

Hair dye contact allergy: quantitative exposure assessment of selected products and clinical cases

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Colouring of hair can cause severe allergic contact dermatitis. The most frequently reported hair dye allergens are *p*-phenylenediamine (PPD) and toluene-2,5-diamine, which are included in, respectively, the patch test standard series and the hairdressers series. The aim of the present study was to identify dye precursors and couplers in hair dyeing products causing clinical hair dye dermatitis and to compare the data with the contents of these compounds in a randomly selected set of similar products. The patient material comprised 9 cases of characteristic clinical allergic hair dye reaction, where exposure history and patch testing had identified a specific hair dye product as the cause of the reaction. The 9 products used by the patients were subjected to chemical analysis. 8 hair dye products contained toluene-2,5-diamine (0.18 to 0.98%). PPD (0.27%) was found in 1 product, and *m*-aminophenol (0.015 to 0.38%) and *p*-aminophenol (0.16 to 2.1%) were found in 3 products. The concentration levels were similar in the patient's products compared to a random sample of 16 hair dye products. The concentration present of toluene-2,5-diamine elicited allergic reactions in concentrations that were 10-fold lower than the legal EU limit of 10%. Hair dye allergy may cause severe clinical reactions, and the current regulation is insufficient in protection of the users. A preventive strategy is needed.

Key words: allergic contact dermatitis; hair dye dermatitis; hair dyes; *m*-aminophenol; *p*-aminophenol; patch test; PPD; *p*-phenylenediamine; quantitative exposure; toluene-2,5-diamine. © Blackwell Munksgaard, 2004.

Accepted for publication 20 April 2004

Colouring of hair is a common fashion trend, and more than 50% of the women in the industrial world use permanent or temporary hair dyes for shorter or longer periods of their life. Even among the male population, colouring of hair is common. Geographical variations in the use are probably wide within Europe.

p-Phenylenediamine (PPD) is, based on a large material of patch tested patients, generally reported as the main sensitizer in hair dye dermatitis (1–7) and is known to be an extremely potent contact sensitizer (8). PPD is allowed at a concentration up to 6% in hair dyes (9). Toluene-2,5-diamine is allowed for use in hair dyes at a concentration up to 10% (9), and it is a well-known sensitizer (1, 3–7). *p*-Aminophenol is a known sensitizer (1, 3–5), but it has not yet been regulated. *m*-Aminophenol is also a sensitizer (5) and is allowed for use in hair dyes at a concentration up to 2% (9). Oxidative hair dye formulations may also contain azo dyes, which might cause

contact allergy either as an individual sensitizing molecule or by crossreactivity in people sensitized to PPD (2–4). Allergic hair dye reactions can be severe, with scalp and facial oedema, spreading to the ears, neck and shoulders (10).

The aim of the present study was to identify and quantify PPD and its derivatives in hair dye products causing clinically characteristic hair dye dermatitis and to compare the results with the content of these substances in a random sample set of similar products bought on the Danish retail market.

Patients

The case group consisted of 9 patients with acute hair dye dermatitis, who were in possession of the product used. They were seen at the Department of Dermatology, Odense University Hospital, and Department of Dermatology, Copenhagen University, over 14 months in 2001–02 (10). They

all had a characteristic clinical hair dye dermatitis with a scalp, facial and neck dermatitis developing within 1–2 days (eventually hours) after hair colouring. The exposure history and patch testing identified a particular product as the cause of reaction. The actually used product was subjected to chemical analysis.

Randomly selected hair dyeing products

The Danish Environmental Protection Agency collected 22 hair dyeing products for non-professional use from Danish retail outlets for compliance testing, in May 2002. Of these, 16 products were hair colouring formulations with 2–3 component systems, which contained precursors and couplers of oxidative hair dyes. The data from the analysis of these 16 products were used for comparison (11).

Chemical analysis

The content of the precursors and couplers in the oxidative hair dye (PPD, toluene-2,5-diamine, *p*-aminophenol, *m*-aminophenol and *o*-aminophenol) formulations were determined using high-performance liquid chromatography (HPLC) with diode array detection, as described earlier (12). In brief, approximately 1 g of formulation containing precursors and coupler, weighed accurately in a dark bottle, was dissolved in 15 ml of acetonitrile and 10 ml of 0.5% sodium ascorbate solution (in 25 mM phosphate buffer). The solution/suspension was filtered through a 0.45- μ m membrane filter and analysed within 24 h. The filtrate was analysed employing HPLC with gradient elution, as described before (12). The target precursors and couplers in a sample were identified by matching the retention time and 220–400 nm UV spectrum of HPLC peaks with those of standard compounds.

Freshly prepared calibration solutions of the target hair dye precursors were analysed together with the sample extracts for the determination of their contents in the hair dye formulation.

Results

A total of 9 different products, each relating to 1 individual patient, were identified (Table 1). The products represented 4 international cosmetic manufacturers. The clinical cases were characteristic and, in most cases, severe. All identified oxidative hair dye precursors and couplers were labelled on the products, except for product no. 6, where PPD was present at a concentration of 0.27%. This product was labelled to contain hydroxyethyl-PPD, which could not be analysed in the present study due to unavailability of the standard chemical.

Toluene-2,5-diamine (0.18 to 0.98%) was present in 8 products, *m*-aminophenol (0.015 to 0.38%) was present in 3 products and *p*-aminophenol (0.16 to 2.1%) was also present in 3 products (Table 1). Comparison of outcome of the chemical analysis from the products used by the patients and data from the analysis of randomly collected samples are summarized in Table 2. Based on this relatively limited material, the 2 data sets give similar concentrations. The chemical analysis was, in all but 1 case, in accordance with the product labelling.

Discussion

This study is the first to analyse chemically the actual content of PPD and selected derivatives in hair dye products causing acute allergic hair dye dermatitis. The outcome of patch testing and product analysis are in accordance with the generally accepted opinion that oxidative hair dye ingredients, PPD and its derivatives toluene-2,5-diamine, *m*-aminophenol and *p*-aminophenol, are common causes of hair dye dermatitis. Chemical analysis of the randomly collected products identified concentrations of the oxidative hair dyes similar to the concentrations in the products used by the patients with hair dye dermatitis. This finding shows that patients can develop hair dye dermatitis from concentrations that are lower than that permitted in the EU Cosmetic directive (9). A product-related factor, e.g. multiple allergens in the same product, may also be of importance. This possibility is exemplified by case nos 4 and 9, both of these reacting to oxidative hair dyes and to Disperse Orange 3 and aminoazobenzene. The explanation for simultaneous patch test reactivity can be either multiple sensitizations or crossreactivity. It is of major scientific interest to develop methods which can differentiate between the 2 conditions. Notwithstanding the mechanistic explanation for multiple reactions, products containing a diversity of chemical allergens may give rise to immunological synergistic clinical effects (13), which may explain the severity of the clinical reactions.

There may exist an individual genetic polymorphism relating to either enzymatic detoxification or hapten activation in the epidermis. A number of such polymorphisms have already been identified to have clinical importance (14–16). It is still unknown whether the reason why for some people do not get an allergic reaction to hair dye is genetic or due to low exposure.

Interestingly, the concentrations of the individual oxidative hair dyes (Tables 1 and 2) are below the limits permitted by the EU Cosmetic directive (9), e.g. toluene-2,5-diamine elicited

Table 1. Hair dye dermatitis cases and exposure assessment based on chemical analysis

Product no. (%)	Patient characteristics	Symptoms	Patch test results*	Results of the chemical analysis of hair dye (%)
Oxidative hair dye (black)	Woman aged 32 years; no previous skin disease; dyed hair for many years Used the product ×3 before. Severe reaction occurring within 24 h after dyeing hair last time	Acute eczema on scalp, severe facial oedema. Sick leave and medical treatment. Symptoms subside over a 14-day period	Standard and hairdressers series without toluene-2,5-diamine. Pos. PPD. Neg. aminophenol	Toluene-2,5-diamine: 0.97 <i>m</i> -Aminophenol: 0.057
Oxidative hair dye (dark blond)	Woman aged 58 years; no previous skin disease; dyed hair for many years Used this product only once. Severe reaction came 2 days later	Acute eczema on scalp, severe facial oedema. No sick leave Medical treatment. Symptoms subside in 1 week	Standard and hairdressers series Pos. PPD and toluene-2,5-diamine Doubtful reaction to nitro-PPD and aminoazobenzene	<i>m</i> -Aminophenol: 0.015 <i>p</i> -Aminophenol: 0.16 Toluene-2,5-diamine: 0.15
Oxidative hair dye (black)	Man aged 21 years; dyed hair ×2, 3rd time; he got symptoms 3 days after the hair colouring	Acute eczema on scalp, periobital oedema Medical treatment. Symptoms subside in 14 days	Standard and hairdressers series Pos. toluene-2,5-diamine and PPD	Toluene-2,5-diamine: 0.63
Oxidative hair dye (dark blonde)	Woman aged 32 years; previously perfume rash; not previously dyed hair Hair dyed at hairdresser. Symptoms days later	Eczema on scalp, facial oedema. Sick leave	Standard and hairdressers series. Pos. PPD, toluene-2,5-diamine, <i>m</i> -aminophenol, Disperse Orange 3 and aminoazobenzene. Pos. perfumes, <i>p</i> -aminophenol, hydroquinone, pyrogallol	Toluene-2,5-diamine: 0.37 <i>m</i> -Aminophenol: 0.067
Oxidative hair dye (light golden brown)	Women aged 20 years; temporary tattoo reaction previously Dyed hair many times before. Symptoms hours after last hair dying	Acute eczema scalp, facial oedema Duty doctor and casualty department	Standard series and treatment series PPD pos. +++ toluene-2,5-diamine not tested	Toluene-2,5-diamine: 0.18 <i>p</i> -Aminophenol: 0.72
Oxidative hair dye (golden blonde)	Women aged 39 years; no previous skin disease dyed hair once a year Now hair dyed at hairdressers. Reaction 3 days later	Eczema at scalp and facial oedema. Asthma. Symptoms persisted for 7–9 days	Standard and hairdressers. Pos. PPD and toluene-2,5-diamine, 4-aminoazobenzene	PPD: 0.27
Oxidative hair dye (black)	Women aged 8 years temporary tattoo with reaction 4 years previously Dyed hair ×10–15 before. First time, used this product. Symptoms within 12 h	Eczema scalp, facial oedema. Eczema spreading to other body sites Casualty. Medical treatment. Sick leave 1 week Symptoms subsided in 14 days	Pos. perfume standard and hairdressers. Pos. PPD +++, toluene-2,5-diamine +++, <i>m</i> -aminophenol +++, <i>p</i> -aminophenol +++, nitro-PPD ++, 4-aminoazobenzene ++	Toluene-2,5-diamine: 0.72 <i>m</i> -Aminophenol: 0.38
Oxidative hair dye (blue black)	Woman aged 53 years; used same hair dye for 1 years. Symptoms shortly after last time	Eczema on scalp, spreading to other body sites. Medical treatment. Sick leave. Emergency department. Symptoms last 6 months	Standard and hairdressers. Pos. PPD ++	Toluene-2,5-diamine: 0.98
Oxidative hair dye (chestnut brown)	Woman aged 56 years. Previously healthy. Used hair dyes for 20 years, ×2–4 a year (at home). Symptoms developed the day after the last hair dye	Eczema scalp, facial oedema. Medical treatment No sick leave. Subsided in 2 weeks	Standard and hairdressers. Pos. PPD, toluene-2,5-diamine, <i>m</i> -aminophenol, <i>p</i> -aminophenol, aminoazobenzene	Toluene-2,5-diamine: 0.61 <i>p</i> -Aminophenol: 2.1

PPD, *p*-phenylenediamine.*Pos. = Positive patch test reaction of at least +. Neg. = Negative patch test result. *m*-Aminophenol is the INCI name for 3-aminophenol, the INCI name being the name that the producer has to write on the product label. *p*-Aminophenol is the INCI name for 4-aminophenol.

Table 2. Comparison of the outcome of chemical analysis of the products used by the patients and randomly collected products

Oxidative hair dye precursor	Concentration % (w/w)	
	Patients' products, <i>n</i> = 9	Randomly collected samples
Toluene-2,5-diamine	0.18–0.98	0.01–3.99
<i>m</i> -Aminophenol	0.015–0.38	0.01–0.94
<i>p</i> -Aminophenol	0.16–2.1	0.31–1.20
<i>p</i> -Phenylenediamine	0.27	–

allergic reactions at concentrations that are 10-fold lower than the legal limit of 10% (9). Special products selected for blonde Scandinavian customers may explain the low use concentration or may be a more general trend.

Exposure to hair dye substances is in epidemiological studies associated with the development of bladder cancer (17, 18) and non-Hodgkin's lymphoma (19). After Gago-Dominguez et al. (17) showed an association between long-term use of oxidative hair dye and bladder cancer in consumers, the European Commission established a task force to review the toxicity of hair dyes. Exposure data as provided in this article are also crucial for the evaluation of other toxicological effects such as cancer. For postmarketing risk assessments of allergy, it is preferable to base the evaluation on clinical data, including threshold studies in sensitized persons in relation to an exposure analysis. In a literature study on 229 hair dye substances, clinical data were only available for 19 substances (20). A suitable quantitative structure–activity relationship model is needed as a screening tool for prospective recognition of potential allergens in hair dyes (20).

In conclusion, hair dye allergy may cause severe clinical reactions and the current regulation is insufficient in protection of the users. A preventive strategy is needed.

Perspectives

Hair dye dermatitis is an important health concern. Studies based only on patch test outcome are insufficient to establish the knowledge needed for an effective preventive strategy. An efficient strategy should include prohibition or further concentration regulation of hair dye substances, product development and identification of particularly sensitive individuals having multiple genetic polymorphisms leading to insufficient and somatic detoxification or allergy formation in the epidermis. The present study, together with the accumulated literature, calls for studies which should include the following data:

- (1) typical cases of hair dye dermatitis;
- (2) patch testing and exposure test with actually used product and product ingredients;
- (3) dose–response studies on allergic persons for estimation of a non-response level of single ingredients for contact allergy;
- (4) chemical analysis of actually used products;
- (5) analysis for known genetic polymorphisms;
- (6) *in vivo* human skin metabolism studies using microdialyses in PPD-sensitive persons and controls;
- (7) use of gene array technology for search of unknown genetic markers for the risk of developing hair dye dermatitis; and
- (8) final risk assessment – based on product-related and individual-related risk factors.

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