



2010 Research Grant Program Winning Abstract

Basophils and Murine Experimental Model of Asthma

By Asifa Zaidi

Asthma is a disorder defined by reversible broncho-constriction, but it is often accompanied by chronic airway hyperresponsiveness (AHR) and inflammation. Asthma is a very heterogeneous and complex disease with multiple pathways and cellular components involved in its pathogenesis. Despite extensive research over the last several decades, there remain many unexplained aspects of asthma pathogenesis, and this is likely an impediment to the development of effective therapy for control of severe asthma. Basophils have recently been discovered to be involved in the initiation and promotion of Th2 immune responses that are associated with asthma. Furthermore, both human and murine basophils secrete large amounts of Th2 cytokines, which are major modulators in the development of asthmatic responses. Although basophils are thought to be involved in inflammatory diseases and accumulate in tissues, the specific role(s) of basophils in the development and effector phase of asthma has not yet been resolved. Interestingly, studies with human subjects demonstrated presence of basophils in significantly increased numbers in the airways of asthmatics and individuals who died from asthma. Furthermore, basophils are recruited to the bronchial mucosa after segmental bronchial allergen challenge in subjects with atopic asthma. We have shown in our preliminary data that basophils are recruited to the lung during immunization phase and present in bronchoalveolar fluid (BAL) after allergen challenge in the experimental model of OVA-induced experimental asthma in mice. However, it is not known whether basophils are recruited to the lung and BAL by all allergens and associated with severity of asthma symptoms (AHR and airway inflammation). We therefore propose that understanding the specific functions of basophils in the asthma pathogenesis (AHR and airway inflammation) is important and may lead to new approaches in asthma therapy. Based on these findings, we hypothesize that basophils are required for all allergen-induced airway hypersensitivity and inflammation. We have successfully used the BD FACSAria™ system in the flow cytometry core at Johns Hopkins University School of Medicine at Baltimore to identify and sort basophils from blood, bone marrow, spleen, and lung tissues of mice (Zaidi and MacGlashan. *J Immunol.* 2010;184:1463).

We aim to determine the contribution of basophils in asthma pathogenesis in two ways. First, we will determine the percentage recruitment of basophils in the lung during allergen immunization phase and presence of basophils in the lung and BAL after allergen challenge. We will use allergens (proteases, fungus, dust mite, cockroach, papain) and intranasal immunization and challenge protocols that are relevant to human asthma. We anticipate that basophils will be recruited in increased numbers during allergen immunization phase as well as after allergen challenge to the lungs in response to all allergens and will be associated with severity of experimental asthma symptoms (AHR and airway inflammation). Basophils will be identified and purified by the BD FACSAria sorter in the lung, blood, bone marrow, and lymphoid tissues after immunization as well as after allergen challenge. We will also determine the activation status of basophils by estimation of CD69/CD200R expression. This is a novel



approach, since currently there is no information on the recruitment of basophils to the lung and BAL in response to all allergens including OVA (shown by us) and their contribution to asthma symptoms (AHR and airway inflammation). Second, we plan to specifically deplete basophils from mice and determine the effect of basophil deficiency on allergen-induced AHR and inflammation. We hypothesize that deficiency of basophils during allergen immunization and challenge will abrogate the allergen-induced AHR and airway inflammation. Overall these experiments will provide insights into the regulation of basophils in allergen-induced AHR and inflammation in an experimental model of asthma in mice. Furthermore, this study will provide the specific contribution of basophils in an experimental model of asthma in mice and provide a potential novel basophil specific approach for further research and development of targeted therapeutics for asthma patients.

A BD Biosciences Research Grant would be an appropriate mechanism to support this research, since the techniques used for this proposed research include flow cytometry, cell sorting with the BD FACS Aria system, and antibodies for blocking, sensitization, detection and sorting of basophils. Also, immunohistochemistry and Western blot will use BD products and supplies. These studies have the potential to provide a novel tool for identifying basophil-dependent targets as disease modifying agents for treatment of asthma and allergic diseases.

The BD Biosciences Research Grant Program aims to reward and enable important research by providing vital funding for scientists pursuing innovative experiments to advance the scientific understanding of disease.

Visit bdbiosciences.com/grant to learn more and apply online.