Azo pigments and quinacridones induce delayed hypersensitivity in red tattoos

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Summary

Background. Induction of delayed hypersensitivity reactions by red tattoos has been occasionally reported. Little is known about the inks used. Azo pigments have been implicated in some instances, but there is only one reported case involving quinacridones.

Objectives. To describe the clinical and pathological features and outcome of skin reactions induced by red tattoo pigments.

Patients, materials, and methods. Six patients with a cutaneous reaction induced by a red tattoo pigment underwent biopsy and prick and patch testing with the inks supplied.

Results. We observed seven reactions in the 6 patients. Histology showed various patterns: three lichenoid, two eczematous, and two pseudolymphomatous. Five reactions occurred with azo pigments, and two with quinacridones, in both cases with Violet 19 and Red 122. Four inks were tested. Only one patch test gave a positive result at a late reading (day 7). Prick tests gave negative results. The reactions required various treatments, including laser treatment for 2 patients. Activation of the reaction in 1 case was transient.

Conclusion. Azo pigments and quinacridones both triggered reactions with similar clinical aspects but with varying histological findings. Patch and prick test results were disappointing with both. Reactions occurred following laser use in 1 case.

Key words: azo dye; hypersensitivity; patch testing; quinacridone; red tattoo.
We acquired data sheets with detailed compositions of the inks from the manufacturers, and asked the tattooists to supply samples of the inks used for cutaneous testing.

**Methods**

Epicutaneous tests were performed with the inks supplied without dilution.

Patch tests with the European baseline series, metal series and a textile dyes series (Chemotechnique Diagnostics, Vellinge, Sweden) were also performed with Finn Chambers® on Scanpor® tape (SmartPractice, Phoenix, AZ, USA) with a 2-day occlusion. Skin reactions were assessed after 2, 4 and 7 days, according to International Contact Dermatitis Research Group criteria.

Prick testing on the inner sides of the patients’ left forearms was also performed with undiluted inks, and skin reactions were assessed after 20 min, 2 days, and 4 days.

Patients were followed up and treated with topical corticosteroids (clobetasol propionate cream under occlusion) or intralesional corticosteroids (triamcinolone acetonide). When the reaction was persistent, treatment with a Q-switched 532-nm Nd:YAG laser was proposed. Patient consent was obtained after a full explanation of possible complications.

**Results**

**Clinical presentation**

The 6 patients developed seven skin reactions, from 2 weeks to 3 years after having a red tattoo. One patient (patient 5) had a reaction in two different tattoos, one of which had been performed 2 months previously and the other 4 months previously, with two different red inks.

All patients had similar presentations, with pruritus, swelling and discomfort in the red portions of their polychromatic tattoos (Fig. 1a, b). The reaction was papular with some crusting.

**Biopsy results**

For all 6 patients, biopsy specimens of the swollen red areas were examined. In 3 patients (1–3), biopsy showed a lichenoid reaction, with a band-like dense lymphocytic infiltrate in the upper dermis and necrotic basal keratinocytes. In 2 patients (4, 5), it showed a pseudolymphomatous reaction: a dermal infiltrate of small to medium-sized CD4+ T cells expressing T follicular helper markers (CD10, CXCL13, Bcl6, and PD1), indicative of a T follicular helper pseudolymphomatous reaction, in patient 3 (Fig. 2), and a T cell pseudolymphomatous reaction with a dense nodular lymphoid infiltrate of small to medium-sized lymphocytes in the reticular dermis, with intermingled pigment deposits, in patient 4. In the latter patient, the lymphoid infiltrate was mainly composed of mature CD3+, CD4+ and CD5+ T lymphocytes. Bcl6 was strongly expressed. B lymphocyte markers were negative. Patient 5 had an eczematous reaction to both tattoos.

**Involved inks**

From the information provided by the tattooists, we were able to obtain the names of the manufacturers and the names and compositions of the inks involved in the seven skin reactions. In five lesions, the colorants used were azo pigments, and in the other two they were quinacridones (Table 1). The trade names of the azo pigments used were as follows: BRIGHT RED (tattoo reaction 1), a mixture of three azo pigments, Red 210 (CI 12477), Yellow 65 (CI 11740), and Orange 13 (CI 21110); RASPBERRY (tattoo reaction 2), Red 63:1 (CI 15880); LIGHT RED, DEEP RED, and DARK RED (tattoo reaction 3), all three corresponding to Red 170 (CI 12475); FLAME RED (tattoo reaction 4), Red 112 (CI 12370); and LIGHT RED (tattoo reaction 5), Red 170 (CI 12475) (Table 2). The trade names of the quinacridone pigments used were as follows: TRUE MAGENTA (tattoo reaction 6), a mixture of two quinacridones, Violet 19 (CI 73 900) and Red 122 (CI 73 915); and DEEP PURPLE AND DEEP MAGENTA (tattoo reaction 7), a mixture of the same two quinacridones, Violet 19 and Red 122 (Table 2). These inks, in addition to organic pigments, contained water, glycerol, and isopropyl alcohol, and some of them contained titanium dioxide, acrylic resin, food-grade gum, phospholipids.
food grade emulsifier, dimethylcarbinol, and witch hazel, according to the Material Safety Data Sheets. There was no mention of preservatives in the composition of the inks supplied by the manufacturers.

**Patch and prick tests**

An ink sample was available for only four reactions from 3 patients, and was patch and prick tested.

Of the 5 patients with reactions to azo pigments, two were patch and prick tested: patch tests with the inks gave negative results, except for patient 1, in whom a reaction was seen on day 7. Patient 5 (tattoo reaction 5) showed ++ reactions to nickel sulfate, potassium dichromate, and cobalt chloride, and a + reaction to fragrance mix 1. Patch tests with metal series including mercury and textile dyes series gave negative results. Prick tests gave negative results (Table 2).

Both patients with reactions to quinacridones were tested: patch and prick tests with the undiluted inks used gave negative results. The results of patch tests with the European baseline series, metal series and textile dyes series for patient 5 (tattoo reaction 6) are mentioned above, and patient 6 showed a + reaction to nickel sulfate (Table 2).

**Treatment**

All of the patients were treated with potent topical corticosteroids (clobetasol propionate 0.05% cream), applied daily under occlusion. Patients 1 and 6 received intralesional triamcinolone. These treatments had only transient efficacy. Subsequently, and because the reaction was very disfiguring, patients 1 and 3 required and accepted Q-switched 532-nm Nd:YAG laser courses. Patient 1 had six courses at 2-weekly intervals in a wide red tattoo. He experienced a severe reaction of the entire lower limb with an initial oedematous erythema, followed by superficial erosion and crust formation that took 2 weeks to heal (Fig. 3a). Curiously, he also suffered a new pruritic and oedematous attack during laser treatment in two distant red tattoos that had been performed several years before but that, initially, were free of any clinical reaction (Fig. 4a, b). These secondary reactions were persistent after the use of topical corticosteroids. Laser treatment was then performed, with no activation of the allergic reaction and no relapse after 2 years of follow-up. Pigmentation disorders, however, remained (Fig. 3b). Patient 3 required only two laser sessions.

**Discussion**

Polychromatic tattoos are now popular worldwide. However, the frequency of delayed reactions to red tattoos is unknown. Information in the literature is scarce, and only a few individual cases and four small series have been documented (1, 2, 4, 5).

The time to the development of a tattoo ink reaction seems to be extremely variable from one patient to
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Table 1. Pigments used in tattooed patients

<table>
<thead>
<tr>
<th>Pigment</th>
<th>Formula</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pigment Red 210 (CI 12 477)*</td>
<td>Reaction mass of 4-[(4-(aminocarbonyl)phenyl)azo]-3-hydroxy-N-(2-methoxyphenyl)napththalene-2-carboxamide and 4-[(4-(aminocarbonyl)phenyl)azo]-N-(2-ethoxyphenyl)-3-hydroxynaphthalene-2-carboxamide</td>
<td></td>
</tr>
<tr>
<td>Pigment Yellow 65 (CI 11 740)</td>
<td>2-[(4-Methoxy-2-nitrophenyl)azo]-N-(2-methoxyphenyl)-3-oxo-butyramide</td>
<td></td>
</tr>
<tr>
<td>Pigment Orange 13 (CI 21 110)b</td>
<td>4,4′-[(3,3′-Dichloro[1,1′-biphenyl]-4,4′-diyl]bis(2,4-dihydro-5-methyl-2-phenyl-3H-pyrazol-3-one)</td>
<td></td>
</tr>
<tr>
<td>Pigment Red 63:1 (CI 15 880)</td>
<td>Calcium 3-hydroxy-4-[(1-sulphonato-2-naphthyl)azo]-2-naphthoate</td>
<td></td>
</tr>
<tr>
<td>Pigment Red 170 (CI 12 475)b</td>
<td>4-[(4-(Aminocarbonyl)phenyl)azo]-N-(2-ethoxyphenyl)-3-hydroxynaphthalene-2-carboxamide</td>
<td></td>
</tr>
<tr>
<td>Pigment Red 112 (CI 12 370)b</td>
<td>3-Hydroxy-N-(2-methylphenyl)-4-[(2,4,5-trichlorophenyl)azo]napththalene-2-carboxamide</td>
<td></td>
</tr>
<tr>
<td>Pigment Violet 19 (CI 73 900)b</td>
<td>5,12-Dihydroquino(2,3-b)acridine-7,14-dione</td>
<td></td>
</tr>
<tr>
<td>Pigment Red 122 (CI 73 915)b</td>
<td>5,12-Dihydro-2,9-dimethylquino(2,3-b)acridine-7,14-dione</td>
<td></td>
</tr>
</tbody>
</table>

*From http://www.dyestuffintermediates.com
bFrom http://www.chemblink.com

another. In our series, reactions to azo pigments containing inks occurred from 15 days to 3 years after tattooing. In other reports, reaction times have ranged from a few weeks to a few months. In our patients, reactions to quinacridone pigments occurred from 2 to 4 months after tattooing. The reaction to quinacridone described by Greve occurred 4 weeks after tattooing (6). In addition, 5 cases of late reactions (after several years) have been reported with inorganic pigments and inks of unknown composition (3, 7–9). One of our patients had an early reaction (2 weeks). This suggests rapid sensitization by the tattoo procedure or previous sensitization to a related pigment. However, this multi-tattooed patient experienced no reaction or other dye contact dermatitis. The 5 other patients had more delayed reactions, from 2 months to 3 years. This time lag could be attributable to several different mechanisms, such as acquired hypersensitivity to the pigment or a tolerance breakdown. The allergens might have been formed by chemical changes in pigment molecules through haptenization in the skin weeks or months after tattooing.

The clinical presentation was similar in our patients, with a pruritic papular reaction and slight crusting restricted to the red areas of the polychromatic tattoos. However, as in other studies, histology patterns were slightly different (2, 3, 6, 7, 10–12). We observed two lichenoid reactions, two pseudolymphomatous reactions and one eczematous reaction with the azo dyes, and one eczematous and one lichenoid reaction with quinacridones. As reported elsewhere, the lichenoid pattern is commonly observed (2, 3, 7, 8, 13, 14). In contrast, there are few reports of pseudolymphomatous
### Table 2. Patient characteristics

<table>
<thead>
<tr>
<th>Patient</th>
<th>Tattoo reaction</th>
<th>Age (years)</th>
<th>Sex</th>
<th>Tattoo site</th>
<th>Interval between tattoo and reaction onset</th>
<th>Histology</th>
<th>Ink manufacturer</th>
<th>Trade name (distributor)</th>
<th>Patch tests</th>
<th>Prick tests</th>
<th>Trade name</th>
<th>Colour index</th>
<th>EBS</th>
<th>Metal series</th>
<th>Textile dye series</th>
<th>Undiluted ink</th>
<th>Undiluted inks</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>M</td>
<td>49</td>
<td>M</td>
<td>Leg</td>
<td>15 days</td>
<td>Lichenoid</td>
<td>Intenze (Rochelle Park, NJ, USA)</td>
<td>BRIGHT RED (Mario Barth Gold label)</td>
<td>Red 210</td>
<td>--</td>
<td>--</td>
<td>Cl 12:477</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>++</td>
<td>--</td>
</tr>
<tr>
<td>2</td>
<td>F</td>
<td>35</td>
<td>F</td>
<td>Ankle</td>
<td>2 months</td>
<td>Lichenoid</td>
<td>Starbrite 2 (Somers, CT, USA)</td>
<td>RASPBERRY</td>
<td>Yellow 65</td>
<td>--</td>
<td>--</td>
<td>Cl 11:740</td>
<td>--</td>
<td>Orange 13</td>
<td>ND</td>
<td>ND</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>M</td>
<td>30</td>
<td>F</td>
<td>Forearm</td>
<td>3 months</td>
<td>Pseudolymphomatous reaction</td>
<td>Eternal (Brighton, MI, USA)</td>
<td>LIGHT RED</td>
<td>Red 170</td>
<td>--</td>
<td>--</td>
<td>Cl 12:475</td>
<td>--</td>
<td>--</td>
<td>ND</td>
<td>ND</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>F</td>
<td>40</td>
<td>F</td>
<td>Chest</td>
<td>3 years</td>
<td>Pseudolymphomatous reaction</td>
<td>Azayaka (Riccione, Italy)</td>
<td>DEEP RED DARK RED FLAME RED</td>
<td>Red 112</td>
<td>Cl 12:370</td>
<td>--</td>
<td>--</td>
<td>ND</td>
<td>--</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>M</td>
<td>44</td>
<td>M</td>
<td>Back</td>
<td>2 months</td>
<td>Eczematous</td>
<td>Eternal (Brighton, MI, USA)</td>
<td>LIGHT RED</td>
<td>Red 170</td>
<td>Ni ++, Cr ++, Co ++, fragrance mix 1 +</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>Cl 12:475</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>6</td>
<td>F</td>
<td>28</td>
<td>F</td>
<td>Back</td>
<td>2 months</td>
<td>Lichenoid</td>
<td>Starbrite (Somers, CT, USA)</td>
<td>DEEP PURPLE</td>
<td>Red 122</td>
<td>--</td>
<td>--</td>
<td>Cl 73:915</td>
<td>--</td>
<td>DEEP MAGENTA</td>
<td>--</td>
<td>--</td>
<td></td>
</tr>
</tbody>
</table>

**EBS**: European baseline series; **F**: female; **M**: male; **ND**: not done; **−**: negative.
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In three of the four published series reporting delayed reactions to red tattoos, there were no details of the inks or colorants used, and in only one series were the inks tested (1). This illustrates the difficulty in obtaining ink details and samples. Of the 15 patients mentioned in the literature who had patch tests with the inks, only 5 had positive results (8, 10, 11, 18, 19). Most authors mention the low sensitivity of the epicutaneous tests, which is probably attributable to the poor penetration into the skin of the applied ink. Patch testing yielded disappointing results in our cases, as in other studies (1, 6, 12, 14–16, 20). However, 1 of our patients had a reaction at D7 after patch testing, which suggests that delayed test reading can be useful. One other author reported a late reaction, at D8 (8). A negative patch test result does not, however, rule out pigment as a cause. Recently, Serup et al. patch tested 90 patients with non-infectious chronic tattoo reactions, with batteries of baseline allergens, disperse dyes/textile allergens, and a selection of tattoo ink products. The tests gave negative results. The authors suggested that the responsible allergen is not present directly in the tattoo ink, but results from chemical changes in the pigment molecule, perhaps weeks or months after tattooing, which would explain why the patch tests gave negative results (21).

Prick tests were performed in only two studies, and only Wenzel et al. reported positive prick test results: skin reactions were evaluated after 4 days in 3 patients with colorants used to make cosmetic tattoos (1). In the 3 patients (four inks) we tested, prick tests gave negative results. Perhaps a later reading would be useful.

Intradermal testing should have been more suitable, as the hypersensitivity reaction was caused by intracutaneous rather than epicutaneous challenge. However, intradermal tests carry the risk of long-lasting skin reactions, and for this reason most authors do not recommend them (12, 14). Likewise, we preferred not to perform intradermal tests. Intradermal testing has been reported in only two studies: in one the test gave a positive result with undiluted ink (15), and in the other only aluminium was tested, and was positive (22).

Metal particles are sometimes reported to be responsible for red tattoo reactions, especially nickel sulfate and mercury sulfide (cinnabar) (10, 18). Although the colorants used in our patients were organic pigments that, according to the manufacturers, contained no metals, we cannot exclude the possibility that they were, in fact, contaminated. Mercuric sulfide is now being replaced by organic pigments, chiefly azo pigments.

Most of the patients in our series reacted to red inks containing azo pigments, in two instances Red 170, as in a previous report (14). Azo pigments belong to a wide class...
of synthetic organic pigments that have the nitrogen azo group N=N as part of their molecular structure. They give bright, high-intensity colours. More than half of all marketed dyes used in textiles, inks, paint and plastics are azo dyes. Because of structural similarities, cross-sensitization between azo pigments and para-amino compounds may occur (23). Hence, patients who experience an allergy to an azo pigment in a tattoo may also be at risk of reactions to everyday products and textiles. Bhardwaj et al. reported the case of a man who developed a reaction to a red tattoo that was successfully treated with a CO₂ laser. Six months later, he experienced a reaction after wearing a red T-shirt. Patch tests with the tattoo colorants and T-shirt dye showed strong positivity (++) in the previously affected area (19). To date, none of our patients has had other reactions, and patch tests with p-phenylenediamine and textile dyes series have all given negative results.

Like azo pigments, quinacridones are light-fast pigments that are used not only in tattoos but also in paints, printing inks, and plastics. They may be associated with azo pigments in red tattoo inks. Bendsoe et al. reported the first case, in 1991, of a patient with a reaction to an ink containing azo pigment and quinacridone (24). To our knowledge, there is only 1 published case of a reaction to quinacridone Red 122 (CI 73 915) pigment (6).

In the present series, we report 2 new cases of reactions to red tattoos with inks containing quinacridones, both involving a mixture of two pigments, Violet 19 (CI 73 900) and Red 122 (CI 73 915), that provide confirmation of the potential involvement of quinacridone pigments in hypersensitivity reactions to red tattoos.

We observed reactions in patient 5 in two different tattoos, with the two types of pigment. Cross-sensitization between azo pigments and quinacridones is unlikely, owing to the absence of chemical similarities, but the reactions may have been attributable to common metabolites produced in the skin, possibly by natural light exposure.

We were unable to identify the allergens involved in our patient reactions. As the skin reactions were restricted to the red areas of the multi-coloured tattoos, we assumed that the culprit allergens were the red pigment and, more specifically, its metabolites. Moreover, other ingredients listed by the manufacturers were the same in other colours. No preservatives were mentioned in the Material Safety Data Sheet enclosed with each ink. In spite of the rarity of positive patch test reactions in other reports, all authors agree in considering colorants to be the causal allergens (3, 6, 7–14). However, the responsibility of other compounds, such as impurities, metal particles, and decomposition products of the inks, cannot be excluded. There are no published data on the decomposition products of quinacridones. It has been shown that the concentration of azo pigments in the skin decreases with time: the pigments are, in part, transported away via the lymphatic system, and in part decomposed in the skin into products, including amino-naphthol-AS and naphthol-AS (25), that could be responsible for adverse reactions. In the literature to date, however, no tests have been performed with these decomposition products. We performed patch tests with naphthol-AS in 3 of our patients, with negative results. It has also been shown that exposure of an azoid, Pigment Red 22, to broadband ultraviolet (UV) B radiation and natural sunlight leads to the formation of the same decomposition products (26, 27). Some authors have described cases worsened by sun exposure (3, 14, 15). In a study of 154 patients with tattoo reactions, 58% were sun-induced, and lasted from hours to days (28). None of our patients noticed any change after sun exposure or complained of worsening, but photoproducts may have been involved in the reaction. We did not perform photo patch testing, because we had no more ink material available. Photo-patch testing with the ink has been mentioned in only two studies, in patients in whom reactions followed sun exposure and for both of whom the tests gave negative results (8, 14).

Our patients experienced severe and long-lasting pruritus. Reduced quality of life was recently reported in patients with tattoo reactions (29), and treatment is difficult. In our series, as elsewhere, treatment with a potent topical steroid and intralesional triamcinolone was unsuccessful (3, 7, 8, 13, 30). Pulsed laser therapy is effective in removing tattoos by selectively blasting the pigment free into the dermis, which is then secondarily removed by macrophages (4, 20, 31). With their consent, 2 of our patients, for whom topical and intralesional corticosteroids were ineffective, were treated with a Q-switched 532-nm Nd:YAG laser. Laser therapy is controversial, because of the toxicity of photoproducts and the risk of worsening the reaction. Vasold et al. showed that laser light decomposes the azo pigments into hazardous products, such as 2-methyl-5-nitroaniline, a mutagen substance, 4-nitrotoluene, which has a genotoxic potential, 1,4-dichlorobenzene, 2,5-dichloroaniline, and methoxy-naphthol-AS (32). One of our patients (patient 1) had a severe reaction during laser treatment. Local and generalized reactions after laser treatment for tattoo removal have been imputed to local or systemic diffusion of pigment (33–35). In our patient, however, in addition to a local reaction, inflammation restricted to the red areas in two distant tattoos was observed. This local reaction in the absence of a generalized reaction cannot be explained by systemic diffusion of the allergen.
Only one article has reported a reaction in a distant tattoo during laser removal of another asymptomatic tattoo (36). The mechanism of this distant activation was unknown. Vasold and Engel showed that UV light, sunlight and laser radiation of azo pigments can induce the production of the same photoproducts (26, 27, 32). We surmised that laser irradiation of the tattoo on the left leg of our patient resulted in local induction of a large amount of photoproducts with enhancement of T specific lymphocytes that, after systemic diffusion, reacted in the red areas of other tattoos on the right leg and the left shoulder against similar photoproducts that were locally induced by sunlight UV. The reactions observed in our patient's brother suggest that the allergen could be produced by natural light exposure and even faster by intense laser light.

The use of lasers for treating tattoo reactions is controversial, surgery should be proposed when topical and intralesional corticosteroids are ineffective on tattoo hypersensitivity reactions. Excision is limited to very small tattoos, because of the risk of scarring. Dermatome shaving of tattoo reactions can be an effective alternative, with acceptable cosmetic results (37, 38), but it is important for all of the pigment to be removed.

There is a lack of legislation concerning tattoo products. The Council of Europe made recommendations, updated in 2008 (ResAP), for governments of the member states concerning the requirements and criteria for the safety of tattoos and permanent makeup (39). In France, legislation has been passed on the basis of the 2008 ResAP resolution. Tattoo and permanent makeup products must not contain or release certain listed aromatic amines, other substances that are prohibited in the composition of cosmetic products, and carcinogenic, mutagenic and reprotoxic substances. They must comply with the maximum allowed concentrations of impurities. In addition, like capillary dyes, they must not contain substances recognized to be sensitizing (40).

Three pigments used in our patients, Violet 19, Red 112 and Red 63, are on the list of the substances that tattoo and permanent make-up products should not contain. In France, there is also now a national vigilance system for monitoring the risk of side-effects resulting from the use of tattoo inks (41).

Conclusion

Reactions to tattoos are long-lasting and difficult to treat. They require special monitoring to identify the substances responsible, and patients need to be followed up to detect new reactions to colorants and inks.

Our observations provide further evidence for the involvement of azo pigments and quinacridone metabolites and photoproducts in hypersensitivity reactions to tattoos. Epicutaneous tests are not predictive, so patients who wish to have new tattoos should be informed of the risk of recurrence.

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