SIGNEC Fifth International Conference on Necrotising Enterocolitis

Some of the speakers and chairpersons at SIGNEC 2017.

The fifth SIGNEC (special interest group in necrotising enterocolitis) international conference was held on 16-17 October 2017 in London. A wide spectrum of topics related to NEC was discussed including, for the first time, a focus on genetic aspects. Many representatives of NEC advocacy and neonatal support attended the meeting including: Bliss Chief Executive Caroline Lee-Davey; Founder of the NEC Society USA Jennifer Canvasser; Susan Spencer from NEC UK, and CEO and Founder of the Instituto PGG in Brazil Simone Rosito. Another first for SIGNEC was a presentation of data from Japan, the country that is known to have the world's lowest incidence of NEC.

Effects of fetal exposure to inflammation on the development of the gastrointestinal tract and its associated microbiome

Dr Steven McElroy has had a longstanding interest in injury and repair mechanisms of the developing small intestine and how these relate to neonatal NEC. Development of maternal chorioamnionitis has been associated with later development of neonatal NEC in offspring, however the mechanisms behind this association are not fully understood. Using a standard mouse model of chorioamnionitis, Dr McElroy's laboratory is attempting to understand the effects of maternal inflammation on the development of the offspring's intestine. His data show that exposure to maternal inflammation during pregnancy induces injury to the placenta and induces significant changes in the overall growth of the offspring, as well as altering the normal development of intestinal innate immunity. These changes occur in pathways critical to the development of NEC and may help to explain why infants exposed to chorioamnionitis are at higher risk of developing NEC.

Novel signaling pathways involved in NEC

The overarching focus of Dr Misty Good's research programme is on the cellular and molecular mechanisms involved in the pathogenesis of NEC. Her laboratory seeks to further characterise the mucosal immune responses in NEC and cross-examine how these responses can be modified or prevented through dietary modifications or targeted intestinal epithelial therapies.

In Dr Good's pre-clinical studies, her laboratory utilises a neonatal mouse model of NEC to understand the signalling pathways and immune cell responses involved in NEC development. Dr Good reviewed several key research techniques performed in her laboratory including human and mouse intestinal epithelial and stem cell culture and the development of 'gut-on-a-chip' technology for a personalised medicine approach. Dr Good and her team have recently filed a patent on a novel therapy for NEC and she is working with the US Food and Drug Administration (FDA) on a future clinical trial.

Sepsis and NEC – more than a differential diagnosis

Preterm delivery is frequently associated with chorioamnionitis and, in contrast to a relatively asymptomatic mother, the fetus

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

Consultant in Neonatal Medicine, Poole Hospital NHS Foundation Trust and Professor of Perinatal Health, Bournemouth University. mineshkhashu@gmail.com

This supplement is based on a two-day conference that was supported by an educational grant from Nutricia Early Life Nutrition.

might suffer from a fetal inflammatory response including umbilical inflammation and increased serum levels of inflammatory cytokines. Antenatal exposure to inflammation places the extremely immature neonate at high risk for adverse gastrointestinal outcomes, such as NEC.

Patients who develop NEC have changes in the gut microbiome that precede the development of NEC and have a low percentage of regulatory T lymphocytes, which are crucial for the control of inflammation. Professor Boris Kramer and his team studied the composition of the gut microbiome in preterm babies and found temporary changes that precede the development of NEC and sepsis, respectively. With the help of an 'electronic nose', changes in the metabolic activity of the microbiome could be detected in babies two days before the development of NEC or sepsis – with the possibility to distinguish NEC and sepsis from each other. Subsequently, the researchers compared the isolated bacteria from blood cultures with the changes in the composition of the gut microbiome. The bacteria species that increase before the development of sepsis are the ones that can be later isolated in the bloodstream. Professor Kramer therefore concluded that the changes in the gut microbiome do not only play a role in the development of NEC but may also be the source of sepsis.

Biobanking for NEC: challenges and opportunities

In her second talk of the day, Dr Misty Good presented the challenges and opportunities of developing an international biorepository for NEC in collaboration with the NEC Society USA.

Recent advances in high-content molecular interrogation and bio-computation (eg genomics, transcriptomics, proteomics and metabolomics) can provide new insights from infants affected by NEC. However, individual centres are limited by the number of cases. Dr Good discussed how the NEC Society Biorepository has

Day 1: Basic science and laboratory research	
Dr Steven McElroy *Associate Professor, University of Iowa, USA*	Effects of fetal exposure to inflammation on the development of the gastrointestinal tract and its associated microbiome
Dr Misty Good *Assistant Professor of Pediatrics, Washington University School of Medicine, St Louis Children's Hospital, Missouri, USA*	1. Novel signaling pathways involved in NEC 2. Biobanking for NEC: challenges and opportunities
Professor Boris Kramer *Professor of Experimental Perinatology, Maastricht UMC, Netherlands*	NEC and sepsis – more than a clinical differential diagnosis
Professor Gail Besner *Nationwide Children's Hospital, Columbus, USA*	1. Update on stem cell and probiotic therapy for NEC 2. Development of a tissue engineered intestine
Dr Venkatesh Sampath *Associate Professor of Pediatrics, Children's Mercy Hospital, Kansas City, USA*	1. Genetic predisposition to NEC: myth or truth? 2. The *SIGIRR* gene in NEC
Day 2: Clinical research and improvements in practice	
Jennifer Canvasser *Founder and Executive Director of NEC Society, USA*	Turning loss into light: the NEC Society's commitment to advocacy, prevention and diverse collaboration
Caroline Lee-Davey *Chief Executive, Bliss, UK*	Bliss report on neonatal transport
Dr Andrew Leslie *Nurse Consultant, CenTre Neonatal Transport, UK*	Transfers of babies with NEC
Panel discussion *Chaired by Dr Eleri Adams, Oxford Newborn Care Unit, UK, with Andrew Leslie, Caroline Lee-Davey and Joanne Ferguson*	How can we improve the transfer of neonates with NEC and the experience of parents?
Professor Tomoaki Taguchi *Kyushu University Hospital, Fukuoka, Japan*	Overview of the nationwide surveys for neonates with NEC in Japan
Dr Nicholas Embleton *Consultant Neonatal Paediatrician, Newcastle Hospitals NHS Foundation Trust, UK*	MAGPIE – mechanisms affecting the gut of preterm infants in enteral feeding trials: a clinician's approach to the use of 'omic' technologies in understanding NEC
Dr Ravi Mangal Patel *Assistant Professor of Pediatrics, Emory University School of Medicine, Atlanta, USA*	New insights into the epidemiology of NEC: role of anaemia and cytomegalovirus
Dr Amit Gupta *Consultant Neonatologist, Oxford University Hospitals NHS Foundation Trust, UK*	Management and outcomes of surgical NEC: a UK-wide cohort study
Professor Neena Modi *Professor of Neonatal Medicine, Imperial College London, UK*	Next steps in NEC clinical trials
Michele Upton *Patient Safety Lead, Maternity and Newborn NHS Improvement, UK*	The national Maternal and Neonatal Health Safety Collaborative: supporting improvements in neonatal outcomes

TABLE 1 Programme of speakers.

advanced collaboration among institutions through the shared use of biological samples and the dedicated pursuit of molecular indicators of disease. She reviewed how streamlining the infrastructure and standard operating procedures across several centres can ensure consistently processed specimens, collection of appropriate clinical data and potential for new discoveries.

Update on stem cell therapy and probiotic administration

Professor Gail Besner updated the audience on two potential novel therapeutic strategies for the prevention of NEC. Laboratory studies recently demonstrated that four different types of stem cells (amniotic fluid-derived mesenchymal, amniotic fluid-derived neural, bone marrow-derived mesenchymal, and enteric neural stem cells) equivalently protect the intestines from NEC. Furthermore, exosomes (small extracellular nanoparticles) secreted by and purified from these four types of stem cells, also equivalently protect the intestines from NEC. These exosomes may therefore represent a non-cell-based therapy for NEC in the future.

Professor Besner provided follow-up on a delivery strategy that improves the ability of probiotics to protect the intestines from NEC. This delivery system involves briefly incubating the probiotic *Lactobacillus reuteri* with microspheres, which stimulate the probiotic to produce a protective biofilm. This preparation significantly reduces the incidence of NEC after just one single dose. Discussions with the FDA for permission to administer this exciting potential therapy to babies will begin in 2018.

Genetic susceptibility to NEC – myth or truth?

Dr Venkatesh Sampath listed several potential risk factors for NEC including use of formula milk, bowel ischaemia, abnormal gut microbial colonisation, and immature intestinal immune responses. However, infants who have these risk factors do not necessarily develop NEC and severe NEC can develop in infants who do not have these risk factors. This suggests that there might be inherent predisposition of certain premature infants to develop NEC when exposed to clinical and environmental risk factors.

Several researchers have attempted to identify genetic risk factors for NEC in premature infants. Some studies have identified that genetic variants in *NOD2*, a gene involved in intracellular anti-bacterial immunity, may increase vulnerability to NEC. Furthermore, very preliminary studies indicate that mutations in genes that inhibit exaggerated innate immune responses, such as *SIGIRR*, may contribute to an increased risk of NEC.

Turning loss into light: the NEC Society's commitment to advocacy, prevention and diverse collaboration

Jennifer Canvasser of the NEC Society USA described how the families of patients can help to inspire innovative solutions to this complex disease. Jennifer's son Micah died just one month before his first birthday from complications of NEC. A collaborative effort between clinicians, investigators and patient-family advocates, the NEC Society continues to create projects, events, and educational materials aimed at improved prevention and treatment options for fragile and premature infants at risk of NEC.

Neonatal transport

Neonatal transport services are a vital part of care for premature and sick babies. Caroline Lee-Davey shared Bliss' important report on neonatal transport in the UK that highlighted the wide variation

Professor Gail Besner.

Jennifer Canvasser.

in transport services. Many areas did not have the resources needed to meet national safety and quality standards.

Dr Andy Leslie from CenTre Neonatal Transport offered an overview of the organisation of neonatal transport in the UK and discussion of transfers for actual or suspected NEC. 'Suspected' NEC is an important distinction for transport services as for many infants the definitive diagnosis is not established until after transfer to a surgical centre.

In the first six months of 2017, CenTre undertook 902 neonatal transfers of which 346 were for uplift of care. Of these, just 27 transfers were of infants in whom NEC was suspected. Dr Leslie and Dr Arthi Mistry looked at a year of CenTre's possible-NEC transfers and traced their final diagnoses. Of 55 infants transferred in whom NEC was considered, 31 had a diagnosis of NEC.

Dr Leslie also discussed the UK readiness for responding to NEC transfers, pointing out that the trend in neonatal transport has been for ever-larger transport services and a concentration of transport resources.

There followed a valuable panel discussion on the difficulties of NEC transfers and the urgent need for further consideration of how best to support parents in this scenario.

Overview of the nationwide surveys for neonates with NEC in Japan

Professor Tomoaki Taguchi explained the data from two national surveys examining NEC incidence, risk factors, and prognosis in Japan – the country with the lowest incidence of NEC. The Neonatal Research Network Japan (NRNJ) survey considered more than 40,000 neonates of birthweight <1,500g. In NRNJ the incidence of NEC was lower than any published data from other networks. Multivariate analyses suggested that the risk factors for NEC are patent ductus arteriosus (PDA), lower gestational age, small head circumference, high frequency oscillatory ventilation, persistent pulmonary hypertension of the newborn, not using indomethacin, and cord blood transfusion.

The second survey, a multicentre study that retrospectively accumulated laparotomy-confirmed NEC cases, indicated that NEC onset correlates with respiratory distress syndrome, PDA, and male sex. NEC mortality correlates with preoperative high C-reactive protein and late surgery. Survival time analyses showed NEC survival was significantly poorer than matched controls.

The two datasets indicate PDA as a risk factor for NEC. Professor Taguchi suggested the Japanese PDA treatment strategy may correlate with the lower incidence of NEC observed. Another possibility is the feeding strategy, ie the slow speed of increasing milk feeds. Both surveys confirm the poor prognosis of NEC, which serves to emphasise the importance of prevention. The reported low incidence of NEC in Japan warrants further exploration in terms of causative and contributory factors.

Mechanisms affecting the gut of preterm infants in enteral feeding trials

In his presentation Dr Nicholas Embleton spoke about the MAGPIE study (mechanisms affecting the gut of preterm infants in enteral feeding trials, www.neonatalresearch.net/magpie.html). He hypothesised that trial interventions involve effects on gut microbial diversity, metabolites (eg short-chain fatty acids) and aspects of host immune function. While current data suggest NEC and/or late-onset sepsis (LOS) are due to a dysregulated immune system in the context of gut dysbiosis, mechanisms have not been systematically studied within large randomised controlled trials (RCTs).

The microbiome and metabolites will be analysed. The trial team will explore differences between disease cases and controls as well as the actions of trial interventions. The impacts of this research will be multiple: translation of knowledge of mechanisms promoting gut health may explain outcomes or suggest alternate strategies to improve health; new non-invasive diagnostic or monitoring techniques and preventative or treatment strategies for NEC or LOS may be identified; and useful data for risk stratification will be revealed. Recruitment is now complete and the MAGPIE trial has samples from 608 babies to analyse.

Development of a tissue engineered intestine

Massive bowel resection for NEC is the most common cause of short bowel syndrome (SBS), which occurs when there is not enough small bowel absorptive surface area to sustain the nutritional needs of a patient. Although bowel lengthening procedures and intestinal transplantation are options, these procedures have associated complications. In the future, tissue engineered intestine produced from the patient's own cells and therefore not subject to immune rejection may represent an alternative therapy for patients with SBS. Professor Gail Besner's second presentation highlighted the techniques currently being used in the laboratory for scaffold preparation and cell seeding for the production of tissue engineered intestine.

New insights into the epidemiology of NEC: role of anaemia and cytomegalovirus

Dr Ravi Mangal Patel presented an update on the epidemiology of NEC including recent studies on the potential role of red cell transfusion, severe anaemia and cytomegalovirus (CMV). There is currently inconsistent evidence to support a causal association between red blood cell transfusion and NEC. Increasing severity of anaemia may be a potential risk factor for NEC and ongoing, large RCTs comparing transfusion thresholds with different tolerance of anaemia in extremely preterm infants will provide new data to guide transfusion practice. These trials may provide higher quality evidence to determine if red blood cell transfusion or severe anaemia increases the risk of NEC. The need for additional studies evaluating characteristics of transfused red blood cells and feeding during transfusion was discussed.

In the second part of his presentation Dr Patel discussed the potential role of CMV infection in NEC. He presented new data showing an increased risk of NEC associated with higher maternal breast milk CMV content.

Management and outcomes of surgical NEC: a UK-wide cohort study

Using the British Association of Paediatric Surgeons congenital anomalies surveillance system, Dr Amit Gupta and colleagues conducted a study of every infant reported to require surgical intervention for NEC in the UK and Ireland between 1 March 2013 and 28 February 2014. The objective was to describe outcomes and investigate factors affecting prognosis for infants with surgical NEC.

Professor Tomoaki Taguchi.

Professor Neena Modi.

Two hundred and thirty-six infants were included in the study. A key factor associated with reduced one-year mortality was older gestational age at birth. Being small for gestational age (SGA) and requiring parenteral nutrition at 28 days post-decision to intervene surgically were associated with increased one-year mortality.

Parents of infants undergoing surgery for NEC should be counselled that there is approximately a 1:3 risk of death in the first post-operative year but that the risk is lower for infants who are of greater gestational age at birth, who are not SGA and who do not require parenteral nutrition at 28 days post-intervention.

Next steps in NEC clinical trials

Professor Neena Modi explored ideas on our approach to neonatal clinical trials and the problem of not getting answers despite the many RCTs that are carried out. She emphasised the importance of collaboration to develop international neonatal consortia and shared an innovative vision of a more efficient model for taking neonatal clinical trials forward.

The national Maternal and Neonatal Health Safety Collaborative

NHS Improvement's Maternal and Neonatal Health Safety Collaborative aims to support neonatal care services by creating conditions for continuous improvement and a culture of patient safety. Michele Upton discussed the ongoing quality improvement work in the neonatal domain of the collaborative that may impact on NEC.

Summary

As in previous years the conference was very well-received by the delegates. The launch of the SIGNEC website (signec.org) was an important highlight of the conference. This has been made possible largely due to the effort and skills of Joanne Ferguson, who has been a champion supporter of SIGNEC from its very inception.

For updates on the sixth SIGNEC conference contact mineshkhashu@gmail.com or visit the recently launched website at signec.org

Neonatal teams are encouraged to use the website for education and signposting for parents. Feedback on the website would be much appreciated.