

Factors associated with *p*-phenylenediamine sensitization: data from the Information Network of Departments of Dermatology, 2008–2013

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Summary

Background. Risk factors for *p*-phenylenediamine (PPD) sensitization include the use of hair dyes, the application of temporary black henna tattoos, working as a hairdresser, and, possibly, exposure to hair dye pretests.

Objectives. To quantify the impact of these (putative) risk factors on PPD sensitization.

Methods. Six items related to PPD exposure were added to the routine Information Network of Departments of Dermatology questionnaire from 2008 to 2013. A retrospective analysis of data from 4314 patients tested with PPD 1% pet. was conducted.

Results. Of the PPD-positive patients (*n* = 271), 80% had their hair dyed, and, of these, 57% subsequently developed scalp dermatitis, whereas only 11% had had a henna tattoo. The self-administrated pretest with hair dye was performed by only a few patients, precluding a more detailed analysis. Hair dyeing [odds ratio (OR) 6.0; 95% confidence interval (CI): 3.9–9.4], henna tattoos (OR 2.4; 95%CI: 1.5–3.7) and being a hairdresser (OR 2.1; 95%CI: 1.3–3.2) increased the risk of PPD sensitization. Neither dyeing of own hair nor application of a temporary henna tattoo seemed to affect PPD sensitization in hairdressers. *p*-Aminoaryl compounds more often gave positive reactions in patients with henna tattoo.

Conclusions. Hair dyeing is the major risk factor for PPD sensitization in this clinical setting, and application of a temporary black henna tattoo may also lead to (strong) PPD sensitization.

Key words: CAS no. 106-50-3; hair dye; hairdresser; *p*-aminoaryl compounds; *p*-phenylenediamine; self-administered hair dye pretest; temporary black henna tattoo.

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Contact sensitization to *p*-phenylenediamine (PPD) (CAS no. 106-50-3) is mainly associated with hair dyeing and is a widespread health issue, with frequencies of 4% in patch tested populations (1, 2) and 0.8% in the general population of Europe (3). Frequent contact with (dark-shaded) hair dyes, (single) application of a temporary black henna tattoo adulterated with PPD and working as hairdresser are established risk factors for the development of contact allergy to PPD, whereas pure henna, made from dried leaves of *Lawsonia inermis*, is barely sensitizing (2, 4, 5).

So-called black henna tattoos contain various, sometimes huge, amounts of PPD (up to 64%, although its use is prohibited in skin cosmetics) or chemically related hair dye chemicals, such as toluene-2,5-diamine (PTD), making the colour darker and longer-lasting, and, moreover, making the product extremely sensitizing (6–8).

Self-administered hair dye pretests, often also referred to as ‘self-tests’ or ‘sensitivity tests’, which are recommended by some hair dye producers, have also been suggested to constitute a risk factor for sensitization to PPD (9–11). In order to collect information about these (proven or putative) risk factors for PPD sensitization, six PPD-related questions were added to the Information Network of Departments of Dermatology (IVDK) questionnaire from 2008 to 2013 (Fig. 1). These questions were to be answered by all patients tested with PPD, which was not included in the baseline series in Germany during the study period, but was tested – in an aimed fashion – in the context of a ‘hair cosmetics’ series. The aim of this study was to estimate independently the impacts of hair dyeing, pretests with hair dyes, hairdresser occupation and temporary black henna tattoos on PPD sensitization, thereby also including IVDK basic information (e.g. occupation) and the evaluation of the six PPD-related questions (Fig. 1).

Q1) Has the patient ever dyed hair, beard or eyebrows?
 if yes
 Q2) Did subsequently an eczematous reaction occur?
 Q3) Has she/he performed a self-test (user test) before hair dyeing?
 if yes
 Q4) Behind the ear?
 Q5) On the forearm?
 Q6) Did the patient ever have a temporary henna tattoo?

Fig. 1. Six additional questions concerning *p*-phenylenediamine exposure integrated into the Information Network of Departments of Dermatology questionnaire from 2008 to 2013 (translation from the German original).

Patients and Methods

The IVDK is a network of, currently, 56 departments of dermatology in Germany, Switzerland, and Austria dedicated to the epidemiology of contact allergy. Its structure and routine operating procedures are described in detail elsewhere (12). Briefly, histories, clinical data and patch test (PT) results of all patients tested in the participating centres are recorded in local databases, and, after anonymization, are transmitted to the IVDK central office at the University of Göttingen twice yearly. Data are subjected to standardized quality control, added to the central IVDK database, and analysed according to international standards (13, 14).

For this retrospective data analysis, we selected data of the years 2008–2013 from the central database of the IVDK. All IVDK members are also members of the German Contact Dermatitis Research Group (DKG). Patch testing and evaluation of reactions is performed according to DKG guidelines (15). For this data analysis, PT reactions on day (D) 3 were taken into account. In a few exceptional cases, when a reading was performed on D4 instead of D3, this reading was selected. Readings coded as +, ++ or +++, that is, positive reactions with erythema, infiltration, papules, and/or (coalescing) vesicles, were rated as positive in dichotomized analyses if not otherwise indicated. PT preparations were purchased from Almirall Hermal (Reinbek, Germany). The PT exposure time was 24 h in 18% of the patients, and 48 h in 82%. With a few (temporary) exceptions, Finn Chambers® (inner diameter 8 mm) on Scanpor® tape were used as test chambers. Of the 77 231 patients patch tested during these years, we selected 13 770 patients who were tested with PPD 1% pet. (Fig. 2). The PPD-related questions were not answered by all PPD-tested patients. In order to avoid any selection bias, we split the data into blocks of single centres and quarterly periods, and limited our analysis to those data blocks for which ≥ 90% of the PPD-tested patients had answered at least question 1 (Fig. 1). Altogether, this resulted in a population of 43 14 patients from 33 departments being eligible for our study, that is, 31% of all patients patch tested with PPD. Most of the patients answered every question, and there was no significant difference between PPD-positive and PPD-negative individuals (data not shown). Hence, a bias of the answers towards problems in PPD-sensitized patients can largely be excluded. Questions 1 and 2 concerned hair dyeing and subsequent dermatitis, and question 3 concerned pretests with hair dyes. Questions 4 and 5 were intended to distinguish two distinct pretest protocols that differ considerably in the application time of the undiluted hair dye formulation and thus, probably, concerning the sensitization risk related to the test (16). Customers

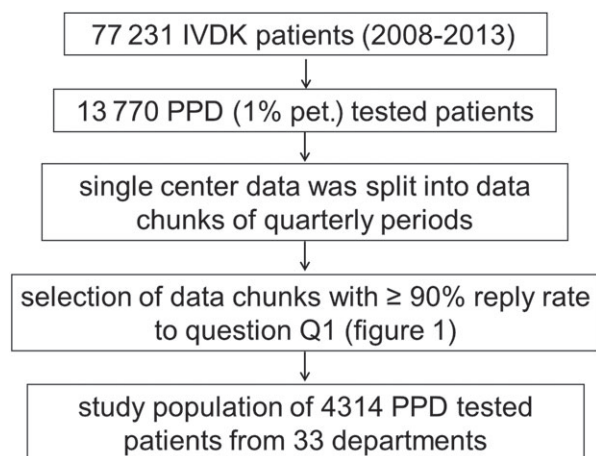


Fig. 2. Outline of patient selection and study population composition. IVDK, Information Network of Departments of Dermatology; PPD, *p*-phenylenediamine.

can easily distinguish between these two protocols by remembering the test localization (behind the ear or on the forearm). Where appropriate, statistical significance ($p < 0.05$) of anamnestic items was calculated with the chi²-test or Fisher's exact test, or was based on non-overlapping 95% confidence intervals (CIs), the latter being used for comparison of crude sensitization frequencies. Logistic regression analyses with dichotomized outcomes were performed, namely (i) strong/extreme positive, (ii) weak positive and (iii) positive reactions to PPD, with negative reaction as a respective reference, and dichotomized explanatory variables (female sex, age ≥ 40 years, history of henna tattoo, hair dyeing and current hairdresser profession). The model was checked with the Hosmer–Lemeshow goodness-of-fit test (H&L) ($p > 0.05$). Results are presented as odds ratio (OR) estimates with profile-likelihood CIs (95%). Data were managed and analysed with the statistical analysis software SAS[®], version 9.4 (SAS Institute, Cary, NC, USA).

Results

By definition, PPD (1% pet.) was tested in every patient in our study cohort ($n = 4314$). Of these, 271 (= 6.3%) tested positive (108 +, 78 ++ and 85 +++), 76 (= 1.7%) had a doubtful/follicular or irritant reaction and 3967 patients (= 92.0%) showed no reaction.

The MOAHLFA indices of the study group and of 271 PPD-positive and 3967 PPD-negative patients are shown in Table 1, together with three additional items. There were more than three times as many hairdressers among PPD-positive patients than among PPD-negative patients

(15.9% versus 4.6%). However, the frequency of hand dermatitis was not increased (34.7% versus 34.1%), whereas there was a seven-fold increase in the frequency of scalp dermatitis (30.3% versus 4.3%). PPD-positive patients mentioned hair cosmetics as the suspected allergen source five times more frequently than PPD-negative patients (59.0% versus 11.7%), and significantly more patients sensitized to PPD were females (79.3% versus 62.7%).

Table 2 shows the proportions of study questions (Fig. 1) answered affirmatively by the study population. Almost 80% of PPD-positive patients dyed their hair, and, among these, the majority (56.9%) suffered from subsequent eczema, and both events occurred significantly more often than in PPD-negative patients. Three times more PPD-positive patients than PPD-negative patients had had a temporary henna tattoo (11.4% versus 3.8%). Strong positive reactions to PPD were significantly more prevalent among patients with a henna tattoo, with 90% (28 of 31 positive reactions) being ++ or +++. In contrast, only 54% (89 of 166) of the positive reactions to PPD among patients who had their hair dyed, but who had not had black henna tattoos, were strong positive reactions ($p < 0.0001$). A self-administered pretest with hair dye was performed by only 69 patients, representing 3.6% of those who had their hair dyed, and this proportion did not differ notably between PPD-positive patients and PPD-negative patients. Hence, no further data analysis was conducted in this respect.

The proportion of PPD-positive patients was significantly higher ($p < 0.0001$) among hairdressers (43/232; 18.5%) than among non-hairdressers (228/4082; 5.6%). In contrast, hairdressers were over-represented among PPD-positive patients compared to PPD-negative patients (15.9% versus 4.6%; $p < 0.0001$). This prompted us to investigate whether consumer behaviour as reflected in our questions had different impacts on PPD sensitization in hairdressers and non-hairdressers (Table 3). Neither dyeing of own hair (with or without subsequent eczema) nor application of a temporary henna tattoo was significantly associated with PPD sensitization among hairdressers, whereas all three items were strongly associated with PPD sensitization in non-hairdressers. As shown in the last column of Table 3, although the proportion of patients using hair dye was comparable between PPD-positive hairdressers and PPD-positive non-hairdressers ($p = 0.12$), eczema after hair dyeing was predominantly seen in PPD-positive non-hairdressers ($p < 0.0001$). Interestingly, only PPD-positive non-hairdressers, and not a single PPD-positive hairdresser, reported a history of temporary henna tattoos ($p = 0.014$). The overlap of these

Table 1. MOAHLEA index and three additional anamnestic items of *p*-phenylenediamine (PPD)-tested patients

Item	Index	PPD-tested (n = 4314)		PPD-positive (n = 271)		PPD-negative (n = 3967)		Chi ² -test
		n	%	n	%	n	%	<i>p</i> -value
Male	M	1563	36.2	56	20.7	1479	37.3	< 0.0001
Occupational dermatitis	O	1044	24.2	77	28.4	941	23.7	0.080
Atopic dermatitis	A	953	22.1	58	21.4	870	21.9	0.81
Hand dermatitis	H	1486	34.4	94	34.7	1352	34.1	0.84
Leg dermatitis	L	360	8.3	8	3.0	349	8.8	0.0008
Face dermatitis	F	625	14.5	26	9.6	588	14.8	0.018
Age ≥ 40 years	A	3075	71.2	178	65.7	2838	71.5	0.039
Present hairdresser profession		232	5.4	43	15.9	182	4.6	< 0.0001
Scalp dermatitis		263	6.1	82	30.3	172	4.3	< 0.0001
Hair cosmetics as suspected allergen source		651	15.1	160	59.0	463	11.7	< 0.0001

p-value = significance level of statistical comparison between patients reacting positively and negatively to PPD (irritant, doubtful or follicular reactions were excluded).

Table 2. Proportions of affirmative responses to the *p*-phenylenediamine (PPD)-related questions

Answer = yes	PPD-tested (n = 4314) % (n)	PPD-positive (n = 271) % (n)	PPD-negative (n = 3967) % (n)	Chi ² -test <i>p</i> -value
Q1: Hair dyed?	43.9 (1893/4314)	79.7 (216/271)	41.3 (1637/3967)	< 0.0001
Q2: Subsequent eczema?	20 (379/1893)	56.9 (123/216)	14.4 (236/1637)	< 0.0001
Q3: Pretest?	3.6 (69/1893)	4.6 (10/216)	3.4 (55/1637)	0.34
Q4: Retroauricular?	21.7 (15/69)	0.0 (0/10)	27.3 (15/55)	0.10
Q5: Forearm?	53.6 (37/69)	80.0 (8/10)	49.1 (27/55)	0.093
Q6: Henna tattoo?	4.3 (185/4314)	11.4 (31/271)	3.8 (151/3967)	< 0.0001

Questions 1–6 were answered with a 'yes' by all patients tested with PPD (1% pet.), as well as by patients reacting positively and negatively to PPD. *p*-value = significance level of statistical comparison (chi²-test) between patients reacting positively and negatively to PPD (irritant, doubtful or follicular reactions were excluded).

three risk factors in PPD-sensitized patients is shown in Fig. 3.

From the literature, it is known that the risk of PPD sensitization increases with age (17). Hence, there are five factors that are definitely or possibly associated with PPD sensitization, that is, sex, age, hair dyeing, exposure to a temporary henna tattoo, and the hairdresser profession, and some of these are strongly related (Table 4). For example, 93.5% of the hairdressers and 91.9% of those who dyed their hair were females, and 85.8% of the hairdressers had dyed their hair.

In order to disentangle the influence of these five determinants and to quantify the risk associated with each factor (controlled for the influence of the other factors mentioned), we performed a logistic regression analysis with these five factors as explanatory (independent) variables and a positive reaction (+ to +++) to PPD (1% pet.) as a dependent variable (Table 5) (H&L, *p* = 0.76). For 3417 patients (79.2% of the study group), information about every explanatory variable was available. Of these, 93 (2.7%) had a weak positive reaction (+) and 138 (4.0%) a strong/extreme positive reaction (++/+++ to PPD, resulting in a total of 231 positive reactions

(6.8%). Application of a temporary henna tattoo [OR 2.4 (95%CI: 1.5–3.7)], hair dyeing [OR 6.0 (95%CI: 3.9–9.4)] and working as hairdresser [OR 2.1 (95%CI: 1.3–3.2)] significantly increased the risk of PPD sensitization, whereas sex and age had no significant influence. We applied two additional logistic regression models, focusing on strong and weak allergic reactions to PPD, respectively, by excluding the respective other group of positive reactions from the dataset (H&L, *p* = 0.67 and *p* = 0.96, respectively). Whereas hair dyeing increased the risk of both weak and strong/extreme allergic reactions (OR 6.3 and OR 5.7, respectively), having a henna tattoo only increased the risk of strong/extreme allergic reactions (OR 3.9) and being a hairdresser only increased the risk of weak positive reactions (OR 2.6) in these subanalyses.

Finally, we analysed concomitant reactions in PPD-positive patients with a history of temporary henna tattoo exposure in comparison with hairdressers and patients who dyed their hair only (Table 6). So-called *p*-aminoaryl compounds (aromatic amines disubstituted in the para-position), such as PTD, *p*-aminophenol, *N*-isopropyl-*N'*-phenyl-*p*-phenylenediamine (IPPD),

Table 3. Selected *p*-phenylenediamine (PPD)-related questions in PPD-positive and PPD-negative hairdressers and non-hairdressers

Questions	Hairdressers (n = 232)							Non-hairdressers (n = 4082)							Comp. PPD-positive
	PPD-positive			PPD-negative			Chi ² -test	PPD-positive			PPD-negative			Chi ² -test	Chi ² -test
	Total	'Yes'	%	Total	'Yes'	%	<i>p</i> -value	Total	'Yes'	%	Total	'Yes'	%	<i>p</i> -value	<i>p</i> -value
Q1: Hair dyed?	43	38	88.4	182	155	85.2	0.59	228	178	78.1	3786	1482	39.1	<0.0001	0.12
Q2: Subsequent eczema?	38	9	23.7	155	24	15.5	0.23	178	114	64.0	1482	212	14.3	<0.0001	<0.0001
Q6: Henna tattoo?	37	0	0.0	165	15	10.4	0.078	216	31	15.5	3622	136	4.5	<0.0001	0.014

p-value = statistical comparison (χ^2 -test) of hairdressers or non-hairdressers reacting positively and negatively to PPD (irritant, doubtful or follicular reactions were excluded). Comp. PPD positive, comparison of PPD-positive hairdressers and non-hairdressers.

Table 4. Risk factors for *p*-phenylenediamine (PPD) sensitization in the study group (n = 4314)

	Female sex	Age ≥ 40 years	Henna tattoo	Hair dyeing	Hairdresser	
Female sex	2751	1965	143	1739	217	n
		71.4	5.2	63.2	7.9	row%
		63.9	77.3	91.9	93.5	col%
Age ≥ 40 years		3075	84	1274	70	n
			2.7	41.4	2.3	row%
			45.4	67.3	30.2	col%
Henna tattoo			185	129	16	n
				69.7	8.6	row%
				6.8	6.9	col%
Hair dyeing				1893	199	n
					10.5	row%
					85.8	col%
Hairdresser					232	

Associations observed between female sex, age (≥ 40 years), history of henna tattoo or hair dyeing and working as hairdresser. The diagonal line lists absolute numbers in the respective category. n, count; row%, percentage of the row; col%, percentage of the column.

hydroquinone, and *tert*-butylhydroquinone, as well as *m*-aminophenol, gave positive reactions significantly more often in patients who had had a temporary henna tattoo than in hairdressers or in patients who had dyed their hair only. Patients with a henna tattoo or hairdressers were significantly younger than hair dye-only users. Hairdressers developed symptoms especially on the hands, and were diagnosed with irritant dermatitis more often than patients in both other groups. Patients with a history of a henna tattoo had predominantly allergic eczemas located on the head, face, or neck, that is, probably reactions to hair dye as a consequence of initial sensitization to PPD-containing henna paste.

Discussion

PPD is the main and indicator allergen for hair dye-associated CA and a 'para substance' (2). Our study concerns a selected subpopulation of patients tested with the DKG hairdressers' series (including PPD 1% pet.). Accordingly, an increased proportion of hair dye users (80%) in comparison with a population of consecutive patients (< 20%) (2) is observed, and generally

the characteristics of the patch tested patients are not comparable to those of consecutively tested patients. Nevertheless, we showed that hair dyeing is associated with a much higher risk of sensitization to PPD [OR 6.0 (95%CI: 3.9–9.4); Table 5] than is being a hairdresser [OR 2.1 (95%CI: 1.3–3.2)] or having a henna tattoo painted [OR 2.4 (95%CI: 1.5–3.7)]. However, according to our data, hair dyeing increases the risk of a weak degree of sensitization, as indicated by a weak positive reaction to PPD and low proportions of concomitant sensitization to other *p*-aminoaryl compounds (except for PTD). In contrast, a henna tattoo increases the risk of strong/extreme PPD sensitization, as reflected by strong/extreme positive reactions to PPD and frequent concomitant reactions to other *p*-aminoaryl compounds. Similarly to hair dye exposure, being a hairdresser increases the risk of a weak allergic PT reaction to PPD, with less frequent broad sensitization to other *p*-aminoaryl compounds. This is in line with an IVDK publication based on data from 2007–2012 comparing hairdressers and clients (18), with a Dutch single-centre study (19), and data from St John's Institute, London, where hairdressers also

Table 5. Logistic regression analyses

Outcome variable	Independent (explanatory) variables, OR (95%CI)				
	Female sex	Age $\geq$ 40 years	Henna tattoo	Hair dyeing	Hairdresser profession
231 positive (+, ++, +++) versus 3186 non-positive	0.69 (0.46–1.05)	1.044 (0.77–1.43)	2.41 (1.54–3.67)	5.99 (3.91–9.37)	2.06 (1.31–3.16)
93 weak positive (+) versus 3048 non-positive	0.84 (0.44–1.69)	1.26 (0.78–2.10)	0.51 (0.12–1.40)	6.34 (3.22–13.3)	2.57 (1.38–4.56)
138 strong/extreme positive (++)/+++ versus 3093 non-positive	0.62 (0.38–1.03)	0.91 (0.62–1.35)	3.87 (2.39–6.12)	5.67 (3.34–9.92)	1.60 (0.89–2.72)

CI, confidence interval; OR, odds ratio; PPD, *p*-phenylenediamine.

Three models were applied, including positive reactions (of different degrees) to PPD as a dependent (outcome) variable, and female sex, age $\geq$ 40 years, history of henna tattoo, hair dyeing and current profession as hairdresser as independent variables. Significant associations are in bold.

Table 6. Concomitant reactions in different subgroups of patients reacting positively to *p*-phenylenediamine (1% pet.)

Allergen	Concentration and vehicle	Henna tattoo, not hairdresser (n = 31)			Hair dyeing only (n = 152)			Hairdresser, no henna tattoo (n = 43)		
		Tested	Positive	% positive (95%CI)	Tested	Positive	% positive (95%CI)	Tested	Positive	% positive (95%CI)
Tolulene-2,5-diamine	1% pet.	27	25	92.6 (75.7–99.1)	114	91	79.8 (71.3–86.8)	42	25	59.5 (43.3–74.4)
<i>p</i> -Aminophenol	1% pet.	26	22	84.6 (65.1–95.6)	112	27	24.1 (16.5–33.1)	43	12	27.9 (15.3–43.7)
<i>m</i> -Aminophenol	1% pet.	26	18	69.2 (48.2–85.7)	110	18	16.4 (10.0–24.6)	42	6	14.3 (5.4–28.5)
Hydroquinone	1% pet.	26	12	46.2 (26.6–66.6)	101	3	3.0 (0.6–8.4)	38	1	2.6 (0.1–13.8)
<i>N</i> -isopropyl- <i>N'</i> -phenyl- <i>p</i> -phenylenediamine	0.1% pet.	30	7	23.3 (9.9–42.3)	143	7	4.9 (2.0–9.8)	35	1	2.9 (0.1–14.9)
Pyrogallol	1% pet.	26	5	19.2 (6.6–39.4)	100	3	3.0 (0.6–8.5)	38	0	0.0 (0.0–9.3)
<i>tert</i> -Butylhydroquinone	1% pet.	19	4	21.1 (6.1–45.6)	93	1	1.1 (0.0–5.8)	32	0	0.0 (0.0–10.9)
Ammonium persulfate	2.5% pet.	23	0	0.0 (0.0–14.8)	100	4	4.0 (1.1–9.9)	42	15	35.7 (21.6–52.0)
First location = head, face, or neck				80.6 (63.7–90.8)			46.8 (40.2–53.4)			6.4 (2.2–17.2)
First location = hands				9.7 (3.4–24.9)			31.0 (25.2–37.5)			85.1 (72.3–92.6)
Chronic irritant dermatitis				0.0 (0.0–11.0)			8.3 (5.3–12.8)			23.4 (13.6–37.2)
Allergic dermatitis				93.5 (79.3–98.2)			68.1 (61.6–73.9)			63.8 (49.5–76.0)
Item		Minimum	Maximum	Mean % (95%CI)	Minimum	Maximum	Mean % (95%CI)	Minimum	Maximum	Mean % (95%CI)
Age (years)		13	50	26.8 (22.0–31.7)	13	82	43.5 (41.4–45.7)	17	60	33.0 (29.3–36.8)

CI, confidence interval.

Crude frequencies of reactions to a selection of allergens and anamnestic items in three subpopulations are shown.

tended to have less stronger reactions to PPD than other professions (20).

At first sight, our results showing a far higher risk of PPD sensitization conferred by hair dyeing ([OR 5.99 (95%CI: 3.91–9.37)] than by henna tattoos [OR 2.41 (95%CI: 1.54–3.67)] seems to contradict the results of a population-based European study, namely, the EDEN study, which was recently published (3). In this study, hair dyeing had no significant effect on PPD sensitization [OR for 'ever hair colorant use' of 0.86 (95%CI: 0.30–2.50)], whereas 'black henna tattoo ever applied' increased the risk nine-fold [OR 9.33 (95%CI: 3.45–25.26)]. There are several differences in study design between the EDEN study and our study, but the two main factors explaining the outcome differences are probably the following.

First, our data were generated by patch testing with PPD 1% pet. in an 8-mm Finn Chamber[®], corresponding to 0.4 mg PPD/cm². In the EDEN study, the TRUE[®] Test with 0.09 mg PPD/cm² was used, which is approximately one-quarter of our dose per unit area. However, the dose per unit area is not the only relevant parameter for the PT outcome (the vehicle also has an influence), albeit a very important one. There is only one study comparing PPD 1% pet. and the TRUE[®] Test with PPD 0.09 mg/cm² intraindividually (21); in this small study, only 7 patients reacted to either of these test preparations. Three had discordant reactions (reaction intensity not mentioned), and 4 had concordantly positive reactions, with 3 of 4 being ++ or +++. Hence, unfortunately, this study cannot provide evidence of diagnostic

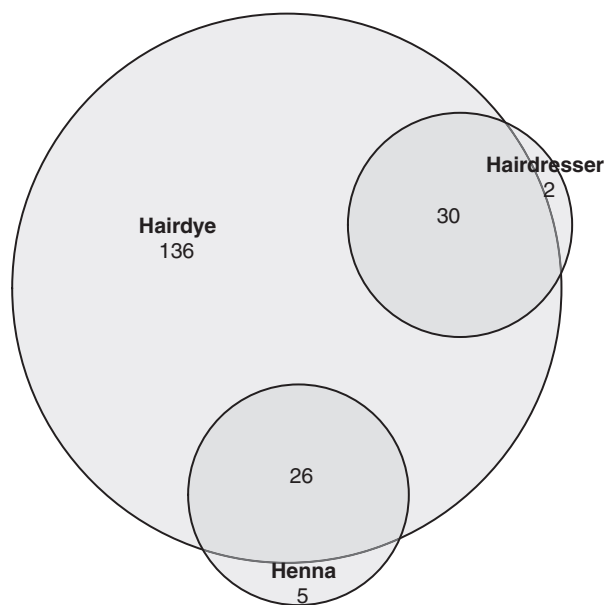


Fig. 3. Area-proportionate Venn (Euler) diagram of exposures (dyeing own hair, having had a temporary henna tattoo, working as a hairdresser) in *p*-phenylenediamine-sensitized patients.

equivalence. From comparative studies with PPD PTs at different concentrations, it is known that lowering the test concentration from 1% pet. to, for example, 0.3% pet. leads to a significant loss of positive reactions, by 25%, and this loss is even more pronounced for weak positive reactions (22, 23). Thus, bearing in mind that hair dyeing mostly leads to a weak degree of PPD sensitization, it is likely that the frequency of PPD sensitization by hair dyeing may have been underestimated in the EDEN study. Second, the EDEN study was a population-based study; consequently, the investigated disease (i.e. sensitization to PPD) was a rare event. Only 22 of 2739 patients tested (0.8%) reacted to PPD. In contrast, our study is based on clinical data, with an overall proportion of 271 of 4314 patients (6.3%) sensitized to PPD. When a rare disease is investigated (with the same test with a given sensitivity and specificity), the relative proportion of false-positive test results increases with decreasing disease prevalence (24). Keeping in mind that PPD reactions in patients sensitized by henna tattoos are often strong or extreme (4), and that these reactions are easily detected by a lower test concentration with the TRUE® Test, it is likely that an overestimation of the risk of PPD sensitization by henna tattoos occurred in the EDEN study. Hence, it seems that an estimation of the relative risk of hair dyeing and having a henna tattoo is more reliable on the basis of clinical data (with a higher prevalence of sensitization) and the established test concentration

of PPD 1% pet. (which is recommended by the ESCD). However, for the clinical diagnosis of (especially young) patients with suspicion of PPD allergy after sensitization to black henna, a gradually increasing PT concentration starting from $\leq 0.05\%$ (to a maximum of 1% pet.) is recommended (25, 26).

Approximately 60% of PPD-positive patients suspected hair cosmetics as the allergen source, and nearly the same proportion (56.9%) developed subsequent eczema reactions after hair dyeing in our analysis (Tables 1 and 2). Hair dyeing was frequent among all women (63.2%), hairdressers (85.8%), and patients who had had a henna tattoo (69.7%) (Table 4). These results are comparable to those of a Danish interview study, in which 74.9% of an unselected female population stated having dyed their hair at least once in a lifetime (27). The risk factor 'hair dyeing' is very common in our study population and, in line with a European study (28), subsequent contact allergic reactions are clear predictors of PPD sensitization (Table 2). However, 180 of 236 PPD-negative patients with eczematous reactions subsequent to hair dyeing were tested with PTD as well, and, in 9 cases, a positive reaction was documented (5.0%). Hence, a small proportion of PPD-negative patients may be 'false negatives', in line with the observation of partial cross-reactivity between PTD and PPD in PTs of approximately 75–80% (2, 29).

Another source of exposure to *p*-aminoaryl compounds is the self-administered pretest, which is encouraged by some hair dye suppliers in order to avoid adverse reactions in already sensitized patients (30). In our study population, the pretest was performed by only 3.6% of the patients, and was not statistically associated with the PPD PT result (Table 2). Although no firm conclusions can be drawn from these data, and these pretests are generally (not just in our study) utilized very infrequently (10), we agree with the statement of the DKG and others that such testing should be avoided, to minimize skin contact with highly potent allergens outside a medical context (9).

In our study, 4.3% of all PPD-tested patients (5.2% of women) stated having had a temporary henna tattoo (Tables 2 and 4). This proportion is comparable to that in a Europe-wide questionnaire study, in which the prevalence of black henna tattoos was 7.2% in female interviewees (3) and 4.8% (weighted average) in an European multicentre study (28). In our investigation, the exact colour of the temporary henna tattoo was not addressed in question 6 (Fig. 1). However, application of a temporary henna tattoo is still a significant risk factor for PPD sensitization (Table 2), owing to the strong allergenic potency of PPD (31).

Contact dermatitis in hairdressers is mostly located on the hands, and that in non-hairdressers is mostly located on the head, face, or neck (18). These circumstances are reflected by our data (Table 6). Of the hairdressers, 85.8% stated that they dyed their hair, but neither hair dyeing ($p = 0.59$) nor a subsequent eczematous reaction ($p = 0.23$) was more common in PPD-positive hairdressers than in PPD-negative hairdressers (Table 3). Furthermore, PPD-positive non-hairdressers significantly more often stated having had an eczematous reaction after hair dyeing or a temporary henna tattoo than PPD-positive hairdressers ($p < 0.0001$ and $p = 0.014$, respectively). Taken together, neither dyeing of own hair, which resulted in eczematous reactions, nor painting of (black) henna tattoos ('private habits') seem to be relevant risk factors for PPD sensitization in hairdressers, whereas these are the main sources of sensitization in the remaining patients ('non-hairdressers'). It is possible that hairdressers, owing to their educational background, are more aware of the hazardous characteristic of PPD in hair dyes or black henna tattoos than the general population ('non-hairdressers'). This probably leads to avoidance of black henna and a preference for unadulterated red henna with low allergenic potency. This would imply that there is still an urgent need for the public to be informed about such health risks.

With regard to concomitant reactions in patients who reacted positively to PPD, we have analysed three groups: cases with henna tattoo (no hairdressers), cases with hair dye only (no henna, no hairdressers), and hairdressers without henna tattoos (Table 6). In accordance with previous results (32), the anamnestic differences between the three groups were reflected not only by the anatomical site affected, but also by the final diagnosis: irritant contact dermatitis was much more frequently diagnosed in hairdressers. Furthermore, the mean ages of hairdressers and that of the henna tattoo group were very low. Especially, the latter group of young PPD-positive patients with temporary henna tattoos showed a significantly

higher frequency of positive PT reactions to a variety of *p*-aminoaryl compounds and *m*-aminophenol. Whereas the frequency of PTD sensitization was increased in comparison with hairdressers only, sensitization to IPPD and sensitization to *tert*-butylhydroquinone were also more frequent than in the subgroup of hair dye users. Several reports have already identified an association between IPPD sensitization and strong or extreme PPD reactions, potentially induced by temporary black henna tattoos (2, 33, 34). *p*-Aminophenol, *m*-aminophenol and hydroquinone reaction prevalences were also higher in cases with a henna tattoo history. Although co-exposure to hair dye chemicals in either hair dyes or black henna preparations cannot be excluded, reactions to substances not utilized in dyes (IPPD and hydroquinone) probably indicate cross-reactivity to PPD (7, 35, 36).

In several reports, increased concomitant reactivity to chemically unrelated baseline series allergens (e.g. metals and essential oils) were noted in PPD-sensitized patients (20, 29, 37). However, we did not find any particular association with the risk factors of PPD sensitization that we had used (data not shown in detail). Hence, such concomitant reactions may indicate independent sensitization, and are possibly attributable to an apparently universal proportion of polysensitized patients in PT populations (38).

Conclusion

By analysing the impact of hair dyeing and henna tattoos on PPD sensitization, independently of each other, we have shown, for the first time, that, generally speaking, hair dyeing bears a considerably greater risk of acquiring PPD sensitization than henna tattoos. However, PPD sensitization acquired via henna tattoos tends to be more intense, resulting in stronger PT reactions and more concomitant reactions to chemically related compounds.

References

- Thyssen J P, White J M. Epidemiological data on consumer allergy to *p*-phenylenediamine. *Contact Dermatitis* 2008; **59**: 327–343.
- Schnuch A, Lessmann H, Frosch P J et al. para-Phenylenediamine: the profile of an important allergen. Results of the IVDK. *Br J Dermatol* 2008; **159**: 379–386.
- Diepgen T L, Naldi L, Bruze M et al. Prevalence of contact allergy to *p*-phenylenediamine in the European general population. *J Invest Dermatol* 2016; **136**: 409–415.
- de Groot A C. Side-effects of henna and semi-permanent 'black henna' tattoos: a full review. *Contact Dermatitis* 2013; **69**: 1–25.
- Warshaw E M, Wang M Z, Mathias C G et al. Occupational contact dermatitis in hairdressers/cosmetologists: retrospective analysis of North American Contact Dermatitis group data, 1994 to 2010. *Dermatitis* 2012; **23**: 258–268.
- Almeida P J, Borrego L, Pulido-Melian E et al. Quantification of *p*-phenylenediamine and 2-hydroxy-1,4-naphthoquinone in henna tattoos. *Contact Dermatitis* 2012; **66**: 33–37.
- Akyüz M, Ata S. Determination of aromatic amines in hair dye and henna

- samples by ion-pair extraction and gas chromatography-mass spectrometry. *J Pharm Biomed Anal* 2008; **47**: 68–80.
- 8 The Commission of the European Communities. Regulation (EC) No. 1223/2009 of the European Parliament and of the Council of 30 November 2009 on cosmetic products. Official Journal of the European Union, 2009. Available at: https://ec.europa.eu/health/sites/health/files/endocrine_disruptors/docs/cosmetic_1223_2009_regulation_en.pdf.
 - 9 Schnuch A, Becker D. Routine preliminary testing of products with oxidative hair colors before hair coloring presents problems and is refused. Position of the leadership of the German contact allergy group. *J Dtsch Dermatol Ges* 2005; **3**: 700–701.
 - 10 Orton D, Basketter D. Hair dye sensitivity testing: a critical commentary. *Contact Dermatitis* 2012; **66**: 312–316.
 - 11 Thyssen J P, Søsted H, Uter W et al. Self-testing for contact sensitization to hair dyes – scientific considerations and clinical concerns of an industry-led screening programme. *Contact Dermatitis* 2012; **66**: 300–311.
 - 12 Schnuch A, Geier J, Lessmann H et al. Surveillance of contact allergies: methods and results of the Information Network of Departments of Dermatology (IVDK). *Allergy* 2012; **67**: 847–857.
 - 13 Uter W, Schnuch A, Gefeller O. Guidelines for the descriptive presentation and statistical analysis of contact allergy data. *Contact Dermatitis* 2004; **51**: 47–56.
 - 14 Uter W, Mackiewicz M, Schnuch A, Geier J. Interne Qualitätssicherung von Epikutantest-Daten des multizentrischen Projektes 'Informationsverbund Dermatologischer Kliniken' (IVDK). *Dermatolo Beruf Umwelt* 2005; **53**: 107–114.
 - 15 Schnuch A, Aberer W, Agathos M et al. Durchführung des Epikutantests mit Kontaktallergenen. Leitlinien der Deutschen Dermatologischen Gesellschaft (DDG) und der Deutschen Gesellschaft für Allergie und Klinische Immunologie (DGAKI). *J Dtsch Dermatol Ges* 2008; **6**: 770–775.
 - 16 European Commission. SCCP (Scientific Committee on Consumer Products). Opinion on sensitivity to hair dyes – consumer self testing, 2007. Available at: https://ec.europa.eu/health/ph_risk/committees/04_sccp/docs/sccp_o_114.pdf
 - 17 Uter W, Gefeller O, Geier J et al. *Untersuchungen zur Abhängigkeit der Sensibilisierung gegen wichtige Allergene von arbeitsbedingten sowie individuellen Faktoren*. Schriftenreihe der Bundesanstalt für Arbeitsschutz und Arbeitsmedizin, Fb 949: Bremerhaven, Wissenschaftsverlag NW, 2002.
 - 18 Uter W, Gefeller O, John S M et al. Contact allergy to ingredients of hair cosmetics – a comparison of female hairdressers and clients based on IVDK 2007–2012 data. *Contact Dermatitis* 2014; **71**: 13–20.
 - 19 Vogel T A, Coenraads P J, Bijkersma L M et al. p-Phenylenediamine exposure in real life – a case-control study on sensitization rate, mode and elicitation reactions in the northern Netherlands. *Contact Dermatitis* 2015; **72**: 355–361.
 - 20 Ho S G, Basketter D A, Jefferies D et al. Analysis of para-phenylenediamine allergic patients in relation to strength of patch test reaction. *Br J Dermatol* 2005; **153**: 364–367.
 - 21 Lazarov A, David M, Abraham D, Trattner A. Comparison of reactivity to allergens using the TRUE Test and IQ chamber system. *Contact Dermatitis* 2007; **56**: 140–145.
 - 22 Geier J, Ballmer-Weber B K, Dickel H et al. Monitoring contact sensitization to p-phenylenediamine (PPD) by patch testing with PPD 0.3% in petrolatum. *Contact Dermatitis* 2013; **69**: 26–31.
 - 23 Hillen U, Dickel H, Löffler H et al. Late reactions to patch test preparations with reduced concentrations of p-phenylenediamine: a multicentre investigation of the German Contact Dermatitis Research Group. *Contact Dermatitis* 2011; **64**: 196–202.
 - 24 Diepgen T L, Coenraads P J. Sensitivity, specificity and positive predictive value of patch testing: the more you test, the more you get? ESCD Working Party on Epidemiology. *Contact Dermatitis* 2000; **42**: 315–317.
 - 25 Goebel C, Coenraads P J, Rothe H et al. Elicitation of the immune response to p-phenylenediamine in allergic patients: the role of dose and exposure time. *Br J Dermatol* 2010; **163**: 1205–1211.
 - 26 Spornraft-Ragaller P, Kammerer E, Gillitzer C et al. Severe allergic reactions to para-phenylenediamine in children and adolescents: should the patch test concentration of PPD be changed? *J Dtsch Dermatol Ges* 2012; **10**: 258–264.
 - 27 Søsted H, Hesse U, Menné T et al. Contact dermatitis to hair dyes in a Danish adult population: an interview-based study. *Br J Dermatol* 2005; **153**: 132–135.
 - 28 Thyssen J P, Andersen K E, Bruze M et al. p-Phenylenediamine sensitization is more prevalent in central and southern European patch test centres than in Scandinavian: results from a multicentre study. *Contact Dermatitis* 2009; **60**: 314–319.
 - 29 Basketter D A, English J. Cross-reactions among hair dye allergens. *Cutan Ocul Toxicol* 2009; **28**: 104–106.
 - 30 Krasteva M, Cottin M, Cristaudo A et al. Sensitivity and specificity of the consumer open skin allergy test as a method of prediction of contact dermatitis to hair dyes. *Eur J Dermatol* 2005; **15**: 18–25.
 - 31 Kligman A M. The identification of contact allergens by human assay. 3. The maximization test: a procedure for screening and rating contact sensitizers. *J Invest Dermatol* 1966; **47**: 393–409.
 - 32 Uter W, Lessmann H, Geier J, Schnuch A. Contact allergy to ingredients of hair cosmetics in female hairdressers and clients – an 8-year analysis of IVDK data. *Contact Dermatitis* 2003; **49**: 236–240.
 - 33 Hald M, Menné T, Johansen J D, Zachariae C. Severe occupational contact dermatitis caused by black rubber as a consequence of p-phenylenediamine allergy resulting from a temporary henna tattoo. *Contact Dermatitis* 2013; **68**: 377–379.
 - 34 Thomas B R, White I R, Mcfadden J P et al. Positive relationship-intensity of response to p-phenylenediamine on patch testing and cross-reactions with related allergens. *Contact Dermatitis* 2014; **71**: 98–101.
 - 35 Uter W, Lessmann H, Geier J et al. The spectrum of allergic (cross-)sensitivity in clinical patch testing with 'para amino' compounds. *Allergy* 2002; **57**: 319–322.
 - 36 Kirchlechner S, Hübner A, Uter W. Survey of sensitizing components of oxidative hair dyes (retail and professional products) in Germany. *J Dtsch Dermatol Ges* 2016; **14**: 707–715.
 - 37 Vogel T A, Heijnen R W, Coenraads P J, Schuttelaar M A. Two decades of p-phenylenediamine and toluene-2,5-diamine patch testing – focus on co-sensitizations in the European baseline series and cross-reactions with chemically related substances. *Contact Dermatitis* 2016; **76**: 81–88.
 - 38 Schwitulla J, Uter W. Multiple contact sensitization. *Hautarzt* 2015; **66**: 680–685.