

Multicentre patch testing with methylisothiazolinone by the European Environmental and Contact Dermatitis Research Group

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doi:10.1111/cod.12220

Key words: adverse reactions; allergic contact dermatitis; contact allergy; dose in $\mu\text{g}/\text{cm}^2$; Kathon CG®; MCI/MI; methylisothiazolinone; micropipette; patch test sensitization; preservative.

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Conflicts of interest: The authors declare no conflict of interests.

Methylisothiazolinone (MI) without methylchloroisothiazolinone has been marketed since early 2000, first in industrial products such as paints and water-based glues, and later also in cosmetics, with a recommended concentration in leave-on products in the range 50–100 ppm.

In the early 2000s, the optimal patch test concentration for MI was not known, which prompted the present study within the European Environmental and Contact Dermatitis Research Group. This contact point is aimed at describing the background on which MI 2000 ppm was recommended to be included in the European baseline series (1).

Eight European dermatology clinics and one US dermatology clinic participated in the study: Amsterdam, The Netherlands; Coimbra, Portugal; Copenhagen (Gentofte), Denmark; Leuven, Belgium; London, United Kingdom; Malmö, Sweden; Odense, Denmark; San Francisco, USA; and Strasbourg, France. The study was conducted over a period of ~9 months from 2006 to 2007.

Three test preparations of MI were prepared from Neolone 950, containing 9.5% MI, at the time kindly supplied by Rohm and Haas (Philadelphia, PA, USA). From a stock solution of MI, aqueous solutions of MI at 1000, 1500 and 2000 ppm were prepared at the Department of Occupational and Environmental Dermatology in Malmö and sent to participating clinics.

Testing was performed with Finn Chambers® (diameter, 8 mm) (Epitest Oy, Tuusula, Finland) on Scanpor® tape (Norgesplaster A/S, Vennesla, Norway) in Coimbra, Gentofte, London, Malmö, Odense, and San Francisco, with van der Bend® Chambers (Brielle, The Netherlands) in Amsterdam and Leuven, and with IQ Chambers® (Chemotechnique Diagnostics, Vellinge, Sweden) in Strasbourg. Volumes of 15 and 20 µl, respectively, were micropipetted onto the filter paper disc in the Finn Chambers® and van der Bend Chambers®, respectively, giving doses of 30, 45 and 60 µg/cm². The corresponding volumes giving the same doses were used for the IQ Chambers®. Patch testing and reading days were in accordance with the routines of the participating clinics. Reading days differed, and for reporting of results we used D3 or D4 readings. Evaluations of the morphology of the patch test results were performed according to the recommendations of the International Contact Dermatitis Research Group (2). Reactions were judged as doubtful when the morphological features of the reaction were consistent with an allergic nature, but when the minimal criteria for an allergic reaction, that is, erythema and infiltration, were not present on the whole test area. Reactions were judged as irritant when they lacked the morphological features consistent with a reaction of an allergic nature. The numbers of consecutively tested dermatitis patients tested with the three test preparations are shown in Table 1.

The numbers of positive reactors for the respective MI concentrations were compared pairwise by

Table 1. Results of patch testing with methylisothiazolinone (MI) in the baseline series within the European Environmental and Contact Dermatitis Research Group. Three thousand one hundred and eighty-six (1 317 males; 1 694 females) patients were tested with MI 1000 ppm, 2820 (954 males; 1 691 females) were tested with MI 1500 ppm, and 1915 (716 males; 1024 females) were tested with MI 2000 ppm*

Participating centres	MI 1000 ppm					MI 1500 ppm					MI 2000 ppm				
	Positive males† (%)	Positive females† (%)	Total tested (% positive)	IR‡	γ§	Positive males† (%)	Positive females† (%)	Total tested (% positive)	IR‡	γ§	Positive males† (%)	Positive females† (%)	Total tested (% positive)	IR‡	γ§
Amsterdam	0	1 (0.6)	420	1	1	0	0	298	1	1	3 (1.1)	6 (3.8)	420	1	0
Strasbourg	1 (0.8)	0	316	1	1	0	0	316	0	1	1 (2.1)	1 (1.3)	124	0	0
Gentofte	1 (0.7)	1 (0.3)	436	5	5	1 (0.6)	2 (0.7)	442	3	7	1 (1.0)	3 (1.4)	315	7	6
Coimbra	2 (3.6)	3 (2.1)	196	0	0	3 (5.4)	3 (2.1)	196	1	3	2 (5.6)	2 (2.5)	116	2	7
Odense	0	1 (0.8)	224	6	6	2 (1.9)	3 (2.5)	224	0	17	0	0	61	5	1
Leuven	1 (0.7)	0	387	3	3	0	0	308	0	3	0	0	218	3	0
Malmö	0	3 (0.7)	697	10	10	0	3 (0.9)	526	0	7	1 (0.8)	3 (1.5)	329	5	0
London	1 (1.0)	0	335	2	2	1 (1.0)	1 (0.4)	335	0	2	1 (2.6)	0	157	1	0
San Francisco*	0	0	175	0	0	0	0	175	0	0	1	0	175	0	0
Total	6 (0.5)	9 (0.5)	3186 (0.5)	28 (0.9)	28	8 (0.8)	12 (0.7)	2820 (0.7)	5 (0.2)	41 (1.5)	10 (1.3)	15 (1.5)	1915 (1.3)	24 (1.3)	14 (0.7)

*The 175 patients from San Francisco are excluded from the male/female percentage calculations for the San Francisco row and in the 'total' row, because they were not retrievable.

†The number of positive males and percentage of tested males in parentheses.

‡The number of positive females and percentage of tested females in parentheses.

§The number of doubtful reactions.

¶The number of irritant reactions.

the use of McNemar's two-sided test. A p -value of <0.05 was considered to be statistically significant. Possible sex differences for the respective concentrations were investigated with Fisher's exact test, two-sided.

The results for the dermatitis patients routinely patch tested in the nine participating clinics are shown in Table 1. In this study, 15 of 3186 patients (0.5%) were diagnosed with contact allergy to MI by patch testing with 1000 ppm, 20 of 2820 patients (0.7%) were diagnosed by patch testing with 1500 ppm, and 25 of 1915 patients (1.3%) were diagnosed by patch testing with 2000 ppm. Doubtful reactions were noted in 0.9% of those tested with MI 1000 ppm, in 1.5% of those tested with MI 1500 ppm, and in 1.3% of those tested with MI 2000 ppm. Few cases of irritant reactions were noted (0.2% for 1500 ppm and 0.7% for 2000 ppm), and no cases of patch test sensitization were observed. On comparison of the outcome of positive reactions for the respective patch test concentrations of MI, no statistical difference was found for 1000 versus 1500 ppm ($p > 0.3$), but statistical differences were found for 1000 versus 2000 ppm ($p = 0.0015$) and for 1500 versus 2000 ppm ($p = 0.0013$). There were almost no sex differences in contact allergy rates, independently of concentration.

Discussion

MI alone was introduced into industrial products in the early 2000s, and in 2005 it was allowed as a preservative in cosmetics (3), with its use increasing since then (4, 5). Even though selected cases of MI contact allergy had been published by then, there was no general recommendation on the ideal patch test preparation for MI.

In this study, it was initially decided to start testing with MI at 1000 ppm, as the department in Malmö had experience with patch testing with MI alone in both humans and animals from the mid-1980s, and before this study started the Malmö clinic had patch tested MI at 475 ppm without adverse reactions in consecutively patch tested dermatitis patients (6). To define an ideal patch test concentration, that is, a patch test concentration that will elicit as many cases of contact allergy as possible without causing any adverse reactions, such as irritation and patch test sensitization, it is necessary to start with a concentration that is safe. This concentration should then be raised stepwise until adverse reactions appear or until a maximal concentration decided in advance has been reached (7). The stepwise increase in patch test concentration of MI, that is, starting

with 1000 ppm and, when no serious adverse reactions are noted, raising the concentration to 1500 ppm and eventually to 2000 ppm, was in line with this. Also testing with two or more concentrations simultaneously gave us a chance to compare test concentrations. The maximum concentration of 2000 ppm was based on experience with other preservatives, suggesting that up to a 20-fold higher concentration than the use concentration in cosmetics can be used for patch testing (8). As we knew that MI at the maximal allowed concentration of 100 ppm would soon be introduced into cosmetics in Europe, we decided to use a test concentration as high as 2000 ppm (8).

Initially, we also considered using petrolatum test preparations of MI, but chemical investigations showed that MI could not be evenly distributed in the pet. test preparations (9). Therefore, only aqueous tests were prepared.

Patch testing MI at 2000 ppm detected the highest frequencies of contact allergy (1.3%), and there were no reports of patch test sensitization. Of the 60 positive reactions to MI, 24 were in males and 36 in females; that is, 50% more females had positive reactions. The sex distribution between males and females was similar to the proportion of males and females being patch tested.

Some reactions were scored as doubtful (0.9–1.5%). One patient with a reaction scored as doubtful when tested with 1000 ppm had a positive reaction to 1500 ppm tested simultaneously. Four patients with a reaction scored as doubtful when tested with 1500 ppm were positive with 2000 ppm. This spoke in favour for the doubtful reactions being weak contact allergies to MI and not irritant reactions.

Few reactions were scored as irritant, and these only occurred when MI 1500 and 2000 ppm was tested. A total of 7921 MI patch tests were applied to the eczema patients. Only 19 tests were scored as irritant, that is, 0.2% of all tests. Concerning both MI 1500 and 2000 ppm, only three of nine clinics noted irritant reactions.

We concluded that MI can be tested at 2000 ppm without any adverse events such as patch test sensitization, and that the dose is accurately dispensed, that is, that a micropipette is used when testing with this preservative, which is always tested as an aqueous solution.

Acknowledgements

The study was financially supported by the former company Rohm and Haas Company, Philadelphia, PA, USA.

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