

Elicitation response characteristics to permanent hair dye in paraphenylenediamine-allergic volunteers

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Background: Elicitation response characteristics to complete permanent hair dye products in paraphenylenediamine (PPD)-allergic volunteers have not previously been explored in detail.

Objectives: To assess the elicitation response characteristics observed in PPD-allergic volunteers upon patch testing with complete hair dyes.

Methods: PPD-allergic volunteers were assigned to 1 of 3 groups depending upon whether they elicited + (group 1), ++ (group 2) or +++ (group 3) reactions following the standard diagnostic procedure. Each group was subsequently patch tested with 2 complete hair dyes (A and B) for 30 min, 1 hr and 24 hr. Patch sites were examined 1 day, 2 days and 3 days after patch removal.

Results: Exposure to either hair dye for 30 min or 1 hr was insufficient to yield positive patch test reactions in all of the PPD-allergic patients in groups 1 or 2. Application of either hair dye for 24 hr was sufficient to yield positive reactions in all of the individuals within groups 2 and 3.

Conclusions: The frequency of positive patch test reactions observed following 24-hr exposure to complete permanent hair dyes is comparable to that observed following 48-hr exposure to 1% PPD/petrolatum in those individuals whose degree of sensitization is such that they typically present ++ or +++ reactions diagnostically.

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There exist 3 common classes of hair dyes (permanent, semi-permanent and temporary), and allergic contact dermatitis has been associated with the use of some chemicals in all 3 classes (1). Permanent (oxidative) hair dyes consist of 2 components that are combined immediately prior to use: a 'dye' component and an oxidative 'developer' component. The dye component typically comprises primary intermediates [e.g. paraphenylenediamine (PPD), toluene-2,5-diamine, *p*-aminophenol or *o*-aminophenol] and a coupler (e.g. *m*-phenylenediamine, *m*-aminophenol or resorcinol). The oxidizing component typically contains hydrogen peroxide, urea peroxide or sodium percarbonate.

While many of the components of permanent hair dyes are contact allergens, PPD is generally recognized as the predominant sensitizer responsible for hair dye dermatitis (2). Predictive animal methods such as the local lymph node assay also clearly identify PPD as a strong sensitizer (3). Despite this, PPD is still used extensively as a primary intermediate in permanent hair dyes. The European Cosmetics Directive 76/768 stipulates that the levels of PPD in this product type must

not exceed 6% (typically resulting in an 'in use' concentration of 3% following combination with the developer component), although in practice, PPD concentrations rarely exceed 4% (typically resulting in an 'in use' concentration of 2%) (4).

Hair dye allergy is typically diagnosed through patch testing with 1% PPD, and this is generally considered a reliable screen. However, there has been much debate about PPD patch testing over recent years. For example, 1% PPD has been found to elicit bullous or erosive patch test reactions, particularly among individuals who were sensitized via temporary black henna tattoos. This has led to the proposal that such patients should be exposed initially to only 0.01% PPD and that this concentration be successively increased by an order of magnitude (up to a maximum of 1%) if no reaction is observed at the first reading (5). In-home or in-salon testing with hair dye products has also been proposed as means to identify hair-dye-allergic individuals before they proceed with a colouring treatment (4), and this process is recommended in leaflets provided with many products currently marketed.

As such, there is continued interest in the elicitation response characteristics observed in hair-dye-allergic individuals. Such studies have been recorded in the literature. Sosted et al. (6) examined PPD elicitation responses at various anatomical regions. When applied to the back, concentrations of PPD ranging from 50 parts per million (p.p.m.) to 10 000 p.p.m. were shown to elicit positive reactions in PPD-allergic patients, with the proportion of individuals found to react ranging from 7% to 87% ($n = 15$). The elicitation 'threshold' (defined as the concentration at which 10% of the patients elicited + or stronger reactions) was also calculated and was found to be similar for the back (38 p.p.m.), upper arm (56 p.p.m.) and the retroauricular region (75 p.p.m.). McFadden et al. (7) examined the effect of patch duration on the elicitation response to different concentrations of PPD. Following exposure for 120 min, 11 of 16 subjects were found to react to 1% PPD versus 2 of 9 who reacted to 0.01% PPD. Furthermore, following exposure for 15 min, 6 of 16 subjects were found to react to 1% PPD versus 0 of 9 who reacted to 0.01% PPD.

Here, we also consider elicitation response characteristics in PPD-allergic individuals, but do so for the first time with complete permanent hair dyes following combination with the oxidizing component in a characterized panel of PPD-allergic volunteers known to elicit reactions of varying severity to 1% PPD in the context of the standard diagnostic procedure. This study facilitates an assessment of the elicitation response characteristics obtained with complete permanent hair dyes.

Materials and Methods

Volunteer selection

The study was conducted at the Institute of Dermatology, 420/7 Rajavithi Road, Bangkok 10400, Thailand, in PPD-allergic volunteers aged ≥ 18 years and in good health. The protocol was approved by the Ethical Review Committee of the Department of Medical Services, Institute of Dermatology.

Test materials

2 permanent black hair dyes (A and B) were used for the study, each comprising a dye and oxidative developer component.

Volunteer recruitment

3 groups of 15 PPD-allergic volunteers who exhibited +, ++ or +++ readings 2 days after patch removal in the standard PPD diagnostic procedure were recruited. Thus, 1% PPD petrolatum (pet.) was applied to the arm for 48 hr in Finn Chambers[®] (Epitest Oy, Tuusula, Finland). Patch site readings were taken 2 days later according to the International Contact Dermatitis Research Groups (ICDRG) scale (8). Based upon this 2-day reading, each volunteer was assigned to the +, ++ or +++ group and sufficient volunteers were recruited to achieve 15 individuals in each group. A naïve panel of volunteers ($n = 15$) without any history of hair dye allergy and negative patch test reaction to 1% PPD were also recruited.

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Test method

Each formulation was prepared by combining the 'dye' and 'developer' components at the required ratio immediately prior to patch application. The samples were then applied to each volunteer's arm in separate Finn Chambers[®] for 30 min, 1 hr and 24 hr. Patch sites were scored for evidence of skin reaction 1 day, 2 days and 3 days after patch removal.

Results

Initially, the effect of the 2 complete hair dye formulations was examined in naïve individuals who were shown to have negative patch test reactions to 1% PPD. No noteworthy skin reactions were observed in any of the 15 volunteers following exposure to either dye for 30 min, 1 hr or 24 hr (Tables 1 and 5).

The effect of the 2 hair dyes was next examined in 3 separate groups of PPD-allergic volunteers, each comprising 15 individuals found to react to the standard diagnostic procedure (48-hr exposure to 1% PPD/pet.), with + (group 1), ++ (group 2) or +++ (group 3) reactions. The objective here was to study the elicitation response characteristics associated with complete permanent hair dyes as a function of exposure time and underlying degree of PPD sensitization. The results are presented in Tables 2–4 and 6–8.

Patch site readings were taken 1 day, 2 days and 3 days after patch removal. As expected, elicitation reactions were typically found to develop between the 1-day and 2-day readings and to persist until the 3-day reading. For ease of interpretation, subsequent data analysis will focus only upon the 2-day reading.

30-min exposure

In all cases, 30-min exposure to hair dye A did not elicit positive skin reactions in all the PPD-allergic volunteers within a given group. In group 1 (+ reactions following 48-hr exposure to 1%

Table 1. Hair dye A – naïve group: no history of hair dye allergy (12 females and 3 males; age 21–60 years)

Exposure time	1-day reading					2-day reading					3-day reading				
	+	++	+++	Total	%	+	++	+++	Total	%	+	++	+++	Total	%
30 min	0	0	0	0/15	0	0	0	0	0/15	0	0	0	0	0/15	0
1 hr	0	0	0	0/15	0	0	0	0	0/15	0	0	0	0	0/15	0
24 hr	0	0	0	0/15	0	0	0	0	0/15	0	0	0	0	0/15	0

Table 2. Hair dye A – group 1: + reaction to 1% paraphenylenediamine (9 females and 6 males; age 28–53 years)

Exposure time	1-day reading					2-day reading					3-day reading				
	+	++	+++	Total	%	+	++	+++	Total	%	+	++	+++	Total	%
30 min	0	0	0	0/15	0	0	0	0	0/15	0	0	0	0	0/15	0
1 hr	0	0	0	0/15	0	1	0	0	1/15	7	0	0	0	0/15	0
24 hr	0	0	0	0/15	0	8	1	0	9/15	60	11	1	0	12/15	80

Table 3. Hair dye A – group 2: ++ reaction to 1% paraphenylenediamine (11 females and 4 males; age 23–55 years)

Exposure time	1-day reading					2-day reading					3-day reading				
	+	++	+++	Total	%	+	++	+++	Total	%	+	++	+++	Total	%
30 min	1	0	0	1/15	7	4	3	0	7/15	47	5	3	0	8/15	53
1 hr	2	0	0	2/15	13	5	3	0	8/15	53	8	2	0	10/15	67
24 hr	7	4	0	11/15	73	4	11	0	15/15	100	6	9	0	15/15	100

Table 4. Hair dye A – group 3: +++ reaction to 1% paraphenylenediamine (9 females and 6 males; age 29–53 years)

Exposure time	1-day reading					2-day reading					3-day reading				
	+	++	+++	Total	%	+	++	+++	Total	%	+	++	+++	Total	%
30 min	6	1	1	8/15	53	4	6	4	14/15	93	5	5	4	14/15	93
1 hr	6	2	1	9/15	60	4	7	3	14/15	93	4	7	3	14/15	93
24 hr	4	5	3	12/15	80	0	4	11	15/15	100	1	5	9	15/15	100

PPD/pet.), none of the volunteers was found to react. Positive patch test reactions were observed in 47% and 93% of the volunteers in groups 2 (++ reactions following 48-hr exposure to 1% PPD/pet.) and 3 (+++ reactions to 1% PPD/pet.), respectively. A similar profile was observed with hair dye B, although the overall number of positive reactions following 30-min exposure was generally higher (0%, 53% and 100% for groups 1, 2 and 3, respectively). By contrast to hair dye A, 30-min exposure to hair dye B was sufficient to identify all the highly sensitized PPD-allergic volunteers in group 3.

1-hr exposure

The impact of increasing the exposure time to 1 hr was slight. Again, hair dye A did not yield positive patch test reactions in all of the PPD-allergic individuals within any group. In group 1, 7% of the volunteers elicited a positive reaction, while the response in groups 2 and 3 was 53% and 93%, respectively. The increased exposure time had a comparable impact with hair dye B, with the fre-

quency of responses observed in groups 1, 2 and 3 increasing to 13%, 67% and 100%, respectively.

24-hr exposure

Increasing the exposure time to 24 hr was associated with a more dramatic increase in the frequency of positive patch test reactions. Under these conditions, hair dye A was found to give positive reactions in 60% of the volunteers in group 1 and 100% of the volunteers in groups 2 and 3. The profile obtained with hair dye B was similar, with positive reactions observed in 73%, 100% and 100% of volunteers in groups 1, 2 and 3, respectively.

Patch test scores

The elicitation responses obtained with the 2 hair dyes was also associated with a general reduction in patch site scores, relative to those obtained originally with 1% PPD/pet. For example, while 24-hr exposure to either hair dye A or B was found to yield positive reactions in all volunteers in group

Table 5. Hair dye B – naïve group: no history of hair dye allergy (12 females and 3 males, age 21–60 years)

Exposure time	1-day reading					2-day reading					3-day reading				
	+	++	+++	Total	%	+	++	+++	Total	%	+	++	+++	Total	%
30 min	0	0	0	0/15	0	0	0	0	0/15	0	0	0	0	0/15	0
1 hr	0	0	0	0/15	0	0	0	0	0/15	0	0	0	0	0/15	0
24 hr	0	0	0	0/15	0	0	0	0	0/15	0	0	0	0	0/15	0

Table 6. Hair dye B – group 1: + reaction to 1% paraphenylenediamine (9 females and 6 males; age 28–53 years)

Exposure time	1-day reading					2-day reading					3-day reading				
	+	++	+++	Total	%	+	++	+++	Total	%	+	++	+++	Total	%
30 min	0	0	0	0/15	0	0	0	0	0/15	0	2	0	0	2/15	13
1 hr	1	0	0	1/15	7	2	0	0	2/15	13	3	0	0	3/15	20
24 hr	2	0	0	2/15	13	11	0	0	11/15	73	12	0	0	12/15	80

Table 7. Hair dye B – group 2: ++ reaction to 1% paraphenylenediamine (11 females and 4 males; age 23–55 years)

Exposure time	1-day reading					2-day reading					3-day reading				
	+	++	+++	Total	%	+	++	+++	Total	%	+	++	+++	Total	%
30 min	6	0	0	6/15	40	5	3	0	8/15	53	6	3	0	9/15	60
1 hr	8	0	0	8/15	53	7	3	0	10/15	67	9	2	0	11/15	73
24 hr	8	5	0	13/15	87	3	12	0	15/15	100	6	8	1	15/15	100

Table 8. Hair dye B – group 3: +++ reaction to 1% paraphenylenediamine (9 females and 6 males; age 29–53 years)

Exposure time	1-day reading					2-day reading					3-day reading				
	+	++	+++	Total	%	+	++	+++	Total	%	+	++	+++	Total	%
30 min	6	5	2	13/15	87	5	6	4	15/15	100	5	6	4	15/15	100
1 hr	8	3	2	13/15	87	4	8	3	15/15	100	4	7	4	15/15	100
24 hr	3	5	4	12/15	80	0	4	11	15/15	100	1	7	7	15/15	100

3, the number of patch test readings lower than +++ was 27%.

Discussion

This study was designed to explore in detail the elicitation response characteristics to complete permanent hair dye products in a characterized panel of PPD-allergic volunteers. There is currently much interest in this area due to concerns associated with the alleged risk of active sensitization occurring as a result of the current diagnostic procedure with 1% PPD/pet. (9–13) and the utility of in-home or in-salon pre-tests for hair dye allergy (4).

The approach taken was to recruit panels of volunteers who were found to elicit +, ++ or +++ reactions to 1% PPD/pet. in the context of the current diagnostic procedure. While it was initially thought possible that irritant skin reactions to the test materials could confound the interpretation of patch tests, no such skin reactions were observed in the panel of naïve volunteers similarly exposed to the complete hair dyes (Table 1 and

Table 5). The impact of patch application time in this study was particularly informative. While the 30-min and 1-hr exposures were insufficient to elicit positive patch test reactions in 100% of the volunteers in groups 1 or 2, the frequency of positive reactions was much higher in group 3 (93% and 100% for hair dyes A and B, respectively). By contrast, 24-hr exposure to either hair dye product did elicit positive patch test reactions in 100% of the individuals in groups 2 and 3. These data demonstrate that the frequency of positive patch test reactions observed following 24-hr exposure to complete permanent hair dye products is comparable to that observed following 48-hr exposure to 1% PPD/pet., particularly in those individuals whose degree of sensitization is such that they typically present ++ or +++ reactions diagnostically.

Interestingly, Ho et al. (14) recently analysed PPD-allergic patients in relation to strength of patch test reactions in the standard diagnostic procedure and found that patients with stronger (++ or +++) reactions are more likely to have a clear history of reacting to hair dye and are less likely to be still dyeing their hair. Taken together,

these observations suggest that 24-hr occluded exposure to permanent hair dye products may be sufficient to identify clinically relevant cases of hair dye allergy (++ and +++ reactors) and approximately two thirds of those individuals who are likely to be still able to dye their hair (+ reactors to 1% PPD/pet.). These observations suggest that exposure to hair dyes for at least 24 hr may be appropriate for in-home testing.

It is also noteworthy that the final concentration of PPD achieved with hair dye A was only 0.5%. While the final concentration of PPD in hair dye B is not known, the higher frequency of positive patch test reactions observed with this product suggests that it may be slightly greater than 0.5%. However, Sosted et al. (6) noted previously that within a population of 15 PPD-allergic individuals, an equivalent frequency of positive patch test reactions was elicited following 48-hr patch testing with 1% PPD or 0.5% PPD (87%). Furthermore, 0.1% PPD and 0.05% PPD still yielded similar frequencies of positive reactions (80% and 73%, respectively). McFadden et al. (7) also found that 2-hr exposure of PPD-allergic volunteers to 1% or 0.3% PPD yielded comparable frequencies of positive patch tests [69% (11/16) and 67% (6/9), respectively]. Furthermore, the patch test scores obtained with these 2 concentrations were also comparable. Nevertheless, allergic individuals should be made aware of the potential for differences in the severity of elicitation response to occur as a result of changing to an alternative hair dye product.

While the current study has in common with other published data, an exploration into the impact of duration of exposure, it does so for the first time using complete permanent hair dye products in the context of allergic volunteers whose response to 1% PPD/pet. in the standard diagnostic procedure has been rigorously characterized. It is hoped that this study will help advance our understanding in this area.

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