

Is benzoquinone the prohaptens in cross-sensitivity among aminobenzene compounds?

PAOLO LISI AND KATHARINA HANSEL

Allergologic and Environmental Dermatology Unit, Department of Medical and Surgical Specialities, University of Perugia, Policlinico Montelucre, Perugia, Italy

Cross-sensitivity in allergic contact dermatitis is a simultaneous allergy to 2 or more contact substances which have in common an antigenic determinant or a metabolic derivative. One of the most notable examples is the cross-sensitivity among aromatic compounds which may be oxidized *in vivo* to benzoquinone (BQ). However, it has also been hypothesized that the allergenicity and cross-sensitization are modulated by the chemical reactivity of the substituents in the para position. A serial dilution of BQ (from 1% to 0.1% in pet.) and three 1,4-substituted benzene derivatives (*p*-aminophenol, hydroquinone, metol), theoretically capable of conversion to BQ by oxidation, were patch tested in 22 *p*-phenylenediamine (PPD) positive patients and in 20 controls. The patients and a further 116 subjects with a positive history of sensitivity to 1 or more aminoaromatic compounds were also tested with some haptens of the para group (PPD, *p*-aminobenzoic acid, *p*-aminodiphenylamine, benzocaine, procaine chloride, *p*-toluenediamine sulfate). The results show that (i) the optimal patch test concentration for BQ was 0.2%, (ii) only 4 of the 22 patients allergic to PPD gave a clearly positive allergic reaction to BQ, and (iii) the number of positive reactions to the aromatic compounds was correlated with the presence of activating ($-\text{NH}_2$, $-\text{OH}$, $-\text{CH}_3$) and deactivating ($-\text{COOH}$) groups in the para position or, perhaps, with their effect on percutaneous penetration. The data suggest that BQ is not the only intermediate in the cross-sensitization of para group haptens. This is probably conditioned by other oxidation products and/or the chemical structure of the substituents in position 4 of the benzene ring.

Key words: aminobenzene compounds; cross-sensitivity; benzoquinone; substituent groups; pro-haptens; chemical reactivity. © Munksgaard, 1998.

Accepted for publication 15 July 1998

Cross-sensitivity in allergic contact dermatitis is generally defined as a simultaneous allergy to 2 or more chemically-related contact substances which have an antigenic determinant in common. However, it is possible that positive patch test reactions to very different compounds are indicative of a true cross-sensitization when they give *in vivo* the same or closely related metabolic derivatives (prohaptens theory). One of the most notable examples of this event is perhaps the cross-sensitivity among aromatic chemicals with an amino substituent in the para position. This has been related to oxidation reduction-hydrolysis processes which lead to the production of iminoquinone and, in the end, of benzoquinone (BQ) (1, 2).

It has also been hypothesized that allergenicity and cross-sensitization are modulated by chemical reactivity of the substituent groups in the para position: some of these are more or less activating

($-\text{NH}_2$, $-\text{OH}$, $-\text{CH}_3$), others deactivating ($-\text{NO}_2$, $-\text{SO}_3$, $-\text{COOH}$) (3).

To verify these hypotheses, mainly formulated on the results of research in guinea pigs, we have investigated the rate of the sensitivity to BQ and some benzene derivatives in subjects with a positive patch test history for at least 1 hapten of the para group.

Materials and Methods

22 volunteer patients sensitized to *p*-phenylenediamine (PPD) (9 male and 13 female, aged between 19 and 72; average age, 46.8 years) were patch tested with a serial dilution of BQ in white petrolatum (pet.) (1%, 0.5%, 0.2%, 0.1%) and with a series of 1,4-substituted benzene derivatives (FIR-MA Diagent, Firenze). Some of these are haptens of procaine chloride 2%, *p*-toluenediamine sulfate

Table 1. Patch test reactions to a serial dilution of benzoquinone in 22 patients sensitized to *p*-phenylenediamine (P) and in 20 controls (C)

Benzoquinone concentrations (%)	Irritant reactions		Positive reactions		Negative reactions	
	P	C	P	C	P	C
1	12	18	5	—	5	2
0.5	7	9	4	—	11	11
0.2	5	2	4	—	13	18
0.1	3	2	1	—	18	18

1%); others are theoretically capable of conversion to BQ either by direct oxidation (*p*-aminophenol 1%, hydroquinone 1%) or following demethylation by methyl transferase enzymes in the skin (metol 1%) (4).

20 consecutive contact dermatitis patients (8 male and 12 female; average age, 44.5 years, range 18–77) without allergy to para compounds, were employed as controls and were tested with the serial dilutions of BQ.

Patch tests with the above-mentioned 1,4-substituted benzene derivatives (without hydroquinone and metol) and with PPD base (1% pet.) were carried out in 116 contact dermatitis patients with a positive history for sensitivity to 1 or more para allergens (29 male and 87 female; average age 43.3 years, range 14–80).

The substances were applied on the upper back with Van der Bend square chambers and removed after 2 days. The readings were carried out on days (D) 2 and 4 and the reactions evaluated according to ICDRG criteria.

Results

The results of patch testing with the serial dilution of BQ in the 22 PPD-positive patients and in the 20 controls are shown in Table 1. Only 1 of the patients had positive reactions to all dilutions of BQ, while 4 reacted to either 1% or 0.2%. In 3 of these, the positive reactions were characterized by erythema, swelling and papules; in the 4th, vesicles were also seen.

Table 2. Positive patch test reactions to 1,4-substituted benzene derivatives in 22 patients sensitized to *p*-phenylenediamine

Tested compounds	Positive reactions
<i>p</i> -aminodiphenylamine	11 ^(a,b)
<i>p</i> -aminophenol	6 ^(a,b)
<i>p</i> -toluenediamine	6 ^(c,d)
metol	5 ^(a,b)
benzocaine	2
<i>p</i> -aminobenzoic acid	—
hydroquinone	—
procaine chloride	—

In brackets, the 4 benzoquinone-positive subjects (a,b,c,d).

In 12 patients, 1% BQ caused an irritant reaction consisting of red-brownish discolouration, almost always associated with erosion. The other dilutions induced less intense skin discolouration and scaling.

In 18 of the controls, irritant reactions to BQ were observed. The morphology and distribution of these in relation to test concentrations were similar to those observed in the patients.

The results of patch testing with 1,4-substituted benzene derivatives in the 22 PPD-positive patients are reported in Table 2. 2 of the 4 BQ-positive subjects reacted to *p*-aminodiphenylamine, *p*-aminophenol and metol; the remaining 2 were positive to *p*-toluenediamine.

The frequency of positive patch test reactions caused by 1,4-substituted benzene derivatives in

Table 3. The frequency of positive reactions to 1,4-substituted benzene derivatives in 116 contact dermatitis patients sensitized to 1 or more para compounds

Tested compounds	Positive reactions		
	no.	%	
<i>p</i> -phenylenediamine base	89	76.7	(-NH ₂)
<i>p</i> -aminodiphenylamine	62	53.4	(-NH-R)
<i>p</i> -aminophenol	31	26.7	(-OH)
<i>p</i> -toluenediamine	29	25.0	(-CH ₃)
benzocaine	22	18.9	(-COO-R)
procaine chloride	8	6.9	(-COO-R)
<i>p</i> -aminobenzoic acid	6	5.2	(-COOH)

In brackets, the substituted group in the para position.

Table 4. The distribution of the 65 cross-reactions among 1,4-substituted benzene derivatives in connection with to their activating (A) and deactivating (D) groups

Positive reactions (no.)	Subjects (no.)	Benzene derivatives		
		A	D	A+D
2	30	27	2	1
3	21	18	—	3
4	10	8	—	2
5	1	—	—	1
6	2	—	—	2
7	1	—	—	1

Activating groups: -NH₂, -OH, -CH₃; deactivating groups: -COOH.

the 116 contact dermatitis patients with anamnestic sensitivity to 1 or more para compounds and the respective substituent group in the para position are indicated in Table 3. This is consistent with the high sensitization potential of PPD and *p*-aminodiphenylamine compared to the other compounds.

In 65 of the 116 patients examined (56.1%), simultaneous reactions were observed; these were caused by more than 2 substances in 35 cases. The connection with activating and deactivating groups in the para position are shown in Table 4.

Discussion

The results of our study on cross-reactivity to the para compounds confirm only partially the theory suggested by Mayer (1) over 40 years ago. According to this author, an aromatic hapten may be converted in vivo to a more reactive species and in particular to BQ (1, 2).

In our investigation and in those of Basketter et al. in both guinea pig (4) and in man (5), only a limited degree of simultaneous reactions between BQ and PPD was observed. These were present in 3 of the 10 PPD-positive subjects examined by Basketter & Lidén (5) and in 4 of our 22. The last 4 were also sensitive to other aminocompounds (*p*-aminodiphenylamine, *p*-toluenediamine), but only 2 showed positive reactions to *p*-aminophenol and metol; these are 1,4-substituted benzene substances which might easily oxidize to BQ. Interestingly, none reacted to hydroquinone, as already reported by others (5, 6), though this compound should easily be oxidized to BQ.

The low number of positive patch test responses to BQ in PPD-sensitized patients suggests that BQ is not the sole nor even the major intermediate in the cross-sensitization of para group haptens. Probably, other oxidation products (7) able to bind epidermal proteins are involved.

It is possible, however, that delayed cross-sensitivity among aminoaromatic compounds is influenced by the chemical structure of substituents in the para position, as indicated by the sensitization rate obtained by Kleniewska & Maibach (3) in guinea pigs. Furthermore, our data on positive patch test reactions in 116 humans, investigated with some simple 1,4-substituted benzenes, agree with this hypothesis. The compound allergenicity was well correlated with the more-or-less important activating potential of substituents in the para position. The incidence of sensitivity to substances with a 2nd amine group in position 4 (PPD, *p*-aminodiphenylamine) was higher than that of compounds with hydroxyl (*p*-aminophenol), methyl (*p*-toluenediamine) and carboxyl groups (benzocaine,

procaine chloride, *p*-aminobenzoic acid). Nevertheless, these substituent groups may also have a significant effect on percutaneous penetration, thus altering the allergenicity.

The activating and deactivating groups also seem to influence the trend of cross-reactions among para group chemicals. These, encountered in 56.1% of the patients, were much more frequent (81.5%) in subjects sensitized to 2 or more benzene derivatives with activating groups. Therefore, chemical substitutions can enhance the sensitization potential and the cross-sensitivity of the aminoaromatic compounds.

A brief mention of the BQ concentration for patch testing should be given. At 1% dilution, the substance is irritant and causes red-brownish discolouration of the skin and erosion, both in PPD-sensitized patients and in controls. The optimal test concentration is 0.2% or, perhaps, 0.1%, as recently proposed by Basketter & Lidén (5). Only these 2 dilutions may elicit an allergic reaction which can clearly be differentiated from the irritant reactions occurring at higher concentrations.

In conclusion, our data supply limited support for the prohaptens theory, because BQ is probably only one of the intermediates in the cross-sensitization of para group compounds. This, however, is also influenced by the chemical structure of the substituents in position 4 of the benzene ring.

References

1. Mayer R L. Group sensitization to compounds of quinone structure and its biochemical base. *Progr Allergy* 1954; 4: 79-82.
2. Dupuis G, Benezra C. *Allergic contact dermatitis to simple chemicals: a molecular approach*. New York: Marcel Dekker; 1982.
3. Kleniewska D, Maibach H. Allergenicity of aminobenzene compounds: structure-function relationships. *Dermatosen* 1980; 28: 11-13.
4. Basketter D A, Goodwin B F J. Investigation of the prohaptens concept: cross reactions between 1,4-substituted benzene derivatives in the guinea pig. *Contact Dermatitis* 1988; 19: 248-253.
5. Basketter D A, Lidén C. Further investigation of the prohaptens concept: reactions to benzene derivatives in man. *Contact Dermatitis* 1992; 27: 90-97.
6. Picardo M, Cannistraci C, Cristaudo A, De Luca C, Santucci B. Study on cross-reactivity to the para group. *Dermatology* 1990; 181: 104-108.
7. Møllgaard B, Hansen J, Kreilgård B, Wildfang I L. Cross sensitization in guinea pigs between *p*-phenylene diamine and oxidation products thereof. *Contact Dermatitis* 1990; 23: 274.

Address:

Paolo Lisi
Dermatologia allergologica e ambientale
Policlinico Monteluce
I 06100 Perugia
Italy