Comparison of oral administration of fructose and glucose on food intake and physiological parameters in broiler chicks

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ABSTRACT Like glucose, fructose is a monosaccharide, but the mechanisms of its absorption and metabolism in the body are very different between the 2 molecules. In this study, we investigated the effects of oral administration of glucose and fructose on food intake, diencephalic gene expression, and plasma metabolite concentrations in broiler chicks. The animals used in this study were 4-day-old male broiler chicks (Ross 308). They were given glucose, fructose (200 mg/ 0.5 mL/ bird), or a similar volume of distilled water orally after 6 h fasting. After treatment, measurements of food intake (at 0, 30, and 60 min), and blood glucose as well as insulin concentrations were measured over time; however, diencephalic (hypothalamus) gene expression and plasma metabolites were measured at 30 min. The results showed that glucose administration suppressed food intake, but fructose administration did not suppress food intake and it was at the same level as distilled water administration. In addition, fructose administration did not increase plasma glucose and insulin levels as did glucose administration. In the diencephalon, expression levels of genes related to the melanocortin system were unaffected by the treatment, while gene expression levels related to intracellular energy regulation, such as AMP-activated protein kinase were affected by the glucose treatment in the fasted chicks. These results suggest that fructose administration does not suppress feeding behavior as a result of possible reduction in the energy levels in the diencephalon and associated energy metabolism.

Key words: monosaccharides, chick, food intake, gene expression, insulin

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INTRODUCTION

Many monosaccharides are naturally present in fruits, however, the level of fructose is higher (Klasing, 1998). Fructose is a monosaccharide similar to glucose. When fructose is taken orally, it is absorbed in the small intestine and transported through the portal vein to the liver where it is metabolized (Hallfrisch, 1990). Absorbed fructose is transported through a facilitated diffusion depending on concentration gradient by glucose transporter-5 (**GLUT5**) in the digestive organs. On the other hand, after absorption in the small intestine, glucose is transported with Na+-dependent glucose transporter (**SGLT**) by active biological transport (Garriga et al., 2004). In the liver, the metabolism of fructose is different from glucose. Glucose becomes fructose-1,6-bisphosphate by insulin-dependent glucokinase and phosphofructokinase and enters the glycolysis, whereas fructose becomes fructose-1-phosphate by insulin-independent fructokinase and is integrated to glycolysis (Franz, 1990; Laia et al., 2011).

In mammals, fructose has a different absorption and metabolic mechanism than glucose. Therefore, fructose causes specific effects in mammals that are different from those of glucose. In particular, there are many reports of the differential effects of glucose and fructose on feeding behavior in mammals (Miller et al., 2002; Jurgens et al., 2005; Cha et al., 2008; Lindqvist et al., 2008; Page et al., 2013; Luo et al., 2015). However, there is no report about the effect on feeding behavior of fructose in poultry.

The aim of present was to examine the effect of oral injection of fructose and glucose on the food intake in broiler chicks. The objective is also to determine the

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differential effects of both monosaccharides on chickens' feeding, brain gene expression, and plasma parameters.

MATERIALS AND METHODS

Animals

All experimental procedures were complied with in accordance with Law No. 105 and Notification No. 6 of the Japanese government and approved by the Animal Experiment Committee of Hiroshima University (authorization No. C19-15).

Day-old male broiler chicks (Ross 308) were obtained from a local hatchery (Fukuda Hatchery, Okayama, Japan). Birds were maintained in a room with 24-h lighting and at a temperature of 30°C. They were given free access to a commercial starter diet (Nichiwa Sangyo Co. Ltd., Kobe, Japan) and water during the pre-experimental period. Chicks (3-day-old) were distributed into experimental groups based on their body weight so that the average body weight was uniform across the treatments. The birds were reared individually in experimental cages (17 × 24 × 24 cm) up to the time of experiments.

Experiment 1. Effects of Oral Administration of Monosaccharide on Food Intake

Oral administration was performed as described elsewhere (Erwan et al., 2014), and the solutions (500 μL) were administered using a 2.5 mL syringe with an elastic-plastic needle. D-Fructose and D-glucose (Sigma, St. Louis, MO) were dissolved in distilled water. Each chicks (4-day-old) were gavaged once only with a similar dose (200 mg/0.5 mL/ bird) of fructose or glucose solution or distilled water (0.5 mL/ bird) as control after 6-h of fasting. Birds were allowed to feed freely for 1 h immediately after each treatment. Food intake was measured by observing the decrease in the amount of preweighed feed at different times (0 min, 30 min, and 60 min). The weight of the food was measured using an electric digital balance with an accuracy of 1 mg.

Experiment 2. Effects of Oral Administration of Monosaccharides on Plasma Glucose and Insulin

Similar to Experiment 1, six hours fasted chicks (4-day-old) were gavaged once only with a similar dose (200 mg/0.5 mL/ bird) of fructose or glucose solution or distilled water (0.5 mL/ bird). Chicks were not provided food after oral administration of monosaccharides. At 15, 30, or 60 min after oral administration, each chick was anesthetized with isoflurane (FUJIFILM Wako Pure Chemical Corporation, Osaka, Japan) and bled by cardiac puncture. Blood was collected into heparinized tubes and centrifuged for 15 min. Harvested plasma was stored at −20°C until assayed. Furthermore, blood samples for ad libitum feeding and fasted birds without administration of glucose or fructose were also collected as comparisons.

Analysis of Plasma Glucose and Insulin

The commercial kit (Glucose CII-Test Wako, Wako Pure Chemical Industries Ltd., Osaka, Japan) was used for the measurement of plasma glucose concentration. The plasma insulin assays were performed according to the previous report (Takahashi et al., 2006; Shiraishi et al., 2011). In brief, chicken insulin concentration was measured by competitive solid-phase immunoassay using Europium (Eu)-labeled chicken insulin in polystyrene microtiter strips (Nunc-Immuno Modules, Thermo Fisher Scientific, Waltham, MA) coated with anti-guinea pig γ-globulin. Chicken insulin (50 μg/100 μL 10 mM bicarbonate saline, pH 9.8) was labeled according to the manufacturer's instructions (Perkin Elmer, Waltham, MA). A diluted antiserum against human insulin (1:20,000) was incubated in each well overnight. After washing the insulin antibody, serial diluted chicken insulin standard (0.1−100 ng/mL) and insulin in chick plasma dissolved in assay buffer was incubated in the wells overnight. EU-labeled insulin was distributed in each well and incubated for 3 h. After washing, 100 μL of the enhancement solution was added to each well and fluorescence in each well was measured with a time-resolved fluorometer (Multilabel Counter, 1420 ALVO, Perkin Elmer). The curve was linearized by the method of least squares on logarithmic amounts of B/B0.

Experiment 3. Hypothalamic Gene Expressions, and Blood Parameters

After being deprived of food for 6 h, each chick (4-day-old) was injected once only with a similar dose (200 mg/0.5 mL/ bird) of fructose or glucose solution or distilled water. After 30 min of oral administration, all chicks were anesthetized with isoflurane (FUJIFILM Wako Pure Chemical Corporation), and blood was collected into heparinized tubes and centrifuged for 15 min. Immediately after blood collection, they were decapitated, and their diencephalons including the hypothalamus were also collected. Harvested plasma was stored at −20°C until assayed. Diencephalic tissue samples were collected and snap frozen in liquid nitrogen and stored at −80°C prior to RNA isolation. The diencephalon was dissected according to the brain atlas of chicks (Kuenzel and Masson, 1988).

Isolation of Total RNA and Quantitative Real-time PCR

RNA was isolated from collected diencephalic tissue samples using Trizol reagent (Thermo Fisher Scientific) and manipulated according to the manufacture's instructions. Isolated total RNA was measured for concentration and purity by nanodrop spectrophotometry

Table 1. Oligonucleotide primer sequences for real-time PCR.

Primer	Forward (5' → 3')	Reverse (5' → 3')	Accession No.
RPS17	AAGCTGCAGGAGGAGGAGAGG	GGTTGGACAGGCTGCCGAAGT	NM_204217
NPY	GGCACTACATCAACCTCATC	CTGTGCTTTCCCTCAACAA	NM_205473
AgRP	AGGCCAGACTTGGATCAGATG	ACTCCAGGAGGCGGACAC	NM_001031457
POMC	ATGGAGCATTTCCGCTGG	TAACTCTCAGCCGACTCCTCGT	NM_001031098
CART	CCGCACTACGAGAAGAAG	AGGCACTTGAGAAGAAGG	BI394769
AMPK	ACATGAATGCGAAGATAGCTGAT	AGCCTTCCAGAGATGACTTCAG	NM_001039603
CPT-1	ACGTCTTTACCACGATTGCC	TTTTGCAAGACCCTCCATTC	NM_001012898
CPT-2	GAAGACCTTCAGGGCTGGTTA	CAAGAGAGTGACGAAGGCACA	NM_001031287
PPARα	GCCATCATTTGCTGTGGAG	CGTCAGGATGGTTGGTTTG	NM_001001464
MCD	GTTGATATGAAGCGTCGAGTGG	TGCTGCTGGAGATATCACTGGT	NC_006098
HADH	GATGAAGCTTGTGGAGGTTGT	CCTGGAGTATCCTTACAACTGACA	NM_001277897

Abbreviations: AMPK, AMP-activated protein kinase; AgRP, agouti-related peptide; CPT-1, carnitine palmitoyltransferase 1; CPT-2, carnitine palmitoyltransferase 2; HADH, hydroxyacyl-CoA dehydrogenase; MCD, malonyl-CoA decarboxylase; NPY, neuropeptide Y; POMC, pro-opiomelanocortin; PPARα, peroxisome proliferator-activated receptor α; RPS17, ribosomal protein S17.

(Thermo Fisher Scientific), and then inactivated for genomic DNA in the sample using DNase I (Thermo Fisher Scientific). Reverse transcription of 500 ng of DNase-treated total RNA was performed using 10μL of 1 × Prime Script RT Enzyme Mix I (Takara, Tokyo, Japan) at 42°C for 15 min. The reactants were used for real-time PCR using a light cycler system (Roche Applied Science, Penzberg, Upper Bavaria, Germany). Real-time PCR reactions were performed using a denaturation step at 95°C for 10 s followed by a thermal protocol of 95°C for 5 s and 60°C for 20 s. 1 × SYBR Premix EX Taq (Takara) and 20 μL buffer containing 0.2 μM of each primer. The primers used for real-time PCR in this study are listed in Table 1. For normalizing the data, ΔC_T was calculated for each sample by subtracting the C_T of RPS17 from the C_T of the target gene. For relative quantitation, ΔC_T for the defined control group was subtracted from the ΔC_T of each experimental sample to generate $\Delta\Delta C_T$. The $\Delta\Delta C_T$ was then used to calculate the approximate fold difference, $2^{-\Delta\Delta CT}$. The results were expressed as the gene of interest mRNA/RPS17 mRNA ratio.

Plasma levels of glucose, non-esterified fatty acid (**NEFA**), total cholesterol (**T-CHO**), triglycerides (**TG**), low-density lipoprotein cholesterol (**LDL**), high-density lipoprotein cholesterol (**HDL**), and lactic acid (**LA**) were determined by Beckman Coulter AU480 automatic biochemistry analysis system (Brea, CA), with reagent kits provided by the manufacturer.

Statistical Analysis

To analyze food intake behavior data, statistical calculations were carried out using a repeated measure two-way analysis of variance (**RM-ANOVA**), using the commercially available package, Stat View (Version 5, SAS Institute, Cary, 1998). This analysis provides *P*-values for differences between treatment, differences over time, and the interaction of treatment with time. All experiments were performed once. When a significant interaction of treatment and time was documented, a post hoc test was used to compare the significance among means at each time point. For comparisons among means of each mRNA level and plasma parameters, one-way ANOVA was done. The Tukey-Kramer test was used to determine overall statistical significance due to treatment. Differences were considered to be significant when P was less than 0.05.

RESULTS

Effects of Oral Administration of Monosaccharides on Food Intake

Figure 1 shows the effect of oral administration of fructose or glucose on food intake in fasted chicks. Although the main effect of treatment was not significant ($P = 0.069$), an interaction between treatment and

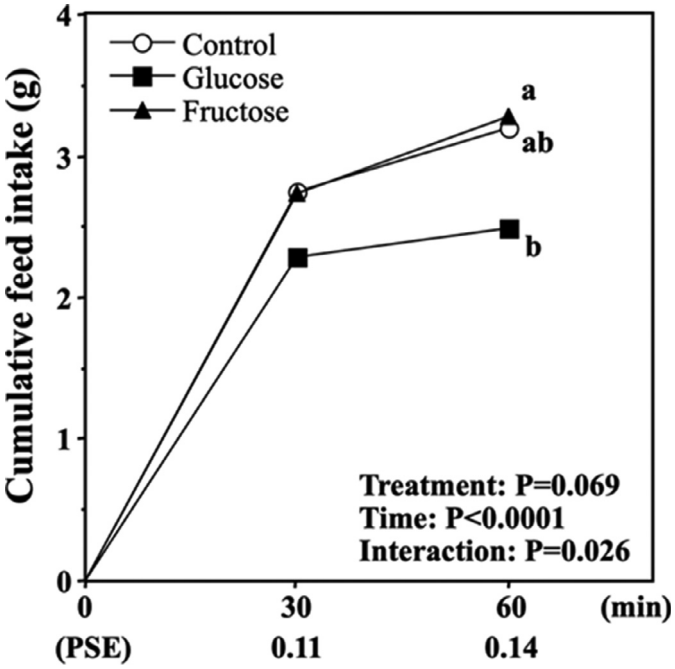

Figure 1. Cumulative food intake of chicks administered orally distilled water, glucose or fructose. PSE, pooled SE. Values are means for each group of 6−7 birds. Means with different letters at each time are significantly different at $P < 0.05$. The white circle show the results for the control group, the black squares show the results for the glucose treated group, and the black triangles show the results for the fructose treated group.

time was significant (P = 0.026). A post hoc test did not detect any difference at 30 min post-administration, but glucose significantly suppressed food intake at 60 min when compared with control or fructose. Food intake was significantly ($P < 0.0001$) increased in all groups with time.

Effect of Oral Administration of Monosaccharides on Levels of Plasma Glucose and Insulin

The effects of monosaccharides on the levels of plasma glucose in fasted chicks after the 15-, 30-, and 60-min oral administration are shown in Figure 2A. The concentrations of plasma glucose in control chicks (distilled water) were significantly lower than that in ad libitum fed chicks at each time point ($P = 0.012$). Glucose administrations caused a rapid increase in plasma glucose concentrations at 15 min ($P = 0.025$; compared with ad libitum fed and fasted chicks), followed by a gradual decrease at 60 min. Although the glucose levels in the fructose group were higher at 30 min after injection ($P = 0.037$) than that in the 6-h fasted group without administration, it was lower in the fructose administered chicks when compared to the ad libitum fed chicks at 15 and 60 min ($P = 0.027$).

Figure 2B shows the effect of monosaccharides on levels of plasma insulin in fasted chicks. In the control group (distilled water administration), there was a significant decrease in plasma insulin compared to the ad libitum fed group at 60 min (P = 0.038), but not 15 and 30 min. As for the glucose-treated chicks, almost similar to the levels of plasma glucose (Figure 2A), the treatment caused a rapid increase in concentrations at 15 and 30 min ($P = 0.021$; compared with ad libitum fed and

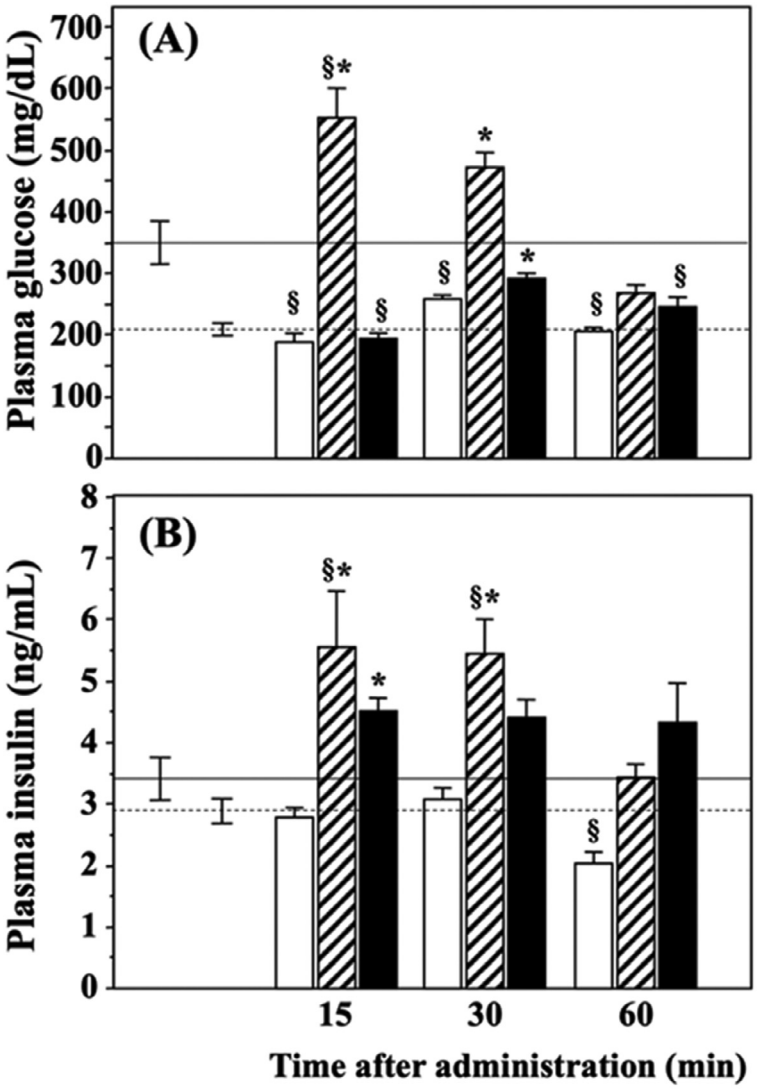

Figure 2. Levels of plasma glucose (A) and insulin (B) in chicks administered orally distilled water (control), glucose or fructose. The vertical line indicates the means with error bars (SEM) for the ad libitum (solid line) and fasted (dashed line) groups. Blank bars: control (distilled water) group, shaded bars: glucose group, blackened bars: fructose group. Each bar value represents mean ± SEM for each group of 6−7 birds. *: $P < 0.05$ compared with fasted group, §: $P < 0.05$ compared with ad libitum group.

fasted chicks), and then the level gradually decreased at 60 min. Oral injection of fructose also facilitated the increase of plasma insulin at 15 min ($P = 0.034$) but there were no significant differences with the 6-h fasted group without administration after 30 min post-administration.

Effects of Oral Administration of Monosaccharides on Hypothalamic Gene Expressions

Figure 3 shows the effect of monosaccharides on hypothalamic gene expressions after the 30-min oral administration. Both monosaccharides did not influence pro-opiomelanocortin (POMC; $P = 0.229$), neuropeptide Y (NPY; $P = 0.515$), agouti-related peptide (AgRP; $P = 0.606$), and cocaine- and amphetamine-regulated transcript (CART; $P = 0.347$) mRNA levels but the gene expression levels of AMP-activated protein kinase (AMPK; $P = 0.014$), malonyl-CoA decarboxylase (MCD; $P = 0.021$), peroxisome proliferator-activated receptor α (PPARα; $P = 0.003$), hydroxyacyl-CoA dehydrogenase (HADH; $P = 0.019$), carnitine palmitoyltransferase 1 (CPT-1; $P = 0.027$), and carnitine palmitoyltransferase (CPT-2; $P = 0.012$) in glucose-treated chicks were lower than those in control and/or fructose-treated chicks.

Effect of Oral Administration of Monosaccharides on Blood Parameters

The effects of monosaccharides on blood parameters in chicks after the 30-min oral administration are shown in Table 2. The levels of NEFA and LDL in both monosaccharide-treated chicks were significantly ($P < 0.001$) lower than that in control chicks. As for the concentration of plasma HDL, there was no significant difference between monosaccharide treatments and control but the level in glucose-treated chicks was lower than that in fructose-treated chicks ($P = 0.032$). The plasma TG

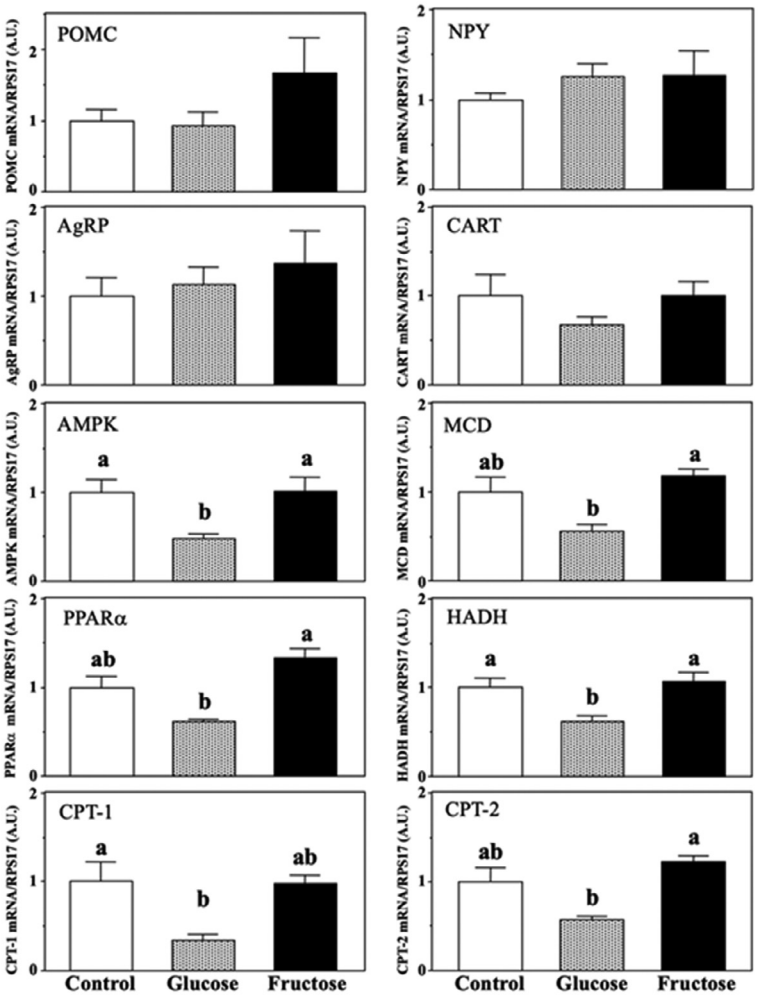

Figure 3. Levels of hypothalamic mRNA expression in chicks as determined by quantitative RT-PCR. Shaded bars: glucose group, blackened bars: fructose group. The values represent mean ± SEM for each group of 5−6 birds. Means with different letters are significantly different at $P < 0.05$. Abbreviations: AgRP, agouti-related peptide; AMPK, AMP-activated protein kinase; CPT-1, carnitine palmitoyltransferase 1; CPT-2, carnitine palmitoyltransferase 2; HADH, Hydroxyacyl-CoA Dehydrogenase; NPY, neuropeptide Y; MCD, malonyl-CoA decarboxylase; PPARα, peroxisome proliferator-activated receptor α; POMC, Pro-opiomelanocortin.

Table 2. Effect of oral administration of monosaccharides on plasma parameters.

Metabolites	Control (n = 6)	Glucose (n = 5)	Fructose (n = 5)	*P*-value
NEFA (mEq/L)	145.8 ± 7.03[a]	86.2 ± 10.26[b]	86.7 ± 8.51[b]	<0.001
T-Cho (mg/dL)	116.9 ± 5.64	100.2 ± 5.10	114.1 ± 6.03	NS
TG (mg/dL)	98.0 ± 3.61[a]	155.8 ± 8.43[b]	111.3 ± 12.07[a]	<0.001
HDL (mg/dL)	112.1 ± 6.28[ab]	103.4 ± 4.33[a]	126.3 ± 4.19[b]	<0.05
LDL (mg/dL)	264.2 ± 29.96[a]	116.7 ± 26.21[b]	111.2 ± 12.31[b]	<0.001
LA (mg/dL)	85.1 ± 7.57[a]	125.6 ± 15.97[ab]	131.8 ± 12.81[b]	<0.05

Abbreviations: HDL, high-density lipoprotein cholesterol; LDL, low-density lipoprotein cholesterol; LA, lactic acid; NEFA, non-esterified fatty acid; NS, not significant; T-Cho, total cholesterol; TG, triglycerides.

Means with different letters are significantly different at $P < 0.05$.

[ab]Values are means ± SEM of the number of chicks in parentheses.

concentration in glucose-treated chicks was higher than that in control and fructose-treated chicks ($P < 0.001$). On the other hand, the level of LA in fructose-, but not glucose-treated chicks were higher than that in control chicks ($P = 0.030$). No significant difference between treatments was observed in plasma T-Cho concentration.

DISCUSSION

In mammals, it is suggested that the fructose metabolism in the central nervous system (**CNS**) regulating feeding behavior and the energy consumption is different from glucose (Cha et al., 2008). Fructose is metabolized mainly in the liver, and kidney (Funari et al., 2005). Fructose is catalyzed by several kinds of proteins including glucose transporter (GLUT2, GLUT5) and phosphofructokinase in glycolysis, and it follows a specific metabolic pathway in the liver, kidney, and small intestine (Funari et al., 2005; Cha et al., 2008). Recently, it has been revealed that these transporters and enzymes exist in the CNS, and that fructose is used as energy source in the mammalian brain (Funari et al., 2005; Funari et al., 2007). However, it is reported that fructose and glucose have different effects on feeding behavior derived from the difference in metabolism. In the experiment of central administration to mice or rats, fructose promoted feeding behavior whereas glucose inhibited it (Miller et al., 2002; Wolfgang, 2007). The present study reveals differences in the central feeding regulation mechanisms of peripheral administration of fructose and glucose in birds (domestic fowl).

The results of food intake in this study confirmed the effect of glucose administration on the suppression of food intake, but showed no effect of fructose administration. As mentioned above, oral fructose administration suppresses food intake in mammals. Feeding behavior is regulated by several pathways. Feeding behavior is often influenced by the energy status of the body. Under excessive energy, ATP level increases and AMP level decreases in the nerve cells. AMP kinase (**AMPK**) is inactivated by decreasing of AMP level because AMP is the activator of AMPK. Inactivation of AMPK causes the inactivation of Acetyl CoA carboxylase (**ACC**) and increasing malonyl CoA. Malonyl CoA increasing causes inactivation of orexigenic neuropeptide expression, it follows that anorexigenic neuropeptide expression increases. It seems that these pathways relate inhibition of feeding behavior by glucose (Loftus et al., 2000; Hu et al., 2003; Gao et al, 2007; Wolfgang, 2007; Cha et al., 2008). When fructose is administered, however, the reverse reaction is caused. Fructose that enters the cell is in the specific pathway through GLUT5 and phosphofructokinase. Fructose uptake causes that intracellular ATP dries up rapidly and AMPK activates. Eventually lead to increasing orexigenic neuropeptide expression (Cha et al., 2008; Lane and Cha, 2009). In this study, AMPK gene expression in the diencephalon of chicks treated with oral glucose was lower than that of control chicks, whereas AMPK gene expression in the diencephalon of chicks treated with oral fructose was at the same level as that of controls. These results suggest that glucose administration increases the amount of ATP and decreases the amount of AMP in the CNS, whereas fructose administration did not cause an increase in ATP or a decrease in AMP as glucose administration. Thereby, no feeding inhibition was observed with fructose administration to chicks. The results of this study showed that the gene expression of AMPK, CPT1, CPT2, MCD, PPARα, and HADH in the diencephalon were higher in the fructose-treated group than in the glucose-treated group. Activated AMPK regulates ATP consumption and activates the glycolytic system and fatty acid oxidation to promote ATP generation (Minokoshi et al., 2002, 2004; Andersson et al., 2004; Kubota et al., 2007). In other words, the activation of AMPK phosphorylates ACC and regulates its activity. As a result, malonyl CoA, a product of acetyl-CoA, is reduced and its inhibitory effect on CPT is lifted, thus increasing CPT activity and fatty acid oxidation (Minokoshi et al., 2002; Hardie, 2007; Towler and Hardie, 2007). In addition, α2AMPK, one of the subunits that make up AMPK, is translocated into the nucleus when AMPK is activated and enhances PPARα expression (Suzuki et al., 2007). MCD converts malonyl-CoA back to acetyl-CoA, and its activity is regulated via AMPK in the liver and skeletal muscle (Saha et al., 2000; Park et al., 2002). Similarly in the CNS, it is assumed that MCD is regulated via AMPK (Blanco Martínez de Morentin et al., 2011). HADH is responsible for β-oxidation of fatty acids, which is transported into the mitochondria by CPT1, and conversion to acetyl CoA (Ramanathan and Ibdah, 2022). This suggests that AMPK is activated in the fructose-treated

group, and ACC is phosphorylated in response to the fructose-induced decrease in CNS ATP, resulting in a more active oxidation of fatty acids and production of ATP in the fructose-treated group than in the glucose-treated group.

Glucose acts directly on the liver to inhibit feeding in layer type chickens (Shutlock and Forbes, 1981; Lacy et al., 1985). However, the effect of glucose was not found in broiler type chickens (Lacy et al., 1985). In this study, oral administration of glucose suppressed food intake in broiler chicks. The broilers used in this study were ROSS strains, which are commonly used in commercial poultry industry today. The studies that examined the feeding suppression effects of glucose mentioned above are about 40 yr old, and the broilers used were New Hampshire and others. Although commercial broiler chickens such as ROSS are said to be based on New Hampshire and others, the physiological characteristics of present broilers do not necessarily match those of the 1990s exactly, just as the characteristics of broilers changed dramatically during the breeding and selection process from the 1950s to the 1990s (Havenstein et al., 1994; Denbow, 1999). A recent study reported that insulin suppresses appetite in 5-day-old ROSS strain broilers via NPY1 and 2 receptors (Yousefvand et al., 2020). Insulin acts to NPY and POMC neurons which are the core of the melanocortin system and regulate feeding behavior (Shiraishi et al., 2008). After glucose administration, insulin, which is secreted accompanying a rise in plasma glucose level, seems to cause inhibition of feeding behavior. However, it seems that fructose does not cause inhibition of feeding behavior by insulin because fructose does not raise plasma glucose levels (Hallfrisch, 1990; Sharon et al., 2002). In fact, in this study, the expression levels of NPY, POMC, and AgRP genes in the diencephalon related to the melanocortin system were similar between the fructose-treated group and the control group. In this study, the glucose treated group showed larger and longer increases in blood glucose and insulin levels than the control and fructose treated groups, which may be one reason for the suppression of food intake in the glucose treated group. Insulin activates lipid synthesis in the liver, and in young chickens, the majority of lipids are synthesized in the liver (Griffin et al., 1992). Lipids synthesized by the liver are released into the blood as triglycerides (**TG**) and transported throughout the body (O'Hea and Leveille, 1968). In this study, NEFA concentrations were lower in the glucose and fructose treated groups than in the control, and in addition, TG concentrations were higher in the glucose treated group than in the others. A previous study also reported that fructose oral administration treatment reduced plasma FFA concentrations in chickens (Komaki et al., 2017). These plasma parameter results are considered to be the result of increased insulin concentrations due to the treatments, in other words, the increase in insulin concentration results in the uptake of NEFA into the liver and activation of TG synthesis in the liver. Since signaling from peripheral organs such as the liver to the central nervous system play an important role in controlling feeding, it is necessary to study the effects of glucose or fructose on the peripheral tissues of chickens in the future.

Fructose absorbed from the digestive tract is transported by the bloodstream to the liver, where it is converted to fructose 1-posphate by fructokinase. The activity of fructokinase is not affected by insulin. Furthermore, fructokinase has no negative feedback mechanism (Fox and Kelley, 1972; Johnson et al., 2009; Khitan and Kim, 2013). Therefore, it is inferred that fructose ingestion causes a continuous supply to the glycolytic system. It is assumed that in this study, the fructose-treated group showed higher plasma lactate levels than the control group, as a result of higher activation of the glycolytic system in the fructose-treated group and the resulting pyruvic acid breakdown. On the other hand, the glucose group did not show higher lactate levels than the control group. In addition, it is reported that 37% of glucose ingested from the diet is converted to lactate during absorption in the intestinal wall (Riesenfeld et al., 1982). This suggests that the high plasma lactate level in the fructose-treated group is the result of energy requirements during fructose absorption in the intestine as well as in the glycolytic system in the liver.

In conclusion, oral administration of fructose in chickens does not induce the same feeding depression as glucose administration, presumably as a result of a fructose-specific mechanism of energy uptake into cells rather than as a result of a dependence on insulin signaling. The results are expected to be useful for the use of fructose-rich fruits and other food wastes in bait. However, high fructose intake has been reported to produce large amounts of uric acid in the body in mammals (Choi et al., 2008), and in chickens, chronically high plasma uric acid levels cause abnormal nail morphology, precipitation of sodium urate in joint synovial fluid, and kidney damage (Hong et al., 2020). When preparing diets for chickens using high fructose-containing food wastes, it is desirable to evaluate it not only by a single fructose feeding test as in this study, but also a long-term feeding trial is needed.

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Authorship Contribution Statement: YO collected the data used in this study, and drafted the manuscript. YK, KS and NF collected the data used in this study, analyzed the data. TS and JS analyzed the data and discussed the data. VC discussed the data and contributed to the development and writing. TB designed the study and analyzed the partial data. YO and TB interpreted the data and reviewed and improved the manuscript.

DISCLOSURES

The authors declare no conflict of interest.

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