stress, illness, pain, or immune challenge, it is important to provide more milk or milk replacer. To encourage intake and prevent digestive problems caused by dietary inconsistency, it is optimal to simply feed a greater daily volume of the same milk or milk replacer mix to which calves are already accustomed. Optimally, the increased volume is delivered by providing at least one additional daily feeding to calves. Alternatively, a concentrated energy source is added to the regular liquid diet or a higher concentration of dried milk replacer powder is fed at the same number of daily feedings. When changes are made in the concentration of milk replacer being fed then this should be done gradually; total solids of liquid feed should not increase by more than 1% at a time (per day) and must never exceed a maximum of 17%. Hyperosmolality and hypernatremia are not issues provided fresh water is available to calves at all times and incremental changes to total solids are made gradually. Table 6.3 shows how the NRC predicts an increasing requirement for the volume of whole milk fed by calf age under cold and warm environmental conditions for an 80 lb birthweight calf. As ambient temperature drops still further into the temperature ranges experienced in the northern US in winter the amount of whole milk being fed would need to be adjusted higher from the volumes detailed in Table 6.3.

TABLE 6.3 Increasing Requirement for Whole Milk in the Diet of a Dairy Calf in Cold Weather*

	Cold: Temperature <50°F	Warm: Temperature 50° to 75°F
4 quarts/day	0–3 days	0–7 days
6 quarts/day	4–10 days	8–14 days
8 quarts/day	11–49 days	15–49 days

*Assuming colostrum administration at a minimum of 10% of calf's bodyweight within first 12 hours of life.

Many use the NRC calculator available online to predict the allowable daily gain from energy and protein for different dairy calf diets. In Table 6.4, the NRC calculator has been used to predict the average daily gain (ADG) of a 2-week-old, 95-lb Holstein calf being fed 6 quarts daily (30 oz of milk replacer powder) using different milk replacer formulations and different environmental temperatures.

By week 3 of age, a Holstein calf on-target to double birth weight by day 56 should be gaining 1.6 lb/day. Table 6.4 shows that neither a 20:20 nor a 22:20 milk replacer is predicted to meet that expectation. At cold temperatures (20° and 40°F), a 6-quart per day diet is both protein and energy deficient, but at the warmer temperatures of 60° and 80°F, the 6 quart per day diet is only protein deficient. With the higher protein milk replacers (24:18 and 28:20), the 6-quart diet is limited by energy intake at colder temperatures. At 20°F, both the 24:18 and 28:20 milk replacers are too low in energy, but at 40°F, only the 24:18 milk replacer is limiting ADG because of lack of energy. If the same 2-week-old, 95-lb Holstein calf is fed 8 quarts of milk replacer per day (40 oz of milk replacer powder), all of the milk replacers shown in Table 6.4 would meet energy and protein requirements to achieve at least an ADG of 1.6 lb/day. Although the tables are focused on NRC predictions for feeding milk or milk replacer at different temperatures, they are useful to guide clinicians in feeding sick, stressed, or immune-challenged calves. It is still recommended to feed sick calves for expected ADG. One can provide more frequent feedings of the typical milk or milk replacer diet to which the calf is accustomed to achieve adequate intake. Milk, milk replacer, and water should be delivered at the temperature to which the calf has been accustomed but it is important to offer it at a temperature of at least 93°F. To encourage feed intake under conditions of illness or stress, it is imperative to provide continuous access to fresh, clean, warm (especially in cold weather) water along with a high quality texturized calf starter with a crude protein content of at least 18%.

TABLE 6.4 National Research Council Predictions of Average Daily Gain (lb/day) for a 2-Week-Old, 95-lb Holstein Calf Fed 6 Quarts of Milk Replacer (30 oz of Milk Replacer Powder) Per Day

	Milk Replacer Crude Protein to Fat Ratio			
Temperature (°F)	20:20	22:20	24:18	28:20
20	1.2	1.2	1.2	1.3
40	1.4	1.5	1.5	1.6
60	1.4	1.5	1.6	1.9
80	1.4	1.5	1.6	1.9

TABLE 6.5 Diagnostic Plan for Workup of Herd Neonatal Diarrhea Problems (<14 Days of Age)

Management

I. Assess success of passive transfer on at least two or three consecutive affected calves
   A. TP, TS, or Brix refractometry
   B. Specific immunoglobulin level on serum from affected calves
II. Discuss management of dry cow
   A. Vaccines
   B. Housing
   C. Calving area (cleanliness and so forth)
   D. Colostrum quality, quantity and feeding
   E. Are affected calves from primiparous or multiparous dams or both?
III. Statistics on morbidity and mortality in calves
IV. What are calves fed after initial colostrum?

Patient

I. Feces collected immediately after onset of diarrhea to diagnostic laboratory on at least two or three consecutive affected calves
   A. Bacterial
      - *Escherichia coli* (type, toxin identification and antibiotic sensitivity) *Salmonella* spp. (type and antibiotic sensitivity)
      - *Clostridium perfringens* (relative numbers, type and toxin identification)
   B. Viral—PCR, EM, ELISA
      - Isolation possibly
      - Rule out BVDV by buffy coat isolation from blood
   C. Parasitic—*Cryptosporidium*
II. CBC, total protein, whole blood selenium
III. Acid–base and serum/plasma chemistries
IV. Hydration status
V. Body condition

Generalities

1. If FPT, ignore *Escherichia coli* unless same organism confirmed also on non-FPT calves.
2. Whenever possible, more than one calf should be sampled before blaming the whole herd problem on a single isolate.
3. Only fresh cases are worth sampling.
4. If patient older than 2 weeks, consider poor replacer rather than infectious diseases.

BVDV, Bovine viral diarrhea virus; *ELISA,* enzyme-linked immunosorbent assay; *EM,* electron microscopy; *FPT,* failure of passive transfer; *GGT,* γ-glutamyl transferase; *PCR,* polymerase chain reaction; *TP,* total protein; *TS,* total solids.

Summary Diagnostic Protocol for Investigation of Neonatal Calf Diarrhea

Table 6.5 gives a diagnostic plan for herd neonatal diarrhea problems. Also see Table 18.2.

Coccidiosis

Etiology

Coccidiosis has become one of the more serious problems encountered in raising dairy calves when the calves are grouped housed either pre- or postweaning. Traditionally, it has been considered a problem in group-housed weaned calves, but the prepatent period is such that clinical disease can be seen before weaning in modern group-reared nursing calf management systems. This style of management currently is very popular because it decreases the labor requirements for many large dairies and is convenient in colder climates.

Although up to 20 species of *Eimeria* may infect cattle, *E. bovis* and *E. zuernii* are considered the major pathogenic species. Sporulated oocysts that are infective for calves and older cattle arise from oocysts passed in the feces of cattle with patent infections. Whereas moisture and cool conditions are conducive to sporulation, extreme heat and dryness are detrimental. Oocysts can remain viable for more than 1 year in favorable conditions that include moisture and absence of temperature extremes. Fecal contamination of feedstuffs, water, or hair coats allows ingestion of infective oocysts by other cattle. Conditions favoring fecal contamination of feed and water exist when calves are grouped in mini free stalls, bedded packs, or other group

housing systems such as is becoming increasingly popular with automated milk feeding of calves. Calves may ingest feces containing oocysts from feed bunks that become contaminated when calves come running up to the bunk to be fed and then splash manure into the bunk or waterers, from calves licking themselves and ingesting feces or fecal-stained hair, or from browsing on contaminated bedding in a housing unit.

Coccidia are quite host-specific intracellular parasites that complete both the asexual and sexual phases of reproduction within the host, but as mentioned previously, sporulation occurs outside the host. Ingested sporulated oocysts excyst in the host, release sporozoites that invade host cells (central lacteals of ileal villi for *E. bovis*, connective tissue cells of ileal lamina propria for *E. zuernii*), and grow to schizonts (meronts) that release merozoites that then infect epithelial cells in the cecum and colon. Second-generation schizonts then form in these cells and subsequently release merozoites that begin the sexual phase of the reproductive cycle by invading yet another host epithelial cell and become microgamonts (male) or macrogamonts (female). Microgamonts release microgametes that seek host cells containing macrogametes (matured macrogamont), fertilization takes place, and a zygote forms and matures to an oocyst that then is released by rupture of the host cell. Invasion of cells and subsequent release of merozoites and oocysts incite varying degrees of pathology within the epithelium of the cecum and colon of affected calves. The magnitude of enteric pathology appears to be related to the dose of oocysts ingested. Small doses of ingested oocysts may result in inapparent infection and eventual induction of immunity. Large doses are more likely to result in clinical disease.

Oocysts are observed in feces (patent infection) approximately 17 to 20 days after infection with *E. bovis* and 16 to 17 days with *E. zuernii.* The numbers of oocysts in the feces do not always correspond with the degree of enteric pathology or clinical signs because even asymptomatic animals may shed fairly large numbers of oocysts. Conversely, some calves become severely ill before the majority of protozoa complete their life cycle and produce oocysts; therefore, fecal oocyst counts may be relatively low despite the serious pathology present in the large intestine.

Recovered calves are thought to be relatively immune to reinfection by the same species of *Eimeria* but at risk for infection by other species. Factors that have a negative influence on the calves' immune competence enhance the pathogenicity of coccidia. Therefore, calves exposed to coccidia oocysts and simultaneously subjected to climatic stress, poor nutrition, exogenous corticosteroids, concurrent inflammatory diseases, or acute or persistent BVDV infection would likely show severe signs of coccidiosis.

Clinical Signs

Classical textbook signs of acute coccidiosis in calves include diarrhea containing mucus and blood, tenesmus, depression, and reduced appetite. Rectal prolapse may occur secondary to proctitis and prolonged tenesmus. Affected calves appear dehydrated and thin and have poor hair coats. Milder cases merely show mild diarrhea without systemic signs and many cases are subclinical.

• **Fig. 6.14** Typical signs of coccidiosis in dairy calves. Some of the calves are well grown and have normal hair coats, but others (especially the heifer that is not in a lock-in stanchion) are undergrown; have a rough, dry, unshed hair coat; and are thin. Many have looser feces than normal for their diet, and the hindquarters are stained by loose manure.

In fact, these textbook signs of acute coccidiosis seldom occur in dairy calves. The predominant signs of coccidiosis in group-raised dairy calves are loose manure, poor condition, poor growth rates, and poor hair coats (Fig. 6.14). The feces seldom contain blood or mucus and tend to have a pea soup consistency. Feces stain the perineum, tail, and hocks of typical cases. Although most calves in the group are infected, only those with severe infection show dramatic signs. Coccidiosis is a perfect example of the "weak sister" law in parasitology; this law states that when a group of animals are parasitized, the most seriously affected bring attention to the problem and act as a signal that the entire group needs treatment. Dairy calves and heifers occasionally show textbook signs of blood- and mucus-stained feces, tenesmus, and inappetence. Tenesmus can be severe and sufficient to prevent the patient from concentrating on eating or drinking.

Coccidiosis is a major disease in group-raised dairy calves and heifers. Heifers raised in confinement groups require prophylactic treatment for coccidiosis, or growth rates can be severely compromised. The age of onset for clinical signs varies. Theoretically, it is possible that calves will show signs by 3 weeks of age based on life cycles of *E. zuernii* and *E. bovis*. Fortunately, this seldom occurs unless newborn calves are put in heavily contaminated environments such as group housing arrangements or hutches that have not been cleaned since previous occupancy by infected calves. In general, coccidiosis becomes a problem for dairy calves at weaning when they are grouped. Weaning and grouping of calves that were previously housed individually induce stress. This stress, combined with an environment that more commonly fosters fecal contamination of feedstuffs, water sources, and hair coats, creates

an ideal situation for coccidiosis. Therefore, clinical signs of coccidiosis are most commonly seen in 8- to 16-week-old calves raised in mini free-stall or automatic lock-in facilities. Occasional outbreaks have been observed in 12- to 18-month-old heifers as well but only very rarely in milking age animals. It would be assumed that older animals showing signs of coccidiosis had not previously developed resistance to the *Eimeria* spp. involved.

Morbidity is often higher than expected because many infected animals remain subclinical or show only mild signs such as diminished weight gain. The mortality rate is low unless the problem is neglected, a severe oocyst loads exist, or if concurrent disease affects the coccidiosis patients.

Nervous coccidiosis has been well described in Canada and the northern United States. Although this form has been observed primarily in beef calves, it may occur in dairy calves as well. Heavy loads of coccidia coupled with severe winter weather seem to be contributing factors to nervous coccidiosis. Affected calves can show a variety of neurologic signs, including (but not limited to) severe tremors, nystagmus, and recumbency. Opisthotonos may be observed and confuse the diagnosis with that of polioencephalomalacia. The mortality rate is high for calves with nervous coccidiosis.

Diagnosis

Clinical signs coupled with fecal flotation (standard flotation or McMaster's flotation) to confirm high numbers of coccidia allow a positive diagnosis. The diagnostic limitations of fecal oocyst counts must be kept in mind:

1. Diarrhea may precede the highest oocyst counts by a few days in acute cases because merozoite damage to the colonic epithelium may cause diarrhea before full patency and maximal oocyst shedding occur. In other words, an animal severely affected by coccidiosis may have a relatively low or even zero oocyst count on a given fecal sample. Necropsy and histopathology may be necessary to confirm the diagnosis in such cases.
2. Nonpathogenic species of coccidia may artificially elevate oocyst counts as they traverse the intestinal tract.
3. Healthy animals may have oocysts in their feces.

In general, oocyst counts of > 5000/g of feces are considered significant when coupled with clinical signs. Several calves should be sampled to confirm the diagnosis because severely affected groups of calves tend to show higher oocyst counts as a population.

The major differential diagnoses are salmonellosis, BVDV infection, endoparasitism as a result of nematodes, and poor nutrition. These diseases should be ruled out when response to treatment for coccidiosis fails to correct the problem. Nervous coccidiosis dictates a much broader differential diagnosis, including polioencephalomalacia, *Histophilus somni* meningoencephalomyelitis, salt poisoning, lead poisoning, and many other neurologic diseases. A CSF tap is a valuable test to rule in or out some of these differential diagnoses.

Calves that die from acute, severe coccidiosis may or may not have gross pathologic lesions in the cecum and colon. Severe infections may cause a diphtheritic membrane from sloughed mucosa, blood, and fibrin. Whole blood clots occasionally are found in the colon, and the mucosa of the cecum, colon, and rectum may be thickened. Small white spots (schizonts) may be apparent on close inspection of the mucosa of the ileum or colon. Microscopic lesions mainly reflect colonic damage secondary to second-generation schizonts and sexual phases of the parasite life cycle. Inflammation, sloughing of epithelial cells, cellular infiltrates, and alteration of the appearance of infected epithelial cells to a less columnar shape may be observed. Whereas schizonts of *E. bovis* tend to be located in the villous tips, *E. zuernii* schizonts are located adjacent to the muscularis layer.

Treatment and Prevention

Treatment and prevention of coccidiosis in calves entail orally administered coccidiostatic or coccidiocidal agents. It is commonplace within the U.S. dairy industry to medicate calves prophylactically because exposure of calves to coccidia is likely. Amprolium, monensin, lasalocid, and decoquinate are the drugs used most commonly to treat groups of affected or at-risk calves. These drugs are added to feed or water at the rates listed in Table 6.6.

Ionophores such as monensin and lasalocid are fed continuously in many calf-raising operations where coccidiosis is known to exist; the same is true for decoquinate. Manufacturers' warnings, dosages, and withdrawal times must be observed and are subject to change. Decoquinate is not toxic to young calves, but ionophores may be. Although various sulfa drugs were the first treatments for coccidiosis in animals, they are not used at present except to treat small groups or individual calves so sick they may not be eating or drinking well enough to ingest therapeutic dosages of drugs added to their feed or water. When sulfa drugs are used for treatment, it is beneficial to treat simultaneously with amprolium at therapeutic levels.

Although parasitologists question the efficacy of the aforementioned drugs in treating clinical coccidiosis, sulfaquinoxaline (13.2 mg/kg orally once daily) in combination with amprolium (10 mg/kg daily) for 5 days appears to provide a good clinical response. Strict adherence to recommended meat withdrawal is required when sulfonamides are used. Individual calves that are severely dehydrated may require supportive fluids; colloids; and, rarely, blood transfusions if colonic hemorrhage has been severe. Tenesmus may be so severe and persistent as to require epidural anesthesia to allow the calf or heifer to rest, eat, and drink.

Although the aforementioned drugs are used widely for prophylaxis in calves at risk for coccidiosis, they should not be thought of as the only means of control. Management practices that allow dirty environments, manure buildup, feeding on ground level, feed and water contamination by

TABLE 6.6 Drugs that Aid in Prevention and Treatment of Coccidiosis

Drug	Name	Use	Dose
Amprolium	Corid*	Prophylactic	5 mg/kg bwt for 21 days
		Therapeutic	10 mg/kg bwt for 5 days
Monensin	Rumensin†	Prophylactic	16.5–33.0 g/ton feed continuously or 1.0 mg/kg bwt for 28 days
Lasalocid	Bovatec‡	Prophylactic	1 mg/kg bwt continuously
Decoquinate	Deccox§	Prophylactic	0.5 mg/kg bwt for 30 days
Sulfamethazine	Several products available	Therapeutic	140 mg/kg bwt loading dose; then 70 mg/kg bwt for 5–7 days
Sulfadimethoxine		Therapeutic	55 mg/kg bwt loading dose; then 27.5 mg/kg bwt for 5–7 days
Sulfaquinoxaline		Therapeutic	13.2 mg/kg orally once daily for 3–5 days

*Corid (amprolium), Merial, Duluth, GA.
†Rumensin 60 (monensin sodium), Elanco Animal Health, Greenfield, IN.
‡Bovatec (lasalocid), Zoetis, Parsippany, NJ.
§Deccox (decoquinate), Zoetis, Parsippany, NJ.
Bwt, Body weight.

manure, and crowding should be corrected. If calves are kept in a clean environment, manure should be scraped away daily to prevent "splashing" of feces into bunks, troughs, waterers, and all over calves' bodies. If premises are cleaned and disinfected between consecutive groups of calves, the risk of coccidiosis is lowered tremendously. Unfortunately, many farmers would rather rely on a drug placed in the feed than do the required cleaning. Many of the coccidiostats are only effective for as long as fed, so calves become at risk if medicated feed is discontinued or there is an interruption in medicated feed or water consumption for some other reason. Therefore, anticoccidial drugs usually are included in the ration for extended periods such as from weaning through breeding age rather than based on manufacturer's recommendations (e.g., 30 days). Increasingly, calf starters and milk replacers including coccidiostats are being marketed for dairy calves. Although these drugs may not be necessary in preweaned calves, it is possible that clinical coccidiosis could occur as early as 2 to 3 weeks of age. Although the rumen is poorly developed in neonatal calves, lasalocid fed from day 1 improves gain and counterbalances coccidial infection (experimentally induced) before weaning. Careful attention to accurate dosing of ionophores in preweaned calves is especially important because this age of animal seems rather prone to toxicity, typically manifested as neurologic signs reminiscent of meningitis or salt poisoning. Certainly, if management conditions allow preweaning coccidiosis, mixed infections of the GI tract would be possible in 2- to 4-week-old calves. *C. parvum,* rotavirus, coronavirus, *E. coli,* or *Salmonella* spp. could also be involved. Obviously mixed infections could worsen the pathology.

Nematodes

Etiology

Intestinal nematodes are an important concern for pastured calves and growing heifers. Although current trends make pasturing of young dairy calves and heifers uncommon, consideration of intestinal parasite burdens is still worthwhile for confined heifers and essential for growing heifers on pasture. A basic understanding of parasites' life cycles and the geographic incidence of the various intestinal parasites is essential when making recommendations to owners. Pastured heifers require planned parasite control programs that include both management and anthelmintic components.

The major nematode parasites of the abomasum include *Ostertagia ostertagi, Trichostrongylus axei,* and *Haemonchus placei*. *O. ostertagi* (brown stomach worm) is the most important nematode in cattle because of its ability to undergo hypobiosis or arrested development of the L4 stage within the abomasum of infected young cattle. Arrested larvae reside in the lumen of gastric glands during seasons of the year that would likely interfere with the parasites' existence outside the host. Therefore, larvae acquired during late fall and early winter at temperatures found in northern climates persist in the host as inhibited larvae for weeks to months before maturing in late winter and spring. In southern temperate zones, larvae acquired during the spring become inhibited and finally mature in late summer or early fall. The biologic purpose of *O. ostertagi* hypobiosis is to avoid exposure of eggs and larval stages to weather not conducive to survival of the parasite. Therefore, harsh winters are avoided in the north, as are hot dry summers in southern zones. In the abomasal wall, hypobiotic larvae are apparently not targeted by the immune system for elimination,

even in previously exposed adult cattle. When arrested larvae emerge from the abomasal glands, they tend to do so with a vengeance that creates severe abomasal pathology and illness known as ostertagiasis type II. Maturation and emergence of large numbers of inhibited L4 larvae cause acute anorexia, weight loss, hypoproteinemia, and severe diarrhea as a result of abomasal mucosal injury, increased abomasal pH because of parietal cell dysfunction, and hyperplasia. The mortality rate is high with this form, although prevalence usually is low. The resultant greatly thickened and nodular abomasal wall has caused pathologists to describe the gross lesion as "Moroccan leather" in appearance. Because ostertagiasis type II occurs in the late winter and spring in northern zones and late summer or fall in southern areas, parasites may not be considered as the cause of illness in affected heifers. Inhibited larvae also are resistant to many commonly used anthelmintics. Ostertagiasis type I is more classically typical of nematode infections because pastured heifers acquire significant loads of larvae that mature to adults over approximately 3 weeks. Type I infections occur during peak pasture seasons and can result in diarrhea, weight loss, hypoproteinemia, or simply poor weight gain.

H. placei and other species found less commonly in cattle also possess the ability to undergo hypobiosis to avoid temperature extremes detrimental to survival outside the host. *H. placei* is pathogenic as a result of blood sucking that can lead to severe anemia.

T. axei may cause injury to the abomasal mucosa that leads to hypoproteinemia, altered digestion and intestinal defense mechanisms, and diarrhea. *Gongylonema* spp. also live in the abomasum and forestomach but are not thought to be major pathogens. Small intestinal nematodes include other *Trichostrongylus* spp., *Cooperia* spp., *Nematodirus helvetianus, Bunostomum phlebotomum, Toxocara vitulorum,* and several other parasites. *Bunostomum phlebotomum* and *Cooperia* spp. are bloodsuckers that damage the intestinal mucosa and create anemia. *N. helvetianus* is an extremely hardy, weather-resistant parasite that causes diarrhea in cattle when present in great numbers.

Large intestinal nematodes include *Oesophagostomum radiatum, Chabertia ovina, Trichuris discolor,* and *Trichuris ovis. O. radiatum* causes diffuse inflammatory reactions in the cecum and colon of cattle. This inflammation, subsequent nodule formation, and hemorrhage cause inflammatory bowel disease that results in diarrhea, weight loss, and hypoproteinemia in heavily parasitized animals. Subsequent long-term pathology and full or partial immunity allow *O. radiatum* nodules to become necrotic or calcified. These chronic lesions are grossly apparent in the bowel serosa and are responsible for the parasite being labeled the "nodular worm." Intussusceptions sometimes occur at the site of chronic *O. radiatum* lesions; therefore, pathologists theorize that the lesions may disrupt or alter normal intestinal motility.

Trichuris spp. occasionally are identified as the cause of severe diarrhea in heifers. *Trichuris* spp. are known as "whipworms" and concentrate in the cecum of cattle hosts.

The aforementioned nematodes all possess pathogenicity of varying degrees in either young or mature animals not previously exposed to parasites. Pastured animals, especially those pastured on lands that are grazed every year, are at risk. The first pasture season of an animal's life presents the greatest risk. Thereafter, partial or full immunity may be present in animals during their adult years. Fortunately, anthelmintics are available to counteract these parasites. Anthelmintics used appropriately, combined with pasture management, allow dairy heifers to be pastured successfully. All authorities agree that parasites are detrimental to calves and heifers, especially those on pasture. Much controversy exists, however, when the topic of worming adult dairy cattle is discussed.

Natural immunity (at least for cows that grazed pasture and acquired exposure to parasites as heifers) should protect adult cattle previously exposed to parasites. Attempts to demonstrate milk yield difference between wormed and nonwormed dairy cattle have given mixed results, and much debate exists as to the relative merits of adult cow deworming, particularly under current confinement housing management systems.

Clinical Signs

As with any parasitic infestation or infection, overt clinical signs may be present only in a few animals within a group. However, all animals in the group will harbor parasite loads. Mild nematode levels simply deter normal growth and gain rates in heifers without causing clinical signs of disease. Moderate levels of nematodes cause some animals in the group to show variable amounts of diarrhea, weight loss, poor hair coats, decreased appetite, hypoproteinemia, and anemia. Heavy nematode levels cause acute appearance of these signs and a greater prevalence of animals showing signs within the group. Appetite depression is consistent and contributory to weight loss or lack of weight gain and, as yet, is unexplained. The predominant types of nematodes in each herd will dictate the signs observed. For example, in type II ostertagiasis, acute hypoproteinemia, severe diarrhea, and inappetence would be observed in some animals within the group, and the time of year would not coincide with a typical pasture associated parasitic problem. If *Haemonchus* spp., *Bunostomum* spp., or *Cooperia* spp. predominate, anemia could be the major sign.

Ancillary Data

Anemia caused by blood loss and hypoproteinemia characterized by hypoalbuminemia are the major abnormalities detected on complete blood count and serum biochemistry. Abomasal pH increases as acid production decreases secondary to parietal cell dysfunction. Pepsinogen is not activated completely to pepsin, a proteolytic enzyme, because this activation requires a low abomasal pH. Therefore, increased plasma pepsinogen levels may be demonstrated when severe abomasal pathology exists as a result of *O. ostertagi* or less commonly *T. axei.* Few, if

any, veterinary diagnostic laboratories in North America routinely offer serum pepsinogen analysis on a commercial basis, however. Eosinophilia may or may not be present in WBC differentials and is not an essential or accurate aid to diagnosis. Fecal flotation and larval culture provide the definitive diagnostic tools. Very severe, acute infections with profuse diarrhea may be associated with electrolyte losses of Na^+, Cl^-, $HCO3^-$, and K^+.

Diagnosis

A definitive diagnosis requires identification of worm eggs or larvae in the feces of cattle having signs consistent with nematodiasis and ruling out other infectious or toxic diseases. When diarrhea is a major sign, salmonellosis, BVDV infection, coccidiosis, and toxicities that result in diarrhea must be ruled out.

Treatment and Control

Minimizing pasture contamination by parasite eggs and larvae is a major component of parasite control for dairy heifers. Heifers should be wormed before turnout and then at 3 and 6 weeks after turnout (or 3 and 8 weeks after turnout if ivermectin is used). This schedule helps to reduce recently ingested worm burdens before mature females begin to contaminate pastures with eggs, and the second treatment should kill ingested overwintered larvae that contaminate the pasture. The two-treatment program helps prevent the dramatic L3 load that generally occurs in northern climate pastures during late summer and early fall. Migration of L3 from manure to herbage occurs earlier during wet summers than dry ones, but L3 loads tend to peak in the fall in northern climates.

Although it may be ideal to select an anthelmintic specifically directed against the major nematodes present on each farm, it is more practical to select broad-spectrum anthelmintics that kill most types of nematodes. Available anthelmintics for nonlactating or nonbreeding-age dairy cattle are listed in Table 6.7. Label recommendations and changes in status of approval may occur with any of the drugs listed in this table. Therefore, practitioners should always verify that label approval exists for dairy animals. Pasture rotation with worming before movement to new pasture is another management tool seldom practiced with dairy operations because of limited acreage.

Although worming and parasite control are definitely beneficial to pastured heifers, the economic benefits of worming adult lactating dairy cows are controversial. Worming programs for lactating cattle may be justified if the cattle are pastured for a significant time each year. Worming would primarily benefit first calf heifers and newly acquired cattle that perhaps had not been pastured as heifers and thus are not likely to have parasite resistance. Dairy herds with a high percentage of first calf heifers and using pasture for a portion of the year probably can justify anthelmintic treatment of lactating cattle.

Economic justification may be lacking for use of anthelmintics in lactating cows in totally confined herds whose heifers never are pastured. Available anthelmintics for lactating dairy cattle are listed in Table 6.8.

TABLE 6.7 Anthelmintics Approved for Nonlactating Dairy Animals (Data Assembled from Information Given on Manufacturer's Labels)

Drug	Dose	Slaughter Withdrawal Time (days)	Spectrum and Comments
Albendazole	10 mg/kg PO	27	GI nematodes, including hypobiotic *Ostertagia* L4, lungworms, *Moniezia* (tapeworms), adult liver flukes (*Fasciola* spp.) Not for use in female dairy cattle of breeding age; potentially teratogenic if administered in early pregnancy
Doramectin injectable	200 µg/kg SC	35	GI nematodes, including hypobiotic *Ostertagia* L4, lungworms, grubs, sucking lice, mites Not for use in female dairy cattle of breeding age or in veal calves
Doramectin (pour-on)	500 µg/kg topically	45	GI nematodes, including hypobiotic *Ostertagia* L4, lungworms, grubs, sucking and biting lice, mites Not for use in female dairy cattle of breeding age or in veal calves
Eprinomectin (pour-on)	500 µg/kg topically	0	GI nematodes, including hypobiotic *Ostertagia* L4, lungworms, grubs *(Hypoderma)*, sucking and biting lice, mites, and horn flies Effective when applied to wet cattle
Eprinomectin (injectable)	1 mg/kg SC 5 mg/kg PO	8–13 (depending on product)	GI nematodes, including hypobiotic *Ostertagia* L4, lungworms Not for use in female dairy cattle 20 months or older or in veal calves
Fenbendazole	10–15 mg/kg PO	8–13 (depending on product)	GI nematodes, including hypobiotic *Ostertagia* L4, lungworms, tapeworms; on rare occasion, fenbendazole may cause bone marrow suppression in calves younger than 1 month of age

TABLE 6.7 Anthelmintics Approved for Nonlactating Dairy Animals (Data Assembled from Information Given on Manufacturer's Labels)—cont'd

Drug	Dose	Slaughter Withdrawal Time (days)	Spectrum and Comments
Ivermectin (injectable)	200 µg/kg SC	35	GI nematodes (adult and larvae), including hypobiotic *Ostertagia* L4, *Dictyocaulus* (lungworm), grubs *(Hypoderma)*, sucking lice, and mites Not for use in female dairy cattle of breeding age or in veal calves
Ivermectin (pour-on)	500 µg/kg topically	48	GI nematodes, including hypobiotic *Ostertagia* L4, *Dictyocaulus* (lungworm), grubs *(Hypoderma)*, sucking and biting lice, mites, and horn flies Not for use in female dairy cattle of breeding age or in veal calves
Levamisole	Varies with formulation	7	GI nematodes (adult), lungworms Not for use in female dairy cattle of breeding age
Moxidectin (injectable)	200 µg/kg SC	21	GI nematodes (adult and larvae), including hypobiotic *Ostertagia* L4, *Dictyocaulus* (lungworm), grubs *(Hypoderma)*, sucking lice, and mites Not for use in female dairy cattle of breeding age or in veal calves <3 months of age
Moxidectin (pour-on)	500 µg/kg topically	0	GI nematodes, including hypobiotic *Ostertagia* L4, *Dictyocaulus* (lungworm), grubs *(Hypoderma)*, sucking and biting lice, mites, and horn flies Not for use in veal calves
Morantel tartrate	0.44 g/100 lb PO	14	GI nematodes (adult)
Oxfendazole	4.5 mg/kg PO	7	GI nematodes, including hypobiotic *Ostertagia* L4, lungworms, tapeworms Not for use in female dairy cattle of breeding age
Clorsulon	7 mg/kg	8	*Fasciola hepatica* adults and late immature larvae
Clorsulon and ivermectin	1 mL/110 lb SC	49	GI nematodes (adult and larvae), including hypobiotic *Ostertagia* L4, lungworms, *F. hepatica* adults, grubs *(Hypoderma)*, sucking lice, and mites Not for use in female dairy cattle of breeding age or in veal calves

GI, Gastrointestinal; *PO,* oral; *SC,* subcutaneous;

TABLE 6.8 Anthelmintics Approved for Lactating Dairy Cattle (Assembled from Manufacturers' Labels)

Drug	Dose	Withdrawal Period for Slaughter and Milk (days)	Spectrum
Eprinomectin (pour-on as Ivomec Eprinex)	500 µg/kg topically	Meat = 0 Milk = 0	GI nematodes, including hypobiotic *Ostertagia* L4, lungworms, grubs *(Hypoderma)*, sucking and biting lice, mites, and horn flies Effective when applied to wet cattle
Fenbendazole	5 mg/kg PO	Meat: 8-13 (depending on product) Milk: 0	GI nematodes (adult), lungworms
Morantel tartrate	0.44 g/100 lb PO	Meat = 14 Milk = 0	GI nematodes (adult)
Moxidectin (pour-on)	0.5 mg/kg topically	Meat = 0 Milk = 0	GI nematodes, including hypobiotic *Ostertagia* L4, lungworms, grubs *(Hypoderma)*, sucking and biting lice, mites, and horn flies

GI, Gastrointestinal; *PO,* oral.

Trematodes

Etiology and Clinical Signs

Liver flukes are a greater problem in beef than dairy cattle, but certain geographic areas harbor flukes and their intermediate hosts, thereby representing risks for pastured dairy heifers and cows. *Fasciola hepatica* is found most commonly in certain areas along the Gulf Coast and western United States; it is rare in the northern United States. Cattle, along with sheep, are the primary definitive hosts of *F. hepatica* and shed eggs in feces. Eggs require a moist environment to hatch miracidia, which find a snail intermediate host. After a complicated reproductive cycle in the snail, the parasite eventually produces metacercariae, which are ingested by grazing animals. Metacercariae invade the duodenum, and immature flukes then penetrate the gut and seek out the liver, where they penetrate the capsule, migrate in the parenchyma, and eventually reside in the bile ducts. This wandering through the gut and liver parenchyma creates a great deal of pathology, which in heavy infestations may cause hypoproteinemia, anemia, reduced appetite, peritonitis, and importantly, predispose to clostridial diseases such as bacillary hemoglobinuria *(C. hemolyticum)*, or black disease (*C. novyi).*

Fascioloides magna, the deer liver fluke, is a large fluke that can infect cattle and sheep. The fluke is found along the Gulf Coast, the Great Lakes region, and the northwestern United States. Although cattle can be infected, the resulting parenchymal cyst does not allow egg release, thus making cattle dead-end hosts. In deer, the natural host, thin-walled parenchymal cysts are formed that allow eggs to emerge into bile ducts. *F. magna* is particularly vicious in sheep because continued migration without encystment is the rule. Because infections are not patent in cattle, liver condemnation or gross postmortem lesions are the major consequences of *F. magna* infection.

Dicrocoelium dendriticum occurs in many geographic regions around the world and in a few areas in the northeastern United States (including the central New York region) and Canada. The life cycle is complex, with both the land snail *(Cionella lubrica)* and the black ant *(Formica fusca)* necessary for transmission back to a definitive host, such as cattle, sheep, goats, horses, pigs, and people. Metacercariae (in ants) ingested by host cattle penetrate the duodenum and directly enter the bile ducts, where they usually cause little detectable illness. However, heavy infestations can occasionally inflame or obstruct biliary ductules and the gallbladder. Most cases are self-limiting.

Fasciola gigantica is limited to tropical regions and will not be discussed here.

Paramphistomum, the rumen fluke, is thought not to be highly pathogenic in the United States but can cause illness in tropical zones. Ingested metacercariae may encyst in the duodenum or migrate to the duodenum wall, where they stay for weeks before migrating upstream to become adults in the rumen and reticulum. Acute paramphistomiasis, which is rare in the United States, refers to duodenal pathology created by large populations of wandering and invading immature flukes.

Diagnosis

Identification of fluke eggs in the feces is possible for *F. hepatica* and *D. dendriticum*. *Paramphistomum* spp. eggs can be confused with *F. hepatica*, especially because both are operculated, but *F. hepatica* eggs are slightly smaller and stained yellow. Routine sampling of feces from 10 to 15 at-risk animals is indicated when *F. hepatica* is suspected. An ELISA serologic test also has been developed to diagnose *F. hepatica* infections. Necropsy specimens are very helpful whenever fluke infestations are suspected.

Treatment

Table 6.7 also lists the available drugs to counteract fluke infestation in nonlactating dairy heifers. Albendazole, clorsulon, and clorsulon–ivermectin are effective against adult and late immature *F. hepatica*. Albendazole is effective against *F. magna* and *D. dendriticum*. Control measures include avoidance of pastures harboring intermediate hosts or killing intermediate hosts such as snails with molluscicides. Veterinarians practicing in endemic fluke regions should be familiar with diagnosis and control measures if their clients pasture heifers. Vaccination against clostridial conditions such as bacillary hemoglobinuria and black disease is prudent in endemic areas.

Cestodiasis

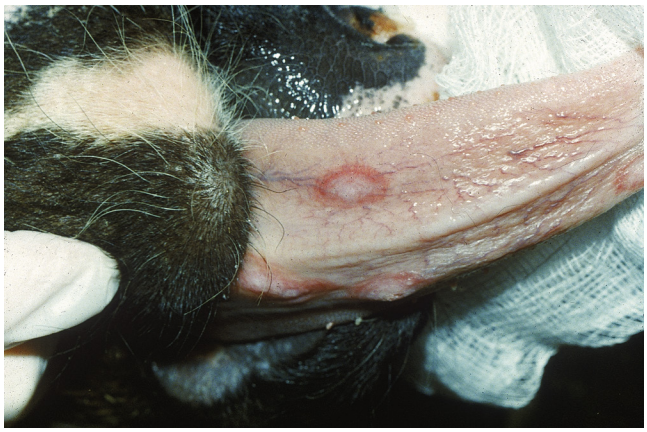
Moniezia benedeni is the small intestinal tapeworm of cattle and is thought to be nonpathogenic. Oribatid mites are the intermediate hosts, and after ingestion of infective cysticercoids by cattle, the worms mature over 2 months and then are shed spontaneously several months later. Treatment with albendazole is effective when necessary. Other tapeworms such as *Taenia saginata,* the beef tapeworm of humans; *Taenia hydatigena* (adults in dogs); and *Echinococcus granulosus* are not major parasites of dairy cattle in the United States and will not be discussed here.

Papular Stomatitis

Etiology

Bovine popular stomatitis virus (BPSV) is a member of the *Parapox* virus genus, within the Pox virus family, and is very closely related, and therefore hard to distinguish from other species members of this genus, including the virus of pseudocowpox and contagious ecthyma virus of sheep and goats. The application of modern molecular diagnostic techniques for more sensitive and specific differentiation of these different species of *Parapox* viruses has been prompted by not just altruistic scientific advance but also by the desire to rapidly identify the precise viral agent causing suspicious vesicular lesions of ruminants and to trace the cause of skin lesions in humans because

• **Fig. 6.15** Typical raised circular lesions of papular stomatitis on the muzzle and lips of a calf.

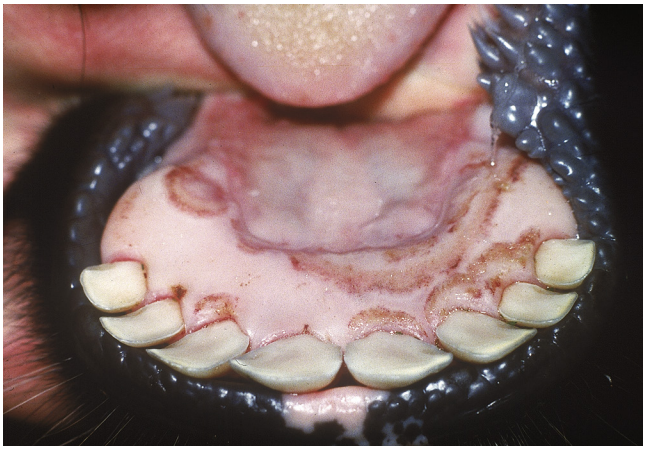


• **Fig. 6.16** Circular papular stomatitis lesions on the ventrum of a calf's tongue.

all of these agents are zoonotic. Such differentiation is now possible using quantitative PCR (qPCR) techniques. In cattle, the disease is spread by contact between infected and noninfected calves or via common feeding devices or containers. The virus is quite contagious in young calves, and it is worth remembering that it also can cause lesions on the skin of humans working with calves. Such zoonotic infection creates lesions similar to the milker's nodules of pseudocowpox in people. Papular stomatitis virus lesions on the muzzle and in the oral cavity of calves frequently are confused with erosions caused by BVDV or potentially other vesicular diseases such as foot and mouth, vesicular stomatitis, or epizootic hemorrhagic disease. It was first described in the United States in 1960.

Clinical Signs

A raised papule on the muzzle or nares is the most commonly observed lesion because of the external nature of the lesion (Fig. 6.15). Papules on the palate, tongue, or lips probably are more common but are less likely to be observed (Fig. 6.16). Although red and raised on the external mucosa, the lesions may appear crusty or brownish-yellow in the oral



• **Fig. 6.17** Papular stomatitis lesions caudal to the incisor teeth and ventral to the tongue in a calf. The lesions are brownish-yellow in color, are slightly raised, and have rough edges.

cavity and have roughened edges (Fig. 6.17). Oral cavity lesions may be flat and therefore confused with erosions. Some papules develop a necrotic white center that sloughs, leaving an ulcerated area within the raised papule. Papular stomatitis lesions may cause an affected calf to show mild salivation and reluctance to nurse or eat, although most calves show no clinical signs associated with the lesions. Such signs usually lead to examination of the oral cavity and subsequent diagnosis. Many, if not most, cases are asymptomatic and go undiagnosed.

Calves from several weeks to several months of age are most commonly affected, but the disease has also been observed in older growing cattle. Lesions also occur in the esophagus and forestomach mucosa but are only detected in those locations during necropsy examination. Such lower alimentary lesions may be confused with infectious bovine rhinotracheitis (IBR)– or BVDV-induced lesions.

Severe illnesses or immunodeficiencies frequently allow extensive proliferation of papular stomatitis virus and subsequently more advanced lesions may be seen. When such severe lesions occur, they are clinically suspected to be BVDV related. In fact, concurrent BVDV infection in immunocompetent calves or poor-doing BVDV-PI calves greatly accentuates papular stomatitis lesions when both viruses are present. Similarly, IBR virus and chronic bacterial infections predispose to worsening of existing papular stomatitis virus lesions. Clinical errors are most common, however, when BVDV-PI calves with a superinfection with cytopathic (CP)-BVDV or chronic concurrent diseases develop amplification of papular stomatitis lesions. Most spontaneous, uncomplicated papular stomatitis lesions resolve within several weeks in immunocompetent animals and are diagnosed by inspection. Severe cases may require biopsy and viral isolation, immunohistochemistry, or PCR to differentiate the lesions from BVDV. Intracytoplasmic, eosinophilic inclusions are typical in the cytoplasm of degenerating cells.

Although the disease may be fatal, most early reports that cited extensive lesions and fatalities probably represented concurrent BVDV infections.

Treatment and Prevention

No specific treatment or method of prevention exists other than to minimize spread by housing calves separately and not using common feeding devices or buckets.

Infectious Diseases Of The Gastrointestinal Tract—Adults

Actinobacillosis

Etiology

Actinobacillus lignieresii, a gram-negative pleomorphic rod, is a normal commensal organism in the oral flora of cattle. Injuries to the oral mucosa or skin that become contaminated with *A. lignieresii* may develop soft tissue infection characterized by an initial cellulitis that evolves into a classical pyogranulomatous infection that can be confused with neoplasia or actinomycosis. Sulfur granules, which are yellow-white cheesy accumulations containing the organism, develop within pus or pyogranulomatous soft tissue lesions associated with *A. lignieresii* infection.

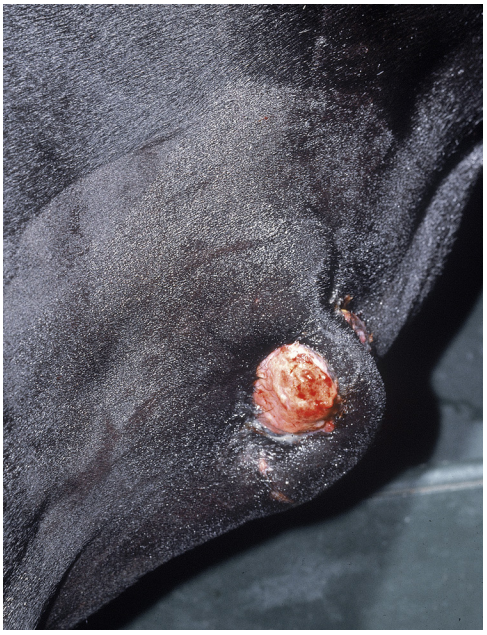
"Wooden tongue," a soft tissue infection of the tongue, is the classical example of *A. lignieresii* infection in cattle, but soft tissue granulomas developing around the head, neck, or other body areas are common as well.

Granulomas of the esophagus, forestomach, and occasionally other visceral locations also are possible. Lymphadenitis, lymph node abscesses, and infectious granulomas originating from lymph nodes may follow soft tissue infections of the oral cavity or pharynx. Extremely fibrous feed material has been incriminated as the cause of mucosal injury that allowed opportunistic *A. lignieresii* infection as a herd problem in dairy heifers. Direct inoculation of the organism into mucosal wounds can occur in the oral cavity, esophagus, and forestomach. Inoculation probably occurs from oral secretions or saliva when soft tissue wounds of the skin are infected at sites distant from the oral cavity.

Clinical Signs

A textbook case of acute wooden tongue in cattle appears as a diffusely swollen, firm tongue that fills the oral cavity. Firm or fluctuant intermandibular swelling usually accompanies the inflammatory enlargement of the tongue (Fig. 6.18). Excessive salivation is observed, and the swollen tongue may protrude from the oral cavity. Anorexia is relative or complete because the tongue has reduced mobility and may be injured by the teeth if chewing is attempted. Fever is present in acute infections but frequently absent in subacute or chronic cases. Distended salivary ducts appearing as ranulae may be observed ventral to the tongue. Although this is the typical "textbook" *A. lignieresii* clinical description, many cases do not involve the tongue but instead cause soft tissue infection in the pharynx or other regions of the mouth. The specific location of the disease is likely associated with an injury to the mucosa in that area. Historically, poor-quality hay containing sticks or briars has been associated with the disease in heifers.

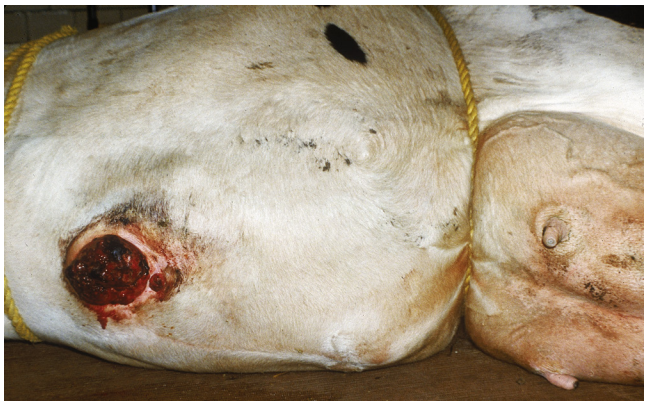
• **Fig. 6.18** Acute wooden tongue with intermandibular cellulitis and salivation.



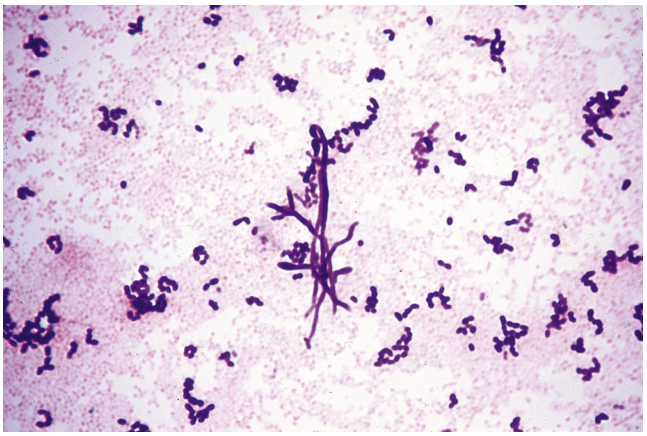
• **Fig. 6.19** Atypical actinobacillosis granuloma in the jugular furrow of a cow secondary to a perivascular administration of concentrated dextrose solution.

Chronic wooden tongue lesions consist of pyogranulomatous masses and fibrosis of the tongue or other soft tissue sites that are infected. Weight loss and marked salivation are common in chronically infected cattle.

Atypical *A. lignieresii* infection is characterized by granulomas, pyogranulomas, or lymph node abscesses. Serous or mucoid nonodorous pus may drain from abscessed infections. Granulomas are raised, red, fleshy to firm in consistency, and contain sulfur granules. Granulomas may occur in the oral cavity, esophagus, forestomach, or other visceral locations. In addition, external granulomas have been observed in the nares, eyelids, face, pharyngeal region, neck, limbs, and abdomen (Fig. 6.19). Infection of abomasopexy toggles or sharp incision sites also has produced these granulomas (Fig. 6.20).



• **Fig. 6.20** Actinobacillosis granuloma at the site of an abomasopexy incision.



• **Fig. 6.21** Gram stain of aspirate from lumpy jaw lesion. Gram-positive branching filamentous rods are characteristic of *Actinomyces bovis*.

Diagnosis

Excision or biopsy of granulomas to provide material for bacterial culture and histopathology is the only means to confirm a diagnosis of *A. lignieresii* infection. Clinical appearances and the presence of sulfur granules are suggestive of diagnosis but not specific because the masses must be differentiated from actinomycoses, botryomycosis, neoplasia, parasitic or foreign body granuloma, and exuberant granulation tissue. Acute wooden tongue can be diagnosed by aspiration of fluid-distended salivary ranulae, the tongue itself, or intermandibular phlegmon when present. Aspirates are submitted for cytology and culture. Chronic wooden tongue lesions are best diagnosed by biopsies submitted for both culture and histopathology.

Treatment

Wooden tongue and other lesions of *A. lignieresii* infection often respond to systemic sodium iodide therapy. Intravenous sodium iodide (20% solution) is an extremely irritating preparation that should be administered IV only by a veterinarian. It is administered at a dose of 70 mg/kg body weight. This dose is repeated at 2- to 3-day intervals until iodism occurs. Alternatively, oral organic iodide can be fed (1 oz/450 kg body weight, daily) after initial IV therapy until iodism occurs. Unfortunately, parenteral iodide preparations are not currently permitted for use in lactating dairy cattle in the US. Signs of iodism include serous lacrimation, seromucoid nasal discharge, and scaly dandruff-like skin appearing on the face and neck of treated cattle. For acute wooden tongue lesions, response to iodine therapy is usually dramatic. Subacute lesions respond more slowly, and chronic lesions carry a guarded prognosis. In calves chronic lesions of the tongue often prevent the patient from eating and starvation may ensue. Sulfonamides, tetracycline, or beta-lactam antibiotics also may be useful and can be used alone or in conjunction with iodine therapy for severe *A. lignieresii* infections of the tongue.

Treatment of *A. lignieresii* granulomas consists of debridement or debulking (if the involved anatomic area allows surgical intervention), coupled with medical therapy as outlined previously. Recurrent or severe lesions can be treated with combined antimicrobial therapy as discussed previously. Cryosurgical treatment has been combined with surgical debulking of some *A. lignieresii* granulomas and appears most effective when mushrooming granulomas attached to a narrow skin base are selected for therapy.

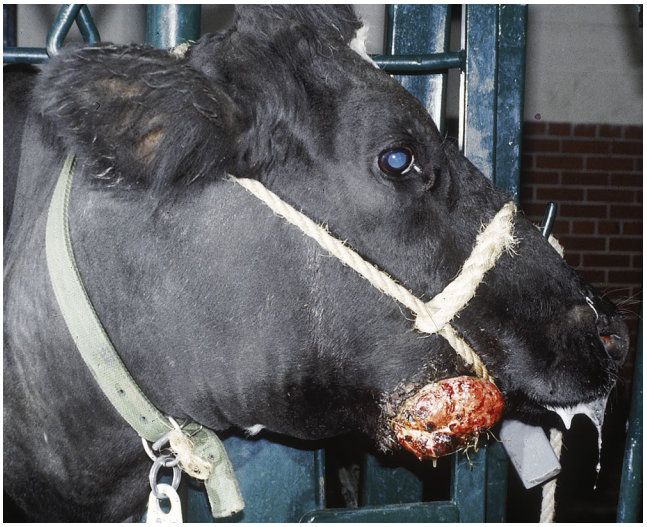
The exact reason that iodides are effective in *A. lignieresii* infection is not known. Suggested mechanisms include penetrance into granulation tissue and destruction of organisms, simple decrease in the granulomatous response, and combinations of activity against *A. lignieresii* and the granulomatous inflammatory response. Iodides are unlikely to cause abortion in cattle, although some commercially available preparations of injectable sodium iodide are labeled with a warning that forbids their use in pregnant cattle. Prevention is best accomplished by feeding hay or other forage that does not contain sticks or other foreign bodies that may damage the oral mucosa.

Actinomycosis

Actinomyces bovis, a gram-positive filamentous organism (Fig. 6.21) that can assume many forms, is the cause of lumpy jaw in cattle and occasionally causes granulomatous infection in other areas of the body. In young cultures, diphtheroid organisms are observed, but in older cultures and crushed preparations of sulfur granules obtained from pus, the organism may be filamentous, branching, coccoid, club-shaped, or diphtheroid. Infection with *A. bovis* typically results in formation of "sulfur granules" in pus or infected tissue. These so-called sulfur granules contain large numbers of the organism. In older literature, the term *actinomycosis* implied granulomatous infections containing sulfur granules and did not differentiate *A. lignieresii* or staphylococcal infections from those caused by *A. bovis.*

The organism is difficult to culture to the degree necessary for bacterial susceptibility testing. This fact contributes to the dearth of scientific information regarding the appropriate therapy for lumpy jaw in cattle. Many other species of *Actinomyces* have been studied in humans, and comparative information from these studies suggests that *A. bovis* is much more resistant to antibiotic therapy than human isolates such as *Actinomyces israeli*.

• **Fig. 6.22** Early lumpy jaw lesion consisting of an edematous soft tissue swelling overlying a painful, firm, bony swelling on the mandible.



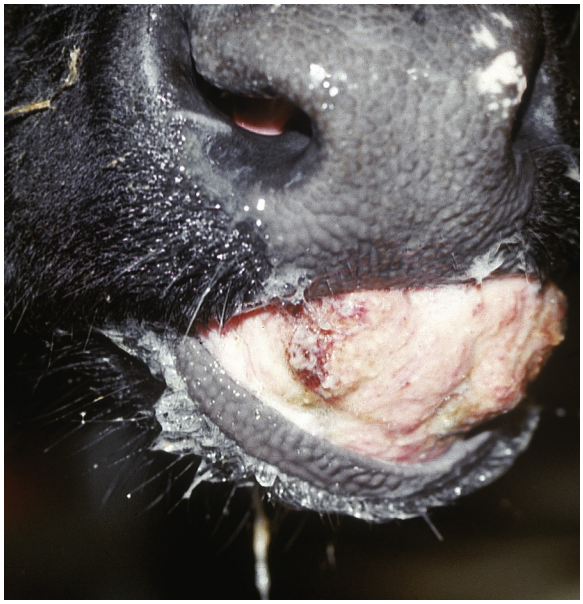
• **Fig. 6.23** Severe *Actinomyces bovis* infection of the mandible with ulceration of the skin and granulomatous proliferation.

Lumpy jaw is a debilitating disease of cattle resulting from infection of the mandible or maxilla by *A. bovis.* The organism has been described as a normal inhabitant of the oral flora and digestive tract of cattle. It is assumed that infection of bones and teeth occurs after injury to the oral mucosa by fibrous feeds or dental eruption (this may be a reason that lumpy jaw seems most common in young adult cattle) and subsequent inoculation of *A. bovis.* The organism also may penetrate around the alveoli of the teeth or contaminate skin wounds via common feed and water troughs. Dr. Rebhun observed one herd with an epidemic of lumpy jaw, with 7 of 60 cows affected. The point source cow had a large, draining, lumpy jaw lesion, and all cows ate silage twice daily from a feed bunk made of coarse boards. Discharge of the organism from the point source cow certainly contaminated the sideboards and bunk. Whether the organism had gained access through the oral cavity, injury to the oral cavity by wood splinters, or skin puncture from wood slivers on the sideboards could not be ascertained. Lumpy jaw usually is a sporadic infection, but as in the aforementioned herd, can be an epidemic or endemic herd problem.

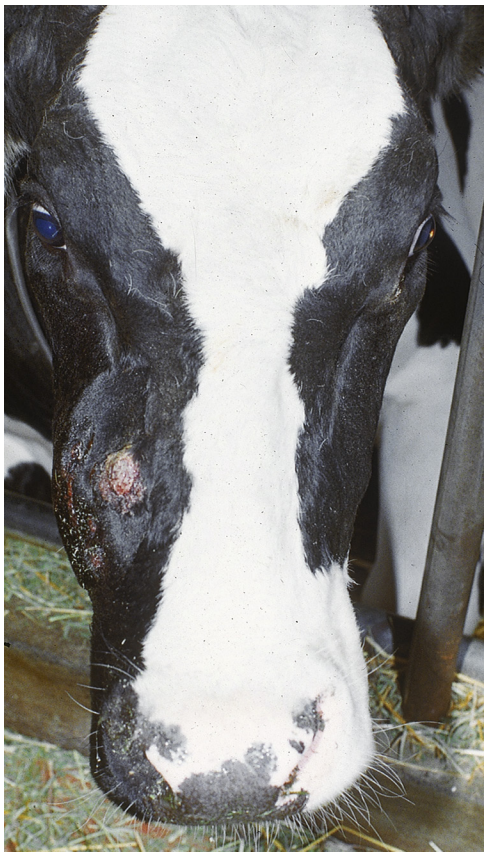
Rarely, *A. bovis* causes infectious granulomas on soft tissues similar to those caused by *A. lignieresii.* Granulomas caused by *A. bovis* have been identified in the trachea, testes, and digestive tract.

Clinical Signs

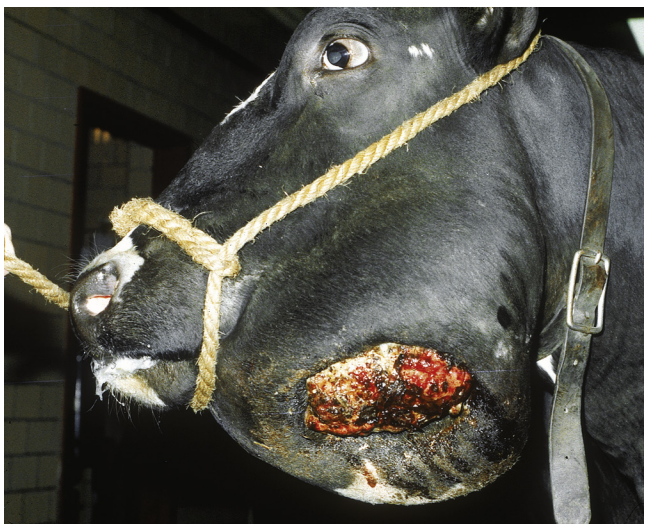
Early *A. bovis* infections of the mandible or maxilla appear as warm, painful swellings consisting of distinct edema overlapping a firm, painful, bony swelling (Fig. 6.22). Such early infections easily could be confused with a traumatic injury. Over a period of weeks, however, bone enlargement becomes obvious and soft tissue edema much less apparent. Salivation and some difficulty in eating may be observed, but inappetence and weight loss seldom are a problem in early cases. After the infection is established in bone, the swelling becomes hard and often painful (Figs. 6.23 to 6.25). Severe cases will have distortion of the teeth anchored in the affected bone when the mouth is examined. External swelling is merely the "tip of the iceberg" as established by skull radiographs that confirm severe osteomyelitis with multifocal radiolucencies caused by rarefaction of bone. Pyogranulomatous infection of bone and associated soft tissues evolves in untreated cases, and these animals will have granulomas develop at the site of draining tracts through the skin or into the oral cavity (Fig. 6.26). Because of distortion, malocclusion, or loss of teeth, eating becomes more difficult for severely affected cows. Salivation, reduced appetite, hesitant attempts to chew, and dropping food from the mouth may be observed. Oral mucosal or tongue lacerations may be apparent. Draining tracts discharge copious quantities of serous or mucopurulent pus that should be considered infectious to other cows.



• **Fig. 6.24** Actinomycosis of the mandibular symphysis region in a cow.



• **Fig. 6.25** Actinomycosis of the maxilla.



• **Fig. 6.26** Advanced lumpy jaw with draining tracts.

Diagnosis

Absolute diagnosis requires a tissue core biopsy or fluid aspirate to identify the causative organism. Core biopsies also allow histologic confirmation. Radiographs confirm osteomyelitis with multiple radiolucent zones and proliferation of periosteal bone (Figs. 6.27 and 6.28). Radiographs also help differentiate lumpy jaw from bony neoplasia; tooth root infections; fractures; sequestra, and when the maxilla is involved, sinusitis.

Treatment

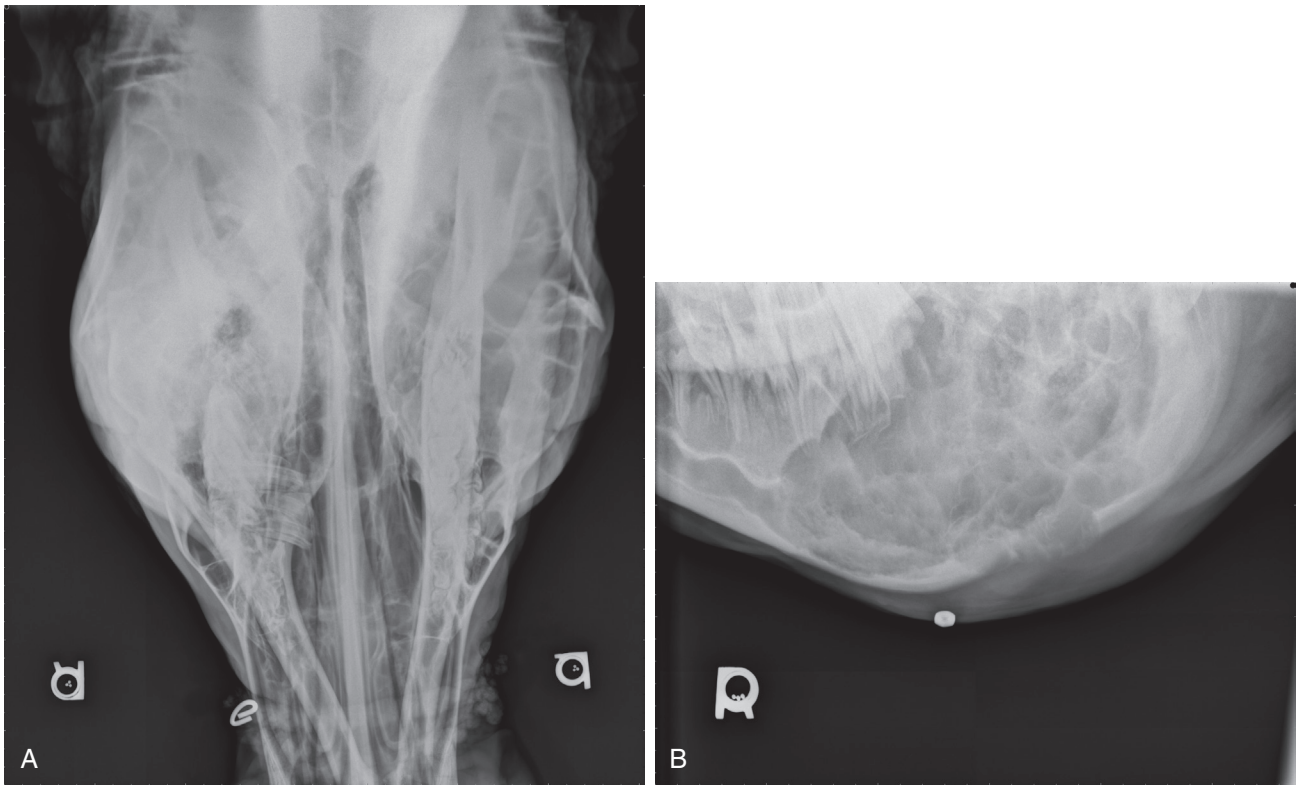
Recommended treatment for lumpy jaw usually includes sodium iodide, but this treatment is often ineffective and should be considered an adjunct, at best, to appropriate antibiotic therapy.

Any discussion of treatment also must allow for the tremendous variation in the severity of osteomyelitis caused by *A. bovis.* Basketball-sized lesions are unlikely to respond to any therapy, but early lesions may be resolved successfully by several protocols. Therefore, treatment is best instituted early. Long-term antibiotic therapy is necessary for well-established infections, and this fact makes owners reluctant to treat cows that do not appear ill and that are still producing well. Ironically, when this same untreated cow eventually becomes ill as the lesion enlarges, many owners will then want something to be done.

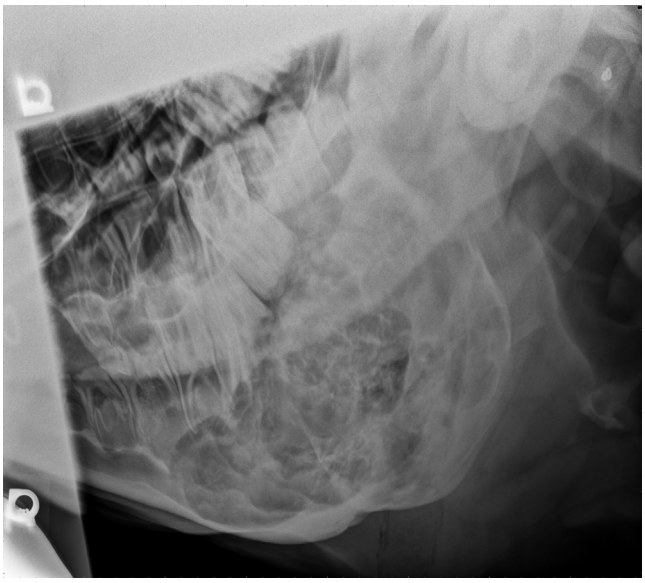
Streptomycin or penicillin–streptomycin combinations have been the drugs of choice for lumpy jaw; unfortunately, streptomycin and penicillin–streptomycin combinations no longer are available for use in cattle in North America. Therefore, other treatments will need to be considered. In one comparative study, erythromycin was active in vitro against *A. bovis* when an MIC of 0.06 to 0.12 was achieved. Therefore, erythromycin may be a good choice. Isoniazid can be used at 10 to 20 mg/kg orally every 24 hours for 30 days, and rifampin can be used at 20 mg/kg orally every 24 hours for 30 days or used at 5 to 10 mg/kg orally every 24 hours and combined with procaine penicillin at 22,000 U/kg SC every 24 hours for 30 days. Isoniazid may cause abortion, should be used with caution in pregnant cattle, and both isoniazid and rifampin represent inappropriate extra-label drug use in the United States. Because antibiotic therapy necessitates prolonged administration and may involve extra-label use of drugs, the implications of such therapy should be discussed with owners before treatment is begun. Use of rifampin in the US currently requires the owner to guarantee that neither milk nor meat from that individual will be sold commercially.

Current recommendations include penicillin 22,000 U/kg once daily and sodium iodide IV (30 g/450 kg) administered once or repeated at 2- to 3-day intervals until iodism occurs. Parenteral iodide preparations are not currently permitted in lactating dairy cattle in the US. Alternatively, organic iodides can be fed at 30 g/450 kg body weight once daily until iodism occurs. Duration of therapy is dependent on the severity of the lesion and response to therapy. We have also had some success in chronic cases, even some with substantial bony involvement via the placement of antibiotic-coated (penicillin or erythromycin) beads into the lesion. Long-term antibiotic therapy has resulted in a surprising cure in a few advanced cases.

Surgery has been suggested and still is used by some as treatment for lumpy jaw of the maxillae. Surgical debulking or removal of large pyogranulomas projecting from the skin of advanced cases may reduce the size of the lesion. Surgical debulking may incite severe hemorrhage. In addition, the affected bone may be further compromised or



• **Fig. 6.27** Dorsoventral (**A**) and lateral (**B**) and skull radiographs of a mature Holstein cow affected with advanced lumpy jaw of the right mandible. Osteomyelitis and characteristic multifocal radiolucencies are present. A draining tract was present at the ventral mandibular margin.



• **Fig. 6.28** Lateral oblique radiograph of 6-year old Brown Swiss cow with an advanced lumpy jaw lesion. Note the bony resorption around the mandibular cheek tooth; oblique views can help "skyline" dental alveoli to assess tooth root involvement.

fractured if overzealous debridement and curettage are performed. Again, the external swelling or masses are just the tip of the iceberg. Loose teeth may require extraction, and fistulous tracts may be flushed with iodine solution as ancillary aids.

Vesicular Stomatitis

Etiology

Vesicular stomatitis virus (VSV) causes lesions that may be indistinguishable from those of foot-and-mouth disease (FMD) in cattle and pigs. Horses also may be infected with VSV, but FMD does not occur in horses. The causative virus of VSV is a member of the genus *Vesiculovirus* in the family Rhabdoviridae, and two distinct serotype groups—Indiana and New Jersey types—are recognized. Each of these major types may be further subdivided into subtypes. The New Jersey VSV type tends to be more pathogenic in cattle. VSV is considered endemic in parts of South America, Central America, and the southwestern United States. In recent years, many livestock operations in the intermountain West have been quarantined after detection of VSV infection in cattle and horses. VSV usually occurs during the summer and fall, but one large epidemic developed during the late fall and early winter in California during 1982 and 1983. In more recent outbreaks, the disease has sporadically occurred in Texas, New Mexico, and Arizona in spring and early summer and then proceeded to spread northward into Colorado, Idaho, Montana, Idaho, Utah, and as far north and west as Nebraska. The general trend for cases to occur in the summer and fall is suggestive of an arthropod vector. Attention has focused on a species of midge, *Culicoides sonorensis,* as a likely vector for VSV because viral replication in multiple tissues of this insect has been documented following experimental VSV infection. Ingestion of infected grasshoppers has also

been shown to induce infection in cattle. Other insects, such as blackflies and sandflies, may act as mechanical vectors for transmission among animals. Many different animals can be infected by VSV, including sheep, goats, wildlife, birds, and insects.

During outbreaks, the virus spreads rapidly from infected animals through secretions and aerosol transmission. Intact skin is not penetrated by VSV, but abrasions or injuries allow infection through the skin and may explain the spread of teat lesions through infected herds by milking machines. When epidemics occur, morbidity is high—especially in dense populations of animals—but mortality usually is low.

Clinical Signs

After VSV is introduced to susceptible animals, clinical signs of varying intensity occur. Classical signs include salivation, fever, lameness, and teat lesions. Fever may precede the more obvious signs because viremia is short-lived. Blanched lesions of the oral mucosa, coronary band, and teats evolve into vesicles that rupture and then slough the involved mucosa to leave denuded surfaces. Within the oral cavity, the lips, tongue, gums, or other areas may be involved. Obvious problems with mastitis occur when teat lesions are widespread. Many animals in some outbreaks have minimal or subclinical lesions that escape detection. When the disease is signaled by a few animals with obvious lesions, physical examination of other animals on the premises frequently will reveal small erosions or ulcers resulting from earlier infection. As with many diseases, the severity of disease varies greatly based on serotype of VSV, density of the population infected, concurrent diseases, and other factors. Many of these signs cannot be distinguished from FMD lesions.

People working with VSV-infected animals or with VSV in laboratories can become infected asymptomatically or develop signs of fever, muscular aches and pains, and possibly lip blisters.

Infected cattle usually recover; mortality rates are low. However, economic losses resulting from decreased production are profound. Oral lesions cause infected cows to eat less, thereby affecting production. Lameness, if present, further deters appetite and access to feed. Teat lesions represent the most disastrous consequence of VSV because mastitis can easily follow incomplete "milk-out" as a consequence of the teats being painful. Therefore, although the mortality rate of the natural disease is low, the cull rate and economic losses can be catastrophic to dairy farmers.

Diagnosis

Whenever a diagnosis of vesicular stomatitis is possible, state and federal regulatory veterinarians should be alerted for help in diagnosis and ruling out FMD. Currently, the diagnosis is definitively arrived at either via serologic tests or identification of virus in tissue samples.

Treatment and Control

Common sense measures such as milking cows with teat lesions last, using aggressive disinfection practices in the milking parlor, reducing animal density, attempting isolation of clinical cases, and reducing stress are the only means of treatment. Softened feeds may be more easily ingested by affected animals. Secondary bacterial infections of lesions may necessitate occasional use of antibiotics.

Control measures and containment must be left to regulatory veterinarians. On Colorado dairies that experienced repeated annual outbreaks, the rate and scope of spread on infected dairies have been curtailed when aggressive insect control measures were initiated soon after detection of the disease. Humans working with infected cattle should wear gloves and perhaps masks.

Bluetongue

Etiology

Bluetongue virus (BTV) is an *Orbivirus* transmitted by *Culicoides* gnats from infected to noninfected ruminants. BTV is almost uniformly asymptomatic in cattle. Cattle and wild ruminants are thought to be reservoirs of BTV, but sheep suffer a more apparent clinical disease characterized by fever, edema, excessive salivation, frothing, and hyperemia of the nasal and buccal mucous membranes. These acute signs in sheep progress to crusting, erosions, and ulcers of the mucous membranes. In addition, affected sheep are lame, resulting from both coronitis and myositis. Pregnant ewes infected with BTV may abort, resorb, or subsequently give birth to lambs with congenital anomalies such as hydranencephaly, cerebellar lesions, spinal cord lesions, retinal dysplasia, and other ocular anomalies or skeletal malformations. The aforementioned clinical signs for sheep summarize a "classical" case, but BTV can be subclinical in sheep as well as in cattle.

In fact, tremendous variation in clinical manifestations and consequences is possible after BTV infection. One reason for this variation worldwide is the multitude of BTV serotypes (at least 24) that have been identified. In addition, there are at least seven serotypes of epizootic hemorrhagic disease virus—a similar *Orbivirus* that primarily affects whitetail deer but can affect cattle causing BTV-like signs—that have been identified. In the United States, 15 BTV serotypes and 3 epizootic hemorrhagic diarrhea virus (EHDV) serotypes have been identified. BTV-positive cattle have not been found in the northeastern United States nor in the upper Midwest states of Wisconsin, Michigan, and Minnesota unless the cows were transported from other regions. Canada remains free of BTV with the exception of southern British Columbia.

Control of the disease is difficult for several reasons:

1. The biologic vector, *Culicoides* spp., is difficult to control, and the virus may overwinter in the larval form of these insects.
2. Infection in cattle, goats, and possibly other wild ruminants can remain subclinical, but infected hosts can act as reservoirs of disease. Sheep are the major species to show clinical signs of disease.
3. Multiple serotypes require specific testing rather than group antigen testing for best detection.

4. Some strains of BTV may cause only subclinical infections in cattle, thereby not arousing clinical suspicion of disease.

Transmission of virus from infected to susceptible animals by *Culicoides* spp. has been studied and results in seroconversion. Transmission of BTV in the United States is predominantly thought to occur via *C. sonorensis* and *C. insignis*, species of midge with a proven role in the epidemiology of the disease; however, there are other species, specifically *C. stellifer* and *C. debilipalpis* that are suspected to play a role in the transmission of both BTV and EHDV. Laboratory or experimental infections in cattle usually do not result in clinical illness, but some reports of field outbreaks describe obvious clinical illness with signs similar to those in sheep with BTV (see also Chapter 16). Therefore, even though most BTV in cattle is thought to be subclinical, certain husbandry or environmental conditions or strains of BTV in field outbreaks appear capable of causing clinical disease. Sunlight may enhance and worsen the clinical signs when sheep are infected with BTV.

Clinical Signs

Most cattle infected with BTV are asymptomatic. When clinical signs are observed in field outbreaks, mucosal and skin lesions predominate. Hyperemia and oral vesicles that ulcerate may involve the mucous membranes of the mouth or tongue. The muzzle may undergo similar vesicular changes that lead to a "burnt" appearance with a dry cracked skin that may slough. Salivation is common, and swelling of lips may occur. Fever is present. Stiffness or lameness is common as a result of both myositis and coronitis. Coronary band hyperemia, ulceration, necrosis, exudates, or sloughing may occur. The skin of the neck and withers may become thickened, exudative, and painful. Therefore, depending on the clinical signs demonstrated, the differential diagnosis for a bovine case of BTV infection might include BVDV, vesicular stomatitis, FMD, malignant catarrhal fever (MCF), IBR, rinderpest, and bovine papular stomatitis. Obviously given the overlap of clinical signs of some BTV infections in cattle with important foreign animal diseases, regulatory officials must be contacted after detection of such lesions.

Reproductive consequences are rare in infected cattle but include fetal death, fetal resorption, abortion, persistent infection of immunotolerant fetuses, and congenital defects such as hydranencephaly, skin disorders, ocular disorders, and skeletal lesions. Infected bulls may become temporarily sterile.

Most cattle with clinical signs recover but may carry the virus for prolonged periods, and others have prolonged lameness or poor condition.

Diagnosis

Clinical signs aid diagnosis and require differentiation from photosensitization, BVDV, MCF, IBR, VSV, EHDV, and FMD. If sheep reside on the premises, clinical signs may be obvious in this species. Regulatory veterinarians should be consulted immediately when BTV is suspected because the differential diagnosis includes exotic diseases.

Absolute diagnosis can be achieved by a variety of diagnostic techniques that include virus isolation or detection of viral antigen, nucleic acid, or antibody. Currently, in many diagnostic laboratories, samples are first screened by conventional or RT-PCR before virus isolation. BTV isolation can be performed on heparinized blood, fetal specimens, spleen, or bone marrow. Samples should not be frozen. In an experimental challenge study BTV could be isolated from the blood of infected cattle for up to 49 days postinoculation; however, infected cattle maintained a level of viremia infective for *Culicoides* spp. for a maximal duration of 3 weeks. Because different strains of BTV may behave differently in cattle, monoclonal FA, virus neutralization, or molecular assays may help further characterize serotypes of virus isolated from blood or tissues. The blood of infected cattle remains positive for viral nucleic acids by RT-PCR for a much longer duration (nearly 4 months) than for virus isolation.

An outer-coat protein, VP2, is responsible for causing virus-neutralizing antibody against BTV in infected animals. Inner-core protein VP7 is a serogroup-specific antigen as are the nonstructural proteins NS1 and NS2. The older complement fixation tests and serum neutralization antibody tests primarily detected group antigens. An immunodiffusion test has also been used, but currently a competitive ELISA is recommended. The ELISA test is quantitative and is serotype-specific through incorporation of monoclonal antibody.

In utero infection resulting in persistent infection of immunotolerant animals can be confirmed only by viral isolation or nucleic acid assays, probably best performed by collecting precolostral blood so that maternal passive antibody in colostrum does not confuse the situation. Precolostral blood for ELISA testing may allow diagnosis of in utero infection and seroconversion of immunocompetent fetuses with or without congenital anomalies. For adult cattle, virus isolation, positive PCR tests, or paired ELISA tests on serum to confirm an increasing titer are required for diagnosis.

Treatment

If clinical cases occur, treatment should be symptomatic. Affected cattle should be kept out of sunlight if possible because sunlight exacerbates skin lesions.

Control

As previously mentioned, control is extremely difficult. Regulatory veterinarians should be consulted. Although a stable positive serum antibody titer only indicates past infection, the economic implications of a positive titer are profound. Export markets, embryo transfer potential, sale of bulls to bull studs, and sale of semen to various markets are negated by positive BTV antibodies in healthy cattle. Geographic

incidence of BTV antibodies varies greatly, with serologic evidence of infection in the Northeast and upper Midwest being rare but relatively common in the southern states, Great Plains, and West.

Epizootic Hemorrhagic Disease

Etiology

Epizootic hemorrhagic disease virus (EHDV), similar to BTV, is an *Orbivirus* from the Reoviridae family that is transmitted between ruminants and maintained in nature via a *Culicoides* vector. Worldwide the virus exists in many tropical and temperate climates and of the seven proposed serotypes of the virus that are identified globally, EHDV-1 and EHDV-2 have been responsible for the majority of cyclical epidemics of the disease in the United States. Over the past decade, a novel EHDV serotype, EHDV-6, which represents a reassortment of endemic EHDV-2 and an exotic EHDV-6 strain, has been repeatedly demonstrated in ruminants in the United States. White tailed deer are the most common hosts for EHDV in North America and the disease can be severe in this species. EHDV has been spreading east and northward in the past few years and has been responsible for high-mortality outbreaks in deer in several midwestern and northeastern states since 2014. *C. sonorensis* is the only confirmed vector of EHDV in the United States but other insect vectors are likely, especially given the fact that the virus has been moving recently into areas of the nation that are not considered part of the natural range of this arthropod species.

Clinical Signs

Clinical disease in cattle has been reported from several midwestern states since 2013, but fortunately, it is a low-morbidity, low-mortality disease in domestic cattle. The primary outcome in cattle is subclinical infection or a mild to moderate transient febrile illness. Occasional cases in the midwestern United States in the past 3 years have demonstrated more obvious systemic illness with inappetence, oral erosions and hypersalivation, diminished milk yield, coronitis, and lameness, in addition to fever. We have seen quite marked dysphagia that has persisted for several days accompanied by fairly severe esophagitis and upper GI erosions. The majority of cattle recover with supportive care (see Chapter 16).

Diagnosis

Exactly the same comments regarding diagnostic testing made in the section on BTV apply for EHDV diagnosis; such are the similarities between the viruses that comparable, but virus specific, serologic, and nucleic acid tests are used by most diagnostic laboratories.

Control

As with BTV, control is very difficult, made even more complicated by white-tailed deer, the natural reservoir of infection in the United States, being so ubiquitous.

Pharyngeal Trauma

Etiology

Natural or iatrogenic pharyngeal trauma commonly results in GI and respiratory consequences in affected cattle. Coarse or fibrous feeds, awns, and metallic foreign bodies occasionally cause pharyngeal punctures or lacerations, but the most common cause of pharyngeal trauma in dairy calves and cattle is iatrogenic injury. Inappropriate, rough, or malicious use of balling guns, paste guns, esophageal feeders, magnet retrievers, Frick speculums, and stomach tubes are the usual causative instruments. Failure of laypeople to lubricate implements, judge appropriate depth of the oral cavity, or hold the animal's head and neck straight when administering oral medication are the most common errors. Purely rough or sadistic treatment also is common.

Acute pharyngeal injury or trauma can have many sequelae. Small punctures may have few acute consequences but eventually result in cellulitis or pharyngeal abscesses. Most acute injuries result in both local and systemic effects. Local effects include pain, reluctance or inability to swallow, salivation, and cellulitis. Systemic effects reflect damage to vagus nerve branches in the pharynx. Such nerve damage can affect rumen motility, eructation, and swallowing, and predisposes to inhalation pneumonia. Bloat resulting from failure of eructation reflects direct or inflammatory injury to vagus nerve branches controlling the complex act of eructation, which requires coordinated activity of larynx, pharynx, and proximal esophageal muscle.

Often cattle that sustain pharyngeal injuries had primary illnesses, the treatment of which included administration of oral medications. Therefore, early clinical signs of pharyngeal injury may be thought to represent failure of response or worsening of the primary condition. Pharyngeal trauma is common in dairy cattle and is underdiagnosed as a cause of illness.

Clinical Signs

The chief complaint in cattle with pharyngeal trauma is anorexia and a suspected abdominal disorder that has not responded to medication (including orally administered medications). Direct tissue trauma is quickly complicated by cellulitis or phlegmon of the retropharyngeal tissue. Clinical signs usually include salivation, a "sore throat" as evidenced by an extended head and neck, fever, fetid breath, soft tissue swelling in the throat latch, and localized or diffuse pharyngeal pain (Fig. 6.29). Most cattle are unable or unwilling to eat. Dysphagia may be present in severe cases and may lead to dehydration because of an inability to drink. Other GI signs caused by varying amounts of direct or indirect damage to vagus nerve branches may occur; these include bloat, rumen stasis, or signs of vagal indigestion. Megaesophagus is an infrequent complication. Respiratory complications include nasal discharge associated with dysphagia, inspiratory stridor, and inhalation pneumonia. Subcutaneous

• **Fig. 6.29** Anxious expression, extended head and neck, salivation, and soft tissue swelling in the throat latch region of a cow with pharyngeal trauma.

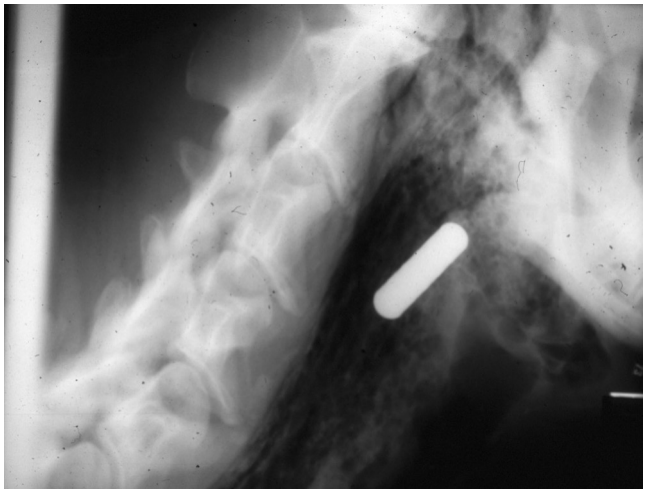
• **Fig. 6.30** Three representative Jersey cows from a herd with an epidemic of pharyngeal trauma associated with mass medication delivered by an owner. Salivation, extended heads and necks because of sore throats, anxious or depressed appearance, dyspnea, inhalation pneumonia, bloat, and subcutaneous emphysema occurred to varying degrees in the affected cattle.

emphysema is present in some patients as air is sucked into the retropharyngeal area and dissects subcutaneously. Although SC emphysema sometimes is limited to the retropharyngeal area, usually air dissects down the neck and reaches the thorax or locations further caudal.

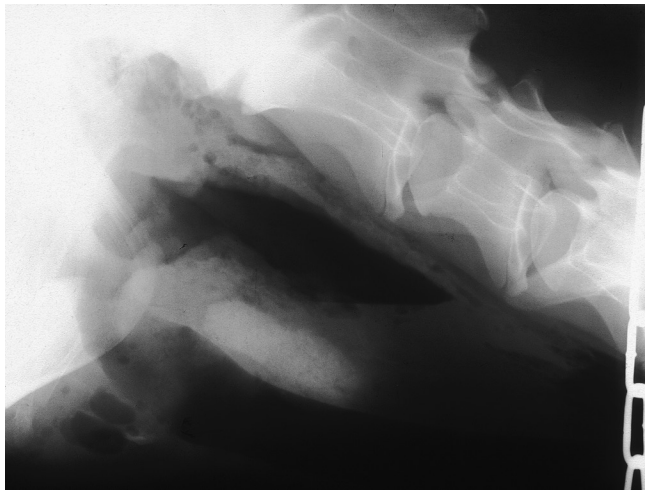
Pharyngeal trauma usually only occurs in one animal in the herd, but herd epidemics have been associated with mass medication (Fig. 6.30).

Diagnosis

Frequently, the clinical signs, coupled with a manual examination of the oral cavity to palpate the pharyngeal laceration, are sufficient for diagnosis. Most injuries are in the caudal pharyngeal region dorsal to the larynx. Severe lacerations also may damage the soft palate or proximal esophagus. Administered boluses or magnets may still be embedded in the retropharyngeal tissues in some cases (Fig. 6.31). An oral speculum and focal light examination also may allow a view of pharyngeal injuries. Manual examination of the oral cavity in affected adult cows can often detect lacerations or abnormal soft tissue swellings.



• **Fig. 6.31** Radiograph of a cow showing a magnet embedded within the retropharyngeal tissues after iatrogenic injury during administration using a balling gun; note evidence of soft tissue swelling and cellulitis in pharyngeal region around the foreign body.



• **Fig. 6.32** Radiograph of the pharyngeal region of a cow that sustained pharyngeal trauma, laceration, and foreign body deposition of a sulfa bolus delivered with a balling gun. A large radiolucent area and tissue emphysema are apparent ventral to the cervical vertebrae, and the bolus can be seen embedded between the air density dorsally and the trachea.

Endoscopy and radiology are very helpful ancillary aids, especially when a manual examination of the oral cavity is inconclusive or when the size of the animal—as with a calf—precludes manual examination. Endoscopy usually allows a view of pharyngeal injuries, but diffuse swelling of the pharynx, larynx, and soft palate sometimes interferes with this procedure. Radiographs are diagnostic of pharyngeal trauma in most cases because air densities and radiolucent retropharyngeal tissues are readily apparent. Pharyngeal foreign bodies and embedded boluses or magnets also are apparent with radiographs (Figs. 6.31 and 6.32).

Treatment

Broad-spectrum antibiotics, analgesics, and supportive measures such as IV fluids are the major components of

therapy for pharyngeal trauma. If an abscess has formed in the pharynx, manual examination and rupture of the abscess into the pharynx is indicated. Whenever possible, it is best to avoid any oral medications until the injury heals. However, sometimes gentle passage of a stomach tube to provide an economical means to hydrate a patient with dysphagia is necessary. Most small pharyngeal lacerations respond to antibiotics such as ceftiofur, tetracycline (9 mg/kg IV every 24 hours), ampicillin (6.6–11.0 mg/kg IM or SC every 12 hours), or other broad-spectrum combinations. Penicillin is not a good initial choice on its own because it seldom is able to control the expected mixed infection. Judicious use of analgesics such as flunixin meglumine (0.5–1.1 mg/kg IV every 24 hours) aids patient comfort, relieves the "sore throat," and may allow an earlier return of appetite. Resolution of dysphagia, when present, is an important positive prognostic sign because the patient can now drink effectively and hydrate herself. Resolution of fever is another positive prognostic sign but may be misleading if the temperature decreased because of concurrent therapy with nonsteroidal antiinflammatory drugs (NSAIDs).

Nursing procedures and ensuring access to fresh clean water and soft feeds such as silage or gruels of soaked alfalfa pellets are helpful. In severe cases, placement of a rumen fistula may be necessary to allow for placement of mashes or liquids directly into the rumen, thereby bypassing the damaged and painful tissues. Antibiotic therapy should be continued 7 to 14 days or longer depending on response to treatment and healing of the pharyngeal wound. Foreign bodies, boluses, or magnets embedded in retropharyngeal locations must be removed.

The prognosis is good for most cases but is guarded for cattle having large lacerations, soft palate lacerations, proximal esophageal lacerations, inhalation pneumonia, or vagal indigestion.

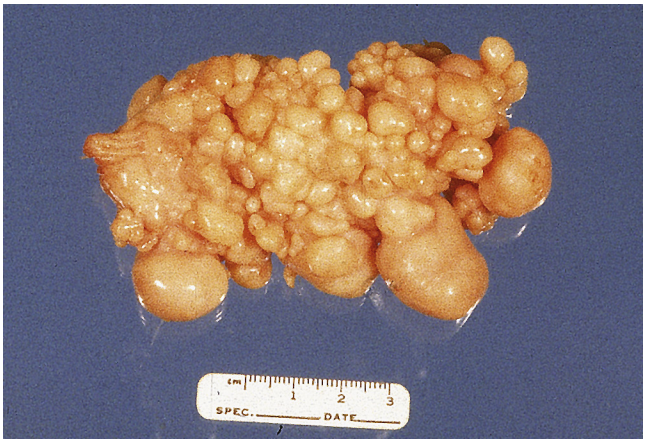
Prevention

Veterinarians should educate laypeople on how to safely administer oral medications. Stomach tubes, specula, esophageal feeder tubes, and balling guns should be inspected after each use to identify any sharp edges or "burrs" that may incite future injury. Proper size specula, tubes, and so on should be based on the animal's size and not "one size fits all."

Alimentary Warts

Etiology

Papillomas and fibropapillomas are observed sporadically in the oral cavity, esophagus, and forestomach of dairy cattle. Oral lesions may occur on the hard palate, soft palate, or tongue. Bovine papilloma viruses (BPV), of which there are currently 14 types recognized, are the suspected cause of these lesions. Because BPV types can be found in normal skin as well as cutaneous papillomas and fibropapillomas, it is uncertain which BPV types actually cause the lesions. Recent studies have identified the DNA of several different BPV types in many cutaneous papilloma and fibropapilloma lesions. Experimentally, cutaneous papillomas can be induced by the inoculation of BPV-1 or BPV-2, but the upper alimentary tract lesions are often histologically classified as squamous papillomas and are thought to be associated with BPV-4 infection. The prevalence of upper GI papillomas can be increased by exposure to bracken fern, which is believed to be immunosuppressive. Bracken fern exposure is not, however, necessary for the development of upper GI squamous papillomas because they occur in the absence of this plant. Squamous fibropapillomas of the upper GI tract are thought due to BPV-2 infection. BPV-2 has also been demonstrated in association with urinary bladder carcinomas in cows with chronic enzootic hematuria. The E5 oncoprotein produced by Deltapapilloma viruses, the group to which BPV-2 belongs, is thought important in the development of these neoplastic bladder lesions.



• **Fig. 6.33** A fibropapilloma that was surgically removed via rumenotomy from a 2-year-old cow with chronic bloat.

Signs

Papillomas and fibropapillomas of the mouth, esophagus, and forestomach create no clinical signs unless they interfere with eructation. Lesions sometimes occur as clusters or along a line in the esophagus, suggesting that trauma from fibrous feed material may facilitate mucosal inoculation of the inciting BPV type. Occasional warts at the cardia or distal esophagus act as a ball valve to interfere with eructation and cause chronic or recurrent bloat, leading to signs of vagal indigestion (Fig. 6.33). Such lesions may be precursors for carcinomas, but this has not been proven except when associated with the ingestion of bracken fern.

Diagnosis

Inspection, endoscopic biopsy, and histopathology are the means of diagnosis. Rumenotomy may be necessary to confirm lesions at the cardia.

Treatment

Lesions are not treated except when discovered during rumenotomy in cattle with failure of eructation. In such cases, removal is curative. In the majority of immunocompetent cattle, upper alimentary papillomas spontaneously resolve within a year of development.

Salmonellosis

Etiology

Much of the discussion regarding salmonellosis has been addressed in the section on calf diarrhea. *Salmonella* spp. cause enterocolitis that varies tremendously in severity in adult cattle. Septicemic salmonellosis may also result in abortion or shedding of the causative organism into milk and colostrum. The organism also may be found in milk secondary to environmental contamination and subsequent mastitis. This latter route appears to be typical of *S.* Dublin mastitis and possibly to lesser degrees for other types.

Salmonella spp. are facultative intracellular organisms that can hide in macrophages, be distributed along with these cells, and occasionally cause bacteremia after invasion of the intestine. Fecal–oral infection is the most common route of infection, but other mucous membranes can be invaded by some serotypes. After ingestion of *Salmonella* organisms, a cow may or may not become clinically ill. Factors that determine pathogenicity include:

1. Virulence of the serotype.
2. Dose of inoculum.
3. Degree of immunity or previous exposure of host to this serotype.
4. Other stressors currently affecting the host.

Given the variability in factors 1 through 4 for most adult dairy cows, it is not perhaps surprising that such a spectrum of clinical signs, prevalence, morbidity, and mortality can be seen. Because *Salmonella* spp. often act as opportunistic pathogens, management, nutritional, and environmental factors that adversely impact the cow's defenses are often at play when the disease becomes problematic on a given operation.

Salmonellosis was primarily a sporadic disease in dairy cattle in the northeastern United States until the 1970s. A single cow within a herd might develop the disease secondary to septic metritis, septic mastitis, BVDV, or other periparturient disorders. Infection seldom spread to other cows. However, in recent decades, larger herds and increased use of free stall housing have changed the clinical epidemiology of salmonellosis, such that herd outbreaks with subsequent endemicity and variable morbidity and mortality are now the rule. Free-stall housing creates a nightmarish setting for diseases such as salmonellosis that are spread by fecal–oral transmission. Stressors include such things as concurrent infection with other bacterial or viral pathogens, transportation, parturition, poor transition cow management, GI stasis or disturbance of the GI flora by recent feed changes, heat or cold, and recent anesthesia or surgery.

S. Dublin is host adapted to cattle, but other types are non–host adapted. A particularly frightening characteristic of *S.* Dublin infection is that infected cows may remain carriers for a long time or even forever. Some shed consistently, others intermittently, and others are "latent" carriers that shed only when stressed. *S.* Dublin also causes mastitis, which tends to be subclinical and persistent. Clinical mastitis caused by *S.* Dublin is thought to originate from environmental contamination of the udder by feces from infected cattle rather than septicemic spread to the udder. Infected calves shed large numbers of organisms, frequently are septicemic, and have respiratory signs coupled with fever that confound the diagnosis and mislead veterinarians unfamiliar with this disease. Other than *S.* Dublin–infected cattle, most cattle infected with non–host adapted serotypes such as *S.* Typhimurium are thought to shed the organism for less than 6 months. However, latent carriers or chronic infection may occur occasionally, and chronic *S.* Typhimurium mastitis has been documented after an enteric epidemic.

Another contributing factor to herd infections is contaminated ration components fed to dairy cattle. Protein source supplements and animal by-product components may be contaminated with *Salmonella*. Improperly ensiled forages that fail to reach a pH <4.5 can also harbor *Salmonella* spp. Birds shedding *Salmonella* can contaminate cut forages or feed bunks to infect adult cattle. This latter pathogenesis has been suspected in several herd outbreaks with type E serotypes, but birds also could transmit other types of *Salmonella* by acting as either biological or mechanical vectors. Farm implements used to handle manure or haul sick or dead animals can be a very efficient means of spreading *Salmonella* if they are used to haul feed, bedding, or apparently healthy animals. The spreading of liquid manure on fields in addition to no-plow planting of crops has caused an increase in forage contamination.

Herd epidemics with an acute onset and high morbidity should be investigated as point source outbreaks of feed or water contamination. Chronic, endemic problems may represent spread of infection by carrier cattle to susceptible or stressed herdmates who then propagate the herd problem by shedding large numbers of organisms in feces during acute disease. It is not unusual to have a herd outbreak in lactating cows without an outbreak in young calves or vice versa.

Salmonella spp. are capable of attachment to, and destruction of, enterocytes. Pathogenic serotypes gain access to the submucosal region of the distal small intestine and colon where their facultative intracellular characteristics guard them against normal defense mechanisms of naive cattle. From this location, the organisms enter lymphatics and may commonly create bacteremia in calves. As with most facultative intracellular bacteria, the host's cell-mediated immune system is essential for effective defense. Diarrhea caused by *Salmonella* spp. is primarily of inflammatory origin with lesser contributions (in some serotypes) by secretory mechanisms. Because mucosal destruction occurs, maldigestion and malabsorption contribute to the diarrhea, and protein loss into the bowel is significant when virulent strains infect cattle. Severe inflammation of the colon is common with resultant fresh blood in the feces or dysentery.

Epidemiology

Domestic and wild animals serve as reservoirs for *Salmonella* spp. Thus, *Salmonella* spp. can be found in the GI tract of a wide range of hosts, often without evidence of clinical disease; humans, cattle, horses, pigs, goats, sheep, poultry, wild birds, dogs, cats, rodents and other mammalian wildlife, reptiles, amphibians, and fish As a result, introduction of *Salmonella* spp. onto a dairy farm can occur through a variety of routes, including purchased cattle, heifers returning to the home farm from off-site raising facilities, wild animals such as birds and rodents, contaminated feed or water, human traffic, and even insects. The presence of *Salmonella* spp. on a dairy farm is therefore not an unexpected finding, and transmission to cattle is primarily via ingestion. The United States Department of Agriculture (USDA) National Animal Health Monitoring System (NAHMS) Dairy 2007 study, based on a single sampling visit to dairy operations in 17 major dairy states, showed that 40% of herds included had at least one cow that was *Salmonella* positive by fecal culture.

In a comprehensive study of more than 800 dairy operations in the northeastern United States, the herd-level incidence rate for laboratory-confirmed salmonellosis was estimated to be 8.6 positive herds per 100 herd-years. However, fewer than 20% of the positive herds accounted for more than 70% of the clinical *Salmonella* cases. Clustering of disease among dairy herds suggests a need for focused efforts to improve bovine health and safeguard public health. The most efficient approach for controlling *Salmonella* at the farm level might be to address prevention and control strategies (e.g., biosecurity and hygiene practices) among the relatively few herds with high frequency of disease, as well as striving to prevent pathogen spread from such herds to those that remain uninfected. In the same study of more than 800 herds, the animal-level incidence rate for salmonellosis among preweaned female calves was estimated to be 8.1 cases per 1000 animal-years compared with 1.8 cases per 1000 animal-years among adult cows. Older heifers rarely developed salmonellosis over the course of the study. Because the mortality rate has been found to be higher among calves than adults with salmonellosis, dairy herd outbreaks that involve calves are an especially important economic concern for herd owners.

Dairy cattle with salmonellosis shed *Salmonella* organisms in the feces while ill and after clinical recovery, and the duration and magnitude of shedding are important determinants of transmission dynamics. The Kaplan-Meier median duration of fecal *Salmonella* shedding among dairy cattle after clinical disease was found to be 50 days, which is well beyond the typical period of clinical signs in cattle with salmonellosis. However, the duration of fecal *Salmonella* shedding exceeded 1 year in some animals. Adult cattle with salmonellosis tend to shed *Salmonella* organisms in their feces longer than calves do, in part because calves often die early in the course of disease. Infected cattle shed copious numbers of *Salmonella* organisms in their feces; the concentration of *Salmonella* within the manure of an infected cow ranges from 10^2 to 10^7 organisms per gram of fresh feces. As adult dairy cattle generate approximately 70 kg of manure per day, this translates into a daily environmental contamination of between 7×10^6 and 7×10^{11} *Salmonella* organisms per cow. This undoubtedly leads to widespread and rapid contamination of the dairy farm environment, and *Salmonella* organisms can survive for prolonged periods in suitable conditions outside the host. Thus, fecal *Salmonella* shedding increases the risk of within-herd transmission and inadvertent spread to other herds. Importantly, in addition to impacting the health and productivity of dairy cattle, these factors lead to an increased risk of zoonotic transmission.

Many *Salmonella* infections in dairy cattle remain subclinical. According to the USDA NAHMS Dairy 2007 study, 14% of cows designated as healthy were *Salmonella* positive based on fecal culture results. As with clinical salmonellosis cases, there is an uneven distribution of subclinical *Salmonella* shedding among dairy herds: about 15% of sampled dairy operations yielded 75% of the positive samples from healthy cows. The prevalence of fecal *Salmonella* shedding among healthy cattle appears to be higher in dairy herds with cases of laboratory-confirmed salmonellosis, as opposed to herds with subclinical *Salmonella* infections only. Multiple studies have shown that the prevalence of fecal *Salmonella* shedding among dairy cattle is highest in summer and fall.

This seasonal association is presumably related to temperature and moisture conditions that prevail in the summer and fall months, but whether these conditions are impacting the bacteria, host species, or both is unclear. The ability of *Salmonella* to thrive in warm, moist environments likely increases the probability of cattle exposure and infection. The effect of heat stress is another potential explanation because such physiologic stress could predispose cattle to intestinal colonization by *Salmonella*. Observations from previous studies also demonstrate variations in the regional prevalence of *Salmonella* burden among dairy cattle in the United States, specifically that there is an increase along a southerly gradient. For example, 33% of more than 700 cows culled from Texas dairy farms were *Salmonella*-positive based on fecal culture results, but only 13% of sampled dairy cows scheduled for culling across the entire the United States were found to be culture-positive for *Salmonella* spp. The underlying mechanisms for this and the seasonal trend are likely similar.

Salmonella spp. have also been isolated from the lymph nodes of dairy cattle, including the superficial cervical and subiliac lymph nodes. Lymph nodes located in fat tissue are not generally removed during carcass processing and thus may be ground with trimmings to produce ground beef. Therefore, superficial cervical and subiliac lymph nodes may be a source of *Salmonella* contamination of ground beef. However, it is unknown whether the presence of *Salmonella* in these lymph nodes has clinical implications for dairy cattle.

Clustering of clinical salmonellosis and subclinical *Salmonella* shedding among dairy farms implies that management practices and other herd-level factors may be associated with increased risk of pathogen introduction, survival, and dissemination. Several studies have reported an association between herd size and fecal *Salmonella* shedding. Herds with at least 400 dairy cows have a higher incidence of clinical salmonellosis than smaller herds. Similarly, herds with more than 500 cows have higher odds of having at least one *Salmonella*-positive cow, relative to smaller herds. Larger herds may have a greater likelihood of purchasing cattle from various outside sources, with the accompanying risk of introducing *Salmonella* via a subclinical shedder that has been stressed by transport. High cattle density may also be a feature of larger herds and could promote *Salmonella* transmission; animal crowding not only enhances contact among cattle but may also emphasize the immunologic and clinical impact of stressful group dynamics. Additionally, larger herds are likely to be characterized by management practices that play a role in increasing the probability of feco-orally transmitted diseases such as *Salmonella*. Unfortunately, herd size is a risk factor that does not easily lend itself to practical intervention because of the management trends and economic constraints that prevail in the modern dairy industry.

Use of free-stall housing may be associated with increased odds of fecal *Salmonella* shedding. Free-stall housing, associated primarily with large herds, presents considerable challenges when combating manure-transmitted pathogens. Freedom of movement in free-stall barns allows cattle to have direct contact with manure from other members of the herd, and it facilitates fecal contamination of common feed and water sources. Disposal of manure in liquid form by irrigation or application of slurry (as opposed to use of a broadcast or solid spreader to discard manure) has also been associated with *Salmonella* shedding. Manure in liquid form may be conducive to *Salmonella* persistence and can be dispersed broadly into the environment, increasing the likelihood of cattle exposure to the organism. Use of sprinklers or misters for heat abatement during the warmer months has also been associated with *Salmonella*-positive herd status. It is possible that this is related to the ability of *Salmonella* to thrive in moist environmental conditions. Alternatively, use of heat abatement practices may simply be a proxy for warmer geographic regions where *Salmonella* prevalence tends to be higher. Other herd-level risk factors for *Salmonella* shedding include lack of an enclosed building for feed storage and cow access to surface water such as ponds and streams, underscoring the importance of biosecurity measures. Finally, feeding anionic salts to cows near the time of parturition was found to be a risk factor for *Salmonella*-positive herd status, although the mechanism underlying this association is unclear. Obviously, some of these herd-level risk factors for *Salmonella* shedding are more amenable to intervention than others.

S. Newport and *S.* Typhimurium are two of the predominant serovars among dairy cattle in the United States. Together, they accounted for 60% (346 of 576) of laboratory-confirmed cases in the Northeast, and both serovars were widespread among dairy farms. Nearly 90% of the *S.* Newport and *S.* Typhimurium isolates were resistant to five or more antimicrobial agents. Similarly, these two serovars accounted for 64% (32 of 50) of salmonellosis cases among hospitalized dairy cattle drawn from the same geographic area of the United States during a similar time frame.

Recent evidence suggests that *Salmonella* Cerro (serotype K) is an emerging pathogen among dairy cattle in the United States. Although rarely detected among clinically ill cattle prior to 2007, *S.* Cerro was isolated from nearly 60% (71 of 120) of the dairy cattle with clinical evidence of salmonellosis in a New York field study. Furthermore, *S.* Cerro is currently the leading serovar among *Salmonella* isolates from clinical bovine samples submitted to veterinary diagnostic laboratories in the northeastern United States. Dairy herd outbreaks of salmonellosis with *S.* Cerro have also recently been reported from the Midwest. To date, antimicrobial resistance has been uncommon among *S.* Cerro isolates from dairy cattle. Isolation of *S.* Newport from dairy cattle with salmonellosis appears to have decreased coincident with the rise in *S.* Cerro. The emergence of *S.* Cerro as a pathogen is particularly noteworthy because previously published reports on this serovar suggested a lack of disease association in cattle.

Similar to serovar Cerro, *S.* Dublin is increasingly being isolated from dairy cattle with salmonellosis in the United States, with a high frequency of multidrug resistance among isolates. *S.* Dublin is a cattle-adapted serovar and thus has unique epidemiologic aspects. Illness occurs primarily in calves younger than 3 months of age, and the predominant clinical manifestation is febrile respiratory illness rather than GI disease. This serovar is particularly challenging because of its tendency to yield chronic, subclinical carriers that continuously or intermittently shed high numbers of organisms into the environment. These carrier animals play a key role in maintaining infection in dairy herds via shedding of *S.* Dublin in feces, milk, and colostrum. Some cattle remain carriers for life. Mature cattle at highest risk for becoming *S.* Dublin carriers include heifers infected between 1 year of age and first calving as well as cows infected near the time of calving. The association between infection around the time of calving and becoming a carrier underscores the potential role of physiologic stress in the pathogenesis of the carrier status. Occurrence of an outbreak of clinical disease has also been reported to be a herd-level risk factor for carrier development. Among dairy herds with a history of previous *S.* Dublin infection, successful control has been associated with avoidance of purchasing cattle from herds identified as *S.* Dublin positive and use of appropriate management strategies in the calving area, such as applying suitable hygienic measures and refraining from placing sick cattle in this area. *S.* Dublin also poses a particular threat to public health, as this serovar has a well-documented predilection for causing invasive disease with relatively high case fatality among human patients.

Trends in antimicrobial resistance among *Salmonella* isolates from clinical dairy cattle samples submitted over an 8-year period in the northeastern United States were recently evaluated by Cummings et al in 2013. The prevalence of resistance to several commonly used veterinary antimicrobial agents (including ampicillin, ceftiofur, florfenicol, neomycin, and various sulfonamides and tetracyclines) decreased significantly over the course of the study period and no increasing trends in antimicrobial resistance were noted. Two antimicrobial agents of particular interest are ciprofloxacin and ceftriaxone because of their importance in treating severe *Salmonella* infections among adults and children, respectively. Resistance to ciprofloxacin among bovine *Salmonella* isolates was not detected during the study period. Nalidixic acid resistance, which is correlated with decreased susceptibility to ciprofloxacin, was also absent among study isolates. In contrast, nearly 40% of bovine *Salmonella* isolates displayed phenotypic resistance to ceftriaxone.

As a leading cause of acute bacterial enteritis in people, *Salmonella* remains a major public health challenge. Disease manifestations may include diarrhea, fever, anorexia, abdominal pain, vomiting, and malaise. Although clinical disease generally resolves within 3 to 7 days, *Salmonella* can also produce invasive infections that may be fatal. Children younger than 5 years of age, elderly adults, and immunocompromised persons are especially susceptible to severe, extraintestinal disease. Foodborne transmission is the most common route and can occur via a variety of sources, including undercooked meat and eggs, raw produce, and unpasteurized dairy products. Bulk-tank milk or milk filter samples from 28% of sampled dairy operations throughout the United States were PCR-positive for *Salmonella* in a study published in 2011, and consumption of unpasteurized milk is reported to be common among dairy farmers and their families. *Salmonella* spp. can also be transmitted by direct contact with the feces of infected dairy cattle, as some veterinarians and farm employees can attest, underscoring the relevance of occupational and environmental exposure for veterinarians, dairy farmers, and those who interact with dairy cattle in public settings.

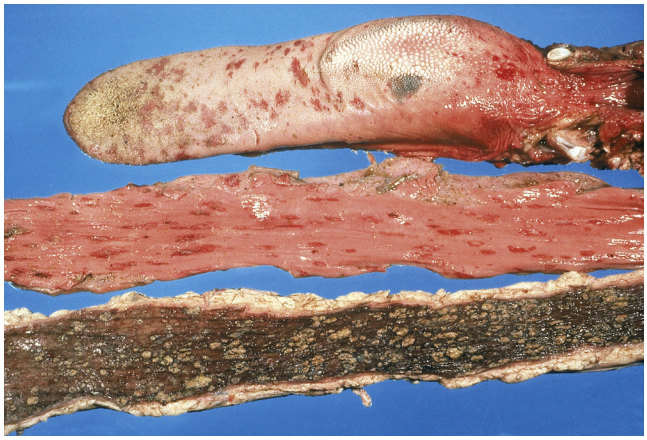
Clinical Signs

As in calves with salmonellosis, adult cattle infected with *Salmonella* spp. may have enteric disease of greatly varying severity. Type E organisms usually cause mild diarrhea, dehydration, fever for 1 to 7 days, and a clinical situation that resembles winter dysentery in that affected cattle appear neither severely dehydrated nor toxic. As a rule, fresh blood is seen less commonly in the feces of type E infections than in types B and C infections. However, the same type E organisms may overwhelm cattle stressed by concurrent infections or metabolic disease due to altered defense mechanisms or those with preexisting acid–base and electrolyte abnormalities.

Fever and diarrhea are consistently expected in salmonellosis, although at the time diarrhea is evident, fever may be absent or have preceded the onset of diarrhea by 24 to 48 hours. This prodromal fever has been confirmed in hospitalized animals that acquired nosocomial salmonellosis. These patients were found to have fever without any signs of illness 24 to 48 hours before developing diarrhea subsequently confirmed as being associated with *Salmonella* types B or C. Fever ranges from 103.0 to 107.0°F (39.4° to 41.7°C) and correlates poorly with other clinical signs as regards severity of illness. However, detection of fever in sick or apparently healthy cows during a herd outbreak is an extremely important aid to diagnosis of an infectious disease rather than a dietary indigestion. Diarrhea is consistent, at least in adult cattle with clinical disease, and may appear as loose manure, watery manure, loose manure with blood clots, or dysentery (Fig. 6.34). Endotoxemia and dehydration accompany diarrhea when virulent strains are encountered or when enteric invasion and bacteremia exist. Anorexia usually accompanies the onset of diarrhea and may be transient in mild cases or prolonged in patients with severe diarrhea and endotoxemia. Feces from cows with type B or C salmonellosis often are foul smelling, containing blood and mucus. Whenever diarrhea with fresh blood and mucus is observed in cattle, salmonellosis should be considered. Recently fresh cows are very susceptible to infection during herd epidemics, and errors in transition cow management often amplify the impact of disease on these cows. Cows with clinical salmonellosis and concurrent abomasal displacements may have a less favorable prognosis after surgical correction of the displacement than surgically repaired cows without salmonellosis. Cows with severe hepatic lipidosis and salmonellosis have an unfavorable prognosis in one author's (TJD) experience. Environmental factors such as heat stress tend to amplify the clinical signs and increase morbidity and mortality. Recording temperatures in apparently healthy cows during a herd outbreak may confirm fevers in some that are about to develop diarrhea or may represent subclinical infections.

• **Fig. 6.34** Fresh blood clots mixed with feces of a cow that had a type C *Salmonella* enterocolitis.

Concurrent infection with *Salmonella* spp. and BVDV after the purchase of herd additions can lead to devastating mortality rates. Dr. Rebhun observed one herd with this combination of acute infections that lost 35 of 130



• **Fig. 6.35** Necropsy specimens from a cow having concurrent bovine viral diarrhea virus (BVDV) and salmonellosis. The tongue *(top)* shows multiple BVDV erosions, the esophagus *(middle)* shows multifocal linear BVDV erosions, and the colon *(bottom)* shows severe inflammatory colitis with mucosal necrosis caused by salmonellosis. (Courtesy of Dr. John M. King.)

adult cattle within 7 days (Fig. 6.35). By comparison, the reported mortality rate in herd outbreaks of *S.* Typhimurium, for example, is approximately 5% to 10%.

Abortions are common, especially when serotypes B, C, or D cause infection and can occur for several reasons:

1. Septicemia with seeding of the fetus and uterus causing fetal infection and death.
2. Endotoxin and other mediator release that cause luteolysis via prostaglandins and apparent alteration in hormonal regulation of pregnancy.
3. High fever or hyperthermia brought about by concurrent fever and heat stress during hot weather.

Cows may abort at any stage of gestation, but as with many causes of abortion, expulsion of 5- to 9-month fetuses is most likely to be observed by dairy personnel.

Salmonella spp. may be found in the milk of infected cattle. With types B, C, and E organisms, this contamination of milk may represent septicemic spread of the organism to the mammary gland, environmental fecal contamination of the milk and milking equipment, or both. Herds infected with *S.* Dublin have chronic mastitis in a percentage of cows infected by this organism. Mastitis caused by *S.* Dublin may be subclinical, and environmental contamination of quarters has been shown to be a more likely cause than septicemic spread to the udder. Occasional cows have chronic mastitis with *Salmonella* serotypes other than *S.* Dublin. Quarters that shed organisms and feces from infected cows create major public health concerns for farm workers and milk consumers. Contaminated milk is a major risk for the entire dairy industry and reason enough to investigate every herd outbreak of diarrhea in dairy cattle with appropriate diagnostic tests. Whereas proper pasteurization reliably eliminates the organism from milk, raw milk should not be consumed.

Diarrhea and illness caused by salmonellosis are common in farm workers and families whenever herd outbreaks occur. It is the veterinarian's obligation to inform clients and workers regarding the public health dangers of salmonellosis and to direct sick farm workers or family members to physicians for treatment.

Ancillary Aids and Diagnosis

Hematology and acid–base electrolyte values are valuable ancillary aids for individual or valuable cattle but are not diagnostic because of the great variation in clinical illness. Fecal cultures are the "gold standard" for diagnosis, and samples from several patients in the early stages of the disease should be submitted to a qualified diagnostic laboratory. Isolates should be typed and antibiotic susceptibility determined. Unlike salmonellosis in horses, *Salmonella* spp. can often be cultured from even a "watery" fecal sample from cattle with salmonellosis. As discussed under the section on calves, serologic confirmation of infection with *S.* Dublin is also available in some European countries and the United States.

Peracute salmonellosis associated with virulent serotypes tends to create a neutropenia with degenerative left shift in the leukogram and metabolic acidosis with Na^+, K^+, and Cl^- values all lowered in affected adult cattle. Elevations in PCV, blood urea nitrogen (BUN), and creatinine can be anticipated in patients with severe diarrhea. Total protein values initially may be elevated because of severe dehydration but are just as likely to be normal or low in time because albumin values decrease quickly as a result of the severe protein-losing enteropathy. BUN and creatinine may be elevated simply because of prerenal azotemia or because of acute nephrosis resulting from septicemia/endotoxemia.

Just as fever precedes the onset of diarrhea in some patients, so may the expected neutropenia with left shift. This has been documented in some cattle that acquire nosocomial hospital infections, although it is unlikely to be detected in field outbreaks because cattle yet unaffected with diarrhea seldom are sampled. Cattle with less than overwhelming acute salmonellosis may have neutropenia, normal WBC numbers, or neutrophilia. Recovering cattle tend to have a neutrophilia.

Sodium, potassium, and chloride tend to be low in most cattle having severe or prolonged diarrhea. As mentioned, peracute severe salmonellosis will result in metabolic acidosis as a result of massive fluid loss and endotoxic shock, but most adult cattle with nonfatal diarrhea do not develop significant acidosis.

The differential diagnosis of salmonellosis in adult cattle is brief if limited to diseases causing fever and diarrhea. BVDV infection and winter dysentery are the primary differentials. Herds with serotypes such as type E or K causing relatively mild signs of fever and diarrhea require differentiation from winter dysentery (depending on the time of year), BVDV infection, and indigestions. Herds with deaths associated with very virulent type B, C or D infections must be differentiated from BVDV infection or toxicities that cause intestinal disease and death. The differential list for cases of more chronic diarrhea includes subacute

ruminal acidosis, internal parasites, Johne's disease, eosinophilic enteritis, lymphosarcoma, chronic peritonitis, and copper deficiency (see Chapters 17 and 18). PCR assays for BVDV should be requested from feces, blood samples, or necropsy specimens to rule out BVDV infection and fecal cultures from multiple patients or necropsy samples evaluated for presence of *Salmonella* spp. Infections caused by *Campylobacter* spp. and *Yersinia* spp. occasionally have been reported in adult cows with fever and diarrhea. The significance and disease incidence associated with these organisms are unknown.

Classical gross necropsy lesions of diffuse or multifocal diphtheritic membranes lining a region of mucosal necrosis in the distal small bowel and colon are present in subacute and chronic cases. In peracute cases, however, minimal gross lesions other than hemorrhage and edema may exist within the involved bowel and enlarged mesenteric lymph nodes. The more acute the death, the less likely gross lesions will be observed. Fibrin casts sometimes are found in the gallbladder and are considered pathognomonic for salmonellosis by some pathologists; bile can be a worthwhile diagnostic sample to submit for culture from necropsy cases if there is a suspicion of salmonellosis.

Treatment

Supportive treatment with IV fluids is necessary for patients that have anorexia, depression, and significant dehydration. Individual patients may be treated aggressively after acid–base and electrolyte assessment. However, outbreaks in field settings seldom allow extensive ancillary workup, and fluid therapy is administered empirically. Use of balanced electrolyte solutions such as lactated Ringer's solution is sufficient for most cattle. Cattle having severe acute diarrhea and >10% dehydration are likely to have metabolic acidosis and may require supplemental bicarbonate therapy. For example, a 600-kg patient judged to be 10% dehydrated and mildly acidotic (base excess = -5.0 mEq/L) should receive 60 L of balanced fluids for correction of dehydration. Rehydration alone may decrease the lactic acid and correct the metabolic acidosis. (The only times that IV bicarbonate therapy is absolutely needed for correction of acidosis in dairy cattle are in the treatment of severe rumen acidosis, enterotoxigenic *E. coli,* or other enteric infections causing excessive production of D-lactate.) If balanced electrolyte fluid therapy does not correct the metabolic acidosis, the cow may however benefit from the administration of bicarbonate. The hypothetical 600 kg cow has 200 kg or L of ECF (0.3 [ECF] X 600), and the base deficit of -5.0 mEq/L implies that each liter of her ECF is in need of 5.0 mEq/L HCO_3^-. Thus 1000 mEq $NaHCO_3$ (0.3 X 600 X 5 mEq) could be added just to make up the existing deficit, and more $NaHCO_3$ would likely be necessary to compensate for anticipated continued losses. This example readily highlights the feeling of helplessness that veterinarians and herd owners experience when a virulent serotype causes serious dehydration in more than a few cows.

Jugular vein catheter placement may allow for repeated administration of IV fluids and repeated IV administration of flunixin meglumine. Hypertonic saline (7.5 times normal) administered at 3 to 5 mL/kg followed by 10 to 20 gallons of oral electrolyte solution, either consumed voluntarily or given by orogastric tube, is a highly practical method of fluid resuscitation in a field setting. This method has become commonplace and is a time- and labor-efficient way of addressing dehydration in grade cattle. Placement of a catheter in an auricular vein may prevent catheter damage from head catches, a common problem with jugular catheters on dairies. Administration of hypertonic saline into smaller diameter veins, such as the auricular vein, may result in phlebitis and catheter failure. When multiple animals merit oral fluid administration during an outbreak of salmonellosis or any other contagious enteric disease or if the same equipment is to be used for drenching of other cattle, laypeople should be aggressively educated as to the possibility of cross-contamination and the need for disinfection between uses. As a crude rule of thumb, cattle that show no voluntary interest in drinking after rapid IV administration of 3 to 5 mL/kg of 7.5 times normal saline solution should provisionally be given at best a guarded prognosis and are mandatory candidates for large-volume oral fluid drenching.

Oral fluids and electrolytes may be somewhat helpful and much cheaper than IV fluids for cattle deemed to be mildly or moderately dehydrated. The effectiveness of oral fluids may be somewhat compromised by malabsorption and maldigestion in patients with salmonellosis but still should be considered useful. Cattle that are willing to drink can have specific electrolytes (NaCl, KCl) added to drinking water to help correct electrolyte deficiencies.

Antibiotic therapy is controversial. Its opponents warn of the potential for emergence of resistant strains that may present great risk for people and animals in the future. Evidence for this phenomenon is sparse except for long-term feed additive antibiotics, and one could argue that antimicrobial use in other species, including humans, represents similar risks. Further opposition states that systemic antibiotics prolong the excretion of *Salmonella* spp. in the feces and may not shorten the clinical course of purely enteric disease. However, discerning animals with infection limited to the gut wall from those animals with gut wall *and* systemic infection is never easy.

Proponents of antibiotic therapy remind us that salmonellosis frequently induces bacteremia (although this is most common in calves), thereby risking septicemic spread of the organism. Clinical differentiation of septicemia versus endotoxemia without septicemia is not easy unless localized infection appears in a joint, eye, the meninges, or lungs. In other words, clinicians can seldom accurately predict which patients with salmonellosis are truly septicemic. In addition, appropriate antibiotic therapy may reduce the total number of organisms shed into the environment by counteracting septicemic spread that allows all bodily secretions,

not just feces, to harbor the organism. These points should be considered by veterinarians and probably dictate against the use of antibiotics in patients with salmonellosis having mild to moderate signs (e.g., low-grade fever, diarrhea, and mild dehydration). Except for valuable cattle that are seriously ill with salmonellosis, systemic antibiotics are seldom administered to adult cows with salmonellosis in the Cornell University or University of Wisconsin Teaching Hospitals.

Therefore, antibiotics are sometimes used when patients appear moderately to severely ill and show signs of fever, dehydration, and have profuse diarrhea or dysentery. These patients usually have elevated heart and respiratory rates, are weak, and appear endotoxemic or septicemic. Given the unpredictable antimicrobial susceptibility patterns for *Salmonella spp.*, antimicrobial therapy should always be guided by culture and susceptibility results. Withdrawal periods should be observed for any nonlabel usage of antibiotics and the Animal Medicinal Drug Use Clarification Act (AMDUCA) guidelines followed at all times. Antibiotics should be continued for 4 to 7 days in patients that are improving.

NSAIDs, especially flunixin meglumine, may be helpful for "antiendotoxic" effects and blockage of various mediators of inflammation and shock. Cattle may be started on 1.1 mg/kg body weight IV every 24 hours and then tapered to 0.5 mg/kg body weight every 24 hours, or the medication may be discontinued after 1 to 2 days. In the United States, this medication must be given intravenously, not intramuscularly or subcutaneously, in dairy cattle. Overdosage or administration of repeated doses of flunixin may cause abomasal or renal pathology and may inhibit intestinal repair. Corticosteroids are contraindicated except as a *one-time* dose of water-soluble corticosteroid for a gravely ill patient in shock. Prednisolone sodium succinate is preferred in this instance.

Isolation of patients with salmonellosis is ideal, albeit difficult, in field settings. Whenever possible, cattle with diarrhea should be confined to an area of the barn away from the rest of the herd. Workers must be educated regarding mechanical transmission of infected feces and other discharges from infected to uninfected cattle. Workers should also be educated regarding the zoonotic implications inherent with salmonellosis.

Prevention and Control

Herd epidemics appear to be increasing in frequency based on confirmed isolations from multiple cow outbreaks identified in New York and the rest of the northeastern United States. Conditions that contribute to an increasing incidence of epidemic salmonellosis include larger herd size; more intensive and crowded husbandry; and the trend for free-stall barns with loose housing, which contributes to fecal contamination of the entire premises. Other major contributing factors include the use of feedstuffs that may be contaminated with *Salmonella* spp. and spreading contaminated manure on unplowed fields. Outbreaks caused by types C and E *Salmonella* spp. have been caused by contaminated feed components, and type E also has been spread by birds that are carriers of the organism.

When salmonellosis has been confirmed in a herd, the following control measures should be considered:

1. Conduct an epidemiologic investigation to help determine the source.
   - Commodities barn or feed storage and handling: Inspect and document source(s) and obtain samples of commodities for culture. Are there other dairies in the area with similar problems? Who hauls the feed onto the farm, and in what? Is this vehicle or trailer used solely for feed transport (not animals, bedding, or manure)? On the farm, how is the feed handled? Is the feed-hauling equipment used for other purposes (e.g., carcass hauling, bedding removal)? Are there other animals or a large population of birds with exposure to the feeds?
   - Water sources: Is there likely fecal contamination? What are the containers used to haul water to pastured cattle, and how/by whom are they transported?
   - Manure handling: Equipment used and destination? What is the flow pattern of flush water? Are the personnel involved in manure handling later handling animals or their feed? Is the manure being spread on unplowed crop fields? Flow patterns of labor, vehicles, water, bedding, and movement of sick and healthy cattle on the dairy should be critiqued. Is there potential cross contamination of feedbunks or feed alleys with contaminated manure by equipment, people or animal movement?
   - Introduction of new animals: Are newly purchased animals quarantined, cultured or serologically investigated for *S.* Dublin infection? How are cattle taken to shows handled on return? Has bulk tank milk been tested for BVDV?
   - Management of cows in the sick pen and maternity pen: Too often, these two sets of cattle are managed and housed together, creating ideal circumstances for infection of fresh cows and heifers! Physical separation and careful allocation of personnel and equipment to each group should be reviewed.
2. Isolate obviously affected animals to one separate group or location if possible.
3. Treat severely affected animals.
4. Institute measures to minimize public health concerns.
   - No raw milk should be consumed.
   - Workers and milkers should wear coveralls, disposable or rubber boots, gloves, and perhaps masks when milking or cleaning barns. Workers and milkers should be encouraged to wash well after work and before eating. Disinfectant footbaths should be placed at exits and entrances to the barn and parlor (for humans and animals), and these footbaths should be maintained diligently.
5. Physically clean the environment, improve hygiene, and disinfect premises (see also the section on calf

salmonellosis). Pressure spraying to physically remove organic matter may seem helpful before disinfection but can be a means of aerosolizing infectious organisms so that both humans and cattle still in that air space at the time of cleaning could be at risk. Because removal of organic debris is incomplete on some surfaces, use of a disinfectant that retains its activity in organic debris and that has documented efficacy against *Salmonella* spp. is optimal. Because shedding is likely to occur from recovered cattle for some time, ongoing efforts at improved hygiene are in order. In particular, protect dry cows and disinfect maternity areas.

6. After resolution of the outbreak or crisis period, a mastitis survey should be conducted that includes bulk tank surveillance. If any *Salmonella* organisms are recovered, culture of the whole herd is indicated to identify carrier cows that should be culled immediately. For *S.* Dublin outbreaks, all cattle should be screened by milk culture, and, if available, serologic testing performed to detect carriers that should be culled, although high prevalence rates in some herds might make immediate culling impractical.

If an epidemic continues despite all of these guidelines, autogenous bacterins may be considered. Although the efficacy and safety of autogenous bacterins are (justifiably) questioned, many practitioners have claimed excellent results when all other measures fail to stop ongoing endemic infections when freshening cows become ill, abortions continue to appear, or calves continue to become ill because of salmonellosis. The siderophore receptor/porin vaccine derived from *S.* Newport (*Salmonella* Newport Bacterial Extract, Zoetis Inc., Kalamazoo, MI) is used by many dairies as a component of *Salmonella* control. The product is typically administered to dry cows as an initial two injection primary series and then boostered annually, although it can be given at any stage of lactation, or to heifers. Although it does not prevent infection, it is often associated with amelioration of disease severity and clinical impact after an outbreak or on endemic farms. The efficacy of J-5 vaccines in salmonellosis control in adult cattle is unknown. Unfortunately, it is difficult to evaluate the efficacy of vaccines used to control endemic salmonellosis in field settings because improvement may be attributed to the vaccine but influenced by herd immunity or alterations in management. (For a further discussion of vaccinations for salmonellosis, see the section on calf salmonellosis.)

Prevention in adults is best accomplished by maintaining a closed herd and maintaining good general health and nutritional management of the late dry period and early lactation cattle. It is inevitable that dairy cattle will be exposed to multiple serotypes of *Salmonella* on many dairies, and the highest risk population for clinical disease are cattle in the late transition period through the first few weeks of lactation. Excellent husbandry, augmented by facilities and practices that allow identification and separation of affected individuals, will position producers to minimize the health and economic impacts of the disease.

Hemorrhagic Bowel Syndrome (Jejunal Hemorrhage Syndrome)

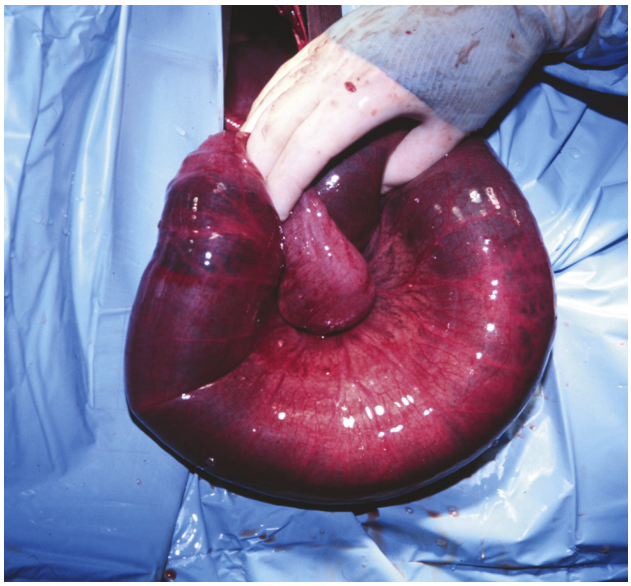
Hemorrhagic bowel syndrome (HBS) is an emerging, often fatal intestinal disease that has been recognized most frequently in adult dairy cows in the United States. Recently, reports of HBS in Canadian dairy and beef cattle have been published, and the condition has also been sporadically identified in dairy cattle from Europe and the Middle East. Other names given to HBS include jejunal hemorrhage syndrome, bloody gut, dead gut, and clostridial enteritis. HBS is characterized by sudden, progressive, and occasionally massive hemorrhage into the jejunum, with subsequent formation of clots within the intestine that create obstruction. Affected areas of the intestine can become necrotic, and affected cows appear to suffer from the combined effects of blood loss, intestinal obstruction, and devitalization of bowel. The disease is seen most commonly in adult dairy cows in the first 4 months of lactation, although cases occasionally occur in late lactation or the dry period. In the United States, the disease is reported to be more common in Brown Swiss cattle.

Etiology

The cause of HBS is currently unknown, and no proven, consistent predisposing factor or factors have been identified. The majority of HBS cases occur during the first 5 months postpartum. In a large survey of American dairy producers, the median parity for cows affected by HBS was reported to be the third lactation, and the median number of days in milk for affected cows was 104 days. During this period, dairy cows experience physiologic stress associated with peak milk yield. In addition, the rations fed during this stage of production are rich in energy and protein and fiber-depleted relative to rations fed later in lactation. These factors have been proposed to place cows at greater risk for HBS, but the events that lead up to the development of this disease remain undetermined. The disease is sporadic, although undoubtedly some "problem" farms experience clusters of cases over relatively short periods followed by quiescent periods of variable duration when no new cases may occur for many months. Identifying potential risk factors that prompt each "outbreak" of new cases can be immensely frustrating for both producers and veterinarians.

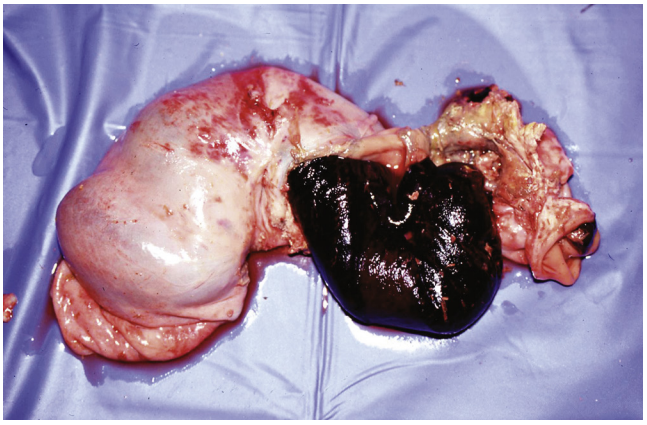
The gross and histologic features of HBS have been described in a few reports. Gross lesions are usually segmental or multifocal in distribution in the small intestine, primarily in the jejunum with occasional involvement of the duodenum or ileum. Affected segments show purple or red discoloration of the intestinal wall, with distension of affected segments caused by intraluminal casts or clots of blood (Figs. 6.36 and 6.37). The intestine orad to these lesions may be distended with fluid and gas, indicating obstruction of affected segments. Fibrin accumulation on the serosal surface of affected intestine may be evident, and affected segments may rupture antemortem or postmortem. The blood clot in affected segments is often tenaciously

• **Fig. 6.36** Fresh field autopsy performed within minutes of death on a mature Holstein cow with hemorrhagic bowel syndrome. Note the purplish discoloration and gas production throughout the small intestine. There was diffuse jejunal involvement.



• **Fig. 6.37** Intraoperative picture of mature Brown Swiss cow with hemorrhagic bowel syndrome. In contrast to the cow in Fig. 6.36, this animal demonstrated the rather more common involvement of just a segment of jejunum with a blood clot obstructing an approximately 12-inch section of bowel. (Courtesy of Dr. Liz Santschi.)

attached to the mucosa, and manual removal of the clot often results in "peeling off" of the surrounding mucosa (Fig. 6.38). On histologic examination of affected bowel, HBS often appears to be a segmental, necrohemorrhagic enteritis, with submucosal edema, mucosal ulceration, transmural hemorrhage, and neutrophil accumulation evident in affected areas. Sloughing of mucosa in affected areas may also be present. A recent retrospective pathologic report demonstrated consistent splitting of the muscularis mucosae or lamina propria from the muscularis layer in the jejunum by hematoma formation without significant inflammation, suggesting that the hemorrhage begins in the lamina propria



• **Fig. 6.38** Resected section of jejunum cut open to show tenacious intraluminal blood clot from an adult Holstein with hemorrhagic bowel syndrome.

secondary to a disturbance in blood or lymphatic flow. As with much of the observational data on this condition, the inability to prospectively reproduce the condition, means that it is hard to draw accurate scientific conclusions about both its the etiology and pathogenesis.

Several reports indicate an association between *C. perfringens* type A and HBS. This association is based on the following observations; (1) affected cows frequently have positive fecal cultures for this organism; (2) *C. perfringens* type A can be readily isolated in heavy growth from blood clots in the jejunum of affected cows; (3) there can be microscopic evidence of intestinal necrosis associated with a dense intraluminal population of large, gram-positive bacteria; and (4) other enteric pathogens associated with hemorrhagic enteritis are rarely identified in tissues or enteric contents of affected cows. In addition, based on anecdotal evidence, reduced monthly incidence of HBS has occurred after administration of autogenous *C. perfringens* vaccines to adult cows on certain dairies. At present, data from controlled studies are not available for evaluation of the effect of such vaccines on the incidence of this disease.

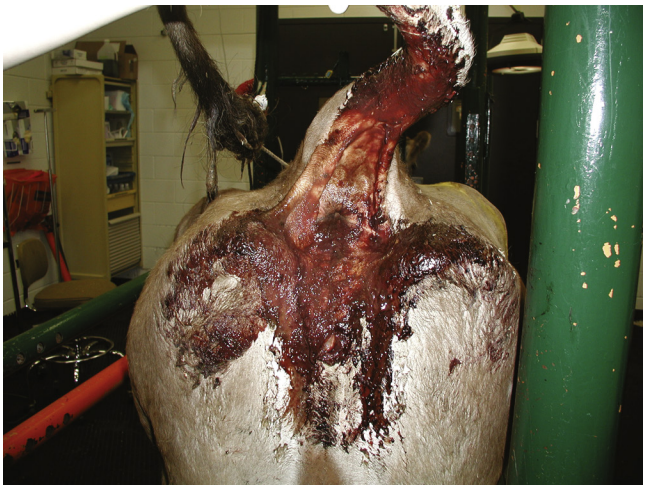
C. perfringens is a large, gram-positive, anaerobic bacillus that is considered to be ubiquitous in the environment and in the GI tracts of most mammals. The rate of isolation of the organism from the GI tracts of cattle may be enhanced by high grain diets. Genetic classification of *C. perfringens* can be performed by real-time PCR. Type A usually produces alpha toxin, although different isolates may produce different quantities of this toxin. Alpha toxin is a calcium-dependent phospholipase that is capable of cleaving phosphatidylcholine in eukaryotic cell membranes. Additionally, the recently discovered beta2 toxin may also be produced by *C. perfringens* type A. Beta2 toxin is also a lethal toxin, and strains of *C. perfringens* with the *cpb2* gene produce variable amounts of beta2 toxin in vitro.

In two studies, *C. perfringens* type A and/or type A + beta2 toxin was isolated from feces and/or intestinal contents of 28 of 32 cows with HBS. These bacteriologic findings are concordant with those of other reports. In the past, veterinary microbiologists have been reluctant to consider

C. perfringens type A as an important disease-causing pathogen of livestock because this organism is part of the normal flora of the cow's intestine. Furthermore, this organism proliferates rapidly in the intestine after death, making isolation from necropsy specimens of questionable diagnostic significance. Because *C. perfringens* type A , with or without beta2 toxin, can be isolated from the gastrointestinal tract of apparently healthy animals, the diagnostic significance of isolation of these organisms from animals with enteric disease is increased if the corresponding toxins can be detected in gastrointestinal contents or blood. In one study, *C. perfringens* types A and A + beta2 toxin were isolated from multiple sites of the intestinal tract of HBS cows at a significantly higher rate than unaffected herdmates (cows with LDA). In addition, intraluminal toxin production was demonstrated in the intestine of HBS cows but not in the intestine of control herdmates with LDA.

It is unclear at present whether enteric proliferation of, and intraluminal toxin production by, *C. perfringens* type A occur as part of the primary insult to the intestine or if these processes occur secondary to another disease or triggering factor. Attempts to reproduce the disease by the experimental inoculation of *C. perfringens* type A into the bowel of early lactation, intensively fed cows have so far failed. Hemorrhage into the intestine from another cause could, in theory, initiate secondary proliferation of the ubiquitous *C. perfringens* because this organism is likely to rapidly multiply when large quantities of soluble protein or carbohydrate is presented to the intestine. In other words, blood certainly could act as a very rich culture medium for this organism. When the organism proliferates, however, the toxins that it releases during rapid growth could contribute to the degradation of the intestinal wall that is characteristic of HBS.

Investigators at Oregon State University at the turn of the century focused on characterizing the potential role of *Aspergillus fumigatus,* a fungus that can be found in livestock feeds. Genetic material of this fungal agent can be detected in the blood and intestine of affected cattle but not in unaffected cattle. Two major hypotheses can be presented regarding the possible participation of *A. fumigatus* in HBS; (1) it acts as a primary contributor to the intestinal lesion or (2) it is involved as an agent that impairs the cow's immune system, thereby facilitating or inciting whatever disease process triggers HBS in the first place. It is also perhaps feasible that DNA of *A. fumigatus* is present both locally and systemically as an opportunist, proliferating and disseminating subsequent to marked intestinal damage associated with some other primary pathology. More recently, Baines et al were able to demonstrate the presence of Shiga toxin producing *E. coli* and mycotoxigenic fungi, including *A. fumigatus,* in the intestinal mucosa and feed respectively, of beef cattle with HBS in Western Canada. There is no classic confirmatory literature fulfilling Koch's postulates to provide definitive proof of a role for any of the theorized etiologic causes at this point in time; rather, observational and retrospective studies provide inconsistent associations between infectious organisms and clinical cases of HBS.

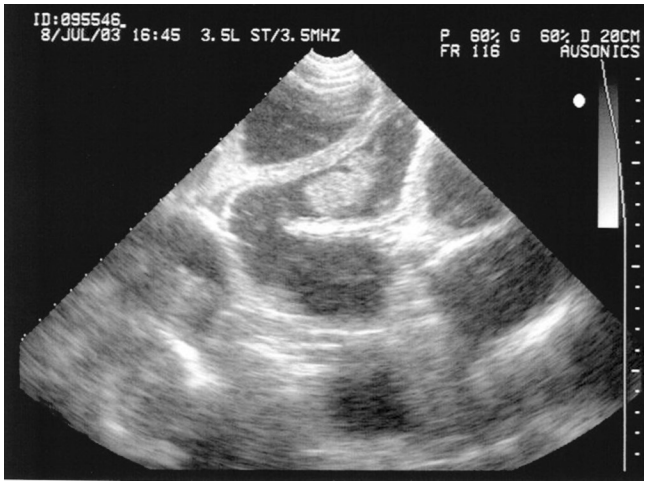


• **Fig. 6.39** Perineum of mature Brown Swiss cow demonstrating the admixture of fresh and digested blood clots typical of hemorrhagic bowel syndrome.

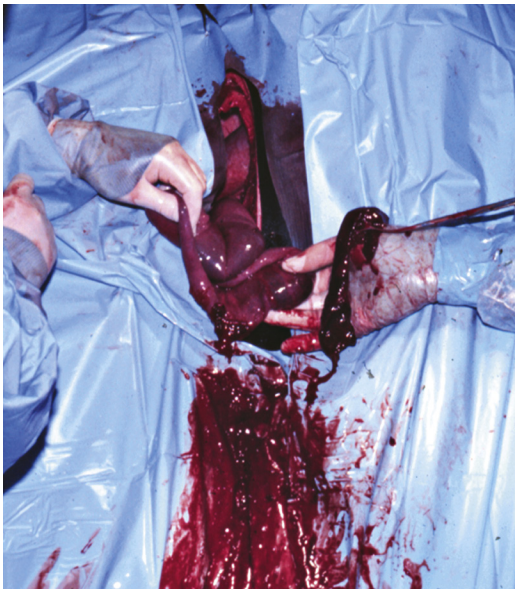
One study worthy of mention attempted to reproduce the condition using a toxigenic isolate of *C. perfringens* type A obtained from a clinical bovine case of HBS in dairy cattle but with no disease noted.

Clinical Signs

Cattle with HBS are typically first noted by owners to be colicky with a rapid pulse and elevated respiratory rate consistent with abdominal pain. The onset is classically peracute, many cattle having been normal, appetent, and high producing at the previous milking. The severity of the colic can be variable from bruxism with kicking at the abdomen through to recumbency and rolling. The cow's extremities are often cool, and the rectal temperature is often below normal. The most significant initial differentials are simple indigestion or obstructive conditions of the small intestine. Retrospective studies have highlighted that this condition tends to be seen during midlactation, affected dairy cows often being between the third and fifth months of lactation. Fecal production and character in affected cattle is very helpful; in the early stages, cattle often have scant to absent manure, but over the following hours, feces become dark and tarlike and may contain dark red to black clots of digested blood (Fig. 6.39). The admixture of fresh, as well as digested blood, distinguishes the condition from bleeding of purely abomasal origin. As clots form in the affected segments of the intestine, the intestine often becomes obstructed, causing some cows to show abdominal distension and subsequently, reduced fecal output. Even in cattle that do continue to pass some feces, bilateral abdominal distension and sucussible fluid in the right ventral quadrant are consistently seen in association with ileus and rumen hypomotility. When viewed from behind, the abdominal contour is typically round or pear shaped in the standing animal. Progressive distension is often appreciated in the lower right abdomen, resulting from accumulation of multiple loops of blood-filled small intestine and ileus in the



• **Fig. 6.40** Transabdominal ultrasound image of lower right quadrant of a cow with hemorrhagic bowel syndrome, demonstrating variably distended loops of small intestine and one small, nonadherent, hyperechoic intraluminal blood clot.



• **Fig. 6.41** Intraoperative image of enterotomy site being used to manually remove and massage obstructing blood clots out of the jejunum in a cow with hemorrhagic bowel syndrome. (Courtesy of Dr. Ryland Edwards.)

ventral abdominal cavity. Scattered, low-pitched "pings" may be evident in the lower right abdomen. In our experience, rectal examination often does not reveal distended loops of intestine because the blood-filled segments of intestine seem to sink to the ventral abdomen, thereby becoming beyond the reach of the examiner. However, small intestinal distension was palpable per rectum in six of eight cows in a Canadian study. Cows with HBS generally have elevated heart rate, normal or subnormal rectal temperature, and little or no interest in feed, but these findings are similar in most diseases that cause small intestinal obstruction. Blood work on affected cattle is neither specific nor prognostically helpful. It is unusual for affected cattle to bleed sufficiently to become anemic, and electrolyte and acid–base changes are usually only mild and typical of other proximal GI conditions in cattle.

Ultrasonography can be used to visualize intestinal distension and clot formation within loops of affected bowel. A 3.5 or 5.0 MHz, sector- or linear-array probe is placed on the abdominal wall at the lower aspect of the right side. Dilated loops of intestine can often be seen stacked on top of one another, and on occasion, material consistent with the appearance of clotted blood can be seen within the distended loops (Fig. 6.40). Some motility is usually retained in these dilated segments when viewed ultrasonographically even if the individual is no longer producing feces.

Differential diagnoses include indigestion, intussusception, intestinal volvulus, enteritis, and abomasal ulcer. Cows with an abomasal ulcer may show melena and shock but do not have the combination of fresher blood with the melena and rarely develop the progressive abdominal distension characteristic of HBS. Indigestion does not progress to feces with melena and blood and does not cause systemic signs associated with shock. Cattle with enteritis continue to pass significant quantities of feces, particularly after treatment with fluids and calcium salts, but cattle with HBS usually do not. Furthermore, when hydration, electrolyte balance, and normocalcemia are restored by fluid therapy, cattle with enteritis typically show resolution of any mild abdominal distension that might have developed as a result of ileus. Differentiation of HBS from intussusception and intestinal volvulus requires exploratory laparotomy.

Treatment

Successful treatment of this disease is difficult. Occasional anecdotal reports exist of successful treatment with fluids, laxatives, antiinflammatory drugs, and antibiotics; however, it appears that such treatment successes with purely medical therapy are quite rare. Cows treated with medical support alone almost inevitably develop ileus, intestinal necrosis with subsequent peritonitis, and shock. Death of affected cattle occurs within several hours to 1 to 2 days after the onset of clinical signs.

The best chances for recovery are offered by a combination of medical treatment and surgery. At surgery, intraoperative findings depend on the extent of clot formation within the intestine and duration of the condition. The serosal surface of affected segments is often dark red, to purple, to black in color, depending on the degree to which the bowel has become compromised. In long-standing cases the affected segments of intestine may be very turgid with luminal blood and highly friable, with the serosal surface already coated with fibrin. The casts of clotted blood within the lumen of the intestine impart a gelatin-like feel to the affected bowel (Fig. 6.41). When there is extensive involvement of multiple segments of jejunum or if extension into the ileum or duodenum has occurred, there will be no opportunity for intestinal resection and anastomosis. Other earlier cases may have fluid

filled jejunum proximal to a short segment of intraluminal clot within bowel that looks much healthier. Techniques for surgical management of HBS cases to date include manipulation of the affected intestine to break down the obstructing clots, enterotomy and removal of the offending clots, and resection and anastomosis of affected segments. At the University of Wisconsin, we routinely attempt manual reduction of the clot(s) without enterotomy at initial surgical exploration and caution owners about the challenges and much poorer outcomes experienced with resection and anastomosis. With manual massage alone, we have a success rate of about 60% to discharge from the hospital, although it should be noted that of these short-term survivors, there is a recurrence rate that approximates 25% over the future life of the cow. Surgical therapy is combined with high-dose penicillin, one to two treatments with flunixin meglumine at 1.1 mg/kg, IV crystalloids, and oral cathartic laxatives. Common reasons for poor surgical outcome include discovery of multiple segments of nonviable bowel, septic peritonitis, and bowel rupture during intestinal manipulation.

Other retrospective studies have not documented such good outcomes, although it is worth noting that these studies examined animals that had been treated by medical therapy alone in some cases or likely those that had been referred after lengthier attempts at treatment on farm than would be typical at our institution. Many Brown Swiss producers, the breed in which this condition has been particularly problematic, have become very adept at recognizing the early signs through hard-won experience and seek veterinary attention promptly.

Prevention

Preventive strategies for HBS remain somewhat speculative at present, given the lack of understanding about the etiopathogenesis of this disease. In addition, controlled studies on the clinical efficacy and economic impact of particular preventive measures have not been completed. Nonetheless, potential risk factors for clostridial overgrowth in the intestine of ruminants have been identified in previous studies, and strategies to reduce those risks might, at least in theory, provide benefits in HBS control. Similarly, the potential role of pathogenic fungi in HBS warrants careful consideration when designing preventive strategies. In short, until more defined information regarding the cause of HBS is published, it may be best to first consider all proposed causes or risk factors (e.g., anaerobic bacteria, fungi, and reduced host disease resistance) and take measures to mitigate these potential risk factors. In so doing, one should consider (1) identifying and correcting management and environmental factors that might impair cow immunity, (2) performing a careful partial budget analysis of the cost of specific preventive measures, and (3) deciding on which specific corrective measure(s) might be most justified for a particular dairy.

To begin with a thorough analysis of transition and fresh cow management should be performed to identify problems with cow comfort, hygiene, nutrition, and disease control that might impact disease resistance during the apparent period of greatest risk for HBS, which is the first 3 to 4 months of lactation. Ration formulation and mixing should be reviewed as well, with due consideration given to such issues as effective fiber and soluble carbohydrate content and their potential dietary influences on gut flora. Feed bunk and pen management should be carefully critiqued to ensure that feed intake is consistent; efforts should focus on identifying and correcting management problems that cause "slug feeding" (e.g., pen overcrowding, poor parlor throughput, and infrequent feeding) and that predispose to subacute rumen acidosis. Silage management, commodity storage, and feed preparation should be examined to determine whether spoilage and mold formation are problematic. Because these critical areas impact numerous facets of cow health other than HBS, identification and correction of problems in these areas will likely provide an overall benefit to cow health. Finally, potential use of feed additives or vaccines directed against specific, potential contributory pathogens should be considered carefully, with the costs of the proposed interventions and their potential efficacy weighed against the prevalence and costs of the disease.

Currently, it is common on farms that have experienced multiple individual cases or occasional sporadic clusters of cases to use either autogenous *C. perfringens* type A vaccines or a commercially available product licensed in the United States (*Clostridium perfringens* type A toxoid, Novartis Animal Health US Inc, Greensboro, NC). There is no reason to believe that commercial toxoids directed against *C. perfringens* types C and D would be helpful in controlling this disease. When autogenous products are used, it is advisable for the manufacturers to use a strain that is both alpha and beta2 toxin producing and to verify that a combination bacterin-toxoid is provided to the client. Anecdotal reports suggest that the incidence of HBS can be reduced on dairies following the introduction of a feed supplement (Omnigen AF, Phibro Animal Health Corporation, Teaneck, NJ) into the ration. This supplement inhibits mold growth and may confer a wider benefit against other mycotoxicoses in dairy cattle to whom it is administered. Other potential supplements that might more generally improve intestinal health such as prebiotics and probiotics may also be considered. There have been no controlled studies to confirm the protective value of any of these immunologic or feed additive approaches to HBS control; very often the enthusiasm for using them is driven by the impact the disease is making currently and the price of milk at the time.

Bovine Viral Diarrhea Virus

Etiology and Background

The disease commonly referred to as bovine virus diarrhea (BVD) was first described by Olafson and Dr. Francis Fox et al in 1946. This initial disease was highly infectious and contagious and imparted high mortality. The causative

organism was later isolated and so began the prolific long-term research into this pathogen of cattle. The initial clinical descriptions of BVD by Fox were of a severe disease characterized by high fever, diarrhea, mucosal lesions, and leukopenia. However, throughout the period from 1950 to 1975, the disease was largely disregarded in parts of the United States—including the northeast—because serologic surveys suggested that most adult cows had serum neutralization titers against BVDV. These results were interpreted to mean that BVDV frequently infected cattle as a subclinical or mild infection and was of little clinical significance. A direct consequence of this thinking was a nearly complete lack of interest in vaccination of dairy cattle against BVDV. The major clinical evidence of BVDV during the years 1950 to 1975 was sporadic subacute or chronic infection in one or more heifers on a farm. These affected animals usually were between 6 and 24 months of age; they developed diarrhea, typical mucosal lesions, fever, and weight loss and survived in poor condition for a variable time before death. Because of the sporadic appearance of such cases, these animals were thought to be immunodeficient and therefore susceptible to BVDV. This theory was tenable for single-case infections but became less believable when four to six heifers on one farm developed similar signs because the likelihood of multiple immunodeficient animals on one farm seemed small.

During that time, the use of modified-live BVDV (ML-BVDV) vaccines occasionally preceded the development of signs of BVD in a group of heifers by 1 to 4 weeks. Although this further discouraged the use of BVDV vaccines, it was explained as an unfortunate circumstance and likely that the heifers had already been incubating field virus. These subacute or chronic cases—usually in heifers—were often called "mucosal disease" because of the easily observable oral erosions and GI lesions found at necropsy, as well as the characteristic clinical signs of fever, weight loss, and diarrhea. Virologic limitations at many diagnostic laboratories during this period added further confusion to the disease clinically referred to as BVD or mucosal disease. Diagnosis was based primarily on serum neutralization titers and FA procedures on tissue samples rather than viral isolation. Current knowledge helps explain why so many of these clinically obvious BVD patients had low or nonexistent serum neutralization (SN) titers against BVDV. Furthermore, the FA techniques used were poor tests that gave erratic results. Therefore, in many cases over this time period, a textbook example of clinical BVD could not be confirmed as BVDV infection.

Reproductive and fetal consequences of infections with the virus were studied during these years (1950–1975), and the implications of BVDV in reproductive failure were suggested clinically but seldom confirmed. The virus was shown to be a potential cause of abortion and congenital anomalies such as cerebellar hypoplasia and ocular defects. Absolute diagnosis of BVDV infection as a cause of clinical reproductive, GI, or other system disease was made difficult by limited laboratory capabilities.

The past 40 years have brought both a wealth of research regarding the virus and the reemergence of BVDV as a major pathogen in cattle. The virus had been classified as a pestivirus within the Togaviridae family because of similarities with hog cholera virus and the virus of Border disease. Recent reclassification finds BVDV as a member of the genus *Pestivirus* within the family Flaviviridae. BVDV is classified in vitro into one of two "biotypes," cytopathic (CP-BVDV) or noncytopathic (NCP-BVDV) based on how each biotype affects cell cultures. Whereas CP-BVDV causes vacuolation and death of certain cell lines within days of inoculation into cell culture, NCP-BVDV inoculation into cell culture results in inapparent infection. NCP-BVDV is the more prevalent biotype in cattle. It serves as the parent virus from which, after genetic recombination, CP-BVDV arises.

In addition, a multitude of "strains" or heterologous isolates exist within each of the BVDV biotypes. The exact number of strains or genetic variation in the virus is not known, but the implications regarding clinical presentations and effective immunization against these multiple strains constitute the major current concerns for BVDV. Furthermore, the strain of virus used to complete a research study may or may not have implications for cattle exposed to a heterologous strain in the "real world." Some strains may be capable of causing congenital anomalies, but others cause severe GI injury. Therefore, the strain chosen for study may have a profound outcome on the study results.

Through genetic sequencing, BVDV can be further classified according to one of two major genotypes (commonly called "types" and sometimes referred to as "species"): 1 and 2. Type 1 strains are considered the classic genotypes banked since the 1950s. Type 2 BVDV was first detected by genetic sequencing of isolates from severe clinical cases in adult cattle and calves in the northeastern United States and eastern Canadian provinces in 1993 to 1994. There are currently 17 recognized subgenotypes of BVDV type 1 (designated 1a–1q) and 3 subgenotypes of type 2 (designated 2a–2c). Viral isolates within a given subgenotype are closely related in nucleotide sequence, sharing > 90% sequence homology. In the United States, three major subtypes have predominated, namely types 1a, 1b, and 2a, although in the past 20 years, the emphasis has shifted from 1a to 1b in terms of prevalence in field cases. On those occasions that type 2 subgenotypes have been identified in the United States, they have been mainly type 2a. In 2014, 3 U.S. isolates of type 2c were identified for the first time. Although severe clinical disease was characteristic of the outbreak of type 2 BVDV in the early 1990s, it should be emphasized that virulent strains of type 1 exist.

Perhaps the most important discovery about BVDV has been the identification and explanation for cattle PI with BVDV. Animals that are BVDV-PI and that have little or no SN antibody against the homologous strain were recognized and later produced experimentally by infecting fetuses between 40 and 120 days of gestation with NCP-BVDV. These researchers were able to cause the PI state by directly

infecting fetuses in seropositive dams (58–125 days) or infecting seronegative dams carrying fetuses (42–114 days) with NCP-BVDV. For unknown reasons, PI cannot be caused by experimental challenge with CP-BVDV.

A brief review of the PI condition is warranted here. Fetuses that are exposed to NCP-BVDV between the approximate ages of 40 and 125 days of gestation may become PI with this strain of virus. These animals are immunotolerant of that NCP strain because immunologically speaking they consider the viral antigens to be self. Such PI fetuses have several potential outcomes; being born normal and growing to adulthood normally; being born apparently normal but succumbing to disease before 1 year of age; or being born weak, small, or dead. However, if a PI animal is challenged by a heterologous CP-BVDV, severe disease may ensue, and in such instances, PI animals usually succumb with signs of acute, subacute, or chronic BVD. Apparently the immunotolerance of the PI animal to its homologous NCP-BVDV renders it unable to mount functional immunologic defenses against certain CP-BVDV strains. This scenario of de novo infection by CP-BVDV in NCP-BVDV-PI animals was assumed by many previous researchers to be the only way animals could get the characteristic "mucosal disease" or fatal clinical BVD. Furthermore, this "superinfection" of PI animals by CP-BVDV strains appeared to explain the outbreaks of BVD that followed use of ML-BVDV vaccines.

More recent studies have shown that animals that develop naturally occurring BVDV-PI often harbor antigenically similar CP and NCP viruses. Genetic studies of these viruses have revealed that insertion of novel RNA into the NCP-BVDV can cause conversion into the CP-BVDV biotype. In other words, a PI animal may develop fulminant CP-BVDV infection from genetic reassortment of its own virus, from transfer of genetic material from a heterologous strain to its own virus, or from exposure to an entirely novel CP or NCP strain. In each of these instances, classic "mucosal disease" may develop in the PI animal.

"Mucosal disease" is often considered as a separate entity from "BVD" by clinicians and researchers. Dr. Rebhun believed strongly that mucosal lesions do not dictate a separate, uniformly fatal entity that is necessarily distinct from BVD and that signs of BVD follow the biologic bell-shaped curve. True, it has been proven that certain CP-BVDV strains can cause superinfection of PI animals, resulting in fatal disease. This fatal disease may follow an acute, subacute, or chronic course and is frequently characterized by fever, diarrhea, weight loss, mucosal ulcerations of the GI tract, digital lesions, or dermatologic lesions. However, clinical experience has shown that naive non BVDV-PI cattle can have mucosal lesions caused by NCP-BVDV infection, yet subsequently survive and form SN titers against this strain. Clinical experience also has shown that fatal BVD has occurred solely as a result of virulent strains of NCP-BVDV and that PI animals are not the only animals that die when exposed to certain CP- or NCP-BVDV. In short, the presence of mucosal lesions is not predictive of death or survival, nor of the PI status. Although the signs of BVD may be more obvious or more profound in superinfected PI than in non-PI animals, the same disease is present.

Similarly, it has been tempting to be "clear-cut" when explaining temporal variation in consequences of fetal exposure to BVDV. Exposure to infected semen may prevent implantation or result in embryonic failure (for reasons that are unclear) until the dam develops immunity against the virus. Infection of the fetus before day 40 may or may not result in fetal death or infertility. Some work suggests embryonic death is likely during this time, but some cattle (or some cattle infected with some strains of virus) can conceive despite acute infection created by oral or IV routes.

Fetuses that are infected with NCP-BVDV before 125 days of gestation are at risk for PI. Fetuses exposed to NCP-BVDV strains between 80 and 150 days may also develop congenital anomalies such as cerebellar hypoplasia, ocular lesions, and many other problems. Because of the overlap between possible PI and congenital lesions, a calf born with a congenital lesion caused by BVD may be either PI or possess a precolostral titer against the BVDV depending upon the gestational age when in utero infection occurred. Experience suggests that the latter (antibody positive, virus negative) is rather more common than the former in calves with congenital lesions. Fetuses exposed to NCP-BVDV after 180 days of gestation are thought to either form antibodies against the virus and survive or be aborted. CP-BVDV strains apparently do not cause PI when pregnant seronegative cows are infected before fetal immunocompetence. Fetal infection by CP-BVDV may cause fetal death, abortion, or the subsequent birth of healthy calves having precolostral antibodies against the infecting CP-BVDV. Congenital lesions may also result from in utero CP-BVDV infections.

The major concern raised by PI animals is constant dissemination of virus because these animals remain a reservoir of BVDV within the herd and shed large amounts of virus in secretions and excretions. Although non-PI herdmates can be vaccinated against BVDV, potential risk to fetuses and young calves remains a concern for herds harboring PI animals. Put simply, PI animals may shed so much virus that the finite immunity in herdmates can be overwhelmed, resulting in infection of non-PI, immunocompetent, and previously exposed or immunized herdmates. Exposure of pregnant herdmates to asymptomatic PI animals is a well-established means of perpetuating endemic BVDV infection in both dairy and beef herds.

Animals being PI explains many heretofore confusing aspects of clinical problems created by BVDV but does not explain the profound variations and patterns of clinical disease caused by BVDV. This variation is more likely explained by multiple strains of NCP-BVDV and CP-BVDV, some of which appear to have a degree of organ specificity. Obviously, previous exposure of cattle to BVDV through natural exposure or vaccination, other diseases that exist concurrent to BVDV exposure, the age and genetics of the cattle, and the strain of BVDV all have a

great influence on the clinical picture created when a group or herd of cattle is exposed. There is no question, however, that within each herd having detectable clinical disease associated with BVDV, the specific clinical signs of disease are repeatable. For example, herds with abortions as a common finding will continue to see abortion, and herds with calves affected with congenital lesions will continue to see such calves without necessarily having cows affected with high fever and diarrhea. Thus, it is unusual to see multiple clinical scenarios within a single herd experiencing disease caused by BVDV. A specific "set" or pattern of signs is more typical, and clinicians never should underestimate the ability of BVDV infection to assume multiple appearances. Future research may allow further distinction of BVDV strains capable of producing specific clinical signs such as thrombocytopenia, specific congenital anomalies, abortions, or GI disease. The disturbing implications of multiple BVDV strains—each possibly possessing individual pathogenicity—center on the consequential need for vaccines that can protect cattle and their fetuses against the heterogenous array of BVD viruses.

The immunosuppressive effects of BVDV infection in cattle are complex and likely contribute significantly to the clinical impact not only of primary BVDV infection but also the clinical outcome for concurrent exposure to other infectious agents, particularly those of the respiratory and GI tract. Immunosuppression that follows BVDV infection occurs because of a combination of lymphocyte depletion and impairment of both innate and acquired immune responses. Highly virulent strains of BVDV are often associated with profound suppression of T helper cell responses alongside increased apoptosis of both B and T cells through downregulation of major histocompatibility complex II and interleukin-2. These effects help explain the enhanced morbidity and mortality seen when herds experience concurrent exposure to other pathogens such as *Mannheimia* and *Salmonella* spp. Immunosuppression may also be an explanation as to why PI animals suddenly develop pneumonia or other infectious diseases when in contact cattle remain normal.

Clinical Signs

A multitude of clinical signs are possible in cattle exposed to BVDV. Frequently, it is emphasized that most naïve cattle or calves experimentally infected with BVDV show little if any evidence of illness yet seroconvert and develop neutralizing antibodies against the infecting strain of BVDV. Such subclinical infection and absence of overt disease also may occur in field situations. However, many other factors such as the age of the animal, concurrent diseases or stresses, relative exposure, dosage, strain and biotype of BVDV, herd and individual cow immune status from previous exposure to BVDV via natural or vaccination means, and presence or absence of PI cattle in the herd must be considered in field situations. As discussed previously, herds experiencing clinical disease because of BVDV will tend to establish a specific pattern of signs rather than variable signs. Clinicians must keep an open mind when considering BVDV as a cause of disease because the signs may be so variable. New signs of BVDV may emerge as more strains evolve. Much of the available experimental data with BVDV has been generated using a limited number of strains. These strains may or may not cause signs similar to wild or field strains. Certain field strains seem capable of causing specific clinical signs. For example, a field strain of NCP-BVDV (genotype 2) found to cause thrombocytopenia clinically was able to create thrombocytopenia in experimentally infected cattle. However, it is obvious that not all strains of BVDV cause thrombocytopenia.

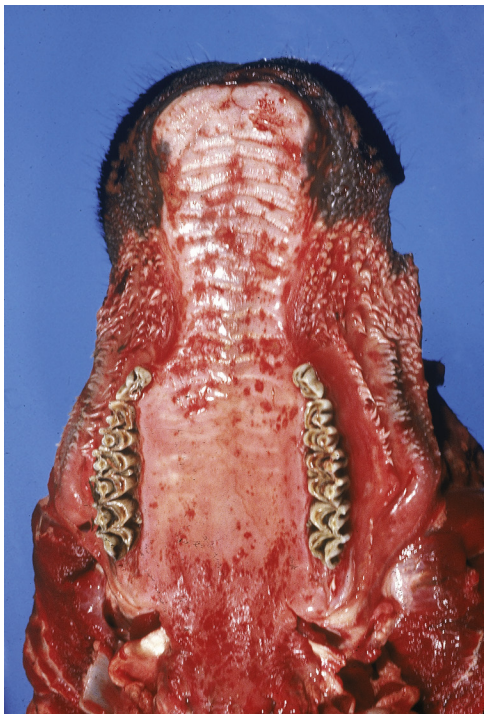
The reported dearth of clinical signs in cattle acutely infected with BVDV is further questioned now that many references to support this theory are quite dated. In addition, the strains responsible for subclinical infections as evidenced by these serologic surveys may not be as prevalent currently as they were 20 to 30 years ago. Clinical signs will be described based on field outbreaks that have been confirmed as BVDV infections.

Acute Illness

Classical signs of fever and diarrhea are possible in naive but immunocompetent calves or adult cattle infected with certain strains of BVDV. Fever and depression usually precede the onset of diarrhea by 2 to 7 days, and fever is frequently biphasic. This biphasic fever starts high (105.0° to 108.0°F [40.6° to 42.2°C]) and diminishes over several days only to recur 5 to 10 days after the onset of the original fever. Diarrhea and GI erosions may be observed during or after the second fever spike, or the patient may recover without showing further signs. Oral erosions will be present in only 30% to 50% of the infected cattle, so absence of oral erosions does not rule out BVDV. Outbreaks of BVDV are most common in 6- to 10-month-old heifers but could occur at any age in naïve populations. A high incidence of clinical disease (mostly high fever) can sometimes be seen in recently fresh cows that are exposed to a new PI animal.

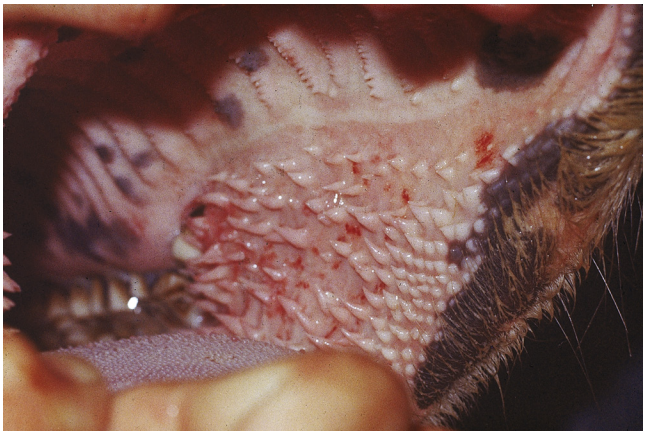
Initial clinical signs in addition to fever include slight to moderate depression and reduced appetite and production. Cattle with a very high initial fever often show tachypnea and may be erroneously diagnosed as having a "viral pneumonia." The tachypnea usually is simply a physiologic response to allow loss of heat caused by fever. If a second fever wave occurs, the clinical signs tend to worsen as appetite and milk production plummet. If GI lesions develop, the cow's appetite is completely suppressed. Few diseases cause the severe degree of anorexia apparent in acute BVDV patients with the severest combination of fever, diarrhea, and GI lesions.

Oral erosions and digital lesions (described later) may be the only "lesions" of BVDV visible to clinicians seeking signs of the disease. Because many, if not most, acutely infected cattle show lesions in neither area, clinicians must maintain an index of suspicion based on other signs (e.g., fever, diarrhea) and examine as many affected animals as possible. In some herds having this form of BVDV, only recently fresh cows develop signs,

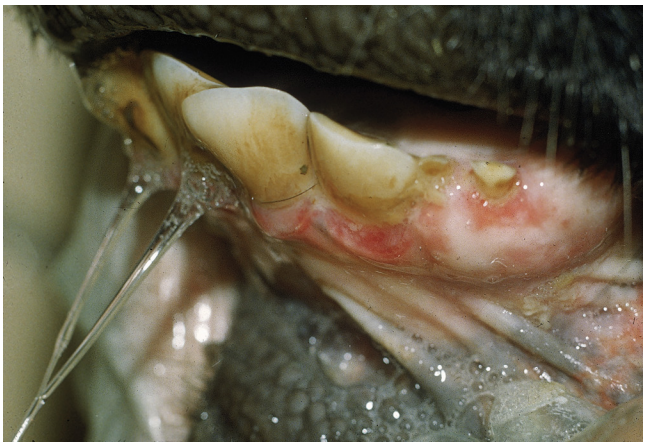


• **Fig. 6.42** Extensive erosions on the soft and hard palate regions of a heifer that died from chronic bovine viral diarrhea virus (BVDV) infection. This heifer was persistently infected with BVDV. Oral erosions in most field cases of BVDV infection involving naïve cattle are not this dramatic or extensive. (Courtesy of Dr. John M. King.)

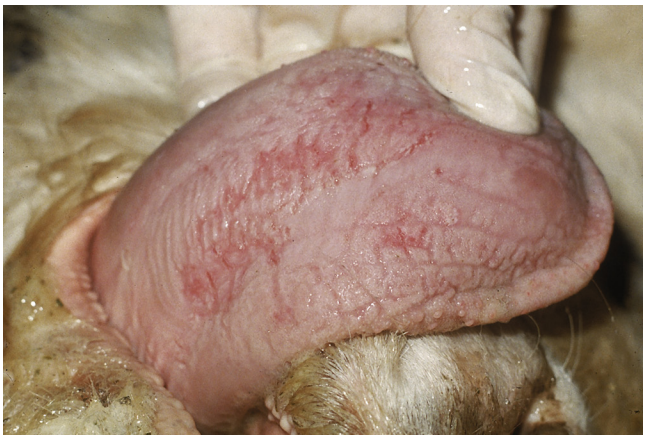
and these affected fresh cows are observed sporadically rather than as an epidemic. Morbidity and mortality levels vary with the classical acute illness but both usually range from 10% to 30%. Occasional catastrophic outbreaks with much higher mortality rates are still encountered in naïve or highly stressed groups of cattle. When present, oral erosions are much less obvious than those observed in pathology texts or in chronic or classic mucosal disease (Fig. 6.42). Focal or multifocal erosions can occur anywhere in the oral cavity and are most common on the hard or soft palates. Hyperemia and erosive changes on the papillae near the lip commissures are sometimes apparent. The papillae may be blunted, shortened, or simply have erosions on the apical portion, causing these areas to appear much more pink or red than the bases (Fig. 6.43). Erosions at the gingival area adjacent to the incisor teeth may occur but sometimes are difficult to interpret because of the natural pink appearance of the gingiva adjacent to the teeth. Close inspection of this area will distinguish sloughing epithelium and erosions from the normal healthy pink mucosa (Fig. 6.44). Both the dorsal and ventral surfaces of the tongue should be examined carefully for ulcers (Fig. 6.45). Slight to moderate salivation may be observed in cattle with oral erosions, and grinding of the teeth may indicate pain caused by other GI lesions. Digital lesions are infrequent in adult cattle experiencing acute BVDV infection, but when present, they appear as coronary band hyperemia, exudation and erosion, or interdigital erosions. Lameness



• **Fig. 6.43** Hyperemia and erosion of the mucosa of the papillae near the lip commissures of an acutely infected naïve cow. The papillae in the middle of the region are eroded, inflamed, and more pink or red than unaffected papillae. Such papillae may or may not appear "blunted."



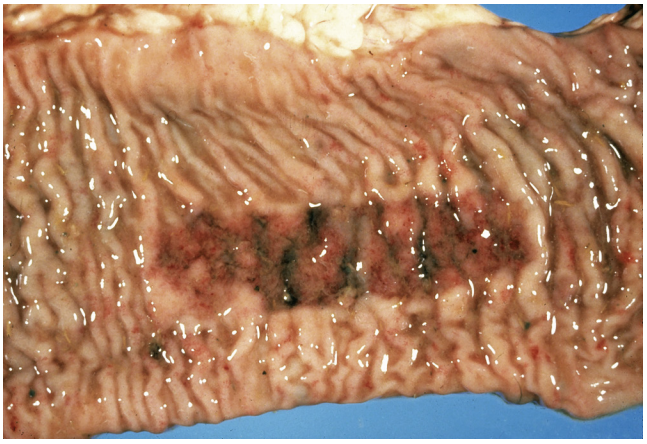
• **Fig. 6.44** Distinct erosions of the mucosa adjacent to the incisor teeth of an acutely infected cow from a herd outbreak of bovine viral diarrhea virus.



• **Fig. 6.45** Erosions on the ventral surface of the tongue in a superinfected bovine viral diarrhea virus persistently infected heifer.

is a distinct sequela to such lesions. The character of the feces in BVDV patients with diarrhea varies from simply loose to watery, and blood or mucus may be apparent in severe cases or in those having thrombocytopenia. Tenesmus may develop secondary to profuse diarrhea and rectal irritation and may be confused with signs of coccidiosis. Leg edema and dermatitis may be noticeable in some PI animals. Ocular discharge may occur in some severely affected cattle.

Immunocompetent seronegative cows exposed to strains of BVDV capable of causing classical acute signs usually seroconvert and survive. However, some seronegative non-PI cows exposed to these viruses become seriously ill and may die. Some NCP-BVDV strains possess sufficient pathogenicity to kill adult, immunocompetent, seronegative cattle. This fact was highlighted by the 1994 epidemic of BVD in Ontario and the northeastern United States. Therefore, a cow or calf does not have to be PI to be killed by a field strain of BVDV. Fatal consequences of BVDV (other than superinfection of PI animals) can occur directly as a result of BVDV-induced thrombocytopenia with subsequent hemorrhage; electrolyte, fluid, and protein losses caused by severe diarrhea; and other causes. Most commonly, however, fatal consequences of BVDV are secondary to opportunistic pathogens creating concurrent infection during BVDV viremia. Even immunocompetent healthy cattle suffer profound alterations in innate and acquired immune defense mechanisms during the time between the onset of BVDV infection and humoral antibody production or recovery. Most healthy cattle exposed to BVDV infection survive this time uneventfully, but less fortunate ones may develop pneumonia, mastitis, metritis, or other bacterial infections while viremic. Temporarily altered cellular immunity affects lymphocytes, neutrophils and macrophages and may predispose to bacteremia or interfere with clearance of circulating microbes. The clinical consequence of this temporary lapse in cellular defenses is an inability of such patients to overcome routine infections. Cattle infected with BVDV experimentally may or may not be exposed to other routine infections, but cattle naturally infected with BVDV are subject to multiple stresses and infections. During the period of viremia and altered cellular defense, dairy calves and cows may succumb to IBR virus, other enteric pathogens (especially *Salmonella* spp.), bacterial mastitis, bacterial pneumonia, and other infections. High mortality rates have been observed when BVDV and *Salmonella* spp. concurrently infect groups of calves or cows. Recently assembled herds or purchased groups of replacement heifers may trigger severe disease by introducing a new strain of BVDV to a resident herd. Immune responsiveness returns to normal as BVDV infection wanes and serum neutralization titers against the virus increase. Therefore, seronegative immunocompetent cattle infected with BVDV do not have any residual or permanent immunodeficiency after resolution of the infection and seroconversion. Both increased severity of concurrent disease and lack of responsiveness to conventional therapy for that disease may be seen during the window of time that a patient is viremic with BVDV. Concurrent infections such as IBR or pneumonia caused by *Mannheimia haemolytica* in animals viremic with BVDV may be so severe as to mask the underlying BVDV because signs of illness or postmortem lesions incriminate respiratory pathogens as the cause of illness. Failure of these more obvious infections to respond to conventional therapy should raise the index of suspicion regarding BVDV infection. For example, a severe outbreak of *M. haemolytica* pneumonia masked underlying BVDV in a herd that had recently added 20 replacement heifers. Cultures obtained from affected cattle via tracheal washes and necropsy, confirmed *M. haemolytica* sensitive to several antibiotics. The indicated antibiotics had been used to treat affected animals, but the expected clinical response was not obtained. Mucosal lesions subsequently were found in a few of the fatal cases, and an NCP-BVDV was isolated from the buffy coat of several affected animals.



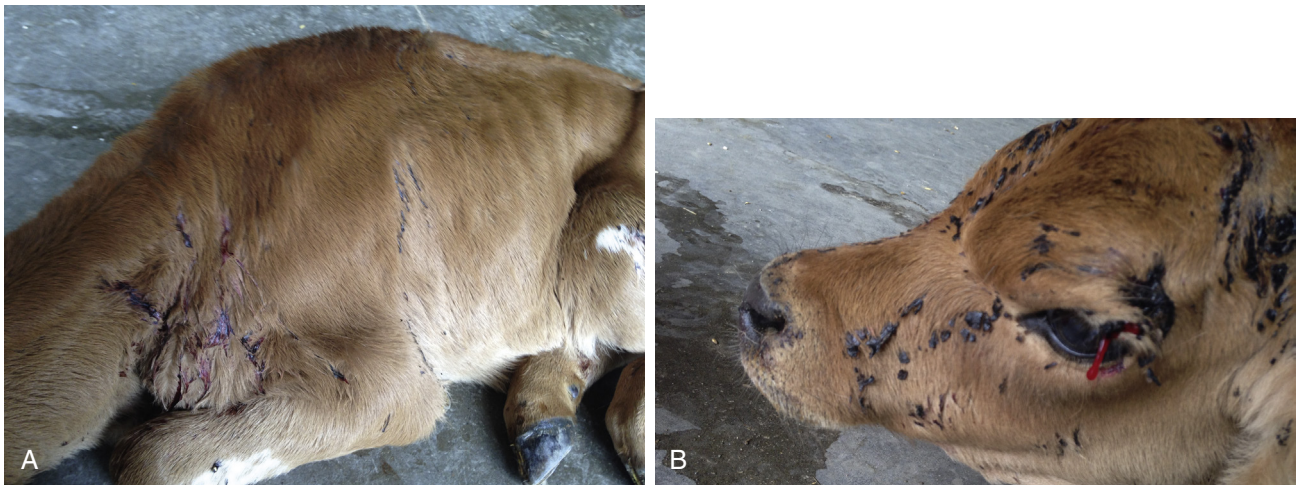
• **Fig. 6.46** Necrosis of Peyer's patch in necropsy specimen of fatal bovine viral diarrhea virus infection. (Courtesy of Dr. John M. King.)

In addition to altered cellular immune responsiveness, acute BVDV usually causes a leukopenia characterized by lymphopenia and sometimes neutropenia. Therefore, not only are WBC functions diminished but their absolute numbers are as well. Leukopenia increases the risk of opportunistic bacterial infection, and neutropenia seems to be associated with increased severity of concurrent diseases.

BVDV also attacks lymphoid tissues such as the spleen, lymph node germinal centers, and Peyer patches and can infect lymphocytes and macrophages (Fig. 6.46).

Combining all the aforementioned negative effects on host immunity helps explain why some non-PI cattle die during acute BVDV infection. Some would argue that these cattle in fact die from *Mannheimia* spp., *Salmonella* spp., or whatever secondary infection overwhelms the animal during the period of transient altered immunity caused by acute BVDV infection rather than from BVDV itself. The net effect, however, is death, and some BVDV strains can kill or contribute to the death of seronegative, immunocompetent adult cattle.

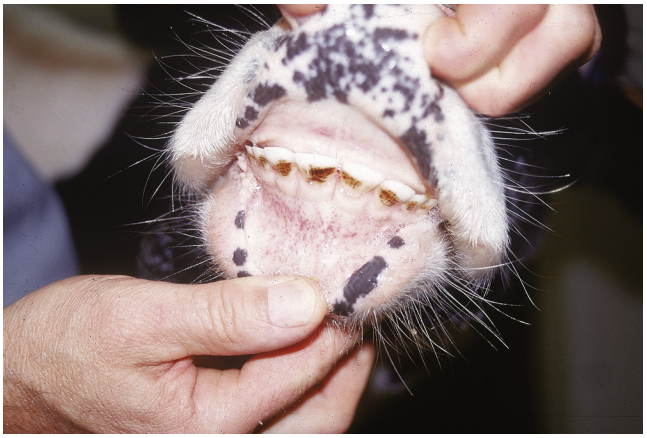
Thrombocytopenia associated with type 2 acute BVDV infection has been observed in adult dairy cattle,



• **Fig. 6.47** **A** and **B,** Two Jersey calves from the same farm affected by type 2 bovine viral diarrhea virus infection causing thrombocytopenia and hemorrhage from trivial trauma such as insect bites during the summer.

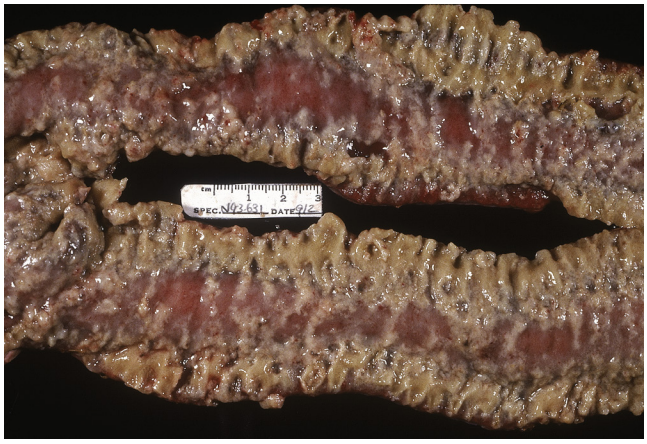
dairy calves, and veal calves. Although platelet counts <100,000/μL are considered abnormal, clinical evidence of bleeding seldom is observed unless the platelet count is <50,000/μL. Conditions such as stress, injections, trauma, or insect bites that may contribute to clinical signs of bleeding in thrombocytopenic clinical patients may not be present in experimental models. Thrombocytopenia associated with bleeding causes blood loss anemia, and is commonly fatal unless treated with fresh whole blood transfusions. Thrombocytopenia occurs as a result of viral infection and destruction of megakaryocytes in bone marrow. Dysfunction of circulating platelets may contribute to clinical signs of impaired coagulation. Field outbreaks of acute BVDV with thrombocytopenia are characterized by one or more of the affected cattle having signs of epistaxis, bloody diarrhea, bleeding from injection or insect bite sites, ecchymoses and petechial hemorrhages on mucous membranes, or hematoma formation (Figs. 6.47 and 6.48). Not all infected cattle show signs of bleeding, and the magnitude of thrombocytopenia varies greatly. In addition, inapparent infection with subsequent seroconversion may occur in some herdmates. However, when bleeding is associated with other clinical signs such as diarrhea, fever unresponsive to antibiotics, GI ulceration, and leukopenia, then BVDV should be strongly suspected. Platelet counts and identification of BVDV by RT- PCR, virus isolation from mononuclear cells in whole blood, or antigen detection by ELISA confirm the diagnosis. Other causes of bleeding can be ruled out by coagulation panels, including assessment of fibrin degradation products.

Acute BVDV infection of naïve, non-PI calves may cause inapparent infection with seroconversion or clinical signs that include fever and diarrhea of varying severity. The greatest risk for calves with acute BVDV infection is concurrent infection with other enteric or respiratory pathogens. Transient reduction of cellular immune



• **Fig. 6.48** Petechiation and severe intestinal bleeding (packed cell volume, 10%) in an 8-month-old heifer having thrombocytopenia associated with acute bovine viral diarrhea virus infection. After a blood transfusion, the heifer recovered.

function and defense mechanisms during BVDV viremia predispose to, and worsen, concurrent infection. Therefore, diarrheic neonatal calves (< 2 to 3 weeks of age) can have acute BVDV infection masked by identification of encapsulated *E. coli*, *Salmonella* spp., rotavirus, coronavirus, or *C. parvum* (Fig. 6.49). Similarly, calves up to several months of age may have overt respiratory disease caused by *Mannheimia* spp., *H. somni*, or respiratory viruses that are isolated from tracheal wash or necropsy specimens. In all of these situations, concurrent BVDV should be suspected when the severity of disease, morbidity, and mortality seem excessive for the identified pathogens. Naïve, non-PI calves born to seropositive cows should acquire passive antibody protection against homologous strains for at least 3 months and in some cases as much as 12 months. However, this passive protection may or may not protect against heterologous strains and may not be protective if calves receive less than adequate amounts of colostrum. In addition, overwhelming exposure to BVDV may override



• **Fig. 6.49** Concurrent *Salmonella* Typhimurium and bovine viral diarrhea virus–induced intestinal lesions in a neonatal calf. (Courtesy of Dr. John M. King.)

any passive protection in some instances. Seronegative calves are at risk at all times. Whenever severe calf mortality associated with enteric or respiratory pathogens occurs, BVDV should be considered and ruled in or out by viral isolation or PCR from blood, necropsy tissue samples, or tracheal wash samples.

Persistent Infection

During the first 18 days of pregnancy, while the bovine embryo is still unattached, if a dam develops viremia because of BVDV infection, no infection of the embryo will typically occur because the zona pellucida prevents viral penetration. Between 30 and approximately 45 days of gestation, it is more likely that embryonic infection will lead to embryonic death; however, between about day 30 and day 125, PI can arise because of fetal infection, but only if the infecting strain is a NCP-BVDV. Such calves are typically born seronegative and PI if the dam is PI. The ability of NCP-BVDV to inhibit the induction of a normal interferon type 1 response by the fetus to viral infection is what gives rise to the PI status. Calves born to a PI dam are always PI themselves, a phenomenon that may be explained by recent research that has identified that BVDV can localize to the oocytes of PI females. Alternatively, a PI calf can be born to a non-PI, immunocompetent dam—the sole requirement is NCP-BVDV infection that creates viremia in the dam of sufficient magnitude to cause transplacental infection at the appropriate time of gestation. PI calves may be transiently seropositive if the dam (PI or not) was infected during pregnancy and passed antibodies to the calf through colostrum; PI dams may generate colostral antibody titers to heterologous strains of BVDV. Surprisingly, many immunocompetent dams that are carrying PI calves actually have very high antibody titers by mid- to late pregnancy because of continual antigenic challenge.

PI calves may appear normal at birth, grow normally, and become productive members of the herd. This situation is perhaps the most frightening because such PI cattle are not easily detected and continue to harbor and shed homologous BVDV through body secretions. Recent data suggest that up to approximately one quarter of PI animals will survive to adulthood (2 years or older), and if one uses an estimate of 1% PI animals in an untested cattle population, it is easy to see how destructive and threatening the PI status can be in both the dairy and beef industries. Apparently healthy PI cattle also reliably reproduce PI offspring that subsequently act as reservoirs of infection for herdmates. PI calves or cattle that are clinically normal may develop signs of acute or chronic ("mucosal disease") BVD if exposed to heterologous strains of CP-BVDV through natural exposure, administration of ML-BVDV vaccines, or genetic recombination of their homologous BVDV strain. In fact, the conversion to CP from NCP biotype probably most commonly occurs by insertions, deletions, and single nucleotide changes of the PI animal's own original strain.

At one time, it was assumed that all heterologous strains of BVDV would cause fatal infections in PI cattle because such cattle would not recognize these strains as foreign. It also was assumed that CP-BVDV strains were necessary to cause disease in PI animals because many workers found both CP- and NCP-BVDV in cattle having chronic or mucosal disease. Not all heterologous strains of BVDV cause PI animals to develop illness, however. Experimental inoculation of PI cattle with certain CP-BVDV strains not only may fail to produce disease but also may be associated with seroconversion against the heterologous CP-BVDV and continued failure of seroconversion against the homologous NCP-BVDV. This situation and that of the PI calf that attains passive-colostrum origin antibodies from its dam constitute two reasons why a PI animal could have serum-neutralizing antibodies against BVDV.

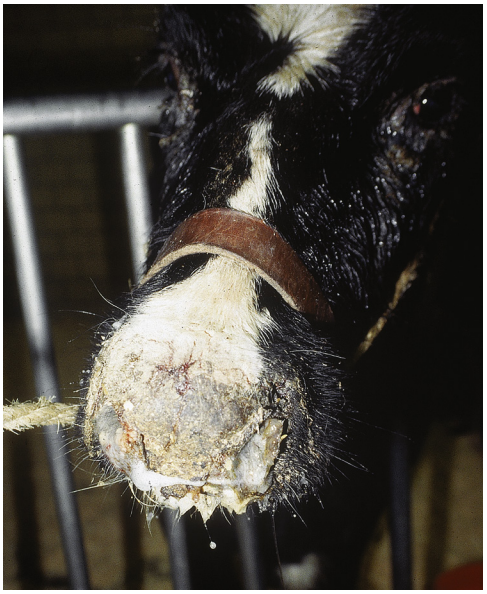
Apparently healthy PI animals often remain in the herd, produce PI offspring, and represent significant sources of perpetuating infection for herdmates and fetuses. Some PI calves are born weak, or small, or die shortly after birth. Weak calves that survive generally succumb to conventional enteric or respiratory pathogens within the first few weeks of life. Clinical signs and gross necropsy findings may not suggest BVDV infection, and death is attributed to enteritis or pneumonia of varying causes. This clinical scenario allows BVDV to escape detection unless blood or tissue samples are submitted for PCR, viral isolation, or antigen detection. Some workers have observed domed skulls and finer-than-normal maxillary shape ("deer noses") in PI calves that are weak, small at birth, and often do not thrive (Fig. 6.50).

The intermediate clinical presentation for PI calves falls somewhere between the apparently healthy PI calf that remains healthy, and the calf that is obviously weak, small, or nonviable at birth. This intermediate type is apparently normal at birth but usually dies before 2 years of age. The cause of death in such PI animals is variable. Recurrent or chronic infections are the hallmark of these calves. Enteritis, pneumonia, ringworm, pinkeye, ectoparasites, or endoparasites may affect such calves, and they may persist or respond

• **Fig. 6.50** A 6-week-old bovine viral diarrhea virus persistently infected calf with poor growth and an abnormally developed skull.

• **Fig. 6.51** A bovine viral diarrhea virus persistently infected yearling heifer that is stunted and has not grown well. The heifer is stanchioned between two healthy herdmates of the same age.

poorly to therapy. Unexplained pneumonia or diarrhea in a single growing heifer on a farm should arouse a suspicion of PI in that animal. Poor growth and stunting compared with herdmates is obvious in these PI animals (Fig. 6.51). Because chronic bacterial, parasitic, or fungal infections typify many of the PI calves in this category, the integrity of immune responses must be questioned. Although PI animals initially were thought to have complete immunocompetence except for the "self" BVDV that they harbor, complete immunocompetence seems unlikely in all cases. There may be a variable expression of cellular or secretory immunity, and other factors, such as the exact time of in utero infection and the strain of NCP-BVDV, may play roles in relative immunocompetence. At least some PI animals appear to have reduced lymphocyte and neutrophil function.

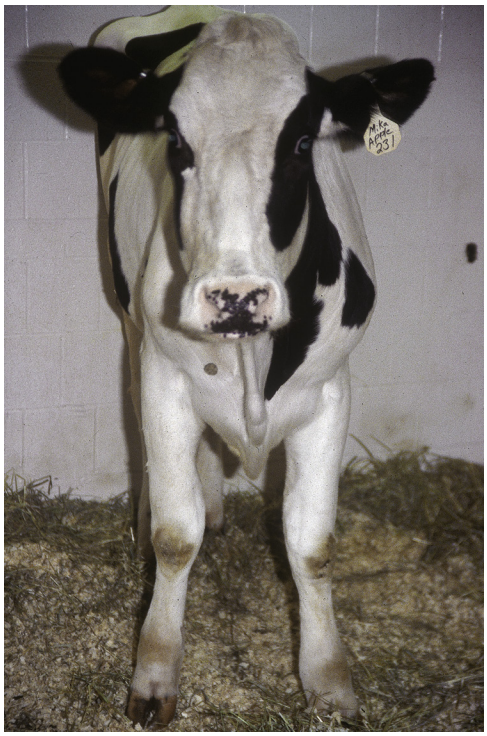


• **Fig. 6.52** "Classic" mucosal disease in a 6-month-old heifer. After contact with "outside" cattle, an entire group of replacement heifers developed fever, diarrhea, and dermatitis, but this heifer that was a persistently infected animal was the only one that died.

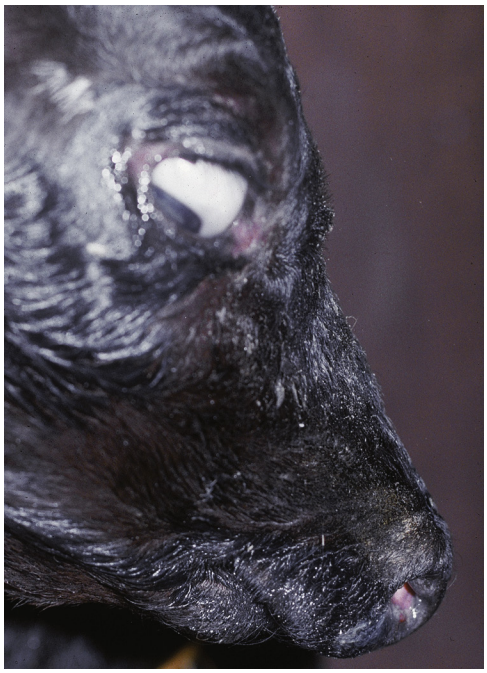
In addition to apparently heightened susceptibility to a variety of opportunistic pathogens, PI animals in this category can succumb to superinfection with CP-BVDV (Fig. 6.52), as discussed previously for classical mucosal disease. In fact, PI animals in this category (e.g., chronic disease, poor-doers, less than 2 years of age) compose the majority of "classic BVD," "chronic BVD," or "mucosal disease" cases. Signs of BVD tend to be profound with diarrhea, poor condition, dehydration, mucosal lesions, and sometimes leg edema (Fig. 6.53), and skin and digital lesions. The course of disease is highly variable—some cases die rapidly, but others linger on as poor-doers. The major differential diagnoses for chronic poor-doer BVDV-PI animals are bovine leukocyte adhesive deficiency (BLAD) and chronic internal abscessation because all of these conditions yield similar gross clinical appearances.

Congenital Lesions

Some BVDV herd infections may only become apparent after the birth of calves with congenital lesions. The teratogenic effects of BVDV are typically manifest after fetal exposure between 80 and 150 days of pregnancy. Again, the individual pattern of disease or set of signs within a specific herd may be unique to that herd. Pregnant adult cattle may experience subclinical infection that results in abortion or the subsequent birth of calves with congenital anomalies such as cerebellar hypoplasia, cataracts, retinal and optic nerve degeneration, hydranencephaly, hypomyelinogenesis, brachygnathism, varying degrees of hairlessness, and other congenital lesions (Figs. 6.54 to 6.56). It must be mentioned here that BVDV is not responsible for all congenital cataracts; there are many other causes. Although a plethora of types of congenital lesions are

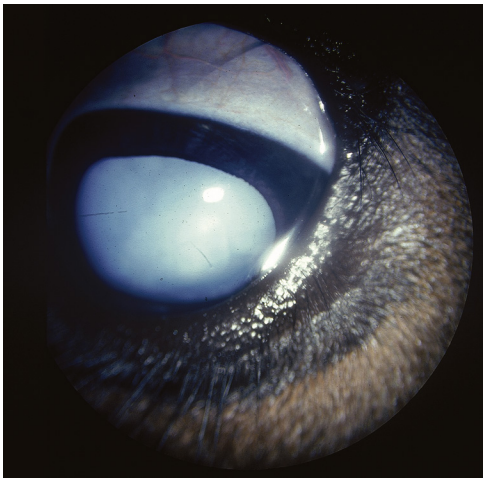


• **Fig. 6.53** A 12-month-old heifer with recurrent fever and edema of all four legs caused by vasculitis and persistent infection with bovine viral diarrhea virus.

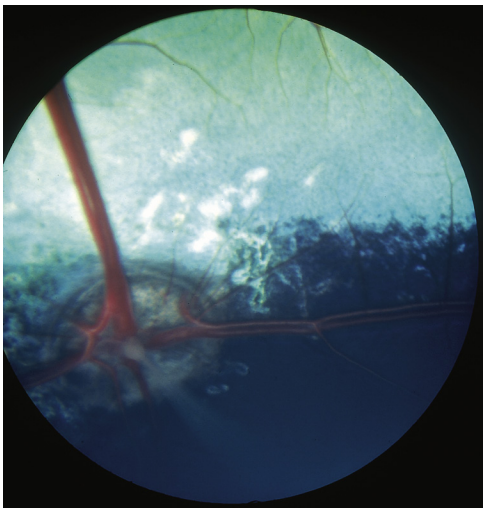


• **Fig. 6.54** Brachygnathism in a calf associated with in utero bovine viral diarrhea virus infection.

possible, only one or two may appear in a single herd and will be repeated in affected calves born over a period of weeks to months. Usually several consecutive calves are affected with the same type of congenital lesion. For example, in a herd that Dr. Rebhun investigated, brachygnathism and cataracts typified the congenital lesions, but

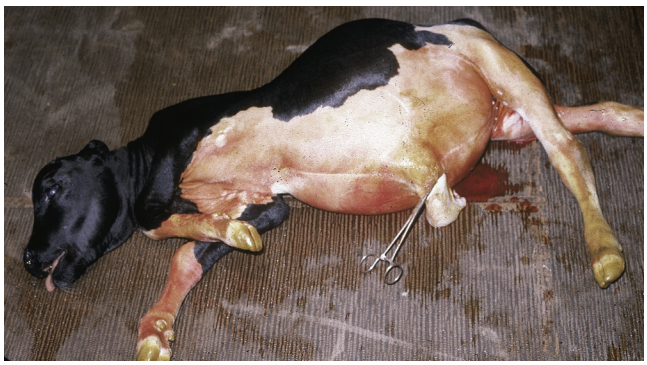


• **Fig. 6.55** Diffuse cataract (bilateral) in a calf that was infected by bovine viral diarrhea virus during the midtrimester of gestation.



• **Fig. 6.56** Optic nerve degeneration and chorioretinal scarring apparent as hyperreflective zones dorsal to the optic disc in a calf infected by bovine viral diarrhea virus during the midtrimester of gestation.

in other herds, other ocular lesions or cerebellar hypoplasia may predominate. The strain of infecting BVDV certainly may play a role in determining the anatomic area of congenital malformation because some strains seem to possess a degree of organ specificity. Both CP and NCP strains are capable of inducing fetal anomalies. Most congenital lesions are thought to indicate in utero infection between days 80 to 150 of gestation. Overlap between this time and the period for persistent infection (40–125 days) exists. Therefore, calves born with congenital lesions may be PI or may be seropositive in precolostral blood samples depending on exactly when the in utero infection occurred and whether NCP or CP BVDV caused the congenital lesion. Calves with congenital lesions should be tested to determine whether they are PI, especially if the congenital lesions are not life threatening and the owner would like to keep the animal.



• **Fig. 6.57** Aborted fetus from a bovine viral diarrhea virus–infected cow.

Reproductive Signs

In addition to fetal congenital defects, BVDV may be associated with a variety of reproductive consequences. Abortion always is a possibility when in utero BVDV infection occurs (Fig. 6.57). Abortion has been observed or caused (experimental infections) at most stages of gestation with CP-BVDV and is possible in the midtrimester or last trimester as a result of NCP-BVDV. Fetuses may be infected several weeks or months before abortion in some instances. Mummification also is possible after in utero BVDV infection.

Perhaps the greatest concerns for future BVDV research revolve around effective protection of fetuses from BVDV. The temporal relationships among PI, congenital lesions, and, to a lesser degree, abortion or mummification seem to have been worked out. However, the consequences of early in utero fetal infection (0–40 days) are not as well known, nor is the degree to which current immunoprophylaxis prevents fetal infection at each stage of pregnancy. The latter becomes vitally important when one remembers that the biggest threat within cattle populations remains PI animals, and the ability to prevent this from happening by vaccination would be a significant step forward in the control of the disease.

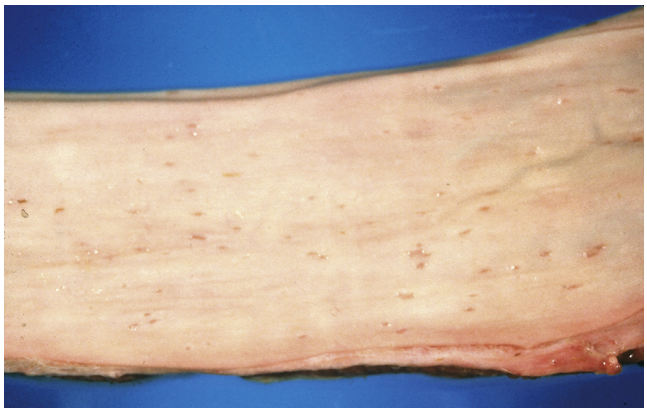
Acutely infected immunocompetent bulls and PI bulls shed BVDV in semen. Insemination with infected semen causes infection and subsequent seroconversion in seronegative female cattle. Cattle infected by such semen tend not to conceive until establishing immunity and seroconversion. Oophoritis has been detected several weeks after experimental infection, and ovarian dysfunction may be responsible for the impaired fertility seen in some infected cow populations. Semen is a possible source of infection and probably has been the occasional cause of reduced fertility and other BVDV related problems in some herds. Frozen semen also has been shown to be capable of BVDV transmission to susceptible cattle. Although PI bulls may have detectable semen abnormalities, these are not consistent, and standard semen testing should not be used in lieu of nucleic acid amplification, viral isolation, or antigen detection to identify infected bulls. Some immunocompetent (non-PI) bulls may also shed the virus in their semen for an extended time after infection. Most commercial bull studs now routinely screen incoming bulls for the PI status, positive serology, or virus shedding before semen collection for AI purposes.

Intrauterine infusion of BVDV at the time of insemination was shown to cause susceptible cows to have early reproductive failure, low pregnancy rates, high return rates, and seroconversion. Reproductive failure occurred as a result of failure of fertilization. However, when either seronegative or seropositive cattle were infected orally or nasally rather than intrauterine, conception was not affected. Thus, the consequences of maternal exposure to BVDV at the times of breeding, fertilization, implantation, or early gestation remain somewhat unknown when infection by routes other than intrauterine occur.

Fluids containing BVDV-contaminated fetal bovine serum used for embryo transfer also can serve as a source of infection in susceptible cattle and be associated with reproductive failure. The potential consequences of PI embryo donors or PI recipients currently dictate rigorous testing of animals to be used for these purposes in embryo transfer.

Diagnosis

In classic cases with fever, diarrhea, mucosal lesions, and digital lesions, diagnosis may be made with some confidence based on the clinical signs, although one should always bear in mind the possibility of other, often notifiable, vesicular diseases depending on location and circumstance. Unfortunately, this represents a distinct minority of the cases. Clinicians must remember that even in epidemic acute disease < 50% of infected cattle may have detectable lesions on clinical examination. In addition, because most cattle infected by strains of BVDV have subclinical or mild infections, signs suggestive of BVDV may be absent. Specific physical examination findings are limited to oral mucosal lesions and digital lesions. Such lesions may be obvious in superinfected PI animals having all of the signs of severe BVD ("mucosal disease") but may be subtle or absent in seronegative animals experiencing acute BVDV infection. Mucosal lesions also may lag behind nonspecific early signs of fever, depression, and reduced milk production. Whenever BVDV infection is suspected, a methodical examination of the oral cavity—aided by focal light illumination—is essential if subtle erosions are to be found. Lesions can be in any area of the oral cavity, but focal erosions of the hard and soft palates, tongue erosions, erosions at gingival border of the incisor teeth, and blunted hyperemic papillae that are eroded at the tip are most commonly seen. Digital lesions are even less common than oral mucosal lesions in field outbreaks of BVDV. When present, coronitis and interdigital erosions are most common. Laminitis usually is observed only secondary to chronic coronitis in PI animals with superinfection. Although not widely practiced, endoscopy to see the esophageal mucosa might allow detection of typical linear erosions that are quite common in both acute and chronic infections (Fig. 6.58).



• **Fig. 6.58** Multifocal linear erosions of the esophageal mucosa caused by acute bovine viral diarrhea virus infection.

Persistence of high fever or biphasic high fever occurring over more than 7 days is found in many acute BVDV infections. Initially, the affected animal may not appear seriously ill and may be thought to have a "respiratory virus." If, however, fever persists and is unresponsive to antibiotics, these same cattle may show more overt anorexia, depression, and dehydration after several days. Diarrhea and mucosal lesions are more common at this time. Few diseases of dairy cattle cause the profound and complete anorexia observed in BVDV-infected cattle having mucosal GI lesions. Oral erosions, esophageal erosions, forestomach erosions, and lower GI lesions contribute to patient pain, discomfort, and subsequent anorexia. Salivation and bruxism also may be observed in these patients.

Bleeding associated with fever and diarrhea in several calves or cows should raise the suspicion of thrombocytopenia associated with acute BVDV infection; it necessitates confirmation of both BVDV infection through appropriate diagnostic tests and thrombocytopenia through taking platelet counts. Similarly, herd reproductive problems such as abortions, mummified fetuses, or dramatically reduced conception rates should be grounds for ruling BVDV in or out as a potential cause. Congenital malformations or lesions in one or a series of calves born within a few weeks or months also should suggest BVDV becomes part of the differential diagnosis.

Routine hematology may suggest BVDV infection but is not reliable as a sole diagnostic aid. For example, leukopenia characterized by lymphopenia is present in most calves and cattle with acute infection with BVDV. Many of these animals are neutropenic as well. Fever of unknown origin coexisting with persistent leukopenia should raise suspicion of acute BVDV but could be mimicked by other diseases such as salmonellosis.

Without question, the most clinically and diagnostically challenging outbreaks of BVDV infection occur with concurrent illness due to *Mannheimia haemolytica* spp. pneumonia, *Salmonella* spp. enterocolitis, or viral respiratory infections such as IBR or BRSV. In such outbreaks, morbidity and mortality may be exceedingly high, and physical findings and lesions at necropsy are predominated by the non-BVDV diseases. Cattle with acute BVDV infection may have had little time to develop pathognomonic gross lesions consistent with BVDV before dying from their concurrent diseases because of the transient immune suppression of cellular defense mechanisms during acute BVDV infection. Necropsy findings in such cases identify overwhelming bronchopneumonia due to *Mannheimia haemolytica*, respiratory pathology consistent with IBR or BRSV, or severe enterocolitis caused by *Salmonella* spp. Lesions consistent with BVDV infection may be absent or only present in a minority of the fatal cases. The temptation for the clinician and pathologist is to accept these gross lesions as sufficient evidence of the primary cause and thus fail to submit samples for appropriate BVDV testing. Many diagnostic laboratories now include BVDV tests, usually in the form of PCR assays, as part of a broader etiologic "panel" when enteric and respiratory disease investigations are carried out using either ante- or postmortem samples.

Similarly, some PI calves or yearlings that are chronic poor-doers and have chronic pneumonia, ringworm lesions, chronic or intermittent diarrhea, chronic parasitism, chronic pinkeye, or other lesions that have not responded to conventional therapy may be written off as having illness caused by the other more obvious infectious diseases if viral lesions are not present or missed. Again, diagnostic testing to demonstrate the presence of BVDV in the animal is essential for positive diagnosis.

Although high mortality calf diarrhea outbreaks are more typically caused by *E. coli,* rotavirus, coronavirus, *Cryptosporidium*, and *Salmonella* spp., occasional outbreaks may have concurrent BVDV infection, and viral isolation should be a part of the diagnostic material submitted from both live and necropsied calves in such cases (see Chapter 18). The differential diagnosis for BVDV infection is lengthy and depends somewhat on the clinical signs present in the affected herd. Acute infections characterized by diarrhea and fever must be differentiated from salmonellosis and other causes of enteritis by bacterial fecal cultures and blood cultures. Abortion epidemics must be differentiated from other bacterial, viral, and protozoan causes of abortion. When hemorrhages are present along with signs of fever and diarrhea, BVDV must be differentiated from bracken fern intoxication, disseminated intravascular coagulation (DIC), other coagulopathies and certain mycotoxicoses.

Other mucosal diseases such as BTV, EHDV and vesicular diseases—both endemic and exotic—must be considered in unusual cases and may necessitate consultation with federal regulatory veterinarians if confusion exists as to the definitive diagnosis.

Concurrent bacterial, viral, or parasitic diseases may confuse or mask the presence of BVDV infection. Whenever

multiple animals fail to respond to conventional therapy for suspected or confirmed bacterial infection, the possibility of BVDV infection should be investigated. Weak or unthrifty PI calves must be differentiated from animals affected by bacterial septicemia, selenium deficiency, and enteric pathogens. The source of illness in chronic "poor-doers" or unthrifty PI calves or yearlings with multiple problems must be differentiated from BLAD, chronic internal abscesses, malnutrition, parasitism, and chronic pneumonia or enteritis.

Because of the variability in clinical signs of BVDV infection, the only absolute proof of BVDV infection is diagnostic testing to demonstrate the presence of virus in tissues or blood. Tracheal wash samples may contain virus in some live calves, and tissues such as intestine, lymph nodes, spleen, and lung may demonstrate virus on necropsy specimens. The presence of virus in blood can be confirmed through submission of whole blood samples for viral isolation from the buffy coat or for detection of viral genetic material through PCR. Virus isolation is still considered the gold standard, but the use of RT-PCR has largely superseded virus isolation as the modern, accurate, and rapid test of choice. PCR is inexpensive, is not restricted to laboratories with cell culture expertise, and is highly sensitive. A variety of samples including whole blood, serum, milk, semen, tracheal fluid, follicular fluid and tissue samples can be tested successfully by RT-PCR. Primers from the 5′ untranslated region of the viral genome allow for identification of either type 1 or type 2 BVDV strains and the test is applicable to either PI or acute infections. A repeat test at least 4 weeks after an initial positive PCR result confirms a PI infection; an acutely infected immunocompetent animal would be expected to have seroconverted and cleared virus by 2 weeks postinfection. A quantitatively high viremia on a single time point sample is however highly suggestive of PI.

RT-PCR can also be used on bulk tank milk samples or pooled serum samples as a means of identifying PI animals within a group of animals. It has been theorized that the maximum herd size that can be tested by bulk tank sampling to identify just one infected animal is 500, although by comparison, it is believed that the pool size for identification of a single positive animal by serum testing is much smaller, probably in the order of 50 individuals. A positive PCR test result from a pooled sample should then direct smaller group testing to identify the infected individual(s). It should be remembered that this positive test result will not distinguish between acute and PI infection and that further repeat testing will be needed to establish the number of infected individuals and specific type of infection present.

Antigen-capture ELISA (AC-ELISA) can also be used in adults and calves older than 6 months of age to detect virus. There are currently AC-ELISA tests available commercially that use serum, milk, or tissue samples. It can be a reliable test for the identification of PI animals but does not have the same sensitivity as RT-PCR when both are compared with the gold standard of virus isolation. AC-ELISA cannot be used with pooled serum samples. AC-ELISA is also considered less reliable in younger calves because colostral antibody may bind to the virus in the blood and limit the ability of antibodies on the ELISA plate to bind to, and therefore detect, the virus. Furthermore, AC-ELISA may not be able to detect the low levels of viremia in some acute infections, so this test may lack sensitivity relative to other viral detection methods for acute cases.

In young calves with colostral antibodies against BVDV, PCR on whole blood is the preferred diagnostic test. Alternatively, skin biopsy (usually ear notches) in formalin or kept cold in saline (check with the diagnostic laboratory) can be submitted for immunohistochemical (IHC) staining, virus isolation, AC-ELISA, or PCR. Ear notch testing has become a very popular test in the United States for the identification of PI animals because virus antigen is consistently found in the ear skin of these animals at any age. Occasionally, acutely infected animals will also show a positive result by ear notch IHC, and this positivity can last for long periods of time, so it is advised that positive animals have a repeat test approximately 4 weeks after a positive IHC ear notch to confirm that they are PI. Because the only appropriate choice is to cull PI animals, it is prudent to run this confirmatory second test, probably a PCR test, particularly in the case of valuable calves. In acute infection of immunocompetent adults, detectable viremia persists for up to 2 weeks. On rare occasions, acutely infected, immunocompetent animals may remain viremic up to 30 to 40 days. The period of viremia tends to be much shorter in subclinically infected animals.

Serology, despite limitations in PI animals, may be helpful when seroconversion can be demonstrated after illness; many animals possess titers > 1:512 following a recent herd epidemic. Paired sera can be obtained at a 14-day interval from animals with clinical signs and/or their penmates; serologic testing for both type 1 and type 2 BVDV should be performed. Obviously, serum titers representing neutralizing antibody levels may be greatly influenced by vaccinations and natural infection. Antibody titers from recently infected, immunocompetent animals are often indistinguishable from vaccination-induced titers.

Positive viral identification coupled with low or nonexistent neutralizing antibody levels suggests acute infection (immunocompetent animal) or persistent infection with BVDV (immunotolerant animal). Generally, immunocompetent animals seroconvert and clear viremia within 2 to 4 weeks, but PI animals remain viremic with low or nonexistent titers to the homologous strain.

Persistently infected animals can be detected by virus isolation or PCR performed on whole blood, and skin biopsies can be submitted for immunohistochemistry, virus isolation, AC-ELISA, or PCR. A positive result on any of these tests may simply reflect acute infection in a normal animal, so PI status is technically confirmed by repeat testing and

detection of the virus at least 3 to 4 weeks after the initial positive result. Animals confirmed as PI should be culled or well isolated from the remainder of the herd because they serve as a constant source of high viral challenge for their herdmates. Again, on occasion, acutely infected, immunocompetent (non-PI) animals can remain viremic for 30 to 40 days. Delaying the second test for 6 weeks may be preferred if the tested animals are of particularly high value; in such cases, false incrimination of an animal as PI would result in significant financial loss. Such animals should be considered PI until proven otherwise by the second test and well isolated from their herdmates. Methods for screening the herd for PI animals are discussed further below in the section on prevention.

Treatment

Cattle with mild clinical disease associated with acute BVDV infection do not require specific therapy but should be offered fresh feed and water and not be subjected to any exogenous stress, transport, or vaccinations. Cattle with specific problems such as diarrhea may require oral or IV fluid therapy if continued diarrhea coupled with relative or absolute anorexia causes dehydration. Clinically ill animals (i.e., those with fever, depression, diarrhea, and dehydration) should not be subjected to any extraneous stress and may benefit from prophylactic bactericidal antibiotics to minimize the potential for opportunistic bacterial infections such as those known to cause pneumonia. Calves with acute BVDV infection are more likely to require supplemental fluids and electrolytes.

In cattle with clinical evidence of bleeding caused by thrombocytopenia, benefit may be derived from fresh whole blood transfusions. Usually 4 L of whole fresh blood collected from a BLV-negative, non-PI-BVDV donor is adequate in an adult unless blood loss has caused life-threatening anemia. In recent years, we have seen this condition more often in calves rather than adults and hence the volume of blood to transfuse will be less; typically, 1 to 2 L is sufficient. Other affected cattle in these herds with thrombocytopenic strains of BVDV may merely be observed if clinical bleeding is not apparent. Such cattle should not be subjected to surgical procedures, parenteral injections, or crowding and should have insect populations controlled to avoid multiple insults that could cause clinical bleeding. Clinical bleeding seldom occurs unless platelet counts are <50,000/μL and trauma to skin or tissues is excessive. Diarrhea may become bloody in some patients in these herds because inflammatory GI lesions may sufficiently irritate the colon to cause bleeding.

Even though most acute BVDV infections are subclinical, this does not hold true for all field epidemics, and some immunocompetent animals do develop severe illness as a result of acute infection with various BVDV strains and thus may benefit from symptomatic therapy. Acute BVDV can be fatal to immunocompetent calves and adult cattle that are naïve to the infecting strain, particularly if other concurrent stressors such as transportation, processing or overcrowding occur. Death caused by BVDV does not implicitly confirm superinfection of PI animals. This is especially true when complications such as thrombocytopenia or secondary bacterial or viral infections befall an immunocompetent individual with transiently depressed cellular immune responses resulting from acute BVDV infection.

Corticosteroids and NSAIDs are contraindicated in cattle with acute BVDV infection because both categories of drug further predispose to digestive tract erosion and ulceration. Animals that are ingesting feed and water are sometimes treated with judicious doses of aspirin as an antipyretic, but aspirin will reduce cytoprotective prostaglandins in the GI tract and kidney. Meloxicam might be a reasonable choice because it is a more selective cyclooxygenase-2 inhibitor with fewer side effects than other commonly used NSAIDs.

Prevention

The currently available diagnostic tests for BVDV facilitate successful BVDV control and possibly even eradication. Test and cull practices in recent years in a growing number of European countries have proven that eradication is achievable but needs central or federal coordination and consistent producer and veterinarian attention to be successful. Even if eradication is not achieved, several countries have been able to reduce the prevalence of PI animals to less than 0.5% from prior levels that were 5 five 10 times as high by using rigorous herd level testing and removal of positive, proven PI animals. Undoubtedly there are now a large number of U.S. dairy herds that individually and informally have some form of PI screening, but it should be part of every herd's biosecurity protocol, independent of cow numbers. Large dairies that use heifer rearers or purchase replacement stock through markets have particular challenges in appropriately screening and "protecting" all pregnant animals from exposure to BVDV during early gestation. The incidence of acute BVDV clinical disease appears to have noticeably decreased over the past 2 decades, presumably because of better vaccination programs and both early detection and elimination of PI calves.

Effective control of BVDV infection in dairy cattle requires four fundamental steps.

1. Improvement of herd immunity through immunization.
2. Identification and removal of PI animals within the herd.
3. Screening of new animals for PI status before introduction into the herd.
4. Implementation of biosecurity practices to prevent fetal exposure to BVDV; in other words, prevention of future PI animals.

Each of these items is detailed in the following section.

1. Improvement of herd immunity through immunization.

The goals of a BVDV immunization program on dairies include; (1) prevention or reduction in severity of acute disease in adults and young stock, (2) prevention or reduction in the rate of fetal infection in pregnant heifers and cows, and (3) enhancement of colostral immunity for protection of newborns. Two fundamental challenges to

effective vaccination for BVDV have existed historically. First, the broad antigenic diversity of BVDV makes it difficult to create a vaccine that induces immunity to all of the potential strains that a herd may encounter over time. Second, the massive amounts of virus shed by PI animals may result in infection even in immunized cattle. The importance of this second issue cannot be overemphasized. Exposure to viremic animals compromises any effort at complete protection of a herd through immunization, and elimination of PI animals from the herd is *the vital step* in BVDV control.

At any one time, there are a plethora of U.S. Department of Agriculture (USDA)-licensed BVDV vaccines commercially available. The practitioner must choose between modified-live and inactivated (killed products). Both have advantages and disadvantages, summarized below:

MLVs—advantages:

- Activation of cellular and humoral immunity
- Long duration of immunity
- Good, albeit incomplete, fetal protection; greater than inactivated vaccines

MLVs—disadvantages:

- Potential for transient immunosuppression
- Potentially unsafe for administration to pregnant cattle (not for all products; some have documented safety for use in pregnant animals)
- Colostrum-derived antibodies may block immune response in calves
- Potential for transient ovarian infection and transient impairment of fertility (not to be used in cattle immediately before breeding)
- May induce acute disease in PI cattle—this should not be a deterrent to use, however

Inactivated (killed) vaccines—advantages:

- Not immunosuppressive
- No risk of fetal or ovarian infection (safe for administration to pregnant cattle and immediately before breeding)

Inactivated (killed) vaccines—disadvantages:

- Primarily activate the humoral immune response (less cell-mediated immunity)
- Require more frequent administration (boostering)
- Shorter duration of immunity
- Variable fetal protection (field versus vaccine strain heterogeneity may limit efficacy)

In the past, MLV vaccines were suspected of inducing clinical disease, including persistent infection, because of insufficient attenuation of the virus used in the product or live virus contamination of vaccine reagents. With greater testing procedures available for extraneous virus testing, as well as higher standards for reagents such as fetal bovine serum for cell culture systems, the risk of live virus contamination of MLV has been greatly reduced compared to the past. To ensure that these precautions are used for a given product, safety data and quality control procedures for vaccine production should be requested from manufacturers of MLV vaccines. There is no doubt that the addition of safer, highly immunogenic MLV vaccines against BVDV in recent years has been a major step forward in the control of acute BVDV infection and the protection of fetuses against in utero exposure and PI. The evolution of BVDV vaccines in the United States has closely followed the antigenic changes in natural infection in the country. In the early 1990s when type 2 BVDV appeared for the first time, there was incorporation of type 2a strains along with the traditional type 1a strains already present into both killed and MLV products. Unfortunately, in recent years, there has been an increase in the prevalence of BVDV type 1b in the United States from natural cases. Although there is good experimental evidence to support cross protection against heterologous strains, one company (Elanco) has now produced a vaccine that contains type 1b BVDV. A recent, thorough meta-analysis examining the effectiveness of currently available BVDV vaccines to prevent reproductive disease concluded that polyvalent vaccines offered greater effective prevention against abortion and fetal infection compared to monovalent products and that the relative risk for abortion and fetal infection was lower with MLV compared with killed products. It is worth pointing out that the beneficial effect of the killed multivalent products was still significant, and it is very important to use MLV products according to label instructions regarding prior vaccination in particular.

A single recommendation for vaccination for dairy herds is unlikely to be uniformly accepted as optimal. This likely reflects different practitioners having varying experiences with a variety of different vaccines. It is likely that this variation in professional opinion occurs because practitioners have been observing a spectrum of BVDV strains challenging a variety of herds over time. The following guidelines are recommended by us and others.

- Replacement heifers (separated from pregnant cows and heifers): Immunize with a MLV product, ideally containing type 1 and type 2 strains at 5 to 6 months of age and again 60 days before breeding. This schedule allows replacement heifers to receive two doses of vaccine before they become pregnant and limits potential problems caused by transient ovarian infection by vaccine virus.
- Adult cows: Administer a MLV vaccine with a label claim for safety in pregnant cattle, once annually 2 to 4 weeks before breeding.

Immunosuppression after MLV BVDV vaccination has been documented, so immunization should be timed to occur during periods of relatively low stress and low pathogen challenge.

If killed products are to be used, the manufacturer's recommendations should be followed regarding the timing of the priming and booster immunization; the interval for these immunizations is typically 2 to 3 weeks. To maximize antibody spectrum, a product containing inactivated type 1 and type 2 virus should be used, or at least one giving demonstrative cross protection against both. Cows and heifers should receive a booster immunization before breeding, in midgestation or midlactation, and for lactating animals,

again at dry-off. Killed products may be optimal for administration to newly purchased, pregnant heifers and cows of unknown previous vaccination status.

Adequately vaccinated dams should impart passive antibody protection to calves through colostrum—at least against homologous strains of BVDV. This passive protection probably dissipates between 3 and 8 months of age in most instances. Therefore, the timing of initial active immunization of calves against BVDV is somewhat controversial. If killed products are used, calves born to vaccinated cows should probably be vaccinated three times—at 12, 14, and 18 weeks of age. Manufacturer's recommendations as regards appropriate intervals between dosing should be followed, but all calves or older cattle of questionable immune status must be vaccinated at least twice to establish primary immunity. Semiannual boosters are then recommended.

Modified-live vaccines are more likely to be blocked by maternal-derived (colostral) antibody in calves, so delaying administration of the first dose until 5 to 6 months is recommended. If protection of younger calves is desired, killed products may be administered at an interval determined by the label.

A common error in vaccination programs is to give only a single killed vaccine to first-calf heifers that have never received previous adequate primary immunization. Management deficiencies allow this mistake to occur more commonly than we realize. Do not assume that dairy farmers have "done it right" and always make directions for use clear-cut when selling vaccine to owners. With more widespread use of computerized records, documentation of proper timing of immunization can be implemented by making BVDV vaccination a recorded "health event" for all cattle.

As dairies become larger, the gap between management and cow-side workers has widened. What the manager perceives to be the standard operating procedure for vaccination and what occurs when cows and calves are vaccinated may be vastly different. Therefore, it is imperative the veterinarian take an active role in training workers on proper vaccine storage, handling, and administration. Personal observation of immunization practices often allows the veterinarian to detect and quickly correct problems. Incomplete protection against BVDV has been documented due to inappropriate storage and handling of vaccines; it is important that farm employees who are responsible for immunization are informed about the correct way to do these tasks. Because labor forces on dairies may turn over rapidly, repeated training sessions on this topic are often required. When necessary, the veterinarian should be willing to assume the role of long-term educator of the workforce.

A mistake we have observed in individually valuable cattle, who are repeatedly used for embryo transfer or oocyte recovery work, is that of linking vaccination to stages in the adult cow's lifecycle; many of these cattle may have very long intervals between dry periods. Consequently, the timing of boosters according to dry off may result in that individual going 18 months or more between boosters and thus being inadequately protected. This can be disastrous when it results in a PI calf but it is also of concern with respect to protection against other antigens frequently combined with BVDV in multivalent vaccines such as BRSV and IBR. A similar phenomenon may present itself in grade cattle that have experienced illness or delayed conception for some other reason. This level of fine detail may escape busy farm managers.

2. Identification and removal of PI animals from the herd.

The prevalence of PI animals is thought to vary greatly from dairy to dairy. Data from a few large prevalence studies indicate that PI dairy cattle typically represent less than 2% of the cattle population. Because the virus may be transmitted vertically, even closed herds may have PI animals.

In the past, serologic screening of the herd after immunization was used to attempt to identify PI animals; the concept was that these animals, being immunotolerant to BVDV, would tend to have low titers, and low-titered animals could then be targeted for testing by virus isolation to confirm PI status. However, in light of the fact that PI animals may mount an immune response to heterologous field or vaccine strains, this method is unlikely to accurately identify all PI animals and is not endorsed.

Tests that detect the presence of virus in the live animal are considered necessary for accurate identification of the PI state. Initially, all animals in the herd, regardless of age or apparent health status, should be included for testing. Tests include virus isolation on whole blood; PCR on whole blood, serum, milk, or tissue; AC-ELISA on blood or milk; and IHC on skin biopsies. AC-ELISA should not be used on calves younger than 6 months of age, owing to potential problems with colostrum-derived antibody interference with viral detection. To prevent confusion between acutely viremic immunocompetent animals and PI animals, it may be necessary to repeat testing on any positive animal a minimum of 3 to 4 weeks (or, for valuable animals, 30–40 days) after the first positive test result. A high level of viremia determined by RT-PCR on an initial blood sample would be suggestive of PI, alternatively BVD antigen positive samples from skin or blood taken prior to colostrum administration in the newborn calf would also be strongly supportive of PI.

To reduce testing costs on large numbers of animals, certain laboratories offer testing on pooled samples; for example, skin biopsies from multiple animals can be placed together in saline, and PCR can be run on the pooled sample to detect the presence of BVDV. Alternatively, composite milk samples can be pooled together and checked for the presence of virus by PCR or virus isolation. Bulk tank samples or string samples can also be used to screen large numbers of lactating cows. Current recommendations state that pooled milk samples should represent fewer than 400 animals to optimize chances of detection of PIs. It is best to check with the regional veterinary diagnostic laboratory for the preferred number of samples to be pooled, shipment requirements, and so on. Obviously, a positive result on a pooled sample would require follow-up testing of the constituent individuals to identify the viremic or PI animal(s).

On rare occasions, infected bulls may shed virus only in the semen. Therefore, to cover this rare yet complication-rich scenario, bulls that test negative for virus in blood or skin biopsy should ideally have their semen screened by virus isolation or PCR.

Ethical considerations regarding the fate of PI animals are worthy of mention. Sale of animals known to be PI at livestock auctions is simply unethical because these animals serve as virus-producing machines that expose many other animals, causing potentially devastating disease on the farms of the unknowing purchasers. The most ethical practice is to euthanize these animals on the premises, although otherwise healthy animals may be considered for slaughter. To our knowledge, no studies currently exist on the persistence of BVDV in properly composted carcasses, but data on the survivability of viruses related to BVDV indicate that long-term environmental persistence is unlikely. Alternatively, carcasses of PI animals can be removed from the premises for rendering.

After all animals in the herd have been tested and PI animals removed, testing should focus on the calves born to gestating, non-PI females. After these calves have been tested and PIs removed, testing should focus on new introductions, show animals, semen and embryos, and heifers raised off-site (see numbers 3 and 4 below). The producer must understand, however, that introduction of a novel strain of BVDV onto a farm may result in fetal infection in non-PI, pregnant females, warranting eventual testing of their offspring.

3. Screening of new animals for PI status before introduction into the herd.

Reducing risk of introduction of BVDV is best accomplished by avoiding the purchase of untested cattle. Without question, the greatest disasters resulting from acute BVDV have followed the purchase of assembled cattle from sales to increase herd size. These purchased animals may be PI. Alternatively, they may be acutely infected—in either case, they represent sources of new virus for the herd. In addition, if newly purchased cows or heifers were exposed to BVDV and became viremic in early pregnancy, they may be carrying PI fetuses. Dr. Joe Brownlie in the United Kingdom, a world-renowned expert on BVDV, aptly refers to these individuals as the "Trojan cow" of BVDV transmission. Therefore, for optimal herd protection, purchased adults should be tested before introduction into the herd; later, the offspring that they were carrying at the time of purchase should be tested for PI status too because PI calves can be born to immunocompetent dams. Whenever possible, new herd introductions should be tested and well isolated from the remainder of the herd for 4 to 6 weeks. During this period, any PI animals in the group of new introductions can be identified and removed, and any acutely infected animals can be given adequate time to recover. Any contact between isolated new introductions and the remainder of the herd, even at fence lines, should be avoided during this period.

Tests to detect virus should be used on newly purchased cattle. Virus isolation, PCR, immunohistochemistry, or AC-ELISA on blood or skin biopsies can be used. Pooling of samples can be considered when large numbers of animals are to be introduced (see number 2). Collection of samples for testing for other diseases in newly purchased stock (e.g., Johne's disease, *Mycoplasma* spp. mastitis) can be performed at the same time.

4. Implementation of biosecurity practices to prevent fetal exposure to BVDV.

Fetal infection leading to the PI state is a critical control point because PI animals represent a massive source of viral challenge for the herd. Even with good vaccination practices, all immunity is finite, and overwhelming viral challenge could theoretically lead to transplacental passage of virus even in immunized, pregnant females. Therefore, protection of pregnant cows and heifers from exposure to high viral challenge is a critical goal of BVDV control within a herd biosecurity program.

Contact with cattle outside the herd should be eliminated or minimized, even at fence lines. Cows and heifers in the first trimester of pregnancy should be considered the most susceptible to creation of the fetal PI state. These animals should be located on the farm in the area that is most protected from contact with outside cattle, new introductions, and show cattle. Pen allocation and pen milking sequence should be critiqued and, if necessary, changed to maximize protection of these animals. Contact of these animals with ill cattle should be minimized whenever possible. The possibility of transmission from wild ruminant species exists, such as white-tailed deer, but this probably represents a very unlikely method of acquiring new infection for most commercial U.S. dairies.

Heifers raised and bred at heifer-raising operations warrant particularly careful scrutiny in a BVDV control program. Heifers from multiple herds are often raised on such operations, and viral challenge from PI animals or acute BVDV infections on that operation could easily induce fetal infection in pregnant heifers. Therefore, young heifers should be tested before transport to heifer-raising operations; if this is not feasible, prompt testing after arrival on such operations is warranted. In addition, all calves born to heifers raised offsite should be considered potential PI animals and tested after birth.

Cattle taken to shows should be considered another source of novel virus on a farm. In the ideal world, show cattle should be tested and confirmed to be non-PI before being taken to shows or sales; this is simply a good ethical practice intended to protect other animals and producers. Show cattle should also be well immunized to limit the likelihood of them developing acute infection while off the premises. Given the shortcomings of vaccines in protecting against the tremendous number of strains of BVDV, even well-vaccinated show cattle should be considered potentially exposed to novel BVDV at shows or sales and ideally kept isolated from the home herd for 4 to 6 weeks on return. If exposed at shows, pregnant show cows and heifers may experience viremia of sufficient magnitude to induce fetal infection, and their calves should be subsequently tested for PI status.

Most reputable sources of semen, embryos, and fetal calf serum have BVDV testing strategies in place. However, nothing should be taken for granted, and the individuals or companies providing bull semen or embryos should be requested to provide documentation of their current control programs and quality control measures for reagents. Control of BVDV in embryo and semen production operations has been recently reviewed. In short, acutely or PI bulls, embryo donors, and embryo recipients are potential animal sources of BVDV. Animals within these populations that shed large amounts of virus may cause fetal infection in others, so BVDV testing of all animals with which bulls, embryo donors, and embryo recipients come in contact during semen or embryo collection, transfer, and pregnancy is necessary. Rarely, infected bulls may shed virus only in semen (i.e., test negative on blood or skin IHC), and semen testing by PCR or virus isolation is considered optimal. All animal-origin reagents used in embryo transfer or in vitro fertilization (including semen and oocytes) should be screened for the presence of BVDV.

Winter Dysentery

Etiology

The etiologic agent responsible for winter dysentery has remained elusive for as long as the disease has existed. In the northern hemisphere, the disease is characterized by explosive herd outbreaks of diarrhea between the months of October and April. *Campylobacter fetus jejuni* long was suspected as a cause, but Koch's postulates never were confirmed. MacPherson, however, was able to infect susceptible cattle using filtered feces from infected cows and therefore believed a virus was involved. Bovine coronavirus has been demonstrated in feces and colonic epithelium of affected cattle, and the same strain that causes diarrhea in calves has been used to experimentally create winter dysentery in adult cows. In Europe, Breda virus (*Torovirus* genus) has been associated with winter dysentery outbreaks. In North America, bovine coronavirus is likely responsible for most outbreaks. It may be introduced into the herd by carrier animals or alternatively an established herd member may be a carrier. Virus shedding in the manure during winter months is thought to allow propagation of the virus and may result in infection and clinical signs in susceptible adult cattle and heifers.

Winter dysentery is of economic importance primarily because of production losses both during the acute outbreak and because some cows do not return to previous production levels for the remainder of the lactation. Death losses are minimal but do occur—almost always in first-calf heifers that develop hemorrhagic diarrhea.

The disease is highly contagious and can spread easily from an affected herd to unaffected herds via fomites—both inanimate and animate. Veterinarians, milk tank drivers, inseminators, salespeople, and other farm visitors frequently are blamed for spreading the disease. Newly purchased cattle and cattle attending shows during the fall, winter, or early spring also can be infected and instigate a herd outbreak. Herds experiencing winter dysentery subsequently appear immune for 2 to 3 years, based on clinical impressions that many herds have an outbreak every third year. Relative age-related resistance is observed, but this protection is incomplete. Cattle infected for the first time tend to have more severe clinical signs than those previously affected.

Clinical Signs

Signs include acute diarrhea in 10% to 30% of the cows within a herd followed by similar signs in another 20% to 70% of the animals within the ensuing 7 to 10 days. The diarrhea is explosive and appears semifluid, dark brown in color, has a pea-soup consistency, is malodorous, and forms bubbles as puddles of manure are formed. Most affected cows have decreased appetite, production losses of 10% to 50%, and become mildly to moderately dehydrated. Some develop cool peripheral parts and sluggish rumen motility suggestive of hypocalcemia. Severely affected animals—especially first lactation cattle experiencing the disease for the first time—have hemorrhagic enterocolitis with dysentery and fresh blood clots in the feces. Tenesmus may be present in these animals, and blood loss anemia may develop. A soft, moist cough is often apparent in several of the affected animals, but the lungs auscult normally in these cattle. Fever usually precedes clinical signs by 24 to 48 hours, and experienced clinicians will detect fever in apparently healthy herdmates that have not yet developed diarrhea. Some cattle have mild fever 103.0° to 104.0°F (39.44° to 40.0°C) accompanying the onset of diarrhea. Herd production decreases commensurate with incidence and severity of disease. Affected cows, especially those in mid or late lactation, may not return to previous production levels for the remainder of their current lactation.

Diagnosis

Winter dysentery must be differentiated from dietary diarrhea, coccidiosis, BVDV, and salmonellosis. Dietary indiscretions that induce diarrhea seldom cause fever and are usually associated with feed changes. Coccidiosis can cause diarrhea, dysentery, whole blood clots, and tenesmus in heifers and on very rare occasions in first lactation animals but does not affect multiparous cows. Fecal smears and flotation allow a diagnosis of coccidiosis. Usually BVDV infection causes leukopenia, higher fever, more prolonged disease, and could be ruled out by PCR or viral isolation. The most likely differential diagnosis is salmonellosis caused by types E, K or mildly pathogenic types of B or C *Salmonella* spp. Salmonellosis of these types can cause fever—frequently preceding the onset of diarrhea—and a variable number of animals may develop diarrhea that contains blood, fibrin strands, or mucus. A neutropenia with left shift in the leukogram would suggest acute salmonellosis but is not a consistent finding. Fecal culture obtained from several acute cases is the only way to rule out salmonellosis.

Confirmation of the presumptive diagnosis requires demonstration of bovine coronavirus in feces by electron microscopy, ELISA, or PCR. IHC staining of colonic tissue may be used on necropsy specimens in acute cases. Outbreaks of diarrhea in North American adult cattle that occur outside the October to April time period are unlikely to be winter dysentery.

Treatment

For most affected cattle, supportive treatment with oral astringents remains the time-tested mode of therapy. Occasional high-producing cattle require parenteral calcium solutions to counteract secondary hypocalcemia or treatment of ketosis secondary to reduced appetite. Oral fluids and electrolytes may be necessary for moderately dehydrated cattle. All cattle should have access to salt and to fresh water.

Severely dehydrated cows occasionally require IV fluid therapy, and first-calf heifers that become anemic because of blood loss require fresh whole blood transfusions in some rare instances. Cattle with tenesmus may necessitate epidural anesthesia to allow rest and reduce rectal and colonic irritation.

Treatment usually is only necessary for 1 to 5 days, by which time most affected cows have recovered their appetites and normal manure consistency. Unfortunately, the disease often dwindles through the herd for 7 to 14 days, such that new cases are still appearing at a time when most cattle are recovered. Although there is no proven efficacy to preventive measures, practicing sound herd biosecurity regarding new herd introductions and show cattle may reduce the likelihood of outbreaks (see descriptions in section on BVDV). Furthermore, disinfected boots and equipment should be required for all visitors to a dairy, as well as clean outer garments. The efficacy of immunization with commercially available coronavirus vaccines is currently unknown.

Campylobacter jejuni

Etiology

Campylobacter jejuni, formerly *Vibrio jejuni,* is a gram-negative, curved to spiral, motile rod capable of causing enterocolitis in many species, including humans, in whom the organism is one of the major causes of bacterial enterocolitis. *C. jejuni* may be present in the normal intestinal flora of many domestic animals and people but is found with greater incidence in diarrhea patients. Because of its ubiquitous nature, the significance of isolation of *C. jejuni* from diarrheic feces of cattle is hard to interpret. However, isolation of *C. jejuni* coupled with failure to isolate other pathogens such as *Salmonella* spp., *Clostridium* spp., or enteric viruses should be considered significant.

Diarrhea and other clinical signs vary from inapparent or mild to fulminant and may be influenced by concurrent diseases, stress, inoculum, strain of *C. jejuni*, other enteric pathogens, and other factors. This variation is highlighted by the reported profound differences observed between experimental infection of gnotobiotic calves and experimental infections of calves with normal GI flora. Gnotobiotic calves had mild catarrhal enteritis with minimal clinical signs, but fever, chronic diarrhea for up to 2 weeks, and some degree of dysentery were observed in nongnotobiotic calves. Although most experimental infections have been conducted in calves, adult cattle are thought to be susceptible as well. Infection in people can occur at any age.

The strain of *C. jejuni* and other factors may influence the site of colonization within the intestine, but most strains affect both the small intestine and colon. *C. jejuni* produces a cholera-like enterotoxin that is an important component of pathogenicity. The organism is mucosa associated but does not appear to be invasive, at least in experimental studies.

Clinical Signs and Diagnosis

Mild or inapparent cases yield little or no detectable signs. Clinical patients with severe signs of diarrhea, fever, dehydration, anorexia, cessation of milk production, and dysentery tend to be sporadic or only represent a low percentage of cattle within a herd. Both adult cows and calves are at risk.

The signs are nonspecific and require differentiation from those indicating salmonellosis, coccidiosis, BVDV infection, and other enteric pathogens. Isolation of *C. jejuni* from diarrheic feces and ruling out other enteric pathogens are essential for diagnosis. Enterocolitis resulting from *C. jejuni* seldom is confused with winter dysentery because of low morbidity in the former contrasted with high morbidity in the latter. Salmonellosis represents the primary differential diagnosis.

Treatment

Calves or cattle with severe diarrhea or dysentery may require 7 to 14 days for recovery. Some diarrhea may persist despite improved vital signs in recovering patients. Oral or IV fluids may be required, and it is best to select fluids following blood acid–base and electrolyte analysis. In humans, antibiotics such as erythromycin, tetracycline, fluoroquinolones, and aminoglycosides are often effective, but penicillin, ampicillin, cephalosporins, and trimethoprim–sulfa combinations appear ineffective. Increasing concern exists regarding *C. jejuni* isolates obtained from human cases of campylobacteriosis and antibiotic resistance (see section on calf diarrhea). Antibiotic susceptibilities are not known for *C. jejuni* infections in cattle and would be best determined by fecal culture and susceptibility results.

Control

Because animals and animal products, such as unpasteurized milk and improperly cooked meat, usually are blamed for *C. jejuni* enterocolitis in humans, a positive diagnosis of *C. jejuni* diarrhea in cattle justifies public health concerns. Infected cattle may recover from the infection over several weeks or may remain carriers. Obviously, many cattle (and people) are asymptomatic carriers, so wide-scale herd testing serves little purpose. However, veterinarians should advise

caution in handling infected cattle and avoidance of unpasteurized milk on farms where the problem is confirmed. *C. jejuni* will grow in milk and may arrive in milk from septicemic spread but is more likely to contaminate milk because of environmental contact.

Enterotoxemia in Adult Dairy Cattle

Etiology

Enterotoxemia thought to be caused by *C. perfringens* has been observed as a sporadic cause of acute death in adult dairy cattle. Some herds have had endemic problems with more than one cow being found dead or agonal over a few months. Premonitory signs are not observed, and, as in calves, the condition is believed to be related to diets exceptionally rich in protein and energy. *C. perfringens* organisms of multiple types that are present in the intestinal tract take advantage of such rich diets and proliferate to produce excessive exotoxins, especially beta-toxin in the case of *C. perfringens* type C. Two recent reports have identified *C. perfringens* type E associated with hemorrhagic enteritis and abomasitis in adult cattle from both North and South America, suggesting a role for the iota toxin elaborated by this particular organism. Because more than one *C. perfringens* type and toxin have been identified from acute necropsy specimens in adult cattle, it is not known which and how many strains of the organism might be responsible for the adult cow disease.

Clinical Signs

Signs are minimal, and most affected cows are found down, agonal, or already dead. Diarrhea may be observed or the animal simply may have abdominal distension, colic, and depression. Another clinical syndrome is one that causes fever, anorexia, small-volume diarrhea, and death, with severe abomasal edema found at necropsy.

Diagnosis

Fresh necropsy specimens must be obtained if *C. perfringens* is suspected as a cause of acute death. Necropsy lesions, as in calves infected by *C. perfringens* type C, may be minimal but might include small intestinal fluid distension, serosal hemorrhages, and an edematous mesentery. Feces and small intestinal content should be cultured for *C. perfringens*; if possible, luminal contents should be tested for the presence of toxins. Samples should be transported in a cooled but unfrozen state. A complete necropsy to rule out other causes of acute death is imperative because the diagnosis of *C. perfringens* enterotoxemia is suggested by exclusion of other diseases such as peracute, virulent salmonellosis.

Treatment and Control

Treatment is seldom possible, but if a specific *C. perfringens* organism and toxins are identified, vaccination with appropriate toxoids would be indicated for potential control. Nutritional management to prevent "slug" feeding is also prudent, especially with high crude protein diets.

Malignant Catarrhal Fever

Etiology

Malignant catarrhal fever has been observed in domestic and wild ruminants worldwide and is caused by a group of gamma herpesviruses. It is a severe lymphoproliferative disease characterized by high fever, corneal edema, mucosal erosions, and lymph node enlargement in clinically affected animals. Lymphocytic vasculitis of a variety of tissues is the classic microscopic lesion. Many ruminant species and pigs are susceptible to MCF viruses, and losses have been incurred on dairies, feed lots, ranches, game farms, zoos, and deer meat–raising facilities. It also causes high mortality rates in bison. Several epidemiologic forms of MCF exist, and they are classically defined by their reservoir ruminant species. In Africa, the causative agent has been isolated and identified as Alcelaphine herpesvirus-1 (AHV1). The term Alcelaphine relates to the subfamily of Bovidae, Alcelaphinae, in which wildebeest, hartebeest, and topi are classified. These species are thought to be the reservoir. The virus apparently is highly cell associated but can be spread during times of stress such as parturition or shipment and may be free in fetal fluids or the young of wildebeest. At times when the virus is released, it becomes infectious for cattle.

In other parts of the world, including the United States, MCF is termed "sheep associated" because sheep appear to be the most likely reservoir of infection. Ovine herpesvirus type 2 (OvHV2) has been identified from ruminants with MCF using PCR, and seroconversion of cattle with MCF to OvHV2 can be demonstrated using competitive inhibition ELISA (CI-ELISA). Recent molecular studies have identified no significant difference in the genome of OvHV2 from sheep and a cow clinically affected by MCF. This suggests that the pathogenesis of sheep associated MCF is not associated with any genomic rearrangement as had been previously theorized. Even though OvHV2 cannot be cultured in vitro, its genome has been sequenced, allowing an increase in the amount and variety of research on the disease and potentially accelerating progress toward a vaccine. In North America, sheep-associated MCF occurs in cattle, bison, pigs, deer, elk, and moose.

Most sporadic or epidemic MCF in cattle has been associated with proximity to sheep. Outbreaks have most frequently occurred when both sheep and cattle were housed on the same farm or cattle were exposed to sheep at fairs. Infection is widespread in North American sheep, and ovine infection is almost always asymptomatic. Cattle and sheep do not have to interact or be in common pastures for the disease to appear. In cattle, cases can be observed at any time of year, although in bison it has tended to be a late winter or early spring disease. In addition, some cattle that develop MCF have no historical direct or indirect exposure to sheep. Asymptomatic, persistent infections with OvHV2 in cattle may occur, and these infections may or may not develop into clinical MCF. Hence, the incubation period for this disease has been difficult to ascertain, with infected cattle developing disease weeks to months after exposure. This

may explain why some cases do not seem linked to exposure to sheep. In one study, most dairy cattle exposed to OvHV2 under natural conditions (close proximity to a sheep feed lot) developed asymptomatic infection rather than overt signs of MCF.

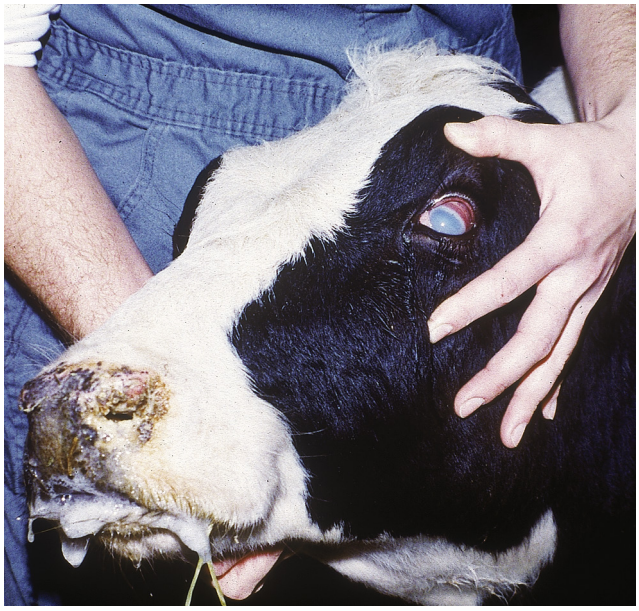
OvHV2 is carried as a lifelong subclinical infection in sheep, and under most husbandry conditions, lambs are not infected as true neonates but after 2 months of age. Rearing sheep from a young age in the absence of adults is a management practice that has proven successful in the reduction of infection prevalence. Nasal shedding by adult sheep is the predominant means of transmission and it is suggested that adolescent sheep (6–9 months of age) shed the most infectious virions; indeed, adult ewes only shed intermittently and at lower amounts then these younger animals. Shedding occurs all year round and contrary to what was often thought there is no association between parturition and increased shedding in ewes.

Clinical Signs

Most cattle affected with MCF have dramatic clinical signs of multisystemic inflammatory disease. Profound pathologic changes can be seen associated with lymphocyte infiltration, diffuse vasculitis, and necrosis throughout the body. Many studies in cattle, bison, and experimental animal infections in rabbits have revealed that CD8 lymphocytes are the predominant cell type associated with vasculitic lesions as well as lymphoid hyperplasia. A great deal of clinical variation is possible, possibly related to the marked variation in tissue tropism demonstrated by OvHV2 at different times in its replication cycle (hence the reason for such difficulty in propagating it in cell culture), and this has caused many authors to categorize MCF based on the predominant organ system involved clinically (i.e., head and eye, enteric, skin). Such categorization is difficult because significant overlap and intermediate clinical situations occur frequently. Sporadic cases are most common in cattle, but herd epidemics have been described in several areas of the United States.

Fever is common in all cases and is high (105.0° to 108.0°F [40.56° to 42.2°C]). Peracute, acute, chronic persistent, and chronic intermittent cases all have fever that usually persists as long as signs are observed. Lymphadenopathy is another finding that is common to most cases. All other signs result from a severe vasculitis that affects many organs but may affect some organs more than others in individual patients. Vasculitis is profound and histologically associated with lymphocytic infiltrates that occasionally can be so extensive as to suggest lymphoreticular neoplasia. Vasculitis affects the GI tract, central nervous system (CNS), eyes, urinary system, liver, skin, upper respiratory tract, and other areas. Hematuria is a common finding if the kidneys and bladder are involved.

Peracute cases may die within 1 to 2 days because of overwhelming viremia and vasculitis of all major organs and yet have minimal clinical signs other than fever, lymphadenopathy, depression, and prostration. Terminal neurologic signs are possible as a result of CNS vasculitis.



• **Fig. 6.59** Adult Holstein cow with the head and eye form of malignant catarrhal fever.

The classical "head and eye" form of MCF is most common in sporadic cases. This form is characterized by persistent high fever (105.0° to 108.0°F [40.56° to 42.22°C]), lymphadenopathy, severe nasal and oral mucosal lesions, ocular lesions, and remarkable depression (Fig. 6.59). In acute cases, extensive mucosal lesions may make it appear as though the animal has had its mucous membranes burned. Frequently, the muzzle and large regions of the oral mucosa appear hyperemic and have a blanched necrotic epithelium that sloughs to leave erosions and ulcers if the patient survives long enough for this to occur. The muzzle may appear dried or sunburned, and the superficial epithelium subsequently may slough away. Sloughing of the nasal mucosa may result in diphtheritic crusts that occlude the airways. Salivation and copious nasal discharge are typical findings. On occasion, oral and nasal lesions are present in the caudal aspect of the mucosal surfaces, out of visual range during physical examination.

Bilateral ophthalmitis results from vasculitis throughout the eyes that spares only the choroid in most cases. Corneal edema is the most common lesion and occurs because of inflammatory changes and exudative cellular deposits on the corneal endothelium that disrupt this layer, thereby allowing overhydration of the corneal stroma. The corneal edema typically begins at the limbus within 2 to 5 days after the onset of fever. The corneal edema then rapidly spreads to the center of the cornea. This centripetal spread of edema distinguishes the ocular features of MCF from contagious keratoconjunctivitis (pinkeye). A severe anterior uveitis, scleritis, conjunctivitis, and retinitis usually coexist. As in other regions of the body, mononuclear cell infiltrates appear in the eyes. Depression is profound because of CNS vasculitis, and CSF confirms a dramatic inflammation characterized by increased protein values and mononuclear cell pleocytosis. Other neurologic signs are possible. Skin lesions

and inflammation of the coronary bands and horn basal epithelium also are possible in those patients that survive more than a few days. The clinical course for most "head and eye" MCF cattle is 48 to 96 hours, although some cases may survive for a longer time, and a few have even been reported to survive.

Acute MCF also may cause severe enterocolitis with diarrhea being a predominant sign. Such cases also are febrile and can have some degree of mucosal lesions, ocular lesions, and other organ involvement. This "enteric form" is again a relative designation because patients frequently have other detectable lesions in addition to diarrhea. However, severe diarrhea may be the most apparent sign and thus may confuse the diagnosis of MCF with BVDV, rinderpest, or other enteric diseases. In bison, enteric signs tend to predominate in acute cases.

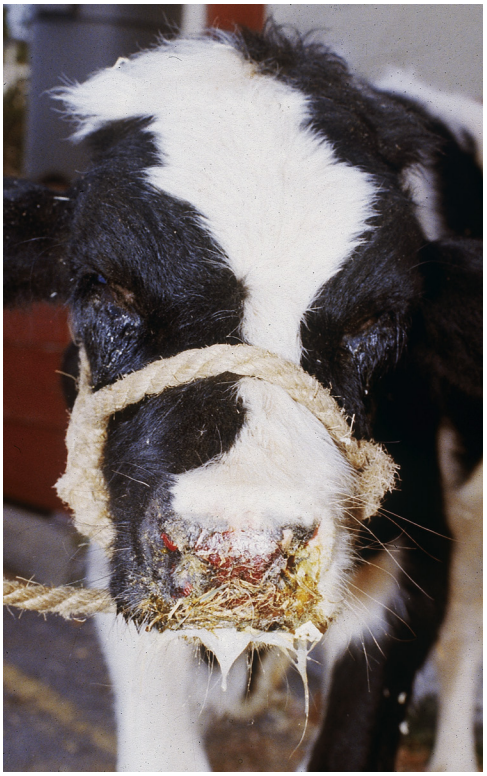
Mild forms of MCF also have been observed. The broad spectrum of potential and observed clinical signs in MCF also makes it likely that some cattle have subclinical mild disease such as enteritis, recover, and respond immunologically to the causative virus. Although previously considered to be a highly fatal disease, up to 50% of MCF cases may survive the acute disease to either recover or become chronic cases.

A rare acute form of the disease presents as a severe hemorrhagic cystitis with hematuria, stranguria, and polyuria. Cattle having this form of acute infection have high fever and only survive 1 to 4 days. Although the most striking clinical signs are limited to the urinary system, histologic evidence of vasculitis and lymphocytic infiltration are generalized on necropsy study.

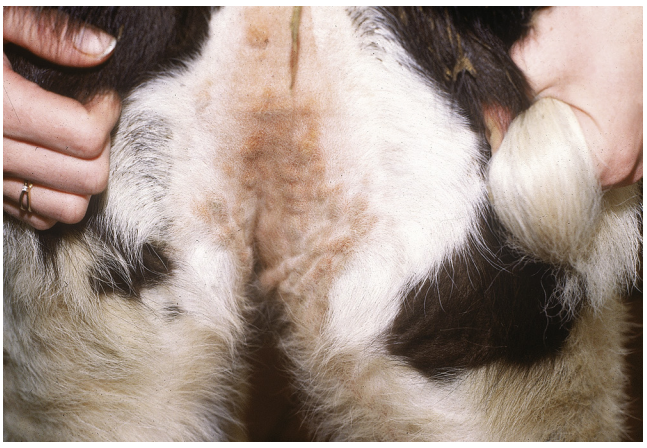
Chronic MCF is characterized by a long clinical course—usually weeks—of high fever, erosive and ulcerative mucosal lesions, bilateral uveitis, papular or hyperkeratotic skin lesions, lymphadenopathy, and digital lesions (Figs. 6.60 and 6.61). Mucosal lesions tend to be severe, slough tissue, and cause salivation and inappetence (Fig. 6.62). Some chronic cases recover only to relapse weeks to months later. Such cases appear healthy between episodes, but recurrence of fever and mucosal, ocular, and skin lesions is debilitating. It is rare for chronic MCF cattle to recover completely and survive.

Diagnosis

Given the wide variability of possible clinical signs of MCF in cattle, the differential diagnosis could include many diseases. Head and eye lesions could be confused with severe IBR associated respiratory and conjunctival infections because corneal edema can occur in some severely affected IBR conjunctivitis cases. However, IBR usually is epidemic and affected animals have characteristic mucosal plaques present on the palpebral conjunctiva and nasal mucosa. Many mucosal diseases such as BVDV, BTV, VSV, EHDV, and FMD may need to be considered depending on the duration, geographic location, and severity of signs. Cattle having severe diarrhea but minimal mucosal lesions could be confused with BVDV infection (see Chapter 16).



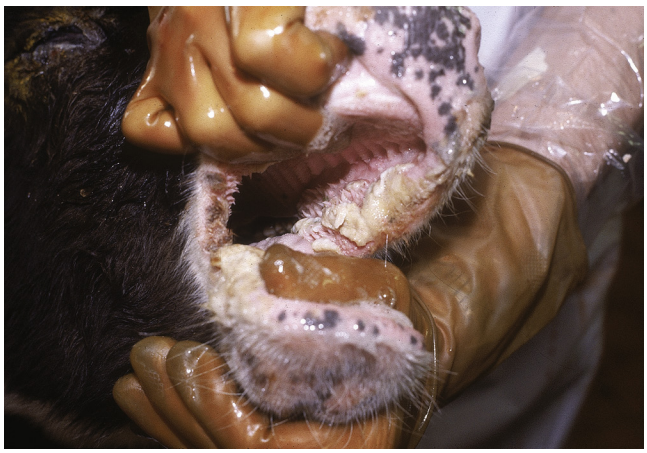
• **Fig. 6.60** A 6-month-old Holstein bull with chronic malignant catarrhal fever.



• **Fig. 6.61** Papular dermatitis in the escutcheon region in a calf with chronic malignant catarrhal fever.

Acute bracken fern intoxications, bacillary hemoglobinuria, and other causes of hematuria may be considered in acute MCF characterized by hemorrhagic cystitis. Acute or subacute mucosal lesions that cause sloughing of muzzle epithelium could be confused with primary or secondary (hepatic) photosensitization.

When ocular lesions are present in MCF patients, the diagnosis is made easier because none of the other mucosal diseases cause severe uveitis and such profound panophthalmitis. As mentioned, IBR conjunctivitis can have corneal edema in severe cases, but intraocular inflammation does not occur with IBR. There are no ocular lesions in acute or chronic postnatal BVDV infections.



• **Fig. 6.62** Chronic necrotic oral and lingual lesions in a yearling heifer with chronic recurrent malignant catarrhal fever.

The acute mucosal lesions of MCF also are unique in classic cases. The oral mucosa is diffusely inflamed and appears as though the patient drank boiling water. The muzzle mucosa appears burnt, crusty, or eroded in these same patients. Unfortunately, these classic mucosal lesions do not occur in all MCF patients, and patients with multifocal erosions or ulcers can be more difficult to differentiate from those with BVDV and other mucosal diseases.

High fever (105.0° to 108.0°F [40.56° to 42.22°C]) that persists through the entire clinical course is characteristic of most acute MCF cases. Chronic MCF cases also have persistent fever that may or may not be as high as that found in acute cases.

Nervous signs suggest a diagnosis of MCF because CNS involvement is rare with other mucosal diseases. However, high fever and terminal prostration are common in fatal cases of most mucosal diseases and could be confused with neurologic signs.

Clinical diagnosis of MCF can be supported by CSF analysis. The characteristic CSF mononuclear cell pleocytosis and elevated protein value found in MCF patients is useful whenever the patient's clinical signs dictate consideration of differential diagnoses. Confirmation of sheep-associated MCF requires demonstration of the viral genome in the blood through PCR analysis of white cells obtained from a whole blood (ethylenediaminetetraacetic acid [EDTA]) sample. Depending on the primers used, PCR for OvHV2 can be specific for that virus and will not detect the genome of AHV1. Alternatively, CI-ELISA can be used to detect MCF antibodies, but this test may not be positive at the time of initial clinical signs. CI-ELISA detects antibodies to either OvHV2 or AHV1 and cannot currently distinguish between wildebeest-and sheep-associated MCF.

Histopathology allows detection of pathognomonic diffuse vasculitis with lymphocytic infiltrates in many organs, including the GI tract, urinary tract, liver, adrenals, CNS, skin, and eyes. Necrotizing vasculitis is present in lymphoid tissues.

Prevention

Prevention of MCF centers on limiting exposure to infected wildebeest and sheep. Airborne transfer of OvHV2 is suspected to occur over a distance of more than 70 m, so segregation of cattle from sheep by greater distances may be protective. This should be considered when housing cattle and sheep at fairs. For cattle and bison herds, Callan recommends a separation distance of 1 mile from sheep. Carrier (asymptomatically infected) cattle may be identified by CI-ELISA on serum and PCR on whole blood. The CI-ELISA test detects seroconversion in exposed individuals that may, owing to varying viral loads in blood, be intermittently negative by PCR on whole blood. Alternatively, acutely infected animals may be positive on PCR but negative on CI-ELISA owing to the delays inherent in generation of an immune response. Therefore, application of both tests may provide optimal sensitivity for detecting infected cattle. Although OvHV2 DNA can be detected in milk, nasal secretions, and ocular secretions of asymptomatic and clinically affected cattle, this viral DNA appears to be cell associated and does not pose a significant risk for horizontal transmission. Transmission from cattle or bison to other animals has not been demonstrated and is considered likely to be a rare event, if it occurs at all.

Johne's Disease (Paratuberculosis)

Etiology

Paratuberculosis (Johne's disease) is a chronic intestinal infection of cattle and other ruminants, caused by *Mycobacterium avium* subsp. *paratuberculosis* (MAP). The disease has a worldwide distribution, and in the United States, recent surveys conducted by the National Animal Health Monitoring system have demonstrated that 70% to 90% of all U.S. dairies have MAP-infected animals.

The etiologic agent is an acid-fast organism that has fastidious in vitro growth requirements requiring special media and may require up to 16 weeks to cultivate from fecal samples. It survives well in farm environments and can survive for 1 year or more in soil and water. The organism is an intracellular pathogen that survives within macrophages. Although regarded as primarily an enteric infection, as the infection progresses MAP may spread via macrophages in blood or lymph to other important sites such as supramammary lymph nodes, mammary gland, (and milk), and uterus (and fetus). Regional surveillance of Pennsylvania and, indirectly, other areas of the northeastern United States confirmed a dairy cow prevalence of up to 7.3% in many areas. With ever increasing dairy herd size, largely the consequence of purchase of cows of unknown status, the herd prevalence will continue to increase. In the United States, MAP-related average costs have been estimated to lie between $22 and $27 per cow per year, but the economic impact of the disease in positive herds versus confirmed negative herds is much higher and may exceed $100 per cow per year when infection progresses to clinical signs of disease. Without question, this disease is of tremendous economic importance to the entire cattle industry and especially to the dairy industry.

Transmission

The most important means of transmission of MAP is by the fecal–oral route. The MAP organisms are shed in the feces of infected cattle and ingested by susceptible animals. Resistance to infection increases with age, so important sites for exposure include maternity pens, cows' udders, colostrum (especially pooled colostrum), milk or milk replacer feeding implements, calf housing areas, and anywhere young calves can be exposed to feces from adult cows. Direct contact is not necessary because studies have shown that in heavily contaminated farm environments, MAP may aerosolize with dust and contaminate surfaces located short distances from adult cow housing areas. Infected cows can also shed MAP directly in milk, and transplacental transmission has been documented in 20% of subclinically infected pregnant cows and up to 40% of cows with clinical signs of Johne's disease.

Older calves have a more variable outcome after infection, and larger doses of MAP are required to cause infections that lead to later onset of clinical signs. Furthermore, young adult or adult cattle seem to have even greater age-related resistance. However, this resistance is relative rather than absolute, and some experimental infections of older calves and adults have been reported. Factors including concurrent diseases, genetics, environment, and other stressors may contribute to increased susceptibility to infection. Semen and reproductive tracts from infected bulls also have yielded *M. paratuberculosis,* but semen rarely appears to be a source of infection, although of course infected bulls will potentially shed the organism in their feces.

Pathogenesis and Progression

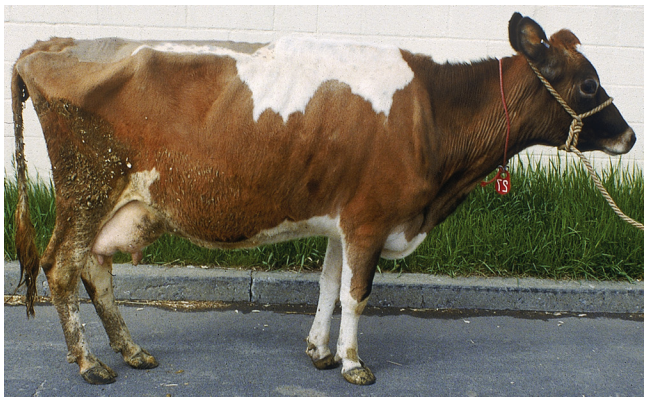
After oral ingestion, MAP organisms invade intestinal epithelial cells, most notably through specialized cells within ileal Peyer patches (M cells), which deliver the organisms to submucosal macrophages. The subsequent course of infection is determined by numerous factors, including the dose of MAP organisms ingested and the animal's individual susceptibility to MAP, which in turn is determined by the age of the animal and the ability of its innate and adaptive immune responses to control MAP. Resistance to MAP infection is estimated to have approximately 10% heritability. Thus, in some exposed animals, MAP organisms will be contained or eliminated by macrophages, and infection will not become established, but in others, the infection will ultimately progress. In the early stages of infection, macrophages are activated by interferon-γ produced by Th1 helper lymphocytes and limit proliferation of the MAP and thereby its spread to other sites. However, the immune response to control MAP results in "collateral damage," inciting a granulomatous response within the intestinal mucosa. This is characterized by progressive infiltration by epithelioid cells, multinucleate giant cells, and lymphocytes surrounding MAP-laden macrophages. In this early stage of infection ("eclipse phase"), during which MAP organisms are slowly proliferating and the inflammatory process is slowly progressing, no clinical signs are observed. The animals are outwardly healthy, fecal shedding of MAP rarely occurs, and serum antibodies are not produced. This makes it very difficult to diagnose the infection at this stage.

Gradually, the ability of the cow to contain the infection wanes, as a Th2 type immune response begins to predominate. Cows remain asymptomatic, but fecal shedding at low levels will begin, and detectable serum antibodies will be produced soon after fecal shedding. Milk production and reproductive performance are not generally affected in these asymptomatic, low-shedding cattle, which can thus serve as a source of MAP for environmental contamination.

Ultimately, spread of the infection accelerates. Fecal shedding at high levels begins, even though there are no clinical signs, and MAP organisms may spread systemically to the mammary gland and fetus. Studies have indicated a 16% reduction in milk production in later stages of asymptomatic infection (i.e., the lactation immediately before culling). Finally, the granulomatous infiltration of the bowel progresses to a severity that results in malabsorption and protein-losing enteropathy, and the cow begins to show the characteristic clinical signs associated with Johne's disease. The entire length of the incubation period before outward signs of illness can be long, often requiring at least 2 to 3 years but up to 10 years in some cases from time of initial infection.

Several points deserve emphasis.

1. Most infected cattle never develop clinical signs before being culled from the herd. Factors that contribute to clinical disease versus asymptomatic infection are not known but probably include organism dose, age at infection, nutrition, concurrent diseases, stresses, and genetics. Infected cattle shed MAP in their manure and transmit the disease to herdmates by MAP contamination of the environment. Herd infection prevalence varies from 20% to 100% in heavily infected herds. Despite this rather high incidence of infection, it is unusual to see clinical signs in more than 5% to 10% of adult cows in the herd per year. Johne's disease has been shown to persist in some herds for more than 10 years with no overt clinical signs of infection.
2. It is widely accepted that cattle that develop clinical signs shed large numbers of organisms and represent the greatest threat to contaminate the environment. Super-shedders may shed MAP in higher concentrations (1–5 million CFU/g of manure) than cattle with clinical disease. Potentially, these animals represent the greatest source of environmental contamination and reservoir for possible transmission to herdmates. Most super-shedders are asymptomatic with no evidence of diarrhea or weight loss yet excrete huge numbers of MAP organisms into the environment.
3. Passive shedding of MAP may occur when noninfected cattle ingest manure contaminated forage or water. With a super-shedder in the herd, ingestion of as little



• **Fig. 6.63** Jersey cow affected with Johne's disease. Poor condition, a dry hair coat, and fecal staining of the hind quarters and tail are apparent.

as 5 mL of manure contamination in forage may result in passive shedding and give rise to a positive fecal culture for a previously uninfected cow. The risk of misclassification of such cattle must be considered when control programs include fecal cultures of all adult animals and culling decisions are based on the results. It is possible that this phenomenon may represent 50% of all culture-positive cattle in the herd when a supershedder is present.

Clinical Signs

The hallmark clinical sign of Johne's Disease is watery, projectile diarrhea. It has been described as pea soup in consistency and often forms bubbles because of the rather liquid consistency. In advanced cases, the diarrhea stains the tail, perineum, and hind limbs. It will stain the rear quarters if the tail switches liquid feces onto the quarters, flanks, and gluteal region. There is typically no blood or mucus present, and the animal is afebrile and does not exhibit tenesmus. However, given today's laxative diets, the diarrhea observed in a patient with Johne's disease is best described as looser compared with herdmates. The animal maintains a good appetite and remains well hydrated but has a significant decline in milk production and rapid loss of body condition. As well as temperature, other vital signs are normal. Moderate to advanced clinical cases have obvious weight loss characterized by muscle wasting, a poor dry hair coat, significant production losses, dehydration, and reduced feed intake, particularly high-energy feedstuffs (Fig. 6.63). Protein-losing enteropathy leads to hypoproteinemia with submandibular and brisket edema. Ventral edema is apparent but may vary in the anatomic area involved. Intermandibular, brisket, ventral, udder, and lower limb edema all are possible (Fig. 6.64). Clinical laboratory tests reveal hypoalbuminemia and possibly anemia of chronic disease. Although most cattle infected with MAP remain asymptomatic, cattle with clinical signs signal the diagnosis and alert both veterinarian and herd owner to the possibility of a herdwide problem.

• **Fig. 6.64** Four-year-old Holstein cow with submandibular edema (bottle jaw) and weight loss caused by Johne's disease. Diarrhea was minimal. The diagnosis was confirmed by right flank laparotomy and ileal lymph node biopsy.

• **Fig. 6.65** A pair of 18-month-old Holstein heifers with advanced Johne's disease. These heifers were representative of an age-grouped epidemic involving 12- to 24-month-old heifers on a single farm. This would imply extremely heavy environmental contamination with *Mycobacterium avium* subspecies *paratuberculosis*.

Despite loose manure, loss of body condition, and diminished milk production, cows with Johne's disease do not appear seriously ill until the terminal stages when finally the appetite is markedly reduced. Occasionally, cattle have diarrhea intermittently rather than continually, but this is unusual. We have also observed cows with Johne's disease with obvious diarrhea that spontaneously reverted to apparently normal manure after shipment to our hospital for diagnosis. Whether stress associated with shipment or a change in diet is responsible for this temporary improvement in fecal consistency is unknown. However, if the animal is not culled, the disease will progress to the point that the animal becomes inappetent, weak, and cachectic, and ultimately death will follow.

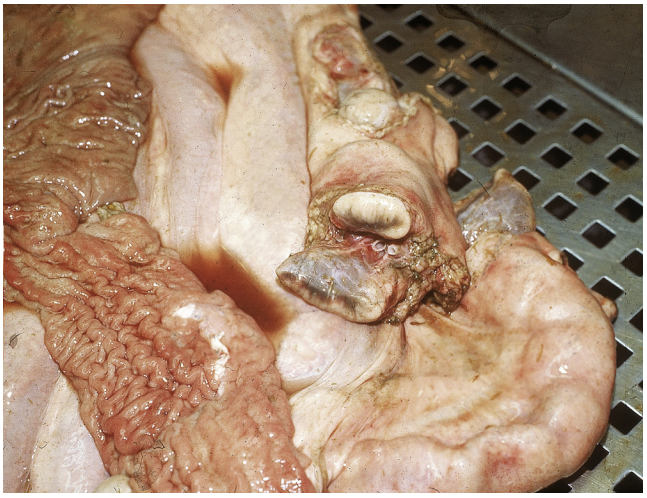
Clinical signs develop only after a prolonged incubation period and usually appear between 2 and 5 years of age. However, signs have been observed in heifers younger than 12 months of age and in mature cows up to 8 to 10 years of age (Fig. 6.65). If several 2-year-old heifers in a herd develop clinical signs of diarrhea, it suggests a rather heavy dose of MAP at an early age, but clinical signs in 5- to 7-year-old

cows suggest a much lower dose of MAP or older age at the time of exposure. Thus, the age of onset of clinical signs will assist an astute clinician as to the severity of the herd problem. Age of onset is probably affected by many factors, such as dose and duration of exposure to infectious organisms, nutrition, genetics, concurrent diseases or stresses, and other factors. The clinical impression that signs frequently develop after the onset of lactation in the first, second, or third lactations suggests lactational stress may be sufficient to amplify subclinical signs and hypoproteinemia to a clinical state. It also is possible that this observation is simply a reflection of closer monitoring of appetite, production, and body condition in lactating animals as opposed to heifers or dry cows. Lactation stress is not a prerequisite to the development of clinical signs, as proven by bulls and steers having clinical Johne's disease. Interestingly, some severely affected bulls and steers with Johne's disease have developed abomasal displacements during the advanced stages of disease.

Many cattle with signs of Johne's disease are culled because of poor production before the diagnosis is confirmed or suspected. This is especially common in free-stall operations in which an individual cow's manure consistency may not be as obvious as it would be in conventional housing and individual stalls. Dairy cattle with confirmed MAP infection have been shown to have higher cull rates than uninfected herdmates because of weight loss and reduced milk yield when clinical disease is evident and decreased production and mastitis when the infection is still, as yet, subclinical. Subclinical infections have also been associated with infertility. Increased mastitis and reproductive failure may be partially explained by hypoproteinemia, negative energy and protein balance, stress, and poor condition. A recent study on two large Minnesota dairy farms identified a threefold increase in the relative risk for culling in MAP fecal culture–positive cattle compared with fecal culture–negative cows. This study is noteworthy in that these herds were typical upper midwestern free stall dairies with an approximately 8% prevalence for MAP fecal culture positive animals. The calculated costs for lost production alongside diminished cull value associated with being fecal culture positive in this study were substantial, in fact greater than $400 per cow.

Diagnosis

Cattle with advanced signs of Johne's disease are easily suspected of having the disease because of diarrhea, hypoproteinemia, production loss, weight loss, and overall deterioration of condition. The only abnormalities detected routinely in serum biochemistry are hypoalbuminemia, hypoproteinemia, and occasionally hyperphosphatemia (>7 mg/dl). Clinical Johne's disease must be differentiated from chronic gastrointestinal parasitism, chronic salmonellosis, toxicities, intestinal neoplasia, copper deficiency, heart failure, glomerulonephropathies, renal amyloidosis, eosinophilic enteritis, and chronic BVDV infections. Postmortem examination of clinically advanced cases reveals grossly visible thickening and increased corrugation of the ileal mucosa (Fig. 6.66), and enlargement of the ileocecal lymph nodes. Histopathology confirms a granulomatous enterocolitis with macrophages and epithelioid cells in the submucosa and lower mucosa. Ziehl-Neelsen staining confirms the presence of MAP in the intestine and lymphatics. However, culture of these same tissues has a much greater sensitivity to detect MAP than does histopathology. Lesions may be present in the cecum and colon of advanced clinical cases and can extend orad from the ileum to more proximal regions of the small intestine. Although MAP may be isolated from other organs such as the liver, uterus, or fetus in some advanced cases, gross lesions consisting of granuloma formation are rare in these organs, and truly disseminated infections having gross lesions are very rare. However, disseminated infections as detected by culture of MAP from lymphatic fluid and lymph nodes such as the prescapular, prefemoral, supramammary, or popliteal lymph nodes do occur in cattle with clinical disease. Aortic calcification has been observed in advanced cases. Mild clinical cases may have a thickened edematous ileum with distended lymphatics on the serosal surface (Fig 6.67). Thickening of the mucosal surface and a raised corrugated appearance is typical. Lymphatic distension is obvious on the serosal surface of the ileum, and the ileocecal lymph nodes, as well as other mesenteric lymph nodes, are enlarged and edematous on cut sections.



• **Fig. 6.66** Necropsy view of thickened ileum and edematous (cut) ileocecal lymph node from a cow with advanced Johne's disease.

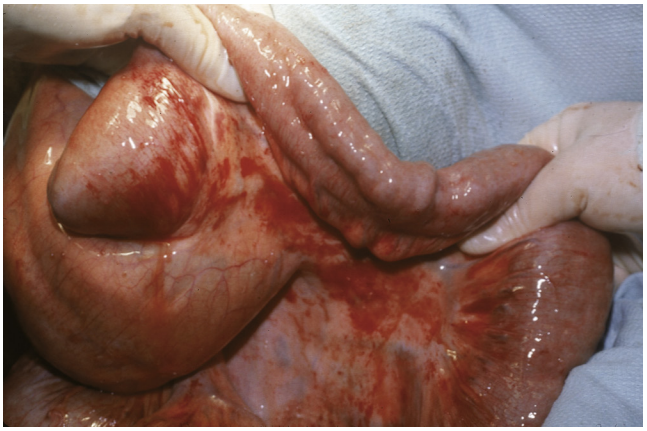
Available confirmatory antemortem diagnostic tests include those that directly detect the organism in feces or those that detect the animal's immune response to MAP infection. Organism detection methods include culture on solid media (Herrold's Egg Yolk Media [HEYM]), culture on liquid media, or detection by PCR. Organism detection tests have the advantage of detection earlier in the course of disease (compared with ELISA) in most cases because fecal shedding usually precedes the development of antibody production. These tests also have the ability to determine the quantity of MAP organisms being shed and thus are helpful for prioritizing animals for culling based on relative impact

on environmental contamination. Culture methods require 4 to 16 weeks for results, depending on the technique used.

Antibody tests used most frequently are serum ELISA or milk ELISA. The complement fixation test and agar gel immunodiffusion tests are inferior in performance, but some nations may still list these tests as being required before importation. The ELISAs have a lower sensitivity to detect asymptomatic fecal shedders (15%) but are often used because they are considerably less expensive and for heavy shedding or clinically affected animals the sensitivity is higher (90%–100% for clinically affected animals).

When presented with an adult animal for which clinical Johne's disease is considered likely, the presence of projectile watery diarrhea, lack of fever, and low plasma protein should raise the index of suspicion, especially if the animal originates from a herd known to harbor the infection. In this situation, a diagnostic test with a rapid turnaround time is essential to aid in clinical decision making. Either ELISA or fecal PCR perform well for this purpose because both have high sensitivity and specificity in evaluation of clinically affected cattle. In contrast, when screening asymptomatic cows in infected herds to assist with culling decisions or biosecurity issues such as maternity pen and colostrum management, organism detection tests are more sensitive than ELISA but may be cost prohibitive and may be positive in passively shedding, noninfected cattle. A cost effective approach is to use milk ELISA, using not just the dichotomous results (i.e., "positive" or "negative") but using the quantitative result (optical density (OD) value or sample to positive (S/P) ratio), recognizing that the strongest reactors are most likely to be heavy fecal shedders and pose the greatest risk for contaminating the environment with MAP. Unfortunately, neither organism detection tests nor antibody tests are useful in screening asymptomatic young cattle (<3 years of age) as herd replacements or for use as embryo transfer recipients because cattle at this age are usually in the "eclipse" phase of infection and will be test negative even if infected. Surgical biopsy of the ileum and ileocecal lymph node could detect MAP infection in these animals but is generally not practical for this screening purpose. The most effective way to prevent introduction of MAP is to purchase animals from herds known to be free of MAP, such as herds that have achieved status 4 level or above in the Voluntary Johne's Disease Control Program.

The gold standard for diagnosis has long been considered culture of ileum, ileocecal lymph nodes, or other mesenteric lymph nodes for cattle with clinical Johne's disease. This technique has been used to identify Johne's disease–infected cattle at slaughterhouses and to gather epidemiologic data regarding prevalence of the disease. Although harvesting ileocecal lymph nodes constitutes an invasive procedure for clinical patients, extremely valuable or individually purchased cows suspected of having Johne's disease may warrant this invasive technique to diagnose the condition definitively, especially when the herd has not been known to have Johne's disease in the past. A right flank exploratory laparotomy is performed to harvest a full-thickness 1.0-cm wedge of ileum and an ileocecal lymph node. The ileal biopsy and half of the lymph node are submitted for culture and histopathology, including a Ziehl-Neelsen stain. The remaining half of the lymph node is used for impression smears that are stained for acid-fast organisms. An absolute diagnosis usually is possible from the impression smears, but if this fails or is questionable, the histopathology generally confirms or denies the diagnosis without the prolonged delay associated with cultures.



• **Fig. 6.67** Thickened, edematous ileum and visibly distended lymphatics on the serosal surface in a cow showing early signs of Johne's disease. A rapid definitive diagnosis was established by biopsy of the ileum and ileocecal lymph node.

Treatment

Cattle with clinical Johne's disease are rarely treated. Occasionally, valuable animals may be treated to allow for salvage of genetic material by embryo transfer or to prolong the life of "pet" cattle. Treatment should not be undertaken to salvage the fetus of a pregnant cow with Johne's disease because in utero infection is likely. Uninfected embryos can be obtained from infected cows, via conventional embryo transfer provided proper embryo washing steps are followed, or after in vitro fertilization subsequent to oocyte pickup.

Owners that wish to undertake treatment should be advised that daily, lifelong treatment is required; treated animals may continue to shed MAP in their feces even if clinical signs are reversed; and relapse will ensue when treatment is stopped. Treatment involves extra-label use of antimicrobial medications for an extended period, which in the United States must comply with regulations set forth in the AMDUCA. Thus, owners should be advised that the treated animal or its products must not enter the human food supply chain.

Monensin is an ionophore antibiotic shown to have activity against MAP and have a beneficial effect in infected cattle. Monensin, approved in the United States as a feed additive, may not be used in an extra-label fashion according to the dictates of AMDUCA. However, if the drug can be legally prescribed for its other indications (coccidiostat, rumen digestion efficiency), it might be used. Clofazimine is an antileprosy drug that has been successfully used to treat Johne's disease at a dose of 2 mg/kg, orally every 24 hours, alone or in combination with other antimicrobials. Resolution of

diarrhea and improvement in plasma total protein can be observed within 2 weeks of starting treatment, although fecal shedding may continue. The drug is no longer available from commercial sources in the United States, so alternative antimycobacterial medications such as isoniazid (10–20 mg/kg orally every 24 hours) combined with rifampin (10 mg/kg orally every 24 hours) are often used. Rifampin usage in the United States currently requires a commitment from the producer that neither milk nor meat from that individual will ever be sold for human consumption, further diminishing the likelihood of dairy cattle ever being treated for this condition.

Control

Herds that are free of MAP can best prevent introduction of the disease by maintaining a closed herd. Purchase of replacement cattle from herds of unknown status poses a great risk for introducing asymptomatic but MAP-infected animals. If herd additions must be purchased, they should be purchased from herds participating in the Voluntary Johne's Disease Control Program that have achieved status level 4 or higher. If this cannot be done, then depending on the age of replacement animals, prepurchase testing for MAP infection may not be a foolproof method of screening new additions (see earlier).

Control of the disease in infected herds will depend in part on the objectives of the herd. Large production herds with no interest in selling replacement cattle may perceive little financial incentive to spend money on controlling the disease because they believe that culling for other reasons will often occur before MAP infection affects production. At the opposite end of the spectrum, eradication may be the goal for a herd from which genetic seed stock will be sold, requiring an aggressive, prolonged, and potentially impressive strategy, including test-and-cull strategies using organism detection tests, with management changes to prevent transmission. Although difficult, eradication of the infection from a herd will require intensive and repeated use of fecal cultures on all animals older than 24 months of age for many years.

Calves should be born in disinfected, cleaned maternity areas and removed from the dam immediately. Calves should be raised completely separately—preferably on a separate farm—from the adult cattle. All calves should be fed colostrum from ELISA-negative, or better, fecal culture–negative cows. Pooled colostrum feeding to calves should be avoided in herds with infected cattle. Colostral replacements could be used as a substitute for colostrum in herds with a high MAP incidence, but their use as a replacement for colostrum may lead to an increased incidence of other diseases in calves such as septicemia or diarrhea. Colostrum that is properly heat treated (60 min at 60°C and with constant stirring) will reduce the number of MAP but might not eliminate the organism completely from the colostrum. If whole milk is fed to calves in known infected herds, pasteurization should be considered. The purchase of replacement animals from herds of unknown Johne's disease status continues to represent the greatest risk to introduce or reintroduce MAP to such herds. Minimizing fecal contamination of feedstuffs, water, pastures, and exposure of calves to adult cow feces are essential and must be evaluated on an individual herd basis. Equipment used for manure removal or that could be contaminated by manure must remain separate from feeding implements and the calf environment. Although these principles for eradication are seemingly straightforward, they may not be practical or affordable in many instances. In between these two extremes, production herds that wish to "contain" the disease, to prevent the prevalence of MAP infection from increasing on the farm, and to reduce the risk of losing animals or milk production to clinical Johne's disease may use an intermediate approach. This may include identifying heavily infected animals through interpretation of milk or serum ELISA alongside management steps to reduce exposure of calves to MAP from feces of adult cows that are potentially shedding the organism. Additionally, these same herds may choose to sell heavy shedders (based on ELISA serology results) or clinical cases immediately, and moderate shedders would be culled at the end of their current lactation.

In the United States, the only licensed vaccine (Mycopar, Boehringer Ingelheim Vetmedica, St. Joseph, MO) is a suspension of heat-killed organisms in an oil adjuvant. Vaccination is shown to prevent or reduce losses from the development of clinical Johne's disease in vaccinated animals. However, it does not prevent infection with MAP, and vaccinated cattle can become infected and shed MAP in feces, although importantly the vaccine does prevent clinical disease in almost all recipients. Any protection from vaccination may be overcome by failure to implement other management changes to reduce exposure to the organism. Vaccination is normally only used in herds experiencing significant losses caused by Johne's disease and should be combined with management changes designed to reduce the spread of infection between cattle. Permission from the state veterinarian is required, and a complete herd tuberculosis (TB) test must be performed before implementation. The vaccine is administered SC in the brisket and frequently predisposes to a local abscess over the next few months or years. Vaccination can result in false-positives on the caudal tail fold test for TB and will also result in positive serology for paratuberculosis. Accidental self-injection of the vaccine can result in a severe painful granulomatous inflammatory response at the site of injection.

Foreign Animal Diseases

Mucosal diseases such as BVDV, BTV, EHDV, MCF, and VSV require differentiation from foreign animal diseases that threaten livestock in the United States (Table 6.9). Extreme vigilance is necessary to prevent entrance of these diseases to this country, and consultation with regulatory state or federal veterinarians is imperative whenever confusion exists. Because a great deal of overlap is possible for the clinical signs present in domestic and foreign mucosal diseases, positive diagnosis including appropriate serologic and virologic confirmation is essential. A description of rinderpest is included in the table for completeness although the World Health Organization declared that it had been completely eradicated globally in 2011.

TABLE 6.9 Foreign or Exotic Animal Diseases Affecting the Gastrointestinal Tract

Disease	Cause	Clinical Signs	Major Differential Diagnosis	Diagnosis	Reference
Foot-and-mouth disease (Aftosa)	FMDV = genus *Aphthovirus*, family Picornaviridae	Fever, salivation, lip smacking, lameness, teat lesions	Vesicular stomatitis	Call regulatory veterinarians	Kahrs (1981)
(Aphthous fever)	Seven distinct serotypes with multiple subtypes	Vesicles progressing to erosions and ulcers of oral mucosa, nasal mucosa, interdigital space, coronary band, teats Abortion	BVDV Bluetongue MCF Rinderpest	Fluid from vesicles Oropharyngeal fluid Tissues Paired sera	Sutmoller (1992)
Rinderpest (cattle plague) *(Peste bovine)*	RV=genus *Morbillivirus*, family Paramyxoviridae	Peracute—high fever, death	BVDV	Call regulatory veterinarians	Kahrs (1981)
	One major serotype with field strains possessing variable pathogenicity	Classic fever, mucous membrane congestion, necrosis, and subsequent erosion	MCF Vesicular stomatitis Foot-and-mouth disease	Samples best obtained from febrile animals with mucosal lesions (early cases)	
		Mucous membrane lesions cause salivation, ocular discharge	Salmonellosis Bluetongue	Serologic testing Viral isolation	Seek and Cook (1992)
		Severe hemorrhagic diarrhea and tenesmus start several days after mucosal lesions	Arsenic poisoning		
		Dehydration, death			
		Subacute or atypical; lower mortality, greater difficulty in distinguishing from differential diagnosis			

BVDV, bovine viral diarrhea virus; *FMDV,* foot and mouth disease virus; *MCF,* malignant catarrhal fever

Liver Abscess

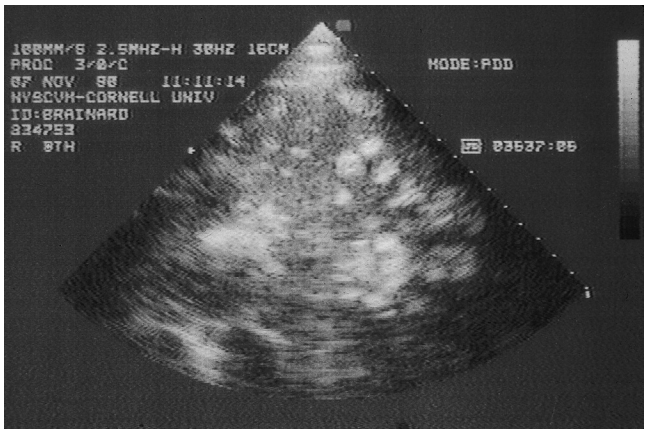
Etiology

Abscesses of the liver occur in all ages of cattle. In calves, liver abscesses are often the result of omphalophlebitis; in older cattle, they most often are secondary to reticulorumenitis. In feedlot cattle, it is well recognized that the change from pasture to a high-concentrate ration causes a rapid increase in rumen fermentation and organic acid production, which may result in erosion and inflammation of the rumen epithelium. Metastasis of bacteria from the inflamed and necrotic rumen wall to the liver occurs via the portal vein. In dairy cattle, similar failure of adaptation of rumen fermentation may occur at the onset of lactation when there is an abrupt increase in the energy content of the diet. Liver abscesses are therefore common potential sequelae to acute or subacute rumen acidosis; indeed, their presence in several cows within a herd should raise concern about nutritional management and the likelihood of subacute rumen acidosis as a herd problem. Liver abscesses also may occur as a result of traumatic reticulitis, where they tend to be an individual cow problem. The most common organisms isolated from hepatic abscesses are *Fusobacterium necrophorum, Bacteroides nodosus,* and *Trueperella pyogenes.* Streptococci and staphylococci also may be isolated from mixed cultures.

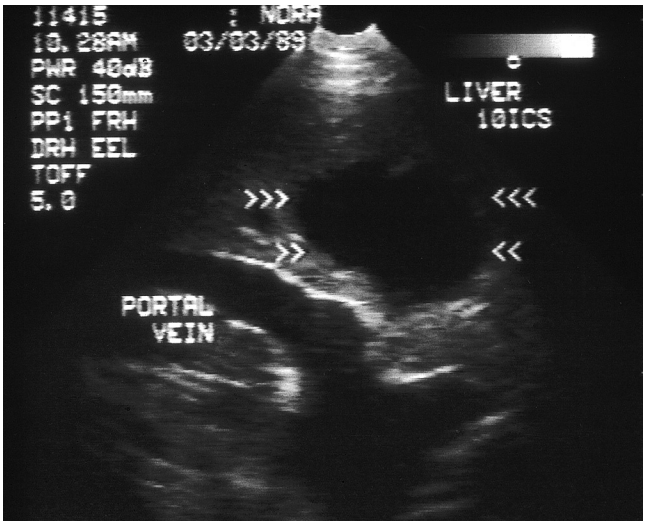
Clinical Signs

Local, circumscribed liver abscesses are characteristically silent clinically and are not associated with systemic abnormalities or with hepatic dysfunction. Such abscesses are found incidentally during the postmortem examination of slaughtered cattle or during abdominal ultrasound examination but are often only of importance economically because of the condemnation of affected livers. As mentioned earlier, the repeated identification of such abscesses either postmortem or by ultrasonography should serve as a sentinel warning regarding nutritional management.

Liver abscesses, when located adjacent to the vena cava, may distort the vessel wall and cause phlebitis and



• **Fig. 6.68** Transabdominal sonogram of the liver in a mature cow with multiple hyperechoic abscesses. The hyperechoic appearance suggests dense purulent exudate, decreasing the chances of successful treatment.



• **Fig. 6.69** Transabdominal sonogram of the liver in a 3-year-old Holstein cow with weight loss and diminished production. A single large hypoechoic abscess can be seen, and the cow recovered after 1 month of systemically administered penicillin treatment.

thrombosis. Septic thromboembolism from the vena cava may cause a respiratory syndrome, referred to as caudal vena caval thrombosis syndrome, characterized by cough, dyspnea, or pulmonary hemorrhage that is described in Chapter 4. In a postmortem series of 6337 slaughtered cattle, liver abscesses were found in 368 (5.8%), and of these, 24% were located in the craniodorsal aspect of the liver with the potential for causing vena caval thrombosis.

Liver abscesses may be associated with constitutional abnormalities that include fever, anorexia, weight loss, and reduced milk production. Neutrophilic leukocytosis and significant increases in serum globulin and fibrinogen are characteristic. Hepatic derived enzymes may not be elevated in the serum of affected cattle, although GGT values may be increased with large abscesses. Growth of a liver abscess near the common bile duct may obstruct bile flow and may result in clinical signs and laboratory abnormalities associated with impeded flow of bile (see below). Liver abscessation also has been recognized as a cause of vagal indigestion.

Ultrasonographic examination of the liver is a valuable diagnostic procedure for determining the location of the abscess(es) and for evaluating prognosis and response to therapy (Figs. 6.68 and 6.69). The lesions may vary in diameter from a few centimeters to more than 20 cm. Characteristically, they may be visualized in three or four adjacent intercostal spaces, and needle aspiration may not be necessary for diagnosis. On rare occasion a large liver abscess can be seen on radiographs displacing the diaphragm (Fig. 6.70).

Treatment

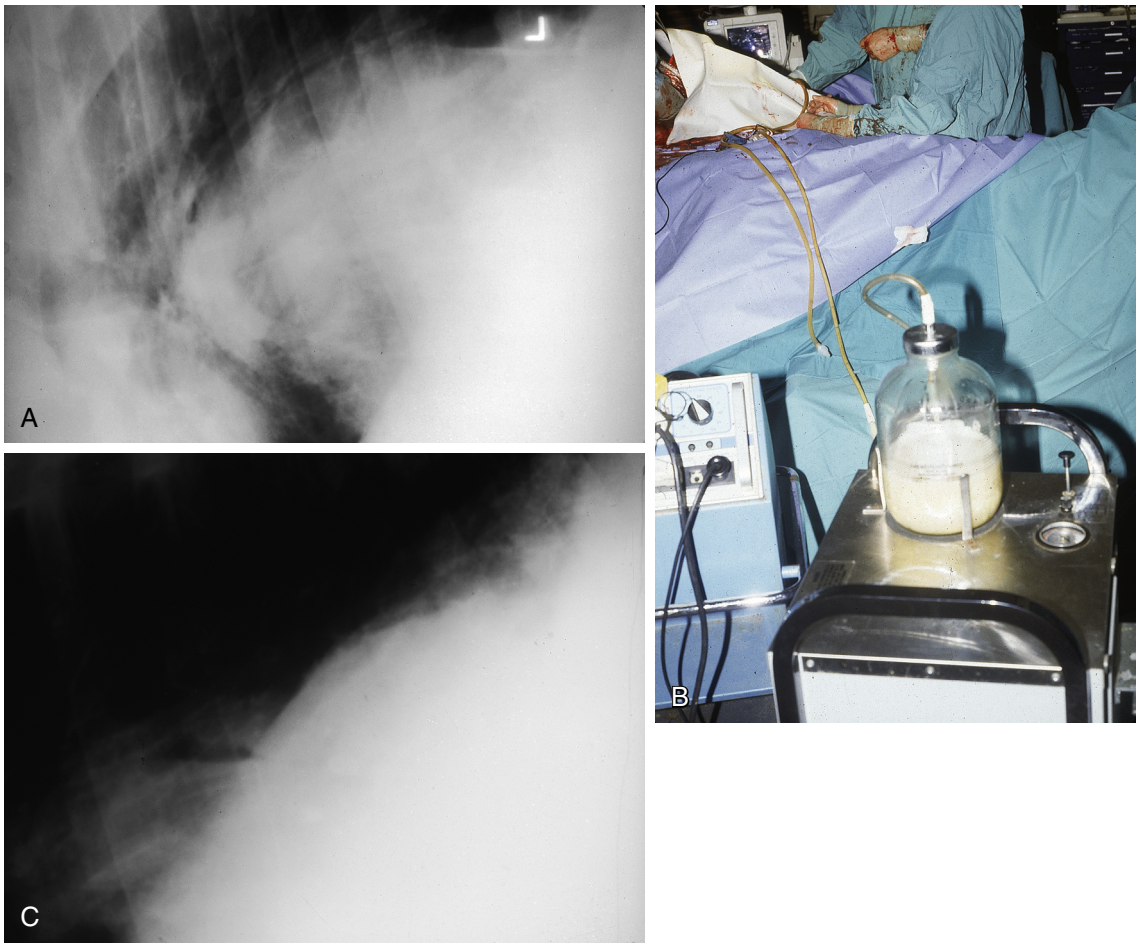
When liver abscesses are recognized clinically and their location identified, it is possible to consider antibiotic therapy, surgical drainage, or both. The decision regarding drainage of liver abscesses depends on the size, location, and the condition of the cow. Penicillin treatment can be successful in some cows with smaller, hypoechoic abscesses, but relapses often occur unless treatment is for 4 or more weeks. Even with surgical drainage, relapses may occur. The prognosis for treatment of liver abscesses that have caused clinical signs is guarded and is least favorable for large and hyperechoic abscesses. Successful surgical treatment of a liver abscess that caused vagal indigestion has been described.

Bile Duct Obstruction and Cholangitis

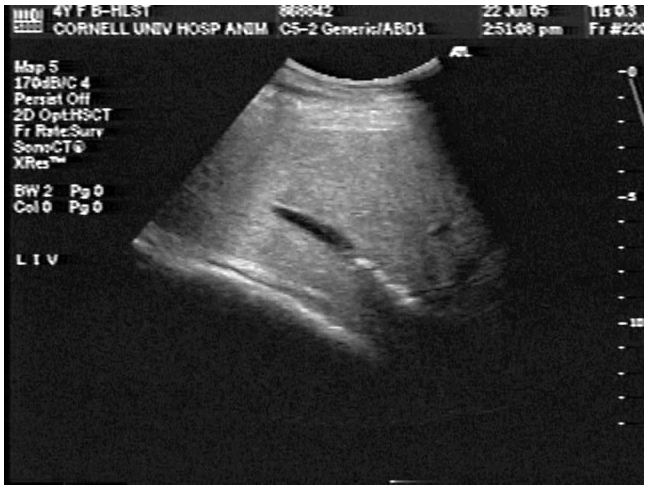
Intrahepatic cholestasis is observed in lactating cattle with severe fatty liver during the periparturient period and is described in Chapter 15. Intrahepatic cholestasis may also occur from "sludge" or stones within hepatic ducts (see Video Clip 6.2). Extrahepatic cholestasis is caused by obstruction of bile flow from choleliths within the common bile duct or by obstruction of flow in the common bile duct as a result of external mechanical pressure exerted on the common bile duct by liver abscesses, by extensive adhesions in the area of the cystic and common bile ducts, or by smaller inflammatory lesions of the common duct near the hilus or the duodenal papilla.

The characteristic sterility of the biliary tract is maintained by the continued production and flow of bile into the intestine. Partial or complete obstruction of bile flow predisposes to ascending infection of the biliary tract by intestinal microorganisms. Infection of the biliary tree causes cholangitis and may result in significant alterations in the physical characteristics of bile, including the accumulation of inspissated products of inflammation and of precipitated bile constituents (bile acids, cholesterol, and even stone formation) (Fig. 6.71), which further impedes the flow of bile.

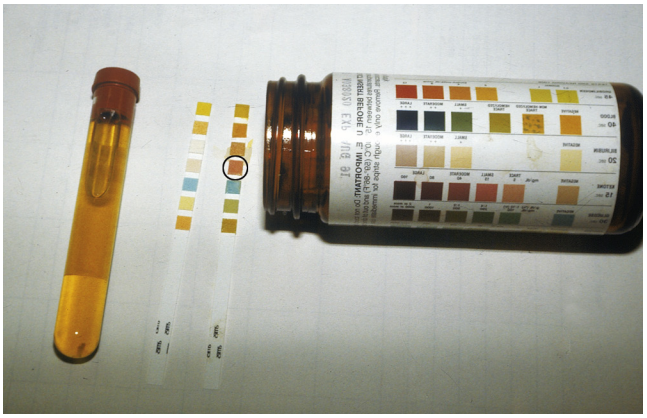
The clinical signs of extrahepatic bile duct obstruction and cholangitis include malaise, colic, fever, icterus with orange-colored urine (Fig. 6.72), and, in some cases photodermatitis (Fig 6.73) secondary to retention of phylloerythrin. Abnormal laboratory findings consist of leukocytosis, hyperfibrinogenemia, hyperbilirubinemia, bilirubinuria, and elevations in serum globulin and the activities of sorbitol dehydrogenase (SDH), aspartate aminotransferase (AST), alkaline phosphatase (AP), and gamma glutamyltransferase (GGT). Ultrasonographic



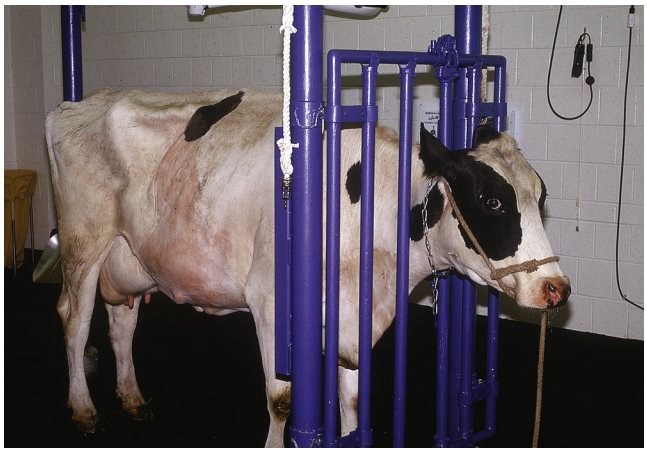
• **Fig. 6.70** **A,** Thoracic radiographs of a 14-month-old Holstein heifer showing a very large mass (liver abscess) displacing the diaphragm. **B,** The liver abscess was drained surgically. **C,** Radiographs repeated after drainage.



• **Fig. 6.71** Abdominal sonogram of an adult Holstein cow hospitalized because of anorexia and mild colic. The cow's γ-glutamyl transferase level was 1500 U/L. Stones are observed in the intrahepatic ducts. There was a marked clinical improvement within 3 days of initiating therapy with penicillin, intravenous fluids, flunixin meglumine, and forced feeding.



• **Fig. 6.72** A sample of urine collected from an adult cow with icteric membranes, fever, anorexia, depression, and hepatogenous photosensitization of the muzzle. The cow responded well to symptomatic treatment similar to the patient described in Fig. 6.71. The urine is orange and positive on multistrip examination for bilirubin. The *circled square* is a positive test result. There is an untested strip to the left.



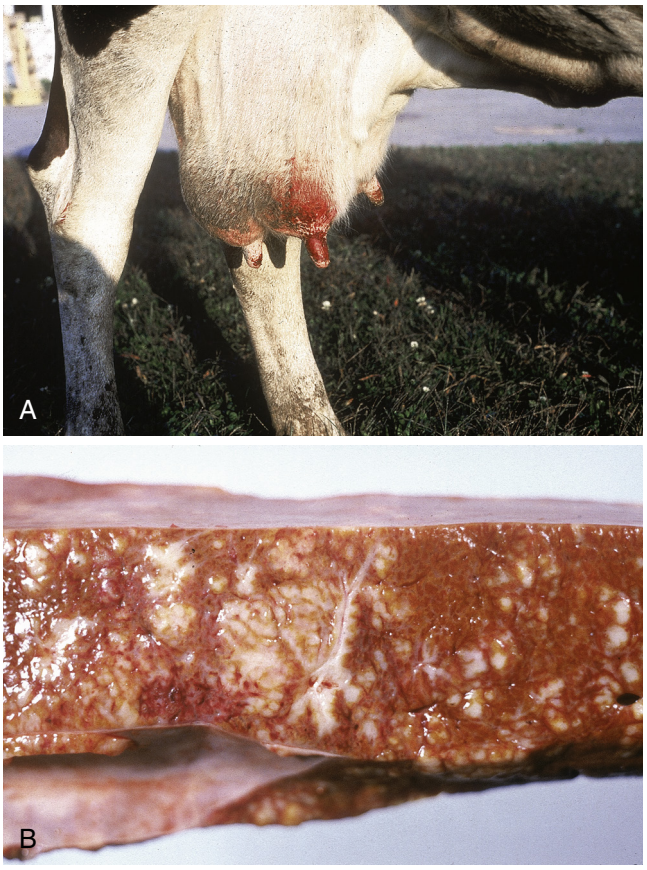
• **Fig. 6.73** The cow described in Fig. 6.72 with pronounced hyperemia (photosensitive dermatitis) of the muzzle.

findings in cows with extrahepatic cholestasis include severe dilatation of the gallbladder, the cystic and common duct, and other major intrahepatic bile ducts. Dilatation of the gallbladder is not specific because in all anorectic cows, the gallbladder may be distended. The diagnostic findings for extrahepatic cholestasis, however, are dilatation of the cystic and common bile ducts and of the major intrahepatic bile ducts (see Video Clip 6.3).

A case of cholelithiasis with cholestasis has been reported by Drs. Rebhun and Cable that was clinically similar to those in which bile duct obstruction is caused by external mechanical pressure on the common bile duct. A laparotomy was performed, and concretions 1 to 3 cm in diameter were palpated in the gallbladder. The choleliths in the gallbladder were crushed manually, and the material was massaged through the distended cystic and common ducts, into the duodenum, and on into the jejunum. After the procedure, there was significant improvement in clinical condition and in liver function test results, although the improvement was transient.

A syndrome of unknown etiology has been observed that is clinically similar to that of extrahepatic obstruction to the flow of bile but in which there is no ultrasonographic or laparotomy evidence of extrahepatic cholestasis. The clinical signs and laboratory test results are similar. When force fed for a few days and treated with penicillin for at least 1 month, there has been gradual improvement in clinical signs and laboratory abnormalities return to normal (see Figs 6.72 and 6.73).

Primary hepatic neoplasms are unusual in cattle but could cause obstruction of bile flow and should be considered in cows with both icterus and photosensitization. In a necropsy series of 66 primary bovine hepatic neoplasms, 40 were classified as hepatocellular carcinomas, 10 as hepatocellular adenomas, and 10 as cholangiocellular tumors. Less frequently observed primary tumors of the liver in this series included hemangiosarcoma, hemangioma, fibroma, and Schwannoma. In the postmortem examination of the livers of 24,169 slaughtered cattle, primary liver tumors of hepatocellular



• **Fig. 6.74** **A,** An 8-year-old Holstein cow with weight loss, inappetence, and photosensitization, which is best seen on the teats and udder. **B,** The cow had a cholangiocarcinoma.

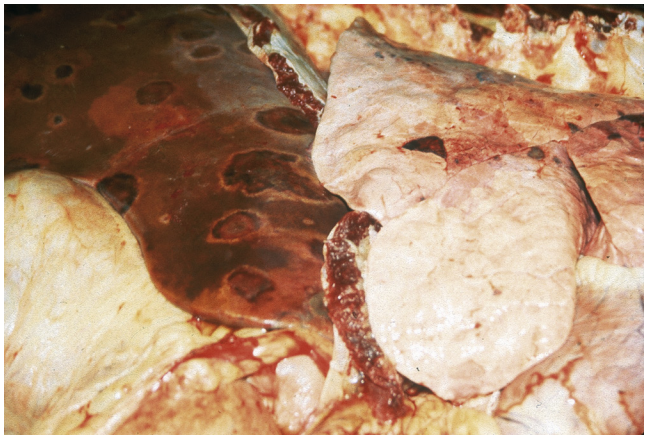
origin were identified in 22 (0.09%). In a third series of 1.3 million livers of cattle examined at slaughter, 36 had primary liver tumors of which 13 were classified as primary hepatocellular neoplasms and 21 as cholangiocarcinomas.

The clinical signs of cattle associated with primary hepatic neoplasms have not been extensively described. The expected clinical signs would be those associated with the growth of an expanding hepatic mass or with metastasis to the lung or to the spleen, both of which have been observed at necropsy. If the tumor obstructs bile flow, then icterus and photosensitization would be expected (Fig. 6.74). One of the authors (SP) has also seen hyperammonemic hepatic encephalopathy associated with an extensive cholangiocarcinoma and complete biliary obstruction in an aged cow. Dermatitis caused by photosensitization is frequently most severe on the teats and muzzle, although it may be more generalized (Fig. 6.75). Ultrasonography should be of value in locating, guiding biopsy, and otherwise assessing the location and prognosis of primary liver tumors.

Hepatic Insufficiency Associated with Sepsis

A syndrome of hepatic insufficiency has been described in lactating cattle after acute septic mastitis or metritis

• **Fig. 6.75** Hepatogenous photosensitization caused by a suspected hepatotoxin.



• **Fig. 6.76** The liver from a 6-year-old Holstein that had intestinal (forestomach and abomasum) and hepatic aspergillosis secondary to generalized sepsis and treatment with broad-spectrum antibiotics.

in which the initial clinical signs were compatible with endotoxemia. Subsequent clinical signs included anorexia; weight loss; reduced milk production; and, in one case, photodermatitis. In addition to increased serum activities of liver enzymes, the cows had remarkable delays in the sulfobromophthalein (BSP) plasma clearance test. Liver biopsies showed hepatocellular vacuolization or necrosis that was attributed to the effects of endotoxemia associated with acute systemic infection. Similar hepatic injury has been reported in humans after endotoxic shock. Five such cases were treated by force feeding and with other symptomatic support. Three of the cows responded satisfactorily to therapy, one failed to respond, and the fifth cow was lost to follow-up evaluation. Based on these observations, it is important to consider the possibility of hepatic injury in the initial management of cows with postpartum sepsis and in their longer term management when there is a sluggish response to therapy of the acute disease. If there is a history of prolonged antimicrobial therapy, intestinal and hepatic mycosis must also be considered (Fig. 6.76). Cows with persistent fever, complete anorexia and elevation of hepatic origin enzymes in the serum after prolonged antimicrobial treatment for diseases such as mastitis or rumenitis may have intestinal and hepatic mycosis. This is usually a fatal disease.

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