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Fluctuating asymmetry in red junglefowl

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Abstract

Fluctuating asymmetry, the random deviation from perfect bilateral symmetry, has recently attracted considerable attention. Levels of asymmetry have been shown to correlate with measures of individual quality. We measured asymmetry in a variety of ornamental and non-ornamental traits in red junglefowl, *Gallus gallus* and examined the patterns of asymmetry among different traits within an individual. All ornamental traits had significantly higher levels of fluctuating asymmetry than did non-ornamental traits. However, inter-trait correlations of asymmetry were low for both ornamental and non-ornamental traits. We then correlated measures of asymmetry with several potential indicators of male quality, including comb size, body size, and body condition. We found little evidence that asymmetry in any measured trait reflected male quality. We measured asymmetry in ornamental traits at several stages of development and found no relationship between male condition and changes in asymmetry over time. Our results indicate that it is necessary to employ caution when choosing traits to be measured in studies of fluctuating asymmetry and that a relationship between asymmetry and individual quality cannot be assumed.

Introduction

Fluctuating asymmetry (FA) is a population parameter which measures the random deviation from perfect symmetry in bilaterally symmetrical traits. Since the same genome directs development of both the right and left sides of the body, deviations from perfect symmetry are thought to arise from an inability of

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individuals to compensate for environmental and genetic stress during development (for reviews, see Palmer and Strobeck, 1986; Parsons, 1990, 1992). This implies that levels of asymmetry between traits within an individual should be correlated. However, many studies have found this not to be the case (e.g., Van Valen, 1962; Mason et al., 1967; Soulé, 1967; Soulé and Baker, 1968; Patterson and Patton, 1990; Leamy, 1992; Evans et al., 1995; Ryan et al., 1995), suggesting that most traits within an individual are subjected to different levels and types of stress during development.

Since some individuals are better able to develop symmetrically in the face of developmental stresses than other individuals, asymmetry can be used to measure individual genetic quality within a population. Studies of sexual selection have utilized asymmetry as an indicator of individual quality, emphasizing FA in ornamental, presumably sexually selected, traits. High quality individuals should be better able to control development and hence are predicted to have relatively low levels of asymmetry, suggesting that sexual selection by females should result in the choice of relatively symmetric mates (Møller and Höglund, 1991; Møller, 1993a; Møller and Pomiankowski, 1993). Several studies have demonstrated sexual selection for individuals with relatively greater symmetry (e.g., Møller, 1992a, 1993b; Swaddle and Cuthill, 1994a, b; Kimball, 1995). However, not all studies have found a relationship between asymmetry in ornamental traits and sexual selection (Møller 1993a; Ligon et al., in press).

Møller and co-authors (Møller and Höglund, 1991; Møller, 1992b, 1993a; Møller and Pomiankowski, 1993) have suggested that male secondary sexual traits which are under directional selection and which reliably reflect male quality will show a negative relationship between the degree of FA and trait size. High quality males will both develop large ornamental traits and relatively symmetrical bilateral traits. Low quality males, in contrast, will produce smaller ornaments and will show higher levels of asymmetry in bilateral traits. Traits known to be under directional sexual selection have been shown to exhibit a negative relationship between trait size and trait FA, while traits not under directional selection exhibit a flat, U-shaped, or positive relationship (Møller and Höglund, 1991; Møller, 1993a; Møller and Pomiankowski, 1993). Therefore, only traits in which there is a negative relationship between trait size and trait asymmetry is asymmetry likely to reflect individual quality.

We measured the degree of FA in several ornamental and non-ornamental traits in male red junglefowl. This allowed us to determine 1) whether asymmetries of different traits within an individual are correlated, and 2) whether ornamental traits have greater asymmetry than non-ornamental traits. Several different traits were measured to estimate male quality, including body size and condition, comb length, and the size of several internal organs such as testis size (Møller, 1988) and spleen size (Møller and Saino, 1994; Møller et al., in press). These data were used to determine 3) whether FA reflects male quality, and 4) whether males in good condition become more symmetrical during development.

Methods

Study species

The red junglefowl is the conspecific ancestor of the domestic fowl (Hutt, 1949; Stevens, 1991). Like many other pheasants, males are elaborately ornamented, with colorful orange, red, and black plumage, and a fleshy red comb, ears, and wattles on the head. The plumage of females is drab and cryptic and the fleshy traits are smaller. Red junglefowl are polygynous and males typically provide no parental care.

Previous studies indicate that mate choice by female red junglefowl is primarily correlated with the color and size of the comb (Zuk et al., 1990a,b, 1995; Ligon and Zwartjes, 1995). These studies have not found a clear or strong preference by females for aspects of male plumage, particularly once the effect of comb size was controlled (e.g., Ligon and Zwartjes, 1995). Comb size and wattle size are known to directly reflect current testosterone levels (Domm, 1939; Collias, 1943; Zuk et al., 1995), and thus are condition-dependent traits (Folstad and Karter, 1992).

The red junglefowl used in this study were descendants of 150 birds obtained from the San Diego Zoo in 1985–1986. The zoo population formed a large, free-ranging flock descended from about 30 individuals of two sub-species which were released on the zoo grounds in 1942 (Collias et al., 1966; Collias and Collias, 1985). All junglefowl used in this study were hatched during a one month period between mid-May and mid-June, 1994. Chicks were fed *ad libitum* and, at about six weeks of age, were removed from the brooders and allowed to free range. In December and January, when males had matured, they were placed into individual pens, where they were provided with fresh water and food *ad libitum*.

Measurement of traits

To examine what factors might affect changes in the levels of asymmetry in a trait, measurements were collected from live males at three different time periods. The first measurements were collected in November 1994, when the birds were about six months of age. At this time, skeletal development was complete and the ornamental wattles and combs were just beginning to enlarge. The second measurement period occurred in January 1995 when the birds were eight months old. Combs and wattles were much larger than in November and the birds had gained weight. The final measurement period was in March 1995 when the roosters were 10 months of age. Although body weights had remained similar, combs and wattles had continued to enlarge (see also Zuk et al., 1990a). In addition, the spurs, used in male-male competition (Ligon et al., 1990), had begun to develop.

During each measurement period we measured the following: 1) weight to the nearest 7 grams; 2) comb length, to the nearest 0.1 mm using dial calipers; 3) wing chord (unflattened) on each side of the body; and 4) tarsus on each side of the

body. In March, we measured two additional bilateral traits: spur length and length of the longest tail feathers (for those males whose longest tail feathers were intact on both sides of the body). To insure accurate and reliable measurement of the spurs, we measured the length of the spur and the width of the tarsus together, as has been done in other studies (e.g., Grahn and von Schantz, 1994).

Three color photographs, a frontal view and both the right and left profiles, were taken of each male during each measurement period. The photographs were digitized into a computer at 150 dpi. Measurements were then collected from these photographs using NIH Image (developed at the U.S. National Institutes of Health, available from the Internet by anonymous FTP from zippy.nimh.nih.gov). Wattle and ear lappet areas were measured by outlining the trait on the profile photographs. Wattle length on each side was obtained from the frontal photograph, measuring from the posterior corner of the mandibles to the bottom tip of the wattle. The measurements were converted into mm or mm^2, based upon a ruler included in each photograph.

Sixteen males were sacrificed after measurements were completed in March 1995. An additional 19 males were sacrificed in early June 1995, at about one year of age. The tarsus, tibia, femur, and humerus from both sides of the body were carefully removed so as to not damage the ends of the bones. After processing, the length of each bone was measured. Two or three capillary tubes of blood collected at the time of death were centrifuged to determine the hematocrit levels (percentage of packed red blood cells). In addition, the number of nits (lice eggs) around the cloaca was scored, with zero representing males with no nits, and 3 representing males heavily covered with nits.

Several internal organs were removed and measured. The right and left testis were removed and individually weighed to the nearest 0.1 g on a digital scale. The heart and liver also were removed, blotted dry, and weighed. Maximum length and width of the spleen was measured using dial calipers and used to compute spleen volume using the equation for an ellipsoid $[(4/3)\pi(\text{width}^2)(\text{length})]$.

All bilateral traits were measured three times on each side of the body to allow determination of measurement error (Palmer and Strobeck, 1986; Swaddle et al., 1994). For wattles and ear lappets, replicate measurements were collected from the same photograph.

Means and standard deviations for the measured traits can be found in Table 1 (bilateral traits) and Table 2 (quality indicators). Since body size showed a high and significant correlation with several traits (Kimball et al., unpubl. data), we statistically controlled for body size by performing partial correlations in analysis involving male quality indicators. Results from partial correlations were the same as those where body size was not controlled, so those results are not included.

Data analysis

To determine the fluctuating asymmetry (FA) of all bilateral traits investigated, the three values for each side were averaged. In this paper, the term FA refers to

Table 1. Means and standard deviations for bilateral traits.

Trait	*n*	*x* (std)
Ornamental traits		
Nov. wattle length (mm)	42	23.2 (3.6)
Jan. wattle length (mm)	39	28.5 (4.3)
Mar. wattle length (mm)	39	32.0 (4.2)
Nov. wattle area (mm^2)	42	348.8 (87.8)
Jan. wattle area (mm^2)	39	559.5 (103.9)
Mar. wattle area (mm^2)	39	641.0 (96.5)
Nov. ear lappet area (mm^2)	42	190.4 (37.8)
Jan. ear lappet area (mm^2)	39	253.8 (37.9)
Mar. ear lappet area (mm^2)	39	266.1 (35.1)
Mar. spur length (mm)	39	19.6 (2.6)
Jun. spur length (mm)	23	22.6 (4.0)
Mar. tail length (cm)	15	37.8 (5.4)
Non-ornamental traits		
Humerus (mm)	37	72.5 (2.3)
Tarsus (mm)	37	83.1 (3.4)
Tibia (mm)	37	113.5 (3.0)
Femur (mm)	37	74.6 (2.5)
Wing chord (cm)	39	23.0 (0.4)

the absolute value of the left minus the right side (unsigned FA). Signed FA refers to the value of the left minus the right side. Since there were no high or significant relationships between trait size and trait FA, we only corrected for trait size in analyses comparing levels of FA between different traits. Relative FA refers to the absolute FA divided by the average length of the trait [FA/((right + left)/2)]. Average FA is the mean FA value for all traits within an individual, except

Table 2. Means and standard deviations for potential quality indicators.

Trait	*n*	*x* (std)
Comb length, Nov. (mm)	43	56.3 (8.7)
Comb length, Jan. (mm)	40	71.3 (5.2)
Comb length, Mar. (mm)	39	75.6 (5.1)
Body condition, Nov. (g/mm)	43	17.8 (1.4)
Body condition, Jan. (g/mm)	40	19.6 (1.4)
Body condition, Mar. (g/mm)	39	20.0 (1.3)
Body size index	37	4.0 (0.1)
Spleen volume (ml)	34	8521 (1856)
Avg. testis wt., Mar. (g)	16	6.4 (1.2)
Avg. testis wt., Jun. (g)	19	6.5 (1.1)
Nit loads	35	1.5 (1.1)
Heart wt. (g)	35	7.4 (0.8)
Liver wt. (g)	35	17.4 (2.4)
Hematocrit (%)	35	38.1 (8.3)

tail length and spur length in June, since samples sizes were smaller for these variables.

We analyzed the data by examining correlations among pairs of traits. This, however, only allows comparison between two traits. Therefore, we also determined whether individuals showed an overall similar level of asymmetry in multiple measured traits by calculating Kendall's coefficient of concordance (Zar, 1984) to determine whether FA's within a suite of traits which develop simultaneously showed a concordant pattern of asymmetry. A principal components analysis (PCA) of the FA data did not yield any additional information, hence the results are not reported.

To compare levels of FA in different traits, we employed two different methods. First, we used an F-test to examine the variance in signed FA, as greater levels of FA exhibit greater variance. Since larger traits can have larger FA, we controlled for trait size by dividing the signed FA by average trait size. We also addressed this problem by use of a paired t-test, again controlling for trait size by dividing absolute FA by average trait size to obtain relative FA. This indicated whether ornamental traits had consistently larger FA within an individual.

To determine trait size for bilateral traits, an average was taken from the three measurements on each side of the body (e.g., three measurements of the left femur plus three of the right). A body size index was calculated by adding together the standardized trait size of the tarsus, tibia, femur, and humerus. Body condition was calculated by dividing weight by tarsus length.

Although average testis size did not differ between March and June, some aspects of the testes differed between those birds sacrificed in March and those in June (Kimball et al., 1997). Therefore, testis size was analyzed separately for the March and June samples. For all other traits, March and June samples were combined.

Non-parametric tests were used in all analyses involving absolute FA, as absolute FA has a half-normal distribution. In addition, comparisons involving the ranked nit loads were analyzed with non-parametric tests. All other analyses involved parametric tests. Analyses were performed on SAS for the PC, ver. 6.4. Power analysis was performed using GPOWER (Faul and Erdfelder, 1992).

To prevent committing a type I error, only those results significant at $p \leq 0.05$ after performing a sequential Bonferroni adjustment (Holm, 1979) were considered significant. In Tables 4 and 5, sequential Bonferroni adjustments were performed separately for each column. We have indicated which results are significant at $p \leq 0.05$ before a sequential Bonferroni adjustment was performed, since a sequential Bonferroni increases the likelihood of committing a type II error.

Results

Do the measured traits exhibit FA?

We examined the statistical properties of the measured traits to determine whether we had measured FA. Descriptive statistics for FA measures are presented

Table 3. Means and standard deviations for absolute FA (|L-R|) and signed FA (L-R).

	|L-R|	(L-R)			Mixed-model ANOVA
	x (std)	x (std)	Skew	Kurtosis	F (df)†
Ornamental tratis					
Nov. wat. length (mm)	1.15 (1.11)	−0.04 (1.61)	−0.84	1.69	21.9 (40, 160)**
Jan. wat. length (mm)	1.77 (1.34)	−0.22 (2.23)	−0.08	−0.24	33.2 (38, 152)**
Mar. wat. length (mm)	1.94 (1.42)	−0.19 (2.40)	0.27	−0.14	69.9 (38, 152)**
Nov. wat. area (mm^2)	39.29 (35.9)	−6.23 (53.2)	−0.48	0.40	30.5 (40, 160)**
Jan. wat. area (mm^2)	78.53 (53.73)	−11.81 (95.2)	−0.24	−0.46	37.0 (38, 152)**
Mar. wat. area (mm^2)	83.88 (65.85)	−53.41 (92.9)**	0.22	0.69	78.5 (38, 152)**
Nov. ear lappet area (mm^2)	39.93 (28.00)	21.06 (44.3)*	−0.28	−0.22	29.4 (40, 160)**
Jan. ear lappet area (mm^2)	45.83 (38.47)	27.44 (53.5)**	−0.12	0.34	29.9 (38, 152)**
Mar. ear lappet area (mm^2)	33.39 (27.88)	−4.60 (43.6)	0.32	0.82	34.4 (38, 152)**
Mar. spur length (mm)	0.55 (0.51)	0.09 (0.75)	0.10	1.10	34.6 (38, 152)**
Jun. spur length (mm)	0.78 (0.52)	0.23 (0.92)	0.01	−0.59	67.2 (22, 88)**
Tail length (cm)	0.92 (0.87)	−0.08 (1.29)	−0.96	1.32	14.1 (14, 56)**
Non-ornamental traits					
Humerus (mm)	0.26 (0.19)	−0.06 (0.32)	0.81	1.22	21.4 (36, 144)**
Tarsus (mm)	0.36 (0.26)	−0.08 (0.44)	−0.58	−0.36	19.1 (36, 144)**
Tibia (mm)	0.42 (0.33)	0.18 (0.51)	−0.01	−0.33	15.3 (36, 144)**
Femur (mm)	0.34 (0.28)	0.10 (0.44)	0.48	−0.16	31.4 (36, 144)**
Wing chord (cm)	0.16 (0.16)	0.00 (0.22)	−0.29	0.92	2.0 (38, 152)*

† Calculated as outlined by Swaddle et al. (1994).
* Significant at $p \leq 0.05$ before performing a sequential Bonferroni.
** Significant at $p \leq 0.05$ after performing a sequential Bonferroni.

in Table 3. Following the recommendations of Palmer and Strobeck (1992), we have included the mean and standard deviation for the absolute value of FA, the signed FA, and the skewness and kurtosis for signed FA. Statistically, signed FA should be normally distributed with a mean of zero. All traits were normally distributed, as measured by a Shapiro-Wilks test. However, ear lappet area in November and January and wattle area in March showed means for signed FA which differed significantly from zero (Tab. 3). Since removal of these three measurements did not alter the conclusions, these three traits have been included for completeness, even though they do not fit the strict definition of FA (see also Ryan et al., 1995).

Since differences between the right and left side often are generally small, even small levels of measurement error can account for much of the variation in FA. We used a mixed-models ANOVA to determine whether measurement error was large, relative to FA, following the procedure suggested by Swaddle et al. (1994). The F-value calculated using the method of Swaddle et al. (1994) is shown in Table 3, and indicates that measurement error was low relative to asymmetry levels for all traits examined here. Only tarsus lengths of live birds had a high level of measurement error (data not included). Measures of tarsus length used in the analyses here are from the bones of sacrificed birds.

FA within an individual

Individual correlations between traits indicate that there are no high or significant correlations between the asymmetry of different traits (range of r_s: -0.38 to 0.40; adjusted $p \gg 0.10$; original $p \geq 0.01$). The statistical power of most of the analyses was low, though the statistical power of several test approached 0.80 (Cohen, 1969). Supporting the non-significant correlations between traits, a concordance test indicated that there was no overall concordance within an individual ($X^2 = 44.06$, df $= 34$, $p = 0.116$) when using the FA of the skeleton traits, and the fleshy traits, wing chord, and spur length measured in March.

It has recently been suggested that inter-trait correlations might be greater if analyses only include traits which develop simultaneously (Watson and Thornhill, 1994), as these traits will be exposed to similar levels of stress. There were three suites of traits which should develop simultaneously (flesh traits, skeletal traits, and plumage traits). No suite of traits showed a significant concordance of FA, though results were significant prior to performing a Bonferroni correction (ornamental flesh traits in March: $X^2 = 58.14$, df $= 38$, $p = 0.019$; skeletal traits: $X^2 = 55.44$, df $= 36$, $p = 0.020$). The two plumage traits, wing chord and tail length, showed little correlation with each other ($r = -0.12$, $n = 15$, $p = 0.666$).

FA in ornamental versus non-ornamental traits

The ornamental traits examined included the fleshy traits (measured in March when development was greatest), spur length measured in June when development was greatest, and tail length. Measurements on the limbs (tarsus, tibia, femur, humerus, and wing chord) were classified as non-ornamental. Results testing each combination of one ornamental and one non-ornamental trait indicate that ornamental traits had greater FA in all 25 comparisons. This was true at both the population level, by use of an F-test on the variance of the signed FA, as well as within an individual, by use of a paired t-test on the relative FA's. All 25 pairwise comparisons were significant even after peforming a sequential Bonferroni on each set of analyses.

FA of male quality

We examined the relationship between trait size and trait FA for all bilaterally symmetrical traits measured. We compared FA to both average trait size, as done by Møller (e.g., 1993a), as well as with the maximum trait size (Evans and Hatchwell, 1993). We found no significant relationships, either positive or negative, when comparing FA with average trait size (Tab. 4). Higher correlations were found when comparing asymmetry to the maximum trait size, though only one is significant after performing a sequential Bonferroni (Tab. 4). The majority of comparisons are positive and do not exhibit the negative relationship predicted for

Table 4. Spearman correlation coefficients of trait FA with trait size and the deviation from the trait mean.

	n	Avg. size	Max. size	Deviation
Ornamental traits				
Mar. wattle length (mm)	39	0.15	0.30	0.05
Mar. wattle area (mm^2)	39	0.05	0.42*	−0.13
Mar. ear lappet area (mm^2)	39	0.22	0.46**	0.00
Mar. spur length (mm)	39	−0.22	−0.17	0.02
Jun. spur length (mm)	23	−0.34	−0.33	0.17
Tail length (cm)	15	0.36	0.40	−0.53*
Non-ornamental traits				
Humerus length (mm)	37	−0.13	−0.09	0.26
Tarsus length (mm)	37	0.17	0.20	0.26
Tibia length (mm)	37	0.08	0.15	−0.06
Femur length (mm)	37	−0.15	−0.09	0.09
Wing chord (mm)	39	0.06	0.25	−0.09

* Significant at $p \leq 0.05$ before performing a sequential Bonferroni.
** Significant at $p \leq 0.05$ after performing a sequential Bonferroni.

traits which indicate male quality. Spur length in June is the only trait which shows a negative relationship in the range predicted by Møller (1993a). The relationship was not significant, though the power of the test was low (0.38) and a larger sample size may have yielded a significant relationship. Regardless of the significance, the relationship may not be biologically relevant, as spurs were still at an early stage of development and the relationship between asymmetry and size may change as the spurs mature.

A U-shaped relationship between trait size and trait FA may occur when traits are under stabilizing selection. To assess possible stabilizing selection, we determined whether there was a positive relationship between the deviation of an individual's trait size from the population mean and trait asymmetry. We did not observe a significant relationship between the absolute value of the deviation from the population mean and trait asymmetry for any of the traits examined (Tab. 4). Many traits showed a negative relationship, contrary to what we expected if high quality individuals had trait sizes near the population mean.

Except for spleen size and nit loads, we would expect a negative relationship between possible indicators of male quality and levels of FA. There was only one significant relationship between a measure of male quality and a measure of trait asymmetry (Tab. 5). In addition, excluding the spleen and nit data, there are more positive correlations than negative (negative 23; positive 36). For spleen volume and nit loads, where a positive relationship would indicate that FA reflected male quality, the relationships are primarily negative (negative 16; positive 8). Averaging the FA's over many traits also did not yield any significant results.

Table 5. Spearman correlation coefficients of FA and quality indicators. Sample sizes in parentheses.

	Comb length	Body condition	Body size	Nit loads	Spleen volume	Testes weight	
Ornamental traits						March	June
Mar. wattle length	0.09 (39)	−0.33 (39)*	−0.26 (35)	0.12 (35)	−0.09 (34)	−0.35 (16)	−0.12 (19)
Mar. wattle area	0.13 (39)	0.05 (39)	0.26 (35)	0.02 (35)	0.06 (34)	−0.12 (16)	0.02 (19)
Mar. ear lappet area	−0.08 (39)	−0.01 (39)	0.00 (35)	−0.05 (35)	−0.04 (34)	−0.57 (16)*	−0.13 (19)
Mar. spur length	0.06 (39)	−0.21 (39)	−0.16 (35)	−0.03 (35)	−0.07 (34)	−0.01 (16)	−0.14 (19)
Jun. spur length	0.32 (23)	0.03 (23)	0.24 (19)	0.03 (19)	−0.42 (18)	–	0.26 (19)
Tail length	−0.05 (15)	0.11 (15)	0.39 (12)	−0.23 (12)	0.40 (12)	0.72 (6)	−0.20 (6)
Non-ornamental traits							
Humerus length	0.13 (35)	0.27 (35)	−0.12 (37)	−0.23 (35)	0.00 (34)	0.01 (16)	−0.11 (19)
Tarsus length	0.11 (35)	0.23 (35)	0.11 (37)	−0.01 (35)	−0.11 (34)	0.01 (16)	0.18 (19)
Tibia length	0.30 (35)	0.24 (35)	0.10 (37)	0.48 (35)**	−0.03 (34)	−0.13 (16)	0.41 (19)
Femur length	0.12 (35)	−0.21 (35)	−0.43 (37)	−0.16 (35)	−0.16 (34)	−0.63 (16)*	0.11 (19)
Wing chord	0.14 (39)	0.14 (39)	0.17 (35)	−0.28 (35)	−0.08 (34)	−0.01 (16)	0.15 (19)
Average FA	0.09 (35)	0.05 (35)	0.19 (35)	−0.02 (35)	0.05 (34)	−0.19 (16)	0.07 (19)

* Significant at $p \leq 0.05$ before performing a sequential Bonferroni.
** Significant at $p \leq 0.05$ after performing a sequential Bonferroni.

Table 6. Spearman correlation coefficients of the change in trait FA (relative FA first measurement – relative FA at second measurement) with comb length and body condition during first measurement period, plus body size. Sample sizes are in parentheses.

	Comb length	Body condition	Body size
a. November to January			
Wattle length	−0.22 (39)	−0.14 (39)	0.10 (35)
Wattle area	−0.11 (39)	0.16 (39)	−0.07 (35)
Ear lappet area	−0.32 (39)	0.19 (39)	0.14 (35)
b. January to March			
Wattle length	−0.15 (39)	−0.02 (38)	0.01 (34)
Wattle area	−0.22 (39)	0.01 (38)	−0.21 (34)
Ear lappet area	−0.17 (39)	−0.10 (38)	−0.18 (34)
c. March to June			
Spur length	−0.04 (23)	−0.24 (23)	−0.17 (19)

The statistical power of most of these tests was low, and only exceeded 0.80 (Cohen, 1969) in two cases. Therefore, we cannot, with certainty, accept the null hypothesis.

There were no significant relationships involving hematocrit levels (range: $r_s = -0.33$ to 0.21, $p > 0.10$ before a Bonferroni adjustment), heart weight (range: $r_s = -0.35$ to 0.35, $p > 0.10$ before a Bonferroni adjustment), or liver weight (range: $r_s = -0.25$ to 0.34, $p > 0.06$ before a Bonferroni adjustment), therefore these data are not considered here.

The degree of deviation from the population mean also did not correlate with any of our measures of male quality (Kimball et al., unpubl. data).

Changes during development

We measured levels of asymmetry in the ornamental fleshy traits and the spurs at different stages of development. If symmetrical development is dependent upon individual quality or condition, we would expect that individuals in good condition would be better able to correct any deviations from perfect symmetry, and hence should show a reduction in asymmetry over time. Although individual asymmetry levels did change, both positively and negatively, during the development of the traits, there was no relationship between the change in asymmetry between measurement periods and comb length, body size, or condition at the time of the first measurement period (Tab. 6). In addition, the relationships are primarily negative whereas a positive relationship would be expected if individuals in good condition developed more symmetrically, as has been found in other studies (Nilsson, 1994; Swaddle and Witter, 1994).

Discussion

Patterns within individuals

Since all traits are under similar genetic control and should have been exposed to similar levels of stress during development, measured levels of FA among different traits within an individual might be expected to be positively correlated (but see Whitlock, 1996). However, we did not find this to be the case for either ornamental or non-ornamental traits, supporting the results of other studies (Van Valen, 1962; Mason et al., 1967; Soulé, 1967; Soulé and Baker, 1968; Patterson and Patton, 1990; Leamy, 1992; Evans et al., 1995). Restricting analyses to those traits which undergo development at the same time (Watson and Thornhill, 1994) increased the concordance, though results were still not significant. Overall, it appears that developmental control is primarily trait-specific, not whole-organism. This has important implications in studies of FA, since measurement of one or a few traits, as is commonly done, may lead to misleading results (Watson and Thornhill, 1994; Ryan et al., 1995).

Ornamental and non-ornamental traits

We found that ornamental traits had greater FA than non-ornamental traits, a pattern consistent with most other studies (Møller and Höglund, 1991; Møller, 1992b, 1994; but see Evans et al., 1995). Either natural selection may exert less pressure for symmetrical development in ornamental traits, or directional selection on ornamental traits may lead to reduced developmental control and hence greater asymmetry (Pomiankowski and Møller, 1995). Regardless of the previous directional selection on red junglefowl, neither wattles nor ear lappets are under current directional selection by female mate choice (Ligon et al., in press), making it unlikely that a recent history of directional selection has led to increased levels of asymmetry in these ornamental traits.

FA and male quality

Since asymmetry arises when an individual is unable to overcome genetic and environmental stresses, it has been suggested that levels of asymmetry reflect individual quality. A relationship has been found between levels of asymmetry and aspects of individual quality, such as reproductive success or survival (e.g., Palmer and Strobeck, 1986; Ueno, 1994; Hutchison and Cheverud, 1995). Asymmetry, particularly in secondary sexual traits, also has been shown to correlate with measures of sexual selection (e.g., Møller, 1992a, 1993b; Swaddle and Cuthill, 1994a; Kimball, 1995).

Previous studies which have examined the relationship between asymmetry and individual quality, particularly in the context of sexual selection, have examined

asymmetry in only one or a few traits. Since levels of asymmetry vary among different traits, the absence of a relationship between asymmetry and individual quality or sexual selection may be due to the choice of traits used in the study (Watson and Thornhill, 1994). To overcome this possible problem, we measured asymmetry in several ornamental and non-ornamental traits and compared them to a variety of non-bilaterally symmetrical traits which might reflect male quality. In spite of the large number of traits examined, we found virtually no evidence that asymmetry of bilaterally paired traits correlated with any of our measures of male quality. In particular, comb size, which reflects current testosterone levels (Zuk et al., 1995) and is a good predictor of both male fighting ability (Ligon et al., 1990) and male mating success (Ligon and Zwartjes, 1995; Ligon et al., in press), showed no significant negative relationships with any measure of asymmetry. Manning and Ockenden (1994) found that averaging the FA's of different traits was a better predictor of individual quality. However, we did not find this to be the case.

One way to determine which traits reliably reflect male quality is to focus on those bilateral traits, particularly ornamental traits, that exhibit a negative correlation between size and FA (Møller and Höglund, 1991; Møller, 1992b, 1993a; Møller and Pomiankowski, 1993). None of the traits we examined, with the possible exception of spur length measured in June, exhibited a negative relationship between trait size and trait FA. Other studies have also failed to find any relationship between trait size and trait FA in sexually selected traits (Evans et al., 1995; Tomkins and Simmons, 1995), indicating that the negative relationship described by Møller and co-authors either depends upon the species examined or on the recent history of selection on the trait in question.

Directional selection, such as often occurs in ornamental traits under sexual selection, may lead to reduced developmental control of the trait (Pomiankowski and Møller, 1995). Once directional selection has reduced trait-specific developmental control, levels of asymmetry in that trait may better reflect whole-organism developmental control (Watson and Thornhill, 1994). Therefore, traits under directional selection will be most likely to exhibit a relationship between FA and quality indicators. Many of the bilateral traits we measured appear to be under little or no current directional sexual selection (Zuk et al., 1990b; Ligon and Zwartjes, 1995; Ligon et al., in press). Thus, it is not surprising that, in this study, we found little relationship between asymmetry and male quality.

Asymmetry of non-ornamental traits also may reveal aspects of individual quality (e.g., Swaddle and Witter, 1994). For traits under stabilizing selection, there should be a positive relationship between the absolute value of the deviation from the population mean and individual asymmetry. Those individuals whose trait sizes are close to the population mean will be of higher quality, and hence should show lower levels of asymmetry. Using this criteria, none of the bilateral traits we examined appeared to reflect measures of male quality.

Changes during development

A relationship between asymmetry in feather traits and nutritional condition during molt has been found (Nilsson, 1994; Swaddle and Witter, 1994), demonstrating that individuals in good condition may develop more symmetrically. We measured several condition indicators during the development of the fleshy ornamental traits and the spurs and found no relationship between the changes in asymmetry levels and any measure of individual quality during the first measurement period, suggesting that those males in good condition were not better able to correct developmental asymmetries than were males in poor condition. However, since our study did not manipulate individual condition, (e.g., by reducing the quantity or quality of food to a subset of males), we cannot be certain that male condition has no effect on the symmetry of bilateral traits in red junglefowl.

General considerations

The ancestors of the red junglefowl used in this study were from a small population released at the San Diego Zoo in 1942. Therefore, these birds may have undergone genetic bottlenecks in their recent evolutionary history. In addition, individuals were raised in capitivity in relatively benign conditions. Both factors may have affected the results obtained here. We did observe variability in all morphological traits examined (Tabs. 1 and 2), though it was not possible to compare these levels of variability with wild individuals to determine if these birds exhibited lower levels of variation. Since all males were raised under the same conditions, the variability we did observe is probably due to genetic variability within the population.

The statistical power for many of these analyses was sufficient to detect many relationships. In fact, had our effect size (correlation coefficients) equalled those of studies which have found significant correlations between asymmetry and individual quality (e.g., Swaddle and Witter, 1994; Ueno, 1994), our sample sizes would have been large enough to detect a significant result if it were present. Furthermore, for the majority of analyses, the direction of the correlation was opposite that which we predicted, and therefore a larger sample size would not cause us to change our conclusions.

Our results indicate that it is important to be cautious when using FA as an indicator of quality in studies of other phenomena. We measured multiple bilateral traits as well as multiple indicators of male quality, and obtained little evidence that asymmetry was related to individual quality. Studies which focus on one or a few traits may be even less likely to observe a significant result between asymmetry and quality, even though such a relationship may exist. Alternatively, the relationships may exist, but may be too subtle to detect without using sample sizes that are much larger than are commonly employed. These results indicate that caution is necessary when choosing traits to measure in studies of FA, as well as in interpreting the biological significance of FA.

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