

SHORT REPORT

Changes in the blood plasma testosterone and cholesterol concentrations during sexual maturation of Pharaoh quails

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Blood plasma concentrations of testosterone and cholesterol were measured during sexual maturation (age 40-66 days) of Pharaoh quail males and females. Blood samples were withdrawn at two-days intervals. In males and females testosterone level increased significantly from week 6 to week 8, with significant differences ($P < 0.01$) being identified between sexes. Plasma cholesterol level was found higher ($P < 0.05$) in males, being, however, not affected by birds' age.

The results presented confirmed that testosterone concentration in the blood plasma may be involved in the physiological control of sexual maturation process in quails of both sexes. Moreover, the results obtained have provided data on biochemical composition of blood plasma in quails during sexual maturation.

KEY WORDS: blood plasma / cholesterol / quail / sexual maturation / testosterone

In many countries an increase in demand for foodstuffs of animal origin has been noticed, mostly for eggs and poultry meat, the latter being characterized by high nutritive and dietary value. This prompted the breeders to put more interest in the

breeding and keeping of quail, the laying performance of which is relatively high. Over the 6 months laying period the total egg production in quails is ten times higher than female's body weight, whereas in chicken such a relation is reached only by the production gathered from 12 months [Richtrova 1999]. The synthesis of secondary egg integuments in quail lasts for about 17 hours, *i.e.* much shorter than in chicken. The effect of the so called rapid "egg synthesis" may be seen in laying two eggs during the same 24 hours period by quails [Rutkowski 2000].

Accordingly, an increase of interest has also been observed in neurohormonal regulation of sexual maturation in quails, which is a process composed of changes at all levels of control axis [Balthazart and Schumacher 1984, Balthazart *et al.* 1998]. The secretion of sexual hormones and their blood concentrations undergo continuous alterations that show regular character. All processes that take place in organism show circadian oscillations, but their phase and amplitude, as well as frequency, may be altered by different environmental factors [Cymborowski 1984].

Considering the importance of testosterone in hormonal regulation of reproductive processes in males and females, the study was aimed at determining the changes in the concentration of this androgen in the blood of Pharaoh quails of both sexes during sexual maturation. Moreover, investigated were the changes in the blood plasma concentration of cholesterol (as the main precursor for the synthesis of steroid hormones, including testosterone).

Material and methods

The study was carried out on 20 Pharaoh quails (10 females and 10 males), coming from the same hatch. The quails were reared on a farm owned by the Department of Poultry and Ornamental Birds Breeding of the Agricultural University of Szczecin. During rearing, the birds were kept under optimum environmental conditions (temperature from 20 to 22°C, 65% relative humidity) in group cages, with free access to feed and water and fed with complete feed mixed diet (loose) prepared according to standards recommended by the Institute of Animal Physiology and Nutrition [1996]. The quails were kept under light regimen of 17 L and 7 D. The light phase started at 5:00 a.m. and lasted until 10:00 p.m. The observations were initiated in week 6 and completed in week 10 of birds' age (day 40-66).

Blood was collected from wing vein of each quail in the morning, always at the same time before feeding, in two days intervals. In the blood plasma, testosterone and cholesterol were determined. Concentration of the former was determined with immunofluorescent method (DelfiaR PERKIN-ELMER, Wallac Oy, Turku, Finland). The method sensitivity and intra-series error amounted to 0.1 ng-cm⁻³ and 5.2%, respectively. The fluorescence was measured using WALLAC 1420 Victor² fluorometer. Cholesterol concentration was determined with enzymatic-colorometric method (ALPHA Diagnostic, Poland). The value of standard amounted to 7.8 mmol-

dm⁻³, and linearity was 20.12 mmol·dm⁻³. The absorbance was measured on a Epoll 20 photometer with wave length 500 nm.

For the parametres examined, mean values and their standard deviations were computed, taking into account successive sample collections as well as division into weekly periods. The comparison of group means (males, females) was made with Duncan's test. The calculations were performed with STATISTICA software package (version 5.5, Statsoft Poland).

Results and discussion

The quail reaches sexual maturity fast, and metabolic transformations that take place in bird's organism are highly dynamic. Figure 1 shows that this dynamics refers also to the synthesis of the hormone. Both in males and females, testosterone concentration was the lowest in week 6, showing then a clear increase until week 9. Over this period, *i.e.* from week 6 to 9 testosterone level in the blood of quails of both sexes was more than two times higher compared to week 6. The increasing secretion of testosterone between week 6 and 9 of life confirms that sexual maturity in Pharaoh quails commences at that time. Testosterone is considered to be one of the major

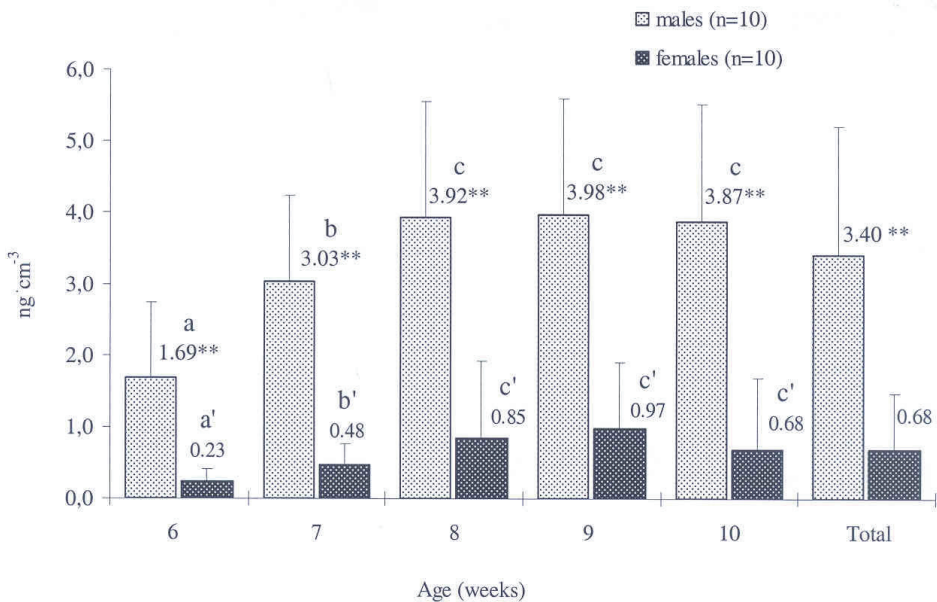


Fig. 1. Concentration of testosterone in the blood plasma of quails from week 6 to week 10 of age (means±SE). Males: means marked with different letters (a, b, c) differ significantly at P<0.01. Females: means marked with different letters (a', b', c') differ significantly at P<0.01. **Differences between means for males and females are significant at P<0.01.

hormones responsible for reproductive processes, and increase in its content of blood plasma is correlated with sexual maturation [Duchala *et al.* 1984, Schlinger and Callard 1989]. In the present study testosterone content of plasma in males appeared similar to that observed in turkey males by Yang *et al.* [1998]. The testosterone level ranged there from 0.4 to 2.0 ng.cm⁻³ until sexual maturity, whereas it amounted to about 4.0 ng.cm⁻³ at the age of 29 weeks. In that time, an increase in the semen production has been observed.

Mean testosterone concentration in males for the whole period analysed, as well as in particular days and weeks, was significantly higher than in females (P<0.01). One can not exclude, however, that the differences between males and females just on day 40, do not occur in the earlier period of life. As Schumacher *et al.* [1988] stated, sexual differences in blood testosterone concentration in Japanese quails occurred just in the first weeks of life.

Figures 2 and 3 show, however, that despite these differences the profile of changes in hormone concentration demonstrated alike upward trend, in particular at the initial observation stage, *i.e.* to the age of 50 days. After this period, it remained in males at similar increased level with some fluctuations, whereas wide differences were found in females, with the upward trend occurring between successive observation days.

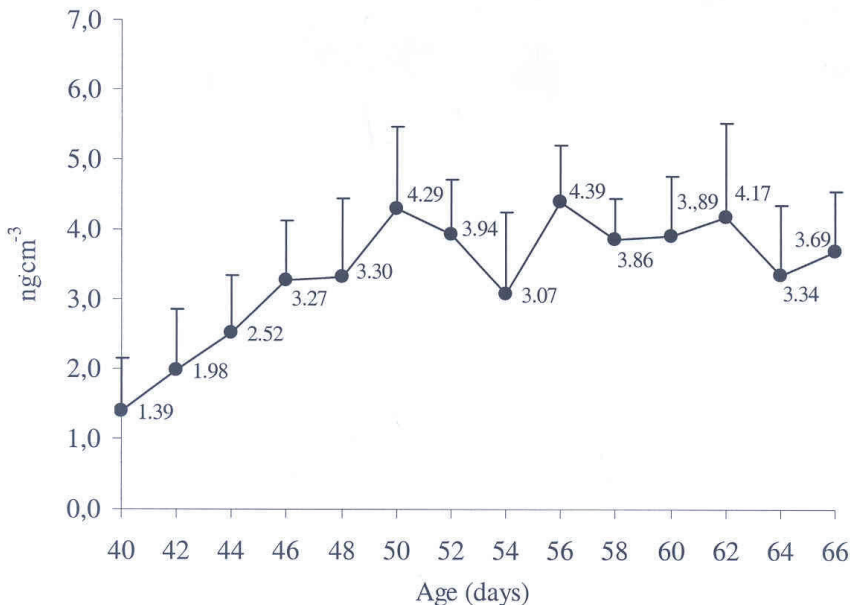


Fig. 2. Changes in testosterone concentration in the blood plasma of male quails from day 40 to day 66 of age (means±SE, n=10).

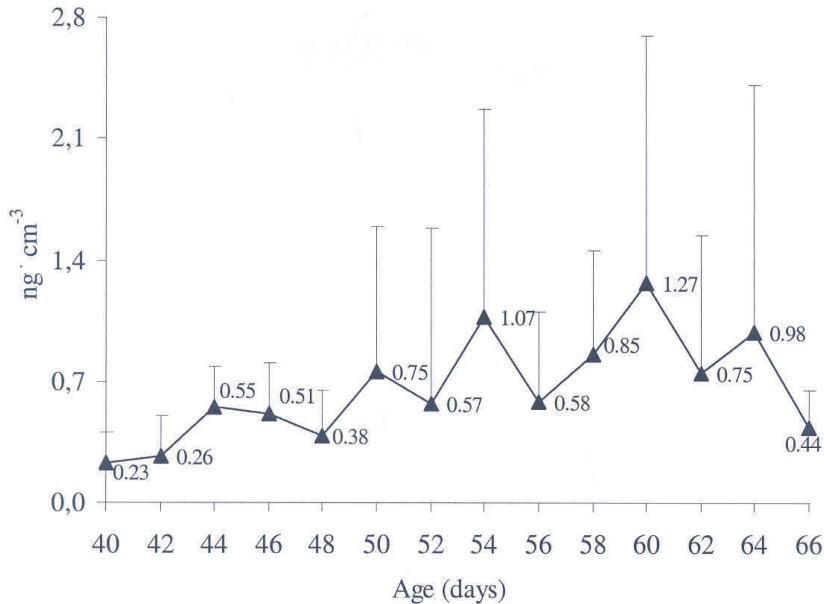


Fig. 3. Changes in testosterone concentration in the blood plasma of female quails from day 40 to day 66 of age (means±SE, n=10).

The important role of testosterone in birds is emphasized not only in males, but also in females [Balthazart *et al.* 198, Schlinger and Callard 1989]. As Rzaşa [1987] stated, testosterone initiated pre-ovulation peak of LH together with other ovarian steroids. Testosterone, together with progesterone, LH and estradiol, is an ovulation-provoking factor, and its blood concentration increase precedes ovulation by several hours [Rzaşa 1987, Brain *et al.* 1988]. Testosterone concentration in quails increases 6-9 hours before ovulation [Gulati *et al.* 1987].

In the present study, in females, there were periods of increased testosterone levels in the form of peaks reaching up to 5.1 ng·cm⁻³ (Fig. 4), which was expressed also by high standard deviation values. Such trends in testosterone secretion were observed in particular up to day 50 of birds' life. Thus, one may suppose that the testosterone peaks found in females had resulted from intensified processes connected with ovulation cycle, and the same with increasing sexual activity. This confirms the opinion cited above Pharaoh females reach sexual maturation, as reflected with ovulation cycles, by about day 50 of life.

Mean cholesterol contents of plasma are presented in Figure 5, showing not marked changes from week to week. Similar observations were made by Peebles *et al.* [1996], who did not show any relation between age and blood lipid concentration in quails during their sexual maturation. In the present study, however, significant

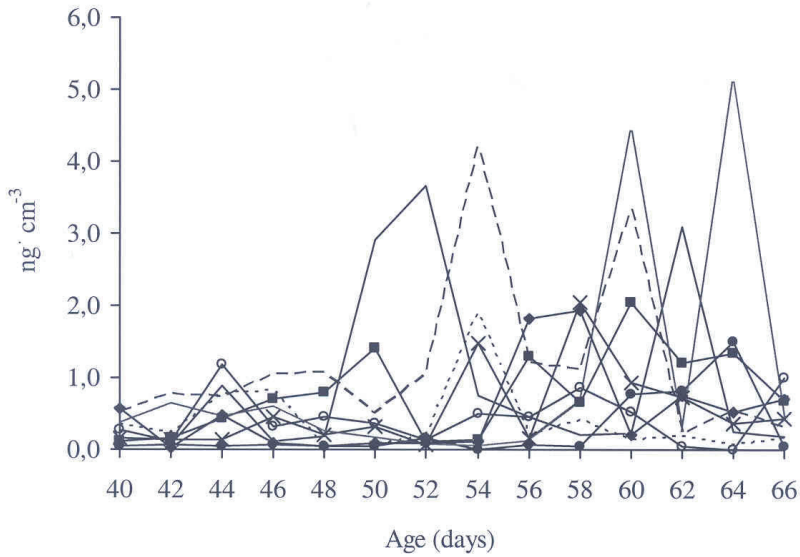


Fig. 4. Changes in testosterone concentration of blood plasma in individual female quails from day 40 to day 66 of life.

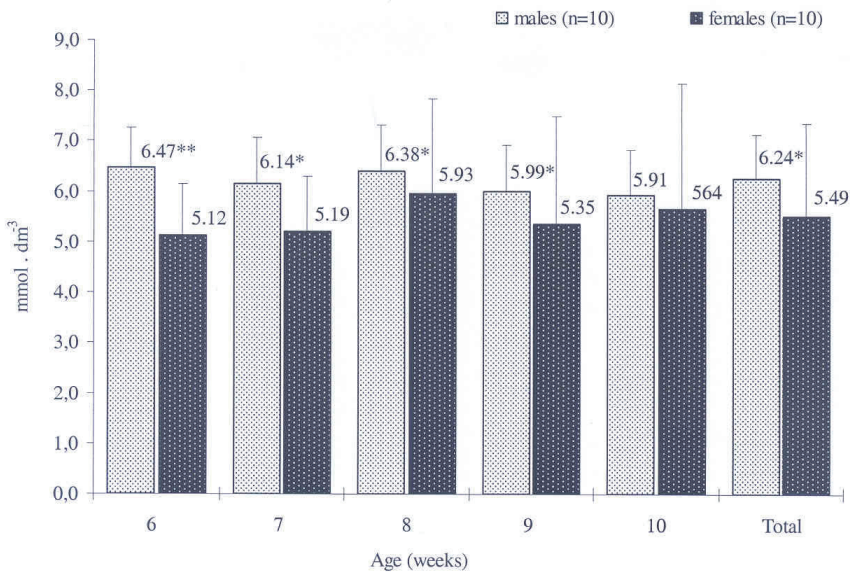


Fig. 5. Concentration of cholesterol in the blood plasma of quails from week 6 to week 10 of age (means±SE). **Differences between means for males and females are significant at $P<0.01$. *Differences between means for males and females are significant at $P<0.05$.

differences were stated in cholesterol concentrations between sexes ($P < 0.05$). Intersexual differences were also showed by Itoh *et al.* [1998] who, contrary to the results of the present study, found higher cholesterol concentration of blood plasma in females than in males. It is very likely that different results arose from the adopted research scheme. In the present study, investigations have been made within chronological frame, withdrawing blood samples from the same bird a number of times, whereas Itoh *et al.* [1998] collected blood samples once. Furthermore, earlier own studies [Błaszczuk *et al.* 2002] suggest the presence of circadian rhythm in the changes of blood cholesterol concentrations. Moreover, content of cholesterol, as well as of other blood lipid components may depend on climatic conditions, feeding, animal condition and sexual activity [Itoh *et al.* 1998]. The cholesterol concentration values stated in the present study may be complementary to investigations on blood plasma lipid content during the sexual maturation of Pharaoh quail.

The results presented here can be summarized as follows.

The increasing testosterone concentration found in successive weeks of observations suggests that Pharaoh quail males reach full sexual activity at the age of 8-9 weeks. The increased frequency of testosterone peaks observed in females in the period of time from the 7th week of life onwards may reflect the ovulation activity starting at that time.

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Zmiany koncentracji testosteronu i cholesterolu w osoczu krwi przepiórek rasy faraon w okresie dojrzewania płciowego

Streszczenie

Określono stężenie testosteronu i cholesterolu w osoczu krwi samców i samic przepiórki rasy faraon podczas dojrzewania płciowego (między 40 a 66 dniem życia). Krew pobierano co dwa dni. W okresie od 6 do 8 tygodnia życia zanotowano u samców i samic znaczący wzrost stężenia testosteronu; ponadto stwierdzono istotne różnice między płciami ($P<0,01$). Podobne różnice zaobserwowano w stężeniu cholesterolu, którego osocze krwi samców zawierało istotnie więcej ($P<0,05$) niż osocze krwi samic. Jednak koncentracja cholesterolu nie zmieniała się z wiekiem.

Przeprowadzone badania wskazują, że testosteron może być elementem kontroli procesu dojrzewania przepiórek obu płci. Uzyskane wyniki mogą także stanowić uzupełnienie danych o biochemicznych wskaźnikach osocza krwi przepiórek w okresie dojrzewania płciowego.

