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Food allergy: Insights into etiology, prevention, and treatment provided by murine models

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Food allergy is a rapidly growing public health concern because of its increasing prevalence and life-threatening potential. Animal models of food allergy have emerged as a tool for identifying mechanisms involved in the development of sensitization to normally harmless food allergens, as well as delineating the critical immune components of the effector phase of allergic reactions to food. However, the role animal models might play in understanding human diseases remains contentious. This review summarizes how animal models have provided insights into the etiology of human food allergy, experimental corroboration for epidemiologic findings that might facilitate prevention strategies, and validation for the utility of new therapies for food allergy. Improved understanding of food allergy from the study of animal models together with human studies is likely to contribute to the development of novel strategies to prevent and treat food allergy. (J Allergy Clin Immunol 2014;133:309-17.)

Key words: *Food allergy, anaphylaxis, murine model, microbiota, regulatory T cells*

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Abbreviations used
AD: Atopic dermatitis
CM: Cow's milk
CT: Cholera toxin
DC: Dendritic cell
FAHF-2: Food Allergy Herbal Formula-2
Foxp3: Forkhead box protein 3
GF: Germ free
LP: Lamina propria
MLN: Mesenteric lymph node
OIT: Oral immunotherapy
PAF: Platelet-activating factor
SEB: Staphylococcal enterotoxin B
Treg: Regulatory T

The genetic revolution over the last decade has had few stronger influences than that on our ability to generate tools from the laboratory mouse. Work stemming from manipulating murine embryonic stem cells earned Drs Mario Capecchi, Martin Evans, and Oliver Smithies the Nobel Prize in Physiology and Medicine in 2007 and led to a new dawn of scientific inquiry that has revolutionized our understanding of most fields of biology, including immunology and allergy. These tools, such as gene deletions, gene insertions, gene reporters, and more, have allowed researchers to define biology in ways previously unobtainable. Despite this, concerns regarding what relevance murine models have in understanding human disease persist.

It goes without saying that mice and human subjects differ in many ways. This was spotlighted recently in an article reporting that the transcriptional responses observed in murine models of endotoxemia, burns, and trauma were not representative of those observed in patients' samples.[1] Although this study has been criticized by leaders in these areas,[2] it raised an important question of whether studies performed in mice (or any animal for that matter) have meaningful bearing on the diseases about which they are intended to inform.

Interestingly, allergy is one field in which this transcriptional analysis approach has shown remarkable consistency between murine and human samples. For example, a recent study using a murine model of atopic dermatitis (AD) included comparisons with data from affected human skin and showed a high degree of homology in the gene expression profile.[3] Using genetically modified mice, the authors definitively showed key roles for T cells and mast cells in disease pathogenesis. Similarly, in a murine model of severe asthma, Yu et al[4] performed transcriptional comparison analysis between the murine lung and patient lung biopsy specimens. Their data elegantly showed a highly significant association in gene expression patterns that was lost in mast cell–deficient mice but restored if mast cells were reconstituted by

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Terms in boldface and italics are defined in the glossary on page 310.

mean of adoptive transfer. There is no doubt that such validation approaches will be an important aspect of mechanistic studies moving forward, especially that researchers in the field of allergy possess a strong collection of tools to study the diseases.

This review aims to outline the role animal models might play in understanding food allergy, as well as to highlight how animal models might contribute to the development of future therapies. However, it is first worth discussing what precisely constitutes an animal model.

There are generally 3 main types of approaches to modeling human disease: homologous (in which the underlying cause, symptoms, and treatments are shared), isomorphic (in which the symptoms and treatments are shared), and predictive (in which symptoms might be different but treatments show efficacy). Within the allergy field, most models are isomorphic.

As we well know from asthma model research, sensitization with intraperitoneal ovalbumin and alum has been a mainstay approach of the airway inflammation community for many years. However, ovalbumin is not an allergen associated with asthma nor do human subjects encounter antigens through the intraperitoneal route or in the context of alum adjuvant. However, because the type 2 immune response and ensuing eosinophilic airway inflammation are highly associated with asthma, this isomorphic model has facilitated significant progress in our understanding of asthma mechanisms helped by the availability of tools such as ovalbumin-specific T-cell receptor transgenic mice, mAbs, and tetramers. In recent years, a shift toward using house dust mite has driven the desire for a more homologous disease model, although there are few data about the physiologic relevance of the levels of house dust mite extract delivered to the mice to elicit pathology. In the field of food allergy, there is insufficient information regarding the nature of food allergens and the mechanisms responsible for loss or lack of tolerance in patients for us to develop a true homologous model at this time because feeding of food allergens to mice elicits oral tolerance, as it does in most human subjects. Instead, mucosal adjuvants, such as ***cholera toxin*** (CT)[5] or ***staphylococcal enterotoxin B*** (SEB),[6,7] or genetically manipulated mouse strains susceptible to enteral sensitization[8] have been used. Interestingly, physiologic exposure to *Staphylococcus aureus*, SEB, or both has been closely connected with many allergic diseases in human subjects,[9] suggesting the potential for a homologous link, although connections between *S aureus* and food allergy remain to be determined. However, the use of these models has already provided significant advances in our understanding of the potential mechanisms of pathogenesis of food allergy and in the development of new therapies.

This review will address how such models can work in synergy with human studies to promote better understanding of the mechanisms, etiology, and potential therapy for food allergy. Key points of this review are listed in Table I.

DEFINING THE ETIOLOGY OF FOOD ALLERGY USING MURINE SYSTEMS

One of the critical advantages of using mouse models to study food allergy is that allergic sensitization or tolerance can be induced to specific allergens under controlled environmental conditions within defined genetic backgrounds, which is not possible in human subjects. This aspect of mouse models allows extensive and precise investigations into the mechanisms involved in disease etiology, such as identification of possible triggers, as well as pathways involved in food allergy. Normally, ingestion of food results in oral tolerance in mice, as in most human subjects. Although the immune mechanisms responsible for breakdown in oral tolerance are not fully understood, increasing evidence from mouse models indicates that alterations in ***regulatory T (Treg) cell*** function and environmental factors, such as microbiota, are likely important contributors to allergic sensitization and food allergy. Increased intestinal permeability has been suggested as a potential cause of food allergy,[10] possibly through increased exposure to intact protein. Loss of oral tolerance can also occur when food antigen is presented through alternative routes, such as the skin, and results in the development of food allergy.

Induction mechanisms of food allergy

To establish tolerance or initiate allergic responses against food antigens, ***dendritic cells*** (DCs) acting as professional antigen-presenting cells must encounter the antigens and bring them to local lymph nodes. Although the function of various intestinal antigen-presenting cell subpopulations to induce tolerance versus sensitization is currently unclear and requires further investigation (for further information, see Pabst and Mowat[11] and Ruiter and Shreffler[12]), under normal conditions, $CD103^+$ DCs have been thought to capture antigen in the lamina propria (LP) and Peyer patches and migrate to the mesenteric lymph nodes

GLOSSARY

CHOLERA TOXIN (CT): A highly toxic protein secreted by the bacterium *Vibrio cholerae*, which causes severe gastric inflammation in animals and is often used to induce an immune response in biological experiments.

DENDRITIC CELLS (DCS): Professional antigen-presenting cells that link the innate and adaptive immune systems by capturing and then presenting antigen to T cells.

FORKHEAD BOX PROTEIN 3 (FOXP3): A transcription factor responsible for the development and function of regulatory T cells.

PLATELET-ACTIVATING FACTOR (PAF): A potent mediator of inflammatory responses that is a regulator of anaphylaxis. Studies have indicated that blocking the effects of PAF prevents fatal anaphylaxis.

REGULATORY T (TREG) CELLS: A subset of T cells that control inflammation and induce tolerance by secreting anti-inflammatory cytokines.

STAPHYLOCOCCAL ENTEROTOXIN B (SEB): A superantigen produced by the bacterium *Staphylococcus aureus* that elicits a massive cytokine release. This severe inflammatory response often serves as a model of inflammation in biological studies.

TGF-β: A cytokine produced by a variety of cells that is involved in the suppression of inflammation by regulating cellular proliferation and differentiation.

THYMIC STROMAL LYMPHOPOIETIN (TSLP): A cytokine that stimulates the maturation of T cells through activation of antigen-presenting cells, such as dendritic cells and macrophages.

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