

Recommendation to include fragrance mix 2 and hydroxyisohexyl 3-cyclohexene carboxaldehyde (Lyal[®]) in the European baseline patch test series

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Background: The currently used fragrance mix in the European baseline patch test series (baseline series) fails to detect a substantial number of clinically relevant fragrance allergies.

Objective: To investigate whether it is justified to include hydroxyisohexyl 3-cyclohexene carboxaldehyde (Lyal[®]) and fragrance mix 2 containing hydroxyisohexyl 3-cyclohexene carboxaldehyde, citral, farnesol, coumarin, citronellol, and α -hexyl cinnamal in the European baseline patch test series.

Methods: Survey of the literature on reported frequencies of contact allergy and allergic contact dermatitis from fragrance mix 2 and hydroxyisohexyl 3-cyclohexene carboxaldehyde (Lyal) as well as reported results of experimental provocation test.

Results: Fragrance mix 2 has been demonstrated to be a useful additional marker of fragrance allergy with contact allergy rates up to 5% when included in various national baseline patch test series. Of the fragrance substances present in fragrance mix 2, hydroxyisohexyl 3-cyclohexene carboxaldehyde is the most common sensitizer. Contact allergy rates between 1.5% and 3% have been reported for hydroxyisohexyl 3-cyclohexene carboxaldehyde in petrolatum (pet.) at 5% from various European centres when tested in consecutive dermatitis patients.

Conclusions: From 2008, pet. preparations of fragrance mix 2 at 14% w/w (5.6 mg/cm²) and hydroxyisohexyl 3-cyclohexene carboxaldehyde at 5% w/w (2.0 mg/cm²) are recommended for inclusion in the baseline series. With the Finn Chamber[®] technique, a dose of 20 mg pet. preparation is recommended. Whenever there is a positive reaction to fragrance mix 2, additional patch testing with the 6 ingredients, 5 if there are simultaneous positive reactions to hydroxyisohexyl 3-cyclohexene carboxaldehyde and fragrance mix 2, is recommended.

Key words: allergic contact dermatitis; contact allergy; epicutaneous testing; EU legislation; fragrances. © Blackwell Munksgaard, 2008.

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The European baseline patch test series consists of the minimum recommended test set to be used during routine diagnostic patch test investigations and that will be ordinarily supplemented by additional series and other allergens as appropriate to adequately investigate the clinical problem. Any baseline patch test series must regularly be re-evaluated for the need of changes in composition. Inclusion of new allergens as well as exclusion of

old substances giving too low yield of relevant positive reactions may be considered. Further, changes in patch test concentration may be warranted (1). For the European baseline patch test series (baseline series), the major responsibility for this process has been delegated to the European Environmental Contact Dermatitis Research Group (EECDRG) by the European Society of Contact Dermatitis (ESCD).

Methyldibromo glutaronitrile 0.5% petrolatum (pet.), an important preservative allergen widely used in household, industrial, pharmaceutical products, but particularly in cosmetics, was the latest patch test substance added to the baseline series (2). However, its use in cosmetics has recently been banned in the EU, not only from leave-on but now also from rinse-off products (3), and this will certainly result in a decrease of contact allergy to methyldibromo glutaronitrile in due course. However, allergen substitution may result in an increase in contact allergy to substances such as the methylchloroisothiazolinone and methylisothiazolinone mixture. Methylisothiazolinone has been recently introduced in cosmetics to replace the mixture as it is not only less allergenic but also less active, so higher concentrations are needed. Methylchloroisothiazolinone- and methylisothiazolinone-sensitive patients may react to methylisothiazolinone alone (4, 5).

2 other allergens in the baseline series have recently been subject to a study evaluating the need to keep them both, i.e. the rubber additives mercaptobenzothiazole (2% in pet.) and mercapto mix (containing mercaptobenzothiazole and 3 other related constituents at 0.5% each in pet.). The study concluded that both the mercapto mix and the mercaptobenzothiazole were required because if the mix had not been tested in the baseline series, on average 22% of patients allergic to a mercapto compound would have been missed, and for mercaptobenzothiazole, this would have been on average 20% (6).

In order to be able to make decisions regarding elimination or inclusion of allergens in the baseline series, the EECDRG encourages representatives of national contact dermatitis groups to collect data and information on patch test results, relevance of the allergens found, etc. as well as comments to be submitted to the EECDRG to be included in the background material on which the final decision will be based.

Currently, this is the case for another group of sensitizers namely fragrances.

Fragrance Mix 2

The currently used fragrance mix (fragrance mix 1) in the baseline series was introduced by Larsen in 1977 (7). It is estimated that at least 15% of relevant cases of perfume allergy are not recognized by patch testing with the fragrance mix 1 in the baseline series (8), but Larsen et al. reported that as much as 33% of fragrance sensitivity may be missed if it is used as the only test substance (9).

Besides the fragrance substances of fragrance mix 1 in the baseline series, a large number of

fragrance substances have been demonstrated to be potential contact sensitizers.

In a European multicentre trial with 14 frequently used fragrance substances, contact allergy to the individual substances was demonstrated in the range 2.7–0.1% (10). 6 of the 14 fragrance substances causing most contact allergy have been tested as fragrance mix 2 in other European multicentre studies (11, 12); the mix contains hydroxyisohexyl 3-cyclohexene carboxaldehyde (Lyrall[®]), citral, farnesol, coumarin, citronellol, and α -hexyl cinnamal. Frosch et al. reported that contact allergy to fragrance mix 2 (14%) was found in 2.9% of the patients and that 1/3 of the patients reacting to fragrance mix 2 were negative to fragrance mix 1 (11). Furthermore, Geier et al. in a large German study reported that 535 of 6968 (7.7%) patients reacted to fragrance mix 1 and 321 of 6968 (4.6%) patients reacted to fragrance mix 2. 151 of them reacted to both allergens, which means that 171 patients (2.5%) tested negatively to fragrance mix 1 but positively to fragrance mix 2 (13). Thus, this finding proves that fragrance mix 2 at 14.0% in pet. has acceptable diagnostic qualities and is helpful as an additional marker for fragrance allergy (13).

Hydroxyisohexyl 3-cyclohexene carboxaldehyde or Lyrall[®]

Of the fragrance substances present in fragrance mix 2, hydroxyisohexyl 3-cyclohexene carboxaldehyde followed by farnesol are the most common sensitizers (12, 14). Hydroxyisohexyl 3-cyclohexene carboxaldehyde is 1 of the fragrance substances published in the Fenn study in 1989 (15). When patch tested in a previous EECDRG study, 3 positive reactions to it were found in 106 patients (2.8%) (16). Similar contact allergy rates, between 1.5% and 3%, have been reported when tested at 5% in pet. from various European centres in consecutive dermatitis patients (10, 14, 17–19). In contrast, in North America, the prevalence was found to be only 0.4%, which was considered to be mainly because of the presence of the ingredient in higher concentrations in deodorants in the EU compared with the USA (20–22). In around 40–50% of patients with contact allergy to the fragrance ingredient, the fragrance mix 1 patch test was negative (10, 14, 18).

Fragrance ingredient allergy, and particularly to citral and hydroxyisohexyl 3-cyclohexene carboxaldehyde, was found to be common in patients with hand eczema (23). However, in an experimental provocation study using immersion of fingers in a dishwashing liquid with hydroxyisohexyl 3-cyclohexene carboxaldehyde in patients with

Table 1. The European Baseline series with the addition of fragrance mix 2 and hydroxyisohexyl 3-cyclohexene-carboxaldehyde

Compound	Concentration % (w/w) in pet. except for those in aqua ^a	Concentration in mg/cm ² *	Concentration in mg/cm ² in hydrophilic dried-in vehicle
Potassium dichromate	0.5	0.2	0.025
4-Phenylenediamine base (<i>p</i> -phenylenediamine)	1.0	0.4	0.090
Thiuram mix	1.0	0.4	0.025 ^b
TMTM	0.25	0.1	
TMTD	0.25	0.1	
TETD	0.25	0.1	
PTD	0.25	0.1	
Neomycin sulfate	20.0	8.0	0.2
Cobalt chloride	1.0	0.4	0.020
Benzocaine	5.0	2.0	0.63 ^c
Nickel sulfate	5.0	2.0	0.20
Clioquinol (chionoform and vioform)	5.0	2.0	0.19 ^d
Colophony	20.0	8.0	1.50
Parabens	16.0	6.4	1.00 ^e
Methylparaben (methyl-4-hydroxybenzoate)	4.0	1.6	
Ethylparaben (ethyl-4-hydroxybenzoate)	4.0	1.6	
Propylparaben (propyl-4-hydroxybenzoate)	4.0	1.6	
Butylparaben (butyl-4-hydroxybenzoate)	4.0	1.6	
<i>N</i> -Isopropyl- <i>N</i> '-phenyl-4-phenylenediamine	0.1	0.04	0.075 ^f
Wool alcohols	30.0	12.0	1.00
Mercapto mix	2.0	0.8	0.075 ^g
<i>N</i> -cyclohexylbenzothiazyl sulfenamide	0.5	0.2	
Mercaptobenzothiazole	0.5	0.2	
Dibenzothiazyl disulfide	0.5	0.2	
Morpholinylmercaptobenzothiazole	0.5	0.2	
Epoxy resin	1.0	0.4	0.050
<i>Myroxylon pereirae</i> (Balsam Peru)	25.0	10.0	0.80
4-tert-Butylphenol formaldehyde resin (PTBP resin)	1.0	0.4	0.040
Mercaptobenzothiazole	2.0	0.8	0.075
Formaldehyde	1.0 ^h	0.3	0.18
Fragrance mix 1	8.0 ^h	3.2	0.45 ⁱ
Cinnamic alcohol	1.0	0.4	
Cinnamal (cinnamic aldehyde)	1.0	0.4	
Hydroxycitronellal	1.0	0.4	
α -Amyl cinnamal (α -Amyl cinnamaldehyde)	1.0	0.4	
Geraniol	1.0	0.4	
Eugenol	1.0	0.4	
Isoeugenol	1.0	0.4	
<i>Evernia prunastri</i> (oakmoss absolute)	1.0	0.4	
Sesquiterpene lactone mix	0.1	0.04	n.a.
Alantolactone	0.033	0.013	
Dehydrocostus lactone and costunolide	0.067	0.027	
Quaternium-15 (Dowicil 200)	1.0	0.4	0.10
Primin	0.01	0.004	n.a.
Methylchloroisothiazolinone and methylisothiazolinone (Kathon CG TM , 100 p.p.m.)	0.01 ^a	0.003	0.004
Budesonide	0.01	0.004	0.001
Tixocortol pivalate	0.1	0.04	0.003
Methyldibromo glutaronitrile	0.5	0.2	n.a.
Fragrance mix 2	14.0	5.6	n.a.
Hydroxyisohexyl 3-cyclohexene carboxaldehyde	2.5	1.0	
Citral	1.0	0.4	
Farnesol	2.5	1.0	
Coumarin	2.5	1.0	
Citronellol	0.5	0.2	
α -hexyl cinnamal (α -hexyl innamaldehyde)	5.0	2.0	
Hydroxyisohexyl 3-cyclohexene carboxaldehyde	5.0	2.0	n.a.

n.a., not available; PTD, dipentamethylenethiuram disulfide; TETD, tetraethylthiuram disulfide; TMTD, tetramethylthiuram disulfide; TMTM, tetramethylthiuram monosulfide.

*Calculations based on the use of the Finn chamber ($\varnothing$ 0.8 cm) technique with application of 20 mg pet. preparation or where appropriate (in water) 15 μ l aqueous test solution (30).

^aIn water.

^bEqual parts of TMTM, TMTD, TETD, and PTD.

^cBenzocaine, tetracaine hydrochloride, dibucaine hydrochloride (5:1:1).

^dClioquinol, chlorquinaldol (1:1).

^eMethyl-4-hydroxybenzoate, ethyl-4-hydroxybenzoate, propyl-4-hydroxybenzoate, butyl-4-hydroxybenzoate, benzyl-4-hydroxybenzoate (1:1:1:1:1).

^f*N*-isopropyl-*N*'-phenyl-4-phenylenediamine, *N*-cyclohexyl-*N*'-phenyl-phenylenediamine, *N,N'*-diphenyl-4-phenylenediamine (2:5:5).

^g*N*-cyclohexylbenzothiazyl sulfenamide, dibenzothiazyl disulfide, morpholinylmercaptobenzothiazole (1:1:1).

^hEmulsifier: Sorbitan sesquioleate 5%.

ⁱCinnamic alcohol, cinnamic aldehyde, hydroxycitronellal, α -amylcinnamic aldehyde, geraniol, eugenol, isoeugenol, oakmoss absolute (4:2:4:1:5:2:1:5).

contact allergy to the substance, no association was found between hand eczema and contact allergy to it (24). In an experimental repeated open application test on the forearms of 18 hydroxyisohexyl 3-cyclohexene carboxaldehyde-hypersensitive patients and 7 controls, 89% of the patients tested positively, while none of the controls reacted (25).

Hydroxyisohexyl 3-cyclohexene carboxaldehyde has been demonstrated in deodorants and perfumes at concentrations up to 0.2% (26, 27). In a recent usage test simulating deodorant use, 15 hydroxyisohexyl 3-cyclohexene carboxaldehyde-hypersensitive patients and 10 healthy controls applied unscented and hydroxyisohexyl 3-cyclohexene carboxaldehyde-scented deodorants in the axillae. None of the controls developed dermatitis, while all 14 patients who completed the study developed axillary dermatitis at common hydroxyisohexyl 3-cyclohexene carboxaldehyde concentrations in the deodorants (28). Currently, there is an International Fragrance Association restriction on the use of hydroxyisohexyl 3-cyclohexene carboxaldehyde, and the recommended maximal concentration is 1.5% (29). Hydroxyisohexyl 3-cyclohexene carboxaldehyde is 1 of the 26 fragrances to be labelled on scented products according to EU legislation. Hydroxyisohexyl 3-cyclohexene carboxaldehyde in pet. at 5.0% w/w fulfils the requirement of a contact sensitizer to be included in a baseline series (1).

Conclusions

Fragrance mix 2 in pet. at 14% w/w (5.6 mg/cm²) and hydroxyisohexyl 3-cyclohexene carboxaldehyde in pet. at 5% w/w (2.0 mg/cm²) are recommended for inclusion in the European baseline series of contact allergens (Table 1). With the Finn Chamber[®] technique, a dose of 20 mg pet. preparation is recommended (30). For the aqueous solutions in the baseline series (Table 1), the recommended volume is 15 µl (30). Whenever there is a positive reaction to fragrance mix 2, additional patch testing with the 6 ingredients, 5 if there are simultaneous positive reactions to hydroxyisohexyl 3-cyclohexene carboxaldehyde and fragrance mix 2, is recommended. Specified knowledge of the contact sensitizer will help heal a present dermatitis and prevent relapses of allergic contact dermatitis as exposure to the 6 fragrance substances in fragrance mix 2 can be avoided as they have to be labelled on cosmetics according to EU legislation. As for other patch test preparations, these preparations should be subjected to constant considerations with regard to modifications needed and justified by new information, such as changes in rates of contact

allergy, allergic contact dermatitis, and adverse reactions, particularly patch test sensitization (1).

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