

Paradigms and perspectives

Targeting maternal immune function during pregnancy for asthma prevention in offspring: Harnessing the “farm effect”?

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Reduction of the community burden of persistent asthma via prevention of early disease onset remains an important but elusive goal among pediatric researchers. Driven by findings from birth cohort studies, a major focus of current research in this area is on therapeutic strategies to enhance the resistance of infants/pre-schoolers to lower respiratory tract infections, which are among the main drivers of early-stage asthma pathogenesis. Acceptance of the scientific validity of this approach from regulators and funding agencies is growing, evidenced by recent support from the US National Institutes of Health (ORBEX; <https://clinicaltrials.gov/ct2/show/NCT02148796>) and the Australian NHMRC (OMPAC; <https://www.anzctr.org.au/#362459>) for trialing immunomodulators for prevention of wheezy lower respiratory tract infections in infants at high risk of asthma.

We strongly endorse these trials, but we believe that an additional/related therapeutic approach merits investigation. This view derives from the seminal epidemiological findings on the low asthma prevalence among offspring of traditional farming families in Europe and the United States.¹ The acquired asthma-resistant phenotype in these children is associated with prolonged oronasal exposure, not only postnatally but also antenatally, to microbial products derived from farming-related activities, including via maternal consumption of unpasteurized dairy products during pregnancy. These prenatal asthma-protective effects have been replicated experimentally using exposure of pregnant mice to various microbial extracts including from barn dust, and this process is TLR-dependent.¹ In contrast, live pathogens have not been associated with transplacental asthma-protective effects, suggesting that the key underlying mechanism involves sustained exposure of the maternal immune system to *benign* microbial stimuli that trigger TLRs, but without the ensuing high-level inflammation accompanying pathogen clearance.

Mechanistic studies on cord blood mononuclear cells from farmer-children demonstrate that they display prenatal “priming” evidenced by precocious upregulation of host defense functions that normally mature postnatally, particularly expression of microbial-sensing receptors,¹ preequipping them for immediate responsiveness to maturation-inducing microbial signals at birth. Consistent with this, the farmer-children display accelerated progression from the neonatal T_H2-skewed immunophenotype associated with “high risk” for infection (and also atopy) to a more balanced/functionally mature state that includes robust T_H1-associated/IRF7-dependent immune capacity required for efficient pathogen clearance, accompanied by enhanced regulatory T-cell activity necessary for calibration of immunoinflammatory responses to pathogens and other environmental stimuli including aeroallergens.¹ Of note in this regard, farmer-children display reduced susceptibility to respiratory infections and inhalant allergies,¹ which are important cofactors in childhood asthma pathogenesis. These findings collectively suggest that maternal farming-associated microbial exposures, reinforced by postnatal exposures of their offspring, serve to mold and calibrate development of immunocompetence in the offspring, ultimately enhancing asthma resistance. Moreover, the effectiveness of this process is correlated with the *diversity* of the microbial stimuli involved.² In this regard, stimulatory signals for immune maturation during infancy are derived from interactions not only with microbial pathogens but also with the gastrointestinal microbiome that is rapidly established after birth. Moreover, the maternal microbiome during pregnancy (which is likely modified by microbial-rich farm exposures) is an important source of immune maturation signals during intrauterine development,³ in addition to contributing to postnatal seeding of the infant microbiome,³ which is also now recognized as a significant modulator of early asthma risk.

TOWARD TRANSLATION OF THE “FARM EFFECT”: PRECLINICAL STUDIES IN PREGNANT AND INFANT MICE

We have been testing the potential benefits of controlled microbial exposure, including during pregnancy, in animal models. The agent used is the immunomodulator OM85-bronchovaxom (OM85BV) (reviewed in Scott et al⁴ and Mincham et al⁵), which comprises an extract of 21 respiratory pathogens containing a diverse array of microbial-derived TLR ligands that may constitute the active agent(s); it has been in widespread clinical use since 1980s for protection against the acute effects and sequelae of severe respiratory infections in infants/children/immunocompromised adults including wheeze, and protection against rhinitis and atopic dermatitis in infants/children,

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Benign microbial exposures (farming environment; OM85BV)



Proposed direct and transplacental effects of maternal “immune training” during pregnancy

Immunological sequelae	Clinical sequelae
Bone marrow myeloid progenitor re-programming; Optimization of regulatory/inflammatory effector cell balance in mucosal tissues; (respiratory; gestational) during local/systemic inflammatory events; Modulation of inflammatory gene networks during microbial/allergen challenge; Reduced levels of circulating pro-inflammatory mediators during acute infections including sepsis.	Stabilization of immune homeostasis in gestational/respiratory tissues at baseline + during local/systemic inflammatory events (infections; asthma exacerbations); Reduced susceptibility to septic shock; Reduced maternal symptom intensity accompanying infections (e.g. influenza); Reduced risk of miscarriage/fetal growth retardation following maternal infection; Reduced risk of severe asthma exacerbations.
Enhanced prenatal maturation of innate immune mechanisms; Increased output of DC from bone marrow during infancy; Accelerated postnatal establishment of functional lung/GIT mucosal DC networks; Increased responsiveness to microbial signals that drive postnatal immune maturation; Increased ability to detect/eradicate respiratory pathogens; Enhanced capacity to calibrate airway inflammatory response intensity; Enhanced control of myeloid progenitor cell responses to bacterial LPS exposure.	Increased ability to clear respiratory infections during “high risk” neonatal/infant period; Reduced susceptibility to septic shock; Reduced risk of early sensitization to aeroallergens; Increased resistance to development of virus-associated wheeze during infancy; Lack/reduction of airway symptoms despite sensitization/exposure to aeroallergens; Reduced risk for development of persistent wheeze/asthma.

A longer term perspective?

The choice of OM85BV for trialing the potential benefits to mother and offspring of maternal immune training during pregnancy is driven not only by the performance of this agent in forerunner human infant and animal studies, but also by the current lack of alternative immunomodulators with a comparable record of human safety/efficacy. The closest possible alternative may appear to be probiotics, however the most recent meta-analyses confirm the limited effectiveness of this approach for asthma prevention. In contrast to probiotics OM85BV is derived from a complex mix of multiple bacterial strains containing multiple TLR ligands, echoing the “diversity” of microbial stimulation provided by the farming environment which is recognized as a strong correlate of the effectiveness of farm exposures in relation to protection against asthma. Future research may eventually identify single-molecular-entity pharmaceuticals that can mimic the pan-specific microbial signals that underlie this farm-associated immune training phenomenon. However, given that OM85BV is administered orally, it is tempting to speculate that similar effects may be achievable via development of appropriate microbial-derived dietary supplements, and if so treatment costs would be considerably lower relative to conventional pharmaceuticals.

FIG 1. Potential benefits to mother and offspring of OM85BV treatment during pregnancy. Based on results from a series of independent experimental model studies, it is proposed that maternal OM85BV treatment during pregnancy will potentially result in a range of benefits to both mother and offspring. DC, Dendritic cell.

and is used in the ORBEX/OMPAC infant trials cited above. We have demonstrated OM85BV-mediated protection against experimental asthma and respiratory infections that is associated with expansion of regulatory (lymphoid/myeloid) cell populations in airway and gestational tissues.^{4,6} Moreover, we have demonstrated that repeated administration of OM85BV is safe in healthy murine pregnancies, and provides effective protection against the inflammatory effects of bacterial LPS and live viral challenge, markedly reducing maternal morbidity and preserving normal maternal/fetal weight gain.^{4,5}

These beneficial treatment effects were associated with preferential attenuation of proinflammatory response pathways in gestational tissues and parallel preservation of both regulatory and antimicrobial functions required for pathogen clearance.^{4,5} The immunoinflammatory phenotype that develops in OM85BV-treated mothers exhibits hallmarks of “immune training,”⁷ a process resembling adaptive immunologic memory but instead involving innate immunity, in which responsive cell populations manifest a prolonged state of enhanced functionality. Crucially, these immune training effects also extend to their offspring, which postnatally display enhanced capacity to regulate airways immunoinflammatory responses.⁵ Immune training is mediated via epigenetic mechanisms and/or alterations in the number/phenotype of precursors of the cell populations involved.⁷ In this regard, bone marrow myeloid progenitors in these offspring, which provide precursors for tissue dendritic cells that mediate immune surveillance, expand markedly by late gestation, and the subsequent postnatal establishment of their airway mucosal dendritic cell networks responsible for maintenance of local immunologic homeostasis is accelerated.⁵ This is in turn associated with reduced susceptibility to early postnatal development of T_H2 -associated experimental asthma,⁵ and enhanced resistance to respiratory viral infection, including during the neonatal period when susceptibility is highest (article in preparation). Moreover, capacity to regulate the intensity of

bone marrow myeloid progenitor responses to LPS (as a mimic for bacterial sepsis) is markedly increased in the offspring,⁵ mirroring the acquired LPS-resistant phenotype observed in their OM85-treated mothers.⁴

PRIMARY PREVENTION OF ASTHMA: SCIENTIFIC RATIONALE FOR PROOF-OF-CONCEPT HUMAN TRIAL IN PREGNANCY

In summary, we hypothesize that maternal OM85BV treatment during pregnancy may provide significant and interrelated benefits to both mother and offspring (Fig 1). Potential maternal benefits include increased resistance to the negative effects of bacterial/viral infections, which can impact fetal growth and increase the risk of miscarriage, as well as attenuation of infection-associated inflammatory symptoms that are commonly exaggerated during pregnancy.⁸ Moreover, on the basis of earlier experimental findings from us and others in sensitized animals (reviewed in Mincham et al⁵), we posit that in pregnant mothers with asthma (the group most likely to produce offspring with asthma), treatment would also reduce susceptibility to asthma exacerbations, which is heightened during pregnancy especially among those with the most severe asthma,⁹ and which is a known risk factor for later asthma development. In addition to mitigation of negative influences on offspring stemming from maternal inflammatory events, we posit a second tier of positive treatment effects resulting from transplacental immune training of microbial-sensing capability, optimizing their responsiveness postnatally to TLR-encoded microbial-exposure signals that drive maturation of the effector/regulatory functions that mediate resistance to both respiratory pathogens and to the inflammatory consequences of aeroallergen sensitization.

We posit that mothers with asthma represent the ideal group for an initial trial, given the high prevalence of asthma

development in their offspring coupled with the high rate of exacerbations expected among the mothers themselves during pregnancy,⁹ and present opportunities to ascertain treatment effects with a sample size that is not prohibitively large. We acknowledge however that issues related to safety in pregnancy need to be formally addressed first given that only 1 relevant population-based surveillance study is currently available, which detected no adverse effects in mothers/offspring.¹⁰ In this regard, OM85BV comprises a mix of natural products that are ubiquitous in the environment, and as such there appears no *a priori* basis for assumption of deleterious effects in pregnancy. We would argue that the 40 years of human safety data available from settings other than pregnancy, coupled with recent studies in animals, provide sufficient body of evidence for justification of an initial human trial. However, patients' views of potential risks/benefits would be a key consideration before proceeding.

In moving forward we need to ensure that entrenched reticence concerning studies in pregnancy does not prevent rational evaluation of this proposal, potentially repeating mistakes of the past exemplified by the decades-long delay in introduction of maternal vaccination during pregnancy, a practice that is now recognized to provide major transplacental benefits to offspring.

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