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REVIEW

Healthcare-associated infections in neonatal units: lessons from contrasting worlds

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Summary Neonatal intensive care units are vulnerable to outbreaks and sporadic incidents of healthcare-associated infections (HAIs). The incidence and outcome of these infections are determined by the degree of immaturity of the neonatal immune system, invasive procedures involved, the aetiological agent and its antimicrobial susceptibility pattern and, above all, infection control policies practised by the unit. It is important to raise awareness of infection control practices in resource-limited settings, since overdependence upon antimicrobial agents and co-existing lack of awareness of infection control is encouraging the emergence of multi-drug-resistant nosocomial pathogens. We reviewed 125 articles regarding HAIs from both advanced and resource-limited neonatal units in order to study risk factors, aetiological agents, antimicrobial susceptibility patterns and reported successes in infection control interventions. The articles include surveillance studies, outbreaks and sporadic incidents. Gram-positive cocci, viruses and fungi predominate in reports from the advanced units, while Gram-negative enteric rods, non-fermenters and fungi are commonly reported from resource-limited settings. Antimicrobial susceptibility patterns from surveillance studies determined the empirical therapy used in each neonatal unit. Most outbreaks, irrespective of the technical facilities available, were traced to specific lack of infection control practices. We discuss infection control interventions, with special emphasis on their applicability in resource-limited settings. Cost-effective measures for implementing these interventions, with particular reference to the recognition of the role of the microbiologist, the infection control team and antibiotic policies are presented.

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Introduction

The neonatal intensive care unit is an ideal situation to incorporate good infection control policy and practice, since it lends itself not only to the spread of severe infections but also to successful interventions. A collaborative effort between neonatologists and clinical microbiologists who take on the role of infection control can successfully mount a defence against healthcare-associated infections (HAIs). Clinical liaison between microbiologist and clinician is well established in developed countries, whereas in the developing countries such practices are yet to be widely recognized. One reason could be that microbiology results are often delayed in less technologically advanced laboratories, thus forcing the clinician to make empirical treatment decisions without consulting or depending on the microbiologist. However, technical advancement is not a prerequisite for appropriate selection of empirical antimicrobial agents, infection control practices or formulating antibiotic policies. In an environment where resources are scarce, it only requires determination and professional cooperation for suitable interventions to work.

This review on healthcare-associated neonatal infections studies the definitions, associated risk factors and the aetiological agents involved with their antimicrobial susceptibility patterns in two contrasting worlds. We discuss the microbiological and infection control intervention strategies that might help, even in resource-limited settings, to prevent the morbidity and mortality associated with HAI.

Levels of neonatal care: the contrast between developed and developing countries

Levels of neonatal care may be classified as shown in Box 1.[1]

A large proportion of neonates in developing countries (63%) and in rural India (83%) are born at home, with poor facilities for safe and clean delivery by unskilled 'dais' or village health workers.[2,3] Even larger hospitals with a high delivery rate do not have access to Level II neonatal care.[3] No sick newborn care unit (SCNU), government or private, is available at district level in many provinces. The equipment and infrastructure are often limited and doctors are forced to select which babies will be admitted and offered facilities such as ventilators.[4] Few state-owned centres are equipped with neonatal intensive care units (NICUs) and these are scattered across the country. Thus, in developing countries we are dealing with neonates with completely different demographic characteristics. Whereas the minimum gestational age of live-born babies managed in a NICU in developed countries is $\leq$25 weeks with birthweights as low as 300 g, the average gestational age of live-born babies in developing countries is $\geq$30 weeks with birthweights $\geq$1000 g. In a study on anthropometry and body composition of south Indian babies at birth, the mean $\pm$ standard deviation (SD) birthweight of all newborns was 2.80 ± 0.44 kg.[5] Financial constraints in developing countries limit the use of technical interventions, due to which very few neonates undergo invasive medical or surgical procedures, unlike reports from developed countries.

The available microbiological diagnostic facilities also vary from centre to centre. Semi-automated and automated culture systems are available only in a handful of tertiary care centres. The cost of providing these services to patients is borne by the family and is often prohibitive; most clinicians treat patients empirically. Microbiological results are particularly important in neonates as signs of sepsis are often non-specific. Hence, while financial constraints are difficult to resolve, we still have the option to utilize cost-effective, alternative interventions such as infection control, which by reducing the incidence of infection will decrease the overall morbidity and mortality in sick neonates. Successful field trials for home-based neonatal care have already been reported.[2] We need to extend these achievements to healthcare settings.

Methods

We searched for articles on the PubMed database, using the index terms 'hospital acquired infection', 'neonate', 'nosocomial infection neonate', 'neonatal care level', 'neonatal care India'. Reference lists of all articles retrieved were searched to obtain literature for the review. The articles were scrutinized to obtain a comparable and standard definition of nosocomial infection in neonates, inclusion and exclusion criteria used, aetiological agents involved, antimicrobial susceptibility patterns and infection control interventions. Within these search results, we reviewed articles mentioning infection control and antibiotic policies, with special reference to neonatal units in developing countries. Full text articles were scrutinized for a majority of English language papers; for a small number of articles we relied only on the published abstract. For foreign language studies we were able to quote only from the abstract published in English.

Box 1 Levels of hospital-based newborn services[1]

Basic neonatal care (level I)

- Well-newborn nursery + evaluation and postnatal care of healthy newborns
- Neonatal resuscitation
- Stabilization of ill newborns until transfer to a facility at which specialty neonatal care is provided

Specialty neonatal care (level II)

- Special care nursery
- Care of preterm infants with birthweight $\geq$1500 g
- Resuscitation and stabilization of preterm and/or ill infants before transfer to a facility at which newborn intensive care is provided

Subspecialty neonatal intensive care (level III)

Level IIIA
- Restriction on type and/or duration of mechanical ventilation

Level IIIB
- No restrictions on type or duration of mechanical ventilation
- No major surgery

Level IIIC
- Major surgery performed on site[a]
- No surgical repair of serious congenital heart anomalies that require cardiopulmonary bypass and/or ECMO for medical conditions

Level IIID
- Major surgery + surgical repair of serious congenital heart anomalies that require cardiopulmonary bypass and/or ECMO for medical conditions

ECMO, extra-corporeal membrane oxidation.
[a] Omphalocele repair, tracheo-oesophageal fistula or esophageal atresia repair, bowel resection, myelomeningocele repair, ventriculoperitoneal shunt.

Normal microbiological flora of neonates

The foetus is exposed to a sterile environment *in utero*, provided no invasive procedures have been carried out on the mother, the membranes are intact until the onset of labour and there is no prolonged rupture of membranes. During the process of delivery, the neonate is exposed to several sources of microbes. These include the maternal genital tract followed by ambient air or water depending on the type of delivery, handling by healthcare personnel and the instruments used at resuscitation.

Rotimi and Duerden studied the development of bacterial flora of neonates during the first week of life.[6] The predominant organisms in the gut, by the end of the first week, were anaerobes. Bifidobacteria were isolated from all the neonates. Bacteroides and clostridia were isolated from 78.3%. Enterococci were isolated from all neonates, enterobacteria from 82.6%, anaerobic cocci from 52.2%. *Staphylococcus aureus* was the predominant species isolated from the umbilicus; it was isolated from 21.7% of neonates on the first day rising to 87% by the sixth day and represented 49% of isolates from this site. Viridans streptococci (31.4% of isolates) were the commonest species recovered from the mouth. They were present from 8 h after birth. The authors also studied the development of microbial flora of preterm neonates.[7] The numbers of infants studied were too small to draw any firm conclusions; their flora predominantly reflected the maternal genital tract.

In contrast, preterm and full-term babies born by Caesarean section were slow to acquire colonizing flora as compared to those born vaginally. The skin of infants born by Caesarean section is sterile soon after birth compared to neonates born by vaginal delivery.[8] Bowel colonization of infants born by Caesarean delivery is also delayed.[9] Colonization with bifidobacterium-like bacteria and lactobacillus-like bacteria reached levels similar to vaginally delivered infants at 1 month and 10 days, respectively.

Many HAIs result directly or indirectly from patient colonization; studies have shown that hospitalized patients are colonized rapidly with hospital flora.[10] Colonizing flora such as *Candida albicans* in the gastrointestinal tract, vagina or perineal area, can precede infection when normal body defences are impaired through underlying disease, immunomodulating therapy, the use of invasive devices, or when the delicate balance of the normal flora is altered through antimicrobial therapy. However, antimicrobial therapy to eradicate colonizing micro-organisms such as *Pseudomonas aeruginosa* is not beneficial and can propagate drug-resistant pathogens.[11]

Immune status of the neonate

A newborn infant, particularly the preterm infant and to some extent the low birthweight infant, does not have a mature immune system and is often unable to mount an effective immune response.[12] Natural barriers, such as the acidity of the stomach or the production of pepsin and trypsin that maintain sterility of the small intestine, are not fully developed until 3–4 weeks after birth. Membrane protective IgA is missing from the respiratory and urinary tracts, and unless the newborn is breast-fed, is absent from the gastrointestinal tract as well. On a cellular level, there is decreased ability of leukocytes to concentrate where necessary. These leukocytes are less bactericidal and phagocytic. At the humoral level, the newborn has low or non-existent levels of the immunoglobulin antibodies IgM, IgE and IgA. The neonate is born with IgG antibodies acquired from the mother. However, it is important to note that passive transfer of maternal antibodies does not take place till 29 weeks of gestation. This has implications for preterm infants born 25–29 weeks of gestation; they are susceptible to infection despite the mother's antibody status. There is a slow rise of immunoglobulin levels after 3 months of age to levels of older children.

Definition of healthcare-associated infection

Before embarking on a review of nosocomial infections, we reviewed the definitions of nosocomial infections used in various studies.

At the outset, the National Nosocomial Infections Surveillance System (NNIS) of the Centers for Disease Control and Prevention (CDC), USA defines nosocomial infection as a localized or systemic condition (1) that results from adverse reaction to the presence of an infectious agent(s) or its toxin(s) and (2) that was not present or incubating at the time of admission to the hospital.[13]

Limitations encountered during the review process

In the neonate, the definition of an HAI is complicated by the fact that neonates can acquire infection from the maternal genital tract during birth. For this reason, neonatal infections are often classified as early onset (usually 0–7 days after birth) and late onset (>7 days after birth).[14] Some authors also classify them as $\leq$72 h after birth and >72 h after birth.[12] It is interesting to note that the CDC includes infections acquired from the maternal genital tract in their surveillance of nosocomial neonatal infection. Several investigators have found these criteria unsatisfactory.

For the purposes of this review, we have considered only those studies that have excluded infections acquired directly from the maternal genital tract; we found that the definition of nosocomial infections varied between studies and were often related to time of acquisition. The Dutch group have modified the CDC criteria to include infections occurring in the neonatal unit 24–48 h after admission.[15] In some studies, infections which manifested after the patient was in the hospital for $\geq$48 h and those infections which developed within a period of 7 days after discharge from the hospital were considered nosocomial.[16–21]

In other studies, all neonates residing for $\geq$3 days in a hospital unit were included.[22–25] Nosocomial transmission of candida in neonates was considered if the neonate showed negative surveillance cultures at birth and positive cultures from one week later, until death or discharge.[26] Shankar *et al.* also recommend using surveillance cultures to differentiate endogenous colonization from nosocomial acquisition.[21]

We believe that a consistent and universally accepted case definition of HAI in the neonate is important because it offers uniformity of data across centres and facilitates a standardized measurement of outcomes. Many studies that we reviewed did not have clear-cut case definitions with clearly stated inclusion and exclusion criteria; when they did, they varied from centre to centre.

Other common limitations were inadequate sample size,[17,24,26–31] or variability of denominator data wherein some authors reported number of infections per 100 patients (attack rate)[32] or the number of infections per 1000 patient-days (incidence density).[16,32] Annual incidence per 100 000 live births and per 100 NICU discharges have also been used.[24] Absence of robust statistical analysis and the inclusion of anecdotal case reports were also limitations.[28,33–37] Some authors acknowledge the lack of technical equipment to report viral, fungal and parasitic causes of HAIs.[38]

Results

Risk factors for HAIs in neonates

Neonates present with their own unique risk factors that predispose them to acquisition of HAI. The vulnerability of the neonate, particularly the preterm neonate, is directly linked to an immature immune system. This is the single most important host-related factor that predisposes them to infection.

Neonatal age itself is a risk factor for HAI [odds ratio (OR) 5.89; 95% confidence interval (CI): 2.96–11.58; $P < 0.05$].[18] In another study, admission to the neonatal unit, rather than age at admission, was associated with increased risk of HAI ($P < 0.001$).[39] The overall nosocomial infection rate was positively correlated with average length of stay in high-risk nurseries ($r = 0.6$, $P < 0.05$).[40]

Preterm gestational age (<32 weeks) was a risk factor in 26–60% neonates with bacterial, viral and fungal HAI.[19,41–46] The percentage of neonates with low birthweight (1.5–2.5 kg) and with very low birthweight (1.0–1.5 kg) who acquired HAI was 55.5 and 28.2 to 29.6% respectively.[47,48] Infection, including HAI, was the most common cause of death in extremely low birthweight (<1.0 kg) neonates and septicaemia (bacterial and fungal) was the most common presentation (68.4%).[25,49] Male sex was a predisposing factor for nosocomial infections ($P < 0.05$).[16,50] The male predominance in neonatal sepsis has suggested the possibility of an X-linked factor in host susceptibility.[12,51]

Underlying medical conditions such as chronic lung disease, gastro-oesophageal reflux, history of neonatal respiratory distress, maternal infection and congenital heart disease predisposed to HAIs in 4.3–26.1% of neonates.[52–55] A high complexity score, which categorizes procedures by severity of illness and technical complexity, was associated with increased incidence of HAI in neonates after cardiac surgery (OR: 4.03; 95% CI: 1.87–8.43; $P < 0.05$).[18] PRISM (Pediatric Risk of Mortality) score of >25 was also related to neonatal HAI (crude OR: 8.90; 95% CI: 3.49–22.76; $P < 0.001$).[56] The Clinical Risk Index for Babies (CRIB) score shows that nosocomial bacteraemia is independently associated with low birthweight and preterm neonates.[19] Lack of maternal antibodies was a risk factor for infection with unusual rotavirus strains.[57]

Factors relating to healthcare personnel, practices and the environment are often overlooked, and yet remain the most obvious and inexpensive area of intervention. Indeed, the most common route of spread of nosocomial pathogens is person-to-person transmission within the unit and during transfer of patients between units. Such incidents have been linked with outbreaks of bacterial and viral infection in the NICU.[55,58–60]

The most common iatrogenic factor contributing to neonatal HAIs is hands of healthcare workers.[22,53,58,61–65] Intervention in the form of simple handwashing procedures and infection control practices has prevented outbreaks, as reported in many studies.[22,42,55,58–60,63,66]

During the process of delivery, the neonate is exposed to several sources of microbes. Medical devices such as umbilical catheters, central venous catheters, urinary catheters and endotracheal tubes are commonly used in the NICU.[23,28,53,54,67,68] Central venous catheters contributed to 48.9% of HAIs in one study[69] and was a significant risk factor ($P < 0.05$) in others.[20,21,23,39] The nosocomial infection rate was higher in neonates subjected to device use ($r = 0.26$, $P < 0.02$).[40] About 10.8% of catheterized patients developed hospital-acquired urinary tract infection (UTI).[70] The duration of ventilation was also related to the acquisition of HAI.[71]

Reuse of single-use items, a common though unsound practice in many units, has led to outbreaks of HAI. Endotracheal tubes and mucous extraction suction catheters soaked in Hibitane were associated with HAI in the labour room and the special care baby unit.[47] Baby placement services, resuscitation equipment and cleansing solutions have also been implicated in HAI.[72]

An environmental risk factor often overlooked is related to the seasonal variation in the incidence of neonatal HAI. Factors such as warm climate have been associated with a rise in colonization rates with *Enterobacter* spp.[73] Increased humidity and increased environmental dew point at the time of use of nursery air conditioners propagates airborne dissemination of *Acinetobacter* spp. and has been associated with acinetobacter-related bloodstream infections.[67] Bacteria in ambient air have been reported to colonize the conjunctiva in neonates.[65]

Agent factors contributing to HAI relate to the aetiological agents implicated in infection. Infection with drug-resistant organisms plays a significant role in the outcome of HAI in all patients, irrespective of their gestational age and underlying condition. Hospitalization leads to colonization of the skin and gastrointestinal tract with resistant flora found in hospitals and subsequent bloodstream infection, when the skin or mucosa is abraded. Studies have reported that administration of prophylactic antibiotics to neonates can increase the incidence of HAI with drug-resistant pathogenic micro-organisms. About 64.8–100% of neonates presenting with HAI had received prior broad-spectrum antibiotics.[16,30,54]

Clinical presentation of HAIs in neonates

A summary of the most commonly reported neonatal HAI is described in Table I.[15,16,18,21,25,31,39,40,44,61,74,75] A review of the findings of these studies is hampered by the variation and sometimes lack of denominator data. The reader is advised to study these reports with caution, taking into consideration the limitations mentioned earlier.

Common aetiological agents of neonatal HAIs

Healthcare-associated infections in the neonatal unit cover the entire spectrum of organisms: bacterial, fungal, viral and rarely parasitic. A review of healthcare-associated bacterial (Table II),[15,17,18,21,23,25,30,33,40,61,76–79] fungal (Table III)[21,25,26,32,46,80–84] and viral (Table IV)[42,43,52,57,60,63,85–94] infections is summarized in the relevant tables.

Fortunately parasitic nosocomial infections are rare. There have been isolated reports of babesiosis transmitted by blood transfusion in neonates.[37] Among four neonates transfused with blood from asymptomatic babesia-infected donors, two (50%) became parasitaemic, of whom only one developed symptoms of babesiosis.

It is interesting to note that Gram-negative fermenters (*E. coli*, *Klebsiella* spp.) and non-fermenting Gram-negative rods such as *Acinetobacter* spp. and *Pseudomonas* spp. have established themselves as predominant causes of serious neonatal infections in the Indian subcontinent (Table II). In contrast, the predominant organisms isolated from invasive neonatal infections in technologically advanced countries are Gram-positive cocci

Table I Clinical presentation of neonatal healthcare-associated infections

Clinical presentation	% of all infections reported
Septicaemia remains the most common cause of neonatal mortality in the NICU. According to the National Neonatal Perinatal Database (2000) in India the incidence of neonatal septicaemia is 24/1000 live births[75]	25–50%[18,21,44,61]
	50–75%[15,25]
	>75%[16,40]
Lower respiratory tract infections	3–10%[16,18,39,61]
	15–30%[15,25,39,44]
	100%[40]
Necrotizing enterocolitis/perforation	2–15%[25,39,44]
	35–75%[40]
Meningitis	1.5–6%[15,25,61]
Skin (central venous catheter site, operation wound, umbilicus) infections	3–10%[16,18,25,44,61]
	11–20%[39]
	30–75%[21,40]
Arthritis	1%[25,31]
Device (ventriculo-peritoneal shunt)	2–6%[18,39]
Urinary tract infections	<1–12%[15,18,21,39,44]
Eye infections	1.8–10%[15,16,21,39,40,74]
	11–40%[15,44,61]
	50–70%[40]
Oropharyngeal infections	3–20%[21,40]
Gastroenteritis	<10%[39]
	20–40%[40]
Upper respiratory tract infections	2.4%[39]
Ear infections	5–20%[40]

Table II Hospital-acquired bacterial infections

Organism	Lessons learnt	% cause of infection
Klebsiella spp.	Nosocomial surgical site infections in neonates following contamination with endogenous flora.[18]	2.5–10%[21,40]
	Also implicated in septicaemia, septic arthritis, meningitis, conjunctivitis, parotitis.	20–60%[18,30,61]
Enterobacter spp.	Bloodstream infection due to contamination of surgical site with endogenous flora.[18]	3–20%[18,40] 50%[61]
Pseudomonas spp.	Cause of septicaemia, pneumonia, urinary tract infection.	10–16%[18,25,61]
Escherichia coli	Rate of antimicrobial resistance was higher in the nosocomial strains of *E. coli* compared to community-acquired strains ($P < 0.05$).[76]	4.3–6%[18,40] >40%[61]
Acinetobacter spp.	Implicated in colonization as well as infection; 56% mortality reported with the latter; mortality in surgical infections 100%.[30]	6–12%[18,33] >25%[23]
Serratia spp.	Cause of septicaemia in neonates with surgical wounds.[18] Also a cause of meningitis, pneumonia, umbilical wound infection, conjunctivitis.	3–35%[18,61]
Stenotrophomonas maltophilia	Cause of septicaemia in neonates with surgical wounds.	4%[18]
Coagulase-negative staphylococcus (CoNS)	Most common pathogen causing HAI in the surgical neonatal unit. Meticillin resistance in CoNS was 92.3% and mortality 16%.[77]	3–11%[18,25] 45–60%[15,21,40]
S. aureus	Meticillin resistance 72.7%. Mortality due to septicaemia 24%.[77] MRSA caused 38.8% of HAI in neonates with mortality of 28.6%.[25] Central venous catheter was the source of infection with *S. aureus* in 7/8 infected neonates.[17]	4–10%[15,18,21,40] >35%[17,25]
Enterococcus spp.	No colonization with vancomycin-resistant enterococci (VRE) was noted in neonates despite prior vancomycin therapy.[78]	5–6%[18,25,40] >23%[21]
Streptococcus mitis	Common cause of bloodstream infection in the surgical neonatal unit.	2.5–10%[21,40]
Bacteroides fragilis	Cause of surgical site infections in neonates.[21]	7.6%[21]
Group B beta haemolytic streptococcus (GBS)	Relatively uncommon cause of HAI in neonates in India. No cases of late-onset disease due to GBS reported from India.[79]	7.9%[40]

HAI, hospital-acquired infection; MRSA, meticillin-resistant *Staphylococcus aureus*.

(coagulase-negative staphylococci, Group B streptococcus).[95] The reason for this difference is probably multifactorial and could be due to gestational age of the babies involved, the use of invasive devices (central vascular catheters and shunts), ambient moisture, humidity and the prevalent flora in the unit. Evidence supporting these risk factors has been discussed elsewhere in the review and probably merits further evaluation.

Of all the fungal infections reported in neonatal patients, *Candida* spp. cause significant mortality and morbidity in the neonatal unit (Table III) and will be discussed in some detail here. Although the source of *C. albicans* infection in the NICU is often considered to be endogenous, molecular typing studies have shown that nosocomial transmission of *C. albicans* is the predominant mode of acquisition.[26,96] The nosocomial acquisition of *C. albicans* is related to cross-contamination via the hands of healthcare workers or parents and the use of contaminated equipment. [26,97] In one study, retrograde medication syringe fluids were significantly more likely to be contaminated with candida than other fluids being administered to the infants ($P < 0.001$). Candidaemia was significantly associated with total parenteral nutrition ($P = 0.04$) and retrograde medication administration ($P = 0.02$).[98] Central vascular catheters, steroid administration, endotracheal intubation and H2-blockers have also been reported as risk factors for systemic

Table III Hospital-acquired fungal infections

Fungi	Lessons learnt
C. albicans[26,32,80,81]	Important causes of non-persistent candidaemia, persistent candidaemia, endocarditis, uveitis. Molecular epidemiology suggests nosocomial rather than maternal transmission of *C. albicans* in neonates.[26]
Non-albicans *Candida* spp.[a,21,25,32,46,80,82]	Cause of candidaemia, endophthalmitis, endocarditis, meningitis, peritonitis. Source of infection was central venous catheter.[82]
Rhodotorula mucilaginosa[83]	Outbreak ($N = 4$) of indwelling catheter-related septicaemia in NICU. Related to birthweight, gestational age, duration of parenteral nutrition, antibiotic therapy and prophylactic fluconazole.
Rhizopus microsporus[84]	Outbreak of cutaneous infection in preterm neonates ($N = 4$). Source traced to wooden tongue depressors used in the nursery as splints for intravenous and arterial cannulation site. The combination of warm, humid conditions in neonatal incubators, particularly in association with occlusive dressings, also favours cutaneous fungal infections.

NICU, neonatal intensive care unit.
[a] Non-albicans *Candida* spp. included *C. parapsilosis*, *C. tropicalis*, *C. lusitaniae*, *C. glabrata*, *C. krusei*, *C. guillermondii*.

fungal infections in neonates.[66,82] Other risk factors include prematurity, low birthweight and use of broad-spectrum antibiotics.[36] Complications of candidaemia such as endocarditis and uveitis have been reported in neonates. The onset of endocarditis was related to persistant candidaemia. Fungal endocarditis was present in 13.7% neonates with persistent disease (>5 days of candidaemia) and 3.7% patients with non-persistent disease (OR: 4.19), while uveitis developed in 3.4% patients. Mortality in neonates with persistent disease was comparable to the mortality in neonates with non-persistent disease.[81]

Viruses account for about 1% of infections in hospitalized neonates.[85] The most common viral infections are due to enterovirus/parechovirus (Table IV). Enteroviruses were responsible for the highest mortality and development of serious sequelae.[85] Respiratory syncytial virus (RSV) is the second most common virus causing infections in hospitalized neonates (14–37%).[42,52,60,85–89] Respiratory viruses were diagnosed in 29.5% of neonates on mechanical ventilators; the most frequent was RSV (14.1%), followed by influenza A virus (10.2%).[88] In another study, nosocomially acquired RSV infection was present in 37% of neonates, 54.3% had an underlying condition predisposing to severe disease and 13% died.[52] Human parainfluenza type 3 is the most common cause of bronchiolitis and pneumonia after respiratory syncytial virus. Parainfluenza type 3 virus was isolated in six of 17 neonates cultured (five symptomatic patients and one asymptomatic patient). Eighteen of 52 nursing personnel had been ill during the previous week, concomitantly with cough and nasal congestion.[93] Nosocomial transmission of rotavirus in neonates has been reported.[43,57,90] The onset of acute diarrhoea due to rotavirus in two neonates was followed by five neonates developing gastroenteritis with the same strain of rotavirus.[90] In another study, in 51% of inpatients with nosocomial gastroenteritis, the causative agent was rotavirus and 26% of those were premature neonates.[43]

HAIs and resistance to antimicrobial agents

Compared to community-acquired infections, HAIs are often caused by multi-drug-resistant pathogens. In this section we concentrate on reports from the subcontinent and other resource-poor settings. In a retrospective study of bacterial isolates from cases of neonatal septicaemia over a period of 5 years, there was a significant rise in the incidence of drug-resistant *Acinetobacter* spp. and *P. aeruginosa*.[99] The incidence rate of acinetobacter septicaemia in another study was 11.1/1000 live births.[100] Other studies have also documented *Acinetobacter* spp. as emerging neonatal pathogens.[23,33,101] Susceptibility tests showed that acinetobacter isolates were resistant to two or more antibiotics, most notably to ampicillin (82.5%), cephalexin (69.6%), gentamicin (66.5%) and cefotaxime (47.8%). Most isolates were susceptible to amikacin (82.6%), ciprofloxacin (73.9%) and piperacillin (69.6%).[33]

Only about 30% of bacterial aetiological agents of neonatal HAI would be covered by an empirical regimen of ampicillin and gentamicin.[102] Gram-negative organisms causing HAI in neonates were

Table IV Hospital-acquired viral infections

Virus	Lessons learnt
Enterovirus/parechovirus[85]	Common cause of hospital-acquired respiratory infection. High mortality and serious sequelae. Infection transmitted from visiting family.
Respiratory syncytial virus (RSV)[42,52,60,85–89]	Nosocomially acquired infection among 2/4 neonates with RSV. Infection control measures successful.[86]
Rotavirus[43,57,85,87,90,91]	Nosocomial transmission of rotavirus in 5/9 neonates with diarrhoea.[90] Winter peak. High morbidity. Among patients with nosocomial rotavirus diarrhoea, 26% were preterm neonates.[43]
Cytomegalovirus (CMV)[85,92]	Transfusion-acquired CMV in 2/21 neonates.[92]
Adenovirus[85]	Gastroenteritis was the main clinical presentation in preterm infants.
Parainfluenza, type 3[85,93]	Outbreak in NICU ($N = 6$). Linked to HCW. Controlled by glove, gown and cohorting.[93]
Herpes simplex virus, rhinovirus, rubellavirus[85]	Infections reported in the NICU.
Influenza A virus[88]	Neonates on mechanical ventilation were nosocomially infected with influenza A virus.
Human coronaviruses[63]	Patient-to-staff and staff-to-patient transmission in NICU. Universal precaution with surface disinfection and handwashing prevent spread of infection.
Echovirus type 7 Coxsackie B3[94]	Nosocomial outbreak ($N = 6$) in special care nursery. Transmission by staff.

NICU, neonatal intensive care unit; HCW, healthcare worker.

less susceptible to the commonly used antibiotics, such as ampicillin (20.7%), amoxicillin (25.4%), gentamicin (56.8%), ceftazidime (28.4%) and cefotaxime (44.8%). These organisms were more susceptible to imipinem (76.4%), amikacin (77.7%), ofloxacin and ciprofloxacin (88.1%).[14,103] Other workers found third-generation cephalosporins and aminoglycosides such as netilmicin to be effective in the treatment of neonatal sepsis.[104] At the same time, studies have also shown that administration of antimicrobial prophylaxis, presumed to prevent HAIs, can be a putative risk factor in itself for HAI.[16,54,78]

Single-centre studies have shown that probiotics containing anaerobic bacteria may reduce the rate and severity of necrotizing enterocolitis.[105]

Antifungal agents

Fluconazole has been recommended as prophylaxis against systemic fungal infections in preterm low birthweight infants.[106,107] However, other workers have found no resurgence of fungal infection after cessation of prophylactic fluconazole use.[108] There is also concern about emergence of resistance to fluconazole. In an investigation into the resurgence of bloodstream infections due to *C. parapsilosis* in one unit, after the institution of fluconazole prophylaxis, primary resistance to fluconazole was not detected.[109] Others propose a twice weekly dosing of prophylactic fluconazole to decrease candida colonization, invasive infection, cost and patient exposure in high-risk preterm infants weighing <1000 g at birth; the lower and less frequent dosing may even delay or prevent the emergence of antifungal resistance.[110]

There are reports of *C. albicans* resistance to fluconazole (12.5%) and amphotericin-B (25%) in studies from India.[80] Newer antifungal agents, including voriconazole and caspofungin, show promise in the treatment of potentially fatal fungal infections in neonates and additional controlled studies are indicated to evaluate their role.[111]

Principles of infection control in the neonatal unit

The existing evidence base for infection control practices specifically for the neonatal unit is described in Table V.[22,29,39,48,55,58–60,63,72,86,87,93,112–118]

Important lessons in infection control can be learnt from published accounts of specific outbreaks. In addition to the outbreaks documented in Tables III and IV, we have selected other outbreak reports that we believe reinforce the infection control message (Table VI).[30,31,58,59,113,114,119–124]

Environmental surveillance is not routinely recommended since pathogens present in the inanimate NICU environment, e.g. floors, walls, sink-drains or furniture are not associated with

Table V Recommended infection control practices

Policy	Practice
Infection control policy and practice	Handwashing, gown, gloving, mask, cohorting uninfected neonates, isolation of infected neonates, short natural fingernails in healthcare staff, thorough cleaning, better patient care facilities, strict winter visiting policies.[22,55,58–60,63,86,87,93,112–114]
Disinfection and maintenance of equipment	Surface disinfection; disinfection of ventilators.[58,60,63,93,112]
Single-use items	Use of disposable endotracheal tubes; mucous extraction suction catheters and hand towels. An expensive option in the resource-poor setting.[47]
Infrastructure and staffing	Regular water supply; improve staff:patient ratio; adequate infrastructure; sick leave policy for staff.[48,59,115,116]
Surveillance and monitoring	Aggressive case finding, notification of contacts; screening cultures for antibiotic resistance; screening for MRSA; surveillance cultures of the environment in outbreak settings; surveillance and monitoring for resistant flora.[29,39,59,72,87,114,116,117]
Antibiotic policy	Adoption of an evidence-based antibiotic policy in the neonatal unit; refers to a 10-point plan on antibiotic use.[118]

MRSA, meticillin-resistant *Staphylococcus aureus*.

HAIs. Only three NICU sites, namely baby placements, resuscitation equipment and various cleansing solutions, were found to be significantly associated with HAIs ($P < 0.001$) in one study.[72] The relative risk of infection was greatest if baby placement sites were colonized (odds ratio = 7.48; $P < 0.01$). This reinforces the need for scrupulous cleaning regimens rather than adopting a policy of routine environmental surveillance.

However, environmental cultures may play a role in specific outbreak situations. Outbreak strains of *Salmonella worthington* were isolated from the baby warmer mattress, baby cot, suction machine bottle and wall of the refrigerator.[114] The role of surveillance cultures to predict the onset of nosocomial infections in neonates undergoing invasive procedures, such as exchange transfusion, has been studied.[29] The authors found that except for staphylococci, the flora from umbilical stump and umbilical vein blood in asymptomatic neonates was similar to the flora from infected neonates.

'Intensive care' need not be synonymous with 'invasive care'.[3] In the presence of constraints such as lack of trained staff, intermittent power supply or lack of disinfection between their use, incubators and other medical devices can be a risk factor for HAI. In these situations kangaroo care provided by the mother has emerged as a cost-effective and widely accepted style of caring for an infant in hospital. In a study from India, there was significant improvement among the kangaroo care group compared with the conventional group, in terms of hypothermia (10/44 vs 21/45, $P < 0.01$), higher oxygen saturations (95.7 vs 94.8%, $P < 0.01$) and decrease in respiratory rates (36.2 vs 40.7, $P < 0.01$). However, there was no statistically significant difference in the incidence of hyperthermia, sepsis, apnoea, onset of breastfeeding and hospital stay in the two groups.[125] Further studies are needed to evaluate the role of kangaroo care and the incidence of HAI in neonates.

Discussion

The role of microbiology in the detection, epidemiological analyses and prevention of HAIs cannot be overemphasized, whether the unit is one that benefits from being resource rich or resource poor. In a setting where most physicians are reluctant to use first-line agents, due to misleading or lack of sufficient susceptibility data, a qualified microbiologist is indispensable. Communication between microbiologist and neonatologist helps in deciding the most probable pathogen and in initiating the most appropriate antimicrobial therapy. The formulation of a mutually agreed antibiotic policy at community, institutional and national levels is imperative.

Table VI Outbreak investigations that provide valuable lessons in infection control

Organism	Outbreak investigation
Klebsiella spp.	Outbreak of septic arthritis ($N = 17$) linked to contaminated cover sheets.[31]
P. aeruginosa	Epidemiological evidence of an association between acquiring *P. aeruginosa* bloodstream infection in neonates and exposure to nurses with long and artificial fingernails. Short natural fingernails is a policy that is essential to reduce the incidence of HAI in neonates.[113]
Serratia marcescens	An outbreak of invasive *S. marcescens* in the NICU ($N = 14$).[119] Molecular tests showed that a vast majority of clinical and environmental isolates (from hands of nurse, handwashes and disinfectants) belonged to the same clonal type. Cohorting of non-infected neonates, isolation of colonized and infected neonates, glove use and handwashing controlled the outbreak. Outbreak ($N = 9$) of *S. marcescens* in the NICU.[58] Epidemic strain isolated from handwashes and doors of incubators. Strict handwashing, disinfection of incubators, cohorting and isolating patients controlled further transmission.
Acinetobacter spp.	During an outbreak, isolates with similar antibiogram were recovered from intravenous catheter and washbasin.[30,120]
Listeria monocytogenes	Neonatal cross-infection due to contaminated equipment resulted in sepsis and central nervous system disease.[121]
Salmonella worthington	Outbreak of seven cases, six fatalities. Equipment and environment were the source of outbreak. Outbreak was controlled through cleaning and fumigation.[114]
Shigella sonnei	Transmission among nursery staff.[59]
Enterotoxigenic *E. coli* (ETEC)	Outbreak involved preterm neonates ($N = 16$); surveillance cultures of swabs from the utensils used to prepare milk feed, culture of the formula feed and all items handled by one particular cook were undertaken. The cook's hand swabs and faecal sample yielded growth of ETEC. The outbreak was controlled by appropriate therapy and institution of proper measures of hygiene.[122]
Enterobacter spp.	Outbreak ($N = 30$ and $N = 10$) of *Enterobacter cloacae* septicaemia traced to preceding bladder catheterization and/or parenteral nutrition solution, respectively.[123,124]

An infection control team (ICT) comprising an infection control nurse or an infection control trained link nurse in the NICU, a neonatologist/physician and a microbiologist must actively participate in outbreak management and infection control policy issues. In turn, it is mandatory that microbiologists balance their focus equally on diagnostic as well as clinical microbiology. Microbiological influence and involvement can be enhanced if the microbiologist joins regular clinical ward rounds and helps to raise awareness among healthcare professionals regarding all aspects of infectious disease management. Education and training is an important remit of the ICT. Besides training of healthcare staff we believe it is important to provide training to empower the mother. As the main carer in the family her education is vital; if she can be made aware of the rationale behind the microbiologist or neonatologist's advice, she will be in a stronger position to participate in the wellbeing of herself and the baby.

Even as huge efforts are underway to halt the misuse of antimicrobial agents, issues regarding antimicrobial resistance in pathogens are less important to the lay public. As long as these essential drugs are available over-the-counter in many countries, all efforts in any other part of the world toward preventing their misuse will be undermined. In addition, a number of privately funded laboratories have sprung up in several cities and towns in developing countries. They lack quality assurance and the personnel who work in identifying pathogens and reporting susceptibility are not trained adequately in quality control methods.

In resource-limited settings, as in technologically advanced units, advising that we wash our hands and use the most appropriate antimicrobial agent may be more valuable than suggesting expensive tools for molecular testing. We provide a simple, resource-efficient template for the instigation and maintenance of infection control in the clinical setting (Box 2). In the present era of global information sharing, professionals working in the area of infection control need not feel isolated. There are several useful web tools that provide practical information and guidance; our own

Box 2 Ten cost-effective steps towards infection control in the neonatal unit

1. Ensure a strict protocol for hygienic handwashing and provision of clinical handwash basins or sinks
2. Involve the microbiologist in the planning stages or when refurbishing the unit; advice on physical setting of the unit and general layout of cots, bays, sinks will impact on infection control
3. Provision of side rooms and bays for the isolation of infected babies or protection of healthy neonates
4. Provide training and advice regarding environmental cleaning; ensuring that all surfaces are maintained clean and dry
5. Create an infection control policy document and a rational antibiotic policy that is constantly reviewed
6. Appoint an infection control team (ICT) comprising a microbiologist, neonatologist, infection control nurse/liaison nurse trained in infection control
7. Support the ICT in the management of infectious diseases and in promoting infection control practices
8. Provide education and training of unit staff in infection control
9. Take the lead in outbreak investigation and control
10. Install a laboratory surveillance system for alert organisms (i.e. important pathogens causing hospital-acquired infections and their susceptibility patterns)

policy is available free of charge at www.infection-controlservices.co.uk/.

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