«Research Note»

Studies on the Role of Arginine Vasotocin in Relation to Oviposition in Laying Hens

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The role of arginine vasotocin (AVT) was investigated in relation to oviposition in laying hens. The posterior pituitary of fowl hens was removed 1-2 h before oviposition of the mid-clutch egg, but all hens so treated laid at the expected time of oviposition. The administration of AVT antiserum (1 ml/fowl hen or 0.3 ml/quail hen) had no effect on oviposition time. It is suggested that AVT released from the posterior pituitary may not act directly to cause the expulsion of the uterine egg.

(*Jpn. Poult. Sci.*, **31** : 358-362, 1994)

Key words : arginine vasotocin, oviposition, fowl hen, quail hen

Introduction

It is well known that there is a high degree of regularity in the time interval between which the successive eggs in a clutch are laid by the hen (ATWOOD, 1929 ; HEYWANG, 1938). Since a close relation between the time of oviposition and a succeeding ovulation in a given clutch has been reported by WARREN and SCOTT (1935) and PHILLIPS and WARREN (1937), the time of ovulation can be predicted within rather narrow range from the individual hen's record of laying. RIDDLE (1921) and BURROWS and BYERLY (1942) demonstrated that the administration of posterior pituitary preparations to the hen caused a premature expulsion of a uterine egg. Vasotocin were found in the posterior pituitary of the hen (MUNSICK *et al.*, 1960) and from *in vitro* and *in vivo* studies, arginine vasotocin (AVT) was found to be the most potent oxytocic substance (MUNSICK *et al.*, 1960 ; HELLER and PICKERING, 1961). The phenomenon of oviposition, *i.e.* the expulsion of the egg from the oviduct, is accompanied by a marked increase in the concentration of blood AVT (TANAKA and NAKAJO, 1962 ; STURKIE and LIN, 1966 ; TANAKA *et al.*, 1984). It has been considered that AVT plays an important role in the induction of oviposition in birds. In earlier works (TANAKA and NAKADA, 1974 ; TANAKA and GOTO, 1976 ; TANAKA, 1976), however, the ovarian oviposition-inducing factor (OOIF) has been found in pre- or post-ovulatory follicles. This OOIF is assumed to be the action of prostaglandins (PGs) (DAY and NALBANDOV, 1977), since they have a potent activity to induce premature oviposition in quail (HERTELENDY, 1972) and fowl (HERTELENDY *et al.*

1975), and were found to be markedly increased in the largest follicle shortly before oviposition and in peripheral blood plasma (OLSON *et al.*, 1986). On the other hand, it is postulated that PGF enhances the release of vasotocin that acts as an additive action to the effects of PGF and further stimulates uterine contractility through binding with the uterine membrane receptor (SHIMADA and SAITO, 1989). No satisfactory conclusion about the mechanisms of oviposition has been established.

The present study was conducted to clarify whether AVT released from posterior pituitary is a direct trigger of oviposition in domestic birds.

Materials and Methods

Animals

Birds from White Leghorn strain, aged 7–14 months and Japanese quail (*Coturnix coturnix japonica*), aged 1–3 months were used. They were maintained in individual laying cages fitted with automatic oviposition recorders and were exposed to a 14-h photoperiod. All birds selected for study were producing clutches of more than 4 eggs with a pause of only a single day between clutches.

Hypophysectomy

The posterior lobe of White leghorn hen was removed according to the method of OPEL (1965). Immediately after the removal that was completed 1–2 h before the expected time of oviposition, hens were returned to their individual cages and oviposition time was recorded. For removal of both the anterior and posterior pituitaries, the posterior lobe was removed first, followed by removal of the anterior lobe through the same surgical hole by sucking. For sham operation, the sphenoid bone was drilled, stopping just before reaching the sella membrane and the dental drill was pulled out, followed by closing the incision. This served as a control. All hens were killed 2–4 h after oviposition and the completeness of the lobectomy was verified in all birds.

Preparation of antiserum to AVT

AVT antiserum (R2-Kyushu) produced on our laboratory (Goro *et al.*, 1986) was used. This antiserum showed 91% binding to AVT at a final dilution of 1 : 100 and about 40% binding at a final dilution of 1 : 100,000, which did not show significant cross-reaction with oxytocin or mesotocin but showed 51% cross-reactivity with vasopressin, when the serum from White leghorn hen was tested. A single dose of 1 ml antiserum/fowl hen or 0.3 ml/quail hen was injected intravenously 8 h before the expected time of oviposition of the mid-clutch egg (Cs). After injection, birds were returned to their individual cages and oviposition time was checked. When oviposition time was more than 2 h late, it was considered a delay.

Results

As shown in Table 1, all of the 6 hens that were subjected to removal of the posterior pituitary 1–2 h before the expected time of oviposition laid at the normal time. Similarly, 4 hens with lack of both anterior and posterior pituitaries were laid at normal oviposition time. After autopsy, posterior pituitaries of 2 hens that were subjected to posterior lobectomy only were found to be incompletely removed. They

laid at the expected time. Sham operated hens also laid at the normal time. Thus, all of the operated hens including control laid normally.

Administration of 1 ml of AVT antiserum into fowl hens 8 h before the estimated time of oviposition had no effect on the time of the lay in any of the 5 hens so treated. The same results were obtained in all of the 9 quail hens, when 0.3 ml of AVT antiserum was given in the same manner as to the fowl hens. No delayed oviposition was observed in this experiment.

Discussion

OPEL (1966) removed the posterior pituitary on the day before expected lay of the terminal (Ct) egg of a sequence with 2 eggs, and he observed that the oviposition time was unaffected by the removal. However, it was considered that his experiment was not a reasonable design, because if the largest follicle destined to ovulate next is removed (TANAKA and NAKADA, 1974) or blockaded from its ovulation by a hormone (NYS, 1987), the uterine Cs egg at the time of treatment is known to behave as a Ct egg in oviposition time. Therefore, the present experiment was conducted to remove the posterior pituitary 1-2 h before the expected oviposition of Cs egg. However, the Cs egg was laid at the expected time in all cases. The report that removal of the posterior pituitary does not affect normal laying (SHIRLEY and NALVANDOV, 1956) supports our results. In this concern, OPEL (1966) has shown that oviposition in neural lobectomized

Table 1. Effect of hypophysectomy on the time of oviposition of C₂ egg in the domestic fowl

Operation*	No. of hens operated	No. of hens laid at expected time	No. of hens laid at delayed time
Removal of posterior pituitary	6	6	0
Removal of both anterior and posterior pituitaries	4	4	0
Incomplete removal of posterior pituitary	2	2	0
Control (Sham operation)	6	6	0

* All hypophysectomies were completed 1-2 h before expected time of oviposition.

Table 2. Effect of injection of AVT antiserum into fowl and quail hens on oviposition time

Birds	Amount of AVT antiserum*	No. of hens injected	No. of hens laid at expected time	No. of hens laid at delayed time
Fowl	1 ml	5	5	0
Quail	0.3 ml	9	9	0

* A single dose of AVT antiserum was injected intravenously 8 h before the expected oviposition time of mid-clutch egg.

hens might account for an abrupt release of AVT from the stalk-median eminence region. In order to rule out such a problem, AVT antiserum was administered into hens. However, oviposition time was unaffected, though the amount of the antiserum given was considered to be enough to neutralize the blood AVT. Since the R2-Kyushu antiserum has a binding activity of 91% at a final dilution of 1 : 100, the amount of the antiserum given, 1 ml/fowl hen or 0.3 ml/quail hen, was considered to be enough to neutralize the blood AVT in both cases. However, determination of hen's blood AVT to see whether it is actually neutralized with AVT antiserum still remains to be tested.

In view of the present results, it is suggested that AVT released from the posterior pituitary may not act directly to cause the expulsion of the uterine egg. As mentioned by SHIMADA and SAITO (1989), the AVT released at oviposition may have an additive action to the effects of PGF. However, further study will be needed to fully understand the mechanism of oviposition, since the origin of PGF at the laying of the Ct egg is not clearly understood.

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鶏及びウズラの放卵に対するアルギニン・バゾトシンの 関連性について

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連産中の鶏及びウズラを用いてアルギニン・バゾトシン (AVT) の放卵に関連する役割を検討した。推定放卵時刻の1~2時間前に雌鶏の下垂体後葉を除去した。しかしすべての個体が推定放卵時刻に放卵した。また、推定放卵時刻の8時間前に AVT 抗血清 1 ml を鶏に、0.3 ml をウズラにそれぞれ静脈投与したが、放卵時刻に影響

は無かった。これらの結果から、下垂体後葉から放出される AVT は放卵を直接誘起する作用はないものと推察された。

(家禽会誌, **31** : 358-362, 1994)

キーワード : アルギニン・バゾトシン, 放卵, 鶏, ウズラ