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Bali, Indonesia, 12–13 October, 2016
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Selected Peer-Reviewed Articles from the First International Joint Conference on Science and Technology (IJCST 2016), Bali, Indonesia, 12–13 October, 2016

The IJCST 2016 is an international scientific conference held in the beautiful island of Bali, Indonesia which was jointly organized by Faculty of Industrial Technology of Universitas Pembangunan Nasional "Veteran" Jawa Timur, Bali State Polytechnic, National Cheng-Kung University, Faculty of Social Science and Law of Universitas Negeri Surabaya, Faculty of Mathematics and Natural Sciences of Universitas Negeri Surabaya, and Faculty of Engineering of UniversitasTrunojoyo Madura.

The event was a place for international researchers, lecturers, students and practitioners to meet, greet, share and exchange their valuable ideas in the advancement of science and technology in the area of engineering, mathematics, natural science, education, and social. The IJCST 2016 believed in the view that to solve current and future potential complicated problems require a multidiscplinary approach and a joint effort of people and resources of various backgrounds.

The parallel session was divided into six groups where each group discussed topics in the domain guided by experts invited to lead. The followings are short descriptions of what was discussed in each group.

1. Engineering

This session discussed the theme "Science and Technology Innovation for Sustainable Development of Globally Competitive and Socio-Environmentally Friendly Industry." Researchers from various backgrounds which are Chemical Engineering, Industrial Engineering, Food Technology, Information Technology and Information Systems, Civil Engineering, and Mechanical Engineering exchanged ideas on how to solve problems from their respective backgrounds in order to sustain the industrial development globally but with less negative excess.

2. Social

The participants in this session came from social discipline and discussed the main theme "Role of Social Sciences, Education, Humanities to Realize Dignified Nation in Global Society." The sub-themes include Intelectual Community Rights, Social Change, Suistanable Development, Cultural Heritage, Media Literacy, Civics Education, Good Electronic Government, Education to Realize Dignified Nation, Role of Humanities Science to Realize Human Welfare, Development of Cross Cultural Communication in Global Society, Sustainable Development for Human Welfare, and Green Tourism.

3. Mathematics, Natural Sciences and Education

The last session discussed "Enhancement and Acceleration on Research and Learning in Mathematics, Natural Sciences and Education" with sub-themes in Education which include Mathematics, Physics, Chemistry, Biology, Science, Computer and Instructional Technologies, and Environment. Other subthemes are Teacher Training, E-Learning, Mathematics, Computer Science, Applied Mathematics and Statistics, Physics, Chemistry, Biology, Geophysics.

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RESEARCH ARTICLE

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Physical Characteristics of Fish Bone Gelatin Extracted Acid

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Gelatin is a hydrocolloid which can be used as gelling, thickeners and stabilizer. Generally, gelatin made from pig bone or cow skin. Some people can't use gelatin from pig because of religion reason. The other alternative to produce gelatin is using fish bone waste. Fishbone waste of tuna, shark, milkfish is a source of gelatin that safe to consumption was a novelty research. Tuna, shark and milkfish are usually consumed only the meat. The objective of the study was to use tuna, shark and milkfish bone waste as a gelatin by acid extraction method. The fishbone gelatin production is conducted in stages degreasing (cleaning), demineralization and extraction. Production gelatin were made from the bones waste of fish (milkfish), sharks and tuna with a concentration of citric acid (9%, 12% and 15%). The best treatment get gelatin from the bones of tuna was at a concentration of 15% citric acid to produce yield 9.27%, gel strength 206.20 g bloom, viscosity 5.90 cP and whiteness of 59.82%.

Keywords: Gelatin, Fish Bones, Shark, Tuna, Milkfish, Citric Acid, Extraction.

1. INTRODUCTION

Fish gelatin is an important alternative gelatin which can be considered as halal and acceptable by all religions. It is made from fish by-products of which fish skin is the most widely used part. The collagen and gelatin-like property of fish bones and scales coupled with their readily availability make it a potential source for development into gelatin products. Gelatin is a protein derivative product of collagen fibers derived from skin, bone and cartilage. Fish bone is a source of gelatin which is safe for human consumption, almost all types of fish and spiny skinned can be used as gelatin. Shark cartilage contains 19.2% protein,[1] tuna fish bones 26.02%[2] and fish belonging to protein toleostei 15–17%.[3] The three types of fish bone contains protein that is high enough so that potential as raw material to production gelatin. The producing of gelatin by hydrolysis method use acid and alkaline. Economically, the process of gelatin using acid is more effective than alkaline hydrolysis. In the acid hydrolysis can use a strong acid and a weak acid. Strong acid is used as Chloride acid. The type of acid used as a marinade solution will affect the physical properties of gelatin, especially color or degree of whiteness.

As described in the introduction, gelatin is derived from collagen which is the principal constituent of connective tissues and bones of vertebrate animals. Collagen is distinctive in that it contains an unusually high level of the cyclic amino acids proline and hydroxyproline. Collagen consists of three helical polypeptide chains wound around each other and connected by intermolecular crosslinks. Gelatin is recovered from collagen by hydrolysis. There are several varieties of gelatin, the composition of which depends on the source of collagen and the hydrolytic treatment use.[4]

The characteristics of gelatin obtained from the skin of tilapia, which manifested that the gelatin from tilapia skins has broad prospects for application.[5] Commercial gelatins are mostly obtained from pig and cow skins. However, the use of gelatin from those resources is restricted due to the outbreaks of bovine spongiform encephalopathy (BSE) or "mad cow disease" and religion reasons. Therefore, there is an increasing interest in the production of fish gelatin as an alternative for mammalian counterpart.[17] Due to its low gel strength of fish gelatin with low gelling temperature, it has been used for film preparation.[18]

Citric acid is a weak organic acid containing three carboxyl groups that can release protons in solution. Proton of citric acid will interact with the carboxyl group of collagen and can disrupt intra and inter molecular bonds tropokolagen so easily converted into gelatin.[11] In the production of fish food grade gelatin, citric acid is widely used because it does not affect the quality of the color or odor.[6] Gelatin quality can generally be assessed from the physical nature and content of certain mineral elements contained in gelatin. The physical properties of gelatin according to colorless or slightly yellow transparent, brittle, odorless, tasteless, sheet-like, flake or powder, soluble in hot water, glycerol and acetic acid and insoluble in solvents organic.[7] Gelatin is an important hydrocolloid which has widespread used in food

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applications. In generally, mammalian gelatin has been utilized due to its high melting, gelling point and thermo-reversibility.[16]

This study was to compare characteristic properties of tuna, shark, milkfish gelatins showing the importance of food applications. The present study was also to differentiate between three types of gelatins based on the studied parameters.

2. METHODOLOGY

2.1. Gelatin Extraction

The stages of the process of gelatin's production as follows:

(1) Preparation of fish bone waste (shark, tuna and milkfish) was separated from the flesh and the skin, then washed using running water.

(2) Degreasing, fish bones are soaked in water at temperature of 70–80 °C for 30 minutes. Remaining fiber and fat still attached to the bones of fish was cleaned again. After it drained and dried fish bones.

(3) Reducing Size, fish bones cut into small pieces with a size of 1–2 cm.

(4) Demineralization, bone fish marinated with citric acid solution (control, 9%, 12% and 15%) in a plastic container with acid resistant weight ratio of the sample and the volume of solvent is 1:3 for 48 hours to form ossein or fish bones were soft.

(5) Laundering, ossein washed to pH 5.0.

(6) Extraction, ossein extracted with distilled water and keep a water bath for 4 hours at a temperature of 70–80 °C.

(7) Filtration, gelatin extract was filtered

(8) Cooling, the filtrate was poured and stored in the refrigerator at a temperature of 4–10 °C for 24 hours.

(9) Drying, the filtrate was poured into a petri dish and dried for 2–3 days at 60 °C using an oven.

(10) Smoothing, a layer of gelatin formed on the entire surface of the petri dish is taken, then mashed.

2.2. Gelatin Gel Properties

2.2.1. Bloom Strength

Gelatin gel was prepared according to the British Standard 757:1975 method (BSI (British Standards Institution), 1975) with a slight modification. Gelatin (2.0 g) was mixed with 30 ml of distilled water in a 50 ml-beaker to obtain a final concentration of 6.67% (w/v). The mixture was stirred thoroughly and left at room temperature for 30 min to allow the gelatin to absorb water and swell. The mixture was then incubated at 42 °C for 30 min in a temperature-controlled water bath with occasional stirring. The beakers were then kept in a temperature-controlled chamber at 10 °C and allowed to stand for 16–18 h before determination of bloom gel strength. Bloom gel strength at 10 °C was determined by a Model TA-XT2i Texture analyzer (Stable Micro System, Surrey, UK) using a load cell of 5 kg equipped with a 1.27 cm diameter flatfaced cylindrical Teflon plunger. The dimensions of the sample were 3.8 cm in diameter and 2.7 cm in height. The maximum force (in grams) taken, when the penetration distance of 4 mm was obtained, was recorded. The speed of the plunger was 0.5 mm/s.[21]

2.2.2. Viscosity of Gelatin Solution

Gelatin solution was prepared by the method[19] with a slight modification.[20] Gelatin was dissolved with distilled water at 60 °C to obtain the final concentration of 6.67% (w/v). The solution was stirred until the gelatin was solubilised completely. Turbidity of gelatin solution was measured by reading the percent transmittance at 360 nm using a double-beam spectrophotometer (UV-16001, SHIMADZU, N.S.W., Australia).

2.2.3. Color Measurement of Gelatin Gel

The gelatin gel was prepared as described in gel preparation for bloom strength measurement. The color of gelatin gel was measured by a Hunter lab colour metre (ColorFlex, Hunter-Lab Reston, USA) and reported by the CIE system L^*, a^* and b^* parameters indicate lightness, redness/greenness and yellowness/blueness, respectively.

2.2.4. Statistical Analysis

All experiments were run in duplicate. Data were subjected to analysis of variance (ANOVA). Comparison of means was carried out by Duncan's multiple range test (Steel and Torrie, 1980). Analysis was performed using a SPSS package (SPSS 10.0 for window, SPSS Inc.

3. RESULTS AND DISCUSSION

3.1. The Yield

Value yield is an important parameter to determine the level of efficiency of the treatment process. Besides the yield can also be used for financial analysis which can be estimated amount of raw materials to manufacture products in a given volume.

The high concentration of citric acid used will result in more strength structures are open and hydrolyzed collagen to gelatin. The highest gelatin yield was obtained from shark bone waste, the higher the citric acid concentration used to hydrolyze the protein, the more gelatin yield produced. The yield of gelatin of tuna bones was much higher than that of milkfish (Fig. 1).

The rate of hydrolysis of collagen is affected by the amount of H^+ ions, the higher the concentration of citric acid solution, the lower pH of the solution, the H^+ ions to produce more. When the H^+ ions generated more so the faster the hydrolysis rate so that the resulting yield will increase. Alkali extraction/acid precipitation process resulted in the highest protein recovery of 98.77% (dry weight basis) and lowest fat content of 0.98% (dry weight basis). The extractable proteins of fish protein isolates (FPI) at various ionic strengths (0 to 0.6 M NaCl) exhibited majority of protein at 38 kDa with trace amount of other.[8] Citric acid derivative (CAD) prepared by modification of citric acid carboxylic

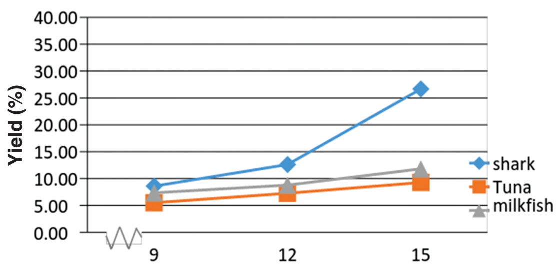

Fig. 1. The relations between fish bone type and concentration of citric acid on gelatin yield.

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Table I. The value gelatin gel strength.

Treatments				Value			
Fishbone	Concentration citric acid (%)	Gel strength (g bloom)	Viscosity (cp)	*L*	*a*	*b*	White degree
Shark	9	$110,55 \pm 0,78^{i}$	$2,90 \pm 0,00^{f}$	77,84	5,95	13,74	$66,12 \pm 0,22^{b}$
	12	$113,10 \pm 0,14^{h}$	$2,90 \pm 0,00^{f}$	75,96	7,13	12,72	$64,88 \pm 0,09^{cd}$
	15	$116,90 \pm 0,14^{g}$	$2,95 \pm 0,07^{f}$	74,69	7,93	11,82	$64,05 \pm 0,16^{d}$
Tuna	9	$185,35 \pm 0,49^{c}$	$4,95 \pm 0,07^{c}$	74,21	7,35	17,56	$61,32 \pm 0,28^{e}$
	12	$202,65 \pm 0,21^{b}$	$5,70 \pm 0,00^{b}$	74,15	7,18	18,96	$60,60 \pm 0,54^{ef}$
	15	$206,20 \pm 0,28^{a}$	$5,90 \pm 0,00^{a}$	73,45	7,69	19,30	$59,82 \pm 0,46^{f}$
Milkfish	9	$133,15 \pm 0,21^{f}$	$3,40 \pm 0,00^{e}$	83,59	4,68	14,28	$70,17 \pm 0,03^{a}$
	12	$156,35 \pm 0,78^{d}$	$4,20 \pm 0,00^{d}$	80,94	5,74	18,69	$65,61 \pm 0,06^{bc}$
	15	$151,40 \pm 0,57^{e}$	$4,05 \pm 0,07^{d}$	79,94	5,25	18,33	$65,56 \pm 0,02^{bc}$

groups with NHS was introduced for cross-linking of gelatine through its amino groups leading to amide bonds formation.[9]

Degradation product, gelatine, taking into account their molecular and submolecular structural properties, possibilities to overcome common problems related to their usage as biomaterial, i.e., the solubility and degradation rate mechanisms, as well as their applications in combination with other types of (bio) polymers.

3.2. Strength Gel

Gelatin gel strength is one of the parameters of the texture and the style to produce a certain deformation.[10] For industrial using, the gel strength into consideration in determining the feasibility of using gelatin.[11] Gelatin gel strength increased at a concentration of citric acid 12%, but the concentration of citric acid 15% strength gelatin gel on the decline (Table I). This was thought to occur in the advanced hydrolysis of fishbone collagen when soaked with a solution of citric acid with a high concentration (15%). In the event of further hydrolysis, all of the hydrogen bond and a covalent bond that links between chains of amino acids to each other will be cut shorter. This caused the molecular weight collagen became smaller resulting in a low gel strength. The gel strength overcome decrease due to the process of termination of the polymer chains of amino acids with increasing acid concentration, so that the bonds between the polymer molecules making up collagen to break into a very short monomer chains and damaged. This causes the gel formation process to be reduced.

Gelatin gel strength in the cartilage of sharks and tuna had increased along with the increasing concentration of citric acid. This was allegedly due to a concentration of 15% citric acid, amino acid chains are formed on both the long and the gelatin has not been damaged. In addition, due to a concentration of 15% citric acid, gelatin cartilage of sharks and tuna have not decreased so that the molecular weight of the gel strength was still stable. The gel strength depends on the length of the amino acid chain. If the condition has been hydrolyzed collagen is perfect, then the gel strength can be increased. This happens because the collagen that has been hydrolyzed can produce long polypeptide chains. Gelatin gel can be stabilized by external pressure covalent bonds are hydrogen bonds, because the covalent bond accelerating melting gel.[12] The each addition of citric acid concentration up to a certain concentration could increase the presence of the amino acids proline and hydroxyproline so that the resulting gel will be more stable.[3]

Gel strength of gelatin in this research was higher of Bloom strength of gelatin hydrolysate bigeye snapper skin (62.6 ± 2.1 g) and similarity gelatin from bigeye snapper skin extracted at 45 °C for 12 h (*P. tayenus*: 227.7 ± 6.20 g, *P. macracanthus*: 254.10 ± 11.13 g).[21]

3.3. Viscosity

Viscosity is one of the physical properties of gelatin which is quite important. The highest viscosity of tuna gelatin with citric acid concentration of 15%, i.e., 5.90 cP (Table I). Fish bone gelatin viscosity are influenced by the content of non-collagen components such as ash and minerals.[13] The presence of minerals that belong to the type of ash in amounts too much influence the characteristics of gelatin gel, such as gel strength, melting point, and viscosity. The low viscosity is also influenced by the distribution of the gelatin molecules in solution as well as the molecular weight of gelatin. If the group of gelatin binds to minerals it will caused the molecular bonds of the gelatin became less so that the distribution of the gelatin molecules faster and viscosity values to be down.

Viscosity fishbone gelatin has increased to citric acid especially milkfish bone gelatin, increasing concentration of citrate acid (12%), but decreased when citric acid concentration increased to 15% (Table I). This is presumably because the amino acid chain fish bones begin to experience disconnection became shorter when the concentration of citric acid to be increased to 15%. The use of citric acid as a marinade solution with a concentration of 12% was the optimum treatment for bone fish gelatin for the concentrations of the amino acid chain of fish bones still long and not damaged, marked the highest viscosity. This is different from gelatin cartilage of sharks and tuna, which continues to increased in viscosity due to the high concentration of citric acid. Allegedly chain amino acids tuna fish bone remains long and not broken until the citric acid concentration of 15%. Bone structure tuna and shark cartilage was harder than the bone fish. That the amino acid chain gelatin will be interrupted due to the increased concentration of the solvent that is characterized by impairment of viscosity.[14]

3.4. White Degree

Whiteness is a general description of the color of gelatin. Generally, the degree of white gelatin is expected to approach 100% for high-grade gelatin is usually colorless so that a wider application. Whiteness of the gelatin will take effect on the application of a product.[15]

Impaired whiteness gelatin along with the increasing concentration of citric acid solution. Solution of citric acid can caused

interactions with protein molecules that affect the color brightness level. The high concentration of citric acid during the demineralization may resulted gelatin becomes darker color.

4. CONCLUSION

The results of this study, the best treatment was the tuna bone gelatin with citric acid concentration of 15%, where the treatment resulted in the yield of 9.27%, gel strength 206.20 g bloom, viscosity 5.90 cP and whiteness of 59.82%.

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