Case Report,

Effects of Cryofrequency at Dermal and Hypodermic Level: Case Study

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Abstract:

Background: Cryofrequency consists of the combination of cryotherapy and radiofrequency, acting simultaneously in the treatment of flaccidity and localized adiposity.

Objective: To investigate the effects of cryofrequency at the dermal and hypodermic levels.

Materials and methods: This is a case study, composed of one woman of 39-year-old, with interest to perform abdominoplasty surgery, presenting adiposity and flaccidity located in infra-umbilical region. Its treatment consists of eight cryofrequency applications.

Results: There was reduction of 7 cm on supra-umbilical plicometry and 6 cm on infra-umbilical; a decrease of 2.7 kg of body weight. The ultrasound data show a decrease of 0.6 mm, in the thickness of the adipose layer. In the histological analysis: thicker epidermis, with more layers and dermis with greater number of fibroblasts and inflammatory cells, greater quantity of neoformed collagen tissue.

Conclusion: The application of cryofrequency promoted reduction of adipose tissue and increased production of collagen.

Level of Evidence: Level III, case-control study.

Keywords: Cryofrequency, Adipose tissue, Heating, Cooling.

Introduction:

Localized adiposity is the accumulation of energy reserves in adipose tissue, determined by caloric intake above that required for body metabolism. Its etiology is due to several factors, such as: physical constitution, heredity, inadequate dressing and location of adrenergic receptors¹,². Generally, an association of localized adiposity with flaccidity is observed, with a decrease in fibroblast activity, associated with a disorganization of the extracellular matrix and reduction of collagen fibers. It can be characterized by a laxity of skin caused by various reasons such as lack of exercise, inadequate diet (low protein and high fat), pregnancy and mainly by increasing and reducing weight in sequence and rapidly³,⁴.

There are numerous therapeutic approaches, being invasive and non-invasive methods, with the purpose of correcting or mitigating these changes. The evolution of technology has enabled the appearance of numerous aesthetic resources; to treat the various changes among them is radiofrequency. The treatment of flaccidity with the use of radiofrequency has been considered a great advance in the aesthetic that allows the correction of signs of aging as wrinkles and
sagging skin. It can be used alone or in association with other procedures to improve sagging facial or body skin\cite{5, 6}.

Cryofrequency consists of the combination of cryotherapy and radiofrequency, simultaneously acting in the treatment of flaccidity and localized fat. Its technology features an ultra-cold tip that cools the skin to minus 10 degrees only in the epidermis (propagated by conduction, from the outside to the inside), while radiofrequency heat reaches the deeper layers of the skin stimulating the formation of new collagen fibers and elastin, increasing metabolism and fat utilization. This new technology produces several thermal shocks, which leads one to believe in the production of a possible instantaneous tension of the skin leading to results against flaccidity and fat in less time\cite{6}.

Cryofrequency consists of a new mode of radiofrequency that has in its applicator two technologies together in the same apparatus, the radiofrequency and the cryotherapy, with cold inducing terms of up to -10 degrees, bringing the issue of thermal shock to the tissue. In the epidermis, while radiofrequency heat reaches deeper layers and the energy released from this electrode leads to better circulatory and nutrient supply, besides tissue hydration, increased oxygenation, acceleration of elimination of catabolites, lipolysis, connective tissue contraction and together this causes contraction of the collagen fibers, simultaneously improving the alterations of the external architecture of the skin and bringing greater comfort to the patient during the application\cite{7, 8}. Therefore, this study aimed to investigate the effects of cryofrequency at the dermal and hypodermic levels.

**Materials and methods:**

This is a case study with a sample of 1 (one) woman who underwent abdominoplasty surgery. The 39-year-old physical activity practitioner had adiposity and flaccidity located in an infra-umbilical region, oriented with preserved local sensibility, and did not present circulatory changes. Its treatment consisted of eight cryofrequency applications, performed at Natal-RN, Brazil. The instruments used were: Physiotherapeutic Evaluation of Localized Adiposity Assessment (PAFAL)\cite{9}. An Ecopalm Wifi Ultrasound Scanner (10MHZ), which analyzes and measures adipose tissue, a Gran duos2 cell phone, Nest KT-311 tripod, Sanny brand adipometer that has a measuring range from 0 to 65 mm and a tape measure of the brand Fiber\cite{10}. Initially, the project was prepared and sent to the Brazilian platform and to the Ethics Committee (CEP) of Potiguar University, UnP, Natal-RN, Brazil, which was analyzed and approved with the present protocol number: 2.565.963. The patient was informed about the procedures performed and to sign the informed consent form (ICF), as well as the validated PAFAL protocol, both of which were attached, for general data collection, anthropometric data evaluation, perimetry using measuring tape to obtain the measure of circumference 12 cm infra-umbilical and 14 in the lateral line of the umbilical scar, the right side being the control side and the left side being treated. The plicometry was performed on the infra-umbilical skin fold three times, 2 cm above the supra-iliac and infra-iliac region, with a result based on the mean values obtained in the three measurements, and body weight assessment. After the initial evaluation, the photographic record was recorded in orthostatism and frontal and lateral view (right and left), and the volunteer with shoulders flexed at 90º. The camera used was placed on a tripod at a height of 70 cm from the ground and placed at a distance of 106 cm from the volunteer. Subsequently, the patient was submitted to an ultrasound examination by a medical specialist. The examination was performed in the infra-umbilical region, an area of 12 cm in height and 14 cm in width, according to the schematic drawing below:

The application of the cryofrequency was done using the apparatus Andrus criofrequênci\cite{10} -
ADOXY MEDICAL (São Paulo, Brazil), performed in dorsal decubitus and with the applicator positioned in the same place where the ultrasonography was performed. The parameters used were: 450 w of frequency, body bipolar mode in the left infra-umbilical region, in an area of 12 cm in height and 14 cm in width, during 08 minutes per area (protocol suggested by the company, there are no data records in the literature). The treatment consisted of an initial evaluation, 1 weekly session of application of the cryofrequency, totaling 8 sessions, after the 4th session, a new evaluation and after the 8th session the final evaluation. In all the evaluations PAFAL was used, besides the ultrasonography, photographic record, verification of body weight, perimetry and plicometry. After the sessions were completed, the patient underwent liposuction and abdominal dermolipectomy: Under epidural anesthesia with sedation. The infiltration with adrenaline solution 1: 3000.00 was performed on the superimposed technique. Aiming at conventional liposuction. Next, the dermolipectomy was performed, starting with the suprapubic incision; displacement in the supraaponeurotic plane up to the xiphoid appendix; performing abdominal muscle straightening; removal of excess skin from the lower abdomen, synthesis by planes with nylon wires and monocril. Maintained negative pressure drain (portovac 4.8). After removal of the skin flap, it was referred for histological analysis. The qualitative analysis was performed using histological microscopy. Hematoxylin and eosin (HE) staining 40x, 100x, 400x, and Masson trichrome 40x, 100x, 400x, and Verhoff 400x were used. To verify the morphology of the dermal, epidermal and hypodermic tissues. After the patient undergoes liposuction and abdominal dermolipectomy. Two tissue samples corresponding to the infra-umbilical area were extracted and used in the histological records.

**Results:**
During the period of application of cryofrequency until abdominoplasty surgery, the patient did not change her diet. Measurement variables before treatment and after 8 sessions are described in Table 1.

| Measurement evaluation | Before (cm) | After 4 sessions on cryofrequency (cm) | After 8 sessions on cryofrequency (cm) | Last (cm) |
|-------------------------|-------------|----------------------------------------|----------------------------------------|-----------|
| Supra-umbilical perimetry | 83          | 86                                     | 70                                     | 84        |
| Infra-umbilical perimetry | 93          | 96                                     | 90                                     | 95        |
| Supra-umbilical plicometry | 25          | 22                                     | 18                                     | 18        |
| Infra-umbilical plicometry | 22          | 22                                     | 20                                     | 16        |
| Weight                  | 67.4 kg     | 67.6 kg                                | 65 kg                                  | 64.7 kg   |
| BMI                     | 25.7 kg/m²  | 25.8 kg/m²                             | 24.8 kg/m²                             | 24.7 kg/m² |

It was verified that the supra-umbilical plicometry presented a reduction of 7 cm in infra-umbilical measurement and reduction of 6 cm in the supra-umbilical measurement. An oscillation of measurements in perimetry, taking into consideration hormonal factors (menstrual period). The mean values obtained in the initial and final evaluation were also presented. Regarding weight, 67.4 kg pre-treatment and 64.7 kg post-treatment were registered, with a decrease of 2,700 kg of body weight and a reduction of BMI from 25.7 to 24.7, to the ideal weight. Quantitative analysis was performed by comparing the treated side and the control side, measuring the adipose tissue, epidermis and collagen. The patient underwent ultrasonographic evaluation at the initial and final moments of the study. The ultrasound data recorded in Table 2 show a decrease when performing the second evaluation, in the thickness of the adipose layer on average of 0.6 mm. There was a reduction of the fat layer of the infra-abdominal region in relation to the values of plicometry, perimetry and ultrasonography. It was observed that at the end of all the treatment, the patient had positive results, confirmed in the table below:
Table 2: Ultrasonography data with measures of thickness of the fatty layer

| Measures                              | Initial Evaluation | Last Evaluation |
|---------------------------------------|--------------------|-----------------|
| Thickness of the adipose layer (Control side) | 12.1 mm            | 12.9 mm         |
| Thickness of the adipose layer (Treated side)  | 11.6 mm            | 11.0 mm         |

Figure 2: Histological sample. (A) Collagen tissue in the dermis and hypodermis side control (B) collagen tissue in the dermis and hypodermis treated side. (C) Adipose tissue control side (D) adipose tissue treated side.

Regarding the histological record, in figure 2A referring to the control side shows the appearance of disorganized fibroblast collagen in the hypodermis. On the treated side (figure 2B) it is possible to observe in more detail thicker epidermis, with more layers and dermis with greater number of fibroblasts and inflammatory cells and more amount of neoformed collagen tissue. Figure 2C, which refers to the control side comparably to Figure 2D referring to the treated side, the integrity of the areas adjacent to the treatment, can be visualized. In Figure 2D there is a decrease in the adipose tissue layer, and degenerating fat cells. Quantitative analysis was performed by comparing the treated side and the control side, measuring the adipose tissue, epidermis and collagen.

The Figure 3A shows the thickness of the epidermis in micrometers, showing in orange the treated side and in blue the control area, it is perceived that the control side presents a smaller thickness in relation to the treated side, which became thicker. It was observed that in the control group at time 1 the epidermis thickness was 34.51 μm and at the final moment it was 34.21 μm.

It was observed that in the group treated at time 1 the epidermis thickness was 75.69 μm and at the final moment it was 77.67 μm.

Figure 3B shows the thickness of the adipose tissue evaluated during the histological analysis, the measurement of this thickness when compared to the control on the treated side shows that the control has a greater thickness. It was observed that in the control group at time 1 the thickness of adipose tissue was 5465.67 μm and at the final moment it was 6040.55 μm. It was observed that in the group treated at time 1 the thickness of adipose tissue was 3645.86 μm and at the final moment was 2497.06 μm.

Figure 3C shows the amount of collagen neoformed in micrometers squared, where the neoformed collagen in the treated area presents in a larger amount in relation to the control that is presented smaller, thus reducing the amount of adipose tissue and increasing the thickness of the epidermis. It was observed that in the control group at time 1 the amount of collagen was 10662.17 μm² and at the final moment it was 23919.93 μm². It was observed that in the group treated at time 1 the amount of collagen was 29214.94 μm² and at the final moment it was 55390.44 μm².
Figure 3: Comparative graphical analysis of the epidermis on the treated side and control side. (A) Thickness of the epidermis; (B) Thickness of Adipose Tissue; (C) Neoformed collagen.

The photographic analysis consisted of 4 steps. Figure 4A was performed in the initial evaluation, Figure 4B in the evaluation after 4 sessions of cryofrequency, Figure 4C after the 8 sessions and Figure 4D in the final evaluation days before the abdominoplasty. It was possible to observe besides better visual result, the improvement of consistency and firmness of the skin.

Figure 4: (A) Photographic report of the first evaluation; (B) report of the second evaluation after 04 sessions of cryofrequency; (C) Photographic report the third evaluation after 08 sessions of cryofrequency; (D) Photographic report days before surgery, after the treatment.
Discussion:
In the present study, the patient did not change her diet and had a 2.7 kg decrease in her body weight throughout the treatment. The produced electromagnetic waves allow the penetration of the heat in the tissue in depth while the cooling is done in the surface for protection of the cutaneous fabric. Depending on the intensity of cold exposure, cell stress induces apoptosis, programmed cell death. Research has also shown that inflammatory stresses, as a consequence of thermal changes, cause the release of HSP60 in adipocytes in humans, achieving the release of pro-inflammatory substances and insulin resistance. They also show that the shock proteins are involved in the activation of the innate cells of the immune system, which results in the infiltration of macrophages into the adipose tissue by the release of Chemokines, such as MCP-1, which regulates the migration of phagocytic cells. Concluding in this study that the cryofrequency generates reduction of fat, not of weight. There are no studies that report and justify weight loss due to preoperative anxiety, but it is believed that this is the main reason for the patient's body weight decrease [8, 10]. The ultrasonography showed a decrease in the second evaluation, in the thickness of the adipose layer on average of 0.6 mm. In the result of the research, in addition to the visualization of the photos, better texture and firmness of the skin, the quantitative data of reduction of measures evaluated in the perimeter, plicometry and ultrasonography confirm the presence of these effects in the skin and adipose tissue. There is a need for a greater understanding of this mechanism of action. One possibility for explanation would lie in the ability to achieve higher temperatures and to obtain not only a comfortable skin flaccidity effect but also effects on adipose tissue through higher superficial and more comfortable superficial temperatures to the patient [7, 11]. It was possible to observe in the histological analysis thicker epidermis, with more layers and dermis with greater number of fibroblasts and inflammatory cells, besides a greater quantity of neoformed collagen tissue. Cryofrequency technology produces several thermal shocks, which suggests a possible instantaneous tensioning of the skin leading to results against flaccidity and fat in less time. Hypoderm is believed to absorb more energy than any other tissue by receiving elevated temperatures (approximately 60°) in a region of twice the size of the head. With the minimum time of 08 minutes lipolysis is expected and can be potentiated according to the amount of energy and time. Using the maximum time of 20 minutes for the same region, it is possible to achieve the effect of apoptosis, ie, cell death. The cryofrequency system, which reaches -10° C, does not prevent bipolar, multipolar and monopolar radiofrequency from being offered to the customer, besides ensuring that it does not burn when producing thermal shocks to the tissues, generating a third physiological effect, destabilizing the metabolism. Such energy mobilizes not only collagen but also fat, and is indicated for tissue flaccidity (facial and body) and localized fat. The combination of cooling and heat would cause a thermal shock generating an instant lifting effect, ie an immediate contraction of the skin with a lasting effect while eliminating localized fat [7, 11]. In the quantitative analysis by means of comparative graphs it presents the highest amount of neoformed collagen in the treated area, thus reducing the amount of adipose tissue and increasing the thickness of the epidermis, It is possible to affirm that the heat generated by the radiofrequency causes an inflammation in the tissue as a mechanism of response to this heating. In the first 24 hours neutrophils appear on the site, moving to the fibrin clot. The epidermis thickens at the edges as a result of the mitotic activity of the basal cells and, within 24 or 48 hours, epithelial cells are projected from the edges that migrate and grow along the incised margin of the dermis, depositing components of the basement membrane as they move. The deepest layer of the skin is affected by high energy and strong heat, while the surface remains cooled and protected, which causes the collagen to contract. Subsequently the formation of neocolagene is obtained, which will produce an improvement in the appearance of the skin [5]. Therefore, with the parameters of this study, it can be observed that the application of cryofrequency promoted reduction of adipose tissue and increased collagen production.

Conclusion:
Therefore, with the parameters of this study, it can be observed that the application of cryofrequency promoted reduction of adipose tissue and increased collagen production.
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