

Review Article

Allergic contact dermatitis: the cellular effectors

IAN KIMBER AND REBECCA J. DEARMAN

Syngenta Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire SK10 4TJ, UK

Contact hypersensitivity reactions are mediated by lymphocytic effector cells. Until recently it was believed that the most important of these were CD4⁺ T lymphocytes. However, there is growing evidence that in many instances the predominant effector cell may be a CD8⁺ T lymphocyte, with in some instances CD4⁺ cells performing a counter-regulatory function. Here we review the roles of CD4⁺ T helper (Th) cells and CD8⁺ T cytotoxic (Tc) cells, and their main functional subpopulations (respectively, Th1 and Th2 cells and Tc1 and Tc2 cells) in the elicitation of contact hypersensitivity reactions and consider the implications of effector cell selectivity for the biology of allergic contact dermatitis.

Key words: CD4⁺ T lymphocytes; CD8⁺ T lymphocytes; contact hypersensitivity; effector cells.
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Skin sensitization resulting in allergic contact dermatitis is dependent upon the initiation of specific T lymphocyte responses. Such responses are provoked initially in lymph nodes draining the site of exposure to a skin sensitizing chemical. Briefly, epidermal Langerhans cells (LC), and possibly other cutaneous dendritic cells (DC), transport allergen from the skin, via afferent lymphatics, to regional lymph nodes where it is presented to responsive T lymphocytes (1, 2). The activation and clonal expansion of allergen-reactive T lymphocytes is the pivotal event in the acquisition of skin sensitization. Following subsequent exposure of the now-sensitized subject to the same chemical allergen, an accelerated and more aggressive secondary immune response will be elicited. This is characterized by the infiltration of effector T lymphocytes into challenged skin sites and the development of a cutaneous inflammatory reaction (3–5). The purpose of this article is to consider the nature of T lymphocytes that effect contact allergic reactions.

For the last 15 years the conceptual basis for cellular immunity has been dominated by the recognition that there exist functional subpopulations of T lymphocytes that serve to determine, in large part, qualitative aspects of adaptive immune responses. First characterized were subsets of CD4⁺ T helper (Th) cells, the most polarized forms of which (designated Th1 and Th2 cells) have been shown to differ with respect to cytokine secretion patterns (6, 7). More recently, a similar heterogeneity among CD8⁺ T cytotoxic (Tc) cells has been

recognized also, with the identification of Tc1 and Tc2 subpopulations (8, 9).

CD4⁺ effector cells

Until recently, the prevailing view was that allergic contact dermatitis, as one form of delayed type hypersensitivity (DTH) reaction, was effected by CD4⁺ Th1-type cells. This view was based on an assumption of a conformity of mechanism among DTH reactions and several experimental observations. Thus, for example, nickel (Ni)-specific CD4⁺ Th1 type cells were identified in the blood and skin of subjects sensitized to nickel (10, 11). Moreover, some investigations in mice found contact hypersensitivity (CHS) to be associated with CD4⁺ T lymphocyte function, and to be compromised when such cells were deleted (12–15). However, in some of these studies it is not always apparent whether CD4⁺ T lymphocytes were necessarily acting as effector cells *per se*, or if the data reflect the fact such cells may be required for the development of other effector populations, such as CD8⁺ T lymphocytes (16). There is increasing interest in the role of these latter cells in allergic contact dermatitis.

Although there is no doubt that CD4⁺ T lymphocytes are able to elicit standard DTH reactions (where conventionally responses are directed at complex protein or cellular antigens, and in which antigenic challenge is performed by intradermal or subcutaneous injection) (17–19), there is

a growing appreciation that DTH and CHS have differential requirements for cellular effectors and that for the elicitation of CHS it is CD8⁺ cells that may play the most important roles (20). A seminal study reported by Gocinski & Tigelaar (21) used monoclonal antibody treatment to deplete selectively either CD4⁺ or CD8⁺ T lymphocytes from mice. The conclusion drawn was that DTH reactions are mediated by CD4⁺ T cells and down-regulated by CD8⁺ cells, whereas the reverse is the case for CHS, although some CD4⁺ lymphocytes were believed to act as CHS effectors also (21). Since then other evidence has emerged to support a pivotal role for CD8⁺ T lymphocytes in the elicitation of contact allergic reactions.

CD8⁺ effector cells and interplay with CD4⁺ cells

In nickel-allergic patients, Ni-specific CD8⁺ can be found (22), and indeed it has been reported that these are the primary effector cells (23). Thus, it was demonstrated that whereas CD4⁺ T lymphocytes from both Ni-allergic and nonallergic subjects would respond to NiSO₄ *in vitro*, Ni-specific CD8⁺ cell responses were observed only with lymphocytes derived from allergic donors (23). These and other studies have suggested that in allergy to nickel at least there are important regulatory CD4⁺ cells that control the production of Th1- and Tc1-type effector cells secondary to production of high levels of the cytokine interleukin (IL) 10 (23, 24). A general model in which CD8⁺ T lymphocytes act as effectors, while allergen-specific CD4⁺ cells perform regulatory functions, is supported by various investigations using mice. For instance, based on an understanding of differential major histocompatibility complex (MHC) class restriction of CD4⁺ and CD8⁺ T lymphocytes, it was shown that mice lacking MHC class I were unable to support CHS responses to the contact allergen dinitrofluorobenzene (DNFB), whereas MHC class II-deficient mice exhibited an exaggerated CHS reaction (25); a reaction shown subsequently to be mediated by CD8⁺ T lymphocytes (26). Consistent with these data are the results of other investigations in which it was found that MHC class I⁺/II⁻ DC were able to prime for contact hypersensitivity, but MHC class I⁻/II⁺ DC were not (27, 28). On the basis of cytokine secretion patterns it is assumed that the CD8⁺ effector T lymphocytes identified in such studies are of a Tc1 phenotype (secreting relatively high levels of the type 1 cytokine interferon (IFN) γ), while the counter-regulatory CD4⁺ lymphocytes are Th2-like (producing comparatively high levels of IL-4 and IL-10 (29)). Although this rather polarized model is attractive, there may in practice be a re-

quirement for both CD4⁺ and CD8⁺ T lymphocytes for the initiation and maintenance of a complete immune response to skin sensitizing chemicals. Certainly this is the implication from the results of recent studies performed in mice by Wang et al. (30). On the basis of secondary depletion experiments from CD4 and CD8 knockout animals, they concluded both CD4⁺ Th1 cells and CD8⁺ Tc1 cells are required for the full development of contact hypersensitivity (30). This is despite the fact that in some cases it appears possible to generate CD8⁺ T lymphocyte responses to skin sensitizing chemicals in the absence of antigen-activated CD4⁺ cells (28, 31). In this context, an investigation of CD4⁺/CD8⁺ selectivity for contact hypersensitivity conducted in immunologically intact mice is instructive (32). Trinitrophenyl (TNP)-derivatized peptides that have affinity for either class I or class II MHC molecules were used. Subcutaneous injection of mice with the class II-binding TNP-peptide (but not with the class I-binding conjugate) in an adjuvant induced low level, but nonetheless discernible, contact hypersensitivity. The implication is that CD4⁺ effector cells mediated the response. In contrast, however, when the same peptides were administered to mice in the context of DC, then only class I-binding TNP-peptides induced contact hypersensitivity, and of a magnitude comparable with that observed following conventional topical sensitization (32). The important conclusions that can be drawn from these experiments are that CD8⁺ may be potent effectors of contact hypersensitivity, but that CD4⁺ may be active also, and the way in which the inducing antigen is delivered to the immune system (presumably related to the mechanism of MHC loading) can have an important influence on whether CD8⁺ or CD4⁺ T lymphocytes predominate as effector cells. On this basis, one may speculate that the ability of chemical haptens to drive CD8⁺ T lymphocyte activation may be associated with their inherent or inducible capacity to interact directly with peptides bound within MHC determinants to create immunogenic trimolecular complexes (33).

Alternatively, or additionally, the apparent dominance of CD8⁺ effector cells in at least some forms of CHS may be associated with their cytotoxic activity.

Effector cell cytotoxic function

The primary pathophysiological role of CD8⁺ T lymphocytes is the lysis of virally infected cells secondary to recognition of virus-encoded peptides in the context of MHC class I determinants. The question is to what extent cytotoxic mechanisms

are required for CHS reactions effected by CD8⁺ cells, or other cellular effectors. The need for cytotoxic activity was investigated in a mouse model of CHS to DNFB in which the predominant or exclusive effectors were found to be CD8⁺ cells (34). Mice lacking perforin and/or Fas/Fas ligand pathways necessary for T cell-mediated cytotoxicity were compared with wild-type animals. It was found that although mice lacking both pathways were able to generate hapten-specific CD8⁺ T lymphocytes, they were unable to mount CHS reactions (34). Similar conclusions regarding the importance of Fas-induced cell death in the pathogenesis of allergic contact dermatitis and other cutaneous inflammatory reactions derived from studies of human skin (35). It is important to acknowledge, however, that cells other than CD8⁺ Tc1-type lymphocytes have the ability to induce cytotoxicity. In a study of human nickel allergy it was shown that Tc1, Tc2 and Th1 cells all had the inherent or inducible ability to induce keratinocyte cytotoxicity (36). The availability of cytokines, and in particular of IFN- γ , is important in determining the breadth and vigour of cytotoxic responses. Recognition by Th1 effector cells of keratinocytes will require the IFN- γ -mediated induction of MHC class II determinants. Furthermore, IFN- γ is known to up-regulate the expression of the Fas receptor and this will increase further susceptibility to cell death. It is hypothesized therefore that where a variety of cytotoxic effector cells are available, the lesion will be initiated by CD8⁺ cells, with the reaction becoming more vigorous, and CD4⁺ effectors playing a role, when IFN- γ has been produced and has rendered keratinocytes more susceptible.

Implications of CD8⁺ effector cells

It may be an accident of hapten encounter, processing and presentation that predisposes toward the initiation of CD8⁺ lymphocyte responses in skin sensitization, but it is nevertheless interesting to consider what implications a predominance of CD8⁺ cells may have for the pathogenesis and natural history of allergic contact dermatitis. Clearly, in the light of the investigations summarized above, the conjecture is that the initiation of damage in the skin will be facilitated by the arrival of specific cytotoxic CD8⁺ cells. However, there is increasing evidence that CD4⁺ and CD8⁺ T cell immunity is regulated differentially, particularly with respect to clonal expansion and the duration of immunological memory (37–39). In antiviral immune responses at least, it has recently been demonstrated that CD8⁺ cells expanded more vigorously in response to antigen than did CD4⁺

cells, and that CD8⁺ memory was more stably maintained and more persistent than CD4⁺ memory (39). Although it may not be justified to do so, if the differential responses displayed by CD4⁺ and CD8⁺ cells in antiviral immunity are extrapolated to contact allergy then the argument would be that predominant CD8⁺ lymphocyte responses would be consistent with persistent or life-long sensitization.

Diversity of effector cell phenotypes: experimental evidence

Finally, it is relevant to consider briefly experimental evidence for the roles of functional subpopulations of CD4⁺ and CD8⁺ lymphocytes in skin sensitization and allergic contact dermatitis. In this laboratory we examined the phenotype of cytokine-secreting draining lymph node cells (LNC) following repeated topical exposure of mice to the contact allergens formaldehyde and dinitrochlorobenzene (DNCB). With both allergens LNC produced high levels of the type 1 cytokine IFN- γ , but only low levels of the type 2 cytokines IL-4 and IL-10. In both instances it was found that the type 2 cytokines were produced by Th2-type CD4⁺ lymphocytes. There was no evidence for induction of Tc2 cells. However, IFN- γ was produced in formaldehyde-sensitized mice by CD4⁺ Th1 cells and in DNCB-sensitized mice by both CD4⁺ Th1 cells and CD8⁺ Tc1 cells (40). These data illustrate that in response to some skin sensitizing chemicals, both CD4⁺ and CD8⁺ cell responses are induced, but that this is not necessarily a universal rule. Somewhat similar results were reported subsequently by Wang et al. (30), who found that in mice sensitized with DNFB both CD4⁺ and CD8⁺ cells produced IFN- γ . In such instances it is assumed that both CD4⁺ Th1 cells and CD8⁺ Tc1 cells contribute to CHS (in the case of DNCB and DNFB) or that (in the case of formaldehyde) the predominant effectors are CD4⁺ Th1 cells. However, it would be inappropriate to conclude that cells of other phenotypes are unable to effect CHS reactions. Recent studies performed in mice with fluorescein isothiocyanate (FITC) illustrate the point. This chemical provokes a selective type 2 T lymphocyte response. Notwithstanding this, FITC does in mice induce delayed-type cutaneous hypersensitivity reactions. Our investigations demonstrated that such reactions were effected by CD4⁺ T lymphocytes with a Th2-type phenotype (41). Taken together with other evidence cited above, it is clear that the major functional subsets of both CD4⁺ and CD8⁺ T lymphocytes are in principle able to induce or participate in CHS reactions.

Concluding comments

In conclusion, the available evidence indicates that a variety of cells may contribute to the development of allergic contact dermatitis. Certainly, it is now apparent that in many instances the most important effectors are CD8⁺ lymphocytes with a Tc1-type phenotype. However, this is not a universal rule and in practice other cells may participate, or even in some circumstances take a leading role. The intriguing possibility is that the nature of skin sensitization responses and the mechanisms through which CHS reactions are effected are dictated at least in part by the nature of the inducing chemical and/or the way in which the chemical hapten is encountered by the immune system. It will be interesting and informative to determine to what extent T lymphocyte responses and effector mechanisms vary between chemical allergens and between sensitized subjects, and whether such variations have an influence on the relative potency of skin sensitizers, or the course and severity of allergic contact dermatitis.

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Address:

Ian Kimber
Syngenta Central Toxicology Laboratory
Alderley Park
Macclesfield
Cheshire, SK10 4TJ
UK
Tel: +44 162 5515408
Fax: +44 162 5590249
e-mail: ian.kimber@syngenta.com