Food sensitivity and the pathogenesis of atopic dermatitis

Hugh A Sampson MD

J R Soc Med 1997;**90**(Suppl. 30):2–8 SECTION OF PAEDIATRICS, 22 OCTOBER 1996

Atopic dermatitis is one form of eczema which frequently develops in early infancy and is distinguished by a characteristic distribution, extreme pruritus, chronically relapsing course, and association with asthma and allergic rhinitis[1]. The distribution of the rash typically varies with age[2], involving the cheeks and extensor surfaces of the arms and legs in infancy, the flexor surfaces in the young child, and flexor surfaces, hands and feet in the teenage patient and young adult. Unlike most dermatoses, atopic dermatitis has no primary skin lesion. The diagnostic criteria of Hanifin and Rajka have been internationally accepted as a standard for diagnosing atopic dermatitis[3]. The incidence of atopic dermatitis has been increasing over the past 40 years and is now estimated to affect between 10% and 12% of the paediatric population[4].

DIFFERENT FORMS OF ATOPIC DERMATITIS

As with asthma, there seem to be two forms of atopic dermatitis—one in which the disease is triggered by allergens with potential IgE-dependency (extrinsic form) and one in which the disease appears to be IgE-independent (intrinsic form). The pathogenic role of antigen-specific IgE antibodies in atopic dermatitis is supported by studies delineating the classical immunopathogenic role of the IgE-mediated cutaneous late-phase response and of the non-classical role of IgE-bearing antigen-presenting cells (APCs), especially Langerhans cells and dendritic cells, in establishing the Th_2 lymphocytic response. In the past, skin biopsy findings from patients with atopic dermatitis were felt to be incompatible with an IgE-mediated mechanism but rather were indicative of a classical type IV, cell-mediated, response. Histological examination of acute skin lesions of atopic dermatitis reveals spongiosis, epidermal hyperplasia, and ballooning of the keratinocytes secondary to intracellular oedema. Mast cell and basophil numbers are normal, and eosinophils are rare[5]. Chronic skin lesions typically lack the spongiosis but have moderate to marked hyperplasia of the epidermis, elongation of the rete ridges, and prominent hyperkeratosis. The number of mast cells and Langerhans cells are significantly increased in chronic lesions whereas eosinophils are reported to be infrequent. Capillary numbers are often increased and capillary walls appear thickened. Cutaneous nerves appear demyelinated and fibrotic at all levels of the dermis. Fibrosis of the upper dermis is a prominent finding in lichenified plaques.

IMMUNOPATHOGENESIS

In both acute and chronic lesions, a prominent lymphocytic infiltrate is seen. Immunohistochemical staining has demonstrated that the infiltrate is comprised predominantly of activated T cells bearing CD3, CD4 and CD45 RO antigens[6,7]. Essentially all T lymphocytes migrating into the skin bear the 'cutaneous lymphocyte antigen' (CLA), which functions as a skin-homing receptor for T cells[8]. Vascular endothelial cells in atopic dermatitis lesions express elevated levels of the receptor, VCAM-1[9], which plays an important role in targeting the homing of CLA^+ T cells to sites of skin inflammation[10]. VCAM-1, which is induced by interleukin (IL)-4 and IL-13[11], also plays an important role in the migration of eosinophils and mononuclear cells into sites of allergic inflammation. Increased numbers of Langerhans cells are found in chronic atopic dermatitis lesions and have been shown to have high-affinity [Fc_EI] and low-affinity [Fc_EII] IgE receptors and surface bound IgE molecules[12]. Mast cell numbers are also increased in chronic atopic dermatitis lesions. Typically mast cells bearing both tryptase and chymase (MC_{TC}) are predominant in normal skin, but in eczematous skin increased numbers of mast cells bearing just tryptase (MC_T—generally seen in the lung and gastrointestinal tract) are present in large numbers[13]. The development of MC_T cells is dependent upon functional T cells, suggesting that infiltrating T cells are responsible for their presence in the eczematous lesions. Keratinocytes also show evidence of cytokine-induced activation. ICAM-1 is expressed on keratinocytes from atopic dermatitis lesions[14], but in contrast to keratinocytes in contact dermatitis (classical delayed-type hypersensitivity (DTH)), HLA-DR is not expressed on the cell surface[15]. Although histologic studies suggested that the eosiniphil (the cell type typically seen at the site of chronic allergic inflammation) was not present in atopic dermatitis lesions, immunocytochemical studies have demonstrated extensive deposition of eosinophil-derived extracellular major basic protein (MBP) in atopic dermatitis lesions but not in normal-appearing skin, and control studies on patients with contact dermatitis revealed no dermal deposition of MBP[16]. More recently increased numbers of $EG2^+$-staining

Division of Allergy/Immunology, Johns Hopkins Hospital, CMSC-1103, 600 North Wolfe Street, Baltimore, MD 21287, USA

eosinophils have been demonstrated in chronic atopic dermatitis lesions, but not in acute lesions or normal-appearing skin[17]. In addition, eosinophil cationic protein (ECP) has been shown to be elevated in the sera of patients with atopic dermatitis and to correlate with disease severity, providing further evidence for eosinophil involvement in atopic dermatitis[18].

The patterns of cytokine expression found in lymphocytes infiltrating skin lesions of contact dermatitis and atopic dermatitis have been evaluated and compared utilizing *in situ* hybridization. In classic DTH reactions (e.g. tuberculin skin reaction), infiltrating T cells express predominantly the Th_1 cytokines, IL-2 and interferon-gamma (IFN-γ)[19], which in turn would account for the expression of HLA-DR antigens on local skin keratinocytes. On the other hand, infiltrating T cells in acute atopic dermatitis lesions express predominantly the Th_2 cytokines, IL-4, IL-5 and IL-13, whereas T cells in the chronic lesions express predominantly IL-5 IL-13[17,20]. The Th_2 profile of cytokines would promote chronic allergic inflammation by upregulating VCAM-1 on vascular endothelial cells, upregulating Fc_E receptors on Langerhans cells and other antigen-presenting cells, recruiting eosinophils and other inflammatory cells to the site, and promoting local synthesis of IgE antibodies. The delineation of the lymphocytic infiltrate associated with allergen IgE-mediated late-phase responses (i.e. T cells expressing IL-3, IL-4, IL-5 and GM-CSF but not IFN-γ)[21], and cloning of antigen-specific T cells (i.e. dust mite and grass pollen) from atopic dermatitis lesions adds further evidence of the pathogenic role of allergens in atopic dermatitis[22,23]. Furthermore, IgE-bearing Langerhans cells have been shown to be 100- to 1000-fold more efficient at presenting allergen (dust mite) to T cells, primarily Th_2 cells, and activating T cell proliferation[24,25]. Finally, it has been demonstrated that patients with IgE-mediated milk-induced skin symptoms possess allergen-specific, CLA^+ T cells in their circulation, which are not present in patients with milk-induced gastrointestinal disease[26]. This suggests that atopic dermatitis patients with milk-induced eczema possess T cells which facilitate the localization of the inflammatory response to the skin. In summary, various recent immunological studies indicate that allergen-specific IgE antibodies may function in several ways to initiate and perpetuate allergic inflammation in the skin.

FOOD ALLERGY AND THE PATHOGENESIS OF ATOPIC DERMATITIS

Whether food allergy is pathogenic in atopic dermatitis has been disputed for nearly a century[27]. Several lines of evidence support a pathogenic role for food hypersensitivity in atopic dermatitis: (1) studies of food allergen avoidance indicate that atopic dermatitis can be prevented in some infants; (2) food challenge studies provoke immunological changes that have been associated with the development of eczematous lesions; and (3) identification and elimination of food allergens leads to clinical improvement and return of abnormal immunologic parameters to normal. One study from the USA reported that approximately one-third of children seen in a university combined dermatology and allergy clinic had food hypersensitivity contributing to their skin symptoms[28], and a second study from France demonstrated that the more severe the atopic dermatitis, the more likely that food allergy is pathogenic[29]. As discussed below, a single isolated ingestion of a food allergen does not provoke an eczematous lesion, but repeated ingestions do result in typical changes of atopic dermatitis.

Since Grulee and Sanford reported that exclusive breast-feeding reduced atopic dermatitis sevenfold compared to cow's milk in a large cohort of infants[30], the effectiveness of breast-feeding and various measures to prevent food allergy and atopic disease has been debated. In a prospective study of 446 unselected preterm neonates, the protective effect of human breast milk or a preterm formula were compared[31]. Infants from atopic families receiving banked human breast milk had significantly less atopic dermatitis at 18 months than infants receiving formula; 16% versus 41%. In a prospective study of 1726 Danish infants, those exclusively breast-fed for more than one month had significantly less cow's milk allergy/intolerance than infants never breast-fed or breast-fed for less than one month; 1.7% versus 4.1%[32]. In two other studies, infants from atopic families whose mothers excluded egg, milk and fish from their diets during lactation (prophylaxis group) were found to have significantly less atopic dermatitis and food allergy at 18 months compared to infants whose mothers' diets were unrestricted[33,34]. At 4-year follow-up, the prophylaxis

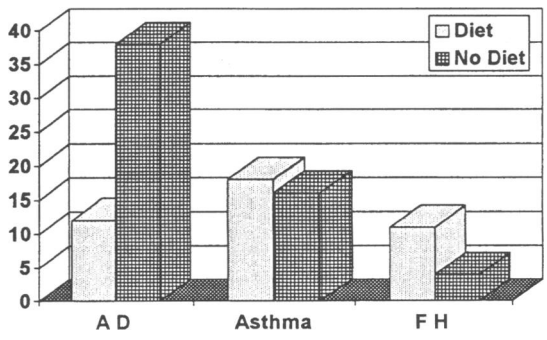

Figure 1 **The effect of eliminating egg, cow's milk and fish from the diets of breast-feeding mothers of infants from atopic families was evaluated for prevention of atopic disease. At 4-year follow-up, children in the prophylaxis group had less atopic dermatitis than similar control children where no dietary intervention was imposed. AD=atopic dermatitis; FH=food hypersensitivity** (Sigurs N, Hattevig G, Kjellman B. *Pediatrics*, 1992;**89**:735—see Ref 35)

group had less atopic dermatitis, as shown in Figure 1, but no difference in food allergy or respiratory allergy was seen[35]. In a prospective non-randomized study of 1265 unselected neonates, the effect of solid food introduction was evaluated over 10 years[36,37]. A significant linear relationship was found between the number of solid foods introduced into the diet by 4 months of age and subsequent atopic dermatitis, with a threefold increase in recurrent eczema at 10 years of age in infants receiving four or more solid foods compared to infants receiving no solid foods prior to 4 months of age. No such relationship was noted between asthma and the introduction of solid foods. A prospective non-randomized study comparing breast-fed infants who first received solid foods at 3 months or 6 months of age, revealed reduced atopic dermatitis and food allergy at 1 year of age in the group avoiding solids for the 6-month period[38], but no significant difference in these parameters at 5 years[39]. Finally, a prospective, randomized allergy prevention trial compared the benefits of maternal and infant food allergen avoidance in the prevention of allergic disease in infants at high risk for atopic disease[40–43]. Breast-feeding was encouraged in both prophylaxis and control groups. In the prophylaxis group, both the diets of lactating mothers and infants were restricted of egg, cow's milk and peanut. The control infants received cow's milk formula for supplementation. The period prevalences of food allergy, cow's milk sensitization and atopic dermatitis in the prophylaxis group were reduced significantly during the first 2 years compared to the control group, but were no longer significantly different beyond 2 years. The cumulative prevalence of food allergy remained lower in the prophylaxis group at 4 and 7-year follow-up, suggesting that the benefits of food allergy preventative measures are of limited duration, primarily because of the frequent remission of food allergy in early childhood[43]. Overall, it appears that food allergen avoidance diets for genetically predisposed infants have their greatest effect on the development of atopic dermatitis, but that this effect may be limited to those with milder disease who are likely to 'outgrow' it after the first few years.

My co-workers and I have studied 470 children and adolescents (median age 4.1 years, range 6 months to 25 years) with atopic dermatitis for evidence of food hypersensitivity. Patients were highly atopic with 80% having elevated total serum IgE concentrations (median 3410 IU/mL), 91% having positive family histories for atopic disease, and 39% having both asthma and allergic rhinitis at the time of initial evaluation (only 23% have atopic dermatitis alone at the initiation of study). Double-blind placebo-controlled food challenges (DBPCFC) provoked a variety of symptoms within minutes to 2 h in nearly 80% of the patients studied. Cutaneous reactions developed in 74% of positive challenges, but symptoms were confined to the skin alone in only 27% of positive responses. During the first evaluation, skin symptoms provoked by DBPCFC generally included a markedly pruritic, erythematous, morbilliform rash which developed in predilection sites for atopic dermatitis. Urticarial lesions were rarely seen. However, urticaria was seen often in follow-up challenges conducted 1 to 2 years later in patients who had adhered to an appropriate allergen elimination diet and had experienced clearing of their eczema, but who remained food sensitive. Although history had not suggested other food-induced complaints, gastrointestinal symptoms developed in 50% of patients (nausea, abdominal cramping, and vomiting and/or diarrhoea), upper respiratory symptoms, especially laryngeal oedema (sensation of itching and tightness in the throat, persistent throat clearing with dry hacking cough, and hoarseness) in 45%, and wheezing in about 15% of positive challenges.

Both immediate and late-phase effects of ingested food allergens were documented during DBPCFC[1,16,44–47]. The pruritic, erythematous morbilliform rash, which is a hallmark of the immediate skin response, generally arose abruptly and persisted for 30–150 min. Several laboratory findings implicated IgE-mediated cutaneous mast cell activation in the pathogenesis of these lesions: (1) a sharp rise in plasma histamine concentration in conjunction with positive DBPCFC (Figure 2); (2) no evidence of immediate activation of circulating basophils as evidenced by lack of change in circulating basophil number, total histamine content, or spontaneous histamine release *in vitro* in association with a positive food challenge; and (3) no significant change in the plasma complement activation products, C3a or C5a, in conjunction with positive food challenges. In addition, several lines of evidence indicated an IgE-mediated late-phase component associated with positive DBPCFC.

During the observation period, patients frequently developed diffuse pruritus and occasionally an erythematous

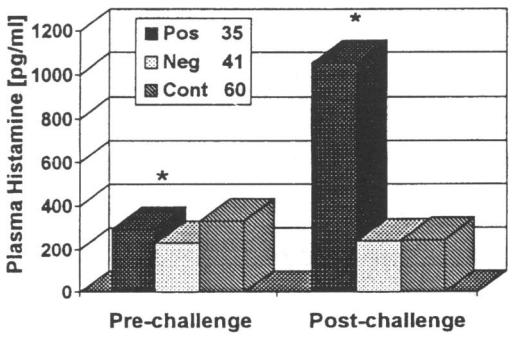

Figure 2 **Mean plasma histamine levels before and after DBPCFC. Pos=positive food challenge; Neg=negative food challenge; Cont=placebo food challenge** (Sampson *et al. N Engl J Med* 1984;**311**:372)

macular rash approximately 4–8 h following primary skin reaction. Peripheral blood eosinophils were found to decrease approximately 6–8 h following a positive food challenge but by 14–18 h post-challenge had returned to pre-challenge levels with a shift to increased numbers of hypodense (activated) eosinophils, as confirmed more recently by others[48]. In addition, serum ECP levels have been reported to rise following a positive food challenge[49]. Skin biopsies obtained at the site of a food challenge-induced morbilliform lesion 10–14 h following a positive challenge revealed eosinophil infiltration and deposition of MBP. Once recruited, eosinophils may release mediators (e.g. LTC_4), several cationic proteins (e.g. MBP, ECP, EDN) which contribute to the pathogenesis of the allergic reaction, and cytokines which may contribute to the inflammatory response (e.g. IL-1β, IL-6, TNF-α, MIP-1) or perpetuate chronic inflammation (e.g. IL-3, IL-5, GM-CSF)[1]. These and other mediators such as leukotrienes, prostaglandins and platelet-activating factor have been reported as prominent in atopic dermatitis and support a pathogenic role for IgE-mediated late phase reaction.

The continuous ingestion of food allergens by children with positive DBPCFC was found to be associated with high levels of 'spontaneous' basophil histamine release *in vitro* and the 'spontaneous' production of histamine-releasing factor (HRF) from mononuclear cells *in vitro*, and most likely *in vivo*[45,50]. The generation of HRF was associated with increased spontaneous basophil histamine release *in vitro*, increased basophil releasibility *in vitro*, and increased cutaneous hyperirritability (state of increased reactivity to a variety of minor non-specific stimuli, e.g. irritants, detergents, heat, cold, etc.). Increased numbers of 'activated' eosinophils (increased numbers of hypodense eosinophils, Figure 3) and basophils ('primed', responsive

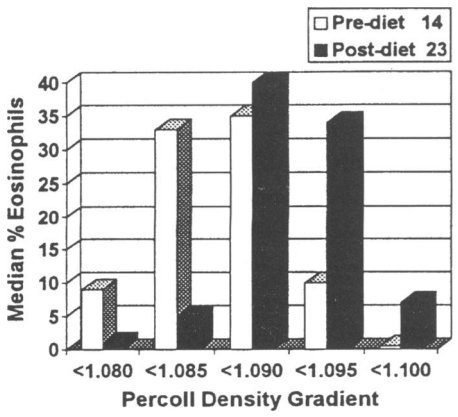

Figure 3 **Eosinophil density profiles in children with atopic dermatitis and food allergy before and after institution of appropriate food allergen elimination diets. 'Normodense' eosinophils (non-activated) are ⩾1.085**

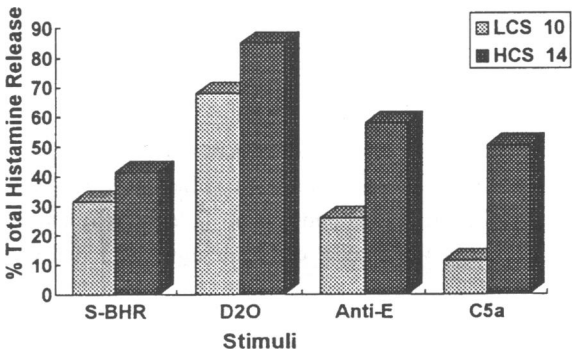

Figure 4 **Basophils from atopic dermatitis patients with high clinical scores (ingesting food allergens) were more releasable and 'activated' ('primed') than those from atopic dermatitis patients with low clinical scores (avoiding appropriate food allergens)** (James JM, Kagey-Sobotka A, Sampson HA. *J Allergy Clin Immunol* 1993;**91**:1155—see Ref 51)

to C5a *in vitro*, Figure 4) were found in food-allergic children with atopic dermatitis ingesting food allergens[51].

In order to better understand the role of the cutaneous mast cell in eczematous lesions, the response of cutaneous mast cells to contact with various allergens was assessed by introducing 'relevant' (food antigen to which the patient was skin test and oral challenge positive) and 'irrelevant' food allergens (food antigen to which patient was skin test positive and oral challenge negative) into skin blister chambers[47]. The chamber blister fluid was then monitored for changes in mediator and cell content. No differences in immediate and late-phase histamine or PGD_2 concentrations were seen when 'relevant' or 'irrelevant' food allergens were placed in the chamber. Since food antigens have been shown to enter the circulation of virtually anyone ingesting food, it was postulated that the difference between sensitized patients with or without clinical reactivity appears to depend upon factors intervening between gastrointestinal antigen absorption and target organ, e.g. modification of the antigen upon absorption, clearing by reticuloendothelial cells or circulating antibodies, etc.

With the institution of appropriate allergen elimination diets, clinical symptoms improved and many immunological indices became normal. Activated eosinophil numbers in the peripheral blood returned to normal (Figure 3), as did basophil activation status (Figure 4)[51]. Spontaneous generation of HRF decreased to background levels over 6–9 months when the food allergens were identified and removed from the diet. In addition, spontaneous basophil histamine release returned to background levels, basophil releasibility became normal, and cutaneous hyperirritability diminished.

To better understand the target organ specificity of food-induced eczematous lesions, peripheral blood mononuclear cells (PBMCs) from seven patients with atopic

dermatitis and milk allergy, 10 with milk-induced gastroenteropathies, and eight normal controls were stimulated *in vitro* with casein and then evaluated for expression of homing receptors[26]. Only patients with atopic dermatitis and milk allergy demonstrated a significant increase in CLA^+ T cells, indicating that the homing-receptor expression on antigen-specific T cells may play a role in determining tissues of involvement in allergic responses.

In a prospective follow-up study of 34 patients with atopic dermatitis depicted in Figure 5, 17 children with food allergy (group 1) who were appropriately diagnosed and placed on an allergen elimination diet experienced a marked and significantly greater improvement in their eczematous rash at 1–2 year and 3–4 year follow-up than 12 similar patients who did not have food allergy (group 2) and 5 children who were food allergic but did not adhere to the elimination diet (group 3)[46]. Only children in group 1 had a significant fall in their serum IgE concentrations over the 3–4 year follow-up (serum IgE concentration correlates roughly with disease severity).

Changes associated with the allergen elimination diet were not confined to the skin in food-allergic patients. Lactulose absorption studies (a measure of gastrointestinal permeability) were abnormal in food-allergic children with atopic dermatitis while ingesting food allergens, but subsequently became normal when the responsible food allergens were eliminated from the diet[52]. In addition, these children were frequently noted to develop improved appetites and experienced some 'catch-up' weight gain. This study indicated that patients with atopic dermatitis and food hypersensitivity had subclinical malabsorption which was reversed when food allergens were removed from the diet.

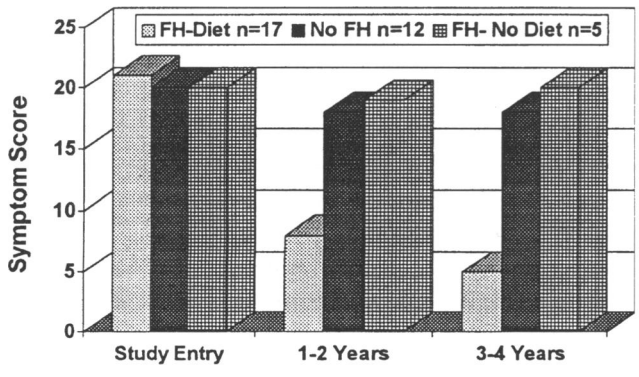

Figure 5 **17 children with atopic dermatitis and food allergy placed on appropriate food-allergen elimination diets (FH-diet) experienced significant improvement in their eczema over the 4-year follow-up compared to 12 children with atopic dermatitis and no food allergy (no FH) and 5 children with atopic dermatitis and food allergy who did not comply with the allergen elimination diet (No diet)**

Five foods were found to account for nearly 90% of the food-induced reactions seen: egg, milk, peanut, soy, and wheat. Egg allergy was the most prevalent, affecting nearly two-thirds of the children with food hypersensitivity. In our highly selected, tertiary referral population, 20% of the children were not allergic to any foods, but 29% were allergic to one food, 23% to two foods, 13% to three foods, 8% to four foods, and 7% to five or more food antigens. Clinical food hypersensitivity was shown to be highly specific. Although specific IgE antibodies were often present to many members of plant and animal families, patients generally reacted only to one or two members of that food family when ingested. For example, significant *in vitro* cross-reactivity was seen among the legume family by prick skin tests, RAST and immunoblots, but little *clinical* cross-reactivity was observed when food antigens were actually ingested[53,54]. Likewise studies with cereal grains showed significant IgE antibody cross-reactivity between grains and grass pollens but little clinical cross-reactivity[55]. Therefore, in order to provide the most nutritionally complete allergen-free diets, DBPCFC or controlled elimination diets should be recommended.

75 patients were rechallenged 1–2 years after completely eliminating the responsible food allergen from their diet. Approximately one-third of symptomatic food allergies had resolved by the time of the second DBPCFC[46]. The likelihood of developing clinical tolerance was dependent upon the food to which the child was reactive; e.g. the development of tolerance to soy was common whereas development of tolerance to peanut was extremely rare. Results of prick skin tests are of little value in predicting loss of clinical reactivity since most remained virtually unchanged for years after the child could tolerate a specific food.

Other investigators utilizing similar food challenge regimens also have demonstrated the pathogenic link between food hypersensitivity and atopic dermatitis. In their studies of children primarily with suspected food hypersensitivity and respiratory allergy, May and Bock reported that 4 of 7 children with a history of eczematous reactions to foods developed rashes within 2 h of administration of a double-blind placebo-controlled oral food challenge[56]. Burks and co-workers have also employed the double-blind placebo-controlled oral food challenge to study 46 children with mild to severe atopic dermatitis presenting to a university dermatology and allergy clinic[28]. As seen in other controlled oral challenge studies, children experienced cutaneous, respiratory and gastrointestinal symptoms. One-third of the children reportedly developed symptoms during the blinded food challenges. Interestingly, no correlation was seen between the likelihood of having a positive food challenge and the severity of the skin symptoms.

CONCLUSION

In summary, IgE antibodies bind high-affinity FcE_I receptors on mast cells, basophils, Langerhans cells and monocytes of atopic individuals as well as low affinity FcE_{II} receptors on antigen-presenting cells (macrophages, monocytes, Langerhans cells, dendritic cells), lymphocytes, eosinophils and platelets. When food allergens penetrate mucosal barriers and reach IgE antibodies bound to mast cells, mediators such as histamine, prostaglandins and leukotrienes are released. The activated mast cells also release cytokines (e.g. IL-4, IL-5, IL-6, TNF-α, PAF) that may promote further inflammation. During the initial 4–8 h of the reaction, primarily eosinophils invade the site of response. These infiltrating cells are activated and release mediators including PAF, eosinophil MBP, eosinophil cationic protein (ECP) and cytokines. In the subsequent 24–48 h, lymphocytes and monocytes infiltrate the area and establish the more chronic inflammatory picture. Proper identification of responsible food allergens and their complete elimination from the diet can result in a reversal of immunopathogenic mechanisms and improvement in the patient's skin condition.

REFERENCES

1 Sampson HA. Atopic dermatitis. *Ann Allergy* 1992;**69**:469–79

2 Hill LW, Sulzberger MG. Evaluation of atopic dermatitis. *Arch Dermatol Syph* 1935;**32**:451–63

3 Hanifin JM, Rajka G. Diagnostic features of atopic dermatitis. *Arch Dermatol Venereol* 1980;**92**:44–7

4 Hanifin JM. Epidemiology of atopic dermatitis. *Monogr Allergy* 1987;**21**:116–31

5 Tong AKF, Mihm MC. The pathology of atopic dermatitis. *Clin Rev Allergy* 1986;**4**:27–42

6 Leung DYM, Bhan AK, Schneeberger EE, *et al.* Characterization of mononuclear cell infiltrate in atopic dermatitis using monoclonal antibodies. *J Allergy Clin Immunol* 1983;**71**:47–56

7 Boss JD, Hagenara C, Das PK, *et al.* Predominance of 'memory' T cells (CD4+, CDw29+) over 'naive' T cells (CD4+, CD45R+) in both normal and diseased human skin. *Arch Dermatol Res* 1989;**81**:24–30

8 Picker LJ, Martin RJ, Trumble A, Newman LS, Collins PA, Bergstresser PR, *et al.* Differential expression of lymphocyte homing receptors by human memory/effector T cells in pulmonary versus cutaneous immune effector sites. *Eur J Immunol* 1994;**24**:1269–77

9 Wakita H, Sakamoto T, Tokura Y, Takigawa M. E-selectin and vascular cell adhesion molecule-1 as critical adhesion molecules for infiltration of T lymphocytes and eosinophils in atopic dermatitis. *J Cutan Pathol* 1994;**21**:33–9

10 Rossiter H, Van Reijsen F, Mudde G. Skin disease-related T cells bind to endothelial selectins: expression of cutaneous lymphocyte antigen (CLA) predicts E-selectin but not P-selectin binding. *Eur J Immunol* 1994;**24**:205–10

11 Bochner BS, Klunk DA, Sterbinsky SA, *et al.* IL-13 selectively induces vascular cell adhesion molecule-1 expression in human endothelial cells. *J Immunol* 1995;**154**:799–803

12 Bruynzeel-Koomen C, van Wichen DF, Toonstra J, *et al.* The presence of IgE molecules on epidermal Langerhans cells in patients with atopic dermatitis. *Arch Derm Res* 1986;**287**:199–205

13 Irani AM, Sampson HA, Schwartz LB. Mast cells in atopic dermatitis. *Allergy* 1989;**44**(suppl 9):31–4

14 Singer KH, Tuck DT, Sampson HA, *et al.* Epidermal keratinocytes express the adhesion molecule intercellular adhesion molecule-1 in inflammatory dermatoses. *J Invest Dermatol* 1989;**92**:746–50

15 Barker J, MacDonald D. Epidermal class II human lymphocyte antigen expression in atopic dermatitis: a comparison with experimental allergic contact dermatitis. *J Am Acad Dermatol* 1987;**16**:1175–9

16 Leiferman K, Ackerman S, Sampson H, Haugen H, Venencie P, Gleich G. Dermal deposition of eosinophil-granule major basic protein in atopic dermatitis. Comparison with onchocerciasis. *N Engl J Med* 1985;**313**:282–5

17 Hamid Q, Boguniewicz M, Leung DYM. Differential *in situ* cytokine gene expression in acute versus chronic atopic dermatitis. *J Clin Invest* 1994;**94**:870–6

18 Kagi MK, Joller-Jemelka H, Wuthrich B. Correlation of eosinophils, eosinophil cationic protein, and soluble interleukin-2 receptor with the clinical activity of atopic dermatitis. *Dermatology* 1992;**185**:88–92

19 Tsicopoulos A, Hamid Q, Varney V, Ying S, Moqbel R, Durham S, *et al.* Preferential messenger RNA expression of Th-1-type cells [IFN-gamma, IL-2] in classical delayed-type (tuberculin) hypersensitivity reactions in human skin. *J Immunol* 1992;**148**:2058–61

20 Hamid Q, Naseer T, Minshall EM, Song YL, Boguniewicz M, Leung DYM. *In vivo* expression of IL-12 and IL-13 in atopic dermatitis. *J Allergy Clin Immunol* 1996;**98**:225–31

21 Kay AB, Ying S, Varney V, *et al.* Messenger RNA expression of cytokine gene cluster, interleukin 3 (IL-3), IL-5 and granulocyte/macrophage colony-stimulating factor, in allergen-induced late-phase cutaneous reactions in atopic subjects. *J Expl Med* 1991;**173**:775–8

22 Van der Heijden F, Wirenga EA, Bos JD, *et al.* High frequency of IL-4 producing CD4+ allergen-specific T lymphocytes in atopic dermatitis lesional skin. *J Invest Dermatol* 1991;**97**:389–94

23 Van Reijsen FC, Bruynzeel-Koomen CA, Kalthoff FS, *et al.* Skin-derived aeroallergen-specific T cell clones of the Th_2 phenotype in patients with atopic dermatitis. *J Allergy Clin Immunol* 1992;**90**:184–93

24 Mudde G, van Reijsen F, Boland G, de Gast G, Bruijnzeel P, Bruijnzeel-Koomen C. Allergen presentation by epidermal Langerhans cells from patients with atopic dermatitis is mediated by IgE. *Immunology* 1990;**69**:335–41

25 Mudde G, Bheekha R, Bruijnzeel-Koomen C. Consequences of IgE/CD23-mediated antigen presentation in allergy. *Immunol Today* 1995;**16**:380–3

26 Abernathy-Carver K, Sampson H, Picker L, Leung D. Milk-induced eczema is associated with the expansion of T cells expressing cutaneous lymphocyte antigen. *J Clin Invest* 1995;**95**:913–18

27 Schloss OM. Allergy to common foods. *Trans Am Pediatr Soc* 1915;**27**:62–8

28 Burks AW, Mallory SB, Williams LW, Shirrell MA. Atopic dermatitis: clinical relevance of food hypersensitivity reactions. *J Pediatr* 1988;**113**:447–51

29 Guillet G, Guillet MH. Natural history of sensitizations in atopic dermatitis. *Arch Dermatol* 1992;**128**:187–92

30 Grulee CG, Sanford HN. The influence of breast-feeding and artificial feeding in infantile eczema. *J Pediatr* 1936;**9**:223–5

31 Lucas A, Brooke O, Morley R, Cole T, Bamford M. Early diet of preterm infants and development of allergic or atopic disease: randomized prospective study. *BMJ* 1990;**300**:837–40

32 Host A, Husby S, Osterballe O. A prospective study of cow's milk allergy in exclusively breast-fed infants. Incidence, pathogenetic role of early inadvertent exposure to cow's milk formula, and characterization of bovine milk protein in human milk. *Acta Paediatr Scand* 1988;**77**:663–70

33 Chandra R, Shakuntla P, Hamed A. Influence of maternal diet during lactation and use of formula feeds on development of atopic eczema in high risk infants. *BMJ* 1989;**299**:228–30

34 Hattevig G, Kjellman B, Bjorksten B, Kjellman N. Effect of maternal avoidance of eggs, cow's milk and fish during lactation upon allergic manifestations in infants. *Clin Expl Allergy* 1989;**19**:27–32

35 Sigurs N, Hattevig G, Kjellman B. Maternal avoidance of eggs, cow's milk and fish during lactation: effect on allergic manifestation, skin prick tests, and specific IgE antibodies in children at age 4 years. *Pediatrics* 1992;**89**:735–9

36 Fergusson D, Horwood L, Shannon F. Asthma and infant diet. *Arch Dis Child* 1983;**58**:48–51

37 Fergusson DM, Horwood LJ, Shannon FT. Early solid feeding and recurrent eczema: a 10-year longitudinal study. *Pediatrics* 1990;**86**: 541–6

38 Kajosaari M, Saarinen UM. Prophylaxis of atopic disease by six months; total solid food elimination. *Arch Paediatr Scand* 1983;**72**:411–14

39 Kajosaari M. Atopy prophylaxis in high-risk infants: prospective 5-year follow-up study of children with six months exclusive breastfeeding and solid food elimination. *Adv Exp Med Biol* 1991;**310**:453–8

40 Zeiger R, Heller S, Mellon M, Forsythe A, O'Connor R, Hamburger R. Effect of combined maternal and infant food-allergen avoidance on development of atopy in early infancy: a randomized study. *J Allergy Clin Immunol* 1989;**84**:72–89

41 Zeiger R. Prevention of food allergy in infancy. *Ann Allergy* 1990;**65**:430–41

42 Zeiger R, Heller S, Mellon M, Halsey J, Hamburger R, Sampson H. Genetic and environmental factors affecting the development of atopy through age 4 in children of atopic parents: a prospective randomized study of food allergen avoidance. *Pediatr Allergy Immunol* 1992;**3**:110–27

43 Zeiger R, Heller S. The development and prediction of atopy in high-risk children: follow-up at seven years in a prospective randomized study of combined maternal and infant food allergen avoidance. *J Allergy Clin Immunol* 1995;**95**:1179–90

44 Sampson HA, McCaskill CC. Food hypersensitivity and atopic dermatitis: evaluation of 113 patients. *J Pediatr* 1985;**107**:669–75

45 Sampson HA, Broadbent KR, Bernhisel-Broadbent J. Spontaneous release of histamine from basophils and histamine-releasing factor in patients with atopic dermatitis and food hypersensitivity. *N Engl J Med* 1989;**321**:228–32

46 Sampson HA, Scanlon SM. Natural history of food hypersensitivity in children with atopic dermatitis. *J Pediatr* 1989;**115**:23–7

47 Charlesworth EN, Kagey-Sobotka A, Norman PS, Lichtenstein LM, Sampson HA. Cutaneous late-phase response in food-allergic children and adolescents with atopic dermatitis. *Clin Exp Allergy* 1993;**23**:391–7

48 Magnarin M, Knowles A, Ventura A, Vita F, Fanti L, Zabucchi G. A role for eosinophils in the pathogenesis of skin lesions in patients with food-sensitive atopic dermatitis. *J Allergy Clin Immunol* 1995;**96**:200–8

49 Suomalainen H, Soppi E, Isolauri E. Evidence for eosinophil activation in cow's milk allergy. *Pediatr Allergy Immunol* 1994;**5**:27–31

50 Sampson HA, MacDonald SM. IgE-dependent histamine-releasing factors. *Springer Semin Immunopathol* 1993;**15**:89–98

51 James JM, Kagey-Sobotka A, Sampson HA. Patients with severe atopic dermatitis have activated circulating basophils. *J Allergy Clin Immunol* 1993;**91**:1155–62

52 Flick J, Sampson H, Perman J. Intestinal permeability to carbohydrates in children with atopic dermatitis and food hypersensitivity. *Pediatr Res* 1988;**23**:303A

53 Bernhisel-Broadbent J, Sampson HA. Cross-allergenicity in the legume botanical family in children with food hypersensitivity. *J Allergy Clin Immunol* 1989;**83**:435–40

54 Bernhisel-Broadbent J, Strause D, Sampson HA. Fish hypersensitivity. II: Clinical relevance of altered fish allergenicity caused by various preparation methods. *J Allergy Clin Immunol* 1992;**90**:622–9

55 Jones SM, Magnolfi CF, Cooke SK, Sampson HA. Immunologic cross-reactivity among cereal grains and grasses in children with food hypersensitivity. *J Allergy Clin Immunol* 1995;**96**:341–51

56 Bock S, Lee W, Remigio L, May C. Studies of hypersensitivity reactions to food in infants and children. *J Allergy Clin Immunol* 1978;**62**:3327–34