

CAN ROOSTERS' HEAD ORNAMENTS SERVE AS A CRITERION FOR THEIR SELECTION AS VALUABLE REPRODUCERS?*

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Abstract

It is widely practiced that before mating the roosters are selected as reproducers mainly on the basis of head ornaments appearance. Relationship between males secondary sexual traits (comb and wattles) with testes size, as all of them are testosterone-dependent traits, were examined. Measurements of head ornaments and testes from two broiler breeder lines and two laying lines of chicken were correlated. The value of fluctuating asymmetry (FA) of bilateral traits was also measured. Positive correlations were found for head ornaments and testes weight in one of broiler breeder lines. No correlations were found for males of laying lines. The degree of FA did not differ between lines, except for FA of wattle length, which was higher (P \leq 0.05) for one of the broiler lines. Results obtained indicated that head ornaments cannot be used as the only criterion for rooster selection as reproducers.

Key words: roosters, head ornaments, secondary sexual traits, testes size

Male birds often have showy and characteristic secondary sexual traits, such as comb, wattles, ear lappets and bright, colourful plumage. Their colour and size might be indicators of males' health, sexual maturity, and status in the flock (Johnsen and Zuk, 1996; Zuk et al., 1995, 1990). Showy ornaments may also affect the mating success as females prefer to mate with males that would give the offspring the best chance of survival (Zuk et al., 1990). This is important especially in the reproductive flocks of poultry, because male selection prior to onset of the reproduction cycle is based on, among others, the development of the head ornaments.

The most influential impact on female mate choice is the comb. Redder and larger combs increased the possibility of mating (Zuk et al., 1995). Neither asymmetry of bilateral ornaments nor manipulation of the other secondary sexual characters, such

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as removing wattles or painting ear lappets, influenced red jungle-fowl female mate choice (Ligon et al., 1998).

Unlike the plumage, the size and colour of comb, wattles, and ear lappets are associated with testosterone level (Balthazart and Hirschberg, 1979; Ligon et al., 1990; Rintamaki et al., 2000; Zuk et al., 1995). Male testes are responsible for secreting testosterone and caponization reduces comb size and body weight (Chen et al., 2009; Chiasson and Carr, 1985).

A positive correlation between circulating testosterone levels and testes weight in broiler breeders was found by Brougher et al. (2005). However, Uller et al. (2005) observed that injecting testosterone into the yolk of Chinese quail eggs before incubation resulted in a decrease of testes mass and length.

Fluctuating asymmetry of bilateral traits, which is considered as a measure of developmental stability (Møller, 1994), might be correlated with the number of eggs laid by hen (Forkmann and Corr, 1996) and with the traits size. According to Møller (1990) the bigger bilateral traits tend to be more symmetrical.

In the present study, in two meat type (broiler breeders) and two laying lines of chicken we examined whether the rate of secondary sexual traits development is related to testes size. We also investigated whether testes, head ornaments size and fluctuating asymmetry differed among chicken types and lines.

Material and methods

Wattles, combs, and testes were collected from roosters in semen production at 40 weeks of age from two laying (ISA Brown and Lohmann Brown) and two broiler breeder (Hubbard Flex and Hubbard F15) lines (n=15 per line), immediately after decapitation. The roosters of all lines were derived from relevant breeding farms where they were kept on a deep litter, under fully controlled environmental conditions and fed with commercial diet, according to standard protocols for particular lines of chicken. The birds of both meat type lines had white plumage, while laying line roosters were dark brown. All males had red, well developed wattles and single comb, however the comb of laying line males were shortened at the day of hatch.

Before slaughter roosters were weighed to the nearest 1 g. Wattles, combs and testes were weighed on a digital scale (wattles together) to the nearest 0.1 g and measured to the nearest 0.1 cm using digital callipers for individual males (Figures 1 and 2). Before measurements wattles and combs were carefully flattened on the table. Testes thickness were measured at their thickest point. Comb width was not measured in roosters from the laying lines because, as mentioned above, they were dubbed at the first day of life. Testes weight in relation to live body weight (relative testes weight) was also calculated.

The average values of body weight, testes size and head ornaments size were analysed using a one-way analysis of variance followed by Duncan's post-hoc test to determine the significant differences between lines using STATISTICA (StatSoft, Inc. 2014; ver. 12). Differences with values of P \leq 0.05 were considered to be statistically significant. The data was also analysed by Pearson's correlation.

Fluctuating asymmetry (FA) was measured for bilateral traits. The degree of relative FA between the left (L) and right (R) trait was calculated according to the following formula: |L - R|/(L + R) (Møller and Swaddle, 1997). These measurements did not deviate from normal distributions with a mean value of zero (Shapiro-Wilks test, P>0.005 in all cases), which indicates that the traits demonstrate a fluctuating asymmetry.

The experiment was approved by the II Local Ethics Commission for Experiments Carried out on Animals at Wrocław University of Environmental and Life Sciences. All procedures contributing to this work complied with the ethical standards.



Figure 1. Head ornaments measurement method (male of Hubbard Flex line)



Figure 2. Testes measurement method

Results

For the laying lines, live body weight was higher for males of ISA Brown line, while the right testis weight and length was higher for Lohmann Brown line (Table 1). The differences were statistically significant (P \leq 0.01). Statistically significant (P \leq 0.05) differences occurred also in testes width. For broiler lines significant differences were found only for comb weight (P \leq 0.01) and comb length (P \leq 0.05) (Table 2). All evaluated traits were higher for the meat type than for the laying type lines, but the differences in comb measurements (weight, length, width) were caused by dubbing procedure that roosters of laying type had undergone at the day of hatch. The average values of relative testes weight are shown in Table 3.

All characteristics were correlated with each other for each line separately (n=15 per line). No correlation between any measured traits in laying lines were observed. The findings revealed strong and positive relationship between testes and head ornaments only in meat type line Hubbard F15 (Table 4).

		ISA Brown				Lohmann Bro	uwo	
Items	weight (g)	length (cm)	width (cm)	thickness (cm)	weight (g)	length (cm)	width (cm)	thickness (cm)
Left testis	11.63 a*±5.15	4.37±0.87	2.21 a±0.48	1.68 ± 0.36	15.86 b±3.54	4.87±0.38	2.60 b±0.30	2.07±0.33
Right testis	$10.48 A \pm 4.68$	4.07 A±0.85	2.17 a±0.45	1.71 ± 0.36	$14.94 B\pm 3.39$	4.77 B±0.32	2.52 b±0.43	1.92 ± 0.22
Left wattle	I	5.79 ± 0.77	4.13 ± 0.97	I	I	6.24 ± 0.48	$4.14{\pm}0.68$	I
Right wattle	I	5.65 ± 1.04	4.09 ± 0.91	I	I	6.34 ± 0.50	4.47 ± 0.66	I
Wattles together	10.66 ± 4.43	I	I	I	12.84 ± 3.17	I	I	I
Comb	14.21 ± 7.07	6.82±1.25	I	I	16.55 ± 6.43	7.21±1.22	I	I
Live body weight	4213.37 A±216.20	I	I	I	3032.33 B±312.55	I	I	I
		Hubbard Fle	2X			Hubbard F1	15	
Items	weight	length	width	thickness	weight	length	width	thickness
	(g)	(cm)	(cm)	(cm)	(g)	(cm)	(cm)	(cm)
Left testis	17.17 ± 5.98	4.88 ± 0.75	2.87±0.45	2.02 ± 0.32	18.28 ± 8.00	4.86 ± 0.99	3.05 ± 0.60	2.13 ± 0.41
Right testis	16.98 ± 5.91	4.81 ± 0.63	2.87±0.42	2.01 ± 0.32	18.00 ± 9.14	4.80 ± 0.91	3.09±0.78	2.13 ± 0.48
Left wattle	Ι	6.30 ± 1.06	4.65 ± 1.10	I	Ι	6.40 ± 1.07	4.94 ± 0.98	Ι
Right wattle	I	6.26 ± 1.67	4.88 ± 1.60	I	Ι	6.25 ± 0.95	4.76±1.18	Ι
Wattles together	13.67 ± 6.34	I	I	I	14.86 ± 5.20	I	I	I
Comb	35.85 A*±13.40	11.84 a±1.65	6.80 ± 1.13	I	51.73 B±17.50	13.05 b±1.18	6.91 ± 0.91	I
Live body weight	5002.67±549.88	I	I	I	4852.67±543.96	Ι	I	I
*Average values	in lines related to the same	e analvsed characteri	stics with different	t letters differ sig	nificantly (A, $B - P \le 0.0$	11; a, b − P≤0.05).		

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Table 3. The values of relative testes weight (%) for four chicken lines (n=15; means; \pm SD)

Items	Hubbard F15	Hubbard Flex	ISA Brown	Lohmann Brown
Relative testes weight	0.74 Aa*±0.31	0.69 Aa±0.25	0.52 Ab±0.23	1.02 B±0.20

*Average values in lines with different letters differ significantly (A, $B - P \le 0.01$; a, $b - P \le 0.05$).

Table 4. Correlation between testes weight (g) and selected measurements (head ornaments and live body weight) for four chicken lines (n=15)

Chicken line	Comb	Comb	Comb	Wattles	Wattles	Live
	weight	length	width	weight	length	body weight
Hubbard F15	r=0.85*	r=0.78*	r=0.57*	r=0.85*	r=0.78*	r=0.45
	p=0.01	p=0.00	p=0.03	p=0.00	p=0.00	p=0.09
Hubbard Flex	r=0.05	r=-0.08	r=0.04	r=0.01	r=0.21	r=0.02
	p=0.86	p=0.77	p=0.87	p=0.97	p=0.46	p=0.60
ISA Brown	_	_	_	r=0.39	r=0.07	r=-0.17
				p=0.15	p=0.35	p=0.54
Lohmann Brown	_	_	_	r=-0.08	r=-0.28	r=0.40
				p=0.78	p=0.32	p=0.14

*Values statistically significant (P≤0.05).

Table 5. Average values of the degree of fluctuating asymmetry (FA) between the left and right bilateral traits of four chicken lines (n=15; means; ± SD)

Items	Hubbard F15	Hubbard Flex	ISA Brown	Lohmann Brown
Testes weight	0.09±0.10	0.06±0.05	0.08 ± 0.04	0.06±0.05
Testes length	0.05 ± 0.05	$0.04{\pm}0.03$	0.05 ± 0.03	0.02 ± 0.02
Testes width	0.05±0.04	0.03 ± 0.02	0.03 ± 0.03	0.05 ± 0.06
Testes thickness	0.04 ± 0.02	0.03 ± 0.02	0.03 ± 0.02	0.05 ± 0.05
Wattles length	0.03a*±0.02	0.06b±0.06	0.04 ab±0.03	0.03 a±0.02
Wattles width	0.06±0.05	0.04±0.03	0.05 ± 0.06	0.06±0.06

*Average values in lines with different letters differ significantly (a, $b - P \le 0.05$).

The mean values of fluctuating asymmetry of bilateral traits characteristics are shown in Table 5. The only difference in FA measurements was found for the wattle length. It was significantly (P \leq 0.05) higher for Hubbard Flex line than for Hubbard F15 and Lohmann Brown lines. The other FA measurements did not differ between the lines.

Discussion

The main result of this study was strong, positive correlation between testes size and size of roosters head ornaments only in meat type line Hubbard F15. Similar results, i.e. a positive relationship between comb area and testes weight, was reported in broiler chickens (McGary et al., 2003 b) and in house sparrow (positive relationship between badge size and testes size) (Møller and Erritzoe, 1988). In contrast, we observed no such correlations for both examined laying lines. Similarly, no such correlations were found for Chinese quail (Uller et al., 2005). However, our observations confirm the results of Vizcarra (2010) and Denk (2006) that there is no correlation between body mass and testes mass, but at the same time are contrary to Pitcher et al. (2005), Garamszegi et al. (2005) and Moller and Pomiankowski (1993) data.

Head ornaments, as they are related to testes mass, and because females tend to choose males by their secondary sexual traits, may also be correlated with sperm characteristics and fertilizing potency. While comb size was negatively correlated with percentage of viable sperm in broiler roosters, there was a positive relationship between comb colour and sperm viability (Navara et al., 2012). However, there was no correlation between comb size and sperm concentration. Other research indicated that broiler breeder males with larger combs had higher frequency of mating, but at the same time lower sperm concentration (Bilcik et al., 2005). Bilcik and Estevez (2005) showed that wattle width was correlated with sperm motility, but they did not find any correlation between morphological traits and male reproductive success. Wattle length and comb width were positively correlated with fertility and sperm penetration through the perivitelline layer in broiler breeders (McGary et al., 2003 a). Similar results showed McGary et al. (2002) for comb area and for testes weight, which were positively correlated with flock fertility. Contrary to above, Prieto et al. (2011) showed that there is no correlation between wattle width and sperm quality variables, and Forkmann and Corr (1996) found that hens mated with roosters with larger wattles laid fewer fertilized eggs. On the other hand, Pizzari et al. (2004) did not find a correlation between comb size and sperm quality in crosses between red jungle and domesticated fowl.

In the presented experiment the sperm quality was not evaluated, as in commercial breeder farms before the onset of reproduction the males are selected only on the basis of body weight and external appearance, including head ornaments development. Therefore, we focused mainly on correlations between head ornaments and testis size, since according to many researchers (Brougher et al., 2005; McGary et al., 2002), the latter is strongly correlated with the reproductive success. Body weight may induce fertility problems, as selection for increased body weight reduced males physical ability to copulate successfully, which was observed especially in turkey industry (Kondra and Shoffner, 1955). Also in broiler breeder flocks the heavier males had lower mating frequency, higher frequency of mating without cloacal contact of female and lower sperm motility (Bilcik et al., 2005). Furthermore, Uller et al. (2005) described that sexual ornaments and testes mass are related to size of eggs laid by parental females. Hens tended to lay larger eggs, but not more eggs, when paired with males with large ornaments and larger testes.

Lack of correlation between combs and testes in both laying lines seems to be obvious, since the combs were dubbed, but the same results were noted also for one of the broiler breeder lines (Hubbard Flex). Moreover, in all lines mentioned above, the lack of correlation between wattles and testes was also stated. The exception in the examined chicken lines was Hubbard F15. The obtained results might suggest that rooster selection for the reproduction flock only by the assessment of the head ornaments, body weight and general male appearance (lack of evident body disorders) is not sufficient, and cannot be the only criteria. This gains particular importance in

broiler breeders, where satisfactory level of fertility becomes a serious problem (Mc-Gary et al., 2003 b). Bilateral ornamental traits are not always the same size (Kimball et al., 1997). Asymmetry of bilateral ornaments did not affect red jungle fowl female mate choice (Ligon et al., 1998). Testes tended to be asymmetric, but this was not correlated to comb or body size in red jungle fowl (Kimball et al., 1997). Asymmetry of roosters' wattles was positively correlated with the number of eggs laid by laying hens (Forkman and Corr, 1996). Moreover, Prieto et al. (2011) did not find a correlation between fluctuating asymmetry and sperm quality variables. Secondary sexual trait asymmetry may be correlated with its size. Larger ornaments tend to be more symmetric. There existed a negative relationship between tail length asymmetry and tail length in swallows (Møller, 1990). Similar results, the negative relationship between the degree of fluctuating asymmetry and ornament size, were noted by Møller and Pomiankowski (1993), but only in taxa with single, not multiple ornaments.

Secondary sexual ornaments may also be connected with immune system in birds. Male barn swallows with larger secondary sexual traits (longer tail) tended to have better immune defence (Saino et al., 1995). Although male house sparrows with larger badges had smaller bursa of Fabricius, which is involved in immune defence (Møller et al., 1996).

Differences in testes size may also depend on ecological and behavioural factors. Pitcher et al. (2005) showed that species that breed colonially and taxa that do not participate in incubation or feeding offspring, had larger testes. Monogamous taxa and nonmigratory avian species had smaller testes and its size increased with clutch size.

In summary, our results indicate that, since in majority of the evaluated lines, the correlations between head ornaments and testes were not stated the secondary sexual traits cannot be used as the predictors of testicular size, and thus probably also of males' reproductive ability.

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Conflict of interest

None of the authors have any conflict of interest to declare.

References

Balthazart J., Hirschberg D. (1979). Testosterone metabolism and sexual behavior in the chick. Horm. Behav., 12: 253–263.

- Bilcik B., Estevez I. (2005). Impact of male-male competition and morphological traits on mating strategies and reproductive success in broiler breeders. Appl. Anim. Behav. Sci., 92: 307–323.
- Bilcik B., Estevez I., Russek-Cohen E. (2005). Reproductive success of broiler breeders in natural mating systems: The effect of male-male competition, sperm quality, and morphological characteristics. Poult. Sci., 84: 1453–1462.

- Brougher S.M., Estevez I., Ottinger M.A. (2005). Can testosterone and corticosterone predict the rate of display of male sexual behaviour, development of secondary sexual characters and fertility potential in primary broiler breeders? Brit. Poultry Sci., 46: 621–625.
- Chen K.L., Tsay S.M., Chiou P.W., Chen T.W., Weng B.C. (2009). Effects of caponization and testosterone implantation on immunity in male chickens. Poult. Sci., 88: 1832–1837.
- Chiasson R.B., Carr B.L. (1985). The effects of castration and/or methimazole feeding on the pituitary-response to temperature extremes by cockerels. Gen. Comp. Endocr., 60: 427–433.
- D e n k A.G., K e m p e n a e r s B. (2006). Testosterone and testes size in mallards (*Anas platyrhynchos*). J. Ornithol., 147: 436–440.
- For k m a n B., Corr S. (1996). Influence of size and asymmetry of sexual characters in the rooster and hen on the number of eggs laid. Appl. Anim. Behav. Sci., 49: 285–291.
- Garamszegi L.Z., Eens M., Hurtrez-Bousses S., Moller A.P. (2005). Testosterone, testes size, and mating success in birds: a comparative study. Horm. Behav., 47: 389–409.
- Johnsen T.S., Zuk M. (1996). Repeatability of mate choice in female red jungle fowl. Behav. Ecol., 7: 243–246.
- K i m b a 11 R.T., L i g o n J.D., M e r o l a Z w a r t j e s M. (1997). Testicular asymmetry and secondary sexual characters in red jungle fowl. Auk, 114: 221–228.
- Kondra P.A., Shoffner R.N. (1955). Heritability of some body measurements and reproductive characters in turkeys. Poult. Sci., 34: 1262–1267.
- Ligon J.D., Thornhill R., Zuk M., Johnson K. (1990). Male competition, ornamentation and the role of testosterone in sexual selection in red jungle fowl. Anim. Behav., 40: 367–373.
- Ligon J.D., Kimball R., Merola-Zwartjes M. (1998). Mate choice by female red jungle fowl: the issues of multiple ornaments and fluctuating asymmetry. Anim. Behav., 55: 41–50.
- M c G a r y S., E s t e v e z I., B a k s t M.R., P ollock D.L. (2002). Phenotypic traits as reliable indicators of fertility in male broiler breeders. Poult. Sci., 81: 102–111.
- M c G a r y S., E s t e v e z I., B a k s t M.R. (2003 a). Potential relationships between physical traits and male broiler breeder fertility. Poult. Sci., 82: 328–337.
- M c G a r y S., E s t e v e z I., R u s s e k C o h e n E. (2003b). Reproductive and aggressive behavior in male broiler breeders with varying fertility levels. Appl. Anim. Behav. Sci., 82: 29–44.
- Møller A.P. (1990). Fluctuating asymmetry in male sexual ornaments may reliably reveal male quality. Anim. Behav., 40: 1185–1187.
- Møller A.P. (1994). Directional selection on directional asymmetry: testes size and secondary sexual characters in birds. Proc. R. Soc. Lond. B, 258: 147–151.
- Møller A.P., Erritzoe J. (1988). Badge, body and testes size in House Sparrows *Passer domesticus*. Ornis Scand., 19: 72–73.
- Møller A.P., Pomiankowski A. (1993). Why have birds got multiple sexual ornaments. Behav. Ecol. Sociobiol., 32: 167–176.
- Møller A.P., Swaddle J.P. (1997). Asymmetry, developmental stability, and evolution. Oxford University Press, New York, NY.
- Møller A.P., Kimball R.T., Erritzoe J. (1996). Sexual ornamentation, condition, and immune defense in the house sparrow *Passer domesticus*. Behav. Ecol. Sociobiol., 39: 317–322.
- N a v a r a K.J., A n d e r s o n E.M., E d w a r d s M.L. (2012). Comb size and color relate to sperm quality: a test of the phenotype-linked fertility hypothesis. Behav. Ecol., 23: 1036–1041.
- Pitcher T.E., Dunn P.O., Whittingham L.A. (2005). Sperm competition and the evolution of testes size in birds. J. Evolut. Biol., 18: 557–567.
- Pizzari T., Jensen P., Cornwallis C.K. (2004). A novel test of the phenotype-linked fertility hypothesis reveals independent components of fertility. Proc. R. Soc. B Biol Sci., 271: 51–58.
- Prieto M.T., Campo J.L., Santiago-Moreno J. (2011). Relationship among fluctuating asymmetry, morphological traits, and sperm quality in layers. Poult. Sci., 90: 2845–54.
- Rintamaki P.T., Hoglund J., Karvonen E., Alatalo R.V., Bjorklund N., Lundberg A., Ratti O., Vouti J. (2000). Combs and sexual selection in black grouse (*Tetrao tetrix*). Behav. Ecol., 11: 465–471.
- Saino N., Møller A.P., Bolzern A.M. (1995). Testosterone effects on the immune system and parasite infestations in the barn swallow (*Hirundo rustica*): An experimental test of the immuno-competence hypothesis. Behav. Ecol., 6: 397–404.

- Uller T., Eklof J., Andersson S. (2005). Female egg investment in relation to male sexual traits and the potential for transgenerational effects in sexual selection. Behav. Ecol. Sociobiol., 57: 584–590.
- Vizcarra J.A., Kirby J.D., Kreider D.L. (2010). Testis development and gonadotropin secretion in broiler breeder males. Poult. Sci., 89: 328–334.
- Z u k M., J o h n s e n T.S., M a c l a r t y T. (1995). Endocrine-immune interactions, ornaments and mate choice in red jungle fowl. Proc. R. Soc. B Biol. Sci., 260: 205–210.
- Zuk M., Thornhill R., Ligon J.D., Johnson K., Austad S., Ligon S.H., Thornhill N.W., Costin C. (1990). The role of male ornaments and courtship behavior in female mate choice of red jungle fowl. Am. Nat., 136: 459–473.

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