



Comparison of the tonic immobility duration, heterophil to lymphocyte ratio, and fluctuating asymmetry of chicks reared with or without a broody hen, and of broody and non-broody hens



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ARTICLE INFO

Article history:

Accepted 12 November 2013

Available online 20 November 2013

Keywords:

Broody hen

Chicks

Tonic immobility duration

Heterophil to lymphocyte ratio

Relative fluctuating asymmetry

ABSTRACT

The purposes of this study were to determine whether rearing with a broody hen influenced duration of tonic immobility, heterophil to lymphocyte ratio, and relative fluctuating asymmetry of chicks and to analyze the differences between broody and non-broody hens in fear and stress indicators. A total of 120 chicks (60 reared with a broody hen and 60 reared without a broody hen), and 48 hens (24 broody and 24 non-broody) were analyzed at 6 and 52 weeks of age, respectively. Broody hens were observed in 12 different Spanish breeds (Blue Andaluza, Black-barred Andaluza, Black Red Andaluza, Black Castellana, White-faced Spanish, Buff Prat, White Prat, Birchen Leonesa, Quail Castellana, Quail Silver Castellana, Red-barred Vasca, and Red Villafranguina), and all the chicks were from an F₁ cross between the Black-barred Andaluza and the Black Castellana breeds. Tonic immobility duration was significantly longer ($P < 0.0001$) in chicks reared without a broody hen than in those reared with a broody hen, suggesting that the presence of a broody hen reduces fear in chicks. There were no significant differences in the heterophil to lymphocyte ratio and the relative fluctuating asymmetry of leg length, leg width, and toe length, although the fluctuating asymmetry of wing length and the combined fluctuating asymmetry value of the 4 traits tended to be significant. These results suggest no effect on stress in chicks in the presence of a broody hen. Tonic immobility duration and heterophil to lymphocyte ratio were similar in broody and non-broody hens, indicating no association of broodiness with the fear and stress levels in hens. In conclusion, the presence of a broody hen during rearing has significant effects on fearfulness of chicks; rearing chicks with a mother being a method to reduce this major behavioral problem. However, broodiness was not associated with fear and stress indicators in hens.

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1. Introduction

In modern poultry husbandry, chicks are typically hatched in brooders and reared without a mother. Under natural conditions, young birds and mammals usually

remain with their parents for a long time. Unlike mammals, chicks can be reared without parents, and providing maternal care is not necessary for commercial practice. However, maternal care can have positive effects on the behavioral development of chicks, affecting behaviors such as aggressiveness, sociality, feather pecking, and activity. Fält (1978) reported that aggressive behavior was higher in the non-brooded chicks than in the brooded ones, and Perré et al. (2002) found that, compared with non-brooded pullets,

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brooded pullets interacted more agonistically when they encountered unfamiliar birds. Rodenburg et al. (2004) and Riber et al. (2007a) found that chicks reared with a mother hen present showed less severe feather pecking and cannibalism than those reared without a mother. Roden and Wechsler (1998), Wauters et al. (2002a,b), Jensen et al. (2006), and Riber et al. (2007b) reported that brooded chicks performed more feeding and general activities than non-brooded chicks; activity being more synchronized in chicks brooded by a broody hen (Jensen et al., 2006; Riber et al., 2007b).

It would be interesting to know if brooded and non-brooded chicks differ in their way to cope with a fear inducing situation or with a stressor. A few experiments have shown that brooded chicks observed in the presence of a mother hen were less fearful than non-brooded chicks. The effect of a broody hen on the duration of tonic immobility (TI), a well-understood defensive anti-predator reaction and a widely used measure of fearfulness in poultry (Forkman et al., 2007), of chicks has been analyzed only in one previous study by Perré et al. (2002). They did not find significant differences for TI or in two other fear measures (the open-field test and the novel object test). Roden and Wechsler (1998) found that chicks reared with a broody hen were less fearful (as indicated by their flight response when the observer stood up near of the pen) than those reared without a hen, flight responses being more frequent in chicks reared without a hen. Rodenburg et al. (2009) found that brooded chicks were more active in the open-field test indicating that they were less fearful. Shimmura et al. (2010) found that chicks' fearfulness, as measured by two behavioral tests (open-field and approach/avoidance), was decreased markedly by the presence of a broody hen.

On the other hand, it would be interesting to know if broody and non-broody hens differ in their fear and stress indicators. Broody hens are exposed to prolonged stress (fasting) associated with incubation, brooding, and rearing of chicks. They generally take only one-fifth of their normal food intake, and on some days they take no food at all, losing up to 20% of their body weight during this period (Wood-Gush, 1971). The broody hen will mutter and growl if disturbed and may even peck to defend their nest behaving in an aggressive way toward other hens (Kent, 1992). Increased plasma prolactin concentration is associated with broodiness (the broodiness hormone; Burke and Dennison, 1980; Zadworny et al., 1985; Sharp et al., 1988). Although prolactin is present in high levels during the incubation period, the care of the young period does not depend upon prolactin (Eisner, 1960; Buntin, 1996). Tactile stimuli, alone or in combination with visual and/or auditory stimuli from newly hatched chicks, are the only cues that induce the transition from the incubation behavior (egg-directed behavior) to the brooding behavior (chick-directed behavior; Richard-Yris et al., 1998). Several hormones related to stress, as the adrenaline, have been identified as mediators linked to prolactin release (Etches, 1996), and corticosterone (a stress hormone) has been shown to inhibit reproduction in birds (Sapolsky et al., 2000; Wingfield and Sapolsky, 2003). Both corticosterone and prolactin are very likely to mediate broodiness;

corticosterone governing the ultimate decision to reduce or suppress broodiness by inhibiting prolactin secretion (Angelier and Chastel, 2009).

The present study aimed to determine whether rearing with a broody hen influenced TI; heterophil to lymphocyte ratio (H:L), a reliable indicator of stress associated with corticosterone concentration (Gross and Siegel, 1983; Davis et al., 2008); and relative fluctuating asymmetry (FA), a measure of developmental instability that has been reported to reflect well-being status and performance of an animal (Van Valen, 1962; Parson, 1990; Møller and Swaddle, 1997), of chicks. Additionally, the differences between broody and non-broody hens in TI and H:L were also analyzed. We hypothesized that brooded chicks would show shorter TI, decreased H:L, and fewer asymmetrical morphological traits, whereas broody hens would show longer TI and increased H:L. With respect to the existing literature, the current study complements the previous experiment analyzing the effect of a broody hen on TI of chicks. To the knowledge of the authors, no study has measured the effects of a broody hen on H:L and FA of chicks, and as far as the authors know, differences between broody and non-broody hens for TI and H:L have not been studied yet.

2. Materials and methods

2.1. Birds and experimental design

There were 12 different breeds housed in 48 litter floor pens (pine-wood shavings; 3 m × 2 m) with nest boxes: 4 pens for each breed, and 20 hens in each pen. These breeds are maintained at the experimental station of El Encín (Madrid, Spain) in a conservation program of Spanish genetic resources started in 1975 (Campo and Orozco, 1982; Campo, 1998). All pens were located in the same henhouse. A total of 24 broody hens were observed. Fifteen broody hens were observed in pens of white shell egg breeds (1 in each of 3, 4, 3, 1, and 4 pens of the Blue Andaluza, Black-barred Andaluza, Black Red Andaluza, Black Castellana, and White-faced Spanish, respectively), 7 broody hens were observed in pens of tinted shell egg breeds (1 in each of 2, 1, 1, 2, and 1 pens of the Buff Prat, White Prat, Birchen Leonesa, Quail Castellana, and Quail Silver Castellana, respectively), and 2 broody hens were observed in pens of brown shell egg breeds (1 in each of 1 pen of the Red-barred Vasca and Red Villafranquina). Broody hens of all breeds exhibited same characteristics of brooding, remaining in the nest, sitting on eggs laid by other hens, with plucked chest and warmer than usual, and muttering, growling, and pecking if disturbed. The hens had been broody for 5 days. Broodiness was induced naturally (change in duration of natural light and temperature in June). TI, H:L, and FA were measured at 52 weeks of age in a total of 48 hens (24 broody and 24 non-broody hens). The broody/non-broody pairs were matched for breed, 1 broody and 1 non-broody hen of each breed in each pen with a broody hen. The non-broody hen of each pen was randomly chosen.

After that, all the non-broody hens were removed from the pens, and the broody hen was confined under an

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