Chapter-3

Development of mouse model of asthma: Total and OVA specific IgG and IgE antibodies in serum and BALF
3.1 INTRODUCTION

Immunoglobulin E (IgE) and mast cells are believed to play important roles in allergic inflammation. Significant progress has been made recently in our understanding of airway inflammation and airway hyperresponsiveness through studies of murine models of asthma. Sensitized mice acutely challenged with inhaled Ovalbumin (OVA) develop allergic airway inflammation, characterized by OVA-specific IgE production, airway eosinophilia, increased pulmonary B and T lymphocytes, and airway hyperreactivity. The present experiment was planned to study the levels of IgE and IgG in OVA-induced experimental asthma in mouse model. Both IgG and IgE were measured in the serum and bronchiolar alveolar lavage fluid (BALF).

3.2 MATERIALS AND METHODS

3.2.1 Animals and Treatments

Female BALB/c mice weighing approximately 25 g were obtained from the animal facility of Indian Institute of Toxicology Research and divided into the following groups. Each group comprised of six mice.

Investigations were done on day 28.

Group I Control mice (SAL/SAL)

Group II Mice sensitized and challenged with OVA. (OVA/OVA mice)

Following protocol was adopted to measure OVA- specific antibodies.

3.2.2 Evaluation of total and antigen-Specific IgG and IgE antibodies

Total and antigen-specific IgG and IgE antibodies in the BALF and sera of OVA/OVA and SAL/SAL mice were evaluated by quantitative ELISA following the
manufacturer's protocol (Mouse IgG ELISA Quantitation kit; Bethyl Laboratories, Inc., Montgomery, Tex., USA).

3.3 RESULTS

The level of total and OVA-specific IgG and IgE in serum and BALF samples from OVA/OVA and SAL/SAL mice are shown in Table 3.1. A significant increase in the level of total and OVA specific IgE and IgG in the serum and BALF of OVA/OVA was observed in comparison to normal mice. Thus, a single sensitization by OVA followed by a series of challenge by the same allergen induced allergic asthma in mice in terms of rise in immunoglobulin level.
Table 3.1

Total and OVA-specific IgG and IgE in normal and asthmatic mice

<table>
<thead>
<tr>
<th>Immunoglobulins</th>
<th>SAL/SAL</th>
<th>OVA/OVA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total serum IgG, mg/ml</td>
<td>3.31 ± 1.89</td>
<td>12.83 ± 1.22*</td>
</tr>
<tr>
<td>OVA-specific serum IgG, mg/ml</td>
<td>2.86 ± 1.03</td>
<td>8.86 ± 2.57*</td>
</tr>
<tr>
<td>OVA-specific BALF IgG, µg/ml</td>
<td>ND</td>
<td>170.56 ± 12.43</td>
</tr>
<tr>
<td>Total serum IgE, µg/ml</td>
<td>5.87 ± 3.41</td>
<td>35.58 ± 5.73*</td>
</tr>
<tr>
<td>OVA-specific serum IgE, µg/ml</td>
<td>2.07 ± 0.50</td>
<td>27.35 ± 3.18*</td>
</tr>
<tr>
<td>OVA-specific BALF IgE, µg/ml</td>
<td>ND</td>
<td>11.93 ± 2.86</td>
</tr>
</tbody>
</table>

Data represent mean ± SD of values from 6 mice in each group. * P < 0.05, statistically significant compared to SAL/SAL.
3.4 DISCUSSION

The immunopathological hallmarks of allergic diseases are: elevated total and allergen specific serum and BALF IgG and IgE, inflammation, airway remodeling and AHR. A wealth of clinical and experimental data suggests that allergic asthma is due to an aberrant lung immune response mediated through T-helper type 2 cells (Th2 cells) and associated cytokine-signaling pathways. The normal lung is able to distinguish between airborne antigens associated with infectious agents, to whom an immunological response is generated, and harmless inhaled antigens, which are usually ignored. In the asthmatic lung, some of these normally harmless antigens activate specific Th2 cells and elicit an inflammatory response characterized by Th2 cytokine production, eosinophilic airway inflammation, airway hyperresponsiveness, and bronchoconstriction (Craig et al., 2004). These pulmonary responses may be accompanied by systemic allergic sensitization, manifested by elevated titers of antigen-specific IgE. Bronchoalveolar lavage fluid (BALF) from mice with allergic airway inflammation contains significant amounts of IgE.

The present results depicted significant high levels of serum and BALF, IgG as well as IgE antibodies (total and OVA specific) in mice treated with OVA/OVA in comparison of control mice (SAL/SAL). Presence of elevated level of IgE and IgG indicates the allergic nature of asthma in mice sensitized and challenged with allergen (OVA). Comparable levels were also measured by Brusselle et al. 1994 after sensitization with Ovalbumin in combination with alum in C57BL/6 mice. Similar results were reported by Arm and Lee in 1992 for elevated levels of IgE in asthmatic mice. It can be concluded that, both antibodies might be taking part in antibody...
dependent pathway which is important for allergic responses. Elevated levels of IgE and IgG are known to be important in the development of an allergen induced airway response. The presence of these characteristics in OVA sensitized and challenged mice confirm asthmatic status of mouse.