

OPINIONS IN ALLERGY

The Hapten–Atopy hypothesis II: the 'cutaneous hapten paradox'

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Summary

One explanation for the striking increase in atopic disease in developed countries over the last 50 years has been the 'Hygiene Hypothesis'; a reduced exposure to pathogenic microorganisms. We have postulated previously that oral and cutaneous exposure to chemicals generally and to haptens in particular, may have also contributed to the increased prevalence of atopic disease; the 'Hapten–Atopy Hypothesis'. The purpose here is to extend further that hypothesis by consideration of the impact interplay between the innate and adaptive immune systems may have on the development of atopic allergy. It is clear that experimental cutaneous exposure to hapten can generate immune responses of different types with regard to T-helper (Th) cell phenotype. Allergic contact dermatitis is frequently associated with a selective Th1 (and Tc1)-driven inflammation, whereas atopic dermatitis is characterized by preferential Th2 cell responses. We postulate here that initial innate immune responses to chemical haptens result in the promotion of Th1 cell responses secondary to stimulation of Toll-like receptor. However, we argue also that under conditions where there is prolonged skin exposure to hapten there will be a shift of Th cell phenotype to selective Th2-type responses. The significance of such interactions is the possibility that repeated low-level skin exposure to certain types of hapten may result in the creation of an immunological environment in which the development of Th2 immune responses to third party antigens is favoured. The hypothesis is advanced that the nature and conditions of skin exposure to common haptens may impact on the quality of cutaneous immune responses such that in some circumstances the development atopic disease is favoured.

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Introduction

The prevalence of atopic disease has risen by 400% over the last 50 years. One proposed basis for this is the 'Hygiene Hypothesis', which argues that increased atopy is associated with a decreased exposure to pathogenic microorganisms in early life [1]. Stimulation (or lack of stimulation) of the innate immune system through Toll-like receptors (TLRs) by microbial agents such as lipopolysaccharide (LPS) has recently been studied in relation to this. We have postulated that increased exposure to chemical allergens ('haptens') via oral encounter with chemicals in food and drugs [2], and *inter alia*, consumer care products through skin, have also contributed to the increased prevalence of atopy over the last 50 years (the 'Hapten–Atopy Hypothesis'). Haptens are usually low molecular weight electrophilic (or potentially lipophilic) chemicals that bind to peptides/proteins, the altered configuration becoming potentially allergenic. They are

encountered most frequently in the context of cutaneous disease as contact allergens, although there are some chemicals, fewer in number, that cause sensitization of the respiratory tract [3]. These agents also have the ability to induce danger signals and interact with the cutaneous innate immune system [4, 5]. Our previous article on the 'hapten-atopy hypothesis', while postulating that both oral and cutaneous exposure to haptens may result in predisposition to atopy and atopic disease [2], concentrated specifically on the effect of oral exposure to haptens. In this article we focus on the potential effect of repeated cutaneous exposure to haptens in the pathogenesis of atopic disease.

Cutaneous exposure of experimental animals to hapten/contact allergens over a relatively short time period of time (days) leads selectively to allergic contact dermatitis (ACD) a form of delayed-type hypersensitivity generally considered to be a T-helper type 1 (Th1)/Tc1-driven condition. In contrast, topical exposure to chemical

respiratory sensitizers, such as trimellitic anhydride, over the same time frame results in preferential Th2 cell activation [6]. Paradoxically, many mouse models of atopic dermatitis (AD) are based on repeated, prolonged cutaneous exposure to the same haptens that initially provoke a Th1/Tc1 profile [7].

Experimental (murine) models of hapten exposure differ markedly in exposure regimen, doses, sensitizing hapten and outcome. For example, even using the same hapten, oxazolone, a single dose can sensitize such that subsequent challenge within 7 days causes an ACD-like reaction, whereas 10 repeated challenges over 22–25 days has been reported to cause an AD-like reaction. It must also be borne in mind that the kinetics of cutaneous immunology can differ markedly between humans and mice. For example while the skin sensitization phase is complete by 7 days in mice this can take as long as 28 days in man [7, 8].

Of particular prominence among danger signals are TLR ligands. It is believed that the induction of adaptive immune responses to haptens has the same or greater need for danger signals and there is evidence to suggest that at least some haptens deliver their 'danger' signal through release of endogenous ligands of TLR. The chemical insult in this way mimics the effects of mechanical insult/injury [9, 10]. Extracellular matrix (ECM)-derived TLR ligands predominantly stimulate TLR4 with the resultant expression of Th1/Th17-promoting cytokines. Release of extra domain A-positive fibronectin (FnEDA⁺), a ligand for TLR4 [11], by recruited Th1 cells [12] provides for a positive feedback mechanism that sustains, and further expands, Th1-driven inflammation. However, with repeated and continued hapten application there is a shift to Th2-driven inflammation [7]. This can be achieved through one or more of the following mechanisms:

- (1) Repeated stimulation of TLR4 has been reported to abrogate the production of Th1/Th17-promoting cytokines [13].
- (2) Extracellular and stress protein ligands will also stimulate TLR2 which, although initially reinforcing a Th1-driven inflammation over time can lead to 'Th2 skewing' [14, 15].
- (3) With prolonged hapten exposure oxidative cellular stress can occur. Picardo et al. [16] showed that *in vitro* the contact allergen *p*-phenylenediamine induces oxidative stress at prolonged, as opposed to short, exposure times (24 vs. 0.5 h). Oxidative stress has also been shown to enhance or cause intracellular TLR signalling as with TLRs 3, 8 and 9 [17–19]. Although stimulation of these TLRs can result in production of type I IFN that could promote Th1 inflammation, TLRs 2, 3, 7, 8, and 9 have each been reported to result in the production of thymic stromal

lymphopoietin (TSLP), a strong promoter of Th2-driven inflammation [20].

- (4) Once Th2 cytokines are produced they will start to down-regulate actively both the capacity of TLR4 to stimulate Th1-promoting cytokines and the positive feedback loop involving FnEDA⁺ [21, 22]. It is therefore reasonable to propose an explanation for the apparently paradoxical observations that short-term, high-level exposure to a hapten can result in selective Th1 driven inflammation, whereas more prolonged and repeated exposure to the same hapten might favour instead preferential Th2-driven inflammation.

Low-grade TLR4 stimulation favours Th2 rather than Th1 responses. Dong et al. [23] looked at the response to ovalbumin (OVA) airways challenge in the presence of low-dose (0.1 µg) and high-dose (100 µg) LPS, a TLR4 ligand. In the presence of high-dose LPS there was a Th1 response, with neutrophilic inflammation and expression of high levels of IFN-γ. However, in the presence of low-dose LPS a Th2 response was observed, with eosinophilic inflammation and high levels of IL-4, IL-5 and IL-13.

In addition to haptens, irritants can also stimulate TLR, or at least synergize in TLR stimulation [24]. It is therefore possible to speculate how lifestyle factors and the conditions of hapten exposure might combine to influence the atopic phenotype. The argument is that repeated low-dose skin exposure to haptens (such as preservatives) and irritants (such as surfactants found in toiletries and detergents) could theoretically result in a low-grade, sub-clinical and antigen-non-specific impact on the regulation of cutaneous adaptive immune function such as to promote the preferential development on Th2-biased responses to third party antigens encountered in the skin.

The paradox: cutaneous hapten causing allergic contact dermatitis and also atopic dermatitis

Allergic contact dermatitis

This has been reviewed elsewhere [25]. In contrast to AD ACD is mediated by Th1/Tc1 cells and expression of Th1 cytokines such as γIFN and IL-12. Cutaneous applications of powerful haptens at a high concentration will cause sensitization; a further challenge will elicit a Th1/Tc1-driven inflammation [26–28]. Th17/Tc17 cells also are present in elicitation phase of ACD in significant numbers [29].

Animal models of atopic dermatitis through cutaneous hapten exposure

Repeated topical exposure of mice to the experimental haptens oxazolone or trinitrochlorobenzene over an

extended period causes skin inflammation to shift from a preferential Th1 phenotype to a selective Th2-type response similar to that observed in human AD [30–32]. Repeated challenges with oxazolone produced a chronic Th2-like skin inflammation. This inflammation consisted of increased numbers of Th2 cells, mast cells and eosinophils, increased IL-4 and elevated IgE levels. Skin barrier abnormality also becomes evident with increased epidermal hyperplasia and decreased expression of filaggrin, increased transepidermal water loss and reduced lamellar membranes, as observed in AD [7, 30, 31].

In such a model of AD provoked through repeated hapten challenge [7] skin perturbation develops with both barrier and epidermal lipid abnormalities as a consequence, and as such increased microbial and other exogenous insults could be expected to lead to some Th1 as well as Th2 element.

As mentioned earlier, there has been interest recently in the role of microbial stimulation of the innate immune system through danger-associated molecular patterns, most notably TLR, and their role in the hygiene hypothesis [33]. It has been postulated that haptens can deliver danger signals, with release of pro-inflammatory cytokines such as TNF- α and IL-1 β [4]. We here consider whether haptens could deliver a 'danger' signal through such TLR stimulation.

Toll-like receptors and endogenous ligands of Toll-like receptors

The innate immune system responds to invading microbes by activation of a pro-inflammatory cascade to engage the immune system. Pattern recognition receptors (PRRs) are crucial parts of this innate immune system. These PRRs can recognize various proteins of invading microorganisms, which then trigger the development of a cytokine cascade. Amongst the various intracellular and membrane PRRs are TLRs and NOD-like receptors [34]. TLRs are the best characterized and to date 10 have been described [34]. In the skin TLRs are found on keratinocytes, dendritic/antigen presenting cells, mast cells and fibroblasts [35]. Bacterial LPS, a major component of gram-negative bacterial cell wall, was first described as a microbial ligand for TLR, in this case TLR4 [36]. Since then a various conserved microbial components have been identified that are able to stimulate different TLRs, including double-stranded RNA, flagellin and peptidoglycan [37].

More recently several endogenous proteins, such as heat shock proteins (HSP), have been found to be activators of the TLRs [37]. To date, six TLRs are known to be stimulated by these endogenous ligands. TLR2 and TLR4 are located on the cell surface (Fig. 1). TLR4 stimulation will lead to release of Th1/Th17-associated cytokines such

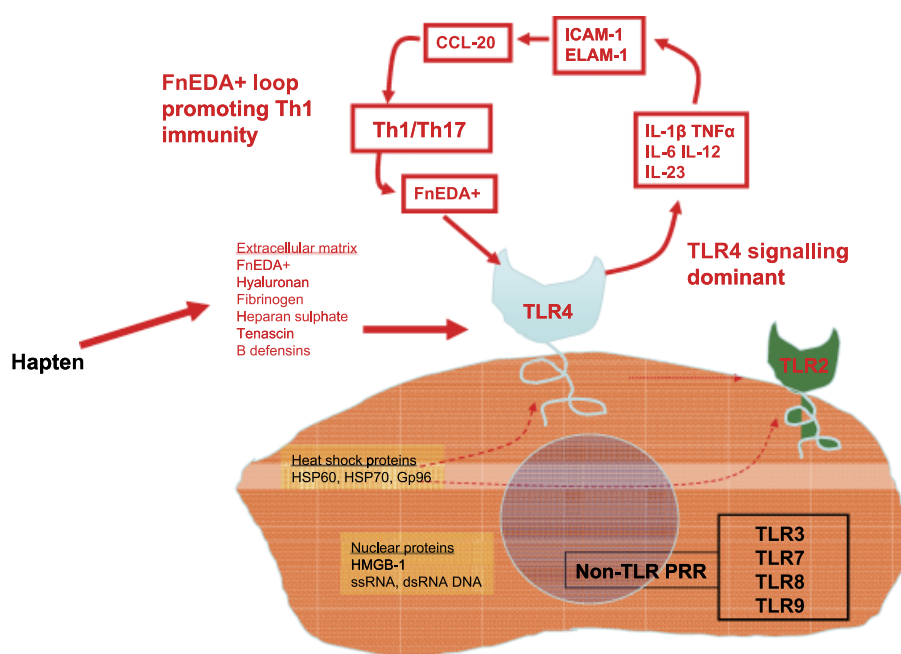


Fig. 1. Cutaneous hapten exposure initially leads to promotion of T-helper type 1 (Th1)-driven immunity. Cutaneous hapten exposure, analogous to mechanical trauma, effects an injury response and expression of extracellular matrix ligands for Toll-like receptor (TLR), such as extra domain A-positive fibronectin (FnEDA⁺) and hyaluronan. This expression leads to TLR4 stimulation as the dominant signal. TLR4 stimulation promotes TLR2 and there is minor TLR2 stimulation. TLR4 stimulation, assisted initially by TLR2, leads to expression of Th1/Th17-associated cytokines. TNF- α and IL-1 β up-regulate endothelial adhesion molecule expression, and together with up-regulation of chemokines such as CCL-20, promote recruitment of Th1/Th17 cells. Th1 cells produce FnEDA⁺ in significant amounts leading to a positive feedback loop for Th1-driven immune response.

as TNF- α , IL-1 β , IL-6, IL-12, IL-18 and IL-23 [11, 32, 38, 39], presumably tailored towards combating extracellular bacteria. Following initial stimulation TLR2 will synergize with TLR4 in driving Th1 stimulation (though this can change with repeated stimulation). TLR3, TLR7, TLR8 and TLR9 are located intracellularly (Fig. 1). Stimulation of intracellular TLRs will initially be characterized by, amongst other cytokines, type 1 IFN production, presumably to provide resistance to viral infection [40, 41].

Known endogenous ligands for TLR are classified as intracellular or extracellular:

- (1) ECM proteins namely hyaluronan, heparin sulphate, tenascin surfactant protein A, fibrinogen, FnEDA⁺ and β -defensin. In addition low-density lipoprotein has been reported to stimulate TLR4 [34, 42].
- (2) Intracellular proteins such as HSPs gp96 HMGB1 (which can also be found in the cytoplasm). Gp96 is the endoplasmic reticulum form of HSP90 [43]. HSPs are thought to stimulate TLR2 and 4 although where this is a direct stimulation has been disputed [38]. HMGB1 is a relatively small protein of 215 amino acid residues [44] which is DNA-binding and is thought to stimulate TLR2 and 4 and another molecular pattern receptor (RAGE) [44]. Messenger RNA, single stranded RNA and DNA act through TLRs 3, 7, 8 and 9, respectively.

Although the ability of haptens to deliver danger signals upon cutaneous application has been recognized for some time [4], the mechanisms by through which this is achieved have not yet been elucidated. However, there is available circumstantial evidence that provision of appropriate danger signals following skin exposure to chemical allergens could be achieved through release of endogenous ligands for TLRs. If this is indeed the case then chemical insults such as this would share a common pathway with other forms of trauma such as mechanical injury [9, 10]. Indeed several endogenous ligands for TLR such as HSP 70 [45], hyaluronan [46] and β defensin [47] are recorded following skin contact with a chemical allergen. In one study an increase in total Fn expression was the one of the earliest changes noted after cutaneous hapten application, preceding any immunological changes (a subset of Fn, FnEDA⁺ is a TLR4 ligand) [11, 48].

Initial cutaneous application of hapten leads to a T-helper type 1 response through Toll-like receptors 4 signalling and an extra domain A-positive fibronectin loop

Hapten-induced expression of endogenous ECM TLR ligands such as hyaluronan and FnEDA⁺ will result in the stimulation of TLR4, and subsequently, the expression of Th1/Th17 cytokines [11, 32, 38, 39]. As some HSP ligands are also up-regulated after cutaneous hapten application

there may also be some TLR2 expression which will initially augment the Th1 response driven by exposure to hapten [49]. There is also some controversy as to whether some ECM TLR ligands can also stimulate TLR2 [42]. TLR4 stimulation has also been reported to up-regulate TLR2 expression [50].

Release of TNF- α and IL-1 β following TLR4 stimulation will cause the enhanced endothelial expression of adhesion molecules ICAM-1 and ELAM-1 with a consequent augmentation of T cell recruitment into skin tissue [51]. Recruited Th1 cells will produce significant amounts of FnEDA⁺, an endogenous ligand for TLR4 [12]. This positive feedback loop will therefore drive and reinforce the Th1 inflammation observed in ACD (see Fig. 1).

Prolonged and repeated exposure to chemical allergens and the evolution of a selective T-helper type 2-type phenotype: role of Toll-like receptors (Fig. 2)

Whereas Th1/Th17 inflammation results following hapten application through TLR4 stimulation and positive FnEDA⁺ feedback loop, repeated and prolonged exposure will lead to a loss of Th1 dominance. This can occur through several mechanisms.

Repeated and prolonged Toll-like receptors 4 stimulation leads to abrogation of the promotion of T-helper type 1/17 inflammation

Repeated stimulation of TLR4 leads to abrogation of the Th1-promoting response [52]. IL-10 may be involved in this reduced responsiveness [53]. It is thought that this is a 'damage limitation' effect and prevents excessive tissue injury and necrosis [13, 54].

Prolonged stimulation of Toll-like receptors 2 stimulation leads to 'T-helper type 2 skewing'

TLR2 activation can trigger secretion of factors such as TNF- α , and has been found to be able in certain experimental conditions to contribute to skin sensitization [49], so that initially TLR2 synergizes with TLR4 stimulation. However, paradoxically, IL-10 released from concomitant TLR-2 stimulation has been shown to block the induction of a subset of Th1 cytokines induced by TLR4 [55], and this could be important in the later stages of inflammation. Bacterial stimulators of TLR2, in contrast to TLR4, can skew the immune system towards a Th2 response, and were found to exacerbate experimental asthma induced by OVA challenge [56]. Other workers have also found a Th2 bias from TLR2 stimulation [15]. Transgenic mice vaccinated in the presence of a TLR2 agonist leads to a Th2 response whereas in the presence of a TLR4 agonist leads to a Th1 response [14]. Exposure to OVA, together

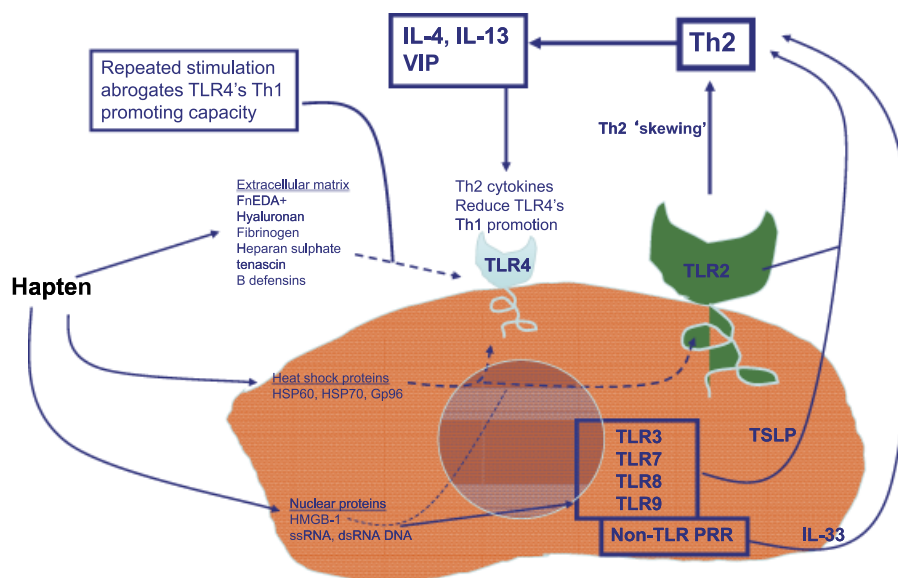


Fig. 2. Chronic cutaneous hapten exposure leads to promotion of T-helper type 2 (Th2)-driven immunity. Repeated stimulation of Toll-like receptor (TLR)4 abrogates its Th1-promoting properties. Prolonged TLR2 stimulation leads to 'Th2 skewing'. Th2 cytokines reduce the Th1-promoting capabilities of TLR4 further and close down the extra domain A-positive fibronectin (FnEDA⁺) Th1-promoting feedback loop. Intracellular stress and intracellular TLR ligand expression and TLR signalling leads to expression of thymic stromal lymphopoietin (TSLP), which promotes Th2 immunity further. Chemical stimulation of some intracellular non-TLR pattern recognition receptors (PRRs), such as NOD2, and IL-33 expression, may also help promote a Th2 response. HSP, heat shock proteins; VIP, vasoactive intestinal peptide.

with a TLR4 agonist adjuvant resulted in the development of a Th1 response in OVA-specific, MHC class II-restricted $\alpha\beta$ TCR transgenic mice whereas similarly sensitized mice, in the presence of a TLR2 adjuvant, developed a Th2 response [14]. Peptidoglycan, which stimulates TLR2, is a potent inducer of granulocyte/macrophage colony stimulating factor (GM-CSF) and cysteinyl leukotriene [57]. The TLR4 activator, LPS, however, does not induce these molecules. This could be of relevance as GM-CSF enhances the capacity of antigen presenting cells from atopic subjects to induce Th2 responses by a mechanism involving cysteinyl leukotriene [58]. Basophils secrete IL-4 and IL-13 in direct response to TLR2 stimulation but not TLR4 stimulation [59]. TLR2 could also promote Th2 responses by abrogating the type 1 IFN amplification of Th1 cell differentiation [60].

Schaub et al. [61] found that for neonatal, adult and mouse peripheral blood mononuclear cells TLR4 stimulation effected a Th1 cytokine response whereas TLR2 signalling resulted in release of both Th1 and Th2 cytokines, the Th2 response being more prominent after longer times of stimulation and with more than minimal TLR2 stimulation.

With respect to vaccination experiments and the role of TLR4 and TLR2 mice, separate experiments have shown that while TLR4 is important for a Th1/Th17 response and diminishes Th2 responses, TLR2 stimulation favours a shift away Th1 to Th2 response to vaccination [62, 63].

Thymic stromal lymphopoietin expression leading to promotion of T-helper type 2 immunity

Extracellular TLR stimulation would be expected to predominate following initial exposure to cutaneous hapten. As alluded previously to prolonged, repeated exposure to haptens with resulting oxidative cellular stress [16] would be expected to lead to intracellular TLR ligand expression and TLR signalling [64, 65]. Like TLR2, intracellular TLRs (TLR 3, 7, 8, 9) can help promote TLR4-induced Th1 responses through IFN augmentation of Th1 cell differentiation [60]. However TSLP, a powerful promoter of Th2 responses, is also induced by TLR2, TLR3, TLR8 and TLR9 [20]. TSLP is highly expressed by keratinocytes in skin lesions of patients with AD [66]. Early production of TSLP precedes infiltration of dendritic cells (DC) expressing its receptor in allergen-induced late-phase cutaneous responses in atopic subjects [67]. TSLP-activated DC secrete Th2-recruiting chemokines but not IL-12 and induce naïve T cells to differentiate into inflammatory Th2 cells producing IL-4, IL-5, IL-13 [20]. Indeed, in an animal/hapten model of AD, production of TSLP corresponded with the switch from Th1 to Th2 cell tissue infiltration; the addition of repeated cutaneous irritant applications in this model further increased the Th2 polarization from repeated hapten challenge and also increased TSLP expression [68]. Irritants can therefore accentuate not only the Th1 response to cutaneous hapten in ACD elicitation and sensitization

responses [69, 70] but also the Th2 response in animal/hapten models of AD.

T-helper type 2 cytokines down-regulate the ability of Toll-like receptors 4 to drive a T-helper type 1-driven phenotype and suppress the T-helper type 1-promoting extra domain A-positive fibronectin feedback loop

Th2 cytokines appear to actively suppress TLR4 function. It has been shown that *in vitro* IL-4 and IL-13 cause a significant decrease in the level of expression of mRNA for TLR4 [22]. This appears to involve the signal transducer and activator of transcription 6 (STAT-6) which is active in atopic disease [71, 72]. Vasoactive intestinal peptide (VIP) is secreted by Th2 cells but not Th1 cells. VIP reverses the expression of TLR4-stimulated pathways; therefore VIP secretion impaired LPS-induced production of IL-6 and RANTES/CCL5 [73] and in another study inhibited TLR4 stimulated macrophage secretion of IL-1, IL-6, IL-12 and TNF- α [21]. A dominant Th2 cytokine phenotype is associated with decreased TLR4 monocyte expression [74]. There is an impaired Th1 response to the TLR4 ligand LPS by T cells derived from atopic asthmatics [75].

Although Th1 cells secrete significant amounts of the TLR4 ligand FnEDA⁺, Th2 cells do not. Thus, a shift from Th1 to Th2 cells will lead to suppression of this positive feedback loop and accelerate Th2 dominance. Consistent with this is the observation in mice that chronic contact inflammation caused by repeated cutaneous application of hapten marked increases in expression of Fn were recorded in the acute phase which was followed by Fn suppression in the chronic phase [76].

Other chemically stimulated intracellular pattern recognition receptors may promote T-helper type 2 cells

Chemical insult, including through hapten exposure, may stimulate both TLR-related and non-TLR related danger signals including inflammasomes [77]. One experiment demonstrated that one avenue of TLR-dependent Th2 polarization was dependent on co-stimulation of the intracellular NOD-like receptor NOD2 contained within the inflammasome [78]. IL-33 is regarded as an alarmin, which is released upon response to tissue injury and is located intracellularly within the nucleus [79]. IL-33 promotes Th2 polarization and is implicated in the pathogenesis of asthma by expanding Th2 cells and is also present in high levels in AD [79].

Thus, the initial innate immune response to cutaneous hapten exposure is primarily a Th1/Th17 response, which could be due to predominance of TLR4 stimulation from ECM TLR ligands. However, prolonged and repeated stimulation will lead towards Th2 polarization through a coalition of multiple effects. The other aspect of daily

cutaneous hapten exposure from household products and toiletries besides its repeated, prolonged nature is that it is usually low dose. Low-dose stimulation of TLR4 again would appear experimentally to favour a Th2 response.

Whereas high-level Toll-like receptors 4 stimulation favours a T-helper type 1 response low-level Toll-like receptors 4 stimulation favours a T-helper type 1 response

We have referred to experimental findings of Dong et al. [23] that while high-level TLR4 stimulation favours a Th1 response, low-dose TLR4 stimulation leads to a Th2 response. In a different experiment a high dose of LPS in similar circumstances, also induced a Th1 response characterized by neutrophil recruitment and an increase in IFN- γ production whereas repeating the experiment with a low dose of LPS induced a Th2 response with eosinophil infiltration [80]. Consistent with that observation, intra-nasal OVA exposure in sensitized mice stimulated a Th1 response in the presence of high doses of LPS but Th2 responses in the presence of low-dose LPS, the Th2 response being TLR4 dependent [81]. These are, therefore consistent, repeatable observations.

Clinical relevance. Is there a role for routine cutaneous hapten exposure in the aetiology of atopic disease?

Between 1964 and 1999, the prevalence of UK childhood AD and asthma has increased by approximately 325% and 500%, respectively [82]. We have already noted that an increase in oral exposure to haptens, in the form of chemicals in processed food and drugs such as antibiotics, has coincided with this increase [2]. There has also been an increase in skin exposure to chemicals contained in toiletries, detergents and household products in Westernized societies over the same period of time (Table 1). In the first half of the 20th century an indoor bathroom with running water was a rarity whereas by 1971 3 million UK households did not have a bathroom; by 2000 indoor

Table 1. Examples of likely cutaneous hapten exposure in pregnant women and infants in first year of life

	Cutaneous hapten exposure	Oral hapten exposure
Pregnant women	Toiletries, household products	Haptenic drugs (e.g. antibiotics, metal salts)
	Fragrances	Processed food and drink
	Hair dyes	
	Metal in jewellery	
	Metal salts in deodorants	
First year of life	Occupational exposure	
	Toiletries including wipes	Processed milk
	Talc	Haptenic drugs Processed food

bathrooms were present in over 99% of households in United Kingdom [83]. Indoor personal showers, which provide for frequent washing and use of skin cleaning products, is a phenomenon of the second half of the 20th century with availability increasing steadily during that time [84]. Fifty years ago exposure to toiletries, detergents and wipes were less common than today, no doubt due partly to lower disposable income ([85] and D. A. Basketter, personal communication). At the same time the number and complexity of chemicals used in skin care products have increased. Notwithstanding this, there is no reason to suppose that such increased opportunities for exposure to chemicals have impacted on the safety of products. In fact the safety profile of these products has changed little, or has frequently improved, as a consequence of company safety assessments and the introduction of new legislation [86, 87]. However, the question here is not one of safety or otherwise of individual products, but rather the more global/holistic impact of exposure to chemicals.

The rise in global use of toiletries is illustrated in Fig. 3. Between 1959 and 1976 there was a 250% increase in the sales of toiletries (all figures converted into 1976 dollar values). Most of the increase, both in relative and absolute terms, occurred in Western countries and sales grew at a rate of nearly double the increase in gross domestic product [85]. The significant increase in the prevalence of atopic diseases during this period of time was also found in these same countries [82]. By 1976 in the UK sales of toiletries stood at US\$581 million an increase of 370% compared with 1959, whereas in the much more populous India sales of toiletries was estimated at US\$74 million, a simple doubling since 1959. It is of interest, therefore, that a 1976 survey of the prevalence of asthma in Birmingham, UK, found a rate of 4.3% in white children born in the United Kingdom but only of 1.1% in children resident in Birmingham but who had been born in the Indian sub-continent [88]. In Japan the sales of toiletries increased

from \$285 million in 1966 to \$1957 million by 1976 [85]. The prevalence of childhood asthma in Japan was estimated in 1966 to be 0.7% but by 1983 this had increased to 7.9% [89].

Permanent and semi-permanent hair dyes usually contain one the powerful aromatic amine group of haptens. Their use by young women has increased substantially. In 1992 a survey by the Japan Soap and Detergent Association found that 6% of women in their twenties reported using hair colouring products [90]. By 2001, this figure had increased to 85% [88].

Some chemicals in skin care products, such as the many surfactants, present in baby wipes soaps and detergents are inherently irritant in nature. Others, such as fragrances and preservatives, also have haptenic potential. As we have reviewed, there are two main immunological responses resulting from cutaneous exposure to hapten i.e. ACD, a Th1-driven disease, and AD, a Th2-driven disease. In clinical practice ACD, often from application of a potent hapten over a defined time limit, is well documented and accepted as a specific entity. But a Th2 response to cutaneous hapten exposure is neither documented nor accepted as a clinical entity. However, it is everyday practice with skin care products to apply haptens repeatedly and at a low dose, which experimentally would favour a Th2 bias.

Some (but not all) of the hapten-induced AD animal models use mice with an inherent Th2 bias. There are certain situations in the normal human lifecycle where there is also a Th2 predisposition, for example the mother during the early and middle gestation, and in neonatal/early life [91, 92]. Th2 dominance during pregnancy is thought to protect the foetus from maternal Th1 attack [92–94]. Predominant Th1-type immunity has been observed in recurrent spontaneous abortion [93, 94] and in preeclampsia [95]. The onset of labour is associated with an increase in cyclo-oxygenase-2 production of prostaglandin E2. As Th2 cytokines inhibit cyclo-oxygenase-2 activity they may permit the pregnancy to continue until term [96]. This persistence of Th2-type immunity required for *in utero* survival and term birth results in a general Th2 dominance in neonates [97]. These are therefore perhaps the time periods when repeated low-dose skin exposure to haptens and/or irritant chemicals may, through the innate TLR system, bias the immune system further towards Th2 polarization and atopy either in the foetus (with the maternal immune system acting as a surrogate), or in the neonate. Indeed one study reports that a stronger maternal Th2 immune profile during the first trimester of pregnancy is associated with subsequent childhood wheezing and atopy at 3 years of life [98]. Some occupations during pregnancy increased the risk of the offspring developing atopic disease [99, 100]. These occupations, namely cooks, dental assistants, electrical assemblers, sewers, bookbinders and nurses involve either increased exposure to

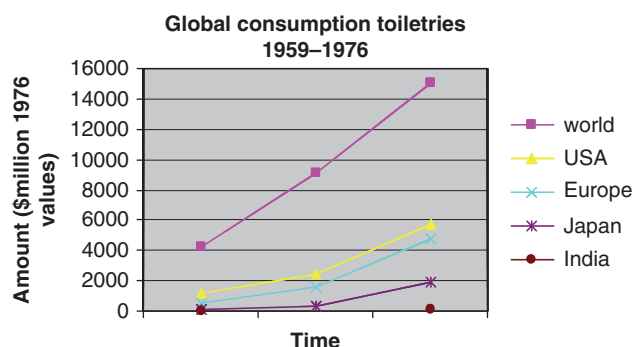


Fig. 3. Changes in global consumption of toiletries 1956–1980. Figures derived from reference Countries and regions with largest increase in atopic disease also showed the highest increase in consumption of skin care products.

chemicals, the necessity for frequent washing of hands and therefore exposure to surfactants and haptens, or repeated low-level mechanical trauma to the skin, all of which may involve some degree of repetitive low-scale stimulation of the skin innate immune system which could promote Th2 immunity. Experimental mechanical irritation polarizes skin DCs to elicit a Th2 response by inducing TSLP [101]. For children in the first year of life modern practices usually involve frequent application of detergents and wipes to the baby's skin. Our survey showed that over 80% of baby skin care products contain haptens [102].

Adaptation of the innate immune system, in particular through TLR4 signalling, by the levels of environmental endotoxin (LPS), has been the subject of much research recently. Furthermore, some atopic protein allergens such as house dust mite for instance, can also stimulate the innate immune system through facilitation of TLR signalling [102]. We postulate here that repeated and prolonged skin exposure to chemical allergens (and possibly certain skin irritants) may stimulate the evolution of a selective Th2 phenotype. This in turn will cause increased suscep-

tibility to atopic allergic disease. Such exposure may be particularly influential at those times when there already exists a selective Th2 bias.

Further work on this theme would include a combined epidemiological immunological approach. Epidemiological work should compare changing prevalences of childhood atopic diseases with the change in hapten exposure in different time 'periods' and countries. Immunological work should include analysis of the changes in innate immune signalling in the mouse models of AD, in particular those associated with the initial Th1 response and to the later Th2 response with repeated hapten exposure.

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