

Studies on Cosmetics and Drugs

Dobrev H. Clinical and instrumental study of the efficacy of a new sebum control cream. *Journal of Cosmetic Dermatology* 2007 (in press)

Background: Some botanical compounds are considered useful to reduce sebum production.

Aim: To evaluate the efficacy of a sebum control cream containing polyphenol-rich extract from saw palmetto, sesame seeds and argan oil in subjects with oily facial skin.

Methods: The study was carried out during the winter months (January and February). A total of 20 healthy volunteers (9 male and 11 female, aged 17-50 years, 16 with oily skin and 4 with combined skin) were studied. The test product was applied twice daily to the face for a period of 4 weeks. A clinical assessment and instrumental measurements were performed before and after the treatment period. Casual sebum level on the forehead and both cheeks was determined with a photometric device (Sebumeter). The quantity of sebum on the mid forehead was determined using sebum collector foils (Sebufix), which were then evaluated with skin camera Visioscope and software SELS (Surface Evaluation of the Living Skin). A subjective evaluation questionnaire regarding the cosmetic characteristics, tolerance and efficacy of the product was filled out by the volunteers at the end of study.

Results: The product was very well accepted by all the volunteers. A visible sebum regulating efficacy was reported in 95% of them. After 4 weeks of treatment, the clinical assessment scores decreased by 33%. There was a significant reduction in the casual sebum level by 20% and area covered with oily spots by 42%. The number of active sebaceous gland remained unaltered.

Conclusion: These results objectively and quantitatively demonstrate the efficacy of the sebum control cream tested to reduce the greasiness and improve the appearance of oily facial skin.

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Dobrev H. Treatment of acne with a new topical preparation. Clinical and instrumental study. 15th Congress of the European Academy of Dermatology & Venereology (EADV), October 4-8, 2006, Rhodes, Greece.

Background: Sepicontrol A5 is a cosmetic active ingredient designated to improve the appearance of oily, acne prone facial skin.

Aim: To evaluate the sebum regulation activity, clinical efficacy and safety of a 3% and 4% Sepicontrol A5 containing cream and gel in subjects with mild to moderate acne.

Methods: A total of 19 volunteers were enrolled in this open-labelled study. The test cream and gel were applied to the face and acne lesions respectively, twice daily for a period of 7 weeks. Counting of acne lesions and acne score method were used for clinical assessment before and after 2, 4 and 7 weeks of treatment. Casual sebum level on the forehead and cheeks was determined with a photometric device (Sebumeter). The activity of sebaceous gland on the forehead was recorded using sebum collector foils (Sebufix), which were then evaluated with skin camera Visioscope and software SELS. In addition, subjective evaluation questionnaire regarding the physical characteristics, tolerance and efficacy of the products and quality of life questionnaires were filled in by the volunteers.

Results: Both products were well tolerated and accepted. A visible improvement was reported in 89% and good to very good clinical response was evaluated in 84% of the subjects. The count of acne lesions decreased significantly by 59% (non-inflammatory - 61%, inflammatory -55%) and the mean clinical assessment scores decreased by 33%. There was a significant reduction in the casual sebum level by 12% and the area covered with oily spots by 27%. The DLQI and CADi mean scores decreased by 46 and 41% respectively.

Conclusion: The offered topical products successfully improve the state of oily acne prone facial skin and raise quality of life of the patients.

Acknowledgements: The author would like to thank Courage+Khazaka, Cologne, Germany for supplying the skin camera Visioscope, tests Sebufix and software SELS, and Solvex Cosmetic Products, Plovdiv, Bulgaria for supplying the preparations.

Dobrev H. The effects of topically applied Matrixyl, natural grape seed and avocado oils on skin surface, hydration and elasticity. III. Spring Symposium of the European Academy of Dermatology and Venereology (EADV), May 19 – May 21, 2005, Sofia, Bulgaria. Book of Abstracts: p73. (P05.4)

Background: Matrixyl is a lipophilic pentapeptide that stimulates the collagen synthesis by fibroblasts in the skin. The grape seed extract is rich in flavonoids which are powerful antioxidants. Avocado oil consists predominantly of unsaturated fatty acid glycerides, vitamins and minerals, and has good emollient properties.

Aim: To evaluate the effects of two 1% Matrixyl, 1% grape seed oil and 2% avocado oil containing creams on aged facial skin using *in vivo* skin bioengineering techniques.

Methods: Measurements were performed on 30 healthy female volunteers (aged 37 to 64 years) before and after once-daily applications for 2 months. Fifteen of the volunteers applied a day cream in the morning and the rest ones - a night cream in the evening. Images of the skin surface at eye corners were obtained with video camera Visioscope and then analyzed with the software SELS. Epidermal hydration and skin mechanical properties at five anatomic regions (forehead, eye corners and cheeks) were measured with a skin capacitance meter (Corneometer) and a suction device (Cutometer). In addition, a subjective evaluation questionnaire regarding the organoleptic characteristics, tolerance and efficacy of the products was given to the volunteers.

Results: The products tolerability and cosmetic properties (consistency, spreadability) were evaluated by 93-100% of the volunteers as “good” to “very good”. The products efficacy (improvement of skin flexibility, anti-wrinkle, smoothing and moisturizing effects) was rated in 67-100% of the volunteers to be “very good” or “good”. The results showed a significant increase in the average epidermal water content (day cream +10%, night cream +11%) and Ur/Uf, skin biological elasticity, (day cream +23%, night cream +16%), while the Uv/Ue, viscoelastic to elastic ratio, was reduced (day cream -3%, night cream -17%). The treatment significantly reduced the skin surface parameters volume (day cream -7%, night cream - 8%), SEr, roughness (day cream -5%, night cream -12%) and the micro relief parameters Rt, Rz, Rm, Ra, Rp (day cream - 8%, night cream -13%), which are indicators for the number, width and depth of the facial wrinkles.

Conclusion: The association of Matrixyl, natural grape seed and avocado oils improves the hydration, elasticity and surface appearance of the aged facial skin. The marked reduction of skin viscoelasticity by night cream suggests the more pronounced stimulating effect on the collagen synthesis compared with day cream.

Acknowledgements: The author thanks Courage+Khazaka Electronic, Köln, Germany for supplying with the Cutometer MPA 580, camera Visioscope and software SELS, and AROMA, Sofia, Bulgaria for supplying with the preparations.

Dobrev H. Evaluation of the efficacy of a Rooibos extract containing antiwrinkle cream. III. Spring Symposium of the European Academy of Dermatology and Venereology (EADV), May 19 – May 21, 2005, Sofia, Bulgaria. Book of Abstracts: p84. (P08.2)

Background: Rooibos plant possesses scientifically proven anti-oxidative, anti-allergic, anti-microbial and anti-inflammatory features.

Aim: To evaluate the efficacy of a Rooibos extract containing cream on aged facial skin using *in vivo* skin bioengineering techniques.

Methods: Measurements were carried out on 21 healthy female volunteers (from 35 to 63 years old) before and after twice-daily applications for 4 weeks. Images of the skin surface at eye corners were obtained with video camera Visioscope and then analyzed with the software SELS (Surface Evaluation of the Living Skin). Skin mechanical properties on five anatomic regions (forehead, eye corners and cheeks) were measured with a suction device Cutometer SEM 474. In addition, a subjective evaluation questionnaire regarding the organoleptic characteristics, tolerance and efficacy of the product was given to the volunteers.

Results: The product tolerability and cosmetic properties (consistency, spreadability) were evaluated by 95% of the volunteers as “good” to “very good”. A visible anti-wrinkle efficacy was reported in 76% of the volunteers. The results showed a significant increase in all mechanical parameters of the skin: Uf, skin distensibility (+7%), Ua/Uf, brut-elasticity (+6%), Ur/Ue, net-elasticity (+23%), Ur/Uf, biological elasticity (+12%) and Uv/Ue, skin viscoelasticity (+21%). The application of a Rooibos containing cream significantly reduced the skin surface parameters SEr, roughness (-13%), SEw, wrinkles (-5%) and the micro relief parameters Rt, Rz, Rm, Ra, Rp (an average reduction of 10%), which are indicators for the number, width and depth of the facial wrinkles.

Conclusion: These results demonstrate with quantitative and objective methods the efficacy of a Rooibos containing cream to improve skin mechanical properties and surface appearance of the aged facial skin.

Acknowledgements: The author thanks Courage+Khazaka Electronic, Köln, Germany for supplying with the camera Visioscope and software SELS, and Bodi-D, Plovdiv, Bulgaria for supplying with the preparation.

Dobrev H. Use of Cutometer to assess epidermal hydration. *Skin Res Technol* 2000, 6 (4): 239-244.

Background/aims The Cutometer equipped with 2-mm diameter suction probe is a device suitable for assessment of epidermal mechanics. The objectives of this study were to determine the sensitivity of cutometer parameters to epidermal hydration, to evaluate the correlation between skin mechanical parameters and skin capacitance, and to study the role of pretension of the skin.

Methods In the first study, skin capacitance (Corneometer) and 10 mechanical parameters were determined before and 60 min after application of 5 different moisturizers on volar forearm. In the second study, measurements of epidermal

mechanics were made without and with 0.1 sec presuction of the skin, before and 120 min after application of petrolatum and glycerin.

Results Hydration of the skin significantly increased the values of capacitance and all rheological parameters. Delayed distension (U_v) and viscoelastic to elastic ratio (U_v/U_e) were the most sensitive cutometer parameters. Significant correlations between the skin capacitance and mechanical parameters were not found. Pretension of the skin did not significantly influence the values of skin mechanical parameters.

Conclusions The non-invasive measurements of skin elasticity are appropriate for an objective and quantitative evaluation of the complex effect of different dermatological and cosmetic products on skin mechanics and hydration.

Dobrev H. Changes in the water content of epidermis following single application of various humidifying agents. *Dermatol Venereol (Bulgaria)* 1997, 36 (2): 11-13.

Effects of single application of pure petrolatum, paraffin oil, glycerin, water-in-oil emulsion (65% lipids) and oil-in-water emulsion (25% lipids) were studied on the volar forearm skin of 20 healthy volunteers. The epidermal hydration was determined with a skin capacitance meter (Corneometer) and values were followed for 120 min. Five minutes after the application of glycerin and O/W emulsion the skin capacitance showed an increase of 54% and 16% as compared with the initial values and slightly decreased after 2 hours to 35% and 12%, respectively ($p < 0.001$). The application of W/O emulsion, paraffin oil and pure petrolatum caused an initial significant reduction of the capacitance, a recovery of the original levels after 30 to 60 min. and an increase of 11-14% after 2 hours ($p < 0.001$). The results illustrated the different effect of the moisturizers tested and the influence of their non-absorbed components on the capacitance values.

Dobrev H. Evaluation of the inhibitory activity of topical indomethacin, betamethasone valerate and emollients on UVL-induced inflammation by means of non-invasive measurements of the skin elasticity. *Photodermatol Photoimmunol Photomed* 2001, 17 (4): 184-188.

Background/Aim Topical indomethacin has been reported to inhibit ultraviolet light-induced erythema. The objective of this study was to verify this assertion and to compare indomethacin 10% ointment to betamethasone valerate 0.1% ointment, water-in-oil emulsion and oil-in-water emulsion by means of non-invasive skin elasticity measurements.

Material/Methods Products were applied on the back skin 60 min and 5 min before and 5 min after UVL irradiation. Untreated test sites served as controls. Clinical evaluation, measurements of epidermal hydration (Corneometer) and mechanical properties of the skin (Cutometer) were made 1 h before and 24 h after exposure.

Results Test areas treated with indomethacin 60 min and 5 min before irradiation showed the significantly lowest visual erythema scores and no significant changes in skin mechanical parameters. At all other test sites, a significant decrease in elasticity parameters (U_e , U_r , U_a/U_f , U_r/U_f) and an increase in viscoelasticity parameters (U_v , U_v/U_e) of the skin were observed. No significant changes of epidermal hydration were found at any of the test sites.

Conclusion The inhibitory action of topical indomethacin on UVL-induced inflammation is superior to betamethasone valerate and emollients. Non-invasive measurement of skin

elasticity could be used as a supplementary tool for objective evaluation and comparison of the photoprotective activity of different topical agents.

Savova J., H. Dobrev, R. Yankova. Evaluation of the photoprotective activity of indomethacin. *Dermatol Venereol (Bulgaria)* 1995, 34 (1), 10-12.

The sun protection factor (SPF) of 10% indomethacin ointment, topically applied prior to exposure to ultraviolet light, is evaluated in a group of 20 persons, using a modification of the well know procedure. Analogical study is carried out on petrolatum ointment and on the so-called cold cream. A considerably higher photoprotective activity of indomethacin (SPF=11.08), as compared to the lower and virtually equivalent activity of the both vehicles (SPF=3.41 and 3.13), is noted. Proceeding from the obtained results, indomethacin is recommended for use as an active local sun protective agent.

Dobrev H. In vivo noninvasive study of the mechanical properties of human skin after a single application of topical corticosteroids. *Folia Med (Plovdiv)* 1996, 38 (2): 11-17.

A noninvasive in vivo suction device for measuring skin elasticity was used to determine the alteration in skin mechanical properties 60 min after an application of 5 commercially available topical corticosteroids as ointments and creams. 25 healthy volunteers (20 females and 5 males, aged 16-55 years, mean age 36 years) were studied. The following relative parameters, independent of skin thickness, were calculated and compared: R2 - gross, R5 - net, R7 - biologic elasticity and R6 - viscoelastic/elastic ratio. Corticosteroid ointments increased the purely elastic parameters R2, R5 ($p < 0.05$) and R7 as well as the parameter of viscoelasticity R6 ($p < 0.001$). Corticosteroid creams altered the elastic parameters weakly and increased the viscoelastic parameter R6 significant ($p < 0.05$). These effects of topical corticosteroid formulations were probably due to the stratum corneum hydration and softening of keratin that improves the epidermal mechanical properties. Moreover, as a result of dermal oedema, the friction between the fibres was reduced and interstitial fluid movement was facilitated through the fibrous network. The noninvasive method applied can be useful for an evaluation and comparison of the effect of different topicals on the elastic and viscoelastic properties of the human skin.

Yankova R., **H. Dobrev**, J. Savova. Photo-conditioned activities of some topical medicines. *International Meeting on Clinical Dermatology - Hot line.* 28-31. 03. 1996, Athens, Greece. Poster 23. Abstracts - p. 148.

We have termed "photo-conditioned", either sensitizing or protective, those characteristics of chemicals applied on the human skin, which are influenced by solar irradiation. In this connection five commercially available preparations: Indomethacin 10% ointment (Pharmachim) (IM), sulfanilic acid 5% in high density emulsion base (Ombroderm, Sopharma) (OD), myconazole 2% plus benzoyl peroxide 5% (Acnidazil cream, Janssen) (AD), tretinoin 0.025% (Acnoten gel, KRKA) (AT) and 0.05% (Retin A cream, Cilag) (RA) were examined for their sun protection factor (SPF) values. Fifty volunteers of skin types I, II and III took part in the essay. The SPF's were evaluated through a modification of the standard method, introduced by R. Yankova (1993). IM showed a high ability to prevent erythema (SPF 7.78; SD 0.85), far distant from OD (SPF 3.83; SD 0.93). Tretinoin both

0.025% and 0.05% did not promote any photoreactivity in the areas treated with AT (SPF 2.20; SD 1.15) and RA (SPF 2.08; SD 1.18). It did not differ from the photo-conditioned action of AD either (SPF 2.44; SD 1.10; $p>0.05$). The data indicate a potential of IM for preventing photodamage. Likewise the results of this study demonstrate a lack of photo-induced erythema by tretinoin.

Dobrev H. Study of human skin mechanical properties by means of cutometer. [Review] *Folia Med (Plovdiv)* 2002, 44 (3): 1-5.

Aim Cutometer is a commercially available non-invasive suction skin elasticity meter. The purpose of the present article is to discuss some aspects of the biological informativeness and interpretation of the results obtained under studying of skin mechanical properties with cutometer.

Methods/Results Results of previously published in the literature studies on the mechanical properties of healthy and diseased skin and their changes after external influences are analyzed

Conclusion The cutometer method of investigation gives an objective and meaningful information on the mechanical properties of healthy and diseased human skin.

Dobrev H. Impact of three different emulsions on skin hydration and elasticity. *II. Spring Symposium of the European Academy of Dermatology and Venereology (EADV), April 29 – May 1, 2004, Budapest, Hungary. J Eur Acad Dermatol Venerol* 2004, 18, Suppl.1: 53.

Background/aim Skin care products are designated to improve skin hydration and elasticity. The objective of this study was to evaluate the moisturizing and plasticising effects of three different emulsions after single application.

Material/methods Effects of two oil-in-water emulsions and a water-in-oil emulsion (Excipial) were studied on the volar forearm skin of 20 healthy volunteers. Epidermal hydration was determined using skin capacitance meter (Corneometer). Mechanical properties of the skin were evaluated using non-invasive suction device (Cutometer). Measurements were made before the application and 10, 30, 60, 90 and 120 minutes later.

Results Ten minutes after the application of oil-in-water emulsions the skin capacitance showed an increase of 32% and 29% compared to initial values and decreased after 2 hours to 26% and 17%, respectively ($P<0.001$). The application of water-in-oil emulsion gradually increased skin capacitance with a maximum of 34% after 1 hour followed by minimal decrease to 31% after 2 hours ($P<0.001$). All three emulsions significantly increased skin viscoelasticity (Uv/Ue), while the water-in-oil emulsion significantly increased the distensibility (Uf) and elasticity (Ua/Uf) of the skin, too.

Conclusion The results confirm the favorable effects of tested skin care products. Values of skin capacitance are influenced by water content and non-absorbed components of the emulsions applied.

Dobrev H. Биоинженерни методи за обективизиране на ефектите на козметичните продукти върху кожата. [Bioengineering methods for documentation of the cosmetic products effects on the skin] *7th National Congress of Dermatology and Venereology*, Sofia (Bulgaria), May 11-13, 2000. Abstracts, pp 31.

Dobrev H. Промени в кожната еластичност след приложение на емулсии съдържащи урея и алфахидрокси киселини. [Changes in skin elasticity after an application of emulsions containing urea and alphas hydroxy acids] **7th National Congress of Dermatology and Venereology**, Sofia (Bulgaria), May 11-13, 2000. Abstracts, pp 31-32. (Abstract)

Dobrev H. Оценка на въздействията на козметични продукти чрез измерване на водното съдържание на епидермиса и остатъчния липиден филм върху кожната повърхност. [Assessment of cosmetical products by means of measurements of the epidermal water content and residual lipid film on the skin surface] **7th National Congress of Dermatology and Venereology**, Sofia (Bulgaria), May 11-13, 2000. Abstracts, pp 20. (Abstract)

Dobrev H. Непосредствени ефекти на мъжка козметична серия “Каро роял” върху водното съдържание и киселинното равновесие на кожата. [Immediate effects of cosmetic series for men “Karo Royal” on the skin water content and pH] **7th National Congress of Dermatology and Venereology**, Sofia (Bulgaria), May 11-13, 2000. Abstracts, pp 21. (Abstract)