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IMPACT OF CHEESE WHEY PROTEIN ON GROWTH PERFORMANCE OF BROILER: AN APPROACH OF CHEESE WHEY UTILIZATION IN POULTRY FEED

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ABSTRACT

Cheese whey is greenish yellow liquid separated during cheese processing. It accounts 80-90% of milk after cheese processing. It is usually wasted by cheese industries, particularly in developing countries like Pakistan that increases BOD and COD of dairy effluent. Various useful components like lactose, proteins, minerals etc. are present in whey. Among proteins, whey proteins are very effective in body muscle anabolism along with other health benefits. Present research utilized whey proteins in the form of protein supplement. Whey protein was precipitated by combination of pH, temperature and salt treatment followed by filtration. Two types of whey protein concentrates (WPCs) were formed. One was creamy textured while the other was in powder form. WPCs were added in broiler feed at the rate of 0.2% in powder form and 2% in the creamy texture form. Growth parameters like feed consumption, body weight and weight gain increased with whey protein supplement while had no effect on feed conversion ratio (FCR). Carcass traits like carcass, breast, thigh, wings, drumstick weight had significantly increased with the incorporation of whey protein while having non-significant effect on liver weight, GIT weight and GIT/carcass ratio. Whey protein supplementation exhibit no significant influence on packed cell volume (PCV), hemoglobin, lymphocytes and polymorphnuclear leukocyte (PMN) while exhibit significant impact on leukocytes and platelets. It is concluded that only 0.2% WPC (powder) exhibit significant impact on carcass growth while 2% WPC (creamy texture) supplementation improved the growth parameters but statistical analysis revealed it non-significant.

Keywords: Whey, Whey Protein Concentrates (WPCs), Muscle anabolism, Essential amino acid, Fast protein, Hematology

INTRODUCTION

The dairy industry is divided into various sectors and each sector produces different kinds of products. Milk, Yoghurt, cheese, butter and ice-cream are common products of dairy industries. During processing of these products different effluents are produced. Each effluent has different characteristics depending upon the process and the product. All these effluents increase biological oxygen demand (BOD) and chemical oxygen demand (COD) of water, when discharged untreated.

Whey is one of the dairy effluents formed during cheese processing. It accounts 80-90% of total milk volume. Apart from being valued as a medicinal agent in the 17th and 18th centuries, whey has primarily been considered a waste by the dairy industry. In the late 20th century, regulations prevented the disposal of untreated whey. At the same time, recognition of the value of whey components accelerated. Modern science has unraveled the secrets of whey proteins and other whey components, and established a sound basis for their nutritional and functional value. Now it is possible to conserve valuable whey components, including lactose, proteins and minerals that results a variety of products available in the market (**Smither, 2008**).

Whey protein is one of the important components of whey. It is one of the two major proteins of milk that accounts 20% of total milk protein while the rest 80% is casein. Most of casein protein becomes the part of the cheese during cheese production while whey proteins left in the whey (the liquid left after cheese production). Whey protein is a protein complex which contains many kinds of proteins and enzymes like beta-lactoglobulin, alpha-lactalbumin, bovine serum albumin (BSA), lactoferrin, immunoglobulins, lactoperoxidase enzymes and glycomacropeptides. These proteins perform many functions. Whey proteins provide all essential and branch chain amino acids, improves body composition, immune modulation and have antimicrobial activity. In addition, whey protein has the ability to act as an antioxidant, antihypertensive, antitumor, hypolipidemic, antiviral, antibacterial and chelating agent. It also enhances strength of muscles; prevent osteoporosis and cardiovascular disease (**Bjorkman*et al.*, 2012**).

Whey proteins have strong position in sport nutrition. Active people take advantage of whey supplements. Whey proteins are considered as “fast protein”, have capability of muscle development during exercise training. Whey protein is also used in enriching some baking products (**Rostamie*t al.*; 2013**). The amino acid composition of these proteins is similar to that of skeletal muscle so they are directly involved in muscle anabolism along with growth and repair. Amino acids provided by whey proteins are efficiently utilized and absorbed. Supplementation of whey proteins provides a higher lean body mass gain and favorable effect on protein metabolism as compared to an iso-nitrogenous casein protein (**Cribb, 2006**).

Whey and whey proteins also found application in poultry feeding. Whey and its products have been reported to contain unidentified growth factor(s) when added to the diet of chickens. Different researches reported that supplementation of liquid whey, up to a certain level, improves broiler growth. Broiler became lactose intolerant at higher concentration of liquid whey (because of the presence of lactose in whey). This condition causes osmotic diarrhea that lower broiler weight (**Al-sadi*et al.*, 2008**). **Shariatmadari and Forbes. (2005**) concluded through various experiments that at least 1.8 parts of whey added to dry food (wet feeding), or whey offered as drinking liquid by diluted with an equal volume of water or used undiluted whey at alternative days with water, were all possibilities for taking advantage of this by-product. Supplementation of commercial whey protein concentrates at two levels i.e. 8g Kg $^{-1}$ and 32g Kg^{-1} increased carcass yield as compared to control group. However, higher level showed better growth performance (**Szczurek*et al.*,2013**).

Whey proteins can be concentrated and isolated through various techniques including ionic selection (including ion-exchange chromatography, gel filtration) and membrane filtration (ultrafiltration, reverse osmosis, gel permeation), polarity base separation (high performance liquid chromatography). Many of these techniques are not applicable in all cases due to the high cost of equipment, poor yield, less productivity and less selectivity during processing. It cannot be affordable for small industries (Jimenez *et al.*, 2012). Combination of pH, heat and chemical treatment is a technique of protein precipitation to avoid the cost of advance technologies. A number of studies reported that chemicals like NaCl, $CaCl_2$, heat treatment up to 100°C and pH adjustment usually 4 to7 were used for

the precipitation of whey proteins (**Bordenave-Juchereau*et* al., 2005; O'Kennedy and Mounsey, 2009; Stanciuc*et al*., 2012**).

The present research was designed to utilize futile whey by forming whey protein concentrates (WPCs) through a combination of pH, heat and chemical treatment. Keeping in view the lactose intolerance of broiler, implemented method reduce lactose content in WPCs as compare to whey powder that had higher lactose content. The amino acid profile of these proteins compelled to study the impact of these proteins on broiler growth performance by supplementation at minimum level.

MATERIAL AND METHODS

Research was conducted according to the following steps. 1) Preparation of whey protein concentrates (WPCs) from cheese whey 2) Bird housing and WPC supplementation in feed 3) Data recording regarding growth performance 4) Statistical analysis

1. Whey was collected during cheese processing from Technology Transfer Center (Processing Hall) National Institute of Food Science and Technology, University of Agriculture, Faisalabad (Pakistan). Whey was chemically treated by the addition of 6mM $CaCl_2$ @ 6mL per liter of whey and pH was adjusted to 7 by 1N NaOH. After chemical addition, it was heated to 90°C for 20 min proteins were denatured and precipitated. Muslin cloth was used for the filtration of precipitates. Most of soluble components, especially lactose was removed as filtrate and whey proteins left as retentate. These retentates were WPC (creamy texture), stored in freezer for supplementation in treatment C. For treatment B, WPC (creamy texture) was dried at 43°C by spreading on aluminum foil. After drying, hard crumbles were ground to fine powder. This was WPC (powder); stored in polythene bag for supplementation in treatment B. Both WPCs were analyzed for its protein content by using Kjeldhal's method (**AOAC, 2000**).
2. Forty five broiler birds (Hubbard strain), day old, of mixed sexes were purchased from a local hatchery. The chicks were weighed and randomly divided into three experimental units A, B and C with three replications each. The birds were placed and reared in deep litter pens each dimension 5 x 3 x 2.5 feet, which were disinfect and white washed before the start of the experiment. A layer of two inches saw dust was used as litter material in each pen which was stirred regularly during experiment to keep it in dry condition. Birds were vaccinated against ND and IBD disease. Commercially available starter (1-3 week) and finisher (4-5 week) rations were used. Feed of each experimental unit was manually supplemented with WPCs on a weekly basis. Birds had free access to feed and water throughout the experimental period. A treatment plan is elaborated in Table 1.Some growth performance parameters like feed consumption, birds' weight, weight gain and feed conversion ratio were recorded on a weekly basis. Performance of fifth week was recorded after five days instead of seven. After 35 days, one bird from each pen was slaughtered according to Islamic Halaal Principles and weights of carcasses and its different cuts were recorded. Blood samples were collected from wing vein two days before slaughtering and sent to Rehmat Laboratory, Faisalabad (Pakistan) for analysis.
3. Weekly data was analyzed LSD under two factor factorial by using Statistic 8.0 software while after slaughter parameters and hematological parameters were analyzed by using CRD with LSD.

Table 1 Treatment plan for supplementation

Treatment	Feed	Water
A (control group)	Normal feed No protein supplementation	Fresh clean water
B	Supplementation of WPC (powder) 0.2%	Fresh clean water
C	Supplementation of WPC (creamy) 2%	Fresh clean water

RESULTS AND DISCUSSION

Protein content in raw whey varied from 0.5-0.9%. Amount of protein in WPC (creamy texture) increased to 21% after filtration while it was further increased to 40% in WPC (powder). Amount of protein in WPC (creamy) was compared to ricotta cheese because of its processing and appearance in accordance with ricotta cheese (**El-Sheikh *et al*., 2010**) while WPC (powder) had a wide range having a protein content i.e.35-80% (**Bylund, 1995**). Protein content of present finding was in the prescribed range.

Weekly growth performance parameters live bird weight, weight gain, feed intake and FCR depicted in Figure 1, 2, 3 and 4 respectively. In figures mean values sharing different subscript differ significantly. It was observed that supplementation of WPCs did not exhibit any significant effect on body weight, weight gain and feed intake from 1-3 week but at 4^{th} and 5^{th} week statistical analysis showed significant difference. It was evident that treatment A and B were significantly differing from each other while mean values of treatment C was in between the mean values of treatment A and B. Amount of protein in treatment C was less than treatment B that is why mean values of treatment C were higher than treatment A but less than treatment B. Supplementation of whey proteins did not exhibit any significant difference on FCR. A little variation had been observed in different treatment groups, but the overall impact was non-significant. Findings of the present study showed an agreement with **Torki and Molanapour (2005), Karimi (2006), Omara (2012) and Abro*et al.* (2012)**. But the results of FCR were different from **Szczurek*et al.* (2013).** who reported that incorporation of whey protein concentrates at the rate of 32g per Kg had a significant effect on FCR. It might be due to higher level of WPCs that exhibit significant impact on FCR.

Parameters regarding different carcass traits were presented in Table 2. Maximum increase in carcass, breast, and heart, wings, thigh and drumstick weight were observed in treatment B. Treatment C also showed an increase in above mentioned parameters than treatment A (controlled group) but less than treatment B. Non-significant difference had been observed in GIT, GIT/Carcass ratio, body fat and liver weight. Amino acid profile of whey proteins is similar to that of skeletal muscles that is why they are directly involved in muscle anabolism (**Cribb*et al.* 2006**). Present research also showed that it was an amino acid profile of whey protein, which exhibited significant impact on wings, thigh, and breast and drumstick muscles. **Majewska*et al.* (2009)**. conveyed that supplementation of liquid whey increase carcass and thigh weight. Similar findings were shown by **Salahuddin*et al*. (2012)**. where 20% additional protein increase dressed, leg and breast weight. In the study of **Huwaida*et al.* (2013).** only 2% difference of protein level in feed resulted an increase in the live bird, carcass, drumstick, thigh, chest, back and wing weight. Supplement WPC @ 8g and 32g per kilogram of feed that increase breast meat yield while liver weight was not affected (**Szczurek*et al.*, 2013**). **Abro*et al.* (2012)**. stated that replacement of plant protein with animal protein had no significant effect on heart weight while **Huwaida*et al.* (2013)**. indicated that heart weight increased with a high proportion of protein.

Blood image is good signal of health status of animals. It helps to identify the severity of infection and the indirect signal of immune status of the birds. Present research reported (Table 3) that supplementation did not distress hemoglobin, polymorphnuclear leukocytes (PMN), lymphocytes and pack cell volume (PCV) while it increased number of white blood cells and platelets. Increase in the number of platelets is a healthy sign. It was due to bioactive components of whey proteins but a higher number of white blood cells indicated sign of any disease. **Ahmed *et al*. (1994), Donkoh*et al*. (1999), Odunsi*et al*. (1999).** reported that hematological parameters were unchanged in protein treatment. Reason behind increase in WBC might be temperature fluctuation during storage. WPC is a sensitive product, especially creamy textured WPC. It was stored in the freezer, but due to unavoidable load shedding it undergoes temperature fluctuation that might cause undesirable changes in it and highest number of white blood cells in treatment B so it is recommended that try to use fresh creamy texture WPC to avoid any harmful effect on hematology. WPC (powder) was preferred to dry in constant temperature to avoid any undesirable changes.

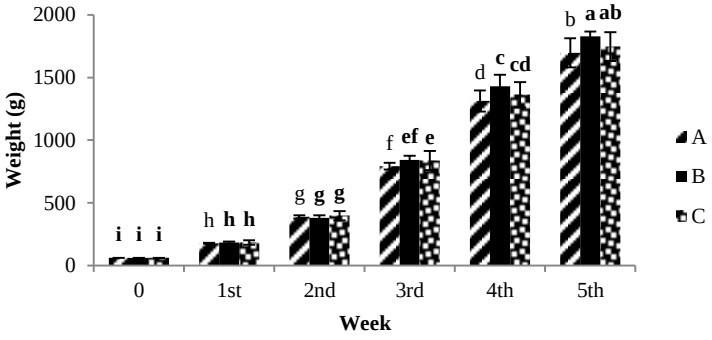

Figure 1 Comparison of means of weekly live bird weight of broiler
A= Normal feed (controlled)
B= Feed supplement with 0.2% whey protein (powder)
C= Feed supplement with 2% whey protein concentrate (creamy)

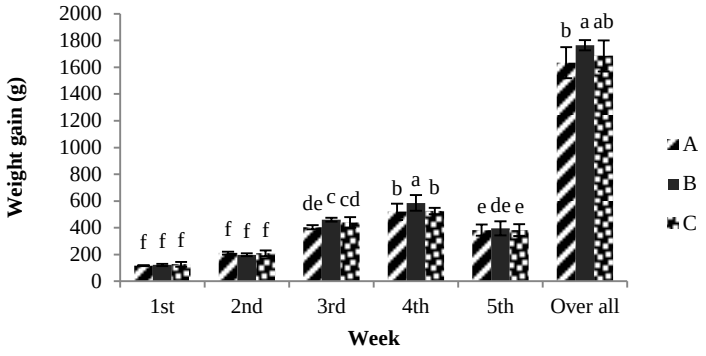

Figure 2 Comparison of means of weekly weight gain of broiler
A= Normal feed (controlled)
B= Feed supplement with 0.2% whey protein (powder)
C= Feed supplement with 2% whey protein concentrate (creamy)

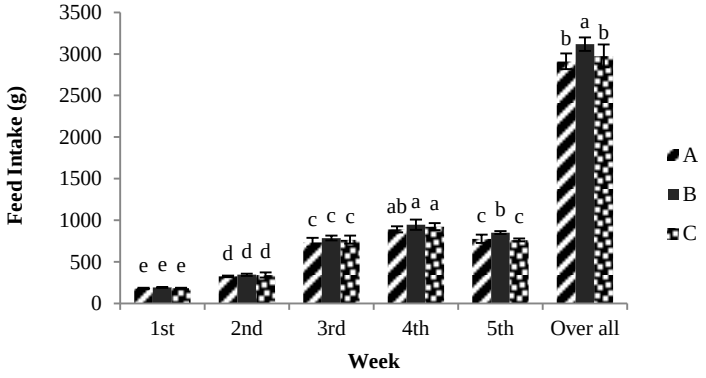

Figure 3 Comparison of means of weekly feed intake of broiler
A= Normal feed (controlled)
B= Feed supplement with 0.2% whey protein (powder)
C= Feed supplement with 2% whey protein concentrate (creamy)

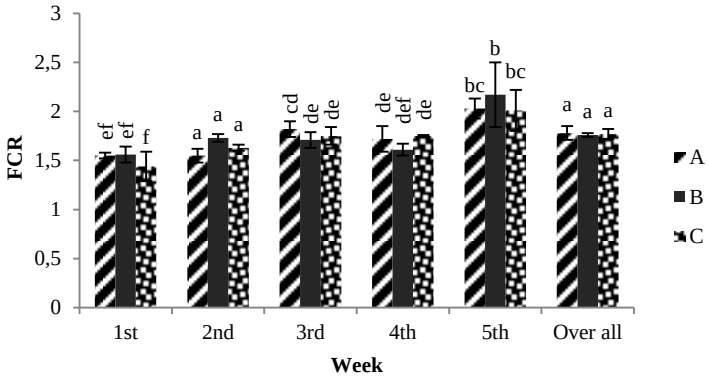

Figure 4 Comparison of means of weekly FCR of broiler
A= Normal feed (controlled)
B= Feed supplement with 0.2% whey protein (powder)
C= Feed supplement with 2% whey protein concentrate (creamy)

Table 2 Comparison of means ±SD of carcass traits

Parameters	A	B	C
Carcass weight	1018±0.04^{b}	1235±0.029^{a}	1083±0.06^{b}
GIT weight	89.16±5.20	101.66±2.88	86.6±12.58
Ratio GIT/carcass	0.087±0.006	0.082±0.002	0.08±0.01
Breast weight	320±12.21^{b}	396±12.219^{a}	348±7.49^{b}
Heart weight	7.06±0.23^{b}	9.63±1.2^{a}	7.33±0.73^{b}
Wings weight	43.60±0.79^{b}	51.76±0.83^{a}	45.2±3.04^{b}
Thigh weight	73.53±3.37^{b}	923±4.40^{a}	81.33±5.50^{b}
Drum stick weight	72.78±3.57^{b}	85.25±3.48^{a}	75.43±8.83$^{a\,b}$
Body fat weight	28.267±4.86	37.03±7.07	33.23±6.54
Liver weight	35±5.96	45±4.86	43±3.18

Table 3 Comparison of means ±SD of hematology

Parameters	A	B	C
Hemoglobin (g/dL)	8.9±1.29	7.9±1.49	10.08±1.19
Polymorphnuclear leukocytes (PMN)	7±3.27	5.6±2.02	4.17±2.02
White Blood Cells (thousands/mm^{3})	8133±2931^{b}	14866±3695^{a}	15833±2010^{ab}
Platelets	9833±1892^{b}	15500±1527^{a}	12400±2291^{ab}
Lymphocytes (%)	91.16±2.84	90.33±0.28	93.33±2.25
Pack Cell Volume (PCV)	28.72±4.73	26.08±3.79	28.72±4.83

CONCLUSION

To date, whey protein concentration (WPC) and its derivatives have not only considered as a good source of essential amino acids but they are a good source of protein substitution in many feeing and food products as well. In case of feeing livestock with a nutrient source like mentioned whey products, it is also effecting on different parameters like, protein amounts of carcasses. WPC is sensitive product especially creamy textured WPC. It was stored in freezer but due to unavoidable load shedding it undergoes temperature fluctuation that might cause undesirable changes in it and highest number of white blood cells in treatment B so it is recommended that try to use fresh creamy texture WPC to avoid any harmful effect on hematology. WPC (powder) was preferred to dry in consistent temperature to avoid any undesirable changes.

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