

Patch testing with components of water-based metalworking fluids: results of a multicentre study with a second series

JOHANNES GEIER¹, HOLGER LESSMANN¹, DETLEF BECKER², MAGNUS BRUZE³, PETER J. FROSCH⁴, THOMAS FUCHS⁵,
UTA JAPPE⁶, PATRICK KOCH⁷, CLAUDIA PFÖHLER⁷ AND CHRISTOPH SKUDLIK⁸

¹Information Network of Departments of Dermatology (IVDK), University of Göttingen, 37075 Göttingen, Germany,

²Department of Dermatology, University of Mainz, SS101 Mainz, Germany, ³Department of Occupational and Environmental Dermatology, Lund University, 20502 Malmö, Sweden, ⁴Department of Dermatology, Klinikum Dortmund and University of Witten/Herdecke, 44137 Dortmund, Germany, ⁵Department of Dermatology, University of Göttingen, 37075 Göttingen, Germany, ⁶Department of Dermatology, University of Heidelberg, 69115 Heidelberg, Germany, ⁷Department of Dermatology, University of Saarland, 66421 Homburg/Saar, Germany, and ⁸Department of Dermatology, Environmental Medicine, and Health Theory, University of Osnabrück, 49069 Osnabrück, Germany

Background: Although many allergens in metalworking fluids (MWF) are identified, there are still some MWF components, which are not sufficiently investigated concerning their sensitizing properties.

Objectives: To investigate sensitization to 10 frequently used MWF components, which are not part of the established MWF test series, in metalworkers with suspected occupational dermatitis due to MWF.

Patients/Methods: Oleyl alcohol, myristyl alcohol, dimethylolurea, 4,4'-methylenebis morpholine, imazalil, 1-amino-2-propanol (monoisopropanolamine; MIPA), 2-amino-2-ethyl-1,3-propanediol (AEPD), 2,5-bis(n-octyldithio)-1,3,4-thiadiazole, zinc alkyl dithiophosphate and dibenzyl disulfide have been patch tested in 144 patients.

Results: 7 patients reacted positively to the formaldehyde releaser 4,4'-methylenebis morpholine, and 6 of these patients also reacted to formaldehyde and/or other formaldehyde releasers. 4 patients reacted positively to myristyl alcohol tested at 10% petrolatum (pet.). Additionally, 20 doubtful or irritant reactions occurred. 1 patient each reacted positively to oleyl alcohol, MIPA, and AEPD. None of the other test substances mentioned above elicited any clear-cut positive reaction. Patch testing with well-known MWF allergens showed proportions of positive reactions, which were comparable to those from other studies, e.g. 11% to monoethanolamine, 8% to colophonium and 3%–5% to various preservatives.

Conclusions: 4,4'-methylenebis morpholine may be an important MWF allergen, although clinical relevance could not be stated definitely in every case. Myristyl alcohol should not be patch tested at 10% pet., but at a lesser concentration, due to irritant properties.

Key words: 2-amino-2-ethyl-1,3-propanediol (AEPD); 1-amino-2-propanol (monoisopropanolamine; MIPA); contact allergy; formaldehyde; metalworking fluids; 4,4'-methylenebis morpholine; myristyl alcohol; occupational; patch testing. © Blackwell Munksgaard, 2006.

Accepted for publication 4 September 2006

Metalworking fluids (MWF) have a complex composition, commonly based on mineral oils or (semi) synthetic hydrocarbon compounds. A broad variety of admixtures such as emulsifiers, solubility enhancers, lubricants, stabilizers, corrosion inhibitors and biocides (bactericides and fungicides) are usually added, according to the respective needs (1–6). A lot of MWF components are known to be skin sensitizers. Based on infor-

mation from the lubricant industry, an interdisciplinary task force on allergy diagnostics in the metalworking branch has published 2 lists of current and former allergens in MWF that are commercially available as patch test substances (3). The German Contact Dermatitis Research Group (DKG) has adopted these allergen panels in their MWF test series. Nevertheless, there are potential allergens in MWF that remain undetected in spite

of patch testing with these series. Therefore, we had tested additional MWF components suspected to have an allergenic potential in metalworkers with occupational dermatitis from 2000 to 2002 (7). As a result of this study, we detected sensitizations to diglycolamine, which meanwhile has been confirmed to be an important MWF allergen by further investigations (8). From June 2004 to June 2005, we continued our studies by patch testing with a second panel of potentially allergenic MWF components that are not commercially available as patch test substances.

Patients, Materials and Methods

In all, 144 metalworkers exposed to MWF, who were patch tested for suspected occupational dermatitis, were included in the study. Of these, 54 were machine operators, 49 metalworkers (not nearer specified), 19 turners, 7 tool makers and 5 grinders. In addition, there were 10 former metalworkers with occupational dermatitis who had already changed their profession. 44 patients were tested in Dortmund, 37 in Malmö, 30 in Osnabrück and the remainder in Homburg/Saar, Mainz, Heidelberg and Göttingen. All German participants are members of the DKG and the Information Network of Departments of Dermatology (IVDK). Of the patients tested, 135 (94%) were males and 29 (20%) had a past or present atopic dermatitis. Hand dermatitis was present in 117 patients (81%), and in 111 patients (77%), a final diagnosis of occupational dermatitis was

stated. The age ranged from 19 to 62 years, with a mean age of 39.5 years.

For this second panel of MWF components, we selected 2 basic MWF components, 2 formaldehyde releasers, 1 fungicide, 2 corrosion inhibitors and emulsifiers, 1 copper corrosion inhibitor and 2 extreme pressure additives (see Table 1). 2 of these substances, i.e. 1-amino-2-propanol (mono-isopropanolamine; MIPA) and 2-amino-2-ethyl-1,3-propanediol (AEPD), had been patch tested at a lower concentration in our first patch test study with MWF components (7). At that time, no reactions to MIPA 1% petrolatum (pet.), and only 1 weak positive reaction to AEPD 1% aqueous (aq.) had been observed (7). In analogy to monoethanolamine (MEA) and diethanolamine (DEA), we increased patch test concentration of both, MIPA and AEPD, to 2% pet. in this study. Test concentrations of the other substances were chosen according to published recommendations (9–15) or by analogy to test concentrations recommended for chemically related substances. Patch test preparations were made at a pharmacy in Göttingen, Germany, from chemicals bought from Sigma-Aldrich, Schnelldorf, Germany (7 substances), and ABCR, Karlsruhe, Germany (4,4'-methylenebis morpholine). Imazalil was a gift from Janssen Pharmaceutica, Beerse, Belgium. Zinc alkyl dithiophosphate 6% pet. was prepared from Lubrizol 1375[®], Lubrizol France, Paris, France. Purity of the raw materials was 95%–100%, except for zinc alkyl dithiophosphate (85%) and oleyl alcohol (83%).

Table 1. Patch test reactions to MWF components in 144 patients

Substance [function in MWF (reference)]	CAS no.	Test concentration* (% pet.)	–	?	+	++	ir	Percent positive
Oleyl alcohol [basic MWF component; solubility aid (9, 10)]	143-28-2	10	138	5	1	0	0	0.7
Myristyl alcohol [basic MWF component; solubility aid (11, 12)]	112-72-1	10	120	16	4	0	4	2.8
Dimethylolurea (biocide; formaldehyde releaser)	140-95-4	1	143	1	0	0	0	0.0
4,4'-Methylenebis morpholine [biocide; formaldehyde releaser (21, 22)]	5625-90-1	1	133	4	4	3	0	4.9
Imazalil [fungicide (22, 32)]	35554-44-0	2	143	1	0	0	0	0.0
1-Amino-2-propanol (mono-isopropanolamine) [corrosion inhibitor; emulsifier (7)]	78-96-6	2	139	4	1	0	0	0.7
2-Amino-2-ethyl-1,3-propanediol [corrosion inhibitor; emulsifier (7, 31)]	115-70-8	2	138	4	1	0	1	0.7
2,5-Bis(n-octyldithio)-1,3,4-thiadiazole [copper corrosion inhibitor; hydrogen sulfide suppressor (33)]	13539-13-4	2	143	1	0	0	0	0.0
Zinc alkyl dithiophosphate [extreme pressure additive in neat oils; ethylhexylzinc dithiophosphate (13–15)]	68649-42-3	6	144	0	0	0	0	0.0
Dibenzyl disulfide (extreme pressure additive; antioxidant)	150-60-7	1	143	1	0	0	0	0.0

MWF, metalworking fluids; pet., petrolatum; ir, irritant reaction.

*% indicates w/w %.

Routine patch testing (standard series, MWF series etc.) in the German centres was done using patch test preparations from Hermal, Reinbek, Germany. In Malmö, Chemotechnique patch test allergens (Tygelsjö, Sweden) and test substances prepared at the department of occupational and environmental dermatology in Malmö were used. Patch test exposure time was 2 days in 130 patients (90%) and 1 day in 14 patients (10%). Patch tests were performed and read according to internationally accepted guidelines (16, 17). For data analysis, generally patch test results at day (D) 3 were selected. In those patients, who did not return for patch test reading at D3 but returned at D4, the latter reading was evaluated.

Data processing at the German centres was done according to the IVDK routine procedure (18). Data from Malmö were transferred anonymously on printed case record forms to the IVDK data centre in Göttingen, Germany, where it was entered into an electronic database and then processed like data from the other participants. For data analysis, the statistical program system SAS (SAS Institute, Cary, NC, USA), version 8.2, was employed.

Results

Patch test results of the MWF components of this second panel are depicted in Table 1. Positive reactions to 4,4'-methylenebis morpholine were observed in 7 patients (4.9%); in Malmö, 3 strong and 2 weak positive reactions were noted, and another 2 weak positive reactions were recorded in Dortmund. In none of these cases, a clinical relevance of these positive reactions in terms of presence of this formaldehyde-releasing biocide in the patient's MWF could be confirmed. However, 6 of the 7 patients with a positive reaction to 4,4'-methylenebis morpholine reacted positively to formaldehyde and/or other formaldehyde releasers too. Additionally, some erythematous reactions occurred (see Table 2).

To myristyl alcohol, 4 patients (2.8%) reacted positively, and another 20 patients showed doubtful or irritant reactions (see Table 1). All the positive reactions, of which none was stronger than a palpable erythema, were interpreted as irritant and not allergic. 1 patient each reacted positively to oleyl alcohol, MIPA and AEPD, respectively. Additionally, to all of these 3 substances, doubtful or irritant reactions occurred in 4 to 5 patients, respectively. To all other substances from our panel, no clear-cut positive reactions were noted, and only a few single erythematous, doubtful reactions occurred.

Tables 3 and 4 show the patch test results to current and former MWF allergens, respectively, of which test preparations are commercially available. As the test series defined by the DKG (8) were tested in the German centres only and the allergen panel tested in Malmö was slightly different, not every single allergen was tested in every single patient. Additionally, in some patients, patch testing with the standard series or the MWF series was omitted due to recent pre-testing by the referring physician. Among the current MWF allergens, reactions to colophonium and MEA were observed most frequently (11 patients each). MEA was tested in the German centres only, leading to a reduced number of patients tested ($n = 99$). Hence, the percentage of positive reactions to MEA 2% pet. was 11.1%. Of the 11 patients reacting to colophonium, 9 also reacted to abietic acid. No reactions to abietic acid without reaction to colophonium occurred. Taken together, 140 patients were patch tested with colophonium and/or abietic acid, and the share of positive reactions to either was 7.9%. Additionally, positive reactions to current MWF allergens were mainly to biocides, ranging from 5.0% for 4,4-dimethyl-1,3-oxazolidine/3,4,4-trimethyl-1,3-oxazolidine (Bioban CS 1135) to 0.7% for sodium-2-pyridinethiol-1-oxide (sodium omdine) and iodopropynyl butylcarbamate, if all

Table 2. Concomitant reactions to formaldehyde or formaldehyde releasers in 6 patients reacting to 4,4'-methylenebis morpholine

Allergen	Patch test concentration	?	+	++
Formaldehyde	1% aq.	3	2	
4,4-Dimethyl-1,3-oxazolidine/3,4,4-trimethyl-1,3-oxazolidine (Bioban CS 1135)	1% pet.		3	1
7-Ethylbicyclooxazolidine (Bioban CS 1246)	1% pet.	1	2	1
1,3,5-Tris(2-hydroxyethyl)-hexahydrotriazine (Grotan BK [®])	1% aq.	1	2	1
N,N'-Methylene-bis-5-methyl-oxazolidine	1% pet.	1	1	
Benzylhemiformal	1% pet.		1	
4-(2-Nitrobutyl)-morpholine/4,4'-(2-ethyl-2-nitromethylene)-dimorpholine (Bioban P 1487)	0.5% pet.	1		
Quaternium 15	1% pet.		4	
Diazolidinyl urea	2% pet.		1	

aq., aqueous; MWF, metalworking fluids; pet., petrolatum.

Table 3. Positive test reactions to current MWF allergens (1, 8)

Allergen	Patch test concentration	Patients tested	Patients with positive reaction	Percent positive
Standard series				
Colophonium	20% pet.	134	11	8.2
Mercaptobenzothiazole	2% pet.	134	4	3.0
MCI/MI	0.01% aq.	98	4	4.1
MCI/MI	0.02% aq.	37	0	0.0
Formaldehyde	1% aq.	135	2	1.5
Amerchol L 101	50% pet.	37	1	2.7
Cetearyl alcohol	20% pet.	97	0	0.0
Zinc diethyldithiocarbamate	1% pet.	97	0	0.0
Lanolin alcohol	30% pet.	99	0	0.0
MWF series (current allergens)				
Monoethanolamine	2% pet.	99	11	11.1
Abietic acid	10% pet.	137	9	6.6
4,4-Dimethyl-1,3-oxazolidine/3,4,4-trimethyl-1,3-oxazolidine (Bioban CS 1135)	1% pet.	139	7	5.0
Diglycolamine	1% pet.	137	5	3.6
7-Ethylbicyclooxazolidine (Bioban CS 1246)	1% pet.	139	4	2.9
1,2-Benzisothiazolinone, sodium salt	0.1% pet.	103	4	3.9
1,2-Benzisothiazolinone, sodium salt	0.05% pet.	37	0	0.0
N,N'-Methylene-bis-5-methyl-oxazolidine	1% pet.	102	3	2.9
1,3,5-Tris(2-hydroxyethyl)-hexahydrotriazine (Grotan BK [®])	1% pet.	99	0	0
1,3,5-Tris(2-hydroxyethyl)-hexahydrotriazine (Grotan BK [®])	1% aq.	37	3	8.1
Diethanolamine	2% pet.	100	2	2.0
p-tert-Butylphenol	1% pet.	100	1	1.0
Benzylhemiformal	1% pet.	102	1	1.0
Sodium-2-pyridinethiol-1-oxide (sodium omadine)	0.1% aq.	135	1	0.7
IPBC	0.2% pet.	101	1	1.0
IPBC	0.1% pet.	37	0	0.0
2-Phenoxyethanol	1% pet.	100	0	0.0
Benzotriazole	1% pet.	110	0	0.0
Octylisothiazolinone	0.025% pet.	102	0	0.0
Octylisothiazolinone	0.01% pet.	37	0	0.0
Ointment base series				
Polyethylene glycol ointment base	Pure	83	3	3.7
Propylene glycol	20% aq.	82	3	3.7
Propylene glycol	5% pet.	37	0	0
Butylhydroxy toluene	2% pet.	86	0	0.0
TEA	2.5% pet.	84	0	0.0
TEA	2% pet.	37	0	0.0
Preservative series				
Triclosan	2% pet.	114	0	0.0

aq., aqueous; IPBC, iodopropynyl butylcarbamate; MCI/MI, 5-chloro-2-methylisothiazolin-3-one/2-methylisothiazolin-3-one; MWF, metalworking fluids; pet., petrolatum; TEA, triethanolamine.

concentrations tested were taken into account. Remarkably, contact allergy rates to 1,3,5-tris(2-hydroxyethyl)-hexahydrotriazine (Grotan BK) differed significantly between Malmö and the German centres (Fisher's exact test; $P = 0.019$). While 3 out of 37 patients (8.1%) reacted to the 1% aq. test preparation in Malmö, no patient (out of 99 tested in Germany) reacted to the 1% pet. preparation. 4 patients (3.0%) had a positive reaction to mercaptobenzothiazole (MBT), and all of these reactions were regarded clinically relevant. In 3 of these cases, however, rubber gloves or rubber shoes were noted as allergen source. To diglycolamine, 5 patients (3.6%) reacted positively, while only 2 patients (2.0%) reacted to DEA. Unfortunately, in none of these cases, information could be obtained as to whether the

corresponding amine was present in the patients' MWF. No reactions were seen with triethanolamine, the preservatives 2-phenoxyethanol, octylisothiazolinone, triclosan and benzotriazole.

Among the former MWF allergens, methyldibromoglutaronitrile (MDBGN) 0.3% pet. elicited positive reactions most frequently (see Table 4). However, in none of these cases, a MWF was the suspected (or even confirmed) allergen source. Fragrance mix and *Myroxylon pereirae* resin (balsam of Peru) elicited positive reactions in 5 and 4 patients, respectively. As with MDBGN, no MWFs were mentioned as allergen sources in these cases. 5 patients reacted positively to morpholinyl mercaptobenzothiazole, and of these, 2 also reacted to MBT. While all of these reactions were regarded clinically relevant, rubber gloves and

Table 4. Positive test reactions to standard series and former MWF allergens (1, 8)

Allergen	Patch test concentration (% pet.)	Patients tested	Patients with positive reaction	Percent positive
Standard series				
MDBGN	0.3	127	8	6.3
MDBGN/phenoxyethanol	1	69	2	2.9
Fragrance mix	8	133	5	3.8
<i>Myroxylon pereirae</i> resin (balsam of Peru)	25	134	4	3.0
Paraben mix	16	134	0	0.0
MWF series (former allergens)				
Morpholinyl mercaptobenzothiazole	0.5	83	5	6.0
Ethylenediamine dihydrochloride	1	110	3	2.7
Chlorocresol	1	115	1	0.9
Chloroxylonol	1	75	0	0.0
Chloroxylonol	0.5	37	0	0.0
Hexamethylenetetramine	1	71	0	0
Dipentene (d,l-limonene)	2	46	0	0
Dipentene (d,l-limonene)	1	37	0	0
2-Hydroxymethyl-2-nitro-1,3-propanediol (Tris Nitro)	1	101	0	0.0
4-(2-Nitrobutyl)-morpholine/4,4'-(2-ethyl-2-nitromethylene)-dimorpholine (Bioban P 1487)	1	91	0	0.0
4-(2-Nitrobutyl)-morpholine/4,4'-(2-ethyl-2-nitromethylene)-dimorpholine (Bioban P 1487)	0.5	37	0	0.0
Ointment base series				
Coconut diethanolamide	0.5	120	3	2.5
Preservative series				
Chloroacetamide	0.2	115	0	0.0
Bronopol (2-bromo-2-nitropropane-1,3-diol)	0.5	90	0	0.0
Bronopol (2-bromo-2-nitropropane-1,3-diol)	0.25	37	0	0.0

aq., aqueous; MDBGN, methyl dibromo glutaronitrile; MWF, metalworking fluids; pet., petrolatum.

rubber shoes were noted as relevant allergen sources in 3 of these patients. A clinical relevance of the 3 positive reactions to ethylenediamine dihydrochloride could not be stated definitely.

Discussion

Our second patch test study with MWF components not yet available as commercial patch test preparations points to 4,4'-methylenebis morpholine possibly being an important MWF allergen.

When interpreting the frequency of positive patch test reactions to the study substances, it is necessary to consider characteristics of the study population and the reaction frequencies obtained with well-known MWF allergens. Our study group consisted of 144 metalworkers, predominantly male (94%). The hands were affected most frequently (81%), and occupational dermatitis was eventually confirmed in 77% of them. The 2 most important MWF allergens, i.e. oxidation products of resin acids tested as colophonium, and MEA, elicited positive reactions in 8% and 11% of the patients tested, respectively. Together with the pattern of test reactions to other allergens of the commercially available MWF series, all these items are indicative for a 'normal' test population of metalworkers exposed to MWF with suspected occupational dermatitis, comparable to published studies (7, 8, 19). Hence, we largely

can exclude a selection bias towards particularly frequently or particularly rarely sensitized patients.

The most interesting results of this study are the positive patch test reactions to 4,4'-methylenebis morpholine in 7 of the 144 patients tested (4.9%). As the presence of this formaldehyde releaser could not definitely be confirmed in any of the MWFs used by these patients, the question of clinical relevance of these reactions raises. First, not in every case an in-depth investigation of the MWFs used at the patient's workplace was possible, especially with respect to additional preservation during the use of the MWF. Hence, 4,4'-methylenebis morpholine might have been present in the MWF although we did not know. Second, concomitant reactions to formaldehyde and/or other formaldehyde releasers occurred in 6 of the 7 patients. As the degree of formaldehyde release from 4,4'-methylenebis morpholine is pH dependent (20), it is presently difficult to tell whether the 4,4'-methylenebis morpholine reactions indicate formaldehyde allergy or not. However, because of the calculated low maximum formaldehyde concentration in the test preparation (about 0.16%), it seems improbable that positive reactions to 4,4'-methylenebis morpholine indicate formaldehyde allergy only. But the observed cluster of reactions points to a group of patients particularly sensitized to the

special group of formaldehyde-liberating biocides. According to the inventory of biocides used in Denmark published in 2001 (21), 4,4'-methylenebis morpholine is used as a biocide in MWF, with a concentration up to 3%. Probably, this is the case in other European countries too. Against this background, it seems likely that the positive reactions observed in our study indicated clinically relevant sensitizations. To our knowledge, up to now, no cases of contact allergy due to 4,4'-methylenebis morpholine have been reported in the literature. However, in 2003, Piebenga and van der Walle reported negative patch tests with 4,4'-methylenebis morpholine 0.3% aq. in 1 patient with contact dermatitis due to MWF and 4 controls (22).

In the report on sensitizing properties of active substances in biocidal products used in the Nordic countries, 4,4'-methylenebis morpholine is grouped into category 1 'no data available on sensitization' (23). However, Zissu could sensitize 10 of 20 animals in a guinea-pig maximization test and thus classified 4,4'-methylenebis morpholine as a moderate sensitizer (24). As 4,4'-methylenebis morpholine is among the notified substances in Annex II of the EC Commission Regulation No. 2032/2003 (25), it will be in use at least until October 2007. Hence, although we would not recommend to add 4,4'-methylenebis morpholine to the MWF test series for the time being, further allergological investigations with this formaldehyde releaser seem worthwhile.

The significant difference in contact allergy rates to Grotan BK throws another light on the difficulties of patch testing with formaldehyde releasers. It seems unlikely that this preservative is so much more in use in the metalworking plants in the Malmö area that this could explain the increased frequency of allergic reactions. In our opinion, a more likely explanation is that different batches and vehicles may have a different bioavailability, concentration and/or degree of degradation. A recent investigation with a different formaldehyde releaser, imidazolidinylurea, revealed great differences in formaldehyde and formaldehyde releaser concentration between aq. and pet.-based test preparations and between different batches, probably resulting in differing contact allergy rates (26).

In contrast, the positive reactions observed to myristyl alcohol 10% pet. probably are false-positive, irritant reactions. In all, weak positive reactions (erythema and infiltration) were elicited in 4 patients, but another 16 patients reacted with erythema only and 4 patients with a distinct irritant reaction. This would mean a reaction index of -0.7 and a positivity ratio of 100%, which is

a typical constellation of an irritant test preparation (27). Probably the test concentration chosen was too high. For myristyl alcohol, various patch test concentrations have been recommended, ranging from 5% pet. to 20% pet. (9). de Groot et al. reported on 3 patients with contact allergy due to myristyl alcohol from cosmetics (12). Their patients reacted to myristyl alcohol 5% pet., but they performed control tests with myristyl alcohol 5% pet. in 60 patients, 10% and 20% pet. in 25 additional patients, and only 1+ reaction was observed to myristyl alcohol 20% pet. while the other tests remained negative (12). Pecegheiro et al. had tested myristyl alcohol at 20% pet., while Tosti et al. preferred 5% pet. (11, 28). Based on these data, we decided to patch test myristyl alcohol at 10% pet. in our study. However, with the reactions observed, we now would recommend a lower test concentration, e.g. 5% pet., for future investigative patch testing. Prompted by the first test results, 2 centres started to patch test myristyl alcohol 10% pet. in addition to the standard series. The results of this investigation will be published separately.

With oleyl alcohol 10% pet., far fewer reactions were observed than that with myristyl alcohol. However, while only 1 patient had a weak positive reaction, erythematous, doubtful reactions occurred in 5 patients. This may indicate too high a test concentration, with irritant erythematous reactions, or too low a test concentration, with only weak reactions in truly sensitized patients. In the literature, 10% pet. as well as 30% pet. are recommended for patch testing with oleyl alcohol, and no overly irritating properties of the higher test concentration have been reported (9, 11, 29). Hence, it might be that we missed some clinically relevant sensitizations to oleyl alcohol by testing at 10% pet. However, as we were not able to find out if oleyl alcohol was present in the MWFs used by the 6 patients with non-negative reactions, we cannot definitely answer this question.

Similar to our first test series, only single positive reactions occurred to MIPA and AEPD, in spite of the increased patch test concentration. With 2% pet., both have now been tested at the same concentration as that of MEA and DEA. We would not recommend to increase the patch test concentration further, as the reaction pattern of both test substances with 4 erythematous reactions each points to an increased irritancy of these preparations. It seems that MIPA and AEPD are far less important allergens in MWFs than MEA or DEA. This may be due to a low sensitizing capacity or a limited usage of these amines in MWF. In a modified Draize test, none of the 150 volunteers could be sensitized by MIPA,

applied as 2% aq. solution (30). AEPD was tested in guinea pigs according to the method by Landsteiner and Jacobs and in a Buehler test. In both settings, AEPD was not sensitizing (31). Hence, it may be true that these amines are no or only weak sensitizers. We have no reliable information about the quantitative usage of MIPA and/or AEPD in MWF.

All other substances of our test panel elicited no clear-cut positive reactions. Some of them have been reported as MWF allergens in case reports before, e.g. imazalil (22). Others, such as dimethylolurea and dibenzyl disulfide, have not been tested before in metalworkers exposed to MWF and with suspected occupational dermatitis. The extreme pressure additive ethylhexylzinc dithiophosphate has been reported to be a sensitizer in MWF in several cases (13–15); however, in our study, we could not find any patient reacting to zinc alkyl dithiophosphate, which is a chemically similar compound.

In conclusion, we would recommend to continue patch testing with 4,4'-methylenebis morpholine and to further investigate clinical relevance of positive reactions observed. Patch test concentration of myristyl alcohol should be reduced to 5% pet. However, as data on these potential MWF allergens still are sparse, it seems too early to test them routinely in a MWF test series. MIPA and AEPD should be tested only when indicated, and one should be aware that the test concentration of 2% pet. may be slightly irritating.

References

- Geier J, Lessmann H. Metalworking fluids. In: *Contact Dermatitis*, 4th edition, Frosch P J, Menné T, Lepoittevin J P (eds): Berlin Heidelberg, Springer, 2006: 569–581.
- Foulds I S. Cutting fluids. In: *Handbook of Occupational Dermatology*, Kanerva L, Elsner P, Wahlberg J E, Maibach H I (eds): Berlin Heidelberg, Springer, 2000: 691–700.
- Geier J, Lessmann H, Schumacher T et al. Vorschlag für die Epikutantestung bei Verdacht auf Kontaktallergie durch Kühlschmierstoffe. 1. Kommerziell erhältliche Testsubstanzen. *Dermatol Beruf Umwelt* 2000; 48: 232–236.
- Geier J, Lessmann H, Schmidt A, Englitz H-G, Schnuch A. Kontaktekzeme durch Kühlschmierstoffe in der Metallindustrie. *Akt Dermatol* 2003; 29: 185–194.
- Sonnenschein G. Kühlschmierstoffe. In: *Handbuch der Arbeitsmedizin*, Konietzko J, Dupuis H. (eds): 20. Ergänzungslieferung 5/98, Kap. IV-2.47.1, Landsberg, ecomed, 1998: 1–15.
- Tiedemann K-H, Zoellner G, Adam M, Becker D et al. Empfehlungen für die Epikutantestung bei Verdacht auf Kontaktallergie durch Kühlschmierstoffe. 2. Hinweise zur Arbeitsstofftestung. *Dermatol Beruf Umwelt* 2002; 50: 180–189.
- Geier J, Lessmann H, Frosch P J et al. Patch testing with components of water-based metalworking fluids. *Contact Dermatitis* 2003; 49: 85–90.
- Geier J, Lessmann H, Dickel H et al. Patch test results with the metalworking fluid series of the German Contact Dermatitis Research Group (DKG). *Contact Dermatitis* 2004; 51: 118–130.
- de Groot A C. *Patch Testing – Test Concentrations and Vehicles for 3700 Chemicals*, 2nd edition. Amsterdam, Elsevier, 1994.
- Koch P. Occupational allergic contact dermatitis from oleyl alcohol and monoethanolamine in a metalworking fluid. *Contact Dermatitis* 1995; 33: 273.
- Tosti A, Vinzenzi C, Guerra L, Andrisano E. Contact dermatitis from fatty alcohols. *Contact Dermatitis* 1996; 35: 287–289.
- de Groot A C, Bruynzeel D P, van Joost T, Weyland J W. Cosmetic allergy from myristyl alcohol. *Contact Dermatitis* 1988; 19: 76–77.
- Isaksson M, Frick M, Gruvberger B, Pontén A, Bruze M. Occupational allergic contact dermatitis from the extreme pressure (EP) additive, zinc, bis ((O, O'-di-2-ethylhexyl) dithiophosphate) in neat oils. *Contact Dermatitis* 2002; 46: 248–249.
- Kanerva L, Tupasela O, Jolanki R. Occupational allergic contact dermatitis from ethylhexylzinc dithiophosphate and fatty acid polydiethanolamide in cutting fluids. *Contact Dermatitis* 2001; 44: 193–194.
- Gruvberger B, Isaksson M, Frick M, Pontén A, Bruze M. Occupational dermatoses in a metalworking plant. *Contact Dermatitis* 2003; 48: 80–86.
- Wahlberg J E, Lindberg M. Patch testing. In: *Contact Dermatitis*, 4th edition, Frosch P J, Menné T, Lepoittevin J P (eds): Berlin Heidelberg, Springer, 2006: 365–390.
- Schnuch A, Aberer W, Agathos M, Brasch J, Frosch P J, Fuchs Th, Richter G; für die Deutsche Kontaktallergie-Gruppe. Leitlinien der Deutschen Dermatologischen Gesellschaft (DDG) zur Durchführung des Epikutantests mit Kontaktallergenen. *Hautarzt* 2001; 52: 864–866.
- Uter W, Schnuch A, Geier J, Frosch PJ. Epidemiology of contact dermatitis. The Berlin Heidelberg: Springer; 2006 Information Network of Departments of Dermatology (IVDK) in Germany. *Eur J Dermatol* 1998; 8: 36–40.
- Geier J, Lessmann H, Schnuch A, Uter W. Contact sensitizations in metalworkers with occupational dermatitis exposed to water-based metalworking fluids. Results of the research project "FaSt". *Int Arch Occup Environ Health* 2004; 77: 543–551.
- Bruze M, Gruvberger B, Geier J. A false negative chromotropic acid test to a formaldehyde-releasing biocide. In manuscript.
- Lassen C, Skarup S, Mikkelsen S H, Kjolholt J, Nielsen P J, Samsoe-Petersen L. *Inventory of Biocides Used in Denmark*. Environmental Project No. 585. Copenhagen, Danish Environmental Protection Agency, 2001.
- Piebenga W P, van der Walle H B. Allergic contact dermatitis from 1-[2-(2,4-dichlorophenyl)-2-(2-propenyloxy)ethyl]-1H-imidazole in a water-based metalworking fluid. *Contact Dermatitis* 2003; 48: 285–286.
- Ingemarsson B, Toft E, Clemenson C. *Sensitising Properties of Active Substances in Biocidal Products Used in the Nordic Countries*. TemaNord 2003:541. Copenhagen, Nordic Council of Ministers, 2003.
- Zissu D. The sensitizing potential of various biocides in the guinea pig maximization test. *Contact Dermatitis* 2002; 46: 224–227.
- EC Commission Regulation No. 2032/2003 of 4 November 2003. *Official Journal of the European Union*, L 307, 2003.
- Rietschel R L, Bruze M, Gruvberger B et al. The relationship of vehicle and concentration of imidazolidinylurea, with attention to formaldehyde allergy status. *Dermatitis* 2006; 17: 48–49.
- Geier J, Uter W, Lessmann H, Schnuch A. The positivity ratio – another parameter to assess the diagnostic quality of a patch test preparation. *Contact Dermatitis* 2003; 48: 280–282.

28. Pecegheiro M, Brandao M, Pinto J, Concalo S. Contact dermatitis to Hirudoid[®] cream. *Contact Dermatitis* 1987; 17: 290–293.
29. Guidetti M S, Vincenzi C, Guerra L, Tosti A. Contact dermatitis due to oleyl alcohol. *Contact Dermatitis* 1994; 31: 260–261.
30. Beyer K H Jr, Bergfeld W F, Berndt W O, Carlton W V, Hoffmann D K, Schroeter A L, Shanik R C. Final report on the safety assessment of diisopropanolamine, triisopropanolamine, isopropanolamine, and mixed isopropanolamine. *J Am Coll Toxicol* 1987; 6: 53–76.
31. Anonymous. 2-Amino-2-ethyl-1,3-propandiol. In: *Gesundheitsschädliche Arbeitsstoffe. Toxikologisch-arbeitsmedizinische Begründungen von MAK-Werten*, Greim H (ed.). 21. Lieferung. Weinheim, VCH, 1995: 1–6.
32. van Hecke E, de Vos L. Contact sensitivity to enilconazole. *Contact Dermatitis* 1983; 9: 144.
33. O'Driscoll J, Beck M, Taylor S. Occupational contact allergy to 2,5-dimercapto-1,3,4-thiadiazole. *Contact Dermatitis* 1990; 23: 268–269.

Address:
Johannes Geier, MD
Information Network of Departments
of Dermatology (IVDK)
University of Göttingen
von-Siebold-Str. 3
37075 Göttingen
Germany
Tel: ++49/(0) 551/39 89 84
Fax: ++49/(0) 551/39 60 95
e-mail: jgeier@ivdk.org