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Article

Effect of anti-prolactin drug and peppermint on broodiness, laying performance and egg quality in indigenous hens

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Abstract: The study was conducted to determine the effect of antiprolactin drug (Bromergon®) and Peppermint (*Mentha piperita* L.) on broodiness, laying performance and egg quality in indigenous hens. The effect of modulation of prolactin concentration on egg production, sequence length and inter sequence pauses were studied by analyzing the oviposition records. Total sixty indigenous laying hens (30-40 weeks of age), were assigned for treatment with peppermint solution and Bromergon®. Sixty laying hens were randomly divided into 4 groups (A, B, C and D) and each group remained 15 hens. Group A was kept for control, Group B was treated with bromocriptine (Bromergon® SANDOZ) orally @ 640 µg per bird per day, Group C with 50% peppermint solution @ 10 g per bird orally and Group D was with peppermint and Bromergon® combined at previous dose. Over the course of trial, incremental dietary peppermint (Group C) significantly ($p>0.05$) increased egg production, body weight of treated indigenous hens than other groups. Egg shell percentage, thickness and haugh unit of hens fed diets supplemented with peppermint were greater than that of hens fed the control diet. However, peppermint supplementation did not influence other egg quality characteristics like albumen and yolk percentages and albumen height. The treated birds had comparatively longer sequences and fewer pauses. It is concluded that the physiological pauses occur during ovulatory sequences can be disrupted effectively using Bromergon® and peppermint. Prolactin levels modulated which may interfere with follicular recruitment and subsequent oviposition thereby improves egg laying potential of the indigenous hens.

Keywords: prolactin; peppermint; broodiness; egg production

1. Introduction

Bangladesh possesses a large variety of chicken mostly of non-descript indigenous type. The indigenous chickens are also called Indigenous chickens (Okada *et al.* 1988). Approximately 20% of the meat and eggs consumed in developing countries are coming from family poultry (Branckaert and Gueye, 2000). It is reported that Indigenous chickens of Bangladesh are low producers and their mature body weights range from 1.0 to 1.2 kg (Barua and Howlider, 1990). Broodiness basically means a hen sits on her eggs for the purpose of hatching the embryos. Physiologically, it has been well established that prolactin (PRL) plays an important role in the onset of incubation of hens (Sharp *et al.,* 1988; March *et al.,* 1994; Ohkubo *et al.,* 1998). Increased plasma PRL concentration is associated with the occurrence of broodiness (Burke and Dennison, 1980; Bacon, 1983). During incubation, PRL mRNA reaches its highest level (Talbot *et al.,* 1991; Karatzas *et al.,* 1997) which infers that PRL is important in the maintenance of broodiness. Still it is not fully elucidated that a change in the concentration of prolactin is responsible for timing of oviposition in domestic hen (Wentworth *et al.,* 1983) or due to longer intervals of LH surges. Also prolactic plays a role in the development of broodiness in poultry with reference to turkey and bantam hens (Sharp *et al.,* 1988) nothing is reported in this breed. The plant family Lamiaceae has received the greatest interest in poultry feed, with peppermint, thyme and oregano as the most popular representatives (Burt, 2004). These medicinal plants active components which are often called phytobiotics or botanicals are secondary metabolites in medicinal plants with positives effects on animal health and productivity (Windisch *et al.,* 2008; Ghazaghi *et al.,* 2014). Peppermint (*Mentha piperita* L.) is widely used in herbal medicine and believed to be particularly beneficial in building the immune system and antimicrobial properties, as well as strong antioxidant properties and enhance appetite, mainly due to its active components (Dorman *et al.,* 2003: Yalçin *et al.,* 2012). There are few research reports available on its practical usage as feed additives in poultry nutrition especially on broilers and the results showed that peppermint leaves had a growth promoting efficacy at an early stage of broilers life (Toghyani *et al.,* 2010) but there is a scarcity reports on its usage in layer diet. Objective of this study is to observe the effect of Bromergon® and peppermint solution on broodiness, laying performance and egg production, to evaluate the growth performance of supplemented with peppermint solution and to find out any adverse effect related to that treatment in laying indigenous hen.

2. Materials and Methods

The experiment was conducted in santibag village, pirganj, Thakurgaon and conducted with total 60 laying hens of local breed was randomly collected by the method described by Nesheim *et al.* (1979). They were divided into 4 groups. Each group kept 15 no. of hens. Group-A for control, Group-B treated by Bromergon®, Group-C treated by peppermint and Group-D treated by Bromergon®, peppermint combinedly. All indigenous birds were collected from nearby village in pirganj, Thakurgaon and were examined to reduce broodiness to increase egg production. Peppermint (*Mentha piperata*) was selected for its efficacy to increase egg weight, shell thickness and laying performance in indigenous hens. Antiprolactin drug Bromocriptine (Bromergon^R 2.5 mg) was purchased from SANDOZ. At first 50gm of peppermint was weighted by the help of electric balance and then thoroughly washed in tap water, cut into small pieces with the help of knife and were grinded with the help of blender. The grinded parts were filtered through filter paper in a bottle and filtrate portion were measured and mixed with distilled water to prepare 100ml solution where 50% peppermint ingredients contain. Finally 5gm iodide salt was added and stored in a refrigerator at 4^{o}C. Bromocriptine (2.5mg) was collected and grinded with pestle and morter. Then divided into 640 µg per day per bird. Then the prepared drugs were administered through orally in daily. After seven days of acclimatization all the 60 were randomly divided into four equal groups (A, B C and D) for assessing the efficacy of antiprolactin and peppermint to reduce broodiness and increase egg production in indigenous hens. Group A: were kept as control Group B were treated with bromocriptine (Bromergon @ 2.5mg) @640 µg per bird per day for twelve weeks, Group C were treated with peppermint solution @ 20ml (50% sol.) /bird for twelve weeks, Group D were treated with antiprolactin @ 640 µg per bird per day and peppermint solution @ 20ml (50% sol.) /bird for twelve weeks.

All the birds of treated and control groups were closely observed for twelve weeks during treatment. Mean initial and weekly body weight of birds for each group were determined and then body weight gain was calculated. By the end of experimental period, five birds from each group were weighed, numbered and then slaughtered. An open sided house was partitioned into four pens of equal size by using expanded wire net, wood, rod and bamboo materials. It was brushed, swiped properly and cleaned with tap water. After washing the pens were disinfected by using chlorine solution (500ppm). The room was left vacant for 14 days. Later, it was again disinfected with solution (1gm/liter) left to dry up properly. During this time all the feeders, waterers and other necessary equipment were properly cleaned, washed and disinfected with finis solution and dried before use. SCOUR-STOP (100gm) was used to prevent respiratory and digestive diseases of poultry with daily supplied feed. PPM also used with water. These were collected from pirganj upazila livestock office in Thakurgaon. No vaccination program was used during the period under observation. The traditional diets were broken rice, wheat and fresh ad libitum water. Diets were purchased from the local market in pirganj. Broody hens were determined by their persistent nesting and rapid return to the nest if removed. Birds active in egg production were determined by observation and by palpation of the shell gland. Egg production was recorded for each hen at the same time each day for a continuous in experimental (12 weeks) period. Egg sequence length and the number of egg sequences were determined from oviposition records following the procedure described by Blake and Ringer (1987). The number of eggs laid on successive days by a particular hen determined the length of each sequence and the number of pauses in each hen's oviposition determined the number of sequences. For each hen, the length of laying sequence was determined on the day the last egg of the current clutch was laid. If a hen did not experience a pause during that period no value was recorded or else the actual number of pauses observed during that period was recorded. Hen-day egg production, feed consumption, egg weight and hen mortality were recorded daily. All production variables were determined for each replicate. The parameters relative to egg quality were evaluated at 30-40 weeks of age. Twelve eggs were randomly collected per treatment (2 egg per replicate) to determine the parameters. The collected eggs were weighed and broken out individually on a glass plate surface and allowed to sit for 5 min. The height of yolk and albumen, and the diameter of yolk were measured using the slide callipers. Yolks were separated from albumen manually and both were weighed. The weight of shell, albumen and yolk were divided by whole egg weight and then multiplied by 100 to determine percentage weight. Eggshell thickness (without inner and outer shell membranes) was measured at the middle part of the eggshell using screw gauge. Haugh units (99 HU) were calculated from the records of albumen height (H) and egg weight (W) using the following formula: HU = 100 log10 (H−1.7 W0.37 + 7.56) according to Haugh (1937). There was no mortality in experimental birds during the experimental period. However, at the end of the experiment (i.e. after12 weeks) postmortem examinations were carried out and there was significant change occur in ovary. In case of treated birds increase number follicle in ovary other than control group. After postmortem examination, morphological changes were examined. In case of broody hen, shrinkage of reproductive tract but in laying hen higher development of reproductive tract and huge number of prominent follicle found in ovary. All the data were analyzed with SPSS statistics 20.0 software. Probability P<0.05 was considered statistically significant.

3. Results

This study was conducted to determine the effects of Antiprolactin (bromocriptine) and Peppermint (*Mentha piperita* L.) on broodiness, laying performance and egg quality in indigenous hen. This experiment was held in my own village santibag, pirganj, Thakurgaon. The experiment was carried out for 12 weeks from January to June, 2015. The results are described based on the following headings.

3.1. Effect of antiprolactin and peppermint on growth in indigenous hen

The observations for live body weight (gm) means of (A), (B), (C) and (D) groups after 12 weeks of the experimental period were 809.0 ± 5.89g, 829.0 ±7.96g, 1200.0 ± 8.51g and 1180.0 ± 1.70g respectively (Table 1). Among all of the experimental groups, group C supplemented with peppermint got the maximum weight (p<0.05) 1200.0 ± 8.51g. These findings regarding on body weight has very close agreements with the study of A.S. Al-Ankari *et al.* (2004) who performed a research on use of habek mint in broiler chicken diet @15 g/kg and reported significant increase in the live body weight of broiler in the treated groups when compared to control group.

3.2. Egg production and pause days

In indigenous hen, supplementation with peppermint leaves showed effect on broodiness, hen-day egg production and egg quality. There was no mortality and the general health status of laying hens was good during the experimental period. The inclusion levels of peppermint leaves and antiprolactin significantly increased the egg production and decreased pause day than control. Bromocriptine increase the number of eggs (48 eggs) in treated birds (group B) compared to the control birds. (Table 2) Mean pause days were significantly (P<0.05) higher in control birds (35 ± 1.02 days) compared to treated birds 9.00 ± 0.45 days (B), 8.00 ± 0.49 days (C) & 8.00 ± 0.52 days (D). Likewise, dietary peppermint leaves (10g/kg) increased hen-day egg production linearly (P<0.05) by 50(group C & D) than control (group A) 24 (Table 2).

3.3. Egg quality

In case of egg quality (Table 3), dietary addition of peppermint leaves (10 g/kg) to laying hens for 12 weeks resulted increase in egg shell percentage group C (8.23) and group D (8.21) than control group A (7.56) and group B (7.55), and increased both linearly (P<0.003) egg shell thickness. Furthermore, with the dietary peppermint leaves (10g/kg), albumin height linearly increased by group C (5.50) and group D (5.54) than control group A (4.91) (Table 3). Haugh unit increased in group C & D than control (A) significantly. However, dietary peppermint leaves had no effects on albumen and yolk percentages in the laying period of indigenous hens.

3.4. Economics of production

Average rearing cost of indigenous hens kept under different treatment groups viz. (A), (B), (C) and (D) were 4512tk, 9015.75tk, 4705.75tk and 9097tk respectively (Table 4). Miscellaneous cost like medication summed up tk. 20 per group. The average live weight of indigenous hen in group (A), (B), (C) and (D) were 809.0 ± 5.89g, 829.0 ± 7.96g, 1200.0 ± 8.51g and 1180.0 ± 1.70g respectively. The indigenous hen was sold in live weight basis at the rate of tk 250/kg. The net profit in the respective group was found tk 2101.75, tk 1273, tk 7274.25 and tk 2808 respectively.

Table 1. Body weight measure during experimental period (monthly).

Groups	0 month (gm)	1st month (gm)	2nd month (gm)	3rd month (gm)
A	$800.0^{a} \pm 0.84$	$804.0^{c} \pm 5.70$	$806.0^{b} \pm 6.85$	$809.0^{b} \pm 5.89$
B	$825.0^{a} \pm 1.05$	$827.0^{b} \pm 10.80$	$828.0^{b} \pm 2.09$	$829.0^{b} \pm 7.96$
C	$825.0^{a} \pm 1.00$	$1020.0^{a} \pm 10.14$	$1120.0^{a} \pm 10.44$	$1200.0^{a} \pm 8.51$
D	$820.0^{a} \pm 12.66$	$1015.0^{a} \pm 1.84$	$1110.0^{a} \pm 2.81$	$1180.0^{a} \pm 1.70$

Values followed by different superscripts in the same column are statistically significant (p>0.05)

Table 2. Total egg production and pause day per bird during experimental period.

Groups	Egg/clutch	Pause day	Total egg production	Total pause day
A	$12.00^{a} \pm 0.63$	$35.00^{a} \pm 1.02$	$24.00^{b} \pm 1.59$	$56.00^{a} \pm 1.09$
B	$10.00^{a} \pm 0.71$	$9.00^{b} \pm 0.45$	$48.00^{a} \pm 1.64$	$36.00^{b} \pm 1.05$
C	$10.00^{a} \pm 0.89$	$8.00^{a} \pm 0.49$	$50.00^{b} \pm 1.52$	$32.00^{c} \pm 1.22$
D	$10.00^{a} \pm 0.71$	$8.00^{b} \pm 0.52$	$50.00^{a} \pm 1.14$	$32.00^{c} \pm 1.64$

Values followed by different superscripts in the same column are statistically significant (p>0.05)

Table 3. Effect of antiprolactin and peppermint on egg quality.

Parameter	A	B	C	D
Egg weight (g/hen)	$37.0^{a} \pm 1.64$	$37.0^{a} \pm 1.03$	$39.0^{a} \pm 2.21$	$39.0^{a} \pm 1.64$
Egg length (cm)	$4.61^{a} \pm 0.24$	$4.60^{a} \pm 0.20$	$4.80^{a} \pm 0.10$	$4.80^{a} \pm 0.11$
Egg width (cm)	$3.70^{a} \pm 0.17$	$3.70^{a} \pm 0.14$	$3.80^{a} \pm 0.12$	$3.80^{a} \pm 0.22$
Albumen (%)	$46.01^{a} \pm 1.22$	$45.52^{a} \pm 1.38$	$46.20^{a} \pm 1.64$	$46.00^{a} \pm 2.30$
Yolk (%)	$34.43^{a} \pm 0.55$	$34.45^{a} \pm 1.41$	$34.61^{a} \pm 0.95$	$34.67^{a} \pm 1.09$
Shell (%)	$7.56^{a} \pm 0.32$	$7.55^{b} \pm 0.27$	$8.23^{a} \pm 0.25$	$8.21^{a} \pm 0.29$
Shell thickness (mm)	$0.29^{a} \pm 0.03$	$0.29^{b} \pm 0.03$	$0.33^{a} \pm 0.01$	$0.33^{a} \pm 0.00$
Albumen height (mm)	$4.91^{a} \pm 0.08$	$4.90^{b} \pm 0.11$	$5.50^{a} \pm 0.18$	$5.54^{a} \pm 0.19$
Haugh unit score	$77.81^{a} \pm 1.84$	$77.74^{a} \pm 2.88$	$81.09^{a} \pm 1.00$	$81.35^{a} \pm 1.70$

Values followed by different superscripts in the same raw are statistically significant (p>0.05

Table 4. Calculation of cost benefit.

Sources		A	B	C	D
Expenditure	Purchase cost of hens	200tk/hen ×15 =3000	206.25tk/hen ×15 = 3093.75	206.25tk/hen ×15 = 3093.75	205tk/hen ×15 = 3075
	Feed cost	1.2tk/day = 1.2×84×15 =1512	1.2tk/day = 1.2×84×15 =1512	1.2tk/day = 1.2×84×15 = 1512	1.2tk/day =1.2×84×15 =1512
	Pudina	-	-	100tk	100tk
	Bromergon	-	3.5tk/day =3.5×84×15 =4410	-	3.5tk/day =3.5×84×15 =4410
	Mortality	-	-	-	-
	Total cost/hen	300.8	601.05	313.72	606.47
Income	egg	24×10×15 =5250	48×10×15 =7200	50×10×15 =7500	50×10×15 =7500
	Spent hen	3033.75	3108.75	4500	4425
Profit	Income – Expenditure	6633.75-4512 =2121.75	10308.75-9015.75 =1293	12000-4705.75 =7294.25	11925-9097 =2828
Net profit	Profit-medication (20)	771.75-20 =2101.75	1293-20 =1273	7294.25-20 =7274.25	2828-20 =2808
Profit/hen		140.12	84.87	484.95	187.2

Supplementation with peppermint was found to be more profitable than the control (A), treatment group (B) and (D) of indigenous hen rearing (Table 6).peppermint (group C) singly used more profitable other than three groups (Table 6). Antiprolactin has no effect on growth but peppermint has significant effect on growth in indigenous hen than control. Antiprolactin and peppermint also combined has significant effect on growth. Antiprolactin and peppermint both has significant effect on broodiness, laying performance and egg quality of indigenous hen than control.

4. Discussion

Poultry production in Bangladesh is dominated by scavenging indigenous chicken. They are dual purpose birds providing both meat and eggs to the farmers. The scavenging birds are raised with little or no inputs and their productivity is very low and irregular (Huque and Stem, 1993). According to previous observations, indigenous hens in the rural scavenging system start to lay at 190-200 days with 3.5 clutches of 12.5 eggs each and an average egg weight of 39 g. After each clutch the hens brood for a period of three weeks and they hatch chicks with an average weight of 27 g. Amin and Bhuiyan (1995) noted in Bangladesh under scavenging system mature live weight, egg production/hen/year and egg weight of indigenous chicken were 1141.48 g, 35-45 and 35-39g respectively.

4.1. Egg sequence

Indigenous birds treated with bromocriptine and peppermint increased the number of laying days in treated birds compared to the control birds with relatively fewer egg sequences. Convincing evidence has been presented implicating increased prolactin secretion as the cause of reduced circulating gonadotrophins, ovarian regression and the shift from egg laying to the incubation phase of reproductive cycle in the hen (Crisostome *et al.*, 1998). In my study I have observed reduced laying pauses and longer sequences in birds treated with bromocriptine and peppermint orally. The increase in egg production is also due to the rate at which follicles enter their final phase of rapid growth, which is also under the influence of prolactin. At high concentration, prolactin interferes with follicular steroid genesis in avian species (Dajee *et al.*, 1998) and only minimal amounts are required for normal growth.

4.2. Laying pauses

Prolactin at high levels suppresses the FSH induced estradiol production through the aromatase enzyme system (Wang *et al.*, 1980) resulting in reduced steroidogenic potential within the follicles. This reduced steroidogenic potential is not able to produce progesterone sufficient to elicit a positive feedback of LH required for ovulation (Dorrington and Gore-Langton, 1981). In support of my statement that modulation of prolactin using bromocriptine overcomes the inhibitory effect of prolactin on follicular development and subsequent oviposition, I have observed at necropsy that ovaries of bromocriptine and peppermint treated birds had greater number of yellow yolk follicles compared to the control group. This may explain the cause for longer sequences and reduced laying pauses in the treated birds. However, the occurrence of more than 8 days (more than 11 days by Reddy *et al.*, 2005; more than 8 days by Reddy *et al.*, 2006) of laying pauses in birds of both groups may be due to the genetic constitution of individual birds. In the present study the treatment with bromocriptine through oral feeding, during laying was able to control egg production in indigenous hens. According to previous observations, indigenous hens were not treated with peppermint resulting scarcity of available reports on effect of peppermint on laying hens, comparison was done with other studies that used herbs of the similar family Lamiaceae. The beneficial effects of dietary inclusion of herbs on gut health, digestion of nutrients and intestinal integrity have been reported earlier (Cross *et al.*, 2007; Brenes and Roura, 2010). These beneficial effects might be directly associated with improvements in laying performance. In the present study, the general health status of laying hens was good during the experimental period (30-40 weeks of age) which could be related to the house environmental system. Generally, the birds given diets containing peppermint leaves had the greatest overall egg production compared with the birds given control diet proving that peppermint leaves had beneficial action in the oviposition process, imperative on the conversion of digested feed into eggs. Moreover, the increase in productive performance of hens due to this medicinal plant could be attributed to its content of essential oils. These active components (cineole, citral, geraniol, linalool and menthol) have shown to possess antimicrobial, antifungal and antioxidant activities as well as improved digestion and absorption of dietary nutrients (Pattnaik *et al.*, 1997; Bupesh *et al.*, 2007; Radwan *et al.*, 2008) that might have improved the performance and production efficiency of hens in this study. Increased feed intake was earlier reported (Al-Ankari *et al.*, 2004) in broilers fed habek mint (*Mentha longifolia*) at 200 g/kg, hence palatability was not a concern as levels in my study was quite low. In the present study, addition of peppermint improved egg weight, egg production of indigenous laying hens. In this study, egg production significantly increased with dietary peppermint leaves. In agreement with (Abdel-Wareth *et al.* 2013) where addition of thyme or oregano leaves at 10, 20, 30 g/kg in hen diets had significantly increased ($P<0.01$) egg weight, egg mass and hen-day-egg production during the periods of 68 to 72 weeks of age, are consistent with our results. This study showed no significant differences in yolk and albumen percentages among treatments. However, there were significant effects on eggshell percentage, eggshell thickness of laying hens at 30-40 weeks of age. Increased shell weight and shell thickness by addition of peppermint leaves may be attributed to its main active components that possess antibacterial and antioxidant activities (Dorman *et al.*, 2003: Yalçin *et al.*, 2012). It could be postulated that peppermint leaves may provide a healthy environment in uterus (e.g. site of calcium deposition) and consequently increase shell weight and shell thickness more than the control treatment. These improvements in egg shell and Haugh unit are important for the egg food industry, because peppermint leaves may enhance the safety and stability of eggs. The haugh unit score is known as an indicator of egg freshness and is related to shelf life (Williams, 1992). My findings is in agreement with earlier study that showed supplementation of thyme improved shell weight and shell thickness compared to hens fed control diet (Ali *et al.*, 2007). To my knowledge, no publication exists on peppermint and combinedly with antiprolactin for its effects on laying periods in indigenous hens. For clear conclusion, further systematic studies might be considered to elucidate the effect of peppermint and antiprolactin at different stages of production in indigenous laying hens.

5. Conclusions

Controlling the prolactin secretion by antiprolactin means resulted to reduce broodiness and increase in egg production over control birds. Peppermint (Group C) alone reduce broodiness, increase egg production, egg weight, shell thickness, egg shell percentage, haugh unit and body weight in indigenous hens and more economical than other groups. The study has few limitations, lack of facilities to test the effect of vitamin, mineral and hormone. More detailed studies are needed in future to test the influence of vitamin, mineral and hormone to determine the optimal dietary inclusion level, and its economic impact. Further such an approach is more promising, practicable and economical without any traces of chemical in meat, egg and blood to improve egg production in backyard poultry farming. It will be help in the improving protein feed in our daily routine meals; cost will be decrease and fulfill our demand. As well as a positive contribution in our national GDP of Bangladesh. It can be recommended that hemato-biochemical and hormonal test can be performed to evaluate the real effect of them.

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Conflict of interest
None to declare.

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