

## **The Use of Nano-Encapsulation of *Morinda citrifolia* Fruit Extract in Drinking Water as Phytobiotic Based Feed Additive in Laying Hens**

**Zainal Choiri<sup>1</sup>, Nanung Danar Dono<sup>1</sup>, Chusnul Hanim<sup>1</sup>, Bambang Ariyadi<sup>1</sup>, Zuprizal<sup>1</sup>**

Faculty of Animal Science, Universitas Gadjah Mada, Yogyakarta, 55281

Corresponding email: zuprizal@ugm.ac.id

### **ABSTRACT**

This study was aimed to analyze the effect of nano-encapsulation of noni fruit extract (NFE) utilization in drinking water as phytobiotic based feed additive in layer phase of laying hens. The experiment was conducted in a completely randomized design consisting of 6 treatments and 5 replicates, with 12 chickens in each replicate pen. The treatments were: P0 = drinking water without feed additive (negative control); P1 = drinking water + Tetracycline 50 mg/hen (positive control); P2 = drinking water + 0.5% extract of noni fruit; P3 = drinking water + 0.5% nano-encapsulation of NFE; P4 = drinking water + 1% nano-encapsulation of NFE; P5 = drinking water + 1.5% nano-encapsulation of NFE. The observed parameter was productive performance of laying hens, which include: daily feed intake, egg production, feed conversion ratio, and egg mass. Data were statistically analyzed using Oneway ANOVA. Results showed that supplementation of nano-encapsulation of noni fruit extract in drinking water with the level of 1.5% did not affect productive performance of laying hens, such as: feed intake, hen day production, egg mass, and feed conversion ratio. Supplementation of antibiotic in low level in current study did not also give any significant effect. It can be concluded that supplementation of 1.5% nano-encapsulation of noni fruit extract via drinking water administration did not influential for maximizing productive performance of laying hens. The use of NFE gave the same result as antibiotic use.

**Keywords:** Laying hens, Nano-encapsulation, Noni fruit extract, Productive performance

### **INTRODUCTION**

The use of in-feed antibiotics was reported to leave residues in eggs with ranges up to several weeks after use (Goetting *et al.*, 2011). Antibiotic residues have negative effects, such as: increased population and colonization of antibiotic-resistant bacteria, increased drug side effects, and even death (Refdanita, 2004). One of alternatives that can replace the presence of antibiotic in the diets is phytobiotic from noni fruit. Noni fruit contains antioxidant substances, such as: vitamin C, kaempferol, quercetin (Deng *et al.*, 2007), narcissoside, nicotifloroside (Su *et al.* 2005; Sang *et al.*, 2001), and nicotifloroside (Sang *et al.*, 2001; Su *et al.*, 2005). Noni fruit was also reported to have antibacterial substances, such as: acubin, L-asperuloside, and alizarin (Wang *et al.*, 2002). However, these polyphenol compounds are highly sensitive to gastrointestinal conditions (alkaline pH, enzymes, and presence of other nutrients), and difficult to dissolve in drinking-water making it difficult to be absorbed, resulting in low bioavailability (Munin and Edwards-lévy, 2011). To maximize the potencies of noni fruit extract, one of the efforts that can be done is by protecting bio-active compound of the extract using nano-encapsulation method with chitosan and sodium tripolyphosphate (Bahreini *et al.*, 2014). The benefits of nano-encapsulation method are: particle sizes and

morphology of the nano-capsule can be manipulated, the efficacy of the active compounds can be protected, and facilitate faster particle diffusion in the mucus and reaching intestinal absorptive cells (Mohanraj and Chen, 2006; Bunglavan *et al.*, 2014). However, there is no available study to show the benefit of nano encapsulation of noni fruit extract in drinking water as an alternative natural antibacterial agent for laying hens.

## MATERIALS AND METHODS

**Experimental diets and parameters observed.** Noni fruit was collected from the Teaching Farm of Faculty of Pharmacy, Gadjah Mada University, Yogyakarta, Indonesia. Chitosan FG Powder (Chimultiguna, Indramayu, Indonesia), Sodium tripolyphosphate (STPP, Brataco, Yogyakarta, Indonesia), three hundred and sixty of twenty weeks ISA Brown laying hens, and basal feed.

**Table 1.** Basal diet composition and nutrients content

Items	Content
Diets composition (% , as fed)	
Corn	59.00
Rice bran	11.00
Meat bone meal	4.00
Dicalcium Phosphate	1.00
Soybean meal	18.00
Phytase	0.02
Sodium bicarbonate	0.30
DL-Methionine	0.10
L-Lysine HCl	0.05
Salt	0.25
Limestone	4.28
Premix vitamins and minerals <sup>a</sup>	2.00
Total	100.00

<sup>a</sup>Premix vitamins and minerals each kg contains: Calcium, 32.5%; Phosphorus, 1.0%; Iron, 6 g; Manganese, 4 g; Iodine, 0.075 g; Copper, 0.3 g; Zinc, 3.75 g; Vitamin B12, 0.5 mg; Vitamin D3, 50,000 IU.

The experiment was conducted in a completely randomized design consisting of 6 treatments and 5 replicates, with 12 chickens in each replicate pen. The treatments were: P0 = drinking water without feed additive (negative control); P1 = drinking water + Tetracycline 50 mg/hen (positive control); P2 = drinking water + 0.5% extract of noni fruit; P3 = drinking water + 0.5% nano-encapsulation of NFE; P4 = drinking water + 1% nano-encapsulation of NFE; P5 = drinking water + 1.5% nano-encapsulation of NFE. The observed parameter was productive performance, which included: daily feed intake, egg production, feed conversion ratio, and egg mass.

**Preparation of noni fruit extract.** A whole of 1.5 kg fresh noni fruits was blended and mixed thoroughly with 1900 ml aquades for 15 minutes. The mixture was then filtered using nylon fabric. The liquid extract was heated at 90 °C for 4 hours to reduce the water content to form a paste. The paste was mixed with 96% alcohol 575 ml with magnetic stirrer (C-MAG HS 7, IKA, Selangor, Malaysia) for 20 minutes. Sediment will be precipitated from the solution after 20 minutes and solution will be ready for nano-encapsulation.

**Nano-encapsulation of noni fruit extract.** Nano-encapsulation procedures were modified from Sundari *et al.* (2014). A number of 3.816 g Chitosan was dissolved in 610 ml acetate 2.5% using magnetic stirrer for 30 minutes. The chitosan solution was added with 382 ml of noni fruit extract and stirred for 20 minutes. In a separate container, 0.125 g STPP was dissolved in 16.7 ml aquades. The STPP solution was then added to the chitosan-extract solution of noni fruit and then homogenized using magnetic stirrer for 20 minutes. The formed solution was heated at 70 °C for 1 hour to evaporate the alcohol.

The data of feed consumption, hen day production, egg mass, and feed conversion ratio were compared between groups by a Completely Randomized Design in Oneway arrangement (Steel and Torrie, 1989). The statistical analysis was conducted with the Statistical Package for the Social Sciences (SPSS for Windows Version 16, SPSS GmbH, Munich, Germany).

## RESULTS AND DISCUSSION

**Feed intake.** Data in Table 2 showed that addition of nano-encapsulation of noni fruit extract via drinking water of laying hens did not give any effect on feed consumption. This might be attributed to the similarity level of energy content between the treatment diets. In poultry, energy content in the diet greatly affects the amount of feed consumption (Rose, 2005). The results of this study were similar to Widiyanto (2016) which reported that 3% noni fruit juice in drinking water did not affect feed consumption in hybrid ducks.

**Table 2.** Productive performance

Items	Treatments						Statistics	
	P0	P1	P2	P3	P4	P5	SEM	<i>P-value</i>
FI	111.60	112.37	109.30	111.06	110.58	110.72	0.762	0.143
HDP	80.76	85.14	75.31	83.48	84.24	83.33	2.468	0.677
EM	44.60	47.19	45.02	46.45	46.54	46.51	1.380	0.748
FCR	2.65	2.45	2.57	2.47	2.49	2.51	0.092	0.652

FI= feed intake; HDP = hen day production; EM = egg mass; FCR = feed conversion ratio.

**Hen day production.** Data in Table 2 showed that addition of nano-encapsulation of noni fruit extract in drinking water of laying hens had no effect on hen day production (HDP). The absence of this effect might be attributed to the health and ability of the chicks to adapt environmental condition of the poultry house. Average temperatures at the moment of research were 25.60 °C in the morning and 31,74°C in the afternoon, while the average of humidity was 67.17% in the morning and 36.71% in the afternoon. Although the temperature during the day tends to be high, but the humidity is low 36.71%. In this condition, the birds were more convenient and more optimal in the process of removing heat from the body during the tropical hot environment. As reported by Gerson *et al.* (2014), high environmental humidity inhibits the heat release process through evaporation and panting. In current study, although the antioxidant activity of nano-encapsulation of noni fruit extract reached 78.4% (Choiri *et al.*, 2016), supplementation of NFE with the level of 1.5% did not affect HDP. The results of this study differ from the findings of Sunder (2013) which stated that addition of 4% noni fruit juice in the feed increased egg production. The different result might be caused by the different methods of noni preparation.

**Egg mass.** Effect of treatment on egg mass variables was shown in Table 2. The data showed that the use of nano-encapsulation of noni fruit extract in drinking water of laying hens has no effect on egg mass. The absence of this effect is suspected because the chickens

used in this research have uniform weight, age, and nutrient feed. Factors that affect the egg mass include: lighting program, the use of unsaturated oil, body weight, age of sexual maturity, and environmental conditions (Joly, 2009). Result in this study was different with the results of Sunder *et al.* (2013) that showed that the addition of noni fruit juice in 4% feed increased egg weight. This difference may be due to differences in the composition of feed ingredients, noni preparation, and the breeds of the birds.

**Feed conversion treatment.** Effect of treatment on FCR variables was shown in Table 2. The data showed that the use of nano-encapsulation of noni fruit extract in drinking water of laying hens has no effect on FCR. This was perhaps caused by chicken used in this research didn't challenge with pathogenic bacteria or biosecurity program was good enough, so pathogenic bacteria didn't grow or develop in the chicken digestive tract. Godoy *et al.* (2012) stated that a good biosecurity program could prevent pathogenic bacteria infections. The absence of pathogenic bacteria made the main function of noni fruit (antibacterial) not appear.

## CONCLUSIONS

Supplementation of 1.5% nano-encapsulation of noni fruit extract via drinking water administration did not influential for maximizing productive performance of laying hens. The use of nfe given the same result as antibiotic use.

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