**INFO**

**Corresponding Author:**
Huyen Thi Thuy Loan, Faculty of Applied Sciences - Health, Dong Nai Technology University, Bien Hoa, Dong Nai, Vietnam.

**E-mail Id:**
huynthithuyloan@dntu.edu.vn

**Orcid Id:**
https://orcid.org/0000-0002-4637-9947

**How to cite this article:**
Duyen NTT, Phuong NTL, Loan HTT et al. Production of Shrimp Pork Powder for Children. *J Adv Res Food Sci Nutr* 2020; 3(1): 50-55.

**Date of Submission:** 2019-12-14

**Date of Acceptance:** 2020-01-28

---

**ABSTRACT**

This research was to study the production of pork powder through hydrolysis by Protamex enzyme with adding naturally sweet flavor from shrimp. Through the research content, factors affect the hydrolysis of protein, including the ratio of enzyme/pork meat (E/S), hydrolysis temperature, hydrolysis time were investigated. The time and temperature factors of the concentration and drying process affect product quality. The Results showed that the properly hydrolysis condition as followed: 2/100 of enzyme/pork meat (E/S) at 55°C. The time of hydrolysis protein was the best quality product about 4 hs. At time, the ratio of shrimp to pork meat is (25:75); the parameters of drying process were as follow: 80°C, 30 minutes. The shrimp pork powder was finally found at 18.8 g/ 100 g total protein content, 1889.2 µg/l soluble protein concentration with highly preferred levels of panelist.

**Keywords:** Hydrolyzed Protein, Protease Enzyme, Pork Powder, Shrimp Powder

---

**Introduction**

Among foods for children, pork meat is also one of the most chosen foods. Dietary food guides, such as the USDA food guide pyramid, are tools to inform the public about diet, nutrition and health (Lachance & Fisher, 2005). In the food guide pyramid, meat is categorized as a protein food group along with poultry, fish and eggs. Undoubtedly, meat is not only a major source of food proteins with high biological value in many countries, it is also an excellent source of some valuable nutrients such as minerals and vitamins (Biesalski, 2005; Chan, 2004; Mulvihill, 2004). In which, some nutrients (e.g., iron, vitamin B12, and folic acid) are either not present or have inferior bioavailability in other foods (Keizo Arihara, 2006). Playing a key role in the contribution of protein for people in Viet Nam, the pig is the fastest growing sub-sectors in Vietnamese agriculture with approximately 90% of meat consumed. To improve digestibility, and modify of sensory quality (such as tex-ture or taste), processes that involve protein hydrolysis steps can be applied. Products of hydrolysis protein are peptide or amino acid but chemical processes, including alkaline or acid hydrolysis, with difficult control characteristics, normally create products with modified amino acids (Olga Luisa Tavano, 2013). Enzymatic hydrolysis can be performed under mild conditions, and could avoid the extreme environments required by chemical treatments. Usually, enzymatic processes avoid side reactions and do not decrease the nutritional value of the protein source (Jose Maldonado, 1998). Additionally, enzymes present substrate specificity which permits the development of protein hydrolysates with better defined chemical and nutritional characteristics (Olga Luisa Tavano, 2013). The study of Fonseca (2016) demonstrates that the Protamex enzyme showed higher capacity when compared to Alcalase and Flavourzyme enzymes to hydrolyze fish proteins (Fonseca, 2016).

One of the important functional properties of protein hydrolysates is solubility (M.I. Mahmoud, 1992). Increasing the degree of hydrolysis enhances the solubility of the total hydrolysates (Souissi, 2007). This is due to the cleavage of proteins into smaller peptides that usually have increased...
solubility (F. Shahidi, 1994). So, this research would evaluate degree of protein hydrolysates through solubility.

For infants younger than 1 year-old, they are more extremely sensitive than adults about flavorings. They need to avoid foods that heavily salted, buttered or sweetened to ensure their long-term good health and lifelong dietary habits. (Robert M. Selig, 2013). Shrimps could be added to improve food sensory. They contain about 18%-20% protein, 75%-80% water, low fat (4.5%), amino acids and minerals such as selenium, copper, zinc,] and calcium in right proportion (Yanar Y, Celik M, 2006). Shrimps also contain polyunsaturated fatty acids necessary to improve human health and are also rich in astaxanthin and vitamin B12 with antioxidant properties (Entsua-Mensah M, 2002).

Especially, naturally sweet flavour of shrimp is one of characteristic which used to improve food sensory. That would be mixed to enrich protein, colour, and flavour for protein hydrolysates power.

Material and Methods

Material

Lean pork and shrimp

Lean meat of porks, were selected because it contains a relatively high proportion of meat muscle and little connective tissue or fat to reduce variability in the hydrolysis reaction to enzyme (Noriza Binti Ahmad, 2016). Lean meat of pork was provided by GreenFeed Viet Nam Foods Joint Stock Company; Shrimp (white shrimp, Litopenaeus vannamei) was bought from BigC's supermarket.

Lean meat of porks and shrimps were transported in ice-filled containers at 0-4°C to the Laboratory of Food Technology at Dong Nai Technology University. Then, they were washed, and connective tissue was removed. After that, Shrimps were beheaded, eviscerated and organs were removed. Then, meat and shrimp were minced (0.5-1 mm) and stored frozen placed in plastic containers at -18 ± 2°C, pending use.

Enzymes

The selected enzymes were Protamex (Serin protease) with 5000 IU/g supplied by Nam Giang Company, at 133/11 Ho Van Hue Street, Ward 9, Phu Nhuan District, Ho Chi Minh City.

Enzyme

The ground pork meat was dispersed in water with a ratio of 1:0.5 (w/w). Then hydrolyzed meat using the ratio of enzyme/pork meat (E/S) was (0.5/100; 1/100; 1.5/100; 2/100; 2.5/100) (w/w). The best enzyme ratio was subsequently used to study the effect of incubation temperature at different temperatures (45°C, 50°C, 55°C, 60°C, 65°C). The best temperature was then used to determine the effect of incubation time at different times (1 h, 2 h, 3 h, 4 h, 5 h).

In shrimp pork powder, the ratio of shrimp to pork meat calculated in 100g powder total (10:90; 15:85; 20:80; 25:75; 30:70) was surveyed. The mixture would be concentrated in 44 minutes, 90°C.

In this operation, the mixture was dried in different drying times (10 minutes, 20 minutes, 30 minutes, 40 minutes, 50 minutes) to convert the hydrolysate to a powder form, which was incorporated into food formulation. The moisture of the final product was chosen 15% to give a safe level, which impacts some technological advantages, such as increased shelf life, ease of handling and transportation, impartment of desirable characteristics such as odor, flavor, color, and taste. These experiences would be conducted to determine the best parameter, which had the most assessment by trained panelists.

Analysis Method

The soluble protein concentration of pork was carried out using Lowry method and Folin - Ciocalteu that used as a standard.

Nitrogen content was analyzed by Kjeldahl standard method according to Vietnam Standards 3705 - 90.

Statistical analysis

Results were expressed as JMP 10 applied for comparison of means (p < 0.05) and Excel.

Sensory Evaluation

Sensory evaluation was conducted on the shrimp pork powder using 20 trained panelists. Quality parameters evaluated were overall acceptability using ranking sensory method. Panellists would assign the sample as follows:

- The most acceptable sample
- The next most acceptable sample
- Neither like nor dislike
- Dislike very much
- Unacceptable sample

Result and Discussion

Effect of the ratio of enzyme/pork meat (E/S) on soluble protein/total protein (P_s/P_t) ratio in hydrolysate

In Figure 1, indicated that the higher E/S ratio, the higher
P_s/P_t ratio, E/S ratio greatly affects the hydrolysis process. Tukey’s HSD test showed that all of P_s/P_t ratio means were statistically different (p < 0.05) except for sample made with 2/100 and 2,5/100 of E/S, which were not statistically different from each other (p > 0.05). Shrimp pork powder had the highest Ps/Pt ratio of 56.89% and 57.11% whereas the lowest P_s/P_t ratio of 41.49%.

The difference in solubility observed among hydrolysates can be due to peptide length and the ratio of hydrophilic/hydrophobic peptides (Souissi, 2007). When there was higher enzyme content, higher smaller peptides were there. This makes solubility of increasing protein hydrolysate.

In this research, E/S ratio of 2/100 (w/w) was selected as a fixed factor to conduct the next experiments.

Effect of hydrolysis time on soluble protein / total protein ratio (P_s/P_t)

As expected, Figure 2 showed that Ps/Pt ratio increased with hydrolysis time, showing then reduced to the stationary phase (Melissa Ferreira SBRDGGID, 2016). In which the degree of hydrolysis became constant at 54.9% and 55.12% for 4 h and 5 h, which were not statistically different from each other (p > 0.05).

Similar the result have been reported for several protein substrates, such as peanut (Jamdar et al., 2010), Jatropha curcas seed cake (Selanon et al., 2014), rice protein (Li et al., 2016) and corn protein (Jin et al., 2016). Such behavior can be related to the reduction in peptide bonds capable of being cleaved, competition between the substrate and the hydrolysis products and enzyme denaturation that decreases its activity (Adler-Nissen, 1986).

According to this result, hydrolysis time of 4 h was selected as a fixed factor to conduct the next experiments.
Effect of hydrolysis temperature on the ratio of soluble protein/total protein ($P_s/P_t$)

Figure 3 shows that the temperature of 55°C gave the highest $P_s/P_t$ (%) compared to other temperatures such as 45°C, 50°C, 60°C, 65°C, which were statistically different from each other ($p < 0.05$). According to the results, $P_s/P_t$ (%) significantly increased ($p<0.05$) when the temperature was increased from 45 to 55°C. And then, it started to decrease at the temperature of 60°C (54.23% down to 50.35%).

Diniz and Martin (1997), studying the hydrolysis of dogfish proteins with the Alcalase 2.4L enzyme, suggested that the constant DH decreasing behavior can happen due to the lack of available peptide bonds combined with a partial deactivation of the enzyme during the course of the hydrolysis.

In this examination, the hydrolysis temperature at 55°C was selected as a fixed factor to conduct the next experiments.

**Effect of Mixed Shrimp Ratio**

**Table 1. Sensory score on shrimp mixing ratio**

| Shrimp: pork meat ratio (%) | 10:90 | 15:85 | 20:80 | 25:75 | 30:70 |
|-----------------------------|-------|-------|-------|-------|-------|
| Sensory score               | 2.1c  | 3.05b | 3.23b | 4.46a | 2.16c |

The results of statistical analysis showed that the proportion of shrimp mixed affecting the senses results of the product is significant at the 95% confidence level ($p <0.05$). Shrimp:pork meat ratio (%) at 90:10 sample has the lowest sensory point 2.1c, the product has an odorless, tasteless smell. The remaining samples have relatively low sensory points, the product has an unsatisfactory smell and taste. Shrimp:pork meat ratio (%) at 30:70 has the highest average score of 4.46a and is statistically different from the rest of the samples showing that when mixing 25% shrimp ratio, shrimp have high calcium, rich protein, and many types of pigments carotenoid called astaxanthin (Dong et al., 2014, Papa et al., 2015, Elagamy et al., 2018, Zuluaga et al., 2018). Astaxanthin has proven to be excellent biological activities including anti-cancer and anti-aging, repairing central nervous system, improve cardiovascular function and protecting eyesight (Wu et al., 2015, Visioli & Artaria, 2017, Zuluaga et al., 2018). Shrimp increases the nutritional such as and sensory value of the product. However it contains trimethylamine, so when supplemented with high content, the product has a slight fishy odor, which reduces sensory value. From the experimental results, we choose the appropriate proportion of shrimp is 25%.

**Effect of Drying Time and Temperature on Product Quality**

The sprinkle product has a typical yellow color, shrimp smell and taste. reasonable.

| Level | Mean            |
|-------|-----------------|
| 55    | 54.230000       |
| 60    | 52.460000       |
| 65    | 50.350000       |
| 50    | 47.250000       |
| 45    | 42.710000       |

Levels not connected by same letter are significantly different.
shrimp pork powder at 80°C - 32 minutes exhibited an acceptable color to the consumer.

The results of the microbial analysis showed that the shrimp pork powder at 80°C were free of mold and coliform growth. They had the total viable count (TVC) of 1 × 10^4 cfu/g, E.Coli (-), Salmonella (-), Staphylococcus aureus (-), Clostridium perfringens (-). Based on the sensory scores, dried at 80°C with 32 minutes had the highest rating in terms of overall acceptability and was acceptable among the different drying conditions.

Table 2. Sensory score on shrimp mixing ratio

| Temperature (°C) – Time (minutes) | 70-37,5 | 80-32 | 90-26,4 | 100-20 |
|----------------------------------|--------|-------|--------|-------|
| Mean sensory scores              | 1.11^d | 3.91^a| 3.08^b | 1.88^c|

Table 2 showed the mean sensory scores of drying with 15% moisture for shrimp pork powder. Highest overall acceptability of drying at 80°C - 32 minutes was observed. It was preferred more than the drying at 70°C - 37,5 minutes, 90°C -26,4 minutes, 100°C -20 minutes. Visual appearance as reported by Zhang et al., 2008 is a vital quality parameter, which influences sales and acceptability of a product, hence

Conclusion

The mechanism of hydrolysis of pork by Protamex enzyme with 2/100 of enzyme/pork meat (E/S), 4 h hydrolysis time and 55°C hydrolysis temperature gained the most effective hydrolysis. At time, the ratio of shrimp to pork meat calculated in 100 g power total was (25:75): the parameters of drying process were as follow: 80°C, 30 minutes reaches 15% moisture and contains a total protein content of 18.8 g/100 g of raw materials. The final product had highly preferred levels of panelist. The results of the project open up new directions for the production of spices with various sources of abundant raw materials in our country.

References

1. Tung DX, Thuy NT, Thang TC. Current Status and Prospects for the Pig sector in Viet Nam: A desk study, Research Report. 2005.
2. Akinyosoye FA, Olowoyo OO, Adetuyi FC. Microorganisms associated with some cassava (Manihot esculenta crantz) products. J Res Rev Sci 2001; 2: 10-14.
3. Santos NN. Hydrolysis of pork muscle sarcoplasmic
proteins by Debaryomyces hansenii. *International Journal of Food Microbiology* 2001; 68: 199-206.
4. Tavano OL. Protein hydrolysis using proteases: An important tool for food biotechnology. *Journal of Molecular Catalysis B: Enzymatic* 2013; 90: 1-11.
5. Maldonado J, Gil A, Narbona E et al. Special formulas in infant nutrition: a review. 1998; 53: S23-S32.
6. Mahmoud MI, Malone WT, Cordle CT. Enzymatic hydrolysis of casein: Effect of degree of hydrolysis on antigenicity and physical properties. *J Food Sci* 1992; 57: 1223-1229.
7. Shahidi F. Sea Food Processing By-Products. In: Seafoods: Chemistry. Processing Technology and Quality. Shahidi F, Botta JR(Eds.), Blackie Academic & Professional, London, UK. 1994; 321-334.
8. Selig RM, Cozza JC, Selig DS. Boosting Calories in Children. Philadelphia, PA 19128. 2013; 215-483-8558.
9. Yanar Y, Celik M. Seasonal amino acid profiles and mineral contents of green tiger shrimp (*Peneaus semisulcatus* De Haan, 1844) and speckled shrimp (*Metapenaeus Monoceros* Fabricus, 1789) from the Eastern Mediterranean. *Food Chem* 2006; 94: 33-36.
10. Entsua-Mensah M, De Graft-Johnson KA, Atikpo MO, Abbey LD (2002) *The lobster, shrimp and prawn industry in Ghana: species, ecology, fishing and landing sites, handling and export*. Food Research Institute/AgSSIP, guineenis.
11. Lachance PA, Fisher MC. Reinvention of the food guide pyramid to promote health. *Advances in Food and Nutrition Research* 2003; 49: 1-39.
12. Biesalski HK. Meat as a component of a healthy diet - are there any risks or benefits if meat is avoided in the diet? *Meat Science* 2005; 70: 509-524.
13. Chan W. Macronutrients in meat. In Jensen WK, Devine C, Dikeman M (Eds.). *Encyclopedia of meat sciences* 2004: 614-618.
14. Mulvihill B. Micronutrients in meat. In Jensen WK, Devine C, Dikeman M(Eds.), Encyclopedia of meat sciences (pp. 618-623). Oxford: Elsevier.
15. Arihara K. Strategies for designing novel functional meat products. *Science direct, Meat Science* 2006; 74: 219-229.
16. Fonseca RAS, Silva CM, Fernandes GR et al. Enzymatic hydrolysis of cobia (*Rachycentron canadum*) meat and wastes using different microbial enzymes. *International Food Research Journal* 2016; 23(1): 152-160.
17. Souissi N, Bougatet A, Triki-Ellouz Y et al. Biochemical and Functional Properties of Sardinella (Sardinella aurita) By-Product Hydrolysates. *Food Biotechnol* 2007; 45: 187-194.
18. Ahmad NB. Beef hydrolysis by zyactinase™ Enzymes. Massey University, Auckland, New Zealand. 2016.
19. Melissa Ferreira SBRDGGID. Marina Silveira MDNTILHA, Vitória Ribeiro Garcia de FIGUEIREDD, Sandra Regina GEDGETTI, Louise Emy KURDZAWA. Influence of the degree of hydrolysis and type of enzyme on antioxidant activity of okara protein hydrolysates. *Food Sci Technol, Campinas* 2016; 36(2): 375-381.
20. Jamdar SN, Rajalakshmi V, Pednekar MD et al. Influence of degree of hydrolysis on functional properties, antioxidant activity and ACE inhibitory activity of peanut. 2010.
21. Selan on D, Saetae D, Sun tornsuk W. Utilization of Jatropha curcas seed cake as a plant growth stimulant. *Biocatalysis and Agricultural Biotechnology* 2014; 3(4): 114-120. http://dx.doi.org/10.1016/j.bcab.2014.08.001.
22. Li S, Yang X, Zhang Y et al. Effects of ultrasound and ultrasound assisted alkaline pretreatments on the enzymolysis and structural characteristics of rice protein. *Ultrasonics Sonochemistry* 2016; 31: 20-28. http://dx.doi.org/10.1016/j.ultsonch.2015.11.019. PMid:26964920.
23. Jin D, Liu X, Zheng X et al. Preparation of antioxidative corn protein hydrolysates purification and evaluation of three novel corn antioxidant peptides. *Food Chemistry* 2016; 204: 427-436. http://dx.doi.org/10.1016/j.foodchem.2016.02.119. PMid:26988521.
24. Zhang G, Arason S, Arnason SV. Physical and sensory properties of heat pump dried shrimp (*Pandalus borealis*). *Trans Chin Soc Agric Eng* 2008; 24: 235-239.
25. Hu J, Lu W, Lv M et al. Extraction and purification of astaxanthin from shrimp shells and the effects of different treatments on its content. *Revista Brasileira de Farmacognosia* 2019; 29: 24-29.