Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

13 Viral Diseases

Sherif R. Zaki • Christopher D. Paddock

INTRODUCTION

Influenza viruses remain the most frequently identified causes of viral infection in the lung. Nonetheless, the diversity of viral agents that cause pulmonary disease is extremely broad, and continues to expand (Table 13-1). Several newly recognized viral pathogens have been identified in the past two decades that are among the most feared and lethal of all emerging infections, including those caused by Hantaviruses, Nipah virus, and SARS coronavirus. Conversely, certain viral infections, particularly those that occur in vulnerable patient cohorts, have diminished during this same interval. For example, the U.S. incidence of varicella pneumonia has declined more than 65% since universal childhood vaccination for varicella was implemented in 1995, and advances in the clinical management of transplant recipients have reduced the incidence of cytomegalovirus pneumonia.

ADENOVIRUSES

Adenoviruses are represented by a ubiquitous and diverse group of at least 51 serotypes found naturally in the upper respiratory tracts and gastrointestinal systems of humans, other mammals, and birds. More than 50% of the known adenovirus serotypes are associated with human diseases. The others are rarely encountered and may or may not cause recognizable disease.

Clinical Features

It is estimated that approximately 5-10% of all pneumonias in infants and young children are caused by adenoviruses. Most pediatric cases of adenovirus pneumonia occur between 6 months and 5 years of age, and serotypes 3, 7, and 21 are the most common causes of pneumonia in this patient cohort. Serotypes 3 and 7 are particularly pathogenic adenoviruses that can cause disseminated and often fatal disease in previously healthy children. In adults, pneumonia is generally associated with serotypes 3, 4, and 7. Periodic epidemics of adenovirus pneumonia in young adults have been identified, particularly among military recruits. In a manner similar to other pathogens, adenoviruses take advantage of impaired or destroyed immune systems to establish persistent and disseminated infections in immunocompromised hosts. Immunocompromised patients are also susceptible to a broader range of different adenovirus serotypes. Because some adenoviruses establish latency in lymphoid tissues and the kidneys of their host, it is believed that many, possibly most, cases of clinical disease caused by adenoviruses in immunocompromised patients are reactivated infections.

Radiologic Features

Chest films typically show bilateral, multifocal, lobar, or segmental consolidations, bronchial wall thickening, hyperaeration, and lobar atelectasis. Pleural effusions and pneumatoceles are reported less frequently.

TABLE 13-1
Viral Infections in the Lung

Family/Agents	Virus
Adenoviridae	Adenovirus
Bunyaviridae	Hantavirus
Coronaviridae	SARS Coronavirus
Herpesviridae	Cytomegalovirus
	Herpes simplex
	Varicella zoster
Orthomyxoviridae	Influenza
Paramyxoviridae	Measles
	Parainfluenza
	Respiratory syncytial virus
	Human metapneumovirus
	Nipah
	Hendra
Viral hemorrhagic fevers	Arenaviruses, bunyaviruses, flaviviruses, and filoviruses

Pathologic Features

Gross Findings

The lungs of patients with adenovirus pneumonia are heavy and edematous, and the bronchi are filled with mucoid, fibrinous, or purulent exudates. The mucosae of the large airways are generally hemorrhagic and congested. Necrotic and inflammatory foci in the pulmonary parenchyma are often represented by yellow palpable nodules.

Microscopic Findings

The primary histopathologic findings include necrotizing bronchitis and bronchiolitis with extensively denuded epithelium, particularly in medium-sized (1-2 mm in diameter) intrapulmonary bronchi (Figure 13-1A). Affected airways may be occluded by homogeneous eosinophilic material, mixed inflammatory cells, detached epithelium, and cellular debris. The lamina propria of bronchi and bronchioles is typically congested and infiltrated by predominantly mononuclear inflammatory cell infiltrates. Bronchial serous and mucous glands are also often involved and show necrosis and mixed inflammatory infiltrates. As the infection progresses, there is involvement of the more distal pulmonary parenchyma, forming foci of bronchocentric necrosis with hemorrhage, neutrophilic and mononuclear cell infiltrates, and karyorrhexis. These findings occur against a background of diffuse alveolar damage. Adenoviruses form intranuclear inclusions in respiratory epithelial cells of the trachea, bronchi, and bronchioles, in the acinar cells of bronchial glands, and in alveolar pneumocytes, and are generally most abundant at the viable edges of necrotic foci. On hematoxylin-eosin stain, early inclusions appear as small, dense, amphophilic structures surrounded by a cleared zone and peripherally marginated chromatin, similar to herpetic inclusions. As the cellular infection progresses, the inclusion becomes larger (as large as 14 microns in some cells) and more basophilic, and the margins of the nuclear membrane become blurred to form the characteristic "smudge cell" (Figure 13-1B).

Ancillary Studies

Various methods can be used to diagnose adenovirus infections, including antigen detection (fluorescence antibody assays and enzyme immunoassays), cell culture, electron microscopy, molecular assays, and serologic testing for group-specific or type-specific antibodies. Immunohistochemical (IHC) staining methods can detect adenovirus-infected cells in formalin-fixed, paraffin-embedded tissues using various commercially available adenovirus group-specific antibodies (Figure 13-1C). Electron microscopy of adenovirus-infected tissues reveals a paracrystalline array of virions represented by icosahedral capsids that measure 70 to 90 nm in diameter (Figure 13-1D). Most adenoviruses can be isolated in cell culture from bronchial washings, tracheal aspirates, or lung biopsy specimens during the early stage of the illness. Molecular assays, particularly gene amplification using polymerase chain reaction (PCR) and in situ hybridization (ISH) methods, have been developed to detect adenovirus nucleic acid in respiratory secretions and in formalin-fixed, paraffin-embedded tissues.

Differential Diagnosis

The differential diagnosis includes those agents that cause necrotizing bronchiolitis, pneumonia, and intranuclear viral inclusions, particularly herpes simplex viruses, varicella-zoster virus, and cytomegalovirus. Histologic clues to distinguish these agents from adenovirus include the presence (herpes simplex virus [HSV] and varicella zoster virus [VZV]) or absence (adenovirus) of multinucleated cells, cytoplasmic inclusions (CMV), or distinctive smudge cells (adenovirus); however, ancillary studies are generally required for confirmation.

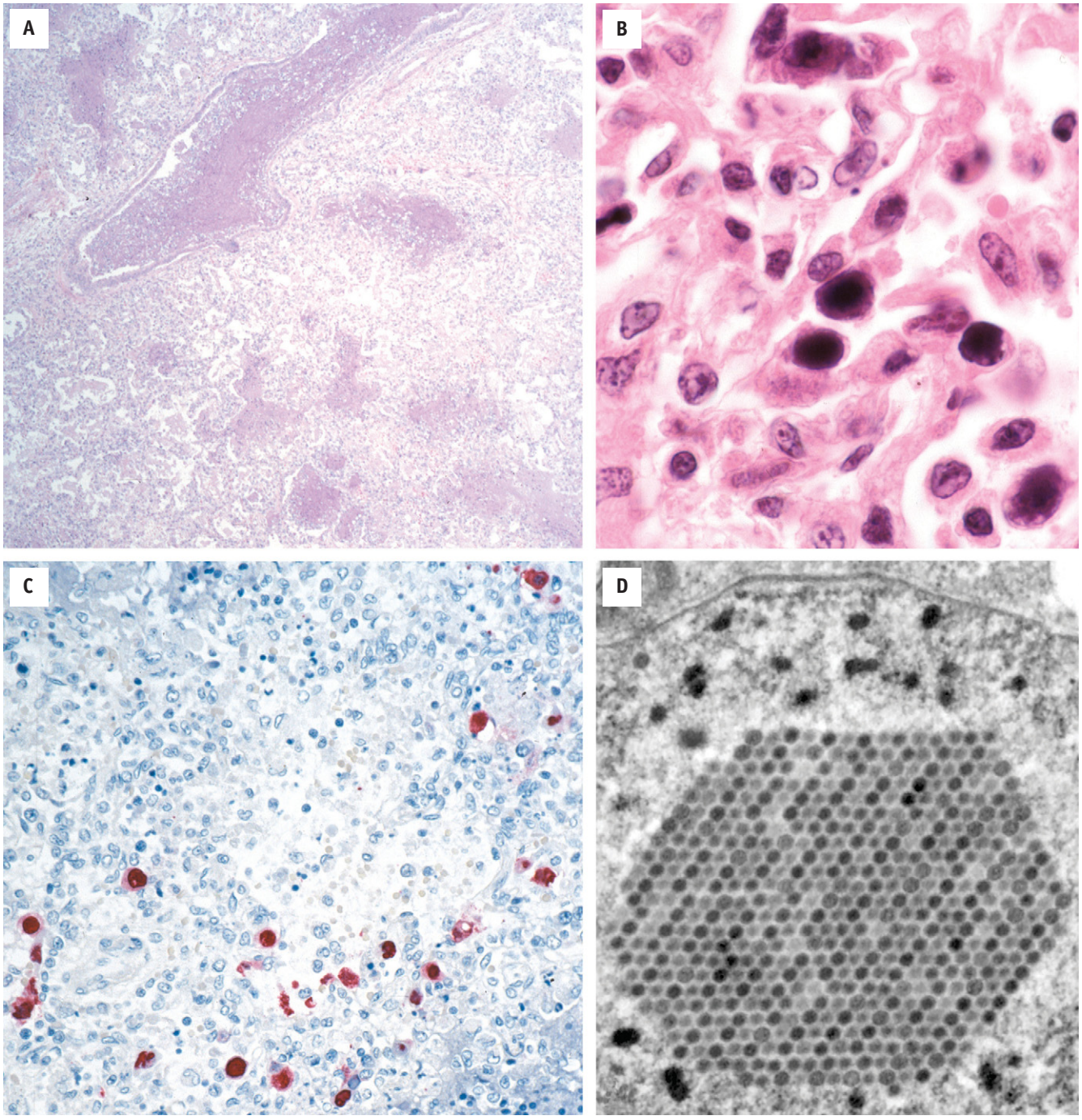

FIGURE 13-1

Adenovirus pneumonia

(A) Bronchioles and scattered alveoli containing necrotic debris comprised of surface epithelium, fibrin, and mixed inflammatory cells. **(B)** Large, basophilic, intranuclear inclusions in alveolar pneumocytes, forming characteristic "smudge cells" that can be observed in advanced adenovirus infections. **(C)** Immunohistochemical localization of adenovirus-infected cells in a patient with fatal adenovirus pneumonia (immunoalkaline phosphatase). **(D)** Paracrystalline arrays of 70-90 nm adenovirus particles in the nucleus of an infected pneumocyte.

Prognosis and Therapy

In immunocompromised patients, the case fatality rate of adenoviral pneumonia approaches 60%, compared with an approximately 15% mortality in immunocompetent patients. There is no proven effective antiviral therapy for adenovirus infections. Most patients receive only supportive care for symptoms of the disease, which includes cessation of immune-suppressing drugs in those patients with iatrogenic immunosuppression.

ADENOVIRUS PNEUMONIA—FACT SHEET

Definition

- Pulmonary infections caused by viruses in the family *Adenoviridae*

Incidence

- Approximately 5-10% of all pneumonias in infants and young children
- Periodic epidemics of adenovirus pneumonia in young adults occur, particularly among military recruits
- Immunocompromised adults, especially transplant recipients, are vulnerable to severe and sometimes fatal pneumonias caused by adenoviruses

Morbidity and Mortality

- In immunocompromised patients, the case fatality rate of adenoviral pneumonia approaches 60%, versus an approximately 15% mortality in immunocompetent patients
- Immunocompromised patients can develop disseminated disease
- Obliterative bronchiolitis is a potential cause of long-term morbidity in some survivors of the infection

Gender, Race, and Age Distribution

- Most pediatric cases of adenovirus pneumonia occur between 6 months and 5 years of age, and are caused by serotypes 3, 7, and 21
- Causes severe disease in neonates that may lead to long-term complications
- In adults, pneumonia is generally associated with serotypes 3, 4, and 7
- No recognized gender or racial predilection

Clinical Features

- Acute upper respiratory tract disease is manifested by tracheobronchitis
- Adenovirus pneumonia presents with signs and symptoms similar to other types of pneumonia, including fever, cough, and chest pain

Radiologic Features

- Bilateral, multifocal, lobar, or segmental consolidations, bronchial wall thickening, hyperaeration, and lobar atelectasis
- Pleural effusions and pneumatoceles are reported less frequently

Prognosis and Therapy

- No proven effective antiviral therapy
- Most patients receive only supportive care for symptoms of the disease
- Severe infections may progress to death in 2 to 3 weeks

ADENOVIRUS PNEUMONIA—PATHOLOGIC FEATURES

Gross Findings

- Lungs are typically heavy and edematous
- Bronchi are generally filled with mucoid, fibrinous, or purulent exudates, and have hemorrhagic and congested mucosae
- Necrotic and inflammatory foci in the pulmonary parenchyma may be represented by palpable yellow nodules

Microscopic Findings

- Necrotizing bronchitis and bronchiolitis with extensive denudation of the surface epithelium, particularly in medium-sized (1 to 2 mm in diameter) bronchi
- Affected airways may be occluded by homogeneous eosinophilic material, mixed inflammatory cells, detached epithelium, and cellular debris
- Bronchial serous and mucous glands are also often involved and show necrosis and mixed inflammatory infiltrates
- Bronchocentric parenchymal necrosis with hemorrhage, neutrophilic and mononuclear cell infiltrates, against a background of exudative diffuse alveolar damage
- Intranuclear inclusions in respiratory epithelial cells and alveolar pneumocytes, generally most abundant at the viable edges of necrotic foci
  - Early inclusions appear as small, dense, amphophilic structures surrounded by a cleared zone and peripherally marginated chromatin, similar to herpetic inclusions
  - Mature inclusions are larger and more basophilic, and the margins of the nuclear membrane become blurred, to form the characteristic "smudge cell"

Immunohistochemical Features

- IHC stains the intranuclear accumulations of virus

Ultrastructural Features

- Intranuclear paracrystalline array of virions represented by icosahedral capsids that measure 70-90 nm in diameter

Pathologic Differential Diagnosis

- Herpes simplex viruses
- Varicella-zoster virus
- Cytomegalovirus

HANTAVIRUSES

Hantaviral diseases in humans are caused by a group of closely related, trisegmented, negative-sense RNA viruses of the genus *Hantavirus*, of the family *Bunyaviridae*. Two classes of hantavirus-associated illnesses have been described: Hemorrhagic Fever Renal Syndrome, (HFRS) for disease in which the kidneys are primarily involved, and Hantavirus Pulmonary Syndrome (HPS), for disease in which the lungs are primarily affected.

Clinical Features

The initial symptoms of HFRS and HPS are similar and resemble those seen in early phases of many other viral diseases. Fever, myalgia, headache, vomiting, weakness, and cough are common symptoms in early phases of both HFRS and HPS. Renal involvement is seen in all cases of HFRS, and the clinical presentation ranges from a mild illness with minimal renal dysfunction to a more severe form with acute renal failure and shock. Only HFRS patients who die during the later phases of renal failure typically show significant pulmonary edema. The clinical picture for HPS is quite different from that for HFRS. The initial prodrome is followed by rapidly progressive pulmonary edema, respiratory insufficiency, and shock. In

fatal cases, the majority of deaths occur within 2 days of hospitalization. Hemorrhages and peripheral signs of vasomotor instability, such as flushing, conjunctival injection, and periorbital edema as seen in HFRS, are extremely rare.

Radiologic Features

Chest radiographs may be normal early in the course of HPS, but evidence of interstitial edema can be observed in the majority of cases within 48 hours of hospitalization.

Pathologic Features

Gross Findings

Large quantities of protein-rich, gelatinous retroperitoneal edema fluid are found in the hypotensive phase of severe HFRS, while all HPS patients have large bilateral pleural effusions and heavy, edematous lungs. In fatal Far Eastern HFRS, a distinctive triad of hemorrhagic necrosis of the junctional zone of the renal medulla, right atrium of the heart, and anterior pituitary can be seen. In patients with HPS, hemorrhages are exceedingly rare, and ischemic necrotic lesions, except those attributed to shock, are not seen.

Microscopic Findings

Histologically, morphologic changes of the endothelium are uncommon but, when seen, consist of prominent and swollen endothelial cells. Vascular thrombi and endothelial cell necrosis are rare. In HFRS, the most severe and characteristic microscopic lesions involve the kidney; however, an interstitial pneumonitis can also be seen in some fatal cases. In contrast, the microscopic changes in HPS are principally seen in the lung and spleen. The lungs (Figure 13-2) show a mild to moderate interstitial pneumonitis characterized by variable degrees of edema and an interstitial mononuclear cell infiltrate composed of a mixture of small and enlarged mononuclear cells with the appearance of immunoblasts. Focal hyaline membranes composed of condensed proteinaceous intra-alveolar edema fluid, fibrin, and variable numbers of inflammatory cells are observed. Typically, neutrophils are scanty and the alveolar pneumocytes are intact with no evidence of cellular debris, nuclear fragmentation, or hyperplasia. In fatal cases, with a prolonged survival interval, tissues show features more characteristic of the exudative and proliferative stages of diffuse alveolar damage (Figure 13-2B). Other characteristic microscopic findings in HPS cases include variable numbers of immunoblasts within the splenic red pulp and periarteriolar white pulp (Figure 13-2E), lymph nodal paracortical zones, hepatic portal triads, and peripheral blood.

Ancillary Studies

Virus-specific diagnosis and confirmation can be achieved through serology, PCR for hantavirus RNA, or IHC for hantaviral antigens. Serologic testing can detect hantavirus-specific immunoglobulin M or rising titers of immunoglobulin G in patient sera and is considered the method of choice for laboratory confirmation of HPS. PCR detects viral RNA in blood and tissues and is extremely useful for diagnostic and epidemiologic purposes. Hantaviral RNA can also be detected in formalin-fixed, paraffin-embedded archival tissues by RT-PCR. IHC testing of formalin-fixed tissues is a sensitive method to confirm hantaviral infections, and viral antigens are found primarily within capillary endothelia throughout various tissues in both HPS and HFRS (Figure 13-2C). In HPS, marked accumulations of hantaviral antigens are found in the pulmonary microvasculature and in splenic and lymph nodal follicular dendritic cells. Electron microscopic studies of HPS lung tissue demonstrate infection of endothelial cells and macrophages. The virus or virus-like particles observed are infrequent and extremely difficult to identify in autopsy tissues; in contrast, typical endothelial granulofilamentous inclusions are seen more frequently (Figure 13-2D).

Differential Diagnosis

HPS should be suspected in cases of acute respiratory distress syndrome (ARDS) without a known precipitating cause among previously healthy individuals. The level of suspicion should be particularly high when patients have a known exposure to rodents in areas where *Peromyscus maniculatus* or other reservoirs of hantavirus are found. Physicians need to differentiate HPS from other common acute respiratory diseases, such as pneumococcal pneumonia, influenza virus, and unexplained ARDS. Diseases that need to be distinguished pathologically from HPS include a relatively large number of different viral, rickettsial, and bacterial infections, as well as various noninfectious disease processes.

Prognosis and Therapy

Recovery in HFRS is usually complete, with no apparent long-term sequelae. Mortality rates for HFRS range from 1-15%, with shock and uremia being the main contributing causes of death, although pulmonary

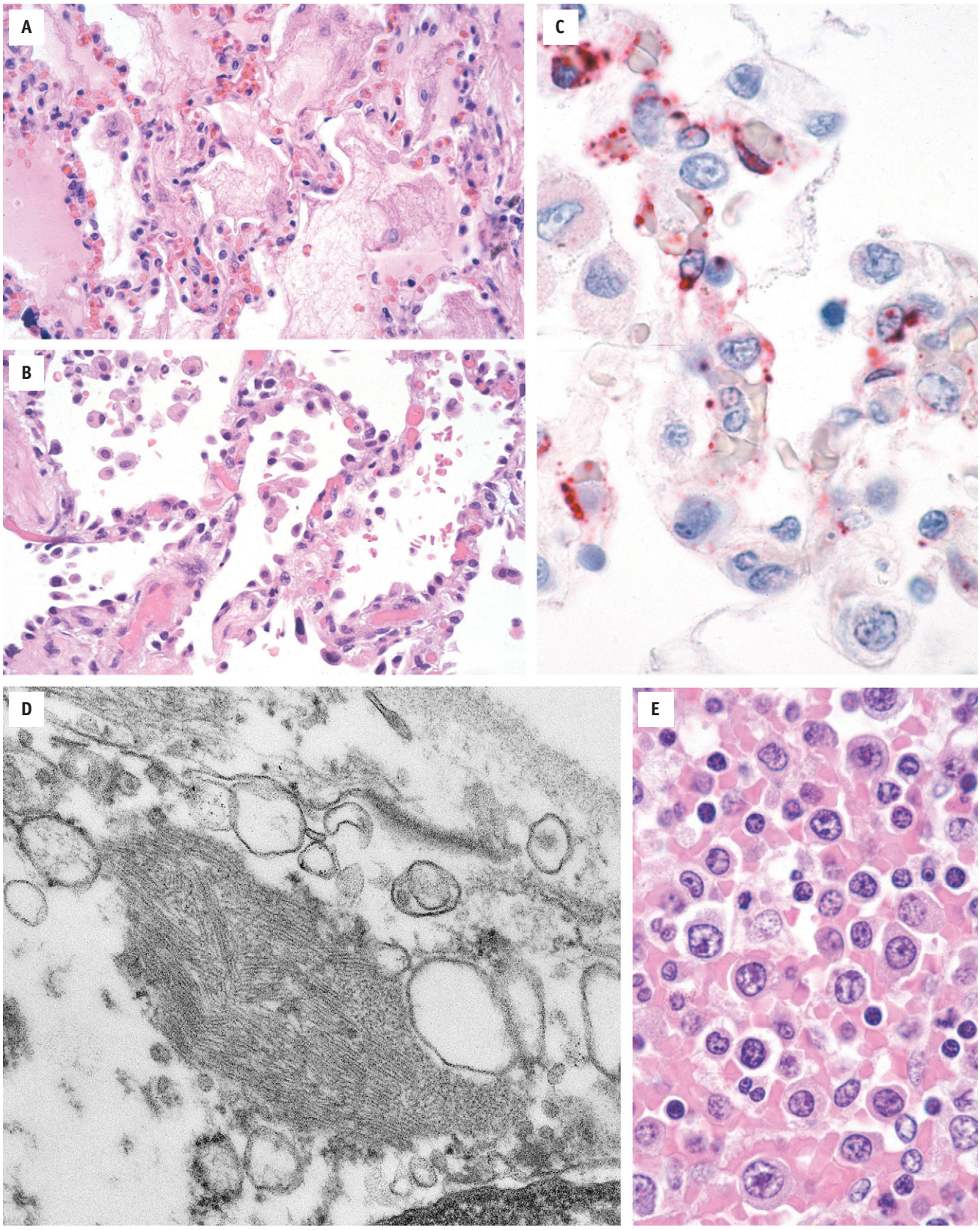

FIGURE 13-2

Hantavirus Pulmonary Syndrome (HPS)

(A) Mild mononuclear interstitial pneumonitis and edema in a typical case of HPS. **(B)** Type II pneumocyte hyperplasia as seen in patients with HPS who die after a prolonged clinical course. **(C)** Widespread immunostaining of hantaviral antigens in the pulmonary microvasculature of an HPS patient (immunoalkaline phosphatase). **(D)** Ultrastructural appearance of a typical granulofilamentous hantavirus inclusion within the pulmonary capillary endothelium. **(E)** Spleen from a fatal case in which immunoblasts are seen in the periarteriolar sheath. Note the prominent nucleoli and high nuclear-to-cytoplasmic ratio.

HANTAVIRUSES—FACT SHEET

Definition

- Hantaviral diseases are caused by closely related, trisegmented, negative-sense RNA viruses of the genus *Hantavirus*, of the family *Bunyaviridae*
- Two classes of hantavirus-associated illnesses have been described: Hemorrhagic Fever Renal Syndrome (HFRS) for disease in which the kidneys are primarily involved and Hantavirus Pulmonary Syndrome (HPS) for disease in which the lungs are primarily affected

Incidence and Location

- Zoonotic viruses maintained in nature by asymptomatic infection of rodents
- Transmission to humans is usually associated with exposure to rodents in and around the home, performing agricultural activities, cleaning animal sheds, sleeping on the ground, and with certain occupations
- Serotypes are distributed throughout the world
- HFRS is more common in Europe and Asia, while HPS is almost exclusively seen in the Americas
- Rare cause of pneumonia

Morbidity and Mortality

- In HFRS, mortality rates range from 1-15%
- In HPS, mortality rates may exceed 50%
- In survivors of HFRS, recovery is usually complete, with no long-term sequelae

Gender, Race, and Age Distribution

- No specific gender, race, or age distribution is generally seen

Clinical Features

- Prodrome of fever, myalgias, headache, vomiting, weakness, and cough is common in both HFRS and HPS
- Renal involvement is seen in all cases of HFRS, and the clinical presentation ranges from a mild illness with minimal renal dysfunction to a more severe form with acute renal failure and shock
- In HPS, the prodrome is followed by rapidly progressive pulmonary edema, respiratory insufficiency, and shock

Radiologic Features

- Interstitial edema without consolidation in the majority of cases within 48 hours of hospitalization
- Pleural effusions are very common

Prognosis and Therapy

- Supportive therapy, such as dialysis and circulatory and respiratory support
- Ribavirin is effective in treatment of HFRS but not HPS

HANTAVIRUSES—PATHOLOGIC FEATURES

Gross Findings

- Severe HFRS: gelatinous retroperitoneal collections, and a distinctive triad of hemorrhagic necrosis of the junctional zone of the renal medulla, right atrium of the heart, and anterior pituitary
- HPS: large bilateral pleural effusions and heavy edematous lungs

Microscopic Findings

- HFRS: the most severe and characteristic microscopic lesions involve the kidney
- HPS:
  - Lungs—an interstitial pneumonitis is seen in most cases, characterized by edema and an interstitial mononuclear cell infiltrate, and sometimes focal hyaline membranes
  - Extrapulmonary-immunoblasts in spleen, lymph nodes, and peripheral blood

Immunohistochemical Features

- IHC testing of formalin-fixed tissues is a sensitive method to confirm infection
- Hantaviral antigens are most commonly detected in endothelial cells of involved organs in HPS and HFRS

Ultrastructural Features

- Virus particles are 70-120 nm in diameter and generally appear spherical to oval in shape
- A lipid envelope containing glycoprotein spikes surrounds a core consisting of the genome nucleocapsids arranged in delicate tangles of filaments
- Granulofilamentous viral inclusions can be seen in endothelial cells
- Viral particles can be extremely difficult to visualize in tissues

Pathologic Differential Diagnosis

- Histopathologic and hematologic findings suggest the diagnosis in HPS and HFRS; however, laboratory confirmation is essential for confirmation of the diagnosis
- Differential diagnosis includes a large number of viral, rickettsial, bacterial infections, as well as non-infectious diseases

edema has been implicated in some patients. In HPS, mortality rates may exceed 50%, depending on serotype involved. Management of patients with HPS or HFRS is often complex and phase-specific. Supportive therapy, such as dialysis and circulatory and respiratory support, is the basis of treatment. Controlled studies suggest that ribavirin, a nucleoside analog, is effective in the treatment of hantaviral infection if administered early. Ribavirin has not proven effective in therapy of HPS.

SEVERE ACUTE RESPIRATORY SYNDROME (SARS) CORONAVIRUS

The causative agent of SARS is an enveloped, positive-stranded RNA virus that is a member of the genus *Coronavirus*, of the family *Coronaviridae.* SARS was recognized during a global outbreak of severe pneumonia that began in late 2002 in Guangdong Province, China, and gained prominence in early 2003 as cases were identified in Asia, Europe, and in North and South America. Initial studies pointed to palm civets as a possible animal reservoir. However, it appears likely that the role of civets and other small mammals is as amplifier hosts within animal markets rather than as the natural reservoir of the virus. More recently, a novel SARS-like coronavirus was found in *Rhinolophus* bats in mainland

China, suggesting that is the more likely species to be the natural reservoir from which the SARS coronavirus emerged.

Clinical Features

The disease causes an influenza-like illness that typically presents with acute onset of fever, myalgia, malaise, and chills, with rhinorrhea and sore throat being less common features. A dry cough is common, but shortness of breath and tachypnea are prominent only later in the course of the disease. Watery diarrhea occurs in some patients, typically associated with clinical deterioration in the second week of illness. People of all ages can develop the illness and children tend to have a much milder clinical course than adults. Transmission is from person to person, and the estimated incubation period is 2 to 14 days.

Radiologic Features

The radiologic features of SARS include the peripheral appearance of lung opacities, lower lobe predominance, and a mixture of ground-glass opacities, interstitial thickening, and bronchiectasis. Pneumomediastinum without preceding positive-pressure ventilation or intubation can be seen later in the disease. Multifocal peripheral subpleural ground-glass opacification or consolidation has been the most commonly observed CT feature at the time of diagnosis in patients with SARS.

Pathologic Features

Gross Findings

In fatal cases of SARS, the lungs are usually heavy and edematous with varying degrees of red and gray hepatization. Multiple bilateral hemorrhagic infarcts are commonly seen in association with subpleural hemorrhages.

Microscopic Findings

The main histopathologic pattern is diffuse alveolar damage (Figure 13-3A). Increased mononuclear cell infiltrates in the interstitium can be seen in some cases. Other findings identified in some patients include focal intra-alveolar hemorrhage, necrotic inflammatory debris in small airways, and organizing pneumonia. In addition, multinucleated syncytial cells may be seen in the alveolar spaces of some patients who die 14 days or more after onset of illness (Figure 13-3B).

Ancillary Studies

ISH and IHC studies of tissues from SARS patients have identified coronavirus in ciliated columnar epithelial cells in the trachea, bronchi, and bronchioles (Figure 13-3C) and in pneumocytes (Figure 13-3D,E) and occasional macrophages in some patients. Antigens are more readily identified in patients who die within the first 2 weeks of onset of illness. Electron microscopic examination can show coronavirus particles and nucleocapsid inclusions in cytoplasmic vesicles and along the cell membranes of pneumocytes, in phagosomes of macrophages, and associated with fibrin in alveolar spaces. Negative stains reveal particles averaging 80-100 nm in size with a characteristic crown-like fringe on the surface (Figure 13-3F).

Differential Diagnosis

The histopathologic findings seen in the lungs of patients who die from SARS are somewhat nonspecific and can also be seen in acute lung injury cases caused by infectious agents, trauma, drugs, or toxic chemicals. Multinucleated syncytial cells similar to those seen in some SARS patients can also be found in many viral infections, including measles, parainfluenza viruses, respiratory syncytial virus, and Nipah virus infections. An unequivocal diagnosis can be made only by laboratory tests such as viral culture, direct fluorescent antibody, serology, PCR, or IHC.

Prognosis and Therapy

Patients with SARS can undergo complete recovery; however, the disease can progress to acute respiratory failure and death in about 5-10% of infected individuals. About 20-30% of all patients need observation in intensive care, and most of these require mechanical ventilation. The clinical management of patients with SARS includes respiratory support with intensive care support as needed. Ribavirin, lopinavir, and type I IFN show inhibition of SARS virus in tissue culture. However, their utility in SARS-infected patients is inconclusive, and they may actually be harmful. Similarly, studies of corticosteroid use are inconclusive and again these agents may possibly cause harm to the patient. Experimental and clinical trials are needed to evaluate the efficacy of various treatments.

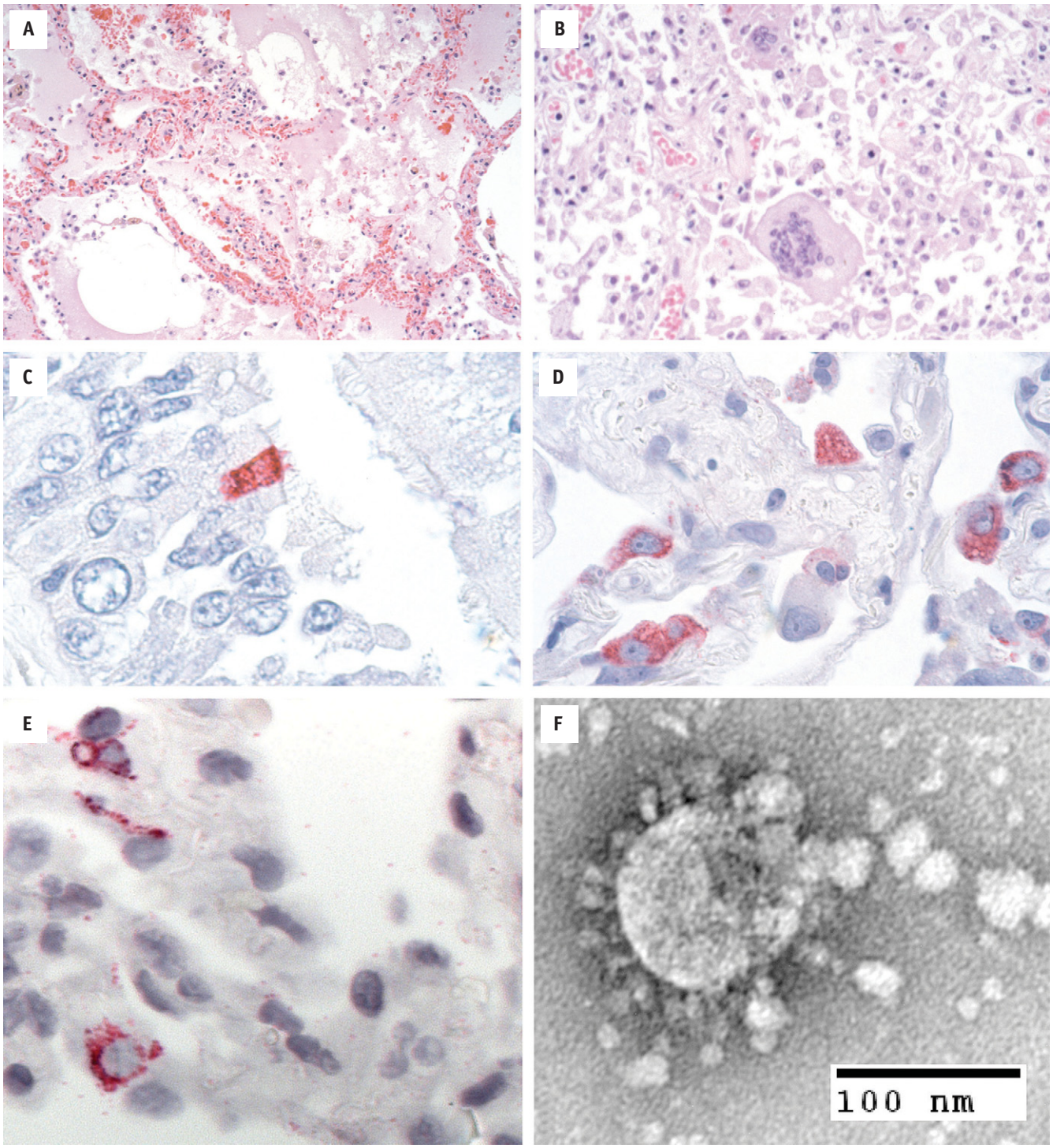

FIGURE 13-3

Severe Acute Respiratory Syndrome (SARS)

(A) Prominent edema, congestion and focal hemorrhage, and mild interstitial inflammation. **(B)** Multinucleated syncytial giant cells as seen in some cases of fatal SARS. Note the absence of discernible viral inclusions. **(C)** Ciliated upper airway epithelial cell containing viral antigens (immunoalkaline phosphatase). **(D)** Immunostaining of coronavirus antigens in alveolar pneumocytes (immunoalkaline phosphatase). **(E)** ISH showing infected pneumocytes and macrophages containing viral nucleic acids (digoxigenin-labeled probe followed by immunoalkaline phosphatase staining). **(F)** Electron micrograph showing 80-100 nm coronavirus particles, named for the characteristic crown-like fringe on their surfaces.

SARS CORONAVIRUS—FACT SHEET

Definition

- SARS coronavirus is an enveloped, positive-stranded RNA virus that is a member of the genus *Coronavirus*, of the family *Coronaviridae*

Incidence and Location

- First reported in Guangdong Province in Southern China in 2002, but rapidly spread to become a worldwide illness in 2003

Morbidity and Mortality

- SARS is fatal in about 5-10% of patients
- In patients who survive the illness, the recovery is usually complete
- Mortality and risk of complications are higher among elderly persons and persons of any age with certain underlying health conditions
- Children have a much milder clinical course than adults
- Secondary bacterial pneumonias with organisms may occur as a complication

Gender, Race, and Age distribution

- People of all ages are vulnerable to SARS
- No recognized gender or racial predilection

Clinical Features

- SARS coronavirus is spread person-to-person, primarily through the coughing and sneezing of infected persons
- Estimated incubation period is 2 to 14 days
- Uncomplicated SARS illness is an influenza-like illness characterized by an abrupt onset of fever, myalgia, headache, malaise, nonproductive cough, sore throat, and rhinitis

Radiologic Features

- Peripheral lung opacities with lower-lobe predominance, and a mixture of ground-glass opacities, interstitial thickening, and bronchiectasis
- CT at the time of diagnosis shows multifocal peripheral subpleural ground-glass opacities
- Pneumomediastinum can be seen later in the disease

Prognosis and Therapy

- Supportive respiratory and intensive care therapy
- Inconclusive studies regarding the efficacy of antivirals and corticosteroids
- Specific antibiotic therapy in cases with secondary bacterial infection
- Prevention of nosocomial transmission is an important strategy for management of cases in a hospital setting

SARS—PATHOLOGIC FEATURES

Gross Findings

- Lungs are usually heavy and edematous with varying degrees of red and gray hepatization
- Multiple bilateral pulmonary hemorrhagic infarcts and subpleural hemorrhages can be seen

Microscopic Findings

- Diffuse alveolar damage
- Hemorrhage
- Edema
- Multinucleated cells in about 10% of cases
- Viral inclusions cannot be identified by light microscopy

Immunohistochemical Features

- IHC reveals SARS-CoV antigens primarily in the respiratory epithelial cells of airways and pneumocytes, particularly in patients who die within the first two weeks of onset of the illness

Ultrastructural Features

- Virions form by alignment of the helical nucleocapsids along the membranes of the endoplasmic reticulum or Golgi complex and acquire an envelope by budding into the cisternae
- Cellular vesicles become filled with virions and progress to the cell surface for release of viral particles
- Negative stains reveal particles averaging 80-100 nm in size with a characteristic crown-like fringe on the surface

Pathologic Differential Diagnosis

- Other causes of diffuse alveolar damage, including many viral, rickettsial, and bacterial infections, as well as non-infectious diseases (trauma, drugs, and toxins)

CYTOMEGALOVIRUS

Cytomegalovirus (CMV), a large, double-stranded DNA virus, is a ubiquitous human pathogen, and in North America infects approximately 50-90% of the population. Like all herpesviruses, CMV remains with its host for life after primary infection and establishes latency in various cell types, including vascular endothelial cells, monocytes and macrophages, neutrophils, and renal and pulmonary epithelial cells. Activation of viral replication occurs in persons with severely compromised immunity.

CLINICAL FEATURES

Most CMV infections are inapparent, although cases of primary infection in otherwise healthy individuals can result in a self-limited mononucleosis syndrome resembling the illness caused by Epstein-Barr virus. Pulmonary involvement in CMV mononucleosis occurs in approximately 6% of these cases. Adults and children with advanced HIV disease and recipients of hematopoietic stem cell and lung transplants are particularly at risk for developing CMV pneumonia. Before the use of CMV screening and effective anti-viral prophylaxis regimens, 10-30% of all patients undergoing allogeneic bone marrow transplantation for leukemia, and 15-55% of solid organ transplants, developed CMV pneumonia with case fatality rates greater than 80% in some series. Neonates are also at risk. Symptomatology includes fever, cough, rales, and hypoxemia. Systemic dissemination and extrapulmonary involvement can occur in some patients.

Radiologic Features

Pulmonary CMV disease typically appears as bilateral nodular or reticular opacities on chest radiographs. Pleural effusions are identified in approximately 10-30% of patients. Because some patients may be co-infected with other pulmonary pathogens, radiologic findings may be confusing. Some patients with documented infection have normal radiographs.

Pathologic Features

Gross Findings

There are several general patterns of pulmonary CMV infection. The lungs are typically heavy and may appear diffusely consolidated, or show scattered nodular foci of hemorrhage and necrosis. Rarely, CMV infection of the lungs manifests as a single pulmonary nodule.

Microscopic Findings

Multiple histopathologic patterns have been reported for CMV pneumonia. Extensive intra-alveolar hemorrhage with scattered cytomegalic cells and relatively scant inflammatory cell infiltrates may occur. In a similar manner, extensive involvement of the alveolar epithelium with minimal inflammation or overt evidence of parenchymal injury has also been described. Other patterns include multifocal or miliary lesions with mixed inflammatory cell infiltrates, hemorrhage, necrosis, and cytomegalic cells, or a diffuse, predominantly mononuclear cell infiltrate, interstitial pneumonitis with intra-alveolar edema and fibrin deposition, and diffusely distributed cytomegalic cells. The cytomegalic changes of CMV-infected cells are evident on standard hematoxylin-eosin staining and are virtually pathognomonic of active CMV infection. The cells are enlarged (25-40 microns) and contain amphophilic to deeply basophilic intranuclear and intracytoplasmic inclusions (Figure 13-4A,B). The single intranuclear inclusion is comprised of viral nucleoprotein and assembled capsids, and is a large (up to 20 microns), round-to-ovoid body with a smoothly contoured border that is generally surrounded by a clear halo that gives the inclusion a distinctive "owl's eye" appearance. Cytoplasmic inclusions are small (1-3 microns), granular bodies that appear after the intranuclear inclusion is well developed and are not uniformly present in all CMV-infected cells. These inclusions represent a mixture of virions and various cellular organelles, and increase in size and number as the infection progresses. Unlike the intranuclear inclusions, the cytoplasmic inclusions stain with periodic acid-Schiff stain and are deeply argyrophilic with methenamine silver stains.

Ancillary Studies

CMV pneumonia is defined by the presence of signs or symptoms of pulmonary disease combined with the detection of CMV in bronchoalveolar lavage fluid or lung tissue samples. Detection methods that support this definition include virus isolation, histopathologic observation of cytomegalic cells, ISH, or IHC stains (Figure 13-4C). Detection by PCR alone is considered too sensitive for the diagnosis of CMV pneumonia and is insufficient for this purpose. CMV is most often cultured in human diploid fibroblasts using a shell vial method to enhance infectivity and can usually yield diagnostic results within 48 hours.

Differential Diagnosis

Because the histopathologic features of CMV pneumonia are varied, the differential diagnosis depends on the predominant pattern of histologic pattern (hemorrhage, miliary inflammatory lesions, or diffuse interstitial pneumonitis). The cytopathologic changes of CMV-infected cells are generally sufficient to establish a diagnosis. CMV inclusions may on occasion, however, be confused with those of other herpesviruses, adenoviruses, or measles, but none of these pathogens collectively shows cytomegaly, a single large nuclear inclusion with a prominent halo, and multiple small cytoplasmic inclusions. Reactive pneumocytes can occasionally show enlarged nuclei, but the nuclei will be immunonegative with IHC for CMV.

Prognosis and Therapy

Ganciclovir, foscarnet, and intravenous CMV immune globulin remain important lines of treatment for CMV pneumonia and have diminished mortality in immunosuppressed patients with this disease. Nonetheless, mortality attributable to CMV pneumonia is approximately 50%.

HERPES SIMPLEX VIRUSES

Human herpes simplex viruses (HSV) are large, enveloped, double-stranded DNA-viruses approximately 100-110 nm in diameter. Two serologic types are recognized, and each is most frequently associated with particular disease syndromes; however, either serotype may cause any of the clinical syndromes associated with either serotype. HSV-1 causes gingivostomatitis, pharyngitis, esophagitis, keratoconjunctivitis, and encephalitis, and is the serotype most commonly associated with adult

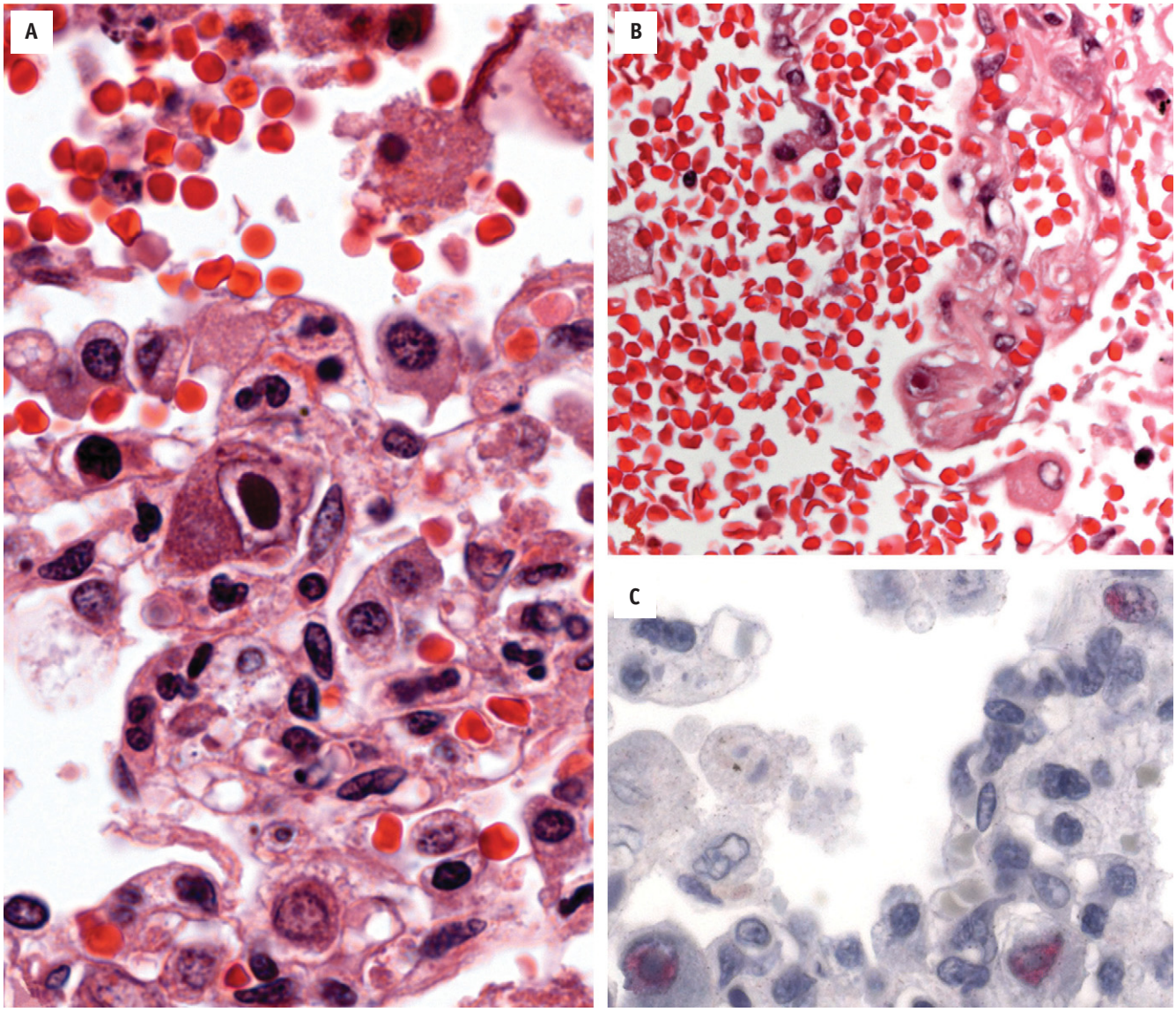

FIGURE 13-4

Cytomegalovirus pneumonia
(A) CMV-infected cell with large, basophilic "owl's eye" intranuclear inclusion and smaller, amphophilic cytoplasmic inclusions. **(B)** Alveolar hemorrhage with relatively little inflammation and CMV-infected pneumocytes. **(C)** Immunohistochemical localization of CMV-infected cells in the pulmonary parenchyma (immunoalkaline phosphatase).

HSV pneumonia. HSV-2 typically infects genital sites and is the serotype associated with approximately 80% of disseminated disease and pulmonary infections in newborn infants.

Clinical Features

HSV, like all herpesviruses, has the ability to persist in an inactive state for varying periods of time and then recur spontaneously following undefined stimuli associated with physical or emotional stress, trauma to nerve roots or ganglia, fever, immunosuppression, or exposure to ultraviolet radiation. Tracheobronchitis and pneumonia are the primary respiratory tract manifestations of HSV infection. In adults, infection of the respiratory tract with HSV may be associated with disseminated herpetic infection, but is more commonly identified as an isolated disease manifestation resulting from reactivation of latent herpetic infections in the oropharynx. Mucocutaneous herpetic infection generally precedes HSV pneumonia, and aspiration of virus-containing secretions into the lower respiratory tract is believed to be the most frequent cause of pulmonary infection with HSV; however, oral lesions may be absent in patients with herpetic laryngotracheitis and bronchopneumonia. Disease can also be associated with airway trauma caused by tracheal intubation or from hematogenous dissemination of HSV. Newborn infants, severely immunosuppressed or burned

CYTOMEGALOVIRUS PNEUMONIA—FACT SHEET

Definition
- Human CMV is a β-herpesvirus with the largest genome (230 kbp) of all the herpesviruses known to infect humans

Incidence and Location
- Common cause of pneumonia in immunocompromised patients
- Woldwide distribution
- CMV is a ubiquitous human pathogen, and in North America infects approximately 50-90% of the population
- Patients with advanced HIV disease and recipients of hematopoietic stem cell and lung transplants are particularly at risk of developing CMV pneumonia

Mortality
- Mortality attributable to CMV pneumonia is approximately 50%

Gender, Race, and Age Distribution
- Can develop in patients of any age
- No apparent gender or racial predilection

Clinical Features
- Fever, nonproductive cough, rales, and hypoxemia
- Disseminated infection may also cause adrenalitis, hepatitis, or encephalitis

Radiologic Features
- Common findings are bilateral nodular or reticular opacities
- Pleural effusions are identified in approximately 10-30% of patients
- Some patients with documented infection have normal radiographs

Therapy
- Ganciclovir, foscarnet, and intravenous CMV immune globulin remain important lines of treatment

CYTOMEGALOVIRUS PNEUMONIA—PATHOLOGIC FEATURES

Gross Findings
- Lungs are typically heavy and may appear diffusely consolidated or show scattered nodular foci of hemorrhage and necrosis
- Rarely, the infection manifests as a single pulmonary nodule

Microscopic Findings
- Multiple histopathologic patterns have been reported, including extensive intra-alveolar hemorrhage, diffuse interstitial pneumonitis, and miliary inflammatory foci with necrosis
- Virally induced cytopathic changes include cytomegaly (25-40 microns) and amphophilic to deeply basophilic intranuclear and intracytoplasmic inclusions in various cell types including macrophages, pneumocytes, glandular epithelium, endothelium, and fibroblasts
  - The single intranuclear inclusion is a large (up to 20 microns), round to ovoid body with a smoothly contoured border that is generally surrounded by a clear halo
  - Cytoplasmic inclusions are small (1-3 microns), stain with periodic acid-Schiff stain, and are deeply argyrophilic with methenamine silver stains

Immunohistochemical Features
- Commercially available antibodies can assist in the diagnosis of CMV

Ultrastructural Features
- Mature enveloped virions from 150- 200 nm

Pathologic Differential Diagnosis
- Herpes simplex viruses, varicella zoster virus, and adenoviruses
- Reactive pneumocytes

patients, and patients with severe trauma are at greatest risk of developing HSV pneumonia. Lower respiratory tract disease in neonates is most commonly associated with disseminated herpetic infections. Most cases of neonatal disease represent primary HSV infections and are acquired during parturition from HSV-infected mothers. The incidence of neonatal HSV infection is approximately 1 in 3,200 deliveries, and disseminated disease develops in approximately 25% of infected neonates. In disseminated infections, signs and symptoms appear a mean of 5 days after birth (range, 0 to 12 days), and approximately 40-50% of these patients develop pneumonia.

Radiologic Features

Chest radiographs of patients with HSV pneumonia show ill-defined nodular or reticular densities of various sizes scattered in both lung fields. During the early stages of disease, these nodules measure 2-5 mm and are best seen in the periphery of the lungs. As the disease progresses, these lesions coalesce and enlarge to form more extensive segmental and subsegmental infiltrates. Computed tomography shows patchy ground-glass opacities with scattered areas of consolidation and nodular densities. Pleural effusions are common.

Pathologic Features

Gross Findings

HSV tracheobronchitis appears as 5-15 mm ulcers covered by fibrinopurulent exudate on the mucous membranes. In HSV pneumonia acquired through the airways, the lungs are heavy and show nodular hemorrhagic foci that are generally distributed around bronchi and bronchioles. In hematogenously acquired HSV pneumonia, hemorrhagic foci usually have a random or miliary distribution.

Microscopic Findings

Herpetic tracheobronchitis is an ulcerative process characterized by large areas of denuded mucosal epithelium and fibrinopurulent exudate containing necrotic cells. Despite extensive tissue damage, cells with intranuclear inclusions may be sparse and are found most often at the margins of the ulcerated epithelium or occasionally in the mucous glands below the ulcerated mucosa. In the lung, herpetic lesions show extensive necrosis and karyorrhectic debris, and are associated with hemorrhage and a sparse-to-moderate neutrophilic infiltrate (Figures 13-5A,B). Intranuclear inclusions are best appreciated in cells at the leading edge of necrotic foci. Inclusions appear either as homogeneous, amphophilic, and glassy (e.g., Cowdry type B inclusions), or as eosinophilic with a halo separating the inclusion from the nuclear membrane (e.g., Cowdry type A inclusions). Other changes associated with HSV, including multinucleation and nuclear molding, ground-glass nuclear chromatin, and ballooning degeneration of the cytoplasm, are more frequently associated with squamous epithelium and less often encountered in the lung.

Ancillary Studies

Virus isolation remains an important diagnostic method; however, because HSV can be isolated from oropharyngeal secretions and occasionally from the lower respiratory tract of patients who lack overt pulmonary disease, virologic cultures must be interpreted in the context of complementary clinical, radiographic, and histopathologic findings as much as possible. PCR methods that amplify HSV DNA from clinical specimens, including tissue and blood, can be particularly useful for distinguishing between HSV-1 and HSV-2 infections. Commercially available antibodies exist for IHC detection of HSV in tissues (Figure 13-5C). Electron microscopy can also be used to demonstrate encapsulated viral particles with a targetoid appearance arranged in a lattice-like pattern (Figure 13-5D).

Differential Diagnosis

HSV, varicella-zoster virus (VZV), adenoviruses, measles virus, and CMV can cause necrotizing hemorrhagic pneumonias, and each produce intranuclear inclusions that may be difficult to differentiate. The viral inclusions of HSV are identical to those of VZV; separation can be accomplished by IHC, molecular methods, or culture. Distinction from adenoviruses can be accomplished if smudge cells are identified (supporting the presence of adenovirus). HSV does not produce cytoplasmic inclusions, which should be seen in CMV and in measles.

Prognosis and Therapy

Prior to the discovery and use of antiviral therapies, 85% of neonates with disseminated HSV disease died from the infection. With early diagnosis and high-dose acyclovir therapy, however, mortality has been reduced to approximately 30%. Foscarnet has been used effectively in some acyclovir-resistant patients.

VARICELLA ZOSTER VIRUS

Varicella zoster virus (VZV), is a human alpha-herpesvirus closely related to HSV. Primary infection causes varicella (chickenpox), and reactivation of latent virus causes herpes zoster (shingles). VZV is ubiquitous in human populations around the world, and humans are the only known host. During the prevaccine era in the United States, approximately 4 million cases, 4,000 to 9,000 hospitalizations, and 50 to 140 deaths were reported annually. VZV-related deaths have declined sharply in the United States, however, since universal childhood vaccination was implemented in 1995.

Clinical Features

Primary infection with VZV occurs by inoculation of respiratory mucosa with infectious aerosols or by direct contact with skin lesions of patients with varicella or herpes zoster. After a primary viremia in the reticuloendothelial system, and secondary viremia in circulating mononuclear cells, the virus is disseminated to the skin, where it initiates a pruritic vesicular rash (chickenpox), and is disseminated back to mucosal sites in the lungs. The attack rate for previously uninfected household contacts exposed to varicella is approximately 90%. VZV also establishes latent infection within satellite cells and neurons of the trigeminal and dorsal root ganglia and can reactivate under various conditions to cause herpes zoster, a painful unilateral vesicular eruption distributed in a dermatomal distribution. Although chickenpox is usually a relatively benign infection in children, adult patients are approximately 25 times more likely than children to develop pneumonia. Pneumonia occurs in approximately 10-15% of adults primarily infected with VZV; however, the incidence of pneumonia in bone marrow transplant recipients and acute leukemia patients may be as high as 30-45%. The greatest risk of severe

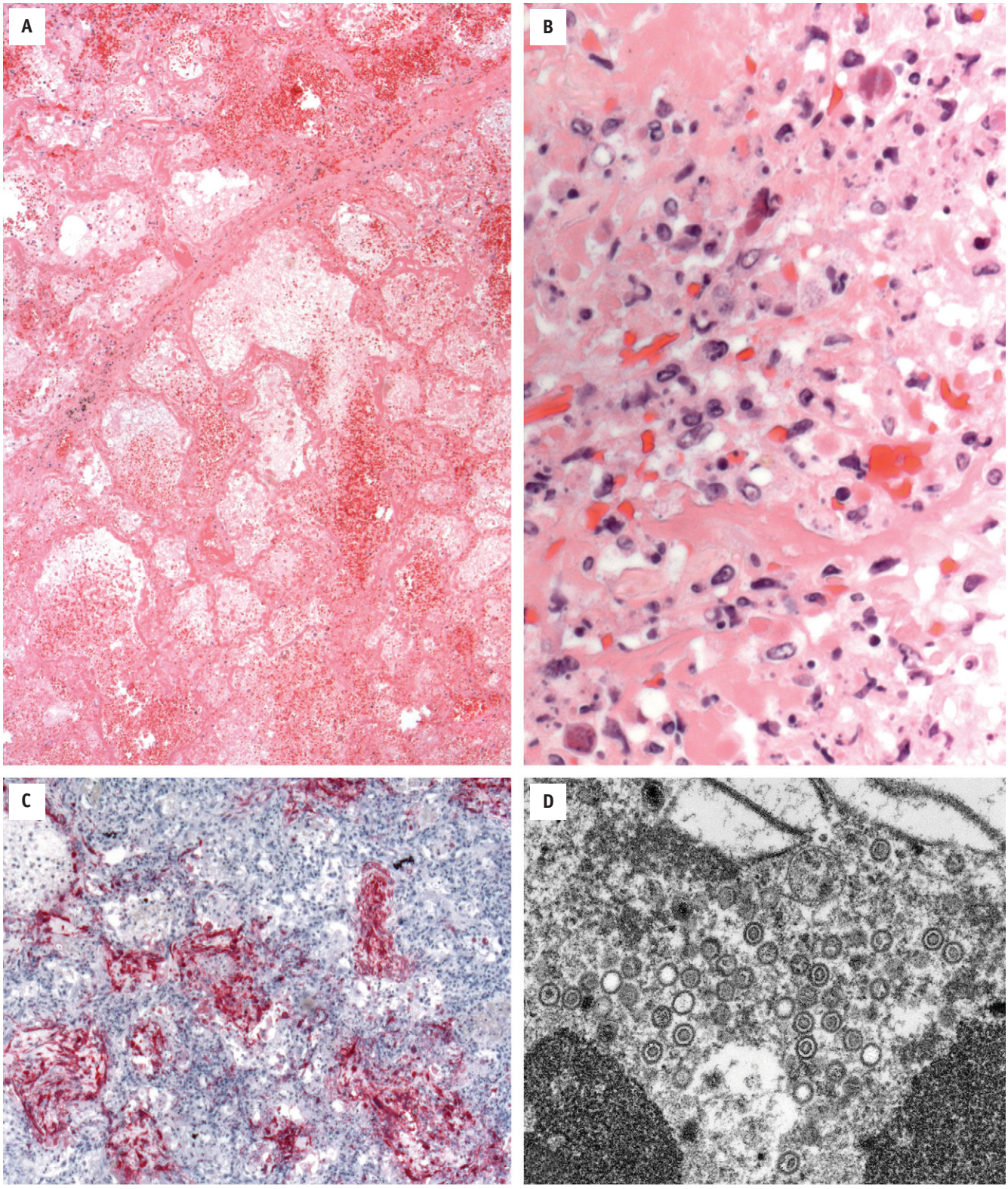

FIGURE 13-5

Herpes simplex virus pneumonia

(A) Extensive necrosis and hemorrhage associated with HSV pneumonia. **(B)** Glassy, amphophilic, intranuclear inclusions in HSV-infected cells at the margin of a necrotic focus in the lung. **(C)** Immunohistochemical localization of HSV in a patient with fatal HSV pneumonia (immunoalkaline phosphatase). **(D)** Ultrastructural view of HSV-infected cell showing complete enveloped virions in the cytoplasm. (Courtesy of Cynthia Goldsmith, Centers for Disease Control and Prevention, Atlanta, Georgia).

HERPES SIMPLEX VIRUSES—FACT SHEET

Definition

- Human herpes simplex viruses (HSV) are large, enveloped, double-stranded DNA-viruses that exist in two serologic types
  - HSV-1 is the serotype most commonly associated with adult HSV pneumonia
  - HSV-2 is the serotype associated with approximately 80% of disseminated disease and pulmonary infections in newborn infants

Incidence and Location

- Worldwide distribution
- Newborn infants, severely immunosuppressed or burned patients, and patients with severe trauma are at greatest risk of developing HSV pneumonia
- Most cases of neonatal disease represent primary HSV infections and are acquired during parturition from HSV-infected mothers; the incidence of neonatal HSV infection is approximately 1 in 3,200 deliveries

Mortality

- Prior to the discovery and use of antiviral therapies, 85% of neonates with disseminated HSV disease died from the infection
- With early diagnosis and high-dose acyclovir therapy, mortality has been reduced to approximately 30%

Gender, Race, and Age Distribution

- People of all ages are susceptible
- No recognized gender or racial predilection

Clinical Features

- In adults, infection of the respiratory tract with HSV may be associated with disseminated herpetic infection, but is more commonly identified as an isolated disease manifestation resulting from reactivation of latent herpetic infections in the oropharynx
- Disseminated disease develops in approximately 25% of infected neonates; approximately 40-50% of these patients develop pneumonia
- Infants with disseminated neonatal HSV infections first show signs and symptoms a mean of 5 days after birth (range, 0 to 12 days). As the disease progresses, the clinical picture often resembles bacterial sepsis, evolving rapidly to pneumonia, shock, and disseminated vascular coagulopathy

Radiologic Features

- Ill-defined nodular or reticular densities of various sizes scattered in both lung fields
- During the early stages of disease, these nodules measure 2-5 mm and are best seen in the periphery of the lungs; as the disease progresses, these lesions coalesce and enlarge to form more extensive segmental and subsegmental infiltrates
- Pleural effusions are commonly identified

Therapy

- Antiviral therapies include acyclovir, valacyclovir, and famcyclovir
- Foscarnet has been used effectively in some acyclovir-resistant patients

HERPES SIMPLEX VIRUSES—PATHOLOGIC FEATURES

Gross Findings

- HSV tracheobronchitis: 5-15 mm ulcers covered by fibrinopurulent exudate on the mucous membranes of the large airways
- HSV pneumonia acquired through the airways: lungs are heavy and show nodular hemorrhagic foci that are generally distributed around bronchi and bronchioles
- Hematogenously acquired HSV pneumonia: hemorrhagic foci have a random or miliary distribution

Microscopic Findings

- HSV tracheobronchitis: large areas of denuded epithelium and exudate containing necrotic cells; cells with intranuclear inclusions may be sparse and are found most often at the margins of the ulcerated epithelium or occasionally in the mucous glands below the ulcerated mucosa
- HSV pneumonia: lesions show hemorrhage and necrosis with karyorrhectic debris; intranuclear inclusions are best appreciated in cells at the edge of necrotic foci
- Inclusions appear as homogeneous, amphophilic, and glassy or as eosinophilic with a halo separating the inclusion from the nuclear membrane
- Multinucleation and nuclear molding, ground-glass nuclear chromatin, and ballooning degeneration of the cytoplasm are more frequently associated with squamous epithelia and less often encountered in the lung

Immunohistochemical Features

- IHC testing of formalin-fixed tissues is a sensitive method to confirm HSV infections; antibodies reactive with both HSV-1 and HSV-2 are commercially available

Ultrastructural Features

- Virus particles are encapsulated and approximately 100-110 nm in diameter
- Individual particles demonstrate a targetoid appearance and are arranged in a lattice-like pattern

Pathologic Differential Diagnosis

- Varicella-zoster virus pneumonia
- Adenovirus pneumonia
- Measles pneumonia
- CMV pneumonia

disease and pneumonia occurs in those patients with chronic lung disease, immune-suppressing conditions, neonates, and pregnant women. The occurrence of pneumonia during herpes zoster is rare, and limited primarily to profoundly immunosuppressed patients, particularly bone marrow transplant recipients. VZV pneumonia develops 2 to 7 days following the onset of rash and is characterized by fever, cough, tachypnea, chest pain, and hemoptysis. Massive pulmonary hemorrhage and pulmonary infarcts are frequent terminal events. Hematopoietic cell transplant recipients may present with signs of visceral dissemination and pneumonia 1 to 4 days before the localized cutaneous eruption of herpes zoster appears, and lower respiratory tract disease has been described in the absence of skin lesions, particularly in neonates and bone marrow transplant recipients.

Radiologic Features

The lungs show multifocal, bilateral, poorly defined nodular densities that measure 5-10 mm in greatest dimension. These opacities may coalesce to form more extensive areas of consolidation. Hilar adenopathy may also occur, but pleural effusions are uncommon. Some patients who survive VZV pneumonia show persistent parenchymal nodules that may mineralize and persist as small (2-3 mm) calcifications, predominantly in the lower zones of the lungs.

Pathologic Features

Gross Findings

The lungs of patients with fatal VZV pneumonia are 2 to 3 times heavier than normal, firm, and plum-colored. There are often multiple necrotic and hemorrhagic lesions on the visceral and parietal pleura that resemble the pox lesions of skin. The trachea and bronchi are generally edematous and erythematous with occasional vesicles on the mucosal surfaces, and there may be lobular consolidation of the lungs as well as randomly distributed hemorrhagic lesions.

Microscopic Findings

The lungs show interstitial pneumonitis and diffuse miliary foci of necrosis and hemorrhage in the pulmonary parenchyma involving alveolar walls, blood vessels, and bronchioles (Figure 13-6A,B). Other findings can include intra-alveolar collections of edema, fibrin, or hemorrhage, diffuse alveolar damage, and septal edema. Virally infected cells with intranuclear inclusions may be identified in respiratory epithelial cells of the trachea and bronchi, pneumocytes, interstitial fibroblasts, or capillary endothelium. Eosinophilic intranuclear inclusions and multinucleated syncytial cells may be difficult to locate but are best identified at the edges of necrotic foci. In cases of disseminated disease, similar necrotizing hemorrhagic lesions and occasional viral cytopathic changes in epithelial cells or fibroblasts may be observed in many other tissues and organs.

Ancillary Studies

Because pulmonary symptoms most often occur several days following the onset of the characteristic rash of varicella, a pathologic diagnosis is seldom required for a real-time diagnosis of VZV pneumonia. Antigen detection kits using fluorescein-conjugated VZV monoclonal antibodies can be helpful for rapid diagnosis of cutaneous VZV infection. Antibodies are also commercially available for IHC detection of VZV in tissue specimens (Figure 13-6C); however, relatively few laboratories are able to provide well-validated assays. Some commercial laboratories offer PCR amplification to detect viral nucleic acid in clinical specimens. Isolation of the virus in cell culture remains the reference standard for the diagnosis of VZV. Infectious VZV is usually recoverable from the clear fluid of cutaneous vesicles of varicella for approximately 3 days after the appearance of these lesions and for approximately 1 week from herpes zoster lesions. By using electron microscopy, VZV has an icosahedral nucleocapsid that is indistinguishable in appearance from other herpesviruses. The enveloped viral particle is pleomorphic to spherical in shape and 180-200 nm in diameter.

Differential Diagnosis

The histopathologic appearance of VZV pneumonia most closely resembles disease caused by HSV with respect to the general pattern of lung injury (e.g., multicentric, necrotizing, and hemorrhagic lesions) and to the appearance of the glassy intranuclear inclusions.

Prognosis and Therapy

The U.S. incidence of varicella pneumonia declined markedly since universal childhood vaccination for varicella was implemented in 1995. Vaccine efficacy at preventing severe disease is approximately 97%. Untreated adult varicella pneumonia is fatal in approximately 10% of cases, but mortality is as high as 25% to 40% in certain high-risk cohorts, including pregnant women, transplant recipients, and neonates. Intravenous acyclovir is recommended for use in all patients for whom the risk of disseminated disease is particularly high or unpredictable, including patients with leukemia, bone marrow transplant recipients, and severely immune suppressed persons.

INFLUENZA VIRUSES

Influenza viruses belong to the *Orthomyxoviridae* family, and include the two important influenza viruses, types A and B, which are associated with significant human disease. All influenza viruses have a segmented, negative-sense RNA core surrounded by a lipid envelope. Influenza A viruses are further classified into subtypes

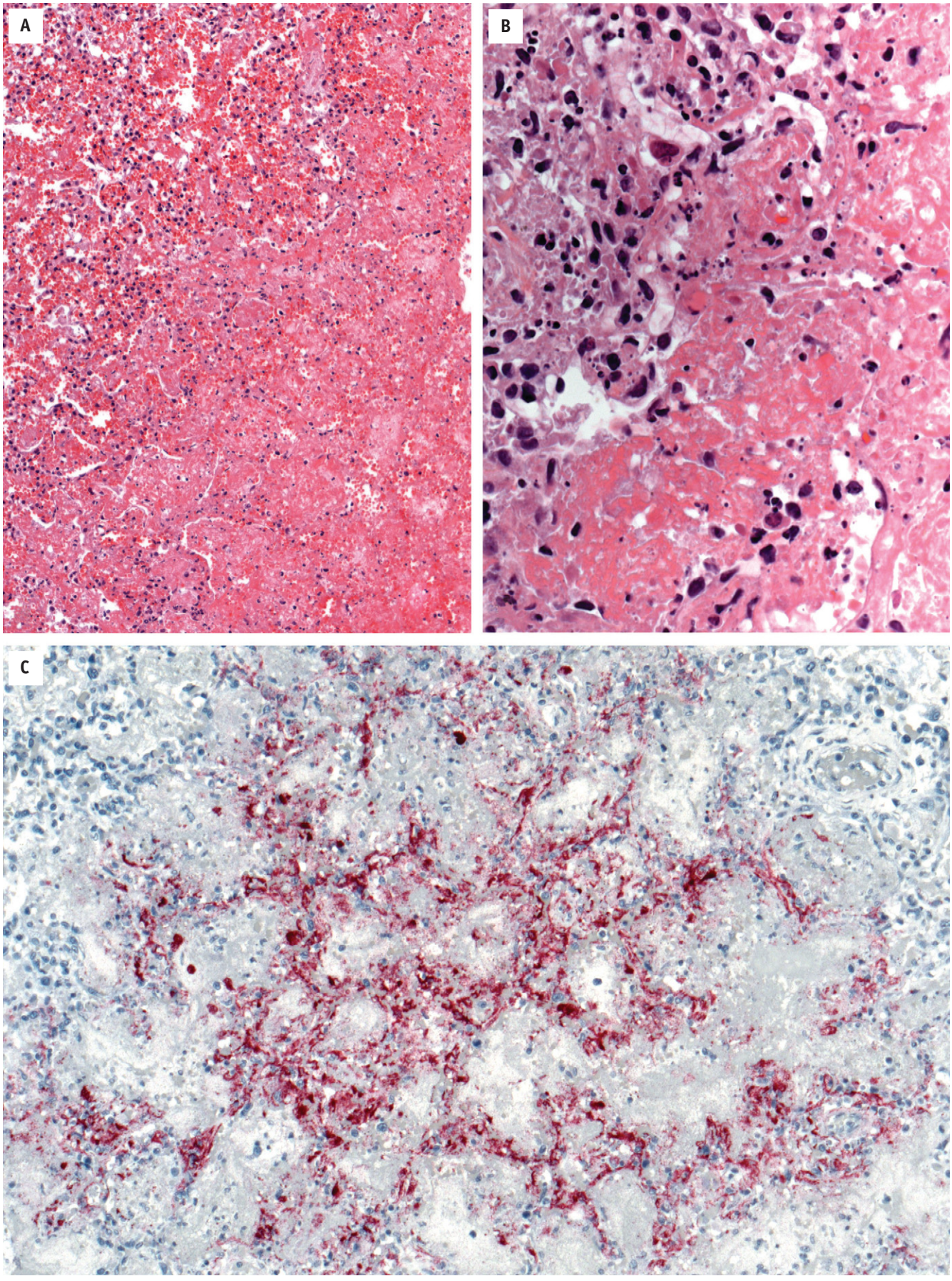

FIGURE 13-6

Varicella zoster virus pneumonia

(A) The periphery of a necrotic and hemorrhagic lesion in a case of fatal VZV pneumonia. **(B)** Eosinophilic intranuclear inclusions in VZV-infected cells at the edge of a necrotic focus. **(C)** Immunohistochemical staining of VZV antigens in a patient with fatal VZV pneumonia (immunoalkaline phosphatase).

VARICELLA ZOSTER VIRUS—FACT SHEET

Definition

- Primary infection causes varicella (chickenpox) and reactivation of latent virus causes herpes zoster (shingles)

Incidence and Location

- Ubiquitous worldwide pathogen, and humans are the only known host
- Highly contagious virus; the attack rate for previously uninfected household contacts exposed to varicella is approximately 90%
- The U.S. incidence of varicella pneumonia has dropped by two-thirds since universal childhood vaccination for varicella was implemented in 1995

Mortality

- Untreated adult varicella pneumonia is fatal in approximately 10% of cases
- Mortality is as high as 25-40% in certain high-risk cohorts, including pregnant women, transplant recipients, and neonates

Gender, Race, and Age Distribution

- Adult patients with varicella are approximately 25 times more likely than children to develop pneumonia; pneumonia occurs in approximately 10-15% of adults infected with VZV
- The greatest risk of severe disease and pneumonia occurs in those patients with chronic lung disease, immune-suppressing conditions, neonates, and pregnant women
- The incidence of pneumonia in bone marrow transplant recipients and acute leukemia patients infected with varicella may be as high as 30-45%
- No apparent gender or racial predilection

Clinical Features

- Primary infection occurs by inoculation of respiratory mucosa with infectious aerosols or by direct contact with skin lesions of patients with varicella or herpes zoster
- VZV pneumonia generally develops within 2 to 7 days following the onset of rash and may be characterized by fever, cough, tachypnea, chest pain, and hemoptysis

Radiologic Features

- Multifocal, bilateral, poorly defined nodular densities that measure 5-10 mm in greatest dimension, and may coalesce to form more extensive areas of consolidation
- Pleural effusions are uncommon
- Some survivors of VZV pneumonia show persistent parenchymal nodules that mineralize and persist as small (2-3 mm) calcifications, predominantly in the lower zones of the lungs

Prognosis and Therapy

- Intravenous acyclovir is recommended for use in all patients for whom the risk of disseminated disease is particularly likely or unpredictable, including patients with leukemia, bone marrow transplant recipients, and severely immune-suppressed persons

VARICELLA-ZOSTER VIRUS—PATHOLOGIC FEATURES

Gross Findings

- Trachea and bronchi are generally edematous and erythematous with occasional vesicles or ulcers on the mucosal surfaces
- The lungs are generally 2 to 3 times heavier than normal, firm, and "plum-colored"
- There are often multiple necrotic and hemorrhagic lesions on the pleura and in the lung parenchyma that resemble the pox lesions of skin

Microscopic Findings

- Interstitial pneumonitis and diffuse miliary foci of necrosis and hemorrhage in the pulmonary parenchyma
- Other findings may include alveolar collections of edema, fibrin, or hemorrhage, diffuse alveolar damage, and septal edema
- Virally infected cells with intranuclear inclusions may be identified in respiratory epithelial cells of the trachea and bronchi, pneumocytes, interstitial fibroblasts, or capillary endothelium
- Eosinophilic intranuclear inclusions and multinucleated syncytial cells may be difficult to locate but are best identified at the edges of necrotic foci
- In disseminated disease, similar necrotizing hemorrhagic lesions and occasional viral cytopathic changes are observed in other tissues and organs

Immunohistochemical Features

- IHC testing of formalin-fixed tissues is a sensitive method to confirm VZV infection and distinguish it from other viral infections, particularly HSV

Ultrastructural Features

- The enveloped viral particle is pleomorphic to spherical and 180-200 nm in diameter
- Viral particles are located within the nuclei of infected cells

Pathologic Differential Diagnosis

- HSV pneumonia (histology is identical)
- Adenovirus pneumonia
- Measles pneumonia
- CMV pneumonia

based on the antigenicity of their hemagglutinin (HA) and neuraminidase (NA) surface glycoproteins. Only one type of HA and one type of NA are recognized for influenza B. Influenza A occurs in both pandemic and interpandemic forms. The epidemiologic pattern of influenza in humans is related to two types of antigenic variation of its envelope glycoproteins, namely antigenic drift and antigenic shift. Fortunately, pandemics, defined as worldwide outbreaks of severe disease, occur infrequently and result from antigenic shift and emergence of new potentially pandemic influenza A viruses that possess a novel HA alone or in combination with a novel NA. Interpandemic influenza occurs virtually every year as a result of antigenic drift resulting from point mutations in the surface glycoproteins, and emergence of new strains related to those circulating in previous epidemics. This enables the virus to evade the immune system, leading to repeated outbreaks during interpandemic years.

Clinical Features

Influenza viruses are spread person-to-person primarily through the coughing and sneezing of infected persons. The typical incubation period is 1 to 4 days. Adults can be infectious from the day before symptoms begin through approximately 5 days after illness onset. Children can be infectious for 10 or more days, and young children can shed virus for several days before their illness onset. Severely immunocompromised persons can shed virus for weeks or months. Respiratory illness caused by influenza is difficult to distinguish from illnesses caused by other respiratory pathogens on the basis of symptoms alone. Uncomplicated influenza illness is characterized by the abrupt onset of constitutional and respiratory signs and symptoms including fever, myalgia, headache, malaise, nonproductive cough, sore throat, and rhinitis. Among children, otitis media, nausea, and vomiting are also commonly reported. Influenza typically resolves after 3 to 7 days in most patients, although cough and malaise can persist for more than 2 weeks. Complications include secondary bacterial pneumonias, febrile seizures, and, uncommonly, encephalopathy, transverse myelitis, Reye's syndrome, myositis, myocarditis, and pericarditis. The risks for complications, hospitalizations, and deaths from influenza are higher among persons aged 65 years or older, young children, and persons of any age with certain underlying health conditions, than among healthy older children and younger adults.

Radiologic Features

The main findings include unilateral or bilateral patchy consolidation of the lungs, which may progress to confluent lung disease. Pleural effusions are uncommon.

Pathologic Features

Gross Findings

Lungs in influenza virus pneumonia, not associated with a bacterial infection, can have different degrees of hemorrhage and edema. Airways can be filled with varying amounts of exudate, and the mucosae of the trachea and large bronchi are hyperemic and swollen. Cross-sections of the lungs show a more or less granular appearance, in which the lower lobes are more affected than the upper lobes. The gross pathologic features in secondary infections depend largely on the specific microbial (usually bacterial) pathogen involved. The mucosae of the large airways can demonstrate hyperemia, hemorrhages, or purulent necrotic debris. In the lungs, the extent of the pathologic process in the lower lobes is generally greater than the upper and may include consolidation, abscesses, hemorrhages, and empyema. Secondary inflammation in the regional lymph nodes may be present. Purulent mediastinitis and pericarditis may also be found in some cases.

Microscopic Findings

The histopathologic features of non-fatal and fatal influenza include necrotizing bronchitis, diffuse alveolar damage, hemorrhage, edema, and thrombi. The pathology is more prominent in larger bronchi, and inflammation may vary in intensity (Figure 13-7A-C). Viral inclusions cannot be identified by light microscopy (Figure 13-7F). Secondary bacterial infections with organisms such as *Streptococcus pneumoniae,* group A Streptococcus, *Staphylococcus aureus*, and *Haemophilus influenzae* may occur as a complication in about 50-75% of fatal cases and make it difficult to recognize the pathologic changes associated with the primary viral infection. The histopathologic features in other organs may include myocarditis, cerebral edema, rhabdomyolysis, and hemophagocytosis (Figures 13-8A,B).

Recent studies suggest that, unlike human influenza viruses, avian virus H5N1 preferentially infects cells in the lower respiratory tract of humans, resulting in extensive damage of the lungs with minimal pathology in the upper respiratory tract (Figures 13-8C,D). This may explain why the H5N1 avian influenza virus is so lethal to humans but so difficult to spread from person to person. These studies show that the avian virus preferentially binds to the α-2,3 galactose receptors, which are found only in and around the alveoli. This is in contrast to the human influenza viruses which preferentially bind to the α-2,6 receptors, which are found throughout the respiratory tract from the nose to the lungs.

Ancillary Studies and Differential Diagnosis

Because of the absence of any characteristic viral inclusions and because the overall pathologic features of influenza may resemble other viral, rickettsial, and certain bacterial infections, an unequivocal diagnosis can be made only by laboratory tests such as viral culture, direct fluorescent antibody and rapid antigen assays, serology, IHC, and ISH. IHC and ISH assays can demonstrate viral antigens and nucleic acids in epithelial cells of small airways (Figure 13-7D,E,G). Antigens are more readily identified in patients who die within 3 to 4 days of onset of illness..

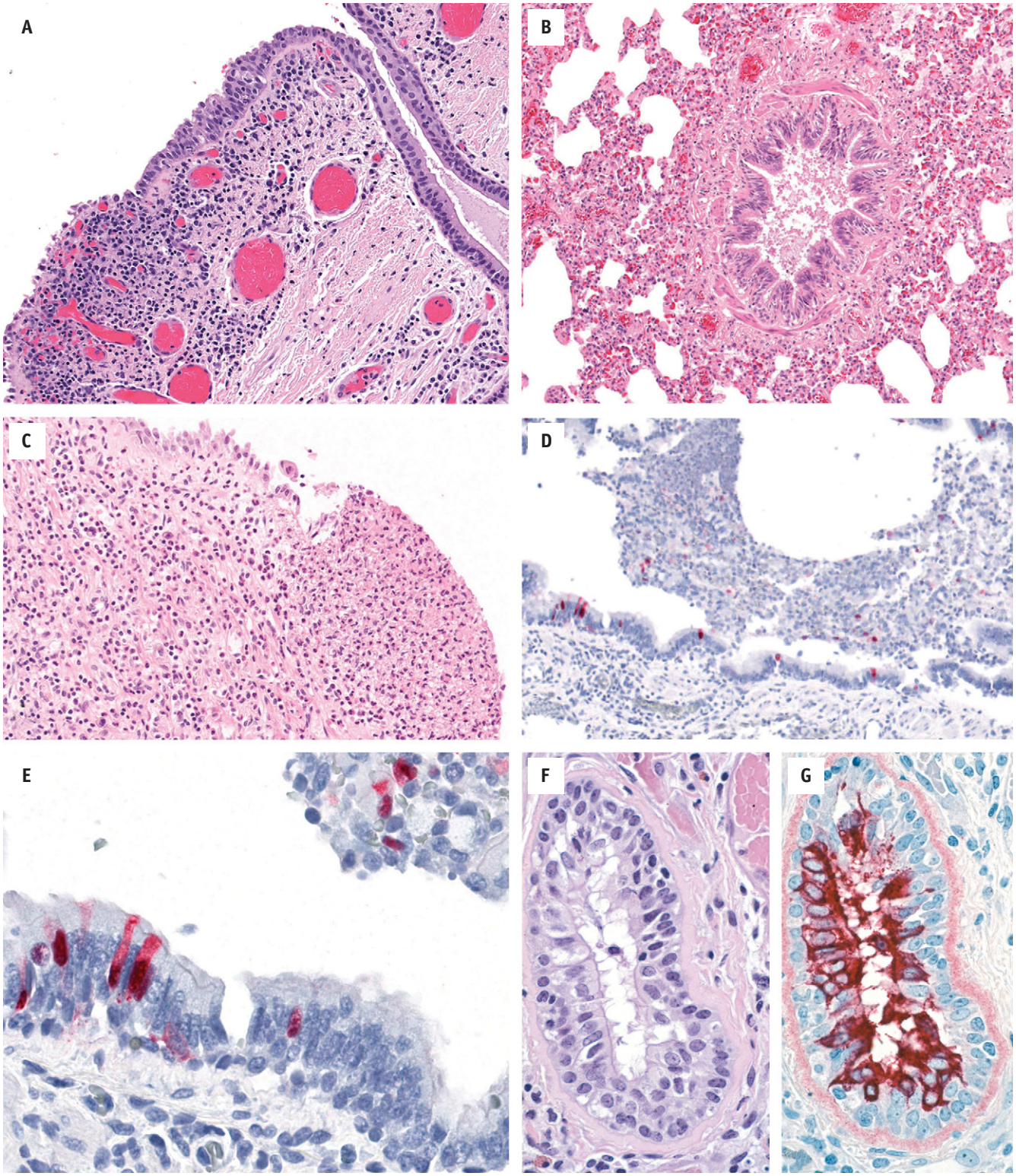

FIGURE 13-7

Influenza

(A) Trachea from a child with fatal influenza B showing congestion and predominantly mononuclear inflammatory cell infiltrates in the lamina propria and submucosa. **(B)** Bronchiole containing necrotic debris and surrounding congested lung parenchyma in a patient with fatal influenza B. **(C)** Ulcerated respiratory epithelium in a large airway of a patient with fatal influenza B. **(D, E)** Immunohistochemical staining of influenza B virus in the respiratory epithelium of an airway (immunoalkaline phosphatase). **(F, G)** Respiratory epithelial cells infected by influenza A, demonstrated with an immunohistochemical stain for hemagglutinin antigen **(G)** (immunoalkaline phosphatase).

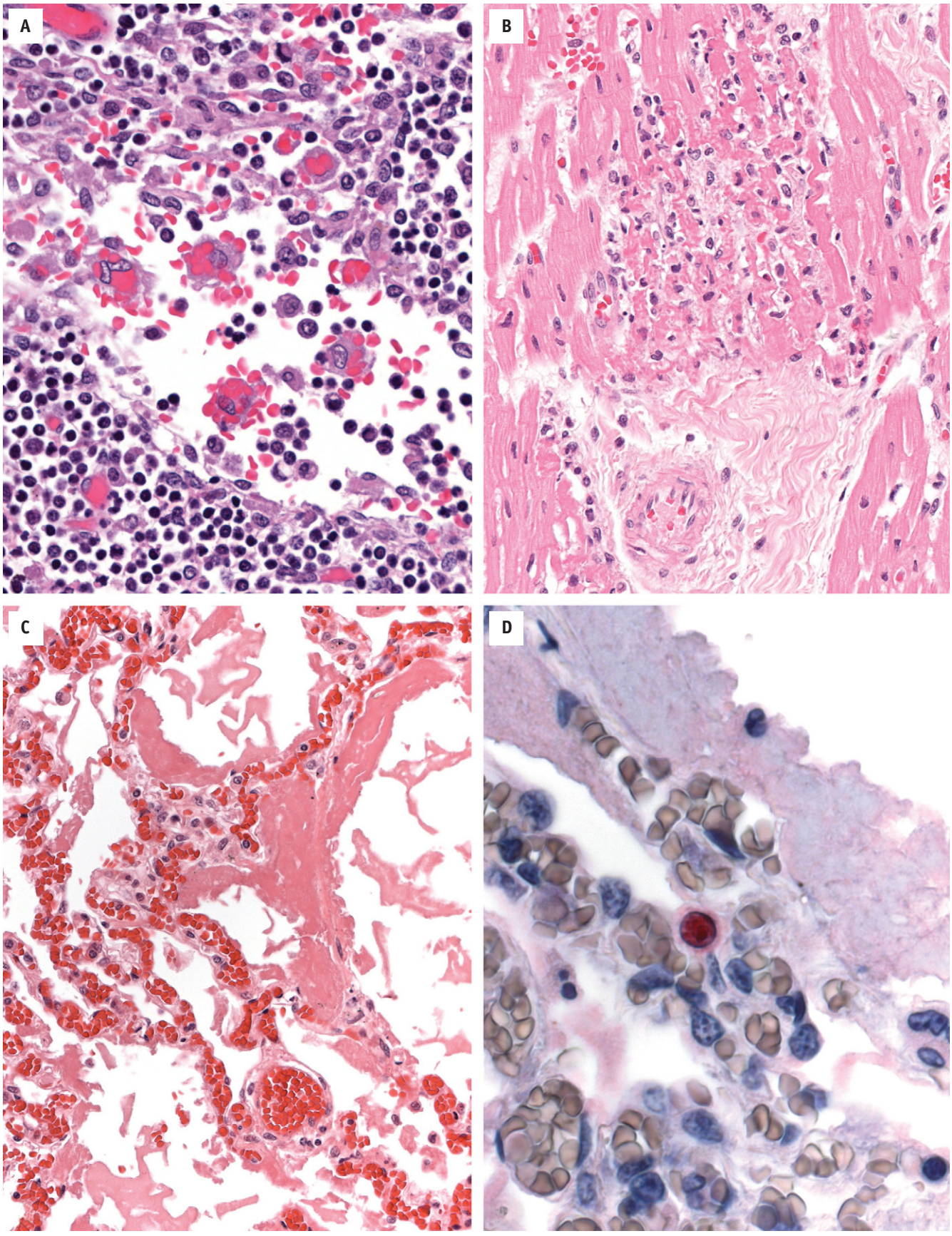

FIGURE 13-8

Influenza

(A) Hemophagocytosis in a peribronchial lymph node in a patient with fatal influenza B. **(B)** Focus of myocyte necrosis accompanied by a mixed inflammatory cell infiltrate in the heart of a patient with fatal influenza B. **(C)** Diffuse alveolar damage in a patient with fatal avian influenza (H5N1) showing hyaline membrane formation and congestion. **(D)** Influenza virus A (H5N1) antigens in the nucleus of a pneumocyte of a patient with fatal avian influenza (immunoalkaline phosphatase). Unlike other influenza viruses that cause disease in humans, H5N1 preferentially infects alveolar epithelial cells, and causes relatively minimal pathology in the upper respiratory tract.

Prognosis and Therapy

Vaccination is an important strategy in the prevention of influenza virus infections. Influenza is seldom fatal in the immunocompetent host, and recovery is usually complete. Supportive management with bed rest, hydration, and antipyretics is the basis of treatment. Antiviral agents may be helpful early in the course of illness. NA, a major antigenic determinant of influenza viruses, catalyses the cleavage of glycosidic linkages to sialic acid and the release of progeny virions from infected cells. Accordingly, it has become an important target for drug inhibitors such as oseltamivir and zanamivir. The M2 surface component and channel of influenza A (not present in influenza B virus) regulates the internal pH of the virus and is blocked by the antiviral drug amantidine.

MEASLES

Measles (rubeola) is an infectious, acute febrile viral illness characterized by upper respiratory tract symptoms, fever, and a maculopapular rash. The causative agent, a member of the genus *Morbillivirus*, of the family *Paramyxoviridae,* is an enveloped virus that contains a negative sense, single-stranded RNA genome. Measles has a worldwide distribution. Although still a significant problem in underdeveloped countries, measles infection became uncommon in the United States after the development and widespread use of an effective measles vaccine. However, a recrudescence of measles infection occurred in several large U.S. urban centers in recent years, associated with reduced use of the vaccine among children and young adults.

Clinical Features

Measles virus is highly contagious and is spread by aerosols and droplets from respiratory secretions of acute cases. Children are usually infected by 6 years of age, resulting in lifelong immunity, and almost all adults are immune either from vaccination or exposure. Clinical infection in children younger than 9 months of age is generally uncommon because of passive protection afforded the infant by the transfer of maternal antibodies, although occasional infections have occurred in this age group. A person with acute measles is infective from just before the onset of symptoms to the end of fever. After an incubation period of about 1 to 2 weeks, the prodromal phase of measles begins with fever, rhinorrhea, cough, and conjunctivitis. Koplik's spots, which are small, irregular red spots with a bluish-white speck in the center, appear on the buccal mucosa in 50-90% of cases shortly before rash onset. An erythematous maculopapular rash begins on the face 3 to 4 days after prodromal symptoms and usually spreads to the trunk and extremities. The symptoms gradually resolve, with the rash lasting for approximately 6 days, fading in the same order as it appeared.

Radiologic Features

Chest radiographs typically show fine reticular and ground-glass opacities as well as nodules and patchy consolidation. Bronchial thickening and peribronchial opacities may also be observed in some patients. Pleural effusions are rare.

Pathologic Features

Gross Findings

In fatal cases, the lungs are heavy and show congestion, hemorrhage, and edema. The gross pathologic features in secondary infections depend largely on the specific microbial (usually bacterial) pathogen involved.

Microscopic Findings

A focal or generalized interstitial pneumonitis, similar to that seen in many other viral infections, is seen in the lungs of measles patients. Histopathologic features include various degrees of peribronchial and interstitial mononuclear cell infiltrates, squamous metaplasia of bronchial endothelium, proliferation of type II pneumocytes, and intra-alveolar edema with or without mononuclear cell exudates and hyaline membranes. Secondary changes created by bacterial or viral superinfection or organizational changes, may alter the original pathology. The hallmark of the disease is the formation of multinucleated epithelial giant cells. These cells, which are often numerous, are formed by fusion of bronchiolar or alveolar lining cells (Figure 13-9A). These cells generally contain characteristic nuclear and cytoplasmic inclusions. The intranuclear inclusions are homogenous, eosinophilic, and surrounded by a slight indistinct halo (Figure 13-9B). The cytoplasmic inclusions are deeply eosinophilic and may form large masses with a "melted tallow" appearance (Figure 13-9B). These giant cells may undergo degenerative changes with progressive loss of cytoplasm, increasing basophilia, and shrinkage of nuclei. The presence of measles virus in these giant cells may be demonstrated by immunofluorescence, IHC, and ISH techniques (Figures 13-9C,D). These giant cells can also be seen in extrapulmonary tissues (Figure 13-9E).

INFLUENZA—FACT SHEET

Definition

- Influenza viruses belong to the *Orthomyxoviridae* family and include the two important influenza virus types, A and B, which are associated with significant human disease
- All influenza viruses have a segmented, negative-sense RNA core surrounded by a lipid envelope
- Influenza A viruses are further classified into subtypes based on the antigenicity of their hemagglutinin (HA) and neuraminidase (NA) surface glycoproteins
- There are 16 recognized HA subtypes and 9 NA subtypes of influenza A virus
- Only one type of HA and one type of NA are recognized for influenza B

Incidence and Location

- Worldwide distributions
- Influenza A occurs in both pandemic and interpandemic forms

Morbidity and Mortality

- Seldom fatal in the immunocompetent host, and recovery is usually complete
- Mortality and risk for complications from influenza are higher among persons aged 65 years or older, young children, and persons of any age with certain underlying health conditions
- Secondary bacterial pneumonias with organisms such as *Streptococcus pneumoniae*, group A streptococcus, *Staphylococcus aureus*, and *Haemophilus influenzae* may occur as a complication

Gender, Race, and Age Distribution

- People of all ages are vulnerable to influenza pneumonia
- No recognized gender or racial predilection

Clinical Features

- Spread primarily through the coughing and sneezing of infected persons
- Typical incubation period is 1 to 4 days
- Uncomplicated influenza illness is characterized by the abrupt onset of fever, myalgia, headache, malaise, nonproductive cough, sore throat, and rhinitis

Radiologic Features

- Unilateral or bilateral consolidation of the lungs
- Rarely associated with pleural effusions

Therapy and Prevention

- Vaccination is an important preventive strategy
- Supportive therapy, such as bed rest, oral hydration, and antipyretics
- Antivirals such as oseltamivir, zanamivir, and amantadine may be helpful early in the course of infection
- Specific antibiotic therapy in cases with secondary bacterial infection

INFLUENZA—PATHOLOGIC FEATURES

Gross Findings

- Airways show hyperemia, hemorrhage, and edema and may be filled with exudate
- Cross-sections of the lungs have a granular appearance, in which the lower lobes are more affected than the upper lobes
- Gross pathologic features in secondary infections depend largely on the specific microbial (usually bacterial) pathogen involved, and include consolidation, abscess formation, hemorrhage, and empyema

Microscopic Findings

- Necrotizing bronchitis and tracheitis
- Diffuse alveolar damage
- Thrombi
- Hemorrhage
- Edema
- Viral inclusions cannot be identified by light microscopy

Immunohistochemical Features

- IHC is extremely valuable for confirming infection
- Influenzaviral antigens are usually sparse and are primarily seen in the epithelial cells of larger airways
- Antigens are more readily identified in patients who die within 3 to 4 days of onset of illness

Ultrastructural Features

- Viral particles are pleomorphic (filamentous and spherical)
- A 10-12 nm layer of HA (rod-shaped) and NA (mushroom-shaped) spikes project radially from the surfaces of the influenza A and B viruses

Pathologic Differential Diagnosis

- A large number of viral, rickettsial, bacterial infections, as well as non-infectious diseases, may have similar histologic features
- Unequivocal diagnosis can be made by laboratory tests such as viral culture, direct fluorescent antibody and rapid antigen assays, serology and IHC

Ancillary Studies

Laboratory confirmation is useful to avoid possible confusion with other rash-causing illnesses. Diagnostic laboratory procedures consist of either direct detection of the virus or viral antigens, usually by indirect immunofluorescence or by serologic methods using hemagglutination inhibition, neutralization, or enzyme immunoassay. Specimens for serologic testing consist of acute and convalescent-phase serum pairs. The presence of specific IgM antibody can be used to diagnose recent infection. IHC and ISH can be performed on tissue specimens. Ultrastructurally, measles virions are pleomorphic, generally spherical, enveloped particles from 120-250 nm in diameter, with a lipid envelope surrounding a helical nucleocapsid composed of RNA and protein.

Differential Diagnosis

In typical cases, the diagnosis of measles can usually be made on the basis of clinical signs and symptoms. Other causes of a similar rash, but without other features of

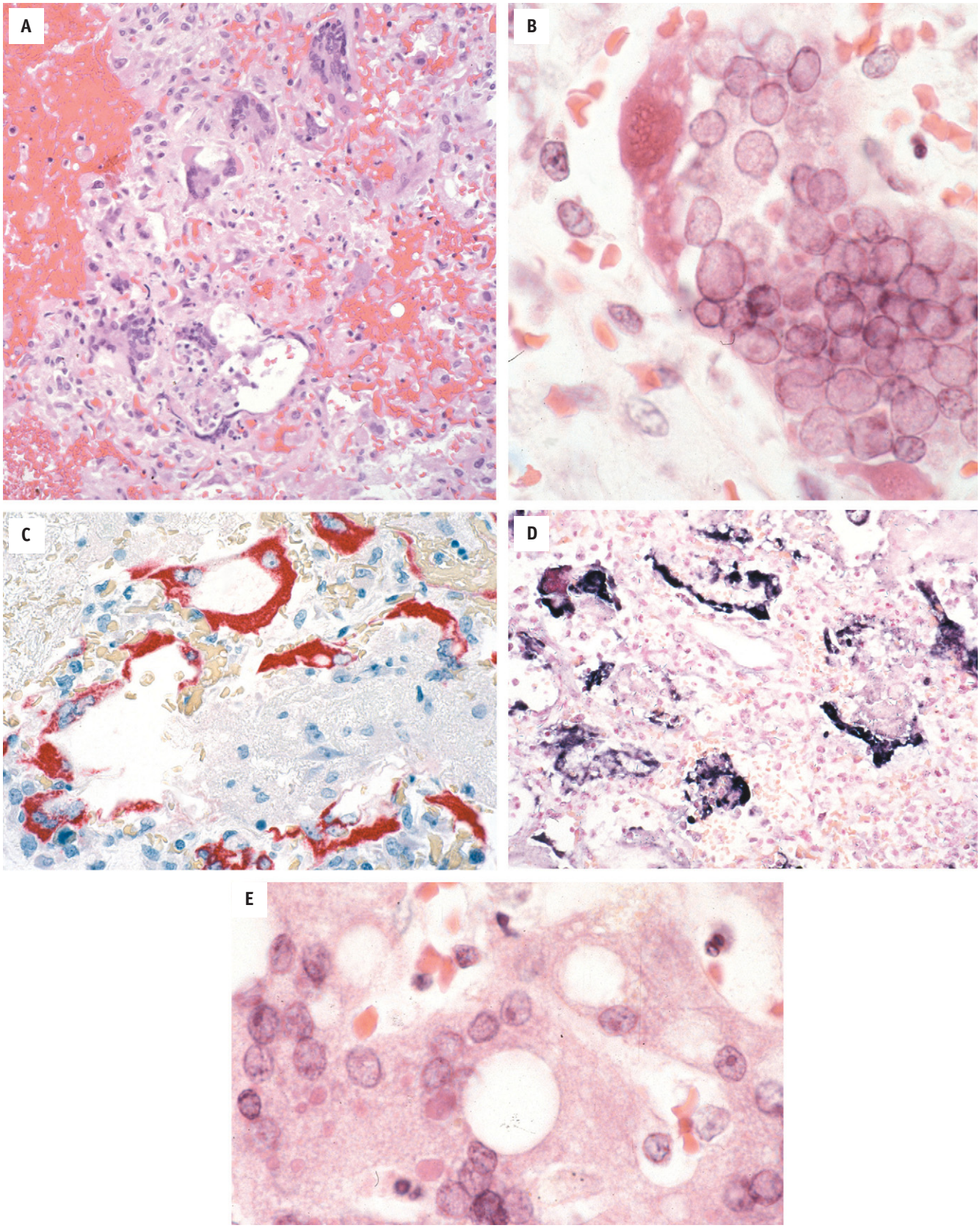

FIGURE 13-9

Measles pneumonia

(A) Multinucleated giant cells generally line the alveoli, although some are found lying free within alveolar spaces. **(B)** Multinucleated giant cell with eosinophilic cytoplasmic inclusions. The inclusions may be large and have a characteristic "melted tallow" appearance. Intranuclear inclusions are also seen, but are ill-defined, eosinophilic, and lack clear circumscription. **(C)** Viral antigens in the cytoplasm of giant cells, highlighted by immunohistochemistry (immunoalkaline phosphatase). **(D)** Numerous measles giant cells are seen by chromogenic in situ hybridization (digoxigenin-labeled probes followed by immunoalkaline phosphatase). **(E)** Multinucleated epithelial giant cells in the liver from a fatal case of measles.

measles, include rubella, dengue virus, enteroviruses, and drug reactions, especially to ampicillin. The histologic diagnosis is facilitated by the identification of the characteristic giant cells in a setting of interstitial pneumonitis. These giant cells are not seen in all cases of measles pneumonia, however, and their absence should not exclude the diagnosis. Furthermore, other viral pathogens, such as respiratory syncytial virus, parainfluenza, metapneumovirus, VZV, and the recently discovered Henipa viruses, may also give rise to pneumonias with giant cells and should be considered in the differential diagnosis. IHC or ISH testing can demonstrate viral antigens or nucleic acids in the majority of cases, assisting in the histologic diagnosis.

Prognosis and Therapy

Supportive therapy, such as bed rest, oral hydration, and antipyretics usually produces rapid and complete recovery. Immune globulins can be useful if treatment is given early in the infection. Vaccination can also be helpful in the treatment regimen if given within 3 days of exposure. In a smaller number of patients, complications can arise as a result of continued and progressive virus replication, bacterial or viral superinfections, or abnormal host immune response. The most common complications are secondary bacterial pneumonia and otitis media. In these settings, specific antibiotic therapy is administered. Other complications include febrile convulsions, encephalitis, liver function abnormalities, chronic diarrhea, and sinusitis. Several pulmonary and central nervous system syndromes that are often fatal have been described. Death occurs in about 1 of every 1000 measles cases; however, the risk of death and other complications is substantially increased in infants, malnourished and immunocompromised individuals, persons with underlying illnesses, and non-immunized populations in underdeveloped countries.

HUMAN PARAINFLUENZA VIRUSES

Human parainfluenza viruses (HPIVs) are second only to respiratory syncytial virus (RSV) as a cause of lower respiratory tract disease in young children. HPIVs are negative-sense, nonsegmented, single-stranded, enveloped RNA viruses that possess fusion and hemagglutinin-neuraminidase glycoprotein "spikes" on their surface. The four serotypes of HPIV belong in the family *Paramyxoviridae*, subfamily *Paramyxovirinae*, and genera *Respirovirus* (HPIV-1 and -3) and *Rubulavirus* (HPIV-2 and -4).

Clinical Features

HPIVs are spread from respiratory secretions through close contact with infected persons or contact with contaminated surfaces or objects. Infection can occur when infectious material contacts mucous membranes of the eyes, mouth, or nose, and possibly through the inhalation of droplets generated by a sneeze or cough. HPIVs are ubiquitous and infect most people during childhood. Serologic surveys have shown that 90-100% of children aged 5 years and older have antibodies to HPIV-3, and about 75% have antibodies to HPIV-1 and -2. The different HPIV serotypes differ in clinical presentations, with HPIV-1 and HPIV-2 most frequently associated with outbreaks of croup and HPIV-3 more often associated with bronchiolitis and pneumonia. HPIV-4 is infrequently detected, possibly because it is less likely to cause severe disease. The incubation period is generally from 1 to 7 days. The HPIVs can also cause repeated infections throughout life, usually manifested by an upper respiratory tract illness (e.g., cold and sore throat). Serious lower respiratory tract disease (e.g., pneumonia, bronchitis, and bronchiolitis) can also occur with repeat infection, especially among the elderly, and among patients with compromised immunity.

Radiologic Features

The main findings associated with HPIV pneumonia include diffuse interstitial opacities, bronchial wall thickening, and peribronchial consolidation. Infection is rarely associated with pleural effusions.

Pathologic Features

Gross Findings

In fatal cases, the lungs are typically heavy and display congestion, hemorrhage, and edema.

Microscopic Findings

In patients with severe HPIV infection, multinucleated giant cells derived from the respiratory epithelium may be seen in association with an interstitial pneumonitis, diffuse alveolar damage, bronchiolitis, and organizing changes (Figure 13-10A,B). These giant cells, which may contain intracytoplasmic eosinophilic inclusions (Figure 13-10B), have also been reported in extrapulmonary tissues such as kidney, bladder, and pancreas.

MEASLES—FACT SHEET

Definition

- Measles virus is a single-stranded RNA virus and a member of the genus *Morbillivirus*, of the family *Paramyxoviridae*

Incidence and Location

- Highly communicable disease of worldwide distribution
- A significant problem in underdeveloped countries
- Uncommon infection in the United States after the widespread use of the vaccine

Morbidity and Mortality

- The most common complications are secondary bacterial pneumonia and otitis media
- Death occurs in about 1 of every 1,000 patients with measles
- The risk of death and other complications is substantially increased in infants, malnourished and immunocompromised individuals, persons with underlying illnesses, and non-immunized populations

Gender, Race, and Age Distribution

- Infection in children younger than 9 months of age is uncommon because of passive protection from immune mothers
- Children (non-vaccinated) are usually infected by 6 years of age
- Natural infection results in lifelong immunity and almost all adults are immune either due to exposure or vaccination
- No recognized gender or racial predilection

Clinical Features

- Typical incubation period is 1 to 2 weeks
- Brief prodrome characterized by fever, rhinorrhea, cough, and conjunctivitis
- Koplik's spots can be seen in the buccal mucosa shortly before rash onset
- An erythematous maculopapular rash begins on the face 3 to 4 days after prodromal symptoms and usually spreads to the trunk and extremities
- The rash lasts for approximately 6 days, fading in the same order as it appeared

Radiologic Features

- Fine reticular and ground-glass opacities in the lungs
- Nodules and patchy consolidation throughout the lungs may be seen
- Rarely associated with pleural effusions

Prognosis and Therapy

- Recovery is rapid and complete in most cases
- Supportive therapy is administered, such as bed rest, oral hydration, and antipyretics
- Immune globulins can be useful if treatment is given early in the infection
- Vaccination can also be helpful if given within 3 days of exposure

MEASLES—PATHOLOGIC FEATURES

Gross Findings

- Lungs are typically heavy, congested, hemorrhagic, and edematous
- Gross findings in cases with secondary infections depend largely on the specific microbial (usually bacterial) pathogen involved, and may include consolidation, abscess formation, hemorrhage, and empyema

Microscopic Findings

- Interstitial pneumonitis with mononuclear cell infiltrates
- Diffuse alveolar damage
- Multinucleated giant cells with characteristic nuclear and cytoplasmic inclusions

Immunohistochemical Features

- Measles antigens can be detected in giant cells and alveolar lining cells

Ultrastructural Features

- Measles virions are pleomorphic, generally spherical, enveloped particles from 120-250 nm in diameter
- A lipid envelope surrounds a helical nucleocapsid composed of RNA and protein

Pathologic Differential Diagnosis

- Other viral pathogens that can cause giant cell pneumonia, such as respiratory syncytial virus, parainfluenza, metapneumovirus, VZV, and Henipa viruses
- Unequivocal diagnosis can be made by laboratory tests such as viral culture, direct fluorescent antibody and rapid antigen assays, serology, and IHC

Ancillary Studies

Diagnosis of infection with HPIVs can be made by virus isolation, direct detection of viral antigens by EIA or IFA in clinical specimens, detection of viral RNA by RT-PCR, demonstration of a rise in specific serum antibodies, or a combination of these approaches. HPIV infections of lung can be confirmed by IHC testing of formalin fixed tissues (Figure 13-10C); viral antigens can be detected in giant cells, pneumocytes, and respiratory epithelial cells. Ultrastructural studies demonstrate variably shaped virions of varying size (ranging from 150-300 nm), with a lipid envelope surrounding a helical nucleocapsid composed of RNA and protein.

Differential Diagnosis

Other viral causes of giant cell pneumonia, including measles and RSV, should be considered in the histopathologic differential diagnosis, and laboratory testing, including IHC, can be useful in determining the correct diagnosis.

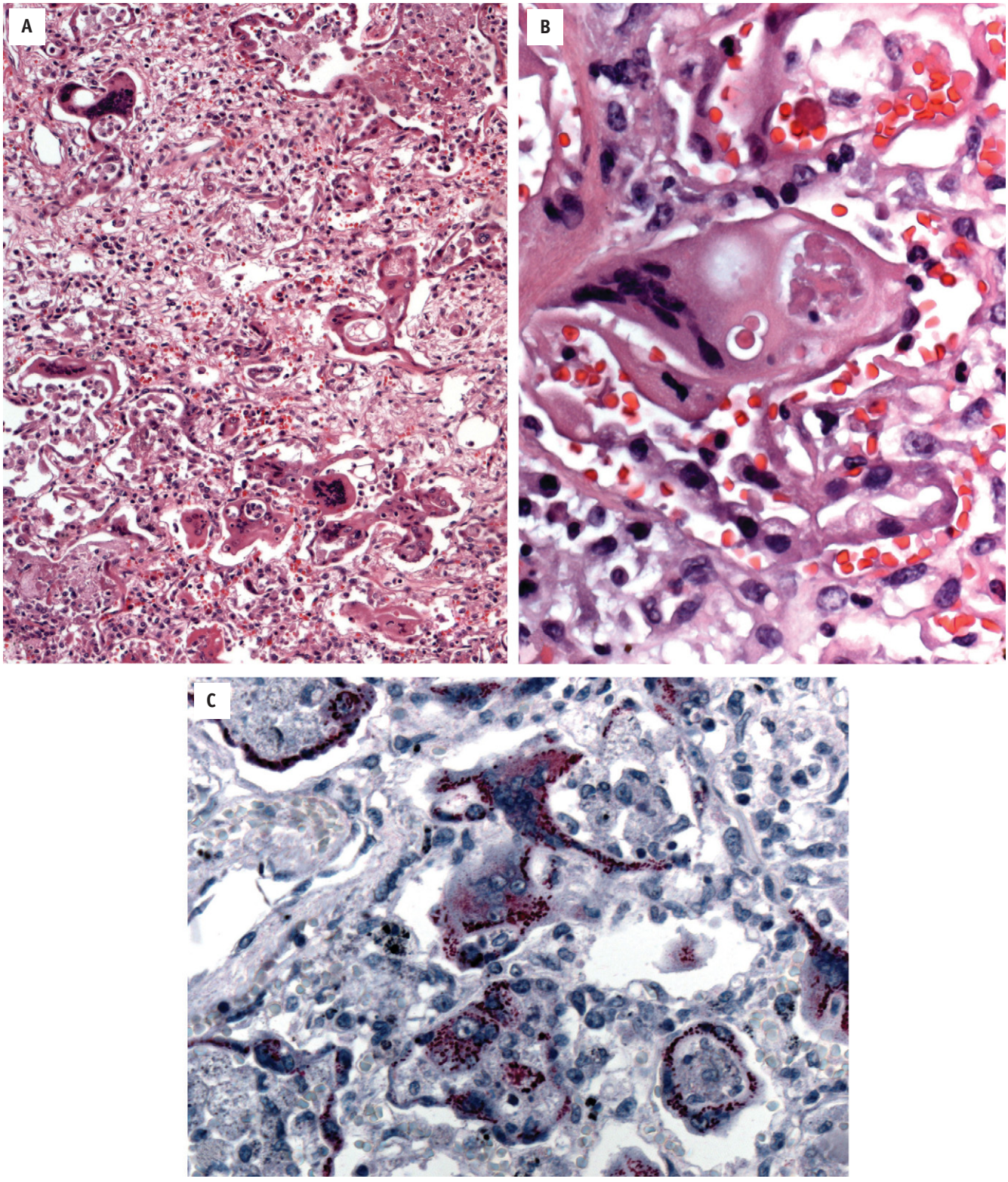

FIGURE 13-10

Parainfluenza virus pneumonia

(A) Giant cell pneumonia showing numerous giant cells, interstitial pneumonitis, and hemorrhage. **(B)** Giant cell containing eosinophilic cytoplasmic inclusions. **(C)** Parainfluenza virus antigens in giant cells localized by IHC (immunoalkaline phosphatase).

Prognosis and Therapy

Most HPIV infections cause a mild, self-limited illness. The highest rates of serious HPIV illnesses occur among young children. HPIV infections are also being increasingly recognized as an important cause of severe morbidity and mortality in immunocompromised adults. The mortality of bone marrow transplant patients with HPIV-3 infection has been reported to be as high as 60% . Supportive management with bed rest, oral hydration, and antipyretics is the basis of treatment. Aerosolized ribavirin has shown some efficacy in the treatment of severe cases of HPIV infection.

RESPIRATORY SYNCYTIAL VIRUS

RSV is a negative-sense, nonsegmented, single-stranded, enveloped RNA virus. RSV is a member of the family *Paramyxoviridae,* and can be further distinguished genetically and antigenically into two subgroups, A and B. The subgroup A strains are usually associated with more severe infections.

Clinical Features

RSV is the most common cause of bronchiolitis and pneumonia among infants and children under 1 year of age. In temperate climates, RSV infections usually occur during annual community outbreaks, often lasting several months, during the late fall, winter, or early spring months. The timing and severity of outbreaks in a community vary from year to year. RSV spreads efficiently during the annual outbreaks, infecting as many as 50% of children in their first year of life. Most children will have serologic evidence of RSV infection by 2 years of age. Illness begins most frequently with fever, runny nose, cough, and sometimes wheezing. During their first RSV infection, between 25% and 40% of infants and young children have signs or symptoms of bronchiolitis or pneumonia, and 0.5-2% require hospitalization. The majority of children hospitalized for RSV infection are under 6 months of age, or are children with cyanotic congenital heart disease, cystic fibrosis, bronchopulmonary dysplasia, or immunosuppression. RSV can also cause repeated infections throughout life, and severe lower respiratory tract disease may occur at any age, especially among the elderly or among those with compromised cardiac, pulmonary, or immune systems.

Radiologic Features

Children with RSV infection most commonly show multifocal air space consolidation and peribronchial thickening. In adults, disease is characterized by bilateral interstitial opacities and multifocal consolidations.

Pathologic Features

Gross Findings

Large and small airways can contain necrotic debris and mucus, and may show ulceration. The lungs may be heavy and diffusely firm and may show areas of hyperexpansion or atelectasis.

Microscopic Findings

The major histopathologic changes described in fatal RSV infections are necrotizing bronchiolitis and interstitial pneumonia. Bronchial lumens and airways are usually filled with necrotic debris and inflammatory cells. Airways show mixed or predominantly mononuclear infiltrates with hyperplastic epithelial changes (Figure 13-11A). These findings may be accompanied by diffuse alveolar damage (Figure 13-11E), and secondary bacterial superinfection. Giant cell pneumonia is seen in some cases (Figure 13-11C). The multinucleated giant cells represent epithelial cells in bronchi, bronchioles, and alveoli, and sometimes contain irregular, intracytoplasmic, eosinophilic inclusions surrounded by a clear halo (Figures 13-11C,D).

Ancillary Studies

Diagnosis of RSV infection can be made by virus isolation, direct detection of viral antigens in clinical specimens by EIA, IFA, or IHC (Figure 13-11B, F), detection of viral RNA by RT-PCR, or demonstration of a rise in RSV-specific serum antibodies. The virus is labile and attempts at culture isolation are often unsuccessful if there is delay or mishandling of the clinical specimen. Ultrastructural studies reveal virions of variable shape and size that range from 120 to 300 nm, with numerous 12-nm glycoprotein spikes.

Differential Diagnosis

Other viral causes of giant cell pneumonia should be considered in the histopathologic differential diagnosis, primarily parainfluenza viruses and measles viruses. Herpes

HUMAN PARAINFLUENZA VIRUSES—FACT SHEET

Definition

- The causative agents are negative-sense, nonsegmented, single-stranded, enveloped RNA viruses of the family *Paramyxoviridae*
- Four serotypes exist: HPIV-1 and 3 are included in the genus *Respirovirus*, and HPIV-2 and through HPIV-4 are included in the genus *Rubulavirus*

Incidence and Location

- Highly communicable disease of worldwide distribution
- Activity varies with serotype: HPIV-1 and 2 peak in the fall, HPIV-3 peaks in the spring and summer, and HPIV-4 is infrequently detected (less likely to cause severe illness)
- Common cause of upper respiratory illness, but an uncommon cause of lower respiratory tract disease

Morbidity and Mortality

- The highest rates of serious HPIV respiratory illnesses occur among young children
- HPIV infections are also an important cause of severe morbidity and mortality in immunocompromised adults

Gender, Race, and Age Distribution

- HPIVs infect most people by 5 years of age, and can cause repeated infections throughout life
- No recognized gender or racial predilection

Clinical Features

- Usually manifests as an upper respiratory illness (cold, sore throat, and croup) with a typical incubation period of 1 to 7 days
- Less commonly presents with symptoms of a lower respiratory illness (pneumonia, bronchitis, and bronchiolitis) in elderly or immunosuppressed patients

Radiologic Features

- Interstitial opacities, bronchial wall thickening, and peribronchial consolidation
- Rarely associated with pleural effusions

Prognosis and Therapy

- Recovery is rapid and complete in most cases
- Supportive management with bed rest, oral hydration, and antipyretics
- Aerosolized ribavirin can be used in immunosuppressed patients with severe illness

HUMAN PARAINFLUENZA VIRUSES—PATHOLOGIC FEATURES

Gross Findings

- Lungs can appear heavy, congested, hemorrhagic, and edematous

Microscopic Findings

- Interstitial pneumonitis with mononuclear cell infiltrates
- Diffuse alveolar damage
- Bronchiolitis
- Organizing pneumonia
- Multinucleated giant cells with characteristic cytoplasmic inclusions

Immunohistochemical Features

- HPIV antigens can be detected in giant cells and alveolar lining cells

Ultrastructural Features

- The virions are variable in shape and size, ranging from 150-300 nm
- A lipid envelope surrounds a helical nucleocapsid composed of RNA and protein
- The virus is morphologically indistinguishable from other members of the *Paramyxoviridae* family when viewed by negative contrast electron microscopy

Pathologic Differential Diagnosis

- Other viral pathogens that can cause giant cell pneumonia, such as measles, respiratory syncytial virus, metapneumovirus, VZV, and Henipa viruses
- Unequivocal diagnosis can be made by laboratory tests such as viral culture, direct fluorescent antibody and rapid antigen assays, serology, and IHC

simplex and varicella zoster viruses less commonly produce multinucleated giant cells in the lung.

Prognosis and Therapy

Mortality in otherwise healthy children hospitalized for RSV pneumonia is less than 1%. However, the disease is fatal in as many as 15-40% of patients with immune suppression or underlying disease. Mortality is greatest in infants with congenital heart disease and pulmonary hypertension, where it approaches 70%. At present, the only antiviral drug with *in vitro* efficacy against RSV is ribavirin.

HUMAN METAPNEUMOVIRUS

Human metapneumovirus (HMPV), first identified in 2001 from clinical specimens obtained from patients with acute respiratory illnesses, is a negative-sense, non-segmented, single-stranded, enveloped RNA virus. HMPV has been categorized in the family *Paramyxoviridae*, subfamily *Pneumovirinae*, genus *Metapneumovirus*. HMPV can be further distinguished genetically and antigenically into two subgroups, A and B.

Clinical Features

HMPV infection is ubiquitous and occurs during infancy and early childhood, with annual epidemic peaks occurring late in the winter and spring months in temperate

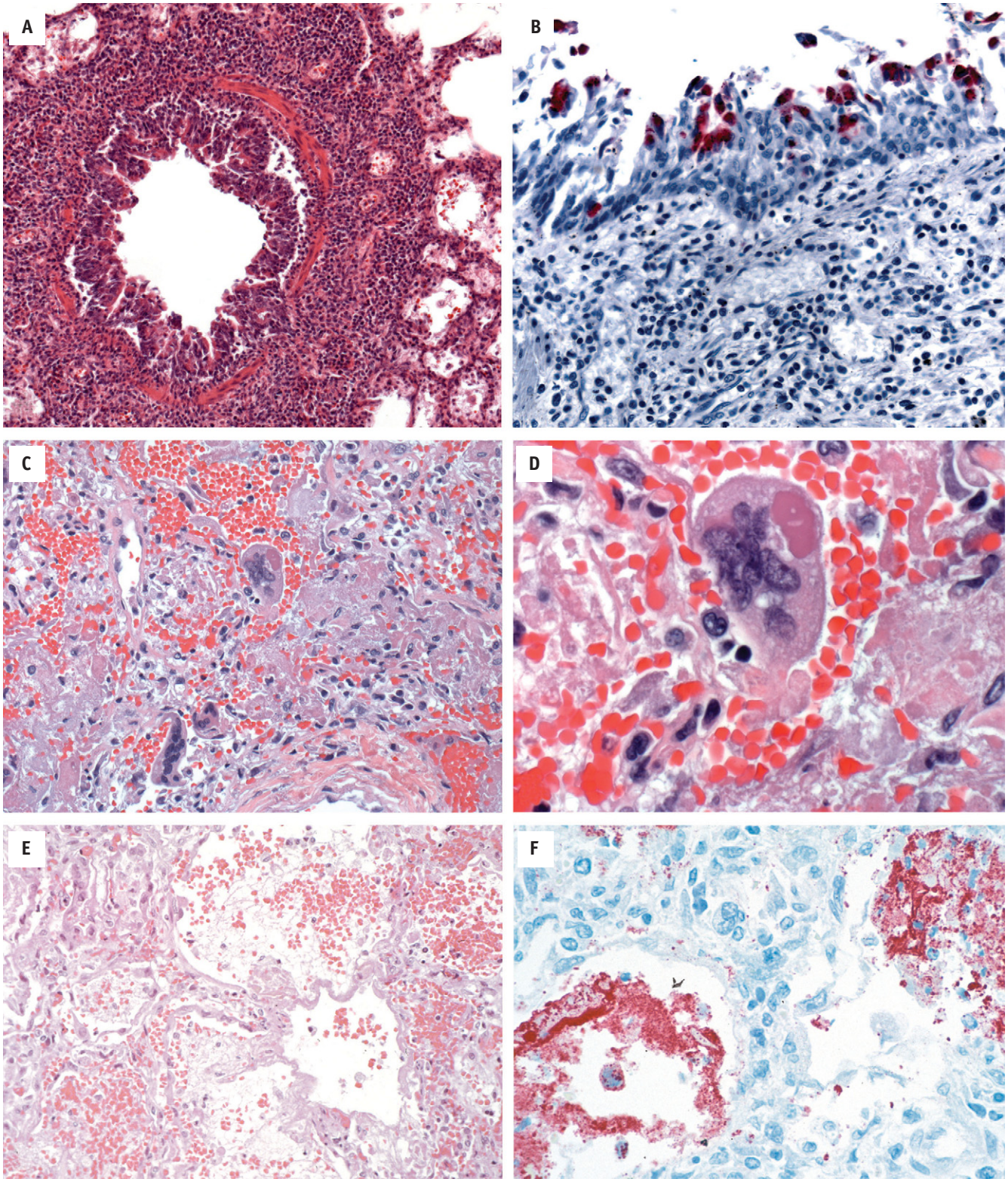

FIGURE 13-11

Respiratory syncytial virus pneumonia

(A) Bronchiolar and peribronchiolar inflammation in RSV infection. **(B)** RSV antigens in the lining epithelial cells of the same bronchiole, highlighted by IHC (immunoalkaline phosphatase). **(C)** Interstitial pneumonitis, hemorrhage, and giant cells in RSV pneumonia. **(D)** Giant cell with "melted tallow"-like inclusion similar to those of measles. **(E)** Diffuse alveolar damage in RSV pneumonia. **(F)** RSV antigens associated with hyaline membranes and necrotic debris (immunoalkaline phosphatase).

RESPIRATORY SYNCYTIAL VIRUS PNEUMONIA—FACT SHEET

Definition

- Respiratory syncytial virus (RSV) is a negative-sense, nonsegmented, single-stranded, enveloped RNA virus that is a member of the family *Paramyxoviridae*

Incidence and Location

- Worldwide distribution
- RSV spreads efficiently among children during annual outbreaks, infecting as many as 50% of children in their first year of life
- Most children will have serologic evidence of RSV infection by 2 years of age
- 0.5-2% of infants and young children infected with RSV are hospitalized
- Most children hospitalized for RSV infection are under 6 months of age or have cyanotic congenital heart disease, cystic fibrosis, bronchopulmonary dysplasia, or immunosuppression

Mortality

- Mortality in otherwise healthy children hospitalized for RSV pneumonia is less than 1%
- RSV pneumonia is fatal in as many as 15-40% of patients with immune suppression or underlying disease
- Mortality is greatest in infants with congenital heart disease associated with pulmonary hypertension, where it approaches 70%

Gender, Race, and Age Distribution

- RSV is the most common cause of bronchiolitis and pneumonia among infants and children under 1 year of age
- No recognized gender or racial predilection

Clinical Features

- Fever, runny nose, cough, sometimes wheezing
- During their first RSV infection, between 25% and 40% of infants and young children have signs or symptoms of bronchiolitis or pneumonia
- RSV can cause repeated infections throughout life, usually associated with moderate-to-severe cold-like symptoms
- Severe lower respiratory tract disease may occur at any age, especially among the elderly or among those with compromised cardiac, pulmonary, or immune systems

Radiologic Features

- Children most commonly show multifocal air space consolidation and peribronchial thickening
- Bilateral interstitial opacities and multifocal consolidations are usually seen in adults

Prognosis and Therapy

- Otherwise healthy children usually recover completely
- Higher mortality in specific patient groups (see above)
- The only antiviral drug with in vitro efficacy against RSV is ribavirin

RESPIRATORY SYNCYTIAL VIRUS PNEUMONIA—PATHOLOGIC FEATURES

Gross Findings

- Large and small airways can contain necrotic debris and mucus and may show ulceration
- The lungs may be heavy and firm, and show areas of hyperexpansion or atelectasis

Microscopic Findings

- Necrotizing bronchiolitis
- Interstitial pneumonia
- Hyperplastic airway epithelium
- Multinucleated giant cells (in some cases) in bronchi, bronchioles, and alveoli, that contain irregular, intracytoplasmic, eosinophilic inclusions surrounded by a clear halo

Immunohistochemical Features

- Viral antigens in multinucleated cells and respiratory epithelial cells

Ultrastructural Features

- The virion is variable in shape and size and ranges from 120 nm to 300 nm
- Particles show numerous 12-nm glycoprotein spikes

Pathologic Differential Diagnosis

- Other viral causes of giant cell pneumonia should be considered, primarily parainfluenza viruses and measles viruses
- Herpes simplex and varicella zoster viruses rarely produce multinucleated cytologic changes in the lung
- Confirmation of diagnosis is by clinical laboratory tests on IHC

regions, often overlapping in part or in whole with the annual RSV epidemic. Seroprevalence studies reveal that 25% of all children aged 6 to 12 months have antibodies to HMPV; by age 5 years, 100% of patients have evidence of past infection. The incubation period is generally from 2 to 8 days. Although HMPV has been associated with a spectrum of respiratory disease, most infections cause a mild, self-limited illness. The patient may be asymptomatic, or symptoms may range from mild upper respiratory tract illness to severe bronchiolitis and pneumonia. During their first HMPV infection, about 10-15% of infants and young children have signs or symptoms of bronchiolitis or pneumonia. About one-half of the cases of lower respiratory illness in children occur in the first 6 months of life, suggesting that young age is a major risk factor for severe disease. Underlying pulmonary disease, especially asthma, may increase the risk of hospitalization for HMPV pneumonia. Like RSV and the HPIVs, studies suggest that HMPV may also contribute to respiratory disease in elderly adults and the immunocompromised.

Radiologic Features

Radiographic findings include interstitial infiltrates with focal consolidation commonly involving the lower lobes of the lung.

Pathologic Features

Gross Findings

In fatal cases, the lungs are typically heavy and display congestion, hemorrhage, and edema.

Microscopic Findings

Histopathologic descriptions are few, and assessment of their validity is complicated by the uncertainty, in some cases, of the clinical significance of detecting this ubiquitous virus. Nonetheless, BAL specimens collected from patients within a few days of a positive HMPV assay show degenerative changes and cytoplasmic inclusions within epithelial cells, multinucleated giant cells, and histiocytes. The intracytoplasmic inclusions are ill-defined, eosinophilic structures that measure 3-4 μm. Necrotizing bronchiolitis may be found on lung biopsy. Lung tissue later in the disease shows chronic airway inflammation, intra-alveolar foamy and hemosiderin-laden macrophages, acute and organizing lung injury, and organizing pneumonia (Figure 13-12A-C). In such cases, typical multinucleated giant cells or viral inclusions cannot be identified. ISH studies on a limited number of human cases suggest infection of alveolar and bronchial epithelial cells.

Ancillary Studies

HMPV is difficult to identify with commonly used viral diagnostic procedures. The virus replicates slowly in primary and tertiary monkey kidney cell lines, and cytopathic effects can be difficult to discern.

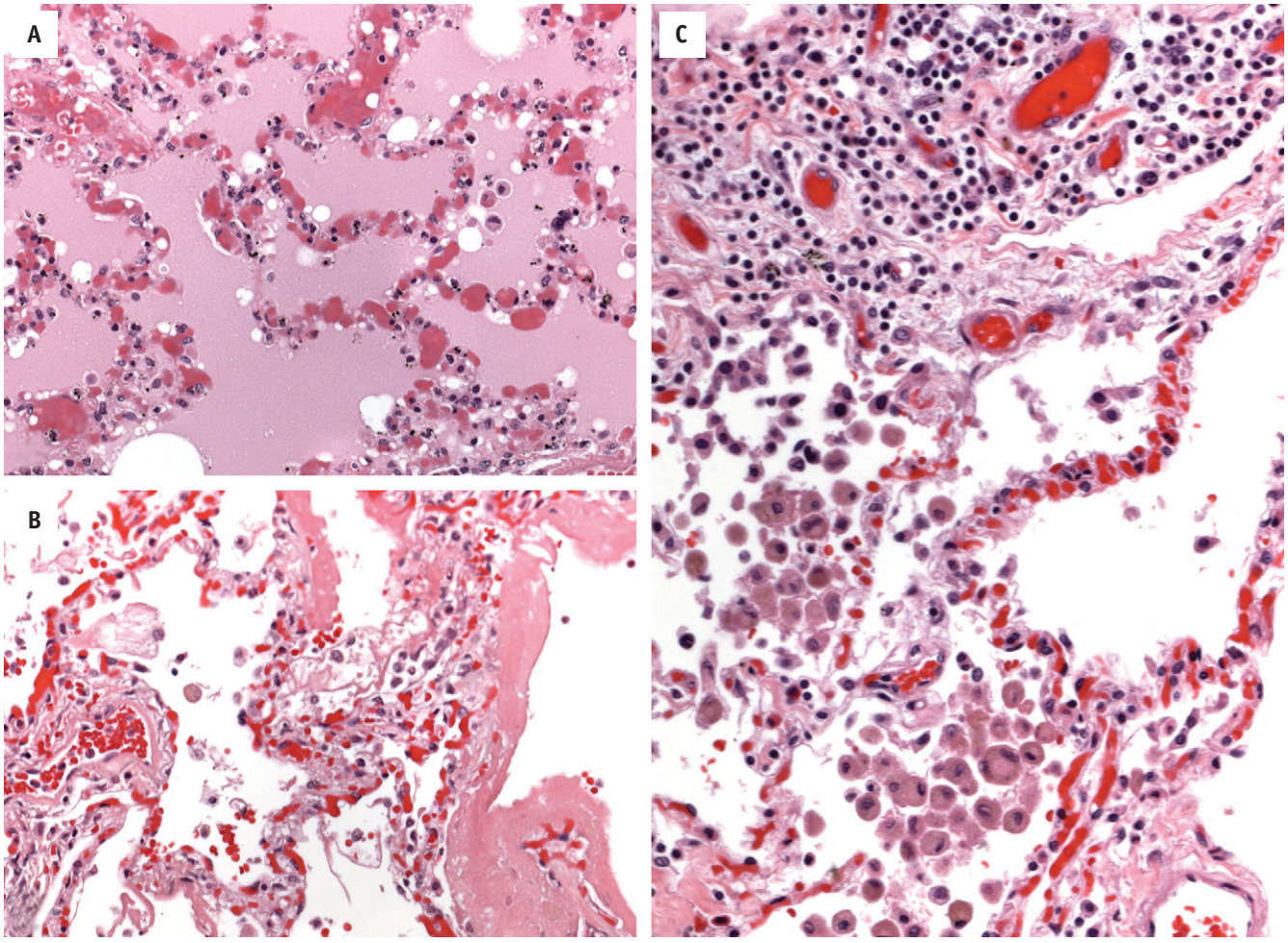

FIGURE 13-12

Human metapneumovirus pneumonia

(A) Severe pulmonary edema in fatal HMPV infection. **(B)** Diffuse alveolar damage in fatal HMPV infection. **(C)** Accumulation of alveolar macrophages in interstitial pneumonia associated with HMPV.

Antibodies to HMPV are not widely available; however, they can be used for identification of the virus by IFA. Most HMPV studies have been conducted using RT-PCR assays or by demonstration of a rise in HMPV-specific serum antibodies. The enveloped virion is variable in shape and size and ranges from 150 to 300 nm.

Differential Diagnosis

Other viral causes of giant cell pneumonia and diffuse alveolar damage, including measles, RSV, HPIV, measles, VZV, and HSV, may be considered, as well as noninfectious causes of diffuse alveolar damage. Laboratory testing, including IHC and ISH, can be useful in making this differentiation possible.

Prognosis and Therapy

Supportive management with bed rest, oral hydration, and antipyretics is the basis of treatment, and usually leads to complete recovery. There are no licensed therapies or prophylactic treatments for HMPV at this time. Ribavirin and intravenous immunoglobulin, which have activity against RSV, were tested against HMPV *in vitro* and were found to have equivalent activity against HMPV and RSV.

HENIPA VIRUSES

Hendra and Nipah viruses belong to the recently designated genus *Henipavirus* within the family *Paramyxoviridae*, subfamily *Paramyxovirinae*, and are nonsegmented, negative-stranded RNA viruses. These zoonotic pathogens were first identified in Australia and Malaysia and have been associated with acute febrile encephalitis and respiratory tract disease.

Clinical Features

Hendra was identified in 1994 when patients who came in close contact with sick horses developed an influenza-like illness with fever, myalgia, headache, lethargy, sore throat, nausea and vomiting. Two patients died with pneumonitis and multiorgan failure. The closely related Nipah virus was identified during an outbreak in Malaysia and Singapore during 1998-1999 that included more than 250 patients. The incubation period ranged from 2 days to 1 month, but in most cases lasted between 1 and 2 weeks. Patients presented with a severe acute encephalitic syndrome, but some also had significant pulmonary manifestations. In Bangladesh in 2001 and 2003, outbreaks of Nipah encephalitis occurred. Similar to the Malaysian outbreak, the most prominent symptoms were fever, headache, vomiting, and an altered level of consciousness. Respiratory illness was much more common in the Bangladesh cases, however, with 64% having cough and dyspnea. Epidemiologic and laboratory investigations identified fruit bats of the *Pteropus* genus as asymptomatic carriers of Hendra and Nipah viruses and possible animal reservoirs. Food-borne transmission has also been reported in an individual who consumed fruit contaminated by *Pteropus* bats.

Radiologic Features

In patients with respiratory illness, chest radiographs reveal bilateral infiltrates consistent with ARDS.

Pathologic Features

Gross Findings

In fatal infections, the lungs are heavy, congested, edematous, and hemorrhagic.

Microscopic Findings

Histopathologic findings in fatal cases of Hendra and Nipah infections are similar, with varying degrees of central nervous system and respiratory tract involvement. Findings include a systemic vasculitis with extensive thrombosis, endothelial cell damage, necrosis, and syncytial giant cell formation in affected vessels (Figures 13-13A,B). Multinucleated giant cells with intranuclear inclusions can occasionally be seen in lung, spleen, lymph nodes, and kidneys. In the lung, vasculitis and fibrinoid necrosis can be seen in the majority of cases (Figure 13-13A). Multinucleated giant cells with intranuclear inclusions are usually noted in alveolar spaces adjacent to necrotic areas (Figure 13-13C).

Ancillary Studies and Differential Diagnosis

The diagnosis of Nipah virus infection, suspected by patient history and clinical manifestations, can be supported by characteristic histopathological findings. The

HUMAN METAPNEUMOVIRUS (HMPV)—FACT SHEET

Definition

- Human metapneumovirus (HMPV) is a negative-sense, nonsegmented, single-stranded, enveloped RNA virus, that is a member of the family *Paramyxoviridae*, subfamily *Pneumovirinae*, genus *Metapneumovirus*
- HMPV can be distinguished genetically and antigenically into two subgroups, A and B

Incidence and Location

- Worldwide distribution
- About 25% of all children aged 6 to 12 months and all children by age 5 years will have serologic evidence of HMPV infection
- Annual epidemic peaks occur late in the winter and spring months in temperate regions, often overlapping in part or in whole with RSV epidemics
- About 4% of all infant hospitalizations for acute respiratory illness or fever are associated with HMPV; the majority of children hospitalized for HMPV infection are under 6 months of age or children with underlying pulmonary disease, especially asthma
- May also contribute to respiratory disease in elderly or immunocompromised adults

Mortality

- Mortality in otherwise healthy children hospitalized for HMPV pneumonia is less than 1%
- Disease may be fatal in as many as 30-40% of patients with immune suppression or underlying disease

Gender, Race, and Age Distribution

- Common cause of bronchiolitis and pneumonia among infants under 1 year of age
- No recognized gender or racial predilection

Clinical Features

- Most infections cause a mild, self-limited upper respiratory tract illness
- During their first HMPV infection, about 10-15% of outpatient infants and young children have signs or symptoms of bronchiolitis or pneumonia
- Severe lower respiratory tract disease (bronchiolitis, pneumonia) may occur at any age, especially among the elderly or among those with compromised cardiac, pulmonary, or immune systems

Radiologic Features

- Bilateral multifocal air space consolidation and interstitial infiltrates

Prognosis and Therapy

- With supportive care, most children recover from illness in 1 to 2 weeks
- No licensed therapies or prophylactic treatments for HMPV
- The only antiviral drug shown to have in vitro efficacy against HMPV is ribavirin

HUMAN METAPNEUMOVIRUS (HMPV)—PATHOLOGIC FEATURES

Gross Findings

- In fatal cases, the lungs are heavy and display congestion, hemorrhage, and edema

Microscopic Findings

- Histopathologic features have received relatively little study
- Necrotizing bronchiolitis that evolves to chronic bronchiolitis has been described, as well as interstitial pneumonitis, acute or organizing diffuse alveolar damage, and increased intra-alveolar macrophages
- Organizing DAD and chronic airway disease in patients who die later in the course of the illness
- BAL specimens may show multinucleated giant cells with cytoplasmic inclusions

Ultrastructural Features

- The virion is variable in shape and size, ranging from 150-300 nm, and is morphologically indistinguishable from other members of the *Paramyxoviridae* family when viewed by negative-stain electron microscopy

Pathologic Differential Diagnosis

- Other viral causes of giant cell pneumonia, necrotizing bronchiolitis, and interstitial pneumonitis and diffuse alveolar damage
- Non-infectious causes of diffuse alveolar damage

most unique histopathologic finding is the presence of syncytial and parenchymal multinucleated endothelial cells. However, this feature occurs in only about one-fourth of the cases and cannot be used as a sensitive criterion for the diagnosis of Henipa virus infections; furthermore, similar cells can also be seen in measles virus, RSV, HPIV, herpesviruses, and other infections. In addition to these viral infections, other non-infectious causes of diffuse alveolar damage may also be considered in the differential diagnosis. Unequivocal diagnosis can be made only by laboratory tests such as IHC, cell culture isolation, PCR, or serology. IHC can reveal widespread presence of Nipah virus antigens in endothelial and smooth muscle cells of blood vessels as well as in various parenchymal cells (Figures 13-13D–F). Ultrastructural studies can also demonstrate the pleomorphic viral particles which are composed of helical nucleocapsids enclosed within an envelope.

Prognosis and Therapy

Only 3 persons are known to have been infected with Hendra virus, and 2 of them died. Death occurs in about 30-40% of patients infected with Nipah virus and is more frequent in patients with rapidly developing severe neurologic signs. Residual neurologic signs are common among survivors. Treatment is supportive, including mechanical ventilation for patients in a deep

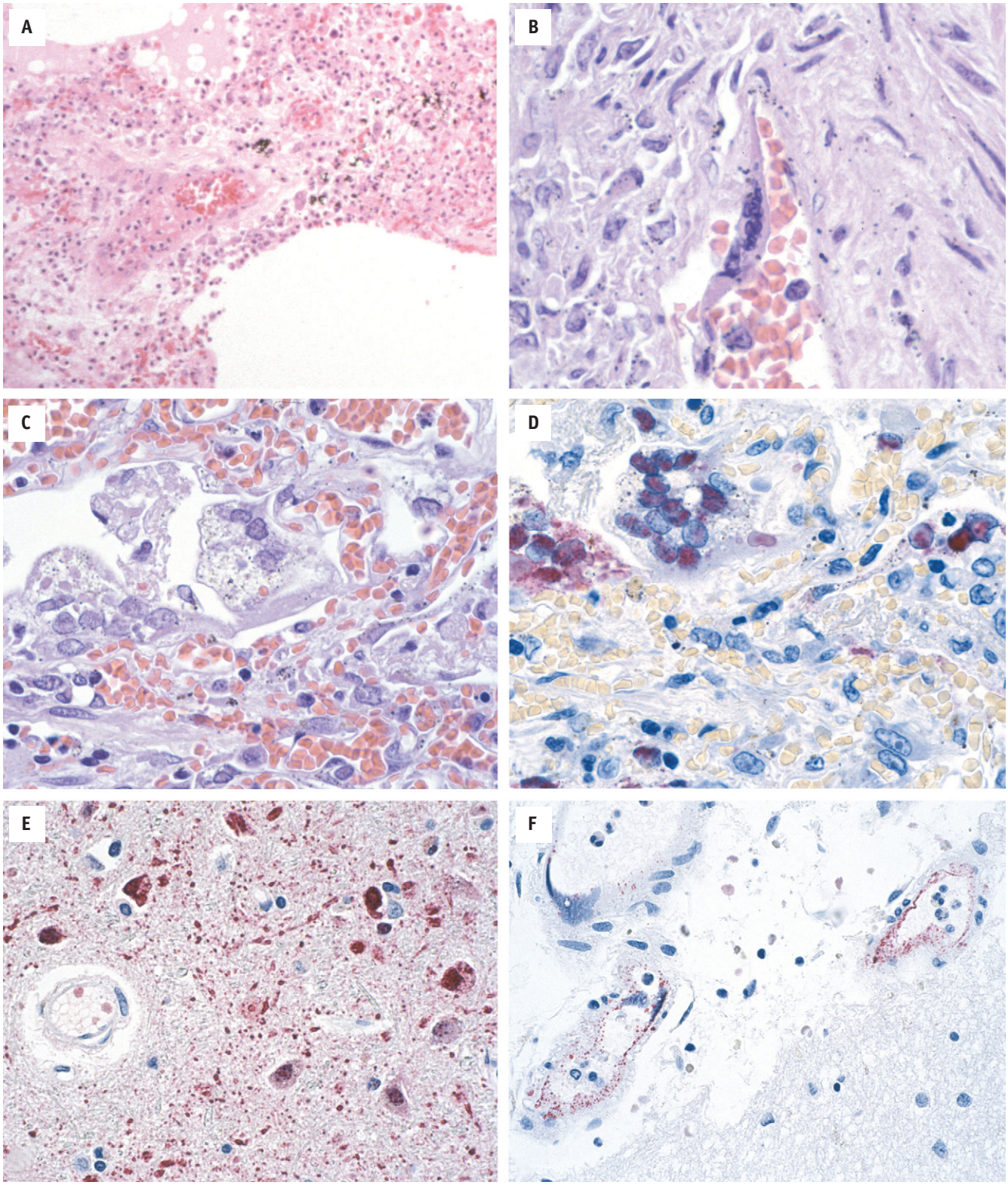

FIGURE 13-13

Nipah virus pneumonia

(A) Interstitial pneumonitis, edema, congestion, and focal thrombosis. **(B)** Pulmonary vessel showing a multinucleated endothelial syncytium. **(C)** Interstitial pneumonitis with intra-alveolar multinucleated giant cells with nuclear inclusions. **(D)** Viral nature of giant cells, in **C,** as evidenced by immunostaining for Nipah viral antigens (immunoalkaline phosphatase). **(E, F)** Immunostaining of Nipah viral antigens in neurons and vascular endothelium in the central nervous system (immunoalkaline phosphatase).

HENIPA VIRUSES—FACT SHEET

Definition

- Hendra and Nipah viruses are nonsegmented, negative-stranded RNA viruses, members of the family *Paramyxoviridae* and subfamily *Paramyxovirinae*

Incidence and Location

- Rare cases have occurred in Australia and Asia throughout the distribution of the animal reservoir, the fruit bat of the *Pteropus* genus
- Infection has occurred primarily in patients who have contact with sick horses and pigs
- Food-borne transmission has also been reported in an individual who consumed fruit contaminated by *Pteropus* bats

Morbidity and Mortality

- Death occurs in about 30-40% of Nipah cases associated with rapidly developing severe neurologic signs
- 2 of 3 of the known Hendra cases died with pneumonitis and multiorgan failure

Gender, Race, and Age Distribution

- Patient demographics depend largely on the mode of exposure, in most cases occupational
- No apparent gender or racial predilection

Clinical Features

- Acute febrile encephalitis and influenza-like illness

Radiologic Features

- Bilateral infiltrates consistent with the acute respiratory distress syndrome

Therapy

- Treatment is supportive, including mechanical ventilation when needed
- Equivocal data regarding the efficacy of ribavirin in treatment

HENIPA VIRUSES—PATHOLOGIC FEATURES

Gross Findings

- In fatal cases, the lungs are heavy and display congestion, hemorrhage, and edema

Microscopic Findings

- Histopathologic involvement of the central nervous system and respiratory system
- Vasculitis, thrombosis, endothelial cell damage, and syncytial cell formation
- Multinucleated giant cells with intranuclear and cytoplasmic inclusions in the brain, lung, and other organs
- Organizing diffuse alveolar damage in patients who die later in the course of the illness

Immunohistochemical Features

- Widespread presence of Henipa virus antigens can be seen by IHC in endothelial and smooth muscle cells of blood vessels, as well as in various parenchymal cells

Ultrastructural Features

- Pleomorphic viral particles composed of helical nucleocapsids enclosed within an envelope

Pathologic Differential Diagnosis

- Other viral causes of encephalitis, giant cell pneumonia, and diffuse alveolar damage, as well as noninfectious causes of diffuse alveolar damage

coma who are unable to maintain airways. Ribavirin was used in humans during the Nipah outbreak in Malaysia, with equivocal results.

HEMORRHAGIC FEVER VIRUSES

The combination of fever and hemorrhage can be caused by different viruses, rickettsiae, bacteria, protozoa, and fungi. However, the term "viral hemorrhagic fever" (VHF) is usually reserved for systemic infections characterized by fever and hemorrhage caused by a special group of viruses transmitted to humans by arthropods and rodents. VHFs are febrile illnesses characterized by abnormal vascular regulation and vascular damage and are caused by small, lipid-enveloped RNA viruses. This syndrome can be caused by RNA viruses belonging to four different families that differ in their genomic structure, replication strategy, and morphologic features (*Arenaviridae, Bunyaviridae, Flaviviridae,* and *Filoviridae*). Arenaviruses, bunyaviruses, and filoviruses are negative-stranded, whereas flaviviruses are positive-stranded RNA viruses. Hemorrhagic fever viruses are distributed worldwide, and the diseases they cause are traditionally named according to the location where they were first described. The oldest and best known is yellow fever virus; others include Lassa fever, lymphocytic choriomeningitis, Ebola, and dengue viruses. The distributions of the individual VHFs are related to the distributions of their specific arthropod and rodent vectors.

Clinical Features

VHF is characterized clinically by its disproportionate effect on the vascular system. Typical manifestations are related to a loss of vascular regulation (vasodilatation and hypotension), vascular damage (leakage of protein into the urine, edema in soft tissues of the face and other loose connective tissues, and petechial hemorrhage in the skin and internal organs), and severe systemic derangement that presents as fever, myalgia, and asthenia proceeding to a state of prostration. Hemorrhage is common with most of these diseases and usually originates from mucosal surfaces. Patients with severe hemorrhagic fever generally develop shock, diffuse bleeding, and central nervous system dysfunction.

Radiologic Features

In patients with respiratory illness, chest radiographs may reveal bilateral interstitial and alveolar edema and hemorrhage.

Pathologic Features

Gross Findings

In fatal VHF, the lungs show congestion, hemorrhage, and edema. Pleural effusion may be found with certain infections.

Microscopic Findings

At autopsy, common findings include widespread petechial hemorrhages and ecchymoses involving skin, mucous membranes, and internal organs. However, in many VHF patients manifestations of bleeding may be minimal or absent. Effusions, occasionally hemorrhagic, are also frequently seen. Widespread, focal, and sometimes massive necrosis is commonly observed in all organ systems and is often ischemic in nature. Necrosis is usually most prominent in the liver and lymphoid tissues. The most consistent microscopic feature is found in the liver and consists of multifocal hepatocellular necrosis with cytoplasmic eosinophilia, Councilman bodies, nuclear pyknosis, and cytolysis (Figure 13-14D). Inflammatory cell infiltrates and necrotic areas are usually mild and, when present, consist of neutrophils and mononuclear cells. Commonly observed histopathologic changes in the lung include various degrees of hemorrhage, intra-alveolar edema, interstitial pneumonitis, and diffuse alveolar damage (Figure 13-14A,E,H).

Ancillary Studies and Differential Diagnosis

The diagnosis of VHF should be suspected in patients with appropriate clinical manifestations returning from an endemic area, particularly if there is travel to rural areas during seasonal or epidemic disease activity. The diagnosis suspected by history and clinical manifestations can also be supported histopathologically. However, because of similar pathologic features seen in VHF and a variety of other viral, rickettsial, and bacterial infections, as well as noninfectious causes of hemorrhage, edema, and diffuse alveolar damage, unequivocal diagnosis can be made only by laboratory tests such as cell culture isolation, serology, PCR, and IHC (Figure 13-14B,F,G,I,J). Ultrastructural studies can also demonstrate the presence of virions. All viruses have a lipid envelope that is acquired by budding at either the cell surface or the internal membranes. The size and shape of these viruses vary from relatively small (35-50 nm), uniform, round particles, as seen with flaviviruses, to more pleomorphic, rod-shaped particles (measuring occasionally up to 15,000 nm) in the case of filoviruses (Figure 13-14C).

Prognosis and Therapy

Case mortality ranges from about 15% with infections such as Lassa fever up to 90% with filovirus infections such as Ebola. Treatment depends on the particular agent and may include the use of passive antibodies, antiviral drugs such as ribavirin, or supportive therapy. Supportive therapy should include the reasonable measures that would be employed in any very ill patient with a fragile vascular bed. Volume replacement may be particularly important in some patients, especially with dengue hemorrhagic fever.

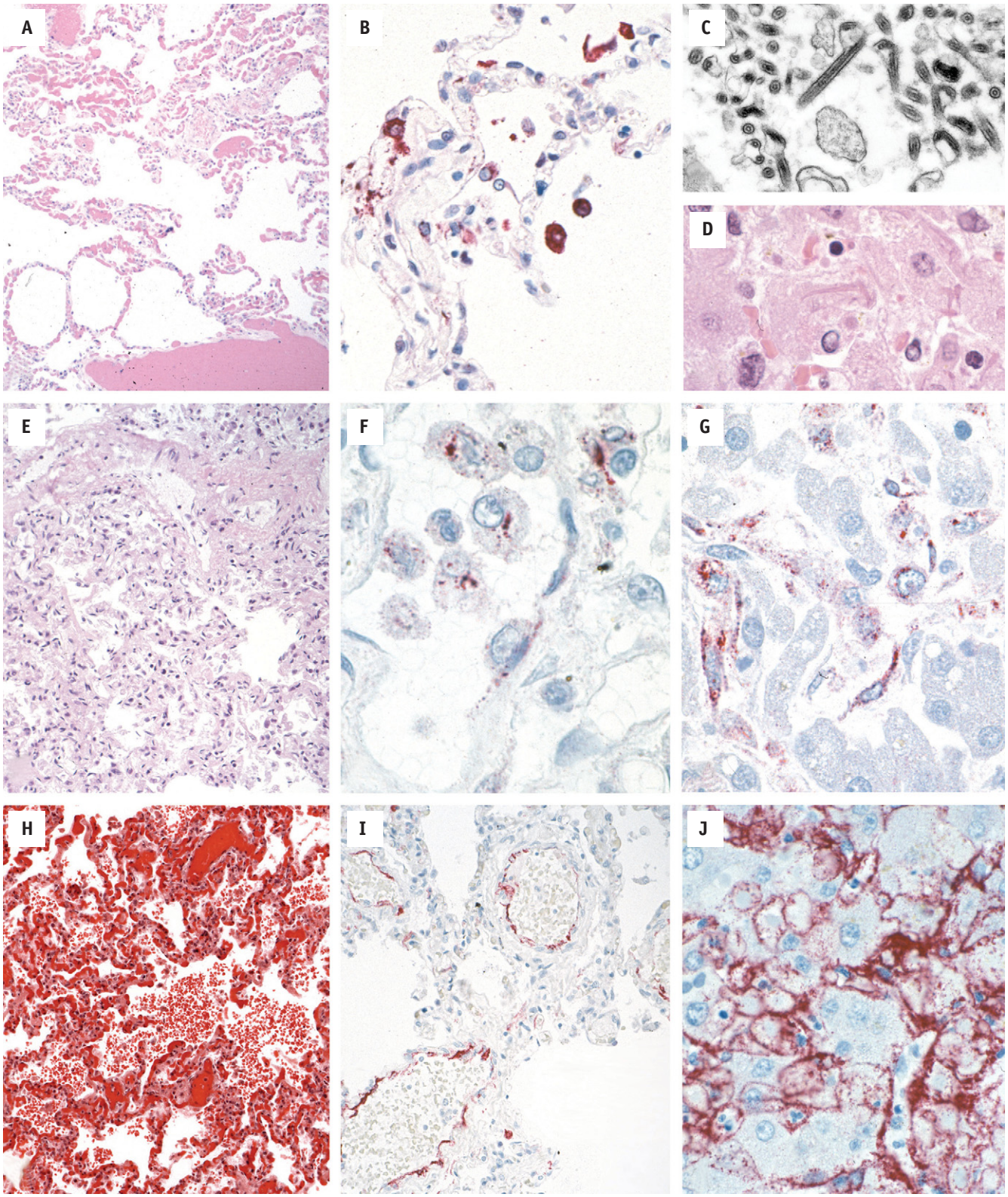

FIGURE 13-14

Viral hemorrhagic fevers

(A) Lung from a fatal Ebola case showing congestion and no significant inflammation. **(B)** Ebola viral antigens in alveolar macrophages, endothelial cells, fibroblasts, and other interstitial cells in the same patient seen in **A** (immunoalkaline phosphatase). **(C)** Ebola viral particles are seen within an intra-alveolar space by electron microscopy. **(D)** Numerous filamentous Ebola virus inclusions are seen within hepatocytes. **(E)** Mild interstitial pneumonitis in a fatal case of dengue hemorrhagic fever. **(F)** Dengue viral antigen-containing circulating mononuclear cells in a pulmonary vessel. Note also the viral antigens in a lining endothelial cell (immunoalkaline phosphatase). **(G)** Immunostaining of liver showing viral antigens predominantly within sinusoidal Kupffer cells. Note the absence of hepatocyte staining (immunoalkaline phosphatase). **(H)** Pulmonary congestion and absence of significant inflammatory response in a case of Lassa fever. **(I)** Lassa viral antigens in endothelial cells lining medium-sized vessels in the lung (immunoalkaline phosphatase). **(J)** IHC reveals Lassa virus antigens in the cytoplasm of hepatocytes and sinusoidal lining cells in association with areas of hepatocellular necrosis (immunoalkaline phosphatase).

VIRAL HEMORRHAGIC FEVERS—FACT SHEET

Definition

- Viral hemorrhagic fevers (VHFs) are systemic infections characterized by fever and hemorrhage, caused by a group of viruses transmitted to humans by arthropods and rodents
- Viruses belong to four different families that differ in their genomic structure, replication strategy, and morphologic features (*Arenaviridae, Bunyaviridae, Flaviviridae,* and *Filoviridae*)
- Arenaviruses, bunyaviruses, and filoviruses are negative-stranded, whereas flaviviruses are positive-stranded RNA viruses

Incidence and Location

- Rare diseases
- Hemorrhagic fever viruses are distributed worldwide, and the diseases they cause are traditionally named according to the location where they were first described
- The distributions of particular VHFs are related to the distributions of their specific arthropods and rodent vectors

Mortality

- Case mortality ranges from about 15% with infections such as Lassa fever up to 90% with filovirus infections such as Ebola

Gender, Race, and Age Distribution

- Patient demographics depend largely on the agent and mode of exposure
- People of all ages are vulnerable to VHFs
- No recognized gender or racial predilection

Clinical Features

- Fever, myalgia, asthenia, and prostration
- Loss of vascular regulation, shock, and central nervous system dysfunction
- Hemorrhage

Radiologic Features

- Bilateral interstitial or alveolar edema and infiltrates

Prognosis and Therapy

- Supportive therapy
- Passive antibodies
- Antivirals (ribavirin)

VIRAL HEMORRHAGIC FEVERS—PATHOLOGIC FEATURES

Gross Findings

- In fatal cases, the lungs display congestion, hemorrhage, and edema
- Widespread petechial hemorrhages and ecchymoses are present systemically

Microscopic Findings

- Histopathologic changes in the lung include varying degrees of hemorrhage, edema, interstitial pneumonitis, and diffuse alveolar damage
- Other systemic pathology as described in the text

Immunohistochemical Features

- Viral antigens can commonly be detected by IHC in the mononuclear phagocytic system and endothelium, as well as in parenchymal cells, depending on the particular VHF agent

Ultrastructural Features

- Features vary for the four families of viruses that cause VHF syndromes
- All viruses have a lipid envelope that is acquired by budding at either the cell surface or the internal membranes
- The sizes and shape of these viruses vary from relatively small (35-50 nm), uniform round particles, as seen with flaviviruses, to more pleomorphic, rod-shaped particles (occasionally measuring up to 15,000 nm) in the case of filoviruses

Pathologic Differential Diagnosis

- Other causes of hemorrhage, edema, and diffuse alveolar damage
- Unequivocal diagnosis can be made only by laboratory tests such as cell culture isolation, serology, PCR, and IHC

SUGGESTED READINGS

General

1. Connor DH, Chandler FW, Schwartz DA, Manz HJ, Lack EE, eds. *Pathology of Infectious Disease,* 1st ed. Connecticut: Appleton & Lange; 1997.
2. Winn WC, Walker DH. Viral infections. In: Dail DH, Hammar SP, eds. *Pulmonary Pathology.* 2nd ed. New York: Springer-Verlag; 1994:429-64.
3. de Roux A, Marcos MA, Garcia E, et al. Viral community-acquired pneumonia in nonimmunocompromised adults. *Chest*. 2004;125: 1343-51.
4. Fields BN, Knipe DM, Howley PM, eds. *Fields Virology*. 4 ed. Philadelphia: Lippincott-Williams & Williams; 2001.
5. Infection unusual pneumonias. In: Katzenstein AL, ed. *Katzenstein and Askin's Surgical Pathology of Non-neoplastic Lung Diseases*. Philadelphia: Saunders Co.; 1997:247-85.
6. Travis WD, Colby TV, Koss MN, et al. Lung infections. In: King DW, ed. *Non-Neoplastic Disorders of the Lower Respiratory Tract.* Washington, DC: American Registry of Pathology and the Armed Forces Institute of Pathology; 2002.
7. Mahy BWJ, Collier L, eds. *Microbiology and Microbial Infections: Virology*. 9 ed. London: Arnold; 1998.
8. Palmer EL, Martin ML. *Electron Microscopy in Viral Diagnosis*. Boca Raton, Florida: CRC Press, Inc.; 1988.

Adenoviruses

9. Becroft DM. Histopathology of fatal adenovirus infection of the respiratory tract in young children. *J Clin Pathol*. 1967;20:561-9.
10. Dudding BA, Wagner SC, Zeller JA, Gmelich JT, French GR, Top FH Jr. Fatal pneumonia associated with adenovirus type 7 in three military trainees. *N Engl J Med*. 1972;286:1289-92.
11. Hierholzer JC. Adenoviruses in the immunocompromised host. *Clin Microbiol Rev*. 1992;5:262-74.
12. Pham TT, Burchette JL Jr, Hale LP. Fatal disseminated adenovirus infections in immunocompromised patients. *Am J Clin Pathol*. 2003;120: 575-83.
13. Zahradnik JM. Adenovirus pneumonia. *Semin Respir Infect*. 1987;2: 104-11.

Hantaviruses

14. Duchin JS, Koster FT, Peters CJ, et al. Hantavirus pulmonary syndrome: a clinical description of 17 patients with a newly recognized disease. The Hantavirus Study Group. *N Engl J Med*. 1994;330:949-55.
15. Lukes RJ. The pathology of thirty-nine fatal cases of epidemic hemorrhagic fever. *Am J Med*. 1954;16:639-50.
16. Nolte KB, Feddersen RM, Foucar K, et al. Hantavirus pulmonary syndrome in the United States: a pathological description of a disease caused by a new agent. *Hum Pathol*. 1995;26:110-20.
17. Zaki SR, Greer PW, Coffield LM, et al. Hantavirus pulmonary syndrome. Pathogenesis of an emerging infectious disease. *Am J Pathol*. 1995;146:552-79.

18. Zaki SR, Khan AS, Goodman RA, et al. Retrospective diagnosis of hantavirus pulmonary syndrome, 1978-1993: implications for emerging infectious diseases. *Arch Pathol Lab Med*. 1996;120:134-9.

SARS Coronavirus

19. Chong PY, Chui P, Ling AE, et al. Analysis of deaths during the severe acute respiratory syndrome (SARS) epidemic in Singapore: challenges in determining a SARS diagnosis. *Arch Pathol Lab Med*. 2004;128:195-204.
20. Ding Y, Wang H, Shen H, et al. The clinical pathology of severe acute respiratory syndrome (SARS): a report from China. *J Pathol*. 2003;200: 282-9.
21. Franks TJ, Chong PY, Chui P, et al. Lung pathology of severe acute respiratory syndrome (SARS): a study of 8 autopsy cases from Singapore. *Hum Pathol*. 2003;34:743-8.
22. Goldsmith CS, Tatti KM, Ksiazek TG, et al. Ultrastructural characterization of SARS coronavirus. *Emerg Infect Dis*. 2004;10:320-6.
23. Ksiazek TG, Erdman D, Goldsmith CS, et al. A novel coronavirus associated with severe acute respiratory syndrome. *N Engl J Med*. 2003;348:1953-66.
24. Nicholls JM, Poon LL, Lee KC, et al. Lung pathology of fatal severe acute respiratory syndrome. *Lancet*. 2003;361:1773-8.
25. Shieh WJ, Hsiao CH, Paddock CD, et al. Immunohistochemical, in situ hybridization, and ultrastructural localization of SARS-associated coronavirus in lung of a fatal case of severe acute respiratory syndrome in Taiwan. *Hum Pathol*. 2005;36:303-9.
26. To KF, Tong JH, Chan PK, et al. Tissue and cellular tropism of the coronavirus associated with severe acute respiratory syndrome: an in-situ hybridization study of fatal cases. *J Pathol*. 2004;202:157-63.

Cytomegalovirus

27. Beschorner WE, Hutchins GM, Burns WH, Saral R, Tutschka PJ, Santos GW. Cytomegalovirus pneumonia in bone marrow transplant recipients: miliary and diffuse patterns. *Am Rev Respir Dis*. 1980;122:107-14.
28. Herry I, Cadranel J, Antoine M, et al. Cytomegalovirus-induced alveolar hemorrhage in patients with AIDS: a new clinical entity? *Clin Infect Dis*. 1996;22:616-20.
29. Ison MG, Fishman JA. Cytomegalovirus pneumonia in transplant recipients. *Clin Chest Med*. 2005;26:691-705, viii.
30. Ljungman P, Griffiths P, Paya C. Definitions of cytomegalovirus infection and disease in transplant recipients. *Clin Infect Dis*. 2002;34:1094-7.
31. Wallace JM, Hannah J. Cytomegalovirus pneumonitis in patients with AIDS: findings in an autopsy series. *Chest*. 1987;92:198-203.

Herpes Simplex Viruses

32. Feldman S, Stokes DC. Varicella zoster and herpes simplex virus pneumonias. *Semin Respir Infect*. 1987;2:84-94.
33. Kimberlin DW. Neonatal herpes simplex infection. *Clin Microbiol Rev*. 2004;17:1-13.
34. Nash G. Necrotizing tracheobronchitis and bronchopneumonia consistent with herpetic infection. *Hum Pathol*. 1972;3:2832-91.
35. Ramsey PG, Fife KH, Hackman RC, Meyers JD, Corey L. Herpes simplex virus pneumonia: clinical, virologic, and pathologic features in 20 patients. *Ann Intern Med*. 1982;97:813-20.
36. Singer DB. Pathology of neonatal herpes simplex virus infection. *Perspect Pediatr Pathol*. 1981;6:243-78.

Varicella Zoster Virus

37. Feldman S. Varicella-zoster virus pneumonitis. *Chest*. 1994;106 :22S-7S.
38. Harger JH, Ernest JM, Thurnau GR, et al. Risk factors and outcome of varicella-zoster virus pneumonia in pregnant women. *J Infect Dis*. 2002;185:422-7.
39. Mermelstein RH, Freireich AW. Varicella pneumonia. *Ann Intern Med*. 1961;55:456-63.
40. Triebwasser JH, Harris RE, Bryant RE, Rhoades ER. Varicella pneumonia in adults: report of seven cases and a review of literature. *Medicine (Baltimore)*. 1967;46:409-23.
41. Waring JJ, Neubuerger K, Geever EF. Severe forms of chickenpox in adults. *Arch Intern Med,* 1942;69:384-408.

Influenza Viruses

42. Guarner J, Paddock CD, Shieh WJ, et al. Histopathologic and immunohistochemical features of fatal influenza virus infections in children during the 2003-2004 season. *Clin Infect Dis*. 2006;43:132-40.
43. Guarner J, Shieh WJ, Dawson J, et al. Immunohistochemical and in situ hybridization studies of influenza A virus infection in human lungs. *Am J Clin Pathol*. 2000;114:227-33.
44. Hers JF, Masurel N, Mulder J. Bacteriology and histopathology of the respiratory tract and lungs in fatal Asian influenza. *Lancet*. 1958;2:1141-3.
45. Hers JFP. Changes in the respiratory mucosa resulting from infection with influenza virus B. *J Pathol Bacteriol*. 1957;73:565-8.
46. Martin CM, Kunin CM, Gottlieb LS, Barnes MW, Liu C, Finland M. Asian influenza A in Boston, 1957-1958: I. Observations in thirty-two influenza-associated fatal cases. *AMA Arch Intern Med*. 1959;103:515-31.
47. Nolte KB, Alakija P, Oty G, et al. Influenza A virus infection complicated by fatal myocarditis. *Am J Forensic Med Pathol*. 2000;21:375-9.
48. Shinya K, Ebina M, Yamada S, Ono M, Kasai N, Kawaoka Y. Avian flu: influenza virus receptors in the human airway. *Nature*. 2006;440: 435-6.
49. Ungchusak K, Auewarakul P, Dowell SF, et al. Probable person-to-person transmission of avian influenza A (H5N1). *N Engl J Med*. 2005;352: 333-40.
50. Van Riel D, Munster VJ, de Wit E, et al. H5N1 virus attachment to lower respiratory tract. *Science*. 2006;312:399.
51. Yeldandi AV, Colby TV. Pathologic features of lung biopsy specimens from influenza pneumonia cases. *Hum Pathol*. 1994;25:47-53.

Measles

52. Akhtar M, Young I. Measles giant cell pneumonia in an adult following long-term chemotherapy. *Arch Pathol*. 1973;96:145-8.
53. Archibald RW, Weller RO, Meadow SR. Measles pneumonia and the nature of the inclusion-bearing giant cells: a light- and electron-microscope study. *J Pathol*. 1971;103:27-34.
54. Breitfeld V, Hashida Y, Sherman FE, Odagiri K, Yunis EJ. Fatal measles infection in children with leukemia. *Lab Invest*. 1973;28:279-91.
55. Kipps A, Kaschula RO. Virus pneumonia following measles: a virological and histological study of autopsy material. *S Afr Med J*. 1976;50: 1083-8.
56. Monafo WJ, Haslam DB, Roberts RL, Zaki SR, Bellini WJ, Coffin CM. Disseminated measles infection after vaccination in a child with a congenital immunodeficiency. *J Pediatr*. 1994;124:273-6.
57. Sata T, Kurata T, Aoyama Y, Sakaguchi M, Yamanouchi K, Takeda K. Analysis of viral antigens in giant cells of measles pneumonia by immunoperoxidase method. *Virchows Arch A Pathol Anat Histopathol*. 1986;410: 133-8.
58. Sobonya RE, Hiller FC, Pingleton W, Watanabe I. Fatal measles (rubeola) pneumonia in adults. *Arch Pathol Lab Med*. 1978;102:366-71.

Parainfluenza

59. Akizuki S, Nasu N, Setoguchi M, Yoshida S, Higuchi Y, Yamamoto S. Parainfluenza virus pneumonitis in an adult. *Arch Pathol Lab Med*. 1991;115:824-6.
60. Butnor KJ, Sporn TA. Human parainfluenza virus giant cell pneumonia following cord blood transplant associated with pulmonary alveolar proteinosis. *Arch Pathol Lab Med*. 2003;127:235-8.
61. Jarvis WR, Middleton PJ, Gelfand EW. Parainfluenza pneumonia in severe combined immunodeficiency disease. *J Pediatr*. 1979;94: 423-5.
62. Little BW, Tihen WS, Dickerman JD, Craighead JE. Giant cell pneumonia associated with parainfluenza virus type 3 infection. *Hum Pathol*. 1981;12:478-81.
63. Madden JF, Burchette JL Jr, Hale LP. Pathology of parainfluenza virus infection in patients with congenital immunodeficiency syndromes. *Hum Pathol*. 2004;35:594-603.
64. Weintrub PS, Sullender WM, Lombard C, Link MP, Arvin A. Giant cell pneumonia caused by parainfluenza type 3 in a patient with acute myelomonocytic leukemia. *Arch Pathol Lab Med*. 1987;111:569-70.
65. Wendt CH, Weisdorf DJ, Jordan MC, Balfour HH Jr, Hertz MI. Parainfluenza virus respiratory infection after bone marrow transplantation. *N Engl J Med*. 1992;326:921-6.

Respiratory Syncytial Virus

66. Delage G, Brochu P, Robillard L, Jasmin G, Joncas JH, Lapointe N. Giant cell pneumonia due to respiratory syncytial virus. Occurrence in severe combined immunodeficiency syndrome. *Arch Pathol Lab Med*. 1984;108: 623-5.
67. Englund JA, Sullivan CJ, Jordan MC, Dehner LP, Vercellotti GM, Balfour HH Jr. Respiratory syncytial virus infection in immunocompromised adults. *Ann Intern Med*. 1988;109:203-8.
68. Neilson KA, Yunis EJ. Demonstration of respiratory syncytial virus in an autopsy series. *Pediatr Pathol*. 1990;10:491-502.

Human Metapneumovirus

69. Cane PA, van den Hoogen BG, Chakrabarti S, Fegan CD, Osterhaus AD. Human metapneumovirus in a haematopoietic stem cell transplant recipient with fatal lower respiratory tract disease. *Bone Marrow Transplant*. 2003;31:309-10.
70. Kuiken T, van den Hoogen BG, van Riel DA, et al. Experimental human metapneumovirus infection of cynomolgus macaques *(Macaca fascicularis)* results in virus replication in ciliated epithelial cells and pneumocytes with associated lesions throughout the respiratory tract. *Am J Pathol*. 2004;164:1893-1900.
71. Sumino KC, Agapov E, Pierce RA, et al. Detection of severe human metapneumovirus infection by real-time polymerase chain reaction and histopathological assessment. *J Infect Dis*. 2005;192:1052-60.
72. van den Hoogen BG, de Jong JC, Groen J, et al. A newly discovered human pneumovirus isolated from young children with respiratory tract disease. *Nat Med*. 2001;7:719-24.

Henipah Viruses

73. Chua KB, Bellini WJ, Rota PA, et al. Nipah virus: a recently emergent deadly paramyxovirus. *Science*. 2000;288:1432-5.
74. Halpin K, Young PL, Field HE, Mackenzie JS. Isolation of Hendra virus from pteropid bats: a natural reservoir of Hendra virus. *J Gen Virol*. 2000;81:1927-32.
75. Hooper P, Zaki S, Daniels P, Middleton D. Comparative pathology of the diseases caused by Hendra and Nipah viruses. *Microbes Infect*. 2001;3:315-22.
76. Murray K, Selleck P, Hooper P, et al. A *Morbillivirus* that caused fatal disease in horses and humans. *Science*. 1995;268:94-7.
77. O'Sullivan JD, Allworth AM, Paterson DL, et al. Fatal encephalitis due to novel paramyxovirus transmitted from horses. *Lancet*. 1997;349: 93-5.
78. Paton NI, Leo YS, Zaki SR, et al. Outbreak of Nipah-virus infection among abattoir workers in Singapore. *Lancet*. 1999;354:1253-6.
79. Wong KT, Shieh WJ, Kumar S, et al. Nipah virus infection: pathology and pathogenesis of an emerging paramyxoviral zoonosis. *Am J Pathol*. 2002;161:2153-67.

Hemorrhagic Fever Viruses (Ebola, Dengue, Lassa)

80. Bhamarapravati N, Tuchinda P, Boonyapaknavik V. Pathology of Thailand haemorrhagic fever: a study of 100 autopsy cases. *Ann Trop Med Parasitol*. 1967;61:500-10.
81. Burke T. Dengue haemorrhagic fever: a pathological study. *Trans R Soc Trop Med Hyg*. 1968;62:682-92.
82. Burt FJ, Swanepoel R, Shieh WJ, et al. Immunohistochemical and in situ localization of Crimean-Congo hemorrhagic fever (CCHF) virus in human tissues and implications for CCHF pathogenesis. *Arch Pathol Lab Med*. 1997;121:839-46.
83. Walker DH, McCormick JB, Johnson KM, et al. Pathologic and virologic study of fatal Lassa fever in man. *Am J Pathol*. 1982;107:349-56.
84. Zaki SR, Peters CJ. Viral hemorrhagic fevers. In: Connor DH, Chandler FW, Schwartz DA, Manz HJ, Lack EE, eds. *Pathology of Infectious Diseases*. Stamford, CT: Appleton and Lange; 1997:347-64.