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BASIC STUDY ON FEEDING REGULATORS USED TO INCREASE CHICKEN MEAT PRODUCTION EFFICIENCY.

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Background:
Glucagon-like peptide-1 (GLP-1) is synthesized from preproglucagon in the pancreas and intestines. [1),2)]. Its amino acid sequence shows it to be homologous to glucagon. Until recently, the only physiological roles attributed to it had been in the inducement of insulin production[3)-8)] and somatostatine secretions[9)]. Lately, GLP-1 has been linked to feeding behaviour[10-11)], and its receptor's existence in the brain has been established[10,12-14)]. Intracerebroventricular administration of GLP-1 has been found to decrease food intake in rats [10)] and chicks [15)]. At present, while all of the regulatory mechanisms involved in feeding have yet to be established, GLP-1 is believed to play an important role in regulating feeding behavior.
The importance of increasing the efficiency of food production goes without saying as the world's population continues to mushroom. It stands to reason that increasing an animal's appetite will also bring about faster gains in weight and increase production efficiency. To date, while there have been many reports dealing with the relation between carbohydrate and protein sources in feeds and their relation to feeding behavior, no useful regulators of feeding behavior have been discovered.

Objectives:
We designed this study in order to examine the effects of various types of protein on the feed intake of chicks, which are regarded as the low appetite animal model, which had been given chicken GLP-1 via an intracerebroventricular route.

Methods:
Single-comb White Leghorn male chicks were used. Room temperature was maintained at 35℃ for the first 7days, then reduced to 32℃ after the 8th day. A 12hr cycle of light (06:00-18:00) and dark was used and water and feed were provided *ad libitum*.
Synthesized chicken GLP-1 (7-36), prepared on the basis of the previous study[18)] was purchased from the Peptide Institute, Inc. (Osaka, Japan). GLP-1 was dissolved in Evans Blue solution, which was prepared in a 0.85% saline solution (Japan Millipore Co. Ltd., Tokyo, Japan).
In Experiment 1, chicks were fed a commercial starter diet (Nihon Nosan Kogyo Co. Ltd., Tokyo, Japan) for10 days. Then, each chick was fixed in a device [17)] and injected with 10 μ l of the solution using a microsyringe. Five concentrations (0, 0.03, 0.1, 0.3, 1.0 μ g) of chicken GLP-1 were injected by the intracerebroventricular route and chicks were immediately started on the same diet. Food intake was determined at 120min after the GLP-1 injection.
In Experiment 2, chicks were distributed into five groups: a group supplied with a 18% soybean protein diet (S); a group supplied with a 18% beef diet (B); a group supplied with a 18% egg diet (E); a group supplied with a 18% casein diet (C); and a group supplied with a 18% wheat protein diet (W). Chicks were fed on their respective diets from the day after birth to 10 days of age. After being fed their respective rations, chicks were injected via the intracerebroventricular route with 0.1 μ g of chicken GLP-1 in saline solution and immediately given each diet. Food intake was measured at 120min after injection. At the end of the experiments, chicks were killed by decapitation, followed by brain sectioning to identify the location of the drug injection. Data regarding the chicks in which the presence of Evans Blue dye in the lateral ventricle could not be verified were deleted. Blood collected from the neck following decapitation was placed into a heparinized tube and centrifuged at 2,000 × g for 10 minutes within 30 minutes after collection. The plasma sample was stored at 4℃ prior to analysis. Plasma glucose was measured with a commercially available kit (Glucose CII-Test Wako, Wako Pure Chemical Ind. Ltd., Osaka). Plasma nonesterified fatty acid (NEFA) was measured with a commercially available kit (NEFA-Test Wako, Wako Pure Chemical Ind. Ltd., Osaka).

Results and discussion:
In Experiment 1, injections of the 0.3 and 1.0 μ g concentrations of GLP-1 greatly decreased the food intake of chicks (data not shown). However, injections of the 0.03 μ g concentration of GLP-1 were not enough to cause a decrease in food intake. Based on this finding, a 0.1 μ g concentration of GLP-1 was used in Experiment 2.
In Experiment 2, in light of the resultant gains in body weight and increase in feed consumption , soybean protein was found to be the most effective and liked by the chicks compared to the other proteins sources used in this study (Table 1). Casein and wheat protein did not appear to appeal to the chicks.
As shown in Table 2, the administration of GLP-1 (0.1 μ g / chick) was enough to decrease the feed intake of chicks irrespective of the dietary groups to which they belonged. Recovery from the inhibition of food intake induced by GLP-1 administration was fastest in the soybean group and slowest in the wheat group. The differences in the recovery times between the soybean group and wheat group may have been caused by the feed preferences of the chicks observed during the feeding period. In addition, recovery of food intake tended to be slower in the beef group. The plasma glucose concentration was significantly lower after the administration of GLP-1 in all groups aside from the beef group. The average plasma glucose concentration in the beef group was lower than in any other group, however the concentration increased to where it exceeded values in all other groups after the GLP-1 injections. The plasma NEFA concentration was significantly higher after GLP-1 administration in all groups with the exception of the casein group,

however, the concentration in the casein group tended to be midrange in all groups. Thus, chicks in groups other than the beef group expressed starvation symptoms, low glucose concentrations, and high NEFA concentrations in plasma.
It may be surmised that the inhibition of food intake is influenced by the taste-appeal of the feed, feed constitution, and other factors besides GLP-1. In this study, changes in plasma glucose levels in chicks induced by GLP-1 injections showed a different pattern in the beef group. One of the reasons for this might be the low plasma glucose concentration observed in the beef group throughout as compared with the other protein source diets, whereby the intracerebroventricular administration of GLP-1 have caused transient hypoglycemia. Therefore, a different type of metabolic action might have occurred in the beef group, which could have affected the feed regulating factors.

Conclusions:
These results suggest that using beef as a protein source in feeds could possibly control the appetite of chicks via influence on energy metabolism. Further studies are necessary to clarify this mechanism prior to its application to meat production.

Pertinent literature:

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Table 1 Effects of dietary protein types on body weight gain and total food intake.

	S	B	E	C	W
Body weight of 8 days (g)	57.2	49.0	52.7	46.7	37.9
Total food intake(g/8days)	835	515	530	315	275

Value are Means. S: soybean protein, B: beef, E: whole egg, C: casein, W: wheat protein.

Table 2 Effects of dietary protein types on food intake, plasma free fatty acid and glucose of chicks administered intracerebroventricularly with chicken glucagon-like peptide-1.

		S	B	E	C	W
Food intake	Sa	1.855±0.12	1.220±0.42	2.190±0.12	0.893±0.14	1.033±0.08
(g/2hour)	G	0.785±0.15**	0.087±0.01*	0.185±0.06**	0.163±0.08*	0.040±0.01**
plasma NEFA	Sa	14.48±1.79	31.09±1.77	32.77±3.09	43.38±3.29	19.86±0.98
(μEq/dl)	G	50.16±5.95**	46.70±3.24**	61.47±4.25**	51.72±3.18	67.67±14.0**
plasma glucose	Sa	268.5±2.4	226.0±3.2	266.0±4.5	270.4±5.4	227.3±8.0
(mg/dl)	G	212.3±3.4**	261.1±11.9	222.8±8.7**	189.0±19.9*	129.9±2.4**

Value are means±SEM. S :soybean protein, E :whole egg, B :beef, C :casein, W :wheat protein.
Sa:administered with saline. G :administered with chicken GLP-1 (0.1μg per chick).
NEFA:nonesterified fatty acid.
Significance compared with saline administration (*:$p<0.05$, **:$p<0.01$).