Stimulating endorphins and sex hormones in the skin

Daniel Schmid, Franziska Wandrey, Fred Zülli - Mibelle Biochemistry, Switzerland

Abstract

The peptide hormone beta-endorphin and the steroid hormone dehydroepiandrosterone (DHEA) play important roles in the skin. The fact that they are also synthesised locally in the skin makes them interesting targets for cosmetic ingredients. An extract of monk's pepper (*Vitex agnus-castus*) was found to perform like beta-endorphin and to stimulate the synthesis of DHEA. In a clinical study, the extract showed significant effects on skin elasticity and density.

DHEA is the precursor for both sex hormones, the female oestrogen as well as the male ones, testosterone and dihydrotestosterone. Circulating DHEA is mainly produced by the adrenal glands. Production is highest when we are in our thirties but then starts to decline leading to the so called 'adrenopause' which occurs in both sexes. In the peripheral tissues, DHEA is transformed to the final sex hormones, depending on the tissue's needs. Oestrogen plays a very important role in skin health and skin ageing. It stimulates the production of collagen and elastin and inhibits the breakdown of the existing fibres. Declining oestrogen levels therefore result in laxity of the skin and a decreased general skin tonicity which leads to sagging and wrinkles. In men, the decrease of the testosterone level results at the skin level in a loss of skin density and in lower elasticity. For a couple of years it has been known that the skin not only represents a target for circulating DHEA, oestrogen, and testosterone but has itself the capacity to produce from the lipid precursor cholesterol the steroid hormone DHEA and all the final sex steroids.[1,2]

Beta-endorphin is a neuropeptide because it is mainly produced in the neurons of the central and peripheral nervous system. Beta-endorphin messengers are agonists of the mu opioid receptor and are used to regulate reactions to stress and pain as well as feelings of euphoria. In the skin the beta-endorphin/mu opioid receptor system is involved in nociception and reactions to inflammation. Beta-endorphin and the mu opioid receptor were found to be expressed in the skin not only in sensory neurons but also in keratinocytes, fibroblasts as well as melanocytes.[3,4,5] There are scientific publications demonstrating a role of beta-endorphin and its receptor in wound healing and skin regeneration.[6]

Here, we present literature data and own experiments which demonstrate a beta-endorphin-like effect as well as stimulation of the synthesis of DHEA by an extract of monk's pepper.

Type of opioid receptor		
mu	kappa	delta
36	22	194

Figure 1: Competition between beta-endorphin and an extract of *Vitex agnus-castus* in binding to opioid receptors. The smaller the indicated IC50-values (µg/ml) the stronger the binding of the extract to the receptor.

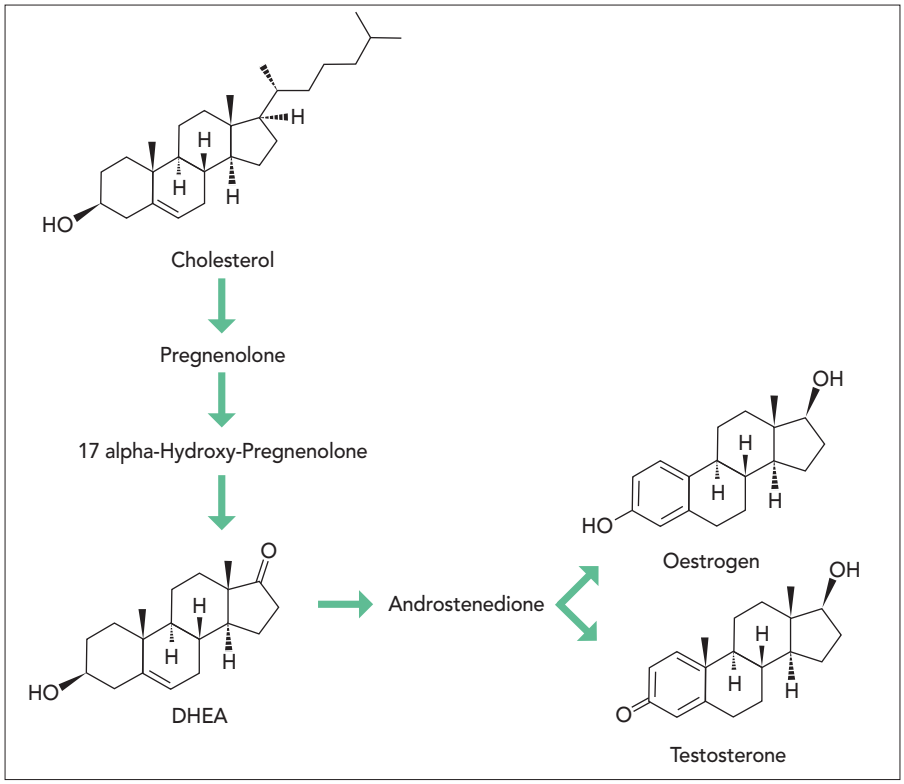

Figure 2: The biosynthesis of the sex hormones from the cholesterol precursor.

Results and discussion

Chaste tree (*Vitex agnus-castus*), also known as monk's pepper, is a large shrub native to the Mediterranean area that produces aromatic berries with a bitter taste. Dried chaste tree berries have been used until now as a pepper substitute and as herbal medicine to treat disorders of the

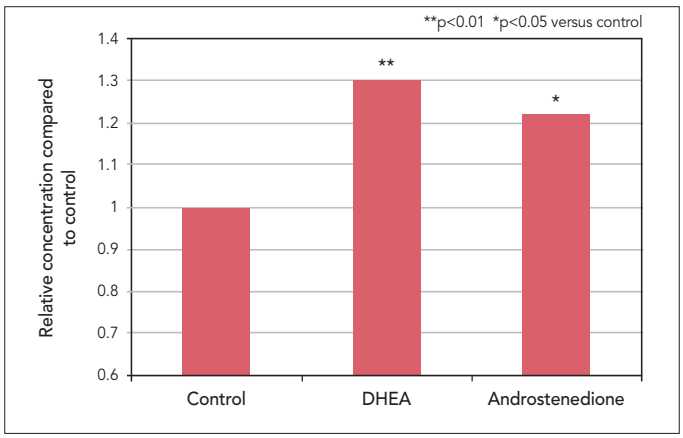

Figure 3: Stimulation of the synthesis of DHEA and androstenedione by the monk's pepper extract at 0.1%.

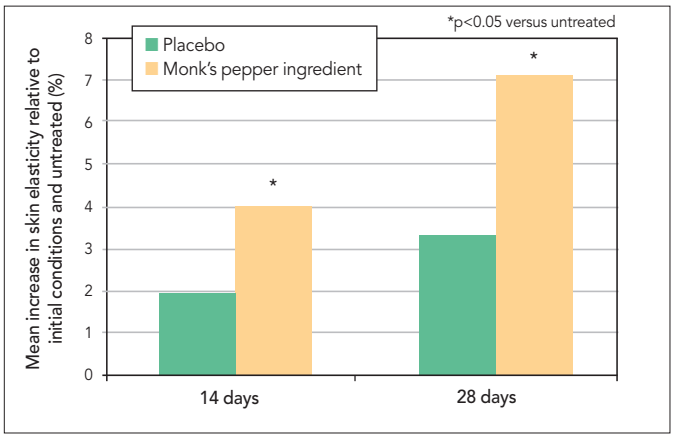

Figure 4: Improvement of skin elasticity after four weeks of treatment with a cream containing 2% monk's pepper ingredient.

female reproductive system. Nowadays it is especially recommended for the treatment of premenstrual syndrome (PMS). The term refers to symptoms such as irritability, tension, anxiety and physical changes that some women experience in the two weeks before the period starts. High levels of the hormone prolactin are thought to play a role in PMS. Chaste tree was found to reduce the release of prolactin, probably through its dopaminergic action. Binding of lipophilic compounds of chaste tree to the dopamine receptor is well documented.[7] Meier *et al.* showed also binding to the opiate receptors, especially to the mu and kappa type (Fig 1).[8] Since anxiety, depression and sleeping problems are important symptoms of PMS and beta-endorphins are known to induce feelings of pleasure and euphoria, the beta-endorphin-like activity might also be involved in the beneficial effect of chaste tree in the treatment of PMS. Binding to the mu and kappa opiate receptor was more pronounced in the lipophilic fraction of an ethanol extract, but the exact molecular nature of the active compounds is not known yet.

The biosynthesis of steroid hormones starts with the conversion of cholesterol to pregnenolone (Fig 2) by a specific cytochrome P450 enzyme (CYP). These enzymes can catalyse monooxygenase reactions leading to the insertion of an oxygen atom into an organic substrate. On the pathway to DHEA another CYP enzyme helps to transform pregnenolone into DHEA by first introducing an hydroxyl group and then deacetylation at the C17. DHEA is then further metabolised to androstenedione with the help of the enzyme 3β-hydroxysteroid dehydrogenase which converts the hydroxyl group at the C3 into a keto configuration. Androstenedione is a direct precursor of the male sex hormone testosterone. However, androstenedione may also be converted directly to oestrogen, the female sex hormone.

In order to assess a potential stimulatory effect on the biosynthesis of sex steroid hormones, the adrenocortical cell line NCI-H295R was used to test a monk's pepper extract. The cell line, derived from an invasive human adrenocortical tumour, was shown to express all the key enzymes of the sex steroid pathway. A defined number of cells was exposed to the monk's pepper extract at different concentrations for 48 hours. The concentration of DHEA and androstenedione in the supernatant was analysed by liquid chromatography and mass spectrometry. Results showed for the monk's pepper extract a significant stimulation of the synthesis of DHEA as well as of androstenedione (Fig 3).

To test in clinical studies, a monk's pepper extract was encapsulated on maltodextrin (Densorphin; INCI: Vitex Agnus Castus Extract, Maltodextrin, Aqua/Water).

A cream, based on an oil in water emulsion, containing 2% monk's pepper ingredient was tested on 15 women and 15 men aged between 52 and 76. The cream with the monk's pepper ingredient was applied twice daily for 28 days on the inner side of the forearm. The other forearm was treated with the placebo cream. Skin elasticity was measured with the Cutometer MPA 580 (Courage & Khazaka GmbH, Cologne). After four weeks of use, and compared to the placebo product, the emulsion with the monk's pepper ingredient clearly improved skin elasticity (Fig 4). In another clinical study, high-frequency ultrasound (Dermascan C Cortex Technology) was used to analyse tissue density in the dermis. The ultrasonographic wave generates an echo when it is reflected at the boundaries between different tissue structures. The intensity of the reflected echoes can be evaluated and visualised in colour images. The collagen and elastin fibre structure of an intact dermis generates many reflections visible as bright colours in the ultrasonographic image. However, disruption of this regular architecture leads to weaker reflections and dark patches (Fig 5). This so-called subepidermal low-echogenic band (SLEB) is characteristic for aged and photo-damaged skin. Changes in SLEB of 15 women and 15 men aged between 50 and 65 were monitored. The volunteers applied a cream with 2% monk's pepper ingredient over 4

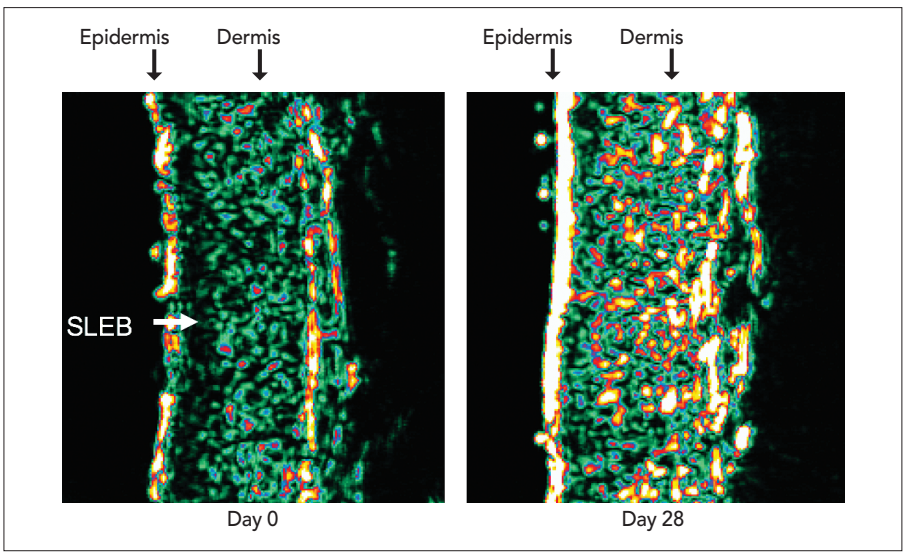

Figure 5: Ultrasonographic images of the forearm of a volunteer before and after treatment. Colour scale: white – yellow – red – green – blue – black; light colours indicating high echogenicity and thus high skin density.

weeks twice daily on the forearm and the placebo cream on the other forearm. Already after four weeks' treatment, the SLEB was much less pronounced indicating a redensification of the dermis (Fig 5). Quantification of the skin density, and compared to the placebo-treated area, showed a statistically significant increase of 9.2%.

Conclusion

The level of circulating DHEA decreases in men between the ages of 40 to 80 years and in women between the age of 30 and the menopause by about 60%.[9, 10] Women at menopause are lacking oestrogens not only because of a missing ovarian activity but also because of reduced adrenal function. Also in men, ageing is accompanied by a decline of testicular as well as adrenal steroid hormones. In the skin, oestrogen stimulates the synthesis of hyaluronic acid and collagen I and III. Declining oestrogen levels are a main cause for skin ageing. Based on the capacity of the skin to synthesise sex steroid hormones from cholesterol, topical application of a monk's pepper extract makes it possible to increase DHEA and androstenedione levels in the skin. Furthermore, the monk's pepper extract was found to contain mu opioid receptor agonist compounds. Plasma levels of beta-endorphin were found to be significantly decreased in postmenopausal women compared to fertile women.[11] Because of the importance of the beta-endorphin / mu opioid receptor system in skin regeneration, the agonist compounds represent the other part of the overall anti-ageing effect of topically applied monk's pepper. PC

References

1 Slominski A, Zbytek B, Nikolakis G, Manna PR, Skobowiat C, Zmijewski M, Li W, Janjetovic Z, Postlethwaite A, Zouboulis CC, Tuckey RC. Steroidogenesis in the skin: implications for local immune functions. J *Steroid Biochem Mol Biol*. 2013; 137:107-23.

2 Pomari E, Dalla Valle L, Pertile P, Colombo L, Thornton MJ. Intracrine sex steroid synthesis and signaling in human epidermal keratinocytes and dermal fibroblasts. *The FASEB Journal*. 2015; 29:508-24.

3 Bigliardi PL, Sumanovski LT, Büchner S, Rufli T, Bigliardi Q. Different Expression of μ-Opiate Receptor in Chronic and Acute Wounds and the Effect of β-Endorphin on Transforming Growth Factor β Type II Receptor and Cytokeratin 16 Expression. *J Invest Dermatol*. 2003; 120:145-52.

4 Schiller M, Raghunath M, Kubitscheck U, Scholzen TE, Fisbeck T, Metze D, Luger TA, Böhm M. Human dermal fibroblasts express prohormone convertases 1 and 2 and produce proopiomelanocortin-derived peptides. *J Invest Dermatol*. 2001; 117:227-35.

5 Kauser S, Schallreuter KU, Thody AJ, Gummer C, Tobin DJ. Regulation of human epidermal melanocyte biology by beta-endorphin. *J Invest Dermatol*. 2003; 120:1073-80.

6 Bigliardi PL, Dancik Y, Neumann C, Bigliardi Q. Opioids and skin homeostasis, regeneration and ageing – What's the evidence? *Exp Dermatol*. 2016; **25** (8):586-91.

7 Wuttke W, Jarry H, Christoffel V, Spengler B, Seidlovai-Wuttke D. Chaste tree (Vitex agnus-castus) – pharmacology and clinical indications. *Phytomedicine* 2003; 10:348-57.

8 Meier B, Berger D, Hoberg E, Sticher O, Schaffner W. Pharmacological activities of Vitex agnus-castus extract in vitro. *Phytomedicine* 2000; 7:373-81.

9 Bélanger A, Candas B, Dupont A, Cusan L, Diamond P, Gomez JL, Labrie F. Changes in serum concentrations of conjugated and unconjugated steroids in 40- to 80-year-old men. J Clin Endocrinol Metab. 1994; **79** (4):1086-90.

10 Labrie F, Luu-The V, Labrie C, Simard J. DHEA and its Transformation into Androgens and Estrogens in Peripheral Target Tissues: Intracrinology. *Frontiers in Neuroendocrinology* 2001; 22:185-212.

11 Genazzani AR, Facchinetti F, Ricci-Danero MG, Parrini D, Petraglia F, La Rosa R, D'Antona N. Beta-lipotropin and beta-endorphin in physiological and surgical menopause. *J. Endocrinol. Invest.* 1981; **375** (4):375-378.