First assessment of the proionic effects resulting from non-thermal application of 448 kHz monopolar radiofrequency for reduction of edema caused by fractional CO\textsubscript{2} laser facial rejuvenation treatments

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Abstract: Among the side effects which occur after treatment with fractional CO\textsubscript{2} laser, one of the most frequent and incapacitating is temporary edema, which is functionally and esthetically incompatible with the patient going immediately back to their social and work life. The 448 kHz capacitive/resistive monopolar radiofrequency proionic system is based on the subthermal electrical stimulation of biological tissues, enabling the restoration of physiological membrane potentials, as well as the ionic balance established through the membrane. This system is capable of improving membrane permeability for an adequate maintenance of cell functions, as well as improving circulation and reducing fluid retention. This study involved one application before laser treatment and one application 24 hours after laser treatment. The results of skin ultrasound presented in this study show that proionic effects help restrict edema progression, thereby reducing recovery time.

Keywords: Radiofrequency-448kHz, Edema, Biostimulation, Post-Laser, Proionic Effects

1. Introduction

One of the main limitations of facial rejuvenation treatments with fractional CO\textsubscript{2} laser (L) is recovery time, which requires the patient to be inactive at least one week[1]. In order to try and reduce edema as well as its discomforts by shortening recovery time, we have worked with the proionic effects resulting from non-thermal application of 448 kilohertz (kHz) monopolar radiofrequency. Proionic effects or proionic system is the name given to the set of phenomena that take place at cellular level and lead to the restoration of the cell membrane’s normal potential, as well as the ionic balance established through the cell membrane. Although radiofrequency (RF) has been and is traditionally used for its thermal effects, Hernández-Bule \textit{et al} \cite{2, 3, 4, 5, 6} have described effects resulting from subjecting various tissues to a 448 kHz electromagnetic field at intensities which are not capable of heating such tissues, showing the occurrence of molecular phenomena other than those caused by the well-known classic thermal effects. Among these, proionic phenomena stand out.

This work involved proionic applications 72 hours prior to treatment with CO\textsubscript{2} laser (in order to prepare the tissue for optimal post-treatment response) and 24 hours after said treatment. In order to prevent contamination of the treated area, the application of an electromagnetic field with radiofrequency was performed through a layer of hydrogel.

This work was intended as a first approach for assessment of the proionic effects of 448 kHz radiofrequency in the reduction of edema caused by facial rejuvenation treatments with fractional CO\textsubscript{2} laser.

2. Materials and Methods

The experimental group included all volunteers who initiated panfacial skin rejuvenation treatment with fractional CO\textsubscript{2} laser between March 1 and April 30, 2014: \textit{n}=12. The control group included patients who had previously undergone...
panfacial skin rejuvenation treatment with fractional CO\textsubscript{2} laser, not subjected to proionic protocols but to conventional post-treatment, with dressing every 24 hours, consistent in the removal of slough with cold compresses immediately after laser for the next 12 hours, Aquaphor topical application every 4-6 hours for 4 days, Valacyclovir and Ciprofloxacin for 7 days and subsequent635nm wavelength Light Emitting Diode (LED) array application in pulsed mode protocol for 10 minutes, with 6-7 mj/cm\textsuperscript{2} fluence\cite{7}.n = 12. The Declaration of Helsinki for ethical principles in medical research with human subjects has been followed.

Inclusion criteria: 40 to 55 years old, Fitzpatrick phototypes I to III, no serious systemic conditions, no treatment within one month, and no smoking within one year. The protocol included: i) treatment session with fractional CO\textsubscript{2} laser (CO\textsubscript{2}RE\textsuperscript{®}, Candela Laser Corp., USA), and ii) pre-treatment and post-treatment sessions with 448 kHz radiofrequency (Elite, Indiba S.A., Spain).

Treatment session with fractional CO\textsubscript{2} laser: Two consecutive passes on all photoaged areas (around the eyes, around the mouth, cheeks, chin, space between the eyebrows, and nose area), except on the front area, where only one pass was performed (given the reduced thickness of the subcutaneous panniculus adiposus). Parameters: 150 microns spot, Deep mode, 80 mJ energy, 1.67 ms pulse duration, 9x9 mm pattern size, and 5\% coverage density. Mean duration: 50 minutes for panfacial coverage (500-700 cm\textsuperscript{2} area).

Pre-treatment and post-treatment sessions with 448 kHz radio frequency (Indiba\textsuperscript{®} Deep Care ELITE, Indiba S.A.): Proionic\textsuperscript{®} protocol. Duration: 30 minutes. Session 24 hours before: for preparation (no edema). Session 24 hours after treatment with fractional CO\textsubscript{2} laser: for treatment (at the time that edema is already settled - moderate edema is beneficial for obtaining clinical results\cite{8}– but before the peak of edema 48 hours after treatment). Parameters: 5-8\% of maximum power (10W to 20W, based on electrical impedance of the skin at the time of treatment), medium-sized resistive electrode, indirect contact (with intact skin in the pre-laser session or with laser-treated areas in the post-laser session): electrode in contact with the back of the hand of the physician, who rubbed his fingers softly on a thin layer of sterile hydrogel on the surface pre-treated with fractional CO\textsubscript{2} laser.

Edema was assessed immediately before and 24 hours after each non-thermal application of 448 kHz monopolar radiofrequency. We used a) a subjective satisfaction scale, and b) Color Doppler Echography with 18-22 MHz linear probe (MyLab\textsuperscript{®} Class C, Esaote SpA) for measurement of skin and subcutaneous panniculus adiposus thickness, because both layers are affected by the edema generated by the Laser treatment \cite{9}. Echographic measurements were taken in the right cheek of patients at the cross section of an imaginary line joining the corner of the mouth with the labium and a second imaginary line perpendicular to the first crossing the outer edge of the right eye.

All treatments were performed by the same physician, and all ultrasound assessments were made by the same specialist.

3. Results

Satisfaction score: 1 very unsatisfied, 2 unsatisfied, 3 indifferent, 4, satisfied, 5 very satisfied. Patients reported a mean satisfaction of 4.58 (SD = 0.67).

\begin{table}[h]
\centering
\begin{tabular}{|c|c|c|}
\hline
Patient & Score & Patient & Score \\
\hline
1 & 4 & 7 & 3 \\
2 & 5 & 8 & 5 \\
3 & 5 & 9 & 5 \\
4 & 5 & 10 & 4 \\
5 & 5 & 11 & 4 \\
6 & 5 & 12 & 5 \\
\hline
\end{tabular}
\caption{Satisfaction score.}
\end{table}

\begin{table}[h]
\centering
\begin{tabular}{|c|c|c|c|c|}
\hline
P & Pre RF (24h post-L) & Post RF (48h post-L) & 24h post-L & 48h post-L \\
\hline
1 & 8.2 & 7.4 & 10.8 & 11.9 \\
2 & 9.8 & 8.8 & 7.6 & 8.3 \\
3 & 10.4 & 9.4 & 11.0 & 12.2 \\
4 & 11.5 & 10.3 & 8.6 & 9.4 \\
5 & 10.1 & 9.0 & 10.9 & 12.0 \\
6 & 10.9 & 9.8 & 11.2 & 12.6 \\
7 & 7.8 & 7.1 & 10.0 & 10.9 \\
8 & 11.3 & 10.2 & 8.4 & 9.2 \\
9 & 11.4 & 10.4 & 10.2 & 11.1 \\
10 & 7.3 & 6.4 & 10.6 & 11.6 \\
11 & 8.9 & 8.1 & 11.7 & 13.2 \\
12 & 11.1 & 10.0 & 10.5 & 11.5 \\
\hline
\end{tabular}
\caption{Subcutaneous adipose pannicule (mm).}
\end{table}
Panniculus adiposus is due to the reduction of edema, which in present at these levels is correlated to a liquid element in the associated with inflammatory response to the protein application of RF. The improvement observed 24 hours after literature[10]. Post-laser edema increase was limited by the coagulation process induced by this type of laser. The this case occurs because of application of the Proionic reduction in the thickness of the dermis and the subcutaneous adipose tissue.

3. Discussion

The edema caused by fractional CO2 laser treatment is associated with inflammatory response to the protein coagulation process induced by this type of laser. The reduction in the thickness of the dermis and the subcutaneous panniculus adiposus is due to the reduction of edema, which in this case occurs because of application of the Proionic protocol. Reduction of the hypoechoic band which was present at these levels is correlated to a liquid element in the literature[10]. Post-laser edema increase was limited by the application of RF. The improvement observed 24 hours after application of the proionic protocol was evidenced as a reduction in the thickness of the dermis and the subcutaneous panniculus adiposus.

The thermal effects of facial rejuvenation treatments with fractional CO2 laser lead to an intradermal inflammatory process which, given the great number of capillaries present at this level, results in edema sufficiently severe to double the thickness of the dermis 48 hours post-treatment (3.33 mm) with respect to baseline thickness (1.65 mm).

The reduction in the Epidermis-Dermis (ED) thickness is statistically significant (p<0.0001) after treatment with the Proionic protocol. ED thickness (Table 3), which is 2.87 mm on average 24 hours post-treatment with fractional CO2 laser, is reduced to 2.02 mm on average 48 hours post-laser if a session with the Proionic protocol is applied and is increased to 3.33 mm on average 48 hours post-laser if this protocol is not applied.

The reduction of the Epidermis-Muscle Fascia (EF) thickness shows non-statistically significant clinical changes, which may be explained by a small sample size or by smaller edema of the subcutaneous panniculus adiposus compared to the dermis (in terms of percentages), which would mean a smaller quantity of flowing plasma on which to act with the Proionic effect. In fact, the ablation and coagulation effects of the fractional CO2 laser do not exceed 1.5 mm skin penetration, suggesting that the action is only intradermal. However, moderate thermal effects will be obtained at the level of the subcutaneous panniculus adiposus justifying a edematizing action of the fractional CO2 laser, smaller as a percentage than in the case of ED thickness, and therefore a smaller therapeutic target for non-thermal application of 448 kHz monopolar radiofrequency. Subcutaneous adipose pannicule thickness (Table 2), which is 9.9 mm on average 24 hours post-treatment with fractional CO2 laser, is reduced to 8.9 mm on average if a session with the Proionic protocol is applied and is increased to 11 mm on average if this protocol is not applied.

These results suggest that proionic effects help shorten patient recovery time by several days compared to the conventional post-fractional CO2 laser treatment protocol. These data should be contrasted by future studies which include a larger sample.

Table 3. Epidermis-Dermis (mm).

<table>
<thead>
<tr>
<th>P</th>
<th>Pre RF (24h post-L)</th>
<th>Post RF (48h post-L)</th>
<th>24h post-L</th>
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</tbody>
</table>

1. Subcutaneous adipose pannicule. Mean (SD).
2. Pre RF vs 24 Post L
Pre RF: 9.89 mm (1.49)
24h post L: 10.13 (1.26)
p=0.68.
2. Pre RF vs Post RF
Pre RF: 9.89 mm (1.49)
PostRF: 8.91 mm (1.37)
p=0.11.
3. 24 Post L vs 48 Post L
24h post L: 10.13 (1.26)
48h post L: 11.16 (1.48)
p=0.08.

2. Epidermis-Dermis. Mean (SD).
1. Pre RF vs Post L
Pre RF: 2.78 (0.51)
24h post L: 2.87 (0.65)
p=0.71.
2. Pre RF vs Post RF
Pre RF: 2.78 (0.51)
PostRF: 1.94 (0.31)
p<0.0001.
3. 24 Post L vs 48 Post L
24h post L: 2.87 (0.65)
48h post L: 3.33 (0.76)
p=0.13.

3. Discussion

The edema caused by fractional CO2 laser treatment is associated with inflammatory response to the protein coagulation process induced by this type of laser. The reduction in the thickness of the dermis and the subcutaneous panniculus adiposus is due to the reduction of edema, which in this case occurs because of application of the Proionic protocol. Reduction of the hypoechoic band which was present at these levels is correlated to a liquid element in the literature[10]. Post-laser edema increase was limited by the application of RF. The improvement observed 24 hours after

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