

POSTER PRESENTATIONS

GREEN EXTRACTION AND COMPARATIVE CHARACTERIZATION OF COLLAGEN FROM DIFFERENT TISSUES OF *Esox lucius*

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Fish processing by-products represent an important renewable source of collagen for biomedical, pharmaceutical, cosmetic, and tissue engineering applications. Compared with mammalian collagen, fish collagen offers excellent biocompatibility, low immunogenicity, and no risk of prion disease transmission. The present study aimed to isolate collagen from different tissues of freshwater pike (*Esox lucius*) and to perform a comprehensive physicochemical, structural, and biological characterization using an environmentally friendly extraction approach.

Collagen was isolated from skin with scales, swim bladder, and bones by cold extraction with 3% acetic acid, followed by beeswax-assisted defatting, dialysis, and freeze-drying; protein content was determined by the Kjeldahl method. Structural characterization was performed using Fourier-transform infrared spectroscopy (FTIR) and scanning electron microscopy (SEM). Amino acid composition was analyzed by HPLC after acid hydrolysis, whereas elemental composition was determined by inductively coupled plasma mass spectrometry (ICP-MS). The biological safety of collagen preparations was evaluated in acute and subchronic toxicity studies in laboratory rats.

The developed extraction procedure effectively preserved the native collagen structure while reducing residual lipid content from $4.6 \pm 0.3\%$ to $0.3 \pm 0.1\%$ without the use of hazardous organic solvents. Collagen yields reached 10.2% from the skin with scales, 6.5% from the swim bladder, and 2.2% from the bones, with protein content ranging from 88.79% to 98.5%. FTIR spectra exhibited characteristic amide I, II, and III absorption bands, confirming the collagen structure. SEM analysis demonstrated well-preserved fibrillar-lamellar morphology with distinct tissue-specific microarchitectural features. HPLC analysis identified twelve amino acids, predominantly glycine and proline, characteristic of fibrillar collagen. ICP-MS revealed significant differences in mineral composition among tissues, with skin-derived collagen containing the highest calcium and phosphorus levels, whereas bone-derived collagen was enriched with magnesium, silicon, and several trace elements. Toxic elements were detected only at trace levels, and acute and repeated-dose toxicity studies confirmed the biological safety of the isolated collagen preparations.

The proposed extraction approach combines high collagen quality with green chemistry principles and sustainable utilization of fish-processing by-products, while beeswax-assisted purification provides an environmentally friendly alternative to conventional solvent defatting. The collagen preparations obtained from different tissues of *Esox lucius* demonstrated favorable physicochemical characteristics, a characteristic amino acid profile, and a high safety profile, indicating their potential for applications in regenerative medicine, pharmaceutical formulations, tissue engineering, cosmetics, and the development of advanced biomaterials.