The Significance of Trans-Epidermal Water Loss After Microneedling and Microneedling-Radiofrequency Procedures: Histological and IRB-Approved Safety Study

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Abstract

Background: Microneedling (MN) and microneedling-radiofrequency (MN-RF) result in skin rejuvenation and skin exposure to pathogens.

Objectives: The aim was to determine histopathological changes of needle-depth injuries in preauricular skin and measure time-dependent repair of transepidermal water loss (TEWL) in subjects.

Methods: MN and MN-RF procedures were performed at 0.5- and 1.5-mm needle depths on preauricular skin strips from a facelift patient. In 10 subjects, MN and MN-RF procedures were performed at 0.5-mm needle lengths on 6 marked opposing face and body sites. MN and MN-RF at 1.5-mm needle lengths were also carried out on each subject's midface skin lateral to the nasolabial fold. TEWL measurements were recorded with a calibrated DermaLab Cortex device (Hadsund, Denmark).

Results: Histological examination confirmed that the penetration depths of microchannels closely approximated the 0.5- or 1.5-mm needle lengths. In addition, MN-RF exhibited zones of coagulation injury at the distal end of the channel. After MN or MN-RF at 0.5-mm needle length, TEWL values were greatest immediately after needling to scalp, midface, neck, chest, arm, and thigh sites and remained slightly higher than baseline throughout the 48-h evaluation period. TEWL measurements after MN or MN-RF at 1.5-mm needle length resulted in the highest- and longest-lasting values throughout the 2-day observation period.

Conclusions: MN and MN-RF devices are novel devices that require further investigation into optimal treatment parameters and protocols, patient selection, and protection against intrusion of external pathogens and reactive cosmeceutical ingredients with barrier repair.

Level of Evidence: 2

Therapeutic

Healthcare professionals have integrated microneedling (MN) in their medical practices as valuable, versatile, and cost-effective skin procedures to treat a number of aesthetic facial and body concerns such as wrinkling, acne scars, striae formations, and alopecia. Recently, the technology of radiofrequency has been incorporated into microneedling devices (microneedling-radiofrequency; MN-RF) to create similar reversible microclefts for passive passage of therapeutic macromolecules from cosmeceuticals, drugs, and cell particles[1-3] and to intensify normal cascades of wound healing

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by amplified zones of coagulative radiofrequency injury that induce additional collagenesis and elastogenesis.[4,5] Although the medical community has recognized the value of needling, physicians are also concerned about the Food & Drug Administration's (FDA) recent recommendations for up-regulating safety and performance with MN and MN-RF devices because of reported adverse events such as granulomatous reactions, infections, and scarring.[6] Physician-knowledge of the onset and recovery period of a penetrated epidermal lipid barrier system would be advantageous for optimizing the passage of safe products such as growth factors and platelet rich plasma through the microchannels after needling. On the other hand, an intentional delayed application of cosmeceuticals, sunscreen blockers, and drugs on a restored barrier system may reduce the incidence of other complications such as product sensitivities and allergies. The anticipated therapeutic benefits derived from MN or MN-RF and intra- and post-treatment products must be balanced by the recovery of the structural and functional disruptions of lipid barriers not only to reestablish a homeostatic seal of the liquid–liquid barrier for proper regulation of our internal fluid system but also to retard undue permeation of toxic cytotoxic substances or pathogens from the external environment. To date, there have been little data to document the normalization of barrier function after the clinical use of MN or MN-RF. The purposes of this study were to: (1) correlate actual preauricular skin penetration and tissue injury in vivo by histology with the use of 2 needle lengths of 0.5 and 1.5 mm by MN and MN-RF devices; and (2) determine the onset, duration, and quantity of elevated transepidermal water loss (TEWL) values, as a measure of lipid-barrier permeability and its renewal, after MN and MN-RF procedures to commonly treated areas on the head, chest, and extremities.

METHODS

Devices

1. The microneedling device (3MD-Microneedling; FDA-cleared, Class I, DP Derm, Miami, FL) consisted of a reusable motor unit hand-piece designed to be attached to a sterile, disposable cartridge that housed a spring-loaded reciprocating piston with an array of 12 needles. Each needle was about 0.2 to 0.3 mm (200–300 μm) in diameter arranged in a fractional pattern design. When activated, the perpendicularly placed needles were power-driven to create thousands of microclefts into the dermis. For the TEWL study, the piston stroke speed frequency was selected at the nonworking end of the hand-piece at a Level 4 (110 Hz) that corresponded to an estimated 1300 micropunctures per second (10 oscillations/s/needle × 12 needles = 1296 punctures/s. Per IRB protocol design, the needle-penetration depth was adjusted on a

dial located at the top of hand-piece for the most commonly used depths at either 0.5 mm length or a 1.5 mm length and excluded the use of the other available needle lengths (0.25, 1.0, 1.5, 2.0, and 2.5 mm) on this device.

2. The microneedling-RF device (INTRAcel; FDA-cleared, Class II, Jeisys, Inc., South Korea) consisted of a disposable sterile tip with an array of 49 partially insulated gold-plated microneedle electrodes (maximal diameter of 200–300 μm) that protruded from the end that was attached to the motorized hand-piece. Insulation of the needle provided thermal protection to the epidermis and dermis except for the exposed 0.3 mm proximal to the tip. The needles were inserted into the skin by a specially designed electronically controlled, smooth motion motor. Per IRB protocol design, the investigator (GHS) selected comparable needle lengths at either 0.5 mm length or 1.5 mm length and disregarded the use of other available needle lengths at 0.8 and 2.0 mm on this device. When needles reached a predefined insertion depth, the RF was emitted with dermal heating while sparing the epidermis by real-time impedance matching. One oscillation was characterized by a needle insertion time of 0.02 s, RF emission time of 0.03 to 0.1 s, and needle extraction time of 0.02 s. The difference in electrical impedance between the epidermis (high impedance) and the dermis (low impedance) further increased selectivity forcing RF energy deeper into the dermis. The RF emission (60-W highest power; 10- to 5000-ms broadest pulse width) was delivered over the entire dermal portion of the noninsulated tip portion (0.3 mm) of each needle, allowing effective and controlled heating.[7]

3. The DermaLabSkinLab Combo Module with Cortex Technology (Hadsund, Denmark) is an established device to measure TEWL. The device was calibrated by the manufacturer with a Declaration of Conformity prior to initiation of the study. The Trans-Epidermal Water Loss probe measured water loss through designated spot of skin with 2 sets of sensors (temperature and humidity) mounted in its diffusion chamber with a diameter of 10 mm covering an area of 0.79 cm2. The water pressure measured in the open chamber was used to calculate the evaporated water over a constant skin area. The TEWL result was designated in $g/m^2/h$ as a function of time as the mean value over the last 5 s. The maximum obtainable value was 250 $g/m^2/h$. Measurement duration could be selected for different time lengths from 1 to 250 s.[8]

Protocol

TEWL Study

The pilot study was approved by an Institutional Review Board (Institute of Regenerative and Cellular Medicine,
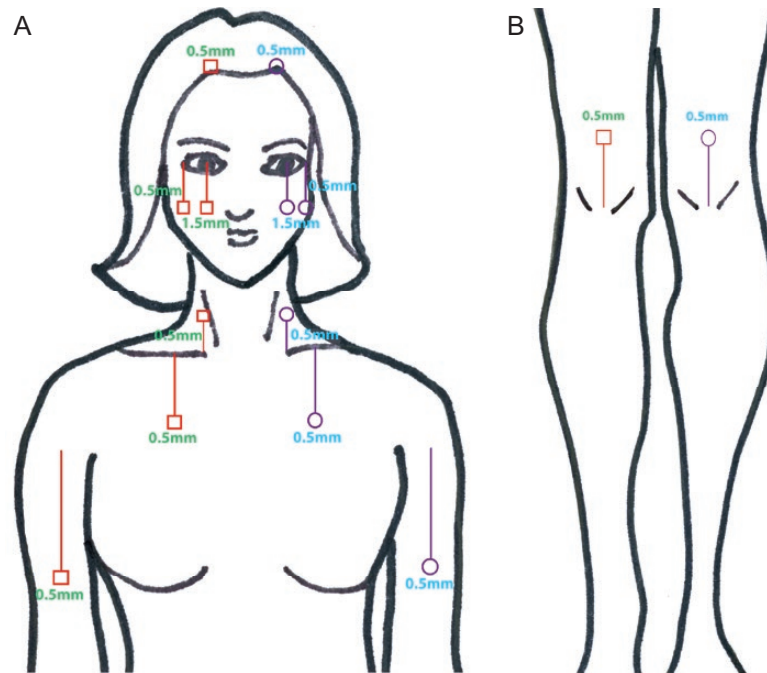
Figure 1. (A) Standardized markings for either MN or MN-RF treatments for the scalp, face, neck, chest, and arms. (B) Standardized markings for either MN or MN-RF treatments for the thighs.

May 2017; and conducted from May 2017 to October 2018). Of more than 25 evaluated subjects (21–55 years of age), only 10 candidates qualified for enrollment in this study who had never received any (1) surgical and nonsurgical procedures or (2) muscle relaxers, fillers, ablative, and nonablative treatments to the proposed microneedling sites within 6 months. Candidates also presented with no active systemic or local infections, acne, pregnancy, autoimmune diseases, or hemorrhagic disorders. After complying with inclusion/exclusion criteria, subjects were provided informed consent prior to participation. Subjects had no history of dermatological diseases and had not been treated with noninvasive or invasive skin procedures for over a year. Subjects were asked to refrain from application of any topical skin care formulations, vigorous physical activity, swimming, saunas, and intake of alcoholic beverages, caffeinated drinks over a period of 48 h prior to and after receiving a single treatment. Subjects did not receive compensation for participation and were not financially responsible for any of the validation studies.

Each subject was asked to draw blindly 1 strip of colored paper from a nontransparent jar that contained a mixture of 5 red and 5 black identical-sized strips of paper. The red strip indicated that MN would be the treatment to the right side, whereas a black strip indicated that MN-RF would be delivered to the right side. The opposing side in each subject received the alternative treatment. Each subject recorded TEWL measurements at baseline and within 5 min, and then at 1, 2, 3, 8, 24, and 48 h after single

treatment to 6 commonly treated sites, as shown in Figure 1 and Tables 1 and 2.

On the procedure day, subjects thoroughly cleansed the treatment sites with Cetaphil liquid soap to remove all cosmetics. Thereafter, subjects rested in a controlled environment study room for 30 min to acclimate their skin to the experimental ambient conditions (relative humidity, 35.1%; range, 29.8%–42.8%) (room temperature, 22.8°C; range, 22.7–24.2°C) before commencing MN and MN-RF treatments and data collection. All subjects received no oral pain or anti-inflammatory medications or topical analgesic gel applications during the entire study. The treatment areas were marked on the skin surface with a permanent marker to ensure that the instrument probe was repositioned in the same place at each evaluation session. The marked sites were gently cleansed with HIBICLENS (Chlorhexidine Gluconate 4.0% w/v) and washed off with sterile saline. The germicide was warmed to 39°C (range, 37–40°C) before skin application. Prior to each procedure, the MN- and MN-RF–motorized hand-pieces and connection sites for the cartridges were wiped down with CaviWipes (17.2% Isopropanol and Ammonium Chloride) to minimize cross-contamination from bacteria, fungi, protozoa, and viruses. Sterile gloves were used to insert each cartridge onto the device. All personnel in the operating room were required to use sterile gloves, operating room masks, and head cover for each procedure. Each subject's skin was stamped with a microneedling tip set at a 0.5-mm needle length (Level 4 at 90 Hz) on the designated 1-cm^2 spot marked on frontal scalp, lateral mid-face,
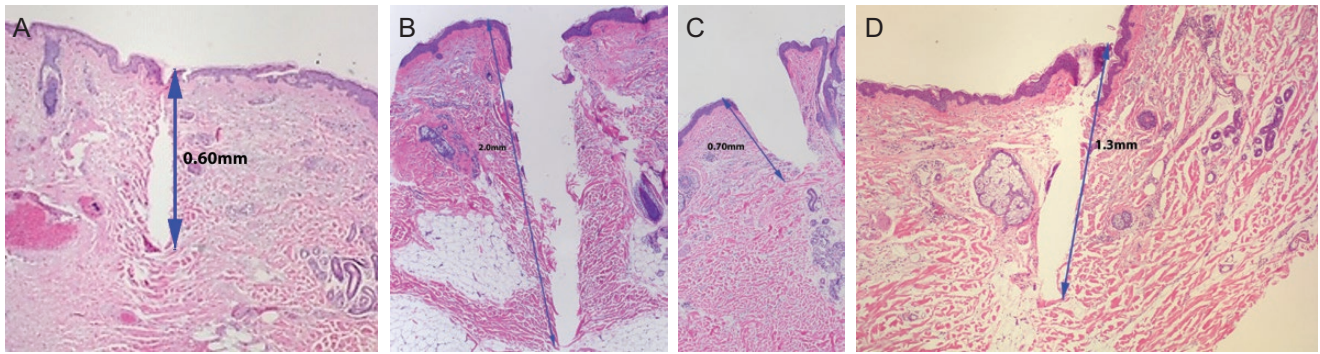
Figure 2. Preauricular excised skin histology samples (H&E, 40 × total magnification) at D_0 at needle lengths of (A) 0.5 mm and (B) 1.5 mm with the MN device. Preauricular excised skin histology samples (H&E, 40 × total magnification) at D_0 at needle lengths of (C) 0.5 and (D) 1.5 mm with the MN-RF device. Micrometer measurements demonstrated the penetration depth of each channel.

Table 1. Right Side (MN) Five Stamping Insertions/cm^2

0.5 mm	1.5 mm
Scalp	—
Midface	Midface
Neck	—
Chest	—
Arm	—
Thigh	—

Table 2. Left Side (MN-RF) Five Stamping Insertions/cm^2

0.5 mm	1.5 mm
Scalp	—
Midface	Midface
Neck	—
Chest	—
Arm	—
Thigh	—

mid-neck, chest, upper inner brachium, and upper inner thigh or stamped with a MN-RF tip set at a 0.5-mm needle length (1 MHz; total RF energy up to 78.4 J/cm^2; level 2 monopolar; 50 ms and 12 W) at mirror-image 1-cm2 treatment sites on the contralateral side. A grounding return pad was applied to the subject's skin while treating in a mono-polar RF mode. In a separate location on the medial aspect of the right or left mid-face, MN or MN-RF was additionally performed in a 1-cm^2 spot, utilizing identical parameter settings with the exception of the use of a 1.5-mm needle length. The investigator (GHS) inserted the array of microneedles in the MN and MN-RF cartridges 5 separate times with a consistent gentle force perpendicular to the skin to minimize surface distortion at all treated sites. Since a stamping technique was employed, the skin was not layered with a gliding gel. The subjects applied no sunscreen blocker, cosmeceuticals, moisturizers, or makeup to the treated sites for 48 h. Subjects were permitted to cleanse their face and bodies with Cetaphil soaps daily during showering and facial washing.

TEWL Evaluation
One technician (Margaret Gaston), experienced in the Cortex technology, performed all TEWL measurements at the same marked skin location at baseline and subsequent posttreatment intervals over 48 h. The technician was blinded to the type of treatment delivered and calculated the average of 5 TEWL measurements per site for statistical purposes. Then the mean averages of set of values from each site in 10 subjects were calculated for the final data points.

Depth-Penetration Validation Study
A 52-year-old healthy female provided informed written consent to treat her intact marked preauricular skin by either MN or MN-RF with 0.5- and 1.5-mm needling using identical parameter settings in the TEWL study in the morning prior to face/neck lift surgery. At the completion of her surgical procedure, each of the four 1- × 1-cm2 needled sites in the skin strip to be removed was excised and pinned to a tongue blade. Specimens were immersed in individually numbered vials containing a 10% formalin solution and transported to an out-sourced hospital pathology department for sectioning (5 μm), staining (Hematoxylin & Eosin), and interpretation. The pathologist was blinded to the type of treatment in order to remove bias in determining needle-penetration depths and tissue injury (Figure 2). The study protocol complied with the ethical guidelines of the 1975 Declaration of Helsinki. The patient enrolled in the investigation did not receive compensation for participation and was not financially responsible for any special studies in the validation research.

Table 3. Demographics for TEWL Study Subjects

Age, years		
Mean	37.7 ± 12.9	
Range	24–53	
Sex	*N*	(%)
Male	0	0%
Female	10	100%
Ethnicity		
Caucasian	3	30%
Hispanic	6	60%
Middle Eastern	1	10%
Fitzpatrick skin types		
II	3	30%
III	4	40%
IV	3	30%

Statistical Analyses

Five readings at individual-treated sites were averaged at each evaluation period over 48 h in each subject. Final values represent the mean average of readings from 10 subjects at each evaluation period and site. A Sensitivity Power Analysis was performed to the predetermined size of 10 subjects. The values used to calculate the analysis were an alpha of 0.05, power of 0.80, computing the mean difference between 2 independent means (2 groups). The analysis was conducted utilizing the G* Power 3.1.9.2 software (Universität Kiel, Germany).

RESULTS

The study enrolled 10 healthy adult female subjects (mean age, 37.7 ± 12.9 years; range, 24–53 years); 3 Caucasians, 6 Hispanics, 1 Middle Eastern; Fitzpatrick Skin Types II-IV), as listed in Table 3.

Each subject was asked to draw blindly 1 strip of colored paper from a nontransparent jar that contained a mixture of 5 red and 5 black identical-sized strips of paper. The red strip indicated that MN would be the treatment to the right side, whereas a black strip indicated that MN-RF would be delivered to the right side. The opposing side in each subject received the alternative treatment. Subjects 1, 2, 4, 5, and 8 were selected to receive MN to the right side and MN-RF to the left side, whereas subjects 3, 6, 7, 9, and 10 were treated with MN-RF to the right side and MN to the left side. In all 10 subjects, baseline TEWL measurements of skin in the scalp, face, neck, chest, upper arm, and thigh, which would be eventually treated with 0.5-mm needling, were measured and averaged between 5.2 ± 2.2 and 10.4 ± 4.5 g/m²/h. Each baseline value was interpreted to be representation of the normal values of Trans-Epidermal Water Loss at that site of intact skin (Table 4). Within 5 min of 0.5-mm MN, mean TEWL values increased higher from baseline levels in treated skin from the scalp (42.8 ± 26.3 g/m²/h) and face (27.2 ± 18.8 g/m²/h) than from treated skin of the neck (18.9 ± 10.0 g/m²h), chest (18.4 ± 12.4 g/m²/h), arm (15.2 ± 7.8 g/m²/h), and thigh (15.6 ± 9.3 g/m²/h), as shown in Table 4. Within 5 min of 0.5-mm MN-RF, mean TEWL measurements rose to higher levels from baseline values in the treated skin of the scalp (36.3 ± 19.7 g/m²/h) and face (29.2 ± 24.0 g/m²/h) than from treated skin of the neck (20.6 ± 14.4 g/m²/h), chest (16.4 ± 7.3 g/m²/h, arm 18.1 ± 10.8 g/m²/h), and thigh (12.1 ± 3.6 g/m²/h), as tabulated in Table 4. In contrast, the use of longer 1.5-mm needles resulted in significantly higher average mean TEWL values from baseline levels within 5 min after MN in midface skin (42.8 ± 15.8 g/m²/h), compared with the average TEWL values after MN with shorter 0.5-mm length needles to adjacent midface skin (27.7 ± 18.8 g/m²/h). Similarly, the use of longer 1.5-mm needles demonstrated significantly higher average mean TEWL values from baseline levels within 5 min after MN-RF in midface skin (39.6 ± 23.2 g/m²/h) compared with average TEWL values after MN-RF with shorter 0.5-mm needles in adjacent midface skin (29.2 ± 24.0 g/m²/h).

Generally, average mean TEWL values in the scalp, neck, chest, arms, and thighs returned towards baseline values 1 to 3 h after 0.5-mm MN or 0.5-mm MN-RF treatments but in a few individual cases remained slightly elevated at 24 to 48 h. TEWL values from facial skin after 1.5-mm MN or MN-RF treatments exhibited the slowest downward trend to baseline levels and still remained elevated in few individual cases up to 48 h. When 0.5-mm MN values or 0.5-mm MN-RF values were added together from 6 different treatment sites (excluding values from the 1.5-mm sites) and averaged at each evaluation period over 48 h, there was no statistically significant difference between them at each interval (Table 5).

H&E microscopy confirmed the actual maximum depths of tissue penetration at 0.5- and 1.5-mm needle settings and the histological reactions to injured skin. The 0.5- and 1.5-mm needle length settings with the MN device correlated closely with the measured depths from the epidermis down to the lowest level of the clefts within the dermis. The 0.5-mm needle penetrated to 0.6-mm tissue depth creating sharp borders along the microchannel (Figure 2A), whereas the 1.5-mm needle extended to a 2.0-mm tissue depth with clean borders along its microchannel (Figure 2B). The 0.5- and 1.5-mm needle length settings with the MN-RF device similarly penetrated to the 0.7- (Figure 2C) and 1.3-mm

Table 4. TEWL Values of 6 MN or MN-RF 0.5-mm Needled 6 Different Sites From 10 Subjects Over 48 Hours (data in the last column [Face 1.5 mm] represents TEWL values of MN or MN-RF 1.5-mm needled sites from the face in 10 subjects over 48 hours)

	Scalp 0.5 mm		Face 0.5 mm		Neck 0.5 mm		Chest 0.5 mm		Arm 0.5 mm		Thigh 0.5 mm		Face 1.5 mm	
	MN	MNRF	MN	MNRF	MN	MNRF	MN	MNRF	MN	MNRF	MN	MNRF	MN	MNRF
Baseline	9.2 ± 2.1	9.1 ± 1.7	9.9 ± 2.1	10.4 ± 4.5	8.6 ± 2.6	8.7 ± 3.4	6.1 ± 1.7	6.5 ± 2.8	7.5 ± 4.8	9.4 ± 8.8	5.4 ± 2.2	5.2 ± 2.2	17.6 ± 4.8	17.2 ± 5.4
Post 5 min	42.8 ± 26.3	36.3 ± 19.7	27.2 ± 18.8	29.2 ± 24.0	18.9 ± 10.0	20.6 ± 14.4	18.4 ± 12.4	16.4 ± 7.3	15.2 ± 7.8	18.1 ± 10.8	15.6 ± 9.3	12.1 ± 3.6	42.0 ± 15.8	39.6 ± 23.2
Post 1 h	15.8 ± 5.8	20.7 ± 18.9	17.9 ± 7.5	16.4 ± 7.9	14.7 ± 6.3	14.1 ± 4.7	12.4 ± 5.4	14.2 ± 7.0	12.2 ± 4.9	12.3 ± 4.4	13.2 ± 7.4	9.7 ± 2.7	30.9 ± 12.4	27.5 ± 12.1
Post 2 h	14.8 ± 8.4	13.5 ± 7.5	15.4 ± 5.8	15.1 ± 4.6	11.7 ± 4.8	12.8 ± 3.5	10.5 ± 3.9	11.9 ± 4.7	12.5 ± 4.7	13.1 ± 5.6	12.5 ± 7.8	10.9 ± 4.0	26.0 ± 10.9	21.7 ± 5.9
Post 3 h	11.8 ± 5.9	11.4 ± 3.8	15.7 ± 5.6	15.0 ± 6.2	12.4 ± 4.5	11.9 ± 3.2	10.2 ± 3.6	11.6 ± 3.8	12.5 ± 5.9	11.9 ± 7.1	12.1 ± 7.3	10.3 ± 3.3	24.9 ± 8.9	20.2 ± 4.8
Post 8 h	13.3 ± 5.5	9.8 ± 1.6	14.4 ± 5.3	12.4 ± 3.2	13.0 ± 6.6	10.9 ± 3.5	9.1 ± 2.4	9.3 ± 2.9	12.1 ± 4.7	12.2 ± 7.1	11.6 ± 6.9	10.2 ± 3.2	24.0 ± 9.0	19.8 ± 5.9
Post 24 h	11.1 ± 2.6	10.9 ± 2.6	12.5 ± 2.7	10.3 ± 2.5	10.2 ± 2.3	8.8 ± 2.8	7.6 ± 2.3	7.0 ± 2.6	9.0 ± 3.7	10.5 ± 7.0	8.6 ± 3.9	7.8 ± 2.0	20.6 ± 6.4	20.1 ± 6.7
Post 48 h	8.7 ± 2.4	10.1 ± 3.9	12.1 ± 3.3	10.9 ± 3.1	11.0 ± 5.2	8.5 ± 2.9	6.2 ± 1.3	5.8 ± 1.4	10.0 ± 5.3	9.7 ± 5.4	7.1 ± 2.3	5.8 ± 1.1	20.5 ± 8.0	19.2 ± 8.3

Table 5. Average Mean TEWL Values After 0.5-mm Needling With MN or MNRF at 6 Different Sites

	MN	MNRF
Baseline	9.2 ± 3.6	9.5 ± 3.4
Post 5 min	25.8 ± 12.2	24.6 ± 10.8
Post 1 h	16.7 ± 6.5	16.4 ± 6.1
Post 2 h	14.8 ± 5.2	14.2 ± 3.9
Post 3 h	14.2 ± 4.9	13.1 ± 3.6
Post 8 h	13.9 ± 4.7	12.1 ± 3.5
Post 24 h	11.4 ± 4.1	10.8 ± 4.0
Post 48 h	10.8 ± 4.3	10.0 ± 4.0

(Figure 2D) tissue depths with clean edges but with mild-to-moderate areas of coagulative necrosis at the bottom of the channels where the needle tips were noninsulated.

Complications

Microneedling and microneedling-RF produced transient minor erythema, swelling, and discomfort during and after treatment for a few hours. Subjects experienced no prolonged itching, burning, stinging, scaling, and dryness over the 6 months follow-up evaluation period. None of the 7 Fitzpatrick Skin Types III and IV developed hyperpigmentation at treated sites. No subjects reported any bacterial, fungal, or herpes zoster, herpes simplex, hepatitis B, hepatitis C, or human immunodeficiency infections.

DISCUSSION

One of the main functions of skin is to maintain a structural and functional barrier between the oftentimes hostile external environment and the internal ecosystem of the host. The skin's layers safeguard us from mechanical/chemical injuries, harmful pathogens, and ultraviolet light damage and enable human to exist in a terrestrial environment by providing a physical and physiological air–liquid barrier against water and electrolyte loss.[9] The permeability barrier is primarily confined to the outer layer of the stratum corneum layer and consists of nonviable corneocytes, cross-linking proteins, and a lipid-enriched bi-layered matrix.[10] After disruption of the permeability barrier, an immediate leakage of extracellular water and calcium occurs through the compromised stratum corneum that triggers lamellar bodies (0.2×0.3 µm) within keratinocytes in the stratum granulosum (SG) and stratum spinosum (SS) to secrete phospholipids, glucosylceramide, sphingomyelin, and cholesterol that eventually contribute to the final extracellular species of ceramides, cholesterol, and free fatty acids in the lipid matrix.[11] Previous publications[12,13] have reported that regional variations in quantity of lipid content exist in intact human skin. The greater amount of lipid content was directly correlated with the degree of barrier permeability.

A number of aesthetic procedures, such as chemical peels and ablation devices, intentionally penetrate, remove, or injure outer skin layers to achieve their epidermal and dermal therapeutic effects. Microneedling with or without radiofrequency represents another analogous procedure that temporarily breaches the epidermal permeability barrier by an array of needles and stimulates dermal wound healing. In MN-RF, insulated or noninsulated needles penetrate the skin either via a monopolar mode that utilizes active electrodes and a grounding pad or via a bipolar mode that employs active electrodes. In monopolar mode, an electrical circuit is formed by electron current flows by tissue resistance from the active electrodes to the grounding pad. In the bipolar mode, an electrical circuit

is released between the active paired electrodes targeting dermal structures with limiting the depth of thermal injury to preserve the epidermis.[14-16]

As a potential treatment for multiple skin conditions, that include rhytids, striae, and scars, future clinical studies will document the efficacy and recovery process after MN and MN-RF treatments. However, adverse events to providers and patients occur with either type of devices from a number of sources such as defective needle cartridges, technical failures of the devices, deficient charger bases, and potential previously contaminated sites on the device that can be transmitted to either the patient or the provider. Other untoward events can result from the use of bio-incompatible procedural and aftercare cosmeceutical and pharmaceutical topical products or from the invasion of bacteria, fungi, and viruses.[17-21] The FDA has drafted recently a nonbinding regulatory consideration for microneedling devices, based on currently available information on "risks with MN to include infection, nerve and blood vessel damage, disease transmission between users, scar formation, hyperpigmentation, skin inflammation, allergic reactions, and skin irritation."[22]

As practitioners continue to use MN and MN-RF devices and apply cosmeceuticals and sunscreens after treatments, adverse events can be expected to increase. From a safety standpoint, the FDA continues to focus on increased safety education to healthcare professionals, authorized clearance and marketing standards of devices, preventive measures to limit of noxious agents into needled skin from patient cross-contaminations, surface pathogens, allergens and impurities in cosmeceuticals and sunscreen formulations, as well as contaminated PRP that might be incurred during the harvesting, processing, and application phases. Such safety measures can be as simple as the use of a protective sleeve around the device and tips, backstops within the device to retard cross-contamination from backwash with biological fluid, and practice of universal safety precautions including proper disposal of tips and blood-related wastes. The results of our TEWL research underscored the benefits and potential drawbacks of resuming the early application of otherwise safe topical agents when the epidermal barrier system is compromised during the initial 24- to 48-h recovery period. The study emphasized the need for all healthcare professionals to understand and apply reasonable safety standards with the use of needling device, including, but not limited to, the implementation of Good Medical Practices for themselves and their patients. Providers should also be aware of appropriate application times with topical ingredients that promote the safety of their patients undergoing such repetitive treatments.

In this study, the validation of actual needle depths and zones of injury after MN or MN-RF was confirmed by histological examination from a single patient's preauricular intact skin specimens treated identically as the subjects in the TEWL study. The MN microchannels penetrated through the lipid barrier layers of the stratum corneum into the dermis at depths that approximated 0.5- or 1.5-mm needle lengths. The MN-RF clefts also reached dermal depths close to the 0.5- or 1.5-mm needle lengths but additionally exhibited coagulation injury at the distal end of the channel. Further studies will be needed to confirm whether MN-RF zones of coagulation would result in a delayed re-establishment of wound closure, retardation and prolongation of TEWL effects, and enhancement of collagenesis and elastogenesis over those observed by MN alone.

The TEWL pilot investigation in this study demonstrated that transepidermal water loss values were greatest immediately after injury and sloped downwards toward baseline values within 2 to 3 h after disruption by MN and MN-RF at a 0.5-mm needle depth to scalp, neck, chest, arm, and thigh sites. However, complete return to baseline values with either treatment type was not universally observed even at the end of the 48-h evaluation period. Of interest, scalp and midface sites after 0.5-mm MN or MN-RF levels, as did midface sites after 1.5-mm MN or MN-RF, registered the highest TEWL values within the first hour after treatment than observed at other sites. The reasons for this observation are unclear but may be supported by studies[12-13] that observed regional variations in lipid content in intact human skin (face > abdomen > leg > plantar stratum corneum) and were directly related to their barrier permeability. In addition, scalp skin is thicker and denser due to increased presence of adnexal structures, such as hair follicles, sweat glands, and sebaceous glands. It is unclear whether these dermal appendages impede the closure of created microchannels after needle-penetrations. On the other hand, the use of a longer needle length of 1.5 mm has been reported to prolong closure of microclefts by TEWL measurement when compared with the use of a shorter needle length of 0.5 mm. Thus, increased levels of TEWL measurements can be expected to occur either when longer needle lengths are used or when treatments are performed in denser and stiffer tissue (scars, scalp), thereby increasing exposure time to noxious external elements. Further studies will be needed to validate these assumptions and speculations.

This pilot investigation was limited to a small sample size of enrolled subjects and narrow selection of most commonly used needle lengths and energies of radiofrequency. Further studies are planned to define the effects of differing needle lengths and needle arrays, number of passes, RF energy impedances, variations of mono-polar and bi-polar delivery modes, as well as the effects of different available cosmeceutical ingredients on the safety and efficacy of MN and MN-RF devices in clinical practice.

CONCLUSION

Microneedling and microneedling-radiofrequency devices are novel devices that require further investigation into optimal treatment parameters and protocols, patient selection,

and timing and ingredients within after-care programs. The significance of barrier replenishment requires additional study to balance the benefits of mechanical–thermal injury and renewed epidermal protection against intrusion of external pathogens. Future TEWL trials will be needed to study differing needle depths, RF energies, and passes that are generally used in the clinical setting.

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