Clinical instrumental evaluation of the efficacy of a cosmetic product in improving skin hydration and skin complexion on hands

TORSTONE SA

RIVOLI

LES MAINS SOIN TOTAL ANTI-AGE

Reference code: lab-01160.13

Complife Italia S.r.l.
info@complifegroup.com
PEC complifeitalia@legalmail.it
complifegroup.com

Sede di Garbagnate Milanese (MI):
Via Guido Rossa, 1
20024 Garbagnate M.se (MI) Italy
+39.02.990.25138
+39.02.990.25007

Sede di S.Martino Siccomario (PV)
Via Monsignor Angelini, 21,
27028 S.Martino Siccomario (PV)
+39.0382.25504
+39.0382.536006

Sede legale: Via Guido Rossa, 1 20024 Garbagnate M.se (MI) Italy, | P.I./C.F. 11093320155
Capitale Sociale € 161.834 i.v., C.C.I.A.A. MI 1437929 - Registro Società N. 342215.

KEY PERSONNEL

Customer

TORSTONE SA
International Center Cointrin
Route de Pré-Bois, 20
Bât. C, 2è étage
CP1913
CH-1215 GENÈVE 15

Principal experimenter

Dr. **Enza CESTONE**
Degree in Medicine and Surgery, Specialist in Dermatology and Venereology
Consultant Complife Italia S.r.l.

Study director - Data analysis and report

Dr. **Eleonora SPARTA'**
Biologist
Complife Italia S.r.l.

Complife Italia S.r.l

Complife Italia S.r.l
Via Angelini, 21
27028 San Martino Siccomario (PV)
tel. +39-0382 25504 - fax +39-0382 536006
Mail: info@complifegroup.com

INDEX

STUDY DESIGN

1.1. Title

Clinical instrumental evaluation of the efficacy of a cosmetic product in improving skin hydration and skin complexion evenness on hands.

1.2. Aim of the study

The study is aimed to evaluate the efficacy of the tested product in improving skin hydration and skin complexion evenness (in terms of reducing the visibility of dark spots and improving skin uniformity) on hands.
In order to reach this goal, a clinical-instrumental study is carried out on 20 healthy female subjects, aged over 40 years old, with dry skin tendency and dark spots on hands. Product efficacy is evaluated after 28 days of its use by means of non-invasive bioengineering techniques able to quantify skin moisturizing index, skin pH and dark spot color. The instrumental analysis is then integrated with the clinical assessment carried out by the Dermatologist and by a self-assessment filled in by the enrolled volunteers.

1.3. Tested product

1.3.1. Information provided by the Customer

☒ Product name: **RIVOLI LES MAINS SOIN TOTAL ANTI-AGE Reference code: lab-01160.13**
☒ Way of use: apply on hands twice a day.
☒ The tested cosmetic products conform to REGULATION (EC) No 1223/2009 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 30 November 2009 on cosmetic products (recast) (Text with EEA relevance) and to its annexes.
☒ The tested cosmetic product was evaluated for its safety of use on human volunteers (safety assessment).
☒ INCI formula:

AQUA, ETHYL MACADAMIATE, BUTYROSPERMUM PARKII BUTTER, CAPRYLIC/CAPRIC TRIGLYCERIDE, PANTHENYL TRIACETATE, CETEARYL ALCOHOL, PROPANEDIOL, HYDROGENATED PHOSPHATIDYLCHOLINE, COCOGLYCERIDES, PENTYLENE GLYCOL, GLYCERIN, PASSIFLORA EDULIS SEED OIL, ETHYL LINOLEATE, TAPIOCA STARCH, PARFUM, CELLULOSE GUM, CETEARYL GLUCOSIDE, C20-22 ALKYL PHOSPHATE, HYDROGENATED COCO-GLYCERIDES, TOCOPHERYL ACETATE, OLEYL ALCOHOL, C20-22 ALCOHOLS, CAPRYLYL GLYCOL, ETHYLHEXYLGLYCERIN, XANTHAN GUM, PHENYLPROPANOL, SODIUM STEAROYL GLUTAMATE, SQUALANE, TAMARINDUS INDICA SEED GUM, SODIUM PHYTATE, TOCOPHEROL, ACETYL RHEUM RHAPONTICUM ROOT EXTRACT, CERAMIDE NP, SODIUM HYDROXIDE, MALIC ACID

1.4. Ethical requirements

The study is carried out in accordance with the following ethical requirements.

1. All the subjects participating in the study are healthy volunteers of at least 18 years old.
2. All of the subjects participating in the study are selected with the supervision of a dermatologist according to inclusion/not inclusion criteria (see § 1.5.1.1.-2.).
3. Volunteers participation in the study was totally free.
4. All of the subjects participating in the study are informed of the aim and the design of the study.
5. All of the subjects participating in the study are informed of the possible risk involved in the study execution.
6. All of the subjects participating in the study give their informed consent signed at the beginning of the study.
7. Before volunteer exposure to the tested product, all relevant safety information about the product itself and each ingredient were collected and evaluated.
8. All of the study procedures are carried out in compliance with the ethical principles for the medical research (Ethical Principles for Medical Research Involving Human Subjects, adopted by the 18th WMA General Assembly Helsinki, Finland, June 1964 and amendments.
9. All of the precautions are taken in consideration in order to avoid excessive skin reactions.
10. If any unexpected/adverse skin reaction occurs, medical investigating specialist evaluates the severity of the reaction (reporting it in the volunteer's data collecting sheet) and proceeds with appropriate therapy.

1.5. Subjects

1.5.1. Subjects selection

The subjects participating in the study were selected by a board certified Dermatologist from a panel of healthy female volunteers, in accordance with the following inclusion/non-inclusion criteria.

1.5.1.1. Inclusion criteria

- ☑ Healthy female subjects
- ☑ Aged over 40 years old
- ☑ Caucasian ethnicity
- ☑ Subjects with dry skin tendency and dark spots on hands
- ☑ Commitment to adhere strictly to the information reported in the study information form
- ☑ Commitment not to use for all the study length other topical products with an activity comparable to the products under study
- ☑ Commitment not to expose to the sun/solar lamps during the study period
- ☑ Subjects aware of the test procedure who have signed an informed consent form.

1.5.1.2. Non-inclusion criteria

- ✘ Subjects who do not fit the inclusion criteria
- ✘ Pregnant or nursing women
- ✘ Subjects that have shown allergies to cosmetic products, toiletries, sunscreens and/or topical drugs
- ✘ Subjects with skin pathologies in the test area
- ✘ Subjects under pharmacological treatment (both locally or systemically) that may interfere with the test execution
- ✘ Positive anamnesis for atopy (if this condition interferes with the test execution).

1.5.1.3. Study withdrawal

- ✘ Subjects not respecting the conditions described in the information form
- ✘ Occurrence of accidents, pathologies or conditions which could interfere with the study
- ✘ Subjects who no longer want to participate in the study.

1.6. Study procedure

The study is carried out as follow:

- **T0**: enrollment of 20 subjects according to inclusion/non-inclusion criteria. Instrumental and clinical analysis of the parameters under study (basal evaluation, T0);
- **SHORT TERM TEST**: instrumental evaluation of skin moisturizing 2 (T2h), 24 (T24h) and 48 (T48h) hours after the single product application + instrumental evaluation of skin pH soon after the first product application (15 minutes after its application the measurement of skin pH is performed, in order to verify that the product doesn't alter skin conditions).
  During the short term test the product is applied by the experimenter on volunteers forearms in order to standardize test conditions (two skin sites are identified and a fixed amount of product is applied to one area while the contralateral side remains untreated and acts as control). Moreover, during the short term test, volunteers are asked to avoid applying any product and to wash their forearms.
- **LONG TERM TEST**: instrumental analysis of skin moisturizing, dark spots colour and skin thickness (ridensifying effect) + clinical analysis of the colour of dark spots after 28 (T28) days of product use. Moreover, volunteers are asked to fill in a self-assessment questionnaire after the first product application and at the end of the study.
  During the long term test (from day 1 to day 28) volunteers apply the product at home, twice a day on hands, according to the instructions provided by the investigator.

1.7. Materials and methods

In the sections here below the materials and methods used in this study are reported. Clinical-instrumental evaluations will be performed at T0 (basal visit) and after 28 days (T28) of product use. All the study procedures are carried out under temperature and humidity controlled conditions (temperature 18-26°C and humidity 50±10%).

1.7.1. Skin moisturization – T0, T2h, T24h, T48h, T28 days

The measurement of the skin moisturization is based on the internationally recognized Corneometer® method. Corneometer® method is based on the dielectric constant of water. The probe shows changes of capacitance according to the moisture content of the measuring object. An electric scatter field penetrates the very first layers of the skin and determines the dielectricity. The used device is a Corneometer CM 825 (Courage+Khazaka, electronic GmbH).

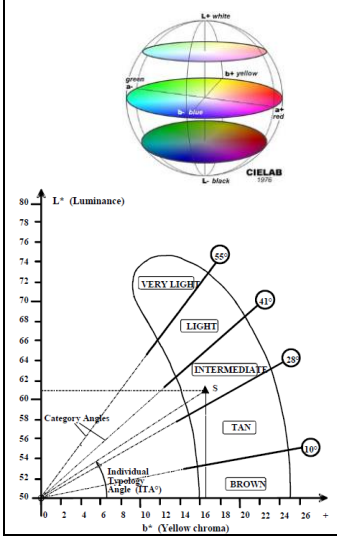
1.7.2. Skin pH evaluation – T0, T15min

The used instrument is the SKIN pH-METER 905®, Courage + Khazaka GmbH. The measure is based on a combined electrode of high quality, in which both the glass electrode sensitive to H+ and the additional reference electrode are placed in the same site. It is connected to a handle probe containing the measurement electronics. Before the measurements, the SKIN pH-meter® 905 (Courage + Khazaka electronic GmbH) is calibrated using two buffer solutions with known pH (pH 4.01 and 1.7) as reference.
Measurement range: 0 to 12; accuracy: ± 0.1 pH.

1.7.3. Reduction of the colour of dark spots, calculation of ITA° (Individual Typology Angle) – T0, T28

The intensity of melanin stain inside the dark spot is measured by means of a spectrophotometer/colorimeter CM-700D (Konica Minolta, Milan, Italy). L* and b* values, which characterize the dark spot, are taken. These data are then interpolated using a mathematical formula that allows to calculate the ITA° (Box. 1). A low ITA° value indicates a brown pigmentation, while a high ITA° value indicates a very light pigmentation.

BOX 1 – Assessment of dark spots colour: ITA angle



The technique. The spectrophotometer CM-700d measures the skin color in the CIELab chromatic space. CIELab is a standardized color space in which the color is defined - under standard illumination conditions (illuminant) and observer angle - by the values called a* and b* that define hue and color saturation and by the value called L* that defines the color brightness. The instrument, which principle of function is the reflectance spectrophotometry, emits an intense white light that re-emitted from the object (at an angle of 10°) is collected by 36 photodiodes each with different spectral sensitivity (from 400 nm to 700 nm). The sensitivity of the photodiodes is regulated according to a "standard observer" simulating the sensitivity of the human eye. This information is then elaborated by a microprocessor and displayed graphically.

ITA° calculation. ITA° is calculated as follows:

$$ITA° = \text{ArcTan}\frac{(L*-50)}{b*} x \frac{180}{3.14159}$$

Based on ITA° it is possible to define the following skin colour categories:

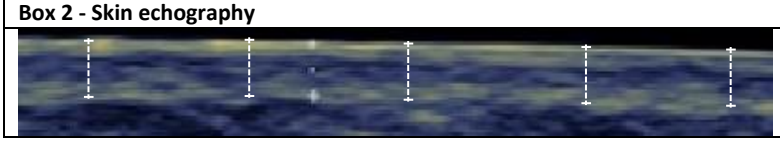
ITA°>55°	Very light
55°>ITA°>41°	Light
41°>ITA°>28°	Intermediate
28°>ITA°>10°	Tanned
ITA°<10°	Brown

COLIPA "Guideline for the colorimetric determination of skin colour typing and prediction of the minimal erythemal dose (MED) without UV exposure".

1.7.4. Evaluation of product "ridensifying" effect – T0, T28

The evaluation of the "ridensifying" effect is carried by means of Aloka alfa6 pro-sound (Hitachi) ultrasound machine. The ultrasound machine allows to measure the skin thickness (epidermidis + dermis). In the analysed skin region, skin thickness is measured in five points (white dotted lines). For further information see box 2.

Box 2 - Skin echography



The 5 white lines represent the 5 measurements of the epidermis + dermis layer (it is the area of the image between two yellowish hyperechogenic lines.

1.7.5. Clinical-dermatological evaluations: reduction of the colour of dark spots – T0, T28

By means of a clinical score scale (reported in box 3) the dermatologist evaluates the improvement of skin complexion uniformity (in terms of reduction of the colour of dark spots) by comparing before and after treatment pictures of the treated areas, acquired by means of a reflex digital camera. Pictures of the three best cases are reported in annex 1.

1.7.6. Self-assessment – T1st application, T28

After the first product application and at the end of the study volunteers are asked to express their personal opinion on product efficacy and properties by answering to a questionnaire.

Box 3 – colour of dark spots: variation vs T0	Score
No variation	**1**
Slight improvement	**2**
Moderate improvement	**3**
Remarkable improvement	**4**

1.8. Results and Statistics

1.8.1 Results

The Results are reported in their respective units in tables.

1) The mean values are calculated as:

$$m = \frac{\sum_{1}^{n} P}{n} \qquad [1]$$

where:

n is the number of subjects who ended the study

P is the value of the parameter to be analyzed.

2) The mean standard error is calculated as:

$$SEM = \frac{\sqrt{\frac{\sum_{1}^{n}(p_i^2) - \frac{\sum_{1}^{n} p_i^2}{n}}{(n-1)}}}{\sqrt{n}} \qquad [2]$$

3) The mean percentage variations were calculated as:

$$\overline{Var(\%)} = \sum_{1}^{n} \frac{P_T - P_0}{P_0} \qquad [3]$$

where

P_o is the value of the parameter to be analyzed at T0;

P_t is the value of the parameter to be analyzed at monitored experimental times.

All the calculations are done using a Microsoft® Excel 2013 (vers. 15.0.4885.1001; Microsoft, USA) worksheet running on Microsoft® Windows 8.1 Professional (Microsoft, USA).

The results of self-assessment questionnaire are calculated as percentage (%) of subjects who assigned a particular judgment (among those proposed). For each question, the number of subjects related to each judgment is counted → (number of subjects) and then divided by the total number of subjects → % of answers.

1.8.2. Statistical analysis

The instrumental data are submitted to 2-way Student's test t for paired data. Statistical analysis is carried out by means of a Microsoft® Excel 2013 (vers. 15.0.4885.1001; Microsoft, USA) worksheet running on Microsoft® Windows 8.1 Professional (Microsoft, USA). The variation is considered statistically significant when p value is <0.05. The statistical analysis foresees the comparison vs T0. Only the data of subjects who ended the study as protocol directed were considered in the statistical analysis.

1.8.3. Interpretation of results

The study here above reported was designed to demonstrate the test product claim(s) in the current framework proposed by Commission Regulation (EU) No 655/2013. Endpoints are measured using techniques currently accepted in the cosmetic field while biases are minimized by procedure(s) standardization according to ISO 9001 Quality Management System. Data are analyzed and interpreted by skilled technician according to both descriptive and inferential statistical analysis procedures. Due to the lack of reference values in the cosmetic field, statistical significance (for instrumental analysis) and percentage of subjects showing an effect (for clinical/sensorial endpoints) are the primary criterion to evaluate the correspondence between the proposed claim(s) and the study output(s). In particular Intragroup (vs. T0) or intergroup (eg. active vs. placebo, treated vs non treated) statistical analysis criterion to reject the null hypothesis (no product effect) is set at p<0.05. For clinical evaluations, the positive effect of the product on the measured parameter is confirmed if more than 50% of the subjects register an improvement. Finally, for the self-assessment questionnaires, the performance and the pleasantness of the product must be perceived by at least 60% of the subjects. Whenever reference values or threshold values exists that values are used to validate product claim(s).

1.9. Start/end date of study

The table here below reports date of beginning and end of the study.

Start date	End date
16/05/2019	05/07/2019

1.10. Report change record

Table here below reports the change log of all approved changes made to the document that make up the course after initial approval.

Rev. no	Date	Description
00	26/08/2019	Release of the first report
01	27/08/2019	Added product reference code

SUBJECTS DEMOGRAPHY

n.	Vol ID	AGE
01	F3656S	61
02	P2820M	68
03	C0085N	62
04	R1784M	59
05	B0012G	70
06	I0170M	68
07	D0668A	53
08	R0329O	66
09	T4004E	52
10	L4183A	67
11	S3831A	63
12	B0015R	64
13	M3265G	57
14	G0138O	65
15	M3661M	62
16	E3159L	56
17	A1462R	72
18	B0014L	65
19	B0526A	62
20	S4174L	56
	Media |Mean	62
	Min	52
	Max	72

RESULTS
SKIN MOISTURIZING – SHORT TERM TEST

TABLES 1a/b. The table below contains the data obtained for each subject taking part in the study for the parameter under study. Data are expressed as corneometric units (a.u.).

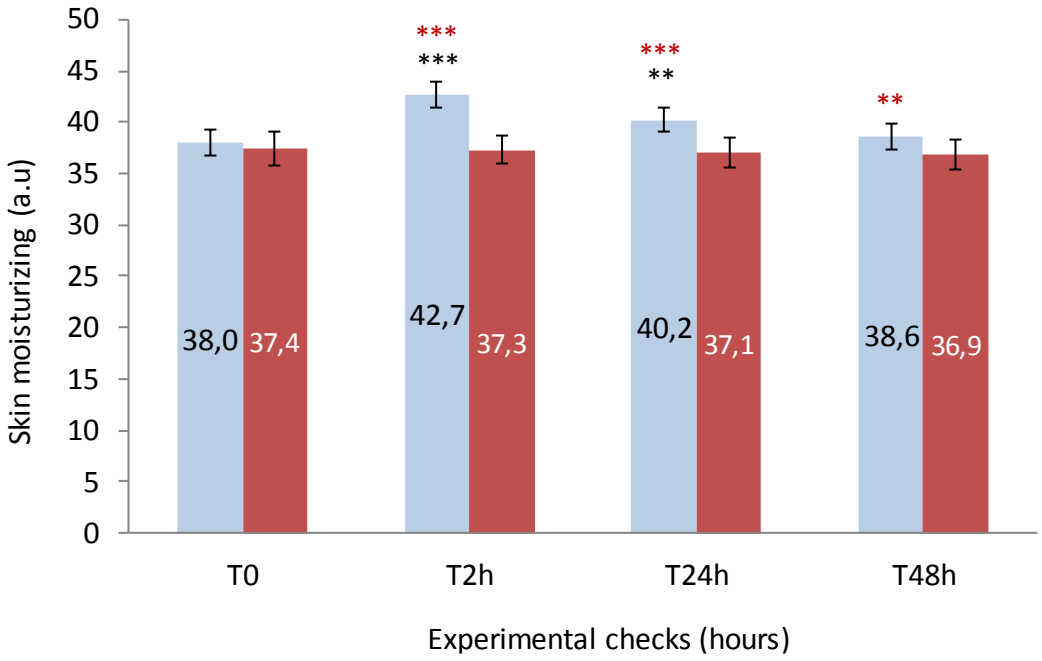
TREATED AREA

n.	Vol ID	T0	T2h	T24h	T48h	Variation % vs T0	T2h	T24h	T48h
01	F3656S	44,8	48,2	44,8	44,2		7,6%	0,0%	-1,3%
02	P2820M	33,5	36,1	34,5	34,5		7,8%	3,0%	3,0%
03	C0085N	40,0	47,5	42,6	42,3		18,8%	6,5%	5,7%
04	R1784M	41,8	43,7	45,3	44,7		4,5%	8,4%	6,9%
05	B0012G	41,3	49,6	48,2	42,4		20,1%	16,7%	2,7%
06	I0170M	37,4	40,9	38,9	37,5		9,4%	4,0%	0,3%
07	D0668A	38,2	43,5	43,1	38,6		13,9%	12,8%	1,0%
08	R0329O	40,2	46,2	40,7	40,5		14,9%	1,2%	0,7%
09	T4004E	33,6	40,5	33,9	33,7		20,5%	0,9%	0,3%
10	L4183A	32,1	39,9	39,6	35,4		24,3%	23,4%	10,3%
11	S3831A	32,2	36,2	32,2	32,2		12,4%	0,0%	0,0%
12	B0015R	43,4	46,4	43,0	42,9		6,9%	-0,9%	-1,2%
13	M3265G	36,5	40,6	41,3	40,7		11,2%	13,2%	11,5%
14	G0138O	54,6	55,6	51,6	54,0		1,8%	-5,5%	-1,1%
15	M3661M	38,3	44,8	43,1	37,6		17,0%	12,5%	-1,8%
16	E3159L	36,4	40,9	38,2	31,0		12,4%	4,9%	-14,8%
17	A1462R	38,5	43,0	38,6	38,4		11,7%	0,3%	-0,3%
18	B0014L	36,3	40,2	37,8	36,2		10,7%	4,1%	-0,3%
19	B0526A	31,6	33,8	33,2	31,9		7,0%	5,1%	0,9%
20	S4174L	28,8	35,9	32,8	34,1		24,7%	13,9%	18,4%
	Mean	38,0	42,7	40,2	38,6		12,9%	6,2%	2,1%
	SEM	1,3	1,2	1,2	1,2	Max	24,7%	23,4%	18,4%
	TEST.t vs T0	--	0,000	0,002	0,199	Min	1,8%	-5,5%	-14,8%
	TEST.t vs UT	0,365	0,000	0,000	0,002				

UNTREATED AREA

n.	Vol ID	T0	T2h	T24h	T48h	Variation % vs T0	T2h	T24h	T48h
01	F3656S	39,8	38,2	42,1	40,1		-4,0%	5,8%	0,8%
02	P2820M	30,3	33,1	30,0	30,6		9,2%	-1,0%	1,0%
03	C0085N	41,7	43,6	41,4	41,2		4,6%	-0,7%	-1,2%
04	R1784M	49,7	47,5	46,1	50,1		-4,4%	-7,2%	0,8%
05	B0012G	43,7	43,2	43,3	42,0		-1,1%	-0,9%	-3,9%
06	I0170M	36,0	36,2	34,7	35,6		0,6%	-3,6%	-1,1%
07	D0668A	35,2	36,5	38,1	36,7		3,7%	8,2%	4,3%
08	R0329O	37,1	38,6	37,8	37,1		4,0%	1,9%	0,0%
09	T4004E	32,4	33,8	32,8	32,6		4,3%	1,2%	0,6%
10	L4183A	29,0	31,7	30,9	31,7		9,3%	6,6%	9,3%
11	S3831A	32,3	33,1	30,2	30,1		2,5%	-6,5%	-6,8%
12	B0015R	41,7	42,9	41,3	40,8		2,9%	-1,0%	-2,2%
13	M3265G	35,9	34,2	38,2	37,7		-4,7%	6,4%	5,0%
14	G0138O	58,2	54,1	53,7	54,1		-7,0%	-7,7%	-7,0%
15	M3661M	37,0	35,7	38,2	36,4		-3,5%	3,2%	-1,6%
16	E3159L	32,5	32,2	30,2	30,6		-0,9%	-7,1%	-5,8%
17	A1462R	39,2	38,9	37,8	37,5		-0,8%	-3,6%	-4,3%
18	B0014L	37,1	36,4	35,9	33,8		-1,9%	-3,2%	-8,9%
19	B0526A	30,6	29,8	30,4	30,4		-2,6%	-0,7%	-0,7%
20	S4174L	27,8	26,6	28,6	29,1		-4,3%	2,9%	4,7%
	Mean	37,4	37,3	37,1	36,9		0,3%	-0,3%	-0,9%
	SEM	1,6	1,4	1,4	1,5	Max	9,3%	8,2%	9,3%
	TEST.t vs T0	--	0,910	0,536	0,249	Min	-7,0%	-7,7%	-8,9%

GRAPH 1. The graph sets out the mean data obtained at each experimental time for the parameter under study. Data are expressed as a mean ± SEM.



COMMENT: tested product application determines a mean statistically significant improvement of skin moisturizing 2 hours after its single application by +12.9%, that lasts up to 24 hours (+6.2%). No significant variations are monitored in untreated (control) area.
The variation recorded at T48h (+2.1%) is not statistically significant compared to baseline but is significant compared to untreated area.

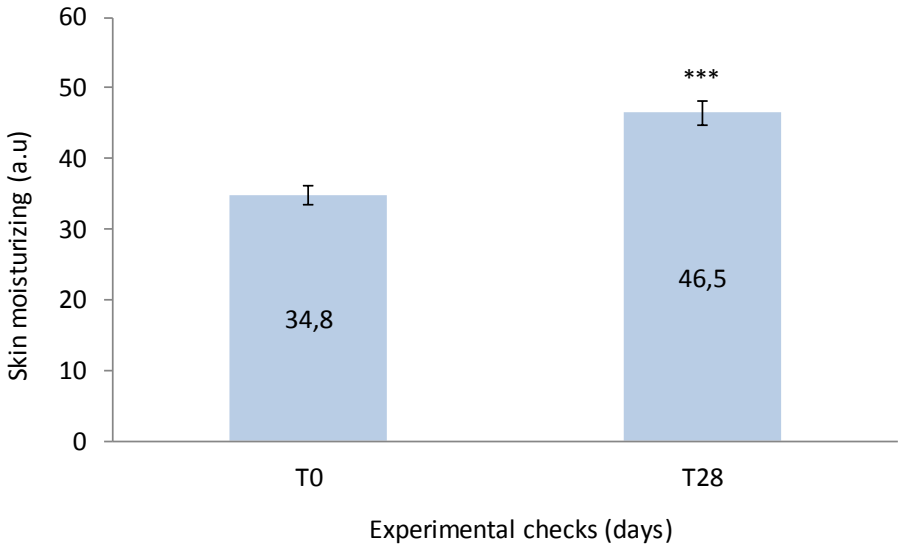
Note: the intra-group statistical analysis (vs. T0) is reported above the error bar in black colour. The inter-group statistical analysis (vs. untreated area) is reported above the error bar in red colour.
Legend: ** p<0.01; *** p<0.001.

SKIN MOISTURIZING – LONG TERM TEST

TABLE 2. The table below contains the data obtained for each subject taking part in the study for the parameter under study. Data are expressed as corneometric units (a.u.).

n.	Vol ID	T0	T28	Variation % vs T0 T28
01	F3656S	23,3	39,0	67,4%
02	P2820M	32,0	39,6	23,8%
03	C0085N	31,2	43,6	39,7%
04	R1784M	45,1	54,2	20,2%
05	B0012G	43,3	47,6	9,9%
06	I0170M	32,8	48,6	48,2%
07	D0668A	26,3	37,7	43,3%
08	R0329O	28,6	47,4	65,7%
09	T4004E	33,4	53,6	60,5%
10	L4183A	32,9	41,3	25,5%
11	S3831A	37,3	58,5	56,8%
12	B0015R	36,8	46,4	26,1%
13	M3265G	39,3	42,7	8,7%
14	G0138O	41,4	64,0	54,6%
15	M3661M	36,6	47,9	30,9%
16	E3159L	44,3	47,9	8,1%
17	A1462R	28,7	30,6	6,6%
18	B0014L	35,4	51,2	44,6%
19	B0526A	39,8	45,2	13,6%
20	S4174L	27,8	43,8	57,6%
	Mean	34,8	46,5	35,6%
	SEM	1,4	1,7	Max 67,4%
	TEST.t vs T0	--	0,000	Min 6,6%

GRAPH 2. The graph sets out the mean data obtained at each experimental time for the parameter under study. Data are expressed as a mean ± SEM.



COMMENT: after 28 days of bi-daily product use tested product determines a mean statistically significant improvement of skin moisturizing +35.6%.

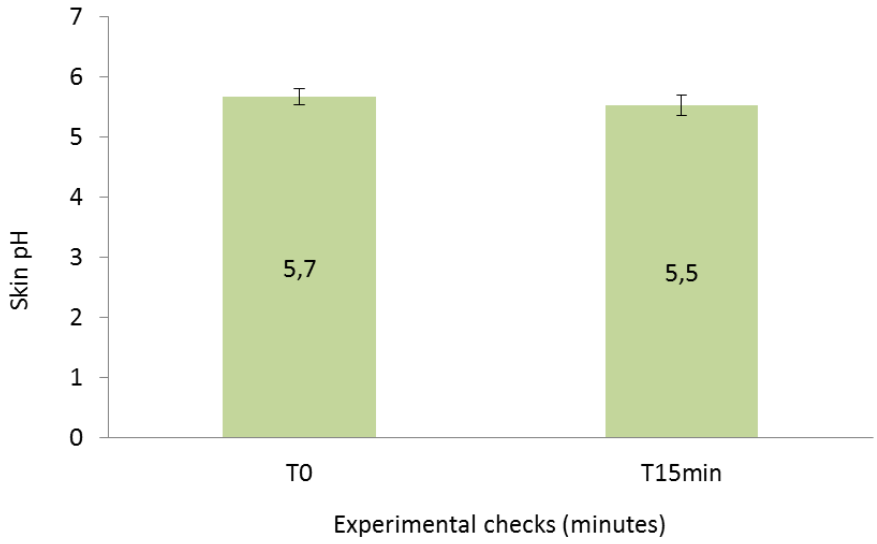
Note: the intra-group statistical analysis (vs. T0) is reported above the error bar in black colour.
Legend: *** p<0.001.

SKIN pH – SHORT TERM TEST

TABLE 3. The table below contains the data obtained for each subject taking part in the study for the parameter under study. Data are expressed as pH values.

n.	Vol ID	T0	T15min	Variation % vs T0 T15min
01	F3656S	5,8	5,2	-10,3%
02	P2820M	5,8	5,5	-5,2%
03	C0085N	6,0	5,5	-8,3%
04	R1784M	4,6	4,7	2,2%
05	B0012G	5,4	5,9	9,3%
06	I0170M	5,5	5,2	-5,5%
07	D0668A	6,7	6,4	-4,5%
08	R0329O	5,8	6,3	8,6%
09	T4004E	5,8	5,4	-6,9%
10	L4183A	6,1	5,4	-11,5%
11	S3831A	5,6	4,9	-12,5%
12	B0015R	6,0	5,5	-8,3%
13	M3265G	5,9	6,2	5,1%
14	G0138O	5,8	6,2	6,9%
15	M3661M	4,2	4,5	7,1%
16	E3159L	5,0	4,7	-6,0%
17	A1462R	5,3	4,9	-7,5%
18	B0014L	6,2	6,9	11,3%
19	B0526A	6,8	6,9	1,5%
20	S4174L	5,3	4,5	-15,1%
	Mean	5,7	5,5	-2,5%
	SEM	0,1	0,2	Max 11,3%
	TEST.t vs T0	--	0,178	Min -15,1%

GRAPH 3. The graph sets out the mean data obtained at each experimental time for the parameter under study. Data are expressed as a mean ± SEM.



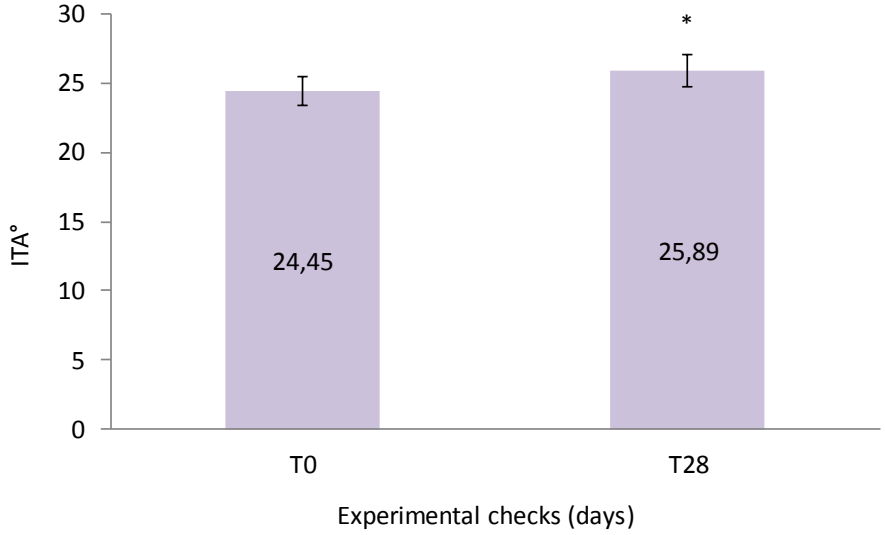
COMMENT: 15 minutes after its single application tested product doesn't alter the basal skin conditions; pH mean value is almost unvaried (a mean variation, not statistically significant, by -2.5% is recorded at T15min).

REDUCTION OF THE COLOUR OF DARK SPOTS, CALCULATION OF ITA° – LONG TERM TEST

TABLE 4. The table below contains the data obtained for each subject taking part in the study for the parameter under study. Data are expressed as degrees (°).

n.	Vol ID	T0	T28	Variation % vs T0	T28
01	F3656S	19,50	20,16		3,4%
02	P2820M	27,28	25,51		-6,5%
03	C0085N	18,35	18,28		-0,4%
04	R1784M	29,89	27,57		-7,8%
05	B0012G	24,04	26,77		11,3%
06	I0170M	31,28	31,59		1,0%
07	D0668A	19,77	25,39		28,4%
08	R0329O	22,29	21,15		-5,1%
09	T4004E	25,06	27,30		8,9%
10	L4183A	20,45	18,88		-7,6%
11	S3831A	35,08	40,17		14,5%
12	B0015R	21,74	25,88		19,0%
13	M3265G	22,45	29,87		33,1%
14	G0138O	28,73	28,54		-0,6%
15	M3661M	22,14	25,06		13,2%
16	E3159L	24,48	23,81		-2,7%
17	A1462R	21,69	26,17		20,7%
18	B0014L	21,30	22,62		6,2%
19	B0526A	31,13	33,13		6,4%
20	S4174L	22,26	20,06		-9,9%
	Mean	24,45	25,89		6,3%
	SEM	1,0	1,2	Max	33,1%
	TEST.t vs T0	--	0,034	Min	-9,9%

GRAPH 4. The graph sets out the mean data obtained at each experimental time for the parameter under study. Data are expressed as a mean ± SEM.



COMMENT: after 28 days of bi-daily product use tested product determines a mean statistically significant improvement of ITA angle by +6.3%. Higher the value of ITA° is, lighter skin pigmentation is.

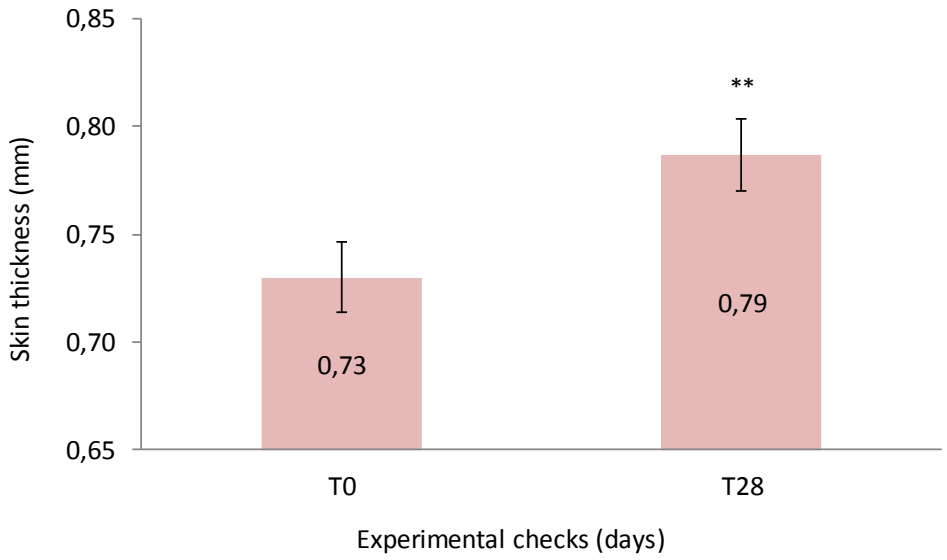
Note: the intra-group statistical analysis (vs. T0) is reported above the error bar in black colour.
Legend: * p<0.05.

PRODUCT "RIDENSIFYING" EFFECT – LONG TERM TEST

TABLE 5. The table below contains the data obtained for each subject taking part in the study for the parameter under study. Data are expressed as millimeters (mm).

n.	Vol ID	T0	T28	Variation % vs T0	T28	Variation (mm) vs T0	T28
01	F3656S	0,76	0,94		23,7%		0,18
02	P2820M	0,66	0,72		9,1%		0,06
03	C0085N	0,74	0,78		5,4%		0,04
04	R1784M	0,60	0,70		16,7%		0,10
05	B0012G	0,66	0,72		9,1%		0,06
06	I0170M	0,78	0,82		5,1%		0,04
07	D0668A	0,74	0,74		0,0%		0,00
08	R0329O	0,74	0,76		2,7%		0,02
09	T4004E	0,64	0,72		12,5%		0,08
10	L4183A	0,72	0,80		11,1%		0,08
11	S3831A	0,82	0,80		-2,4%		-0,02
12	B0015R	0,82	0,76		-7,3%		-0,06
13	M3265G	0,82	0,76		-7,3%		-0,06
14	G0138O	0,60	0,70		16,7%		0,10
15	M3661M	0,78	0,94		20,5%		0,16
16	E3159L	0,84	0,84		0,0%		0,00
17	A1462R	0,70	0,82		17,1%		0,12
18	B0014L	0,68	0,74		8,8%		0,06
19	B0526A	0,72	0,76		5,6%		0,04
20	S4174L	0,78	0,92		17,9%		0,14
	Mean	0,73	0,79		8,2%		0,06
	SEM	0,0	0,0	Max	23,7%	Max	0,18
	TEST.t vs T0	--	0,001	Min	-7,3%	Min	-0,06

GRAPH 5. The graph sets out the mean data obtained at each experimental time for the parameter under study. Data are expressed as a mean ± SEM.



COMMENT: after 28 days of bi-daily product use tested product determines a mean statistically significant improvement of skin thickness by +8.2% (+0.06mm).

Note: the intra-group statistical analysis (vs. T0) is reported above the error bar in black colour.
Legend: ** p<0.01.

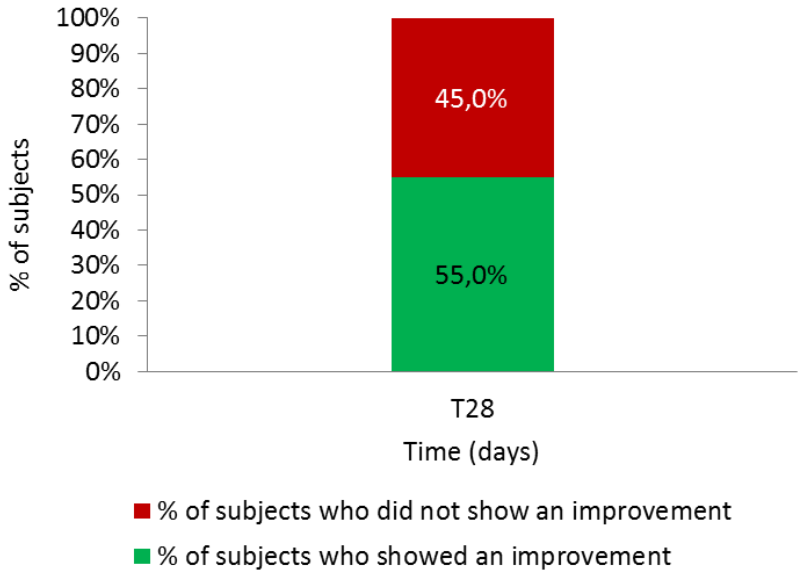
REDUCTION OF THE COLOUR OF DARK SPOTS – LONG TERM TEST

TABLE 6. In the table below clinical scores obtained for each volunteer after 28 days of product use are reported.

n.	Vol. ID	T28
01	F3656S	2
02	P2820M	1
03	C0085N	1
04	R1784M	1
05	B0012G	2
06	I0170M	1
07	D0668A	3
08	R0329O	1
09	T4004E	2
10	L4183A	1
11	S3831A	2
12	B0015R	2
13	M3265G	4
14	G0138O	1
15	M3661M	2
16	E3159L	1
17	A1462R	3
18	B0014L	2
19	B0526A	2
20	S4174L	1
	Mean	1,8
	SEM	0,2

LEGEND: 1. No variation; **2.** Slight improvement; **3.** Moderate improvement; **4**. Remarkable improvement.

GRAPH 6. The graph reports the percentage of subjects related to the effect of the product.



COMMENT: an improvement of skin complexion evenness (in terms of reduction of the colour of dark spots) was clinically observed after 28 days of use in 55% of the enrolled subjects.

SELF ASSESSMENT QUESTIONNAIRE

Here below the results obtained from the questionnaire filled in by each volunteer after the first product application are summarized. The results are expressed as percentage (%) of subjects who expressed the same opinion among those proposed.

No.	After first product application	Very pleasant	Pleasant	Neither pleasant nor unpleasant	Unpleasant	Very unpleasant	Positive answers
01	What do you think about the product aspect?	20,0%	75,0%	5,0%	0,0%	0,0%	95,0%
02	What do you think about the product texture?	30,0%	65,0%	5,0%	0,0%	0,0%	95,0%
03	What do you think about the product fragrance?	50,0%	45,0%	5,0%	0,0%	0,0%	95,0%
04	What do you think about the product spreadability?	15,0%	85,0%	0,0%	0,0%	0,0%	100,0%
No.		Very good	Good	Not good	Bad		Positive answers
05	What do you think about product penetration?	15,0%	70,0%	15,0%	0,0%		85,0%
No.		Silky	Soft	Sticky	Oily		Positive answers
06	What is the after feel on the skin?	25,0%	55,0%	15,0%	5,0%		80,0%
No.		Very good	Good	Not good	Bad		Positive answers
07	What is your overall appreciation of this product?	15,0%	85,0%	0,0%	0,0%		100,0%
No.	After the application	Intense	Moderate	Slightly	Not at all		Positive answers
08	Have you noticed an amelioration of skin hydration?	25,0%	55,0%	20,0%	0,0%		80,0%
09	Have you noticed an amelioration of skin smoothness?	25,0%	60,0%	15,0%	0,0%		85,0%
No.	After the application, have you noticed that:	Agree	Moderately agree	Not completely agree	Not agree		Positive answers
10a	Your skin is more moisturized	30,0%	65,0%	5,0%	0,0%		95,0%
10b	You skin is smoother	25,0%	70,0%	5,0%	0,0%		95,0%
10c	Skin is more protected	10,0%	85,0%	5,0%	0,0%		95,0%
10d	Fine lines are less visible	0,0%	50,0%	45,0%	5,0%		50,0%

Here below the results obtained from the questionnaire filled in by each volunteer after 28 days of product use are summarized. The results are expressed as percentage (%) of subjects who expressed the same opinion among those proposed.

No.	After 4 weeks of application, have you noticed that:	Agree	Moderately agree	Not completely agree	Not agree	Positive answers
1a	Your skin is more moisturized	30,0%	70,0%	0,0%	0,0%	100,0%
1b	Your skin is smoother	20,0%	70,0%	10,0%	0,0%	90,0%
1c	Fine lines and wrinkles are less visible	20,0%	60,0%	20,0%	0,0%	80,0%
1d	Your skin looks re-densified (plumped)	10,0%	70,0%	20,0%	0,0%	80,0%
1e	Dark spots look less visible	5,0%	80,0%	15,0%	0,0%	85,0%
1f	Skin complexion looks more even	15,0%	70,0%	15,0%	0,0%	85,0%
No.	After application, the following issues seem improved:	Strongly improved	Improved	Unchanged	Aggravated	Positive answers
2a	Dehydration	5,0%	90,0%	5,0%	0,0%	95,0%
2b	Dark spots	5,0%	80,0%	15,0%	0,0%	85,0%
2c	Uneven look	10,0%	60,0%	30,0%	0,0%	70,0%
2d	Fine lines and wrinkles	10,0%	55,0%	35,0%	0,0%	65,0%
2e	Thinned skin (bony)	5,0%	65,0%	30,0%	0,0%	70,0%
No.		Yes	No			Positive answers
03	When you have used the product, have you felt uncomfortable sensations?	0,0%	100,0%			100,0%
No.		Yes	No			Positive answers
4a	If yes, have you stopped the application?	--	--			--
4b	If the treatment has been stopped, has it been stopped for a skin reaction?	--	--			--
4c	Have you stopped the treatment for other reasons? If yes, specify why.	0,0%	100,0%			100,0%
No.	Subsequent use of the product	Yes	No			Positive answers
05	Would you continue using this product?	95,0%	5,0%			95,0%
No.		Luxe	Masstige	Mass market		Positive answers
06	According to you, this product is sold in which market seg	15,0%	85,0%	0,0%		--
No.		Absolutely yes	Maybe	Probably not	Absolutely not	Positive answers
07	Would you buy this product independently from price?	45,0%	55,0%	0,0%	0,0%	100,0%

CONCLUSIONS

On the basis of the obtained results, it is possible to conclude that the product:

TORSTONE SA

RIVOLI
LES MAINS SOIN TOTAL ANTI-AGE
Reference code: lab-01160.13

determines a general improvement of the analysed skin parameters both after its first application and after 28 days of its daily use.
In particular the product determines:

- An increase of skin moisturizing by +12.9% 2 hours after its single application, by +6.2% 24 hours after its single application and by 35.6% after 28 days of use.
- An increase of ITA° by +6.3% after 28 days of use (higher is the value of ITA°, lighter is the skin pigmentation).
- An increase of skin thickness (epidermidis + dermis) by 8.2% after 28 days of product use.

The reported % are referred to the mean variations of the analysed parameters versus T0.
Reported variations are statistically significant variation versus T0.

- 15 minutes after its single application tested product doesn't alter the basal skin conditions; pH mean value is almost unvaried (a mean variation, not statistically significant, by -2.5% is recorded at T15min).

The clinical analysis performed by the Dermatologist shows an improvement of the evenness complexion (in terms of reduction of the colour of dark spots) recorded in 55% of the enrolled subjects.

Moreover, the most part of the enrolled volunteer positively judged the product for the most part of the investigated aspects.

Study Director – Data analysis and report

Dr Eleonora Spartà

Digitally signed by Eleonora Spartà
Date: 2019.08.28 11:59:16 +02'00'

Principal Investigator

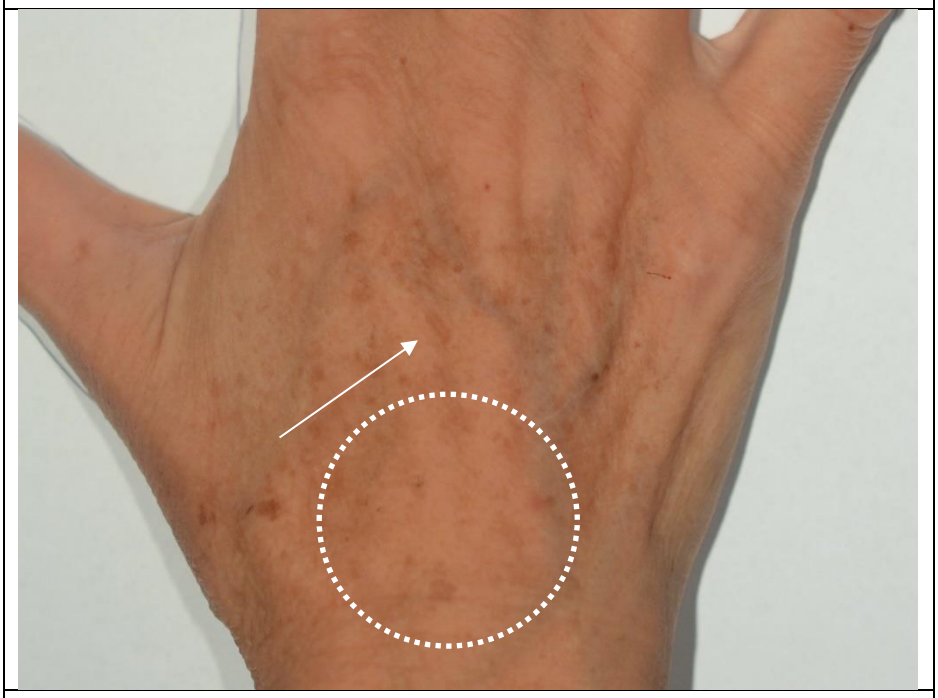
Dr Enza Cestone

DOTT.SSA ENZA CESTONE
MEDICO CHIRURGO
SPECIALISTA IN DERMATOLOGIA E VENEREOLOGIA
Via Mentana, 27 - 27100 PAVIA
Partita IVA 02541110183
C.F. CST NZE 80S42M109Z
Parere O.d.M. PV n. 32 del 03/11/2009

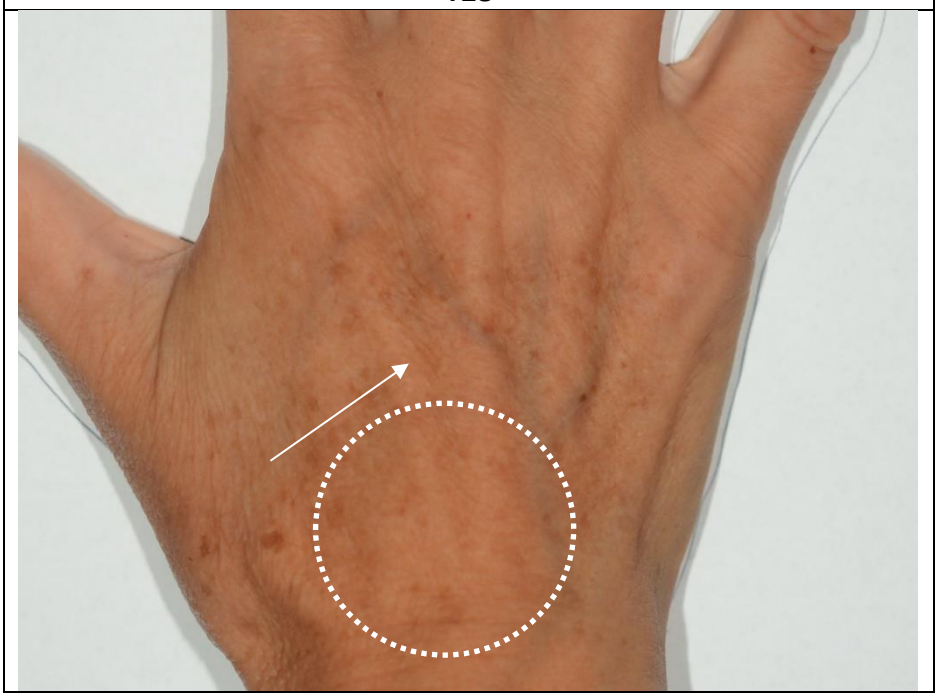
Digitally signed by Enza Cestone
Date: 2019.08.28 10:34:21 +02'00'

ANNEX 1 – DIGITAL PICTURES

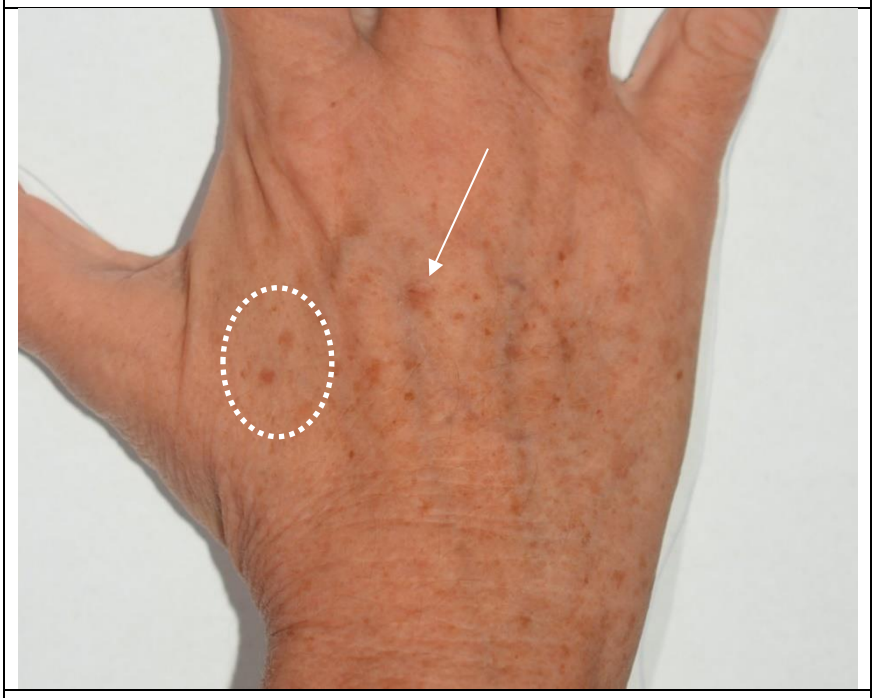
T0 – vol #7



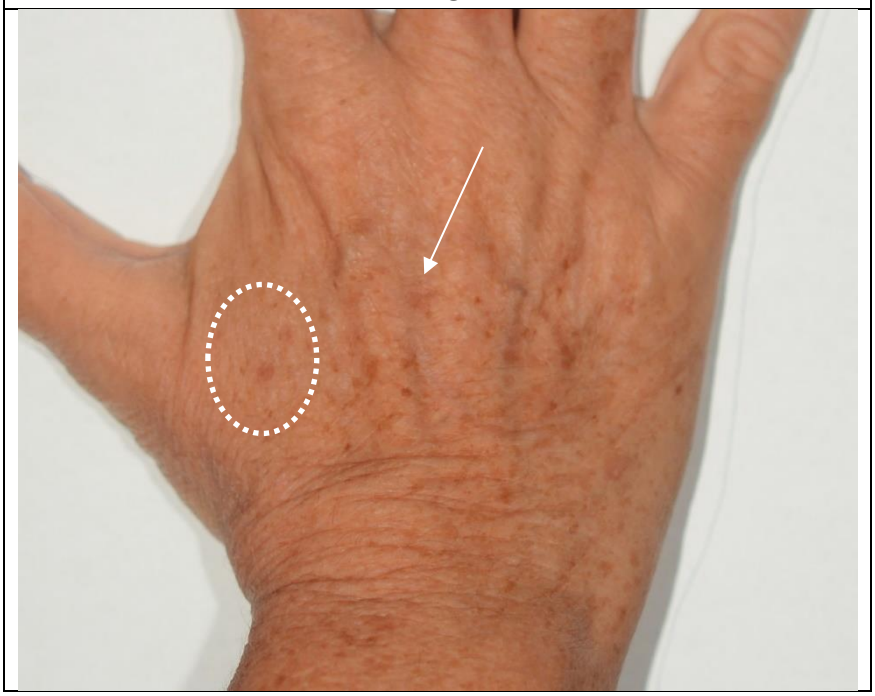
T28



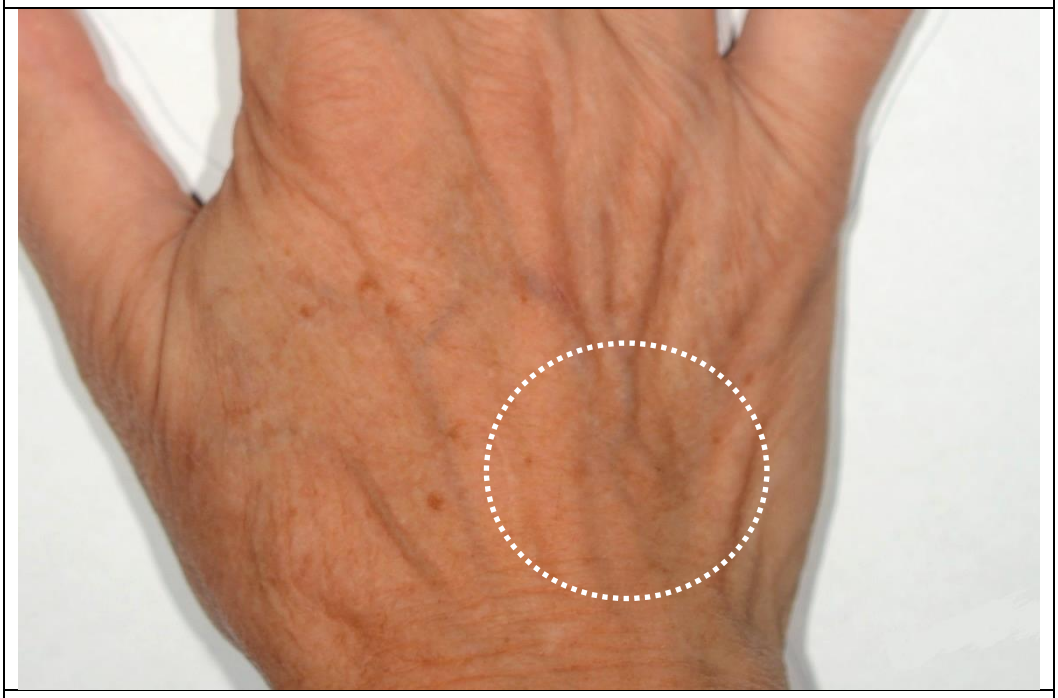
T0 – vol #13



T28



T0 – vol #17



T28