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Characteristics of Shrimp Head Protein Hydrolysate Flour (*Litopenaeus Vannamei*) with Addition of Different Papain Enzym Concentrations

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A B S T R A C T

Shrimp head was one of the high protein wastes produced by the shrimp meat processing industry, one form of utilization was protein hydrolysate flour. The papain enzyme was chosen as the hydrolyzing agent because of its ability to specifically hydrolyze proteins without causing damage. Foam-mat drying was a method of drying food by bubbling to dryness, which was relatively cheap and simple method. The aimed of this study was to determine the effect of papain enzyme addition on the properties of shrimp head protein hydrolysate flour. This research method used a completely randomized design (CRD) with different enzyme concentration treatments (control, 1%, 2% and 3%). The data obtained were tested with normality test, homogeneity test, ANOVA and Tukey analysis to determine the difference between of treatments. The results of data analysis showed that the addition of different concentrations of papain enzyme affected all the test parameters, namely yield, degree of hydrolysis, protein content, water content, glutamic acid and color. Based on the results of the study, the best shrimp head protein hydrolysate product was the addition of 3% papain enzyme concentration with a yield value of 16.36%, degree of hydrolysis 68.40%, protein content 39.91%, moisture content 4.83%, glutamic acid 18.78%, and a slightly light brown color with L (lightness) value (-35.03), a value (15.72) and b value (27.66).

Keywords: Shrimp head, protein hydrolysate, papain enzyme, foam-mat drying

1. Introduction

White shrimp (*Litopenaeus vannamei*) was one of leading fishery commodities that's growing rapidly in Indonesia and has a high economic value. Based on ITC Trademap, shrimp was still a superior commodity with an export value of nearly USD 2,04 billion or 8.8% of the import value of world shrimp in 2020 (Ministry of Marine Affairs and Fisheries, 2021). The solid waste produced from shrimp freezing industry can reach 50-60% which consists of head, offal, shell, etc. (Senphan and Bjakul, 2012; Nirmal *et al.,* 2020). Shrimp head parts that are not optimally utilized will result in environmental pollution, so it is necessary to implement a zerowaste system. Main components on the shrimp head consists of proteins, lipids, minerals and other components that can be reused as functional materials (Trang and Pham, 2012; Trang and Huynh, 2015; Trung *et al.,* 2022). One forms of shrimp head utilization was protein hydrolysate flour.

Protein hydrolysate was a concentrated and pure form of protein produced through chemical or enzymatic hydrolysis (Wangkheirakpam *et al.,* 2019). Chemical hydrolysis was the chemical breakdown of a molecule through the binding of water to produce small molecules using acids and bases such as sulfuric acid (H_2SO_4), perchloric acid, HCl, and NaOH. While the enzymatic hydrolysis uses enzymes to produce hydrolysate products that avoid changes and damage to the product. This hydrolysis reaction will produce high quality protein hydrolysates because the concentration of hydrolyzing components, pH, temperature, and hydrolysis time are controlled (Wangkheirakpam *et al.,* 2019). Hydrolysis process can use protease enzyme, one of them was papain enzyme because it was classified as exopeptidase or endopeptidase (Likhar and Chudasama, 2021). The purpose of this study was to determine the effect of adding papain enzyme concentration on the characteristics of shrimp head protein hydrolysate (yeald, degree of hydrolysis, protein content, moisture content, glutamic acid and color) and to determine the best concentration of papain enzyme oh shrimp head protein hydrolysate.

2. Materials and Methods

The materials used to make protein hydrolysate included shrimp heads, papain enzyme, aquades, CH_3COOH, NaOH, egg white, and maltodextrin. The main ingredient of shrimp head waste was obtained from Rejomulyo Fish Market, Semarang City.

Shrimp head protein hydrolysate flour was prepared by enzymatic hydrolysis using papain enzyme. Shrimp head was homogenized and mixed with aquades in a ratio of 1:4. Addition of papain enzyme with concentrations of 0%, 1%, 2%, and 3% for the hydrolysis process. Hydrolysis using a waterbath (WTB24 Memmert, Amerika Serikat) with 55ºC temperature for 6 hours. pH was adjusted to neutral (pH=7) using pH meter (Hanna HI2003-02, Amerika Serikat), where CH_3COOH as the acidic atmosphere regulator and NaOH as the basic atmosphere regulator. Enzyme inactivation was performed at 90ºC temperature for 20 minutes. Centrifugation at 4ºC for 15 minutes with 3500 rpm speed to obtain the solution fraction in the form of shrimp head protein hydrolysate using centrifuge (Gemmy, Taiwan). Shrimp head protein hydrolysate was dried with foam mat drying method using oven (Sharp Eo-28Lp, Jepang), where 5% egg white as foaming agent and 2% maltodextrin as a filler. The protein hydrolysate flour was stored in 50 ml airtight plastic packaging and then analyzed. There are 3 replicates treatments of papain enzyme concentration addition (0%, 1%, 2%, 3%) using Completely Randomized Design (CRD). The analysis parameters of protein hydrolysate consist of yield, degree of hydrolysis, protein content, moisture content, amino acids, and color. Data was analyzed using SPSS version 16 which consisted of normality test, homogeneity test, ANOVA, and Tukey analysis.

3. Result and Discussion

3.1 Proximate Composition of Shrimp Head

Table 1 - Proximate Content of Shrimp Head

Proximate	Average (wb) ± SD (%)
Moisture content	80.15 ± 1.02
Total Protein content	14.67 ± 0.65
Total Fat content	0.93 ± 0.63
Total Ash content	2.64 ± 0.05

Moisture content of shrimp head was relatively high up to 80.15% in line with the high moisture content of all types of marine biota. According to Rostini and Pratama (2018), moisture content of fresh vannamei shrimp reached 77.59%. Moisture content was an important element in food product because it can determine freshness, appearance, texture, and taste. Moisture content also determines the quality and acceptability of a food material.

The total protein content of shrimp heads analyzed reached 14.67%, this result shows that shrimp heads have protein content that are not much different when compared with protein content in body of seawater shrimp in general, as mentioned by Ali *et al.* (2017), that's the average protein content of seawater shrimp reached 17.10%. Protein contained was composed of amino acid which the role of building materials in the body and regulate metabolism in living cells. According to Jin *et al.* (2022), amino acid has a significant influence to the flavor of shrimp, because it was contain glutamic acid that can provide umami flavor.

The total fat content which is contained in vannamei shrimp head (*Litopenaeus vannamei*) reached 0.93%. These results are not to different with the research of Li *et al.* (2021) that the crode fat content in vannamei shrimp ranges between 0.8%-1.1%. The difference in fat content was influenced by shrimp type and life stage of shrimp when harvested. Shrimp that are in the moulting stage have higher fat content (Alhajee *et al.,* 2020).

Total ash content in the analyzed shrimp head reached 2.64%, these results are lower when compare to findings of Ghorai *et al.* (2016) on proximate analysis of the shrimp head which show that the ash content reached 19.82%. This difference occurs because ash content in food materials related to mineral content in the material. According to Liu *et al.* (2021), ash content reflects the content of inorganic compounds in biological samples to same extent. High ash content in shrimp indicated that the shrimp was rich in mineral element.

3.2 Characteristics of Protein Hydrolysate

Table 2 - Characteristics of Shrimp Head Protein Hydrolysate with Papain Enzyme Addition

Papain Enzyme Concentrations (%)	Yield (%)	Degree of Hidrolysis (%)	Protein Content (%)	Moisture Content (%)	Glutamic Acid (%)
0	9.80 ± 0.18^a	62.41 ± 0.10^a	34.83 ± 0.04^a	7.22 ± 0.06^c	16.76 ± 0.09^a
1	11.70 ± 0.29^b	65.69 ± 0.03^b	34.85 ± 0.05^a	8.63 ± 0.05^d	17.68 ± 0.10^b
2	14.28 ± 0.34^c	66.82 ± 0.15^c	37.07 ± 0.04^b	6.84 ± 0.15^b	18.42 ± 0.10^c
3	16.36 ± 0.30^d	68.40 ± 0.04^d	39.91 ± 0.06^c	4.83 ± 0.05^a	18.78 ± 0.11^d

Note:

- Data are the mean of 3 replicates ± standard deviation.

- Data followed by different superscript letters in the same column indicate significant differences (P < 5%).

a. Yield

The yield of shrimp head protein hydrolysate produced showed increasing results as the concentration of papain enzyme. These result according to the opinion of Barekat and Soltanizadeh (2017), that papain enzyme is one of plant enzyme which can degrade myofibrillar protein and connective tissue, thereby accelerating the process of osmosis and facilitating the release of water from tissues. The highest yield value was obtained from the treatment of 3% papain enzyme addition. The research result shows that the addition of different papain enzyme concentration affected to the yield of shrimp head protein hydrolysate. On the research of Noman *et al.* (2018) about protein hydrolysate of sturgeon fish with 3% papain enzyme obtained 17.47% yield value. The yield value on this research was also detected to be lower than Idowu *et al.* (2018) research about protein hydrolysate of salmon fish with 3% alcalase and papain enzyme addition which produced yield value with a range of 24.05% - 26.39%. The purpose of calculating the yield value was to find out the productivity of shrimp head protein hydrolysate with differences of methods used.

b. Degree of Hydrolysis

The addition of papain enzyme at different concentrations had a significant effect on the degree of hydrolysis of shrimp head protein hydrolysate. The highest degree of hydrolysis produced on shrimp head protein hydrolysate with addition of 3% papain enzyme, while the lowest degree of hydrolysis shown in control treatment. Papain enzyme used can be effectively break down proteins composed of amino acid known as polypeptides, in additional the enzymatic hydrolysis method can produce hydrolysate products that avoid changes and demage to nonhydrolyzed products (Noman *et al.* 2018). The degree of hydrolysis produced by shrimp head protein hydrolysate at the optimum condition of 68.40%, higher than whiting waste protein hydrolysate (Me*rlangius merlangus*) on the research of Korkmaz and Tokur (2022) amounting to 50.92% and lower than protein hydrolysate of trout fish waste (Onc*horynchus mykiss*) amounting to 74.30%.

Increased hydrolysis value influenced by several factors, such as enzyme concentration, enzyme type used, pH, and hydrolysis time. The higher the enzyme concentration, the higher the dissolved nitrogen value. Li *et al.* (2022), explained that total free amino acid increased along with the higher concentration of papain enzyme due to the breaking of the peptides bone that convert proteins into simpler forms containing the element N. Hydrolysis process with addition of enzyme at optimal condition can produced the higher degree of hydrolysis (Islam *et al.,* 2020).

c. Total Protein Content

The different addition of papain enzyme to shrimp head protein hydrolysate shown the highest result protein content in addition 3% papain enzyme. Protein content increases along with addition of papain enzyme concentration. The result on this research according to the opinion of Li *et al.* (2016), that addition papain enzyme treatment significantly increasing the soluble protein content and reducing gliadin levels in wheat flour. The higher protein content of protein hydrolysate also caused by the use of drying tool with foam mat drying method, where proteins will be easily damaged by heat, but in foam mat drying method the protein will be safe because the drying process used foam and low temperature. The total protein content in line with the research of Hardy and Jideani (2017), that explained the foam mat drying method can accelerates water evaporation and carried out with low temperature, so as not to demage cell tissue and lower nutritional value.

The protein content of shrimp head protein hydrolysate with addition of 3% papain enzyme was lower than the research by Idowu *et al*. (2018), that salmon fish protein hydrolysate with addition of alcalase enzyme and 3% papain enzyme amounting to 82.01% and also lower compared to the research by Nomat *et al.* (2018), that protein hydrolysate of sturgeon fish (*Acipenser sinensis*) with addition 3% papain enzyme amounting to 79.67%. This indicates that the sample and enzyme concentration was very impactful to hydrolysis product, because related to soluble nutritional value from the raw material used.

d. Moisture Content

The addition of different concentration papain enzyme significant effect on moisture content of shrimp head protein hydrolysate (P<5%). Karn and Kumar (2015) explained that increased of moisture content value because proteolytic power of papain enzyme tends to be more active, resulting in increased moisture content, the more active the proteolytic protease enzyme causing water demand to increase.

The moisture content in this research lower than the research by Annisa *et al.* (2017) that examined tilapia protein hydrolysate in addition 5% papain enzyme which has a moisture content of 9.8% with spray drying method. These differences may be due to the different kinds of sample and drying method used. According to Peng *et al.* (2023), the hygroscopic nature of protein hydrolysate greatly affects product quality.The moisture content increased due to absorption of moisture from the environment causes food products to deteriorate. Moisture content on the surface of material affected by relative humidity of surrounding air space.

The moisture content of protein hydrolysate resulting from this research was lower because foam mat drying method used to absorbing water, so the higher drying temperature, the water will be lost and the moisture content will be lower. Moisture content in this research in line with the opinion of Tontul and Topuz (2017) that explained the highest drying temperature resulting in the product with lower moisture content. The moisture content in a product affected by the length of the drying process, the extent of the evaporation process, and the drying process.

e. Glutamic Acid

Papain enzyme acts as a biocatalyst which accelerates the reaction of protein breakdown into amino acids, one of the being glutamic acid. This was in line with the opinion of Leni *et al.* (2020), that breaking reaction speed will be higher, so that the peptide bone will hydrolyzed a lot and the protein will more hydrolyzed into amino acids.

The highest glutamic acid content in shrimp head protein hydrolysate was found in addition of 3% papain enzyme concentration amounting to 18.78%, which was lower than the research by Ryu *et al*. (2021) for Atlantic Salmon Fish protein hyrolysate with a glutamic acid content amounting to 29% in addition of pepsin and pancreatin enzymes, while shrimp head protein hydrolysate higher than protein hydrolysate of red lionfish muscle in addition of alcalase enzyme which has a glutamic acid content of 13.24%. The differences glutamic acid content of the product affected by raw material type and not all amino acids can be easily hydrolyzed. In addition, a decrease in the rate of protein hydrolysis reaction is caused by several things, namely a decrease in the specific peptide bond for the enzyme, product inhibition, enzyme inactivation and the stability of the enzyme molecule which affects the binding of the enzyme to the substrate.

Glutamic acid was an amino acid type most commonly found on the fisheries product as a flavoring agent. According to Li *et al*. (2021), amino acid contained in protein hydrolysate with fish raw materials will produce amino acid with delicious flavor and aroma. Amino acid plays a role in addition of flavor is glutamic acid, aspartic acid, glycine and alanine.

3.3 Color of Shrimp Head Protein Hydrolysate

The results of the color analysis of shrimp head protein hydrolysate with different concentrations of papain enzyme are shown in Table 7. The addition of different enzyme concentrations had a significant effect (P < 5%) on the L (lightness), a (red/green) and b (yellow/blue) values of shrimp head protein hydrolysate color. The highest L value was obtained in the control treatment, while the lowest value was shown in addition of 2% papain enzyme concentration. Based on the brightness value test, the shrimp head protein hydrolysate with addition of enzyme concentration was included in the indication of dark color because it was below 50. According to Hunterlab (2012), the L-value (brightness) with low numbers (0-50) indicates darkness of color, while the L-value with high numbers (51-100) indicates brightness of color.

The results of a value test that has been carried out, it was found that addition of 3% papain enzyme concentration has the highest value (15.72), while the lowest value was in the control treatment, so the type of color possessed was included in the red color group. According to Sharma (2017), the value of a indicates chromatic color of red-green mixture with a value of +a (positive) from 0 to +100 for red and -a (negative) from 0 to -80 for green.

The b-value test obtained by adding 1% papain enzyme concentration had the highest b value of 31.49, while the lowest value was found in the control treatment. These results show that the b value test had a significant effect on all treatments. The results of b value on shrimp head protein hydrolysate with the addition of 1% papain enzyme concentration will have a yellowish color. The higher b value, the more yellow the sample. This is reinforced by Pathare *et al.* (2013) where the b* value indicates a yellowish color and -b* indicates a bluish color.

According to Table 7, the color of the control treatment was black-brown, the addition of 1% papain was slightly dark-brown, and the addition of 2% and 3% papain was slightly light-brown. The color of the protein hydrolysate was also determined by the pigments found in the fish used as raw materials. The color of the fish protein hydrolysate was also affected by the Maillard reaction during the hydrolysis process. The higher concentration of papain enzyme used, make higher the dextrose content. This results in more hydroxyl groups from reducing sugars, thus increasing the potential to react with amino groups from proteins to form Maillard products and cause a brown color (Poojary and Lund, 2022).

Elavarasan and Shamasundar (2016) stated that there was an interaction between free amino groups and aldehydes, so that aldehydes are formed during hydrolysis process. The hydrolysis was performed at 50°C, where lipids can undergo oxidation. In addition, the drying process (80°C) will contribute to the interaction of free amino groups and aldehydes resulting in browning of the product (non-enzymatic browning).

Table 6 - Glutamic Acid of Shrimp Head Protein Hydrolysate

Treatments	**Color Output**	**L**	**a**	**b**	Description
Control		-19.72^{d}	9.06^{a}	21.75^{a}	Blackish brown
1%		-28.94^{bc}	15.33^{b}	31.49^{cd}	Slightly dark brown

2%		-38.29^{a}	15.66^{b}	26.41^{b}	Slightly bright brown
3%		-35.03^{ab}	15.72^{b}	27.66^{bc}	Slightly bright brown

Note:

- Data are the mean of 3 replicates ± standard deviation.

- Data followed by different superscript letters in the same column indicate significant differences (P < 5%).

4. Conclusion

The best shrimp head protein hydrolysate flour was produced by the enzymatic hydrolysis process with addition of 3% papain enzyme concentration. The results of yield value were (16.36%), degree of hydrolysis (68.40%), protein content (39.91%), moisture content (4.83%), glutamic acid (18.78%), with a slightly light brown color.

Conflict of Interest

The authors declare no conflict of interest.

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References

Alhajee, S. A., Alnoor, S. S., and Sultan, E. (2020). Moulting cycle related changes in biochemical, antioxidants genes and feeding rates in commercial penaeidae shrimps *Metapenaeus affinis* (H. Milne Edwards, 1837). *Plant Archives*, 20(2), 17-24.

Ali, S. S. R., Ramachandran, M., Chakma, S. K., and Sheriff, M. A. (2017). Proximate composition of commercially important marine fishes and shrimps from The Chennai Coast, India. *International Journal of Fisheries and Aquatic Studies*, 5(5), 113-119.

Annisa, S., Darmanto, Y. S., and Amalia, U. (2017). The effect of various fish species on fish protein hydrolysate with the addition of papain enzyme. *Journal of Fisheries Science and Technology*, 13(1), 24-30.

Barekat, S., and Soltanizadeh, N. (2017). Improvement of meat tenderness by simultaneous application of high-intensity ultrasonic radiation and papain treatment. *Innovative Food Science and Emerging Technologies*, 39, 223-229.

Elavarasan, K. and Shamasundar, B. A. (2016). Effect of oven drying and freeze drying on the antioxidant and functional properties of protein hydrolysates derived from freshwater fish (*Cirrhinus mrigala*) using papain enzyme. *Journal of Food Science and Technology*, 53, 1303-1311.

Ghorai, T., Sarkar, S., and Dora, K. C. (2016). Proximate composition analysis of shrimp head waste (SHW) fermented by convetional and biofermenter methods. *Journal Environment and Ecology*, 34(1), 74-77.

Hardy, Z. and Jideani, V. A. (2017). Foam-mat drying technology: A review. *Critical Reviews in Food Science and Nutrition*, 57(12), 2560-2572.

Hunterlab. (2012). Hunter L, a, b, vs CIE L*, a*, b*: Measuring Color Using Hunter L, a, b, versus CIE 1976 L*, a*, b*. *Hunter Associates Laboratory Inc*.

Idowu, A. T., Benjakul, S., and Sinthusamran, S. (2018). Protein hydrolysate from salmon frames: Production, characteristics and antioxidative activity. *Journal of Food Biochemistry.* 1-12.

Islam, M. S., Hongxin, W., Admassu, H., Noman, A., Ma, C., and An Wei, F. (2021). Degree of hydrolysis, functional and antioxidant properties of protein hydrolysates from grass turtle (*Chinemys reevesii*) as influenced by enzymatic hydrolysis conditions. *Food Science and Nutrition*, 9(8), 4031-4047.

Jin, Y., Xu, M., Jin, Y., Deng, S., Tao, N., and Qiu, W. (2022). Simultaneous detection and analysis of free amino acids and glutathione in different shrimp. *Foods*, 11(17), 2599.

Karn, S. K. and Kumar, A. (2015). Hydrolytic enzyme protease in sludge: Recovery and its application. *Biotechnology and Bioprocess Engineering*, 20, 652-661.

Korkmaz, K. and Tokur, B. (2022). Optimization of hydrolysis conditions for the production of protein hydrolysates from fish wastes using response surface methodology. *Food Bioscience*, 45, 101312.

Leni, G., Soetemans, L., Caligiani, A., Sforza, S., and Bastiaens, L. (2020). Degree of hydrolysis affects the techno-functional properties of lesser mealworm protein hydrolysates. *Foods*, 9(4), 381.

Li, L., Chen, H., Lü, X., Gong, J. and Xiao, G. (2022). Effects of papain concentration, coagulation temperature, and coagulation time on the properties of model soft cheese during ripening. *LWT-Food Science and Technology*, 161, 113404.

Li, X., Wang, Y., Li, H., Jiang, X., Ji, L., Liu, T. and Sun, Y. (2021). Chemical and quality evaluation of pacific white shrimp *Litopenaeus vannamei*: Influence of strains on flesh nutrition. *Food Science and Nutrition*, 9(10), 5352-5360.

Li, Y., Yu, J., Goktepe, I. and Ahmedna, M. (2016). The potential of papain and alcalase enzymes and process optimizations to reduce allergenic gliadins in wheat flour. *Food Chemistry*, 196, 1338-1345.

Li, Y., Wang, X., Xue, Y., Ruan, S., Zhou, A., Huang, S., and Ma, H. (2021). The preparation and identification of characteristic flavour compounds of maillard reaction products of protein hydrolysate from grass carp (*Ctenopharyngodon idella*) bone. *Journal of Food Quality*, 2021(1), 8394152.

Likhar, V. and Chudasama, B. J. (2021). Seafood enzymes and their potential industrial applications. *Journal Entomology and Zoology Studies*, 9(1), 1410-1417.

Ministry of Marine Affairs and Fisheries. (2021). Indonesia's rank as a world exporter of fishery products increases during the pandemic. can be accessed on https://kkp.go.id/djpdspkp/artikel/33334-peringkat-indonesia-sebagai-eksportir-produk-perikanan-dunia-meningkat-di-masa- pandemi. Accesed by August 4th, 2024.

Nirmal, N. P., Santivarangkna, C., Rajput, M. S., and Benjakul, S. (2020). Trends in shrimp processing waste utilization: an industrial prospective. *Trends in Food Science and Technology*, 103, 20-35.

Liu, Z., Liu, Q., Zhang, D., Wei, S., Sun, Q., Xia, and Liu, S. (2021). Comparison of the proximate composition and nutritional profile of byproducts and edible parts of five species of shrimp. *Foods*, 10(11), 2603.

Noman, A., Y. Xu, W. Q. Al-Bukhari, S. M. Abed, A. H. Ali, A. H. Ramadhan, and W. Xia. (2018). Influence of enzymatic hydolysis conditions on the degree of hydrolysis and functional properties of protein hydrolysate obtained from chinese sturgeon (*Acipenser sinensis*) by using papain enzyme. *Journal Process Biochemistry*, 67, 19-28.

Pathare, P. B., Opara, U. L. and Al-Said, F. A. J. (2013). Colour measurement and analysis in fresh and processed foods: A review. *Food and Bioprocess Technology,* 6, 36-60.

Peng, X., Liu, C., Wang, B., Kong, L., Wen, R., Zhang, H., Xiaobo Y., Yun B., and Aera J. (2023). Hygroscopic properties of whey protein hydrolysates and their effects on water retention in pork patties during repeated freeze–thaw cycles. *LWT-Food Science and Technology*, 184, 114984.

Poojary, M. M. and Lund, M. N. (2022). Chemical stability of proteins in foods: Oxidation and the maillard reaction. *Annual Review of Food Science and Technology*, 13(1), 35-58.

Rostini, I., and Pratama, R. I. (2018). Effect of steaming on physical and chemical characteristics white shrimp (*Litopenaeus vannamei*) from Indramayu waters. *In IOP Conference Series: Earth and Environmental Science,* 176(1), p. 012046.

Ryu, B., Shin, K. H., and Kim, S. K. (2021). Muscle protein hydrolysates and amino acid composition in fish. *Marine Drugs,* 19(7), 377.

Senphan, T., and Benjakul, S. (2012). Compositions and yield of lipids extracted from hepatopancreas of pacific white shrimp (*Litopenaeus vannamei*) as affected by prior autolysis. *Food Chemistry*, 134(2), 829-835.

Sharma, G. (2017). Color Fundamentals for Digital Imaging. In Digital Color Imaging Handbook (pp. 1-114). CRC press.

Tontul, I., and Topuz, A. (2017). Spray-drying of fruit and vegetable juices: effect of drying conditions on the product yield and physical properties. *Trends in Food Science and Technology*, 63, 91-102.

Trang, S. T. and Huynh, N. D. B. (2015). Physicochemical properties and antioxidant activity of chitin and chitosan prepared from pacific white shrimp waste. *International Journal of Carbohydrate Chemistry*

Trang, S. T. and Pham, T. D. P. (2012). Bioactive compounds from by-products of shrimp processing industry in Vietnam. *Journal Food Drug Anaysisl.* 20, 194–197.

Trung, T. S., Tran, H. V., Le, M. H., Ky, P. X., Brown, P. B., and Van Ngo, M. (2022). Growth performance, haematological parameters and proximate composition of rainbow trout *Oncorhynchus mykiss* Fed Varying Dietary Levels of Protein Hydrolysate from Heads of *Penaeus monodon* shrimp processing industry. *Regional Studies in Marine Science*, 55, 102643.

Wangkheirakpam, M. R., Mahanand, S. S., Majumdar, R. K., Sharma, S., Hidangmayum, D. D. and Netam, S. (2019). Fish waste utilization with reference to fish protein hydrolisate-A review. *Fishery Technology*, 56(3), 169-178.