

Physicochemical properties of acid-soluble collagen extracted from silver carp skin

Bartłomiej Milanowski, Hanna Wosicka-Fraćkowiak, Stanisław Woźny,
Kornelia Poniedziałek, Martyna Nyga

GENERICA R&D Lab, Regionalne Centrum Zdrowia Sp. z o.o., 3 Na Kępie St., 64-360 Zbąszyn, Poland

Introduction

Collagen is the most abundant protein in our body, comprising approximately 30% of the total protein content. Collagen and its hydrolysates have broad applications in many fields, such as food, pharmaceutical, cosmetic, biomedical materials, photographic and leather industries.

Twenty-eight types of collagen have been reported so far, but about 85% of the dermal collagen is type I, which represents the fibrillar collagens. The molecular structure of collagen is defined by three polypeptide α -chains forming a right-handed triple helix.

Porcine and bovine collagen sources have been the most popular so far. However, bovine spongiform encephalopathy (BSE) and transmissible spongiform encephalopathy (TSE) still create anxiety, which may clear the way for fish collagen. Although marine collagen's physical and chemical properties may differ from mammalian ones, fish collagen is unlikely to cause BSE or TSE and is not forbidden for religious reasons. Moreover, fish skin is a huge byproduct of fishery processing worldwide and its utilization as a source of collagen seems a decent choice.

Materials and methods

All reagents were of analytical grade. Purified Type I Bovine Collagen Solution 3 mg/mL (TeloCol®-3, Advanced BioMatrix) was purchased from CellSystems GmbH, Germany.

Collagen gel was obtained from the defrosted and prepared skin of silver carp (*Hypophthalmichthys molitrix*) by the in-house acid extraction process yielding a

telopeptide-intact collagen. After that, the telocollagen gel was purified by multistage filtration.

The final product (~ 100 L) was characterized with different techniques, including organoleptic, pH, conductivity, viscosity, weight loss on drying, identity by SDS-polyacrylamide gel electrophoresis (SDS-PAGE), protein content by biuret assay, nativity and agglomerate content using size exclusion chromatography with UV detection (SEC-UV), lactic acid content using HPLC-UV method, microbiological purity and long-term stability.

Results and discussion

Characteristics of the telocollagen hydrogel obtained from silver carp skin are given in Fig. 1 and Fig. 2 and Table 1.

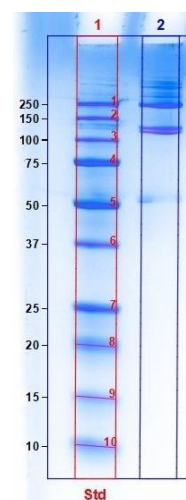


Fig. 1. SDS-PAGE pattern of Protein Standard (lane 1) and telocollagen gel from silver carp skin (lane 2).

Table 1. Physicochemical properties of acid-soluble collagen extracted from silver carp skin.

Parameter, Testing, and Method	Result
Physical form (organoleptic)	Gel
Structure (organoleptic)	Smooth, homogeneous without lumps
Mechanical impurities (organoleptic)	Lack
Color (organoleptic)	Light grey
Smell (organoleptic)	Characteristic fishy
pH (Ph. Eur., 2.2.3)	3.0
Conductivity (Ph. Eur., 2.2.38)	103 mS/m
Viscosity	25760 mPas
Weight loss on drying (gravimetrically)	97.2 %
Identity (SDS-PAGE)	Bands around 250, 125 and 118 kDa are found (Fig. 1)
Protein content (Biuret assay)	3.0 %
Nativity (SEC-UV)	Compliance of the retention times of the tested sample with the retention times of the collagen type I standard (Fig. 2)
Lactic acid content (HPLC-UV)	0.9% w/w
Microbiological purity (Ph. Eur., 5.1.4)	TAMC <10 CFU/g TYMC <10 CFU/g Lack of <i>P. aeruginosa</i> and <i>S. aureus</i>
Shelf life	Minimum of 36 months at 5±3 °C

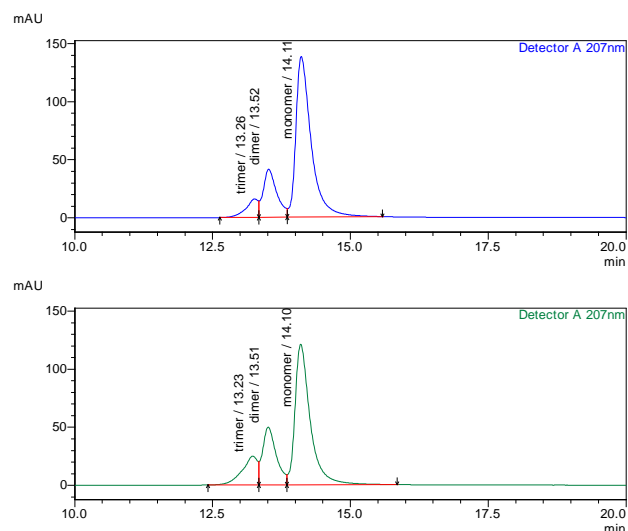


Fig. 2. SEC-UV chromatograms of purified type I bovine collagen standard solution (at the top) and telocollagen gel from silver carp skin (at the bottom).

Conclusion

Acid-soluble collagen from the skin of silver carp was isolated and purified and some properties of this product were investigated. SDS-PAGE patterns showed it was type I collagen. SEC-UV indicated that the collagen was in its native form (triple helix). The protein content in the gel was around 3% and the product met the criteria of microbiological purity. In addition, it was stable for 36 months when stored at a temperature 5±3 °C.

Acknowledgements: The authors are grateful for the financial support received within the Polish National Centre for Research and Development grant number POIR.01.01.01-00-1004/18.

References

- Zhang, J., Duan, R., Tian, Y., Konno, K., 2009. Characterisation of acid-soluble collagen from skin of silver carp (*Hypophthalmichthys molitrix*). *Food Chemistry*, 116(1), 318-322. <https://doi.org/10.1016/j.foodchem.2009.02.053>
- Amirrah, I.N., Lokanathan, Y., Zulkiflee, I., Wee, M.M.R., Motta, A., Fauzi, M.B., 2022. A comprehensive review on collagen type I development of biomaterials for tissue engineering: From biosynthesis to bioscaffold. *Biomedicine*, 10(9), 2307. <https://doi.org/10.3390/biomedicine10092307>