

Continuing Medical Education Examination— Body Contouring

Analysis of the Cutaneous and Systemic Effects of Endermologie® in the Porcine Model

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Learning Objectives:

The reader is presumed to have a broad understanding of plastic surgical procedures and concepts. After studying the article, the participant should be able to:

1. Understand the design of a study to examine the effect of Endermologie® treatments on the Yucatan mini-pig model.
2. Realize the histologic changes produced by Endermologie® treatments in this study animal whose cutaneous and subcutaneous tissues have similarities with human tissues.

Physicians may earn 1 hour of Category 1 CME credit by successfully completing the examination based on material covered in this article. The examination begins on page 421.

Endermologie® has been touted as an alternative to or adjunctive therapy with liposuction for the removal of subcutaneous fat, body contouring, and tissue toning. In this study we examined the effects of Endermologie® treatment with the Yucatan mini-pig model. Pigs were divided into three treatment groups (n = 4) and underwent 4, 10, or 20 treatment sessions, respectively. Blood and urine analysis was conducted during the study to obtain evidence of tissue trauma or breakdown and excretion of subcutaneous fatty tissue. Matched en bloc tissue sections from treated and untreated areas were examined for possible wound healing, inflammatory response, and tissue architecture changes. Examination of tissues revealed changes of architecture with accumulation of dense, longitudinal collagen bands in the mid and deep subdermal tissue layer; some distortion and disruption of adipocyte cell membranes were also observed. The degree of tissue change found was dependent on the number of treatment sessions performed. Endermologie® treatments did not elicit a classical wound healing or inflammatory response, with no evidence of either increased tissue vascularity or cell division. No evidence of mobilization of fatty tissue was found, and no fat breakdown products or metabolites were detectable in blood or urine. Treatments did not cause a decrease in subcutaneous tissue thickness. No evidence of skin or muscle injury was noted. Further

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Supported by grants from the Aesthetic Surgery Education and Research Foundation, The American Society for Aesthetic Plastic Surgery, and National Institutes of Health GM 40437 (LBN) and P30 AR41943 (LBN). The ES1 Therapeutic Massager® was loaned by LPG USA, Fort Lauderdale, FL.

Accepted for publication June 25, 1998.

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studies are warranted to determine whether the changes noted are short term or permanent.

Endermologie® (LPG Systems, Valence, France) is the use of a mechanical device that, when applied to the skin, lifts the skin by use of suction and massages it between rollers. The device is designed to automate the knead and roll technique of massage used by massage and physical therapists. Benefits from the use of Endermologie® reportedly include softening of burn scars (Costagliola M, unpublished data, 1995), improved recovery from muscle fatigue (Portero P, Canon F, Duforez F, unpublished data, 1996), and as a treatment for cellulite.^{1,2} Other effects of Endermologie® that have been claimed to date include skin toning, increased skin and interstitial blood and lymphatic flow,³ skin hyperoxygenation, improved cellular nutrition and elimination of waste products, and softening and restructuring of connective tissue. Physicians are now reporting the use of Endermologie® as an adjunct to or substitute for suction lipoplasty for the purposes of body contouring.^{4,5} However, there have been no reports of studies undertaken to scientifically evaluate the systemic and histologic effects of Endermologie®. This study was designed to document the reduction of fatty tissue in the subcutaneous layer of porcine skin, examine the dermis and superficial fascia for wound healing responses, and study the kinetics, urinary excretion of metabolites, and systemic responses after the use of Endermologie®.

Material and Methods

Endermologie Device

The Endermologie® device used in this study was the ES1 Therapeutic Massager® from LPG USA (Fort Lauderdale, FL). Endermologie® treatments were performed by a single operator who was trained in the use of the device by a representative of the manufacturer. The device was maintained in accordance with the recommendations of the manufacturer throughout the course of the study.

Animal Care

Twelve age-matched (4 months of age) Yucatan miniature swine (Harlan Sprague Dawley, Columbia, MO) were randomly assigned to three experimental groups, with four animals in each group ($n = 4$). During the course of the study the animals were housed and fed in accordance with the guidelines of the Animal Care

Committee of Vanderbilt University Medical Center, Nashville, TN.

Endermologie® Treatments

Subjects were administered Endermologie® treatments once or twice a week for a total of 4, 10, or 20 treatments. A series of standardized Endermologie® treatment maneuvers was performed during each session and was chosen, after consultation with the representative of LPG USA, to represent the current clinical practice of Endermologie®. The duration of each session was exactly 10 minutes. For each subject, one side was randomly chosen as the treatment side, with the opposite side untreated for the duration of the study and used as the control. Power settings were chosen for each maneuver at the onset of the study and were gradually increased as the study progressed, mimicking current clinical practice. During each session the subjects received light inhalational anesthesia with 1.0% to 1.5% isoflurane (Aerrane®; Fort Dodge Animal Health, Fort Dodge, IA) so that they were not flaccid but exhibited muscle tone and limb movement. Body weights were measured once weekly after treatment. Skin temperature was measured with an infrared temperature scanner (model DT-1001; Exergen Corp., Newton, MA), at the point midway between the posterior scapula and the hind limb trochanter on the treated side and on the midspine (untreated area) immediately before and after each treatment.

Laboratory Data

Baseline blood and urine laboratory values were obtained for each subject before the study began (Quest Diagnostic Laboratories, Nashville, TN) (Table). Blood was collected immediately after treatment once a week: in the subjects undergoing long-term treatment, blood was also collected once a week 12 to 20 hours after treatment for determination of creatine kinase levels. Blood was also obtained at the time of subject sacrifice and tissue fixation. Urine was obtained via a urine collection bag (Hollister pediatric U-Bag) 1 to 6 hours after treatment.

Tissue Fixation and Staining

At the time of sacrifice the subjects were anesthetized with ketamine 25 mg/kg (Phoenix Pharmaceutical, St. Joseph, MO) and acepromazine 0.7 mg/kg (Vedco, St. Joseph, MO) intramuscular injection and administered 2.5% isoflurane inhalational vapor. The right carotid artery and internal jugular vein were exposed and cannulated. After administration of heparin sulfate (125 units/kg intravenous), the subjects were perfused with normal saline solution (100 mL/kg) via the carotid artery

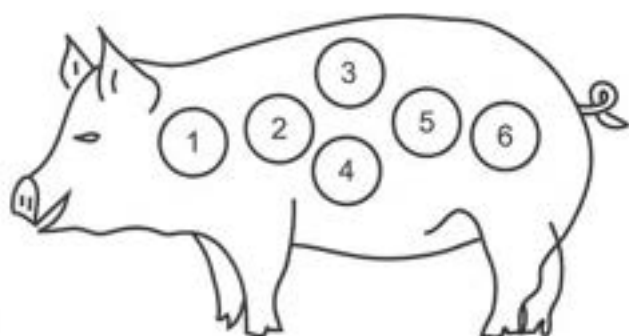


Figure 1. Sites of full-thickness tissue harvesting.

and allowed to drain via the jugular vein. After saline solution perfusion the subjects were perfused with 4% paraformaldehyde (125 mL/kg) and kept in cold storage for 24 hours to allow for in situ tissue fixation. At the end of 24 hours, full-thickness sections of tissue including skeletal muscle, measuring 10 cm in diameter, were excised en bloc from matched sites of treated and untreated areas (Figure 1). Tissue samples then underwent further fixation in 4% paraformaldehyde before sectioning. At the time of tissue sectioning for histologic and immunohistochemical study, two tissue specimens were cut from each of the six tissue blocs harvested from each side of the subjects (six treated blocs, six untreated blocs), providing 12 treated and 12 untreated (control) samples of tissue for each subject (A-L untreated, M-X treated). Paraffin-embedded sections were stained with Gomori's trichrome, Verhoeff-van Gieson elastic stain, and immunostaining with factor VIII to highlight vessels. Proliferating cell nuclear antigen (PCNA) immunostaining was performed to identify cell populations undergoing mitosis.

Quantitative Morphometric Analysis and Photography

Thickness of the subcutaneous fat-connective tissue layer positioned between the deep dermis to the muscle fascia was measured on a Zeiss Axioplan 2 microscope at original magnification $\times 1.25$ with Zeiss Image Pro-Plus Software. Colorimetric measurements of the percentage of collagen and the percentage of elastic fibers in the subcutaneous fat-connective tissue layer were assessed at original magnification $\times 2.5$ in the standard fashion, with the same Zeiss software. Digital photographs of tissues were captured by use of a Kontorn Elektronik® 3008 camera mounted on an Olympus® AH-2 light microscope; photographs were cropped and arranged by use of Adobe Photoshop® and Quark Express® software.

Table. Laboratory data: blood chemistry and urinalysis

Electrolytes
Liver functions
Blood urea nitrogen/creatinine
Protein/albumin
Creatine kinase
Amylase/lipase
Cholesterol and triglycerides
Urine specific gravity
Urine proteins
Urine glucose/ketones
Urine bilirubin/urobilinogen
Urine cells/bacteria
Urine crystals (including urate and cholesterol)

Results

During the study the subjects remained healthy, gaining weight at a steady pace throughout the course of their treatments, as anticipated from data on porcine growth supplied by Harlan Sprague Dawley. During the performance of each Endermologie® session, noticeable hyperemia occurred only within the skin receiving treatment. This hyperemia was transient and visibly resolved within 30 minutes after treatment. However, no consistent changes in skin temperature were recorded in the period immediately after treatment. Visual inspection during the course of the treatments and at the conclusion of treatments did not show any noticeable differences between treated and untreated regions of the subjects. Specifically, there were no changes to the skin or visual evidence of tissue toning consistently seen. However, as the weeks of treatment progressed, the person performing the treatments noted a subjective loosening or relaxation of the skin that became more noticeable during each session.

Laboratory Results

No abnormalities were detected in serum electrolytes, proteins, triglycerides, cholesterol, or complete blood count during the study. Creatine kinase, measured 12 to 18 hours after treatment, was never elevated. Consistent elevations of serum lactate dehydrogenase and intermittent elevations of other liver function test results (serum glutamic-oxaloacetic transaminase, serum glutamic-pyruvic transaminase) were detected in all subjects in all three treatment groups, but bilirubin levels were normal. Urinalysis revealed intermittently elevated urine specific

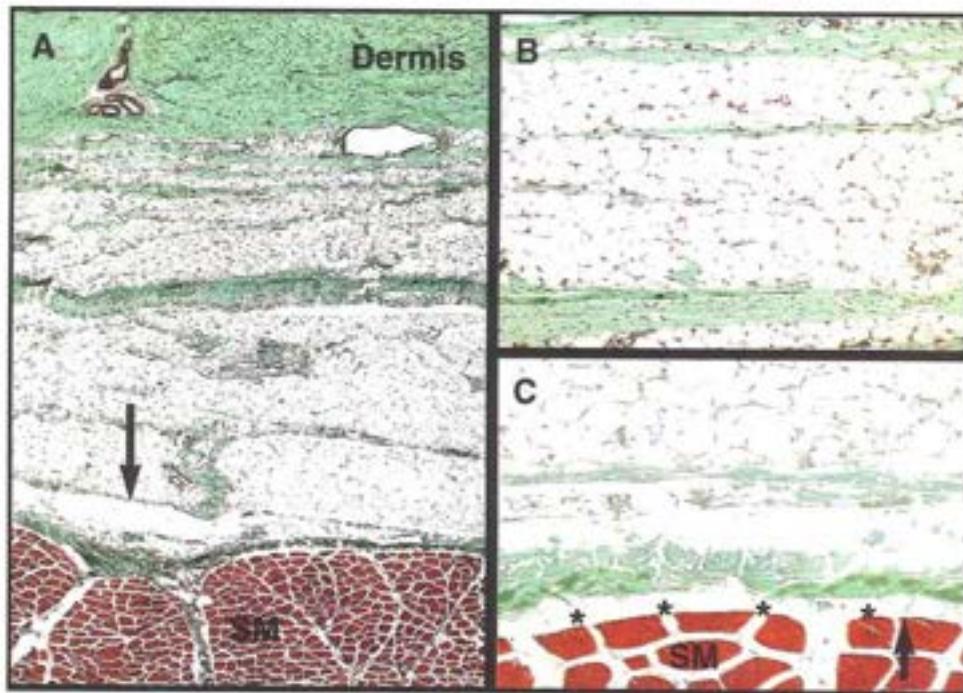


Figure 2. Photomicrographs of subcutaneous tissue from a subject undergoing 10 treatment sessions (intermediate group). **A**, Low-power view with intact vessels notable. Arrow points to tissue tear. **B**, Superficial subdermal layer with minimal adipocyte distortion. **C**, Adipocyte distortion and rupture. Torn fascia separated from skeletal muscle is notable (asterisks and arrow). SM, Skeletal muscle.

gravity and rarely detectable urine protein, with no occurrence of urine lipids, tissue breakdown products, crystals, globins, or blood.

Histologic Examination of Skin

Inspection of the skin revealed no evidence of trauma or injury, either grossly or microscopically. PCNA-stained tissues showed no increases in proliferation of epidermal, fibroblastic, or endothelial cell populations when compared with untreated skin. No inflammatory infiltrates were found in the dermis or epidermis of treated skin, and the skin architecture appeared unchanged in all subjects. No increase or decrease of skin vascularity was observed.

Histologic Examination of the Subcutaneous Tissue and Muscle

Inspection of the subdermal tissues revealed significant changes to tissue architecture. In the short-term (four treatments) group there were no noticeable changes. However, in the intermediate-term subjects (10 treatments), changes were seen primarily at the deepest subcutaneous level. At the region superficial to skeletal muscle and fascia, adipocyte cell membrane distortion and cell deformity were evident (Figure 2). The degree of cellular

distortion seemed to lessen at the more superficial subcutaneous layer. The appearance of increased collagen accumulation in the subdermal tissue was also more prominent in the deeper portion of the subcutaneous fatty layer. In the tissues from long-term subjects (20 treatments) these changes were much more pronounced. Adipocyte distortion and cell membrane rupture were common features seen in the deepest subdermal region and occurred less frequently at the superficial level. Dense longitudinal collagen bands were prominently displayed, especially in the mid to deep subdermal tissues. The fascia overlying the skeletal muscle was frequently separated off of the muscle and was markedly thickened. However, there were no obvious morphologic changes in or distortions to the muscle itself (Figure 3). Factor VIII immunostaining, which specifically highlights endothelial cells, did not show any evidence of neovascularity in the subcutaneous tissues. No proliferative fibroblasts were detected in the subcutaneous region by PCNA immunostaining.

Subcutaneous Tissue Thickness

Normally, regional variations in subcutaneous tissue thickness are present in the porcine model, with subcutaneous tissues closer to the cephalic end of the subject (neck,

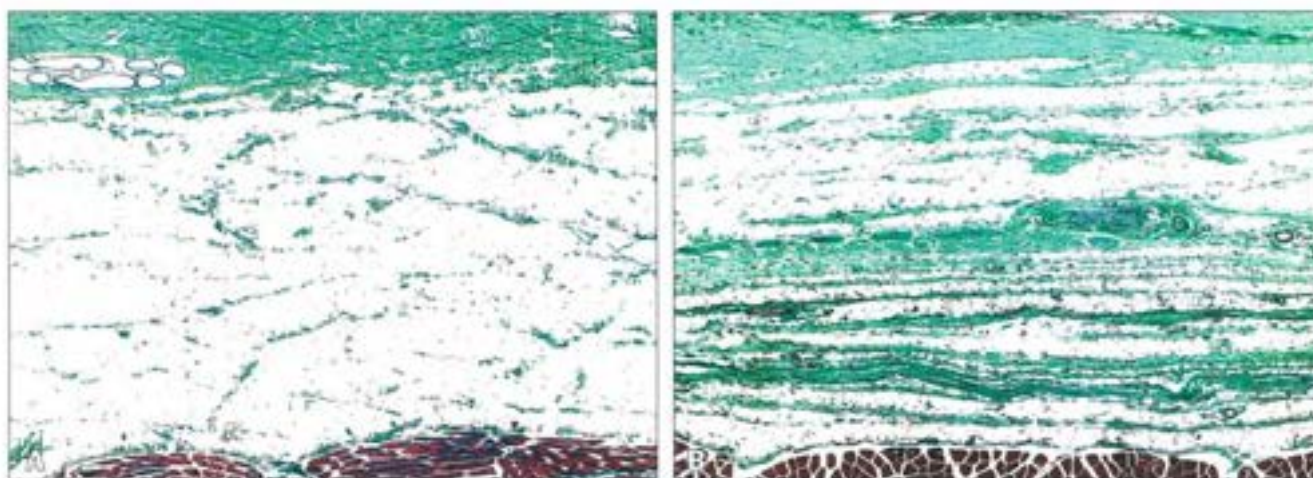


Figure 3. Photomicrographs of subcutaneous tissue from a subject undergoing 20 treatment sessions (long-term group). **A,** Untreated tissue. **B,** Treated area demonstrates dense longitudinal collagen banding, most prominently in the mid to deep subdermal regions.

shoulder) generally thicker than at the caudal end (posterior flank and hip). The same pattern of tissue thickness occurred in animals of all three treatment groups. No significant differences in thickness of the subcutaneous tissues were found when comparing site-matched nontreated with treated tissues in short-term, intermediate-term, or long-term treated subjects (Figure 4).

Subcutaneous Tissue Collagen and Elastin Content

Colorimetric analysis of subcutaneous tissues revealed significant increases in the percentage of tissue collagen content in treated tissues from both intermediate-term and long-term subjects (Figure 5). The collagen content of the subcutaneous tissue increased on average from 27% to as high as 130% in the long-term subjects. Lower average increases of tissue collagen were also measured in intermediate-term subjects, but short-term subjects showed no increase in tissue collagen content. No quantitative or qualitative changes in subcutaneous tissue elastin were seen.

Discussion

Since 1991 Endermologie® has been touted as a treatment for cellulite and more recently as an adjunct to liposuction. Efficacy of the treatments for cellulite reduction, tissue toning, and treatment of body contour irregularities has been proposed on the basis of the use of photogrammetry, which reportedly documents the claims made about the benefits of Endermologie®. However, none of the reports used experimental control subjects or explained the mechanism by which the treatments had an effect. There are several studies that seem to indicate the

efficacy of Endermologie® in the treatment of scars, and it has been used as a physical therapy adjunct in Europe for more than 10 years with apparent safety. However, until now, no study has been undertaken to examine the systemic metabolic responses and histologic changes that result from treatment with Endermologie®.

The young Yucatan mini-pig is an excellent model for use in this type of study, even though there is not an animal model that exactly duplicates human skin and subcutaneous tissue. As in human beings, the Yucatan mini-pig has a bilayered subcutaneous fat layer and lacks a panniculus carnosus. It shares metabolic and skin architectural and compositional similarities with human beings. Although the younger animals do not have an extremely thick deposition of subdermal fat, this layer does exhibit regional variations in thickness as it does in humans. The problems with fibrosis and calcification of cutaneous and subcutaneous tissues seen in older pigs are not encountered in younger animals. Treatment of one side of each subject and use of the other side as the untreated control subject provided a way to note any changes in tissue architecture or appearance resulting from Endermologie® treatments, even though the subjects were growing and gaining weight. It also allowed us to examine the effects of Endermologie® treatment on regions of various tissue pliability and thickness that occur in this model and are found in human subjects.

Other aspects in the treatment protocol were also chosen to mimic what would occur in human beings undergoing treatment with Endermologie®. The subjects were minimally sedated and could move during the treatments,

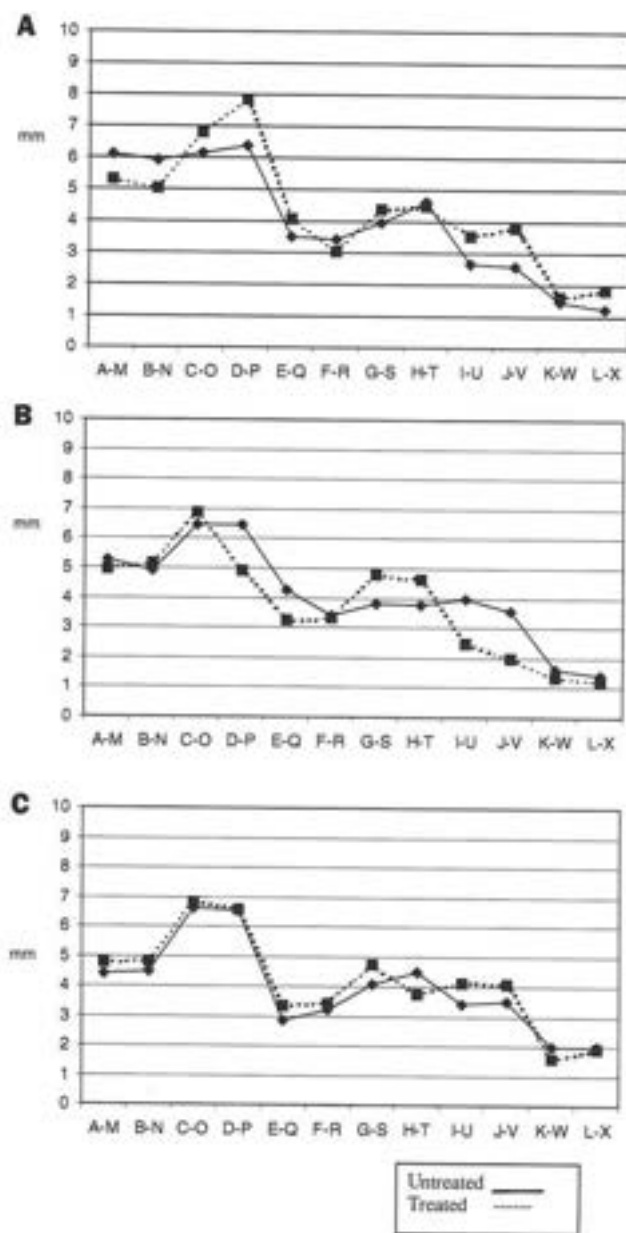


Figure 4. Measurement of subdermal tissue thickness in millimeters, comparing treated and untreated areas. **A**, Short-term group (four treatments). **B**, Intermediate-term group (10 treatments). **C**, Long-term group (20 treatments).

more closely approximating the muscle tone of awake human beings undergoing treatment. As has been observed in human beings, porcine subjects exhibit transient skin hyperemia with treatment, but this observation does not correspond with consistent increases in skin temperature. When skin temperature was measured, an area of untreated exposed skin was used as the control. The temperature was not taken on the control side because it was down against the table and would pre-

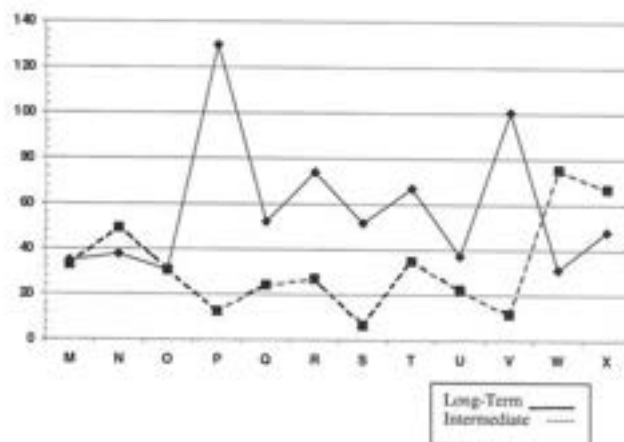


Figure 5. Percentage increase of subdermal tissue collagen content in treated tissues (M through X) of intermediate- and long-term subjects.

dictably become warmer because of contact against the table and lack of air flow over its surface. We postulate that this occurrence is due to the cooling effect of air as it flows into the powered treatment head over the skin surface. Transient hyperemia was the only skin change noted in our subjects. No subjective changes in the skin tone or body contour on the treated side were observed.

No biochemical evidence for fatty tissue breakdown, or mobilization of breakdown byproducts was found in serum or urine. Thus the biochemical data support our histologic and gross observations that fat is not removed from the tissues. The only significant abnormalities in blood chemistry were elevated liver function test results. These are commonly elevated transiently after anesthesia with halogenated inhalational agents. We believe that the intermittently elevated urine-specific gravity, the only urinalysis result that was consistently abnormal, was due to the timing of urine collection. Treatments were most routinely performed in the morning, before the subjects had been fed and watered, resulting in the collection of more concentrated urine. Again, no fatty tissue breakdown products or metabolites were detected in urine.

The lack of a classical wound healing response to Endermologie® treatment is significant. Initially, when evidence of trauma to the deep subdermal tissue was seen, it was believed that some tissue inflammatory or wound repair response would also be detectable. However, the response of the tissues to the trauma of treatments seems to be limited to alterations in subcutaneous tissue architecture; specifically, adipocytes are deformed or their cell membranes are occasionally dis-

rupted, primarily at the deep subdermal level. This occurrence at a deep level is consistent with the way in which an Endermologie® device exerts forces on the tissues. When the tissues are lifted by the suction applied through the powered treatment head, there is probably a sheering and lifting force that separates the tissues at the subcutaneous tissue-muscle fascia interface. The tissue separation occurs at this level where the relatively fixed skeletal muscle and the mobile subcutaneous tissues meet. The lateral forces applied to the tissue between the rollers should remain constant, dependent on the spring tension that allows the rollers to spread apart to accommodate tissues of varying thickness. The third force generated on the tissue is caused by any twisting action that occurs during the various maneuvers performed by the machine operator. Which of these three forces acting on the tissues is primarily responsible for the changes that we have described cannot be determined from this study. The net result is the accumulation of thick, longitudinal bands of collagen, again occurring primarily in the deep subdermal layer and running throughout the tissue. This banding is dependent on the number of treatments, being much more prominent in subjects that underwent 20 treatments than in those receiving only 10 treatments; it was not detected in subjects that underwent four treatments. We speculate that the smoothing effect and subjective appearance of decreased cellulite, which has been described in previous reports,^{1,2,5} is the result of these tissue architecture changes. Cellulite has been described as the unsightly appearance of skin, caused by tethering of skin and excessive fat to underlying fascia. The redistribution of the vertical tethering force vector, via collagen bands running parallel to the skin surface, coupled with tearing of vertical bands by the lifting action used during Endermologie® maneuvers, may account for the skin smoothing and the appearance of decreased cellulite in treated tissues. Whether these tissue changes are permanent or temporary remains uncertain. Further research is needed to ascertain whether architectural changes in tissue persist months after treatments have ended.

Conclusions

Our study with a porcine model demonstrates that Endermologie® treatment does not cause fatty tissues to be broken down, mobilized, and excreted. A standard regime of Endermologie® sessions does not precipitate an inflammatory or classical wound healing response. Increased cell proliferation, neovascularity, or injury to the dermis and epidermis does not occur. Nevertheless, changes in subdermal tissue architecture result from Endermologie® treatment, and these changes are proportional to the number of treatments performed. Although adipocyte injury occurs, there is no net decrease in subcutaneous tissue thickness. However, significant accumulation of collagen occurs mainly in the deep subdermal layer, primarily in the form of dense longitudinal collagen bands. These bands may be largely responsible for the smoothing effect to the skin that has been reported after Endermologie® treatments. The question of whether these changes are temporary or permanent must be addressed by future long-term studies. ■

We thank Ms. Nancy Cardwell, Ms. Mary McKissack, Mr. Jesse Britton, Professor Karem Jabbour, Mr. Phil Williams, Ms. Donna Schot, and Ms. Brittina Murphy for their invaluable technical assistance during the course of this study.

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Continuing Medical Education Examination—Body Contouring: Cutaneous and Systemic Effects of Endermologie® in the Porcine Model

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Multiple Choice

- The young Yucatan mini-pig model is ideal for this type of study for all of these reasons, *except*:
 - Their skin and subcutaneous tissue is identical to that of humans
 - They are metabolically similar to humans
 - They do not exhibit fibrosis or calcification of their tissues when traumatized
 - They exhibit variations of skin and subcutaneous tissue thickness
- Gross skin changes attributed to Endermologie® treatment in this study include:
 - Toning
 - Smoothing
 - Increased temperature after treatments
 - Hyperemia
 - All of the above
- Histologic skin changes noted in this study include:
 - Mild inflammation
 - Neovascularity
 - Increased skin thickness
 - Decreased skin thickness
 - None of the above
- The greatest histologic change attributable to Endermologie® treatment occurred:
 - At the dermal-epidermal junction
 - At the dermal-subcutaneous tissue junction
 - At the subcutaneous tissue-muscle fascia junction
 - Throughout the subcutaneous tissue
 - To arteries, veins, and lymphatics
- Histologic changes to subcutaneous tissue included:
 - Fascia separation, elevation, and thickening
 - Vessel wall thickening
 - Decreased subcutaneous tissue thickness
 - Neovascularity
- Changes to tissue attributed to Endermologie® treatment included:
 - Increased dermal and subdermal collagen content
 - Increased deep subdermal collagen content
 - Increased subdermal collagen and elastin content
 - Increased collagen and decreased elastin content
- Of the forces generated on the tissue by Endermologie® treatment, those that appeared to have the greatest effect and cause tissue changes are all of the following, *except*:
 - The machine suction setting
 - The spring tension of the compressing rollers
 - The various Endermologie® maneuvers
 - The downward pressure applied by the machine operator



This Continuing Medical Education activity is made possible by an educational grant from the Aesthetic Surgery Education and Research Foundation.

8. From this study it may be proposed that the effect of Endermologie® treatment on cellulite is:
- To break down cellulite fat, with absorption and excretion of fat in the stool and urine
 - The redirection of forces in the tissue from vertical to horizontal, resulting in a smoothed and toned appearance to the skin
 - An increase in fat metabolism and tissue oxygenation, with a resulting decrease in subcutaneous fat
 - The initiation of an inflammatory process that leads to fat breakdown and increased collagen/scar formation
9. This study proves that the effects of Endermologie® treatment:
- Are permanent (long-term) when intermittent follow-up treatments are continued
 - Appear to be safe in the short-term, but long-term effects and results are unknown
 - Are temporary (short-term) and disappear several months after the last treatment
 - Are unsafe and detrimental to the skin and subcutaneous tissues
10. Endermologie® treatment:
- Is designed to automate the knead and roll technique of massage
 - Has been proven as an effective and safe alternative to liposuction
 - Is a safe and effective method for weight reduction
 - Is equally effective for body contouring whether 10 or 20 treatments are performed
 - Causes visible tissue trauma with bruising and discomfort
11. Subcutaneous tissue thickness was unchanged by Endermologie® treatments.
T F
12. Increased collagen accumulation in the subcutaneous tissue was the most prominent histologic change that was noted, and this occurred throughout the subcutaneous tissue layer.
T F
13. Although no histologic changes were noted in the epidermis, the deep dermis showed increased mitotic figures and cellular distortion.
T F
14. Elevated liver function test results, elevated urine specific gravity, and abnormalities in urine and serum proteins were attributable to the effects of Endermologie® treatment.
T F
15. Histologic changes noted in this study were accompanied by weight loss in the subjects.
T F
16. Characteristics of the young Yucatan mini-pigs, shared with human beings and making them desirable for use in a study of this type, are the absence of a true panniculus carnosus and weight stability during the course of the study.
T F
17. This study showed no biochemical evidence for fatty tissue breakdown or mobilization of fat breakdown products.
T F
18. PCNA (proliferating cell nuclear antigen) and factor VIII immunostaining revealed no evidence of increased skin mitosis or neovascularity resulting from Endermologie® treatment.
T F
19. Trauma to the skin, evidenced by hyperemia, limited the length of each Endermologie® treatment to 10 minutes.
T F
20. Cellular distortion and cell membrane disruption were most prominent in the deepest subcutaneous tissue layer.
T F

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