

Effect of dietary arginine on organ weight and feed intake in Japanese quail (*Coturnix japonica*)

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SUMMARY

This study showed the effects of different dietary arginine levels on organ weight and feed intake in growing Japanese quail. Quails were provided with diets containing low, control, or high arginine for a fourteen-day experimental period. Dietary arginine levels had no significant effect on relative liver weight in both sexes, whereas low arginine intake was associated with increased relative brain weight. These findings suggest tissue-specific responses, with the liver exhibiting metabolic resilience and the brain being preferentially maintained under nutrient limitation. Feed intake was reduced under low dietary arginine during the second week, indicating sensitivity to arginine availability at this developmental stage. Variations in feed intake across studies suggest that arginine's effects are context-dependent, influenced by physiological status, environmental conditions, and baseline diet composition. Overall, our findings highlight the role of dietary arginine in organ weight and feed intake, reflecting both direct effects on organ development and indirect effects through feed intake regulation.

Keywords: Arginine, Japanese quail, organ weight, feed intake

INTRODUCTION

Adequate early nutrition is essential for optimal growth, and a deficiency leads to impaired development and affects overall fitness (Fidgett and Gardner, 2014). Amino acids are essential nutrient components influencing multiple physiological processes in birds. Adequate dietary amino acid intake is necessary for growth, reproduction, and development, whereas an amino acid imbalance can lead to nutrition-related disorders (Alagawany et al., 2021; Kidd et al., 2021). During diet formulation, species-specific amino acid requirements must therefore be carefully considered (Herring et al., 2021).

In birds, arginine plays a particularly important role as a precursor for nitric oxide, which is critical for avian physiology, protein synthesis, and metabolism (Wu, 2009), especially during the grower phase. Unlike mammals, birds lack a complete urea cycle and therefore cannot synthesise arginine endogenously, making it an essential dietary amino acid (Abdullah et al., 2019; Fathima et al., 2024; Khajali and Wideman, 2010). Supplementation of arginine has been shown to improve carcass traits and overall performance in Japanese quail (Al-Tamimy et al., 2025). Conversely, a deficiency of this limiting nutrient negatively impacts body weight, feed intake, and egg production. In particular, reduced dietary arginine intake has been directly associated with decreased body weight in Japanese quail (Sousa et al., 2022). Moreover, Reis et al. (2012) reported that individual supplementation of arginine does not enhance the performance of quail;

rather, its interaction with other amino acids, such as lysine, improves overall performance. In contrast, Morais et al. (2022) and Tuesta et al. (2018) found that certain arginine-to-lysine ratios do not significantly affect production traits in Japanese quail during the grower phase. These findings suggest a complex relationship, indicating that specific nutritional formulations may be required to adequately meet the metabolic needs of growing quail. In addition, arginine contributes to the synthesis of key biological molecules, which are essential for regulating blood flow and improving nutrient absorption, both of which are necessary for optimal growth in quail (Atakisi et al., 2009; Al-Daraji et al., 2012). Similarly, its importance in poultry nutrition is underscored by evidence showing that arginine supplementation can improve physiological performance, including enhanced feed conversion ratios (Tuba, 2015).

Although arginine is recognised as an essential amino acid for Japanese quail, the extent to which dietary arginine influences growth performance, organ development, and feed intake remains insufficiently understood. Previous studies have reported mixed findings: some have demonstrated improvements in carcass traits and nutrient utilisation with arginine supplementation, whereas others have shown limited or no benefit when arginine is provided alone or when its ratio with lysine is adjusted (Morais et al., 2022; Reis et al., 2012; Tuesta et al., 2018). In addition, arginine serves as a precursor for nitric oxide, polyamine, and creatine molecules essential for blood flow regulation, nutrient absorption, and cellular growth (Al-Daraji and

Salih, 2012; Atakisi et al., 2009; Wu, 2009). Given this background, we hypothesised that short-term adjustments in dietary arginine supply would influence early feed intake patterns and relative organ development in growing Japanese quail. Because both insufficient and excessive arginine can disturb metabolic balance during the rapid starter-growth phase, understanding these immediate physiological responses is essential for formulating appropriate diets. Therefore, evaluating how different arginine levels affect feed intake and organ weights over a short growth period is necessary to clarify previous inconsistencies and provide more accurate, species-specific nutritional guidance.

MATERIALS AND METHODS

Experimental animal management

The study was carried out at the Animal Experimental Farm of the Institute of Animal Science,

Biotechnology, and Nature Conservation, University of Debrecen, Hungary. Japanese quail were hatched on the farm using an industrial incubator (WQ-63 Model 2021 Version 2, AGROFORTEL, Budapest, Hungary) under standard conditions (Ndunguru et al., 2024). The birds were provided with unrestricted access to feed and water until they reached two weeks of age. During the first four days, infrared lamps maintained a rearing temperature of 37 °C, which was then gradually reduced by 3 °C every four days until reaching 24 °C at the end of the second week. The relative humidity in the cages was maintained and ranged from 60 to 65%. At the beginning of the trial, the room conditions were set at 24 ± 3 °C with 60–75% relative humidity and a 12:12 h light-dark cycle. The quails were offered a grower diet formulated from corn, wheat, corn germ meal, corn gluten meal, and fishmeal, providing 22% crude protein and 12.13 MJ/kg metabolisable energy (Table 1).

Table 1. Feed composition and calculated nutrient content of the experimental diets

Ingredients	Treatments (with inclusion rate%)		
	Control	Low arginine	High arginine
Corn	3.40	3.40	3.40
Wheat	30.00	30.00	30.00
Corn germ meal	39.19	39.19	39.19
Corn gluten meal	10.90	10.90	10.90
Fishmeal	3.74	3.74	3.74
Sunflower oil	8.08	8.08	8.08
Limestone	1.40	1.40	1.40
MCP	0.42	0.42	0.42
L-Lys	0.77	0.77	0.77
DL-Met	0.06	0.06	0.06
L-Thr	0.28	0.28	0.28
L-Trp	0.06	0.06	0.06
L-Ile	0.20	0.20	0.20
L-Arg	0.39	0	0.78
Inert (kaolin)	0.39	0.78	0
Salt	0.22	0.22	0.22
Premix ^a	0.50	0.50	0.50
Nutrient content (%)			
ME (MJ/kg)	12.13	12.13	12.13
Crude protein	22.00	22.00	22.00
Lys	1.30	1.30	1.30
Met	0.50	0.50	0.50
Thr	1.02	1.02	1.02
Trp	0.22	0.22	0.22
Leu	2.16	2.16	2.16
Ile	0.98	0.98	0.98
Arg	1.25	0.94	1.56
Leu/Ile	2.20	2.20	2.20
Ca	0.80	0.80	0.80
P	0.56	0.56	0.56
non phytate P	0.30	0.30	0.30
Na	0.15	0.15	0.15

^a The premix provided the following per kilogram of complete diet: 5000 IU vitamin A, 1000 IU vitamin D₃, 24.5 mg/kg vitamin E, 1 mg vitamin K₃, 0.75 mg vitamin B₁, 2.5 mg vitamin B₂, 6 mg Ca-d-Pantothenate, 2 mg vitamin B₆, 10 µg vitamin B₁₂, 55 µg biotin, 12.5 mg niacin, 0.3 mg folic acid, 1500 mg choline chloride, 66 mg Zn, 9.6 mg Cu, 48.1 mg Fe, 66 mg Mn, 0.9 mg I, 0.21 mg Se, 60 µg Co. MCP: monocalcium phosphate; ME: metabolisable energy.

Experimental design and sampling

When they were two weeks of age, 153 unsexed Japanese quail, each weighing approximately 53.66 ± 0.55 g, were selected and randomly assigned to three treatment groups. The experiment followed a randomised complete design with three treatments and three replicates, each containing seventeen birds. The dietary treatments included a control group: a diet formulated per NRC (1994) with a high-arginine group (control diet supplemented with 25% arginine above the recommended amount (1.56%)) and a low-arginine group (control diet restricted by 25% of arginine from the recommended amount (0.94%)) (Table 1). Quails were housed in groups in identical cages (40 cm × 50 cm × 40 cm; length × width × height) within the same facility to ensure consistent environmental conditions. Feed was provided in specially designed feeders to minimise spillage. The trial was conducted for 14 days (Reda et al., 2024). Daily fresh feed was replaced between 08:00 and 09:00, and daily feed intake was recorded.

At the end of the experiment, 48 birds (eight per treatment group, both male and female) were euthanised through cervical dislocation for tissue sampling, and liver and brain weights were measured using a digital balance (± 0.01 g accuracy).

Statistical analysis

Statistical analyses were performed using R v. 4.2.2 (R Core Team, 2024). Data visualisation was conducted using the 'ggplot2' package (version 3.4.3). To evaluate the effects of treatment, sex, and their interaction on relative liver and brain weight over the 14-day trial period, we used a linear model. Liver and brain weights were measured to assess the effects of

arginine in growing Japanese quail. The calculation of relative organ weight was calculated as relative organ weight = (organ weight/body weight) × 100 (Adomeh, 2024; Iheanacho et al., 2022). For relative liver and brain weight, treatment and sex were included as fixed factors. We used linear mixed models (LMMs) using the 'lmer' function from the 'lme4' package (Bates et al., 2015) to examine the effect of treatments and time period on feed intake, with cage number treated as a random effect. The significance of fixed effects was assessed using the 'lmerTest' package (version 3.1.3) to compute p-values using two-way ANOVA (Kuznetsova et al., 2017). Statistical significance was set at the 5% level.

Ethical approval

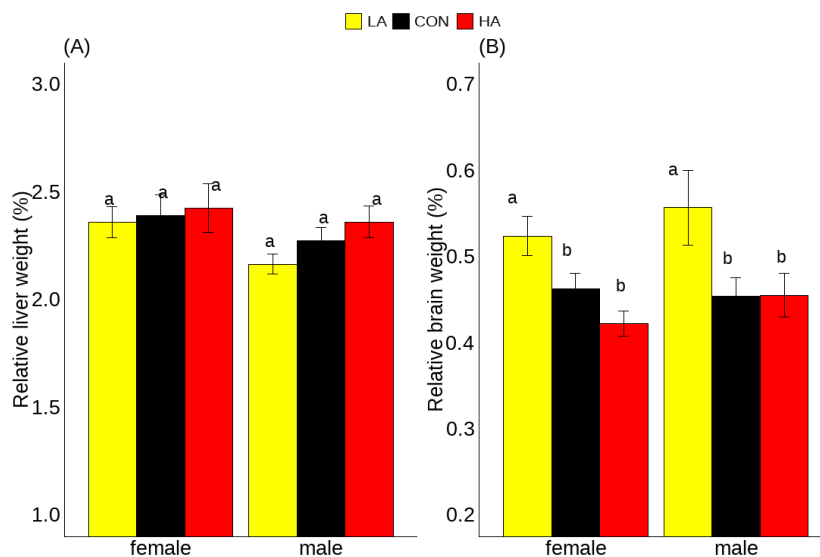
The experiment was carried out in accordance with EU Directive 2010/63/EU, which governs the use of animals for scientific research. Ethical approval was granted by the Animal Use Ethical Committee of the University of Debrecen, Hungary (Protocol No. 5/2021/DEMAB). All experimental procedures followed institutional rules and regulations. Reporting of this study was performed in line with the ARRIVE (Animal Research: Reporting of In Vivo Experiments) guidelines.

RESULTS AND DISCUSSION

Effect of treatment and sex on relative organ weight

Dietary arginine had no noticeable effect on relative liver weight (Figure 1A; Table 2), while low arginine intake was associated with higher relative brain weight in both sexes of growing Japanese quail (Figure 1B; Table 2).

Figure 1. The effect of treatment and sex on relative liver and brain weight



A.: Effect of treatment and sex on relative liver weight

B.: Effect of treatment and sex on relative brain weight

Data are means \pm s.e.m. from 8 birds per group and were analysed using ANOVA. Different letters indicate significant differences at a given time point at $p < 0.05$. LA, low arginine; CON, control; HA, high arginine.

Table 2. ANOVA for the effect of treatment, sex and their interaction

Effect	Df	F-value	P value
Treatment	2	1.326	0.276
Sex	1	3.689	0.061
Treatment: sex	2	0.337	0.715
Residuals	42		
Treatment	2	8.595	<0.001
Sex	1	0.790	0.378
Treatment: sex	2	0.433	0.650
Residuals	42		

Dietary arginine serves as a substrate for protein synthesis, functions as a precursor for nitric oxide, polyamines, creatine, and other metabolites that influence tissue development (Fathima et al., 2024; Wu et al., 2009). Supplementation of dietary arginine has been reported to increase tibia bone growth and organ weights, such as the spleen and bursa (Kheiri and Landy, 2020; Luqman et al., 2020). However, Al-Tamimy et al. (2025) reported that dietary arginine supplementation did not increase liver weight in Japanese quail, whereas it increased liver weight in guinea fowl (Lala et al., 2022), suggesting a species-specific role of dietary arginine. As evidenced by the findings that dietary arginine levels did not significantly affect relative liver weight in both sexes suggests that hepatic growth is relatively conserved and maintained even under conditions of arginine restriction or excess. This may be because the liver, as a central metabolic organ, has high priority for nutrient allocation; thus, its structural growth is less sensitive to moderate fluctuations in dietary arginine as long as basal metabolic demands are met (Zaefarian et al., 2019).

In contrast, the finding that low dietary arginine increased relative brain weight in both sexes may reflect a compensatory allocation mechanism, whereby nutrient restriction slows somatic growth more than neural development, resulting in proportionally larger brains relative to body weight. Nutrient insufficiency can impair brain structure and development (Kumar et al., 2024). However, in unpredictable environments, birds face trade-offs that can drive brain size evolution (Fristoe and Botero, 2019). Although brain and body mass usually show an allometric relationship in vertebrates, we used the brain-to-body mass ratio as an index of relative brain size because the birds in our study were similar in age and body size. This measure provides a practical way to compare groups within the same species while reducing the influence of size differences (Smaers et al., 2012). The brain is often considered a "priority organ" during early growth, is maintained even under nutritional stress, and brain development is intimately linked to metabolic allocation (Bennett and Harvey, 1985). Therefore, arginine restriction may cause adaptive changes that give priority to maintaining the brain at the expense of peripheral tissues, increasing the relative weight of the brain. Therefore, liver exhibiting metabolic resilience and the brain being preferentially spared under dietary stress, the overall lack of effect on relative liver weight and the increase in relative brain weight due to loss of mass during the experiment under arginine restriction highlight tissue-specific priorities in resource allocation.

Effect of treatment on feed intake

The results showed that the interaction between treatment and time period had a significant effect on feed intake (Table 3). In the second week of the experimental period, feed intake was reduced under low dietary arginine (Figure 2).

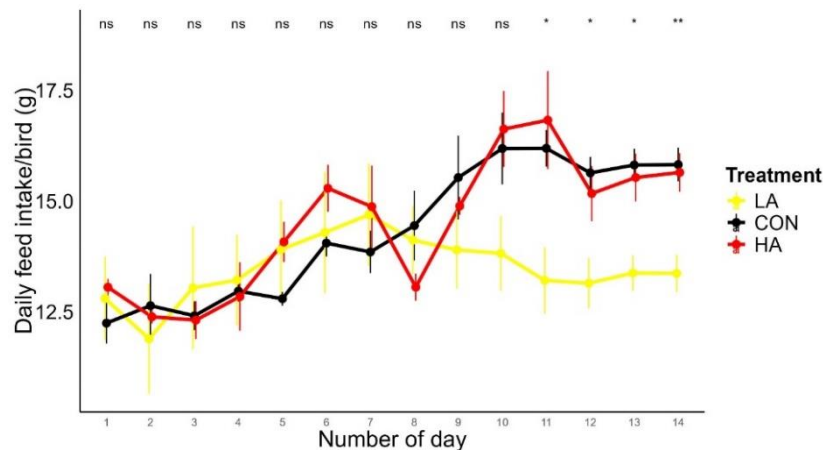
Table 3. ANOVA results for the effect of treatment and day on feed intake

Effect	Sum Sq	Mean Sq	NumDF	DenDF	F- value	P value
Treatment	1.721	0.860	2	6	1.040	0.409
Day	137.137	10.549	13	78	12.748	<0.001
Treatment: day	62.228	2.393	26	78	2.892	<0.001

The effect of arginine on feed intake has been reported with somewhat inconsistent results in avian species. In some studies, supplementation of arginine increased feed intake, suggesting a possible stimulatory effect on appetite or improved nutrient utilisation (Lala et al., 2022). However, under normal circumstances, supplementation of arginine did not increase feed intake in Japanese quail (de Lima et al., 2022; Reis et al., 2012; Sousa et al., 2022). By contrast, under heat stress, supplementation improved feed intake (Kalvandi et al., 2022). These annotations suggest that the effect of arginine on feeding behaviour may depend on different factors, including physiological status, environmental conditions, and the diet itself. In our study, the observed reduced feed intake in grower

Japanese quail under low dietary arginine may have resulted from nutrient deficiencies, which can disturb key metabolic pathways and appetite regulation. Such a deficiency could impair growth, reduce feed intake, and lower nutrient utilisation efficiency. In addition, decreased arginine availability may affect metabolic pathways that regulate feeding behaviour, further contributing to the observed reduction in intake. Thus, these findings support that feed intake responses to different levels of dietary arginine are different and modulated not only by dietary sufficiency but also by the broader physiological and environmental context, such as heat stress (Banerjee & Chaturvedi, 2018; Bilal et al., 2021; Nassar et al., 2023).

Figure 2. Effect of treatment and time period on feed intake



Data are means \pm s.e.m. from 51 birds per group and were analysed using ANOVA. Asterisks indicate the significant difference: * at 0.05, ** at 0.01, *** at $<$ 0.001, and ns indicates no significant difference. LA, low arginine; CON, control; HA, high arginine.

CONCLUSIONS

In conclusion, dietary arginine influenced relative brain weight, whereas relative liver weight remained unchanged. This pattern may reflect resource allocation and prioritisation of specific tissues under varying levels of dietary arginine. In addition, dietary arginine affected feed intake, indicating that growing Japanese quails are particularly sensitive to arginine supply during this developmental stage.

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