《Research Note》

Increase in Plasma Luteinizing Hormone and Estradiol-17 β Concentrations following an injection of Luteinizing Hormone-Releasing Hormone in Incubating and Brooding Gifujidori Hens

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Chicken luteinizing hormone-releasing hormone- I (cLHRH- I) was injected intravenously into incubating and brooding Gifujidori hens, and the plasma concentrations of luteinizing hormone (LH) and estradiol-17 β (E₂) were measured by a radio-immunoassay. The LH concentration increased within 15 min after the injection, and the E₂ concentration increased within 60 min. The results suggest that the pituitary of incubating and brooding hens is capable to respond to cLHRH-I and the ovary is also capable to respond to LH released by cLHRH- I.

(*Jpn. Poult. Sci.*, **34** : 404-409, 1997)

Key words : LH, estradiol-17 β , LHRH, incubating hen, brooding hen

Introduction

Hens of some breeds of the chicken such as Japanese native breeds (Gifujidori, Bantam, etc.) incubate eggs for 21 days and subsequently brood chicks for about two to three months following the lay of eggs successively for a certain period of days. During the incubating and the brooding period, the secretion of luteinizing hormone (LH) (SHARP *et al.*, 1979, 1988 ; KUWAYAMA *et al.*, 1992 ; KANSAKU *et al.*, 1994) from the pituitary and estradiol-17 β (E₂) (ZADWORNÝ *et al.*, 1988 ; KUWAYAMA *et al.*, 1992) from the ovary is at a lower level. Whether the lower level of LH and E₂ is due to a decreased responsiveness of the pituitary and the ovary to their stimulatory factors is obscure. The present experiments were performed to elucidate whether the pituitary of incubating and brooding Gifujidori hens is capable to release LH in response to chicken luteinizing hormone-releasing hormone-I (cLHRH-I) and whether the ovary is capable to secrete E₂ in response to LH released by the cLHRH-I.

Materials and Methods

Gifujidori hens (0.8 to 1.0 kg body wt ; 12 months of age) were kept in individual floor pens (4 birds each in a room of 57 cm W, 57 cm D, and 57 cm H) in a temperature controlled house ($20 \pm 2^\circ\text{C}$). In each room of the house, one nest box (41 cm W, 21 cm D, and 41 cm H) with a sensor of recorder for incubation behavior was placed. Feed and water were provided *ad libitum* consumption under 14h (05 : 00 to 19 : 00) light daily. The hens which had continued to incubate eggs for one week after the onset of incubation behavior (sit on egg for more than 23h of a 24h day) and those continued to brood chicks for one week after hatch were used for experiments.

cLHRH-I (Gln⁸-LHRH ; Sigma Chem. Co., St. Louis, MO, USA) was dissolved in 0.9% NaCl solution and injected intravenously into the incubating and the brooding hens in a dose of $20\mu\text{g}$ per bird. The volume of injection was 0.5 ml. Control hens received an injection of the saline solution. Blood (1.0 ml) was collected from a wing vein using a heparinized syringe immediately before the injection ("0" min.) and at 15, 30, 60 and 120 min after the injection successively from same hens, and the plasma was obtained and stored at -20°C . Within one month, the plasma was assayed for LH using a homologous radioimmunoassay (RIA) (HATTORI and WAKABAYASHI, 1979), and also for E_2 using a routine RIA (KONO *et al.*, 1981). The intra-assay coefficient of variation (CV) was 7.0% (LH) and 7.3% (E_2), respectively. The inter-assay CV was less than 8.0% in both assays.

Results and Discussion

In both incubating and brooding hens, an increase in the plasma concentration of LH was found at 15 min after the injection of cLHRH-I (Fig.1). In control (saline injected) hens, no appreciable change was observed up to 120 min. The rate of increase from the level at "0" min. (immediately before injection) was greater in the incubating hens (approximately 6-fold increase) than in the brooding hens (approximately 4-fold increase). The greater responsiveness of the pituitary to cLHRH-I for the release of LH in the incubating hens than in the brooding hens is assumed to be due to a difference in the period of deficiency of an ovarian steroid hormone which stimulates the production of LH in the pituitary cells (KAWASHIMA *et al.*, 1982). This assumption is based on the fact that the time of sampling the plasma from the brooding hens was 3 weeks later from the time of the sampling from the incubating hens and the ovarian steroid hormone secretion is known to be at a low level throughout the incubating and brooding periods (SHARP *et al.*, 1979 ; ZADWORN *et al.*, 1988 ; KUWAYAMA *et al.*, 1992). SHARP and LEA (1981) reported on Bantam hens that the responsiveness of the pituitary to cLHRH-I for the release of LH was greater in the brooding hens at 39-49 days after hatching than in the incubating hens on 5-20 days of incubation. It is assumed that the pituitary responsiveness to cLHRH-I may increase at a later stage of the brooding period. At any rate, the increase in the plasma LH concentration in response to cLHRH-I may imply that the pituitary of incubating and brooding Gifujidori hens is capable to release LH if a certain stimulation is provided.

Fig. 1. The plasma concentration of Luteinizing Hormone (LH) in (a) incubating and (b) brooding Gifu-jidori hens after i.v. injection of 20 µg of chicken luteinizing hormone-releasing hormone-I (cLHRH-I) (○) or 0.5 ml of saline solution (●).

The vertical bars represent SEM of 4 birds. Asterisks indicate significant difference (*P < 0.05, **P < 0.01) from the value at "0" min. (immediately before injection).

The plasma E_2 concentration was increased following the injection of cLHRH-I and reached a peak at 60 min after the injection in both incubating and brooding hens (Fig.2). In control hens, no appreciable change was observed up to 120 min. The increase may be due to an indirect effect of cLHRH-I via the pituitary LH release, because the secretion of E_2 is stimulated by LH (CAMPER and BURK, 1977 ; ZADWORNÝ *et al.*, 1989), and because no evidence for the direct action of cLHRH on the ovary of hens has been reported thus far.

The results of the present study suggest that the pituitary of incubating and brooding Gifu-jidori hens is capable to respond to cLHRH-I and that the ovary is also capable to respond to LH released by the effect of cLHRH-I. The reported low level of

Fig. 2. The plasma concentration of estradiol-17 β in (a) incubating and (b) brooding Gifu-jidori hens after i.v. injection of 20 μ g of chicken luteinizing hormone-releasing hormone-I (cLHRH-I) (○) or 0.5 ml of saline solution (●).

The vertical bars represent SEM of 4 birds. Asterisks indicate significant difference (*P < 0.05) from the value at "0" min. (immediately before injection).

plasma LH and E₂ during the incubating and the brooding period (KUWAYAMA *et al.*, 1992) may not be due to the inability of the pituitary to secrete LH and of the ovary to secrete E₂ but to the deficiency of stimulation of the pituitary and the ovary.

Acknowledgments

This study was supported by Grant-in-Aid for Incentive Research (A) from Ministry of Education, Science and Culture (No. 06856034) and for Joint Research of Monbusho International Scientific Research Program (No. 07044190 to K.S.). This work was reported on XX World's Poultry Congress (New Delhi, India, September 1996) with the aid of the Travel Grant of Japan Poultry Science Association.

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抱卵および育雛中の岐阜地鶏母鶏における黄体形成ホルモン
放出ホルモン投与後の血漿黄体形成ホルモンおよび
エストラジオール濃度の増加

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抱卵中および育雛中の岐阜地鶏母鶏の静脈内にニトリ黄体形成ホルモン放出ホルモン (cLHRH-I) を投与し, 血漿黄体形成ホルモン (LH) とエストラジオール (E_2) 濃度をラジオイムノアッセイにより測定した。LH 濃度は cLHRH-I の投与 15 分以内に上昇し, E_2 濃度は 60 分以内に上昇することが認められ, 抱卵中および育雛

中の岐阜地鶏母鶏の脳下垂体は cLHRH-I に反応し得ること, また卵巣も cLHRH-I により放出された LH に反応し得ることが示唆された。

(家禽会誌, **34**: 404-409, 1997)

キーワード: 黄体形成ホルモン, エストラジオール, 黄体形成ホルモン放出ホルモン, 抱卵, 育雛