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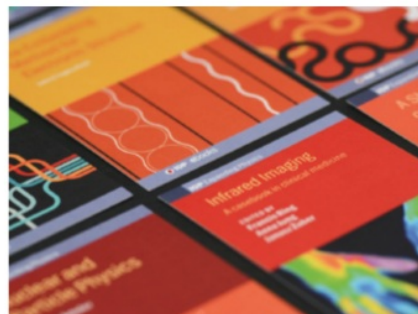
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Nutritive and antioxidative properties of some selected agro-industrial by-products fermented with the fungus *Chrysonillia crassa* as alternative feedstuffs for poultry

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Abstract. Agricultural by-products used as poultry feed in many developing countries. However, their use for poultry feed is limited due to high fiber and lack of nutritional substances (e.g., crude protein). The objective of this study was to evaluate the antioxidant activity and chemical composition of the agriculture by-products after they are fermented with fungi *Chrysonillia crassa*. Materials were three kinds by-product e.i rice bran, tofu, and palm kernel. Five hundred grams of three kinds of sterilized, dried byproducts placed in a plastic bag then added with 250 ml of aquades (for tofu and rice bran byproducts) and 500 ml to palm kernel byproduct. Five dishes of culture isolates of *Chrysonillia crassa* with two days old were put and mixed thoroughly in the sterilized by-products and incubated for two days then were dried. All treatments were three replicated. The antioxidant activity and chemical composition were in vitro analyzed. Results showed that the antioxidant activity of tofu improved that is before and after fermentation was 12330.08 ± 0.00 and 1466.96 ± 10.70 , respectively. The fermentation also improved protein content. The increased of the crude protein for palm kernel, rice bran and tofu before and after fermentation were 11.81 ± 0.00 to 12.22 ± 0.05 ; 9.35 ± 0.00 to 10.57 ± 0.09 ; 168 ± 0.00 to 21.48 ± 0.27 , respectively. On the contrary fungal fermentation decreased the crude fibre of palm kernel and rice bran but not for tofu. The decreasing of crude fibre before and after fermentation of palm kernel and rice bran were 31.54 ± 0.00 to 27.04 ± 2.60 and 29.67 ± 0.00 to 27.01 ± 0.18 , respectively. The fat content of the by-product increased only for rice bran that is 7.45 ± 0.00 to 11.39 ± 1.86 , for tofu decreased, and palm kernel was not affected. In conclusion, it was fungal fermentation. Palm kernel, rice bran, and tofu using *Chrysonillia crassa* increased in fat, crude protein, and decreased crude fibre, however for antioxidant activity was variation in results.

1. Introduction

The use of alternative feedstuffs in poultry rations is needed in response to the increased price of the conventional feed ingredients [22]. Some selected agro-industrial by-products have been explored for their potential as the alternative feedstuffs for poultry, including palm kernel, rice bran, tofu waste, etc. Apart from their cost-benefit, there are general problems associated with the use of agro-industrial by-products, i.e., the high fibre and low protein contents that may limit the inclusion levels of such by-products in poultry rations [23]. Fungal fermentation has been practiced to improve the nutritive characteristics as well as the antioxidative properties [27] of feed ingredients. Fermentation may degrade the complex carbohydrates into simple sugars and also increase the biosynthesis of protein (microbial protein) [27]. In this present study, some selected agro-industrial by-products were fermented using the fungus *Chrysonillia crassa*. The latter filamentous fungus was employed considering its fibrinolytic and protein-enhancing activities [27]. The fungus *C. crassa* has also been reported to improve the antioxidant

activity in rice bran as previously be reported by Sugiharto et al. [27]. The current work aimed to evaluate the nutritive and antioxidative properties of some selected agro-industrial by-products after fermentation with the fungus *Chrysonillia crassa*.

2. Materials and methods

Five hundred grams of three kinds of sterilized, dried byproducts (tofu, rice bran and palm kernel) placed in a plastic bag then added with 250 mL of aquades (for tofu and rice bran byproducts) and 500 ml to palm kernel byproduct. Five dishes of culture isolates of *Chrysonillia crassa* with two days old were put and mixed thoroughly in the sterilized by-products. All treatments were three replicated and incubated for two days then were dried. Test of antioxidant activity used DPPH+radical scavenging assay. [8], Total phenolic and tannins contents in the supernatant from the three byproducts fermented were evaluated according to [14] based on the Folin-Ciocalteu method with few modifications. The proximate compositions of the samples of byproduct were determined using standard analytical methods. All measurements were presented in percentages. The water content of all samples of byproducts fermented was determined using the AOAC method [2]. Fat was determined using the soxhlet fat extraction method [15]. Determined of crude fibre content was used Wende's method [7]. Determined of protein of the sample was using the Kjeldahl method [17]. All data collected were analyzed by analysis of variance (ANOVA) followed by Duncan's multiple range test to assess the difference between mean values.

3. Result

3.1 Antioxidant activity phenolic and tannins content of the three by-products before and after treatments

Table.1 showed that antioxidant activity of the three by-products appeared highly significant differed ($p < 0.01$) between before and after treatments. Antioxidant activity for tofu was improved after treatments, on the contrary for rice bran was decrease and for palm kernel was not any affected by the treatments. For phenolic content of the three by-products showed highly significant differed ($p < 0.01$) between before and after treatments. The phenolic content for palm kernel was improved after treatments, conversely for rice bran and tofu were decrease. Tannins content of three by-products showed highly significant differed ($p < 0.01$) between before and after treatments. The tannins content for palm kernel and tofu were improved after treatments, whereas tannins in rice bran was not any effect by the treatments.

Table 1. Antioxidant activity, phenolic and tannins contents of the three by-products before and after treatments

by-product	Antioxidant activity		Phenolic Content		Tannins Content	
	Before treatment	After treatment	Before treatment	After treatment	Before treatment	After treatment
palm kernel	371.50±10.45	434.16±26.86	0.06±0.00 ^b	0.34±0.01 ^a	0.25±0.00 ^b	0.42±0.00 ^a
rice bran	955.54±0.00 ^a	678.33±24.59 ^b	0.14±0.00 ^a	0.09±0.00 ^b	0.14±0.00	0.16±0.01
tofu	12330.08±0.00 ^b	1466.96±10.70 ^a	0.41±0.00 ^a	0.39±0.00 ^b	0.09±0.00 ^b	0.34±0.00 ^a

^{a,b} Values with different letters within the same rows were significantly different ($p < 0.01$).

3.2 Antioxidant activity, phenolic and tannins contents of the three by-products before and after treatments

Chemical contents of the three by-products are presented in Table 2. The chemical contents were significantly differed ($p < 0.01$ and < 0.05) between before and after treatments. Results showed that crude fiber of the three by-products decreased after treatment except for tofu, otherwise crude protein of all by-products improved after treatment.

For the moisture and ash content of all by-products were not different between before and after treatments. For the fat content showed variety in results, for rice bran was improved, but tofu was decrease and for palm kernel was not affected by the fermentation.

Table 2. Chemical contents of three by-products before and after treatments

Byproduct	Treatment	Moisture %	Ash %	Fat %	Crude Fibre %	Crude Protein %
Palm kernel	Before	8.37±0.00	4.69±0.00	8.30±0.00	31.54±0.00 ^a	11.81±0.00 ^B
	After	9.48±0.40	4.55±0.40	6.75±1.01	27.04±2.60 ^b	12.22±0.05 ^A
Rice bran	Before	11.45±0.00	10.11±0.00	7.45±0.00 ^b	29.67±0.00 ^A	9.35±0.00 ^b
	After	8.79±0.24	12.00±1.22	11.39±1.86 ^a	27.01±0.18 ^B	10.57±0.09 ^a
Tofu	Before	10.80±0.00	4.16±0.00	5.35±0.00 ^a	13.83±0.00	19.68±0.00 ^B
	After	9.05±0.06	4.55±0.26	4.26±0.62 ^b	13.40±0.41	21.48±0.27 ^A

^{A,B,a,b} Values with different letters within the same column in each byproduct were differ ($p < 0.01$ and $p < 0.05$).

4. Discussion

The present study got antioxidant activity of by-products improved by fungal fermentation. As showed in Table. 1 that antioxidant activity of tofu improved before and after fermentation those was 12330.08±8.00 and 1466.96±10.70 respectively. This finding is same as found by [19] and [6]. [19] found that solid state fermentation of rice bran with the *R. oryzae* increased free phenolic content by more than 100%. [6] suggested that fermentation can improve antioxidant activity and it may be due to the microbial hydrolysis or breakdown of plant cell walls into the various antioxidant compounds i.e phenolic compounds and flavonoids. The increasing of the antioxidant activity of tofu was contributed by increasing of tannins content and might be the tannin contents is condensed tannin. This opinion is supported by [5] that in fact fermentation can improve the mass or mycelia of fungi and the mycelia are rich source of antioxidant compounds such as phenols like condensed tannins and flavonoids. In the contrary antioxidant activity of rice bran decreased. This decreasing is reasonable because the antioxidant compounds i.e phenolic content decreased while tannin was not effected by fermentation. For antioxidant activity of palm kernel was remain constant even though the phenolic and tannin contents increased. Thus might be the flavonoid content in the phenolic compound was low and the tannins content was not condensed tannin but hydrolysalisable tannins. As mention by [5] that tannin which is contained in antioxidant compounds is belong to condensed tannins

The study showed that fungal fermentation could improve protein content of all the three by-products. The increased of the crude protein for palm kernel, rice bran and tofu before and after fermentation were 11.81±0.00 to 12.22±0.05; 9.35±0.00 to 10.57±0.09; 168±0.00 to 21.48±0.18 respectively. The result is contributed by findings of [1] and [27]. [1] found that fermentation by the fungus *Trichoderma harzianum* could successfully produce fungal biomass protein using rice polishings. Same finding also got by [27] that fermentation on the used rice using *Chrysonillia crassa* or *Monascus purpureus* increased in content of amino acids and crude protein. In accordance with [11] showing the increased protein content in cassava flour following fermentation with the use of yeast. Likewise, [25] also showed that the protein contents of sterile fermented cassava pulp could be enriched using *Aspergillus niger*, after 8 days incubation. According to [16] that increasing protein production by filamentous fungi related to exploiting the extraordinary extracellular enzyme synthesis and secretion machinery of industrial strains to produce single recombinant protein products. [10] explained that the increased protein in the fermented products may be due to the increase in the extracellular protein produced by the fungi growing on substrates. [3] further suggested that the increased protein in the fermented product may be due to the potency of the fungi in producing an enzyme capable of degrading starch and polysaccharides into monosaccharides. The latter compound may easily be processed to protein by the fungi. [12] explained that the increase in protein was thought to be due to the addition of proteins donated by microbial cells due to its growth, which produced a single cell protein product (SCP)

or cell biomass containing about 40-65% protein. The study showed that fungal fermentation could decrease in the crude fibre of the two by-products e.g palm kernel and rice bran but not for tofu.

The decreasing of crude fibre before and after fermentation of palm kernek and rice bran were 31.54 ± 0.00 to 27.04 ± 2.60 and 29.67 ± 0.00 to 27.01 ± 0.73 , respectively. The finding is contributed by [20] and [12]. [20] found that the combination between *Trichoderma harzianum* fermentation for 3 days and followed by *Saccharomyces cerevisiae* for 7 days fermentation could decreased the crude fibre content of duckweed. According to [12] that the decreasing of crude fiber content was the effect of the high cellulose activity which is done by more microbes and this cellulose enzyme will degrade cellulose to glucose so that at the end of fermentation the crude fiber content decreases. The study resulted that the fermentation increased of fat content of the by-product of rice bran that is 7.45 ± 0.00 to 11.39 ± 1.86 . This result is same as got by [18]. [18] found that a newly isolated oleaginous fungus, *Mucor circinelloides* Q531, was able to convert mulberry branches into lipids. The highest yield and the maximum lipid content produced by the fungal cells were 42.43 ± 4.01 mg per gram dry substrate (gds) and $28.8 \pm 2.85\%$, respectively. [18] suggested that the increasing of the fat content was affected by the enzyme activity of the isolated fungus was capable of converting the cellulose and hemicelluloses to available sugar monomers which are beneficial for the production of lipids.

5. Conclusion

Fungal fermentation of the three kinds of by-products i.e. palm kernel, rice bran and tofu using *Chrysonillia crassa* could increase in fat, crude protein also antioxidant activity and can decrease in crude fibre.

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