The role of early life infection on the programming of CD4+ T-cells

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Figure 7.3.3c  Levels of immunoglobulin, total IgE in serum of mice infected as a neonate with *S. typhimurium* or sham-infected.

Figure 7.3.4a  Levels of IL-5 (A, D) unstimulated, (B, E) OVAp stimulated, (C, F) CD3/CD28 stimulated, (A,B,C) splenocytes and (D,E,F) PBLN of neonatally sham-infected or *S. typhimurium* infected mice.

Figure 7.3.4b  Levels of IL-10 (A, D) unstimulated, (B, E) OVAp stimulated, (C, F) CD3/CD28 stimulated, (A,B,C) splenocytes and (D,E,F) PBLN of neonatally sham-infected or *S. typhimurium* infected mice.

Figure 7.3.4c  Levels of IL-4 (A, D) unstimulated, (B, E) OVAp stimulated, (C, F) CD3/CD28 stimulated, (A,B,C) splenocytes and (D,E,F) PBLN of neonatally sham-infected or *S. typhimurium* infected mice.

Figure 7.3.4d  Levels of IL-13 (A, D) unstimulated, (B, E) OVAp stimulated, (C, F) CD3/CD28 stimulated, (A,B,C) splenocytes and (D,E,F) PBLN of neonatally sham-infected or *S. typhimurium* infected mice.

Figure 7.3.4e  Levels of IFN-γ (A, D) unstimulated, (B, E) OVAp stimulated, (C, F) CD3/CD28 stimulated, (A,B,C) splenocytes and (D,E,F) PBLN of neonatally sham-infected or *S. typhimurium* infected mice.

Figure 7.3.5a  Percentage of (A) CD4+ cells (B) CD8+ cells and KJ1-26+ cells as a % of total (C) CD4+ cells and (D) CD8+ cells in splenocytes from mice that were sham or *S. typhimurium* infected as neonates.

Figure 7.3.5b  Percentage of (A) CD4+ cells (B) CD8+ cells and KJ1-26+ cells as a % of total (C) CD4+ cells and (D) CD8+ cells in PBLN from mice that were sham or BCG infected as neonates.
List of Abbreviations

2-ME         2-mercaptoethanol
AAI         allergic airways inflammation
AAD         allergic airways disease
Ab          antibody
ACCM        animal cell culture medium
AHR         airways hyper-responsiveness
ANOVA       analysis of variance
APC         antigen-presenting cell
Aro         aromatic prechorismate pathway
BALF        broncho-alveolar lavage fluid
BCA         bicinchoninic acid
BCG         Mycobacterium bovis (Bacille Calmette Guerin)
BSA         bovine serum albumin
CD          cluster of differentiation
CFU         colony forming units
COPD        chronic obstructive pulmonary disease
d          day
DC          dendritic cell
ELISA       enzyme linked immunosorbant assay
FACS        fluorescence-activated cell sorting
FCS         foetal calf serum
G           gravity
GI          gastro-intestinal
GINA        global initiative for asthma
GM-CSF      granulocyte-macrophage colony stimulating factor
HBSS        Hank’s buffered salt solution
HEPES       4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid
IFN         interferon
Ig          immunoglobulin
IL          interleukin
i.n.        intranasal
i.p.        intraperitoneal
iTreg       inducible T-regulatory cell
i.v.        intravenous
IVC         individually ventilated cage
LB          Luria-Burtani
LH          lung homogenate
LN          lymph node
LPS         lipopolysaccharide
m           monoclonal
M-cells     microfold cells
mDC         myeloid dendritic cell
MSC         mucous secreting cell
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<th>Abbreviation</th>
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<tr>
<td>NEA</td>
<td>non-eosinophilic asthma</td>
</tr>
<tr>
<td>NK</td>
<td>natural killer</td>
</tr>
<tr>
<td>NKT</td>
<td>natural killer T-cell</td>
</tr>
<tr>
<td>nTreg</td>
<td>natural T-regulatory cell</td>
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<tr>
<td>OD</td>
<td>optical density</td>
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<td>OVA</td>
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<tr>
<td>PAMP</td>
<td>pathogen associated molecular pattern</td>
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<td>periodic acid Schiff</td>
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<td>PBLN</td>
<td>peri-bronchial lymph node</td>
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<td>PBS</td>
<td>phosphate buffered saline</td>
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<tr>
<td>pDC</td>
<td>plasmacytoid dendritic cell</td>
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<tr>
<td>PMN</td>
<td>polymorphonuclear</td>
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<tr>
<td>PP</td>
<td>Peyer’s patch</td>
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<tr>
<td>PRR</td>
<td>pathogen recognition receptor</td>
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<tr>
<td>RBC</td>
<td>red blood cell</td>
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<tr>
<td>RPMI</td>
<td>Roswell Park Memorial Institute</td>
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<td>RT</td>
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<td>SEM</td>
<td>standard error of the mean</td>
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<td>SPBS</td>
<td>sterile phosphate buffered saline</td>
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<td>SPF</td>
<td>specific pathogen free</td>
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<td>STAT-6</td>
<td>Signal transducer and activator of transcription-6</td>
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<tr>
<td>Tg</td>
<td>transgenic</td>
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<td>TCR</td>
<td>T-cell receptor</td>
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<td>TGF</td>
<td>transforming growth factor</td>
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<td>Th</td>
<td>T helper</td>
</tr>
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<td>TLR</td>
<td>Toll-like receptor</td>
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<tr>
<td>TMB</td>
<td>Tetra-methyl benzidine</td>
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<tr>
<td>TNF</td>
<td>tumour necrosis factor</td>
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<td>regulatory T-cell</td>
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<tr>
<td>VEGF</td>
<td>vascular endothelial growth factor</td>
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<td>white blood cell</td>
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<td>WCC</td>
<td>white cell count</td>
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<td>WHO</td>
<td>World Health Organisation</td>
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<td>WT</td>
<td>wild-type</td>
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Conference proceedings arising from this work

Conference presentations


Conference poster presentations


Competitive grants arising from this work

Professor Paul Foster, **Ms Angela Ferguson**. (2006)- ‘CD4+T-cell programming in early life and neonatal infection’. GSKA Post Graduate Support Grant, GlaxoSmithKline Australia, Boronia, Vic. ($A25, 000 over two years).
ABSTRACT

Asthma is a chronic inflammatory disease of the airways that is characterised by activation of CD4+ T-helper 2 type (Th) cells and eosinophils. The cause of this aberrant Th2 response is unknown but lack of early life infection is thought to play a significant role. The timing of infection and the type of pathogen may be critical to programming the immune response to a protective Th1, or destructive Th2, phenotype.

The immune responses to infection with Salmonella typhimurium and Mycobacterium bovis Bacille Calmette Guerin (BCG) have been identified as targets for reprogramming or preventing the development of asthma. However, the role of these infections in contributing to a Th2-Th1 switch or suppression of this response remains limited. In this investigation ovalbumin (OVA) T-cell receptor (TCR) transgenic (Tg) mice in combination with these bacterial strains expressing OVA have been used to specifically track the affects of each infection as well as OVA exposure on the T-cell response and the development of allergic airways disease (AAD) in the mouse model.

BCG infection as an adult and a neonate prior to OVA challenge induced significant reductions in eosinophils in broncho-alveolar lavage fluid (BALF) and lung tissue compared to sham-infected mice that received OVA challenge. However, high levels of both Th1 (interferon gamma (IFN-γ)) and Th2 (interleukin (IL)-4, IL-5, IL-13) cytokines from supernatants of cultured peri-bronchial lymph node (PBLN) cells and splenocytes were found in all groups examined. Further studies tracking the development of the immune system after BCG infection at birth without OVA exposure revealed significant decreases in lung tissue eosinophils and decreased immunoglobulin (Ig) G1, IgG2a and IgE levels from serum compared to sham-infected controls. This coincided with decreased numbers of CD4+ and CD8+ T-cells in the spleens and PBLN cells. Levels of cytokines in splenocytes and PBLN cell cultures failed to show significant trends toward either a polarised Th1 or Th2, leaving a mixed Th1/Th2 phenotype.

Infection with S.typhimurium lowered eosinophil levels in BALF, and mucous secreting cell (MSC) and eosinophil number in lung tissue after challenge with