Topical effects of a novel blend of natural products (DermaStem™) on human skin.

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Objective

The objective for this research was to document the effects of DermaStem™on specific modes of action on primary human dermal fibroblasts and inflammatory cells in vitro, and on hydration status, skin elasticity, and wrinkle reduction in healthy human facial skin in vivo.

Background

Skin health and protection from premature ageing associated with oxidative stress, inflammation, and reduced stem cell repair, is a complex interplay of different biological functions. The healthy proliferation and migratory capacity of dermal fibroblasts, their matrix deposition, and protection from damage by free radicals, are important factors in prevention of skin ageing and loss of elasticity, that leads to dry and wrinkled skin.

The aim in the development of DermaStem™ was to identify natural compounds that would have an effect on the proliferation and differentiation of dermal cells, and would therefore support the actual restructuring of the skin, leading to greater moisture retention, greater elasticity, and consequently a reduction in fine lines and wrinkles.

DermaStem™

The formulation for DermaStem™ was based on selected ingredients proven to have effects on human stem cell biology [Jensen et al., 2007; Irhimeh et al., 2007; Bush et al., 2010; Yang et al., 2011]. These key ingredients were combined with a broad range of botanical ingredients and extracts, each separately having a longstanding traditional use in rejuvenation and repair.

In vitro testing of ingredient properties on human dermal fibroblasts

A panel of in vitro tests was performed to document effects on primary adult human dermal fibroblasts:

- Support of skin cell proliferation;
- Matrix deposition (collagen production).

Multiple ingredients in DermaStem™ supported the proliferation, migration, and matrix deposition of primary human dermal fibroblasts.

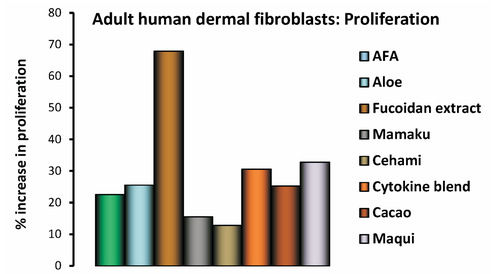

Figure 1. *Aphanizomenon flos-aquae* (AFA), Aloe vera, fucoidan extracted from *Undaria pinnatifida*, Cyathea medullaris (Mamaku), *Centipeda cunninghamii* (Cehami), cytokine blend, *Theobroma cacao* (cacao), and *Aristotelia chilensis* (Maqui) berry significantly increase dermal fibroblast proliferation. Vanilla bourbon and colostrum did not significantly increase dermal fibroblast proliferation on their own but synergistically potentiated the effect of AFA. A blend of cytokines (growth factors) including epidermal growth factor, fibroblast growth factors, keratinocytes growth factor, hepatocyte growth factors and Stem Cell Factor led to substantial increase in dermal fibroblast proliferation above baseline.

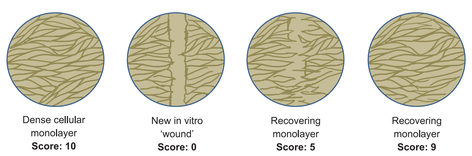

Diagram: An in vitro cellular bioassay was used as a model for wound healing, where adult human dermal fibroblasts were cultured until they formed a dense monolayer. A scratch was created through the monolayer. The cell cultures were treated with different DermaStem™ ingredients, and the recovery followed for the next 36 hours.

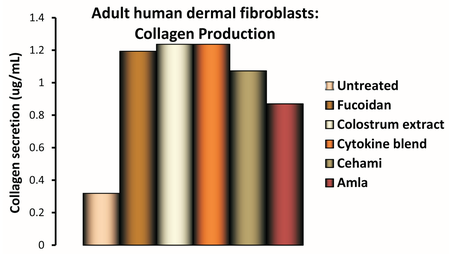

Figure 2. Collagen production by primary adult human dermal fibroblasts in culture was increased by fucoidan, bovine colostrum extract, cytokines, Cehami, and Amla.

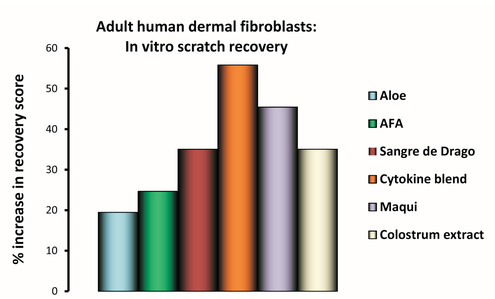

Figure 3. The effects of ingredients on in vitro scratch recovery (a cellular model for wound healing) was examined. A blend of cytokines (growth factors) including epidermal growth factor, fibroblast growth factors, keratinocytes growth factor, hepatocyte growth factors and Stem Cell Factor led to substantial increase in dermal fibroblast migration and accelerated recovery of the in vitro scratch. In addition, *Aloe vera, Aphanizomenon flos-aquae* (AFA), Sangre de Drago, Maqui, and bovine colostrum extract supported accelerated recovery.

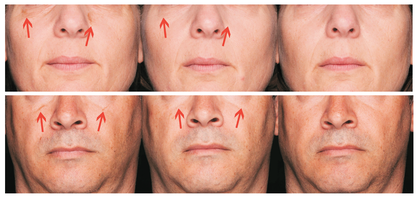

Figure 4. Examples of rapid changes in wrinkle depth, darkness under eyes, and skin hydration.

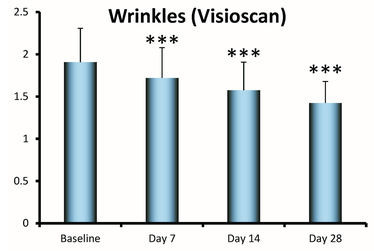

Figure 5. DermaStem™ demonstrated dramatic decreases in the Visioscan parameters of surface roughness associated with the depth of fine and coarse wrinkles. The reductions below baseline were highly significant already at 7 days ($p<0.001$), and remained highly significant at 14 and 28 days of product use (as indicated by the triple asterisks) ($p<0.02$). The average reductions were 9.8%, 17.4% and 25.3 % after 7, 14 and 28 days of use, respectively, with maximum % improvement reaching 39.3%.

Human clinical testing on skin health

In an open-label study, 10 individuals (5 males and 5 females) used DermaStem™ twice daily for 28 days. Measurements of skin elasticity and moisture along with surface skin analysis, were performed at 0, 7, 14, and 28 days.

Methodology

- An Institutional Review Board (IRB) approval was obtained for an open-label 4-week study on 10 people (5 males and 5 females), 35-50 years of age;
- DermaStem™ was applied twice daily on delicate areas of the face and full facial skin;
- Wrinkle reduction was assessed by surface evaluation of living skin, conducted instrumentally using a Visioscan image analysis system;
- Assessment of elasticity and viscoelastic properties of the skin were measured as a function of flexibility and firmness using a cutometer;
- Hydration of the skin was measured using a Nova Dermal Phase meter.

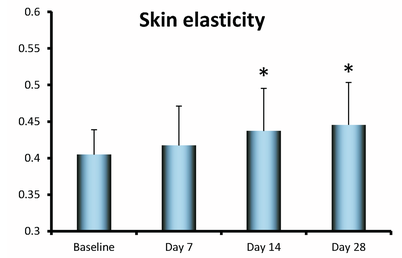

Figure 6. Evaluation of skin elasticity/flexibility via Cutometer indicated an increase in biological elasticity on the test sites treated with the test product. The increases were statistically significant from baseline after 14 and 28 days of use (as indicated by the asterisks), and averaged a 10.0% increase in elasticity, with maximum % improvement reaching 31.9%.

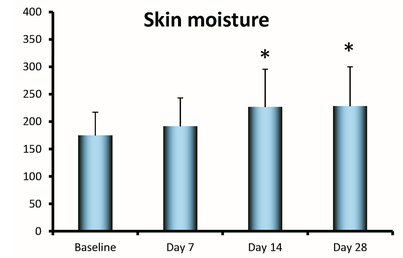

Figure 7. Novameter readings demonstrated that the test product dramatically increased the skin moisture content. The increases were statistically significant from baseline after 14 and 28 days of use (as indicated by asterisks) with average increases of 29.7% and 30.5%

Antioxidant and anti-inflammatory properties

Additional in vitro bioassays using primary human blood cells were used to document:
• Cellular antioxidant protection under oxidative stress (CAP-e test); • Inhibition of free radical formation by inflammatory cells.

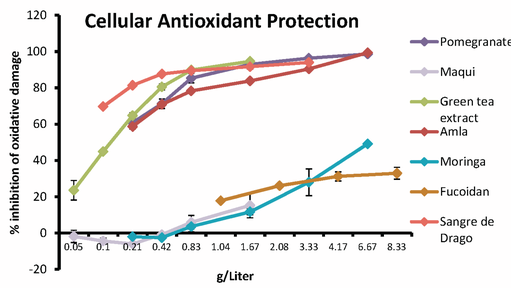

Figure 8. The cellular antioxidant protection capacity of each ingredient in DermaStem™ was tested in the CAP-e bioassay. The in vitro data showed potent antioxidant bioavailability at the cellular level by Indian Gooseberry (Amla), pomegranate, Sangre de Drago, and green tea extract.

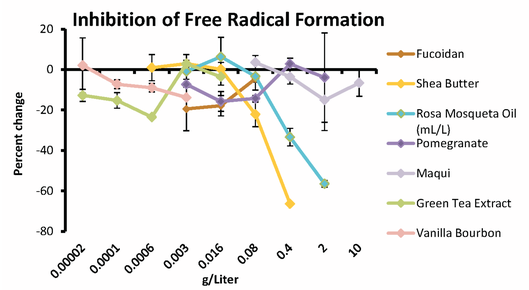

Figure 9. DermaStem™ ingredients were evaluated for the ability to inhibit free radical formation by inflammatory cells placed under oxidative stress conditions. Reduced free radical production by ingredients in DermaStem™ included fucoidan extract, Shea butter, Rosa mosqueta, Maqui, pomegranate, Vanilla bourbon, and green tea extract.

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