

Research article

Serobiochemical changes induced by various concentrations of ethanol through drinking water in broiler chicks

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Abstract

There is a common practice of using ethanol in drinking water to subside the respiratory problems, increase weight gain and improve feed conversion ratio in broiler birds, in Pakistan. A study was planned to investigate the ethanol effects on broilers chicks through drinking water. To ascertain the effects of ethanol, 150 day old broiler chicks were randomly divided into 5 equal groups from 1 to 5 at the age of one week. Ethanol (2, 4, 8, 16 and 0% v/v) was offered in drinking water from 8 to 42 days of age daily. Blood samples were collected from experimental birds and studied for parameters which showed significant increase in serum concentration of alkaline phosphatase, urea and creatinine in treated groups as compare to control group. Gross and histopathological examination of organs showed no significant changes except intestine that showed gross and microscopic hemorrhages at higher ethanol dose. The study suggests that ethanol intake in poultry affects liver and kidney, therefore serum concentration of urea, creatinine and alkaline phosphatase are changed in treated broiler.

Keywords: Alkaline phosphatase, Broiler, Creatinine, Ethanol, Urea.

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Introduction

Poultry is an important sector providing employment to millions of people. It has a great share in GDP of Pakistan. Poultry is producing 6.07×10^8 kg meat and 13,813 million eggs [1]. This sector has made progress but still it is facing some serious problems like disease outbreak. Farmers are dealing with such problems by doing some tricks. One such practice is using ethanol for treating respiratory problems and getting better FCR [2-4].

Ethanol is primarily absorbed through oral route, starting from oral mucosa but small intestine is the place where the maximum absorption is carried out. Absorbed alcohol is then transported to liver through portal vein before distributing it to whole circulatory system [5]. Body starts to dispose it of immediately after the ethanol is absorbed. More than 90% of absorbed ethanol is catabolized in liver and remaining is metabolized by other tissues including kidney [6] and excreted in urine and expired air [7]. Liver metabolizes absorbed ethanol by three main enzymatic pathways: 1) alcohol dehydrogenase (ADH): –main system– that couples the ethanol oxidation into acetaldehyde reducing nicotinamide adenine dinucleotide (NAD⁺), 2) the microsomal ethanol-oxidizing system (MEOS): connecting

ethanol and oxidation of NAD⁺ phosphate to oxygen reduction into hydrogen peroxide (H₂O₂), in the presence of cytochrome P-450, 3) catalase (CAT) pathway: where, ethanol is oxidized into acetaldehyde with simultaneous breakdown of H₂O₂ in a CAT catalyzed reaction [8, 9]. The metabolites of these pathways are mainly acetaldehyde and free radicals [10] along with acetate, hydrogen ions, carbon dioxide and water [11]. Renal function is reliably monitored by serum creatinine and urea level those are sensitive biochemical indicators [12]. Impairment of kidney function is suspected if serum urea and creatinine levels are high [5]. Elevated level of this alkaline phosphatase is often linked to liver damage, toxicity, malnutrition and captivity stress [13-15].

Effect of ethanol depends upon dose rate, route, dosage, genetics and physical status of individual [16]. Ethanol effects hepatic tissue, heart muscle, thyroid, brain, respiratory system, hematology, serum electrolyte, pancreas, lymphoid organs and gastrointestinal tract.

These known effects and use of ethanol in poultry were main factors provoking authors to plan a study to determine the effects of ethanol on blood parameters of poultry.

Materials and Methods

In this experiment one hundred and fifty day old broiler chicks were obtained from a hatchery and reared them under optimum and identical conditions of brooding and management. Recommended vaccination schedule was adopted. Feed and drinking water was provided *ad libitum*. Recommended vaccination schedule was followed. After one week these chicks were randomly divided into 5 equal groups (1 to 5). Ethanol (2, 4, 8, 16, 0% v/v) in drinking water offered from day 8th up to 42. To collect the blood and organ samples, 50 birds, 10 from each group were slaughtered humanely at the age of 14, 28 and 42 days.

Parameters selected for study include blood urea determination, creatinine and alkaline phosphatase in serum. Blood urea nitrogen was determined via spectrometer by Berthelot's reaction using a commercially available kit (Gokhan laboratuvar SAN. TIC A.S. Izmir, CAT.NO.G-500). Blood creatinine was determined via spectrometer using a commercially available kit (Gokhan laboratuvar SAN. TIC A.S. Izmir, CAT.NO.G-321). Serum alkaline phosphatase was determined via spectrometer using a commercially available AP kit (Gokhan laboratuvar SAN. TIC A.S. Izmir, CAT.NO.G-041).

Statistical analysis

Data analysis was performed using SAS statistical software (SAS, 1996). Variance techniques were used for analyzing data and means were compared using LSD test.

Results

Serum alkaline phosphatase

Alkaline phosphatase concentrations (μL) obtained at each sampling in treatment and control groups are given in Table 1. At the end of first week post treatment, group 2 showed significant ($p < 0.05$) increase, while decrease was significant in group 3 serum Alkaline phosphatase concentration as compare to control group. Significant increase ($p < 0.05$) was also observed at the end of 3rd week post treatment in group 1 and 2. At 5th week there was no

significant difference observed in all treatment groups.

Table 1: Serum Alkaline phosphatase concentration of chicks given different dilutions of ethanol in drinking water

Group	Age in weeks		
	1 st	3 rd	5 th
1	1.289±0.07	1.354±0.148*	0.453±0.292
2	1.359±0.003*	1.398±0.090*	0.330±0.113
3	0.612±0.04*	0.620±0.392	0.553±0.594
4	0.412±0.034	0.392±0.156	0.513±0.361
5	1.092±0.097	0.329±0.017	0.610±0.429

Notes *significantly different $p \leq 0.05$ as compare to control. Group 1: receiving 2% ethanol water; Group 2: receiving 4% ethanol water; Group 3: receiving 8% ethanol water, Group 4: receiving 16% ethanol water; Group 5: receiving pure water (control group)

Serum urea concentration

Serum urea concentrations (mg/dl) observed at in five experimental groups are given in Table 2. At the end of first week post treatment, there was significant rise ($p < 0.05$) in serum urea concentration in group 1 and 3 as compare to control group, while the findings remained non-significant in all understudy groups at the end of 3rd and 5th week.

Table 2: Serum urea concentration of chicks given different dilutions of ethanol in drinking water

Group	Age in weeks		
	1 st	3 rd	5 th
1	5.701±0.23*	3.495±0.590	5.59±3.160
2	4.618±0.327	4.292±0.573	5.250±0.221
3	6.642±0.208*	7.495±1.909	4.615±0.149
4	5.937±0.213	7.327±3.770	4.671±1.134
5	4.010±0.033	5.476±2.090	6.219±2.172

*significantly different $p \leq 0.05$ as compare to normal. Group 1: receiving 2% ethanol water; Group 2: receiving 4% ethanol water; Group 3: receiving 8% ethanol water, Group 4: receiving 16% ethanol water; Group 5: receiving pure water (control group)

Serum creatinine concentration

Serum urea concentrations (mg/dl) of five experimental groups throughout the treatment period are given in Table 3. At the end of first week post treatment, significant increase ($p < 0.05$) in serum creatinine level was observed in all treatment groups as compare to control group. This significant increase ($p < 0.05$) was also observed at the end of 3rd week post treatment in group 1, 2 and 3. At the end of study period there was no significant difference observed among all experimental groups.

Table 3: Serum creatinine concentration of chicks given different dilutions of ethanol in drinking water

Group	Age in weeks		
	1 st	3 rd	5 th
1	32.375±0.403*	11.477±2.436*	28.4±8.494
2	37.09±0.749*	15.485±1.322*	13.995±3.91
3	26.99±0.89*	15.040±2.022*	19.715±20.23
4	27.38±0.73*	26.400±0.862	18.585±12.02
5	17.320±0.607	26.89±0.622	12.15±4.762

*significantly different $p \leq 0.05$ as compare to normal. Group 1: receiving 2% ethanol water; Group 2: receiving 4% ethanol water; Group 3: receiving 8% ethanol water, Group 4: receiving 16% ethanol water; Group 5: receiving pure water (control group)

Histopathology

There were no gross and microscopic lesions evident in observed organs except intestine. Intestinal mucosal surface showed hemorrhages either mild or slightly severe in all treatment groups.

Discussion

There were significant ($p < 0.05$) changes in concentration of alkaline phosphatase, group 1 showed increased level at end of 3rd week and group 2 at 1st and 3rd week. The results were in line with the findings of Dino and Nechifor [17] who conducted the trial on ethanol treated rats to determine the activity of liver enzymes in serum. This changed level of alkaline phosphatase in serum indicates that there is liver damage to some extent, although there were no histopathological lesions seen with light microscope.

At the end of 1st week post-treatment, significant increase ($p < 0.05$) in serum creatinine was observed in all treated groups as compare to that of control group, at end of 3rd week group 1, 2 and 3 revealed significant results, while at 5th week of treatment period, no group showed significant results, the birds seem to adopt the effect of continuous ethanol ingestion. Adaramoye and Aluko [5] also found increased serum creatinine level in ethanol intoxicated wistar rats and related it with kidney damage caused by chronic ethanol toxicity. Sayali et al. [18] reported significant increase in serum creatinine level in heavy alcoholic males, but in present study highest alcohol concentration treated group (16%) showed significant increase only at the

end of 1st week the reason may the specie difference and study design.

Serum urea level was significantly higher ($p < 0.05$) in group 1 and 3 at the end of 1st week of treatment, whilst all other groups showed non-significant results. Raised level of urea and creatinine was documented in rats given intravenous injections of ethanol [19]. Similar findings were reported in female wistar rats when the mammals were given oral ethanol dose [20]. Another result supporting study have been conducted on rabbits [21]. Alimi et al. [22] reported kidney damage (severe congestion, renal necrosis, glomeruli damage and unadorned degenerative alteration in renal tubules) in ethanol treated wistar rats they observed high level of serum creatinine and urea. Dehydration, heavy protein intake and kidney dysfunction causes rise in blood urea and creatinine content in avian specie [23, 24].

It can be hypothesized that intake of ethanol restricted birds to drink ample amount of water because after intake of ethanol birds were sleepy; this low intake induced dehydration and thickening of blood -increased level of hemoglobin, erythrocyte count and pack cell volume (PCV) [2, 25]- ultimately showing high levels of creatinine and urea in blood. The high level of serum creatinine and urea -as in present case- shows impairment of kidney function [5].

Conversely, Yuan et al. [26] reported protective effect of ethanol on kidney in mice where they observed lower plasma level of urea and creatinine following the intravenous injection of ethanol. This change was, probably due to difference in specie and treatment methodology of present experiment.

Conclusions

The findings of present studies revealed that ethanol use in poultry does not enhance production potential rather it alters serum urea, creatinine and alkaline phosphatase concentrations indicating liver and kidney damage. So the use of ethanol with these concentration may be discouraged to combat various ailments of respiratory in nature.

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