International Journal of Agriculture and Food Sciences Research
ISSN: 3006-7685 (Print), ISSN: 3006-3736 (Online) | Volume 1, Number 3, Year 2024
DOI: https://doi.org/10.62051/ijafsr.v1n3.02
Journal homepage: https://wepub.org/index.php/IJAFSR/index

Development and Evaluation of a Novel Fish-Derived Protein Peptide Conditioner Formulated with Pneumatophorus japonicus Heads Peptides

Haijing Zhang [1, 6], Runhui Liu [1, 6], Xichang Wang [1, 2, 3, 4], Miaosi Hong [5],

Changhua Xu [1, 2, 3, 4, *]

[1] College of Food Science & Technology, Shanghai Ocean University, Shanghai 201306, PR China

[2] Shanghai Engineering Research Center of Aquatic-Product Processing & Preservation, Shanghai 201306, China

[3] Laboratory of Quality and Safety Risk Assessment for Aquatic Products on Storage and Preservation (Shanghai), Ministry of Agriculture, Shanghai 201306, China

[4] National R&D Branch Center for Freshwater Aquatic Products Processing Technology (Shanghai), Shanghai 201306, China

[5] Shanghai Jiao Tong University Affiliated Sixth People's Hospital, Shanghai 201306, PR China

[6] Suzhou Qinpu Biotechnology Pte Ltd, Suzhou, Jiangsu Province 215215, China

*Corresponding Author: chxu@shou.edu.cn

ABSTRACT

Given the unique advantage of hair conditioners formulated with marine-derived proteins and their hydrolysates in terms of green, effective, sustainable, and environmentally friendly. This study presents a novel fish-derived protein peptide-based hair conditioner that featuring Pneumatophorus japonicus heads peptides (PHP). Results demonstrate that the PHP conditioner exhibits favorable sensory properties, improves the surface morphology and structure of damaged hair, reduce the degree of fragmentation and roughness, improve the gloss and make hair smoother. PHP penetrates into the hair shaft, interacting with hair fiber keratin to enhance the integrity and stability of the α-conformation of hair keratin. Furthermore, the PHP conditioner treatment increased total amino acid content by 4.98% and hydrophobic amino acid content by 2.67%, approaching the levels observed in virgin hair. Therefore, the PHP conditioner effectively permeates the surface and interior of hair fibers, filling voids and ameliorating changes in both the microstructural and compositional aspects of the hair. These improvements surpass those observed with a commercially available wheat protein conditioner. Overall, the PHP conditioner effectively repairs damaged hair, offering a new strategy for the development of innovative natural hair care products.

KEYWORDS

Fish-Derived protein peptide; Hair conditioner; Damaged hair; Hair care

1. INTRODUCTION

Hair plays a significant role in personal image and self-expression. However, it is frequently exposed to external stressors from various unfavorable chemical or physical styling salon procedures and environmental factors, which can lead to varying degrees of damage, resulting in a dull and rough appearance [1]. While these adverse effects can be reduced by the use of hair conditioners to keep the hair and scalp clean, preserving hair health, repair or at least mitigate external damage, and improve the physical and aesthetic properties of the hair [2]. However, most of the hair conditioners on the market contain limited natural active ingredients, many chemical components, false slippery, superficial smoothness, and poor lasting moisturizing effects. Additionally, the cosmetics industry faces numerous challenges stemming from consumer attitudes, including cultural or religious considerations, health or safety concerns, and environmentally friendly practices [3].

The cosmetics industry is witnessing a clear trend towards incorporating marine-derived ingredients in formulations due to their ability to cater to diverse consumer demands and possess a wide array of bioactivities, including antioxidant, moisturizing, UV-protective, and restorative effects [4]. Given the increasing consumer preference for natural active substances in hair care products, the development of fish-derived protein peptide hair care products holds significant research potential. Protein and peptide-based hair care ingredients repair damaged hair mainly through the two mechanisms of surface film formation and internal penetration. Macromolecular proteins and peptides mainly deposit on the surface of damaged hair through electrostatic interactions or chemical cross-linking, forming a protein film that protects against external stressors [5]. Conversely, small molecular weight peptides and proteins possess better penetration capabilities, enabling them to traverse the lipid layer on the surface of hair fibers and reach the hair shaft, thereby improving hair quality [6]. Research has shown that fish-derived collagen hydrolysates extracted from Mozambique tilapia (*Oreochromis mossambicus*) scales exhibit potential for promoting hair growth and enhancing skin health both in vitro and in vivo [7]. Several cosmetic companies have incorporated marine collagen into hair care products, leveraging the properties and functionalities of protein to improve, restore, and modify damaged hair fibers and strengthen the scalp [8]. Miller et al. have developed a mussel coating protein-derived complex flocculant capable of mitigating frictional surface damage, showcasing significant potential for hair care applications[9]. Therefore, designing and producing novel protein peptide-based hair conditioners holds immense promise for developing cost-effective and sustainable products for the hair care industry.

This study focuses on the development of a fish-derived protein peptide hair conditioner using enzymatically hydrolyzed *Pneumatophorus japonicus* heads peptides (PHP) as the active ingredient, formulated with other hair care product materials. The research investigates the reparative effects of the PHP conditioner on damaged hair and compares its efficacy to a commercially available wheat protein conditioner.

2. MATERIALS AND METHODS

2.1. Materials and Reagents

Fresh fish (*Pneumatophorus japonicus*) heads, were provided by Zhejiang Industrial Group Co., Ltd. (Zhoushan, Zhejiang, China). A commercial wheat protein conditioner was purchased from Shanghai Bee Flower Daily Necessities Co., Ltd. (Shanghai, China). The main hair conditioner raw materials ceteartrimonium chloride, cetearyl alcohol, EDTA-2Na, methylchloroisothiazolinone, methylisothiazolinone, essence, CL12490, CL11680, and citric acid were purchased from local commercial establishments and were analytical grade.

Damaged hair (DH) samples were collected from Shanghai Xinsheng hair transplantation Hospital. Virgin hair (NH) samples from volunteers of Shanghai Ocean university, healthy hair, forehead hairline with no signs of decline, not for bleaching, dyeing, ironing hair processing, such as age is 20-40 years old.

2.2. Preparation of Pneumatophorus japonicus Heads Peptides (PHP)

The *Pneumatophorus japonicus* heads were first defatted and chopped into distilled water (water: bones=4: 1), and then alkaline protease and trypsin (enzyme activity ratio 1: 3) were selected for the hydrolysis process. Enzymatic hydrolysis was carried out at 51°C and pH 7.0 for 4h under water bath oscillation (120 rpm). The supernatant was obtained by enzyme inactivation (90°C, 15 min), centrifugation (10000 rpm, 10 min), 1% activated carbon decolorization (30 min), and filtration.

2.3. Preparation of Fish-derived Protein Peptide Conditioner

According to Table 1, the phase A component mixed water bath is heated to 90℃, and placed at a constant temperature, and removed when used. Components of phase B were mixed and heated in a water bath to melt at 75°C. Phase A was then added to phase B. The mixture was homogenized using a homogenizer at 10000 rpm for 25 min, followed by stirring with a magnetic stirrer at the highest speed for 25 min until a homogeneous mixture was achieved. When the system is cooled to 45℃, add the active ingredient PHP, and finally add each component of the C phase one by one and stir until uniform.

Table 1. Fish-derived protein peptide conditioner formula sheet

Constituent	Raw material	Function	Content (g)
A	Water	Solvent	88.1
B	Ceteartrimonium Chloride	Cationic surfactant; Antistatic; Antibacterial agent	7
	Cetearyl Alcohol	Emulsifier; Softening agent; Thickener	3
	EDTA-2Na	Chelating agent	0.1
C	Methylisothiazolinone	Formaldehyde	0.2
	Methylchloroisothiazolinone	Formaldehyde	0.2
	Pneumatophorus japonicus head peptides	Active ingredient	0.5
	Essence	Essence and fragrance	0.2
	CL12490	Colorant	0.2
	CL11680	Colorant	0.2
	Citric acid	PH regulator (PH < 5)	0.3

2.4. Hair Sample Pretreatment

The VH and DH were rinsed three times by oscillating with ultra-pure water for 1 min each time, and the impurities attached to the hair surface were removed and then air dried naturally. DH samples were divided into 4 groups, 1 group was used as negative control (no treatment), the other 3 groups were applied with conventional conditioner, commercially available wheat protein conditioner, and PHP-containing conditioner for 1 h, and VH was used as positive control group. After treatment with the above different types of conditioners, samples were rinsed three times with ultrapure water, and then dry it naturally. Hair fibers were gently removed with tweezers, washed for 3 times and left for natural air drying at room temperature for 12 h. Samples were then sorted and stored successively according to the numbering order.

2.5. Hair Optical Microscope

The hair samples of each treatment group were mounted on glass slides and observed under an inverted microscope using a 10× eyepiece and a 40× objective lens. The morphology, surface gloss, and appearance of the hair samples and treated with different conditioners were visually assessed and documented through photographic imaging.

2.6. Scanning Electron Microscopy (SEM) of Hair

Hair samples from each treatment group were trimmed to approximately 1 cm in length. Individual hair strands were carefully extracted using tweezers and mounted onto a sample stage covered with conductive adhesive tape. After gold spraying for 50 s, the hair samples were scanned using a scanning electron microscope at an accelerating voltage of 5 kV. Scanning was performed at magnifications of 1000×, 2000×, and 10000× to capture the external morphology of the hair.

2.7. Differential Scanning Calorimetry (DSC) Analysis of Hair

Hair samples from each treatment group were equilibrated at 25°C and 65% relative humidity for 48 hours. Samples were then cut into fragments less than 1 mm in length, and 5-6 mg of each sample was weighed accurately and placed in an aluminum pan. The experiments were conducted under a nitrogen flow rate of 50 mL/min. The temperature program involved initial heating from room temperature to 60°C balanced for 30 min to remove surface moisture. Subsequently, the samples were cooled to 30°C at a rate of 10°C/min and then reheated to 270°C at the same rate. The heat absorption curves were recorded, and each treatment group was subjected to three replicate experiments. The average peak temperature and peak area were calculated. The relative helix content (RHC) of the hair keratin was determined using the following formula:

$$\mathrm{RHC} = \frac{\Delta \mathrm{H_d}}{\Delta \mathrm{H_{d0}}} \times 100\%$$

ΔH_d is the enthalpy of denaturation (J/g). ΔH_{d0} is the enthalpy of denaturation of the untreated unhealthy hair. The peak temperature represents the denaturation temperature of the α-helix peak (Td, ℃).

2.8. Amino Acid Content Determination in Hair Samples

Hair samples from each treatment group were cut into fragments approximately 5 mm in length. Approximately 15-20 mg of each hair sample was accurately weighed and placed in a hydrolysis tube. 10 mL of 6 mol/L hydrochloric acid was added to the tube, which was then cooled in a 4°C refrigerator for 5 min before being sealed under vacuum. Hydrolysis was performed at 110°C for 22 h in an oven. After cooling, the hydrolysate was repeatedly rinsed with distilled water, filtered, and made up to 50 mL in a volumetric flask. 1 mL of the filtered hydrolysate was transferred to a petri dish and dried under vacuum at 50°C. The residue was dissolved in 1 mL of distilled water and dried under vacuum again until completely dry. 2 mL of a pH=2.2 sodium citrate solution was added to the test tube, dissolving the residue completely. The solution was then vortexed to ensure homogeneity and filtered through a 0.22 μm pore-size aqueous filter membrane. The filtrate was collected and transferred to a 2 mL autosampler vial and stored at -20°C until analysis using an amino acid analyzer.

2.9. Statistical Analysis

All experimental studies were expressed as mean ± standard deviation. The data were analyzed by ANOVA test (SPSS 21.0 statistical software). Tukey test was used to analyze the significant difference between the mean values of each parameter. Relevant data charts were generated using Origin 2024 and Microsoft Excel 2019.

3. RESULTS AND DISCUSSION

3.1. Analysis of Fish-Derived Protein Peptide Conditioner Formulation

It is widely acknowledged that conditioners enhance the softness and smoothness of damaged hair by providing moisture [10]. Conditioners function by neutralizing the negative charge of hair fibers and reducing the hydrophilicity of the cuticle layer through the addition of positively charged agents and lubricants. These agents typically include anti-static and lubricating substances such as polymers, oils, waxes, and cationic surfactants [11]. A visual assessment was conducted on the physical properties of three formulations: a conventional conditioner, a commercially available wheat protein conditioner, and a conditioner incorporating PHP. As shown in Figure 1, all formulations exhibited stability, with no observable impurities, layering, solidification, or signs of degradation. The commercially available wheat protein conditioner displayed a yellow hue, while the conditioner without PHP was orange. The PHP conditioner exhibited a slightly deeper color compared to the formulation without PHP, but no unusual coloration or particulate matter was observed. All formulations were also evaluated for odor, with no unusual or unpleasant scents detected. Therefore, the prepared conditioners demonstrated good sensory properties.

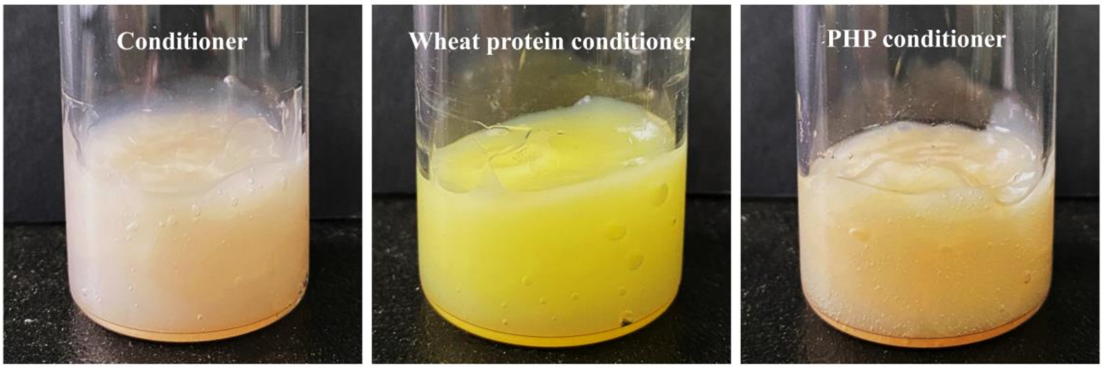

Figure 1. Visualization of the results of different conditioner configurations

3.2. Optical Microscopy Analysis of Hair Surface Morphology

The surface morphology of DH treated with conventional conditioner, wheat protein conditioner, and PHP conditioner was examined using optical microscopy. Images were captured at different locations along the hair shaft to compare the effects of the conditioners (Figure 2). VH exhibited a smoother surface with a glossy appearance and flattened, non-raised edges. In contrast, DH presented a dull surface, uneven edges, and raised cuticular scales. Conditioner treatment generally improved the condition of the hair cuticle to a certain extent. However, conventional conditioners, lacking protein-based active ingredients, primarily addressed hair surface roughness and raised cuticles, showing limited improvement in glossiness. Conditioners incorporating wheat protein and PHP, in addition to smoothing the hair surface, demonstrated a notable enhancement in hair shine. Most conventional hair conditioner formulations consist of supramolecular complexes formed by the interaction of cationic polyelectrolytes and surfactants [12]. However, the cuticle layer of hair fibers hinders the penetration of these large molecule chemicals, resulting in minimal deposition on the hair surface. These residual compounds on the cuticle surface can only temporarily fill and smooth damaged cuticle layers, failing to penetrate the cortex for deep repair [13]. Wheat protein, PHP, and other protein and peptide-based ingredients exhibit high chemical affinity with hair keratin. These large molecules can adhere to the hair surface, while smaller molecules can diffuse into the hair interior, even penetrating the cortex. Consequently, such conditioners demonstrate superior repair efficacy compared to conventional conditioners lacking protein-based active ingredients.

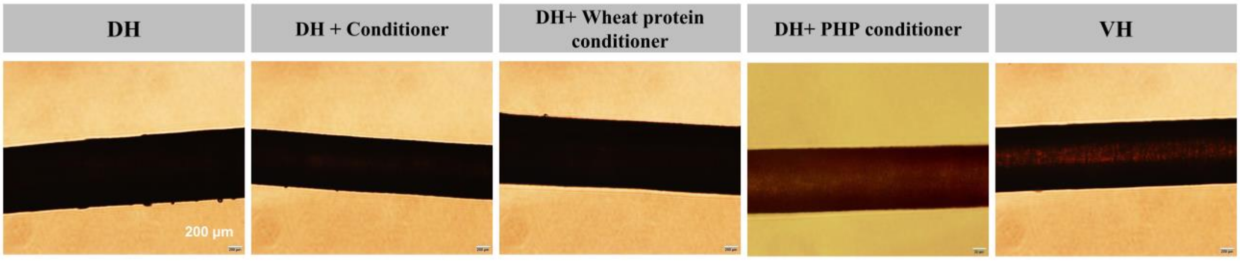

Figure 2. Microscopic observation of hair samples of each group after treatment with different hair conditioners

3.3. SEM Analysis of Hair Surface Morphology

The hair cuticle typically comprises multiple layers of overlapping proteins, cross-linked through disulfide bonds, forming a highly stable structure [14]. This structure exhibits a characteristic scale-like morphology resembling roof tiles, effectively protecting the hair from damage [15]. Figure 3 presents SEM images of DH and VH, magnified 1000×, 2000×, and 5000×, following 1 h treatment with different conditioners. SEM images provide a detailed view of the hair surface microstructure, revealing the arrangement and repair status of the cuticular scales. In order to compare the repair effect of ordinary conditioner, wheat protein conditioner and PHP conditioner on the hair cuticle. SEM observations revealed that DH exhibited disorganized cuticular scales, with some being raised or even fractured and detached. The surface texture appeared indistinct. Magnification at 5000× revealed the presence of residual material accumulating along the edges of the cuticular scales following conditioner treatment. Conventional conditioner treatment resulted in a greater amount of residual material and thicker scale edges, followed by wheat protein conditioner. PHP-containing conditioner demonstrated a relatively favorable outcome. After treatment, the scales of DH were arranged more neatly, with less residual material. The cuticle exhibited a satisfactory degree of closure, approaching the tiled-like stacking and well-defined texture observed in VH. These findings corroborate the conclusions drawn from optical microscopy. The large molecule chemical components in conventional conditioners accumulate on the hair surface without penetrating the cortex, while peptides, with their lower molecular weight compared to protein-based active substances, not only improve the surface morphology but also penetrate the hair fiber cortex, thereby exerting their beneficial effects.

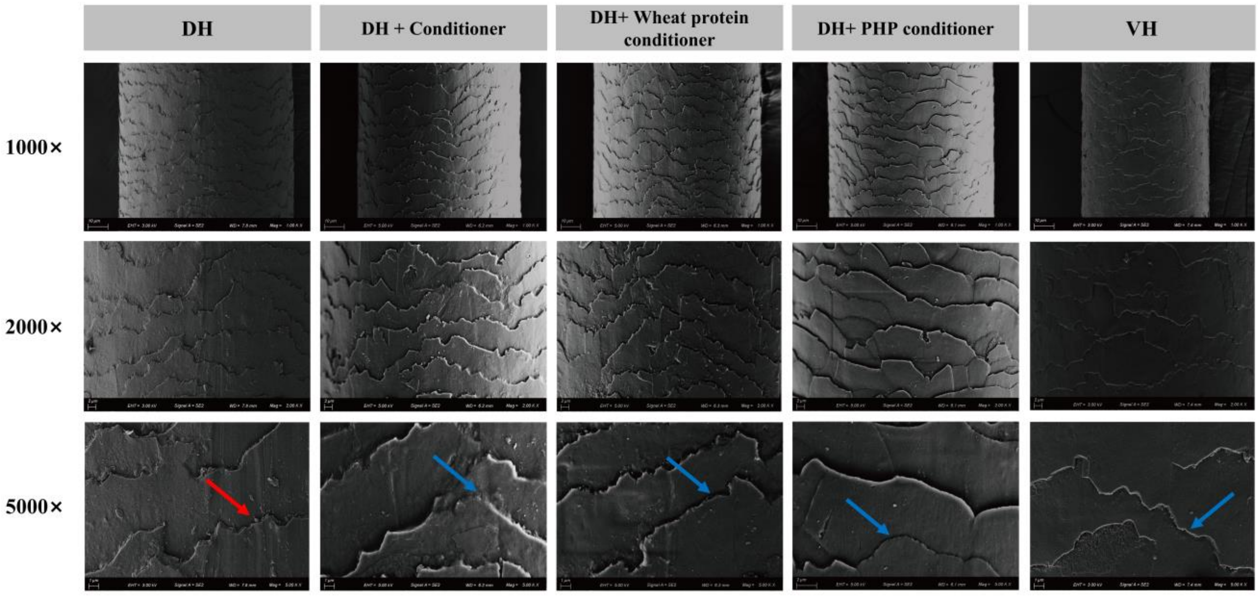

Figure 3. Scanning electron microscopy of the surface of hair samples of each group after treatment with different hair conditioners

3.4. DSC Analysis of Hair Thermal Decomposition

DSC can not only explore the moisture retention effect of hair, but also enables the investigation of keratin folding and stability by monitoring the heat changes occurring in hair fibers as a function of temperature. Hair samples exhibit thermal transitions as temperature increases, resulting in a peak on the DSC curve corresponding to the protein melting temperature. α-keratin displays a distinct α-helix absorption peak in the range of 230-250°C [16], primarily reflecting the structural integrity of α-helical proteins within microfibrils. Table 2 presents the influence of different protein peptide conditioners on the α-helix peak of DH. Notably, VH exhibits a higher ΔH_d compared to DH, indicating significant disruption of the α-helix protein structure in DH. Treatment with various conditioner resulted in gradual increases in ΔH_d for DH, with RHC increasing by 3.57%, 1.44%, 3.78%, and 5.74% respectively. The highest RHC value was observed after treatment with PHP-containing conditioner approaching that of NH. The porous and less compact structure of DH allows PHP to penetrate the hair interior in a layer-by-layer manner, interacting with hair fiber keratin. This interaction enhances the integrity and stability of the α-conformation structure of hair keratin, leading to a more stable and robust hair fiber structure. These findings demonstrate that PHP conditioner exhibits optimal repair efficacy for damaged hair and has obvious improvement on hair quality.

Table 2. Effect of different conditioners on α-helical peak content in DH and VH

Item	DH	DH + Conditioner	DH+Wheat protein conditioner	DH+PHP conditioner	VH
Td (℃)	225.09	223.72	223.79	224.39	223.22
⊿H_d (J/g)	6.781	7.0635	6.895	7.080	7.235
RHC (%)	85.74	89.31	87.18	89.52	91.48

3.5. Amino acid content analysis in hair interior

The hair cortex is primarily composed of α-keratin, which is consist of various amino acids. Notably, it contains high levels of cysteine and glutamic acid [17]. Table 3 presents the amino acid types and content in VH, DH, and DH treated with different conditioner. A comparison of the data reveals higher levels of cysteine, serine, and glutamic acid in VH. Conversely, the reduced cysteine and glutamic acid content in DH indicates specific damage to the hair fibers. Research suggests that glutamic acid, methionine, alanine, and leucine contribute to the formation of α-helical structures, crucial for hair stability [18]. VH exhibits significantly higher levels of hydrophobic amino acids and total amino acids compared to DH. Treatment with wheat protein conditioner and PHP conditioner increased the total amino acid content in DH, further supporting the penetration and absorption of proteins and peptides into the hair. Hydrophobic interactions play a primary role in maintaining the coiled-coil structure of keratin during intermediate filament formation. These interactions enhance the overall binding affinity of peptide-hair interactions, with a higher hydrophobic amino acid content correlating to a stronger affinity with hair [19]. PHP conditioner demonstrates superior repair activity compared to wheat protein conditioner. After treatment, DH exhibits an increase in total amino acids by 4.98% and hydrophobic amino acids by 2.67%, approaching the levels observed in VH. Therefore, peptide-based active ingredients in PHP conditioner can penetrate and fill gaps at different levels, both on the hair surface and within the hair interior, leading to improvements in the internal composition and subtle structural changes of the hair.

Table 3. Effect of different conditioners on amino acid species and content in DH and VH

Content (g/100g) / Amino acids	DH	DH + Conditioner	DH + Wheat protein conditioner	DH + PHP conditioner	VH
Aspartic acid	3.89 ± 0.26	3.81 ± 0.01	4.37 ± 0.01	4.67 ± 0.23	4.84 ± 0.04
Threonine	4.76 ± 0.31	4.58 ± 0.02	4.99 ± 0.00	4.93 ± 0.24	5.47 ± 0.02
Serine	6.73 ± 0.44	6.54 ± 0.01	7.28 ± 0.01	7.62 ± 0.37	7.87 ± 0.00
Glutamic acid	9.63 ± 0.65	9.59 ± 0.01	10.78 ± 0.01	10.71 ± 0.53	11.86 ± 0.02
Glycine	2.44 ± 0.16	2.51 ± 0.00	2.73 ± 0.00	2.72 ± 0.13	2.88 ± 0.02
Alanine#	2.16 ± 0.14	2.38 ± 0.00	2.71 ± 0.00	2.58 ± 0.12	3.03 ± 0.07
Cysteine	6.10 ± 0.71	4.27 ± 0.04	4.80 ± 0.01	9.15 ± 0.48	5.54 ± 0.03
Valine	3.09 ± 0.20	3.70 ± 0.01	4.17 ± 0.01	2.52 ± 0.12	4.56 ± 0.05
Methionine#	0.40 ± 0.03	0.77 ± 0.01	0.82 ± 0.01	0.62 ± 0.03	0.89 ± 0.06
Isoleucine#	2.05 ± 0.13	1.94 ± 0.01	2.21 ± 0.01	1.72 ± 0.08	2.45 ± 0.03
Leucine#	5.06 ± 0.26	4.59 ± 0.02	5.20 ± 0.02	6.37 ± 0.26	5.79 ± 0.05
Tyrosine	2.63 ± 0.13	2.44 ± 0.00	2.82 ± 0.00	3.72 ± 0.18	3.14 ± 0.05
Phenylalanine#	1.67 ± 0.13	1.63 ± 0.03	1.83 ± 0.00	2.62 ± 0.11	2.01 ± 0.00
Lysine	2.22 ± 0.13	2.11 ± 0.00	2.35 ± 0.01	2.31 ± 0.10	2.61 ± 0.01
Histidine	0.99 ± 0.08	7.84 ± 0.19	8.77 ± 0.11	1.04 ± 0.05	10.18 ± 0.43
Arginine	5.99 ± 0.42	6.01 ± 0.02	6.62 ± 0.01	6.45 ± 0.34	7.31 ± 0.00
Proline	4.91 ± 0.30	4.93 ± 0.03	5.33 ± 0.03	4.97 ± 0.50	5.70 ± 0.04
Hydrophobic amino acid	16.21	16.24	18.1	18.88	19.87
Total amino acid	69.74	69.63	77.78	74.72	86.14
Note: # is a hydrophobic amino acid.					

4. CONCLUSION

A novel fish-derived protein peptide conditioner incorporating *Pneumatophorus japonicus* heads peptides (PHP) as the active ingredient was formulated. The hair surface morphology, chemical composition, and other relevant parameters were evaluated to determine the repair efficacy of this PHP conditioner on damaged hair. The findings demonstrate that the PHP conditioner can effectively repair the surface morphology and structure of damaged hair. In terms of internal composition, differential scanning calorimetry analysis revealed that PHP conditioner treatment resulted in improved integrity and stability of the α-helical structure of keratin in damaged hair. Concurrently, the total amino acid content and hydrophobic amino acid content also exhibited significant increases. These results are attributed to the adhesive properties of PHP on the hair surface, forming a protective film that is gradually absorbed and utilized by the hair interior. This research offers promising insights and strategies for developing environmentally friendly hair care formulations based on proteins.

ACKNOWLEDGEMENTS

This work is financially supported by the National Natural Science Foundation of China (32371321).

REFERENCES

[1] R. D. Sinclair. Healthy Hair: What Is it? [J]. Journal of Investigative Dermatology Symposium Proceedings, 2007, 12(2):2-5.

[2] L. Fernández-Peña and E. Guzmán. Physicochemical Aspects of the Performance of Hair-Conditioning Formulations [J]. 2020, 7(2):26.

[3] G. Liobikienė and J. Bernatonienė. Why determinants of green purchase cannot be treated equally? The case of green cosmetics: Literature review [J]. Journal of Cleaner Production, 2017, 162:109-120.

[4] A. Martins, H. Vieira, H. Gaspar, et al. Marketed Marine Natural Products in the Pharmaceutical and Cosmeceutical Industries: Tips for Success [J]. 2014, 12(2):1066-1101.

[5] E. Malinauskyte, R. Shrestha, P. A. Cornwell, et al. Penetration of different molecular weight hydrolysed keratins into hair fibres and their effects on the physical properties of textured hair [J]. International Journal of Cosmetic Science, 2021, 43(1):26-37.

[6] A. L. Vazquez Villa, M. R. Senrra Aragao, E. P. Dos Santos, et al. Feather keratin hydrolysates obtained from microbial keratinases: effect on hair fiber [J]. Bmc Biotechnology, 2013, 13.

[7] S. B. Hwang, H. J. Park and B.-H. Lee. Collagen Hydrolysate from the Scales of Mozambique Tilapia (Oreochromis mossambicus) Improve Hair and Skin Health by Alleviating Oxidative Stress and Inflammation and Promoting Hair Growth and Extracellular Matrix Factors [J]. 2023, 21(9):475.

[8] C. G. Sotelo, M. Blanco, P. Ramos, et al. Sustainable Sources from Aquatic Organisms for Cosmeceuticals Ingredients [J]. 2021, 8(2):48.

[9] D. R. Miller, S. Das, K.-Y. Huang, et al. Mussel Coating Protein-Derived Complex Coacervates Mitigate Frictional Surface Damage [J]. ACS Biomaterials Science & Engineering, 2015, 1(11):1121-1128.

[10] S. B. Ruetsch, Y. K. Kamath, L. Kintrup, et al. Effects of conditioners on surface hardness of hair fibers: An investigation using atomic force microscopy [J]. Journal of Cosmetic Science, 2003, 54(6):579-588.

[11] M. F. R. Gavazzoni Dias. Hair Cosmetics: An Overview [J]. International Journal of Trichology, 2015, 7(1).

[12] M. J. L. Pereira, E. M. R. Neto, F. J. G. Junior, et al. Evaluation of the Behaviour of Violet Acid Pigment 43 in Formulations of Shampoos and Conditioners in Bleached Hair [J]. 2018, 10(3):282-287.

[13] Bobde, Anup, Sonchal, et al. Technical Evaluation of Leave-in type Hair Conditioner Formulations [J]. 2013.

[14] B. Bhushan, G. Wei and P. Haddad. Friction and wear studies of human hair and skin [J]. Wear, 2005, 259(7):1012-1021.

[15] T. Takahashi. A highly resistant structure between the cuticle and the cortex of human hair. II. CARB, a penetration barrier [J]. International Journal of Cosmetic Science, 2019, 41(1):28-35.

[16] C. R. Ribeiro De Castro Lima, L. D. Brocardo Machado, M. V. Robles Velasco, et al. DSC measurements applied to hair studies [J]. Journal of Thermal Analysis and Calorimetry, 2018, 132(3):1429-1437.

[17] K. Song, H. Xu, K. Xie, et al. Effects of chemical structures of polycarboxylic acids on molecular and performance manipulation of hair keratin [J]. 2016, 6(63):58594-58603.

[18] T. Takahashi and S. Yoshida. A highly resistant structure between cuticle and cortex of human hair [J]. International Journal of Cosmetic Science, 2017, 39(3):327-336.

[19] C. F. Cruz, N. G. Azoia, T. Matama, et al. Peptide protein interactions within human hair keratins [J]. International Journal of Biological Macromolecules, 2017, 101:805-814.