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Full Length Research Paper

Effect of transient postpubertal hypo- and hyperthyroidism on reproductive parameters of Iranian broiler breeder hens

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One hundred and thirty two 26-week-old broiler breeder hens were randomly assigned into one of three treatments as control, hypothyroid (HYPO; propylthiouracil (PTU)-treated) or hyperthyroid (HYPER; thyroxine (T₄)-treated) group. PTU and T₄ were administered between weeks 30 and 33 of age. Blood sampling was started at week 29, and repeated every week until week 35, coinciding with weekly body weighing. Using ELISA, plasma levels of T₃, T₄ and estradiol were assayed. Egg number, fertility, hatchability, grading of day-old chicks and embryonic developmental stage of unhatched eggs were determined for individual artificially inseminated hen. Effects of PTU and T_4 treatment on plasma T_4 levels were significant (P < 0.05), but T_3 levels were only affected by PTU. No significant effect was observed in plasma estradiol levels. Increased body weight following PTU treatment was not observed in other groups. Weekly egg number of HYPER was significantly lower (P = 0.0435) than other two groups, while the effect of PTU was not significant. Hatchability of fertile eggs (%) in HYPO was significantly decreased (0.00%, P < 0.0001); while other groups did not significantly differ. The number of grade 1 chicks in control was greater than in HYPER group. Fertility (%) was not affected by PTU or T₄. In unhatched eggs, percent of non-pipping or internal pipping stage of embryonic life in HYPO was more than other groups; but external piping was not affected by treatments (P > 0.05). In conclusion, among the different reproductive parameters in this study, hatchability and weekly egg production were the most responsive parameters to decreased or increased plasma thyroid hormone levels, respectively.

Key words: Hypothyroidism, hyperthyroidism, broiler breeder hens, reproduction, egg production, fertility, hatchability.

INTRODUCTION

Thyroid hormones (thyroxine or T_4 and triiodothyronine or T_3) are necessary for reproductive system development and function in birds (McNabb, 2000). Role of thyroid hormones on various processes in birds, including reproduction and gonadal development was reviewed by McNabb (2007), indicating the critical effects of these hormones on different avian bodily system, as also shown in other species (Hulbert, 2000). Thyroid hormones seem to play an important role in testicular development in rats (Cooke, 1991) and are said to affect the capacity of testis to produce sperm (Mendis-Handagama and Ariyaratne, 2001). Buzzard et al. (2000) reported an increased number in Sertoli and germ cells following the neonatal hypothyroidism in rats, but opposite results were reported for hyperthyroidism.

In poultry, thyroidectomy resulted in gonadal aplasia and considerable diminution in egg production in hens and testicular atrophy in cocks (Falconer, 1971). Transient prepubertal hypothyroidism induced with the goitro-

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gen propylthiouracil (PTU), resulted in decreased postpubertal body weight coincided with a transient increase in serum levels of testosterone and sperm production in Iranian indigenous cockerels (Akhlaghi and Zamiri, 2007). Kirby et al. (1996) suggested that appropriately timed PTU treatment may result in a permanent increase in testis size and sperm production in domestic fowl.

Large doses of thyroxine (5 mg/kg diet), had an inhibittory effect on reproductive function in broiler cockerels (*Gallus domesticus*) aged 96 weeks (Jacquet et al., 1993). Thyroid hormones are essential for initiation and maintaining the ovarian function in turkey hens (Lien and Siopes, 1991), although it appears that the presence of thyroid hormones as well as long days is required for initiating and maintaining photorefractoriness in turkey hens and hypothyroidism terminated the photorefractoriness and reinitiated lay in turkey breeder hens (Siopes, 1997).

Although there are reports on maternal thyroid hormone effects on embryonic development or function in quail (Wilson and McNabb, 1997) and turkey (Christensen and Davis, 2001), sex-associated differences in dependency on the thyroid for gonadal function made it difficult to formulate a generalized relationship (Knowlton et al., 1999).

There are few reports on reproductive effects of hyperthyroidism compared to those of hypothyroidism in broiler breeder hens; therefore, this experiment was conducted to determine the effects of transient alterations, either increase or decrease, in plasma thyroid hormone levels on reproductive performance of broiler breeder hens with particular emphasis on egg production, fertility and hatchability as functional assessments of avian reproduction and chick quality.

MATERIALS AND METHODS

Birds and experimental treatments

One hundred and thirty two 26-week-old Iranian broiler breeder hens (Arian strain, Babol-Kenar Line Breeding Center, Iran) were randomly assigned into one of three treatments as control, hypothyroid (HYPO) and hyperthyroid (HYPER) with 4 replicates of 11 hens in each treatment. 6-N-propyl-2-thiouracil (PTU) and thyroxine (T₄) were administered at a level of 100 and 1 mg/lit of drinking water for HYPO and HYPER groups, respectively (starting at week 30 up to the end of week 33); while control group received normal drinking water. Pens were covered with the litter materials and equipped with laying trap nests for daily egg production recording of individually wing-tagged hens. All hens were normally fed with a pelleted diet formulated according to the specifications of the National Research Council (USA, 1994). The lighting schedule and the house temperature was 15 h light: 9 hr darkness and 21 °C respectively throughout the experiment (week 26 - 35) according to Arian broiler breeder management guidelines.

Artificial insemination and blood sampling

Roosters (26-week-old; n = 20) were abdominally massaged to habituate for semen collection. Hens were artificially inseminated

with semen diluted in homogenized and pasteurized low-fat milk (at the ratio of 1-4). Eggs were collected and numbered during the last 13 days of treatment period and stored 12 °C and 75% relative humidity, before being transferred into a hatchery. Blood samples (2 ml) from 6 hens of each replicate (24 hens in each treatment) were drawn from the wing vein, using a 2.5 ml syringe, and EDTA-coated tubes, for seven successive weeks (29-35). Blood sample was centrifuged (3000 RPM for 12 min) and plasma was separated and stored at -20 °C until analyzed for T₃, T₄ and estradiol (E₂) assays, using ELISA (Sauer et al., 1982). The intra- and inter-assay co-efficients of variation were 3.7 and 4.3 for T₃, 3.3 and 4.6 for T₄, and 4.8 and 5.4 for estradiol respectively.

Hatchery

Eggs (n = 1023) were transferred into a hatchery to determine the fertility, hatchability and chick quality. All eggs were set in a same setter/hatchery machine to avoid interassay variability. Using light candling, fertile eggs were separated to continue the hatching process. All candled infertile eggs were broken out for visual verify-cation of the absence of an embryo. On day of transfer from setter to the hatchery machine (early on day 19), eggs from each hen were placed in a separate special hatching basket for determination of hatchability and separate grading of newly hatched chicks [as grade 1 (healthy) or 2 (unhealthy)]. Unhatched eggs were examined to determine the embryonic developmental stages which were categorized as non- (NP), internal (IP) or external (EP) pipping stages.

Statistical analysis

Data were subjected to the procedure GLM, but repeated measure data were analyzed by the procedure mixed of SAS 9.1 (2002 - 2003, SAS Institute, NC). The effects of treatment, age and treatment by age interaction were included in a model with a covariate structure known as autoregressive (AR-1) was fitted. Body weight was included in the model as a covariate for analysis of variance and the mean separation was done by the Duncan's Multiple Range Test (DMRT) or Student-Newman-Keuls (SNK) test (P<0.05). T-test was used to compare each treatment to the control group for different parameters.

RESULTS

Body weight

Overall means of body weight in control (3315 g) and HYPER (3362 g) groups did not significantly differ (P > 0.05); although that of HYPO (3410 g) group was significantly greater than other two (P = 0.035). The significant effect of age (P < 0.0001) on body weight in all three groups was accompanied by a significant (P = 0.0283) effect of PTU treatment x age interaction, but a non-significant (P > 0.05) effect of T₄ treatment x age interaction, in HYPO and HYPER groups respectively. There was a gradual increase in overall means of body weight in all three groups from 29 - 35 weeks of age with the highest value (3671 g) at week 35 in HYPO group (Table 1).

Plasma hormones assay

Plasma T₃ levels were significantly affected by PTU treat-

Table 1. Mean (±SEM) and summary analysis of variance of the effect of propylthiouracil (PTU) or thyroxine (T₄) treatment, age, and treatment x age interaction on body weight, plasma levels of thyroid hormones and estradiol, and weekly egg number of Arian broiler breeder hens.

Parameter	Treatment *			P**					
				Treatment		Age ***		Treatment x Age	
	Control	НҮРО	HYPER	ΗΥΡΟ	HYPER	ΗΥΡΟ	HYPER	ΗΥΡΟ	HYPER
Body weight (g)	3335 ± 79.3 ^b	3422 ± 81.3 ^a	3361 ± 93.3 ^b	0.035	NS	< 0.0001	< 0.0001	0.0283	NS
T ₃ (ng/ml)	1.601± 0.212 ^a	0.871 ± 0.06 ^b	1.625 ± 0.07 ^a	< 0.0001	NS	< 0.0001	NS	< 0.0001	NS
T₄ (ng/ml)	11.878 ± 0.77 ^b	8.005 ± 2.54 [°]	36.085 ± 3.76 ^a	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001
Estradiol (pg/ml)	200.3 ± 18.63 ^a	207.9 ± 25.37 ^a	207.2 ± 26.14 ^a	NS	NS	NS	NS	NS	NS
Weekly Egg Production (No./hen)	5.395 ± 0.70 ^a	5.199 ± 0.82 ^a	4.986 ± 0.84 ^b	NS	0.0435	< 0.0001	< 0.0001	NS	NS

Within each row, treatments with common superscript are not significantly different at P < 0.05 (DMRT).

*100 mg/lit propylthiouracil in hypoyhyroid (HYPO) and 1 mg/lit thyroxine in hyperthyroid (HYPER) and common water in control treatment were used.

**Within each effect, values were reported for each parameter after individual comparison of HYPO or HYPER to control group.

***26 to 35 weeks of age for weekly egg number and 29 to 35 weeks of age for other parameters were considered.

NS: Non-significant.

ment, age, and PTU x age interaction (P < 0.0001) where the lowest mean was recorded for HYPO group (0.871 ng/ml) being significantly smaller than for the control (1.601 ng/ml) or HYPER (1.625 ng/ml) groups (Table 1). T₄ treatment did not result in a significant increase in plasma T₃ levels compared to those of the control group (Table 1). No significant age-dependent changes in plas-ma T₃ levels were observed in control or HYPER group. Both PTU and T₄ treatment resulted in significant effects on plasma T₄ levels, as well as the effects of age and treatment x age interaction (P < 0.0001). Mean plasma T_4 levels were 36.085, 11.878 and 8.005 ng/ml for HYPER, control, and HYPO groups respectively. The effect of age on T₄ levels for control group was not significant (P > 0.05); but, a gradual significant increase in overall mean values was found from week 30 (13.841 ng/ml) to week 33 (25.439 ng/ml) which decreased to lower levels at week 35 (13.887 ng/ml).

The differences in plasma E_2 levels among control (200.343 pg/ml), HYPO (207.886 pg/ml), and HYPER (207.205 pg/ml) groups were not signifi-

cant (Table 1). Similarly, the effects of age or treatment x age interaction were not significant (P>0.05). The highest level of plasma E_2 was recorded at week 33 (216.571 pg/ml) was not significantly different from the lowest value found at week 35 (191.286 pg/ml).

Weekly and total egg production

Weekly egg production (Table 1) was significantly reduced by T₄ treatment (4.79 egg number/week/ hen, P = 0.0435); but, the effect of PTU was not significant (5.42 *vs.* 5.02 egg number/week/hen in control and HYPO groups, respectively; P > 0.05). Age had a significant effect on this parameter (P < 0.0001) for which the highest and lowest rates were recorded at week 27 and 26 (5.96 *vs.* 4.11 egg number/week/hen) respectively.

Treatment x age interaction did not significantly affect this parameter (P > 0.05). Total 10-week-long egg production of the control, HYPO and HYPER groups did not significantly differ (P > 0.05).

Fertility, hatchability and chick quality

Fertility percentage was not significantly affected by PTU (96.5%) or T₄ (96.3%) treatment compared to that of control (98.1%) group (P > 0.05). Although the hatchability percentage of fertile eggs in HYPER was lower (78.2%) than control group (83.1%), but they were not significantly different (P > 0.05). None of eggs produced by PTU-treated hens hatched (Table 2).

Quality grading of newly hatched chicks (Table 2) showed that the grade 1 chick production (as a percent of fertile eggs) in control group (75.4%) was significantly higher than HYPER group (63.7%; P < 0.001). T₄ treatment resulted in higher but non-significant grade 2 chicks compared to that of control group (7.7% *vs.* 14.5% for control and HYPER groups respectively; P > 0.05).

Unhatched eggs and pipping stage

As noted earlier, percentage of unhatched eggs (as a percentage of fertile eggs) in HYPO group

Parameter*		P***			
	Control	НҮРО	HYPER	ΗΥΡΟ	HYPER
Fertility (%)	98.1 ± 0.70 ^a	96.5 ± 1.10 ^a	96.3 ± 1.00 ^a	NS	NS
Hatchability (%)	83.1 ± 3.5 ^a	00.0 ^b	78.2 ± 3.9 ^a	< 0.0001	NS
Grade 1Chicks (%)	75.4 ± 4.3 ^a	00.0 ^c	63.7 ± 3.8 ^b	< 0.0001	0.0438
Grade 2 Chicks (%)	7.7 ± 3.4 ^a	0.00 ^b	14.5 ± 2.8 ^a	< 0.0001	NS
Unhatched Eggs (%)	16.9 ± 3.5 ^b	100.00 ± 0.0 ^a	21.8 ± 3.9 ^b	< 0.0001	NS
NP (%)	7.0 ± 2.0 ^b	60.0 ± 4.9 ^a	4.5 ± 1.5 ^b	< 0.0001	NS
IP (%)	3.3 ± 1.3 ^b	27.7 ± 4.1 ^a	6.2 ± 1.7 ^b	< 0.0001	NS
EP (%)	6.6 ± 2.6^{a}	12.3 ± 2.2 ^a	11.1 ± 2.5 ^a	NS	NS

Table 2. Mean (\pm SEM) and summary analysis of variance of the effect of propylthiouracil (PTU) or thyroxine (T₄) treatment on fertility, hatchability, newly hatched chicks quality, and pipping status of unhatched eggs of Arian broiler breeder hens.

Within each row, treatments with common superscript are not significantly different at P< 0.05 (DMRT).

*Values in hatchability and unhatched eggs are as percentages of fertility.

**100 mg/lit propylthiouracil in hypoyhyroid (HYPO) and 1 mg/lit thyroxine in hyperthyroid (HYPER) and common water in control treatment were used.

***Values were reported for each parameter after individual comparison of HYPO or HYPER to control group. NP: Non-pipping; IP: Internal Pipping; EP: External Pipping; NS: Non-significant.

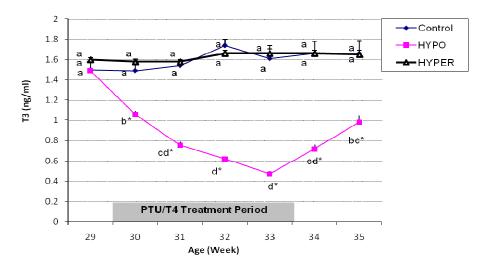


Figure 1. Mean (±SEM) plasma levels of triiodothyronine (T₃) in control, hypothyroid (HYPO) and hyperthyroid (HYPER) Arian broiler breeder hens (n=24 hens/treatment/week). Within each curve, means with common superscript(s) are not significantly different at P < 0.05 (SNK test). Within each age (week), asterisks (*) designate significant differences in HYPO or HYPER group compared to the corresponding values for control group (T-test). HYPO: 100 mg/lit propylthiouracil (PTU) in drinking water; HYPER: 1 mg/lit thyroxine in drinking water; Control: Drinking water without any additives.

was 100% (Table 2). Furthermore, the corresponding values for control (16.9%) and HYPER (21.8%) groups did not significantly differ (P > 0.05). In control group, unhatched eggs (16.9%) included 7, 3.3 and 6.6% of NP, IP and EP stages of embryonic development respectively; while 4.5, 6.2 and 11.1% were recorded for HYPER group. Respective values for the totally unhatched (100%) eggs of PTU-treated hens consisted of 60.0, 27.7 and 12.3%. T₄ treatment had no significant effect on

NP, IP or EP percentages (P > 0.05); but PTU-treated hens showed a significant increase in NP and IP percentages (P < 0.0001); the EP percentage was not significantly affected in these hens (P > 0.05).

DISCUSSION

Figures 1 and 2 show the trends of changes in plasma T₃

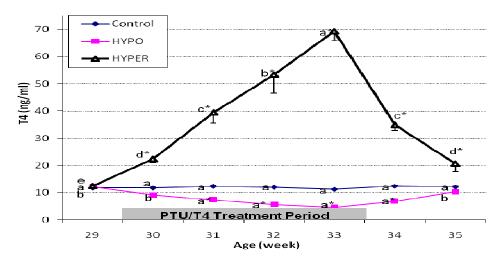


Figure 2. Mean (+/-SEM) plasma levels of thyroxine (T₄) in control, hypothyroid (HYPO) and hyperthyroid (HYPER) Arian broiler breeder hens (n=24 hens/treatment/week). Within each curve, means with common superscript(s) are not significantly different at P < 0.05 (SNK test). Within each age (week), asterisks (*) designate significant differences in HYPO or HYPER group compared to the corresponding values for control group (T-test). HYPO: 100 mg/lit propylthiouracil (PTU) in drinking water; HYPER: 1 mg/lit thyroxine in drinking water; Control: Drinking water without any additives.

and T₄ levels in different experimental groups. In present study, the antithyroid drug, PTU (Ganong, 1993), could significantly decrease the plasma T_3 and T_4 levels in HYPO group. PTU interferes with thyroid hormone biosynthesis at two points. First, thyroid peroxidase inhibition by which PTU as an antioxidant agent is readily oxidized and thereby prevents oxidation of iodide which is required for thyroid hormone synthesis. Secondlyseww, 5'-deiodinase inhibition in peripheral tissues by which PTU prevents conversion of T₄ to T₃ (Ganong, 1993; Taurog, 1996). It is also suggested that H₂O₂ generation, a limiting step in thyroid hormone biosynthesis, is partially inhibited by PTU treatment through the inhibition of Ca²⁺/NADPH-dependent oxidase (Ferreira et al., 2003). By contrast, exogenous T₄ treatment resulted in a significant increase in plasma T₄ levels in HYPER birds compared to those of control; but no significant changes were observed in plasma T₃ levels which can be attributed to status in which excess exogenous T₄ rapidly and almost totally converted to the metabolically inactive reverse-T₃ (rT₃) (Decuypere et al., 1987).

In the present study, PTU treatment resulted in a significant increase in body weight of HYPO group; whilst, there was no significant difference between body weights of T₄-treated and control groups. Thyroid hormones are required for growth in birds (McNabb, 2007) and stimulate growth hormone (GH) secretion in birds, reptiles, rats, and human (Roussean et al., 2002). Thyroidectomy resulted in considerable reduction in growth rate in young chickens (McOre et al., 1984, Cogburn et al., 2000) or in chick embryo (McNabb et al., 1984; Cogburn et al. 2000). Pandy and Bajaj (1974) reported a very low growth rate

in both sexes of thyroidectomized or hypothyroid 4-weekold chicks. PTU-induced prepubertal hypothyroidism resulted in a decreased adult body weight in Iranian indigenous cockerels (Akhlaghi and Zamiri, 2007). Similarly, weight loss after PTU treatment was reported in cockerels (Jacquet et al, 1993) and rats (Mendis-Handagama and Ariyaratne, 2001). A decrease in hepatic mRNA expression of GH receptor and insulin-like growth factor-I (IGF-I) in PTU-treated chickens was reported by Tsukada et al. (1996). T_4 treatment in adult male hypothyroid rdw rats resulted in increased body weight (Jiang et al., 2000). In sex-linked dwarf chickens which are deficient in hepatic 5'-deiodinase enzyme (which converts T₄ to more biologically active thyroid hormone, T_3) and thus, are T_3 deficient (Decuypere and Kühn, 1987), T₃ or T₄ administration had a stimulatory effect on growth (Scanes et al., 1983; Marsh et al., 1984); this stimulatory effect in growing chicks can be mediated either by increases in circulating concentrations of IGF-I and/or by direct effects of T_3 on the growing tissues (Cogburn et al., 2000). Within certain ranges, growth in birds is related to thyroid hormone exposure; however, at circulating thyroid hormone concentrations, either above or below this range, growth is decreased (McNabb and King, 1993; Cogburn et al., 2000) as were reported in continuous dietary T₃ administration in growing chickens (Decuypere et al., 1987) or different thyroid manipulating treatments in domestic ducks (Bishop et al., 2000). The effect of thyroid hormone on protein and lipid metabolism is of a biphasic nature in which in low physiological concentration they are anabolic while at higher concentration they are catabolic (Decuypere et al., 2005) and inhibit growth in many tis-

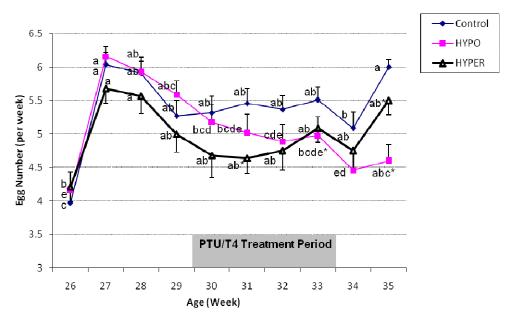


Figure 3. Mean (+/-SEM) weekly egg production (No. /hen) in control, hypothyroid (HYPO) or hyperthyroid (HYPER) Arian broiler breeder hens (n= 44 hens/treatment/week). Within each curve, means with common superscript(s) are not significantly different at P < 0.05 (SNK test). Within each age (week), asterisks (*) designate significant differences in HYPO or HYPER group compared to the corresponding values for control group (T-test). HYPO: 100 mg/lit propylthiouracil (PTU) in drinking water; HYPER: 1 mg/lit thyroxine in drinking water; Control: Drinking water without any additives.

sues (Hulbert, 2000). In an 8-week period of thyroid hormone treatment in domestic ducks, Bishop et al. (1995) reported no increase in muscle or body mass which is in agreement with present study. It may seem that the most of these reports are in contrast to the present study which reports higher body weight in hypothyroid hens; but, indeed, these reports included different treatments in nonadult animals or poultry.

Although the growth is a relatively insensitive indicator of thyroid disruption (McNabb, 2007), the picture for adults can be different. Increased body weight (partly fluid) in hypothyroid and decreased one in hyperthyroid adult human are observable symptoms of abnormal thyroid function (Wartofsky,1990; Ganong, 1993), although decreased growth is common in young hypothyroids (Wartofsky, 1990). During long-term thyroid hormone deficiency, some weight gain usually occurs which is coincided with an accumulation in mucopolysaccharids in subcutaneous tissues and other organs termed myxedema in human (Griffin, 2004). Usual decreased appetite (hypophagia) in hypothyroids (Wartofsky, 1990; Griffin, 2004) could not affect the body weights of hypothyroid hens in the present study; because, restricted feeding regimen which is a common practice for preventing fatness in broiler breeders made all hens in different groups of the present study to completely consume their daily rations. Overall, it can be stated that increased body weight in hypothyroid hens of the present study can be attributed to higher accumulation of water in body and in to some extent to increased adipose tissue content (Wentworth and Ringer, 1986) or decreased mobilization and meta-bolism of lipids (McNabb, 2007).

Treatment with T_4 decreased the weekly egg production, while no significant effect was observed in PTU-treated hens, as compared to the control; however, the total egg production during the 10-week-long experimental period did not significantly differ among different groups (Figure 3). Thyroid hormones are required for normal reproductive system development (Decuypere and Verheyen, 1986) and reproductive activity in female birds (McNabb, 2007). Literatures on hyperthyroidism effects on egg production in poultry are limited compared to those of hypothyroidism or thyroidectomy.

Thyroxine treatment resulted in testicular regression and decreased hypothalamic gonadotropin releasing hormone (GnRH) in male starlings, indicating that T_4 mimics the effects of long-day length (Boulakoud and Goldsmith, 1991). Feeding dessicated thyroid to laying hens suppressed the rate of growth of the ovum and reduced the size of the mature yolk (Asmundson and Pinsky, 1935) in which the inhibitory effects of thyroid hormones on ovarian growth may result from a depression of concentrations of yolk precursors for the ovary (van Tienhoven, 1961). Decrease in egg production of T_4 -treated hens can be attributed to antigonadal effects of high concentrations of thyroid hormones (Decuypere and Verheyen, 1986). In case of decreased or removed thyroid hormones, there are several reports on decreased or ceased egg laying either in hypothyroid or thyroidectomized females as early as 1930s. Thyroidectomy induces delayed gonadal maturation (Blivaiss, 1947) and decreased egg production (Winchester, 1939; Blivaiss, 1947) in female domestic fowl and decreases egg production and ovarian weight in ducks and egg production in hens (Berg and Bearse, 1951). Hypothyroidism results in decreased egg production and egg weight in poultry (Wentworth and Ringer, 1986); so that, temporary treatment with goitrogen has been used as a strategy for altering the timing and performance of egg laying in chickens (Lien and Siopes, 1993a, b). Wilson and McNabb (1997) reported that treatment with another antithyroid drug, methimazole (8 mg/day for 40 days), following a former treating period (4 mg/day for 30 days) resulted in ceased laying in Japanese quails.

Furthermore, treatment of developing female chickens with thiouracil from 0-6 or 6-16 weeks of age decreased egg production (Peebles et al., 1994). Thyroid inhibition in adult hens is associated with complete cessation of egg laying in galliforme birds (Decuypere et al., 1991). In present study, however, treatment with PTU had no significant effect on egg laying which may be attributed to lower dose of PTU (100 mg/lit) administered in drinking water. This dose was sufficient enough to decrease the plasma levels of thyroid hormones (8.005 and 0.871 ng/ ml for T₄ and T₃, respectively, compared to corresponding levels of 11.878 and 1.601 ng/ml in control group). Besides, in reports mentioned above, treating period was longer (6 or more compared to 4 weeks in present study). Herein, we aimed to study the egg characteristics of PTU-treated hens in which treatment with PTU at a dose of 100 mg/lit for 4 weeks coincided with both significant decreased plasma levels of thyroid hormones and unaffected egg laying, so that the aim was met.

Neither plasma levels of E₂ nor fertility was affected by hypo- or hyperthyroidism in the present study. However, the hatchability percentage of fertile eggs was significantly reduced in PTU-treated hens. Hatchability of eggs in T₄-treated hens was lower, but not significantly, than the control group; T₄ treatment increased the number of grade 2 (lower guality) hatchlings compared to that of control. There are several reports indicating that maternal thyroid hormones are transferred into the egg prior to laying (Hilfer and Searls, 1980; Sechman and Bobecks, 1988; Prati et al., 1992; Wilson and McNabb, 1997; McNabb, 2002). Plasma lipoproteins and thyroxinebinding prealbumin (transthyretin) are important for transporting the circulatory maternal thyroid hormones into the volk (McNabb, 2002) and embryo (Noble, 1991; Southwell et al., 1991; Speake et al., 1993). Hens deposit thyroid hormones in their eggs in relation to their own thyroid status; so maternal hormones are available in the egg prior to the time when the embryonic thyroid gland is producing and releasing appreciable thyroid hormone (almost on the 9th day of the incubation). Thyroid hormones exist not only in the yolk, but also in the albumen

(McNabb and Wilson, 1997). Embryonic plasma T₃/T₄ ratio rises in conjunction with internal pipping into the air cell and remains high throughout the remainder of the perihatch period (McNabb, 2007), which led to the historic idea that T₃ was the "hatching hormone" (Freeman, 1974). Increased secretion of embryonic hypothalamic corticotrophin-releasing hormone (CRH) (Vanderborne et al., 2005) and resultant increase in secretion of growth hormone from pituitary (De Groef et al., 2008) which results in down-regulation of the deiodinating enzyme, 5D-III, (decreased T_3 degradation) and a slower increase in 5⁻D-I activity (increased T₃ production from T₄) (Galton</sup> and Hiebert, 1987) along with increased number of pituitary thyrotropes (Nakamura et al., 2004; Muchow et al., 2005) and increasing size of thyroid gland (De Groef et al., 2006) have been considered as the major reasons for increased embryonic plasma T₃ levels during the perihatch period. Maternal thyroid hormones could influence embryonic development in Japanese quail (Wilson and McNabb, 1997) and treatment of the hens (Christensen and Donaldson, 1994; Decuypere et al., 1982; McNabb, 2007) or the eggs (Decuypere et al., 1988) with goitrogens and resultant decrease in maternal/embryonic thyroid hormones was associated with delayed or failed hatching; while small amounts of exogenous T₄ introduced into turkey eggs prior to incubation could improve hatchability (Christensen, 1985). Elevated thyroid hormone, however, can be fatal (Christensen and Davis, 2001). In the present study, none of the eggs produce by hypothyroid hens hatched which can be due to decreased maternal and/or embryonic circulatory levels of thyroid hormones; since PTU could decrease maternal plasma T_4 and T_3 levels which result in decreased yolk levels of thyroid hormones.

Alternatively, this antithyroid drug can be transmitted into the egg and affect the embryonic thyroid gland (Moseley and Landauer, 2003). Thyroid hormones stimulate a variety of metabolic and developmental processes necessary for successful hatching (McNabb, 2007). Incubation period in eggs which are produced by goitrogen-treated hens has been longer (Christensen and Donaldson, 1994). As the incubation period in the present study was 21-day; it can be stated that these embryos had not enough time to complete hatching process; therefore, no eggs hatched. This is supported by pipping stages data (Table 2) of this group for which most of the embryonic loss was recorded at NP (60%) and IP (27.7%) stages, i.e. before starting the external pipping.

Conclusion

Although the picture may be different for treatments that using different birds, doses and duration of treatments in other treating dose/length and other experimental birds, among the different reproductive parameters of broiler breeder hens investigated in this study, hatchability and weekly egg production were the most sensitive parameters to decreased or increased plasma thyroid hormone levels, respectively.

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