

TEST REPORT N. 10/000394462

date of issue 07/12/2010

Customer ID 0053426

Kind attention

PELIN DOGAL BAKIM VE
KOZMETIK SAN. TIC. LTD. STI
GIRNE MAH. IRMAK SOK.
KUCUKYALL ISMERKEZI G
BLOK NO: 37 ERENKOY VD
34852 MALTEPE/ISTANBUL

Turchia

Sample information

Acceptance number 10.097039.0001
Delivered by Our technician Dr.ssa Alessandra Conte on 19/10/2010
Place of origin PELIN DOGAL BAKIM VE KOZMETIK SAN. TIC. LTD. STI GIRNE MAH. IRMAK SOK.
KUCUKYALL ISMERKEZI G BLOK NO: 37 ERENKOY VD 34852 MALTEPE/ISTANBUL
Sample description PELINTY'S BITKI DERMAN ANTIAGING BAKIM SETI

Sampling data

Sampled by Customer

EVALUATION OF THE EFFICACY OF ANTI-WRINKLE COSMETIC PRODUCTS**INDEX**

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INTERNATIONAL NOMENCLATURE COSMETIC INGREDIENT (INCI) LABELLING OF THE SAMPLES (PROVIDED BY THE CUSTOMER)**INGREDIENTS:****ANTIAGING GECE BAKIM KREMI 40+**

Aqua, White Mineral Oil, Petrolatum, Butyrospermum Parkii, C12-C15 Alkyl Benzoate, Glyceryl Stearate (and) Ceteareth-20 (and) Ceteareth-12 (and) Cetearyl Alcohol (and) Cetyl Palmitate, Water (aqua), Glycerin, Pentapeptide-18, Caprylic Glycol, Cetyl Stearyl Alcohol, Althea Officinalis Extract, Lavandula Stoechas, Agropyrum Repens, Orbignya Oleifera Seed Oil, Laurdimonium Hydroxypropyl Hydrolyzed Wheat Protein, Cucumis Sativus Extract, Tocopherol Acetate, Allantoin, Fragrance, Carbomer, Aqua, Diazolidinyl Üre, Sodyum Benzoat, Potasyum Sorbat.

ARINDIRICI VE CANLANDIRICI TONIK

Aqua, Water, Glycerin, Pentapeptide-3, Althea Officinalis Extract, Lavandula stoechas, Agropyrum repens, Laurdimonium Hydroxypropyl Hydrolyzed Wheat Protein, Cucumis sativus Extract, Tocopherol Acetate, Allantoin, Fragrance, Aqua, Diazolidinyl Üre, Sodyum benzoat, Potasyum Sorbat..

METHOD DESCRIPTION**1. AIM OF THE TEST**

The aim of the study is the in vivo evaluation of possible changes of skin micro-relief (**wrinkles**), after the repeated application of a cosmetic treatment on intact skin.

2. SUBJECTS UNDERGOING THE TREATMENT

20 volunteers, sex: F, age: **40 – 50**.

Criteria of volunteers selection:

- Healthy subjects.
- Absence of skin pathologies.
- Absence of pharmacological treatments.
- Negative anamnesis for atopy.
- Obligation to avoid the use of other topical products in the testing zone during the test and to avoid the application of body creams in the zone to be tested for at least 3 days before the beginning of the test.
- Exclusion: pregnant women and women in lactation period, minors.

Each volunteer is given a form to fill in and subscribe, in order to express free consent to the test and to the above mentioned conditions.

The volunteers, during the measurements, have not to apply any make up, cream or lotion in order to avoid possible interferences in the measurements and in the test results.

3. EXECUTION OF THE TEST

As agreed with the Customer, the 20 volunteers have applied the samples twice a day (morning and evening) for 30 days: ANTIAGING GECE BAKIM KREMI 40+ in the morning and ARINDIRICI VE CANLANDIRICI TONIK + ANTIAGING GECE BAKIM KREMI 40+ in the evening.

Before the beginning of the test, the volunteers have received all the instructions given by the Customer (correct use of the product, safety, INCI list, etc.).

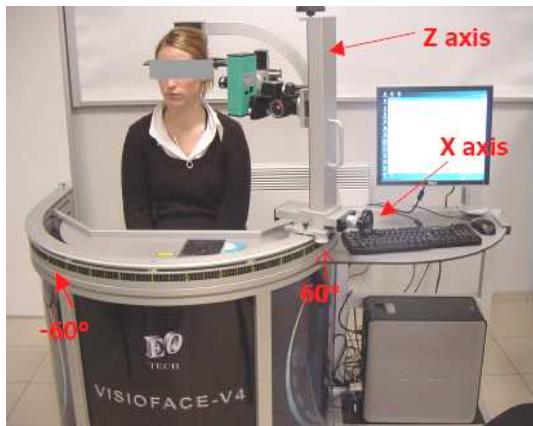
- The **instrumental controls** have been performed:

1. T0: before the beginning of the test. Observation of the skin basal values.
2. T1: after 15 days of application.
3. T2: after 30 days of application.

- **Autoevaluation:** to each of the **20** volunteers has been also given an autoevaluation questionnaire to obtain a judgement from potential customers on product performances.

INSTRUMENTS AND PRINCIPLES OF MEASURE

CUTANEOUS MICRO-RELIEF



The measurement is carried out by DERMATOP BLUE (EOTECH, France) using Breuckmann technology.

DERMATOP BLUE is a skin scanner to perform tests for the evaluation of the skin structure. It compares the results obtained evaluating the volunteers at time 0 and after the treatment with the product to be tested. The instrument measures very precisely the performances of cosmetic products formulated to reduce skin imperfections such as: wrinkles.

A sensor system identifies with great precision the zone to be tested, makes a skin scanning that will detect the position, area and volume of relieves with microscopic (pores) or macroscopic (mattress skin due to cellulite) dimensions. The scanning is performed before and after the treatment and finally the differences are registered, photographed and statistically treated.

The skin is optically monitored by using a digitalisation system of the image without the use of silicon moulds.

The system is based on the graphic representation of in vivo skin illuminated by a blue source and on the electronic elaboration and evaluation of this image according to the clinical parameters. The parameters qualitatively and quantitatively correspond to the physiological condition of the skin and have been derived by the conventional parameters for wrinkledness.

For the evaluation of skin before and after the treatment the 2 most representative parameters have been chosen from the file **Results.csv**. that contains 50 profiles of "2D-roughness statics" for each volunteer and for each considered time.

- Rz (μm): Average Maximum profile height difference (Rptm):** represents the average width of the negative peak.
- Ra (μm): Linear average profile roughness:** represents the arithmetical average of the parameters of wrinkledness calculated on the micro profile of the skin.

CALCULATIONS AND STATISTICS

The average values obtained from the instrumental evaluations and the relative decreases/increases with respect to the basal value, at the 3 fixed values:

- T0 (before the beginning of the test)
- T1 (after 15 days of application)
- T2 (after 30 days of application)

It has been calculated the % variation of the medium instrumental detections with reference to the basal values at the fixed times according to the following formula:

$$\text{variation \%} = \left[(v_1 - v_2) / v_2 \right] \times 100$$

where v_1 is the detection at the considered time and v_2 is the corresponding basal value (T0).

To perform the analysis of the significance, the variance analysis has been applied to the significance level $\alpha=0,05$.

RESULTS**EVALUATION OF THE ANTI-WRINKLE EFFICACY
before and after application ON PERIOCULAR AREA
EVALUATION OF THE TREATED AREA BY DERMATOP BLUE**

The following tables report the **R_a** and **R_z** values for each volunteer at the 3 considered times

Volunteers nr	T0		T1		T2	
	RA (μm)	RZ (μm)	RA (μm)	RZ (μm)	RA (μm)	RZ (μm)
1	0,041	0,133	0,033	0,107	0,039	0,119
2	0,026	0,082	0,025	0,076	0,025	0,078
3	0,035	0,125	0,034	0,108	0,030	0,097
4	0,071	0,124	0,056	0,116	0,054	0,109
5	0,044	0,108	0,033	0,099	0,034	0,093
6	0,041	0,104	0,037	0,096	0,039	0,103
7	0,042	0,103	0,035	0,085	0,034	0,092
8	0,055	0,133	0,045	0,117	0,040	0,117
9	0,033	0,098	0,030	0,093	0,028	0,092
10	0,019	0,060	0,018	0,059	0,019	0,052
11	0,031	0,093	0,027	0,089	0,026	0,093
12	0,054	0,170	0,047	0,145	0,046	0,144
13	0,036	0,119	0,036	0,115	0,036	0,107
14	0,045	0,129	0,041	0,106	0,037	0,106
15	0,029	0,091	0,026	0,086	0,025	0,077
16	0,035	0,090	0,033	0,087	0,026	0,079
17	0,033	0,115	0,028	0,089	0,033	0,095
18	0,037	0,124	0,034	0,101	0,033	0,101
19	0,044	0,124	0,042	0,108	0,041	0,106
20	0,029	0,107	0,026	0,075	0,028	0,092
AVERAGE	0,039	0,112	0,034	0,098	0,034	0,098

In addition, we have calculated the percentage decrease of each volunteer at T1 and T2 with respect to the basal values T0:

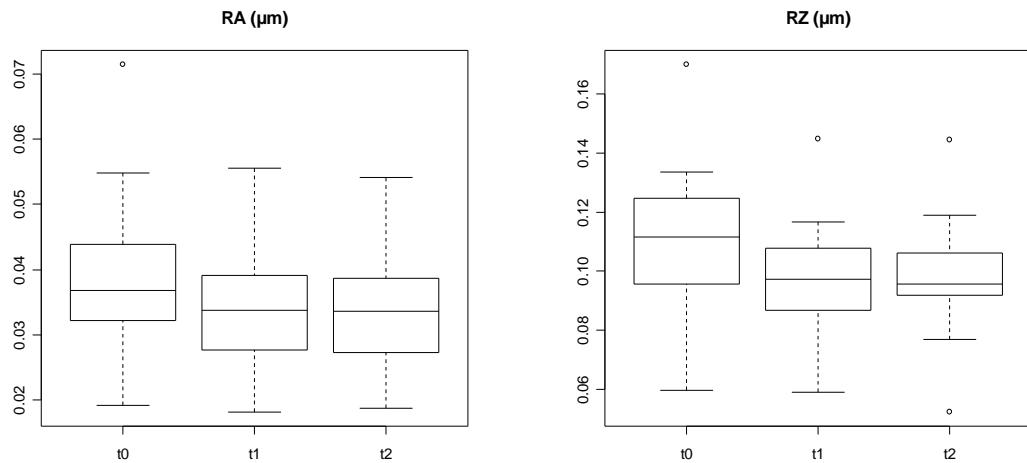
Volunteers nr	DECREMENTS %			
	T1 VS T0		T2 VS T0	
	RA (µm)	RZ (µm)	RA (µm)	RZ (µm)
1	-18,6	-19,8	-5,5	-10,5
2	-3,4	-7,5	-1,8	-4,6
3	-2,9	-13,7	-12,8	-22,8
4	-22,3	-7,0	-24,2	-12,2
5	-23,8	-8,7	-22,5	-13,5
6	-10,3	-7,7	-6,3	-1,1
7	-16,3	-16,9	-19,6	-10,8
8	-18,0	-12,5	-27,9	-12,4
9	-8,6	-5,6	-14,6	-6,7
10	-5,2	-0,8	-2,0	-12,1
11	-14,2	-4,2	-16,1	-0,2
12	-13,0	-14,7	-14,3	-14,9
13	-0,6	-3,0	-0,8	-10,1
14	-9,7	-17,7	-19,1	-18,1
15	-10,6	-5,2	-15,1	-15,1
16	-4,1	-3,3	-26,1	-12,0
17	-15,2	-23,0	-1,7	-17,4
18	-7,9	-18,3	-10,9	-18,4
19	-4,5	-12,9	-6,4	-14,6
20	-11,2	-29,7	-1,4	-14,5
AVERAGE	-11,0	-11,6	-12,5	-12,1

ANALYSIS OF SIGNIFICATIVENESS

The table below reports the significativeness indexes obtained after the application of variance analysis (level of significativeness $\alpha=5\%$).

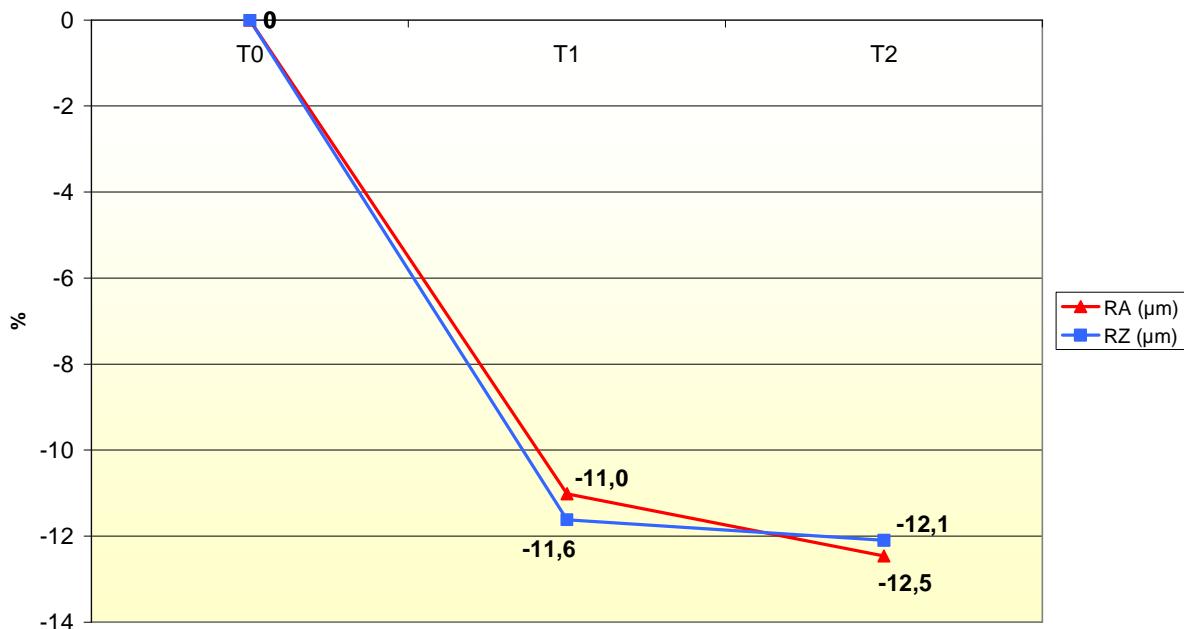
	T0 vs T1	T0 vs T2
Ra (μm)	0,16	0,10
p-value<0,05 <i>significant</i> p-value $\geq 0,05$ <i>not significant</i>	<i>not significant</i>	<i>not significant</i>
Rz (μm)	0,046	0,044
p-value<0,05 <i>significant</i> p-value $\geq 0,05$ <i>not significant</i>	<i>significant</i>	<i>significant</i>

BOXPLOT: distribution of Ra(μm) values and Rz(μm) values among the observed times (T0, T1, T2).

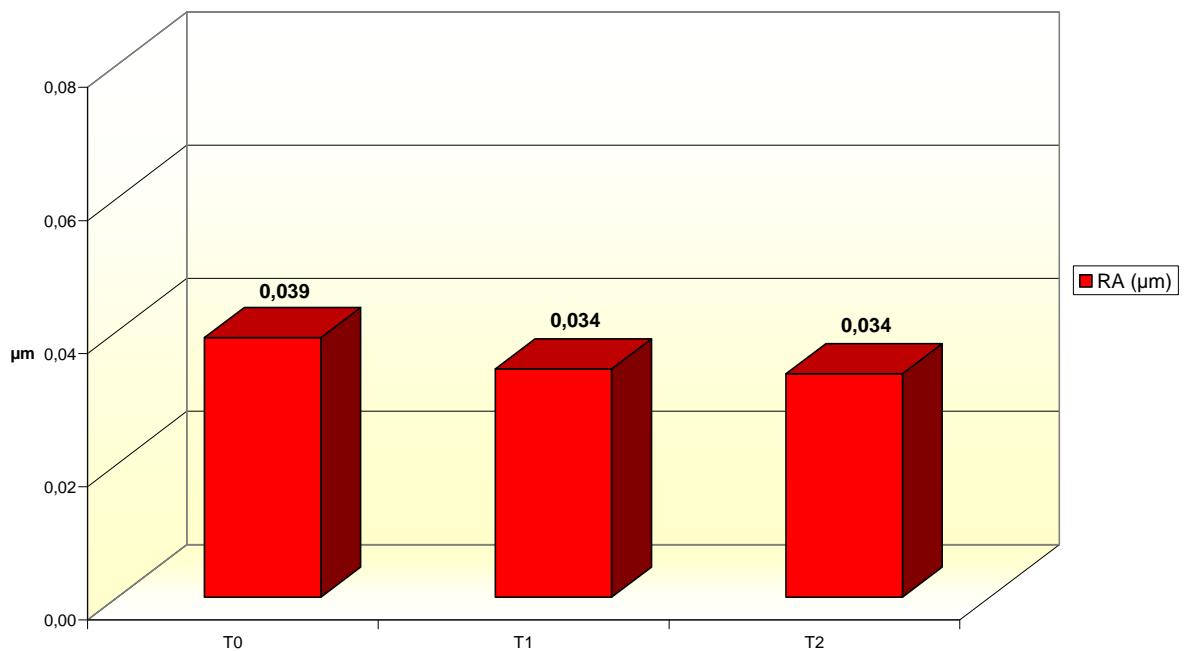


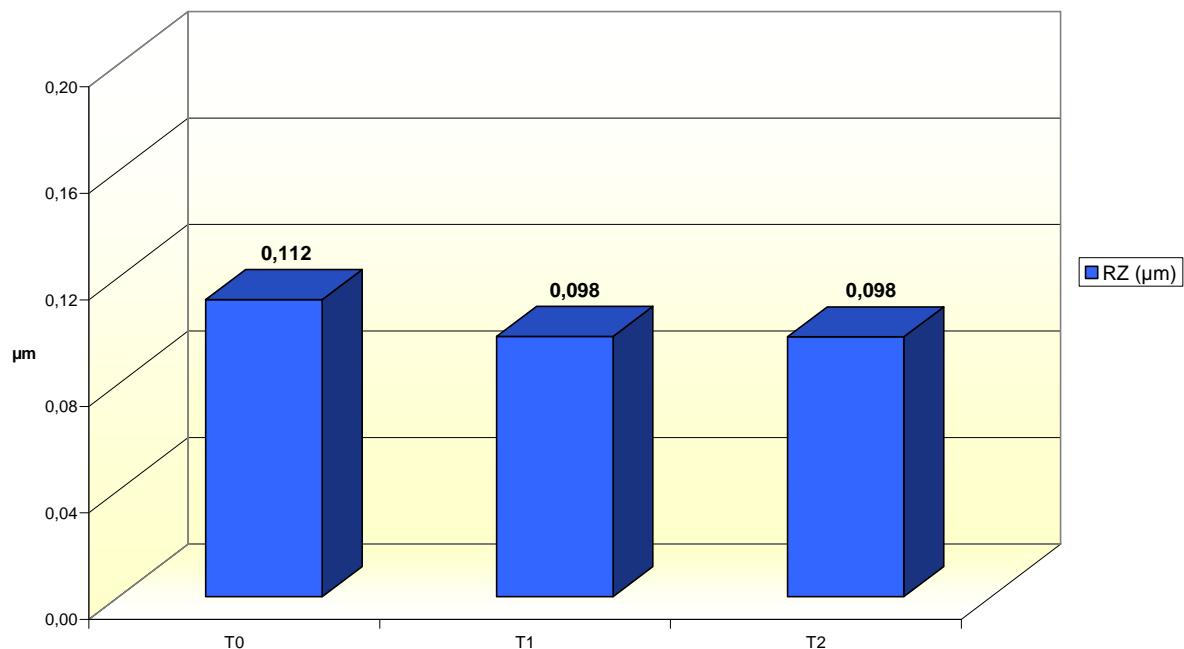
GRAPHS

AVERAGE PERCENTAGE DECREMENTS



RA(µm) AVERAGE REDUCTION



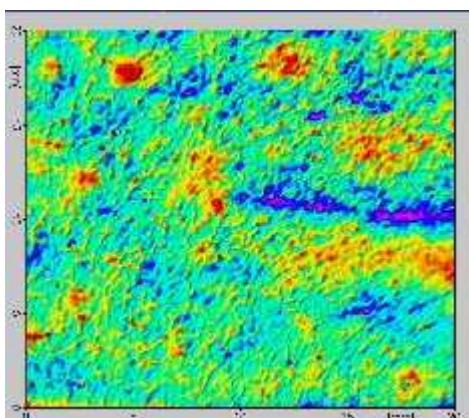
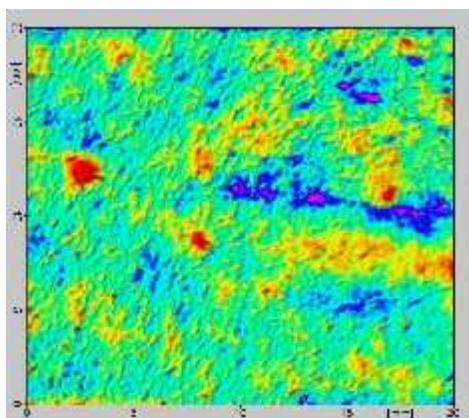
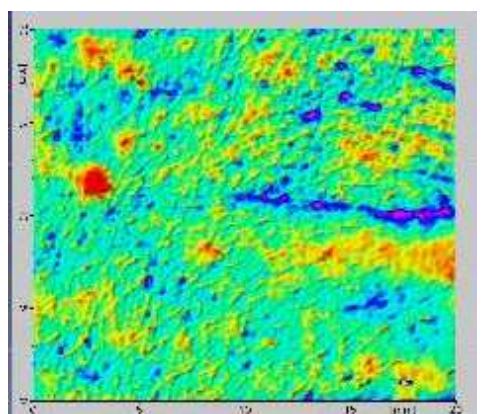
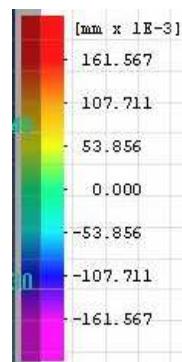
RZ(µm) AVERAGE REDUCTION

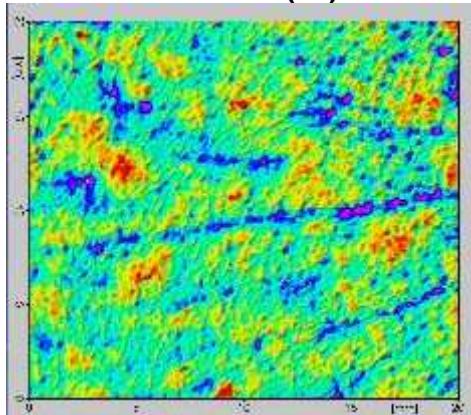
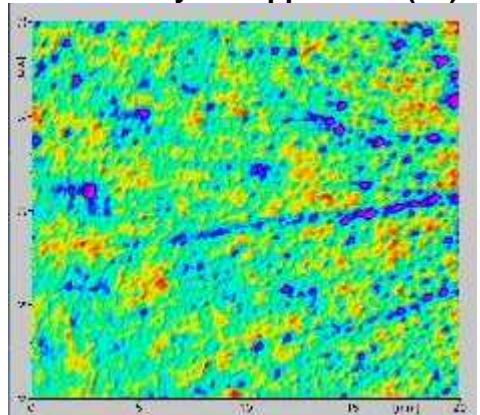
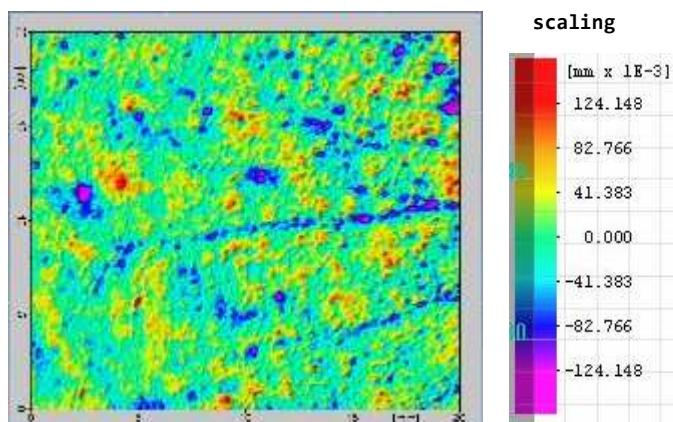
IMAGES CREATED BY DERMATOP SOFTWARE

Here we have reported an example of images re-elaborated by DERMASURF software (skin topography). Such software permits the skin evaluation by a modification of the image acquired by DERMATOP BLUE instrument.

From the image previously registered by DERMATOP BLUE instrument, we have first extracted an area 20x20 mm width; such area has been visualized as indicated in the image below and after it has been applied the colour scale ("scaling").

Here we have reported the most significant elaboration respect to T0, T1 and T2.

VOLUNTEERS NR 1**Before treatment (T0)****After 15 days of application(T1)****After 30 days of application (T2)****scaling**

VOLUNTEERS NR 2**Before treatment (T0)****After 15 days of application(T1)****After 30 days of application (T2)**

AUTOEVALUATION QUESTIONNAIRE**1. TOTAL RESULTS****GENERAL INFORMATION****1. HOW IS YOUR SKIN?**

dry	10%
normal	15%
mixed	60%
fat	15%

2. HOW OFTEN DO YOU USE COSMETIC PRODUCTS SPECIFIC FO WRINKLES?

never	20%
sometimes	45%
often	20%
always	15%

3. WHERE DO YOU PREVALENTLY SPEND YOUR DAYS?

outdoor	15%
indoor	20%
mainly outdoor and shortly indoor	5%
mainly indoor and shortly outdoor	35%
equally indoor and outdoor	25%

4. DO YOU USE A SPF CREAM DURING THE SUN EXPOSITION?

never	10%
sometimes	25%
always	65%

5. HOW OFTEN DO YOU USE TANNING LAMPS?

never	70%
sometimes	30%
often (once a week)	0%
always (more than twice a week)	0%

BEFORE USE**6. IN YOUR OPINION, HOW DO YOU FIND YOUR WRINKLES BEFORE THE APPLICATION OF THE COSMETIC PRODUCT?**

light	35%
moderate	55%
deep	10%

7. IN YOUR OPINION HOW DO YOU FIND YOUR SKIN BEFORE THE APPLICATION OF THE COSMETIC PRODUCT?

	HYDRATED	LUMINOUS	SOFT	ELASTIC	SMOOTH	COMPACT
not at all	10%	20%	0%	0%	10%	15%
poorly	50%	45%	35%	50%	60%	30%
fairly	40%	35%	65%	50%	30%	55%
much	0%	0%	0%	0%	0%	0%

AFTER USE**8. AFTER THE TREATMENT WITH THE COSMETIC PRODUCT, HAS YOUR SKIN IMPROVED?**

yes	60%
no	40%

9. IF SO, HOW MUCH?

poorly	25%
sufficiently	58%
much	17%

10. HOW DO YOU FIND YOUR WRINKLES AFTER THE APPLICATION OF THE COSMETIC PRODUCT?

not attenuated at all	45%
lightly attenuated	40%
attenuated	15%

11. IN YOUR OPINION HOW DO YOU FIND YOUR SKIN AFTER THE APPLICATION OF THE COSMETIC PRODUCT?

	HYDRATED	LUMINOUS	SOFT	ELASTIC	SMOOTH	COMPACT
not at all	5%	5%	0%	0%	0%	0%
poorly	20%	30%	20%	25%	35%	40%
fairly	65%	55%	60%	60%	55%	55%
much	10%	10%	20%	15%	10%	5%

12. EXPRESS A COMPREHENSIVE JUDGEMENT ABOUT THE EFFICACY OF THE COSMETIC PRODUCT

not at all satisfied	35%
quite satisfied	40%
satisfied	15%
very satisfied	10%

ANTIAGING GECE BAKIM KREMI 40+**1. HOW DO YOU JUDGE THE PERFUME OF THE COSMETIC PRODUCT?**

very pleasant	35%
pleasant	40%
no pleasant/ no unpleasant	15%
unpleasant	10%
very unpleasant	0%

2. CAN YOU EASILY SMEAR THE COSMETIC PRODUCT?

yes	95%
no	5%

3. DOES THE COSMETIC PRODUCT EASILY ABSORB?

yes	35%
no	65%

4. HOW DO YOU JUDGE THE SENSORY ASPECT (COLOUR, PERFUME, TEXTURE) OF THE COSMETIC PRODUCT DURING THE APPLICATION?

very pleasant	0%
pleasant	45%
no pleasant/ no unpleasant	45%
unpleasant	10%
very unpleasant	0%

ARINDIRICI VE CANLANDIRICI TONIK**1. HOW DO YOU JUDGE THE PERFUME OF THE COSMETIC PRODUCT?**

very pleasant	15%
pleasant	65%
no pleasant/ no unpleasant	15%
unpleasant	5%
very unpleasant	0%

2. CAN YOU EASILY SMEAR THE COSMETIC PRODUCT?

yes	100%
no	0%

3. DOES THE COSMETIC PRODUCT EASILY ABSORB?

yes	100%
no	0%

4. HOW DO YOU JUDGE THE SENSORY ASPECT (COLOUR, PERFUME, TEXTURE) OF THE COSMETIC PRODUCT DURING THE APPLICATION?

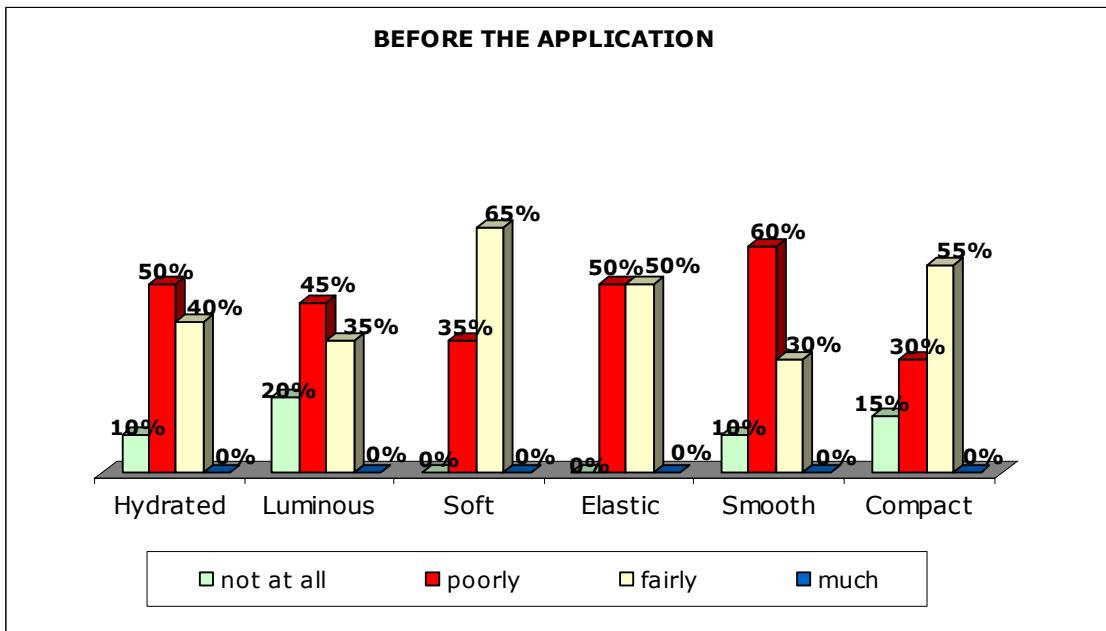
very pleasant	10%
pleasant	85%
no pleasant/ no unpleasant	5%
unpleasant	0%
very unpleasant	0%

Notes:

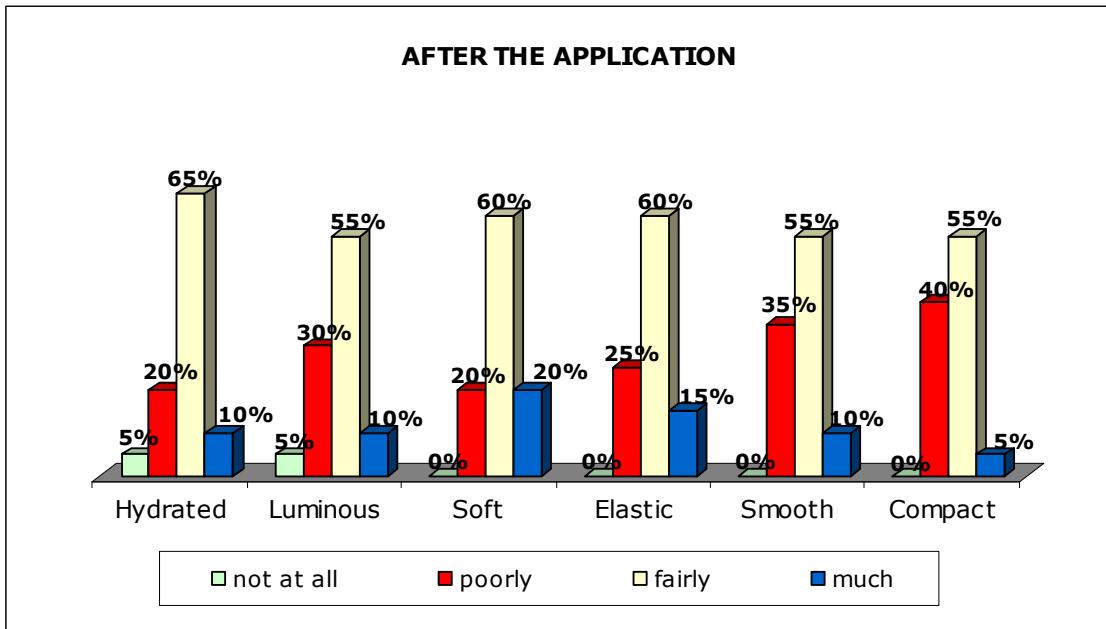
11 volunteers on 20 (the 55% of the panel) have noticed that the cream (antiaging gece bakim kremi 40+) is too greasy, so the skin is glossy/fat.

2. COMPARISON OF THE CHARACTERISTICS OF THE SKIN BEFORE AND AFTER THE APPLICATION OF TREATMENT

GRAPH – Comparison of the skin condition **before** the treatment

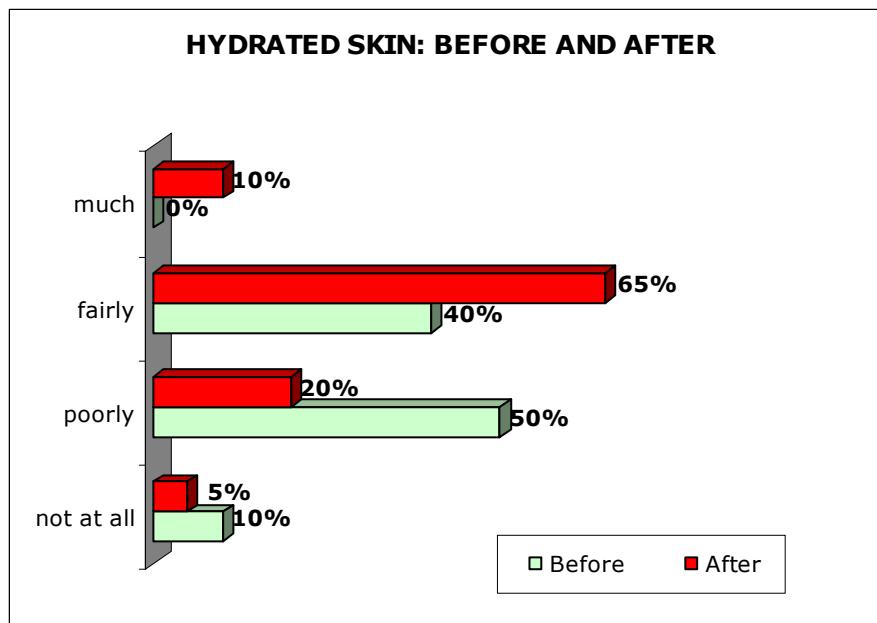


GRAPH – Comparison of the skin condition **after** the treatment

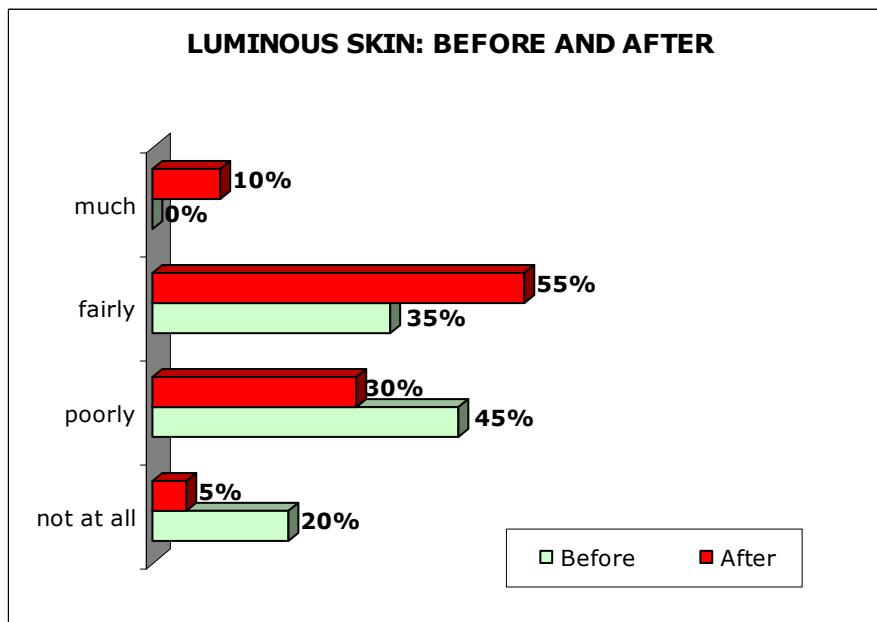


In the specific:

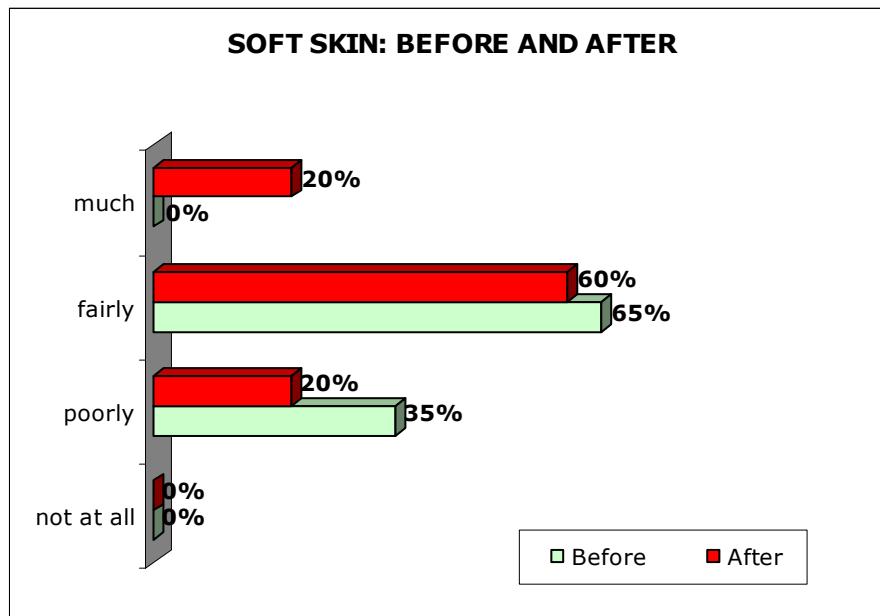
GRAPH – Hydrated skin: comparison before and after the application of cream



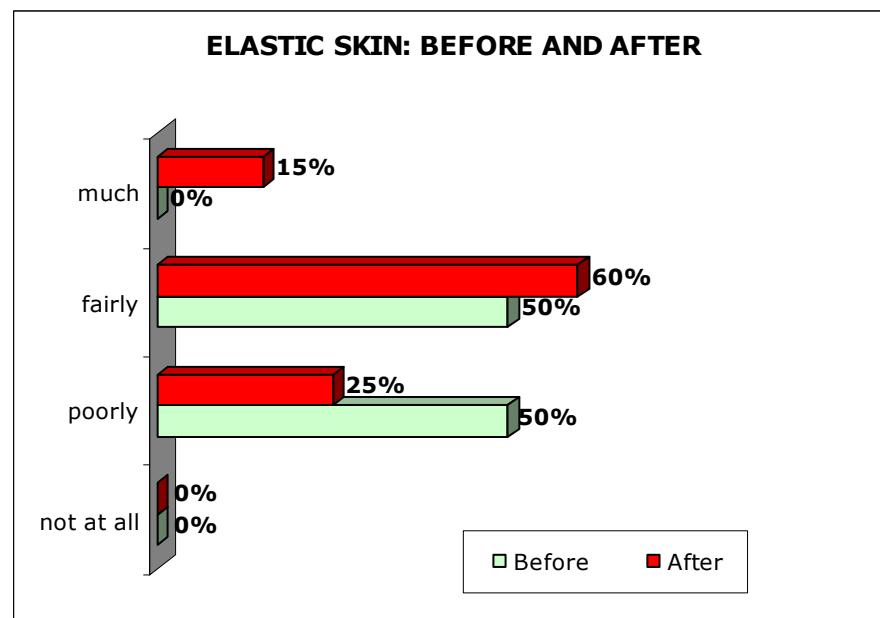
GRAPH – Luminous skin: comparison before and after the application of cream



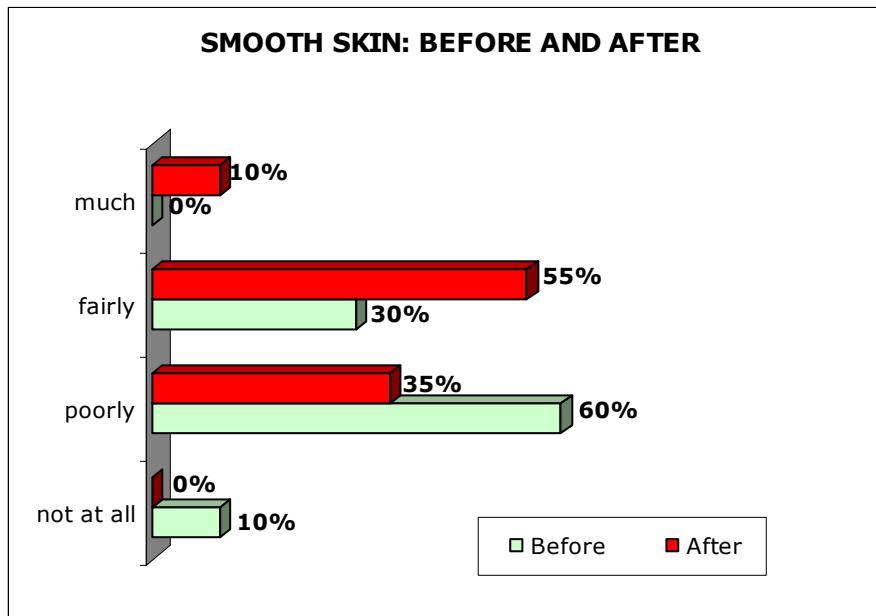
GRAPH – Soft skin: comparison before and after the application of cream



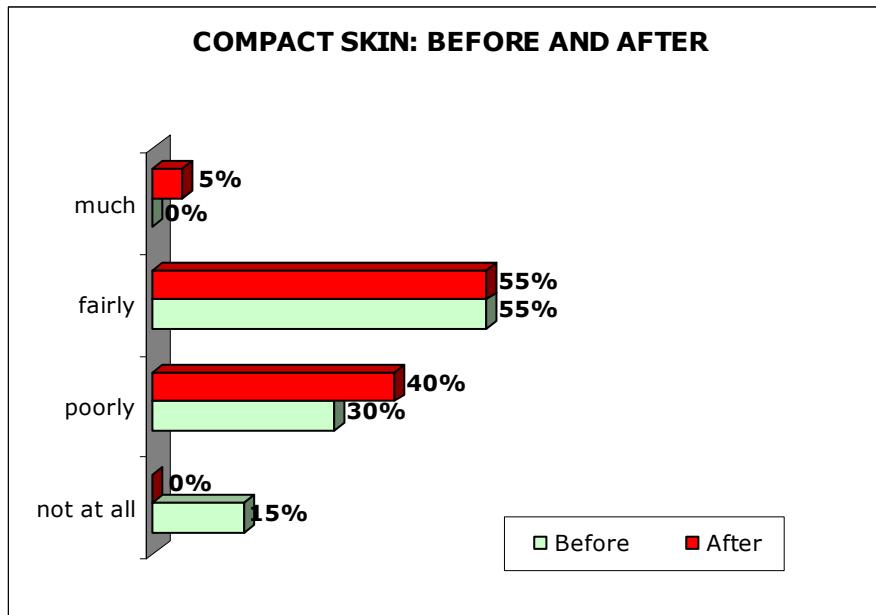
GRAPH – Elastic skin: comparison before and after the application of cream



GRAPH – Smooth skin: comparison before and after the application of cream



GRAPH – Compact skin: comparison before and after the application of cream



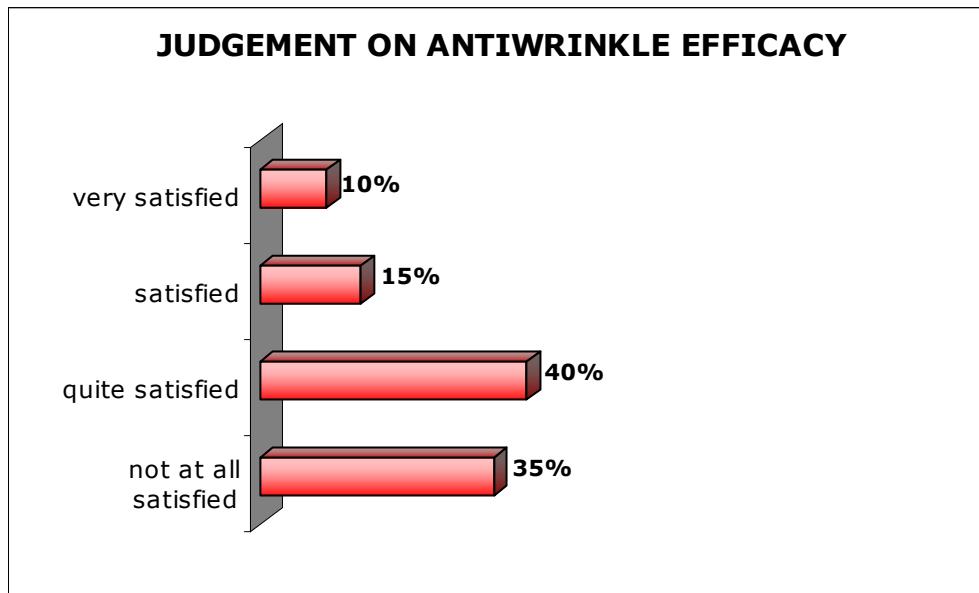
Tab. – Evaluation of the skin before and after the application of the treatment

	Judgement	BEFORE	AFTER
HYDRATED	fairly + much	40%	75%
	not at all + poorly	60%	25%
LUMINOUS	fairly + much	35%	65%
	not at all + poorly	65%	35%
SOFT	fairly + much	65%	80%
	not at all + poorly	35%	20%
ELASTIC	fairly + much	50%	75%
	not at all + poorly	50%	25%
SMOOTH	fairly + much	30%	65%
	not at all + poorly	70%	35%
COMPACT	fairly + much	55%	60%
	not at all + poorly	45%	40%

The table, that divides in 2 groups the conditions “much”, “fairly”, “poorly” and “not at all”, points out the improvement of the skin after the application of the products.

3. COMPREHENSIVE JUDGEMENT ABOUT THE ANTIWRINKLE EFFECT OF THE TREATMENT

GRAPH – Comprehensive judgement about the efficacy of antiwrinkle product



CONCLUSIONS

Considered the obtained results following the indicated protocol, it is possible to draft these conclusions:

INSTRUMENTAL CONTROL

Considering the results obtained we can conclude that – in the conditions expected by the test - the treatment ANTIAGING GECE BAKIM KREMI 40+ and ARINDIRICI VE CANLANDIRICI TONIK is able to change the micro-relief of the skin, in terms of reduction of wrinkles.

In fact, with the continuous application of the product twice a day for 30 days we have pointed out an average reduction of R parameter (wrinkledness). **Ra parameter** - which represents an arithmetical average of wrinkledness parameters calculated on the micro profile of the skin is reduced by 12,5% on the average. **Rz parameter** – which represents the average width of the negative peak is reduced by 12,1% on the average.

AUTOEVALUATION STUDY

From the study performed on 20 questionnaires, we can conclude as follows:

After 30 days of constant application, the 60% of the volunteers have noticed an improvement of the skin aspect.

The answers to the questionnaires have shown that the volunteers have appreciated the smooth, compact, hydrated and elastic effect of the products on their skin.

On the whole, the volunteers are satisfied of the efficacy of the antiwrinkle treatment. In particular, the 40% is quite satisfied, the 15% is satisfied and the 10% is very satisfied.

BIBLIOGRAPHY

- "Guidelines for the evaluation of the efficacy of cosmetic products", The European Cosmetic, Toiletry and Perfumery Association – COLIPA.
- T. Callaghan, M. Brand, K.- P. Wilhelm: Microrelief & Wrinkle measurements – SOFW-Journal 134-3/2008
- Substantiating Anti aging Product Claims Loraine M. Harnisch, m. K. Raheja, Leslie K. Lockhart and Alessandra Pagnoni Cosmetics & Toiletries Vol. 114, No 10/October 1999
- Skin topography measurement by interference fringe projection: a technical validation J.M. Lagarde, C.Rouvrais, D.Black, S.Diridollou and Y. Gall. Skin Research and technology 2001; 7: 112-121.
- Linee Guida EEMCO : Valutazione della Topografia Cutanea JL Léveque Cosm Technol 2000 3[5] 11-20.
- EEMCO guidance for the in vivo assessment of tensile functional Properties of the Skin. Skin Pharmacol Appl Skin Physiol 2001; 14: 52-67
- E.Berardesca EEMCO Guidance for the assessment of stratum corneum hydration: electrical method Skin Research and Technology 1997; 3: 126-132
- KP Wilhelm: Misurazioni dell' idratazione cutanea. Considerazioni generali e potenziali difficoltà Cosmetic Technology 1999; 2 [4].
- V. Rogiers, M.P. Derde, G.Verleye : Standardized Condition Needed for skin Surface Hydration Measurement 1990 vol.105: 73-81.
- Council of Europe: Recommendation N°R(90)3, adopted 4th February 1990.
- OECD (1998): development of OECD Test Guidelines for Use in Tests with Human Volunteers; ENV/MC/CHEM/RD (98) 27th Joint Meeting of the Chemicals Group and Management Committee, 11-13 Feb 1998.

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