CHIEF SCIENTIST OFFICE

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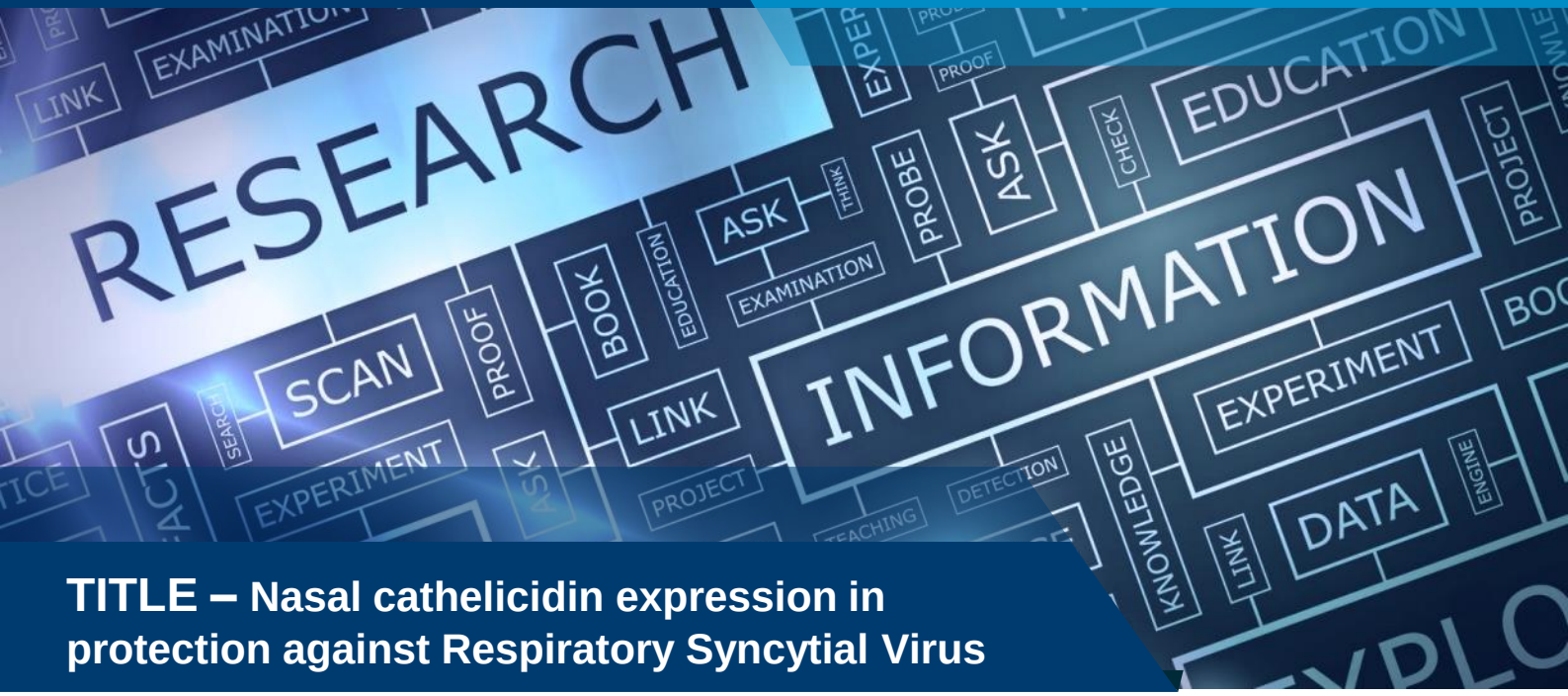
RESEARCH PROJECT BRIEFING



TITLE – Nasal cathelicidin expression in protection against Respiratory Syncytial Virus

AIMS

Over 100,000 children under five years old die each year worldwide from infection with Respiratory Syncytial Virus (RSV). This project focused on cathelicidin, a substance naturally-produced by our bodies, which may act as an 'anti-viral shield' in the nose, blocking infection with this virus. By studying lining fluid from the noses of premature and full-term babies and young children in Scotland and The Netherlands, we aimed to find out when cathelicidin is first made; track its levels over time; work out how this was affected by bacteria living in the nose; understand how cathelicidin levels influenced the risk of severe RSV infection; and evaluate the protective potential of compounds that could stimulate cathelicidin production.

KEY FINDINGS

- Infant and adult nasal cathelicidin expression was measured - this established the normal ranges, and demonstrated that production began soon after birth, with low levels in infants.
- Nasal cathelicidin levels were variable between individuals, with levels affected by a range of host and environmental factors - this demonstrated the existence of nasal cathelicidin-low/deficient individuals who might be future targets for boosting cathelicidin.
- Prematurity of birth did not affect early nasal cathelicidin levels – this showed that low cathelicidin in the nose is not specific to premature infants, despite their increased RSV risk.
- Nasal cathelicidin levels were related to inflammation – this suggested possible mechanisms.
- In certain age groups, the type of bacteria found in the nose was related to the amount of cathelicidin found – this suggested additional mechanisms and treatment approaches.
- Infants who could most effectively increase the amount of cathelicidin in their noses when they got RSV infection, had the best clinical outcomes – this demonstrated the important nature of this process and likely value of cathelicidin boosting treatments in RSV infection.
- Compounds that could stimulate cathelicidin production in cells in the laboratory were able to prevent RSV infection and/or spread of infection in those cells – this can stimulate development of future possible treatments and preventative approaches for RSV infection.

WHAT DID THE STUDY INVOLVE?

Samples were collected from the noses of a range of infant and adult groups, including i) the Edinburgh Theirworld Birth Cohort, with both premature and term infants, with samples collected at birth, the age a given preterm was expected to have been born, 9 months and 2 years, ii) The RESCEU Longitudinal Cohort of healthy term infants, with samples at birth and again during and after mild community RSV infection, iii) a cohort of infants, under 1 year old, hospitalised with severe RSV, with samples at admission and recovery, and iv) healthy younger adults and the elderly. These samples were used to study cathelicidin levels, inflammation, the bacteria present in the nose, and the interaction between these and other clinical factors. In addition, studies used cells in the laboratory to look for protective effects, against RSV infection and spread, of compounds that could induce cathelicidin production.

Engagement events and regular newsletters were delivered for families involved in the Edinburgh Theirworld Birth Cohort, and public engagement activities were developed and delivered around understanding viral lung infections and our immune responses (including creating online animations, school outreach events and newspaper articles).

WHAT WERE THE RESULTS AND WHAT DO THEY MEAN?

Respiratory syncytial virus (RSV) is a major cause of ill health and death, particularly in the very young and in premature infants, but also in the elderly. It is the leading cause of infant hospitalisation in the developed world, causes ~3 million hospital admissions per year in children <5 years old globally, and ~160,000 deaths per year worldwide. There are currently no good therapies for RSV infection. Therefore, there is an important unmet need to find new approaches to prevent and treat RSV infection.

Cathelicidin is a key, naturally-produced, antiviral, Host Defence Peptide made by our bodies and produced in increased amounts during infection. We have previously shown that cathelicidin has antiviral activity against RSV. Furthermore, we have shown the importance, in mice, of both i) natural cathelicidin production in protection against RSV, and ii) a protective impact of applying cathelicidin the lungs of mice infected with RSV.

Using Scottish and Dutch clinical cohorts to collect human samples, this project defined, for the first time, the normal range of nasal cathelicidin expression levels in infants and adults. We showed that infants were born with negligible nasal expression of cathelicidin, but that they upregulated cathelicidin expression over a time course that was similar to the development and stabilisation of the bacterial populations in their respiratory tracts, compatible with an interaction between these factors. These observations complement additional (separately funded) studies in mice, which suggest that increasing age and specific types of bacteria can regulate cathelicidin expression and consequently susceptibility to severe RSV infection.

Stabilised, baseline nasal cathelicidin levels in young children were variable between individuals and were affected by factors including age and season of birth. This demonstrated the existence of nasal cathelicidin-low/deficient individuals, who may be appropriate targets for future interventions designed to boost expression of protective nasal cathelicidin.

We found that prematurity of birth did not affect early nasal cathelicidin levels, with negligible nasal cathelicidin expression observed around birth in both term and preterm infants. This was followed by a highly variable expression at term equivalent age (the gestational date a preterm infants was expected to have been born at), which approached average adult levels. No difference was observed in the expression levels between premature and term infants at 9 months after birth, by which time both group approximated healthy adult levels. These data show that premature infants do not have a developmental defect in nasal cathelicidin expression, despite their increased risk of RSV disease.

The baseline nasal expression levels of cathelicidin at 9 months and 2 years of age in healthy term infants was found to have increased and settled at levels comparable to healthy adults. No substantial difference were observed between younger adults and elderly participants. This is compatible with production of nasal cathelicidin starting after birth, in response to natural introduction of the various bacteria (microbiota) that come to populate the airways after birth, and then stabilising once a stable airway microbiota has been formed, at levels specifically related to the types of bacteria present.

Nasal cathelicidin expression was found to be higher in infants who also had signs of inflammation in the nose. This provides insight into which cells are producing cathelicidin, and how it is regulated. This is under ongoing evaluation, but may inform therapeutic strategies.

Studying the microbiota in the nose showed the expected effects of age and prematurity, but also led to discoveries about cathelicidin. In healthy 9 month old infants (but not at birth), there was a novel, specific relationship between the levels of nasal cathelicidin and the types of bacteria present in the upper respiratory tract. The causality of this relationship is not yet known; cathelicidin may contribute to defining which types of bacteria survive and populate the infant's nose and, in turn, those bacteria may feed back to influence how much cathelicidin is being produced. Both factors can affect susceptibility to infection with RSV and could be targets for new therapeutic and preventative strategies.

Critically, studies using samples from the noses of infants with RSV infections showed i) that effective upregulation and/or higher baseline levels of nasal cathelicidin (seen in mild community acquired cases of RSV infection) was associated with protection from severe disease, and ii) that the lack of an effective nasal cathelicidin upregulation (seen in severe, hospitalised cases) was associated with severe RSV disease. This important observation implies that boosting cathelicidin levels in "at risk" infants and those with constitutively low nasal cathelicidin levels could be valuable to future treatment strategies. This is compatible with published descriptions of low systemic (blood) levels of cathelicidin being associated with severe RSV disease in hospitalised infants in the USA.

Studies using cells in the laboratory found that boosting cathelicidin production (using a range of novel compounds and established cathelicidin inducers, such as Vitamin D3 and Phenylbutyrate) could prevent RSV infection and/or the spread of the virus between these cells. These experiments implicated two mechanisms involved in this process, suitable for future targeting.

Public Engagement

A range of public engagement initiatives were conducted as part of this study, although these were substantially changed as a result of the opportunities available during the pandemic. These included i) regular participation events and newsletters for the families involved in Edinburgh Birth Cohort study, ii) working with a freelance journalist on a feature on this topic for the Daily Mail (https://www.dailymail.co.uk/health/article-8906815/Breakthrough-defeat-cold-virus-thats-second-biggest-global-killer-infants.html), iii) working with Dr Lana Woolford, to develop a series of lay animations explaining the immune response to pulmonary viral infections and vaccinations (https://www.ed.ac.uk/inflammation-research/information-public/videos-resources/immune-memory-coronavirus), and iv) ongoing development (with elements of consultation/co-creation with local teachers and pupils) of an online schools' resource on the relationship between microbiome, infection, immunomodulation, antibiotics and vaccines, featuring a digital game, quizzes, information videos, animations, and teacher extension packs (https://www.ed.ac.uk/inflammation-research/information-public/public-engagement-news-events/cir-visits-castlebrae-high-school-to-co-produce-ne; https://www.ed.ac.uk/inflammation-research/information-public/public-engagement-news-events/cir-eca-collab).

WHAT IMPACT COULD THE FINDINGS HAVE?

These studies:

- describe the normal range of nasal cathelicidin expression in a range of age groups
- establish the existence of nasal cathelicidin-low/deficient individuals
- establish relationships between nasal cathelicidin expression and RSV disease severity, as well as with the types of bacteria present in the nose
- reveal the potential to boost protective cathelicidin expression in RSV infection

The findings of this study therefore have the potential to impact:

- RSV disease prediction and risk stratification of infants based on a minimally invasive nasal absorptive membrane sampling to establish nasal cathelicidin levels
- the development of novel RSV prevention and/or treatment strategies, based on boosting nasal cathelicidin in cathelicidin-low/deficient infants (and potentially elderly individuals) using drug-based or microbiota-based interventions

The significance of these impacts, if realised, will be to enable both early targeted prevention of RSV infection and to develop new treatment approaches for RSV disease.

HOW WILL THE OUTCOMES BE DISSEMINATED?

Two scientific papers have been published (open access) so far:

- Sintoris S, Binkowska JM, Gillan JL, Zuurbier RP, Twynam-Perkins J, Kristensen M, Melrose L, Parga PL, Rodriguez AR, Chu ML, van Boeckel SR, Wildenbeest JG, Bowdish DME, Currie AJ, Thwaites RS, Schwarze J, van Houten MA, Boardman JP, Cunningham S, Bogaert D, Davidson DJ. (2024) Nasal cathelicidin is expressed in early life and is increased during mild, but not severe respiratory syncytial virus infection. Sci Rep. 14(1):13928. doi: 10.1038/s41598-024-64446-1.PMID: 38886476
- Boardman, J. P., Hall, J., Thrippleton, M. J., Reynolds, R. M., Bogaert, D., Davidson, D. J., Schwarze, J. , Drake, A. J., Chandran, S., Bastin, M. E., Fletcher-Watson, S. (2020) Impact of preterm birth on brain development and long-term outcome: protocol for a cohort study in Scotland BMJ Open Sci. 10(3):e035854. doi: 10.1136/bmjopen-2019-035854 PMID: 32139495

The results were presented for the first time in an invited presentation at the Gordon Research Conference of Antimicrobial Peptides 2023 (the preeminent conference in this field):

D. J. Davidson (2023) Gordon Conference on Antimicrobial Peptides, Il Ciocco, Barga, Italy "The Antiviral Significance of Cathelicidins in the Lung“

The details of these studies have been publicly disseminated via the Institute website and by lay summaries on social media platforms, and will be included in future public engagement events and newsletters for Edinburgh Birth Cohort families.

Prof. Davidson has recently retired, so further research and dissemination will be contingent upon other researchers (locally or internationally) choosing to develop this line of study. Future work should i) examine the directionality of the relationship between nasal cathelicidin expression and microbiota (including the impact of specific microbial species), ii) study the cellular targets (and inflammatory significance) of cathelicidin induction, both naturally occurring and also with regard to novel interventions, iii) undertake animal modelling of cathelicidin induction in prevention and/or treatment of RSV infections, iv) pursue further engagement with broader audiences including clinicians, PPI groups, and the wider public.

CONCLUSION

Over 100,000 children under five years old die each year worldwide from infection with Respiratory Syncytial Virus (RSV). This CSO-funded project found that cathelicidin, a substance naturally-produced by our bodies, may act as an important ‘anti-viral shield’ in the nose, to protect against this infection. By studying lining fluid from the noses of premature and full-term infants in Scotland and The Netherlands, the team discovered that cathelicidin is first made soon after birth. They found that infants who effectively ramped-up cathelicidin levels when faced with RSV infection, developed less severe disease. This research suggests new ways to boost cathelicidin levels in the nose, and approaches to determining at-risk infants, highlighting new strategies for prevention and treatment of this disease.

RESEARCH TEAM & CONTACT

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Additional Information

Completion date 28th February 2023 – following pandemic-based extensions.

We would like to thank the CSO for providing costed and no-cost extensions for this project to ameliorate the significant complications caused by the COVID-19 pandemic, and for the CSO staff understanding and flexibility in helping us to maximise the value of this project under those circumstances. This was enormously valuable and appreciated.