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Contrast lipocryolysis: pre and post session tempering improves clinical results.

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Abstract.

Alternative crystal structures are possible for all lipids and each different crystal structure is called a polymorphic form. Inter-conversion between polymorphisms would imply the possibility of leaning crystal formation towards the most effective polymorphism for adipocyte destruction. Food industry has been tempering lipids for decades. Tempering technology applied to lipocryolysis gave birth to “contrast lipocryolysis”, which involves pre and post lipocryolysis fat layer heating as part of a specific tempering protocol. In this study, we evaluated the skinfold thickness of 10 subjects after a single Contrast Lipocryolysis session and witnessed important and fast reductions.

Key words. Contrast, lipocryolysis, tempering, conditioning, adipocytes.

Introduction

The original works that gave birth to lipocryolysis claimed that localized-fat-reduction was the result of local apoptotic adipocyte destruction as a consequence of a heat extraction triggering stimulus.[1 2] Since then, this therapy has walked a long way. The empiric results witnessed and the absence of inflammation provided a "keyhole" where apoptosis was the only key that could fit-in.[3] Still, in every study we performed, we were able to identify non-apoptotic cell death up to some extent, normally in a very small proportion.[4 5 6] These findings opened our eyes to the fact that there were at least two processes coexisting beneath the action of lipocryolysis and that even not fully understood, both where susceptible of modifications in order to achieve better clinical results.

Lipocryolysis is a treatment that combines adipose tissue heat extraction with vacuum. It is a safe technology[7 8] that is effective for localized fat reduction.[9] Today, much more is known about every aspect of lipocryolysis as pioneer research is opening new gates to promising edges that will improve clinical outcome. One of these new technical developments is the second generation of lipocryolysis, also known as contrast lipocryolisis, which has already proved to be effective in *in vitro* adipocyte models.[3] Apoptotic adipocytolysis as a consequence of intracellular changes was the first and most logical action mechanism proposed for lipocryolysis.[1 8 9 10] It was assumed that adipocytolysis was the biological consequence of intracellular lipid crystallization. A number of

alternative crystal structures are a characteristic property of all lipids.[11] This is due to the fact that there are a number of different possibilities of packing the long hydrocarbon chain into a crystal lattice. This phenomenon is called polymorphism and each different crystal structure is called a polymorphic form of the lipid.[11] Iconographic evidence already backed-up the process of natural fat crystallization after lipocryolysis.[6] But the kinetics and thermodynamics that drive the formation, growth, stabilization, melting and destruction of lipid crystals are extremely complicated.[6] Inter-conversion between the typical three polymorphisms is an extremely appealing process because it implies the possibility of leaning crystal formation towards the most effective polymorphism for adipocyte destruction. If controlled, this should mean an exciting breakthrough towards clinical outcome improvement. Food industry has been tempering lipids for decades and for different reasons.[11] Lipid mixtures with two or three compounds –phases- cannot be compared to multiphase *in vivo* systems where the lipid variety is almost infinite and where biological limitations when heating or cooling living tissues play a major role.[4] Yet, triglyceride polymerization, crystal kinetic and thermodynamic principles remain the same.

Although the exact correlation between lipocryolysis, crystallization, gel-like behavior, apoptosis, necrosis and inflammation remains hidden,[4] different tempering sequences had already been tested[4] and, in comparison to conventional lipocryolysis, some of them proved to: a) destroy more adipocytes, b) crystallize an increased number of cells and c) produce larger crystal structures.[3] Tempering technology applied to lipocryolysis gave birth to the

"contrast lipocryolysis" technology. To evaluate the clinical results obtained after a single session of contrast lipocryolisis was the aim of this study.

Results

M1 mean skinfold was 3.79 cm (SD 0.78). M2 mean skinfold was 3.05 cm (SD 0.62) and M3 mean skinfold was 2.80 cm (SD 0.61). All values of M1, M2 and M3 are plotted in figure 2. Thicker panicles showed larger reductions in absolute numbers. The maximum fat layer reduction observed represented a 31% reduction of the original adipose panicle thickness and the minimum fat layer reduction observed represented a 23% reduction of the original adipose panicle thickness. The mean fat layer reduction for the whole sample was 26.6% (SD 2.72). The difference observed between the means of M1 and M3 was statistically significant ($p<0.01$).

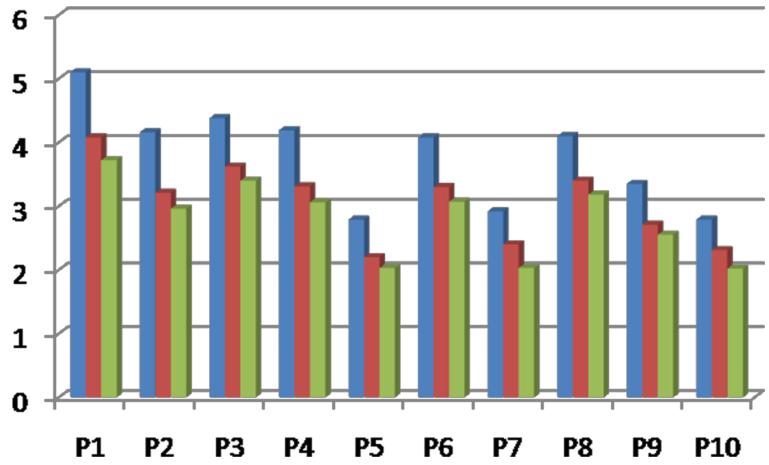

***Figure 1: skin fold**. M1, baseline measurement (blue); M2, 15 days post-session measurement (red); M3, 30 days post-session measurement (green); P: patient.*
Y axis: initial skinfold thickness (cm).

Discussion

1. Contrast lipocryolysis seems to be more effective than conventional lipocryolysis, though further evidence is needed.

It seems logical to assume that contrast lipocryolysis will be the natural evolution of conventional lipocryolysis. In a previous study we evaluated 16 women and we found a statistically significant skinfold reduction of 6,95 mm (SD 2.45) after a single conventional lipocryolisis session.[5] In the present study we observed a mean 9.9 mm reduction (SD 6,1) after a single contrast lipocryolysis session. When comparing both studies, the fat layer reduction achieved with contrast lipocryolysis represented a 42.45% improvement towards fat layer reduction observed with conventional lipocryolysis. Still, though both studies were methodologically very similar, comparable experiments using exactly the same conditions and evaluation days for both treatments should be conducted.

Another study conducted in *in vitro* adipocyte models showed that pre and post lipocryolysis temperature conditioning provided huge increments in adipocyte destruction and in crystal formation.[3] This study compared conventional lipocryolysis to 4 different tempering patterns and concluded that precondition for 5 minutes at 40ºC followed by 30 minutes at 8ºC and post condition at 38ºC for 10 minutes was the best tried tempering protocol. This remains the actual tempering protocol for contrast lipocryolysis, though further research, with larger amples and exhaust follow-up, should provide more data in order to evaluate and optimize other tempering protocols.

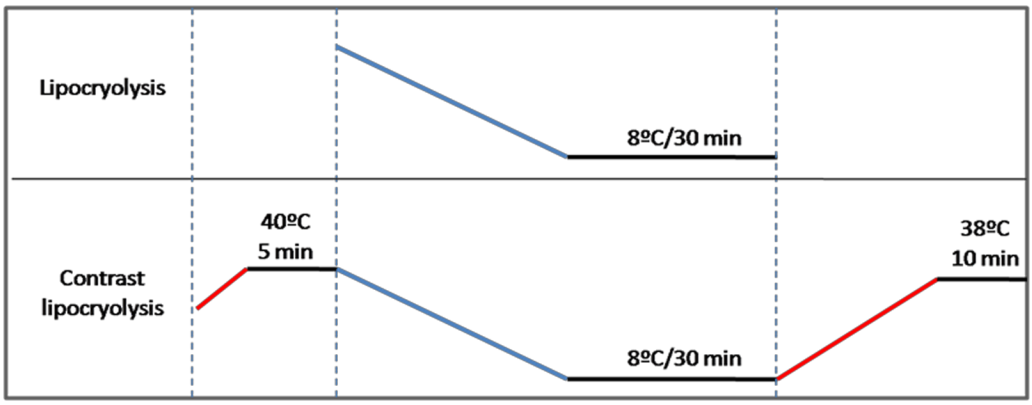

Figure 2. Contrast lipocryolysis and conventional lipocryolysis. *Heating (red line) speed is dual: when temperature is below 36ºC, blood flow restoration naturally enhances heating speed, resulting in a mean heating speed of 8.25ºC/min. When temperature is around 36ºC, blood flow plays a role against further heating, resulting in a mean heating speed of: 2ºC/min. Cooling (blue line) speed is 3ºC/min. Target temperature in adipocytes (black line): 40ºC during*

5 minutes for pre-conditioning, <10ºC during 30 minutes for conventional lipocryolysis and 38ºC during 10 minutes for post-conditioning. Whole contrast lipocryolysis procedure lasted 60 minutes.

2. Fat panicle thickness.

Contrary to intuition, we saw that thicker fat panicles reached the treatment temperature faster. Thicker fat panicles were easily cooled down than thinner ones, probably due to the tissue irrigation differences. This may result in an added challenge for contrast lipocryolysis machines, as fat layer thickness may determine individual protocols and affect session time. Comparable experiments with a larger number of subjects should be conducted, since this fact might be important for the clinical application of contrast lipocryolisis.

Material and methods

Sample consisted of 10 volunteer women recruited consecutively between November the 15th 2013 and December the 15th 2013, with a mean age of 48.1 (Standard deviation –SD- 9.73) years old. This study is in accordance with the standards set by the Helsinski Declaration of 1975. Inclusion criteria were: a) no systemic pathologies, b) not under chronic medication protocols, c) not pregnant nor breastfeeding, e) with no contraindications for lipocryolysis application, f) >2cm skinfold, g) Body Mass Index between 22 and 27. Between 30 days prior and 45 days after the session, patients did not follow any other treatment for localized fat reduction neither for body weight reduction. Each session was performed in the lower abdominal area by the same personnel. The application of "contrast lipocryolysis" was performed with Lipocontrast® (Clinipro, Sant Cugat del Vallés, Spain). Heat and cold extraction energy was fully and automatically deployed by Lipocontrast® throughout the whole procedure (figure 1), resembling the best tempering protocol according to the results obtained in previous studies.[3] Skinfold thickness was assessed with a plicometer Harpenden Skinfold Caliper® (Baty International, Burgess Hill, UK). The baseline plicometry measurement (M1) was taken immediately before the therapeutic session. The second (M2) and the third (M3) measurements were taken 15 and 30 days after the therapeutic session respectively.

Normal distribution assumption was verified with a Shapiro-Wilk test and homocedasticity assumption was verified with a Levene test. M1, M2 and M3 means were compared with a t-Student test. Statistical analysis was performed with SPSS version 17 for Windows (IBM Corporation, Armonk, NY, USA).

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Conflict of Interests

Dr. Hernán Pinto is an external medical advisor to Clinipro SL.

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