Effect of dietary supplementation of probiotics and enzymes on the haematology of rabbits reared under two housing systems

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Abstract

Aim : To study the influence of housing system and dietary supplementation of probiotics and enzymes on haematological parameters of rabbits.

Materials and Methods: A total of 144 weaned rabbits were divided into 2 groups of 72 in each group and housed under conventional cage system and backyard system. The rabbits in each housing system were divided into 4 groups of 18 in each group and the diets were supplemented with probiotics, enzymes and both.

Results: The housing system and supplementation of probiotics and enzymes did not exert significant influence on any of the haematological parameters studied. However, there was slight positive influence of probiotic and enzyme supplementation on the health status of rabbits as revealed by haematological parameters. The overall mean Total erythrocyte count, total leucocyte count, lymphocytes, neutrophils, eosinophils, monocytes, haemoglobin and packed cell volume were 7.52, 6.29 (103/mm3), 60.27%, 35.71%, 1.35%, 1.92%, 10.67 g/dl and 34.25%, respectively.

Conclusion: Rabbits can be reared on low input backyard system without any adverse effect on health and supplementation of probiotics and enzymes had a positive influence on health status of rabbits.

Key words: dietary supplementation, enzymes, haematology, housing systems, probiotics, rabbits

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Introduction

The role of rabbits to provide a regular supply of high quality protein and income under sustainable systems that utilize renewable resources at minimal costs is presently recognized in many parts of the world, especially in the wake of emerging diseases associated with other species like Avian Influenza, Swine flu etc.. Rabbits have number of characteristics such as small body size, short generation interval, high reproductive potential, rapid growth rate, genetic diversity and the ability to utilize forages and byproducts as major diet components that make them suitable as meat-producing small livestock [1]. Combes [2] reported that rabbit meat contains 21.0 +1.5% protein, 1.2 + 0.1% minerals and 59 mg/100g cholesterol. A ratio of omega 6/omega 3 of 5.9 make rabbit meat attractive for health purposes. Rabbit meat is a highly digestible, low calorie food, often recommended by nutritionists over other meats [3].

The world rabbit meat production is estimated to be 1.69 million tons in the year 2010 [4]. However, the

share of India is negligible. In developing countries such as India, where enormous meat shortages exist, the potential for rabbit production is greatest. Rabbit rearing is gaining momentum because of the fact that meat from rabbit is low in cholesterol and delicious. In fact, backyard rabbitries are best suited for our country to increase the per capita income and protein availability. In view of the higher initial capital investment in providing cage system of housing, the proposed study is aimed to evaluate the effect of raising rabbits under back yard system of rearing. Comparative analysis of the production/reproduction parameters vis-a-vis production cost in the 3 groups favoured the non-conventional housing system (bare floor), which is cost effective in terms of production and also guarantees lower mortality and higher production [5].

Of late, use of antibiotics as growth stimulators is being discouraged. The use of microorganism (probiotics) as feed additives is currently widely promoted as an "alternative" to antibiotic growth promoters [6]. Similarly, use of Fibrolytic, proteolytic and lipolytic enzymes to improve digestibility of nutrients is being explored [7]. The level of nutrition greatly influences productivity in livestock. It is therefore pertinent to assess the health status of the animals used in various feed trial, and one of the best way to assess it, is the use of hematological parameters [8,9]. Therefore this study was undertaken to investigate the effects of supplementation of probiotics and enzymes on hematological parameters and performance of broiler rabbits in two different housing systems.

Materials and Methods

Permission from Animal Ethics Committee: The research work pertaining to the above article has been carried out with the permission of Institutional Animal Ethics Committee, as per the laws in force.

A total of 144 weaned rabbits (28 days) belonging to 3 breeds viz. New Zealand White, Grey Giant and Flemish Giant, maintained at the "Rabbit Production for Meat" scheme of the Department of Animal Genetics and Breeding, College of Veterinary Science, Hyderabad were divided into two groups and were reared in conventional cage system and backyard rearing system, until 16 weeks of age. Each group was subdivided into 4 groups each consisting of 18 rabbits. The three breeds were equally represented in all the groups. Rations supplemented with probiotics and enzymes were fed to three groups of rabbits while one group was fed control ration.

Housing and management: Bunnies under cage system of rearing were housed in galvanized iron wire net cages arranged in rows on an iron frame in "M" shape at a height of about 2.5 feet from the floor. Each bunny was provided a cage floor area of 1 square foot by accommodating 6 bunnies in one cage of 3.0 feet length, 2.0 feet width and 1.25 feet height. Cages were equipped with automatic waterers and earthen pots were used as feeders. The asbestos roof of the rabbitry was covered with a thin layer of paddy straw, which was wetted with overhead water sprinklers during hot periods of the day. Gunny cloth curtains were hung around the sheds to protect the animals from extreme temperatures.

For backyard system, a thatched roof shed with gravel floor and chain link wire mesh walls was erected and rabbits were provided floor space of about 2 square feet per bunny. Concentrate feed and fresh and clean drinking water were made available adlibitum by using earthen bowls.

Experimental rations and feeding: The four

experimental rations were – T1 was control ration without any supplement; T2 was T1 + Probiotic (*Saccharomyces boulardi* 50% and *Pediococcus acidilacticii* 50%, 109 CFU/gm of feed; T3 was T1 + Kemzyme HF @ 500 gms/Ton of feed and T4 was T1 + Probiotic + Enzyme at above levels. Kemzyme HF contained Cellulase (>1,00,00,000), Xylanase (>26,00,000), Pectinase (>2,50,000), glucanase (>10,00,000), -Amylase (>7,00,000), Protease (>6,00,000) and Lipase added kemin Units per kg product.

The concentrate feed mixture contained Maize (50%), Groundnut cake (24%), Wheat bran (25%) and Mineral mixture (1%). The CP and DCP content of concentrate feed was 15.5 and 12% respectively with a digestibility coefficient of 78. Coccidiostat and Vitamins A, D, E and C were added to the feed mixture at recommended levels. Feed and water were made available *ad-libitum* to all the experimental animals. About 200 gms of Lucerne green fodder was offered to each rabbit daily.

Hematological traits studied: At 16 weeks of age, blood was collected into vaccutainers from ear vein. Hematological parameters such as total erythrocyte count, total leucocyte count, hemoglobin, differential leucocyte count and packed cell volume were estimated by automatic whole blood analyzer (Humacount, Medsource Ozone Biomedical Pvt. Ltd). The data recorded on different traits were subjected to least squares analysis [10] using the General Linear Model procedure of SPSS version 15.

Results

The mean hematological parameters in broiler rabbits as affected by rations in rabbits reared under cage and backyard systems of housing are presented in Tables-1.

Total erythrocyte count (TEC): The overall mean total erythrocyte count of rabbits was 7.52 ± 0.12 and ranged from 7.49 ± 0.16 in cage system to 7.54 ± 0.19 (106/mm3) in backyard systems with non-significant differences. The mean TEC in rabbits fed experimental rations T1, T2, T3 and T4 were 7.03 ± 0.19 , 7.22 ± 0.14 , 7.78 ± 0.15 and 7.28 ± 0.26 (x106/mm3) in cage system and 7.47 ± 0.24 , 7.12 ± 0.14 , 7.62 ± 0.32 and 7.95 ± 0.10 in back yard system, respectively. Slightly higher values were observed in rabbits fed experimental rations but, the differences were non-significant statistically.

Total leucocyte count (TLC): The mean total leucocyte count of rabbits was 6.29 ± 0.08 and ranged from 6.13 ± 0.10 in cage system to 6.46 ± 0.13

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Table-1. Mean haematological parameters of rabbits reared under cage and backyard systems as affected by experimental rations

Parameter	System	Overall		T1		T2		Т3		T4	
		Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
TEC (10 ⁶ /mm3)	Cage	7.49	0.16	7.03	0.19	7.22	0.14	7.78	0.15	7.28	0.26
	Backyard	7.54	0.19	7.47	0.24	7.12	0.14	7.62	0.32	7.95	0.10
TLC (10 ³ /mm3)	Cage	6.13	0.10	5.94	0.16	6.02	0.19	6.34	0.20	6.24	0.26
	Backyard	6.46	0.13	6.21	0.26	6.50	0.19	6.43	0.31	6.68	0.34
Lymphocytes (%)	Cage	57.83	1.73	59.33	3.60	60.33	4.12	56.33	3.52	55.33	3.05
	Backyard	62.71	2.08	62.20	3.10	66.83	2.12	64.00	4.73	57.83	5.89
Neutrophils (%)	Cage	35.46	1.44	36.17	3.30	34.83	2.89	34.17	3.56	36.67	2.38
	Backyard	35.96	1.50	37.10	4.04	38.00	2.11	33.22	3.15	35.50	2.49
Eosinophils (%)	Cage	1.38	0.19	1.17	0.48	1.83	0.31	1.00	0.37	1.50	0.43
	Backyard	1.33	0.19	1.00	0.37	2.00	0.52	1.33	0.21	1.00	0.37
Monocytes (%)	Cage	2.00	0.17	1.92	0.17	2.17	0.31	2.06	0.37	1.83	0.17
	Backyard	1.83	0.21	1.83	0.17	2.00	0.31	1.77	0.40	1.66	0.17
Hb (g/dl)	Cage	10.51	0.08	10.38	0.24	10.60	0.18	10.58	0.17	10.73	0.13
	Backyard	10.83	0.10	10.88	0.10	10.78	0.14	10.74	0.17	10.92	0.11
PCV (%)	Cage	34.17	0.49	34.00	0.68	34.12	0.65	33.84	0.61	34.60	0.63
	Backyard	7.54	0.19	7.47	0.24	7.12	0.14	7.62	0.32	7.95	0.10

(x103/mm3) in backyard system of rearing with nonsignificant differences among the leucocyte count of rabbits reared under the two housing systems. Mean TLC in cage reared rabbits ranged from 5.94 ± 0.16 in T1 to 6.34 ± 0.20 in T3. No significant differences were observed in the TLC due to the influence of feeding experimental rations, though slightly higher values were observed in those fed experimental rations. Under backyard system of housing, the mean TLC in rabbits fed experimental rations T1, T2, T3 and T4 were 6.21 ± 0.26 , 6.50 ± 0.19 , 6.43 ± 0.31 and 6.68 ± 0.34 , respectively. No significant differences were observed due to the influence of feeding experimental rations.

Lymphocytes: The overall least squares mean lymphocyte per cent was 60.27 ± 1.38 and ranged from 57.83 ± 1.73 in cage system to 62.71 ± 2.08 in backyard system of rearing with non-significant differences among the rabbits reared under the two housing systems. The influence of experimental rations was found to be non-significant on the lymphocyte per cent in rabbits reared under both the housing systems. The lymphocyte per cent ranged from 55.33 ± 3.05 in (T4) to 60.33 ± 4.12 (T2) in cage system and from 57.83 ± 5.89 (T4) to 66.83 ± 2.12 (T2) in backyard system.

Neutrophils: The mean neutrophil per cent in rabbit blood was 35.71 ± 1.03 and ranged from 35.46 ± 1.44 in cage system to 35.96 ± 1.50 in backyard system with non-significant differences among the rabbits under the two housing systems. Mean neutrophil per cent in broiler rabbits ranged from 34.17 ± 3.56 to 36.67 ± 2.38 under cage system and from 33.22 ± 3.15 to 38.00 ± 2.11 under backyard system. The influence of experimental rations was found to be non-significant on the neutrophil per cent in blood.

Eosinophils: The mean eosinophil per cent in rabbits was 1.35 ± 0.14 and ranged from 1.33 ± 0.19 in backyard system to 1.38 ± 0.19 in cage system of rearing. The effect of housing system was non-significant on eosinophil per cent. The eosinophil per cent in broiler rabbits reared under cage system of housing ranged from 1.00 ± 0.37 to 1.83 ± 0.31 while it ranged from 1.00 ± 0.37 to 2.00 ± 0.52 in backyard system of rearing. The influence of experimental rations was found to be non-significant under both the systems.

Monocytes: The mean monocyte per cent in rabbits was 1.92 ± 0.13 and ranged from 1.83 ± 0.21 in backyard system to 2.00 ± 0.17 in cage system. The influence of housing system and experimental rations was non-significant on the monocyte per cent. The mean monocyte per cent in rabbits ranged from 1.83 ± 0.17 to 2.17 ± 0.31 under cage system and from 1.66 ± 0.17 to 2.00 ± 0.31 under backyard system.

Hemoglobin: The mean Hemoglobin content of rabbits was 10.77 ± 0.06 and ranged from 10.51 ± 0.08 in cage system to 11.03 ± 0.10 (g/dl) in backyard system of rearing with nonsignificant differences among the rabbits reared under the two housing systems. The influence of experimental rations was also non-significant in rabbits reared under both the housing systems. The hemoglobin content ranged from 10.38 ± 0.24 to 10.73 ± 0.13 in cage reared rabbits and from 10.74 ± 0.17 to 10.92 ± 0.11 in backyard system of rearing. No significant differences were observed among the rabbits fed different experimental rations.

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nt Packed Cell Volume (PCV): The mean PCV of

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rabbits was 34.25 ± 0.37 and ranged from 34.17 ± 0.49 in cage system to $34.33 \pm 0.57\%$ in backyard system of rearing with nonsignificant differences among the rabbits reared under the two housing systems. The PCV ranged from 33.84 ± 0.61 in (T3) to 34.60 ± 0.63 (T4) in cage reared rabbits while it ranged from 33.50 ± 1.00 (T3) to 34.67 ± 0.80 (T2) in backyard reared rabbits. The effect of supplementation of enzymes and probiotics was non-significant on the PCV under both the housing systems.

Discussion

Total erythrocyte count: Housing system did not have any significant effect on the total erythrocyte count of rabbits. The mean TEC of rabbits was $7.49 \pm$ 0.16 and 7.54 \pm 0.19 (x10⁶/mm3) in cage and backyard systems of housing respectively which is within the normal range of $3.8 - 7.9 \times 10^6$ /mm3 [11]. However, in both the housing systems, slightly higher values were observed in rabbits fed probiotics but, the differences were non-significant statistically. The results are in accordance with findings of [12] who reported that TEC increased when broiler chicks were fed diets containing probiotic organisms S.boulardi and P.acidilacticii, indicating a positive influence on the hematological indices like TEC. The results of the present investigation are supported by findings of [13, 14] who recorded highest mean values of TEC in broiler chicks fed Diet supplemented with probiotics. The results also corroborate the findings of [21-23] who observed non significant effect of yeast culture at different levels on total erythrocyte count in broiler chicks while [24] did not report any effect of probiotics supplementation on blood characteristics in pigs. Enzyme supplemented group of rabbits in both the systems of housing showed a non-significant increase in erythrocyte count, similar to the findings of [16, 23].

Total leucocyte count: Leucocytes are cells of immune system chiefly involved in defending the body against both infectious diseases and foreign material. No significant differences were observed in broiler rabbits with respect to leucocyte count in two systems of housing. There was a non significant increase in the TLC in broilers fed rations supplemented with probiotics in both the systems of housing and ranged from 5.94 ± 0.16 in (T1) to $6.34 \pm 0.20(x103/mm3)$ in (T3) in cage system and 6.21 ± 0.26 in (T1) to 6.68 ± 0.34 (x103/mm3) in (T4) under backyard system. The TLC obtained in the present study was within the normal range of 5-8 x 103/mm3 [11]. The results obtained in the present study are in accordance with the findings of [17] who have

reported a significantly higher total WBC count in broiler rabbits fed diets supplemented with probiotics. The results also corroborate the finding of [18] who reported significantly higher leucocyte counts in broiler chicks fed bio active Yeast. While [24] reported no significant effect of addition of probiotics on blood characteristics. The improvement in blood profile with probiotic containing *S. boulardii* indicates its beneficial effect on haemopoiesis and immunostimulatory effect as reported by [19] in broiler chicken. The results also corroborate the findings of [21].

Lymphocytes: No significant differences were observed with respect to mean lymphocyte per cent in broiler rabbits reared under two systems of housing. Rations supplemented with either enzymes, probiotics or both did not exert any significant influence on the mean lymphocyte per cent in broiler rabbits reared under cage or backyard system of housing. The mean lymphocyte percent observed in the present experiment are within the normal physiological range as reported by [11]. The results are in accordance with the findings of [22]. The results of the present investigation are contrary to the findings of [17] who reported significantly higher absolute lymphocyte count in rabbits fed ration containing bioactive yeast. The normal lymphocyte count in the present study is a good indicator, as increased lymphocyte count is a sign of viral infection or any other disease condition. This infers that the broiler rabbits in the present experiment were in a state of sound health.

Neutrophils: There were no significant differences in the mean neutrophil percent in rabbit blood due to the influence of housing system. Supplementation of enzymes, probiotics or both did not have any significant effect on neutrophil per cent either in cage or backyard system. The observations of the present study were in the normal range of 35.02 to 43.2 %, as reported by [11]. Normal neutrophil count observed in the present study indicates that the rabbits are free from any infection or inflammatory conditions as in such cases, there will be a general increase in the neutrophil count.

Eosinophils: Housing system and dietary supplementation of probiotics and enzymes did not exert significant influence on the eosonophil count in broiler rabbits. The mean eosinophil per cent in rabbits was 1.35 ± 0.14 and ranged from 1.33 ± 0.19 in backyard system to 1.38 ± 0.19 in cage system of rearing. The Eosnophil count in the present experiment was within the normal range of 0 to 4% as reported by [11]. Increased eosonophil counts are generally indicative of allergic conditions and parasitic infestation and the normal values obtained in the present study hint at the good health status of the rabbits.

Monocytes: Both the housing system and experimental rations did not exert significant influence on monocyte count. The overall mean monocyte per cent in broiler rabbits was 1.92 ± 0.13 The results of the present experiment were within the normal range of monocytes reported by [11]. As was observed with other hematological parameters, normal range of monocytes is a good indicator of health. Monocytosis may be indicative of various disease states or chronic inflammation.

Hemoglobin: The effect of housing system and experimental rations was found to be nonsignificant on the hemoglobin content of rabbits. Mean hemoglobin content was 10.51 ± 0.08 and 11.03 + 0.10 g/dl in rabbits fed experimental rations and reared in cage and backyard system of rearing, respectively. [11] also reported similar range of haemoglobin content (10 to 15 g/dl) in healthy rabbits.

Packed Cell Volume (PCV): Packed cell volume or erythrocyte volume fraction (EVF) is the percentage of blood volume that is occupied by red blood cells. Mean PCV ranged from 34.17 ± 0.49 in cage system to $34.33 \pm 0.57\%$ in backyard system of rearing with nonsignificant differences among the rabbits reared under the two housing systems. The experimental rations had a non-significant effect on PCV of rabbits reared under both the housing systems. [11, 20] reported mean PCV ranges of 31 to 40 and 30 to 50 per cent, respectively. Abnormal PCV values are indicators of hemorrhagic (low PCV) or hypoxic conditions (high PCV). Normal values of PCV obtained in the present study indicate healthiness of the rabbits.

Conclusion

It can be concluded from the above experiment, that rabbits can be profitably reared under backyard system of housing without any adverse effect on health. The dietary inclusion of probiotics and enzymes will have a positive influence on the health and immune status of rabbits.

Author's Contribution

SCA and MM designed the experiment and implemented the design; GPM analyzed the data and prepared the manuscript; KRK provided the probiotics and enzymes and revised the manuscript. All authors have read the final version of the article and approved.

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Authors declare that they have no conflict of interest

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