Necrotizing Enterocolitis: Relationship to Innate Immunity, Clinical Features, and Strategies for Prevention

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Necrotizing Enterocolitis: Relationship to Innate Immunity, Clinical Features, and Strategies for Prevention

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Objectives After completing this article, readers should be able to:

1. Define the pathophysiology of necrotizing enterocolitis (NEC).
2. Explain the relationship among minimal enteral nutrition, subsequent feedings, and NEC.
3. List the factors that render human milk enteral feedings protective against NEC.
4. Explain what measures should promote advances in mortality and morbidity associated with NEC in the near future.
5. Describe the complications of NEC.

Introduction
Necrotizing enterocolitis (NEC) is one of the most serious and devastating diseases encountered in the neonatal intensive care unit (NICU). It is the most common gastrointestinal malady in neonates. It affects 1% to 8% of all infants admitted to the NICU and has a mortality rate of 10% to 50%. NEC accounts for at least 1,000 deaths annually in the United States. The increased incidence of NEC over the past few decades may be attributable to advancements in perinatal care, which have allowed very preterm infants to survive long enough to develop NEC.

The only consistently defined risk factor for NEC is prematurity; the incidence varies inversely with birthweight and gestational age. NEC strikes 4% to 13% of all very low-birthweight babies (<1,500 g). Although NEC primarily affects preterm infants, 5% to 28% of cases occur in term and near-term babies. The disease is rare in older children.

Asphyxia, congenital heart disease, polycythemia, umbilical catheterization, and the use of indomethacin and methylxanthines have been proposed as risk factors, although none has been shown to have a consistent association with NEC.

The age of onset of NEC also varies inversely with gestational age. In term infants, the median age at onset is 2 days; the preterm infant may develop the disease at several weeks of age. The risk remains high in preterm infants until they reach 35 to 36 weeks postconceptional age. The disparity in age of onset between term and preterm infants suggests that the condition in term infants may be due to a disease process different from that seen in preterm infants.

Clinical Presentation
The clinical presentation of NEC is highly variable, ranging from mild feeding intolerance or abdominal distention to fulminant shock and death. The classic description involves abdominal distention, microscopically-to-grossly bloody stools, gastric residuals, and possibly localizing abdominal signs. Laboratory evaluation may reveal a normal-to-elevated white blood cell count with a shift to immature precursors, neutropenia, thrombocytopenia, and disseminated intravascular coagulation (DIC). Electrolyte abnormalities and metabolic acidosis are also common. Initial radiographic evaluation may reveal a fixed, dilated loop of bowel. The characteristic radiologic finding is pneumatosis intestinalis, which is believed to be intraluminal gas produced by bacte-

Abbreviations

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Definition</th>
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<tbody>
<tr>
<td>DIC</td>
<td>disseminated intravascular coagulation</td>
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<tr>
<td>Ig</td>
<td>immunoglobulin</td>
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<tr>
<td>LPS</td>
<td>lipopolysaccharide</td>
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<tr>
<td>NEC</td>
<td>necrotizing enterocolitis</td>
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<tr>
<td>NICU</td>
<td>neonatal intensive care unit</td>
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<tr>
<td>NPO</td>
<td>nil per os</td>
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<tr>
<td>PAF</td>
<td>platelet-activating factor</td>
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<td>TLR</td>
<td>toll-like receptor</td>
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rrial fermentation. Pneumatosis intestinalis is found in 70% to 80% of confirmed cases of NEC. Portal venous gas or pneumoperitoneum may be seen with severe cases.

Staging of NEC is according to a modification of the system first proposed by Bell and colleagues in 1978. The staging system is based on clinical, laboratory, and radiologic findings. Stage I signs are nonspecific and can be due to a number of problems, including sepsis and simple feeding intolerance. Stage II and III criteria are more specific for NEC: pneumatosis intestinalis, pneumoperitoneum, grossly bloody stools, DIC, and metabolic acidosis.

**Histology/Gross Pathology**

Histologically, NEC is characterized by mucosal edema, inflammation, hemorrhage, coagulation necrosis, and mucosal ulceration. The terminal ileum and proximal colon are the sections of bowel affected most commonly, although more extensive disease is possible from the stomach to the rectum.

**Pathophysiology**

The pathophysiology of NEC, as yet incompletely understood, has been the subject of recent study. In simple terms, the condition can be considered an aberrant response of the immature gut and immune system in the setting of enteral nutrition and the presence of bacteria.

**Feeding**

The introduction of enteral nutrition is a key component in the pathogenesis of NEC. Unfed infants rarely develop the disease; indeed, 90% to 95% of infants who develop NEC have been exposed to recent enteral volume advancement. Enteral nutrition may predispose to NEC by disrupting mucosal integrity, reducing gut motility, or altering gastrointestinal blood flow. Appropriate feeding advancements have not been elucidated for preterm infants, but it appears that aggressively increasing feedings increases the risk of NEC. The type of feeding also is critical. The fetal intestine, despite being exposed to high volumes of amniotic fluid, is not prone to NEC. Human milk appears to decrease the risk of NEC compared with formula. Finally, the osmolality of formulas is important, with those that have high osmotic loads being associated with NEC.

**Bacteria/Infection**

Considerable attention has been directed to the relationship of intestinal microbes and the pathogenesis of NEC. Most of the attention to date has focused on microorganisms as causative of NEC. More recent evidence is accumulating to suggest that a lack of normal intestinal commensal microflora might play a critical role in pathogenesis. On the one hand, bacteremia occurs in up to 35% of cases of NEC. It is unclear whether bacteria play a primary role in the pathogenesis of NEC or bacterial translocation occurs due to an already compromised intestinal barrier. Either way, the fact that cases may occur in clusters suggests that infectious agents play an important role in the disease process.

No pathogen is found consistently in NEC. Bacteria reported to be associated with NEC include *Escherichia coli*, *Klebsiella*, *Enterobacter*, *Pseudomonas*, *Salmonella*, *Clostridium*, coagulase-negative *Staphylococcus*, and *Enterococcus* sp. Gram-positive organisms are more common in stages I and II disease, and enteric organisms predominate in more severe cases. Viral pathogens, namely, rotavirus, coronavirus, and enteroviruses, also have been described in association with NEC. On the other hand, bacteria also play a primary role in normal intestinal homeostasis. There are approximately $10^{13}$ commensal microbes in the adult human gut. At birth, however, the gut is sterile. In the newborn period, the gut becomes colonized with *E. coli* and streptococci from the maternal vaginal flora. The type of nutrition that a baby receives appears to have an impact on subsequent colonization patterns, with formula-fed infants showing an early increase in *Enterobacteriaceae* colonization and the guts of breast-fed infants colonized early with *Enterobacteriaceae* and *Bifidobacterium*.

The composition of the intestinal microflora has been found to be important not only in the pathogenesis of NEC but also in intestinal homeostasis. Recent studies have shown that toll-like receptors (TLRs) located on the intestinal epithelia interact both with commensal organisms and with pathogenic organisms. The interaction between these TLRs and pathogenic organisms may initiate a cascade that translates into gut injury. However, as has been shown in mouse models, if certain TLRs such as TLR2, TLR4, or a downstream signaling molecule (MyD88) are “knocked out,” the gut becomes more susceptible to injury (Fig. 1). Interestingly, the absence of any microorganisms also mediates an aberrant response, and treatment with TLR agonists (ligands) such as lipoteichoic acid or lipopolysaccharide (LPS) actually may prevent intestinal injury. In addition, the relationship between the presence of commensal microflora and the development of immunity in the form of T-cell function is being clarified. Therefore, the presence of commensal organisms, and not just the absence of possible pathogens, is important for maintaining intestinal...
integrity. This raises the question of the potentially del-
etious role of broad-spectrum antibiotics, commonly
initiated in the preterm population, which likely dramat-
ically alters the intestinal microflora. The disruption
of normal flora, combined with a breach in intestinal
epithelial integrity, causes an increase in inflammatory
mediators, including platelet-activating factor (PAF),
LPS, tumor necrosis factor-alpha, and nuclear factor-
kappa-B. The release of cytokines and reactive oxygen
species further compromises intestinal health.

Immature Gut
An immature gastrointestinal tract and immune response
predispose preterm infants to NEC. The preterm intesti-
te demonstrates increased permeability to high-
molecular weight molecules, decreased motility, and
decreased mucus production compared with the gastro-
intestinal tract of more mature infants. Figure 2 shows a
scheme of how this could result in not only intestinal
injury such as NEC, but also distal organ damage
through initiation of systemic inflammation. In addition,
preterm infants show an inappropriate cellular and hu-
moral response to intestinal pathogens. Lastly, the im-
mature gastrointestinal tract shows impaired circulatory
dynamics in response to an enteral volume load.

Role of Hypoxia–Ischemia
The relationship between hypoxic-
ischemic events and NEC is contro-
versial. Hypoxia–ischemia was long
believed to play a prominent role
in the pathogenesis of NEC, but
much of the supporting evidence
was anecdotal. Unfortunately, this
has been one of the major reasons
for neonatologists withholding en-
teral feeding from babies who have
experienced low or even borderline
low Apgar scores. There is, in real-
ity, a dearth of literature to imply
icate hypoxic–ischemia as a primary
contributor to cases of NEC in pre-
term infants. The late occurrence
of NEC in preterm infants who
do not have preceding evidence of
ischemia argues against hypoxia–
ischemia as a primary causative
agent in the development of NEC.
However, histologic examination
does reveal coagulation necrosis,
suggesting the occurrence of isch-
emia, perhaps as a secondary event
in the pathogenesis of NEC. Epidemiologic studies also
have not shown an association between NEC and as-
phyxial events.

Treatment
Medical Management
Prompt medical intervention is critical in the initial man-
agement of NEC. The patient should be made nil per os
(NPO), with gastric decompression undertaken with an
orogastric tube. For confirmed cases (Stages II or III),
NPO status should continue for 7 to 14 days; for stage I
NEC, clinical judgment may dictate a shorter course.
During this period, parenteral nutrition is required to
optimize nutritional support. Enteral feedings may be
resumed cautiously when the baby stabilizes, as evi-
denced by normal abdominal examination results, nor-
mal bowel sounds, and no blood or gastric residuals.

Infants who develop severe NEC may require inten-
sive support. Hypotension and respiratory failure may
develop, requiring intervention. The potential for coagu-
lopathy and electrolyte disturbance requires close moni-
toring and correction, as necessary.

Parenteral antibiotics should be administered expedi-
tiously to cover enterococci, staphylococci, and coli-
forms. If perforation is suspected, anaerobic coverage
also should be initiated. Local resistance patterns dictate specific antibiotic selection.

**Surgical Indications/Management**

Surgical intervention is required in 27% to 63% of cases of confirmed NEC. Pneumoperitoneum is the only absolute indication for surgical intervention. Clinical deterioration despite maximal medical intervention also is considered by most to warrant surgery. Relative indications include increased abdominal distention or discoloration, portal vein gas, or a fixed intestinal loop.

Laparotomy with resection of nonviable bowel remains the current standard of care. Peritoneal drainage also has been employed as a bridge to laparotomy when an infant is judged too sick to tolerate laparotomy. Peritoneal drainage without laparotomy or primary peritoneal drainage also has been suggested as definitive therapy for infants who have complicated NEC. Two ongoing randomized, controlled trials should shed more light on the issue.

**Prevention**

**Infection Control**

Good hygiene practice (eg, handwashing) has been demonstrated to be effective in controlling outbreaks of NEC.

**Trophic Feedings**

Minimal enteral nutrition, also referred to as trophic, trickle, or hypocaloric feedings, is the term applied to topical nutrition of the small intestine administered in the first week or two after birth. During this time, enteral feedings are limited for several days, with most nutrition provided parenterally. After this initial period, enteral feedings are advanced judiciously. Infants exposed to minimal enteral feedings have not shown an increased risk of NEC compared with infants kept NPO. This “gut priming” has been shown to increase gut motility, reduce the incidence of cholestasis, and improve tolerance of subsequent feedings. There is increasing evidence that not introducing at least small amounts of food to the gastrointestinal tract results in atrophy and predisposes the intestine to inflammation and likely translocation of bacteria (Fig. 3).

Minimal enteral feedings appear to be safe, even in babies receiving mechanical ventilation or catheterized with umbilical lines. Although yet unproven, this approach may be protective against NEC and sepsis in the very preterm population (Fig. 4).

**Human Milk**

Enteral feeding with human milk has been shown to decrease the severity of NEC compared with formula feeding. Fresh human milk contains many immunoprotective factors, such as immunoglobulins (Igs), lactoferrin, neutrophils, lymphocytes, lysozyme, and PAF acetylhydrolase (which inhibits PAF). Human milk also is believed to promote intestinal colonization with *Lactobacillus*. The efficacy of banked human milk is less clear because freezing and pasteurization reduce the cellular components and Igs of the milk.

**Antenatal Steroids**

Antenatal steroids are not administered to expectant mothers as prophylaxis against NEC, but when administered to prevent respiratory distress syndrome, antenatal steroids decrease the risk for NEC. The mechanism by which steroids protect against NEC is unclear but probably involves a decrease in inflammatory mediators,
increased activity of enzymes involved in digestion and absorption, and potentially maturation of the microvillus membrane. A protective effect of postnatal glucocorticoids has not been demonstrated clearly, although their long-term use to treat chronic lung disease has been associated with adverse neurodevelopmental effects.

Oral Antibiotics

Oral antibiotics, primarily aminoglycosides, clearly reduce the incidence of NEC and death due to NEC. However, concerns regarding the emergence of bacterial resistance have rendered oral antibiotic prophylaxis an inappropriate intervention.

Probiotics

The human gut is normally colonized with commensal organisms such as *Bifidobacterium* and *Lactobacillus* sp. As mentioned previously, the widespread use of broad-spectrum antibiotics in the NICU disrupts development of normal microflora and allows for growth of pathogenic organisms. Promising recent studies have shown that probiotic supplementation has a positive impact on the incidence of NEC in preterm infants. Further study is necessary to determine which probiotic formulation provides the maximal protection. Recent evidence suggests that probiotic bacteria may not need to be alive to impart a positive effect; even bacterial products, such as TLR ligands or CpG segments of DNA, may be effective in ameliorating inflammation in the intestine. This is a very fertile area for additional research.

Immunoglobulins

Another possible preventive measure is oral supplementation with Ig. One randomized trial showed protection against NEC in preterm infants given an IgA-IgG preparation. However, a recent Cochrane review, which examined five studies on Ig supplementation, found no significant difference in NEC or death from NEC with IgG or IgG-IgA preparations. Of note, no randomized, controlled trials have evaluated IgA-only preparations.

Acidification

Preterm infants have been shown to have decreased gastric acid produc-
tion in response to enteral feeding. An acid environment is an important barrier to many pathogenic microorganisms. One prospective, randomized study demonstrated a lower incidence of NEC in infants whose enteral feedings were supplemented with acid to keep the pH between 3 and 4. Although promising, these results need follow-up confirmation before routine supplementation can be recommended.

**Glutamine**
Glutamine has been shown to be important in immunity and intestinal function. Previous studies in animals and critically ill adults have shown improved outcomes with glutamine supplementation. Although the studies in human infants, including two large multicenter trials, have not demonstrated decreased NEC with glutamine supplementation, it is important to remember that none were designed to address this specific issue. Furthermore, a couple of single-center studies in human neonates have shown decreased sepsis and improved intestinal function with glutamine supplementation. Two larger multicenter trials (one using the enteral and the other using the parenteral route of glutamine supplementation) did not show a decrease in sepsis, but the trial of enteral supplementation suggested a lower prevalence of intraventricular hemorrhage and periventricular leukomalacia. Animal and human studies have demonstrated decreased intestinal inflammation with glutamine administration, which may be germane to the prevention of not only intestinal injury but also distal organ injury that occurs as a result of intestinal inflammation. More needs to be learned about this important amino acid as it relates to preterm infants.

**Arginine**
Arginine is a substrate for nitric oxide, which demonstrates vasodilatory and anti-inflammatory properties. Arginine also is a precursor for other amino acids that are important in intestinal homeostasis, including glutamine and glutamate. Although initial results suggest that arginine supplementation may play a role in protecting against NEC, a recent Cochrane review noted that further study is needed before a recommendation for arginine supplementation can be made.

**Complications/Outcomes**

**Strictures**
Infants who survive episodes of NEC are at risk for serious complications, the most common of which is stricture formation. Up to 39% of affected patients develop stricture(s). Strictures may form as soon as 2 weeks following acute disease. Most strictures are colonic. Occasionally, partial strictures may present with recurring episodes of sepsis and feeding intolerance, which should be ruled out with appropriate radiologic contrast studies.

**Malabsorption/Short Gut**
Although short gut syndrome and malabsorption are expected when significant amounts of bowel need to be resected, malabsorption also occurs in cases of NEC that do not require resection. This may be due to persistent mucosal injury from an ongoing inflammatory process.

**Complications Associated with Total Parenteral Nutrition**
Most infants who have confirmed NEC require central catheterization for prolonged parenteral nutrition. The catheters employed for total parenteral nutrition are associated with significant risks of infection, vessel perforation, and iatrogenic pleural and pericardial effusions. The use of parenteral nutrition and NPO status also place the patient at risk for cholestasis and delay in intestinal maturation.

**Recurrence**
As many as 6% of infants who develop NEC experience disease recurrence.

**Extraintestinal Complications**
Infants who recover from an episode of NEC have been shown to be at increased risk of developing extraintestinal complications, such as sepsis, bronchopulmonary dysplasia, and neurodevelopmental delay. A recent retrospective study found that infants who had experienced an episode of NEC are at increased risk for microcephaly, short stature, and serious developmental delay. An increased incidence of spastic diplegia and hemiplegia suggests periventricular damage. It is intriguing that an event long believed to be limited to the gastrointestinal tract may have broader consequences.

**Conclusion**
NEC continues to be a source of considerable consternation in the NICU. Despite wonderful advances in the care of the preterm neonate over the past decades, there has been little improvement in the morbidity and mortality associated with NEC. Ongoing research, which continues to elucidate the pathophysiology of this disease process, should provide novel avenues for intervention. Rather than therapies initiated after NEC strikes, preventive strategies targeting at-risk populations appear
to hold the most promise for improving outcomes in the future.

Suggested Reading
Hoyos AB. Reduced incidence of necrotizing enterocolitis associated with enteral administration of *Lactobacillus acidophilus* and *Bifidobacterium infantis* to neonates in an intensive care unit. *Int J Infect Dis*. 1999;3:197–202
Neu J. The “myth” of asphyxia and hypoxia-ischemia as primary causes of necrotizing enterocolitis. *Biol Neonate*. 2005;87:97–99
NeoReviews Quiz

4. Necrotizing enterocolitis (NEC) is the most common gastrointestinal malady in neonates. Of the following, the most consistent risk factor for NEC is:
   A. Birth asphyxia.
   B. Congenital heart disease.
   C. Indomethacin administration.
   D. Preterm gestation.
   E. Umbilical catheterization.

5. NEC is characterized histologically by mucosal edema, inflammation, hemorrhage, coagulation necrosis, and mucosal ulceration. Of the following, the section of the bowel most commonly affected in NEC is the:
   A. Distal colon.
   B. Duodenum.
   C. Jejunum.
   D. Stomach.
   E. Terminal ileum.

6. NEC is considered an aberrant response of the immature gut and immune system in the setting of enteral nutrition and the presence of bacteria. Of the following, the most accurate statement regarding the epidemiology, pathophysiology, and treatment of NEC is that:
   A. Age of onset of NEC varies directly with gestational age.
   B. Enterally unfed infants rarely develop NEC.
   C. Glutamine supplementation decreases the occurrence of NEC.
   D. Gram-positive organisms predominate in severe cases of NEC.
   E. Hypoxia–ischemia is a primary contributor to NEC in preterm infants.