Effect of Freeze Dried Protein Hydrolysate from Yellowstripe Scad (*Selaroides Leptolepis*) in Reducing Oil Uptake in Fried Seafood Product

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***Abstract*—Fried products are widely preferred by consumer for appealing crust formation, colour, texture and flavour, but the product is less healthy due to high oil uptake as well as high calorie level. Protein is used to study oil reduction due to the ability in film forming and thermal gelation properties. The objective of this paper is to determine the effect of freeze dried protein hydrolysate from Yellowstripe Scad (*Selaroides leptolepis*) in reducing oil uptake in fried seafood product. Protein hydrolysate was produced using potassium buffer, 2% Alcalase and 2h of hydrolysis time. Protein hydrolysate was incorporated at different percentage (0%, 5%, 10%, 15% and 20%) in the batter. Oil uptake was conducted using Soxhlet method, while moisture content was conducted using gravimetric method according to AOAC, 2000. Result depicts that the reduction of oil uptake in fried seafood showed an increasing trend (6% to 32%) as the amount of incorporated protein hydrolysate was increased. However, the acceptable amount of incorporated protein hyrolysate was 10%. The water retained in the sample in 5% and 10% of incorporation were 33.5% and 32.9% while 15% and 20% incorporation had 16% and -5.15%, respectively.**

***Index Terms*—frying, Alcalase enzyme, water retention, batter incorporation**

I. Introduction

Yellowstripe scad (Fig. 1) belongs to the small pelagic group which is categorised as low value fishes, is one of the plentiful marine source in Vietnam sea area [1]. In Malaysia, this species is considered underutilised, where these Yellowstripe scad has been used in fish burger, with acceptable favour, household cooking or used as animal feed [2]. This species is distinguished by its prominent lateral yellow band and smaller eye, differing from scads of *Selar* [1]. These pelagic fishes generally attain a maximum weight of less than 500 gram each. Yellowstripe scad is a small species fish with a maximum length of 22cm but more commonly less than 15cm [3]. This species has a typical scad body shape, with compressed elongate, oblong body with the dorsal and ventral profiles equally curved [4].

Figure 1. Yellowstripe scad with prominent lateral yellow band

In order to increase the value and utilization of low value proteinacious fish, processes such as protein hydrolysis via enzymatic hydrolysis is used to produce a more marketable and functional protein hydrolysate [5]. Fish protein hydrolysate produced by controlled enzymatic hydrolysis, is considered to be the best fish protein hydrolysate due to its nutritional properties of well balanced amino acids composition and these hydrolysate is highly digestible by consumers [6]. Hydrolysis affects hydrophobicity, polar groups and molecular weight (size) which directly influence the functional properties of hydrolysate as food ingredient [7]. Freeze drying process has two main principles low temperature and the vacuum atmosphere where samples are dried by removing water from the matrix at very low temperature via sublimation of frozen water to vapour in vacuum chamber [8]. This drying method is considered to be the most suitable method to inhibit denaturation of protein compared to other drying methods.

There are studies regarding protein on oil uptake reduction due to the ability in film forming and thermal gelation properties [9]. Hydrophilic and lipophilic side chains of protein has the ability to hydrate or hold water and posses emulsifying properties that is linked to oil uptake during frying process [10]. Cross linking of protein also reduce porosity in sample, subsequently reduce oil absorption up to 30%.

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Lipid such as fats and oils have crucial impact in functional and sensory properties of fried products through enhancing, carrying and releasing flavour, also to produce suitable texture and mouth-feel characteristics of product [11], [12]. Fat is a naturally palatable *par excellence* when hot frying fat replaces water loss, exerting tenderizing effect on the crust for flavour, crispness and pleasant taste [13].

Deep frying is an immersion frying where food is cooked by immersing it completely in hot edible oil to retain flavours and juices of product [9]. This method involves physical and chemical changes in food through starch gelatinization, protein denaturation, crust formation and water vaporization which affect mass and heat transfer of oil and water in product [14]. Deep frying causes water in product to vaporize, moving away from product through surrounding hot oil [15], [16]. Batters are often used in frying to improve quality of fried products by improving texture, flavour, weight and volume [17], [10]. Apparently, the function of batter as a coating is to reduce water loss during frying, subsequently reduce oil absorption, improve structural integrity of product, retard gas transport, also to carry ingredients and to prevent flavour absorption [18]

Deep frying causes water in product to vaporize, moving away from product through surrounding hot oil [15], [16]. Water removal causes increase of porosity in fried products. Higher amount of water in product leads to more water loss during frying, leaving more pores that facilitate oil absorption [19]. Batter coating is used to reduce moisture loss during frying, reducing oil absorption too [20]. Thus, this paper aimed to determine the effect of incorporation of freeze dried protein hydrolysate from yellowstripe scad (*Selaroides leptolepis*) in reducing oil uptake in fried seafood product.

II. Materials and Methods

A. Protein Extraction

Protein extraction in this study involve edible portion of Yellowstripe scad. The edible portion of fish was obtained by removing the head, viscera, tails and fins. The fish was grinded using a blender.

Fifty grams of fish meat was deactivated by immersing into water bath at 90 ℃ for 10 min. It was centrifuged at 3500 rpm for 20 min for oil separation. Then it was mixed with 100ml of potassium buffer and adjusted to pH 8 using 2.0M sodium hydroxide. 2.0% of enzyme concentration was added. The hydrolysis was conducted for 2h in a water bath. The solutions were centrifuged at 10000 rpm for 20 min and filtered. The liquid hydrolysate was then dried using freeze dryer.

B. Drying Methods

The samples were freeze dried using Labconco Freeze Dryer (USA) with Stoppering Tray. It was operated at -54 ℃ with vacuum condition of 0.250 mbar. The samples were pre-frozen prior to freeze drying at -80 ℃.

C. Sample Preparation for Frying

Carrier sample was squid and the batter was incorporated with protein hydrolysate at different percentage (0%, 5%, 10%, 15% and 20%). The sample was deep fried and drained for 30 min. Oil and moisture analysis were conducted to determine the percentage of oil uptake and water retention.

D. Oil Uptake

In crude fat determination using Soxhlet method [21], about 1g of sample was weighed and wrapped into a filter paper before placing it into a thimble.

Pre-dried extraction cup was weighed and filled with 40ml of petroleum ether. Then the extraction cup was placed into the extraction unit of the Soxhtec machine (Labtec ST310, Sweden). The system started with boiling, continued with rinsing, recovery and pre-drying steps. The extraction cup was removed and dried in oven at 103 ℃ for 2h. Then, the extraction was cooled in desiccators for 1 hour before weighing. The percentage of fat was determined by (1).

$$\text{Percentage of fat content (\%)} = \frac{\text{Weight of fat (g)}}{\text{Weight of smple (g)}} \times 100 \quad (1)$$

Percentage of oil uptake in coated sample relative to uncoated sample was calculated based on Lipid Content (LC) determination shown in (2):

$$\text{Percentage of oil uptake (\%)} = \frac{\text{LC (after incorporation)} - \text{LC (before incorporation)}}{\text{LC (before incorporation)}} \times 100 \quad (2)$$

E. Water Retention

Moisture content determination of samples was determined with gravimetric method, using drying oven as the main instrument [21]. Pre-dried crucible and lid was recorded before adding sample. Approximately, two grams of sample was weighed and placed into the crucible. Then, it was dried at 105±5 ℃ until the weight remained constant. The dried sample was cooled and weighed again. The difference in weight before and after was the percentage of dried weight (3 and 4).

$$\text{\% Dried sample} = \frac{\text{weight after drying}}{\text{weight before drying}} \times 100\% \quad (3)$$

$$\text{\% Moisture of sample} = 100\% - \text{\% of dried sample} \quad (4)$$

The percentage of water retention was calculated by using Water Content (WC) of sample before coating and after coating according to the formula shown in (5).

$$\text{\% water retention} = \frac{\text{WC (after incorporation)} - \text{WC (before incorporation)}}{\text{WC (before incorporation)}} X\, 100 \quad (5)$$

III. Results and Discussion

A. Protein Hydrolysate and Yield

Liquid fish protein hydrolysate produced from enzymatic hydrolysis was subjected to freeze drying at which the yield obtained was 13 to 16%. The best condition selected was 2h of hydrolysis with the highest water holding capacity of 37% (unpublished data). Water holding capacity has major influence as oil uptake is closely related to moisture retained in the sample. Water holding capacity, also influence the viscosity of batter, which directly influence batter pick up and coating on the sample.

Freeze dried powdered protein hydrolysate was incorporated into batter at different percentage (0%, 5%, 10%, 15% and 20%) to determine the effect of protein hydrolysate in reducing oil uptake.

B. Oil Uptake

The results obtained from oil uptake showed the percentage of oil absorbed as compared to the control which is 0% of protein hydrolysate incorporation. Fig. 2 illustrates the reducing trend of oil uptake as the percentage of incorporation increased. As the incorporation increased from 5% to 20%, oil reduction increased from 10% up to 32%.

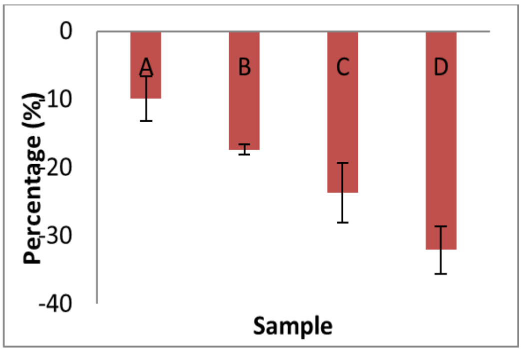

Figure 2. The effect of protein hydrolysate incorporation on oil uptake. (A=5%, B=10%, C=15%, D=20%)

However, the mechanism of oil uptake is closely related moisture loss in the sample. Mohamed *et al.* (1998) [20] reported that batter coating reduces moisture loss during frying, subsequently reduces oil absorption. Oil uptake showed a reducing trend as the amount of protein hydrolysate increased because of the functional properties of protein to reduce moisture loss during frying.

Protein has been focused in oil uptake reduction due to the ability in film forming and thermal gelation properties [9]. Cross linking of protein could also reduce the porosity in sample, which will reduce oil absorption (up to 30%). Creusot *et al.* (2011) [22] reported the effectiveness of oil reduction by protein as an ingredient of coating, where they deduced that oil reduction is closely related to gel formation of protein.

The results also show that effect of protein in reducing oil uptake had similarity with previous studies, where oil reduction up to 44% in fried potato slice [23] and egg albumin reduce oil uptake by 27% [24].

Oil reduction in C and D, 15% and 20% of protein hydrolysate incorporation, respectively, showed higher amount of oil reduction, but they are not preferable as the reduction might be due to lesser crust formed during frying. The batter formed from formulation C and D showed less viscous and lower batter pick up. Thus, lesser oil was retained in the crust. Although the amount of fish protein hydrolysate increased, different amount of other batter ingredients (such as flour, salt and water) give different adhesion power between coating suspension and sample, surface characteristics and frying conditions [25]. A uniform coating formation is essential to minimise mass transfer during frying [26].

C. Water Retention

Water retention is the percentage of water retained in the sample after deep frying process. Fig. 3 shows the percentage of water retained in the fried sample as incorporation of protein hydrolysate increased. As the percentage of incorporation increased to 10%, water retention increased to 38% but dropped to 16% then to -5%. This showed that crust successfully retained water up to 10% of incorporation and positive oil reduction. This is because water loss is closely related to oil absorption.

The increase in water retention and reduce in oil absorption due to incorporation of ingredient as coating film was also agreed by Freitas *et al.* (2009) [27]. Malak (2016) [28] reported that addition of soy protein isolate as coating films in deep frying also increase water retention. During deep frying, water is evaporated via steam, leaving empty pores on the surface of the crust which causes oil to be absorbed (water replacement) [29]. In addition, partial amount of moisture in the sample that is converted into steam during frying causes pressure gradient, where the steam escaped through capillaries and channels in the cellular structure. Oil is drawn into the sample as the internal vapour pressure decreases upon cooling [30].

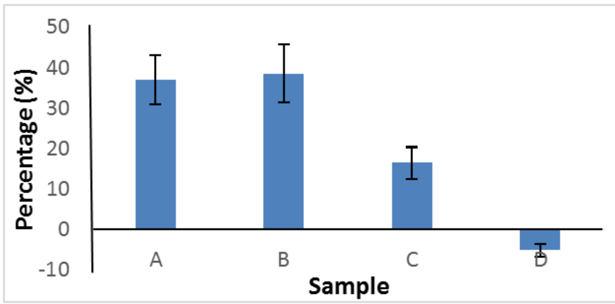

Figure 3. The effect of protein hydrolysate on water retention (A=5%, B=10%, C=15%, D=20%)

Thus, incorporation of protein hydrolysate up to 10% showed positive effect in reducing oil uptake but 15% and 20% gave negative effect. Lower water retention could be caused by lesser crust formed. This could be due to the properties of protein hydrolysate as the batter formed was less viscous as the percentage of incorporation increases. Crust formation is required to limit mass transfer during frying. Thus, lesser crust formation could not absorb oil and retain water in the sample.

IV. Conclusion

The study clearly shows that 10% of incorporation in batter, is the most preferable with reduction of oil uptake by 17% with highest water retention (38%). Although 15% and 20% incorporation have lower oil uptake, 24% and 32%, respectively, the water retention for both are low, showing that, lesser oil uptake was probably due to lesser amount of crust formed. Lesser crust formed could not absorb oil, but the crust could not retain water. Thus, 10% of fish protein hydrolysate incorporated in the batter is recommended to give a uniform batter that is essential to minimise mass transfer during frying.

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