# Electrophoretic Mobility of Some Tattoo Dyes as an Approach to Remove Their Subcutaneous Traces

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## Synopsis

The electrokinetic ( $\zeta$ , zeta) potential was determined for a series of commercial tattoo pigments. A standard experimental method involving the measuring of the level difference formed in a U-shaped tube filled with a solution containing the dye after application of some potential difference was used to find  $\zeta$ -potential values. All of them were negative and sufficiently large to ensure electrophoretic mobility of the pigment particles in a special gelatin-based electrophoretic bed. Gelatin-based beds, one containing a pigment and the other without the pigment, were set side by side in a microelectrophoretic cell. The application of relatively low potential difference (20–25 V) provoked the migration of the pigment in the gelatin bed without pigment for as much as 10 mm after a 40-minute long electrophoresis. The intensity of the color of the pigment did decrease noticeably. These results seem to indicate the potential applicability of the reported method for the elimination of old and/or unwanted tattoo and of tattoo traces left after previous manipulations.

## INTRODUCTION

As tattooing becomes very popular all over the world, the necessity to remove old, no longer needed, blurred, deformed tattoo pictures is becoming more and more widespread too. Although "temporal" tattoo can be removed without any serious troubles, classical subepidermal patterns are very tenacious and sometimes require more time and effort to eliminate than was spent on their preparation.

There is a wide variety of approaches that can be used to remove or discolor the classical tattoo. They range from the simple mechanical scraping of skin layers together with the pigment particles fixed in them (1) to more advanced laser destruction (discoloration) of the pigment inside the patient's skin (1–3). It should be noted that all these approaches are more or less traumatic and may leave traces in the form of scars, hypo/hyperpigmentation of the skin, and changes in the skin texture and may provoke its contamination or

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persisting inflammation (4,5). Even the most gentle methods may leave some pigment traces in the form of shades or blurred spots. These problems may push patients to add new tattoos over leftovers of the old ones to hide these cosmetic defects.

Therefore, it is obvious that the need of complete removal of old tattoo pigments with minimal skin damage is undoubtedly acute. This need will become even greater when tattoo carriers decide that their tattoo pictures should be removed or corrected. In this context, we consider possible approaches that may lead to new solutions to be used in the technologies of tattoo removal.

Organic tattoo pigments are particles usually sized from 200 nm to 5 µm (6), which are electrophoretically mobile. Their electrokinetic (zeta) potential can be determined using appropriate methods such as the classical measuring of the difference in liquid levels reached in the U-shaped pipe after application of some fixed external DC-voltage during some period of time. This method is still extensively used in many fields of applied technique, chemistry, and biology (7–9).

Then, when the determined zeta potential value is sufficient to expect any substantial mobility of the particles, the possibility of tattoo removal can be tested further in direct experiments involving a model of the human skin and, finally, be verified in the experiments with real skin in volunteers.

Therefore, it is important to determine some typical zeta potentials of the tattoo pigments in isotonic solutions.

## MATERIALS AND METHODS

## PIGMENTS

A series of unbranded dyes for tattooing (Figure 1) was purchased at an online store and then used for the preparation of the solutions and other mixtures to investigate their electrophoretic mobility. Only four color samples: black, white, red, and green were selected for this investigation because they are the most popular for tattoo pictures.

## PREPARATION OF PIGMENT SUSPENSIONS

All the pigments were suspended in isotonic solution (0.9% NaCl) to reach the concentrations described in the corresponding manuals (0.25 mL/L). As a result, four brightly colored solutions were obtained and then each of them was placed in the U-shaped glass tube (Figure 2, 1) for measuring its zeta potential.

# DETERMINATION OF THE $\zeta$ POTENTIAL

Following the classical method of determination (10), each solution was poured into the tube in such a way to fill it over the main faucets (Figure 2, 2) level. Then both faucets were closed and excessive colored solution removed from the upper parts of the device (Figure 2, 3), which then was filled with the coupling fluid (same isotonic solution of



Figure 1. Samples of tattooing pigments used for investigation of the electrophoretic activity.

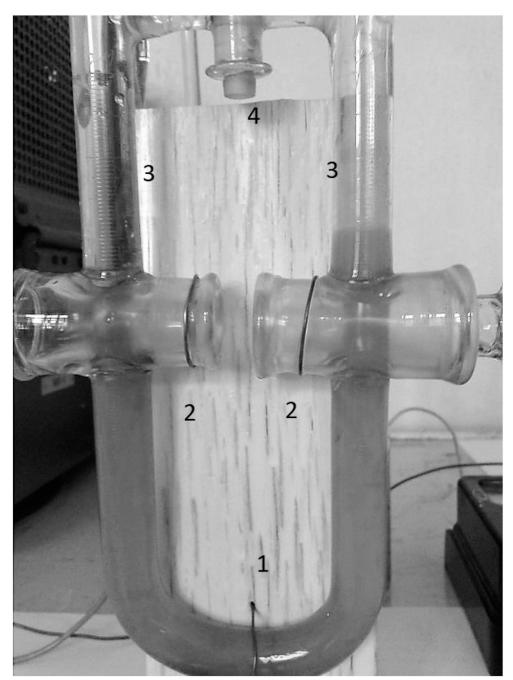
NaCl containing no dissolved dyes). Finally, the balancing faucet (Figure 2, 4) was opened to equalize the coupling fluid levels in both parts of the tube.

Afterward, both extremities of the U-shaped tube were connected electrically to the power supply through additional agar—agar bridges installed between each bend of the tube and the power supply outputs. Then the main faucets (2) were opened and voltage was applied between both parts of the glass tube.

Because the dye grains acquire some electric charge in the isotonic solution, they will move toward one of the poles causing a level difference between working and coupling liquids. Having measured the difference between the margins in each bend, the zeta potential can be calculated by the following formula (11):

$$\zeta = \frac{\eta b l}{E \varepsilon \varepsilon_0 t},$$

where  $\eta$  is the viscosity of working solution (because all working solutions were quite diluted, it was taken as equal to the viscosity of water  $1 \times 10^{-3}$  Pa s); b is the final difference between the solution levels in each bend, m; l is the distance between the axes of two parts of the U-shaped tube (0.58 m); E is the electric voltage applied; (260 V throughout all experiments);  $E_0$  is the standard vacuum dielectric permittivity constant (8.85  $\times$   $10^{-12}$  F/m); E is the relative dielectric permissivity of water (equal to 81); and t is the duration of electrophoresis (s).



**Figure 2.** U-shaped tube for experimental determination of zeta potential. A difference in the liquid levels was achieved as a result of electrophoresis. (1) Working part filled with the dye solution; (2) main faucets; (3) coupling fluid parts and (4) balancing faucet.

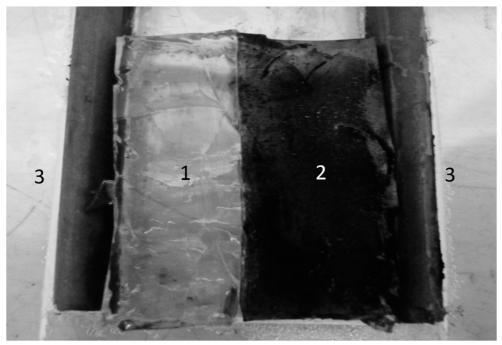


Figure 3. Microelectrophoresis cell with the gelatin samples: (1) raw sample; (2) the sample consisting some dye; (3) graphite electrodes. Let us compare positions of the boundary between the colored and not-colored samples for various gelatin/black dye samples.

The dye solution level did rise in the "positive" bend (Figure 2, right side) for all four samples meaning that the dye grains for all colors used in the framework of our experiments acquired a negative charge when dissolved in the isotonic NaCl solution.

#### ANALYSIS OF THE ELECTROPHORETIC MOBILITY OF PIGMENTS

Because the zeta potential of the black pigments was the largest, it was selected for the investigations related to applicability of electrophoresis to extract samples of "real" pigments samples from tissues. No human or animal tissue samples were used because of ethical restrictions and gelatin-based mixtures were used at that stage.

Twenty-five milligrams of unbranded gelatin was dissolved in 500 mL of hot water to obtain such mixtures. Then the solution was divided into two parts; one of them was left for cooling and hardening, whereas the required amount of the pigment suspension was added to the second one to make its concentration equal to 0.25 mL/L. Then it was also left for cooling and hardening.

Parallelepipedal samples were cut out of each part and placed into a plastic microelectrophoretic cell (Figure 3). The cell was designed in such a way (12) that each sample was in contact with the graphite electrodes located on the outer edges of the cell and connected to an external electric power supply, whereas the inner sides of the samples were in contact with each other ensuring transferability of the dye particles between the samples. Because the dye particles can move from the colored sample toward the not-colored one under the applied external electric voltage, this process can be visualized by distortion or shifting of the margin between the samples. In case it moves noticeably, the principal applicability of this method to extract the tattoo dyes out of human skin tissues can be expected.

All microelectrophoretic experiments involving gelatin samples were carried out under the following conditions: DC voltage applied, 20–25 V; current, 5–7 mA; and process duration, 2,400 s. It should be noted that such, or close, potential values are used regularly in practice at various medical electrophoretic manipulations or biochemical investigations (13).

## **RESULTS**

Average White

Average

All experimental results related to determination of the dyes' zeta potentials are shown in Table I.

As seen from the average zeta potential values given in Table I, the difference between the potentials of various dyes was larger than the experimental error. It means that the dye composition is an influential parameter governing the value of its electrokinetic potential. Because the zeta potential of the most popular black pigment was the largest, it should be the most mobile and the most suitable for electrophoretic extraction. Therefore, this pigment was selected for further experiments.

As seen from Figures 4 and 5, the black dye's mobility was quite significant and it reached 10 mm even after the 40-min-long electrophoresis. Besides, it can be noticed that the

Dye	Electrophoresis duration, s	Level difference, m	ζ, V
Red	1,200	0.020	-0.135
		0.020	-0.135
		0.024	-0.162
Average		0.021	$-0.142 \pm 0.0119$
Black	1,200	0.020	-0.135
		0.024	-0.162
		0.022	-0.148
		0.026	-0.175
		0.020	-0.135
Average		0.024	$-0.151 \pm 0.014$
Green	1,200	0.018	-0.121
		0.016	-0.108
		0.022	-0.148
		0.020	0.135

0.026

0.0204

0.012

0.016

0.02

0.02

0.014 0.064

0.175

 $-0.138 \pm 0.0194$ 

-0.0810

-0.108

-0.135

-0.135 -0.0944

 $-0.111 \pm 0.0194$ 

Table I

Experimental Data and Calculated Zeta Potentials for the Dyes

1,200



Figure 4. Mobility of the black dye particles after the 2,400-s electrophoresis. Left, before; right, after electrophoresis. The boundary has shifted for approx. 10 mm.

color intensity of the black pigment becomes weaker after the electrophoresis. It is quite obvious because some portions of the dye were migrating away from the sample, so its concentration and coloring intensity should decrease.

## DISCUSSION

All tested tattoo samples have revealed the negative zeta potentials, although their values were sufficient to maintain quite significant mobility of the dye grains inside the faux skin samples. The margin between colored and not-colored gelatin samples has moved for about 10 mm after 40-min-long application of the 20–25 V potential difference. These results prove that the method presented in this work looks quite promising for elimination of the old or unwanted tattoos alone or in combination with the others. In the latter case, it should be used at the final stage to eliminate the residual traces and shadows of the tattoos. Low voltages and comparatively short application time tested in the framework of present investigation allow one to expect that the volunteer requesting the removal of the tattoo should not experience serious discomfort because of local skin overheating during the treatment. However, a series of thorough experiments is recommended with tattooed animal skin to determine the exact regimes of electrophoresis before testing this approach on human volunteers.

This result promises potential applicability of this method in technologies of elimination of old, blurred, or unwanted tattoos.

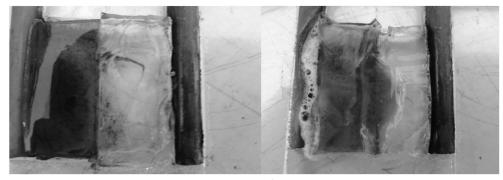


Figure 5. Mobility of the black dye particles after the 2,400-s electrophoresis. Left, before; right, after electrophoresis. The boundary has shifted for approx. 8 mm.

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