

EFFECT OF PARTICLE SIZE AND PROCESSING CONDITIONS ON THE PRODUCTION OF POWDERED COLLAGEN FROM ARAPAIMA GIGAS SCALES AND ITS APPLICATION IN FUNCTIONAL PRODUCTS

Berita Dávila Ruiz¹, Christiaan E. Moreno¹, Fiorella V. Güere Salazar², William C. Santos Tello^{3*}, Jhonn R. Ruiz De la Cruz⁴, Kathy Flores Cabrera De Ruiz⁴, Luis Angel Oñate Trigozo⁴, Vanessa J. Caldas Garnique⁵

¹ Universidad Tecnológica del Perú, Calle. Natalio Sánchez N° 125, Lima, Perú

² Universidad Nacional Mayor de San Marcos, Ciudad Universitaria, Lima 15081, Perú

³ Universidad Continental, Av. Alfredo Mendiola 5210, Los Olivos 15306, Perú

⁴ Universidad Nacional de Ucayali, Carretera Federico Basadre Km 6.2, Pucallpa, Perú

⁵ Asesoría y Consultoría de Proyectos Ambientales S.A.C., Martinete Mz K Lote 8, Lima, Perú

*Corresponding: wsantos@continental.edu.pe

Abstract. This research aimed to determine the effect of particle size and processing conditions on the yield and physicochemical characteristics of powdered collagen obtained from paiche scales (Arapaima gigas). For this purpose, hydrobiological residues from the Peruvian Amazon were used, considering their valorization as a sustainable alternative for obtaining value-added functional ingredients.

The experimental procedure included washing, removal of non-collagenous proteins, demineralization, thermal hydrolysis, and drying under controlled conditions. Different particle sizes and processing temperatures were evaluated during the process. The variables analyzed included extraction yield, protein content, moisture percentage, solubility, and antioxidant activity of the powdered collagen. The results showed statistically significant differences among the treatments applied ($p < 0.05$), demonstrating that particle size reduction directly influences recovery efficiency and the functional properties of the final product.

The most favorable experimental condition achieved an extraction yield of 23.8%, protein content of 89.6%, moisture content of 5.4%, and solubility of 82.1%, while antioxidant activity reached approximately 68%. Likewise, the statistical analysis presented a coefficient of determination of $R^2 = 0.80$, evidencing a significant relationship between processing conditions and the functional quality of the obtained collagen. Overall, the results confirm the potential of Arapaima gigas scales as a sustainable raw material for the production of powdered collagen intended for nutraceutical and functional food applications.

1. Introduction

The fishing industry generates large volumes of waste during the processing of aquatic resources, including scales, skins, bones, and viscera, which are frequently discarded or used in low-value applications. However, these by-products contain bioactive compounds of industrial interest, particularly structural proteins such as collagen, whose valorization contributes to the integral utilization of fishery resources and the reduction of environmental impacts associated with inadequate waste disposal [1–3].

Arapaima gigas scales constitute an important source of collagen due to their high proportion of fibrillar proteins and minerals. In recent years, collagen of aquatic origin has attracted increasing interest in the food, pharmaceutical, and nutraceutical sectors because of its biocompatibility, bioavailability, and functional potential as an ingredient for the development of health-promoting products [2,4].

Furthermore, the utilization of residues from Amazonian species represents a sustainable alternative for generating added value within regional production chains.

Several studies have demonstrated that processing conditions significantly influence extraction efficiency and the physicochemical characteristics of the collagen obtained. Among the most relevant factors are temperature, processing time, and raw material particle size, since these parameters affect mass transfer, protein release, and collagen structural stability during extraction and drying stages [3,5]. Adequate particle size reduction increases the surface area available for chemical and thermal treatments, promoting protein recovery and improving the functional properties of the final product.

* Corresponding author: wsantos@continental.edu.pe

In this context, the production of powdered collagen from *Arapaima gigas* scales represents a technological alternative with potential for the development of functional ingredients intended for nutraceutical and food applications. Therefore, the present study evaluated the effect of particle size and processing conditions on extraction yield and the physicochemical properties of powdered collagen obtained from *Arapaima gigas* scales, aiming to identify the most favorable conditions for its utilization as a high-value functional ingredient.

2. Materials and Methods

2.1. Raw Material Preparation and Collagen Extraction

The *Arapaima gigas* scales used in this study were obtained from facilities dedicated to the processing of aquatic resources in the Peruvian Amazon. Initially, the raw material underwent a manual selection process to remove impurities, muscle tissue residues, and foreign materials that could affect extraction quality.

Subsequently, the scales were washed with running water and conditioned through controlled drying until suitable conditions for mechanical processing were achieved. Once dried, they were ground and classified according to particle size to obtain different granulometric fractions, which were used to evaluate their influence on extraction yield and the physicochemical characteristics of the collagen obtained.

The collagen extraction process included the removal of non-collagenous proteins, demineralization, thermal hydrolysis, and drying. The elimination of non-collagenous proteins reduced interfering compounds present in the raw material, while demineralization facilitated the release of the protein matrix contained in the scales.

Subsequently, the material was subjected to thermal hydrolysis under different processing conditions to promote collagen extraction. Finally, the extract obtained was dried and conditioned to produce powdered collagen for subsequent analyses.

2.2. Evaluation of Physicochemical Properties of Collagen

The powdered collagen obtained was characterized through the determination of physicochemical and functional variables considered relevant for evaluating its potential application in nutraceutical products and functional foods. The variables analyzed were extraction yield (%), protein content (%), moisture (%), solubility (%), and antioxidant activity (%).

Extraction yield was calculated as the percentage ratio between the mass of recovered collagen and the initial mass of processed scales. Protein content was used as an indicator of the purity and quality of the product obtained, while moisture content was used to evaluate the stability and preservation conditions of the powdered collagen. Solubility was employed to determine the functional behavior of the product in aqueous systems, and antioxidant activity was used to estimate its potential capacity to neutralize free radicals.

The experimental data obtained were subjected to statistical analysis to determine the effect of particle size and processing conditions on the evaluated variables. Analysis of variance (ANOVA) was applied to identify significant differences among treatments, considering a significance level of 5% ($p < 0.05$). Additionally, regression analysis was performed to establish the relationship between processing conditions and the functional quality of powdered collagen through the estimation of the coefficient of determination (R^2).

3. Results and Discussion

The results obtained demonstrated that particle size and processing conditions significantly influenced extraction yield and the physicochemical properties of powdered collagen obtained from *Arapaima gigas* scales. Analysis of variance revealed statistically significant differences among the evaluated treatments ($p < 0.05$), indicating that the modifications applied during raw material conditioning and processing had a direct effect on the quality of the final product.

Particle size reduction promoted greater exposure of the protein matrix within the scales, facilitating mass transfer during demineralization and thermal hydrolysis stages. Consequently, collagen recovery efficiency increased and some of its functional properties, particularly solubility and antioxidant activity, improved. Likewise, processing conditions exerted a significant influence on the stability of the extracted proteins and on the final characteristics of the product obtained. These findings are consistent with previous studies reporting improvements in collagen extraction yield and functional properties when the available surface area is increased through particle size reduction [6,8].

The most favorable experimental condition presented an extraction yield of 23.8%, protein content of 89.6%, moisture content of 5.4%, solubility of 82.1%, and antioxidant activity of approximately 68%. These results demonstrate efficient recovery of protein compounds and confirm the potential of *Arapaima gigas* scales as a raw material for the production of high-value functional ingredients. The values obtained are consistent with studies conducted on collagen from aquatic sources, which report high protein contents and bioactive properties associated with its potential application in nutraceutical products and functional foods [5,8].

Furthermore, regression analysis showed a positive relationship between processing conditions and the functional quality of powdered collagen, yielding a coefficient of determination of $R^2 = 0.80$, which evidences a strong association between processing variables and the functional quality of the collagen obtained.

Table 1. Physicochemical Properties of Powdered Collagen Obtained from Arapaima gigas Scales Under Optimal Experimental Conditions.

Parameter	Unit	Value
Extraction yield	%	23.8
Protein content	%	89.6
Moisture	%	5.4
Solubility	%	82.1
Antioxidant activity	%	68

Table 2 presents the behavior of the physicochemical properties of powdered collagen obtained from Arapaima gigas scales under different particle sizes and processing conditions. An increasing trend was observed in extraction yield, protein content, solubility, and antioxidant activity as particle size decreased, with the best results achieved in treatment T4. In contrast, moisture content showed a progressive decrease, indicating greater drying efficiency and improved potential product stability.

These results demonstrate that an adequate reduction in particle size promotes the release and recovery of protein compounds while simultaneously improving the functional properties of powdered collagen obtained from Amazonian aquatic residues.

Table 2. Physicochemical Properties of Powdered Collagen Obtained from Arapaima gigas Scales under Different Processing Conditions

Treat ment	Part icle Size	Extra ction Yield (%) (%)	Prot ein Cont ent (%)	Mois ture (%)	Solub ility (%)	Antioxi dant Activit y (%)
T1	Coar se	16.8 ± 1.2	80.4 ± 1.6	7.8 ± 0.4	68.5 ± 1.8	54.2 ± 1.5
T2	Med ium	19.7 ± 1.1	84.3 ± 1.4	6.7 ± 0.3	74.8 ± 1.5	59.6 ± 1.3
T3	Fine	21.9 ± 0.9	87.1 ± 1.2	5.9 ± 0.2	79.3 ± 1.2	64.1 ± 1.1
T4	Very fine	23.8 ± 0.8	89.6 ± 1.1	5.4 ± 0.2	82.1 ± 1.0	68.0 ± 1.0

Values correspond to mean ± standard deviation of the evaluated variables.

4. Mathematical Relationships and Process Evaluation

The properties evaluated during powdered collagen production were determined through mathematical relationships used to quantify extraction efficiency and the functional characteristics of the product obtained.

$$RE = \frac{M_c}{M_i} \times 100$$

Where:

RE = extraction yield (%)

Mc = mass of collagen obtained (g)

Mi = initial mass of processed scales (g)

$$H = \frac{M_h - M_s}{M_h} \times 100$$

Where:

H = moisture (%)

Mh = wet mass of the sample (g)

Ms = dry mass of the sample (g)

$$R^2 = 1 - \frac{\sum(y_i - \hat{y}_i)^2}{\sum(y_i - \bar{y})^2}$$

Where:

R^2 = coefficient of determination

y_i = observed values

$\hat{y}_i$ = values estimated by the regression model

$\bar{y}$ = mean of observed values

Equation (1) was used to determine the extraction yield of collagen obtained from Arapaima gigas scales. Equation (2) was used to evaluate the moisture content of the final product, while Equation (3) was employed to analyze the relationship between particle size, processing conditions, and the functional properties of powdered collagen. Regression analysis yielded a coefficient of determination of $R^2 = 0.80$, demonstrating a significant relationship between processing variables and the functional quality of the product obtained.

Conclusions

Particle size and processing conditions significantly influenced extraction yield and the physicochemical properties of powdered collagen obtained from Arapaima gigas scales, with statistically significant differences observed among the evaluated treatments ($p < 0.05$).

The most favorable experimental condition achieved an extraction yield of 23.8%, protein content of 89.6%, moisture content of 5.4%, solubility of 82.1%, and antioxidant activity of approximately 68%, demonstrating that appropriate particle size reduction promotes the recovery of protein compounds and improves the functional properties of the final product.

Likewise, regression analysis showed a coefficient of determination of $R^2 = 0.80$, indicating that the variations observed in the functional quality of powdered collagen are closely related to the processing conditions applied during extraction and raw material conditioning.

The results obtained confirm that *Arapaima gigas* scales constitute a sustainable and technically viable source for the production of powdered collagen with potential applications in nutraceutical products and functional foods, contributing to the integral utilization of Amazonian aquatic residues and the generation of high-value ingredients.

Acknowledgements

The authors express their gratitude to the Ucayali Aquaculture Technical Board for the academic support and facilities provided for the execution of this research.

References

- [1] Jafari, H., Lista, A., Siekapen, M.M., Ghaffari-Bohlouli, P., Nie, L., Alimoradi, H., Shavandi, A. (2020) Fish Collagen: Extraction, Characterization, and Applications for Biomaterials Engineering. *Polymers*, 12(10), 2230.
- [2] Coppola, D., Oliviero, M., Vitale, G.A., Lauritano, C., D'Ambra, I., Iannace, S., de Pascale, D. (2020) Marine Collagen from Alternative and Sustainable Sources: Extraction, Processing and Applications. *Marine Drugs*, 18(4), 214.
- [3] Karim, A.A., Bhat, R. (2009) Fish Gelatin: Properties, Challenges, and Prospects as an Alternative to Mammalian Gelatins. *Food Hydrocolloids*, 23(3), 563–576.
- [4] Liu, D., Nikoo, M., Boran, G., Zhou, P., Regenstein, J.M. (2015) Collagen and Gelatin. *Annual Review of Food Science and Technology*, 6, 527–557.
- [5] Gómez-Guillén, M.C., Giménez, B., López-Caballero, M.E., Montero, M.P. (2011) Functional and Bioactive Properties of Collagen and Gelatin from Alternative Sources: A Review. *Food Hydrocolloids*, 25(8), 1813–1827.
- [6] Huang, Y.R., Shiau, C.Y., Chen, H.H., Huang, B.C. (2015) Isolation and Characterization of Fish Scale Collagen from *Tilapia* (*Oreochromis* sp.) by a Novel Extrusion–Hydro-Extraction Process. *Food Chemistry*, 190, 997–1006.
- [7] Wang, L., An, X., Yang, F., Xin, Z., Zhao, L., Hu, Q. (2008) Isolation and Characterization of Collagen from the Skin, Scale and Bone of Deep-Sea Redfish (*Sebastes mentella*). *Food Chemistry*, 108(2), 616–623.
- [8] Wang, B., Wang, Y.M., Chi, C.F., Luo, H.Y., Deng, S.G., Ma, J.Y. (2013) Isolation and Characterization of Collagen and Antioxidant Collagen Peptides from Scales of Croceine Croaker (*Pseudosciaena crocea*). *Marine Drugs*, 11(11), 4641–4661.