SIGNEC U.K. and the Second International Conference on Necrotising Enterocolitis

The SIGNEC U.K. second international conference on necrotising enterocolitis, held at BMA House, London, on 3-4 September 2014.

Introduction

Established by Professor Minesh Khashu to help advance a field of neonatal medicine that has seen little progress in the last two decades, the special interest group for necrotising enterocolitis (SIGNEC U.K.) provides a platform for involvement of a wide range of healthcare professionals to facilitate knowledge sharing, networking and collaboration for optimising research and improvements in practice. Initially comprising healthcare professionals from the UK, the group now has international involvement and includes neonatologists, paediatricians, surgeons, transfusion medicine specialists, epidemiologists, basic science researchers, nurses, dietitians, trainees and other healthcare professionals with an interest in necrotising enterocolitis (NEC) and health improvement.

NEC continues to be the major cause of mortality and morbidity for preterm newborns. Despite some recent advances in the understanding of NEC pathogenesis, the condition is still poorly understood.

The second SIGNEC U.K. conference was held in London in September 2014. Akin to the first conference in 2013,[1] distinguished clinicians and researchers from the USA, Canada, the UK and Europe presented their cutting edge work. NEC was discussed in great breadth and depth including breakthroughs in basic science, for example, the development of Toll-like receptor 4 (TLR4) inhibitors, as well as intestinal microbial diversity, genetic, maturational and environmental predispositions. Results from the recent PiPS trial and the UK Neonatal Collaborative NEC study were deliberated. Discussions regarding ethical conundrums and NEC patient information leaflets provided useful insights.

Probiotics: is it time for routine supplementation?

Professor Kate Costeloe opened the presentations by reviewing the various clinical trials that have evaluated the therapeutic role of probiotics. Despite meta-analyses suggesting that probiotics protect the preterm baby from NEC and death, their use is limited – probably because clinicians doubt the validity of extending the conclusions to their patients.[2,3] The largest contributor to the meta-analyses is the ProPrems trial, which showed no evidence of benefit on mortality and no decrease of NEC in infants born at <28 weeks' gestation or <1,000g birth weight.[4] The unpublished UK PiPS trial found no evidence of efficacy[5] and found high rates of cross colonisation of the placebo group, emphasising the importance of infection control measures. Professor Costeloe concluded that current evidence does not answer the important questions: which babies might benefit and what product to use? Therefore, until

Minesh Khashu MBBS, MD, FRCPCH, Fellowship in Neonatal Intensive Care

Consultant in Neonatal Medicine, Poole Hospital NHS Foundation Trust and Visiting Professor, Centre for Midwifery, Maternal and Perinatal Health, Bournemouth University. mineshkhashu@gmail.com

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further evidence becomes available, the safest and most effective way to provide both pre- and probiotics may be through early use of colostrum.

Immune regulation of the intestinal stem cells in the pathogenesis of NEC

In the first of his two presentations, Professor David Hackam highlighted how the underlying molecular and cellular causes of NEC are not fully understood. Evidence suggests that there is an important role for bacterial-mucosal interactions in disease pathogenesis. Professor Hackam's team has recently discovered that the receptor for Gram negative bacteria, namely TLR4, plays a critical role in NEC pathogenesis. Specifically, TLR4 activation within the intestinal mucosa leads to the death of intestinal epithelial cells, impaired intestinal stem cell proliferation and reduced mucosal perfusion. Moreover, TLR4 expression is elevated in the intestinal tract of the premature infant, which is a consequence of the role of TLR4 in regulating normal gut differentiation during embryonic development. The premature intestine, therefore, is characterised by excessive TLR4 expression, leading to the development of NEC when colonising microbes activate TLR4.

Professor Hackam's team has extended these findings through the development of novel small molecule inhibitors of TLR4, which effectively treat NEC in small and large animal models. Taken together, these findings provide insights into the pathogenesis of this disease and offer novel and exciting therapeutic approaches.

The role of the mucosal microbiome in NEC

Dr Jörn-Hendrik Weitkamp summarised the findings of his research on the mucosal microbiome in NEC. His group sought to determine how faecal microbial populations in preterm infants correlate with those detected in the intestinal tissue and whether NEC infants harbour distinct mucosal microbiota, possibly contributing to the disease.

In patients with NEC, faecal and tissue samples demonstrate dramatically reduced bacterial diversity and stool samples do not

Day 1: Basic science and laboratory research	
Professor Kate Costeloe *Professor of Paediatrics, Queen Mary University of London, UK*	Probiotics, a clinical perspective: is it time for routine supplementation?
Professor David J. Hackam *Chief of Pediatric Surgery, John Hopkins Children's Center, USA*	Immune regulation of the intestinal stem cells in the pathogenesis of NEC
Dr Jörn-Hendrik Weitkamp *Assistant Professor of Pediatrics, Vanderbilt University, USA*	The role of the mucosal microbiome in NEC
Dr Kathleen Sim *Clinical Research Fellow, Imperial College London, UK*	Dysbiosis with characteristic microbial signature anticipating necrotising enterocolitis in very premature infants
Dr Paolo De Coppi *Head of Surgery Unit, University College London, UK*	Role of amniotic fluid stem cells in NEC and tissue engineering of the intestine
Professor Boris W. Kramer *Professor of Experimental Perinatology, Maastricht UMC, Netherlands*	Endotoxin tolerance of the fetus – a double-edged sword for NEC
Day 2: NEC: Clinical research and improvements in practice	
Zoe Chivers *Bliss Head of Innovations and Campaigns, UK* Catherine Miles *Bliss Volunteer*	Those three letters – a parent's view of NEC
Professor Shoo K. Lee *Professor of Paediatrics, Obstetrics and Gynaecology and Public Health, Toronto, Canada*	Decreasing the incidence of NEC through breast milk: the Canadian experience
Professor Neil Marlow *Professor of Neonatal Medicine, University College London, UK*	The contribution of NEC to the burden of long-term impairment after preterm birth
Professor Susan G. Albersheim *Clinical Professor, University of British Columbia, Canada*	How short is too short in 2014?
Professor Minesh Khashu *Consultant in Neonatal Medicine, Poole Hospital, UK*	Development of parent information leaflets for NEC
Dr Karen D. Fairchild *Associate Professor of Pediatrics, University of Virginia, USA*	Heart rate and cardiorespiratory analysis for early detection of NEC
Professor David J. Hackam *Chief of Pediatric Surgery, John Hopkins Children's Center, USA*	Novel clinical approaches to immune regulation in NEC: small molecules, amniotic fluid and new formulae
Dr Cheryl Battersby *Clinical Research Fellow, Neonatal Data Analysis Unit, Imperial College London, UK*	The UK Neonatal Collaborative NEC Study

TABLE 1 Programme of speakers.

represent all phyla detected in tissue. Compared to gestational age and age-matched surgical controls, NEC faecal and mucosal samples show a loss of Actinobacteria and a statistically significant overrepresentation of Firmicutes, specifically *Staphylococcus*. Patients with the highest *Staphylococcus* abundance in resected tissue had earlier onset of NEC (mean 12 versus 30 days) and were delivered by caesarean section, supporting findings by others that risk factors for early and late NEC may be different.[6]

Dysbiosis anticipating NEC in very premature infants

Dr Kathleen Sim described her team's work into early diagnosis of NEC via faecal microbiota signatures.[7] Premature infants (<32 weeks'gestation) have been recruited on to a two-year cohort study at Imperial College NHS Trust. Over 10,000 daily faecal samples have been collected together with detailed daily clinical information seeking a microbial association with NEC in pre-diagnosis samples. Sequencing of rRNA gene regions was used to characterise the microbiota of faecal samples preceding diagnosis from neonates who developed NEC (Bell's stage I and II/III), and matched controls. Two faecal microbiota signatures were associated with increased risk of NEC: bloom of *Clostridium perfringens* and dominance of Enterobacteriaceae. A screening tool based on the detection of significant shifts in the faecal microbial spectrum, or pathognomonic signature, merits further exploration for early diagnosis of NEC and targeted treatment.

The role of amniotic fluid stem cells

Dr Paolo De Coppi described how stem cells taken from amniotic fluid were used to restore gut structure and function following intestinal damage in rodents. The findings pave the way for a new form of cell therapy to reverse serious damage from inflammation in the intestines of infants. Stem cells are known to have anti-inflammatory effects, but this is the first time it has been shown that amniotic fluid stem cells can repair damage in the intestines. This modality holds significant promise and needs to be explored further.

Endotoxin tolerance of the fetus – a double-edged sword for NEC

Professor Boris Kramer described how chorioamnionitis resulting from microbial invasion of the amniotic cavity is, perhaps, the most frequent cause of preterm birth. Although chorioamnionitis is associated with an increased risk of NEC, the mechanisms underlying this association remain largely unknown. Professor Kramer hypothesised that developmental alterations of the fetal immune system in the course of chorioamnionitis determine the risk of development of NEC. The consequences of chorioamnionitis on fetal gut development are adverse and experimental models of chorioamnionitis with ureaplasma show a severe impairment of gut structure associated with a complex dysregulation of the fetal immune system, resulting in a hypo-responsiveness of immune cells to endotoxin from *E. coli*. Taken together, the intrauterine conditioning of the fetal immune system may predispose an already injured gut to subsequent injury resulting in NEC.

Those three letters – a parent's view of NEC

In the first presentation of Day 2, Zoe Chivers, Head of Innovations and Campaigns at Bliss, introduced the charity and its work in supporting the families of babies with NEC by driving improvement in practice, aiding research and providing information to parents. Catherine Miles, a Bliss volunteer and parent of a child who suffered from NEC, spoke of her family's journey following the premature birth of their son: traumatic emergency surgery for NEC, reversal surgery for an ileostomy, a subsequent diagnosis of mild cerebral palsy and an uncertain future. The presentation raised questions around the need for more knowledge of long-term outcomes, better parent information from reliable sources and improved family-centred care, especially during the post-surgical period.

Catherine Miles, Bliss volunteer and parent.

Conference organiser Professor Minesh Khashu.

Decreasing the incidence of NEC through breast milk: the Canadian experience

Professor Shoo K. Lee reviewed the literature on the benefits of human milk for sick babies in the neonatal unit giving special attention to preterm babies who are at risk of NEC. He discussed how and why human milk might help to prevent the condition and examined the results of various studies as well as 'real life experience' in centres that have adopted this strategy. Professor Lee shared the positive results from a study of Canadian units where human donor milk was introduced routinely for preterm infants whose mothers were unable to supply breast milk for their babies. He demonstrated how effective leadership and engagement at a national level in Canada has yielded very encouraging results.

NEC and long-term impairment after preterm birth

In his talk, Professor Neil Marlow reminded the audience that NEC remains an important cause of neonatal morbidity and mortality – NEC requiring surgery occurred in 8% of the EPICure2 cohort and was responsible for 12% of the neonatal deaths. At follow-up, infants who have had NEC are at higher risk of cerebral palsy and developmental problems. In the ORACLE trial, children who had suspected/confirmed NEC had more mild functional impairments and also bowel problems in middle childhood. Surgical NEC is a risk factor for poor school performance, behaviour problems and growth impairment in the original EPICure cohort.

NEC is accompanied by a rise in cytokine levels and clinical evidence of sepsis, both of which are independently associated with poor long-term outcomes. In terms of motor and cognitive disability after very preterm birth, NEC is a significant marker of risk, alongside sepsis and hypoxia. The progression from inflammation to disability is probably mediated through neuronal and glial cell pathways resulting in brain white matter injury.

How short is too short?

Professor Susan Albersheim discussed the hospital course of a patient with severe NEC from an ethical perspective. She highlighted the importance of considering outcome data, both in the literature and for individual neonatal units, in terms of mortality and morbidity, including short- and long-term quality of life. This is particularly relevant given emerging diagnostic and treatment modalities. Standards of practice dictate certain decisions when outcomes are clear, but how are medical uncertainties considered in terms of survival and prediction of eventual intestinal autonomy? Furthermore, are home parenteral nutrition and single or double organ transplantations standard of practice? Professor Albersheim presented several frameworks for decision making to aid the clinician and promote informed parental consent.

Panel discussion

Continuing along the theme of 'ethical conundrums', a panel of experts considered various ethical issues including parental disagreement with medical recommendations. The panelists included Dr David Hackam, Professors Neil Marlow and Susan Albersheim, and Ms Joanne Ferguson, a parent who experienced difficult decision-making scenarios when one of her twin sons developed and subsequently died from NEC.

Development of parent information leaflets for NEC

Following lunch, the delegates worked in multiple facilitated groups to explore ideas and content for NEC patient information leaflets, intended for families. Draft documents were produced that will be refined through collaborative work with parent groups and Bliss; the parent information leaflets will be available by April 2015.

Heart rate and cardiorespiratory analysis for early detection of NEC

Dr Karen Fairchild discussed how abnormal heart rate characteristics of reduced beat-to-beat variability and repetitive, transient decelerations occur in sepsis and NEC, reflecting derangements of autonomic nervous system activation.

A specially designed monitor – the 'HeRO' monitor – calculates a heart rate characteristics index that indicates the risk that an infant will be diagnosed with sepsis in the next 24 hours. In a study of 3,003 very low birthweight infants, mortality was significantly reduced when clinicians could see the HeRO score.[8] In a subgroup of preterm infants with NEC, there was a significant rise in the HeRO score 16 hours before clinicians diagnosed NEC requiring surgery. More recent studies by Dr Fairchild's group indicate that abnormal respiratory patterns, in particular an increase in the amount of time spent in periodic breathing (regular, repetitive short apnoeas), also occur in some infants prior to clinical deterioration from NEC or sepsis. It is hoped that stronger evidence over the coming years will establish the impact of predictive cardiorespiratory monitoring on early diagnosis and NEC outcomes.

The UK Neonatal Collaborative NEC Study

The UK Neonatal Collaborative NEC Study is the first population study to report the incidence of NEC in England. All 163 neonatal units in England have contributed data to this two-year prospective population cohort study of infants born and admitted to neonatal care in England in 2012 and 2013. Although the final analyses are still underway, Dr Cheryl Battersby was able to share some of the initial findings. With the lack of a case definition for NEC, only infants severely affected by NEC (defined as receiving surgery or died as a result of NEC) were included. With this definition, there was more than a two-fold variation in the crude incidence rates of NEC across the 23 neonatal networks in England, ranging from 2-5% in infants born at <32 weeks' gestation. The final analyses will explore the possible reasons for the variation in crude NEC incidence across the country, including whether feeding exposures influence the susceptibility to developing severe NEC.

Professor Shoo K. Lee.

Summary

While researchers grapple with the various avenues for prevention, early diagnosis and treatment of NEC, parallel efforts are required to improve practice based on current evidence and to precisely delineate the intermediate and long-term impact of NEC. Above all, special emphasis on family-centred care and better information/communication for parents is required.

The second international conference on NEC was very well received with feedback from all delegates confirming it to be a useful platform for learning, networking and collaboration. Plans are already underway for the next annual SIGNEC U.K. conference in September 2015.

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For updates on the third SIGNEC U.K. conference, planned for September 2015, contact melanie.thomas-lee@danone.com