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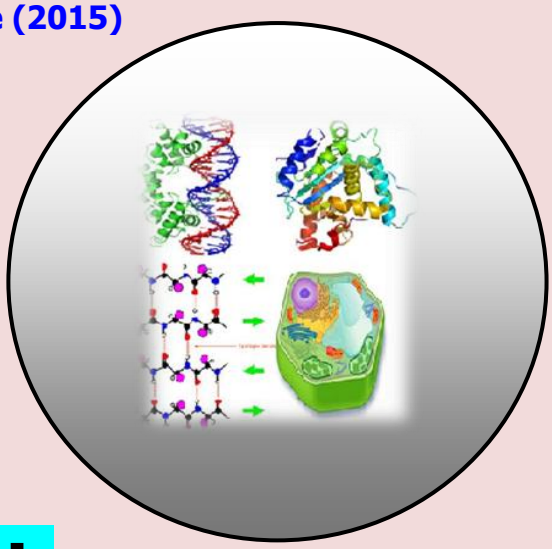

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RESEARCH PAPER

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Effect of Water Extract of Garlic Cloves (*Allium sativum*) on Egg Production and Yolk Cholesterol Levels in Egg Laying Hens

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ABSTRACT

The present study was conducted to determine the effects of Garlic cloves (Allium sativum) water extract on the egg production and yolk cholesterol level of 40-week old hens. One hundried and twenty 40-wk-old hens were colony caged in an environmentally controlled house to evaluate the effect of Garlic cloves water extract administration on laying hens. Garlic extract was prepared by macerating Garlic cloves in distilled water (1:1, w/w). Hens were randomly divided into four equal groups: one served as a control and was administered with drinking water only. Theother three groups were administered 2%, 4%, and 6%% water extract of Garlic cloves, respectively. The results showed that the addition of 0.20-0.60% garlic extract (Allium sativum) in drinking water had no significant effect (P>0,05) to feed and drink consumption, and egg shell thickness. However, significantly (P<0.05) increased egg weight, number of eggs, egg yolk, hen-day production, and feed efficiency. However, it decreased significantly (P<0.05) cholesterol levels in egg yolks. It can be concluded that Garlic cloves water extract increased egg production, but decreasing yolk cholesterol contents of egg laying hens.
Keywords: Phytochemicals, Allicin, Cholesterol and Egg Laying Hens.

INTRODUCTION

Garlic (*Allium sativum*) gained the confidence of many scientists and medications around the world for the prevention and treatment of many diseases and is widespread and consumed as herbs and herbs for thousands of years. Recent studies have validated many of the medicinal properties associated with garlic and its potential to reduce disease risk (Mahmoud *et al*., 2010), have been used for medicinal purposes-proven to have efficacy as antibiotic, antiviral, and antifungal, antibacterial, and potent inhibitors of food pathogens (Silvam, 2001; Lee *et al*., 2003). He *et al.* (2009) states that *Allyl trisulfide* is a major sulfur compound in garlic oil that has biological activity, such as antibiotics and anticancer found in animal and human studies. The function of garlic oil, such as antidiabetes and antiobesity is poorly understood.

The effect of phytochemical compounds to lower fat and cholesterol content in eggs is the use of garlic (*Allium sativum*). Garlic is believed by many scientists and medications around the world for the prevention and treatment of many diseases, and is widely dispersed and consumed as herbs and herbs from thousands of years ago. Recent studies have shown medicinal properties of garlic attributed and its potential to reduce the risk of disease. Garlic has been shown to have anti-thrombotic activity, blood lipids, blood pressure, and has a protective effect on the heart (Kasuga *et al*., 2001), antibacterial properties, and potent food pathogen inhibitors (Lee *et al*., 2003). The mechanism of garlic has been shown to be an effective antioxidant and its ability to stimulate the immune response (Lim *et al*., 2006).

Garlic shows efficacy as a broad-spectrum antibiotic against gram-positive and gram-negative bacteria. Other properties of Garlic are as a decrease in cholesterol levels in serum and eggs in chickens (Bidura *et al*., 2017), blood pressure, cancer prevention, immune system, and infection treatment, as well as antioxidants (Ao *et al*., 2010).

Previous research has shown that this function is mainly due to the bioactive components of garlic, including sulfur-containing compounds, such as *alliin*, *diallylsulphides, allicin, flavonoids, saponins*, and *fructans* (Amagase *et al*., 2001; Silvam, 2001). Therefore, different garlic preparations used in various studies may be one of the reasons for inconsistent results (Ao *et al*., 2010). Allicin is primarily responsible for the sharp smell of garlic and is produced from inert chemicals in raw garlic called alliina derivatives of cysteine by the action of enzymes, allinases in the presence of pyridoxal phosphates (Silvam, 2001). Garlic produces allicin to protect itself from bacteria and other diseases and antioxidants (Lee *et al.,* 2003). Studies on Garlic as an alternative to growth promoters in livestock production were carried out and their beneficial effects on growth, digestibility, and carcass properties have been reported (Nusairat, 2007; Bampids *et al*., 2005; Amooz and Dastar, 2009).

Garlic is studied in various forms of extract, such as water extract, ethanol, and dry powder. Garlic contains various organosulfur compounds, such as allicin, S-allylcysteine, diallyl disulfide, S-methylcysteine sulfoxide and S-allylcysteine (Lim *et al*., 2006). Studies on garlic as an alternative to the growth promoters of livestock production were performed, proving beneficial effects on growth and digestability (Bampids *et al*., 2005). Obochi *et al*. (2009) showed that garlic extract stimulates the secretion of gonadotropin and hormones produced by the ovaries, and can inhibit the proliferation of cancer cells. Al Aqil (2016) reported that adding 1.0% Garlic powder into laying hen diets had some positive effect on productive performance and egg quality parameters from 52 to 60 weeks of age.

Among the most potent and recognized active components in garlic are allicin. Allicin is unstable and difficult to absorb from the digestive tract. According to Bampids *et al.* (2005), the process of heating or dissolving garlic turned out to enable the enzyme allinase. Combining cloves of garlic with distilled water is available enough time for allinase to be liberated and form allicin of alliin. The reason has inspired researchers to study the effects of garlic clove extract in drinking water on productive performance and cholesterol content in eggs in laying hens.

MATERIALS AND METHODS

Animals, treatments, and experimental design: This study used 120 Lohmann Brown hens, 40 weeks of age, with a homogeneous body weight of 1675.26 ± 20.35 grams obtained from a commercial poultry farm. All chickens were given commercial feed specific for laying hens containing 2750 kcal/kg of Metabolizable Energy (ME); 17% of CP; 3.6% of Ca; and available phosphor of 0.48%. For the treatments, hens were placed into three groups each containing 5 hens: (i) hens were only given water as a drink, (ii) hens were given 2 cc of Garlic cloves extract in 100 cc of drinking water; (ii) hens were given 4 cc of Garlic cloves extract in 100 cc of drinking water; and (iv) hens were given 6 cc of Garlic cloves extract in 100 cc of drinking water, respectively.

Each treatment was repeated six times for a total of 120 hens. Food and drinking liquid were given *ad libitum*. The individual hens were weighted weekly, and feed consumption and egg production was recorded daily.

Preparation of Garlic (*Allium sativum*) Extract: Prepare 100 g of fresh garlic cloves (*Allium sativum*) obtained from Local Market for use in research. The garlic cloves are finely ground and mixed in 100 ml of water and then macerated (heated in 45^0C) for 30 minutes and stored overnight (Bidura *et al*., 2017). The blended extract of Garlic was then filtered using a cheese cloth. This extract was used for the treatment, which is given to chickens in drinking water each at level: 2 cc/100 cc; 4 cc/100 cc; and 6 cc/100 cc of drinking water, respectively.

Performance, Egg Quality Metrics, and Laboratory Analysis

Eggs were collected and labeled on a daily basis at 10.00 h and 15.00 h throughout the experimental period. The percent egg production was calculated. Once every two weeks, the eggsfrom three consecutive days were used to measure egg weight and quality. Yolk cholesterol content was analyzed for two consecutive weeks. The content of fat and cholesterol in eggs were analyzed following the Liberman-Burchard methods (Lieberman and Burchard, 1980).

Statistical analysis: All data were analyzed with ANOVA to determine the differences among treatments. If differences were found, then further analysis was performed with Duncan's multiple range test.

RESULTS

The results are presented in Table 1. It is known that the addition of 0.20-0.60% of garlic extract (*Allium sativum*) in drinking water has no significant effect (P>0.05) on feed and drink consumption. However, there was a significant effect (P<0.05) on egg weight per head (g/head), the average number of eggs produced, the average hen-day production, and feed efficiency.

The addition of 0.20-0.60% of garlic cloves extract (*Allium sativum*) in drinking water did not affect the thickness of the eggshell, but significantly (P<0.05) increased the yolk color and significantly (P<0.05) lowers fat content and cholesterol content in egg yolks. The addition of Garlic cloves water extract at the level of 2-6 cc/100 cc in drinking water, apparently (P<0.05) can increase egg weight per head (g/head), the average number of eggs produced, and hen-day production.

Table 1. The effect of Garlic (*Allium sativum*) cloves water extract added in drinking water and administered to 40-50 weeks aged of egg laying hens to the egg production and yolk cholesterol level.

Variables	Treatments[1)]				SEM[2)]
	A	B	C	D	
Feed Consumption (g/head/days)	160.73a	157.82a	154,16a	154,28a	2,705
Water consumption (ml/head/days)	405,81a	386,27a	378,42a	390,61a	10,504
Egg weight (g/head)	57,61b[3)]	61,17a	61,42a	60,98a	0,351
The numer of eggs (egg/70 days)	56,73b	61,43a	60,87a	61.12a	1,082
Hen-day production (%)	81,04a	87,75b	86,95b	87,32b	1,208
Feed conversion ratio (feed consumption: total egg weight)	2,79a	2,58b	2,51b	2,53b	0,013
The thickness of eggshells (mm)	0,341a	0,335a	0,339a	0,327a	0,019
Yolk colour (1-15)	6,25b	8,04a	8,28a	8,35a	0,205
Yolk cholesterol (mg/dl)	175,92a	163,69b	164,27b	164,05b	3,072

Notes:

1. A: drinking water without Garlic cloves extract as control; (B): drinking water with 2 cc/100 cc Garlic cloves extract; (C): drinking water with 4 cc/100 cc Garlic cloves extract, and (D) drinking water with 6 cc/100 cc Garlic cloves extract, respectively.
2. SEM: Standard Error of Treatment Means
3. Means with different superscripts within raw values are significantly different (P<0.05)

The average value of FCR (feed consumption: total egg weight) over ten weeks of observation in the control group was 2.79/head (Table 1). This was significantly different (P<0.05) from hens in treatment groups B and C an average of 7.53%; 10.04%; and 9.32%, respectively lower than control (A).

The mean value of egg yolk color in the control chicken group was 6.25 (Table 1). Addition of garlic clove water extract at level 2 cc/100 cc in drinking water (B); 4 cc/100 cc in drinking water (C), and 6 cc/100 cc in drinking water (D), an average of 28.64%; 32.48%; and 33.60%, respectively significantly different (P<0.05) higher than control.

In Table 1, the total cholesterol content in the egg yolk in the control chicken group was 175.92 m /dl egg yolks. The addition of garlic clove water extract at levels of 2-6 cc/100 cc in drinking water significantly (P<0.05) were decreased the yolk cholesterol content. The cholesterol content in the egg yolk of treatments B, C, and D, an average of 6.95%; 6.62%, and 6.75%, respectively lower than control (A).

DISCUSSION

The results showed that the addition of garlic clove water extract at the level of 2-6 cc/100 cc in drinking water did not affect the consumption of feed and drinking water. This is due to the feed energy content of all treatments is the same. Chickens consume feed to meet energy needs. The results of this study are supported by Wibawa *et al.* (2016) and Issa and Omar (2012) who reported that giving Garlic extract in drinking water at the level of 2.5-5% in drinking water did not significantly affect feed consumption and drinking water, but significantly improved feed efficiency.

The results showed that the addition of garlic clove water extract (Allium sativum) in drinking water can increase egg weight, egg count, and hen-day production. Same as reported by Yalcin *et al*. (2006) that the addition of garlic powder at the level of 5 or 10 g/kg showed an increase in egg production. The same thing was reported by Khan *et al*. (2007) that laying hens given Garlic flour (2-8%) showed higher egg production. According to Adibmoradi *et al*. (2006) that active compounds in Garlic can increase villus height and depth of criptus, as well as decrease in epithelial thickness and number of villi cells in the duodenum, jejunum, and ileum of poultry. Increased villus height, as well as epithelial thickness and goblet in the duodenum, jejunum, and ileum can improve nutrient uptake (Nusairate, 2007). Similar results were reported by Ramakrishna *et al.* (2003), that Garlic supplementation may increase pancreatic enzyme activity and micro-environment conditions for better nutrient utilization in mice. Yalcin *et al.* (2006) showed that the addition of Garlic powder at the level of 5 or 10 g/kg of ration showed an increase in egg production. Also, Khan *et al*. (2007) reported that laying hens given dry garlic (2-8%) showed higher egg production intensity. Raeesi *et al*. (2010) reported that the administration of 1% Garlic flour in the ration significantly resulted in higher breast meat than others. This result is contrary to the research of Ao *et al*. (2010) who found that giving 1% -3% of fermented Garlic flour in food had no effect on egg production and egg weight.

The color of egg yolks in chickens that drank Garlic extract increased significantly. This suggests that the presence of vitamin A or carotenoid pigments are efficiently absorbed and utilized by chickens. Ao *et al*. (2010) reported that giving 1-3% of the fermented Garlic flour in real feed increased the color of the egg yolks compared to the controls. The increase in yolk color is due to the high content of beta-carotene in herbal extracts. Beta-carotene and vitamin E are natural antioxidants.

It is known that antioxidants have an important role in inhibiting and suppressing free radicals. In addition, flavonoids and vitamin C (Andarwulan, 2012) and other phenolic compounds (Nahak and Sahu, 2010) can also act as natural antioxidants. Al Aqil (2016) reported that hens fed diets containing 0.4; 0.8; or 1.0% Garlic powder showed lower (better) feed conversion ratio and lower egg yolk color than those fed diets control. The addition of Garlic extract (*Allium sativum*) at levels of 2-6 cc/100 cc in drinking water significantly decreases the cholesterol content in the egg yolks. The decrease in egg cholesterol is due to the presence of phytochemical compounds in Garlic, such as beta-carotene and *Allyl trisulfide* is a major sulfur compound in Garlic oil having biological activity, such as antibiotics and anticancer found in animal and human studies (He *et al*. 2009). The function of Garlic oil, such as antidiabetes and antiobesity is poorly understood, and decreases glucose levels in normal men (Zhang *et al*., 2001). Increased consumption of beta-carotene can lower cholesterol levels in carcass meat, because beta-carotene can inhibit the enzyme HMG-CoA reductase (Hydroxy metyl glutaryl-CoA) that play a role in mevalonic formation. Mevalonic is needed in the process of cholesterol synthesis by inhibiting the enzyme, thus inhibiting cholesterol formation (Syahruddin *et al*., 2013). Also reported by Issa and Omar (2012) that the reduction of triglycerides is due to the inhibition of Acetyl CoA synthetase enzymes necessary for the biosynthesis of fatty acids. This is due to the possible mechanism of action of hypocholesterolemic and hypolipidemia in Garlic products which can suppress the activity of lipogenic and cholesterogen enzymes from the liver, such as malate enzymes, fatty acid synthase, glucose-6-phosphatase dehydrogenase (Cavallito *et al.,* 1994) and reductase enzymes 3-hydroxyl-3-methyl-glutaryl-CoA. The results of Sakine and Onbasilar (2006) reported that Garlic supplementation could lower plasma cholesterol concentration when laying hens were fed 0.50% Garlic flour and 1.0% in mice significantly lowered cholesterol and triglyceride (Eidi *et al*. 2006). Wibawa *et al.* (2016) stated that the giving of Garlic extract through drinking water at a concentration of 2.5-5% significantly lowered the serum blood fat and cholesterol level. Beta-carotene derived from Sauropus leaf extract of 5 cc/100 cc drinking water significantly decreased cholesterol levels in serum and chicken egg yolks (Bidura *et al*. (2017). The use of compounds in Garlic in feed can reduce plasma cholesterol concentration when laying hens were given Garlic as much as 0.5-1.0% (Sakine and Onbasilar, 2006). Bidura *et al*. (2017) reported that 5.0% of Garlic water extract (*Allium sativum*) in drinking water can increase egg production and can lower serum cholesterol levels and in egg yolks in chickens. The addition of real fermented Garlic powder significantly reduces plasma cholesterol levels and does not cause adverse effects on production (Chowdhury *et al*., 2002; Ao *et al.,* 2010; and Mahmoud *et al*., 2010). Prasad *et al.* (2009) reported similar findings, namely total cholesterol and triglycerides, significantly decreased with the presence of Garlic in rations. Contrary to the results of the study Birrenkott *et al*. (2000) showed that a 3% supplement of Garlic flour in ration was ineffective in lowering yolk cholesterol (mg/g) or other lipid components of serum layers, even when fed for up to 8 months and in broiler chickens (Amooz and Dastar, 2009). Similarly, Yalcin *et al.* (2006) reported that total yolk cholesterol was not affected by Garlic supplementation. Similar findings were reported in mice in which garlic powder failed to affect lipid profiles in mice (Islam and Choi, 2008). Raeesi *et al*. (2010) reported that Garlic at 1% and 3% levels did not have a significant effect on relative carcass weight and pad-fat in chicken.

CONCLUSION

It can be concluded that the addition of 0.20-0.60% Garlic extract (*Allium sativum*) in drinking water has no significant effect on feed and drinking water consumption, and egg shell thickness. However, it can increase egg weight, the numer of eggs, hen-day production, egg yolk, feed efficiency and lower yolk cholesterol levels in laying hens 40-50 weeks old.

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