EFFECT OF IN OVO ADMINISTRATION OF EXOGENOUS GHRELIN ON WEIGHT OF INTERNAL ORGANS IN NEWLY-HATCHED CHICKS

Alireza Lotfi* and Habib Aghdam Shahryar**
*Ilkhchi Branch, Islamic Azad University, Ilkhchi, Iran
**Department of Animal Science, Shabestar Branch, Islamic Azad University, Shabestar, Iran
(Email: arlotfi@gmail.com)

ABSTRACT
The present study was performed for investigation on possible effects of exogenous ghrelin (GH-releasing peptide) on liver, heart, gizzard (proventriculus+gizzard), intestine and breast muscle weights. One hundred fertilized eggs were divided into two groups includes control or group1 (without any in ovo ghrelin administration) and group2 (in ovo administration of 100ng/egg ghrelin at day-5). In ovo injection process was done by 22g needles and infusion of 0.5 ml solution in albumen. All of eggs were incubated under normal hatchery conditions. At end of incubation, ten chicks from each group were decollated and internal organs were separated and weighed. The results had shown that exogenous ghrelin only had significant effect on intestine weight (group1: 2.05g and group2: 2.30g) (P<0.01) and minor positive effect on gizzard weight (group1:2.20g and group2: 2.26), too. There was not any significant positive effect on liver, heart and breast muscle weights via in ovo injection of ghrelin. With attention to ghrelin’s gastrointestinal origin (synthesis in gastrointestinal cells) and physiological potential for GH releasing, so its effect on gastrointestinal organs of chicken (such as gizzard and intestine) may be because of its specific regulatory roles in development of these organs during embryonic life. Present findings are in agreement with past findings about effects of in ovo injected ghrelin on chicken intestinal enzymes activity. As conclusion, in ovo ghrelin can affect embryonic development of gastrointestinal organs and their weight gain in chicks.

KEY WORDS: GH-releasing peptides, ghrelin, in ovo administration, internal organs, newly-hatched chickens.

INTRODUCTION
Ghrelin is an endocrine regulatory peptide with GH-releasing function. This peptide was isolated by Kojima et al., (1999) in gastric cells of laboratory rat, for first time. After identification of mammalian ghrelin, so many relative studies were conducted on its physiological functions in the body. Studies had shown that ghrelin is strong GH-releasing factor, and also has ability to stimulate appetite, increase body weight and regulation of energy metabolism (Nakazato et al., 2001; Toshinai et al., 2001; Vicennati et al., 2007). About avian ghrelin, relative studies are limited in published literatures when it compared with mammalian ghrelin’s studies. The chicken ghrelin has GH-releasing effect, too (Kaiya et al., 2009). The chicken ghrelin includes 26 amino acid is shorter than human and rat ghrelin (28 amino acid) (Kaiya et al., 2002) (Figure1).

![Peptide structure of rat ghrelin; shows sequence of amino acids and acylation with a medium-chain fatty acid.](image)

Figure 1. Peptide structure of rat ghrelin; shows sequence of amino acids and acylation with a medium-chain fatty acid.

Budat and Harvey, (2003) had shown that chicken ghrelin has in vivo growth stimulatory effect and can increase somatotrophs proliferation in in vitro condition. Expression of ghrelin gene in avian embryo caused to suggest that maternal ghrelin has developmental effect in chicken (Sirotkin et al., 2006; 2008). In this regard, Yoshimura et al., (2009) had identified ghrelin peptide in yolk and albumen of chicken egg. Gahr et al., (2004) had reported that ghrelin mRNA has increases after embryonic day-4 in chicken embryo, considerably. In ovo experiment that conducted by Turkish researchers (Deprem and Gulmez, 2007) shows in ovo infusion of IGF-1 (insulin like growth factor) cause...
muscular development and hypertrophy in neonatal Japanese quails. Currently, Liu et al., (2012) shows similar findings in duck. First in ovo studies with ghrelin infusion (Lotfi et al., 2011a; 2011b) had reported insulin and prolactin increases in plasma of hatched chicks. So, with attention to developmental effects of maternal ghrelin, in present study, possible effect of in ovo ghrelin administration on liver, heart, gizzard (proventriculus+gizzard), intestine and breast muscle size were investigated.

MATERIALS AND METHODS

One hundred fertilized eggs obtained from commercial broiler breeder flock was divided into two groups includes control or group1 (without any in ovo ghrelin administration) and group2 or (in ovo administration of 100ng/egg ghrelin at day-5). All of eggs were incubated under normal hatchery conditions. The lyophilized tube of rat Ghrelin were obtained from Sigma-Aldrich Company. Homogenous solution of ghrelin (1% acetic acid) was prepared and in ovo injection procedure was done via 22g needle at a dark room with 37 °C temperature. Embryo mortality was detected by daily candling and separated from hatchery for healthy incubation. At end of incubation, ten chicks from each group were decollated and internal organs were separated and weighed. The obtained data were analyzed by SAS software and unpaired t-test was applied for detection of significant differences.

RESULTS

Data obtained from weighting separated internal organs of newly hatched chicks were presented as table1. Group 2 (100 ng/egg ghrelin) had significantly greater intestine weight in comparison with control (intact) group. There were no any significant difference between groups for pectoral muscle, preventriculus, gizzard, heart and liver.

Table 1. Weights (g) of internal organs of newly hatched chicks subjected to in ovo ghrelin administration at day-5.

<table>
<thead>
<tr>
<th>Groups</th>
<th>dosage/ injection time</th>
<th>liver</th>
<th>heart</th>
<th>proventriculus+gizzard</th>
<th>intestine</th>
<th>Breast muscle</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Control (without injection)</td>
<td>1.13</td>
<td>0.35</td>
<td>2.20</td>
<td>2.05 ±b</td>
<td>1.59</td>
</tr>
<tr>
<td>2</td>
<td>100 ng/egg day-5 of incubation</td>
<td>1.09</td>
<td>0.34</td>
<td>2.26</td>
<td>2.30 ±a</td>
<td>1.50</td>
</tr>
<tr>
<td>P value</td>
<td></td>
<td>1.131</td>
<td>0.102</td>
<td>0.831</td>
<td>0.013</td>
<td>0.061</td>
</tr>
<tr>
<td>SEM</td>
<td></td>
<td>0.015</td>
<td>0.008</td>
<td>0.134</td>
<td>0.047</td>
<td>0.034</td>
</tr>
</tbody>
</table>

- different letters (a or b) shows significant difference between groups.
- SEM: standard error of the mean.

DISCUSSION

The phenomenon in relation to avian ghrelin is discussed that ghrelin may transfer to egg internal material via oviduct during egg formation and has considerable role in developmental regulations at embryonic life (Kaiya et al., 2009). In other words, findings of Yoshimura et al., (2009) on maternal ghrelin in fertile eggs and Kaiya et al., (2009) suggests are according to similar reports on mammalian ghrelin (Nakahara et al., 2007), that show regulatory roles of ghrelin in mammalian embryonic development and organogenesis. In present study, given the bioactivity of endogenous ghrelin, the administrated exogenous ghrelin has no excessive effect on weights of liver, heart and pectoral muscle. Significant increase in intestine weight may be referred to ghrelin specific roles in gastrointestinal development, whereas these tissues are main source of ghrelin synthesis and releasing. In this regard, Kitazawa et al., (2007) had stated that ghrelin has considerable effect on gastrointestinal motility especially in gizzard and colon and has minor effect on intestine motility. Present study that suggests in ovo ghrelin effect on intestine and gizzard size is in agreement with Kitazawa et al., (2007) report. In the other relative reports in pigs, injection of 7.5 and 15 microgram ghrelin/ kg BW caused increasing gastric weight (Slupecka and Wolinski, 2007). Ghiasi gahleh-kandi et al., (2011) had reported increase in intestinal enzyme activity follow in ovo ghrelin administration. In present investigation, unlike Deprem and Gulmez (2007), in ovo injection of another growth stimulator peptide (ghrelin) was no cause of increase in chicken pectoral muscle size.

It seems that mentioned difference may be because of difference in physiological roles of these two peptides. In endocrinological term, IGF-1 is a post-GH, and ghrelin is pre-GH growth stimulator factor, in other words, ghrelin act is highly dependent to GH releasing, but IGF-1 is independent largely, when it administrated into eggs. Exogenous ghrelin may be able to stimulate development of chicken embryo, where endogenous ghrelin is sufficient for this critical act and cause optimum organogenesis.
REFERENCES


