



REVIEW

Recent advances in understanding necrotizing enterocolitis [version 1; referees: 2 approved]

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v1 **First published:** 25 Jan 2019, 8(F1000 Faculty Rev):107 (<https://doi.org/10.12688/f1000research.17228.1>)
Latest published: 25 Jan 2019, 8(F1000 Faculty Rev):107 (<https://doi.org/10.12688/f1000research.17228.1>)

Abstract


Necrotizing enterocolitis is a devastating intestinal disease affecting preterm infants. In spite of ongoing research and advancement in neonatal care, mortality remains high, especially in infants with advanced disease. The mechanism of disease development, the progression of intestinal injury, and management remain areas of ongoing research and controversy. In this review, we examine our current understanding of the disease, its epidemiology, the risk factors associated with the development of the disease, and its pathophysiology. We also describe current management and new emerging research highlighting potential future directions.

Keywords

necrotizing enterocolitis (NEC), pathophysiology, premature, neonates

Open Peer Review

Referee Status:  

	Invited Referees	
	1	2
version 1 published 25 Jan 2019		

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Author roles: **Alganabi M:** Conceptualization, Writing – Original Draft Preparation, Writing – Review & Editing; **Lee C:** Writing – Review & Editing; **Bindi E:** Writing – Review & Editing; **Li B:** Conceptualization, Writing – Original Draft Preparation, Writing – Review & Editing; **Pierro A:** Conceptualization, Writing – Review & Editing

Competing interests: No competing interests were disclosed.

Grant information: This work is supported by a Canadian Institutes of Health Research (CIHR) Foundation Grant 353857. AP is the Robert M. Filler Chair of Surgery at The Hospital for Sick Children (HSC).

The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

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How to cite this article: Alganabi M, Lee C, Bindi E *et al.* **Recent advances in understanding necrotizing enterocolitis [version 1; referees: 2 approved]** *F1000Research* 2019, **8**(F1000 Faculty Rev):107 (<https://doi.org/10.12688/f1000research.17228.1>)

First published: 25 Jan 2019, **8**(F1000 Faculty Rev):107 (<https://doi.org/10.12688/f1000research.17228.1>)

Introduction

Necrotizing enterocolitis (NEC) is an inflammatory intestinal disease that affects 5–7% of preterm neonates¹. It is characterized by variable intestinal injury from epithelial injury to transmural involvement and perforation. It is also marked by inflammation and often bacterial invasion². NEC is one of the leading causes of morbidity in preterm infants³. It affects nearly 10% of preterm infants with a birth weight of <1,500 grams⁴. The mortality rate for preterm infants who have extremely low birth weight (<1,000 grams) is 30–50% and for infant with a very low birth weight (VLBW) (<1,500 grams) is 10–30%, and there has not been a significant change in the past 20 years⁵.

To assess the severity of NEC, Bell’s classification was proposed in 1978. It categorizes the severity of NEC based on clinical and radiographic signs and remains the most widely used tool in early assessment. In recent years, however, this staging criterion has been modified as our understanding of the disease has improved, yet there continues to be controversy about the validity of this staging system at lower gestational ages (GAs)⁶. Severity of NEC plays a key role in both the management and the outcome of affected neonates. Neonates with proven or advanced NEC, categorized as Bell’s stage II and III, respectively, are at risk of developing peritonitis, sepsis, bowel perforation, and other severe systematic complications including capillary leak syndrome and multi-system organ failure⁷.

The pathophysiology of NEC is multifactorial and remains not fully understood. The risk factors for the development of the disease are multiple and some are controversial. This leads to difficulty in establishing novel strategies to prevent the development of NEC and its progression. Herein we discuss some of our latest understanding of the disease’s epidemiology, risk factors, pathophysiology, treatment strategies, and future directions.

Epidemiology

The incidence of proven NEC (Bell’s stage II and III) in preterm babies depends on both the degree of prematurity and the geographic location of the patient. A recent systematic review on the incidence of NEC in high-income countries found variation in the incidence of the disease based on GA, birth weight, and country^{8,9}. Overall, NEC incidence was highest among the most preterm infants. In infants born at a GA of <28 weeks, the lowest reported incidence of NEC was in Japan (2%) and the highest in Australia, Canada, and Italy (7–9%)⁸. In neonates with a GA of between 28 and 31 weeks, reported NEC incidence was also lowest in Japan (0.2%), while other developed nations had incidence rates ranging from 2–3%. Similarly, for VLBW infants, NEC incidence ranged from 2% in Japan to 6–7% in the USA and 9% in Poland^{8–10}. These findings collectively indicate that the degree of prematurity and low birth weight are important factors in developing NEC. The varied incidence rates between countries suggest various factors influencing the development of NEC including environment, diet, and genetic predisposition.

Risk factors

The only consistently described risk factors for NEC are formula feeding, intestinal dysbiosis, low birth weight, and prematurity¹¹.

Low birth weight and prematurity are the most commonly reported risk factors for NEC, with the lowest birth weights and GAs having the highest incidence of NEC¹². Maternal factors such as chorioamnionitis, cocaine abuse, in-utero growth restriction, increased body mass index, intrahepatic cholestasis during pregnancy, lack of prenatal steroids, mode of delivery, placental abruption, preeclampsia, and smoking have inconsistently been implicated in the development of NEC^{13–17}. In addition, many other risk factors for the development of NEC have been reported including administration of acid-suppressing medications, acute hypoxia, antibiotic exposure, blood transfusions, cardiac anomalies, neonatal anemia, and poor intestinal perfusion^{18–21}. Finally, prolonged use of indomethacin to promote the closure of patent ductus arteriosus (PDA) has been shown to be associated with the development of NEC^{22,23}. However, the incidence of NEC was not lower in infants who underwent primary surgical closure of PDA compared to infants treated with indomethacin²⁴ (Table 1).

Pathophysiology

NEC affects multiple organs, and its pathophysiology appears to be multifactorial. The classical understanding of NEC pathophysiology suggests that intra-luminal bacteria disrupt and invade the intestinal epithelium at the tips of intestinal villi²⁵. Endotoxin

Table 1. Risk factors for the development of necrotizing enterocolitis.

Maternal factors
Chorioamnionitis
Cocaine abuse
In-utero growth restriction
Increased body mass index
Intrahepatic cholestasis during pregnancy
Lack of prenatal steroids
Mode of delivery
Placental abruption
Preeclampsia
Smoking
Main risk factors
Low birth weight
Prematurity
Formula feeding
Intestinal dysbiosis
Other risk factors
Acid-suppressing medications
Acute hypoxia
Antibiotic exposure
Blood transfusions
Cardiac anomalies
Neonatal anemia
Poor intestinal perfusion
Prolonged use of indomethacin for patent ductus arteriosus closure

from these bacteria binds to Toll-like receptor 4 (TLR4) found on the intestinal epithelial cells, activating pathogen-associated molecular pattern (PAMP) receptors, which facilitate the breakdown of the gut barrier and allow bacteria to translocate²⁶. This process subsequently leads to an intense inflammatory response in the lamina propria mediated by tumor necrosis factor-alpha (TNF- α), interleukin-1 β (IL-1 β), and other inflammatory cytokines²⁷. Vasoactive substances are also released in the intestine, and those associated with NEC include platelet-activating factor (PAF), endothelin-1 (ET-1), and nitric oxide (NO)²⁸. Intestinal inflammation also activates complement and coagulation systems. In these systems, leukocytes and platelets adhere to the endothelium, preventing blood flow in the microvascular structure of the small intestine and leading to tissue injury. Additional damage to the endothelium from adherent neutrophils and platelets also impairs NO generation needed for vasorelaxation^{29,30}. Through the efforts of various research laboratories, including ours, potential mechanisms have been explored to investigate how the disease may develop and how it may be treated (Table 2).

Nitric oxide

The pathogenesis of NEC is likely initiated by postnatal insults on the immature intestine in the presence of some of the previously mentioned risk factors. These factors lead to the initial epithelial injury, which causes an intestinal inflammatory response and release of inflammatory mediators³¹. Ford *et al.*³² first highlighted the role of NO and that of inducible NO synthase (iNOS)-derived NO in NEC development. NO plays a critical role in NEC development, and the details of its role have been extensively explored. Once intestinal barrier failure occurs, the lamina propria is exposed to increased levels of endotoxins and other bacterial products owing to bacterial translocation³³. The net result is activation of the neonatal immune system, which triggers an inflammatory cascade that leads to a severe pro-inflammatory response characterized by the release of NO, cytokines, and prostanooids. NO interacts with superoxide and leads to the production of peroxynitrite, a potent oxidant³⁴. This subsequently leads to enterocyte apoptosis or necrosis as well as impairment of both enterocyte proliferation and epithelial repair through enterocyte migration. The imbalance between tissue injury and repair further catalyzes the inflammatory cascade involved in NEC development. The ultimate consequence of these insults is further epithelial injury with risk for bacterial translocation resulting in sepsis, a vicious cycle which can lead to severe inflammation, bowel necrosis, perforation, and death.

Table 2. Factors involved in necrotizing enterocolitis pathophysiology.

Nitric oxide
Toll-like receptor 4
Microvascular blood flow (intestinal ischemia)
Dysbiosis
Reduced activity of intestinal stem cells

Toll-like receptor 4

The premature infant intestine is characterized by elevated expression of TLR4 on the intestinal epithelium^{35,36}. TLRs play an essential role in the activation of innate immunity by recognizing specific patterns of microbial components³⁷. TLR4 expression was increased in mice and humans with NEC^{38,39}, and mutations in the TLR4 signaling pathways have been described in human NEC^{40–42}. Additionally, TLR4 knockout mice were protected from NEC induction⁴³. TLR4 is activated by lipopolysaccharides on Gram-negative bacteria³⁶. Activation of TLR4 by intestinal lumen microbes results in barrier injury and impaired intestinal repair³⁹, which consequently allows the translocation of the luminal bacteria, vasoconstriction, intestinal ischemia, and NEC⁴⁴. TLR4 can also be inhibited by probiotic bacteria that activate TLR9⁴⁵ and can prevent goblet cell differentiation⁴³, which are needed to maintain the physical mucous intestinal barrier to pathogenic bacteria.

Microvascular blood flow

NEC is a disease often characterized by areas of intestinal ischemia with insufficient blood supply. The development of NEC has been associated with generalized neonatal hypoxia and exchange transfusions⁴⁶. Derangement of the intestinal microcirculation in NEC leads to areas of poor blood flow which may help facilitate the inflammatory cascade, resulting in intestinal injury⁴⁷. Feeding and postprandial hypoxia have been shown to synergistically induce intestinal hypoxia in experimental NEC, highlighting the important balance between oxygen supply and demand⁴⁸. The role of circulation in the pathogenesis of NEC continues to be a topic of research interest.

Microbiota

Dysbiosis is a disruption of gut microbiota development and its homeostasis, which has been associated with the development of NEC⁴⁹. This pathological process involves a lack of beneficial commensal microbes combined with a low diversity of bacteria, which allows the overgrowth of pathogenic bacteria that induce an inflammatory response⁵⁰. A meta-analysis of intestinal dysbiosis in preterm infants preceding NEC found an increased relative abundance of Proteobacteria and a decreased relative abundance of Firmicutes and Bacteroides⁵¹. Dysbiosis has been associated with the use of antibiotics and/or antacids in the NICU, formula feeding, and inflammatory response dysregulation⁵². NEC and matched control fecal samples tested for bacterial diversity and clustering showed that NEC patients tend to have less-diverse microbiomes and different distributions of the intestinal bacteria^{53–56}. Proteobacteria may trigger a strong inflammatory response and colonization by anaerobic bacteria, which have been associated with NEC⁵⁷. However, since Proteobacteria are also common constituents in the intestinal microbiome of preterm neonates who do not develop NEC, the role of microbiota remains unclear and represents only part of the complex pathogenesis⁵⁸. While prophylactic probiotics have been shown to reduce the incidence of NEC when baseline incidence levels are high⁵⁹, not all probiotic preparations are equally efficacious⁶⁰. Comparing the efficacy of different strains, mixtures of multiple strains and long-term safety remain areas of current research^{61,62}.

Intestinal stem cells and epithelial regeneration

The small intestinal epithelium renews every three to six days, a rate driven by vigorous proliferation within the intestinal crypts towards the villus tip⁶³. Within the crypts there is a distinct stem cell zone containing intestinal stem cells (ISCs). ISCs are responsible for producing progenitor cells that differentiate into various types of epithelial cells including enterocytes, goblet cells, entero-endocrine cells, and Paneth cells⁶⁴. ISCs are activated and replicate in response to intestinal injury. To measure the number of ISCs, leucine-rich repeat-containing G-protein-coupled receptor 5 (Lgr5), a well-established wingless integrated (Wnt)-associated stem cell marker⁶⁵, has been used. NEC has been characterized by a decrease in Lgr5-positive ISCs^{63,66}, and restoration of ISC activity appears to have a beneficial effect⁶⁶. The mechanism by which ISC viability is disrupted in NEC remains poorly understood and is currently being investigated.

Treatment strategies

Preventative treatments

Breast feeding has been shown to reduce the incidence of NEC relative to formula feeding^{59,67}. In examining which components of breast milk help in reducing NEC incidence, breast milk-derived exosomes⁶⁸ and human milk oligosaccharides (HMOs)⁶⁹ were investigated using an experimental model of NEC. Breast milk-derived exosomes have been shown to promote intestinal epithelial cell viability and stimulate intestinal stem cell activity⁶⁸. HMOs increased the number of goblet cells and mucin expression, stabilizing the intestinal barrier⁶⁹. In infants with a birth weight of <1,500 grams, the use of prophylactic probiotics reduced the incidence of NEC (RR 0.34, 95% CI of 0.23–0.50)⁷⁰, the risk of NEC-associated mortality (RR 0.56, 95% CI of 0.34–0.93)⁷¹, and the total length of hospital stay⁷². Notably, however, probiotic administration did not significantly reduce the risk of developing NEC in infants with a birth weight of <1,000 grams⁷³ or the risk of developing NEC requiring surgery in infants of any size (RR 0.56, 95% CI of 0.56–1.25)⁷⁰. Despite remaining uncertainties, the American Pediatric Surgical Association (APSA) Outcomes and Clinical Trials Committee Cochrane Review supports the prophylactic use of probiotics in preterm infants with a birth weight of <2,500 grams to reduce the risk of NEC in addition to the use of human breast milk rather than formula whenever possible⁷⁰.

Medical and surgical treatments

Suspicion of NEC is frequently based on clinical presentation, which can include feeding intolerance, abdominal distention, bloody stools, emesis, and gastric retention. These signs lead to further workup including blood work to detect potential thrombocytopenia or metabolic acidosis and imaging studies to identify dilated loops of bowel, intestinal perforation, pneumatosis intestinalis, and portal venous gas. Depending on imaging

findings and clinical presentation, surgical intervention is considered. Abdominal X-ray and ultrasound have been shown to be useful in helping to monitor the progression of the disease and detecting the presence of NEC⁷⁴. In general, for Bell stage I (suspected NEC), supportive medical management alone is provided. For Bell stage II (proven NEC), medical management is usually tried first. This includes antibiotic treatment, nasogastric decompression, and total parenteral nutrition. If the patient fails to respond to medical treatment, surgical management is considered⁷⁵. Patients with Bell stage III (advanced NEC) can be treated medically and may require inotropic support. However, neonates who develop intestinal perforation, have suspected bowel necrosis, or fail to respond to medical treatment require surgical treatment. Among VLBW infants, 27–52% require surgical intervention⁷⁶.

Future directions

Current research using both animal models and human tissue has yielded novel potential therapeutic avenues (Table 3). For example, the prospects of using stem cell therapy⁷⁷ and breast-milk derived exosomes⁶⁸ appear to be promising. Amniotic fluid stem cells given by intraperitoneal injection migrated to the intestinal villi and colonized almost exclusively the damaged intestine of NEC rat pups, where they promoted auto-regeneration of the intestinal epithelium⁷⁸. Stem cell-derived exosomes have also reduced the incidence and severity of experimental NEC⁷⁹. In addition, milk-derived exosomes have recently been shown to reduce intestinal epithelial injury⁶⁸. Safety and efficacy trials still need to take place before some of these potential treatments may become clinically available. Considering the lack of improvement and the unchanged mortality associated with NEC, it is promising to see that several avenues are being explored in both disease prevention and treatment.

Table 3. Future potential therapy for necrotizing enterocolitis.

Breast milk component (human milk oligosaccharides or exosomes) administration
Prophylactic probiotics
Stem cell administration

Grant information

This work is supported by a Canadian Institutes of Health Research (CIHR) Foundation Grant 353857. AP is the Robert M. Filler Chair of Surgery at The Hospital for Sick Children (HSC).

The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

References

1. Neu J, Walker WA: **Necrotizing enterocolitis**. *N Engl J Med*. 2011; **364**(3): 255–64. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
2. Thompson AM, Bizzarro MJ: **Necrotizing enterocolitis in newborns:**

pathogenesis, prevention and management. *Drugs*. 2008; **68**(9): 1227–38. [PubMed Abstract](#) | [Publisher Full Text](#)

3. Cotten CM, Oh W, McDonald S, et al.: **Prolonged hospital stay for extremely**



preterm infants: risk factors, center differences, and the impact of mortality on selecting a best-performing center. *J Perinatol.* 2005; 25(10): 650–5.
[PubMed Abstract](#) | [Publisher Full Text](#)

4. Luig M, Lui K, NSW & ACT NICUS Group: **Epidemiology of necrotizing enterocolitis—Part II: Risks and susceptibility of premature infants during the surfactant era: a regional study.** *J Paediatr Child Health.* 2005; 41(4): 174–9.
[PubMed Abstract](#) | [Publisher Full Text](#)
5. Fitzgibbons SC, Ching Y, Yu D, *et al.*: **Mortality of necrotizing enterocolitis expressed by birth weight categories.** *J Pediatr Surg.* 2009; 44(6): 1072–5; discussion 1075–6.
[PubMed Abstract](#) | [Publisher Full Text](#)
6. Gordon PV, Swanson JR, Attridge JT, *et al.*: **Emerging trends in acquired neonatal intestinal disease: is it time to abandon Bell's criteria?** *J Perinatol.* 2007; 27(11): 661–71.
[PubMed Abstract](#) | [Publisher Full Text](#)
7. Sonntag J, Wagner MH, Waldschmidt J, *et al.*: **Multisystem organ failure and capillary leak syndrome in severe necrotizing enterocolitis of very low birth weight infants.** *J Pediatr Surg.* 1998; 33(3): 481–4.
[PubMed Abstract](#) | [Publisher Full Text](#)
8. **F** Battersby C, Santhalingam T, Costeloe K, *et al.*: **Incidence of neonatal necrotizing enterocolitis in high-income countries: a systematic review.** *Arch Dis Child Fetal Neonatal Ed.* 2018; 103(2): F182–F189.
[PubMed Abstract](#) | [Publisher Full Text](#) | [F1000 Recommendation](#)
9. Kosloske AM: **Epidemiology of necrotizing enterocolitis.** *Acta Paediatr Suppl.* 1994; 396: 2–7.
[PubMed Abstract](#) | [Publisher Full Text](#)
10. Wójkowska-Mach J, Różańska A, Borszewska-Kornacka M, *et al.*: **Necrotizing enterocolitis in preterm infants: epidemiology and antibiotic consumption in the Polish neonatology network neonatal intensive care units in 2009.** *PLoS One.* 2014; 9(3): e92865.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
11. **F** Rose AT, Patel RM: **A critical analysis of risk factors for necrotizing enterocolitis.** *Semin Fetal Neonatal Med.* 2018; 23(6): 374–9.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | [F1000 Recommendation](#)
12. **F** Samuels N, van de Graaf RA, de Jonge RCJ, *et al.*: **Risk factors for necrotizing enterocolitis in neonates: a systematic review of prognostic studies.** *BMC Pediatr.* 2017; 17(1): 105.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | [F1000 Recommendation](#)
13. **F** Travers CP, Clark RH, Spitzer AR, *et al.*: **Exposure to any antenatal corticosteroids and outcomes in preterm infants by gestational age: prospective cohort study.** *BMJ.* 2017; 356: j1039.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | [F1000 Recommendation](#)
14. **F** Been JV, Lieveense S, Zimmermann LJ, *et al.*: **Chorioamnionitis as a risk factor for necrotizing enterocolitis: a systematic review and meta-analysis.** *J Pediatr.* 2013; 162(2): 236–42.e2.
[PubMed Abstract](#) | [Publisher Full Text](#) | [F1000 Recommendation](#)
15. **F** Lu Q, Cheng S, Zhou M, *et al.*: **Risk Factors for Necrotizing Enterocolitis in Neonates: A Retrospective Case-Control Study.** *Pediatr Neonatol.* 2017; 58(2): 165–70.
[PubMed Abstract](#) | [Publisher Full Text](#) | [F1000 Recommendation](#)
16. Czyrko C, Del Pin CA, O'Neill JA Jr, *et al.*: **Maternal cocaine abuse and necrotizing enterocolitis: outcome and survival.** *J Pediatr Surg.* 1991; 26(4): 414–8; discussion 419–21.
[PubMed Abstract](#) | [Publisher Full Text](#)
17. Downard CD, Grant SN, Maki AC, *et al.*: **Maternal cigarette smoking and the development of necrotizing enterocolitis.** *Pediatrics.* 2012; 130(1): 78–82.
[PubMed Abstract](#) | [Publisher Full Text](#)
18. **F** Denning NL, Prince JM: **Neonatal intestinal dysbiosis in necrotizing enterocolitis.** *Mol Med.* 2018; 24(1): 4.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | [F1000 Recommendation](#)
19. Mueller NT, Bakacs E, Combellick J, *et al.*: **The infant microbiome development: mom matters.** *Trends Mol Med.* 2015; 21(2): 109–17.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
20. **F** Safe M, Chan WH, Leach ST, *et al.*: **Widespread use of gastric acid inhibitors in infants: Are they needed? Are they safe?** *World J Gastrointest Pharmacol Ther.* 2016; 7(4): 531–539.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | [F1000 Recommendation](#)
21. Singh R, Visintainer PF, Frantz ID 3rd, *et al.*: **Association of necrotizing enterocolitis with anemia and packed red blood cell transfusions in preterm infants.** *J Perinatol.* 2011; 31(3): 176–82.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
22. Herrera C, Holberton J, Davis P: **Prolonged versus short course of indomethacin for the treatment of patent ductus arteriosus in preterm infants.** *Cochrane Database Syst Rev.* 2007; (2): CD003480.
[PubMed Abstract](#) | [Publisher Full Text](#)
23. Jain A, Shah PS: **Diagnosis, Evaluation, and Management of Patent Ductus Arteriosus in Preterm Neonates.** *JAMA Pediatr.* 2015; 169(9): 863–72.
[PubMed Abstract](#) | [Publisher Full Text](#)
24. Yee WH, Scotland J, Evidence-based Practice for Improving Quality (EPIQ) Evidence Review Group: **Does primary surgical closure of the patent ductus arteriosus in infants <1500 g or ≤32 weeks' gestation reduce the incidence of necrotizing enterocolitis?** *Paediatr Child Health.* 2012; 17(3): 125–8.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
25. Zhang C, Sherman MP, Prince LS, *et al.*: **Paneth cell ablation in the presence of *Klebsiella pneumoniae* induces necrotizing enterocolitis (NEC)-like injury in the small intestine of immature mice.** *Dis Model Mech.* 2012; 5(4): 522–32.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
26. Molteni M, Gemma S, Rossetti C: **The Role of Toll-Like Receptor 4 in Infectious and Noninfectious Inflammation.** *Mediators Inflamm.* 2016; 2016: 6978936.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
27. De Plaen IG: **Inflammatory signaling in necrotizing enterocolitis.** *Clin Perinatol.* 2013; 40(1): 109–24.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
28. Schnabl KL, Van Aerde JE, Thomson AB, *et al.*: **Necrotizing enterocolitis: a multifactorial disease with no cure.** *World J Gastroenterol.* 2008; 14(14): 2142–61.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
29. Claud EC, Keegan KP, Brulic JM, *et al.*: **Bacterial community structure and functional contributions to emergence of health or necrotizing enterocolitis in preterm infants.** *Microbiome.* 2013; 1(1): 20.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
30. Watkins DJ, Besner GE: **The role of the intestinal microcirculation in necrotizing enterocolitis.** *Semin Pediatr Surg.* 2013; 22(2): 83–7.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
31. Nowicki PT, Reber KM, Giannone PJ, *et al.*: **Intestinal O₂ consumption in necrotizing enterocolitis: role of nitric oxide.** *Pediatr Res.* 2006; 59(4 Pt 1): 500–5.
[PubMed Abstract](#) | [Publisher Full Text](#)
32. Ford H, Watkins S, Reblock K, *et al.*: **The role of inflammatory cytokines and nitric oxide in the pathogenesis of necrotizing enterocolitis.** *J Pediatr Surg.* 1997; 32(2): 275–82.
[PubMed Abstract](#) | [Publisher Full Text](#)
33. Ciftci I, Ozdemir M, Aktan M, *et al.*: **Bacterial translocation and intestinal injury in experimental necrotizing enterocolitis model.** *Bratisl Lek Listy.* 2012; 113(4): 206–10.
[PubMed Abstract](#) | [Publisher Full Text](#)
34. **F** Aceti A, Beghetti I, Martini S, *et al.*: **Oxidative Stress and Necrotizing Enterocolitis: Pathogenetic Mechanisms, Opportunities for Intervention, and Role of Human Milk.** *Oxid Med Cell Longev.* 2018; 2018: 7397659.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | [F1000 Recommendation](#)
35. **F** Kuzmich NN, Sivak KV, Chubarev VN, *et al.*: **TLR4 Signaling Pathway Modulators as Potential Therapeutics in Inflammation and Sepsis.** *Vaccines (Basel).* 2017; 5(4): pii: E34.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | [F1000 Recommendation](#)
36. **F** Hackam DJ, Sodhi CP: **Toll-Like Receptor-Mediated Intestinal Inflammatory Imbalance in the Pathogenesis of Necrotizing Enterocolitis.** *Cell Mol Gastroenterol Hepatol.* 2018; 6(2): 229–238.e1.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | [F1000 Recommendation](#)
37. Takeda K, Akira S: **TLR signaling pathways.** *Semin Immunol.* 2004; 16(1): 3–9.
[PubMed Abstract](#) | [Publisher Full Text](#)
38. **F** Egan CE, Sodhi CP, Good M, *et al.*: **Toll-like receptor 4-mediated lymphocyte influx induces neonatal necrotizing enterocolitis.** *J Clin Invest.* 2016; 126(2): 495–508.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | [F1000 Recommendation](#)
39. Leaphart CL, Cavallo J, Gribar SC, *et al.*: **A critical role for TLR4 in the pathogenesis of necrotizing enterocolitis by modulating intestinal injury and repair.** *J Immunol.* 2007; 179(7): 4808–20.
[PubMed Abstract](#) | [Publisher Full Text](#)
40. Sampath V, Menden H, Helbling D, *et al.*: **SIGIRR genetic variants in premature infants with necrotizing enterocolitis.** *Pediatrics.* 2015; 135(6): e1530–4.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
41. **F** Fawley J, Cuna A, Menden HL, *et al.*: **Single-Immunoglobulin Interleukin-1-Related Receptor regulates vulnerability to TLR4-mediated necrotizing enterocolitis in a mouse model.** *Pediatr Res.* 2017; 83(1–1): 164–74.
[PubMed Abstract](#) | [Publisher Full Text](#) | [F1000 Recommendation](#)
42. Ganguli K, Meng D, Rautava S, *et al.*: **Probiotics prevent necrotizing enterocolitis by modulating enterocyte genes that regulate innate immune-mediated inflammation.** *Am J Physiol Gastrointest Liver Physiol.* 2013; 304(2): G132–G141.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
43. Sodhi CP, Neal MD, Siggers R, *et al.*: **Intestinal epithelial Toll-like receptor 4 regulates goblet cell development and is required for necrotizing enterocolitis in mice.** *Gastroenterology.* 2012; 143(3): 708–718.e5.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
44. **F** Lu P, Sodhi CP, Hackam DJ: **Toll-like receptor regulation of intestinal development and inflammation in the pathogenesis of necrotizing enterocolitis.** *Pathophysiology.* 2014; 21(1): 81–93.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | [F1000 Recommendation](#)
45. de Kivit S, Tobin MC, Forsyth CB, *et al.*: **Regulation of Intestinal Immune Responses through TLR Activation: Implications for Pro- and Prebiotics.** *Front Immunol.* 2014; 5: 60.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

46. Beeby PJ, Jeffery H: **Risk factors for necrotising enterocolitis: the influence of gestational age.** *Arch Dis Child.* 1992; 67(4 Spec No): 432–5.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

47. Young CM, Kingma SD, Neu J: **Ischemia-reperfusion and neonatal intestinal injury.** *J Pediatr.* 2011; 158(2 Suppl): e25–8.
[PubMed Abstract](#) | [Publisher Full Text](#)

48. Chen Y, Koike Y, Miyake H, *et al.*: **Formula feeding and systemic hypoxia synergistically induce intestinal hypoxia in experimental necrotizing enterocolitis.** *Pediatr Surg Int.* 2016; 32(12): 1115–9.
[PubMed Abstract](#) | [Publisher Full Text](#)

49. Cho I, Blaser MJ: **The human microbiome: at the interface of health and disease.** *Nat Rev Genet.* 2012; 13(4): 260–70.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

50. **F** Patel RM, Denning PW: **Intestinal microbiota and its relationship with necrotizing enterocolitis.** *Pediatr Res.* 2015; 78(3): 232–8.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | **F1000 Recommendation**

51. **F** Pammi M, Cope J, Tarr PI, *et al.*: **Intestinal dysbiosis in preterm infants preceding necrotizing enterocolitis: a systematic review and meta-analysis.** *Microbiome.* 2017; 5(1): 31.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | **F1000 Recommendation**

52. **F** Cassir N, Simeoni U, La Scola B: **Gut microbiota and the pathogenesis of necrotizing enterocolitis in preterm neonates.** *Future Microbiol.* 2016; 11(2): 273–92.
[PubMed Abstract](#) | [Publisher Full Text](#) | **F1000 Recommendation**

53. Wang Y, Hoenig JD, Malin KJ, *et al.*: **16S rRNA gene-based analysis of fecal microbiota from preterm infants with and without necrotizing enterocolitis.** *ISME J.* 2009; 3(8): 944–54.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

54. Mai V, Young CM, Ukhanova M, *et al.*: **Fecal microbiota in premature infants prior to necrotizing enterocolitis.** *PLoS One.* 2011; 6(6): e20647.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

55. Morrow AL, Lagomarcino AJ, Schibler KR, *et al.*: **Early microbial and metabolomic signatures predict later onset of necrotizing enterocolitis in preterm infants.** *Microbiome.* 2013; 1(1): 13.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

56. Torrazza RM, Ukhanova M, Wang X, *et al.*: **Intestinal microbial ecology and environmental factors affecting necrotizing enterocolitis.** *PLoS One.* 2013; 8(12): e83304.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

57. Torrazza RM, Neu J: **The altered gut microbiome and necrotizing enterocolitis.** *Clin Perinatol.* 2013; 40(1): 93–108.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

58. **F** La Rosa PS, Warner BB, Zhou Y, *et al.*: **Patterned progression of bacterial populations in the premature infant gut.** *Proc Natl Acad Sci U S A.* 2014; 111(34): 12522–7.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | **F1000 Recommendation**

59. Hackam DJ, Good M, Sodhi CP: **Mechanisms of gut barrier failure in the pathogenesis of necrotizing enterocolitis: Toll-like receptors throw the switch.** *Semin Pediatr Surg.* 2013; 22(2): 76–82.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

60. **F** Patel RM, Underwood MA: **Probiotics and necrotizing enterocolitis.** *Semin Pediatr Surg.* 2018; 27(1): 39–46.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | **F1000 Recommendation**

61. **F** Chang HY, Chen JH, Chang JH, *et al.*: **Multiple strains probiotics appear to be the most effective probiotics in the prevention of necrotizing enterocolitis and mortality: An updated meta-analysis.** *PLoS One.* 2017; 12(2): e0171579.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | **F1000 Recommendation**

62. **F** Dermysli E, Wang Y, Yan C, *et al.*: **The “Golden Age” of Probiotics: A Systematic Review and Meta-Analysis of Randomized and Observational Studies in Preterm Infants.** *Neonatology.* 2017; 112(1): 9–23.
[PubMed Abstract](#) | [Publisher Full Text](#) | **F1000 Recommendation**

63. Gassler N: **Paneth cells in intestinal physiology and pathophysiology.** *World J Gastrointest Pathophysiol.* 2017; 8(4): 150–60.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

64. Umar S: **Intestinal stem cells.** *Curr Gastroenterol Rep.* 2010; 12(5): 340–8.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

65. **F** Khan Z, Orr A, Michalopoulos GK, *et al.*: **Immunohistochemical Analysis of the Stem Cell Marker LGR5 in Pediatric Liver Disease.** *Pediatr Dev Pathol.* 2017; 20(1): 16–27.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | **F1000 Recommendation**

66. Niño DF, Sodhi CP, Egan CE, *et al.*: **Retinoic Acid Improves Incidence and Severity of Necrotizing Enterocolitis by Lymphocyte Balance Restitution and Repopulation of LGR5+ Intestinal Stem Cells.** *Shock.* 2017; 47(1): 22–32.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

67. Herrmann K, Carroll K: **An exclusively human milk diet reduces necrotizing enterocolitis.** *Breastfeed Med.* 2014; 9(4): 184–90.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

68. Hock A, Miyake H, Li B, *et al.*: **Breast milk-derived exosomes promote intestinal epithelial cell growth.** *J Pediatr Surg.* 2017; 52(5): 755–9.
[PubMed Abstract](#) | [Publisher Full Text](#)

69. Wu RY, Li B, Koike Y, *et al.*: **Human Milk Oligosaccharides Increase Mucin Expression in Experimental Necrotizing Enterocolitis.** *Mol Nutr Food Res.* 2018; e1800658.
[PubMed Abstract](#) | [Publisher Full Text](#)

70. Downard CD, Renaud E, St Peter SD, *et al.*: **Treatment of necrotizing enterocolitis: an American Pediatric Surgical Association Outcomes and Clinical Trials Committee systematic review.** *J Pediatr Surg.* 2012; 47(11): 2111–22.
[PubMed Abstract](#) | [Publisher Full Text](#)

71. Rees CM, Hall NJ, Fleming P, *et al.*: **Probiotics for the prevention of surgical necrotizing enterocolitis: systematic review and meta-analysis.** *BMJ Paediatr Open.* 2017; 1(1): e000066.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

72. **F** Aceti A, Gori D, Barone G, *et al.*: **Probiotics and Time to Achieve Full Enteral Feeding in Human Milk-Fed and Formula-Fed Preterm Infants: Systematic Review and Meta-Analysis.** *Nutrients.* 2016; 8(8): pii: E471.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | **F1000 Recommendation**

73. **F** Sawh SC, Deshpande S, Jansen S, *et al.*: **Prevention of necrotizing enterocolitis with probiotics: a systematic review and meta-analysis.** *PeerJ.* 2016; 4: e2429.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | **F1000 Recommendation**

74. Janssen Lok M, Miyake H, Hock A, *et al.*: **Value of abdominal ultrasound in management of necrotizing enterocolitis: a systematic review and meta-analysis.** *Pediatr Surg Int.* 2018; 34(6): 589–612.
[PubMed Abstract](#) | [Publisher Full Text](#)

75. Rees CM, Hall NJ, Eaton S, *et al.*: **Surgical strategies for necrotising enterocolitis: a survey of practice in the United Kingdom.** *Arch Dis Child Fetal Neonatal Ed.* 2005; 90(2): F152–5.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

76. **F** Robinson JR, Rellinger EJ, Hatch LD, *et al.*: **Surgical necrotizing enterocolitis.** *Semin Perinatol.* 2017; 41(1): 70–9.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | **F1000 Recommendation**

77. Drucker NA, McCulloh CJ, Li B, *et al.*: **Stem cell therapy in necrotizing enterocolitis: Current state and future directions.** *Semin Pediatr Surg.* 2018; 27(1): 57–64.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

78. Zani A, Cananzi M, Fascetti-Leon F, *et al.*: **Amniotic fluid stem cells improve survival and enhance repair of damaged intestine in necrotising enterocolitis via a COX-2 dependent mechanism.** *Gut.* 2013; 63(2): 300–9.
[PubMed Abstract](#) | [Publisher Full Text](#)

79. **F** McCulloh CJ, Olson JK, Wang Y, *et al.*: **Treatment of experimental necrotizing enterocolitis with stem cell-derived exosomes.** *J Pediatr Surg.* 2018; 53(6): 1215–20.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#) | **F1000 Recommendation**

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Version 1

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Competing Interests: No competing interests were disclosed.
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Competing Interests: No competing interests were disclosed.

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