Skin Pigmentation and Texture Changes after Hair Removal with the Normal-mode Ruby Laser

Evaluations by Skin Reflectance, Profilometry, and Ultrasonography

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Promising clinical results have been obtained with the normal mode ruby laser for removal of unwanted hair. Melanin within the hair follicles is thought to act as target for the ruby laser pulses, whereas epidermal melanin is thought to be a competitive chromophore, responsible for potential side effects. This study aimed (i) to objectify postoperative changes in skin pigmentation and texture and (ii) to evaluate the importance of variations in preoperative skin pigmentation for the development of side effects 12 weeks after 1 treatment with the normal-mode ruby laser. A total of 17 volunteers (skin types I – IV) were included. Each volunteer was exposed to a range of normal-mode ruby laser fluences on 3 different test areas located at the proximal part of the hairy pubic region (n=51 test areas). A shaved test area served as control. Skin reflectance spectroscopic measurements, 3-dimensional surface contour analysis and ultrasonography objectified postoperative changes in skin pigmentation and texture. Blinded clinical assessments revealed postoperative hyperpigmentation (2% of test areas) and hypopigmentation (10%), whereas no textural changes were seen. Reflectance spectroscopically-determined pigmentary changes depended on the degree of preoperative skin pigmentation, fairly pigmented skin types experiencing subclinical hyperpigmentation and darkly pigmented skin types experiencing subclinical hypopigmentation. Three-dimensional surface profilometry documented similar pre- and postoperative surface contour parameters, indicating that the skin surface texture is preserved after laser exposure. Ultrasonography revealed similar skin thicknesses in laser-exposed and untreated control areas. It is concluded that normal-mode ruby laser treatment is safe for hair removal in skin types I – IV. Key words: laser-assisted hair removal; experimental study; hyperpigmentation; hypopigmentation.

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Unwanted hair growth is a common problem. Recently, optical devices have been developed to remove hair, and promising clinical results have been obtained with different lasers (1 – 3) and with non-coherent intense light sources (4). Within the newest technology, 2 entirely different techniques have been developed to provide selective photothermolysis of hair follicles: in the first technique, light is absorbed by an exogenous carbon-based paint (Nd: YAG laser 1064 nm) (2, 5, 6). In the second technique, melanin in the follicular epithelium acts as an absorbing chromophore for photons delivered by either the long-pulsed ruby laser (694 nm), the long-pulsed alexandrite laser (755 nm), or the non-coherent intense light source (1, 3, 4, 7, 8). Clinical studies with the long-pulse ruby laser have shown different treatment outcomes: 3 studies have published mean regrowth values from 40 – 80% 3 months after the laser exposure (1, 9, 10) and 1 study has reported that no difference in hair regrowth could be detected between the ruby-laser-treated area and the control area 12 weeks postoperatively (11). When repetitive treatments are performed with the normal-mode ruby laser, the mean percentage regrowth is found to be 44 – 65.5% after the first treatment, 33 – 41% after the second treatment and 29 – 34% after the third treatment (10, 12). Moreover, the question of permanency or long-lasting hair removal has recently been addressed with the normal-mode ruby laser as laser-induced alopecia occurred in 4 out of 7 participants 2 years after a single laser exposure (13).

An overall satisfactory treatment outcome after laser-assisted hair removal requires a high clinical efficacy and a low occurrence of postoperative side effects. Side effects are attributed to non-specific, thermal damage of epidermis and dermal extrafollicular structures and may be due to competitive absorption by the epidermal melanin (14). The absorption spectrum for melanin is broad and includes the emission spectrum for the long-pulsed ruby laser (15). In ruby laser-assisted hair removal it is, therefore, important to avoid epidermal melanin absorption, which may result in transient or permanent side effects such as pigmentary or textural changes.

This study intended (i) to objectify postoperative changes in skin pigmentation and texture and (ii) to evaluate the importance of preoperative variations in skin pigmentation for the development of side effects 12 weeks after 1 treatment with the normal-mode ruby laser.

MATERIALS AND METHODS

Subjects and protocol

A total of 17 healthy, adult Caucasians (9 females, 8 males) were enrolled in the study after informed consent was given. The skin type classification described by Fitzpatrick was used (16): according to that, 2 volunteers had skin type I, 7 skin type II, 5 skin type III, and 3 had skin type IV. Blonde, red, brown and black hair colours were included. Each volunteer was exposed to a range of normal-mode ruby laser fluences on 3 different test areas located at the proximal part of the hairy pubic region (n=51). In addition, there was a shaved control test area (n=17). Treatment modalities were selected according to prior randomization to the test areas that were shaved the day before laser exposure.

Preoperatively and 12 weeks postoperatively the following assessments were performed: (i) clinical evaluations by a blinded physician; (ii) skin reflectance spectroscopic measurements to objectify preoperative skin pigmentation and postoperative laser-induced pigmentation changes; (iii) 3-dimensional skin surface contour analyses to
quantify laser-induced surface contour changes; and (iv) blinded high-frequency ultrasound examinations to objectify changes in skin thickness.

Laser techniques
A Chromos 694 depilation ruby laser (SLS/Biophile, Wales, UK) was used for the treatments.

The ruby laser operates at a wavelength of 694 nm in the free-running, long pulse mode. The pulse duration was 0.8 ms. Laser treatments were performed once with 3 fluence levels delivered to each volunteer, 15–20 and 25 J/cm². The spot diameter was 5 mm and the repetition rate of the laser was 1 Hz. Nine laser spots were applied to each test area (1 cm²). An external laser energy meter was used to monitor fluences (Ophir Nova 10P, Ophir, Israel). No local anaesthesia was used during the laser treatments.

Skin reflectance
Pre- and postoperative skin pigmentation was measured by a reflectance spectrometer (UV-Optimize, Matic, Herlev, Denmark). The equipment quantifies skin pigmentation and skin redness independently on relative biological scales from 0% to 100% (17). Zero percent pigmentation is found in white skin with no pigment at all and 100% corresponds to the pigmentation in theoretically black skin with no light reflection. The method has previously been used to quantify skin pigmentation before and after laser exposure (18, 19).

Three-dimensional surface contour analysis
A total of 64 silicone replicas were obtained preoperatively and 12 weeks postoperatively from laser-exposed and untreated control areas (Silflo, Flexico, UK). The replicas were scanned by a laser-optical profilometer equipped with an optical triangulation sensor (OTM) and operating with a sample density of 25 points/mm (UBM Messtechnik GmbH, Ettlingen, Germany). The measured area could be identified with an accuracy of 40 μm. Skin surface topography was described by means of 3-dimensional parameters: The interdependent amplitude parameters $S_a$ (the arithmetic mean deviation) and $S_q$ (the root mean square of $S_a$) describe the average deviation of the profile from the mean line and give a first impression of the average roughness. $S_q$ represents the height of the core material portion, i.e. the bearing area, $S_{vk}$ and $S_{pk}$ the surface valley proportion and surface peak proportion, respectively, representing the material below and above the core surface (20).

Ultrasonography
Cross-sectional B-mode scans of the skin were obtained with a 20-MHz ultrasonograph (Dermascan C1, Cortex Technology, Hadsund, Denmark) (21, 22). The gain compensation curve was kept constant throughout the investigation. The transducer used has a lateral resolution of 130 μm, an axial resolution of 60 μm and a maximum depth of the viewing field of 30 mm. The velocity of ultrasound in the skin is 1580 m/s. The ultrasonic wave is partially reflected at the boundary between adjacent structures and generates echoes of different intensities, which after digital processing are visualized as A-scans. Skin thickness (mm) was measured as an average of all 224 A-scans contributing to the B-mode image, corresponding to the resolution of 60 μm and a maximum depth of the viewing field of 30 mm. The velocity of ultrasound in the skin is 1580 m/s. The ultrasonic wave is partially reflected at the boundary between adjacent structures and generates echoes of different intensities, which after digital processing are visualized as A-scans. Skin thickness (mm) was measured as an average of all 224 A-scans contributing to the B-mode image, corresponding to the

\[ r = 0.52, P = 0.03 \]

\[ r = 0.50, P = 0.04 \]

\[ r = 0.49, P = 0.05 \]

Fig. 1. Clinical scores of postoperative hypopigmentation (■) and hyperpigmentation (□) 12 weeks postoperatively vs. the applied fluence levels. Only test areas with clinically visible pigmentary changes are illustrated.

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\[ \Delta \text{pigm}\% = \text{postoperative pigmentation}\% - \text{preoperative pigmentation}\% \]

\[ r = 0.52, P = 0.03 \]

\[ r = 0.50, P = 0.04 \]

\[ r = 0.49, P = 0.05 \]

Fig. 2. Skin reflectance-determined postoperative pigmentary alterations vs. the preoperative skin pigmentation. The laser-induced pigmentary changes (Δ pigm%) were calculated as: postoperative pigmentation%–preoperative pigmentation%, positive values indicating hyperpigmentation, negative values hypopigmentation. Linear regression lines with 95% CI lines, correlation coefficients (r) and test for regression line slopes different from zero (P) are indicated for each fluence level.
distance from the epidermal entrance echo to the interface between the skin and subcutaneous fat. Because ultrasonography is not an entirely objective measurement, the subsequent image analysis was performed in a blinded fashion. The laser-induced change in skin thickness was calculated as a percentage: \( \frac{(\text{Thickness}_{\text{postoperative}} - \text{Thickness}_{\text{preoperative}})}{\text{Thickness}_{\text{preoperative}}} \times 100 \).

**Statistics**

Before-and-after comparisons and comparisons with the untreated control area were analysed by the paired t-test, since data passed the Kolmogorov-Smirnov normality t-test (skin pigmentation, skin thickness and surface contour parameters). Linear regression analysis was used to evaluate the relation between preoperative skin pigmentation and laser-induced postoperative pigmentary changes. \( p < 0.05 \) was regarded as statistically significant.

**RESULTS**

Laser-induced side effects were evaluated by skin reflectance measurements to objectify changes in skin pigmentation, and by ultrasonography and 3-dimensional contour analysis to objectify textural changes. Clinical evaluations were performed by a physician, blinded to the treatment data.

**Pigmentary changes**

**Clinical evaluation.** Five test areas responded with hypopigmentation (10%) and one with hyperpigmentation (2%) (Fig. 1). The most intense pigmentary alterations were experienced at the highest fluence levels and in dark skin types.

**Skin reflectance.** The postoperative reflectance-determined skin pigmentation differed from the preoperative skin pigmentation in 47 of 51 test areas (92%), indicating the presence of both clinical and subclinical hyper- or hypopigmentation. The intensity of reflectance-determined pigmentary changes depended on the degree of preoperative skin pigmentation (Fig. 2), lightly pigmented skin types experiencing clinical or subclinical hyperpigmentation and dark skin types experiencing clinical or subclinical hypopigmentation.

**Textural changes**

**Clinical evaluation.** No test areas developed epidermal alterations, atrophy, or hypertrophy.

**Three-dimensional surface profilometry.** The overall surface topography appeared similar before and after treatment, corresponding to similar before-and-after values of the surface contour parameters. Each fluence level (15, 20, 25 J/cm\(^2\)) was tested separately and no significant differences were observed between the tested parameters (\( S_a, S_q, S_k, S_p, S_n \)). The parameters also remained constant in the untreated control areas.

**Ultrasonography.** The calculated percentage of change in skin thickness was not significantly different in the laser-exposed test areas (15, 20, 25 J/cm\(^2\)) vs. the untreated control area.

**DISCUSSION**

This is, to our knowledge, the first study that applies non-invasive, objective methods in the evaluation of postoperative side effects from laser-assisted hair removal. The study provides information about the effects on skin pigmentation, thickness and surface contour after 1 treatment with the long-pulsed ruby laser for hair removal. Skin reflectance measurements documented the presence of subclinical, postoperative pigmentary changes, since the postoperative reflectance-determined skin pigmentation differed from the preoperative skin pigmentation in 47 out of 51 treated test areas and clinically visible pigmentary changes were only visible in 6 of the treated test areas. No linear dose-response relationship was observed, which may be due to inhomogeneity in preoperative skin characteristics (hair colour and density, collagen density). However, light-pigmented skin experienced postoperative subclinical hyperpigmentation and dark-pigmented skin experienced subclinical hypopigmentation. This finding is in accordance with the fact that ruby laser light in dark-pigmented skin is strongly absorbed by the epidermal melanin, leading to damage of melanocytes (14), whereas thermal effects in fairly-pigmented skin may provoke post-inflammatory hyperpigmentation. In contrast to the laser-induced postoperative pigmentary changes, surface profilometry and ultrasonography documented that the surface texture and skin thickness are preserved after ruby laser treatment and, moreover, that increasing preoperative skin pigmentation poses no risk of inducing postoperative texture changes in skin types I–IV.

By clinical examination, 2% of the test areas responded with postoperative hyperpigmentation and 10% responded with postoperative hypopigmentation. No epidermal alterations, atrophy, or hypertrophy was observed. In general, these results are in accordance with the incidences of side effects reported in the literature: Transient pigmentary changes have been the most frequently reported side effect from ruby laser treatment, hypopigmentation occurring in 3–23% of the treated patients and hypopigmentation in 0–15% (1, 7, 10, 23). In a follow-up study with a 2-year observation period no permanent pigmentary changes were observed, thus supporting the theory that ruby laser-induced pigmentary changes are temporary (13). No significant scarring has been reported in any of the clinical studies with ruby laser-assisted hair removal. The fact that a remarkable variation is reported concerning the incidences of postoperative pigmentary changes may be explained by the circumstance that the referred studies have not been carried out under standardized conditions: different physical laser parameters have been used, the follow-up period varied from 90 days to 2 years, the preoperative skin characteristics were not standardized (hair colour, skin pigmentation, anatomical region), and the majority of papers estimated the incidences of side effects by subjective clinical evaluations. The presented study represents the first approach to objectify side effects from laser-assisted hair removal.

The mode of action of ruby laser-assisted hair removal is selective photothermolysis of melanin-containing structures in the hair follicles (24). It is therefore evident that patients with lightly pigmented grey, white, yellow or red hair obtain a reduced percentage of hair follicles, which are permanently damaged, and thus have a limited success rate compared with patients with brown or black hair (1). Preoperative hair colour may also influence the occurrence of laser-induced side effects as dark and densely located hair follicles absorb more laser energy than lightly pigmented and loosely arranged hair follicles. Dark hair, therefore, may be at higher risk of
thermal-induced side effects. In the present study no measurements were performed on hair colour and, therefore, no relationship could be established between preoperative hair colour and postoperative side effects. In order to obtain long-lasting hair removal from ruby laser treatment, it is necessary to treat the skin several times. In this study only 1 treatment was performed and the side effects may be underestimated for repetitive treatments.

Before laser-assisted hair removal it is important to consider the suitability of the skin for the laser procedure and to recognize if lightly pigmented hair or dark skin colour constitute a limitation for an optimal treatment outcome. However, from the results in this study, it is concluded that normal-mode ruby laser treatment is safe for hair removal in skin types I–IV.

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REFERENCES