



The Effect of Addition of Squid Ink (*Loligo* sp.) to Pizza Dough on Its Organoleptic Properties and Antioxidant Activity

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ABSTRACT

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The use of squid ink (*Loligo* sp.) in food can be an innovative opportunity for the culinary community in the manufacture of processed food products and their antioxidant content. This study aims to identify and analyze the organoleptic properties and antioxidant content of squid ink pizza dough (*Loligo* sp.). Organoleptic testing was carried out on pizza dough with 10, 20, and 30% addition of squid ink involving 30 semi-trained students as the panelists. Further, the samples with the best organoleptic test result were used as squid ink pizza dough samples tested for antioxidant activity analysis using the DPPH method. The antioxidant test involved three treatments, namely pizza dough with squid ink (*Loligo* sp.) without the baking process, pizza dough without squid ink (*Loligo* sp.) with the baking process, and pizza dough with squid ink (*Loligo* sp.) and baking process. The results showed no significant effect on organoleptic properties in hedonic testing, but there was a significant effect from the addition of 10, 20, and 30% squid ink (*Loligo* sp.) on color and taste in hedonic quality testing. Meanwhile, the antioxidant activity on pizza dough with the addition of squid ink (*Loligo* sp.) has an inactive antioxidant activity value ($IC_{50} > 200$ ppm). The results of this study can be used as a reference for a food product that incorporates squid ink (*Loligo* sp.).

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I. Introduction

The monthly increase in squid (*Loligo* sp.) production results in a rise in waste, as there are several unprocessed squid parts, including the ink (Direktorat Jenderal Pengelolaan Ruang Laut, 2020). In Indonesia, the utilization of squid ink waste (*Loligo* sp.) is still relatively low. However, the results of previous studies suggested that squid ink (*Loligo* sp.) can be beneficial for health and consists mainly of pigments that produce a dark color in ink (Derby, 2014), more commonly known as melanin compounds that correlate with antioxidant activity (Riyad et al., 2020). Antioxidants in squid ink are derived from polysaccharides, which are long chains (Luo, 2013) of combined sugar molecules found to protect against free radicals (Li et al., 2018). Besides, The results of phytochemical tests in a previous study indicated the presence of flavonoid and phenolic compounds in squid ink (*Loligo* sp.) as an antioxidant (Jeyasanta & Patterson, 2019).

The addition of squid ink (*Loligo* sp.) to pizza dough is one of the most recent culinary innovations that aim to increase the antioxidant content of food products, serving as an opportunity for the community in the culinary field to enhance the content of nutrients, particularly antioxidants.

Pizza is typically categorized as fast food because it requires less preparation time, and therefore the process of preparing pizza can be completed in a relatively short amount of time. Fast food typically has high calories, sodium, and fat so excessive consumption can cause degenerative diseases. However, according to research conducted by (Schmiele et al., 2011), pizza dough contains a low concentration of nutrients and bioactive compounds because it is made from refined wheat flour. In contrast, the addition of meat sauce and other toppings affects the pizza's nutritional content, as evidenced by the findings of Gallus et al. (2006), that pizza has a relatively high

nutritional content, as it contains a source of protein, complex carbohydrates, vitamins, and minerals, as well as a sufficient calorie density, making it a food with a good source of energy.

II. Method

This experimental study employed a completely randomized design (CRD) with three levels of squid ink and pizza dough formulation, as well as one treatment. The research design included testing the organoleptic properties of pizza dough with 10, 20, and 30% squid ink by involving 30 semi-trained panelists from nutrition students at Universitas Singaperbangsa, Karawang (UNSIKA). The organoleptic test aimed to identify and analyze the pizza dough's organoleptic properties, such as flavor, texture, aroma, and color, along with the panelist preferences. Further, following the organoleptic results, the most preferred pizza dough with squid ink underwent an antioxidant test.

Three pizza dough formulations were tested for antioxidant activity in the laboratory. Those formulations consisted of the baked pizza dough, squid ink pizza dough (*Loligo* sp.) that was not baked, and baked squid ink pizza dough (*Loligo* sp.). This was examined to determine whether the baking process and the addition of squid ink affected the antioxidant activity. This test was completed at the Food Technology Laboratory of Pasundan University (UNPAS).

III. Results and Discussion

A. Organoleptic Properties

In this study, the organoleptic properties of squid ink pizza dough (*Loligo* sp.), covering color, aroma, flavor, and texture, were evaluated subjectively by using the capacity or sensitivity of the human sensory system (Negara et al., 2016). Three types of organoleptic properties of the squid ink pizza dough were carried out in this study, including a sample selection test that assessed the organoleptic quality (hedonic quality test) and an acceptance test to determine the subject's preference for the squid ink pizza dough (*Loligo* sp.) (Tarwendah et al., 2017).

The results of hedonic testing indicated that the addition of squid ink (10, 20, and 30%) had no significant effect on the color, aroma, flavor, and texture of the squid ink pizza dough (*Loligo* sp.), nor on the panelists' preference for the sample. This is because the only difference between pizza formulations in the study was the percentage of squid ink (*Loligo* sp.) addition, while the other ingredients used to make pizza dough were identical. The subsequent hedonic test results are presented in Table 1.

Table 1. Hedonic Test of Squid Ink Pizza dough (*Loligo* sp.)

| Parameter | Mean Score of Hedonic Test | | | | P-Value |
|-----------|----------------------------|--------------------|--------------------|--|---------|
| | F1 (10%) | F2 (20%) | F3 (30%) | | |
| Color | 6.63 | 6.60 | 6.50 | | 0.908 |
| | 1.273 ^a | 1.221 ^a | 1.408 ^a | | |
| Aroma | 5.73 | 6.10 | 5.43 | | 0.298 |
| | 1.741 ^a | 1.423 ^a | 1.924 ^a | | |
| Flavor | 6.30 | 6.33 | 6.07 | | 0.966 |
| | 1.643 ^a | 1.493 ^a | 2.116 ^a | | |
| Texture | 6.20 | 6.43 | 6.23 | | 0.740 |
| | 1.562 ^a | 1.851 ^a | 1.851 ^a | | |

^aRemark: nine assessment criteria (1= strongly dislike; 2= immensely dislike; 3= dislike; 4=moderately dislike; 5=between like and dislike; 6=a little like; 7=like; 8=very much like and 9=strongly like); and the significance value of P>0.05 indicates that H0 is accepted, and Ha is rejected

In contrast to the hedonic quality test on squid ink pizza dough (*Loligo* sp.), there was a significant effect of squid ink addition (10, 20, and 30%) on pizza's color and taste. This is because squid ink has a brilliantly dark black hue. Therefore, the higher concentration of squid ink (*Loligo* sp.) added to pizza dough produces the darker color of the final processed food. The melanin compound in squid ink (*Loligo* sp.) is responsible for its dark black hue (Derby, 2014). Besides, a higher concentration of squid ink on pizza dough can result in a saltier to savory flavor. This finding is caused by squid ink's umami properties derived from its high glutamate content and amino acid,

producing a saltier taste (Derby, 2014). The effect of increasing squid ink concentration on pizza's color and flavor can affect the organoleptic quality, even if its aroma and texture are not significantly affected. The addition of squid ink at a low concentration produces a fishy smell, so increasing the squid ink concentration will increase the produced aroma. With the same amount of wheat flour (90 g per treatment sample), the pizza's odor and organoleptic quality were not significantly affected. Table 2 displays the results of the hedonic quality test.

Table 2. Average Hedonic Quality Test on Squid Ink Pizza Dough (*Loligo* sp.)

| Parameter | Quality Hedonic Test Mean | | | | P-Value |
|-----------|----------------------------|------------------------------|------------------------------|---|---------|
| | F1 | F2 | F3 | | |
| Color | 3.07 1.048 ^a | ± 2.43 0.898 ^b | ± 2.37 0.999 ^b | ± | 0.025 |
| Aroma | 4.77 1.357 ^a | ± 4.40 ± 1.380 ^a | 3.93 ± 1.780 ^a | ± | 0.068 |
| Flavor | 4.87 1.106 ^a | ± 5.00 ± 1.145 ^a | 3.67 1.184 ^b | ± | 0.000 |
| Texture | 4.13 1.279 ^a | ± 4.17 ± 1.487 ^a | 4.47 ± 1.795 ^a | ± | 0.839 |

^bRemark: assessment criteria (color, 2= very black; 3= black); (aroma, 3= having strong squid ink fragrant; 4= slightly having squid ink fragrant); (flavor, 3= salty; 4= slightly salty; 5= between salty and flavorless); (texture, 4= slightly soft)

a.b = equivalent letter notation. There is no statistically significant difference at the Mann-Whitney test level of 5%. The significance level is $P > 0.05$ (H_0 is accepted while H_a is rejected), $P < 0.05$ (H_a is rejected) (H_0 is rejected and H_a is accepted).

In testing the classification of samples, the pizza dough containing 20% of squid ink attained the highest level of preference for squid ink (*Loligo* sp.) pizza dough due to its very black color, slightly squid ink aroma, slightly salty to bland taste, and slightly soft texture. Table 3 presents the results of the ranking tests of the squid ink pizza dough (*Loligo* sp.).

Table 3. Average Ranking Test on Squid Ink Pizza dough (*Loligo* sp.)

| Parameter | Treatment | | |
|---------------|-----------|----------|----------|
| | F1 (10%) | F2 (20%) | F3 (30%) |
| Color | 2.07 | 1.80 | 2.13 |
| Aroma | 2.03 | 1.87 | 2.10 |
| Flavor | 2.00 | 1.80 | 2.20 |
| Texture | 1.97 | 1.77 | 2.27 |
| Overall score | 1.97 | 1.80 | 2.23 |

^cRemark: Average ranking test (1= strongly preferable; 2=preferable; 3=less preferable)

Pizza dough with a 20% addition of squid ink (*Loligo* sp.) attained the highest overall average organoleptic value. Further, laboratory tests were then carried out to analyze the value of antioxidant activity using the DPPH method on the three pizza dough samples.

B. Antioxidant Activity

The antioxidant test results on the squid-ink pizza dough (*Loligo* sp.) indicated antioxidant activity with an inactive intensity assumed to be caused by several reasons, including the addition of squid (*Loligo* sp.) The selection of squid ink from the neighborhood fish market was motivated by the low concentration of bioactive components that support antioxidant activity and the manufacturing procedure for squid ink pizza dough (*Loligo* sp.), affecting the sample's high IC_{50} value. The results of the antioxidant activity test are shown in Table 4.

Table 4. Antioxidant Activity Testing on Pizza dough

| Code | Sample | Testing Parameter | Testing Results (ppm) |
|------|--------------------------------|--|-----------------------|
| A | Baked Pizza Dough | | 5520,37 |
| B | Baked Squid Ink Pizza Dough | Antioxidant activity test using DPPH method with a parameter value of IC ₅₀ | 3639,28 |
| C | No Baked Squid Ink Pizza Dough | | 2899,54 |

^dRemark: Intensity of antioxidant activity (very active <50 ppm; active 50-100 ppm; moderate 101-250 ppm; weak 250-500 ppm; inactive >500 ppm)

From the results, it may be inferred that squid ink (*Loligo* sp.) addition to pizza dough can increase antioxidant activity, but the heating procedure used also influenced the antioxidant activity's value. In general, pizza dough can be baked at a maximum temperature of 200°C for a maximum of 15 minutes. However, the use of a high temperature and long baking time on pizza dough with the addition of squid ink (*Loligo* sp.) can result in low organoleptic quality because of the Maillard reaction (caramelization) on pizza dough and the loss of antioxidant activity (Muchtadi & Sugiyono, 2013). Therefore, in this study, Pizza dough with the addition of squid ink that was prepared using the baking process and without the baking process presented antioxidant activity value with an inactive intensity. Besides, The squid ink addition caused a significant difference in antioxidant activity. As a result, the decrease in temperature and duration of time used to avoid loss of antioxidant activity value did not produce good results. This is in accordance with research conducted by e(Lin et al., 2020) which states that high storage temperatures reduce antioxidant activity faster than low temperatures

This suggests that the addition of squid ink (*Loligo* sp.) to pizza dough can boost antioxidant activity but that the baking process can affect the antioxidant activity's usefulness. In general, pizza dough can be baked for a maximum of 15 minutes at a temperature of 200°C. However, the addition of squid ink (*Loligo* sp.) and baking procedure at a high temperature for an extended period of time might lead to the inferior organoleptic quality of the pizza (*ISBN 9781435481374 - Le Cordon Bleu Cuisine Foundations Direct Textbook*, n.d.) because of the Maillard reaction (Tamanna & Mahmood, 2015) (caramelization) on the pizza and the reduction in antioxidant activity value (Muchtadi & Sugiyono, 2013). Since there is a significant difference in antioxidant activity between the two formulations with the squid ink addition, this study concluded that pizza dough added with squid ink that was prepared through the baking process and without the baking process had an antioxidant activity with inactive intensity. In order to prevent the loss of antioxidant activity, the temperature was decreased, but due to the length of baking time, this process still did not produce positive outcomes.

IV. Conclusion

The hedonic test results on the squid ink pizza dough (*Loligo* sp.), which covered a test in the level of panelists' acceptance or preference, showed that the 10, 20, and 30% squid ink addition carried no significant effect on the pizza's organoleptic quality. However, the squid ink addition had a significant impact on the pizza's color and taste. Therefore, the ranking test positioned the sample with the addition of 20% squid ink as the most preferred percentage and was tested for its antioxidant activity. In the analysis of the antioxidant activity of the three pizza dough formulations, the antioxidant activity with an inactive intensity was observed in the formulation. This finding was presumed to be caused by several factors that caused the loss of antioxidant activity. The squid (*Loligo* sp.) used in this study was obtained directly from the local fish market so that they had a low level of bioactive compounds contributing to antioxidant activity and the processing process in making squid ink pizza dough (*Loligo* sp.) which affects the high IC₅₀ value.

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