



A study on evaluation of dietary supplementation of organic acids on growth performance and nutrient utilisation in New Zealand White rabbits[#]

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Citation: Dharavath, S., George, S.K., K. Ally, K. Shyama, K., Dipu, M.T. & Thomas, M. (2025). A study on evaluation of dietary supplementation of organic acids on growth performance and nutrient utilisation in New Zealand White rabbits. *Journal of Veterinary and Animal Sciences* 57 (1), 79-84
<https://doi.org/10.51966/jvas.2026.57.1.79-84>

Received: 12.09.2025

Accepted: 03.11.2025

Published: 31.03.2026

Abstract

A study was conducted to evaluate the effect of dietary supplementation of organic acids on growth performance and nutrient utilisation in New Zealand White rabbits. Feeding experiment was conducted in 24 weaned rabbits of either sex, aged four weeks at Rabbit Breeding Station, College of Veterinary and Animal Sciences, Mannuthy. Animals were randomly allotted into four dietary treatments following completely randomised design with six replicates in each group, and were reared under uniform managerial conditions for a period of four months. All the experimental rabbits were offered with concentrate mixture (18% CP and 2700 kcal of DE) and green fodder as per ICAR 2013 feeding standards. Treatment groups comprised of T1 (un supplemented), T2 (supplemented with 1 per cent fumaric acid), T3 (supplemented with 0.5 per cent formic acid) and T4 (supplemented with 0.5 per cent fumaric acid and 0.25 per cent formic acid). A digestibility trial of three days duration was conducted at the end of feeding experiment. Parameters recorded during the study included daily feed and fodder intake, weekly body weight, average daily gain (ADG), feed conversion ratio, nutrient digestibility and faecal pH. Results revealed no significant ($p>0.05$) differences in dry matter (DM) intake, average daily gain and feed conversion ratio (FCR) among the treatment groups. Digestibility coefficients of nutrients except nitrogen free extract (NFE) differed significantly ($p<0.05$) among treatment groups. Dry matter digestibility was highest for the group supplemented with 0.5 percent formic acid in the diet (T3). Crude protein (CP) and crude fibre (CF) digestibility values were higher ($p<0.05$) for organic acid supplemented groups with marked improvement in T3 and T4 groups. The digestibility of ether extract (EE) also improved ($p<0.05$) with organic acid supplementation and the values were similar ($p>0.05$) among the supplemented groups. Faecal analysis demonstrated lowered ($p<0.05$) pH in organic acid supplemented groups. Study revealed that dietary supplementation with formic acid (0.5%) proved most effective in enhancing the utilisation of various nutrients, suggesting its potential use as a feed additive in rabbit production.

Keywords: Organic acids, fumaric acid, formic acid, nutrient digestibility, faecal pH

[#]Part of MVSc thesis submitted to Kerala Veterinary and Animal Sciences University, Pookode, Wayanad, Kerala

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Rabbits have tremendous potential for sustainable production of animal protein due to short generation interval, prolific production and high feed conversion ratio (Cheeke, 1986). Rabbit meat is high in protein and low in fat, cholesterol and sodium compared to other animal meats. Rabbits are strict herbivores and hindgut fermenters, relying primarily on a high-fibre diet for optimal digestive health. They consume a wide variety of forages, grasses, leafy vegetables and fibrous roughages, which help maintain proper gut motility and prevent digestive disturbances. A balanced rabbit diet typically consists of concentrates rich in carbohydrates, proteins and minerals, supplemented with roughages to ensure adequate fibre intake. Fibre plays a crucial role in stimulating caecal fermentation, promoting beneficial microbial growth and enhancing nutrient utilisation (Gidenne, 2015). Rabbits also exhibit caecotrophy, a unique physiological behaviour in which they re-ingest soft faeces (caecotrophs) directly from the anus to recover essential nutrients such as B vitamins, microbial protein, and volatile fatty acids produced during caecal fermentation (Blas & Wiseman, 2010). This process significantly improves their nutrient efficiency and contributes to better growth and health.

Rabbits are sensitive to multifactorial digestive disorders, which can cause high mortality and morbidity rate, and seem to be related to microbial dysbiosis in the caecum. During the first growing period, in fact, changes in feeding behaviour and in-housing conditions together with an incomplete maturation of the digestive immune system could promote the development of potentially pathogenic microflora, which could cause digestive troubles and lower zootechnical performance (Cesari et al., 2008). Antibiotics in feed were used in animal production to prevent enteric diseases and to reduce the economic losses. However, concerns about human health and legal restrictions have stimulated the search for non-antibiotic growth promoters in animal feeding.

Organic acids are included in the diets of monogastric animals, especially pigs and poultry as alternative to antibiotic growth promoters (Papatsiros et al., 2012). Dietary supplementation of organic acids has beneficial effects like enhancement of growth rate by improving gut health through the reduction of pH, promoting beneficial bacterial growth and increasing the digestibility of nutrients. However, most of the studies on dietary supplementation of organic acids have focussed on poultry and pigs and very little research has been conducted in rabbits. Hence, the present study was conducted to evaluate dietary supplementation of organic acids such as fumaric acid and formic acid on growth performance and nutrient utilisation in growing New Zealand White rabbits.

Materials and methods

Location of study

The study was conducted at the department of Animal Nutrition and Rabbit Breeding Station, College of Veterinary and Animal Sciences, Mannuthy, Thrissur, Kerala, India.

Selection and feeding management of experimental animals

Twenty-four New Zealand White growing rabbits of either sex aged four weeks were selected for the study from the Rabbit Breeding Station, College of Veterinary and Animal Sciences, Mannuthy. The rabbits were divided into four groups of six animals each in a completely randomised design. All the experimental animals were housed individually in metallic cages having facilities for feeding and watering.

Feeding regimen

The rabbits were allotted to four dietary treatments and offered concentrate mixture (18% CP and 2700 kcal DE) and green fodder as per ICAR (2013) recommendations. The treatments consisted of T1 (un-supplemented), T2 (supplemented with 1 per cent fumaric acid), T3 (supplemented with 0.5 per cent formic acid), and T4 (supplemented with 0.5 per cent fumaric acid and 0.25 per cent formic acid). Ingredient composition

Table 1. Ingredient composition of concentrate mixture, %

Ingredients	Percentage
Yellow maize	37.5
Corn gluten fibre	8
Black gram husk	7
Wheat bran	11
Deoiled rice bran	11.5
Rice polish	9.05
Alfalfa pellet	9
Gingely oil cake	6
Salt	0.5
Supplevite-M *	0.25
Toxfia**	0.2
Total	100

* Supplevite-M (250 gram) contains 5,00,000 IU of vitamin A, 1,00,000 IU of vitamin D3, 0.2 g of vitamin B2, 75 units of vitamin E, 0.1 g of vitamin K, 0.25 g of calcium pantothenate, 1 g of nicotinamide, 0.6 g of vitamin B12, 15 g of choline chloride, 75 g of calcium, 2.75 g of manganese, 0.1 g of iodine, 0.75 g of iron, 1.5 g of zinc, 0.2 g of copper, 0.045 g of cobalt.

**Toxfia contains specially treated hydrated sodium calcium aluminosilicates (HSCAS), MOS, choline chloride with buffered organic acids like propionic acid.

of concentrate mixture is given in Table 1. Concentrate feeds were offered in the morning at 8.00 AM and fodder was given in the evening at 3.00 PM daily to each rabbit. The left-over quantities of the feed and green grass were weighed daily and their moisture content was determined to calculate the dry matter intake. Clean drinking water was made available to all animals throughout the experiment. The animals were maintained under identical conditions of feeding and management throughout the experimental period of four months.

Data collection

Daily feed intake and weekly body weight of all the rabbits were recorded throughout the experimental period. A 3-day digestibility trial was conducted at the end of the feeding phase to determine nutrient digestibility, faecal pH, and faecal microbial counts. The price of ingredients used for the study was fixed as per rate contract fixed by Revolving Fund Feed mill, Department of Animal Nutrition, College of Veterinary and Animal Sciences, Mannuthy.

Analysis of feed fodder and faecal samples

Proximate analysis of feed, green grass and faecal samples was done as per the standard procedures (AOAC, 2016). Calcium and phosphorus in the feed were analysed by conventional precipitation and titration method (AOAC, 2016).

Statistical analysis

The data were statistically analysed as per Snedecor and Cochran (1994). Parametric data obtained from different observations during the experiment were analysed using one-way ANOVA, and significant differences between means were separated using Duncan's multiple range test (DMRT) by statistical package for the social sciences (SPSS), version 24.0.

Results and discussion

Chemical composition of feed and fodder

The per cent chemical compositions of the experimental diet and fodder provided to rabbits under four dietary treatments are presented in Table 2. The concentrate mixture contained 92.68 ± 0.02 per cent DM, 18.18 ± 0.11 per cent CP and 8.51 ± 0.02 per cent CF, with a calculated digestible energy (DE) value of 2704.26 kcal/kg. Chandran *et al.* (2023) reported a similar proximate composition for rabbit feed, containing 91.32 per cent DM, 18.74 per cent CP, 8.31 per cent CF and 5.50 per cent EE. The green grass offered to the rabbits contained 24.27 ± 0.13 per cent DM, 10.69 ± 0.39 per cent CP, 27.48 ± 0.41 per cent CF, 2.75 ± 0.05 per cent EE, 11.14 ± 0.10 per cent total ash, 47.95 ± 0.70 per cent NFE, 1.31 ± 0.01 per cent acid-insoluble ash, 0.69 ± 0.05 per cent calcium, and 0.35 ± 0.03 per cent phosphorus. The proximate composition of fodder recorded in the present study was in agreement with the results reported by Kamalhasan *et al.* (2018).

Growth performance of rabbits maintained on four dietary treatments

The growth performance of rabbits maintained on four dietary treatments is summarised in Table 3 and the results revealed similar final weight and daily weight gain among all the treatment groups. Cardinali *et al.* (2008) conducted a study on weaned New Zealand White rabbits experimentally infected with *Escherichia coli*, evaluating the effect of a dietary blend of formic acid, citric acid, and essential oils at 1.0 g/kg feed. Rabbits in the supplemented group showed significantly higher ($p < 0.05$) body weight gain (1267 g) compared to the control group (1180 g). Lin *et al.* (2023) conducted a study in Ira breed rabbits, evaluating the effects of dietary supplementation with 1000 mg/kg of mixed organic acids (including butyric and citric acids). Rabbits in the supplemented group showed significantly higher ($p < 0.05$) body weight gain (2142 g) compared to the control group (1987 g). However, growth performance of rabbits in the present study was not influenced by organic acid supplementation and it might be due to variations with regard to the type and inclusion levels of organic acids in the diet.

Table 2. Chemical composition¹ of concentrate mixture and green fodder fed to New Zealand White growing rabbits, %

SI. No	Parameters	Experimental feed	Fodder
1	Dry matter	92.68±0.02	24.27±0.13
2	Crude protein	18.18±0.11	10.69±0.39
3	Crude fibre	8.51±0.02	27.48±0.41
4	Ether extract	5.43±0.07	2.75±0.05
5	Total ash	7.29±0.01	11.14±0.10
6	Nitrogen free extract	60.60±0.19	47.95±0.70
7	Acid insoluble ash	1.09±0.01	1.31±0.01
8	Calcium	1.10 ±0.13	0.69±0.05
9	Phosphorus	0.89±0.01	0.35±0.03

¹Mean of six replicates with SE; Values from 2-9 expressed on DM basis

Table 3. Growth performance¹ of rabbits maintained on four dietary treatments

Parameters	Dietary treatments				p-value
	T1	T2	T3	T4	
Initial body weight, g	447.00±35.89	458.83±31.82	470.00±17.95	444.33±19.53	0.905 ^{ns}
Final body weight, g	2505.00±80.80	2571.50±110.76	2695.17±107.66	2577.50±77.49	0.570 ^{ns}
Average daily gain, g	18.38±0.66	18.86±0.97	19.87±0.83	19.07±0.66	0.613 ^{ns}
Total body weight gain, gm	2058.00±73.61	2112.67±108.92	2225.16±92.93	2133.17±74.45	0.613 ^{ns}
Total dry matter consumed (g/animal)	9466.80±0.25	9084.48±0.27	8455.60±0.15	8745.99±0.21	0.281 ^{ns}
Average daily dry matter consumed (g/animal)	78.89±2.5	75.70±2.3	70.46±2.1	72.88±2.2	0.281 ^{ns}
Feed conversion ratio	4.61±0.42	4.32±0.40	3.82±0.38	4.13±0.41	0.182 ^{ns}

¹Mean values of six replicates with SE; ns-non significant ($p>0.05$)

Digestibility of nutrients in rabbits maintained on four dietary treatments

Table 4. Digestibility coefficients of nutrients¹ of four experimental diets, %

Parameters	Dietary treatments				p-value
	T1	T2	T3	T4	
Dry matter	63.30±1.56 ^a	70.01±1.08 ^b	75.13±0.99 ^c	73.35±3.36 ^b	0.001 [*]
Crude protein	63.27±1.43 ^a	70.82±0.95 ^b	76.98±0.96 ^c	74.13±3.45 ^{bc}	0.001 [*]
Crude fibre	42.87±3.87 ^a	48.59±1.94 ^b	62.42±1.56 ^c	53.49±3.03 ^{bc}	0.001 [*]
Ether extract	73.63±2.23 ^a	80.63±1.35 ^b	85.04±0.79 ^b	83.30±2.22 ^b	0.001 [*]
NFE	73.10±0.99	75.30±0.90	77.01±1.71	77.80±2.81	0.280 ^{ns}

¹Mean of six replicates with SE; Means having different small letters as superscript differ significantly within a row; ns- non significant ($p>0.05$), *-significant ($p<0.05$)

The total dry matter intake and average daily feed intake reported in the study was similar ($p>0.05$) across treatment groups. Michelan et al. (2002) supplemented diets with fumaric acid (0.15 %) in growing rabbits and reported no significant ($p>0.05$) effect on the intake of dry matter. Cesari et al. (2008) concluded that dietary inclusion of a formic and lactic acid blend (0.5 %) in rabbit diets did not significantly influence feed intake or growth performance, but it contributed in maintaining gut health by limiting undesirable microbial activity. On contrary, Suiryanarayana et al. (2015) found that butyric acid at 0.4 per cent level in piglet diet significantly increased ADG ($p<0.05$), which was accompanied by improved nutrient utilisation and intake.

Feed conversion efficiency (g feed per g gain) of rabbits maintained on four experimental diets T1, T2, T3 and T4 were 4.61 ± 0.42 , 4.32 ± 0.40 , 3.82 ± 0.38 and 4.13 ± 0.41 respectively. Similarly, Cesari et al. (2008) evaluated a blend of formic acid and lactic acid in rabbit diet (0.25%) and reported no significant effect ($p>0.05$) on feed efficiency, with FCR values of 3.72 in the control group and 3.68 in the supplemented group. In contrary, Lin et al. (2023) tested a dietary blend of formic acid, lactic acid and citric acid at 0.25 per cent in New Zealand White

rabbits and reported significant improvement in FCR ($p<0.05$), with values of 3.85 in the control and 3.55 in the supplemented group.

The data on digestibility of nutrients is presented in Table 4. The digestibility coefficients of DM obtained in the present study were 63.30 ± 1.56 , 70.01 ± 1.08 , 75.13 ± 0.99 and 73.35 ± 3.36 per cents for diets T1, T2, T3 and T4, respectively. A significant improvement ($p<0.05$) was observed in the supplemented groups compared to the non-supplemented group. The highest value was recorded in the T3 group, while the lowest was noted in the T1 group fed with the control ration alone. The T2 and T4 groups exhibited comparable DM digestibility, which was higher than that of T1 but lower than T3. Similarly, Lin et al. (2023) reported that supplementation of a blend of organic acids (formic, lactic, and citric acids) at 0.25 per cent level in the diet of New Zealand White rabbits significantly ($p<0.05$) improved DM digestibility, increasing from 65.8 per cent in the control group to 72.6 per cent in the supplemented group.

The digestibility coefficients of CP recorded in this study were 63.27 ± 1.43 , 70.82 ± 0.95 , 76.98 ± 0.96 and 74.13 ± 3.45 per cents for T1, T2, T3 and T4, respectively.

Table 5. Data on faecal pH of rabbits maintained on four dietary treatments

Parameters	Treatments				p value
	T1	T2	T3	T4	
pH	7.48±0.21 ^b	7.02±0.05 ^a	6.93±0.06 ^a	7.08±0.09 ^a	0.027*

¹Mean of six replicates with SE; Means having different small letters as superscript differ significantly within a row;

*-significant ($p < 0.05$)

A significant improvement ($p < 0.05$) was observed in the supplemented groups compared to the non-supplemented group. Higher CP digestibility values were recorded in T3 and T4 groups compared to T1 and T2. Although the CP digestibility in the T4 group was higher than in T1, it was comparable with that of T2 and T3. Similarly, Lin et al. (2023) reported a significant improvement ($p < 0.05$) in protein digestibility when supplemented with a blend of formic, lactic and citric acids at 0.25 per cent level, with digestibility values increasing from 70.5 per cent in the control group to 77.4 per cent in the treated group.

The digestibility coefficients of CF obtained in the present study were 42.87 ± 3.87 , 48.59 ± 1.94 , 62.42 ± 1.56 and 53.49 ± 3.03 per cents for T1, T2, T3 and T4, respectively, following a trend similar to that of crude protein digestibility. Abdel-Hamid and Omar (2019) observed a marked improvement ($p < 0.01$) in CF digestibility with the inclusion of formic acid at 0.5 per cent level, reporting values of 35.7 per cent in the control group and 42.6 per cent in the treated group.

The digestibility coefficients of EE recorded in this study were 73.63 ± 2.23 , 80.63 ± 1.35 , 85.04 ± 0.79 and 83.30 ± 2.22 per cents for T1, T2, T3 and T4, respectively. Significant improvement ($p < 0.05$) was observed in the supplemented groups compared to the non-supplemented group. Similarly, Abdel-Hamid and Omar (2019) reported that formic acid supplementation at 0.5 per cent significantly ($p < 0.01$) enhanced EE digestibility from 69.2 per cent in the control group to 75.8 per cent in the treated group.

The digestibility coefficients for NFE obtained in this study were 73.10 ± 0.99 , 75.30 ± 0.90 , 77.01 ± 1.71 and 77.80 ± 2.81 per cents for T1, T2, T3 and T4, respectively with no significant difference ($p > 0.05$) between the dietary treatments. Chowdhury et al. (2014) also reported no significant effect of citric acid in rabbit diet (0.5%), with NFE digestibility of 75.9 per cent in control and 76.6 per cent in treated group. In contrary, Abdel-Hamid and Omar (2019) reported that formic acid at 0.5 per cent level in rabbit diet significantly increased NFE digestibility ($p < 0.01$) from 75.2 per cent in control to 80.6 per cent in treated group. Lin et al. (2023) also found that a blend of formic, lactic and citric acids at 0.25 per cent level in rabbit diet significantly improved NFE digestibility ($p < 0.05$), with values of 76.0 per cent in control and 81.2 per cent in supplemented group.

Faecal pH

The average faecal pH of rabbits maintained on the four experimental diets T1, T2, T3, and T4 were 7.48 ± 0.21 , 7.02 ± 0.05 , 6.93 ± 0.06 , and 7.08 ± 0.09 respectively, as presented in Table 5. The data revealed a significant ($p < 0.05$) reduction in faecal pH in the organic acid-supplemented groups. Similarly, Lin et al. (2023) reported that supplementation with mixed organic acids in New Zealand White rabbits reduced faecal pH in a dose-dependent manner, with both 1000 mg/kg and 2000 mg/kg levels showing significant reductions.

Conclusion

From the findings of present study, it can be concluded that dietary supplementation of organic acids, particularly formic acid (0.5%), significantly improved nutrient digestibility and faecal pH in New Zealand White rabbits without adversely affecting growth performance, feed intake and feed conversion ratio. Therefore, formic acid at 0.5 per cent level can be recommended as a dietary supplement in rabbit production.

Acknowledgement

The authors are grateful to the Dean, College of Veterinary and Animal Sciences, Mannuthy, for extending the facilities required to conduct this study.

Conflict of interest

The authors declare that they have no conflict of interest.

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