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An approach to allergy risk assessments for e-liquid ingredients

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ABSTRACT

Many flavours and fragrances are known allergens. Their selection and inclusion levels in e-liquids must therefore be guided by toxicological principles, taking into account the exposure pattern and inhalation route of exposure. For contact sensitisation, a general, agreed quantitative risk assessment approach to prevent dermal sensitisation exists. Here we propose exposure parameters and safety factors to apply this approach to e-liquid ingredients. Additionally, as a risk management approach for pre-sensitised individuals, we derive a threshold of 0.1% for indicating the presence of a contact sensitiser in e-liquid. Risk assessment for respiratory sensitisation is not well established. Occupational exposure limits that protect against respiratory allergy are generally very low. Cocoa shell extract is used as a case study to discuss the issues. A tolerable exposure level is derived and estimates of consumer exposure are presented, leading to the practical risk management approach of excluding respiratory sensitisers as e-liquid ingredients. Related to this, if natural extracts are used as flavourings in e-liquids, we recommend only protein-free versions are used. Additionally, we recommend the presence of any potential food allergens should be noted on the product information.

1. Introduction

The use of various flavours in e-cigarettes and e-liquids can lead to concerns over the potential for allergic responses from the use of these vaping products. Different types of allergy can be relevant to vaping exposures. Skin or contact sensitisation, also known as type IV delayed cell-mediated type hypersensitivity, needs to be considered due to the use of flavours or fragrances, and sometimes other compounds, such as preservatives, many of which are known potential contact sensitisers. Dermal quantitative risk assessment for contact sensitisation is well established (Api et al., 2008), and this paper proposes the details for an analogous approach for vaping exposures. Early thinking in this area has been briefly described in a poster presentation elsewhere (Costigan, 2014).

Because the main intended exposure from vaping products is via inhalation, respiratory allergy (also referred to as type I immediate IgE-mediated hypersensitivity) also requires consideration. Although chemical respiratory allergy is much less common than contact sensitisation, the potential adverse effects are much more severe. We have previously introduced respiratory sensitisation as an exclusion criterion for ingredient selection (Costigan and Meredith, 2015). In this Position Paper, we expand upon that by discussing cocoa shell extract as a case study to illustrate the risk assessment process and related challenges, and include some practical resources for identifying type I allergies. Additionally, we propose to extend this exclusion criterion to risk management of ingredients associated with IgE-mediated food allergies and discuss various food immunological responses.

2. Type IV delayed cell-mediated hypersensitivity

Vaping transforms e-liquid into an aerosol that enters the consumer's mouth and respiratory tract. E-liquid formulations generally contain nicotine, solvents and flavourings, and might contain other ingredients, such as preservatives. Some flavour ingredients, whether natural extracts or synthetic, can induce contact sensitisation. Thus, a hazard identification approach is needed to decide whether an intended ingredient has that potential. And if it does, whether the risk of allergy responses from the expected exposures is acceptably low.

Sensitisation has two basic phases, induction and elicitation of a reaction (OECD, 2012). Induction 'primes' an individual's immune system to a specific substance by inducing specialised

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immunological memory on exposure to an allergen. Induction might take weeks to years of exposure to develop and, after induction, further exposure can elicit the classic inflammatory reaction associated with allergic contact dermatitis. Both induction and elicitation are threshold mechanisms. The threshold for elicitation, however, is typically lower than that for induction. The network of Langerhans cells in oral tissue, means that oral exposure is also relevant. However, contact sensitisation reactions are not known to extend into the respiratory tract.

Typical types of compounds with contact sensitisation potential are flavourings and preservatives. Flavourings often contain natural flavours, which are effectively mixtures of constituents. To assess the risks associated with single ingredients and constituents of naturals, three questions should be addressed:

1. Does the ingredient/constituent have any sensitisation potential?
2. If yes, is the sensitiser present at a sufficiently low level that it is not expected to elicit reactions even in pre-sensitised individuals?
3. If present at higher levels, are the levels expected or not expected to induce sensitisation?

In this article, we derive a risk assessment approach specific to vaping products. Fig. 1 summarises the resultant practical process for assessment of contact sensitisation of e-liquid ingredients.

2.1. Contact sensitisation hazard identification

The main toxicological test for determining sensitisation potential of a substance is the local lymph node assay (OECD, 2010). This test measures the concentration needed to stimulate a threefold increase in lymph node cell proliferation in mice. Other classic animal methods are the Buehler and guinea pig maximisation tests. The classic clinical study, which shows an absence of sensitisation induction in humans at specific levels, is the human repeat insult patch test (Basketter, 2008).

Good progress has been made with *in silico* and *in vitro* methods for contact sensitisation hazard identification. The OECD has published two new *in vitro* test guidelines (442C and 442D) that address key events in the adverse outcome pathway for contact sensitisation and, therefore, can be part of a weight of evidence approach to hazard assessment. No *in vitro* test is validated to assess the relative potency of sensitisers.

2.2. Quantitative risk assessment (QRA)

The QRA methodology for contact sensitisation is well established and has been adopted by industry (e.g. International Fragrance Association and Research Institute for Fragrance Materials) and regulators (e.g. REACh). The method is based on defining a no expected sensitisation induction level (NESIL) and applying appropriate uncertainty factors to establish a level where the risk is acceptably low—the acceptable exposure level (AEL) (Api et al., 2008). The consumer exposure level (CEL) is then estimated and compared against the AEL. If AEL/CEL=>1, the proposed use of the compound is deemed supportable. NESILs can be derived from animal and/or human data and are quantitative measures of the potency of the sensitiser. Compilations of appropriate sensitisation data for NESILs derived from flavours and fragrances have been published (Gerberick et al., 2005; Kern et al., 2010).

Although elicitation is also of some interest, the main interest of the QRA is in avoiding sensitising consumers at all. The usual approach to protecting pre-sensitised consumers is to define levels above which consumers should be informed of the presence of known sensitisers, so they may decide whether to use the product if they know of, or suspect a sensitivity. This approach is analogous to that taken in the EU Cosmetics Directive 76/768/EEC which requires known sensitisers at concentrations greater than 0.001% in leave-on products and 0.01% in rinse-off products to be mentioned on the label (European Commission, 1976). In the REACh, CLP legislation the threshold for mentioning sensitisers is 0.1%.

How to practically risk assess multiple sensitizers with a similar chemistry in one product, that might exhibit some level of cross-reactivity, still requires further debate. Quantitative evidence for levels of cross-reactivity exists for only a limited number of compounds and is not currently available for the bulk of flavour ingredients used in e-liquids. Where such information is available, this should be considered in the risk assessment.

2.2.1. Acceptable exposure level (AEL)

The AEL is the weight of evidence NESIL divided by the product safety assessment factor (SAF). The product SAF was defined by Api et al., (2008) and actually combines three separate SAFs: inter-individual variation, product-specific matrix effects and product-specific use. A further refinement for sensitising fragrance ingredients (Basketter and Safford, 2016) suggested decreasing the product matrix SAF range from 1 to 10 to 0.3−3 and expanding the product use SAF into three different SAFs: frequency or duration of product use, potential presence of occlusion and skin condition/site. However, specifically for the risk assessment of e-liquids, we consider the SAF defined by Api et al. (2008) to be more conservative and applicable. Occlusion, such as from clothing, is not applicable to the oral and respiratory exposure from e-liquids. The proposed skin condition/site factor considers pre-existing inflammation, particularly in areas prone to increased levels of inflammation (e.g. hands, underarms, peri-anal and peri-ocular regions). The mouth, however, can normally sustain considerable abrasion forces from chewing and salivary enzymes and, hence, is not particularly prone to inflammation. The possible accumulation of a product through repeated use, which is part of the product use SAF proposed by Basketter and Safford, is incorporated in the calculation of CELs presented here.

As vapers represent a general, adult population, we recommend a default inter-individual variation SAF of 10. The matrix SAF accounts for the compound having been tested in a different vehicle than the e-liquid aerosol to which the consumer will be exposed. As the presence of irritants or certain solvents in the e-liquid aerosol might increase epithelial penetration, we recommend a matrix SAF of 3. The product-specific use SAF takes in account exposure-related matters such as site of contact, epithelial integrity and duration of exposure. During vaping, a brief peak of oral exposure occurs during the actual puff and holding of the aerosol in the mouth when the formulation is deposited in the oral mucosal cavity. Lips and parts of the mouth are highly vascular, which results in increased absorption of compounds, although rapid dispersion and salivary dilution significantly limit exposure. For such exposure, we recommend a product-specific use SAF of 3 (Api et al., 2008). The overall product SAF, therefore, is 10*3*3 = 90, meaning the AEL for a compound is the “weight of evidence NESIL”/90.

2.2.2. Consumer exposure level (CEL)

For contact sensitisation, the CEL represents the quantity of compound per exposed surface area (Kimber et al., 2008). A worst case approach is to estimate exposure to an ingredient during the initial mouth-hold phase and assume that all the ingredient in the puff deposits in the mouth. This is extremely conservative as it would mean no product deposition in the lungs whatsoever.

Few reports on adult mouth surface area are available (Collins