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Continuous usage of a hair dye product containing 2-methoxymethyl-p-phenylenediamine by hair dye allergic individuals

Running head: Responses to 2-methoxymethyl-p-phenylenediamine containing hair dye in allergic individuals

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article keywords: permanent hair dyeing, cross-elicitation, p-phenylenediamine (PPD), 2-methoxymethyl-p-phenylenediamine (ME-PPD), allergic contact dermatitis

Conflict of interest:

The authors Carsten Goebel and Monika Kock are employees of the Procter & Gamble Company. The hair dye ingredient studied in this paper is currently used in commercial products marketed by the Procter & Gamble Company. The authors Brunhilde Blömeke and Pieter-Jan Coenraads participated as experts in skin sensitization. The authors alone are responsible for the content and writing of the paper.

Bulleted statements:

What's already known about this topic?

PPD allergic individuals had a lower response rate to forearm testing simulating hair dye use when the hair dye product contained ME-PPD instead of PPD.

What does this study add?

This study investigates if PPD allergic individuals tolerate full hair dyeing with their selected ME-PPD hair dye shade, if they did not respond to forearm pre-testing with that hair dye at day 2.

Abstract

Background: Despite a positive patch test reaction to *p*-phenylenediamine (PPD) and/or toluene-2,5-diamine (PTD) many people attempt to continue dyeing their hair with products containing PPD or its derivatives.

Objectives: Investigation of elicitation reactions among PPD/PTD allergic individuals to hair dye products containing the less-sensitizing PPD-derivative 2-methoxymethyl-p-phenylenediamine (ME-PPD).

Methods: Elicitation reactions were studied in 43 PPD/PTD allergic individuals by a 45-min pre-test with a ME-PPD containing hair dye on their forearm and upon negative result followed by exposure to subsequent hair colour treatment(s).

Results: 38/43 PPD/PTD allergic individuals did not develop an elicitation reaction during the pre-test with ME-PPD containing hair dye products and were eligible for subsequent hair colour treatments. Of these 38 PPD/PTD allergic individuals 29 tolerated subsequent hair dyeing with ME-PPD containing hair dye products while 9 showed mild to moderate allergic reactions upon the first hair colour treatment.

Conclusions: Hair dye products with the less sensitizing ME-PPD were tolerated by 29/43 (67%) PPD/PTD allergic individuals throughout continued hair dyeing with an average of 9 treatments per year. Five individuals reacted upon pre-testing while only mild to moderate skin reactions occurred upon hair dyeing in 9 individuals who were not identified by the pre-test. To our knowledge this is the first study among PPD/PTD allergic individuals indicating that a negative 45 min pre-testing with a hair dye product helps to avoid severe allergic reactions.

Introduction

Allergic contact dermatitis related to the use of hair dye products is an important health concern for a certain subgroup among consumers. This is mainly related to the use of the primary intermediates *p*-phenylenediamine (PPD) and toluene-2,5-diamine (PTD) providing excellent hair coloring performance while being considered as major contact allergens among hair dye precursors. Less frequently, also other aromatic amine hair dyes are involved.¹

Following the idea of primary prevention of skin sensitisation and of the onset of contact dermatitis², the recently developed precursor 2-methoxymethyl-*p*-phenylenediamine (ME-PPD)³ provides excellent hair coloring performance^{4,5} with significantly lower skin sensitizing properties compared to PPD or the structurally related compound PTD³ (for chemical structures see Fig. 1). Consequently, based on the available pre-clinical data the risk of allergy induction through ME-PPD is considered very low for hair color consumers because the maximal on-head exposure is more than 100 fold lower than the allergy induction threshold (8.8 versus 1075 µg/cm²). For PPD and PTD the values are much closer to the threshold (i.e. 16.1 versus 27.5 and 22.7 versus 41.3 µg/cm², respectively).³

In contrast to sensitization induction, there is no standard predictive approach to the identification of safe exposure levels for elicitation in hair dye allergic individuals,⁶ nor can this be predicted from knowledge of the relative potency for the induction of skin sensitization.⁷ For oxidative hair dyes,

cross-reactions between PPD, PTD and other para-substituted benzene components are frequently reported.^{1,6,8,9} Consequently, dermatologists generally recommend discontinuing the use of oxidative hair colors for individuals allergic to PPD or PTD to avoid cross reaction with other para-substituted benzene compounds. Nevertheless, many hair dye product consumers consider hair dyeing as an important personal-care need¹⁰ in particular when related to grey hair coverage and continue to use hair dyes even when they have been advised to discontinue using respective hair colorants.^{6,11}

In a recent study in the Netherlands our group investigated if PPD-allergic individuals (with a documented history of hair dye-related allergy) develop cross-reactions to ME-PPD containing hair dye test products when exposing their forearm simulating typical oxidative hair dye usage conditions (i.e. application of the hair colour tint mixed with hydrogen peroxide for approximately 30-45 min followed by rinsing). Cross-elicitation to the ME-PPD containing hair dye test product occurred in 9/30 (30%) while 70% did not react.¹²

In the present study in Germany, we analyzed if hair dye allergic individuals that did not respond to a 45 min exposure on their forearm (simulating hair dyeing conditions) would tolerate repeated and continued full hair colour treatments at their hairdresser. We report the results of the (cross-) elicitation pre-test as well as the experience with continuous hair colour treatments over an observation period of one year.

Materials and Methods

Cases

Individuals (n=43 females) who had contacted our consumer service because they were interested in the PPD/PTD alternatives due to their history of allergic contact dermatitis related to hair dyeing were recruited in Germany. Inclusion criteria were intolerance of hair dye usage and a PPD and/or PTD positive diagnostic patch test(s). For all 43 individuals a relevant contact allergy to PPD and/or PTD was documented by a dermatologist in their individual allergy card. For 17/43 individuals the allergy card contained information on the strength of the diagnostic patch test reaction to PPD and/or PTD and for 26/43 individuals the strength of the patch test response was not specified (referred to by the abbreviation nsp).

Pre-testing with ME-PPD containing hair dye products

The participants were asked to choose their preferred shade of 10 existing ME-PPD containing hair dye products with a ME-PPD on-head concentration range (free base) from 0.15% $\pm$ 0.2 to 1.0% $\pm$ 0.2. These shades were grouped into three tonal shade levels (dark to medium-dark, medium-light, light) with appropriate concentration ranges of ME-PPD (see Table 1). The formulation base was equivalent to conventional hair dye products, comparable to Blömeke *et al.*¹² and Goebel *et al.*¹³,

additionally containing fragrance. In addition to ME-PPD the following hair dye precursors were used according to the desired shade: m-aminophenol, resorcinol, 2-methylresorcinol, hydroxyethyl-3,4-methylene-dioxylaniline HCL, 1-hydroxyethyl-4,5-diamino pyrazole sulfate, 2,4-diaminophenoxyethanol HCL, 4-amino-2-hydroxytoluene, 2-methyl-5-hydroxyethylaminophenol. Shades and developer (hydrogen peroxide solution 6% w/w; Welloxon) were supplied by Procter & Gamble, Schwalbach, Germany. Materials were sent to the individual dermatologist who conducted product pre-testing with the shade of choice to assess cross-elicitation reactions to ME-PPD as well as reactions to other dye or non-dye ingredients. The ready-to-use colour cream was prepared immediately before testing by mixing equal amounts of the colour tint and the developer. By providing detailed written instructions (following the protocol of the Allergy Alert Test, see http://www.colourwell-colourwise.eu/allergy_alert.html) and syringes (5 ml, 1 ml), the dermatologist was guided to apply an amount of 0.2 ml of the ready-to-use colour cream on the inner forearm to an application area of 1 cm² for 45 minutes (Fig. 2). Readings were performed at 45 minutes directly after the colour cream had been removed by rinsing (with wet tissue) and after 48 hours (D2). Responses were graded in reaction categories according to the German Contact Dermatitis Research Group (DKG) comparable to the International Contact Dermatitis Research Group (ICDRG) criteria.^{14,15} For documentation of the results an evaluation sheet including a scoring scheme (see Table 2) was provided.

Full hair colour treatments with ME-PPD containing hair dye products

Individuals with a negative pre-testing result of the shade of choice were sent to their hairdresser to receive a full hair colour treatment with that shade. Supply with the shade through the hairdresser continued for the entire study period between February 2013 and May 2014. For the purpose of continued product supply the individual's hairdresser had to provide feedback (written or by phone call) about the number of colour treatments applied and the corresponding compatibility. All participants were instructed to report to their dermatologist or directly to the study coordinator at any time should any adverse reaction occur.

At termination of the study (April to May 2014) all individuals were queried by telephone about continuous product compatibility. Finally, all individuals and their hairdresser received the information that a corresponding ME-PPD hair dye product is now commercially available in combination with emphasizing once more that careful observation of the product compatibility is of utmost importance for them in collaboration with their dermatologist and hairdresser.

Results

Diagnostic patch test information at study onset and conduct of study

The study population was comprised of 43 females with a documented history of contact dermatitis to PPD and/or PTD related to hair dyes. The age range was 36-80 years (mean age 62 years), and for one individual no age indication was available. For 17 individuals details of the strength of their diagnostic patch test response for PPD and/or PTD were available: for PPD (13=+++, 2=++) and/or

PTD (3=+++ , 5=++ , 1=+) according to ICDRG criteria, while for 26 no further specification (nsp) of the strength of the positive patch test response was available from the allergy card provided by their dermatologist (see Fig 3 for a flow diagram with an overview of the study).

Shade selection

Each individual selected the ME-PPD containing shade of choice according to tone levels from black to very light blond (see Table 1): 26 selected dark to medium-dark shades, 10 selected medium-light shades and 7 preferred light shades containing $1.0\% \pm 0.2$, $0.5\% \pm 0.1$, and $0.15\% \pm 0.1$ ME-PPD, respectively.

Pre-testing

The dermatologist of each of the 43 individuals received the selected shade plus developer for the 45-min pre-testing on the forearm together with detailed guidance how to generate and apply 0.2 ml of the ready to use hair dye cream (see Fig. 2). Consequently, the applied dose was considered to be 200 mg hair dye product/cm² equivalent to an average applied ME-PPD dose of 2000, 1000 and 200 µg/cm², respectively. D2 reading was chosen, because it is the current industry recommendation for allergy alert testing on hair color products (see http://www.colourwellcolourwise.eu/allergy_alert.html) taking into account that readings at further days in practice are considered to affect compliance negatively. In addition, readings at 45 minutes directly after the colour cream was removed were considered useful to investigate if immediate contact reactions occur. At the 45 min reading time no reactions were observed. At the 48 h reading time (D2) there was a reaction in 4 individuals (9%) (see Table 3). At day 5 after the pre-testing exposure, one individual showed a positive reaction that was not obvious at D2.

These 5 individuals did not meet the criteria for hair colour treatments with the selected shade. Thus, 38 individuals were eligible to receive the first full hair colour treatment at their hairdresser (see Fig 3).

Full hair colour treatments

Upon the first and subsequent full hair colour treatments, 29 of the 38 PPD/PTD allergic individuals tolerated continuous treatments with the selected ME-PPD containing hair dye product. The number of full hair colour treatments within the one year observation period per individual related to the shade used (corresponding to the ME-PPD concentrations) is depicted in Fig. 4(a), indicating that up to 15 treatments per individual with the dark to medium-dark shade were well tolerated. The average number of full hair colour treatments per individual was almost 9. For 7 individuals detailed results for PPD and/or PTD on their initial diagnostic patch test were available: 5 were +++ graded to PPD, some also with positive results for PTD, see Table 4. For 2 individuals positive reactions to PTD only were documented, graded ++ and +, respectively.

The total number of full hair colour treatments per ME-PPD concentration level is provided in Fig. 4(b) indicating that dark to medium-dark shades were selected for 134 treatments representing the majority, i.e. 53%, while medium-light and light shades were used to a lesser degree (26% and 20%, respectively).

Upon the first full hair color treatment, 9 individuals developed allergic contact dermatitis symptoms after first hair dyeing even though their pre-test reaction was negative (see Table 5). The symptoms occurred mostly between 8 h and 2 days. In 2 cases symptoms were recognized either shortly after rinsing or as late as one week after the treatment, respectively. For 6 of these 9 individuals details of their initial diagnostic patch test strength to PPD were available: 4 with +++ and 2 with ++, the latter also reacted ++ to PTD.

Detailed information about the symptoms was gathered by a telephone interview with each individual. Redness, papules and itching generally occurred on scalp and adjacent skin areas between 8 and 24 hours after the hair color treatment. In addition 4 individuals mentioned swelling, therefrom 1 individual reported in addition development of vesicles. In 2 cases a dermatologist and in 1 case a general practitioner was consulted. They prescribed local treatment. In 6 cases no physician was consulted. In 5 of these 6 cases self-treatment had been applied: 3 used topical agents, while 1 had access to oral antihistamines and 1 treated herself with oral steroids. In one case antihistamines had been used for other health reasons prior to the hair dye treatment.

The severity of the allergic reaction was evaluated by the clinical symptoms and the intensity of the applied therapy: local therapy (mild reaction); oral therapy (self-treatment)/no physician (moderate reaction); oral therapy and physician (marked reaction); intravenous therapy and/or hospitalization (severe reaction). Based on this evaluation the degree of the contact dermatitis manifestation was considered to be mild to moderate for all 9 cases.

A comparison of all responding and non-responding individuals regarding the ME-PPD shades used combining pre-testing and full hair color treatment results is provided in Fig 5. Positive reactions occurred in 46% (12/26) of the individuals exposed to dark to medium dark shades, in 20% (2/10) exposed to medium light shades and in none of the individuals (0/7) exposed to light shades.

Discussion

With the introduction of ME-PPD into commercial hair dye products, a PPD/PTD alternative has become available that is considered to avoid de-novo generation of hair dye contact allergy for hair dye users.³ However, despite warnings to not use ME-PPD hair colorants in case of hair dye allergy, individuals with existing allergy to PPD and/or PTD are likely considering the use of ME-PPD containing hair dye products as an alternative. Consequently, in our present study we extended our

previously reported analysis of the cross-elicitation response to ME-PPD in PPD allergic individuals¹²: We asked whether PPD/PTD allergic individuals tolerate full hair colour treatments with their preferred ME-PPD containing shade upon negative pre-testing on their forearm. Through requests reaching our customer service, we recruited 43 PPD and/or PTD allergic individuals with a documented history of hair dye-related allergic contact dermatitis to participate in this study.

Continuous hair color exposures with ME-PPD containing hair dye products with concentrations up to 1% $\pm$ 0.2 were tolerated by 67% (29/43) of the individuals in intervals as short as 4 weeks over the observation time of the one year study period (Fig. 4). The limited cross elicitation rate to the full hair color treatments of 33% (14/43) is comparable to the cross-elicitation rate of 30% to a 30 min forearm exposure simulating hair dyeing with the maximized ME-PPD concentration of 2%.¹²

Among the 38 individuals with negative pre-testing results, additional 9 individuals showed skin reactions with symptoms ranging from mild to moderate following the first full hair colour treatment with their selected ME-PPD containing hair dye product (see Table 5). The reasons for this difference are not clear but one likely explanation may be that the pre-testing challenge on the forearm was not sufficient to trigger an elicitation response. When subsequently the scalp and hairline skin were exposed upon the first hair coloring it appears likely that the repeated availability of the antigen was sufficient to overcome the (cross-) elicitation threshold for memory T cells leading to an allergic contact dermatitis^{16,17} in these individuals.

Furthermore, the individual sensitivity as well as the exposure concentration affects the time until an allergic individual (cross-)reacts as was shown for isoeugenol.¹⁸ Thus, differences regarding the re-activation threshold may also have impacted the individual onset of the (cross-)elicitation responses observed in two individuals with symptoms occurring within 15 minutes or as late as 1 week after the hair color treatment (see Table 5). This is in line with the recently described variability of time intervals between hair dyeing and dermatitis onset (one day up to one month).¹⁹

Ho *et al.*²⁰ analyzed tolerance of PPD containing hair dye products in PPD allergic individuals based on the strength of their previous response to PPD under diagnostic patch testing: Tolerance was recorded in 73 % of 55 cases with a mild strength (+) and 49% of 61 patients with a moderate strength (++) but in none of 14 with a severe strength (+++). In contrast, we found tolerance to ME-PPD containing hair colorants in five cases with a strong diagnostic patch test reaction to PPD (+++), for three of them with a frequency of full hair color treatments of every 4 to 7 weeks for dark to medium-dark and medium-light shades, respectively (see Table 4). These findings indicate that the addition of the methoxymethyl side chain to PPD yielding ME-PPD can strongly reduce or completely prevent T-cell recognition even in strongly PPD-sensitized individuals.

Apart from limited T cell recognition, low exposure (e.g. through short contact time and professional coloring techniques reported by Chan *et al.*¹⁰) may have been relevant for the clinical tolerance observed (see Fig. 4). This is further supported by our finding that the percentage of positive elicitation reactions increased with increasing ME-PPD concentrations of the shades used (see Fig. 5). For the individuals without specified reaction strength, low PPD contact sensitivity may be additionally considered a reason for the clinical tolerance observed.

So far, our results indicate that hair dyeing with ME-PPD containing hair color is associated with a certain risk for PPD/PTD- allergic hair dye consumers, with a (cross)-elicitation rate of approximately 30%. Based on the experience with the small number of 43 allergic individuals, pre-testing with the hair color product of choice can be considered helpful to provide an alert of a strong (cross-) reactivity, but is insufficient to provide an indication of the absence of mild to moderate symptoms.

A commercially available pre-testing kit containing solely PPD has been reported to fail in a PTD/PPD allergic individual with a past D2 diagnostic patch test strength of +++ on PTD and ++ on PPD.^{21,22} Although PPD is considered an acceptable screening agent,⁹ other hair color precursors^{23,24} and formulation ingredients²⁵ were described as further responsible allergens in allergic contact dermatitis to hair dye products. Therefore we consider pre-testing with the hair color product selected by the consumer as advantageous, because it contains all possible allergens.

In summary, we found that pre-testing with the ME-PPD containing hair color product of choice was predictive for 77% of PPD/PTD-allergic individuals (33/43) and 67% (29/43) tolerated continuous hair coloring during the observation period of approximately one year. This indicates that due to its structural difference the recognition of ME-PPD by the immune system of a considerable number of allergic individuals is reduced or is possibly completely lacking in some cases.¹² This is in line with the primary prevention benefit of ME-PPD in terms of significantly reduced skin sensitization potency for all hair color consumers.³

In terms of secondary prevention the recommendation for hair dye allergic individuals to avoid hair coloring including PPD/PTD alternatives such as ME-PPD is a relevant measure because of a (cross)-elicitation rate of approximately 30%. However, avoidance of hair dyeing by allergic individuals is known to be a challenge, since patients have been described who don't want to avoid hair dyeing disregarding their dermatologists' advice and continue conventional hair dyeing ignoring their symptoms.^{10,20,26} Our study for the first time correlates two critical steps for hair dye allergic individuals: forearm (pre)-testing with the selected hair dye shade at the individual's dermatologist and in case of a negative result subsequent professional hair colour treatment(s) (see Fig. 3). We consider this approach as providing relevant guidance for the allergic individuals helping to avoid the

underestimation of risks associated with hair dyeing and that consultation with the dermatologist is of major importance for them prior considering the use of hair colours.

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Figure Legends

Fig 1. Chemical structures of the hair dye primary intermediates

Fig 2. Instructions for pre-testing under hair dye use condition (Allergy Alert Test)

Fig 3. Study overview

Fig 4. Number of tolerated full hair colour treatments related to ME-PPD concentration within one year observation period (a) per individual and (b) total number.

Fig 5. Total number of negative and positive elicitation reactions per tonal shade level from pre-testing and full hair color treatments

Tables

Table 1 Grouping of the ME-PPD containing hair dye shades in different tonal shade levels with corresponding concentration ranges of ME-PPD

Shade tone levels	Shade name	Average applied ME-PPD concentration
Dark / Medium dark	Black	1.0% $\pm$ 0.2
	Dark Brown	
	Medium Brown	
	Light Brown	
	Light Brunette Blond	
Medium light	Dark Blond	0.5% $\pm$ 0.1
	Medium Blond	
	Medium Brunette Blond	
Light	Light Blond	0.15% $\pm$ 0.1
	Very Light Blond	

Table 2 Scoring scheme: Assessment guidance of epicutaneous test reactions based on reaction criteria according to the German Contact Dermatitis Research Group (DKG) with reference to the International Contact Dermatitis Research Group (ICDRG) recommendations. "Irritation (Ir)" and "not tested (nt)" were removed from the table as these criteria were not considered to be relevant.

Grade	Description of response	Meaning
-	No reaction	Negative
?	Erythema only, no infiltration	doubtful reaction
+	Erythema, infiltration, possibly discrete papules	positive, allergic reaction
++	Erythema, infiltration, papules, vesicles	double positive, allergic reaction
+++	Erythema, infiltration, confluent vesicles	triple -positive allergic reaction

Table 3 Overview of elicitation reactions following 45 minutes pre-testing on forearm to ME-PPD containing hair dye shade of choice compared to initial diagnostic PPD/PTD patch testing results.

Indivi-dual ID	Initial diagnostic PPD/PTD patch test result **	Reaction pre-test *	
		45min	48h
5	nsp/nd	-	++
24	+++ /nsp	-	+++
26	+++ /+++	-	+
67	+++ /(+)	-	_***
71	+++ /+++	-	+++

* dark to medium dark tone level with ME-PPD range of 1% $\pm$ 0.2 was chosen in all cases;

** initial PPD/PTD diagnostic patch test results (if more than one diagnostic patch test result was available, highest score is presented);

*** no reaction was diagnosed at 48 hours, reaction was reported by individual at day 5

nsp: no further specification; nd: not documented

Table 4: PPD/PTD allergic individuals tolerating full hair colour treatments with ME-PPD containing hair dye with detailed diagnostic patch test results available

Individual ID	Initial diagnostic PPD/PTD patch test result	Average applied ME-PPD concentration [%]	Number of colourations	Interval of colourations [weeks]
52	+++ / ++	1	11	4-5
54	+++ / ++	1	9	6-7
13	+++ / +++	0.5	9	6-7
12	+++ / na	0.15	4	10-11
23	+++ / na	0.15	3	16-17

Table 5: Overview of elicitation reactions after first professional full hair colour treatment in individuals with negative pre-testing compared to initial diagnostic PPD/PTD patch testing results (nsp: no further specification; nd: not documented).

Individual ID	Initial diagnostic patch test result PPD/PTD	ME PPD hair dye shade	Severity of symptoms	Time point of 1 st symptoms after rinsing
18	++ / ++	Medium light	mild	8 hours
21	+++ / nsp	Dark / Medium dark	mild	2 days
25	+++ / nsp	Dark / Medium dark	moderate	1 week
28	++ / ++	Dark / Medium dark	mild	1 day
50	nsp / nsp	Dark / Medium dark	moderate	8 hours
51	+++ / nd	Dark / Medium dark	mild	1 day
63	nsp / nd	Medium light	mild	within 15 minutes
72	+++ / nsp	Dark / Medium dark	mild	1 day
74	nsp / nd	Dark / Medium dark	mild	8 hours

Fig 1. Chemical structures of the hair dye primary intermediates

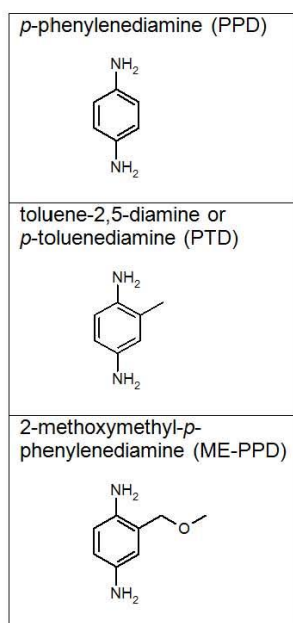


Fig 3. Study overview

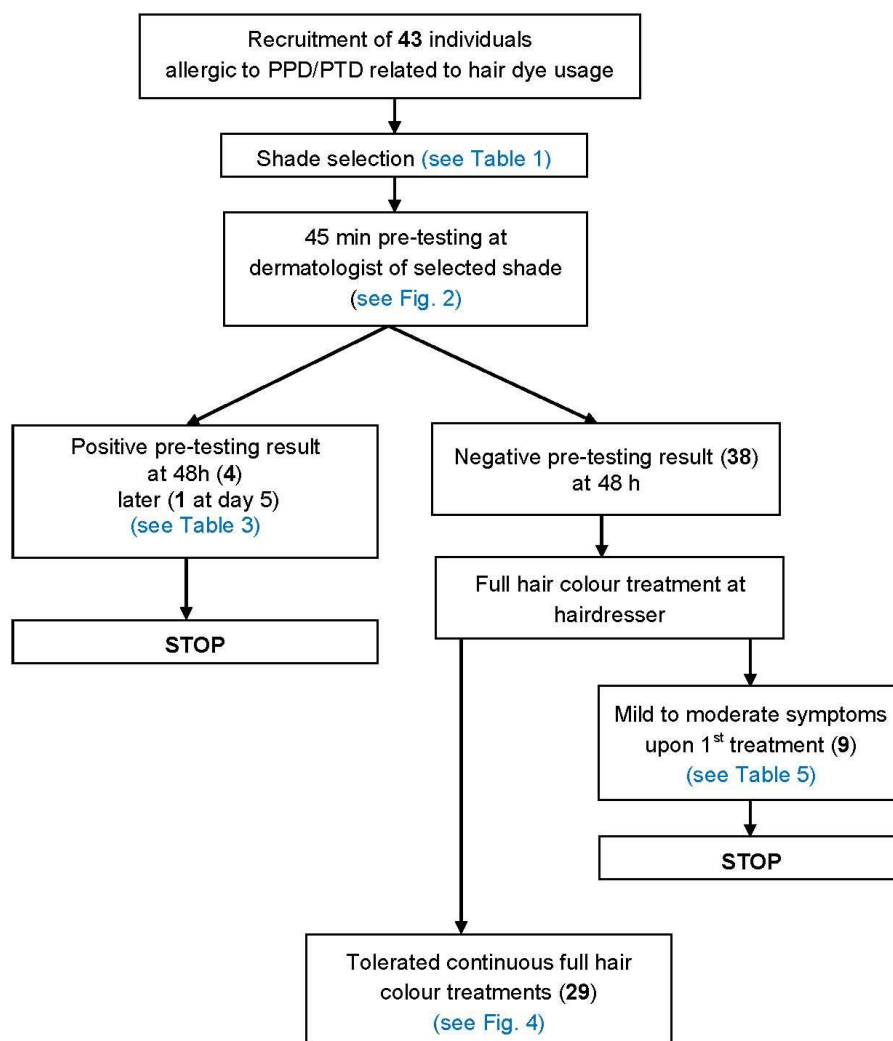


Fig 4. Number of tolerated full hair colour treatments related to ME-PPD concentration within one year observation period (a) per individual and (b) total number

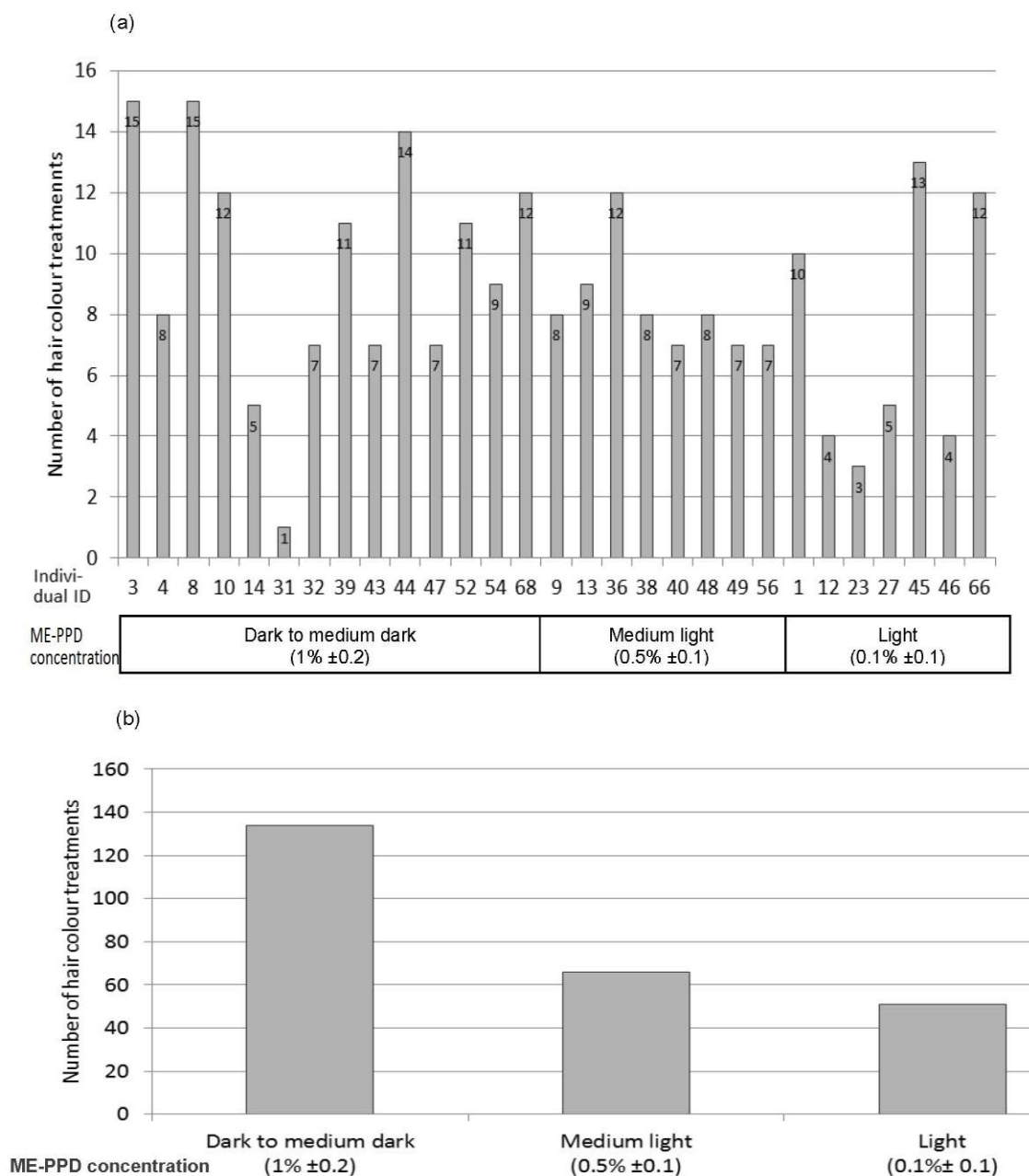


Fig 5. Total number of negative and positive elicitation reactions per tonal shade level from pre-testing and full hair color treatments

