Anti-ageing properties of phytoglycogen

KEYWORDS: Phytoglycogen, anti-aging, skin care, skin penetration, cellular growth.

INTRODUCTION

Glycogen, a branched glucose polysaccharide, is one of the body’s primary sources of stored energy. It is naturally found in the tissues of most living organisms and continuously undergoes a cycle of degradation and synthesis as required (Figure 1) through a regulated process involving several enzymes and hormones (1, 2).

Although most of the body’s glycogen is stored in the liver and muscles, a small amount has been found in the epidermis. The same enzymes that synthesize and break down glycogen internally are also found in the skin, although their activity and regulation differs (3, 4). In a study where the concentration of the end products of glycogen metabolism were determined in the skin, it was found that the majority of glucose is degraded to lactic acid (~75%) through glycolysis (1, 5, 6).

The glycogen concentration in the epidermis has been measured to be 0.465 µg/mg of epidermis and remains largely the same over time. However, the concentration increases when the epidermis is stressed, such as when sunburned or stripped, and has also been found in higher quantities in psoriasis lesions. These findings suggest the involvement of glycogen in cell growth and wound healing (5, 7).

Phytoglycogen is chemically identical to the glycogen stored in animal cells, but is produced and stored in plants. Phytoglycogen isolated from non-GMO sweet corn (PhytoSpherix®, Mirexus Inc.) is in the form of monodisperse, 70 nm diameter particles (measured by dynamic light scattering). Other commercial sources of glycogen include purified marine glycogen (Glycoenergizer, Cobioba; Dermosaccharides® GY, Laboratoires Sérobiologiques S. A.), and a synthetic analogue, produced enzymatically from starch (Bioglycogen™, Glico Nutrition Co., Ltd.) (8-10). Phytoglycogen particles are hydrophilic and can tightly bind water to enhance the moisturizing properties of skincare formulations (11).

Beyond this activity, the cellular breakdown of phytoglycogen in the skin has the potential to provide anti-aging benefits in topical applications. The aim of this study was to investigate the effect of phytoglycogen on the skin and evaluate its potential as an anti-aging ingredient for cosmetics applications.

MATERIALS AND METHODS

Materials

Phytoglycogen studies were performed using PhytoSpherix® (Mirexus Inc., distributed by Active Box SRL in Italian market**).

Cellular uptake and response

To investigate the impact of phytoglycogen on the growth rate and function of skin cells, the response of human fibroblasts (HFF-1) was studied. 500 µg/ml phytoglycogen was added to growth medium containing 0.2 g/L glucose and the growth of cells was compared to those grown under the same conditions but in medium containing no phytoglycogen, replaced with an equal amount of sodium pyruvate. After 36 hours of incubation, the proliferation rate, collagen production, and hyaluronic acid (HA) production of cells grown under both conditions were measured.

Cell proliferation was evaluated by manual counting using a hemocytometer. An enzyme linked immunosorbent (ELISA) assay was used to quantify the amount of HA produced. Collagen production was assessed by immunohistochemistry (using an anti-type collagen 1 antibody) where fluorescence intensity correlated with collagen quantity.

Skin penetration

Skin penetration studies were performed using full thickness human breast skin, obtained with permission from donors undergoing mammoplasty surgery. Tissues were collected within 2 hours of surgery and stored at -20 °C until use. A set of 9 mm diameter Bronaugh-type Teflon flow-through diffusion cell (Permagear, Inc., Hellertown, PA) were used for the skin penetration study. Phosphate buffered saline (pH 7.2) was used as the receptor fluid and maintained at 37 °C.

The skin samples were mounted onto the diffusion cell and a set of 9 mm diameter Bronaugh-type Teflon flow-through diffusion cell (Permagear, Inc., Hellertown, PA) were used for the skin penetration study. Phosphate buffered saline (pH 7.2) was used as the receptor fluid and maintained at 37 °C. The skin samples were mounted onto the diffusion cell and a solution of 0.2% fluorescently tagged phytoglycogen...
(with Rhodamine-B) was administered to the upward facing epidermis. The samples were covered with a Teflon cap to prevent dehydration, then incubated for 24 hr, followed by a thorough rinse with water and tape stripped 2x to remove surface bound residue, or 10x to remove the stratum corneum layer.

Penetration of the fluorescently labelled phytoglycogen was evaluated using a Zeiss LSM 710 confocal microscope. Samples were cryosectioned with a Leica CM1850 cryostat to expose a cross-sectional surface before imaging. The fluorescence intensity profile was used to estimate the penetration depth of phytoglycogen.

**Clinical trials**

The anti-aging effect of phytoglycogen on the appearance of the skin was assessed in a double-blind clinical trial conducted by Thomas J. Stephens & Associates (Richardson, TX). Groups of female volunteers, 49-70 years of age, were randomly assigned to one of three treatment groups: a base cream containing no phytoglycogen (placebo), the same base cream with 0.1% phytoglycogen, or 0.3% phytoglycogen. Base formulations contained water, glycerin, prunus amygdalus (almond) oil, persea gratissima (avocado) oil, butyrospermum parkii (shea) butter, cetyl alcohol, dimethicone, sorbitan stearate, stearyl alcohol, xanthan gum, phenoxyethanol, sorbic acid, capryly glycol, tocopherol acetate, and triethanolamine. Each group consisted of 30-32 volunteers (93 total volunteers in the study). Following a one week washout period and baseline assessment, each group was asked to apply the test formulation to their entire face twice daily for 6 weeks. Volunteers returned to the test site after 2 weeks and 6 weeks for a follow-up assessment. At these visits, an expert grader evaluated their skin condition in nine categories based on a 10 point scale: overall appearance, fine lines, coarse wrinkles, smoothness/roughness (tactile), smoothness/roughness (visual), firmness (visual), global hyperpigmentation, evenness of skin tone, and clarity. A binomial sign test was used to test for statistical significance.

**RESULTS AND DISCUSSION**

**Cellular uptake and response**

The results of the cellular response study are shown in Figure 2. Cell proliferation was found to increase by 20% when cells were grown in the presence of phytoglycogen compared to the control. HA and collagen production were also found to increase by 83% and 60% respectively. These characteristics suggest that phytoglycogen may provide an anti-aging benefit to the skin, as cellular growth rate becomes slower and HA and collagen production decrease with age (12-14).

**Skin penetration**

The skin is a barrier between the body and the outside environment. It selectively allows substances across but is generally very difficult to penetrate. The skin is comprised of layers, with the stratum corneum being the outermost layer made up of dead skin cells and the limiting factor in penetration (in humans, ~10-20 µm thickness) (15-17). Below this is the epidermis (~60-150 µm thickness) and dermis (<1-5 mm thickness), which is vascularized (15). Living cells reside in the dermis and are gradually keratinized as they are pushed towards the surface by new cells growing below them (16). In order for active ingredients to realistically impact the health of the skin, they must be able to penetrate to the dermis layer where they can affect living cells.

Figure 3 shows the fluorescence intensity profile of the incubated skin sample. The maximum penetration depth is estimated to be about 150-170 µm based on the length scale, which indicates penetration through the stratum corneum and into the dermal layer of the skin.

**Clinical trials**

The average improvement between the baseline and week 6 for each of the treatment groups in all expert grading categories is shown in Figure 4. An example of the visible improvement in skin condition is given in Figure 5. To determine the statistical significance of these results, a binominal signed test was used. When all nine grading categories are considered together, both the 0.1% and 0.3% outperformed the placebo in all evaluated categories, corresponding to an improvement in performance with a p value of 0.004, a statistically significant result.

A comparison between 0.1% and 0.3% phytoglycogen treatments indicated that the 0.3% group outperformed the 0.1% group in 8 of the 9 categories, indicating a more effective dose (p value of 0.02). From these results it was determined that both the 0.1% and 0.3% phytoglycogen creams improved the condition of the skin compared to the placebo, and that the 0.3% treatment performed better than the 0.1% treatment after 6 weeks. A summary of these results are shown in Table 1.
CONCLUSION

Phytoglycogen is a safe and natural material that promotes the skin’s health and enhances its appearance. The important role of glycogen in cell growth, wound healing, and moisture retention has been known for many years, but new data demonstrates its potential as an anti-aging ingredient for personal care products. Clinical data indicates that visual improvements can be seen in 6 weeks, resulting in an improvement in overall appearance, fine lines, coarse wrinkles, global hyperpigmentation, smoothness/roughness (visual), smoothness/roughness (tactile), firmness (visual), evenness of skin tone, and clarity. The mechanism of action can be inferred from complementary in vitro studies, which show that topically applied phytoglycogen can penetrate the skin to the dermis, where living cells are able to take up the material and use it to promote cell proliferation, HA production, and collagen production – all of which act to improve the health of the skin.

REFERENCES AND NOTES

**Active Box SRL: www.activebox.it


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