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Effect of Peripheral Administration of Ghrelin on Serum Insulin, T_3, T_4 and Some Biochemical Parameters in Geese

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ABSTRACT: Nowadays, ghrelin has been identified in six species of birds (includes chickens, turkey, emu, goose, duck and Japanese quail). In present study, effect of exogenous ghrelin on serum insulin, T_3, T_4 and some biochemical parameters of geese were investigated. Forty eight 28d old geese were assigned in to 3 treatments and 4 replicates (include 4 birds in each replicate). The experiment was arranged in completely randomized design (CRD). The injected dosage of ghrelin was different in treatments: 0, 50 and 100 µg ghrelin/kg BW. Injection procedure was conducted on d 28 of age and before onset of experimental rearing period. 12 h after ghrelin injection, two birds from each replicate of each treatment that had BW close to the mean replicate was subjected to collection of blood sample. Analysis of serum samples show that the injection of ghrelin caused insulin declines in serum (G50 and G100 in comparison with G0). There is lower level of T_4 was observed for ghrelin-injected groups which group with high dosage of ghrelin (G100) show lowest level of T4. Uric acid had minor elevation for G50 and dramatic elevation for G100 when compared with G0. Other parameters include T_3, Ca, P and Total protein haven't any changes via peripheral ghrelin administration. In conclusion, ghrelin administration in goose can decrease insulin and T_4 levels and increases uric acid concentrations in serum. Further investigations can be suggested to identification of goose ghrelin roles.

Key words: Ghrelin, Peripheral administration, Regulatory peptides, Serum biochemical parameters, Geese.

INTRODUCTION

Ghrelin passed 15-years from its discovery in mammalian (Kojima *et al*., 1999). Published literatures have demonstrated major regulatory functions for ghrelin, such as growth hormone-releasing activity (Hashizume *et al*., 2005); food intake, weight gain and energy balance (Vizcarra *et al*., 2007). In birds, chicken ghrelin with 26-amino acids and is shorter than human or rat ghrelin, that firstly identified by Kaiya *et al*. (2002), and until recently ghrelin has been identified in six species of birds includes chickens, turkey, emu, goose, duck and Japanese quail (Kaiya *et al.*, 2008).

Ghrelin is multifunctional endocrine peptide which can affect other endocrine hormones and related serum biochemical factors. Mammalian ghrelin has considerrable role in glucose homeostasis by modulation of insulin releasing (Ahima, 2006; Castañeda *et al*., 2010). Ghrelin increases blood glucose (Dezaki *et al*., 2004) and decreases plasma insulin levels in humans and rodents models (Broglio *et al*., 2001; Dezaki *et al*., 2004).

Some different studies such as Granata (2008) reported that both acylated and non-acylated ghrelin stimulated glucose-induced insulin releasing by ß-cells. Also, Lee *et al*. (2002) observed that intravenous administration of ghrelin stimulates insulin secretion in free-feeding rats. Khazali (2005) with ventricle infusion of ghrelin in mammalian model reported that ghrelin may increase the mean plasma concentration of T_3 and T_4. In human, the effect of ghrelin on hypothalamus-pituitary-thyroid axis and circulated T_4 was documented.

Our previous studies (Lotfi *et al*., 2011; Aghdam Shahryar & Lotfi, 2013) show considerable effect of *in ovo* ghrelin on chicken insulin and thyroid hormones. High dosage of *in ovo* administrated ghrelin can elevate plasma T_4 level (Aghdam Shahryar and Lotfi, 2013). Ghrelin may cause insulin-resistance affect in chicken to maximize glucose concentration at hatching. But there is no any evidence for other species of birds.

In present study, effect of exogenous ghrelin injection on serum insulin, T_3, T_4 and some biochemical parameters of geese were investigated.

MATERIALS AND METHODS

Forty eight 28d old geese were assigned in to 3 treatments and 4 replicates (include 4 birds in each replicate). The experiment was arranged in completely randomized design (CRD). The lyophilized rat ghrelin was purchased from Sigma-Aldrich Co. (USA), dissolved in 1% acetic acid solvent and desired concentrations of ghrelin were prepared. The injected dosage of ghrelin was different in treatments: 0, 50 and 100 µg ghrelin/kg BW. Injection procedure was conducted on d 28 of age and 12 h after ghrelin injection, two birds from each replicate of each treatment that had BW close to the mean replicate was selected. Blood sample was collected from the wing vein using sterilized syringes.

Blood samples were centrifuged and the serum was separated and collected in to microtubes. Serum biochemical parameters and thyroid hormones were measured by Pars Azmoon© Elisa kits (Pars Azmoon Co., Tehran), and insulin was measured by Glory© Elisa kit. Collected data were analyzed by SAS software (Ver. 9.1) and statistical comparisons were conducted by Duncan (1955) multiple range test.

RESULTS AND DISCUSSION

The hormonal and biochemical parameters are presented in Table 1 and Table 2, respectively. In present study, injection of ghrelin caused insulin declines in serum (G50 and G100 in comparison with G0). There is lower level of T_4 was observed for ghrelin-injected groups which group with high dosage of ghrelin (G100) show lowest level of T4 (Table 1). Uric acid had minor elevation for G50 and dramatic elevation for G100 when compared with G0. Other parameters include T_3, Ca, P and Total protein haven't any changes via peripheral ghrelin administration.

Ghrelin has hyper-glycemic effect in avian (Lotfi *et al*., 2011) and it can raises basal glucose levels in mammalian (Broglio *et al*., 2003), because of this it's so natural that insulin has declines after ghrelin injection. In present study, decreasing in T_4 following ghrelin injection (Table1) may be because of ghrelin effects on basal metabolism that lower T_4 enhances slower metabolism and its direct effect on pituitary thyroid (HPT) axis and its role in the regulation of energy homeostasis (Khazali, 2009).

Table 1. Effect of IP-injection of rat ghrelin on some of serum hormonal characterizes in geese.

Treatments	Injection dosage (ng/kg BW)	insulin	T_3 ng/dl	T_4 ng/dl
(control)	0	4.62[a]	1.4	4.00[a]
G50	50	2.62[b]	1.85	1.97[b]
G100	100	1.55[c]	1.45	2.15[b]
P-value		0.010	0.433	0.004
SEM		0.70	0.14	0.23

Means within columns with different superscript letters are significantly different (P < 0.05).

Table 2. Effect of IP-injection of rat ghrelin on some of serum biochemical parameters in geese.

Treatments	Injection dosage (ng/kg BW)	Ca mg/dl	P mg/dl	Uric acid mg/dl	Total protein g/dl
(control)	0	10.32	5.62	1.40[b]	3.30
G50	50	10.45	6.17	2.87[ab]	3.77
G100	100	9.92	7.05	3.52[a]	3.50
P-value		0.933	0.297	0.024	0.292
SEM		0.45	0.36	0.52	0.21

Means within columns with different superscript letters are significantly different (P < 0.05).

Also, increases in uric acid levels can reflect skewing to more protein metabolism (Khazali, 2009) may for reserving glucose stores in the body. In other hand, present findings about insulin hormone can be in agreement with our previous reports on in ovo ghrelin effects which neonatal chickens have higher glucose concentration (Lotfi *et al*., 2011). As conclusion, ghrelin administration in goose can decrease insulin and T_4 levels and increases uric acid concentrations in serum. It seem that these hormonal changes may be in relation to ghrelin act in glucose hemostasis and less consumption of energy resources specially glucose resource. Further investigations can be useful to solving ghrelin puzzle in goose.

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