REVIEW ARTICLE

Drugs 2008; 68 (9): 1227-1238
0012-6667/08/0009-1227/$53.45/0

Necrotizing Enterocolitis in Newborns
Pathogenesis, Prevention and Management

Alecia M. Thompson and *Matthew J. Bizzarro*

Department of Pediatrics, Yale University School of Medicine, New Haven, Connecticut, USA

Contents

Abstract

Necrotizing enterocolitis (NEC) is primarily a disease process of the gastrointestinal (GI) tract of premature neonates that results in inflammation and bacterial invasion of the bowel wall. Despite advances in the care of premature infants, NEC remains one of the leading causes of morbidity and mortality in this population. It occurs in 1–5% of all neonatal intensive care admissions and 5–10% of all very low birthweight (<1500 g) infants. Although research has presented an interesting array of potential contributing factors, the precise aetiology of this multifactorial disease process remains elusive. Historically, it was believed that NEC arose predominantly from ischaemic injury to the immature GI tract, yet alternate plausible hypotheses indicate that many factors are likely to be involved. These may include issues related to the introduction and advancement of enteric feeding, alterations in the normal bacterial colonization of the GI tract, bacterial translocation and activation of the cytokine cascade, decreased epidermal growth factor, increased platelet activating factor, and mucosal damage from free radical production.

Clinical manifestations of NEC may be vague, including increased episodes of apnoea, desaturations, bradycardia, lethargy and temperature instability. There may also be GI-specific symptoms such as feeding intolerance, emesis, bloody stools, abdominal distention and tenderness, and abdominal wall discolouration.

Laboratory values may be indicative of infection, coagulation abnormalities and fluid retention. Radiographic signs may include ileus, dilated or fixed intestinal loops, air in the intestinal wall or free air in the abdomen. Medical treatment typically consists of bowel rest and decompression, antibacterial therapy, and management of other haematological or electrolyte imbalances. Increased respiratory and cardiovascular support is sometimes needed. In neonates who do not respond adequately to medical management, or if pneumoperitoneum is present, surgical intervention may occur with either use of a peritoneal drain or laparotomy.

Advances in antenatal and neonatal care have resulted in increased survival of extremely preterm neonates. As this at-risk population continues to increase, an effective preventative strategy for NEC is needed. One preventative strategy is the use of antenatal corticosteroids to enhance maturation of the fetus if preterm delivery is likely. Recommendation of use of breast milk, early initiation of trophic feeds and judicoius advancement of enteric feeds are current postnatal strategies. Other preventative strategies that have been investigated include the use of oral antibacterials, antioxidants, supplementation of arginine and epidermal growth factor, none of which have changed clinical practice. Recent promising data indicate that prophylactic use of probiotics may play a role in preventing the onset of NEC. However, more large-scale, definitive studies are needed.

Necrotizing enterocolitis (NEC) is a multifactorial disease process of the gastrointestinal (GI) tract of newborns and infants that may result in mucosal and/or transmural necrosis. Although the precise aetiology of the disorder is unknown, NEC is primarily a disease of premature neonates, with 90–95% of cases occurring in infants born before 36 weeks' gestational age. It occurs in approximately 1–5% of all admissions to the newborn intensive care unit and approximately 5–10% of all very low birthweight (<1500 g) neonates.[1-4]

Despite recent advances in antenatal and postnatal care of premature infants, NEC remains one of the leading causes of significant morbidity and mortality in this population. Current management strategies primarily consist of medical stabilization and efforts aimed at halting progression of disease once it has occurred. Recently, novel therapeutic strategies for prevention of onset of the disease, including the use of probiotics, have been investigated with some intriguing results.

1. Pathogenesis

Although NEC typically occurs in premature neonates, the exact aetiology of its onset remains uncertain. Although many antenatal and postnatal risk factors for NEC have been identified, the majority of these factors are related to one or more of the following: (i) hypoxic-ischaemic injury to the GI tract; (ii) physiological immaturity of the GI tract; and (iii) alterations in the normal microbiological flora of the intestines (figure 1).

1.1 Hypoxic-Ischaemic Injury

Ischaemic injury to the GI tract is believed to be a major contributing and potentially inciting factor in NEC. Any process that results in hypoperfusion and subsequent hypoxic injury to the GI tract, including, but not limited to, the presence of a patent ductus arteriosus, sepsis, polycythaemia, *in utero* cocaine exposure, peri- or postnatal asphyxia, respiratory distress syndrome, congenital heart disease, the presence of umbilical catheters and exchange transfusion, may predispose the neonate to developing NEC.[1,3,4] In the setting of hypoperfusion, the Herring-Breur or 'diving reflex' is initiated, whereby blood is shunted away from 'less important' organs, such as the intestines, towards critical organs, such as the brain and heart. This process may result in hypoxic-ischaemic injury to the intestines.[1,4] Reperfusion may then trigger the proinflammatory cascade, resulting in damage to the mucosal barrier

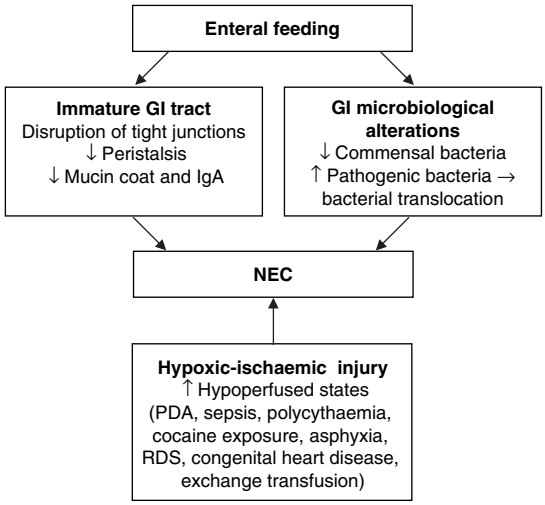

Fig. 1. Proposed multifactorial pathophysiology culminating in the onset of necrotizing enterocolitis (NEC). **GI** = gastrointestinal; **PDA** = patent ductus arteriosus; **RDS** = respiratory distress syndrome; ↑ indicates increased; ↓ indicates decreased; → indicates leads to.

of the intestines and creating the potential for bacterial invasion and translocation. Although this proposed mechanism might explain many cases of NEC, in others, no inciting hypoxic-ischaemic event is identifiable. In addition, most infants who do have evidence of hypoxic-ischaemic injury do not develop NEC.[1,3,5,6] Therefore, it is likely that several factors, some known and some unknown, contribute to this disease process.

1.2 Physiological Immaturity of the Gastrointestinal (GI) Tract

NEC is almost exclusively a disease process of preterm neonates. Therefore, it is likely that factors related to the physiological immaturity of the GI tract are involved. Anand et al.[7] proposed the following three major altered components of the intestinal barrier of preterm neonates that may contribute to the onset of NEC: (i) disruption of the integrity of epithelial tight junctions; (ii) impaired peristalsis; and (iii) deficiencies in components of the mucus coat.

Tight junctions between epithelial cells of the intestines serve to create a barrier to fluid and other molecules. Release of proinflammatory cytokines after reperfusion injury, for example, may disrupt the integrity of this barrier and result in intestinal hyperpermeability. Among other things, this may lead to bacterial invasion and translocation via the intestinal wall. Peristalsis is also impaired in preterm neonates until approximately the eighth month of gestation.[8] Peristalsis decreases the amount of time that bacterial antigens are in contact with enterocytes. Thus, impaired peristalsis may not only allow an increased carbohydrate load from enteral feeding to serve as bacterial substrate, but may also prolong exposure of the gut to bacterial antigens, thereby inducing an inflammatory response. In addition to tight junctions and peristalsis, the mucus coat is an important structural barrier of the GI tract, which produces, among other things, IgA. IgA, present in mucous secretions, provides protection against pathogenic organisms. Deficient IgA production in premature infants may ultimately facilitate bacterial translocation across the intestinal mucosa.[2]

Other attributes of the premature GI system that may predispose an infant to NEC include decreased production of epidermal growth factor, an enzyme important for repair of GI cells.[9-11] Increased presence of platelet activating factor (PAF), a potent inflammatory and vasoconstrictive mediator, and a decreased ability to neutralize PAF via its acetylhydrolase degrading enzyme, are additional factors present in premature neonates that may predispose them to NEC.[12] Nitric oxide, a vasodilator and free radical molecule, has also been implicated in NEC. Its upregulation in ill premature neonates causes damage to the intestinal epithelium by direct membrane peroxidation and induction of apoptosis.[6,7,9-12]

1.3 Alterations in Normal GI Microbiological Flora and the Role of Enteral Feeding

The aetiology of NEC is often thought to be due to an infectious agent because clusters of NEC can occur. Abnormal colonization of the intestinal tract of premature neonates may contribute to the onset of NEC. Commensal species of bacteria and fungi normally inhabit the GI tract and aid in certain aspects of digestion. Preterm neonates, given their high risk of infection, are frequently exposed to broad-spectrum antibacterials, which may alter the normal flora of their GI tract. The predominant species of bacte-

ria found in the GI tract of healthy, term, breast fed infants is *Bifidobacteria*.[13] In contrast, species of *Staphylococcus*, *Enterobacter*, *Enterococcus* and *Clostridia* are the predominant faecal bacterial species in premature neonates undergoing intensive care, with very little colonization with *Bifidobacteria*.[13,14] Species of *Escherichia* and other bacterial, viral and fungal pathogens, including rotavirus and species of *Candida*, have also been implicated in the aetiology of NEC.

In the susceptible premature intestine with decreased motility, absorption and mucosal integrity, incomplete immunological defences, and decreased commensal bacteria, proliferation of more pathogenic bacteria may occur.[1,6,7] Bacteria, both commensal and pathogenic, use carbohydrates from enteral feedings as a substrate to grow and proliferate.[4] Indeed, enteral feedings were reported as having been administered prior to the onset of the disease in >90% of patients with NEC.[2,3] There are distinct associations between volume, rate, type of feeding and the development of NEC. The proposed rationale behind these associations is that excessive volume and/or a rapid rate of increase in feeding volume may overwhelm the ability of the GI tract to propel and digest its contents. Large volumes of substrate in a premature GI tract with decreased motility and digestive capabilities, and increased pathogenic bacteria, may result in stasis and proliferation of pathogenic bacteria.[15-19] Bacterial proliferation may then result in invasion and bacterial translocation into the bowel wall, with subsequent production of inflammatory cytokines and endotoxins, which may result in mucosal and/or transmural necrosis.[4]

Table I. Clinical presentation of necrotizing enterocolitis

Signs and symptoms
Apnoea
Bradycardia
Oxygen desaturation
Lethargy
Irritability
Temperature instability
Abdominal distension
Abdominal tenderness
Absent bowel sounds
Feeding intolerance
Abdominal wall cellulitis
Blood in stool
Emesis
Increased gastric residuals
Laboratory abnormalities
Metabolic acidosis
Thrombocytopenia
Leukopenia
Leukocytosis with ↑ bandcells
Glucose instability
Hyponatraemia
Coagulation abnormalities (↑ PT/PTT, ↓ fibrinogen)
↑ C-reactive protein
Radiographic abnormalities
Intestinal dilatation
Presence of fixed loop of dilated bowel
Thickening of the intestinal wall
Ascites
Pneumatosis intestinalis
Air in the portal venous system
Pneumoperitoneum

PT = prothrombin time; **PTT** = partial thromboplastin time; ↑ indicates increased, ↓ indicates decreased.

2. Presentation and Staging

NEC is a disease process that typically presents in the first 2 weeks of life, with a slight predilection for the male sex and Black race.[1-4] The onset of disease can manifest in a myriad of ways, including clinical signs and symptoms, and laboratory and radiographic findings (table I).

Some of the signs and symptoms of early, suspected NEC are relatively nonspecific and common to other neonatal disease processes, and, therefore, may result in varying management strategies. These may include systemic signs such as increased apnoea, bradycardia and/or episodes of oxygen desaturation, onset of lethargy or irritability, and temperature instability. Abdominal signs related to the pathophysiology of the GI tract, but still somewhat nonspecific for NEC, may include abdominal distension and tenderness, absent bowel sounds, presence of blood in the stool, emesis and increased gastric feeding residuals.[20]

Abnormalities on laboratory evaluation can also be found in association with NEC and may include the onset of metabolic acidosis, thrombocytopenia, leukopenia, leukocytosis with an increase in the proportion of immature white blood cells, glucose

instability, hyponatraemia, coagulation abnormalities (including disseminated intravascular coagulation) and increased C-reactive protein level.[1,20]

Radiographic evidence of NEC can also be helpful in both diagnosing and staging disease, and may include intestinal dilatation, bowel wall thickening and pneumatosis intestinalis. Pneumatosis intestinalis is caused by bacterial translocation into the bowel wall with production of hydrogen gas. This can then be seen radiographically as linear or circular lucencies within the intestinal wall. In more severe NEC, gas in the portal venous system and pneumoperitoneum may occur.[20,21]

Based on a combination of these findings, the severity of NEC can be staged according to the modified Bell's criteria (table II), with higher stages of disease being associated with greater risk of an adverse outcome.[20]

3. Adverse Outcomes Associated with Necrotizing Enterocolitis

NEC is a common cause of death in preterm neonates, with a higher predilection for those most premature and with more advanced stages of disease. Despite early diagnosis and optimal management, 25–33% of all infants with NEC will die.[1,6] In addition to the threat of death, NEC is the second most common cause of significant morbidity in premature newborns. These morbidities primarily include short- and long-term GI problems and neurodevelopmental impairment.[6,20,23]

GI morbidities related to NEC primarily include malabsorption and failure to thrive because of lost or dysfunctional bowel, and problems related to long-term use of parenteral nutrition, including catheter-related blood stream infections, cholestasis and liver failure.[24] Stricture formation from damaged intestinal mucosa may occur in up to 20% of all patients with proven NEC and usually requires surgery. Liver and/or intestinal transplants are becoming more of an option for managing the long-term complications of NEC. Between 14% and 43% of all causes of intestinal failure leading to referral for liver and intestinal transplant in the US are as a result of NEC.[24,25]

In addition to GI-related complications, significant neurological morbidity may occur in neonates surviving NEC. Increased adverse neurological se-

Table II. Modified Bell's staging criteria for necrotizing enterocolitis (reproduced from Kliegman and Walsh,[20] © 1987 with permission from Elsevier, and Bell et al.,[22] with permission)

Stage	Systemic signs	Abdominal signs	Radiographic signs	Treatment
IA: suspected	Temperature instability, apnoea, bradycardia, lethargy	Gastric retention, abdominal distension, emesis, guaiac-positive stool	Normal or intestinal dilation, mild ileus	NPO, antibacterials for 3 days
IB: suspected	Same as for stage IA	Grossly bloody stool	Same as for stage IA	Same as for stage IA
IIA: definite, mildly ill	Same as for stage IA	Same as for stage IB, plus absent bowel sounds with or without abdominal tenderness	Intestinal dilation, ileus, pneumatosis intestinalis	NPO, antibacterials for 7–10 days
IIB: definite, moderately ill	Same as for stage IA, plus mild metabolic acidosis and thrombocytopenia	Same as for stage IIA, plus absent bowel sounds, definite tenderness, with or without abdominal cellulitis or right lower quadrant mass	Same as for stage IIA, plus ascites	NPO, antibacterials for 14 days
IIIA: advanced, severely ill, intact bowel	Same as for stage IIB, plus hypotension, bradycardia, severe apnoea, combined respiratory and metabolic acidosis, DIC and neutropenia	Same as for stage IIB, plus signs of peritonitis, marked tenderness and abdominal distension	Same as for stage IIB	NPO, antibacterials for 14 days, fluid resuscitation, inotropic support, ventilator therapy, paracentesis
IIIB: advanced, severely ill, perforated bowel	Same as for stage IIIA	Same as for stage IIIA	Same as for stage IIA, plus pneumoperitoneum	Same as for stage IIIA, plus surgery

DIC = disseminated intravascular coagulation; **NPO** = *nil per os* (nothing by mouth).

quelae have been observed in neonates with NEC requiring surgical intervention compared with infants treated medically or the premature neonatal population in general.[26] Rees et al.[23] performed a systematic review of ten studies comparing outcome data at a median of 20 months from 7843 extremely low birthweight infants, 821 of whom were diagnosed with NEC. Neurodevelopmental impairment was seen in 45% of neonates with NEC compared with 35% (odds ratio [OR] 1.60; 95% CI 1.30, 2.00) of those with similar gestational age and birthweight but without NEC. In neonates with NEC, 20% developed cerebral palsy, 3% visual impairment, 3% hearing impairment, 36% cognitive impairment and 35% developed psychomotor impairment. The rates of impairment were higher in neonates with more advanced stages of disease. Surgical NEC carried an increased risk of cerebral palsy (OR 2.74; 95% CI 1.44, 5.21) and psychomotor impairment (OR 1.90; 95% CI 1.10, 3.20) compared with those who were managed medically.

4. Management

4.1 Medical Management

Most therapeutic options for NEC are aimed at treating the disease process once it has occurred and limiting the progression of disease. NEC can be treated both medically and surgically, depending on the stage of disease. In earlier stages of disease (stage ≤IIIA; see table II), treatment often consists of bowel rest and administration of parenteral antibacterials to limit bacterial invasion and translocation. Initially, affected neonates are given nothing by mouth and total parenteral nutrition is administered for nutritional support. A full evaluation for systemic infection via bacterial translocation should be performed, including sampling of the blood, urine and cerebrospinal fluid, if possible, for culture. Parenteral antibacterials should also be administered. Antibacterial coverage is aimed at aerobic and anaerobic enteric species of bacteria and may include ampicillin or vancomycin for coverage of Gram-positive species; an aminoglycoside, such as gentamicin, or a third-generation cephalosporin, such as cefotaxime, for coverage of Gram-negative aerobes; and metronidazole or clindamycin for coverage of anaerobic organisms (table III). When the diagnosis of NEC is considered definite (stage ≥IIA; see table II), parenteral antibacterials are administered, in conjunction with bowel rest, for a period of 7–14 days.[4,20]

In addition to clinical monitoring and cardiopulmonary support, when needed, radiographic evaluations are routinely performed to evaluate for progression of disease. This may initially include scheduled abdominal radiographs every 4–8 hours or as needed based on changes in clinical status. Ultrasound has recently been evaluated for its utility in diagnosing and monitoring patients with NEC. Ultrasound may be able to help diagnose patients earlier than traditional abdominal radiographs,[21] and may also be useful for detecting pneumatosis intestinalis, portal venous gas, intraperitoneal gas, abnormal calibre of the bowel wall and lack of perfusion to the bowel.[21,27-29] However, use of ultrasound is currently limited because of its high inter-operator variability and lack of availability at all times of the day and night.

In addition to clinical and radiographic assessment, laboratory values should be monitored closely based on the instability of the infant. Interventions, such as administration of packed red cells to improve oxygen carrying capacity, and platelet and fresh frozen plasma transfusions to lessen the risk of haemorrhage, should be initiated to correct abnormalities that may contribute to progression of the disease process. Electrolyte levels and acid-base status should also be monitored and corrected as needed.

Table III. Proposed antimicrobials for treatment of necrotizing enterocolitis

Antibacterial	Type of bacterial coverage
Ampicillin (or other penicillin derivative)	Gram-positive enteric organisms
Vancomycin	Gram-positive enteric organisms
Gentamicin (or other aminoglycoside)	Synergy for *Enterococcus*; Gram-negative enteric organisms
Cefotaxime (or other third-generation cephalosporin)	Gram-negative enteric organisms
Metronidazole	Anaerobic enteric organisms
Clindamycin	Anaerobic enteric organisms

4.2 Surgical Management

If the infant does not improve significantly despite medical treatment for several days, if pneumoperitoneum is present, or if clinical, laboratory and/or radiographic disease worsen, surgical management if often required. In fact, 20–60% of neonates with NEC will require some type of surgical intervention.[1,16,30] For many neonates with intestinal perforation, laparotomy is performed with resection of the necrosed portion of intestine and an ostomy is created at the end proximal to the resected bowel. The distal end of the resected region may be used to create a mucous fistula. Alternatively, some centres may elect to perform resection and primary anastomosis.[31,32] In some situations, a second laparotomy may be performed to re-evaluate areas of intestine with questionable viability. This 'second look' may occur at 24–48 hours after the initial procedure to verify that perfusion is adequate or to determine if that area has succumbed to necrosis and needs to be resected. The bowel may then be re-anastomozed several weeks after recovery.[4,20]

The decision to perform a surgical procedure may be affected by the relative instability of the neonate and his or her size, as this affects the ability of the neonate to tolerate surgery and anaesthesia. Up to 50% of extremely low birthweight (<1000 g) infants who undergo surgery for NEC may die.[20] Extremely low birthweight and unstable infants with intestinal perforation may have similar outcomes with peritoneal drainage as with surgery.[25,30,33] Peritoneal drainage facilitates removal of infected or inflammatory fluid, and, in conjunction with the medical management strategies outlined in section 4.1, aids in resolution of the peritonitis. Drainage is typically performed for approximately 1 week, at which time the drain is removed if the patient improves. If no improvement is seen, the patient may still undergo laparotomy.[25,33]

5. Prevention

As NEC represents the most common neonatal GI emergency, much work has been done to evaluate potential antenatal and postnatal preventative strategies aimed at the proposed pathophysiological mechanisms of disease.

5.1 Antenatal Corticosteroids

Administration of maternal antenatal corticosteroids for threatened preterm delivery has been shown to decrease the incidence of NEC.[34] The proposed mechanism is similar to that involved in improvement of fetal lung maturation with administration of corticosteroids because the GI tract also matures under the influences of exogenous corticosteroids. The intestinal barrier is presumably strengthened, thus decreasing the potential for translocation of bacteria and bacterial endotoxins. The anti-inflammatory properties of corticosteroids may also exert some protective effect on the GI tract. Roberts and Dalziel[34] reviewed eight randomized controlled trials assessing the effect of antenatal corticosteroids on NEC in a total of 1675 infants. Their results indicated that this therapy was beneficial, with a relative risk reduction of NEC of 0.46 (95% CI 0.29, 0.74). Administration of maternal antenatal corticosteroids for threatened preterm labour has now become a standard therapy because of these and other benefits to the neonate.

5.2 Trophic Feedings

'Trophic', 'non-nutritive' or 'priming feeds' are initiated to stimulate peristalsis of the GI tract and induce appropriate enzymatic activity for digestion. In 2003, Berseth et al.[15] published findings indicating that trophic feeds decreased the incidence of NEC. A total of 71 infants were fed 20 mL/kg/day of unfortified breast milk or preterm formula for 10 days. In the second group, 70 infants were fed 20 mL/kg/day initially, advancing subsequently by 20 mL/kg/day each day starting on day 2 to a maximum of 140 mL/kg/day. A total of 10% in the advancing group developed NEC compared with approximately 1% in the trophic-fed group ($p = 0.03$). Many alternate strategies for enteral feeding initiation and advancement are utilized in preterm neonates, but the introduction of non-nutritive feedings for some period of time prior to advancement is often utilized and may be beneficial in the prevention of NEC.

5.3 Breast Milk

Breast milk contains maternal IgA, epidermal growth factor, PAF acetylhydrolase (which inacti-

vates PAF), prebiotic elements (which favour the replication of commensal GI bacteria) and anti-inflammatory cytokines such as interleukin-10. These substances combine to facilitate digestion and replication of protective bacteria, and to provide protection to the mucosal barrier of the GI tract.[7,10,16-18,35] One consistently observed difference between breast milk-fed and formula-fed infants is that breast milk-fed infants have lower counts of potentially pathogenic bacteria, such as *Clostridia*, *Enterococcus* and *Enterobacter*, and higher counts of more typical commensal species, such as *Staphylococcus* and *Bifidobacteria*.[35] In a multicentre case-control study, it was noted that all 53 cases of NEC had begun enteral milk feeding prior to the diagnosis.[19] A total of 70% of patients with stage II/III NEC received breast milk versus 92% of matched controls who received formula (OR 0.19; 95% CI 0.05, 0.73).

In 1990, Lucas and Cole[36] performed a prospective multicentre study to evaluate the role that early feeding choices played in the development of NEC. Their findings indicated that of neonates who were fed formula exclusively, confirmed NEC was 6-fold more common than in neonates who were fed breast milk alone. Furthermore, the risk of NEC was 3.5-fold higher in neonates who were fed formula exclusively than in those who were fed a combination of breast milk and formula.

Sisk et al.[16] evaluated whether high proportions (≥50%) of enteral feeds comprised of human milk fed within the first 14 days of life were protective against NEC in very low birthweight infants. 202 infants were grouped into those being fed with low (<50%) or high (≥50%) proportions of human milk. Five of 46 (10%) of the low human milk group developed NEC compared with 5 of 156 (3%) of the high human milk group (OR 0.17; 95% CI 0.04, 0.68). Based on these findings, the use of breast milk in the preterm population is preferable.

5.4 Oral Antibacterials

The pathophysiology of NEC may include an overabundance of pathogenic bacteria and, therefore, oral antibacterials may be of some use in preventing the onset of disease. In 1998, Siu et al.[37] published results of a double-blind, randomized, controlled study using oral vancomycin versus placebo to prevent NEC. Nine of 71 (13%) premature neonates developed NEC in the vancomycin group compared with 19 of 69 (28%) in the placebo group ($p = 0.035$). Gentamicin has also been evaluated for prophylactic use against NEC. In one investigation, oral gentamicin was administered for 1 week in premature neonates to prevent NEC. In this double-blind, randomized, controlled trial, 20 neonates received gentamicin, while 22 received placebo.[38] None of the 20 patients administered gentamicin developed NEC compared with 4 of 22 (18%) who received placebo ($p = 0.05$).

Bury and Tudehope[39] performed a meta-analysis of five studies involving 456 infants in order to evaluate the use of enteral antibacterials for prevention of NEC. Their main finding was that there was a significant reduction in the rate of NEC with prophylactic administration of an enteral aminoglycoside (relative risk [RR] 0.47; 95% CI 0.28, 0.78). There was also a significant reduction in NEC-related mortality in infants who received oral antibacterials (RR 0.32; 95% CI 0.10, 0.96). However, a significant increase in rates of colonization with resistant bacteria was also noted in the group administered enteral antibacterials (RR 1.73; 95% CI 1.00, 2.97).[39]

Although potentially effective, prophylactic use of oral antibacterials to prevent NEC has not been widely adopted for several reasons. NEC may involve bacterial invasion with any or a number of Gram-positive and aerobic or anaerobic Gram-negative species. Therefore, treatment with one antibacterial is unlikely to provide adequate antimicrobial coverage in all cases. In addition, the widespread use of antibacterials may be associated with the emergence of antibacterial-resistant organisms. As a consequence, this strategy is not currently recommended.

5.5 Probiotics

One of the most promising interventions for the prevention of NEC may be the use of probiotics. Probiotics are oral supplements containing microbes that are usually indigenous to the healthy human body. Probiotics are believed to provide a benefit to preterm neonates by enhancing the IgA mucosal response, improving the mucosal protective barrier, increasing the production of anti-inflammatory cy-

tokines, decreasing intestinal wall permeability and competitively excluding pathogenic microbes in the GI tract.[40] Some probiotics produce lactic acid, which inhibits proliferation of some species of pathogenic bacteria, while preferentially enhancing growth of most commensal species. Probiotics also play a beneficial role in protein and carbohydrate digestion.[40] Clinically, these effects may manifest as improved feeding tolerance, decreased time to achieving full enteral feedings, decreased incidence of NEC and decreased incidence of systemic infections in the populations studied. Studies have shown that supplementation with probiotics, mostly *Bifidobacteria*, *Lactobacillus* and *Saccharomyces*, have reduced the incidence and severity of NEC with few adverse effects.[41-45]

Various studies have specifically assessed the colonization pattern of premature neonates supplemented with various probiotics. The underlying hypothesis is that NEC is precipitated by colonization of the immature GI tract with pathogenic species, thereby inciting a systemic inflammatory response. Therefore, supplementation with probiotics may confer a benefit by allowing flora that are usually indigenous to the GI tract to flourish and compete with more pathogenic species. Costalos et al.[41] randomly assigned 87 neonates born at a gestational age of 28–32 weeks to receive either the probiotic *S. boulardii* (n = 55) or maltodextrins (n = 36) for 30 days. Their aim was to investigate the ability of *S. boulardii* to modify neonatal intestinal flora and its function. The numbers of *Bifidobacteria* cultured were significantly higher in the group that received *S. boulardii*. Conversely, stool colonies of *E. coli* and species of *Enterococcus* were significantly lower in the *S. boulardii* group. NEC occurred in 10% of the probiotic group compared with 16% in the control group (OR 0.50; 95% CI 0.15, 1.98). Bacteraemia occurred in 6% of the probiotic group compared with 8% of the placebo group (OR 0.70; CI 0.13, 3.6). However, *S. boulardii* was not isolated from any positive blood culture.

A large, multicentre, double-blinded study by Dani et al.[42] in 2002 in Italy evaluated the efficacy of *Lactobacillus rhamnosus* supplementation in reducing the incidence of urinary tract infection, bacterial sepsis and NEC in preterm infants. Infants <32 weeks' gestational age or <1500 g birthweight were randomized to receive either *Lactobacillus* or placebo. 585 patients were randomized, of whom 295 received probiotics and 290 received placebo. Urinary tract infections and NEC occurred in lower proportions of supplemented infants, but the differences were not statistically significant.

In 2005, a study published by Lin et al.[43] evaluated the use of probiotics in reducing the incidence and severity of NEC in very low birthweight neonates fed enterally and surviving beyond the 7th day of life. 367 neonates were enrolled, of whom 180 were randomized to the probiotic group and 187 to the control group. The probiotic used was a combination of *L. acidophilus* and *B. infantis*. The incidence of NEC, defined as greater than stage II, in the probiotic groups was 2 of 180 (1%) compared with 10 of 187 (5%) in the control group (p = 0.04). The numbers needed to treat to prevent one case of NEC and one death due to NEC were 27 and 31, respectively. No differences in severity of NEC were observed between groups. No blood cultures were positive for *B. infantis* or *L. acidophilus* during the study period.

In 2005, Bin-Nun et al.[44] evaluated the use of probiotics to decrease the incidence and severity of NEC in enterally fed very low birthweight neonates. Neonates either received no supplementation or a daily feeding of a mixture of *B. infantis*, *Streptococcus thermophilus* and *B. bifidus*. The probiotic and control groups contained 72 and 73 infants, respectively. NEC occurred in 4% of patients in the probiotic group and 16% in the control group (p = 0.03). In addition, the severity of NEC was significantly reduced in the probiotic group (mean stage 1.50 vs 2.30 in the control group; p = 0.005).

In 2007, Deshpande et al.[45] performed a systematic review of randomized controlled trials, including some of those discussed here,[41-44] to evaluate the efficacy of probiotics in preventing stage ≥II NEC in neonates <33 weeks' gestational age and <1500 g birthweight. Their analysis consisted of seven studies that initiated probiotic supplementation within the first 10 days of life. The probiotics used included strains of *Lactobacillus*, *Saccharomyces* and *Bifidobacteria*. 1393 neonates were analysed, consisting of 690 controls and 703 in the probiotic group. In the control group, 38 of 690 (6%)

infants developed NEC compared with 15 of 703 (2%) of infants who were administered probiotics (RR 0.36; 95% CI 0.20, 0.65). A reduction in the risk of all-cause mortality was also observed in the probiotic group based on data from five trials (n = 1268; RR 0.47; 95% CI 0.30, 0.73). The potential harm of probiotics was also evaluated by comparing the risk of bacteraemia in the treatment and placebo groups. Data from six trials were included (n = 1355) and analysis revealed no significant difference in the risk of bacteraemia between neonates given probiotics and those who did not receive probiotics (RR 0.94; 95% CI 0.74, 1.20).

Although probiotics may be a promising approach for prevention and decreased severity of NEC, issues exist regarding the standardization of an appropriate probiotic supplement for neonates. Most studies have utilized various combinations of bacteria and amounts of culture-forming units for different lengths of time. These differences in methodology have created difficulties in elucidating the most beneficial probiotic supplement for the premature population. Questions remain concerning the strains or combinations of strains that offer the best benefit. Potential exists for a significant difference in the magnitude of the benefit when administered to formula versus breast-fed neonates. There are also uncertainties over the optimal time to start probiotics in order to confer maximal benefit, and the long-term consequences of probiotic supplementation. A multicentre, randomized, controlled trial currently in development by the US National Institute of Child Health and Human Development Neonatal Research Network may answer some of these questions.

5.6 Prebiotics

There are concerns about the potential infectious threat of probiotics to neonates.[46] This has resulted in investigation of the use of prebiotics. These are compounds that selectively increase the population of existing commensal GI bacteria. They act as substrates peferentially utilized by commensal bacterial to proliferate.[47]

Kapiki et al.[35] compared the effect of use of a prebiotic, fructo-oligosaccharide supplemented formula on the gut flora of preterm neonates with that of a placebo-supplemented formula. At the end of 7 days, the number of *Bifidobacteria* in the stools was determined to be significantly higher in the prebiotic group. The percentage of infants colonized with *Bifidobacteria* was also significantly higher in the prebiotic group (90% vs 70%, respectively; $p < 0.05$). Conversely, the numbers of *E. coli* and *Enterococcus* in the stools were significantly lower than in the placebo group. This study did not report any findings on the incidence of NEC in their study populations.

5.7 Other

Other preventative strategies have been researched with no significant effect on the prevention of NEC. Administration of oral IgG and IgA was theorized to offer protective effects in neonates in light of their relative immunoglobulin deficiency. Multiple studies were therefore conducted and reviewed, but no effect on the rates of NEC was observed.[48,49] Superoxide dismutase, a naturally occurring antioxidant that prevents damage from free oxygen radicals, and N-acetylcysteine, another antioxidant, were evaluated for their potential to prevent NEC. No protective effects were seen with either antioxidant.[50,51] Epidermal growth factor plays a role in the repair and maintenance of the GI tract, and is present in the breast milk of humans and other mammals. The results of epidermal growth factor supplementation were promising in an animal model, but its administration to infants diagnosed with NEC resulted in only minor improvement.[52,53] Arginine was considered a potential agent for prevention of NEC when it was discovered that levels of arginine may be decreased in affected neonates.[54,55] The proposed effect of arginine in this setting is a reversal of vasoconstriction induced by nitric oxide deficiency in premature infants under stress. A decreased incidence of all stages of NEC in premature infants supplemented with arginine has been observed in one study.[56] However, these findings have yet to be confirmed.

6. Conclusions

NEC is a multifactorial disorder of the GI tract of premature neonates with a variety of inciting events culminating in a similar terminal pathway and clinical picture. The incidence and mortality of NEC

have remained the same in recent years as have the management strategies, which are primarily aimed at limiting progression of disease once it has occurred. Recent efforts have focused on prevention of the onset of disease. One such strategy, the prophylactic use of probiotic supplementation, seems promising. However, many issues need to be addressed with respect to standardizing this supplement for general use in the premature population and assessing the potential risks of its generalized use.

Acknowledgements

No sources of funding were used to assist in the preparation of this review. The authors have no conflicts of interest directly or indirectly related to the content of this review.

References

1. Lin PW, Stoll BJ. Necrotising enterocolitis. Lancet 2006; 368 (9543): 1271-83
2. Gibbs K, Lin J, Holzman IR. Necrotising enterocolitis: the state of the science. Indian J Pediatr 2007; 74 (1): 67-72
3. Kliegman RM. Models of the pathogenesis of necrotizing enterocolitis. J Pediatr 1990 Jul; 117 (1 Pt 2): S2-5
4. Panigrahi P. Necrotizing enterocolitis: a practical guide to its prevention and management. Pediatr Drugs 2006; 8 (3): 151-65
5. Hsueh W, Caplan MS, Qu X, et al. Neonatal necrotizing enterocolitis: clinical considerations and pathogenic concepts. Pediatr Dev Path 2002; 6 (1): 6-23
6. Henry M, Moss RL. Current issues in the management of necrotizing enterocolitis. Sem Perinatol 2004; 28 (3): 221-33
7. Anand RJ, Leaphart CL, Mollen KP, et al. The role of the intestinal barrier in the pathogenesis of necrotizing enterocolitis. Shock 2007; 27 (2): 124-33
8. Berseth CL. Gestational evolution of small intestine motility in preterm and term infants. J Pediatr 1989; 115 (4): 646-51
9. Warner BW, Warner BB. Role of epidermal growth factor in the pathogenesis of neonatal necrotizing enterocolitis. Semin Pediatr Surg 2005; 14 (3): 175-80
10. Warner BB, Ryan AL, Seeger K, et al. Ontogeny of salivary epidermal growth factor and necrotizing enterocolitis. J Pediatr 2007; 150 (4): 358-63
11. Shin CE, Falcone RA, Stuart L, et al. Diminished epidermal growth factor levels in infants with necrotizing enterocolitis. J Pediatr Surg 2000; 35 (2): 173-7
12. Rabinowitz SS, Dzakpasu P, Piecuch S, et al. Platelet-activating factor in infants at risk for necrotizing enterocolitis. J Pediatr 2001; 138 (1): 81-6
13. Millar M, Wilks M, Costletoe K. Probiotics for preterm infants? Arch Dis Child Fetal Neonatal Ed 2003; 88 (5): 354-8
14. Harmsen HJ, Wildeboer-Veloo AC, Raangs GC, et al. Analysis of intestinal flora development in breast-fed and formula-fed infants by using molecular identification and detection methods. J Pediatr Gastroenterol Nutr 2000; 30 (1): 61-7
15. Berseth CL, Bisquera JA, Paje VU. Prolonging small feeding volumes early in life decreases the incidence of necrotizing enterocolitis in very low birth weight infants. Pediatrics 2003; 111 (3): 529-34
16. Sisk PM, Lovelady CA, Dillard RG, et al. Early human milk feeding is associated with a lower risk of necrotizing enterocolitis in very low birth weight infants. J Perinat 2007; 27 (12): 428-33
17. Lee JS, Polin RA. Treatment and prevention of necrotizing enterocolitis. Sem Neonatol 2003; 8 (6): 449-59
18. Updegrove K. Necrotizing enterocolitis: the evidence for use of human milk in prevention and treatment. J Hum Lact 2004; 20 (3): 335-9
19. Henderson G, Craig S, Brocklehurst P, et al. Enteral feeding regimens and necrotizing enterocolitis in preterm infants: multi-center case-control study. Arch Dis Child Fetal Neonatal Ed. Epub 2007 Sep 3
20. Kliegman RM, Walsh MC. Neonatal necrotizing enterocolitis: pathogenesis, classification, and spectrum of disease. Curr Prob Pediatr 1987; 17 (4): 213-88
21. Epelman M, Daneman A, Navarro OM, et al. Necrotizing enterocolitis: review of state-of-the-art imaging findings with pathologic correlation. Radiographics 2007; 27 (2): 285-305
22. Bell MJ, Ternberg JL, Feigin RD, et al. Neonatal necrotizing enterocolitis: therapeutic decisions based upon clinical staging. Ann Surg 1978; 187 (1): 1-7
23. Rees CM, Pierro A, Eaton S. Neurodevelopmental outcomes of neonates with medically and surgically treated necrotizing enterocolitis. Arch Dis Child Fetal Neonatal Ed 2007; 92 (3): 193-8
24. Goulet O, Sauvat F. Short bowel syndrome and intestinal transplantation in children. Curr Opin Clin Nutr Metab Care 2006; 9 (3): 304-13
25. Blakely ML, Tyson JE, Lally KP, et al. Laparotomy versus peritoneal drainage for necrotizing enterocolitis or isolated intestinal perforation in extremely low birth weight infants: outcomes through 18 months adjusted age. Pediatrics 2006; 117 (4): 680-7
26. Walsh MC, Kliegman RM, Hack M. Severity of necrotizing enterocolitis: influence on outcome at 2 years of age. Pediatrics 1989; 84 (5): 808-14
27. Merritt CR, Goldsmith JP, Sharp MJ. Sonographic detection of portal venous gas in infants with necrotizing enterocolitis. Am J Roentgenol 1984; 143 (5): 1059-62
28. Faingold R, Daneman A, Tomlinson G, et al. Necrotizing enterocolitis: assessment of bowel viability with color doppler ultrasound. Radiology 2005; 235 (2): 587-94
29. Silva CT, Daneman A, Navarro OM, et al. Correlation of sonographic findings and outcome in necrotizing enterocolitis. Pediatr Radiology 2007; 37 (3): 274-82
30. Moss RL, Dimmitt RA, Henry MC, et al. A meta-analysis of peritoneal drainage versus laparotomy for perforated necrotizing enterocolitis. J Pediatr Surg 2001 Aug; 36 (8): 1210-3
31. Hall NJ, Curry J, Drake DP, et al. Resection and primary anastomosed is a valid surgical option for infants with necrotizing enterocolitis who weight less than 1000 g. Arch Surg 2005; 140 (12): 1149-51
32. Griffiths DM, Forbes DA, Pemberton PJ, et al. Primary reanastomosis for necrotising enterocolitis: a 12-year experience. J Pediatr Surg 1989; 24 (6): 515-8
33. Moss RL, Dimmitt RA, Blakely ML, et al. Laparotomy versus peritoneal drainage for necrotizing enterocolitis and perforation. N Engl J Med 2006; 354 (21): 2225-34
34. Roberts D, Dalziel S. Antenatal corticosteroids for accelerating fetal lung maturation for women at risk of preterm birth. Cochrane Database Syst Rev 2006; (3): CD004454
35. Kapiki A, Costalos C, Oikonomidou C, et al. The effect of a fructo-oligosaccharide supplemented formula on gut flora of preterm infants. Early Hum Dev 2007; 83 (5): 335-9
36. Lucas A, Cole TJ. Breast milk and neonatal necrotizing enterocolitis. Lancet 1990; 336 (8730): 1519-23

37. Siu YK, Ng PC, Fung SCK, et al. Double blind, randomized, placebo controlled study of oral vancomycin in prevention of necrotizing enterocolitis in preterm, very low birthweight infants. Arch Dis Child Fetal Neonatal Ed 1998; 79 (2): 105-9
38. Grylack LJ, Scanlon JW. Oral gentamicin therapy in the prevention of neonatal necrotizing enterocolitis: a controlled double-blind trial. Am J Dis Child 1978; 132 (12): 1192-4
39. Bury RG, Tudehope D. Enteral antibiotics for preventing necrotizing enterocolitis in low birthweight or preterm infants. Cochrane Database Syst Rev 2001; (1): CD000405
40. Hammerman C, Bin-Nun A, Caplan M. Germ warfare: probiotics in defense of the premature gut. Clin Perinatol 2004; 31 (3): 489-500
41. Costalos C, Skouteri V, Gounaris A, et al. Enteral feeding of premature infants with *Saccharomyces boulardii*. Early Hum Dev 2003; 74 (2): 89-96
42. Dani C, Biadaioli R, Bertini G, et al. Probiotics feeding in prevention of urinary tract infection, bacterial sepsis and necrotizing enterocolitis in preterm infants: a prospective study. Biol Neonate 2002; 82 (2): 103-8
43. Lin HC, Su BH, Chen AC, et al. Oral probiotics reduce the incidence and severity of necrotizing enterocolitis in very low birth weight infants. Pediatrics 2005; 115 (1): 1-4
44. Bin-Nun A, Bromiker R, Wilschanski M, et al. Oral probiotics prevent necrotizing enterocolitis in very low birth weight neonates. J Pediatr 2005; 147 (2): 192-6
45. Deshpande G, Rao S, Patole S. Probiotics for prevention of necrotizing enterocolitis in preterm infants with very low birthweight: a systematic review of randomized controlled trials. Lancet 2007; 369 (9573): 1614-20
46. Kunz AN, Noel JM, Fairchok MP. Two cases of lactobacillus bacteremia during probiotic treatment of short gut syndrome. J Pediatr Gastroenterol Nutr 2004; 38 (4): 457-8
47. Parracho H, McCartney AL, Gibson GR. Probiotics and prebiotics in infant nutrition. Proc Nutr Soc 2007; 66 (3): 405-11
48. Lawrence G, Tudehope D, Baumann K, et al. Enteral human IgG for prevention of necrotizing enterocolitis: a placebo-controlled, randomized trial. Lancet 2001; 357 (9274): 2090-4
49. Foster J, Cole M. Oral immunoglobulin for preventing necrotizing enterocolitis in preterm and low birth-weight neonates. Cochrane Database Syst Rev 2004; (1): CD001816
50. Davis JM, Rosenfeld WN, Richter SE, et al. Safety and pharmacokinetics of multiple doses of recombinant human CuZn superoxide dismutase administered intratracheally to premature neonates with respiratory distress syndrome. Pediatrics 1997; 100 (1): 24-30
51. Soghier LM, Brion LP. Cysteine, cystine or N-acetylcysteine supplementation in parenterally fed neonates. Cochrane Database Syst Rev 2006; (4): CD004869
52. Dvorak B, Halpern MD, Holubec H, et al. Epidermal growth factor reduces the development of necrotizing enterocolitis in a neonatal rat model. Am J Physiol Gastrointest Liver Physiol 2002; 282 (1): G156-64
53. Sullivan PB, Lewindon PJ, Cheng C, et al. Intestinal mucosa remodeling by recombinant human epidermal growth factor 1-48 in neonates with severe necrotizing enterocolitis. J Pediatr Surg 2007; 42 (3): 462-9
54. Becker RM, Wu G, Galanko JA, et al. Reduced serum amino acids concentrations in infants with necrotizing enterocolitis. J Pediatr 2000; 137 (6): 785-93
55. Zamora SA, Amin HJ, McMillan DD, et al. Plasma L-arginine concentrations in premature infants with necrotizing enterocolitis. J Pediatr 1997; 131 (2): 226-32
56. Amin HJ, Zamora SA, McMillan DD, et al. Arginine supplementation prevents necrotizing enterocolitis in the premature infant. J Pediatric 2002; 140 (4): 425-31

Correspondence: Dr *Matthew J. Bizzarro*, Department of Pediatrics, Yale University School of Medicine, 333 Cedar Street, P.O. Box 208064, New Haven, CT 06520-8064, USA. E-mail: matthew.bizzarro@yale.edu