



Using Garlic Powder Supplements in the Diet of Growing Crossbred Rabbits; Evaluation of Weight Gain, Physiological and Biochemical Blood Parameters and the Excretion of *Escherichia coli* in Feces

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ABSTRACT

The experiment was conducted on 160 growing crossbred rabbits (New Zealand White × Local), consisting of 80 bucks and 80 does, with an initial weight of 505g. The experiment was arranged in a completely randomized design with five treatments and eight replications. The five treatments corresponded to five levels of garlic powder supplementation in the rabbit diet (%DM): no garlic powder (GP0), 0.5% garlic powder (GP0.5), 1% garlic powder (GP1.0), 1.5% garlic powder (GP1.5), and 2% garlic powder (GP2%). The results showed that rabbits fed with GP0.5 and GP1.0 diets had better final weight, daily weight gain, and FCR than GP0 ($P<0.05$). Improvements were observed in some physiological and biochemical blood parameters, including RBC, HGB, WBC, NEU, LYM, and globulin, in the GP0.5 group compared to GP2.0 and GP0 ($P<0.05$). Increasing the level of garlic powder in the diet also elevated HDL-C compared to the no garlic powder group ($P<0.05$). Additionally, the density of *E. coli* in the feces was lower in the GP0.5 and GP2.0 groups compared to GP0 ($P<0.001$). These results indicate that supplementing 0.5% garlic powder in the diet of growing crossbred rabbits enhanced nutrient intakes, daily weight gain, and improved *E. coli* disease resistance.

Key words: *Escherichia coli*, Garlic powder, Blood physiology, Blood biochemistry, Growing rabbits.

INTRODUCTION

In recent years, rabbit farming has gained increasing attention in the Mekong Delta. According to statistics of 2023, all 13 provinces in the region have developed rabbit farming on a medium and small scale, with a total meat production of over 1,000 tons, accounting for more than 20% of the national output, doubling the amount from 2021 (GSOV 2024).

The current trend in animal husbandry focuses on improving livestock productivity by enhancing weight gain and reducing feed costs. Consequently, research on incorporating probiotics (Truong et al. 2024), prebiotics (Ishaq and Canogullari 2024), organic acids, enzymes (Eid et al. 2024) and phytogetic (Al-Sagheer et al. 2023) into diets has been prioritized.

Garlic is one of the commonly used phytogetics which numerous studies have shown that the active components of garlic have beneficial effects on livestock and poultry, including antimicrobial, cholesterol-lowering, antioxidant, and growth-promoting properties (Ogbuewu et al. 2019; Farag et al. 2021; Ademolue et al. 2024).

The use of garlic in treating *E. coli* infection in rabbits improved their growth, biochemical parameters, and immune response while also decreasing fecal shedding and histopathological lesions (Farag et al. 2021). Chen et al. (2021) found that incorporating dried garlic (0.01%, 0.1%, and 1%) into broiler diets increased average daily weight gain, particularly at the 0.01% and 0.1% levels in the first 21 days. Another study showed that adding 1% garlic to broiler feed significantly enhanced body weight gain and could be a viable alternative to antibiotics. Administering Korean aged garlic extract at a concentration of 0.1% and/or 0.2% by *Leuconostoc citreum* led to improvements in broiler performance, including increased body weight gain and decreased feed conversion ratio, liver weight, and cecal *E. coli* count. While adding garlic to the water did not reduce abdominal fat or serum cholesterol levels, supplementing with garlic and soybean oil improved the final weight and breast and back weights of broilers fed a soybean diet. Furthermore, incorporating allicin into the diet enhanced antioxidant capacity and reduced blood lipid levels in chickens.

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According to Cho et al. (2020), garlic and allicin have been utilized to enhance the health and performance of livestock. The diet containing 0.5% garlic and 5% dandelion was discovered to be the most effective in enhancing the growth performance, carcass quality traits, and various parameters related to the nutritional fat quality of pigs (Samolińska et al. 2020).

According to the findings of Castillo-Lopez et al. (2021), adding garlic oil to the diet decreased the levels of total short-chain fatty acids in the reticulum and lowered the ratio of acetate to propionate in the rumen, without impacting rumen pH in cattle. Moreover, *in vitro* experiments showed that adding 0.5mL of garlic juice per 100mL of solution could increase propionate production and decrease the acetate to propionate ratio, indicating a possible reduction in hydrogen availability for methane-producing microorganisms (Kekana et al. 2021). These findings suggest that supplementing calves with garlic extract may improve their performance.

According to findings of studies, garlic has significant potential to be used instead of antibiotics, leading to producing meat yield and free from drug residues (Navidshad et al. 2018), enhancing animal welfare, and supporting sustainable development in animal farming (Chen et al. 2021). The objective of this study was to determine the effects of different levels of garlic powder supplementation in the diet of growing crossbred rabbits on weight gain, physiological and biochemical blood parameters, and the excretion of *E. coli* in feces.

MATERIALS AND METHODS

Study location

The experiment was conducted at Cam Nhung farm in Thoi Hoa ward, O Mon district, Can Tho city, Vietnam from November 2023 to April 2024. The chemical composition of experimental diets was analyzed at laboratory E205 of the Faculty of Animal Sciences, Agriculture University of Can Tho University.

Animals and experimental design

Sixty growing crossbred rabbits (New Zealand White × Local), consisting of 80 bucks and 80 does, with an average weight of 505 ± 15.2 g (Mean \pm SD), were randomly assigned to five treatments with eight replicates (2 bucks and 2 does per experimental unit). The treatments were five levels of garlic powder (GP) supplementation in the diet (%DM): 0% (GP0), 0.5% (GP0.5), 1% (GP1.0), 1.5% (GP1.5), and 2% (GP2.0) (Table 1).

Feeds and feeding

Before the experiment, the rabbits were fed *ad libitum* for one week to determine dry matter (DM) intake requirements and to calculate the diet. The feed provided to the animals was weighed daily, and the residue feeds were reweighed the following morning. Fresh water was free to access.

The rabbits were fed a mixture of soya waste, soybean extraction meal, and garlic powder (corresponding to each treatment) in the morning, followed by *Operculina turpethum*. Elephant grass was provided *ad libitum*. Garlic was peeled, dried and finely ground before being used in rabbit diets. The feed intake requirements were recalculated weekly to match the experimental design and

the growing requirements of the experimental rabbits. The chemical composition of the ingredients used in this experiment was analyzed and presented in Table 2.

Table 1: Ingredient composition of the diet

Ingredient composition, %	Treatments				
	GP0	GP0.5	GP1.0	GP1.5	GP2.0
DM					
Soya waste	36.0	36.0	36.0	36.0	36.0
Soybean extraction meal	18.0	17.5	17.0	16.5	16.0
<i>Operculina turpethum</i>	12.0	12.0	12.0	12.0	12.0
Garlic powder	0.00	0.50	1.00	1.50	2.00
Elephant grass	34.0	34.0	34.0	34.0	34.0
Total	100	100	100	100	100
OM	90.9	90.9	91.0	91.0	91.0
CP	19.8	19.7	19.5	19.4	19.3
NDF	45.3	45.1	45.0	44.9	44.7
ADF	29.4	29.3	29.2	29.1	29.0
EE	6.72	6.71	6.70	6.69	6.67
Ash	9.10	9.05	9.00	8.95	8.90

DM=Dry Matter; OM=Organic Matter; CP=Crude Protein; EE=Ether Extract; NDF=Neutral Detergent Fiber; ADF=Acid Detergent Fiber; Ash=Total mineral; GP0=0% Garlic powder; GP0.5=0.5% Garlic powder; GP1.0=1% Garlic powder; GP1.5=1.5% Garlic powder; GP2.0=2% Garlic powder

Table 2: Chemical composition of ingredients (%DM)

Feeds	DM	OM	CP	NDF	ADF	EE	Ash
Soya waste	12.7	94.2	19.2	36.6	28.2	9.87	5.80
Soybean extraction meal	90.2	89.7	43.2	27.8	19.6	2.50	10.3
<i>Operculina turpethum</i>	12.2	88.6	14.7	39.6	26.1	5.10	11.4
Elephant grass	16.8	88.9	9.84	65.7	36.9	6.20	11.2
Garlic powder	89.4	96.6	15.9	-	-	0.67	3.41

Measurements

The weight gain of rabbits was recorded weekly at a specific time. Feed intake, daily weight gain, and FCR were monitored and calculated throughout the 12-week experiment. The amount of feed offered and refusals were documented daily in the morning.

The nutrient digestibility of the diet was assessed following the method described by McDonald et al. (2010). The digestibility trial was conducted in the 10th week of the experiment and lasted for seven consecutive days. Feed offered, refusals, feces, and urine were continuously collected over seven days to determine nutrient digestibility and nitrogen retention.

Chemical analysis

The samples were dried at 60°C for 72 hours, and then crushed through a 1mm mesh (Cutting Mill SM100, Retsch, Germany) to analyze for dry matter (DM), organic matter (OM), crude protein (CP), ether extract (EE), and Ash contents according to the procedures of AOAC (2000). However, neutral detergent fiber (NDF) and acid detergent fiber (ADF) were analyzed by the procedure of Van Soest et al. (1991).

Blood collection

Blood samples were collected at the end of the 12-week growth experiment. Blood samples were drawn by humane puncture of the ear vein in the early morning, before feeding, from rabbits that had fasted overnight. The blood was then divided into two test tubes - one containing EDTA and the other containing heparin. Immediately after collection, the blood samples were transferred to Center Lab Vietnam for analysis. The hematological parameters were

analyzed by machine: Cell-DynR 1700 (manufacturer: Abot). The biochemical parameters were analyzed by machine: Humalyer 2000 (manufacturer: Humen).

Table 3: Feed and nutrient intakes of the experimental rabbit

Item	Treatments					SEM P	
	GP0	GP0.5	GP1.0	GP1.5	GP2.0		
Feed intake, g DM/animal/ day							
Soya waste	24.6	24.8	24.8	24.8	24.7	0.650	0.999
<i>Operculina turpethum</i>	9.87	9.87	9.87	9.87	9.87	0.269	1.000
Elephant grass	8.21 ^b	9.30 ^a	9.31 ^a	9.97 ^a	9.63 ^a	0.255	0.001
Soybean extraction meal	14.6	14.4	13.8	13.9	13.4	0.355	0.117
Garlic powder	0.00 ^c	0.31 ^d	0.63 ^c	0.95 ^b	1.27 ^a	0.021	0.001
Nutrient intakes, g DM/animal/day							
DM	57.3	58.7	58.3	59.4	58.9	1.269	0.824
OM	52.3	53.6	53.3	54.3	53.8	1.165	0.818
CP	13.3	13.4	13.2	13.3	13.1	0.314	0.974
NDF	22.4	23.1	22.9	23.4	23.0	0.447	0.618
ADF	15.4	15.8	15.7	16.0	15.7	0.321	0.811
EE	3.81	3.89	3.87	3.92	3.88	0.086	0.923
Ash	4.98	5.09	5.04	5.13	5.05	0.104	0.867

Mean values within rows with different superscripts are different at $P < 0.05$. SEM=Standard error of the mean. DM=Dry Matter; OM=Organic Matter; CP=Crude Protein; EE=Ether Extract; NDF=Neutral Detergent Fiber; ADF=Acid Detergent Fiber; Ash=Total mineral; GP0=0% Garlic powder; GP0.5=0.5% Garlic powder; GP1.0=1% Garlic powder; GP1.5=1.5% Garlic powder; GP2.0=2% Garlic powder.

Table 4: The nutrient digestibility (%) and digestible nutrients (g/day) of experimental rabbits in the nutrient digestibility trial

Item	Treatments					SEM P	
	GP0	GP0.5	GP1.0	GP1.5	GP2.0		
The nutrient digestibility (%)							
DMD	80.5	83.6	82.9	81.3	81.5	1.745	0.725
OMD	81.4	84.4	83.9	82.5	82.8	1.657	0.729
CPD	86.4	87.6	86.9	87.7	84.9	1.426	0.659
EED	79.3	82.2	81.7	80.0	80.4	2.054	0.848
NDFD	53.4	57.6	56.6	54.7	55.8	2.889	0.862
ADFD	42.1	45.6	45.1	44.6	44.5	2.691	0.912

SEM=Standard error of the mean. DMD=Dry Matter Digestibility; OMD=Organic Matter Digestibility; CPD=Crude Protein Digestibility; EED=Ether Extract Digestibility; NDFD=Neutral Detergent Fiber Digestibility; ADFD=Acid Detergent Fiber Digestibility; GP0=0% Garlic powder; GP0.5=0.5% Garlic powder; GP1.0=1% Garlic powder; GP1.5=1.5% Garlic powder; GP2.0=2% Garlic powder.

Table 5: Nitrogen balance

Item	Treatments					SEM P	
	GP0	GP0.5	GP1.0	GP1.5	GP2.0		
N intake, g	2.63	2.62	2.60	2.60	2.56	0.029	0.580
N retention, g	1.60 ^{ab}	1.83 ^a	1.41 ^b	1.59 ^{ab}	1.44 ^{ab}	0.096	0.039
%NR/Ni	60.9 ^{ab}	70.0 ^a	54.2 ^b	61.1 ^{ab}	56.4 ^{ab}	3.514	0.038
NI/W ^{0.75}	1.55 ^a	1.49 ^{ab}	1.46 ^{ab}	1.43 ^b	1.46 ^{ab}	0.029	0.038
NR/W ^{0.75}	0.95	1.05	0.79	0.87	0.82	0.063	0.056

^{a,b} Mean values within rows with different superscripts are different at $P < 0.05$. SEM=Standard error of the mean; GP0=0% Garlic powder; GP0.5=0.5% Garlic powder; GP1.0=1% Garlic powder; GP1.5=1.5% Garlic powder; GP2.0=2% Garlic powder.

Quantification of *E. coli*

The bacterial quantification of *E. coli* in the feces of the experimental rabbits was performed by counting the colonies using the ISO standard method (ISO, 16649-1:2018). After counting the colonies on the plates, the next

step was to calculate the number of *E. coli* in 1 gram of feces using the following formula:

$$N = \frac{\sum C}{Vd(n1 + 0.1 \times n2)}$$

Where:

N: Total number of *E. coli* in 1 gram of feces (CFU/g)

$\sum C$: Total number of colonies counted at different dilutions

V: Volume of the dilution used to culture the sample (ml)

d: Dilution concentration where colonies were counted

n1: Number of plates counted at dilution

n2: Number of plates counted at the next dilution.

Statistical Analysis

Data were analyzed for mean values and standard errors using Microsoft Excel 2010. ANOVA was performed with the General Linear Model in Minitab 16.1.0 (Minitab 2016) and Tukey's test was applied for pairwise comparisons (Minitab 2016). Statistical significance was set at $P < 0.05$. *E. coli* quantification results were compared using the chi-square test with 99% confidence.

RESULTS

Feed and nutrient intakes

Soy waste, soybean extraction meal, and *Operculina turpethum* were fed at the same values, resulting in no difference between treatments ($P > 0.05$). The intake of elephant grass was higher in treatments supplemented with garlic powder than in those without it ($P < 0.05$). Moreover, the quantity of elephant grass intake increased, which led to a higher nutrient intake in the diets incorporating garlic powder. However, this finding had no statistical significance ($P > 0.05$) (Table 3).

Digestibility and nitrogen balance

Digestibility of DMD, OMD, EED, NDFD, and ADFD tended to be higher in the garlic powder-supplemented diets than in the control, although nutrient intake was not significantly different among the treatments ($P > 0.05$) (Table 4). NI/W^{0.75} value was higher in the unsupplemented diet group than in the garlic powder group ($P < 0.05$). In contrast, N retention and %NR/Ni were highest in the 0.5% garlic powder diet group and lowest in the 1.0% garlic powder diet group ($P < 0.05$) (Table 5).

Daily weight gain

The average weight gain of rabbits on the four diets supplemented with garlic powder tended to decrease from 20.3 to 18.8g/day but was still higher than that of the control diet at 17.8g/day ($P < 0.05$). After 12 weeks of feeding, the live weights ranged from 1971 to 2209g/head, with the lowest being in the unsupplemented diet and the highest in the 0.5 and 1.0% garlic powder supplementation levels ($P < 0.05$) (Table 6).

Physiological and biochemical blood indicators

The physiological indices (Table 7) and blood biochemistry (Table 8) show that the experimental rabbit blood parameters were all within the normal physiological range. Some blood physiological indices such as WBC, NEU, LYM, RBC, and HGB were highest in the GP0.5 diet ($P < 0.05$). The indices related to platelets and the remaining

Table 6: Final live weight, daily weight gain, and FCR of the experimental rabbits

Item	Treatments					SEM	P
	GP0	GP0.5	GP1.0	GP1.5	GP2.0		
Initial live weight, g/head	472	497	507	522	527	22.64	0.460
Final live weight, g/head	1971 ^b	2209 ^a	2200 ^a	2135 ^{ab}	2108 ^{ab}	47.81	0.012
Daily weight gain, (g/head/day)	17.8 ^b	20.3 ^a	20.1 ^a	19.2 ^{ab}	18.8 ^{ab}	0.407	0.001
FCR	3.23 ^a	2.88 ^b	2.89 ^b	3.07 ^{ab}	3.10 ^{ab}	0.071	0.007

Mean values within rows with different superscripts are different at $P < 0.05$. SEM=Standard error of the mean; FCR=Feed conversion ratio; GP0=0% Garlic powder; GP0.5=0.5% Garlic powder; GP1.0=1% Garlic powder; GP1.5=1.5% Garlic powder; GP2.0=2% Garlic powder.

Table 7: Physiological parameters in the blood of experimental rabbits

Item	Treatments					SEM	P
	GP0	GP0.5	GP1.0	GP1.5	GP2.0		
WBC, $10^9/L$	5.01 ^b	7.54 ^a	3.69 ^b	6.47 ^a	5.03 ^b	0.305	0.001
NEU, $10^9/L$	3.03 ^{bc}	4.40 ^a	2.00 ^c	3.42 ^{ab}	2.85 ^{bc}	0.235	0.001
LYM, $10^9/L$	1.92 ^b	3.01 ^a	1.62 ^b	2.28 ^{ab}	2.01 ^b	0.206	0.007
MONO, $10^9/L$	0.06	0.10	0.05	0.43	0.13	0.101	0.121
EOS, $10^9/L$	0.00	0.07	0.01	0.07	0.00	0.020	0.062
BASO, $10^9/L$	0.07	0.07	0.01	0.29	0.03	0.074	0.137
RBC, $10^{12}/L$	5.67 ^{ab}	6.19 ^a	5.55 ^{ab}	5.56 ^{ab}	5.24 ^b	0.147	0.013
HGB, g/dL	11.9 ^{ab}	12.7 ^a	11.3 ^b	11.4 ^b	11.0 ^b	0.278	0.011
HCT, %	36.2	38.8	34.9	35.0	33.9	1.045	0.062
MCV, fL	63.9	62.6	62.9	62.8	64.8	0.875	0.420
MCH, pg	20.9	20.6	20.4	20.5	21.0	0.295	0.554
MCHC, g/dL	32.7	32.9	32.5	32.6	32.4	0.520	0.962
RDW, %	13.2	13.5	14.2	14.0	14.9	0.606	0.360
PLT, $10^9/L$	268	300	327	271	215	46.74	0.556
MPV, fL	6.87	6.86	5.38	4.11	3.89	0.820	0.187
PDW, fL	14.6	12.9	14.6	13.9	14.0	0.744	0.521
PCT, %	0.19	0.21	0.19	0.11	0.09	0.046	0.316

Mean values within rows with different superscripts are different at $P < 0.05$. SEM=Standard error of the mean; GP0=0% Garlic powder; GP0.5=0.5% Garlic powder; GP1.0=1% Garlic powder; GP1.5=1.5% Garlic powder; GP2.0=2% Garlic powder; WBC=White Blood Cells; NEU=Neutrophils; LYM=Lymphocytes; MONO=Monocytes; EOS=Eosinophils; BASO=Basophils; RBC=Red Blood Cell Count; HGB=Hemoglobin; HCT=Hematocrit; MCV=Mean Corpuscular Volume; MCH=Mean Corpuscular Hemoglobin; MCHC=Mean Corpuscular Hemoglobin Concentration; RDW=Red Cell Distribution Width; PLT=Platelet Count; MPV=Mean Platelet Volume; PDW=Platelet Distribution Width; PCT=Plateletcrit.

parameters related to red blood cells and white blood cells did not show significant differences between the experimental diets ($P > 0.05$). The HDL-C index was higher in the diets supplemented with garlic powder compared to the control ($P < 0.05$). However, the AST and ALT indices showed large fluctuations with ranges of 29.6-49.8 and 44.6-77.1 U/L, respectively, but there was no statistical significance ($P > 0.05$). The GLO index tended to decrease linearly when increasing the garlic powder ratio in the diet from 0.5-2.0%. The results also recorded the highest globulin content in the GP0.5 diet and lower copper content in the GP0, GP1.5, and GP2.0 diets ($P < 0.05$).

Quantification of *E. coli* concentration in feces of experimental rabbits

The results of *E. coli* quantification in rabbit feces were monitored over 12 weeks of the experiment (Table 9), which showed a difference in density between the diet groups ($P < 0.01$). The highest *E. coli* density was recorded in the control group ($4.90 \pm 1.79 \times 10^5$ CFU/g), which was 2.7 times higher than the density found in the diet supplemented with 0.5% garlic powder. However, the *E.*

coli density found in the experiment was not at a level that caused disease in rabbits, and no cases of disease were recorded in experimental rabbits during the experiment.

Table 8: Biochemistry of blood of experimental rabbits

Item	Treatments					SEM	P
	GP0	GP0.5	GP1.0	GP1.5	GP2.0		
HDL-C, mmol/L	0.23 ^b	0.37 ^{ab}	0.60 ^a	0.50 ^{ab}	0.60 ^a	0.068	0.011
AST, U/L	43.2	29.6	39.7	41.8	49.8	9.948	0.708
ALT, U/L	44.6	45.3	64.3	65.0	77.1	11.88	0.304
GLO, g/L	20.4 ^b	25.9 ^a	22.2 ^{ab}	20.8 ^b	20.1 ^b	0.942	0.007

Mean values within rows with different superscripts are different at $P < 0.05$. SEM=Standard error of the mean; GP0=0% Garlic powder; GP0.5=0.5% Garlic powder; GP1.0=1% Garlic powder; GP1.5=1.5% Garlic powder; GP2.0=2% Garlic powder; AST=Aspartate Aminotransferase; ALT=Alanine Aminotransferase; GLO=Globulin in plasma; HDL-C=High Density Lipoprotein Cholesterol.

Table 9: Quantitative results of *E. coli* on experimental rabbit feces

Treatments	Number of test samples	$N \times 10^5 \pm SD$ (CFU/g)
GP0	12	$4.90 \pm 1.79a$
GP0.5	12	$1.82 \pm 1.02c$
GP1.0	12	$3.90 \pm 2.57ab$
GP1.5	12	$2.97 \pm 2.36bc$
GP2.0	12	$2.24 \pm 0.84c$
Total	60	$P = 0.001$

Values in the column bearing at least one common symbol do not differ at $P < 0.05$. N=average number of colonies per gram of sample; GP0=0% Garlic powder; GP0.5=0.5% Garlic powder; GP1.0=1% Garlic powder; GP1.5=1.5% Garlic powder; GP2.0=2% Garlic powder.

DISCUSSION

In recent years, garlic has been considered a phytobiotic with growth-promoting and natural antibiotic effects used in pig and chicken farming (Alagbe and Oluwafemi 2019; Chen et al. 2021). In the present research, results showed significant improvements in daily weight gain, final weight, and feed conversion ratio in rabbit groups using diets supplemented with garlic powder compared to the control. According to the publication by Chimbaka and Walubita (2020), when adding 0.3-0.9% garlic powder to the diet, similar improvements were shown, and the authors also hypothesized that increasing the proportion of garlic powder in the diet would continue to increase growth performance in rabbits. At the same time, higher feed intake was noted when using diets containing 6g of garlic powder/kg of feed according to Ademolue et al. (2024). Also, according to Ademolue et al. (2024), garlic helps enhance the absorption of important nutrients by improving feed intake, stimulating digestive enzyme secretion, and increasing gastric and intestinal

motility. Studies seem to lead to the general hypothesis that increasing the level of garlic powder in the diet will bring about better growth results. However, our feeding results only recorded about 0.5-1.0% garlic powder supplementation in the diet with different effects, while higher supplementation levels did not find positive improvements.

Although there were no differences in nutrient intake, digestibility, and digestible nutrients, NR and %NR/NI were highest in the 0.5% garlic powder diet. This suggests that garlic powder inclusion at 0.5% resulted in positive improvements in nitrogen metabolism and growth retention. A similar effect was explained by Kamruzzaman et al. (2014) using garlic (stems and leaves) supplementation in sheep, where the improvement in nitrogen metabolism was due to the effect of garlic on the nitrogen conversion capacity of beneficial microorganisms. This was because garlic had a high content of inulin (Teferra 2021), a prebiotic that selectively stimulates the growth of beneficial microorganisms in human and animal intestines (Gibson et al. 2017). In contrast, the digestion process in rabbits occurs mainly in the cecum, which plays an important role. Therefore, enhancing beneficial microflora would contribute to improving the nitrogen metabolism capacity of rabbits.

In addition, the growth of beneficial microorganisms produced short-chain fatty acids that reduced intestinal pH, thereby inhibiting harmful microorganisms. According to Saulnier et al. (2009), lactic acid bacteria inhibit the growth of *E. coli*, and inulin played an indirect role in this process. The prebiotic effect of inulin in protecting intestinal health and treating digestive disorders was also published by Akram et al. (2019) and Guarino et al. (2020). This contributed to explaining the lower *E. coli* density in feces found in the garlic powder-supplemented treatments compared to the control in the study. Similar results have been reported in another study on pigs (Sun and Kim 2020).

Significantly higher red blood cell counts, hemoglobin levels, WBC, LYM, NEU, and globulin indices were recorded in diets containing 0.5% garlic powder. Hemoglobin played a role in transporting oxygen to cells, thereby promoting metabolism, improving digestion, and absorption (Washington and Van Hoosier 2012). WBC and globulin indices reflect the animal's immune capacity. In addition to inulin, garlic also contained the compound allicin, which acted as an antibacterial agent, enhancing the immune system by improving various physiological parameters, including RNA synthesis (Rahman 2007).

According to Jang et al. (2018), garlic powder supplementation improved immune factors, reduced FCR in animals, and lowered serum cholesterol and cortisol levels. Inulin present during intestinal fermentation produced beneficial metabolites, such as propionate, which interfered with and inhibited cholesterol production in the host liver (Qin et al. 2023). This finding was supported by the increase in blood HDL-C concentrations when the proportion of garlic powder in the diet was increased. HDL-C aids in removing excess cholesterol from the body and transporting it to the liver for processing and excretion. However, intolerance to inulin or excessive use of garlic powder in the diet may not yield positive and long-term effects. Evidence shows that ALT and AST indices

fluctuate, which are signs reflecting the level of liver cell damage, possibly due to pathology or excessive physiological activity in the liver during the body's detoxification process. Previous studies have reported gastrointestinal dysfunction with high inulin intake in humans (Le Bastard et al. 2020; Tawfick et al. 2022). Currently, there have been few assessments of the adverse effects of garlic powder on animals, but based on the results of the current study, we recommend caution when using high garlic powder diets for rabbits.

Conclusion

Adding 0.5% garlic powder to the diet of growing crossbred rabbits had a beneficial impact. It improved feed consumption, nutrient absorption, final weight, and daily weight gain. The higher levels of garlic powder in the diets led to increased HDL-C levels in the blood and enhanced the FCR. The inclusion of garlic powder resulted in lower *E. coli* levels compared to diets without garlic powder. Garlic powder shows promise as a potential alternative to antibiotics for enhancing animal farming practices in alignment with the principles of organic animal husbandry in modern agriculture.

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Ethics Statement: The study was conducted with approval for animal care, housing, and sample collection under the Animal Welfare Assessments of the Animal Ethics Committee of Can Tho University, Vietnam (CTU-AEC24002).

Author's Contribution: T.T.Trung. Conceived, designed, performed the experiments, and analyzed the data; P.Nhan wrote the paper; all authors reviewed and approved the final manuscript.

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