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Use of high protein dietary leaves substitute protein source in organic laying hens feed

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Abstract The results showed that comparing egg percentage, egg weight, egg shell thickness eggshell weight Yolk weight egg white height and hog unit of laying hens fed diet using plant leaf substitute protein source from organic soybean meal were not statistically differed ($p>0.05$). In terms of egg white and yolk quality of organic chicken eggs can be used high protein plant leaves instead of organic soybean meal protein. It was found that cassava leaves was more effective than acacia leaves and mulberry leaves based on egg white weight, and the color of the egg yolk was significantly higher than the leaves of other plants. It can be concluded that the use of cassava leaves as raw material to replace protein sources from soybean meal in organic laying hen diet. Because there was no difference in egg percentage, egg weight and eggshell characteristics, but the egg white height was higher. The color of the egg yolk was significantly higher than the raw material from other plant leaves, and the egg quality level was AA level. Cassava leaves are also readily available raw materials and it is cheaper than using organic soybean meal.

Keywords: Protein dietary leaves, Laying hens

Introduction

Organic livestock is produced using an organic farming system, avoiding using chemical drugs, hormones, genetically modified plants, and animals. In addition, animal welfare is taken care of, and stress reduction, including avoiding using by-products from animals as animal feed. Emphasizes local wisdom from experience combined with modern science that relies on nature to solve problems and bring about agricultural sustainability (Åkerfeldt *et al.,* 2021). Animal husbandry feeds must be certified organic produce, and the producer must

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prepare a feeding plan for the animals, and can be checked at any time (Organic agriculture certification Thailand, 2017).

Over 90% of the world's commercial poultry products are commercially raised chickens (European Commission, 2016). Due to the continuous demand for higher yields, most farmers use intensive farming methods raised in a limited area, with water control, feed control, and many farm factors to reduce costs and increase productivity. In addition, they are using chemicals such as various minerals and antibiotics, etc. This farming system does not consider animal welfare and food safety as it should (Mattioli *et al.,* 2021). However, nowadays, consumers are beginning to realize and pay more attention to organic chicken continuously. According to Gálvez *et al*. (2020) whose comparison of nutrients in a chicken-fed intensive commercial system, free-range and organic methods, it was found that organic chicken meat had a lower fat content and cholesterol than other farming systems. It contained more important nutrients than other systems, significantly, namely eicosapentanoic acid (EPA), docosapentanoic acid (DPA), and docosahexanoic acid (DHA).

However, raising in the organic system, the cost of animal feed is considered the highest cost of raising. Therefore, using local ingredients as feed sources can reduce costs, and high-protein leaves can be used as feed for chickens (Olukomaiya *et al.*, 2019). Since those forage leaves have an average protein of about 20%, they can be used as a protein source to replace soybean meal. It is very important that farmers can grow them for their use or can be harvested. Therefore, it is suitable for raising laying hens to reduce feed costs. Plant leaves that can be used as a source of protein are white popinac, coral tree, agasta, cassava, hemp, gourd, mulberry, etc. Using these plant leaves as feed for laying hens in an organic system will reduce feed costs. It is another way to support living organisms in the organic egg farm ecosystem.

Materials and methods

This study was conducted from October of 2021 to September 2022, at the Department of Animal Production and Technology, Faculty of Agro-Industrial Technology, Rajamangala University of Technology Tawan-ok Chanthaburi Campus in Thailand.

Experimental design and animal grouping

Experimental design

The experiment was performed by using Completely Randomized Design (CRD) with 120 commercial hybrid laying hens which were divided into 4 grops and 3 repeated trials.

Animal grouping

Animals in this study were separated into 4 groups. They were fed with different food supplies according to the experimental plan. The chicken's diets contained protein from organic soybean meal. Protein from soybean meal in the diets was replaced by using 10% of 3 types of high protein plants that were horse tamarind leaves, mulberry leaves and cassava leaves. Treatment 1 was reatment: protein from soybean meal. Treatment 2 was protein replacement from horse tamarind leaves. Treatment 3 was protein replacement from mulberry leaves. Treatment 4 was protein replacement from cassava leaves.

Animals

This study was performed by using 120 commercial hybrid laying hens. The hens were divided into 4 groups. In each group had 3 repeated trials with 10 hens.

Experimental method

This research was conducted using 15-week-old commercial hybrid laying hens. All birds had been habituated for 6 weeks. Diets fed before the experimental period composed with 18% of crude protein that provided energy 2,800 Kcal / Kilogram. The birds participated in the experiment when they were 21-week-old and dietary treatments were given as planned. The diets were produced under Thai Agricultural Standard (TAS 9000- volume 2, 2011) Each treatment equally contained 2,800 Kcal / Kilogram. The experimental period ran until the birds aged 32 weeks. Throughout the experiment, the hens were fed under ad libitum feeding and were vaccinated according to the suggestion of Department of Livestock Development.

The birds were housed following Thai Agricultural Standard TAS 9000 Part 2-2011: Organic Livestock. The hens were reared on floor within 2x5 square meter open housing. The floor was covered with 2 inches of organic chaff. All birds were released to 5x10 meter yard at 6am to 6 pm every day.

Data collection

The total number of eggs and egg weight were recorded every day in order to calculate percentage of egg production. Eggs from each group were randomly picked and stored in a fridge (5 degrees). Data of outer egg quality such as egg weight, eggshell weight and eggshell density were recorded. Also, data of inner egg quality such as yolk weigh, albumin weight, albumin height, yolk color were recorded. Haugh Unit was calculated as following:-

$$\text{Haugh Unit (HU)} = 100 \log (H - 1.7W^{0.37} + 7.57)$$

when H = albumin height (mm); W = egg weight (gram)

Statistical analysis

Data were checked and analyzed by using the analysis of variance with Proc. GLM (SAS, 1985). Each treatment was compared to find averages.

Results

Egg production, egg weight, eggshell density and eggshell weight of the hens fed with protein replacement dietary from high protein plants

The results showed that egg production from laying hens that consumed four experimental diets - soybean meal, horse tamarind leaves, mulberry leaves and cassava leaves- had no statistical difference ($p>0.05$). The averages of egg production were 73.97 %, 70.48 %, 72.44 % and 72.48 % respectively. In addition, egg weight was not statistical difference ($p>0.05$). The averages of egg weight were 174.50, 172.47, 176.70 and 169.93 respectively as shown in Table 1.

Table 1. Comparison of egg production, egg weight, eggshell thickness and eggshell weight from laying hens fed with protein replacement from 3 types of leaves

Factors	Organic soybean meal	Horse tamarind leaves	Mulberry leaves	Cassava leaves	P-value
Egg production (%)	73.97	70.48	72.44	72.48	0.676
Egg weight (gram)	174.50	172.47	176.70	169.93	0.841
Eggshell thickness (mm)	0.38	0.36	0.39	0.39	0.133
Eggshell weight (gram)	5.74	6.15	5.83	5.82	0.142

* **:Remark**= statistically different at a significant level of 0.05
** =statistically different at a significant level of 0.01

In term of eggshell quality, there were not statistical differences on thickness. The averages were 0.38, 0.36, 0.39 and 0.39 mm respectively; moreover, eggshell weight was not statistical significance ($p>0.05$) as the averaged data were 5.74, 6.15, 5.83 and 5.82 gram respectively (Table 1). According to the information above, it can be concluded that replacing source of protein in dietaries for laying hen from soybean meal to 3 types of high protein leaves did not show any effects on egg production, egg weight, eggshell thickness and eggshell weight.

Quatilty of albumin and egg yolks from laying hens fed with protein replacement dietary from high protein plants

The results found that albumin weight from the hens that fed with cassava leaves remained the same quality as the hens fed with organic soybean meal, feeding the hens with cassava leaves showed better result for albumin weight comparing with horse tamarind leaves and mulberry leaves (Table 2). It was statistically significance differed ($p<0.05$) as the averaged 154.28, 148.91, 151.84 and 153.26 in protein source from organic soybean meal, horse tamarind leaves, mulberry leaves and cassava leaves respectively. There were not significantly differed in egg yolks ($p>0.05$) and the averages were 15.25, 15.09, 14.43 and 14.8 respectively. The results for color of egg yolks showed that eggs from the hens fed with cassava leaves had the highest color intensity ($p<0.05$) with an average of 9.00. The averages of albumin height were 8.61, 8.77, 9.68 and 9.32 respectively. The averages of HU were 67.44, 69.31, 75.80 and 74.41 respectively.

Table 2. Comparison of albumin quality and egg yolk quality from laying hens fed with protein replacement from 3 types of leaves

Factor	Organic soybean meal	Horse tamarind leaves	Mulberry leaves	Cassava leaves	P-value
yolk weigh	15.25	15.09	14.43	14.8	0.462
albumin weigh	154.28^{A}	148.91^{C}	151.84^{B}	153.26^{A}	0.000**
albumin height	8.61	8.77	9.68	9.32	0.124
yolk color	6.17^{C}	7.33^{B}	8.67^{A}	9.00^{A}	0.000**
Haugh unit (%)	67.44	69.31	75.80	74.41	0.07

* **:Remark**= statistically different at a significant level of 0.05
** =statistically different at a significant level of 0.01

It could be assumed that cassava leaves could be effectively used to replace organic soybean meal because albumin height remained the same quality comparing with soybean meal and the result was still significantly higher than other leaves and color intensity of egg yolks. Cassava leaves were found easily in Thailand and and could reduce cost since they were cheaper than soybean meal.

Discussion

According to this study, protein replacement from organic soybean meal to 3 types of high protein leaves in chicken's dietary was investigated. The results of egg production, egg weight, eggshell thickness and eggshell weight from

laying hens fed with 3 types of leaves were not significant differed. The results was also described by Panja and Srichana (2013) who reported the comparison between commercial chicken food supplements and food supplements with mulberry leaves addition. It stated that there were not significantly differed among egg mass, egg weight, shell weight, shell thickness and shell strength. Zhang *et al.* (2009) who studied about increasing 5% of mulberry leaves in food supplements for Hyline brown chickens reported that egg weight and shell characteristics from the hens consuming both types of supplements (adding 5% of mulberry leaves and soybean meal) was not differed. There are many research reports on using horse tamarind leaves as source of protein in chicken food dietary. For example, Atawodi *et al*. (2008) who studied eggs in Nigeria reported that the averages of size and specific gravity of eggs between commercial food and food mixed with horse tamarind leaves showed no statistical difference. A report from Abou-Elezz *et al.* (2011) stated that egg laying rate, egg mass, egg weight, shell thickness and egg shape index of Rhode Island Red laying hens in Mexico had no differences. Moreover, Eichie *et al.* (2015) reported that using horse tamarind leaves as protein source in food dietary for broiler chicken stated that the result of Feed Conversion Ratio found no significant difference as compared to soybean meal as protein source.

The results showed that replacing soybean meal with cassava leaves in food supplements for laying hens showed the same results as written by Saparattananan *et al*. (2005) and Morgan and Choct (2016). They reported that cassava leaves could be effectively used as main source of protein instead of soybean meal or shredded fish. Beside the 3 types of leaves, there are many kinds of local leaves that are suitable for using as protein for chicken food dietary such as moringa leaves. According to the research of Abou-Elezz *et al.* (2011), they stated that the averages of egg weight and FCR remained the same quality as using soybean meal. Thongwittaya (2006) who studied about replacing soybean meal with sesame meal in food dietary for laying duck found that percentage of egg product and FCR showed no differences.

According to the information above, it can be concluded that using local plant leaves instead of soybean meal in food supplement for laying hens did not affect quality of eggs. Egg production, egg weight, shell thickness and shell weight remained the same characteristics.

In term of albumin and yolk quality of eggs from laying hens fed with 3 types of leaves instead of soybean meal, the results found that cassava leaves, comparing with horse tamarind and mulberry leaf showed better rates of albumin weight and yolk color. However, yolk weight, albumin height and HU showed no statistical differences. According to the research from Techakriengkrai *et al.* (2022) who studied about using cassava leaves in food dietary for chicken. They

found that cassava leaves gave higher color intensity of yolk comparing with other leaves. Khajarern and Khajarern (1992) who studied the effect of coloring agent from cassava leaves in food dietary for De Kalb chicken stated that the higher proportion of cassava leaves, the darker of yolks. Beside cassava leaves, there are several kinds of plants that help to increase color intensity of yolk such as annatto seeds (Kado *et al*., 2015; Yoosi and Tangkawanit, 2009). In addition, cassava leaves are rich of vitamins such as B_1, B_2 and C. They contain high protein that provide 1,600-1,800 Kcal/Kg (Ravindran, 1991; Khajarern and Khajarern, 2007). Cassava leaves contain antioxidants and glutathione that play an important role to slow down cell deterioration, promote detoxification and increase immune system in animals.

When comparing HU in this research, there was not statistically differed between soybean meal and 3 types of leaves (horse tamarind, mulberry and cassava). The averages were 67.44, 69.31, 75.80 and 74.41 respectively. However, when considering egg quality following the standard of the Ministry of Agriculture and Cooperatives in Thailand, Pornchaloempong and Rattanapanone (2022), there were 3 levels of egg quality that were AA (H.U > 72), A (H.U. = 60-71) and B (H.U. < 60). This study showed that egg quality from laying hens fed with cassava leaves and mulberry leaves stood in the highest level of the standard.

In conclusion, it could be said that cassava leaves are shown to be proper to replace soybean meal in food supplement for laying hens because egg quality such as egg production, weight and shell characteristics. Moreover, cassava leaves gave better results in albumin height and yolk color than other leaves. Besides, the quality of egg stand in the highest level (AA) and cassava leaves are found easily in Thailand. It helps to reuse wastes and reduce cost because organic soybean meal is quite difficult to find in Thailand.

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