

INVESTIGATIVE REPORT

Contact Allergy to the Isomers of Diglycidyl Ether of Bisphenol F

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Diglycidyl ether of bisphenol A (DGEBA) is the monomer and main allergen in the most common epoxy resin, DGEBA-R. Epoxy resin(s) based on bisphenol F (DGEBF-R) contains the 3 isomers of the diglycidyl ether of bisphenol F. Contact allergy to these isomers (p,p'-DGEBF, o,p'-DGEBF and o,o'-DGEBF) has not previously been described in humans. To investigate contact allergies to the DGEBF isomers in patients with contact allergy to DGEBA-R, 9 patients were patch-tested with dilution series of the isomers. All 9 were found to have contact allergy to p,p'-DGEBF and 7/9 to o,p'-DGEBF. Additionally, of 20 workers employed in epoxy-based manufacture and with contact allergy to DGEBA-R and DGEBF-R, 18 reacted to p,p'-DGEBF and 15/20 to o,p'-DGEBF. According to a guinea pig maximization test, all 3 DGEBF isomers are strong sensitizers. Still, o,o'-DGEBF elicited only 3 positive reactions in all participants. **Key words:** bisphenol F; contact allergy; DGEBA-R; DGEBF-R; epoxy resin; epoxy resin of the bisphenol F-type; isomer.

(Accepted August 11, 2003.)

Acta Derm Venereol 2004; 84: 12–17.

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Epoxy resins are frequently reported occupational contact allergens (1). The most common resins are based on bisphenol A. The monomer of this type of resin is the diglycidyl ether of bisphenol A (DGEBA), which is also the most important contact allergen in epoxy resin(s) produced by the reaction of bisphenol A and epichlorohydrin (DGEBA-R) (2). DGEBA-R is used in most standard patch-test series. Epoxy resin(s) manufactured with methods similar to those of DGEBA-R, but produced by the reaction between bisphenol F and epichlorohydrin (DGEBF-R), are also frequently present in epoxy resin systems (3). Epoxy resin(s) can also be manufactured from phenolic novolac resins and epichlorohydrin (DGEBF(n)-R) (3). Several commercial DGEBF-R and DGEBF(n)-R have been analysed in our department and found to contain varying concentrations of monomers, which are the three isomers of diglycidyl ether of bisphenol F (DGEBF) (to

be published). The isomers are p,p'-DGEBF, o,p'-DGEBF and o,o'-DGEBF (Fig. 1). In the following, these 3 isomers, unless otherwise specified, are referred to as the DGEBF isomers. Epoxy resins, which contain the DGEBF isomers (DGEBF-R and DGEBF(n)-R) are referred to as epoxy resins of the bisphenol F-type. The production of DGEBA-R is designed to yield virtually only one isomer of DGEBA (p,p'-DGEBA) (Fig. 1) (3). The molecular weights (MW) of DGEBA and the DGEBF isomers are 340 and 312, respectively. In epoxy resin systems, DGEBA-R and DGEBF-R are sometimes mixed.

When consecutively patch-tested patients with dermatitis were tested with a DGEBF-R, it was found that contact allergy to DGEBF-R was at least as common as that to DGEBA-R (4). It was also found that 14 of 21 patients with contact allergy to DGEBF-R simultaneously reacted to DGEBA-R (4). However, contact allergy to the isolated, purified and chemically identified DGEBF isomers has not previously been described in humans, and it has not been known whether the frequency of contact allergy to the individual DGEBF isomers differs from one to the other and to DGEBA. In the present study, dermatitis patients with known contact allergy to both DGEBA-R and epoxy resin(s) of the bisphenol F-type were patch-tested with dilution series of p,p'-DGEBF, o,p'-DGEBF, o,o'-DGEBF and DGEBA. Furthermore, a group of workers exposed to

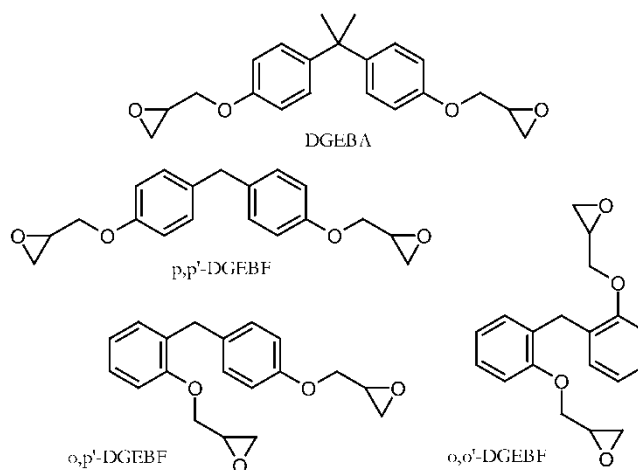


Fig. 1. DGEBA: diglycidyl ether of bisphenol A; p,p'-DGEBF: p,p'-diglycidyl ether of bisphenol F; o,p'-DGEBF: o,p'-diglycidyl ether of bisphenol F; o,o'-DGEBF: o,o'-diglycidyl ether of bisphenol F.

epoxy resin systems and diagnosed with contact allergy to DGEBA-R and epoxy resin(s) of the bisphenol F-type were patch-tested with p,p'-DGEBA, o,p'-DGEBA and o,o'-DGEBA.

MATERIALS AND METHODS

Study population

The study population was composed of 2 groups of individuals (groups A and B) with contact allergy to DGEBA-R and epoxy resin(s) of the bisphenol F-type. Group A consisted of 9 patients (4 men, 5 women, mean age 48.3 years, range 28–75) among consecutive patients with dermatitis investigated at our department and patch-tested with the standard patch-test series of the department, which also contained an epoxy resin of the bisphenol F-type.

Group B consisted of 20 workers (19 men, 1 woman, mean age 43.9 years, range 29–64) diagnosed with contact allergy to DGEBA-R and epoxy resin(s) of the bisphenol F-type. They participated in an investigation of occupational contact allergies performed on location at a Danish wind turbine factory with epoxy-based manufacture (to be published). The study was approved by the Ethics Committee of Lund University Medical Faculty (group A) and the Danish Scientific Research Ethics Committee (group B). Informed consent was obtained from each patient and worker.

Test substances

DGEBA, p,p'-DGEBA, o,p'-DGEBA and o,o'-DGEBA were isolated, purified and chemically identified at our laboratory, as described elsewhere (to be published). Analytical high pressure liquid chromatography (HPLC) was used to investigate the contamination of DGEBA, p,p'-DGEBA, o,p'-DGEBA and o,o'-DGEBA in each other (5). The patch-test preparation of DGEBA-R 1.0% in petrolatum was bought from Chemotechnique Diagnostics, Malmö, Sweden (groups A and B). The DGEBA-R (Rütapox 0161) was provided by Bakelite Gesellschaft mbH, Duisburg, Germany (group A) and by Shell Chemicals, Bromma, Sweden (Epicote 862) (group B). The DGEBA(n)-R (D.E.N. 431) was provided by Dow Epoxy Products, Stockholm, Sweden (groups A and B). In group B, the epoxy resins were patch-tested as part of an investigation of occupational contact allergies (to be published).

Patch-testing

The patch tests were left on the back for 48 h and scored according to the ICDRG criteria. Concentrations of the test substances in petrolatum and acetone are given in % w/w and % w/v, respectively.

The patients (group A) were patch-tested with dilution series of the individual DGEBA isomers and DGEBA. The dilution series were prepared from stock solutions in acetone of the respective substance in equimolar concentrations to DGEBA 1.0% w/v ($0.029 \text{ mol} \times 1^{-1}$) and DGEBA 0.92% w/v ($0.029 \text{ mol} \times 1^{-1}$) and 9 tenfold serial dilutions. Finn chambers (Epitest Ltd Oy, Tuusula, Finland) on Scanpor (Norgesplaster A/S, Vennessla, Norway) were used; 15 μl of the solutions was applied on a filter paper disc in each test chamber. The patch tests were read on D3 and D7. In the event of strong patch test reactions on D3, the area in question was treated with topical corticosteroids and the treated patch-test reaction was not further read.

The workers (group B) were patch-tested with IQ chambers (Chemotechnique Diagnostics, Malmö, Sweden). For practical reasons, petrolatum was used for the patch-test preparation within group B and no dilution series of the DGEBA isomers were tested. The patch tests with DGEBA-R (1.0% w/w), DGEBA-R (0.25% w/w) and DGEBA(n)-R (0.25% w/w) were read on D3 and D7. The patch tests with the DGEBA isomers (1.0% w/w) were read only on D3, because of the limited time period during which the investigation took place.

Controls

Twenty controls among consecutive patients with dermatitis were patch-tested with the individual DGEBA isomers. To minimize the risk of patch-test sensitization, the concentration was 0.1% w/v in acetone.

RESULTS

Patch test reactions

Group A. All patch test reactions are given in Table I. The numbers of reactors to the patch-test preparations in the dilution series with DGEBA, p,p'-DGEBA and o,p'-DGEBA are shown in Fig. 2. All 9 patients reacted positively to p,p'-DGEBA 0.92% w/v, 7/9 reacted to o,p'-DGEBA 0.92% w/v and 0/9 patients reacted positively to o,o'-DGEBA 0.92% w/v. The only positive reaction to o,o'-DGEBA was a weak one on D7 to 0.092% w/v. The reaction to o,o'-DGEBA was weak compared to the reactions to DGEBA, p,p'-DGEBA and o,p'-DGEBA in the same patient (no. 9). DGEBA elicited patch-test reactions in 8/9 patients. There was one patient (no. 4) who reacted positively only to p,p'-DGEBA and only to the highest concentration. The lowest of the patch-test concentrations to elicit a positive reaction were 0.001% w/v ($0.000029 \text{ mol} \times 1^{-1}$) DGEBA (no. 2) and 0.00092% w/v ($0.000029 \text{ mol} \times 1^{-1}$) p,p'-DGEBA (nos. 2 and 7). Few reactions were read on D7, since many of the participants were treated with topical corticosteroids. However, 4 patients had patch tests with p,p'-DGEBA that were negative or doubtful on D3 and positive on D7, which was not the case for any of the patch tests with DGEBA. Two patients (nos. 3 and 5) had positive reactions to o,p'-DGEBA on D7 that were doubtful on D3.

Group B. The results for DGEBA-R, DGEBA-R and DGEBA(n)-R on D3 and D7, as well as for the DGEBA isomers on D3 are given in Table II. p,p'-DGEBA 1% w/w gave positive reactions in 18/20 workers, 15/20 reacted positively to o,p'-DGEBA 1% w/w, whereas 2/20 workers reacted to o,o'-DGEBA 1% w/w. Slightly fewer workers reacted to DGEBA-R and DGEBA(n)-R compared to DGEBA-R.

Controls. None of the DGEBA isomers gave any reaction.

Table I. Group A. Patch test reactions to dilution series of DGEBA and the DGEBF isomers on day 3 (D3) and day 7 (D7)

Case	Conc. ^c	DGEBA ^a		p,p'-DGEBF ^b		o,p'-DGEBF ^b		o,o'-DGEBF ^b	
		D3	D7	D3	D7	D3	D7	D3	D7
1	1.0	++	++	++	+++	++	+++	-	-
	0.1	++	++	++	++	+	++	-	-
	0.01	-	(+)	(+)	(+)	(+)	(+)	-	-
	0.001	-	-	-	-	-	-	-	-
2	1.0	+++	NR	+++	NR	+++	NR	-	(+)
	0.1	+++	NR	+++	NR	-	(+)	-	(+)
	0.01	+++	NR	+++	NR	-	(+)	-	-
	0.001	++	+	++	++	-	-	-	-
	0.0001	-	-	-	-	-	-	-	-
3	1.0	++	++	+	++	(+)	++	-	-
	0.1	(+)	(+)	+	++	-	-	-	-
	0.01	-	-	-	(+)	-	-	-	-
	0.001	-	-	-	-	-	-	-	-
4	1.0	-	-	(+)	++	-	-	-	-
	0.1	-	-	-	(+)	-	-	-	-
	0.01	-	-	-	-	-	-	-	-
5	1.0	++	++	+++	++	++	+++	-	-
	0.1	++	+	+++	++	++	+++	-	-
	0.01	-	-	-	-	(+)	++	-	-
	0.001	-	-	-	-	-	?	-	-
	0.0001	-	-	-	-	-	(+)	-	-
	0.00001	-	-	-	-	-	-	-	-
6	1.0	+++	NR	+++	NR	+++	NR	-	-
	0.1	+++	NR	+++	NR	+++	NR	-	-
	0.01	+++	NR	+++	++	-	-	-	-
	0.001	-	-	-	(+)	-	-	-	-
	0.0001	-	-	-	-	-	-	-	-
7	1.0	+++	NR	+++	NR	+++	NR	-	-
	0.1	+++	NR	+++	NR	+++	NR	-	-
	0.01	+++	NR	+++	NR	+++	NR	-	-
	0.001	(+)	(+)	(+)	+	-	(+)	-	-
	0.0001	-	-	-	(+)	-	-	-	-
	0.00001	-	-	-	-	-	-	-	-
8	1.0	+++	NR	+++	NR	-	-	-	-
	0.1	++	+	(+)	+	-	-	-	-
	0.01	(+)	-	-	(+)	-	-	-	-
	0.001	-	-	-	-	-	-	-	-
9	1.0	+++	NR	+++	NR	++	+++	-	-
	0.1	+++	NR	+++	NR	++	+++	-	+
	0.01	++	++	(+)	+++	-	-	-	-
	0.001	-	-	-	-	-	-	-	-

DGEBA=diglycidyl ether of bisphenol A; p,p'-DGEBF=p,p'-diglycidyl ether of bisphenol F; o,p'-DGEBF=o,p'-diglycidyl ether of bisphenol F; o,o'-DGEBF=o,o'-diglycidyl ether of bisphenol F.

+++; ++; +=positive reaction of allergic nature. (+) Doubtful reaction. - Negative reaction. NR=not read due to treatment of the patch-test reaction with topical corticosteroids. ^aPatch-tested in a dilution series prepared from a stock solution of 1.0% w/v ($0.029 \text{ mol} \times 10^{-1}$). ^bPatch-tested in a dilution series with equimolar concentrations to DGEBA: 0.92% w/v ($0.029 \text{ mol} \times 10^{-1}$). ^cPatch-test concentrations equimolar to DGEBA.

Purity

The contamination of p,p'-DGEBF, o,p'-DGEBF, o,o'-DGEBF and DGEBA in each other is given in Table III. The highest detectable contamination was 0.1% w/w of p,p'-DGEBF in o,p'-DGEBF. The indicated purity of these 4 substances, as estimated by integration of the peaks in the HPLC chromatograms, was 99.4–99.9% w/w (unpublished data).

DISCUSSION

Contact allergy to the DGEBF isomers

In this study, contact allergies to the DGEBF isomers are described for the first time in humans. A very low number of positive patch-test reactions to o,o'-DGEBF were found, although all participants in groups A and B previously had positive reactions to DGEBF-R and/or

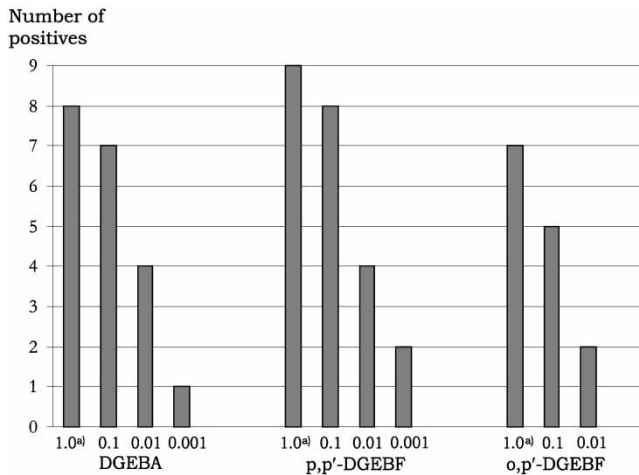


Fig. 2. Group A (n=9). Number of patients with positive reactions to equimolar dilution series of DGEBA, p,p'-DGEBF and o,p'-DGEBF. DGEBA=diglycidylether of biphenol A. p,p'-DGEBF=p,p'-diglycidyl ether of bisphenol F. o,p'-DGEBF=o,p'-diglycidyl ether of bisphenol F. ^aPatch tests in equimolar concentrations: DGEBA, 1.0% w/v (0.029 mol × 1⁻¹); DGEBF, 0.92% w/v (0.029 mol × 1⁻¹).

DGEBF(n)-R. In both groups A and B, the few cases with positive reactions to o,o'-DGEBF had strong (+++) patch-test reactions to the other simultaneously

Table III. Impurities in patch-test substances

Substance	Contaminant (% w/w)			
	DGEBA	p,p'-DGEBF	o,p'-DGEBF	o,o'-DGEBF
DGEBA		^a <0.02	<0.02	<0.02
p,p'-DGEBF	<0.06		<0.06	<0.06
o,p'-DGEBF	<0.001	0.1		0.03
o,o'-DGEBF	<0.03	0.08	0.03	

DGEBA=diglycidyl ether of bisphenol A; p,p'-DGEBF=p,p'-diglycidyl ether of bisphenol F; o,p'-DGEBF=o,p'-diglycidyl ether of bisphenol F; o,o'-DGEBF=o,o'-diglycidyl ether of bisphenol F. ^aWhen no contaminant was identified, the detection level for the investigated contaminant is indicated.

patch-tested DGEBF isomers and epoxy resins. No patch-test reaction could be explained by contamination of the other patch-tested substances. The test reaction to o,o'-DGEBF in group A is difficult to evaluate with regard to contact allergy, since the same patient did not react to the highest patch-tested concentration. The workers in group B were not patch-tested with dilution series of the DGEBF isomers and therefore the importance of contamination is difficult to evaluate fully. The results of a guinea pig maximization test (GPMT), in which it has been shown

Table II. Group B. Patch-test reactions to DGEBA-R, DGEBF-R, DGEBF(n)-R and the DGEBF isomers in workers exposed to epoxy resins

Case	DGEBA-R 1.0% w/w		DGEBF-R 0.25% w/w		DGEBF(n)-R 0.25% w/w		p,p'-DGEBF 1.0% w/w	o,p'-DGEBF 1.0% w/w	o,o'-DGEBF 1.0% w/w
	D3	D7	D3	D7	D3	D7	D3	D3	D3
1	++	++	++	+++	++	++	+++	++	-
2	++	NR	++	NR	++	NR	+++	+++	(+)
3	++	+	++	++	++	+	+++	++	-
4	+++	++	+++	+++	++	+++	+++	+++	++
5	++	++	+++	+++	+++	+++	+++	+++	+
6	++	+++	+	+++	-	+	++	++	-
7	++	++	++	++	++	++	++	+	-
8	+	++	++	++	+	+	++	+	-
9	++	+	-	+	-	+	++	+	-
10	++	++	(+) ^a	(+)	-	(+) ^a	++	+	(+)
11	++	+++	+	++	+	++	++	+	-
12	-	++	-	++	-	++	++	++	-
13	-	+	-	+	-	+	++	++	-
14	++	++	+	(+)	(+) ^a	(+)	++	(+)	-
15	+	++	+	++	+	+	+	(+)	-
16	+++	++	++	++	+	++	+	+	-
17	+	++	+	++	+	++	+	+	-
18	(+)	(+) ^a	+	(+)	+	(+)	+	(+)	-
19	+	+	(+) ^a	(+)	(+) ^a	(+)	(+)	-	-
20	+	+	+	+	(+)	(+) ^b	-	-	-
Sum	17	18 (1 NR)	15	15 (1 NR)	12	14 (1 NR)	18	15	2

DGEBA-R=epoxy resin based on bisphenol A. DGEBF-R=epoxy resin based on bisphenol F. DGEBF(n)-R=epoxy resin based on phenolic novolac resin. DGEBA=diglycidyl ether of bisphenol A; p,p'-DGEBF=p,p'-diglycidyl ether of bisphenol F; o,p'-DGEBF=o,p'-diglycidyl ether of bisphenol F; o,o'-DGEBF=o,o'-diglycidyl ether of bisphenol F. +++; ++; += positive reaction of allergic nature. (+) doubtful reaction. - negative reaction. NR=not read due to treatment of the patch-test reaction with topical corticosteroids. ^aPositive patch-test reaction when re-tested with the same substance. ^bNegative patch-test reaction when re-tested with the same substance.

that all 3 DGEBF isomers are strong sensitizers (5), indicate a discrepancy between the GPMT and the present study concerning o,o'-DGEBF. Twenty controls tested negatively to the individual DGEBF isomers and so there are no obvious indications of false-positive, i.e. irritant, reactions. Therefore, the few reactions to o,o'-DGEBF probably represent true contact allergy. In the same GPMT it was shown that DGEBA cross-reacts with p,p'-DGEBF and o,p'-DGEBF, but not with o,o'-DGEBF (5). The cross-reaction pattern found in the GPMT thus suggests an explanation of the comparatively few reactions to o,o'-DGEBF, if it is assumed, as in the present study, that a majority of individuals with contact allergy to DGEBA-R, which is the most common epoxy resin, have been primarily sensitized to DGEBA. Another explanation for the low frequency of contact allergy to o,o'-DGEBF in humans could be the lesser content of o,o'-DGEBF compared to the other 2 DGEBF isomers in commercial resins. The epoxy resins of the bisphenol F-type contained approximately 5–8 times more of the summarized content of p,p'-DGEBF and o,p'-DGEBF than of o,o'-DGEBF, with the exception of the DGEBF-R (Rütapox), which contained 15 times more of the former 2 DGEBF isomers than of o,o'-DGEBF (to be published). However, the dose–response relationships concerning the sensitizing capacities of the individual DGEBF isomers are unknown. It is thus difficult to know whether the lower content of o,o'-DGEBF is of any importance when an individual is sensitized exclusively with epoxy resins of the bisphenol F-type. There may be alternative explanations to the patch-test results of the DGEBF isomers, such as differences in penetration and metabolism. However, the penetration seems not to differ to an extent that would explain the differences in the patch-test results. The octanol/water partition coefficient ($\log P_{o/w}$) is judged to be the most important parameter for modelling the skin permeability of chemicals (6). The calculated $\log P_{o/w}$ is 3.84 for DGEBA and 3.26 for the isomers of DGEBF (7). For values of $\log P_{o/w} > 3$ the rate of increased permeability levels off (6).

The GPMT referred to earlier also showed that sensitization with DGEBA, p,p'-DGEBF or o,p'-DGEBF would probably result in contact allergy to the other 2 substances (5). Therefore, if the results from the GPMT are valid for humans, it is impossible to know, based on the results from patch-testing with DGEBA and the DGEBF isomers, whether the contact allergy originates from a DGEBA-R or an epoxy resin of the bisphenol F-type.

In the GPMT, p,p'-DGEBF was found to be a slightly stronger sensitizer than DGEBA (5). It was also found that more animals, although sensitized with DGEBA, tested positively to p,p'-DGEBF than to

DGEBA itself (5). For most of the dilution steps, a higher number of patients reacted to p,p'-DGEBF than to DGEBA (Fig. 2). One patient reacted exclusively to p,p'-DGEBF, although had previously shown to have had contact allergy to DGEBA-R. These observations indicate that p,p'-DGEBF is possibly a slightly stronger sensitizer in humans than DGEBA. o,p'-DGEBF elicited reactions in approximately 75% of the participants within both groups A and B, compared to 90–100% for p,p'-DGEBF, indicating that o,p'-DGEBF might be a weaker sensitizer in humans compared to p,p'-DGEBF.

Patch-testing with corticosteroids does not affect the strength of the reactions of adjacent patch tests (8). It is therefore assumed that patch tests adjacent to patch-test reactions treated with topical corticosteroids are also unaffected.

It should be mentioned that the 2 epoxy groups in the molecules of DGEBA and the 3 DGEBF isomers contain a stereogenic centre, i.e. a carbon atom bonded to 4 different groups. Each of these compounds is therefore a mixture of stereoisomers. Whether that influences their behaviour as antigens is unknown. It has hitherto been impossible to separate the stereoisomers chemically and therefore DGEBA and the 3 isomers of DGEBF are discussed as if each were a single substance.

Contact allergy to DGEBF-R and DGEBF(n)-R

According to the results of the present study, o,o'-DGEBF does not contribute to the patch-test reactivity of epoxy resins of the bisphenol F-type to any large extent. The content of the DGEBF isomers in several DGEBF-R and DGEBF(n)-R has been analysed at our department (to be published). The summarized concentration of p,p'-DGEBF and o,p'-DGEBF in the DGEBF-R and DGEBF(n)-R patch-test preparations used within group B (Table II) could be estimated to 0.2% w/w and 0.1% w/w, respectively, and the concentration of DGEBA in the DGEBA-R patch-test preparation to 0.9% w/w. Possibly, the differences in the number of workers who reacted to DGEBA-R, DGEBF-R and DGEBF(n)-R can be explained by the differences in concentrations of the 2 DGEBF isomers and DGEBA in the patch-tested epoxy resins.

In a recent study, the frequency of simultaneous contact allergies to epoxy resins of the bisphenol F-type among patients with contact allergy to DGEBA-R was found to be high (>92%) (9). No differences between DGEBF-R and DGEBF(n)-R were found. Additionally, 6 patients who were not included in the mentioned study had contact allergy only to DGEBF-R/DGEBF(n)-R (9). The patch-test concentrations used in the referred study were 1.0%.

In addition to the DGEBF isomers, other oligomers are present in DGEBF-R and DGEBF(n)-R (3). For

DGEBA-R no allergic reactions to oligomers with higher MW than DGEBA (MW 340) have been demonstrated (2). It is unknown whether oligomers in DGEBF-R and DGEBF(n)-R with higher MW than the DGEBF isomers (MW 312) are sensitizers in humans.

In conclusion, contact allergies in humans to the main constituents in epoxy resins of the bisphenol F-type, i.e. the DGEBF isomers, are described for the first time in the present study. It is found that both p,p'-DGEBF and o,p'-DGEBF are sensitizers, whereas o,o'-DGEBF elicited very few reactions. The results indicate that p,p'-DGEBF is a sensitizer in humans, which is at least as strong as DGEBA.

ACKNOWLEDGEMENTS

We thank Östen Sörensen for skilful technical assistance. The study was supported by grants from the Swedish Council for Work Life Research and the Swedish Foundation for Health Care Sciences and Allergy Research.

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