

Original article

The spectrum of allergic (cross-)sensitivity in clinical patch testing with 'para amino' compounds

Background: Allergic contact sensitization to 'para amino' compounds is frequent and the spectrum of cross-reactivity between members of this chemical group is variable.

Methods: A retrospective analysis of clinical patch test data obtained with a special test series in the centres of the Information Network of Departments of Dermatology (IVDK) between 1995 and 1999.

Results: In the 638 patients tested with the above test panel positive reactions were observed most often to *p*-aminoazobenzene (16.2%), *p*-phenylenediamine (14.1%), *p*-toluylenediamine (10.0%), followed by 4,4'-diaminodiphenylmethane (8.5%), Disperse Orange 3 (8.4%) and *p*-aminophenol (3.1%). Among the 544 patients tested with *p*-phenylenediamine and all seven additional 'para amino' compounds, concordance between reactions varied greatly. The stronger the positive test reaction to *p*-phenylenediamine, *p*-toluylenediamine or *p*-aminoazobenzene, the more frequently additional positive reactions to the other compounds were observed.

Conclusions: A screening employing several 'para amino' compounds is necessary to describe the individual spectrum of allergic contact sensitization, as there is no reliable marker substance. Further research should aim at (i) establishing the mechanism of cross-reactivity to 'para amino' compounds and (ii) identifying exposures in the environment.

W. Uter^{1,2}, H. Lessmann², J. Geier²,
D. Becker³, T. Fuchs⁴, G. Richter⁵
(for the IVDK Study Group*,
German Contact Dermatitis
Research Group (DKG))

¹Department of Medical Informatics, Biometry and Epidemiology, Friedrich-Alexander University Erlangen-Nürnberg, Erlangen; ²Information Network of Departments of Dermatology (IVDK), Georg August University, Department of Dermatology, Göttingen; ³Department of Dermatology, University of Mainz; ⁴Department of Dermatology, Allergy Unit, Georg August University, Göttingen; ⁵Department of Dermatology, Allergy Unit, Technical University of Dresden, Germany

Key words: contact dermatitis; cross-reactivity; patch tests; *p*-aminoazobenzene; *p*-phenylenediamine; *p*-toluylenediamine.

Wolfgang Uter, M.D.
Department of Medical Informatics, Biometry and Epidemiology
Friedrich-Alexander University, Erlangen-Nürnberg
Waldstr. 6
D-91054 Erlangen
Germany

Accepted for publication 13 November 2001

Some 70 years ago, Mayer reported on experiments

* Centres of the IVDK contributing to this analysis (in alphabetical order): Aachen (H. Dickel), Augsburg (O. Hirschsteiner, A. Ludwig), Berlin UKRV (J. Grabbe, T. Zuberbier), Bochum (C. Szliska), Dortmund (P.J. Frosch, B. Pilz, C. Pirker), Dresden (G. Richter), Duisburg (J. Schaller), Erlangen (K. P. Peters, M. Fartasch), Essen (H.-M. Ockenfels, U. Hillen), Göttingen (Th. Fuchs), Graz (W. Aberer, B. Kränke), Halle (G. Gaber, D. Lübke), Heidelberg (A. Schulze-Dirks, M. Hartmann), Jena (M. Gebhardt, A. Bauer), Kiel (J. Brasch), Lübeck (J. Kreusch, J. Grabbe), Magdeburg (U. Jappe, E. Weisshaar), Marburg (I. Effendy), München TU (J. Rakoski), Nürnberg (I. Müller), Osnabrück (W. Uter), Ulm (H. Gall (Deceased)), Wuppertal (O. Mainusch, J. Raguz).

indicative of allergic cross sensitivity to different chemicals with a *para* di-substituted structure (1), which are often termed 'para amino' compounds (2). The exact mechanism leading to the metabolic formation of common haptens is still enigmatic for this group of substances, apart from some hypotheses, e.g. the formation of reactive, nucleophilic quinone imines as 'common denominator'. However, it is very well known that the degree of allergenic cross-reactivity between different 'para amino' compounds varies a lot, both in well controlled animal experiments and in clinical patch testing. Thus, the German Contact Dermatitis Research Group (Deutsche Kontaktallergie-Gruppe, DKG) considered it important to develop a diagnostic tool to

Table 1. DKG test series 'para amino' compounds, results 1995–1999 (readings at 72 h)

Allergen	Concentration (%)	N (test)	Number			% Positive
			?/R	+	++/+++	
<i>p</i> -aminobenzoic acid (PABA)	10.0	613	2	0	1	0.2
<i>p</i> -aminophenol (CI 76550)	1.0	613	5	17	2	3.1
<i>p</i> -toluylenediamine	1.0	613	47	32	29	10.0
sulfanilamide	5.0	619	2	1	1	0.3
Disperse Orange 3 (CI 11005)	1.0	618	8	21	31	8.4
<i>p</i> -aminoazobenzene (CI 11000)	1.0	617	32	51	49	16.2
4,4'-diaminodiphenylmethane	0.5	613	11	31	21	8.5
Tested in 573 of these patients: <i>p</i> -phenylenediamine	1.0		15	37	44	14.1

All allergens in petrolatum.

describe precisely the spectrum of individual reactivity, instead of relying on just one or two marker substances. Results with a special patch test series conceived for this purpose in 1995 (3) are presented in this paper.

Material and methods

In the centres of the Information Network of Departments of Dermatology (IVDK) listed above, being represented in the DKG at the same time, both the complete patch test results and a short standardized history of all patients patch tested in these centres are regularly collected and transferred to the Göttingen data centre (<http://www.ivdk.gwdg.de>) for pooled analysis (4). Proportions of sensitizations were calculated based on the 72 h readings, which followed ICDRG guidelines (5). Patch test substances (see Table 1) were obtained from Hermal, Reinbek, Germany.

The dichotomised patch test outcome considered here was defined as 'positive', i.e. '+', '+ +', '+ + +' reaction, vs 'not positive', i.e. negative, questionable or irritant reaction. To address the question of concordance, we calculated Cohen's kappa, accompanied by the respective 95% confidence interval (CI). This standard measure in such analyses quantifies the chance-corrected proportion of agreement between two discrete ratings and gives a more informative quantitative description of concordance than the crude proportion of agreement for skewed distributions of ratings (6) as in this case, with considerably more negative than positive reading results. For data management and analysis, the statistical software package SAS 8.0 (SAS Institute, Cary, NC) was employed.

Results

Between January 1995 and December 1999 altogether 49252 patients were patch tested in the IVDK centres. Since its introduction in spring 1995, the special 'para amino' test series was applied to 638 patients; results are shown in Table 1 (due to temporary unavailability or omission in individual cases the actual number of patients tested with the allergens is lower). In 544 patients all seven allergens and *p*-phenylenediamine were patch tested; this group of patients was considered in the further analyses, with results shown in Table 2, however, omitting *p*-aminobenzoic acid and sulfanilamide because they caused very few positive reactions (Table 1).

Concordance, as quantified with Cohen's kappa, was 'fair to moderate' (>0.50) for six combinations: Disperse Orange 3 with *p*-phenylenediamine, *p*-aminophenol, *p*-toluylenediamine and *p*-aminoazobenzene, as well as *p*-phenylenediamine + *p*-toluylenediamine and in particular *p*-phenylenediamine + *p*-aminoazobenzene (0.64, 95% CI: 0.55–0.74; the maximum concordance observed). All combinations with 4,4'-diaminodiphenylmethane displayed 'fair' concordance (range: 0.20, 95% CI: 0.07–0.33 with *p*-toluylenediamine to 0.38, 95% CI: 0.26–0.50 with *p*-phenylenediamine).

Table 2. Pattern of concomitant reactivity to seven 'para amino' compounds and *p*-phenylenediamine in 544 patients patch tested synchronously with all 8 allergens

		Percentages of all patients reacting positively to the allergen†					
Percentage positive concomitant reaction	†	PPD n = 74	PAP n = 11	PTD n = 49	DOR3 n = 31	PAAB n = 68	DDM n = 36
<i>p</i> -phenylenediamine	PPD	—	90.9 *	69.4 **	93.6 **	72.1 **	66.7 *
<i>p</i> -aminophenol	PAP	13.5	—	22.5 *	35.5 **	16.2 *	22.2 *
<i>p</i> -toluylenediamine	PTD	46.0	100.0	—	77.4 **	45.6 **	30.6 *
Disperse Orange 3	DOR3	39.2	100.0	49.0	—	44.1 **	30.6 *
<i>p</i> -aminoazobenzene	PAAB	66.2	100.0	63.3	96.8	—	52.8 *
4,4'-diaminodiphenylmethane	DDM	32.4	72.7	22.5	35.5	27.9	—
Isolated reactions		13.5	—	24.5	—	20.6	19.4

† Patients reacting positively to allergen reacted to the compounds in the column.

Agreement according to Cohen's kappa (κ) denoted to three categories: * excellent (κ > 0.75); ** fair to good (κ 0.75–0.40); *** poor (κ < 0.40) agreement beyond chance (6).

Allergic (cross-)sensitivity in patch testing with 'para-amino' compounds

Table 3. Proportions of positive reactions to 'para amino' compounds (left column) in patients with non-positive and different grades of positive reactions to *p*-phenylenediamine, *p*-aminoazobenzene and *p*-toluylenediamine, respectively, in 544 patients

Allergen (% positive reaction)	<i>p</i> -phenylenediamine				
	Negative/IR	?	+	++	+++
<i>p</i> -toluylenediamine	2.9	14.3	36.1	38.5	91.7
<i>p</i> -aminophenol	0.2	0.0	2.8	11.5	50.0
Disperse Orange 3	0.4	0.0	16.7	42.3	100.0
<i>p</i> -aminoazobenzene	3.7	14.3	55.6	69.2	91.7
4,4'-diaminodiphenylmethane	2.6	0.0	33.3	30.7	33.3

	<i>p</i> -aminoazobenzene				
	Negative/IR	?	+	++	+++
<i>p</i> -toluylenediamine	3.6	7.7	18.4	77.3	87.5
<i>p</i> -aminophenol	0.0	0.0	7.9	22.7	37.5
Disperse Orange 3	0.2	0.0	21.1	63.6	100.0
<i>p</i> -phenylenediamine	4.3	23.1	55.3	90.9	100.0
4,4'-diaminodiphenylmethane	3.6	3.9	34.2	18.2	25.0

	<i>p</i> -toluylenediamine				
	Negative/IR	?	+	++	+++
<i>p</i> -phenylenediamine	7.2	16.2	53.6	87.5	100.0
<i>p</i> -aminophenol	0.0	0.0	14.3	25.0	60.0
Disperse Orange 3	1.3	2.7	35.7	56.3	100.0
<i>p</i> -aminoazobenzene	7.0	13.5	46.4	81.3	100.0
4,4'-diaminodiphenylmethane	4.4	13.5	14.3	31.3	40.0

The remaining combinations had kappa coefficients between 0.21 and 0.47. Not infrequently, singular reactions were observed to the 'para amino' compounds (Table 2, bottom row). The strength of the patch test reaction to one of the three most common allergens in our analysis (*p*-phenylenediamine, *p*-toluylenediamine and *p*-aminoazobenzene) was significantly correlated with the proportion of positive reactions to the other 'para amino' allergens considered (Table 3, $P < 0.0001$ for all cross tabulations, Cochran–Armitage trend test).

Discussion

In line with previous results, we found a heterogeneous pattern of reactivity to different 'para amino' compounds. Simultaneous occurrence of positive patch tests to different allergens, also referred to as clinical cross-reactivity, cannot automatically be equalled with allergenic cross-reactivity, as previous (co)sensitization due to exposure to the other compound(s) must always be considered as an alternative explanation. As a consequence, allergic cross-sensitivity will be over-estimated, if such combined exposure actually occurs. Conversely, negative reactivity to a structurally related allergen can confidently be interpreted as indicative of a lack of allergic cross-sensitivity.

Our data clearly illustrate that there is not just one

marker allergen, e.g. *p*-phenylenediamine that can detect 'para amino' compound allergy with a sufficient sensitivity. Instead, a range of other such compounds should usually be tested in addition, like the set of substances presented here (Table 1). This set could, in future, exclude the rare allergens sulfanilamide and *p*-aminobenzoic acid (which may, however, be relevant to individual cases of allergic reactions to topical drugs or sunscreens). Interestingly, *p*-aminoazobenzene appears to be a more sensitive screening allergen for 'para amino' compound allergy than *p*-phenylenediamine, at least if the total proportion is considered (Table 1), but not necessarily regarding the allergic cross-sensitivity pattern (Table 2). A similar observation has been made previously (7). Remarkably, a notable proportion of cases of active sensitization caused by *p*-aminoazobenzene had been found in that study (the test concentration of *p*-aminoazobenzene was not given). Late readings (beyond D4) are not performed regularly in the IVDK centres. However, among those patients whose tests were read later ($n = 373$), we found only four cases of late reactivity to *p*-aminoazobenzene which could possibly be interpreted as active sensitization. Thus, the present test concentration (1% in petrolatum) and the risk of active sensitization does not seem to be excessively high.

Although the strength of a positive patch test reaction should generally not be over-interpreted, the

observation of a significantly increasing proportion of other positive test reactions to 'para amino' compounds with increasing degree of a positive reaction to *p*-phenylenediamine, *p*-toluylenediamine and *p*-aminoazobenzene, respectively, stimulates the following research question: do patients with strong and multiple patch test reactions represent a subset which is genetically more prone to be sensitized (and elicited)? This notion may be supported by recent findings indicating an association between the NAT2*4 haplotype (rapid acetylators) and contact sensitization to *para*-substituted aryl compounds (8). As a consequence for clinical practice, those 'para amino' allergens should probably be omitted from the individual test tray which regularly caused positive test reactions in case of a (previous) '+++' reaction to one of the respective 'leading allergens', namely *p*-phenylenediamine,

p-toluylenediamine or *p*-aminoazobenzene (see Table 3), to lessen the risk of presumable 'spill-over' effects, i.e. 'angry back' phenomena.

In conclusion, the present test series appears adequate to diagnose, with patch testing, the spectrum of 'para amino' compounds capable of eliciting allergic contact dermatitis, assuming that these allergens are reliable markers of the respective subclasses of this group of substances. However, further research should probably go into the molecular mechanism of 'para amino' allergic cross-sensitivity – including aspects like genetically determined variants of relevant metabolic pathways. In addition, the sources of exposure to these compounds (beyond well known sources like oxidative or azoic dyes) in the workplace and other environments should be identified better.

References

1. MAYER RL. Die Überempfindlichkeit gegen Körper von Chinonstruktur. Arch Dermatol Syph 1928;**156**:331–354.
2. BRUZE M. The chemical basis of para-amino compounds. Derm Beruf Umwelt 1984;**32**:174–175.
3. RICHTER G. Zur Problematik sogenannter Paragruppen-Allergien am Beispiel der N-Phenyl-N-isopropyl-p-phenyldiamin-Sensibilisierung. Derm Beruf Umwelt 1995;**43**:210–214.
4. UTER W, SCHNUCH A, GEIER J, FROSCH PJ. The epidemiology of contact dermatitis. The information network of departments of dermatology (IVDK) in Germany. Eur J Dermatol 1998;**8**:36–40.
5. WAHLBERG JE. Patch testing. In: RYCROFT, RJG, MENNÉ, T, FROSCH, PJ, LEPOITTEVIN, J-P, , editors. Textbook of Contact Dermatitis, 3rd edn. Berlin: Springer, 2001, 435–468.
6. FLEISS JL. Statistical Methods for Rates and Proportions, 2nd edn. New York: Wiley, 1981.
7. ARNOLD WP, VAN JOOST T, VAN DER VALK PGM. Adding p-aminoazobenzene may increase the sensitivity of the European standard series in detecting contact allergy to dyes, but carries the risk of active sensitization. Contact Dermatitis 1995;**33**:444.
8. WESTPHAL GA, REICH K, SCHULZ TG, NEUMANN C, HALLIER E, SCHNUCH A. N-acetyltransferase 1 and 2 polymorphisms in para-substituted arylamine-induced contact allergy. Br J Dermatol 2000;**142**:1121–1127.