Interactions between histamine H1 and H3 and dopamine D1 receptors on feeding behavior in chicken

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Abstract:

**BACKGROUND:** Brain monoamines (such as histamine and dopamine) play an important role in emotions, cognition, reward and feeding behavior. The interactions between histamine and dopamine were studied in many physiological functions but this correlation is unclear in feeding behavior of chickens. **OBJECTIVES:** The aim of this study was to investigate the interaction of central histaminergic and dopaminergic systems on food intake in broiler chicken. **METHODS:** In this study intracerebroventricular (ICV) injection was used for manipulation of histaminergic and dopaminergic systems. In Experiment 1, 3 h-fasted chicks were given an ICV injection of histamine, SCH23390, a D1 receptors antagonist and co-injection of histamine and SCH23390. Experiments 2-5 were similar to experiment 1 except birds were injected with AMI-193, D2 receptors antagonist; NGB2904, D3 receptors antagonist; L-741,742, D4 receptors antagonist and 6-OHDA, 6-hydroxydopamine instead of SCH 23390, respectively. In experiment 6, ICV injection of dopamine, chlorpheniramine, H1 receptors antagonist and co-administration of dopamine and chlorpheniramine were done. Experiments 7-9 were similar to experiment 6, except birds ICV injected with famotidine, H2 receptors antagonist; thioperamide, H3 receptors antagonist and alpha-FMH, alpha-fluoromethylhistidine in place of chlorpheniramine, respectively. Then cumulative food intake (g) was measured at 30, 60 and 120 min after the injection. **RESULTS:** Histamine decreased food intake compared to the control chicks indicating an inhibitory effect of histamine on food intake and SCH23390 attenuated the effect of histamine on food intake (p<0.001). In addition, hypophagic effect of histamine decreased by 6-OHDA (p<0.001). Chlorpheniramine and alpha-FMH significantly attenuated dopamine induced hypophagia (p<0.001). However, thioperamide amplified the inhibitory effect of dopamine on food intake(p<0.001). **CONCLUSIONS:** These results suggest there is relationship between histaminergic and dopaminergic systems on food intake in chicken and H1, H3 and D1 receptors are involved in this interaction.
Introduction

Birds, like mammals, have complex mechanisms regulating food intake. Given a choice between more than one diet, turkeys, broilers, layers, and other avian species display the ability to self-select a diet adequate for growth or production (Denbow, 1999). Although compensation may not be complete, if exposed to severe feed restriction early in life, birds compensate by increasing their intake in order to increase weight gain following the restriction. In contrast, force feeding birds twice their ad libitum food intake causes Leghorns to stop eating for 7-10 days until their fat stores approach pre-force-feeding levels. Therefore, clearly, birds have the ability to regulate food intake (Denbow, 1999). A number of peptides comprise a complex network that regulates feeding behavior in avian (Kaneko et al., 2012).

Histamine is one of the well-known amines which contribute in many physiological functions in the central nervous system (CNS) (Blandina et al., 2012). It is reported that paraventricular nucleus (PVN) and ventromedial hypothalamus (VMH) receive afferent projected axons of histaminergic (HA-ergic) neurons from tuberomammillary nucleus (TMN). They send projections organized in functionally distinct circuits impinging on different brain regions and their firing frequency changes according to the behavioral state (Giannoni et al., 2009). Four subtypes of histamine receptors have been classified (H1, H2, H3 and H4), all of these subtypes distributed in several part of CNS (Schneider et al., 2014).

Brain histamine plays a vital role in eating behavior and has been considered as satiety signal that is released during food intake (Rozov et al., 2014). It is known ICV administration of histamine decreases food intake whereas ICV injection of chlorpheniramine (histamine H1 receptor antagonist), or alpha-fluoromethylhistidine (α-FMH, selective inhibitor of histidine decarboxylase) increases food intake in rats (Morimoto et al., 2001) and chicken (Kawakami et al., 2000; Taati et al., 2009).

Dopamine is the predominant catecholamine neurotransmitter in the mammalian brain which controls a variety of functions including locomotor activity, cognition, emotion, positive reinforcement and food intake (Ikemoto, 2007). Dopamine is a key anorexigenic neurotransmitter modulating reward which acts mainly through its projections from the ventral tegmental area (VTA) into the nucleus accumbens (NAcc) and arcuate nucleus (ARC) (Volkow et al., 2011). Dopaminergic neurons (DAergic) of the Substantia Nigra (SN), Pars Compacta, VTA and hypothalamus give origin to three main pathways, Nigrostriatal, Mesolimbocortical and Tuberoinfundibular in CNS (Cadet et al., 2010). The inhibitory effect of DA on food intake was decreased by SCH23390 pretreatment in chicken (Bun go et al., 2010; Zendehdel et al., 2014). The same observation was reported in mammals in which ICV injection of SKF 38393 (D1 receptors agonist) and apomorphine (D2 receptors agonists) decreased cumulative food intake in rats (Kuo, 2002).

Based on the reports, there is correlation between DAergic and HAergic systems where L-DOPA activates histaminergic neurons in CNS (Yanovsky et al., 2011). For instance, H3 receptor interacts with D1 and D2 receptors in the striatum of patients suffering Parkinson’s disease (Yanovsky et al., 2011). It is shown the dopamine D1 and...
D2 receptors in the basolateral amygdaloid (BLA) nucleus may be involved in the anxiogenic-like effects induced by histamine in rat model (Bananej et al., 2012). Histaminergic projections innervate VTA, which consists of the cell bodies of dopamine neurons (Montoro et al. 2013). The modulation of monoamines and other neurotransmitters via H3 heteroreceptors has also been described (Schlicker et al. 1994). It is possible that the H3 receptor functions as a heteroreceptor to release monoamines such as NE, and DA.

To date, much progress has been done to identify mediatory effect of neurotransmitters on feeding behavior in mammals but food intake regulation in domestic poultry is not fully studied (Zendehdel and Hassanpour 2014; Hassanpour et al., 2015). It is well documented that central feeding behavior is not regulated via a single neuropeptide and a wide distributed neural network interacts with diversity of neurotransmitters on feeding status (Zendehdel et al., 2016).

The interactions between histamine and dopamine were studied in many physiological functions but this correlation is unclear in feeding behavior of chickens. So, the aim of this study was to investigate the interaction of central histaminergic and dopaminergic systems on feeding behavior in broiler chicken.

Materials and Methods

Animals: A total of 432 one-day-old broiler chickens (Ross 308) were purchased from a local hatchery (Eshragh Co. Iran). At 2 d of age birds were randomly transferred to individual cages and kept at a temperature of 30 ± 1°C with 50 ± 2 percent humidity (Olanrewaju et al., 2006). Birds were housed according to a completely randomized design. During the study birds had ad libitum access to fresh water and a starter diet containing 21% crude protein and 2850 kcal/kg of metabolizable energy (Animal Science Research Institute Co., Iran). Three hours prior to the injections, birds were food deprived (FD3) but had free access to water. Animal handling and experimental procedures were performed according to the Guide for the Care and Use of Laboratory Animals by the National Institutes of Health (USA) and the current laws of the Iranian government for animal care.

Experimental drugs: The histamine, SCH 23390 (D1 receptors antagonist), AMI-193 (D2 receptors antagonist), NGB2904 (D3 receptors antagonist), L-741,742 (D4 receptors antagonist), 6-OHDA (6-hydroxydopamine), dopamine, chlorpheniramine (H1 receptors antagonist), famotidine (H2 receptors antagonist), thioperamide (H3 receptors antagonist), α-FMH (alpha-fluoro-methylhistidine) and Evans blue purchased from Sigma Co. (Sigma, USA). Drugs were first dissolved in absolute dimethyl sulfoxide (DMSO) then diluted with 0.85% saline containing Evans blue at a ratio of 1/250. DMSO with this ratio does not have a cytotoxic effect (Blevins et al., 2002; Qi et al., 2008).

Intracerebroventricular injection procedures: Intracerebroventricular (ICV) injections conducted at 5 days of age. A total of 9 experiments were designed to investigate the interconnection of histaminergic and dopaminergic systems on food intake in neonatal birds. Each experiment included 4 treatment groups with 12 replicates per group (n = 48 chickens per experiment). In each experiment, chicks were weighed and allocated into experimental groups based on
their body weight so that the average weight (45±5 g) between treatment groups was as uniform as possible. Each chicken received ICV injected once in each experiment. Injections were done using a microsyringe (Hamilton, Switzerland) without anesthesia based on the method described by Davis et al., (1979) and Furuse et al., (1997). In this technique, the head of the chick was held with an acrylic device in which the bill holder was at 45° and the calvarium was parallel to the surface of table (Van Tienhoven and Juhasz, 1962). An orifice was made in a plate that was located over the skull immediately over the right lateral ventricle. The microsyringe was inserted into the ventricle through the orifice in the plate. The tip of the needle perforated only 4 mm below the skin of the skull (Jonaidi and Noori, 2012). There is no physiological stress using this technique in neonatal chicken (Saito et al., 2005). Injections were done in a volume of 10 μl (Furuse et al., 1999). The control group received control solution as 10 μl of saline containing Evan’s blue (Furuse et al., 1999). At the end of the experiments, to recognize the accuracy of injection, chicken were killed by decapitation. Accuracy of placement of the injection in the ventricle was verified by the presence of Evans blue followed by slicing the frozen brain tissue. Only data from individuals in which dye was present in their lateral ventricle were used for analysis. All experimental procedures were done from 8:00 A.M. until 3:30 P.M. Also, the time course of food consumption was selected by previous studies (Bungo et al., 2010; Zendehdel et al., 2008, 2014, 2016).

**Food intake measurement procedure:** In this study, 9 experiments were designed, each with 4 treatment groups (n=48). In Experiment 1, 3 h-fasted chicks were given an ICV injection of histamine (300 nmol), SCH23390 (5 nmol), a D1 receptors antagonist and co-injection of histamine and SCH23390. Experiments 2-5 were similar to experiment 1 except birds were injected with AMI-193 (5 nmol), D2 receptors antagonist; NGB2904 (6.4 nmol), D3 receptors antagonist; L-741,742(6 nmol), D4 receptors antagonist and 6-OHDA (2.5 nmol), 6-hydroxydopamine instead of SCH 23390, respectively. In experiment 6, ICV injection of dopamine (40 nmol), chlorpheniramine (300 nmol), H1 receptors antagonist and co-administration of dopamine and chlorpheniramine were done. Experiments 7-9 were similar to experiment 6, except birds ICV injected with famotidine (82 nmol), H2 receptors antagonist; thioperamide (300 nmol), H3 receptors antagonist and α-FMH (250 nmol), alpha-fluoromethylhistidine in place of chlorpheniramine, respectively. To find the possible relationship between these two systems, effective and sub-effective doses of pharmacologic agents were administered to confront nullifying effects of the agents. In other words, when an effective dose of a system is administered, the sub-effective dose of the other system is considered. Immediately after injection, chickens were returned to their individual cages and provided with access to pre-weighed food ad libitum and water. Cumulative food intake (g) was measured at 30, 60 and 120 min after the injection. Food consumption was calculated as a percentage of body weight (%BW) to minimize impact of body weight on the amount of food intake. Each bird was just used once in each experimental group.

**Statistical analysis:** Data is presented as mean ± SEM (standard error of the mean). Cumulative food intake (as percentage
Effect of ICV injection of 300 nmol chlorpheniramine, 40 nmol dopamine and their combination on cumulative food intake in neonatal chicken is presented in Fig. 6. According to the results, the ICV injection of 40 nmol dopamine significantly decreased food intake compared with control group (p<0.001) (Fig. 6). Chlorpheniramine administration also significantly decreased the effect of dopamine on food intake in birds (p<0.001) (Fig. 6).

As seen in Fig. 7, ICV injection of sub effective dose of famotidine (82 nmol) had no effect on food intake (p>0.05). Also, ICV injection of 40 nmol dopamine significantly decreased food intake in neonatal broiler chicken (p<0.001). Co-injection of dopamine and famotidine had no effect on food intake compared with dopamine group (p>0.05) (Fig. 7).

Based on the results in experiment 8, 300 nmol thioperamide administration significantly amplified dopamine-induced hypophagia in chickens (p<0.001) (Fig. 8); but ICV injection of thioperamide alone could not alter food intake in comparison with control group (p>0.05) (Fig. 8).

As observed in Fig. 9, ICV injection of sub effective dose of α-FMH (250 nmol) had no effect on feeding behavior in neonatal broiler chicken (p>0.05). Also, ICV injection of dopamine (40 nmol) significantly decreased food intake in neonatal broiler chicken (p<0.001). Co-injection of dopamine and α-FMH decreased dopamine-induced hypophagia in neonatal broiler chick-
en (p<0.001) (Fig. 9). These results showed that dopamine-induced hypophagia is mediated via histaminergic H1 and H3 receptors in neonatal chicks.

**Discussion**

Based on the literature this paper is the first report on the relationship between DAergic and HAergic systems on food intake in FD3 neonatal broiler chickens. According to the results, there is relationship between two systems via D1, H1 and H3 receptors on central food intake regulation in neonatal broiler chicken. According to the previous studies, the ICV injection of dopamine and L-DOPA (precursor of dopamine) significantly decreased food intake in FD3 broiler cockerels (Zendehdel et al., 2014).
of body weight) was analyzed by repeated measure two-way analysis of variance (ANOVA) using SPSS 16.0 for Windows (SPSS, Inc., Chicago, IL, USA). For treatment, showing a main effect by ANOVA means was compared by Tukey-Kramer test. p<0.05 was considered significant difference between treatments.

Results

The results of interaction between central DAergic and HAergic systems on food intake control in neonatal broiler chicks are presented in Figs. 1-9. In experiment 1, ICV injection of an effective dose of histamine (300 nmol) significantly decreased food consumption at 30, 60 and 120 min post-injection (p<0.001). ICV injection of sub effective dose of SCH23390 (5 nmol) had no significant effect on food intake compared to control group in FD3 neonatal broiler (p>0.05); while co-injection of SCH23390 (5 nmol) and histamine (300 nmol) significantly attenuated histamine-induced hypophagia at 30, 60 and 120 min post-injection (p<0.001)(Fig. 1).

As seen in Fig. 2, the ICV injection of sub effective dose of AMI-193 (5 nmol) had no significant effect on food intake compared to control group in FD3 neonatal broiler (p>0.05). Also, co-injection of AMI-193 and histamine had no effect on histamine-induced hypophagia in chicken (p>0.05) (Fig. 2).

As seen in Figs. 3 and 4 ICV injection of sub effective doses of NGB2904 (6.4 nmol) and L-741,742 (6 nmol) could not alter food intake induced by 300 nmol histamine in neonatal chick (p>0.05) (Figs. 3 and 4).

According to the results obtained from experiment 5, post hoc analysis revealed that 6-OHDA (2.5 nmol) administration significantly decreased hypophagic effect of histamine (p<0.001) (Fig. 5); however, ICV injection of 6-OHDA (2.5 nmol) alone had no effect on food intake compared with control group(p>0.05) (Fig. 5). The results obtained from experiments 1-5 revealed that hypophagic effect of histamine is mediated via dopaminergic D1 receptors in neonatal chicks.
A daily decrease on cumulative food intake was reported using D1 (SKF 38393) and D2 (apomorphine) receptors agonists in rats (Kuo 2002). Also, previous studies showed that central injection of histamine H1 receptor antagonist increased cumulative food intake in rat (Morimoto et al., 2001), layer and broiler chickens (2010). Perhaps, histamine mediates food intake in CNS using H1 receptors, but the direct location of this receptor is still unclear in chicken (Taati et al., 2006). Also, in this study, we used sub effective doses of antagonists to determine possible interaction of HAergic and DAergic systems on feeding behavior in neonatal chicken. As observed, sub effective doses of H1-H4 receptors antagonists had no role on food intake in neonatal broiler chicken. Thioperamide is histamine H3 receptor antagonist which increases histaminergic neurons activity. Perhaps thioperamide-induced histaminergic neurons activity occurs via disencumbering negative feedback on histidine decarboxylase enzyme and/or release of histamine (Sakata et al., 1997).

As seen in this study, a relationship was identified between two systems via D1, H1 and H3 receptors on central food intake regulation in neonatal broiler chicken. H3 receptors antagonists are able to stimulate monoamine neurotransmission. Recently it was revealed modulation of DAergic neurons play an important role in functional interactions between histamine and other monoamines (Flik et al., 2015). The ICV injection of histamine into the VTA mimicked the stimulatory effect of thioperamide on DAergic neurons (Flik et al., 2015). So, it seems interaction exists between central DAergic and HAergic systems, our results are in agreement with previous reports.

On the other hand, the stimulatory effect of thiopiramid on DAergic system is mediated by activation of dopamine neurons probably via excitatory histamine H1 receptors (Flik et al., 2011). The ICV injection H1 receptor agonist stimulates mesolimbic and mesocortical dopamine release (Lapa et al., 2005). Thus, activation of the HAergic system by L-DOPA increased histamine and dopamine levels in forebrain areas (Yanovsky et al., 2011). The results of the current study demonstrate that chlorpheniramine attenuated dopamine induced hypophagia while thioperamide increased the effect of dopamine on food intake. This finding is consistent with previous observations. It was reported that thioperamide potentiated L-DOPA and methamphetamine induced DA release in striatum and nucleus accumbens, respectively (Nowak et al., 2008). Other antagonists of H3 receptors, ABT-239 and GSK189254, increased DA levels in PFC and cingulate cortex, respectively (Medhurst et al., 2007).

It was recently shown that H3 receptors form heteromers either with D1 or D2 receptors in the striatum, where histamine is able to decrease sensitivity of the receptor to dopaminergic drugs, while an H3 receptors antagonist increases its sensitivity (Ferrada et al., 2009). Also, there are H3 receptors on dopaminergic neurons terminals as hetero-receptors and modulate dopamine release (Flik et al., 2015). However, scarce information exists on interaction of DAergic and HAergic systems on feeding behavior. So, we are not able to compare our results with them. However, it is reported there are other interactions between DAergic and HAergic systems via GABAergic and/or serotonin where GABA and serotonergic neurons contribute with dopamine neurons in modulation of anxiety behavior (Zarrindast and...
Khakpai, 2015). For instance, thioperamide increased serotonin, norepinephrine and dopamine levels in the rat prefrontal cortex (Flik et al., 2015). Also, the H3 receptors are ligands on serotoninergic and DAergic neurotransmission (Flik et al., 2015).

In conclusion these results suggest there is a relationship between histaminergic and dopaminergic systems on food intake in chicken and H1, H3 and D1 receptors are involved in this interaction. Based on our knowledge, there was no report on molecular and cellular interaction between DAergic and HAergic systems on feeding behavior in avian. So, further researches are needed to determine molecular pathway(s) for this relationship. Presumably, obtained results can be as base information for further studies on the possible involvement of DAergic and HAergic systems with other neurotransmitters on feeding behavior.

Acknowledgments

This research was supported by a grant from the Research Council of the Faculty of Veterinary Medicine, University of Tehran, Iran.

References
Histamine and dopamine interaction

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اتر اثرات تقابلی گیرندههای $D_1$ و $H_3$ هیستامینی با گیرندههای $D_3$ دوپامینی بر رفتار

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چکیده

منوآمین های مغزی (مثل هیستامین و دوپامین) نقش مهمی را در حواس، شناخت، پاداش و تنظیم اشتها بازی می کنند. بسیاری از اعمال فیزیولوژی تقابل بین سیستم هیستامینرژیک و دوپامینرژیک بررسی شده است اما تداخل عمل آنها در تنظیم اشتها در جوجهها ناشی از هیستامین و مغزی دوپامینی مطرح است. هدف از این مطالعه بررسی تداخل سیستم های دوپامینرژیک و هیستامینرژیک در تنظیم اشتها در جوجهها. مطالعه در ۱۳۹۵ میلادی انجام شد و مطالعه بر روی ۴ گروه جداگانه صورت گرفت. سایر موارد انجام شد.

روش کار:

پس از ایمنی بررسی های اولیه و در بهتری از این، مطالعه در ۱۳۹۵ میلادی انجام شد و مطالعه بر روی ۴ گروه جداگانه صورت گرفت. سایر موارد انجام شد. در مطالعه محاسبه اثرات متقابل گیرنده های دوپامینرژیک و هیستامینرژیک در تنظیم اشتها در جوجه های دوپامینی و هیستامینی انجام شد. در این مطالعه، گروه اول و گروه دوم به ترتیب با آنتاگونیست گیرنده $D_1$ و دوپامین تزریق شدند. در گروه سوم، جوجه ها به ترتیب با آنتاگونیست گیرنده $H_3$ و هیستامین تزریق شدند. در گروه چهارم، جوجه ها به ترتیب با آنتاگونیست گیرنده $H_1$ و دوپامین تزریق شدند. در این مطالعه، تزریق داخل مغزی هیستامین به جلوگیری از اثرات متقابل گیرنده های دوپامینرژیک و هیستامینرژیک در تنظیم اشتها در جوجه های دوپامینی و هیستامینی کمک کرد.