Pakistan J. Zool., pp 1-8, 2024. **DOI**: https://dx.doi.org/10.17582/journal.pjz/20230912065722

Effect of Aqueous Extract of *Cichorium intybus* as Alternative Antibiotic Growth Promotor on Growth Performance, Lipid and Mineral Profiles of Broilers

Muhammad Yousaf[1,2], Muhammad Sarwar[1], Irfan Ahmed[2,3]*, Zahid Kamran[2], Dalia Fouad[4], Shahzad Ashraf[1], Farid Shokry Ataya[5], Shakeel Ahmad[6], Abdur Rahman[7], Mubarik Mahmood[7], Ahmad Kamran Khan[8] and Hafiz Ishfaq Ahmad[9]

[1]*Institute of Animal Nutrition and Feed Technology, University of Agriculture, Faisalabad, Pakistan*
[2]*Department of Animal Nutrition, Faculty of Veterinary and Animal Sciences, The Islamia University of Bahawalpur, Bahawalpur, Pakistan*
[3]*Yunnan Provincial Key Laboratory of Animal Nutrition and Feed, Yunnan Agricultural University, Kunming 650201, Yunnan Province, People's Republic of China*
[4]*Department of Zoology, College of Science, King Saud University, PO Box 22452, Riyadh 11495, Saudi Arabia*
[5]*Department of Biochemistry, College of Science, King Saud University, PO Box 2455, Riyadh 11451, Saudi Arabia*
[6]*Department of Poultry Science, Faculty of Veterinary and Animal Sciences, The Islamia University of Bahawalpur, Bahawalpur, Pakistan*
[7]*Department of Animal Sciences, CVAS-Jhang 35200, University of Veterinary and Animal Sciences, Lahore 54000, Pakistan*
[8]*Department of Plant Protection, Ghazi University, Dera Ghazi Khan, Punjab, Pakistan*
[9]*Department of Animal Breeding and Genetics, Faculty of Veterinary and Animal Sciences, Bahawalpur, Pakistan*

Article Information
Received 12 September 2023
Revised 15 November 2023
Accepted 04 December 2023
Available online 28 February 2024
(early access)

Authors' Contribution
Data curation was made by MY, IA AKK, MM, AR. Formal analysis was made by ZK, SA, HIA, SA. Methodology was made by MY, MS, IA, FSA. Software was assigned to ZK, AKK, SA, AR, MM. Writing original draft was done by MY, IA, DF, HIA. Writing review and editing was done by MY, IA.

Key words
Antibiotic growth promoter, Antibiotics, Chicory, Growth performance, Lipid profile, Broiler

ABSTRACT

This study aimed to determine the beneficial effect of chicory leaves extract on growth performance, lipids and minerals profiles of broilers at different pH levels. The influence of *Cichorium intybus* (chicory) leaf extract was determined on day-old chicks (n=150) that were divided into 15 groups, each of 10, for 35 days. These replicates were allotted to five treatment groups, each receiving three replicates. Two experimental diets (starter and finisher) were formulated with and without antibiotic growth promoter (AGP) Enradin and coccidiostat Salinomycin. Birds of positive control (PC) were offered commercial ration supplemented with AGP and Salinomycin along with water without any supplementation, while birds of negative control were fed ration without AGPs but given fresh water without any extract. Moreover, birds of acidic chicory leave extract, neutral chicory leave extract and basic chicory leave extract (BCLE) groups were fed diets without AGPs but given water supplemented with chicory leave aqueous extract, at pH 3, 7 and 12, respectively. Supplementation of chicory leaf extract significantly influenced ($P<0.05$) the weight gain and FCR in BCLE economically better than controls. However, feed intake, serum minerals (Ca, P, Na, and K), and mortality were found non-significantly ($P>0.05$) affected by the chicory leaf aqueous extract. Replacement of AGPs with chicory leaves aqueous extract, extracted at different pH, revealed better performance of broilers in terms of weight gain and efficiency of feed utilization along with better profit margin. Therefore, the use of chicory extract in broiler production may be recommended as an inexpensive but efficient alternative to AGPs.

* Corresponding author: irfanahmad166@yahoo.com
0030-9923/2024/0001-0001 $ 9.00/0

INTRODUCTION

Recent studies indicated that because of extensive use of antibiotics as growth promoters, there is a risk of developing cross-resistance and multiple-antibiotic resistance in harmful bacteria and many other harmful effects (Verdonk *et al*., 2005; Mahdi *et al*., 2022). Antibiotics like tetracycline are the major cause of liver injury which not only alter liver functions but also disturb the metabolism

of primary nutrients (Andrade and Tulkens, 2011). It has been noticed that blood triglyceride concentration may be altered because of an injured or affected liver. Antibiotics have also been reported to disrupt the gut microflora and may allow growth of pathogens leading to compromised production (Levy, 2000). Reduced absorption of minerals has also been reported in birds receiving antibiotics as a growth promoter, which might be attributed to changes in gut environment because of antibiotics failure (Klevay and Milne, 2002). Due to these adverse effects of antibiotics on health and its transmission to the human food chain, the European Commission has phased out and finally banned the selling and use of antibiotics as growth promoters in feed (EC Regulation No. 1831/2003). Poultry nutritionists now-a-days are looking for new substitutes to antibiotic growth promoters that have minimum side effects for poultry which could promote growth and can be equally effective against pathogenic microorganisms of gastrointestinal tract by enhancing weight gain and feed efficiency (Ahmed *et al*., 2022). Due to ban on the use of antibiotics, there is a shift in focus and the use of biological products, such as prebiotics, probiotics, enzymes, organic acids, synbiotics and plant extracts (phytobiotics), as alternatives to antibiotic feed additives in diets for non-ruminant or monogastric animals, has found place in poultry feed (Bedford, 2000). Amongst the various alternatives, plant aqueous extracts are valuable to reducing these problems. Some plants have natural effects like tonics, stimulants, carminative, antiparasitic, antimicrobial, antibacterial, antifungal, and antiseptic (Ferdous *et al*., 2019).

Cichorium intybus (Chicory) is an herbal medicinal plant of the Asteraceae family (Ivarsson *et al*., 2011), containing inulin, flavonoids, coumarins, fructo-oligosaccharides and many vitamins that have anti-bacterial, anti-inflammatory, anti-hypercholesterolemia, hepatoprotective, anti-oxidant, anti-fungal, digestive, diuretic, immuno-stimulant, anti-cancer, gastroprotective, and laxative properties without any significant harmful effects (Kalantari and Rastmanesh, 2009). *C. intybus* is a health-promoting feed ingredient that benefits the host by selectively stimulating the growth and or activity of one or many naturally present or introduced bacterial species in the intestine that can regulate appetite and lipid-glucose metabolism (Glibowski and Skrzypczak, 2017; Song *et al*., 2018). Chicory has shown the ability to decrease colonization of undesirable bacteria such as *Escherichia coli* and *Salmonella* and increase the growth of desirable microorganisms. Prebiotics in poultry maintain normal intestinal microflora by competitive prohibition and antagonism, alter metabolism by decreasing bacterial enzyme activity and increasing digestive enzyme activity (Yaqoob *et al*., 2021; Jin *et al*., 2000; Islam *et al*., 2004). Plant extracts like flavonoids, carotenoids and other substances affect fatty acid accumulation in tissues and control the changes during meat storage (Koreleski and Swiatkiewicz, 2007; Ponnampalam *et al*., 2022).

The negative effects of antibiotics on health have encouraged scientists to find new and safe alternatives to these antibiotics, which may act as growth promoters with limited or no side effects. In this regard, an aqueous extract of *C. intybus* leaves may serve as a potential candidate. Therefore, this study was planned to examine the effect of aqueous extract of chicory leaves extracted at different pH on blood lipid and mineral profiles, feed conversion ratio and weight gain in broiler chicks.

MATERIALS AND METHODS

The study was conducted at the Raja Muhammad Akram Nutrition Research Center, Institute of Animal Nutrition and Feed Technology, University of Agriculture, Faisalabad, to investigate the effect of *C. intybus* leaves extract on growth performance, blood lipids, and minerals profile in broilers. The various performance parameters, such as initial body weight, weekly body weight gain, weekly feed consumption, feed conversion ratio, daily water consumption, mortality, blood lipids (triglycerides, HDL, and LDL), and blood minerals (Ca, P, Na, and K) were recorded.

Preparation of chicory leaves extract

Fresh chicory/Kasni (*Cichorium intybus*) leaves were collected, sliced, meshed into small pieces, and dried under shade. The dried chicory leaves were ground into powder form. The leaf powder was mixed in distilled water @ 2 g / 100ml at three different pH levels, i.e., 3 pH (HCl), 7 pH (distilled water) and 12 pH (NaOH), respectively and then extracted at 80 °C for 3 h (Mavumengwana, 2004). After that, the extract was filtered and then neutralized with HCl or NaOH. These extracts were kept for 48 h at room temperature to be used for the experiment.

Experimental birds

One hundred fifty day-old-broiler chicks (Hubbard) of mixed sexes were purchased from a local hatchery. The chicks were randomly divided into 5 treatments with five replicates experimental units of 10 chicks each. These replicates were further allotted to five treatment groups positive control group (PC, diet with antibiotic Enracin, coccidiostat Salinomycin and fresh water without chicory extract), acidic chicory leave extract group (ACLE, diet without antibiotic but

Table I. Ingredient composition (%) and chemical composition of broiler experimental diets.

	Starter diet	Finisher diet
Ingredients		
Corn	44.47	48.86
Millet	6.50	6.00
Rice polish	13.00	13.00
Wheat bran	7.50	5.20
Corn gluten 30%	2.50	2.50
Corn gluten 60%	2.00	2.50
Rapeseed meal	3.50	3.00
Soybean meal	9.83	8.40
Fish meal	7.50	7.50
Limestone	1.16	1.16
MDCP[1]	0.17	0.17
L-Lysine 78.8%	0.60	0.48
DL-Methionine 99.5%	0.25	0.21
L-threonine	0.05	0.05
Premix	0.30	0.30
Salt	0.067	0.067
Antibiotic (Enracin)	100 g/ton	100 g/ton
Coccidiostat (Salinomycin)	500 g/ton	500 g/ton
Total	100	100
Chemical composition		
Protein %	19.65	18.75
ME (Kcal/Kg)	2850	2965
Fat %	5.26	5.31
Ash %	6.86	6.64
Lysine %	1.20	1.10
Methionine %	0.52	0.52
Ca%	0.98	0.90
Av. Phosphorus%	0.40	0.39
Crude fiber %	5.56	5.02

[1]Monodicalcium phosphate.

fresh water will contain chicory extract), neutral chicory leave extract group (NCLE, diet without antibiotic but fresh water will contain chicory extract), basic chicory leave extract group (BCLE, diet without antibiotic but fresh water will contain chicory extract) and negative control group (NC, diet without antibiotic and fresh water without chicory extract) in such a way that each treatment had three replicates in the trial. Nutrients composition is mentioned in Table I. On 35th day 3 ml blood was collected by wing vein puncture from three birds/replicate. Blood was allowed to clot in syringes (without anticoagulant), and serum was separated and stored in 1.5 ml Eppendorf tubes. Serum samples were stored refrigerated till submitted for analysis of minerals (Na, K, Ca and P) by atomic absorption spectrophotometry (Stef and Gergen, 2012) and lipids (triglycerides, LDL and HDL) by automatic biochemical analyzer Selectra (Jafari *et al*., 2011). Data collected was subjected to statistical analysis using the analysis of variance technique (ANOVA) under completely randomized design (CRD). Treatment means were compared by Duncan's New Multiple Range tests (Steel *et al*., 1996).

RESULTS AND DISCUSSION

Effect on performance characteristics

Table II shows that addition of various chicory leaves extract did not exhibit any significant ($P<0.05$) effect on total feed consumption of the birds of all treatment groups as compared to control groups (positive and negative). However, average feed consumption was numerically higher in the groups given water supplemented with chicory leaves extracts. These findings are in agreement with those observed by Asia and Gultekin (2012) who reported that feed consumption was not affected significantly in broilers treated with chicory leaves extract. It may be due to using chicory extracts in water instead of feed. Contrary to the results of the present study, Safamehr *et al*. (2013), Waldroup *et al*. (2013) and Behboud *et al*. (2011) found increased feed consumption in broilers treated with chicory by using different forms (pulp, powder) as feed additives and it may be due to its better palatability when mixed in feed.

The study results revealed that body weight gain was recorded significantly ($P<0.05$) higher in treatment BCLE, given water supplemented with chicory leaves extract, extracted at 12 pH, compared to other treatment groups. However, the difference in weight gain of the birds given water supplemented with chicory extracts, extracted at 3 pH, 7 pH, and antibiotic-supplemented feed was found to be non-significant ($P>0.05$). The higher body weight gain in birds treated with chicory leaves extract may be due to its complex carbohydrates such as oligofructose and oligosaccharides (Sangoh and Park, 2012; Chambers *et al*., 2011). The use of chicory has been shown to improve body weight gain, which probably is due to improved digestibility of nutrients by reducing microbial competition in the absorption of nutrients from host birds (Yousfi *et al*., 2017) and reducing ammonia production and other growth-depressing metabolites of microbes (Nabizadeh, 2012; Anderson *et al*., 2000).

Table II. Performance characteristics, blood lipids profile and blood minerals profile of broiler birds receiving different levels of aqueous extract of *Cichorium intybus* leaves.

Parameters	Treatment					SEM	P value
	PC[1]	ACLE[2]	NCLE[3]	BCLE[4]	NC[5]		
Performance characteristics							
Feed intake (g)	3060.23	3030.48	3000.76	3066.42	2982.89	31.01	
Weight gain (g)	1818.73^{bc}	1845.96^{ab}	1791.23^{bc}	1966.77^{a}	1690.00^{c}	29.27	
FCR	1.68b^{c}	1.64^{ab}	1.67^{bc}	1.56^{a}	1.76^{c}	0.02	
Water intake	7100.66	7227.66	7368.00	7156.33	7026.00	54.97	
Mortality %	0.66	0.33	0.66	0.66	0.33	0.13	
Blood lipids profile							
Triglycerides (mg/dl)	25.33^{b}	30.00^{b}	40.00^{a}	38.33^{a}	18.00^{c}	2.26	0.00
HDL (mg/dl)	48.33	47.33	49.66	48.33	50.66	0.63	0.54
LDL (mg/dl)	64.66^{b}	84.33^{a}	68.00^{b}	70.33^{b}	40.66^{c}	3.86	0.00
Blood minerals profile							
Ca (mg/dl)	8.76	8.76	8.46	9.00	8.60	0.07	0.21
P (mg/dl)	5.40	5.27	5.23	5.46	5.64	0.09	0.75
Na (mmol/Lit.)	141.33	140.73	141.13	142.13	139.76	0.32	0.23
K (mmol/Lit.)	3.43	3.37	3.47	3.39	3.41	0.01	0.26

Values within the same row which have different superscripts are significantly different (P<0.05). [1]Positive control, [2]Acidic chicory leave extract, [3]Neutral chicory leave extract, [4]Basic chicory leave extract, [5]Negative control.

These findings are similar to those observed by Silava *et al*. (2011), who reported increased body weight gain in broilers treated with chicory leaf extracts. The chicory leaves extract, at pH 12 showed better weight gain than those given water treated with chicory extracts, at pH3 and neutral pH. Maximum body weight gain due to chicory extract, at pH 12, may be due to the reduction in anti-nutritional factors of feed ingredients when treated with an alkaline solution of chicory, as observed by Wah *et al*. (1977). Chicory extracts may also have reduced pathogenic bacterial load in water and gastrointestinal tract, which improves the body weight gain by reduced intestinal thickness, which is very helpful in digestion and absorption of nutrients (Safamehr *et al*., 2013). Moreover, chicory has also been known to improve mucosal growth, villus height, width, crypt depth, and villus height to crypt depth ratio. These factors may stabilize nutrients and increase the digestion and absorption of these nutrients, thus enhancing body weight gain (Awad *et al*., 2011; Waldroup *et al*., 2013).

The addition of various *C. intybus* leaves extract in drinking water exhibited significant (P<0.05) effect on the FCR of the birds of the BCLE group as compared to PC, NCLE, and NC groups. The study's results revealed significant (P<0.05) improvement in FCR of the birds in the treatment BCLE group, which were given water supplemented with *C. intybus* leaves extract, extracted at 12 pH as compared to those of other treatment groups except the ACLE group.

These findings are compatible with those observed by Silava *et al*. (2011), who reported improved FCR in broilers treated with *C. intybus* leaves extract. An improving trend was found in FCR due to the chicory leaves extract, at 12 compared to pH 3 and neutral pH levels. *C. intybus* extracts may have reduced pathogenic bacterial load in water and gastrointestinal tract which improved the body weight gain by reducing intestinal thickness and increasing intestinal length, which is very helpful in digestion and absorption of nutrients (Safamehr *et al*., 2013).

Mortality

The highest mortality was recorded in treatments PC, NCLE and BCLE groups (6.6%), followed by ACLE and NC groups (3.3%). Postmortem findings depicted that the cause of mortality was ascites in almost all the birds because of rapid growth, all these conditions require high oxygen levels in the blood (Kiiskinen, 1985). It was also observed that acidosis affects cellular membrane

Table III. Economic analysis of Broilers receiving different levels of aqueous extract of *Cichorium intybus* leaves.

Parameters	Treatment					SEM
	PC[1]	ACLE[2]	NCLE[3]	BCLE[4]	NC[5]	
Total feed consumed (kg / bird)	3.06	3.03	2.99	3.06	2.98	0.03
Cost of total feed consumed/bird (PKR)	128.52	121.20	119.86	122.66	119.20	1.49
Cost/kg weight (PKR)	71.01^{c}	65.85^{ab}	67.03^{bc}	62.62^{a}	70.81^{c}	0.96
Cost differential/kg gain (PKR)	0.00	5.15^{ab}	3.98^{b}	8.39^{a}	0.19^{c}	0.95
Relative cost benefit/kg gain (%)	100.00^{c}	107.85^{ab}	106.02^{bc}	113.52^{a}	100.38^{c}	1.52

Values within the same row which have different superscripts are significantly different (P<0.05), [1]Positive control, [2]Acidic chicory leave extract, [3]Neutral chicory leave extract, [4]Basic chicory leave extract, [5]Negative control.

integrity and reduce free radical elimination. Resultantly, leakage of blood vessels occurred and accumulated in the abdominal cavity, resulting in the development of ascites (Moschandreou, 2012).

Blood lipids profile

The NC group had the lowest (p<0.05) blood triglycerides, followed by the PC, ACLE, NCLE and BCLE groups. Higher serum triglyceride contents might be due to the increased digestibility of nutrients by the addition of chicory leaves extract, so as a result, more accumulation of triglycerides in the serum. Similar results were reported by Miao *et al*. (2008) in which they found increased levels of serum triglycerides at 0.6% and 1% inclusion levels of chicory leaves extract in broiler birds. In contrast, the results of Safamehr *et al*. (2013) were contrary to a recent study where they observed non-significant differences among all treatment groups with different chicory inclusion levels in bird's diet. Asia and Gultekin (2012) found decreased serum triglycerides by the addition of chicory. Velasco *et al*. (2012) investigated a non-significance decrease in serum triglycerides by supplementing chicory root inclusions. Jafari *et al*. (2011) reported a significant decreasing trend of serum triglycerides when chicory was added to the feed. Navid and Mahmoud (2011) concluded that triglyceride concentrations were markedly reduced (p<0.05) in groups fed 1.5% and 2% chicory compared to the control, as shown in Table II.

The NC group had the lowest (p<0.05) blood triglycerides, followed by the PC, NCLE, BCLE, and ACLE groups. The results of Miao *et al*. (2008) favor our results, where they report an increased level of serum LDL at 0.6% and 1% inclusion of chicory extract in broiler birds. However, findings of Velasco *et al*. (2012) were in contrary to current study results in investigating a significant decrease in serum LDL with the supplementation of chicory root inclusions. Behboud *et al*. (2011) concluded that there was a decrease in the serum LDL with the supplementation of chicory and black seeds together. Jafari *et al*. (2011) observed a significant difference in serum LDL when chicory was added in the feed. Navid and Mahmoud (2011) concluded that serum LDL concentrations were markedly reduced (p<0.05) in groups fed 1.5% and 2% chicory compared to control.

Blood minerals profile

The BCLE group has higher (p>0.05) serum Ca concentration as compared to other treatment groups. The NC group has the highest (p>0.05) serum P concentration which is 5.64 compared to other treatment groups. Numerically maximum serum Na concentration was observed in BCLE group while the lowest in NC group. No significant difference was observed in serum K in all treatment groups. However, the maximum value was observed in the NCLE group, while the lowest value was observed in the ACLE group. The average values for Ca, P, Na, and K are given in Table II.

The results of the present study are in line with the findings of Safamehr *et al*. (2013), who reported that dietary inclusion of *C. intybus* was more beneficial in broiler production. Behboud *et al*. (2011) also reported a reduction in the cost of feed consumed at higher inclusion levels of *C. intybus* leaf pulp. However, in contrast, Liu *et al*. (2011) observed the high broiler production cost by including *C. intybus* as shown in Table III because of the high production, harvesting and processing cost of chicory leaves.

CONCLUSION

Based upon this study, it is concluded that replacement of AGPs with chicory leave aqueous extract, extracted at different pH revealed better performance of broilers in terms of weight gain, increased FCR, good assimilation

of feed ingredients, low mortality rate, better feed intake along with better profit margin in comparison to control group of birds fed with untreated feed. Therefore, the use of chicory extract in broiler feed may be recommended as an inexpensive feed ingredient or additive, but an efficient alternative to AGPs as chicory leaves extract showed no pronounced side effects.

Funding

The authors extend their appreciation to Researchers Supporting Project number (RSPD2023R965), King Saud University, Riyadh, Saudi Arabia for funding this research.

IRB approval

The research was approved from Institutional Review Board.

Ethical statement

The rearing, sampling and culling of the birds were followed by the principles of Animal Ethical Committee of University of Agriculture, Faisalabad.

Statement of conflict of interest

The authors have declared no conflict of interest.

REFERENCES

Ahmed, I., Li, Z., Shahzad, S., Naveed, S., Khan, A.K., Ahmed, A., Kamran, Z., Yousaf, M., Ahmad, S., Afzal, G. and Ahmad, H.I., 2022. Potential probiotics role in excluding antibiotic resistance. *J. Fd. Qual*., pp. 1-20. https://doi.org/10.1155/2022/5590004

Anderson, D.B., Mccracken, V.J., Aminov, R.I., Simpson, J.M., Mackie, R.I., Verstegen, M.W.A. and Gaskins, H.R., 2000. Gut microbiology and growth-promoting antibiotics in Swine. *Nutr. Abst. Rev*., 70: 101-108.

Andrade, R.J. and Tulkenes, P.M., 2011. Hepatic safety of antibiotic used in primary care. *J. Anti-Microb. Chemother*., **66**: 1431-1446. https://doi.org/10.1093/jac/dkr159

Asia, S.E. and Gultekin, Y., 2012. Effects of inulin and β-glucan supplementation in broiler diets on growth performance, serum cholesterol, intestinal length, and immune system. *J. Vet. Anim. Sci*., **36**: 388-394.

Awad, W., Ghareeb, K. and Bohm, J., 2011. Evaluation of the chicory innulin efficacy on ameliorating the intestinal morphology and modulating the intestinal electrophysiological properties in broiler chickens. *J. Anim. Physiol. Anim. Nutr*., **95**: 65-72. https://doi.org/10.1111/j.1439-0396.2010.00999.x

Bedford, M., 2000. Removal of antibiotic growth promoters from poultry diets: Implications and strategies to minimize subsequent problems. *World's Poult. Sci*., **56**: 347-365. https://doi.org/10.1079/WPS20000024

Behboud, J., Ali, R. and Elmira, H., 2011. Comparative effect of chicory (*Cichorium intybus*) and *Nigella sativa* extract with an antibiotic on different parameters of broiler chickens. *J. appl. Environ. Biol. Sci*., **1**: 525-528.

Chambers, J.R., Spencer, J.L. and Modler, H.W., 2011. The influence of complex carbohydrates on *Salmonella typhimurium* colonization, pH, and density of broiler ceca. *J. Poult. Sci*. **76**: 445-451. https://doi.org/10.1093/ps/76.3.445

Ferdous, M.F., Arefin, M.S., Rahman, M.M., Ripon, M.M.R., Rashid, M.H., Sultana, M.R., Hossain, M.T., Ahammad, M.U. and Rafiq, K., 2019. Beneficial effects of probiotic and phytobiotic as growth promoter alternative to antibiotic for safe broiler production. *J. Adv. Vet. Anim. Res*. **6**: 409. https://doi.org/10.5455/javar.2019.f361

Glibowski, P. and Skrzypczak, K. 2017. Prebiotic and synbiotic foods. In: *Microbial production of food ingredients and additives*. Academic Press. pp. 155-188. https://doi.org/10.1016/B978-0-12-811520-6.00006-4

Islam, M.W., Rahman, M.M., Kabir, S.M.L., Kamruzzaman, S. and Islam, M.N., 2004. Effects of probiotics supplementation on growth performance and certain hemato-biochemical parameters in broiler chickens. *J. Vet. Med*., **2**: 39-43. https://doi.org/10.3329/bjvm.v2i1.1933

Ivarsson, E., Frankow-Lindberg, B.E., Andersson, H.K. and Lindberg, J.E., 2011. Growth performance, digestibility and faecal coliform bacteria in weaned piglets fed a cereal-based diet including either chicory (*Cichorium intybus*) or ribwort (*Plantago lanceolata*) forage. *Animals*, **5**: 558–564. https://doi.org/10.1017/S1751731110002193

Jafari, B., Ali, R. and Elmira, H., 2011. Comparative effect of chicory (*Cichoriumintybus*) and *Nigella sativa* extract with an antibiotic on different parameters of broiler chickens. *J. appl. environ. biol. Sci*., **1**: 525-528.

Jin, L.Z., Ho, Y.W., Abdullah, N. and Jalaludin, S., 2000. Digestive and bacterial enzyme activities in broiler fed diets supplemented with *Lactobacillus* cultures. *J. Poult. Sci*., **79**: 886-891. https://doi.org/10.1093/ps/79.6.886

Kalantari, H. and Rastmanesh, M., 2009. Protective property of *Cichorium intybus* in CCl4 induced liver damage in mice. *Arch. Iran. Med*., **3**: 46-47.

Kiiskinen, N.T., 1985. The effect of diet supplementation with reagent rapeseed meal on performance of broiler chicks. *Nutr. Abst. Rev*., **55**: 42.

Klevay, L.M. and Milne, D.B., 2002. Low density magnesium increases supraventricular ectopy. *Am. J. clin. Nutr*., **75**: 550-554. https://doi.org/10.1093/ajcn/75.3.550

Koreleski, J. and Swiatkiawicz, S., 2007. Dietary supplementation with plant extracts, xanthophylls and synthetic antioxidant: Effect on fatty acid profile and oxidative stability of frozen stored chicken breast meat. *J. Anim. Feed Sci*., **16**: 463–471. https://doi.org/10.22358/jafs/66802/2007

Levy, J., 2000. The effects of antibiotic use on gastrointestinal function. *Am. J. Gastroenterol*., **95**: 8-10. https://doi.org/10.1016/S0002-9270(99)00808-4

Liu, H.Y., Iverson, E., Jonsson, L., Holm, L., Lundh, T. and Lindberg, J.E., 2011. Growth performance, digestibility, and gut development of broiler chickens on diets with inclusion of chicory (*Cichorium intybus L.*). *Poult. Sci*., **90**: 815-823. https://doi.org/10.3382/ps.2010-01181

Mahdi, I., Fahsi, N., Hijri, M. and Sobeh, M., 2022. Antibiotic resistance in plant growth promoting bacteria: A comprehensive review and future perspectives to mitigate potential gene invasion risks. *Front. Microbiol*., **13**: 999988. https://doi.org/10.3389/fmicb.2022.999988

Mavumengwana, V.B., 2004. Isolation, purification and characterization of inulin and fructooligosaccharides from *Chicorium intybus* and inulinase from *Aspergillus niger,* MS dissertation, Rhodes University. pp. 112.

Miao, X., Tianming, H. U., Cunlin, Z., Quanzhen, W., Changhui S. and Weize, S., 2008. Effect of water-soluble extract of chicory on slaughter performance and lipid metabolism of broilers. *Acad. J. Electron. Mag. Northw. A&F Univ. Yangling Shaanxi*, 712100, DFA. 31650.

Nabizadeh, A., 2012. The effect of inulin on broiler chicken intestinal microflora, gut morphology and performance. *J. Anim. Feed Sci*., **21**: 725-734. https://doi.org/10.22358/jafs/66144/2012

Navid, H.M. and Mahmoud, P.M., 2011. Comparative effect of using neem, nishyinda and papaya and probiotic on performance and serum composition of broiler chickens. *Annls biol. Res*., **2**: 373-378.

Ponnampalam, E.N., Kiani, A., Santhiravel, S., Holman, B.W., Lauridsen, C. and Dunshea, F.R., 2022. The importance of dietary antioxidants on oxidative stress, meat and milk production, and their preservative aspects in farm animals: Antioxidant action, animal health, and product quality-Invited review. *Animals*, **12**: 3279. https://doi.org/10.3390/ani12233279

Safamehr, A., Fallah, F. and Nobakht, A., 2013. Response of broilers to the diets consisting of chicory and nettle. *Iran. J. appl. Anim. Sci*., **3**: 131–137.

Sangoh, P. and Park, B.S., 2012. Effect of feeding inulin oligosaccharides on cecum bacteria, egg quality and egg production in laying hens. *Afr. J. Biotech*., **11**: 9516-9521. https://doi.org/10.5897/AJB12.5250

Silava, W.T.M., Nunes, R.V. and Pozza, P.C., 2011. Evaluation of inulin and probiotic for broiler chickens. *J. Anim. Sci*., **33**: 19-24.

Song, J., Li, Q., Li, P., Liu, R., Cui, H., Zheng, M., Everaert, N., Zhao, G. and Wen, J., 2018. The effects of inulin on the mucosal morphology and immune status of specific pathogen-free chickens. *Poult. Sci*., **97**: 3938-3946. https://doi.org/10.3382/ps/pey260

Steel, R.G.D., Torrie, J.H. and Dickey, D.A., 1996. *Principles and procedures of statistics. A biometric approach* (3rd ed.). McGraw Hill Book Co. Inc. New York. USA. pp. 666.

Stef, D.S. and Gergen, I., 2012. Effect of mineral-enriched diet and medicinal herbs on Fe, Mn, Zn, and Cu uptake in chicken. *Chem. Central J*., **6**: 1-9. https://doi.org/10.1186/1752-153X-6-19

Velasco, G., Sánchez, C. and Guzmán, M., 2012. Towards the use of cannabinoids as anti-tumour agents. *Nat. Rev. Cancer*, **12**: 436-444. https://doi.org/10.1038/nrc3247

Verdonk, J.M., Shim., A.J., van, S.B., Leeuwen, P. and Verstegen, M.W.A., 2005. Applications of inulin-type fructans in animal feed and pet food. *Br. J. Nutr*., **93**: 125-138. https://doi.org/10.1079/BJN20041355

Wah, C.S., Sharma, K. and Jackson, M.G., 1977. Studies of various chemical treatment of Sal-seed-meal to remove or inactivate tannins. *Indian J. Anim. Sci*., **47**: 8.

Waldroup, A.L., Skinner, J.T., Heirholzer, R.E. and Waldroup, P.W., 2013. An evaluation of fructo-oligosaccharide in diets for broiler chickens and effects on *Salmonella* contamination of carcasses. *J. Poult. Sci*., **89**: 1651-1662.

Yaqoob, M.U., Abd El-Hack, M.E., Hassan, F., El-

Saadony, M.T., Khafaga, A.F., Batiha, G.E., Yehia, N., Elnesr, S.S., Alagawany, M., El-Tarabily, K.A. and Wang, M., 2021. The potential mechanistic insights and future implications for the effect of prebiotics on poultry performance, gut microbiome, and intestinal morphology. *Poult. Sci*., **100**: 101143. https://doi.org/10.1016/j.psj.2021.101143

Yousfi, Z., Kazemi, F., Rezaei, M. and Ansari, P.Z., 2017. Effect of chicory extract and probiotic on performance, caracas characteristics, blood parameters, intestinal microflora and immune response of broiler chickens. *Iran. J. Anim. Sci. Res*., **9**: 185-195.