THE INFLUENCE OF COLLAGEN FROM VARIOUS SOURCES ON SKIN PARAMETERS

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Abstract

Collagen is the main component of connective tissue – it represents 30% of total proteins in the animal body. This protein occurs in a wide range of tissues, e.g. in bone, skin, tendon, ligaments and cornea. It provides structural integrity, strength, resistance to tensile stress and elasticity. Due to its excellent biocompatibility and controlled biodegradability, collagen has found diverse application in the biomedical field such as wound dressing, drug carrier and tissue engineering. However, concerns about contamination of mammalian collagen have stimulated the search of another source of this biopolymer. Fish wastes are thought to be an attractive and safe new source of collagen. Fish and mammalian collagen differ in physical and chemical properties.

The aim of this work was to examine the influence of collagen extracted from different sources (rat tail tendons, fish scales of northern pike (Esox lucius) and fish skin of Brama australis) on skin parameters such as hydration, colour, pH and skin's barrier quality. The measurements had been taken on the skin surface before as well as after application of the collagen solutions.

The most harmful effect on skin parameters was observed after application of rat tail collagen solution. Collagen extracted from scales of Esox lucius showed the most favourable effect on the skin parameters.

The source of collagen has a significant influence on its effectiveness. The greatest virtues for human body were observed in the case of fish collagen extracted from Esox lucius scales.

***Keywords:** fish collagen, Brama australis, Esox lucius, rat tail tendons, skin parameters*

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Introduction

Collagen, as the main component in the extracellular matrix and connective tissue, it is the most abundant protein in mammals – it constitutes nearly 30% of total proteins in the animal body [1,2]. During the last years, several different collagenous proteins have been isolated and characterized, which lead to expanding the collagen family to at least 29 types of collagen. They are marked as types I - XXIX [3,4]. The various types of collagen differ in amino acid sequence, structure and function.

Collagenous proteins are characterized by a triple-helix structure in which three left-handed polypeptide chains are supercoiled into a right-handed triple helix [5,6]. Each polypeptide chain consists of about 1000 amino acids and they are formed of a repeating triplet: Gly-X-Y, where Gly is glycine, X is generally proline and Y is hydroxyproline [4]. This sequence stabilizes the triple helix and is responsible for its specific shape. Proline and hydroxyproline are heterocyclic amino acids containing a nitrogen atom, which causes a significant effect on the structure of the triple helix. Collagen helix also contains polar amino acids, such as lysine, arginine, glutamic acid and aspartic acid [7]. The collagen comprises non-helical fragments, i.e. telopeptides, which are located at the ends of the collagen molecule or are embedded in the superhelix [8].

This biopolymer is the main structural protein responsible for the structural integrity of the connective tissue of many multicellular organisms. A wide range of tissues from tendons and ligaments to skin, cornea, bone and dentin have divergent mechanical requirements. Some of them need to be elastic and other require to be stiff and tough. Therefore, collagen as a building material shows a broad versatility - it provides mechanical stability, toughness, tensile strength and elasticity [9,10]. The skin contains mainly type I, III and V collagen, which determine the tension, elasticity, durability and hydration of skin [11].

Type I collagen is distributed in bone, skin, tendon, ligaments, cornea and other organs [1]. For that reason, the most common raw materials for collagen extraction are skin, bones, tendons and cartilage [12]. Due to the potential risk for viral and prion contamination of collagen derived from animal tissues, non-mammalian sources of this protein are sought. Collagen extracted from fish wastes presents an attractive new source of collagen as it is a by-product of food production. Mammalian and fish collagens differ in thermal stability and the amino acid composition – fish collagen is characterised by a lower proline and hydroxyproline content and a lower denaturation temperature [13-15].

The paper focuses on comparing the influence of collagen extracted from different species (rat tail tendons, fish scales of northern pike (*Esox lucius)* and skin of *Brama australis*) on skin parameters such as hydration, colour, pH and skin's barrier quality.

Materials and Methods

Collagen from rat tail tendon [16] as well as collagens from fish tissues – scales of *Esox lucius* [15] and skin of *Brama australis* [17] were prepared in our laboratory.

Afterwards, 0.1% solutions of each collagen were prepared and applied on the forearm skin of voluntarily probants. Then, the evaluation of skin condition after the application of collagen solutions was made, including hydration, pH, colour and skin's barrier quality. The measurements were conducted with the participation of five probants (women, aged 22 years).

The hydration level of the skin surface (*stratum corneum*) was determined using Corneometer CM 825 (Courage+Khazaka, Germany). Skin's pH was tested using pH-meter (Elmetron, Poland), skin's barrier quality (TEWL - Transepidermal Water Loss) was examined using Tewameter TM 300 (Courage+Khazaka, Germany) and skin colour was measured by Skin-Colorimeter CL 400 (Courage+Khazaka, Germany).

The measurements had been taken on the skin surface in three places before application and after 10, 20, 30, 60, 120 and 180 min from application of the collagens solutions. The results of these measurements were averaged and the standard deviation was calculated. All measurements were performed in the laboratory in controlled temperature and humidity conditions (20-22°C, relative humidity 40-60%).

Results and Discussion

The results of evaluation of skin condition after the application of collagen solutions are shown in FIGs. 1-4.

The application of obtained collagen from rat tail tendons and fish tissues (marine *Brama australis* and fresh water *Esox lucius*) had initially deteriorated the skin's barrier quality manifesting itself as the increase in TEWL (FIG. 1). The highest TEWL value was observed after application of collagen from rat tail tendons. The level of TEWL had returned to the initial level 120 min after application of collagen solutions from rat tail tendons and *Esox lucius* scales and after 180 min in the case of collagen from the skin of *Brama australis*. The solution of collagen extracted from the scales of *Esox lucius* improved the skin's barrier quality – 180 min after application of this solution the level of TEWL decreased below the preliminary level.

Slight redness of skin appeared after the application of collagen solutions (FIG. 2). Collagen from rat tail tendons made the skin the more red and irritated. After 120 min from the application of collagen solution from *Esox lucius* scales the skin colour solely had returned to the initial level.

The application of collagens solution had improved the hydration of the outer skin layers (FIG. 3). After 30 min the level of hydration of the skin surface decreased, however, within three hours of the study it remained at a higher level than the initial one regardless of the source of collagen. The solution of collagen from *Esox lucius* scales have the best long-term moisturizing properties. The increase in TEWL could have an indirect impact on corneometric measurements.

Application of rat collagen solution had increased the skin pH, while a solution of fish scale collagen had slightly decreased the pH of skin (FIG. 4).

The most harmful effect on skin parameters was observed after application of rat tail collagen solution. Application of rat collagen irritated the skin and deteriorated the skin's barrier quality the most. While collagen extracted from scales of *Esox lucius* showed the most favourable effect on the skin parameters. Collagen from *Esox lucius* improved the skin's barrier quality and have the best long-term moisturizing properties.

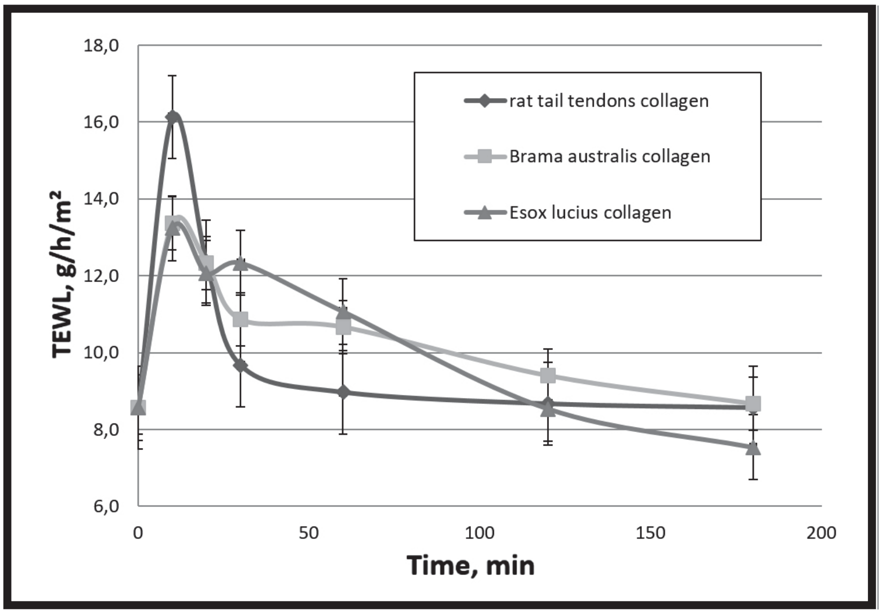

FIG. 1. The graph of dependence of skin's barrier quality (manifesting as Transepidermal water loss – TEWL) from time after application of collagen from different sources – rat tail tendons, scales of *Esox lucius* and skin of *Brama australis.*

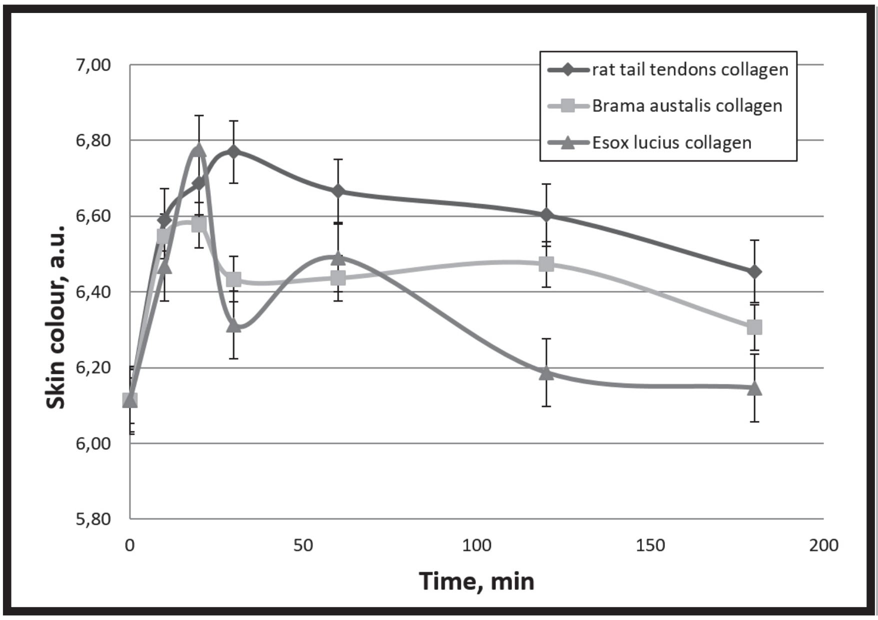

FIG. 2. The graph of dependence of skin colour from time after application of collagen from different sources – rat tail tendons, scales of *Esox lucius* and skin of *Brama australis.*

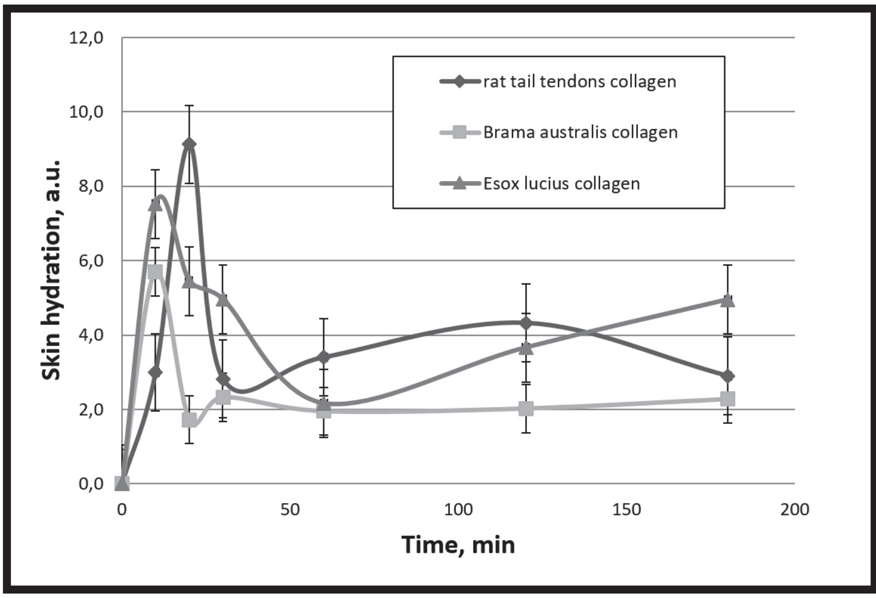

FIG. 3. The graph of dependence of skin hydration from time after application of collagen from different sources – rat tail tendons, scales of *Esox lucius* and skin of *Brama australis.*

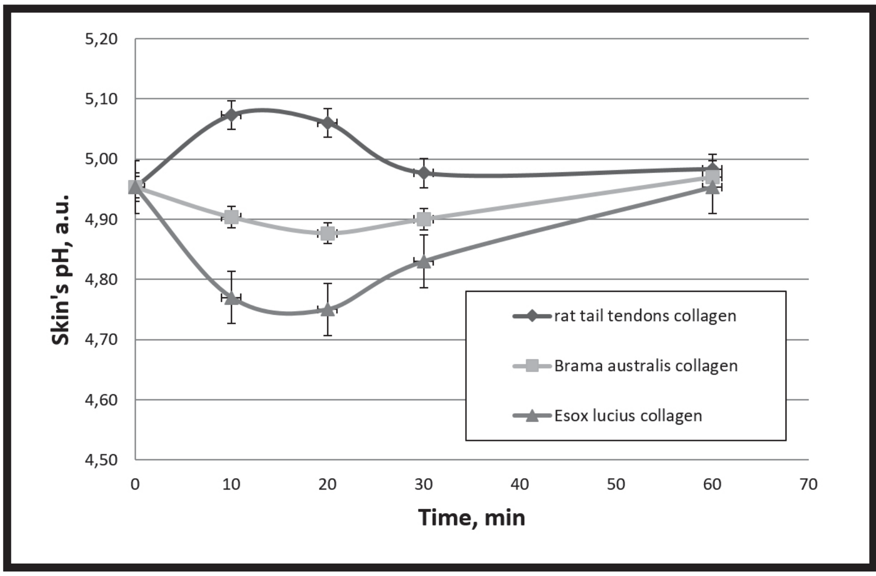

FIG. 4. The graph of dependence of skin's pH from time after application of collagen from different sources – rat tail tendons, scales of *Esox lucius* and skin of *Brama australis.*

Conclusions

The source of collagen has a significant influence on its effectiveness. The greatest virtues for human body were observed in the case of fish collagen extracted from *Esox lucius* scales.

Collagen, due to its biocompatibility, biodegradability and non-toxicity, is widely used for cosmetic, pharmaceutical and biomedical applications. Fish collagen may be a good base for the production of collagen matrices for the skin applications (e.g. for wound dressings) because it exhibits a positive active effect on the skin. Moreover, other active substances can be incorporated into these matrices. For that reasons, fish collagen may be an attractive alternative to mammalian collagen for biomaterials production.

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