www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; SP-11(11): 643-648

www.thepharmajournal.com
Received: 07-08-2022
Accepted: 11-09-2022

Insha Afzal
Division of Livestock Production and Management, Faculty of Veterinary Sciences and Animal Husbandry, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Shuhama, Alusteng, Srinagar, Jammu & Kashmir, India

Azmat Alam Khan
Division of Livestock Production and Management, Faculty of Veterinary Sciences and Animal Husbandry, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Shuhama, Alusteng, Srinagar, Jammu & Kashmir, India

Sofi Umer Bashir
Division of Livestock Production and Management, Faculty of Veterinary Sciences and Animal Husbandry, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Shuhama, Alusteng, Srinagar, Jammu & Kashmir, India

Masood Saleem Mir
Division of Livestock Production and Management, Faculty of Veterinary Sciences and Animal Husbandry, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Shuhama, Alusteng, Srinagar, Jammu & Kashmir, India

Umar Amin
Division of Livestock Production and Management, Faculty of Veterinary Sciences and Animal Husbandry, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Shuhama, Alusteng, Srinagar, Jammu & Kashmir, India

Corresponding Author:
Insha Afzal
Division of Livestock Production and Management, Faculty of Veterinary Sciences and Animal Husbandry, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Shuhama, Alusteng, Srinagar, Jammu & Kashmir, India

Histomorphological and biochemical studies of bovine colostrum fed broiler chicken

Insha Afzal, Azmat Alam Khan, Sofi Umer Bashir, Masood Saleem Mir and Umar Amin

Abstract
The aim of the study was to determine the effect of dietary supplementation of Bovine Colostrum on the liver, intestinal histomorphology and serum biochemistry of broiler chicken. The birds in the control (T_1) group were fed the basal diet whereas in other treatment groups basal diet was supplemented with 1% (T_2), 1.5% (T_3), 2% (T_4) Bovine Colostrum respectively. The results revealed that there was a significant difference in the villus height and thickness of lamina muscular is across the groups whereas no significant difference was found in the depth of crypts. The histomorphological analysis of liver didn't reveal any significant difference between the control group and the colostrum fed groups. The results of serum biochemistry with respect to glucose, total protein and uric acid revealed significant differences in total protein and uric acid levels between the colostrum fed groups and the control group whereas no significant difference with respect to glucose concentration was reported.

Keywords: Bovine colostrum, histomorphology, biochemical, broiler

1. Introduction
Broiler chicks have been shown to benefit from immediate access to feed. Although the focus of nutrition has been on provision of energy, chicks would benefit from a more balanced nutrient profile. To cope with market demand for protein (meat), modern broilers are reaching market age sooner each year [1]. Therefore advances in nutrition will be fundamental to securing this rapid growth achievement and maintaining sustainable broiler production. Accordingly, the common focus of nutrition, to simply supply nutrients for maintenance and growth has become obsolete. Specialist areas such as immune nutrition, are rapidly gaining attention [2, 3]. Therefore during broiler diet formulation, choosing ingredients to maximize nutrient availability, rather than simply meeting energy or amino acid levels, is necessary. Feed additives are substances added to basal diet used in animal nutrition for improving the nutrient utilization or to enhance a particular metabolic activity to improve animal's performance. Feed additives are a group of feed ingredients that can cause desired animal response in a non-nutrient role such as pH shift, growth or metabolic modifiers [4]. Following the ban on the use of antibiotics as growth promoters in animal nutrition by the European Union (EU) in 2006, the nutritionists and researchers attempted other alternatives claiming to enhance the performance of broiler chicken. Common feed additives used in poultry diets include antimicrobial, antioxidants, pH control agents and enzymes [5].
Colostrum is the "early" milk produced by female mammals and in case of bovines it is the first four days of milk post parturition [6]. In addition to proteins, carbohydrates, fats, vitamins and minerals it contains various growth factors, cytokines and nucleosides. It is rich in oligosaccharides, natural antimicrobials, immune regulation factors [7] and anti-oxidative factors. Bovine colostrum contains immuno-globulins such as IgG, IgM, IgA, IgD, and IgE. IgG and IgM play role in control of systemic infections, while as IgA functions within internal body surfaces such as the intestine [8]. Colostrum is a proven effective nutraceutical for the enhancement of immune function in a diverse range of animal species including cattle, horse, pig, sheep, cats, mice, hamsters, ferrets and lizards [9]. However, scanty literature about the use of bovine colostrum in found in poultry.
Since much work has not been reported regarding the use of bovine colostrum in broiler diets, therefore present study was envisioned to study the effect of using bovine colostrum as feed additive in the diets of broiler chicken on the histomorphology of their liver and intestine.

2. Materials and Methods

Bovine Colostrum from Six Crossbred Jersey Cows secreted immediately after calving was procured and stored in sterile containers in packs of 250 ml or less and frozen immediately till use. A biological trial duly approved by Institutional Animal Ethics Committee (IAEC) was conducted on one hundred and ninety two, three days old commercial broiler chicks (Rathi cob) procured from a reputed hatchery and reared for a period of 36 days. The birds were reared on deep litter throughout the experimental period. The chicks were distributed randomly into four treatment groups (T_1, T_2, T_3 and T_4) and each treatment comprised of 4 replicates of 12 birds each. Group T_1 was offered basal diet comprising of commercially available broiler pre-starter and starter rations while T_2, T_3 and T_4 were offered basal diets supplemented with 1, 1.5 and 2% of colostrum/kg feed.

2.1. Histomorphology

For the histomorphological analysis, the tissue samples from the duodenum, jejunum, and the ileum were collected from the slaughtered birds and fixed in 10% buffered formalin saline. Tissues were dehydrated by immersing through a series of alcohols of increasing concentrations (from 70% to absolute), infiltrated with xylene, and embedded in paraffin. Casting of blocks was carried out in Lmolds (two Lshaped pieces) which facilitated the manipulation of size as per the requirement. The rotary type microtome was used for cutting the paraffin sections. The blocks were properly trimmed and the sections of 5 mm thickness were cut. Continuous ribbons (67 inches long) of the material were cut and laid on the surface of constant temperature water bath (around 55 °C). The sections were separated with a heated scalpel after they spread completely. The cut sections were mounted on the clean glass slides using Mayer's egg albumin as the section adhesive. The mounted slides were dried in paraffin oven at 60 °C for one hour. The tissue sections were stained by the Harris haematoxylin and eosin staining method. The paraffin sections were deparaffinised with the xylene before hydration through graded alcohol to distilled water. This was followed by the dehydration in ascending grades of alcohol. The clearing was performed in the xylene and a drop of Distrene Plasticiser Xylene (DPX) mount ant was placed on a cover slip and the section on the slide pressed on it. The slide was inverted and the cover slip was pressed with a rod to remove the air bubbles, if any trapped. The values were measured with an oculometer at a magnification of 4x, 10x and 40x under a light microscope fitted with the stage micro meter. Same procedure was followed for histomorphological analysis of liver samples collected at the time of slaughter.

2.2. Blood Biochemistry

Blood was collected from the birds at 5 weeks of age, in the vials containing EDTA as an anticoagulant. Plasma was harvested following centrifugation at 5000 rpm for 5 minutes, and stored in multiple aliquots, at -40 ºC until used. The parameters given below were estimated from the serum samples with the aid of auto analyser equipment (MS 4) by using respective biochemical kits.

2.2.1. Glucose

Glucose was estimated by Glucose Oxidase-Peroxidise/GOD-POD (Trinder's) method using a diagnostic kit manufactured by Dia Sys Diagnostics India Private Limited.
The assay procedure is summarized in Table-1.

Table 1: Test procedure for glucose

Pipette into test tubes	Blank	Standard	Test
Standard	-	10 µl	-
Sample	-	-	10 µl
Reagent	1000 µl	1000 µl	1000 µl

Mixed well, and incubated for 10min. at 37 °C. Glucose concentration read from auto-analyser (MS 4) calibrated for the purpose against reagent blank at wavelength 505 nm

2.2.2. Total protein

Total Protein was estimated by Biuret method using a diagnostic kit manufactured by DiaSys Diagnostics India Private Limited. The assay procedure is summarized in Table-2.

Table 2: Test procedure for total protein

Pipette into test tubes	Blank	Standard	Test
Standard	-	20 µl	-
Sample	-	-	20 µl
Reagent	1000 µl	1000 µl	1000 µl

Mixed well, and incubated for 5 minutes at room temperature. Total Protein concentration read from auto-analyser (MS 4) calibrated for the purpose against reagent blank at wavelength 546 nm.

2.2.3. Uric acid

Uric acid was estimated by Enzymatic, Colorimetric, Uri case/Peroxidase method using a diagnostic kit manufactured by DiaSys Diagnostics India Private Limited. The assay procedure is summarized in Table-3.

Table 3: Test procedure for uric acid

Blank	Standard	Sample
Enzyme 1.00 ml	1.00 ml	1.00 ml
Solution -	-	-
Sample -	-	0.025 ml
Standard -	0.025 ml	-

Mixed well and incubated for 5 min. at 37 °C. Uric acid concentration read from auto-analyser (MS 4) calibrated for the purpose against reagent blank at within 30 min.

Statistical analysis

The analysis of the data was carried out by using Analysis of Variance (ANOVA). The data generated was classified treatment wise and averages were drawn. After performing statistical analysis the tests were referred by p-values. Any p-value less than or equivalent to 0.05 ($p \leq 0.05$) was taken as statistically significant. The comprehensive statistical package for social sciences (SPSS ver.15.00) Chicago U.S.A. for Windows was used for analysis.

3. Results

3.1 Histomorphology

3.1.1 Liver

Table-4 shows the effect of incorporation of BC on histomorphology of liver of broiler birds. Liver from control group (T_1) revealed mild degeneration with occasional vacuolar changes in hepatocytes associated with kupffer cell hyperplasia. Also, focal mononuclear cell accumulations could be seen in liver parenchyma (Plate 1).

Liver from group T_2 (group supplemented with 1% BC) revealed sinusoidal congestion. Mild hepatocellular degeneration with increased eosinophilia and rounding of cells could be seen. Also, kupffer cell hyperplasia could be observed (Plate 2).

Liver from group T_3 (group supplemented with 1.5% BC) revealed mild to moderate sinusoidal congestion with slight dilatation of sinusoids. Hepatocellular degeneration associated with focal to diffuse mononuclear and neutrophil infiltration was seen in parenchyma (Plate 3).

Liver from group T_4 (group supplemented with 2% BC) revealed mild congestion with normal hepatocytes and mild kupffer cell proliferation (Plate 4).

Table 4: Effect of incorporation of Bovine colostrum in broiler chicken diet on Histomorphology of liver

	Treatment groups			
	T_1 (0% colostrum)	**T_2 (1% colostrum)**	**T_3 (1.5% colostrum)**	**T_4 (2% colostrum**
Congestion	-	-	+++	+
Hepatocellular degeneration	+	+	+	-
Infiltration	+	++	++	+
Kupffer cell hyperplasia	+	+	-	

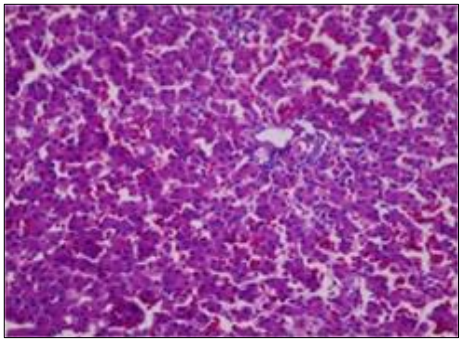

Plate 1: Histological section of liver of control group (T_1). HE x 40 (oz x 4)

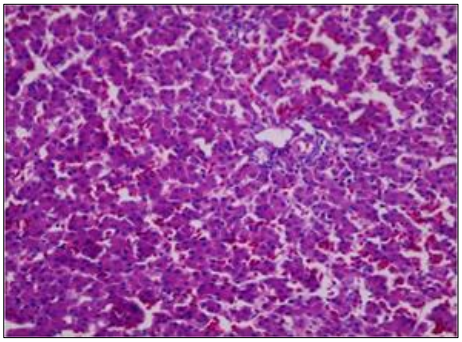

Plate 2: Histological section of liver of T_2.HE x 40(oz x 4)

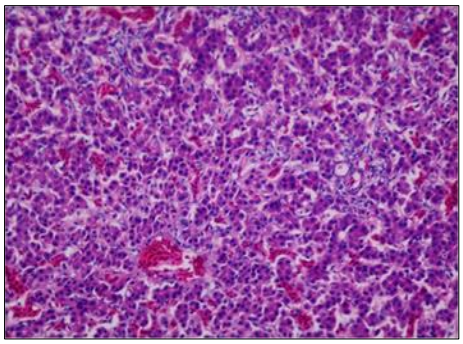

Plate 3: Histological section of liver of T_3.HE x 40(oz x 4)

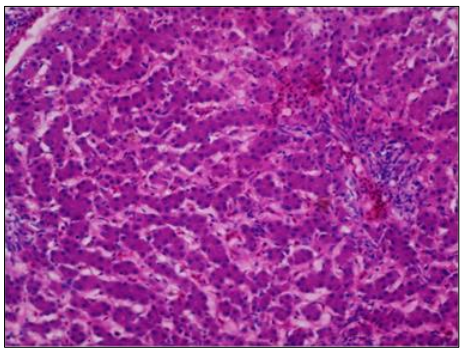

Plate 4: Histological section of liver of T_4. HE x 40(oz x 4)

3.1.2 Intestine

Table-5 shows the effect of incorporation of BC on histomorphology of different intestinal segments of broiler birds. The villus height of groups T_1, T_2, T_3 and T_4 was 509.35±13.61, 699.74±19.00, 706.08±23.98 and 962.00±13. 1µm respectively and thickness of lamina muscular is in the groups was 130.34±3.69, 171.27±3.94, 193.45±1.01 and 221. 43±8.41 µm respectively. Height of villi and thickness of lamina muscular is showed increasing trend and were significantly higher in T_2, T_3 and T_4 as compared to T_1, highest being in T_4. Crypt depth was 86.47±2.44, 76.55±1.72, 72.96±3.29 and 71.12±1.58µm in the groups T_1, T_2, T_3 and T_4 respectively. Crypt depth showed no significant difference between different groups (Plates 5-8).

Table 5: Effect of incorporation of Bovine colostrum in broiler chicken diet on Histomorphology of intestine

	Treatment groups			
	T_1 (0% colostrum))	**T_2 (1% colostrum)**	**T_3 (1.5% colostrum)**	**T_4 (2% colostrum**
Villus height(µm)	509.35±13.61^a	699.74±19.00^b	706.08±23.98^c	962.00±13.1^d
Crypt depth(µm)	86.47±2.44	76.55±1.72	72.96±3.29	71.12±1.58
Lamina muscular is (µm)	130.34±3.69^a	171.27±3.94^b	193.45±1.01^b	221.43±8.41^c

Means within the same row with different superscripts are significantly different ($p \leq 0.05$)

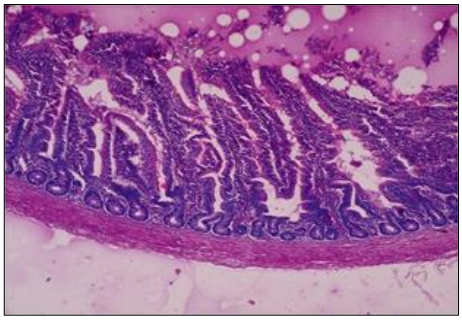

Plate 5: Histological section of intestine of T_1. HE x 10(oz x 4)

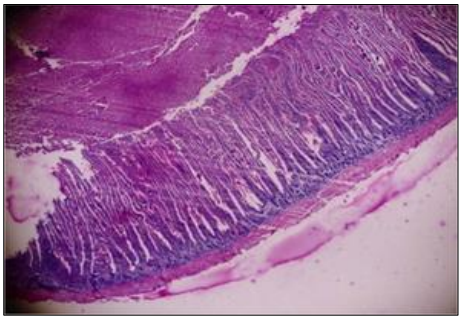

Plate 6: Histological section of intestine of T_2. HE x 4 (oz x 4)

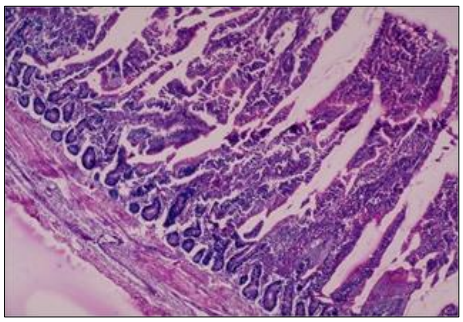

Plate 7: Histological section of intestine of T_3. HE x 10 (oz x 4)

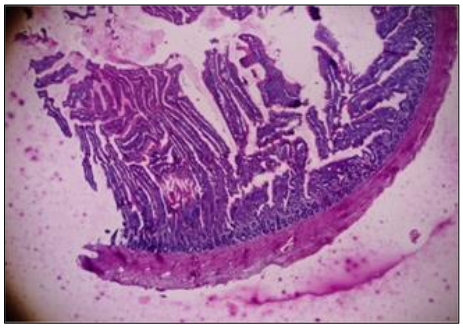

Plate 8: Histological section of intestine of T_4. HE x 4 (oz x 4)

3.1.3. Blood Biochemical Studies

Data pertaining to serum constituents in broiler chicken fed diet with different percentages of BC have been set out in Table-6.

The serum glucose levels in different treatment groups varied between 221.32±13.24 and 319.14±27.99 mg/dl with no statistical significance ($p<0.05$) in serum of broilers among the various treatment groups.

Values for serum uric acid ranged between 4.66±0.55 and 7.21±0.96 ml/dl. A significant difference was observed in birds fed with diets supplemented with 1.5% (T_3) and 2% (T_4) as compared to the control group (T_1). Among the various BC treated groups no significant difference was found with respect to serum uric acid level.

With respect to total protein concentration in serum,T_4 (group supplemented with 2% BC in diet) was found to have significantly higher serum protein concentration as compared to the control group (T_1) as well as among the various BC treated groups. Values ranged between 3.73±0.23 and 10.42±1.17 g/dl.

Table 6: Effect of incorporation of Bovine Colostrum in broiler chicken diet on blood biochemistry

Parameter	**Treatment groups**			
	T_1 (0% colostrum)	**T_2 (1% colostrum)**	**T_3 (1.5% colostrum)**	**T_4 (2% colostrum**
Total protein(g/dl)	3.73±0.23^a	4.89±0.24^a	5.48±0.26^a	10.42±1.17^b
Uric Acid(ml/dl)	4.66±0.55^a	6.03±0.56^{ab}	7.21±0.96^b	7.18±0.48^b
Glucose(mg/dl)	269.96±48.18^a	319.14±27.99^a	221.32±13.24^a	259.36±25.32^a

Means within the same row with different superscripts are significantly different ($p \leq 0.05$)

4. Discussion

4.1. Histomorphology

The beneficial effect of BC supplementation on intestinal histomorphology in terms of intestinal villi and lamina muscular is but no effect on histomorphology of liver was observed in our experiment. Several scientific studies have shown that the addition of probiotic products to broiler diets can have a positive effect on gut morphology and consequently, on performance parameters. These histomorphological changes are represented by elongated villi and a higher villi/crypt ratio, which indicates a lower rate of enterocyte-cell migration from the crypt to the villus. When less energy is needed for renewal of the gut epithelium, more energy is available for growth [10]. The result of this study is in agreement with that of [11] who reported that the addition of the probiotic *E. faecium* increased villus height in the ileum of broiler chicken and that of [12] who found an increase in the number of villi per unit length in duodenum of birds given dietary treatment of antibiotic growth promoter, Virginamyc in as compared to the control. In piglets, BC has been shown to enhance mucosa restoration by stimulating migration of epithelial cells along the crypt-villous axis in intestine, and by decreasing apical cell apoptosis [13]. Bovine colostrum helped to preserve intestinal integrity after weaning, especially in the duodenum where bovine colostrum maintained higher villi (+ 10 to 20%) and reduced crypt depth (- 6%), the first week PW compared to a control treatment [13, 14, 15]. The reduction in crypt depth observed in piglets offered the BC diet suggests that the greater villus height observed in these animals is attributable to decreased villus atrophy rather than increased epithelial mitosis [16]. Similarly, [17] found increase in thickness of muscle layer (TML) in catfish, *Pseudoplatystoma fasciatum* on BC supplementation. An improvement in histology of liver on BC supplementation has been reported [18] that was not found in our study. The beneficial effect of BC on intestinal histology could be attributed to the various growth factors present in BC like IGF, PDGF, TGF'S, etc which promote cell growth and differentiation. Increasing the villus height would suggest an increased surface area capable of greater absorption of available nutrients, and enterocytes have more time to fully differentiate and to fulfil their digestive and absorptive functions. The villus crypt is considered as the villus factory, and deeper crypts indicate fast tissue turnover to renew the villus in response to normal sloughing or inflammation resulting from pathogens or their toxins. Because the energy required to maintain the gut accounts for some 25 percent of the total basal metabolic needs of an animal, any reduction of need for renewal of gut tissue can have a significant impact on the amount of energy available for growth and caloric conversion efficiency [10].

4.2. Blood Biochemistry

There was a significant ($p \leq 0.05$) effect on blood biochemical parameters like total protein (TP) and uric acid but no significant effect on glucose. Total protein concentration showed an increasing trend with the increase in BC supplementation across the groups. The results of this study are in agreement with those [19] who found that the concentration of total protein in serum of blood collected on day 21 was greater ($p \leq 0.05$) for Dried Whey (DW) fed birds compared with the control group of chickens. The results of this study also coincide with those [20] who reported a significant increase in serum total protein concentration on supplementation of 2% natural spices and suggested that elevated serum TP level may be due to high level of protein and other nutrients in *T. foenum graceum* seeds and increased body weight gain. Therefore, it can be concluded that increased total protein concentration in blood in our study could be due to high nutritional value of BC and increased body weight gain. Also, serum protein levels are sensitive to nutritional influences [21]. In some cases, increased serum TP may accompany acute inflammatory states, dehydration, or secondary to certain types of tissue damage [22] so further investigation is required. However, results as obtained under the present study are in contradiction to those reported by [23] who reported that the treatment with Dried Whey (DW) significantly reduced plasma content of total protein with increasing DW level in the diet. The result of this study also didn't agree with other workers [24] who reported significantly lower concentration of serum uric acid levels on *Curcuma longa* supplementation. Uric acid is the major avian nitrogenous waste product and an important anti oxidative agent [25, 26]. Any changes in protein catabolism are mainly reflected in serum uric acid concentrations. Since the findings of this study reveal increased total protein concentration in blood, this might have resulted in increased blood uric acid levels due to higher protein catabolism. The results of this study regarding blood glucose concentration are not in agreement with those workers [27] who observed that the supplementation of 500 mug/kg Cr decreased the serum glucose ($p \leq 0.05$) concentration in blood and who observed that Cinnamon powder, at its lowest level, significantly increased average glucose level in blood at 7, 42 and 49 days of age [28].

5. Conclusion

It was thus concluded that bovine colostrum when incorporated in broiler diets has a beneficial effect on intestinal morphology with no adverse effect on liver histomorphology. The serum uric acid and total protein concentration were increased by BC supplementation.

6. References

1. Kleyn R, Chrystal P. Feeding the young broiler chicken in practice: a review 23rd World's Poultry Congress. Brisbane, Australia; c2008.
2. Field CJ, Johnson I, Pratt VC. Glutamine and arginine: immune nutrients for improved health. Medicine & Science in Sports & Exercise. 2000;32:S377-S388.
3. Okamoto Y, Okano K, Izuishi K, Usuki H, Wakabayashi H, Suzuki Y. Attenuation of the systemic inflammatory response and infectious complications after gastrectomy with preoperative oral arginine and ω3 fatty acids supplemented immune nutrition World Journal of Surgery. 2009;33:1815-1821.
4. Hashemi SR, Davoodi H. Phytogenics as new class offeed additive in poultry industry. Journal of Animal and Veterinary Advances. 2010;9(17):2295-2304.
5. Fulton RM, Nersessian BN, Reed WM. Prevention of *Salmonella enteritidis* infection in commercial ducklings by oral chicken egg-derived antibody alone or in combination with probiotics. International Journal of Poultry Science. 2002;81(1):34-40.
6. Gopal PK, Gill HS. Oligosaccharides and glycoconjugates in bovine milk and colostrum. British Journal of Nutrition. 2000;84(S1):69-74.
7. Kelly GS. Bovine colostrums: A review of clinical uses. Alternate Medicine Reviews. 2003;8:378-394.

8. Muller LD, Ellinger DK. Colostral immunoglobin concentrations among breeds of dairy cattle. Journal of Dairy Science. 1981;64:1727-1730.
9. Taillon C, Andreasen A. Veterinary Nutraceutical Medicine. Canadian Veterinary Journal. 2000;41:231-234.
10. Mohnl M. Improving poultry's intestinal morphology, performance with feed additives; c2011. www.wattagnet.com.
11. Samli HE, Senkoylu N, Koc F, Kanter M, Agma A. Effects of Enterococcus faecium and dried whey on broiler performance, gut histomorphology and intestinal microbiota. Archives of Animal Nutrition. 2007;61(1):429.
12. Miles RD, Butcher GD, Henry PR, Littell RC. Effect of antibiotic growth promoters on broiler performance, intestinal growth parameters, and quantitative morphology. Poultry Science. 2006;85(3):476-85.
13. Huguet A, Le Normand L, Fauquant J, Kaeffer B, Le Huerou-Luron I. Influence of bovine colostrum on restoration of intestinal mucosa in weaned piglets. Livestock Science. 2007;108:20-22.
14. Le Huërou-Luron I, Marion J, Bebin K, Thomas F, Ganier P, Thibault JN, *et al*. Supplementation of a weaning diet with a bovine colostrum extract minimize post-weaning duodenal structural, but not functional, alterations in 7-d old piglet. In: 9th International symposium on digestive physiology in pigs, May 14-17, 2003, Banff, Alberta, Canada. Edmonton, Canada: University of Alberta. 2003;2:238-240.
15. King M, Morel P, Pluske J, Hendriks W. A comparison of the effects of dietary spray dried bovine colostrum and animal plasma on growth and intestinal histology in weaner pigs. Livestock Science. 2008;119:167-173.
16. Al-Mukhtar MYT, Polak JM, Bloom SR, Wright NA. The search for appropriate measurements of proliferative and morphological status in studies on intestinal adaptation. In: Mechanisms of Intestinal Adaptation. (Ed. J. W. L. Robinson, R. H. Dowling and E. O. Riecken) MTP Press: Lancaster; v1982. p. 3-27.
17. Rodrigues APO, Patricia P, Kindlein L, Delgado EF, Cyrino JEP, Machado-Neto R. Intestinal histo morphology in *Pseudoplatystoma fasciatum* fed bovine colostrum as source of protein and bioactive peptides. Scientia Agricola. 2010;67(5):524-530.
18. Abdelmoemin AR, Mahamoud EM, Ghalab EM, Abdelrahman MK. The potential effect of some dairy products on liver functions, immunity and intestinal microbiota in rats. Agricultura. 2008;6:30-37.
19. Szczurek W, Szymczyk B, Arczewska-W Osek A, Jozefiak D, Alloui MN. The effects of dietary whey protein concentrate level on performance, selected intestinal tract and blood parameters, and thiobarbituric acid reactive substances in the liver and breast meat of broiler chickens. Journal of Animal Feed Science. 2013; 22:342-353.
20. Elagib HAA, Nabiela EM, Abbass SA, Genawi TAN. Effect of Natural Spices on Plasma Proteins in Broiler Chicks. Journal of Nutrition and Food Sciences. 2012;2:7.
21. Hoffman WS. The Biochemistry of Clinical Medicine; c1966.
22. Murray RK, Granner DW, Mayes PA, Rodwell VW. Harper Biochemistry. Applton and Lang; c2000.
23. Ashour EA, Abd El-Hack ME, Osman AO. Growth Performance, Carcass Characteristics and Blood Serum Biochemistry of Broiler Chickens Fed Different Levels of Dried Whey. International Journal of Current Research in Biosciences and Plant Biology. 2015;2(6):1-8.
24. Kumari P, Gupta MK, Ranjan R, Singh KK, Yadava R. *Curcuma longa* as feed additive in broiler birds and its patho-physiological effects. Indian Journal of Experimental Biology. 2007;45:272-277.
25. Harr KE. Clinical chemistry of companion avian species: a review. Veterinary Clinical Pathology. 2002;31:140-151.
26. Jurani M, Vyboh P, Zeman M, Lamosova D, Kost'al L, BlaIeek P. Post-hatching dynamics of plasma biochemistry in free-living European starlings (*Sturnus vulgaris*). Comparative Biochemistry and Physiology Part A. 2004;138:89-95.
27. Króliczewska B, Zawadzki W, Dobrzanski Z, Kaczmarek-Oliwa A. Changes in selected serum parameters of broiler chicken fed supplemental chromium. Journal of Animal Physiology and Animal Nutrition. 2004;88(11-12):393-400.
28. Koochaksaraie RR, Irani M, Gharavysi S. The effects of cinnamon powder feeding on some blood metabolites in broiler chicks. Revista Brasileira de Ciência Avícola. 2011;13(3):197-202.