Ablative Targeting of Fatty-Tissue Using a High-Powered Diode Laser

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Background and Objective: Concerning current clinical practice, laser-assisted lipoplasty is still secondary to other procedures. In order to evaluate effects of thermal interaction with fatty-tissue, a near infrared diode laser was examined under reproducible conditions.

Methods: Based on optical spectroscopy of fatty-tissue, a high-powered diode laser (\(\lambda = 940 \text{ nm}\)) was used to irradiate \(n = 59\) fat samples of fresh corpses in non-contact mode. Thermal effects were histologically evaluated by computer based metric measurements. Calculated values included ablation rate (AR) and the ratio of cavity diameter to diameter of collateral damage (CCD\(_{\text{ratio}}\)). Pearson’s correlation and analysis of covariance (ANCOVA) were used for statistical evaluation. \(P\) values of less than 0.05 were considered to indicate statistical significance.

Results: Regarding the conditions examined, irradiances from 250 to 400 W/cm\(^2\) revealed both increased ablation capacities and decreased collateral damages. An average irradiance of 370 W/cm\(^2\) shows an average CCD\(_{\text{ratio}}\) of 2:1 and an average AR of 9.98 mm\(^3\)/second.


Key words: diode laser ablation of fat; lipoplasty; optical properties of fatty-tissue

INTRODUCTION

Soft-tissue reconstruction using free or pedicled flaps often require several flap debulking procedures [1–3]. At present, precise flap contouring by means of laser-assisted lipoplasty does not fit clinical requirements [4–6]. However, little is known about diode laser ablation of fat. Diode lasers provide some benefits for ablative fat removal. Proposed advantages include: fiber conduction of laser light for endoscopic purposes, an intensity range of \(10^2\)–\(10^6\) W/cm\(^2\) creating thermal interaction, preventing photochemical interaction (\(10^6\) W/cm\(^2\)), plasma inductive ablation (\(10^9\) W/cm\(^2\)), photodisruptive interaction (\(10^{15}\) W/cm\(^2\)). Further advantages are a near infrared (NIR) spectrum, deep surface penetration of several millimeters, high efficiency, and finally low price [7–10].

In this study, based on spectroscopic measurements of optical absorption properties of fatty-tissue, a continuous waved (cw) high-powered diode laser set at a wavelength of \(\lambda = 940 \text{ nm}\) was used to irradiate human samples of adipose tissue. Thermal effects were histologically evaluated by computer based metric measurements.

METHODS

Examination of Optical Properties of Fatty-Tissue

Measurements of absorption properties of human adipose tissue taken from fresh and unfixxed female body donors were carried out. Fat samples were mechanically homogenized and centrifuged for 6 minutes at 10,000 rpm. The solid cellular components were discarded. The liquid (oily) cellular components were separated. Two different specimens were examined: (1) homogenized “not bloody tinged fatty-tissue” (NBFT) separated from fat samples lacking of blood (appearing yellow in color); (2) homogenized “bloody tinged fatty-tissue” (BFT) separated from fat samples covered with blood (appearing yellow to light red in color). Absorption property of water (isotonic sodium chloride) was also evaluated.

Optical transmission \(T(\lambda)\) of each specimen was determined using a Perkin-Elmer Lambda25 grating spectrometer (\(\lambda = 190–1,100 \text{ nm}\)) covering the visible and the near UV range. Liquids were placed in the optical path using cuvettes for spectroscopy (Kartell, UV range disposable PMMA

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cuvettes, physical size 45×12.5 mm (h×w). Optical absorption \( A_{\lambda} \) was determined by using the equation:

\[
A_{\lambda} = -\log(T_{\lambda})
\]

**Laser Ablation of Fat and Histological Examination**

Fifty-nine lipocutaneous samples of fat taken from body donors, harvested from the antero-medial thigh region of eight fresh and unfixed female human corpses, were examined. Harvested samples underwent laser irradiation within 48 hours after patients’ death. During laser irradiation fat samples were put into a custom-made device. The laser beam was lined up parallel to and below the skin, striking the subcutaneous fatty-tissue from the side. Fat samples were continuously irrigated with a controlled, constant flow of tap water. A burette was used for irrigation.

A high-powered diode laser (Laserline LDF-600 Diode) set at a NIR wavelength of \( \lambda = 940 \) nm was used. The laser delivered 500 W at maximum power in cw-mode. The laser light was conducted through a High-YAG plugged, polished fiber (core diameter 1,000 \( \mu \)m ± 2\%, Teflon Jacket, external diameter 1,600 \( \mu \)m). The emitted free laser beam was defocused in front of each fat sample by an anti-reflex coated quartz-glass lens system (two plano-convex lenses; \( f = 65 \) mm). Irradiance ranged from 93 to 1,579 W/cm² based on an output power of 100–500 W and a laser spot size ranging from 0.17 to 1.08 cm². Total duration of exposure was 30–90 seconds.

Histological evaluation of more than 1,000 sections was performed. Tissue staining was completed using a modified trichrome Masson–Goldner technique (nuclei stain brownish-black; the cytoplasm ranges from a bright red to green; connective tissue takes on a green stain whereas elastic fibers appear violet). This technique proved to be particularly well-suited for differentially evaluating thermal damage of adipose tissue. Cross-sections, taken 300 \( \mu \)m apart, were made with a thickness of 6 \( \mu \)m. Light microscopy was used for histological examination. Computer based metric measurements (software DISKUS 32 picture representation, version 4.25.6) were taken to quantify the thermal impact of laser energy on fatty-tissue. Measurements were performed within different depth of the fat ablated cavities at 0%, 50%, 90%, and 100% cavity depth. Measured parameters (mm) included cavity width (CWI), cavity depth (CDE), border of carbonization (BC), border of moderate collateral damage (BMCD), zone of septalized collateral damage (ZSECD), and zone of spotted collateral damage (ZSPCD). In addition, calculated parameters included collateral damage (CD = BC + BMCD + ZSECD + ZSPCD) measured in mm, cavity volume (CV) geometrically based on a cylinder and measured in mm³, ablation rate (AR) measured in mm³/second, and the ratio of cavity diameter to diameter of collateral damage (CD \( \text{ratio} = \text{CWI} : (\text{BC} + \text{BMCD} + \text{ZSPCD} + \text{ZSECD})). AR was defined as vaporized volume of fat (CV), measured in mm³, per second [s]. AR should briefly supplement information concerning varying ablative capacities of the diode laser used.

**Statistical Evaluation**

Statistical evaluation was carried out for exploratory purposes. \( P \) values of less than 0.05 were considered to indicate statistical significance. Values are given as means ± SD.

Pearson correlation coefficient (\( r \)) was assessed in order to examine associations between energy density, irradiance as well as duration of exposure (30, 40, 45, 60, 75, 90 seconds), defined as independent variables, and corresponding measured parameters, defined as dependent variables. In addition, analysis of covariance (ANCOVA) was used for multivariate evaluation. Separate three-way and four-way analyses of covariance were done followed by \( f \)-tests and post hoc \( t \)-tests in order to proof statistical significance. Finally, residual analyses were done to evaluate validity of models assumptions.

Statistical analyses, approved by the Institute of Medical Statistics of the University Hospital Aachen, were done using SAS for Windows (SAS Institute Version 9.1, Cary, NC).

**RESULTS**

**Evaluation of Optical Properties of Fatty-Tissue and Water**

Separated oily component of fat cells showed a yellow to light red color. Spectroscopic analyses demonstrated three chromophoric absorption peaks of different amplitudes of the oily component of fatty-tissue shown in Figure 1. The highest peak was found at a visible light wavelength of \( \lambda = 460 \) nm. The other two peaks were lower, seen at

![Fig. 1. Absorption properties of the oily component of fatty-tissue relative absorption (abbreviated units) as a function of wavelength; three absorption peaks at a wavelength of \( \lambda = 460 \) nm, \( \lambda = 930 \) nm and \( \lambda = 1,030 \) nm for the NBFT and BFT specimens, as well as the peak of water (isotonic sodium chloride) at a wavelength of \( \lambda = 975 \) nm Abbreviations: NBFT, not bloody tinged fatty-tissue; BFT, bloody tinged fatty-tissue; H₂O, water.](image-url)
wavelengths within the NIR range of $\lambda = 930$ nm and $\lambda = 1,030$ nm. Water revealed an absorption peak at a wavelength of $\lambda = 975$ nm. The operating wavelength of the high-powered diode laser used, $\lambda = 940$ nm, approaches both the NIR absorption peak of fat and of water.

**Evaluation of Laser Ablation of Fatty-Tissue**

Histological sections, stained in modified trichrome Masson–Goldner technique, demonstrated diverse morphological changes corresponding to different laser settings (see Fig. 2).

Samples showed characteristic features of collateral damage (CD) in comparison to control specimens. Vacuolated and condensed zones were seen surrounding the central cavity caused by laser ablation. Lack of permanent irrigation caused severe collateral tissue damage exemplarily shown in Figure 2 Panel B. Based on these specimens, total CD was defined by three main circular regions surrounding the central cavity: an inner layer appearing bright black (BC), a compact middle layer staining bright red (border of excessive collateral damage (BECD)), and an outer layer staining light red (BMCD) characterized

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![Fig. 2. Histological studies of fatty-tissue stained in modified trichrome Masson–Goldner technique Images (A–F) show exemplary light-microscopic views of adjacent thermal impairment of fatty-tissue caused by different irradiances (W/cm²). Duration of exposure was 30 seconds. Borders and zones of collateral tissue damage are outlined by arrowheads. Original magnification 2.5× A (above left): control sample; thermally unimpaired fatty-tissue B (above right): 1,300 W/cm²; histological example for severe collateral damage (CD) following irradiation without continuous irrigation; note the broad BC (stained black) and severe CD (stained strongly red). C (middle left): 370 W/cm²; note the increased width of cavity and minor CD. D (middle right): 370 W/cm²; detailed view; carbonization is absent; only minor CD is evident without loss of nuclear staining. E (below left): 1,250 W/cm²; effects of distinct heat conduction showing an increased border of CD and a decreased cavity width. F (below right): 1,250 W/cm²; detailed view; note the vacuolated tissue, specific red staining and loss of nuclear staining. BC, border of carbonization; BECD, border of excessive CD; BMCD, border of moderate CD; ZSPCD, zone of spotted CD; ZSECD, zone of septalized CD. [Figure can be viewed in color online via www.interscience.wiley.com.]
by tissue vacuolization and disruption of cell membranes through expansion of water and fat. In addition, two radiating zones were defined: a ZSPCD and a ZSECD. Both of these zones appeared dark to light red in color and were found in between connective tissue septa.

The onset of fat ablation was delayed up to 10 seconds after the laser was applied to the tissue. The CCD_ratio shown in Figure 3 (Panel D) was 3:1 using irradiances of about 300 W/cm², whereas irradiances of about 1,300 W/cm² approaches an inverse ratio of 1:2. High irradiances may therefore cause disproportionately more heat conduction and CD in relation to the amount of fat ablated. This effect may result from both increased absorption and decreased transmission properties of fatty-tissue following irradiation. As a result, change of optical properties of fatty-tissue may then reduce ablative capacity of the diode-laser. Concerning value

Fig. 3. A–D: Scatter plots (n = 18; duration of exposure was 30 seconds). Panel A (above left): dependence of energy density on ablation rate (AR); note the clear decrease of AR beyond 60 J/mm³ corresponding to irradiance values beyond 370 W/cm²; Panel B (above right): dependence of irradiance on CV; note increase of ablation up to 370 W/cm² and decrease beyond 370 W/cm²; Panel C (middle left): dependence of irradiance on AR; note the fading of AR beyond 370 W/cm²; Panel D (middle right): dependence of irradiance on the ratio (CCD_ratio) of cavity diameter (mm) to diameter (mm) of CD; Panel E (below left): dependence of irradiance on CD; total CD increases with higher irradiances; Panel F (below left): dependence of irradiance on border of carbonization; in case of occurrence intensity of carbonization increases with higher irradiances. [Figure can be viewed in color online via www.interscience.wiley.com.]
averages, an irradiance of 370 ± 0 W/cm² corresponding to an energy density of 58.96 ± 41.38 J/mm³ (Fig. 3A), showed the best overall results for ablative fat removal. This irradiance level, reflected by a favorable average CCD ratio of 2:1, was characterized by an AR of 9.98 ± 7.65 mm²/seconds.

Linking up all effects seen, irradiances from 250 to 400 W/cm² may reflect the ideal power setting for ablative fat removal regarding the conditions examined (see Fig. 3D).

**Statistical Results**

Concerning Pearson’s correlation coefficient both irradiance (r ranging between –0.2284 and 0.4805) and energy density (r ranging between –0.1325 and 0.4911) showed a poor to moderate correlation to the dependent variables. Strongest associations were found regarding total CD and ZSECD.

Duration of exposure demonstrated a statistical significant effect on AR (P = 0.0061), CWI (P = 0.0056) and CV (P = 0.0329). Further more, irradiance as well as energy density showed statistical significant effects on CWI (P = 0.0440, P = 0.0333) and especially on the CCD ratio (P = 0.0066, P = 0.0228). However, neither irradiance nor energy density or duration of exposure proved a significant effect on total CD (all P > 0.05).

Residual analysis showed that the model assumptions were approximately valid in all 10 models.

**DISCUSSION**

Following soft-tissue coverage with flaps, liposuction-assisted lipectomy is the most commonly performed method to improve contour at the recipient area. Regarding fatty-tissue surgery, using lasers may provide benefits for precise fat removal. Although at present laser-assisted procedures cannot compete with other techniques such as ultrasonic-assisted liposuction [4,5,11,13]. Unfortunately, laser ablation of fatty-tissue is complicated by the fact that fat does not respond as homogeneously as other tissues—such as muscle—to laser energy [12]. The heterogeneous response of fatty-tissue to laser energy depends on energy requirements of phase changes of water from liquid to steam, tissue desiccation formed by rapid expansion of steam vacuoles, fat liquefaction/vaporization, and changes in optical properties of fatty-tissue.

Badin et al. [6], though, highlights certain advantages of laser lipolysis over ultrasonic-assisted liposuction using a Nd:YAG laser device, operating at a wavelength of λ = 1,064 nm. Neira et al. [14] suggest an optimum wavelength of λ = 630 to λ = 640 nm for laser liquefaction of fat.

Concerning our results a wavelength of λ = 940 nm achieves an increased absorption of both fatty-tissue and tissue water while maintaining a penetration depth of the laser of several millimeters. Requirements for a constant ablation process, however, include largely unvarying transmission properties of the tissue. Transmission, though, is complicated by carbonization decreasing penetration depth and increasing surface temperature [15–17]. Carbonization, therefore, promotes a mismatch between ablation and CD in favor of CD concerning amount and/or intensity (see Fig. 3E,F). This relationship shown in Figure 3 (Panel D) was reflected by the CCD ratio (see Fig. 3D). Cooling of the irradiated surface using water opposes a disproportional increase in surface temperature [16,18,19].

Nevertheless, the low water content of fat, measured to be 35 ± 6 ml/mol triglyceride [20], and its high flash point, stated to be 189 °C [21], makes high energy necessary for vaporization of fat. In contrast, vaporization of water begins through temperatures above 100 °C and therefore reflects the most important part of the ablation process [15,18,22]. Localized water deposits within connective tissue septa, known to contain nearly 70% of water [23,24], may be responsible for both side effects observed namely that of zones of septralized and that of spotted CD (ZSECD and ZSPCD). Such deposits show an increased affinity to conducted heat than surrounding fat cells which contain less water. Pulsed lasers, therefore, are proposed to be beneficial by reducing energy spread [22]. Collateral tissue damage, though, cannot be avoided [25]. Concerning histology, similar results were recently reported by Ichikawa et al. [26] examining a NIR, pulsed Nd:YAG laser device. However, improved efficiency of laser ablation should result from a wavelength matching the absorption properties of the tissue [18,22].

Finally, useful diode-laser ablation of fat, characterized by both an effective AR and minor CD, appears to be challenging but possible in vitro. Results may therefore justify some further exploration of that supplementary technique for use in soft-tissue surgery.

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