

Handling, ACTH, ACTH₁₋₂₄, and Naloxone Effects on Preening Behavior in Domestic Chickens

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WILLIAMS, N. S. AND D. L. SCAMPOLI. *Handling, ACTH, ACTH₁₋₂₄, and naloxone effects on preening behavior of domestic chickens.* PHARMACOL BIOCHEM BEHAV 20(5) 681-682, 1984.—Excessive grooming behavior has been well documented in various animals as a result of stress or intraventricularly administered ACTH and ACTH₁₋₂₄. In the present study, handling, ACTH, and ACTH₁₋₂₄ were associated with significant increases in preening of domestic chickens when compared to saline-treated controls. Naloxone-treated birds showed preening levels similar to those of unhandled and saline-treated birds.

ACTH ACTH₁₋₂₄ Naloxone Handling Stress Preening

ANIMALS under stress often exhibit higher levels of grooming behavior than those in relatively unstressed situations. Delius *et al.* [5] suggested that this may be a way of dispersing the boost in energy brought about by the activation of the hypothalamus-pituitary-adrenal cortex system. Numerous investigators have shown significant increases in grooming behavior in rats after intraventricular administration of ACTH and ACTH₁₋₂₄ [4, 7, 8, 11, 13]. A yawning and stretching syndrome was exhibited in dogs, cats and rabbits after receiving ACTH intracisternally [9]. A significant increase in yawning and head shaking has also been observed in pigeons given ACTH intraventricularly [5]. Green *et al.* [12] showed that stress-enhanced grooming in rats, following exposure to a novel environment, was blocked by naloxone.

The present study was conducted to determine how a known stressor, handling [10], affects specific kinds of behavior in chickens; if the behavior(s) could be blocked by naloxone; and if they could be produced by treatment with ACTH and ACTH₁₋₂₄.

METHOD

Two male and two female, heavy-hybrid chickens (1-2 kg; 8 months old; Cobbs Poultry Farm, Littleton, MA) were housed in separate cages (78×78×75 cm). They were exposed to a 14:10 light/dark cycle (lights on at 7:00) and given food and water ad lib. Chickens under Chloropent (0.015 cc/10 g body weight; IM) and Ketelar (Ketamine Hydrochloride; 0.08 cc/10 g, IP) were implanted with a polyethylene cannula into the lateral ventricle of the forebrain using the following coordinates from van Tienhoven and Juhasz [14]: AP, 8.0 mm; L, 1.5 mm; and DV, 8.1 mm. Cannula were fixed into place with dental cement. Testing began after at least four days of recovery and occurred between 12:00 and 15:00 hours.

Six testing situations were used: no handling, 10 min of handling, 2 mg naloxone/0.05 ml 0.75% saline (IP) followed by 10 min of handling, 1 IU ACTH/2 μl saline (ICV), 8 μg ACTH₁₋₂₄/2 μl saline (ICV) and 2 μl saline (ICV). The naloxone dosage is that used by Green *et al.* [12]; ACTH and ACTH₁₋₂₄ levels are similar to those reported by Delius *et al.* [5] and Williams *et al.* [16].

These procedures were repeated five times per bird in random order using a double blind procedure where appropriate so that birds received treatments in different orders and the treatment was unknown to the investigator. Only one type of injection was administered to each bird on any given day. Injections were carried out with a minimum of handling. After a 10 min delay, which we have found to be the best time to begin observing behavioral changes after handling, behavior was recorded for 15 min in 30 sec intervals by a white-coated observer. For each time interval, all behaviors that occurred within it were recorded. These included all the maintenance behaviors (feeding, drinking, stretching, and various kinds of grooming) as well as pecking, sitting, walking and sleeping. Preening behavior was defined as grooming behavior that involved use of the beak.

Data were analyzed using the analysis of variance for single factor experiments with repeated measures; differences between all possible pairs of means were tested by the Newman-Keuls method [15].

RESULTS

Preliminary inspection of the data indicated that no sex effect was obtained and therefore data were combined. The only sizeable difference in behavior involved preening behavior. The largest percent change in preening as compared to non-handling occurred for handled, ACTH and ACTH₁₋₂₄ treated birds (Table 1). Naloxone/handled and saline-treated

TABLE 1
PREENING SCORES OF CHICKENS RECEIVING SIX TREATMENTS

Treatment (N=4)	Mean \pm Standard Error	% Change*
No Handling	5.8 \pm 2.6	—
Handling	9.9 \pm 2.8 [†]	70.1
Naloxone Plus Handling	6.5 \pm 1.5	12.1
Saline	7.1 \pm 3.0	22.4
ACTH	15.6 \pm 1.7 [‡]	169.0
ACTH ₁₋₂₄	14.8 \pm 2.3 [‡]	155.2

*Each treatment compared to no handling.

[†]Significantly different from no handling, $p < 0.05$, Newman-Keuls method.

[‡]Significantly different from no handling, naloxone plus handling, saline, and handling, $p < 0.05$, Newman-Keuls method.

birds showed levels of preening similar to those of non-handled birds. In computing the analysis of variance, the mean value of the five replicates for each chicken and each treatment was used. The analysis of variance applied to these data revealed that the distributions were non-homogenous, $F(5,15)=18.9$, $p < 0.01$.

Results of the Newman-Keuls method for comparing treatment effects are also summarized in Table 1. Significant increases in preening were found in handled, ACTH and

ACTH₁₋₂₄ treated birds as compared to unhandled controls. No significant differences were seen in the preening of saline-treated, non-handled and naloxone/handled birds. The lower level of preening in naloxone/handled versus handled birds seems to indicate that naloxone treatment may block the increase seen after handling, although the difference was not statistically significant.

While handling, ACTH and ACTH₁₋₂₄ were all associated with increased preening and no significant difference was found between ACTH and ACTH₁₋₂₄ treated birds, the effect of handling was found to be significantly different from that of both ACTH and ACTH₁₋₂₄. This suggests that the administered amounts of ACTH and ACTH₁₋₂₄ were in excess of those produced by the animals in their reaction to handling and indicates the need for further studies to provide dose-response information.

These results implicate the role of ACTH, specifically the first 24 amino acids of the compound, in influencing excessive grooming in yet another class of vertebrates. The marked increases in preening scores after ACTH, ACTH₁₋₂₄ and handling would imply that the elevation in this behavior is stress-induced and that preening behavior may be a good index of the level of stress in chickens.

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