Laboratory scale study on the effect of feeding quinoa (*Chenopodium quinoa*) as meal on serum biochemistry of broiler

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**Abstract**

Quinoa is a pseudo-cereal with the variety of vitamins, minerals contents, important proteins, essential unsaturated fatty acids and carbohydrates with less glycemic index. The current study was designed to evaluate the effects of quinoa seed meal on serum biochemistry of broiler birds. Seventy (one day old) chicks were raised and divided into groups (n=6) with ten birds (n=10) in each group. The total duration of the experiment was 42 days and all the birds were fed with commercially available feed from market (Starter) from day 1 to day 14. From day 15 to day 42 all the birds in group 1 were fed with the diet containing 50g/kg quinoa seed meal. Group 2 birds were fed with the diet containing 100g/kg quinoa seed meal. Group 3 birds were fed with 200g/kg quinoa seeds meal as a protein source. From day 21 to day 42 all birds from group 4 were fed with 50g/kg quinoa seed meal. Group 5 chicks were fed with the diet containing 100g/kg quinoa seeds meal and group 6 chicks were fed with diet contain quinoa seed meal (200g/kg) as a protein source. Control birds were fed with 100% commercially available feed. In results, significant effects of quinoa meal in the trial. 2 and trial 3 improved kidney functions (ALT & AST) and liver functions (Urea & Creatinine). The quinoa phytochemicals indicate major role as protein meal in the poultry feed through a multiplicity of effects.

**Keywords**: Kidney function; Liver function; Poultry; Quinoa

**Introduction**

Nutritive balanced feeding is most important for economic poultry production. Numerous feed additives used to increase poultry production including various drugs, but antibiotic-resistant and excessive use of drugs cause a variety of health risk to consumers [1]. Use of antibiotics in poultry feed as a growth promoter is reduced globally [2]. Scientist prefers natural plant-based...
meals because of its elevated crude protein content to be used as a potential animal feedstuff the nutritive content of plant seeds makes them an essential part of animal feed [3]. Phytobiotics originate from plants are proved to be excellent growth promoters [4]. Chemical composition revealed that quinoa seeds are a useful supply of protein, raw dietary fiber and calcium supplements [5]. Being gluten-free quinoa has health beneficial characteristics [6]. Quinoa has great nutritional value due to vitamins (B, C, and E) and lipids. In developing countries, quinoa has become a fashionable food, rather than other cereals [7]. Quinoa (Chenopodium quinoa) provide great alternatives to soybean as financial durability associated due to top quality proteins, macro as well as micronutrients because tend to be great causes of bioactive non-nutrient substances [8].

Quinoa with high nutritive contents might be suitable as livestock feed [9]. Quinoa seeds meal protect animals from oxidative stress by raising their antioxidant capacity and dropping lipid peroxidation in plasma and different tissues [10]. Quinoa attained high biological value due to the presence of lysine and sulphur containing amino acid and protein-rich quinoa contain excellent amino acid contents [11]. High levels of energy, calcium, iron, phosphorous, Vitamin E and Vitamin B are present in quinoa than corn, barley, wheat and oats [12]. Additionally, zinc and calcium are present in quinoa in high amount [13]. Quinoa body fat higher in α-linoleic acid and an important omega 3 fatty acid, as compared to oat as well as soya natural oils. Due to its high inherited variability quinoa is not native to the Andes but also cultivated in Europe, North America, North Africa, Pakistan and China [14].

The current study was designed to observe the effects of quinoa seed as a partial protein source and its impact on broiler health by studying its effects on liver functioning, kidney functions as well as histological analysis of different organs (heart, lungs, kidney, and spleen) after feeding the birds with varying amounts of quinoa seeds meal.

**Materials and methods**

**Plant material**

Quinoa seeds meal were obtained from University of Agriculture, Faisalabad. Freshly harvested seeds were washed with distilled water and dried in the air for three to four days under the shed until constant weight in order to use as a protein source as partial protein replacement in the commercially available feed. Total 3 different meals prepared using quinoa seed meal as a protein source (Table 1). For trial 1 and trial 4 (50g/kg quinoa seed meal partially replaced as a protein source). For trial 2 and trial 5 (100g/kg quinoa seed meal used as the protein source). For trial 3 and trial 6 (200g/kg quinoa seed meal used as the protein source) (Table 2).

**Table 1. Diet composition for different groups for studying quinoa seeds effect**

<table>
<thead>
<tr>
<th>Trials</th>
<th>Treatment Day</th>
<th>Quinoa seed as the protein meal replacement (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1</td>
<td>14-42</td>
<td>50g/kg</td>
</tr>
<tr>
<td>Group 2</td>
<td>14-42</td>
<td>100g/kg</td>
</tr>
<tr>
<td>Group 3</td>
<td>14-42</td>
<td>200g/kg</td>
</tr>
<tr>
<td>Group 4</td>
<td>21-42</td>
<td>50g/kg</td>
</tr>
<tr>
<td>Group 5</td>
<td>21-42</td>
<td>100g/kg</td>
</tr>
<tr>
<td>Group 6</td>
<td>21-42</td>
<td>200g/kg</td>
</tr>
<tr>
<td>Control group</td>
<td>1-42</td>
<td>0%</td>
</tr>
</tbody>
</table>
Table 2. Quinoa seed meal feeding trials in different groups of the broiler

<table>
<thead>
<tr>
<th>Trials</th>
<th>Composition</th>
</tr>
</thead>
<tbody>
<tr>
<td>T.1</td>
<td>50g/kg quinoa seed meal partially replaced as a protein source</td>
</tr>
<tr>
<td>T.2</td>
<td>100g/kg quinoa seed meal used as the protein source</td>
</tr>
<tr>
<td>T.3</td>
<td>200g/kg quinoa seed meal used as the protein source</td>
</tr>
<tr>
<td>T.4</td>
<td>50g/kg quinoa seed meal partially replaced as a protein source</td>
</tr>
<tr>
<td>T.5</td>
<td>100g/kg quinoa seed meal used as the protein source</td>
</tr>
<tr>
<td>T.6</td>
<td>200g/kg quinoa seed meal used as the protein source</td>
</tr>
<tr>
<td>Controls</td>
<td>100% commercially available feed</td>
</tr>
</tbody>
</table>

Rearing of birds
To study the dietary effects of quinoa seed meal on broiler 70 healthy chicks (one day old) were purchased from the hatchery. Chicks were kept in cages under controlled temperature. Sugar water (5%) given to chicks on the weekly bases as an energy boost for the first two weeks. Rodents and pets were also kept away from chicks. The experiment went on for 42 days for all six trials as well as for control. Ten chicks were used for each experimental dietary group. One group was fed with commercially available poultry feed as control while other groups were fed with diet supplemented with quinoa seeds. Feed, water, and uniform light provided round the clock during the study period. To study the effects on liver function and kidney functioning analysis all the birds were slaughtered at day forty-two (42) for the collection of blood.

Blood collection
From jugular vein of birds, almost 3ml of blood were taken from each bird. The blood samples of each bird were collected into two well-labeled sets of without anticoagulant vacutainers separately.

Biochemical assay
The blood samples without anticoagulant were in a centrifuged at 3,000 rpm for 15 minutes and separated serum stored at -10°C for further analysis. The serum was used for the estimation of biochemical constituents. Serum AST, ALT, Urea, Creatinine investigated with automatic analyzer Microlab 300 (Merck®, Germany) available at Department of Poultry Science, the University of Agriculture Faisalabad.

Statistical analysis
The data was analyzed using GraphPadPrism 7 software (USA). Results were expressed as mean ± SD. One-way analysis of variance (ANOVA) followed by Dunnett post-test was used for analysis. The difference at p <0.05 considered statistically significant.

Results
Effects of quinoa diet on broiler health the effects on Liver function and Kidney function studied and measured by estimating the levels of ALT and AST for liver function analysis. For kidney function analysis Urea and Creatinine test were performed. Laboratory investigations of ALT (U/L) of chicks with different Quinoa seeds trials showed following trends as Trial 4 (25.67±2) > Control (21.00±2) > Trial 1 (17.67±3) > Trial 6 (17.00±2) > Trial 5 (14.33±3) > Trial 3 (10.33±0.5) > Trial 2 (8.667±1). Trial 4 showed the highest elevated level of ALT in comparison with other trials. Trial 2, Trial 3 and Trial 5 were significant (P ≤ 0.001) as compared to control samples. Trial 1, Trial 4 and trial 6 were nonsignificant (P > 0.05) as compared with the control sample. Trial 2, Trial 3 and Trial 4 were significant (P ≤ 0.001) in comparison with Trial 1 (Fig.1-a). Laboratory investigations of AST (U/L) level of chicks with different Quinoa seeds diets showed the following trends as Trial 4 (228.3 ± 37) > Trial 2 (197.0 ± 56) > Trial 1 (184.0 ± 47) > Trial 5 (176.0 ± 83) > Trial 6 (166.7 ± 15) > Trial 3 (154.7 ± 28) > Control (141 ±
10.82). Trial 4 (In which the birds supplemented with 50g/kg quinoa seeds from day 21 to day 42) elevated the AST level in comparison with other trials (Fig.1-b).

Laboratory investigations of serum creatinine (mg/dl) of chicks fed with different Quinoa seeds meal showed different effects in following trends as Trial 4 (1.2± 0.1) > Trial 2 (1.1± 0.1) > Trial 1 (1.0± 0.1) > Trial 6 (1.0± 0.1) > Trial 5 (0.9± 0.1) > control (0.9± 0.1) > Trial.3 (0.9± 0.2). Trial 4 showed the highest effect on broiler serum creatinine level as compared to other trials (Fig.1-c). Trial 4 exhibited most significant effect on serum creatinine level in comparison with other trials that showed non-significant (P > 0.05) in comparison with control sample (Fig.1-c).

Laboratory investigations of Urea (mg/dl) of broiler with different Quinoa seeds meal trials showed following trends Trial 4 (45.00 ± 5) > Control (42.33 ± 1) > Trial 1 (41.00 ± 3) > Trial 5 (31.33 ± 6) > Trial 6 (31.33 ± 1.5) > Trial 2 (30.33 ± 3.512) > Trial 3 (26.67 ± 2). Trial 4 showed the significant (P > 0.05) level of serum urea in comparison with other trials (Fig. 1-d).

Figure 1. Showing the effects of quinoa seed meals on serum (a) Alanine Amino transferase (b) Asparate Aminotransferase (c) Creatinine (d) Urea level of broiler. Values were mean ± SD of ten replicates. *Significant, **High significant, ***highly significant, ns Not significant.
Discussion
The pseudocereal quinoa (Chenopodium quinoa) contains a high quantity health beneficial nutrients with protein contents (12-23%), vitamin (B, C, and E), minerals such as (Iron, Zinc, Phosphorus and Potassium), fat content (7-9%) and dietary fiber [10]. The current study was designed to investigate the effects of quinoa seeds as partial protein source on broiler health by studying its effects on liver and kidney functions measuring the level of AST, ALT, Creatine, and Urea after slaughtering the birds at day 42 to study the quinoa seeds effect as an alternative protein source of soybean meal for broiler feed. Studies reported the use of quinoa seeds as a potential nutrient for poultry feed. Quinoa seeds meal supplementation proved beneficial in trials using quantity less than 150g/kg [15].
ALT is a crucial enzyme in the biotransformation and detoxing numerous toxicants, reactive oxygen species as well as endobiotics and xenobiotics [16]. In the present study ALT level was not significantly elevated (P > 0.05) in trial 1, trial 4 and trial 5 as compared to control. ALT level declined significant (P > 0.05) in Trial 2, Trial 3 and trial 5. Our results showed non-significant (P > 0.05) level of ALT in trial 6 in which birds were fed with 200g/kg quinoa seeds as protein source from day 21 to day 42 in comparison with control. According to results of ALT, our findings suggest that quinoa (Chenopodium quinoa) seeds as a partial replacement of protein supplement (100g/kg) is beneficial and save in use for broiler feed without deleterious effects. The use of Quinoa seeds more than 100g/kg in broiler feed affect blood biochemical parameters of broiler. In the present study non-significant high level of ALT was found among broiler showed an indication of muscular or liver damage. In our study, no abnormal elevation in the level of ALT enzyme was observed indicating that Quinoa as partial replacement of protein source can be implemented in broiler diet agreeing with Cruz et al. who reported the addition of quinoa seeds in broiler feed without any anti nutritional results [17].
AST used in liver function activity used to determine hepatocellular toxicity [18] but in the current study, AST are not significantly (P > 0.05) affected by dietary treatments showed significant differences among the different treatment.
In the current study, no hyperuricemia happens in birds due to quinoa and renal illness. The normal concentration of urea predicts the quality of protein in the diet. Any kind of modifications within proteins catabolism tends to be primarily associated with serum urea. In the present study significantly declined values of urea were found in trial 2 trial, trial 3, trial 5 and trial 6 chicks fed with quinoa seeds as the protein source compared to control. In our results, we did not observe any increase in reference value area in case of all treatments.
In the present study concentration of creatinine was not significant (P > 0.05) among all broiler birds fed with 50g/kg quinoa seeds as partial protein source in trial 1. Serum creatinine level is a sign associated with proteins metabolic process, a byproduct of phosphocreatine break down within muscle cells [19] in current study, creatinine level showed phosphocreatine break down within muscle cells.
In our results trial 1 and trial 4 showed highly significant values of all biochemical parameters in which the birds were fed with 50g/kg quinoa seeds as a protein source. According to our study on all biochemical parameters, our findings suggest that quinoa (Chenopodium quinoa) seeds as a protein supplement in quantity less than 100g/kg are beneficial and save in use for broiler feed with no deleterious effects on liver functions ALT and AST. Similarly, kidney functions Urea and Creatinine were also normal.
Conclusion
In the current study, we observed no harmful effects of quinoa supplemented meals on liver functions as AST and ALT level was in desirable range and kidney functions analysis of serum Urea and Creatinine were also in the normal range in which the birds were fed with different quinoa seeds meal as partial replacement of protein source. Trial 6 showed the absence of certain features and hemorrhagic areas in heart of trial 6 chick might be due to the high percentage of protein fed diet. Although no abnormalities were observed suggesting that quinoa (Chenopodium quinoa) seeds as a protein supplement in quantity less than 200g/kg are beneficial.

Ethical approval
All experimental trial approved by the ethical board of Government College University Women University Faisalabad, Pakistan and performed in accordance with relevant guidelines and regulations.

Authors’ contributions
Conceived and designed the experiments: A Riaz, MS Masoud & S Mustafa, Performed the experiments: S Mustafa & Z Tariq, Analyzed the data: MA Riaz & A Riaz, Contributed materials/analysis/tools: A Riaz & M Ilyas, Wrote the paper: S Nazar, AU Haq, Z Sabir, A Riaz, MA Riaz & M Ilyas.

References