

## CONTROL OF HYDATIDOSIS IN RABBITS THROUGH FEEDING LOCAL PLANTS

AKHTAR TANVEER, RIFFAT MUSTAFA AND ZAHEER ANWAR

Department of Zoology, University of the Punjab, Quaid-e-Azam Campus,  
Lahore-54590, Pakistan

**Abstract:** Out of twenty-six healthy rabbits acclimatized to the optimal laboratory conditions for fifteen days, 4 were considered as normal control and twenty-two were inoculated (intraperitoneally and subcutaneously) with 45000 protoscoleces of *Echinococcus granulosus* of sheep origin. They were at random divided into 4 groups. Groups 1-3 were orally given (2g/day) mashed leaves of *Aloe vera*, powdered *Punica granatum* (unripped ovary part) and fruit of *Azadirachta indica* respectively. Group 4 was considered as infected control. All the rabbits were fed on seasonal green fodder and ordinary tap water *ad libitum*. Their blood samples were pooled after every 10 days upto 70 days for biochemical analyses. Increased ( $P < 0.001$ ) body weight was noted in experimental rabbits. It was clear from the biochemical results that activities of acid phosphatase, alkaline phosphatase and glucose decreased ( $P < 0.001$ ) in rabbits treated with *A. vera*, *P. granatum* and *A. indica* as compared to normal control and the values of glutamic oxaloacetic transaminase, glutamic pyruvic transaminase, cholesterol and total protein improved ( $P < 0.001$ ) as a result of herbal treatment. The bilirubin value improved only in rabbits treated with *A. vera* and *A. indica* ( $P < 0.001$ ). The results of our present investigation showed that alterations in different enzymes and biochemical metabolites induced by liver protoscoleces were improved in the rabbits orally given *A. vera*, *P. granatum* and *A. indica*. Although the extent of improvement varies in these plants, even then the present results are promising and it is suggested that the active ingredients of these plants should be checked for further use.

**Key words:** Hydatidosis, herbal control, blood biochemistry, rabbits.

### INTRODUCTION

**H** ydatidosis is medically and economically one of the most important disease and both unilocular (*Echinococcus granulosus*) and multilocular (*E. multilocularis*) of the disease are widely and actively extending their range into areas previously considered to be free of this infection (Schwabe, 1986). Sometimes it look like an out break in different localities like England and Wales (Stallbaumer *et al.*, 1986), in Morocco (Pandey *et al.*, 1988) and in China (Chi *et al.*, 1990). It is also an important zoonosis and the chances of its high infection in man are more in those areas where sheep and cattle grazing is carried out with the help of dogs (FAO report, 1993) as in Pakistan. Its wide prevalence in the live stock of Pakistan has been given consideration by Khan and Haseeb (1984), Iqbal *et al.* (1986), Pal and Jamil (1986) and Iqbal *et al.* (1989). Its seriousness in human beings is also far greater than what the published record shows and its incidence is much higher then what is generally believed to be (Junejo *et al.*, 1995). Its pathology resulted due to various haematological and biochemical alterations have been given consideration (Pandey, 1971; Schantz,

1984) with the ultimate finding that such changes were attributed to the hydatid cyst fluid that seeps out of the hydatid cyst wall thereby causing necrosis of the surrounding cells with the ultimate result in the increase of enzymatic level in the blood.

Cyst can be removed only by surgery (Schantz, 1984) that is expensive, debilitating and entail risk. Therefore, programs have been implemented to control the parasite in regions where it is endemic. Most control programmes are aimed at eliminating the source of infection to man and other intermediate hosts. Besides this two benzimidazole derivatives, namely mebendazole and albendazole are now routinely used in cystic echinococcus (Davis *et al.*, 1986; Horton, 1989; Matossian *et al.*, 1992). Many plant drugs have also been in practice since ancient times for the treatment of parasitic infection in man and animals (Nadkarni, 1954; Chopra *et al.*, 1956; Said, 1969).

Since Pakistan has a great variety of herbs and medicinal plants grown widely in the northern hilly areas. Unfortunately a large number of the indigenous medicinal plants and herbs have not been investigated by the modern phytochemical and pharmacological methods except Said (1969), Ikram and Hussain (1978), Ohigashi *et al.* (1982), Sanyal *et al.* (1985), Akhtar (1986).

Akhtar (1986) studied the anthelmintic efficacies of indigenous plant and herbs allegedly used to treat various parasitic infections by the practitioners of old and traditional medicines so as to find a scientific use of these herbs for veterinary therapeutic purposes. He also studied the anthelmintic activities of several powdered plant drugs and their extracts in goats, buffaloes and calves. Akhtar (1987) found out a number of plant drugs and their extracts that possess interesting potent and safe anthelmintic principle for veterinary usage. Anwar *et al.* (1997a) tested two indigenous plants to control hydatidosis in experimentally infected rabbits along with the common fodder and their effects were studied on different haematological and biochemical parameters. Beside this plants extract was used to control the larval stages of *E. granulosus* (Anwar *et al.*, 1997).

In the present work 3 plants (*Aloe vera*, *Punica granatum*, *Azadirachta indica*) were used to control this disease. These plants were orally given to experimentally infected rabbits with common fodder, and their effects were studied in terms of their biochemical parameters which may give a clear picture of the changes they have produced in curing the disease.

## MATERIALS AND METHODS

### *Maintenance of rabbits*

Twenty-six healthy rabbits maintained under optimal conditions were fed twice a day on seasonal green fodder. Few crystals of  $\text{KMnO}_4$  were added to their drinking water to minimize the chances of infection.  $\text{KMnO}_4$  solution was used to disinfect the utensils and cages. Rabbits were individually weighed in the beginning of experiment and then before each inoculation.

### *Hydatid cyst fluid*

Hydatid cyst fluid (HCF) was aspirated from the cyst present in liver, lungs and spleen of infected sheeps at local slaughter house and only viable protoscoleces were selected for inoculation. Hydatid cyst fluid was centrifuged at 500 rpm for 5 minutes and the pellet thus formed was washed with saline solution, containing the Penicillin and Streptomycin sulfate as described by Ohnishi (1985). The protoscoleces were counted in the chamber of white blood cells of haemocytometer. Two inoculums each having 22500 protoscoleces were injected sub-cutaneously and intra-peritoneally in the left flank of rabbit after taking all antiseptic precautions.

### *Grouping of rabbits*

Rabbits were divided into 5 groups. For each plant treatment 3 treated (six rabbits each) and one experimental control (having 4 rabbits) along with one normal control (n=4) were maintained. Following plants and their parts were used:

The leaves of *Aloe vera* (Kwargandal), ovary part of *Punica granatum* (Anar) and fruit of *Azadirachta indica* (Neem) were used for feeding animals.

### *Preparation of plant/herbal extracts*

The dried plants and their parts (Anar, Neem) were ground to fine powder. 2 g of each powdered plant was mixed with 25 ml of tap water and then given to rabbits. The fresh leaves of kwargandal were mashed and 2 g mixed with 25 ml of water was given to rabbits. Three treated groups were orally given 2 g/day of *A. vera*, *P. granatum* and *A. indica* up to 70 days. The experimental control (without herbal treatment) and normal control were also maintained.

### *Blood sampling and biochemical analyses*

After every 10 days about 4 ml blood was pooled from the marginal vein of each rabbit and refrigerated at 4°C for 15 minutes and then centrifuged at 3000 rpm for 20 minutes. Unhaemolysed and clear serum was used for estimating acid phosphatase, alkaline phosphatase, glutamic oxaloacetic transaminase (GOT), glutamic pyruvic transaminase (GPT), bilirubin, cholesterol, glucose and total protein according to standard kits (Randox, U.K.), following Dacie and Lewis (1991).

## RESULTS

### *Group I (treated with leaves of Aloe vera)*

The initial body weight (g) before any treatment in normal control, experimental control and treated rabbits was  $1056 \pm 65.23$ ,  $1075 \pm 68.23$  and  $1054.16 \pm 164.63$  respectively. It decreased (-0.35%) after 10 days in normal control but increased (0.76% and 0.39%) in experimental control and rabbits orally given leaves of *A. vera* (2 g/day)

( $P < 0.001$ , Table 1, Fig.1). Afterwards, body weight of normal control rabbits fluctuated a lot with an overall increase of 29.37%, till the end. In case of experimental control and treated rabbits body weight increased upto 26.5% and 13.6% after 50 days. After 60 days their body weight decreased upto -5.65% and -5.15%. After 70 days there was again an increase (29.34% and 23.58%) in the body weight of both experimental control and treated rabbits ( $P < 0.001$ ) (Fig.1).

After 10 days the specific growth rate (%) increased by 0.02% and 0.07% in both normal control and experimental controls. It was noted that specific growth rate (%) of treated rabbits faced a slight decrease (0.04%) after 10 days, (0.11%) 20 days, followed by an increase of 0.78%, 0.40%, 0.6% after 30, 40 and 50 days, respectively. The values again decreased (-4.9%) after 60 days with an increasing trend (1.73%) after 70 days.

### Blood biochemistry

The biochemical effect of *A. vera* on the blood of rabbits have been studied in terms of changes it produced in the activity of some enzymes such as acid phosphatase (AcP), alkaline phosphatase (Ap), glutamic oxaloacetic transaminase (GOT), glutamic pyruvic transaminase (GPT). The changes produced in the concentration of certain biochemical metabolites like bilirubin, cholesterol, glucose and total protein were also studied.

The initial value of AcP (IU/dl) for normal control, experimental control and treated rabbits was  $80.25 \pm 1.29$ ,  $81.14 \pm 1.83$  and  $96.96 \pm 2.11$ , respectively. Later on, there were great fluctuations in these values and after 70 days the AcP value increased by 9.65% for normal control along with a prominent decrease in experimental control and treated rabbits (-245.80% and -90.45%) ( $P < 0.001$ , Fig.2). The value of Ap (IU/dl) gradually decreased from  $13.29 \pm 7.69$  to  $10.02 \pm 5.06$  for normal control rabbits. However, the overall values for Ap in the experimental and treated rabbits showed sharp decline (-689.7% and -200.7%) ( $P < 0.001$ , Fig.2). The GOT value (IU/dl) of normal control rabbits showed an overall increase (16.55%) while the similar values for the experimental control declined upto -58.5%. In case of treated rabbits GOT value improved by 5.29% in the end of treatment ( $P < 0.001$ , Fig.2). There was overall increase (13.15% and 10.48%) in the GPT value (IU/dl) for both normal control and treated rabbits while experimental control faced decline by -8.80%. Changes in GPT values were found statistically significant ( $P < 0.001$ ) (Fig.2).

In the beginning of experiment the average bilirubin value (mg/dl) for normal control rabbits was  $0.86 \pm 0.67$ . This group, however, faced decline (-26.31%) till the termination of experiment. In case of experimental control rabbits great fluctuations were noted in the bilirubin value with an overall decline of -15.20%. Moreover, the bilirubin values for the treated rabbits showed an overall increase (34.7%) ( $P < 0.001$ , Fig.2). It was noted that cholesterol level (mg/dl) increased continuously from  $70.34 \pm 10.23$  to  $80.49 \pm 13.30$  for normal control rabbits. Although experimental control rabbits showed great fluctuations but their cholesterol contents increased by 80.3% till the end of experiment. The treated rabbits also faced increased (59.45%) cholesterol values ( $P < 0.001$ , Fig.2). Glucose level (mg/dl) showed variations with an overall

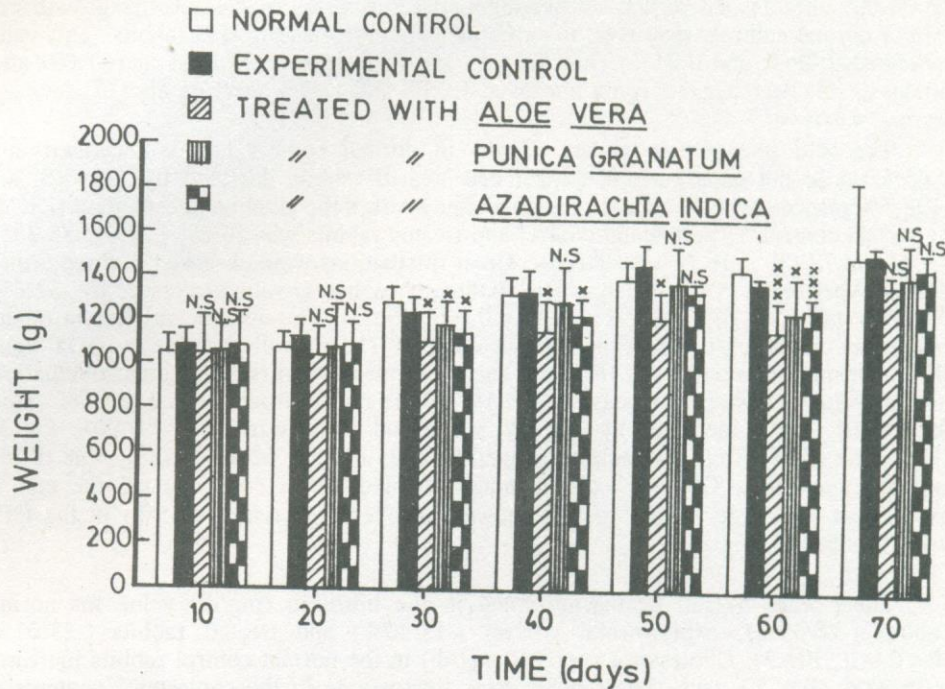


Fig. 1: Changes in the body weight of rabbits, normal control, experimental control and treated with *Aloe vera* leaves, *Punica granatum* fruit and ovary part and *Azadirachta indica* fruit (2 g/day). Values given are mean  $\pm$  S.D. of 4 control, 3 experimental control and six treated rabbits for 70 days. The statistical significance has been determined by Student's 't' test and probability represented by stars: \*  $P < 0.05$ ; \*\*  $P < 0.01$ ; \*\*\*  $P < 0.001$ .

decrease in all the rabbits. Glucose level for the normal control and experimental control groups decreased by -103.42% and -195.7% while for the treated rabbits the decrease noted was -34.73% till the end of experiment ( $P < 0.001$ , Fig.2). The total protein value (g/dl) at the start of experiment was  $6.025 \pm 0.434$  and it changed a little till the end for normal control. In case of experimental control variations were noted in the values with an overall increase of 17.20%. The amount of total protein increased by 11.23% for treated rabbits ( $P < 0.001$ , Fig.2).

#### Group II (treated with ovary part and flowers of *Punica granatum*)

The average weight (g) of normal control, experimental control and treated rabbits increased from  $1056 \pm 65.23$  to  $1512.5 \pm 335.8$ ,  $1075.0 \pm 68.23$  to  $1533.33 \pm 14.43$  and  $1056.5 \pm 184.83$  to  $1433.33 \pm 137.53$  respectively. The body weight of treated rabbits increased by 22.9% after 50 days and decreased by -11.66% after 60 days.

Again there were increasing trend (24.99%) in the body weight of treated rabbits ( $P < 0.001$ , Fig.1). There was an overall increase (0.41%) in the specific growth rate (%) of normal control. However, in experimental control and treated rabbits. This value increased (0.96% and 0.81%) after 50 days and then decreased (-0.51 and -1.09) after 60 days. This decrease was again improved 1.05% and 1.37% increase after 70 days.

The acid phosphatase values (IU/dl) in normal control rabbits increased upto 9.65% while the experimental control and treated rabbits declined by -245.8% and -192.5% respectively ( $P < 0.001$ , Fig.3). After 10 days the alkaline phosphatase (I.U/dl) in normal control, experimental control and treated rabbits was  $13.29 \pm 7.69$ ,  $55.33 \pm 43.92$  and  $72.00 \pm 18.50$  respectively. Great fluctuations were observed in three groups during experiment. After 70 days their alkaline phosphatase value decreased by -32.6%, -689.7% and -263.26% respectively ( $P < 0.001$ , Fig.3). The decline was less in treated rabbits as compared to the experimental control. The overall increase in GOT value (IU/l) for normal control was 16.55%. The experimental control have great fluctuations in their values showing a decrease of -58.5% till the end of experiment. The GOT values for treated rabbits increased by 38.63% at the end of experiment ( $P < 0.001$ , Fig.3). GPT value (IU/l) of normal rabbits changed from  $25.25 \pm 2.62$  to  $35.62 \pm 3.68$  till the end of experiment. GPT of experimental control declined (-8.80%) till the end of experiment while the treated rabbits showed an overall increase (9.45%) in the GPT value ( $P < 0.001$ , Fig.3).

There were overall decreasing trend in the bilirubin (mg/dl) value for normal control (-26.31%), experimental control (-15.20%) and treated rabbits (-23.07%) ( $P < 0.001$ , Fig.3). Cholesterol contents (mg/dl) in the normal control rabbits increased by 5.87% after 70 days. There were great fluctuations in the cholesterol contents of experimental control and treated rabbits and their values increased by 80.3% and 52.00% respectively ( $P < 0.001$ ) (Fig.3). The glucose level (mg/dl) at the start of experiment for normal control, experimental control and treated rabbits was  $179.52 \pm 12.86$ ,  $153.83 \pm 13.04$  and  $109.52 \pm 9.53$  respectively. There were great fluctuations in their glucose values during the experiment with a decline of -103.42%, -195.7% and -38.79% at the end of experiment ( $P < 0.001$ , Fig.3). Initial and final total protein (g/dl) values were almost the same ( $6.025 \pm 0.434$  to  $6.28 \pm 1.24$ ,  $8.182 \pm 2.94$  to  $9.88 \pm 1.38$ ) for both normal control and experimental control. In case of treated rabbits the total protein value increased by 2.29% at the end of experiment. The results were found statistically significant ( $P < 0.001$ ) (Fig.3).

### *Group III (treated with fruit of Azadirachta indica)*

The body weight (g) of both experimental control and treated rabbits increased till 50 days and it was followed by decrease (-5.65% and -5.88%) after 60 days. After 70 days their body weight again increased (29.34% and 26.27%) ( $P < 0.001$ ) (Fig.1). There were continuous increase (0.58%, 0.96% and 0.87%) in the specific growth rate (%) of normal control, experimental control and treated rabbits respectively upto 50 days. After 60 days it decreased (-0.51% and -0.49%) followed by an increase of 1.05% and 1.56% at the end of experiment for both experimental control and treated rabbits.

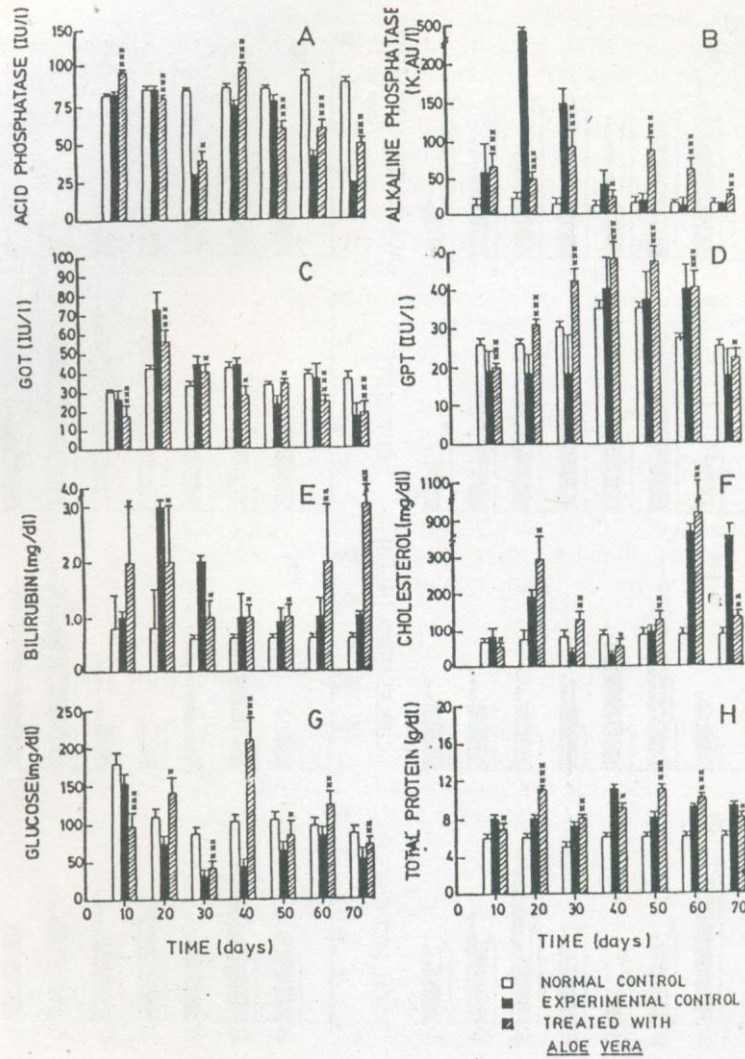


Fig. 2: Biochemical changes in rabbits (intraperitoneally and sub-cutaneously injected with 45000 protoscolec) after oral administration of *Aloe vera* leaves (2 g/day). Values given are mean  $\pm$  S.D. of 4 control, 3 experimental control and six treated rabbits for 70 days. The statistical significance has been determined by Student's 't' test and probability represented by stars: \* P < 0.05; \*\* P < 0.01; \*\*\* P < 0.001.

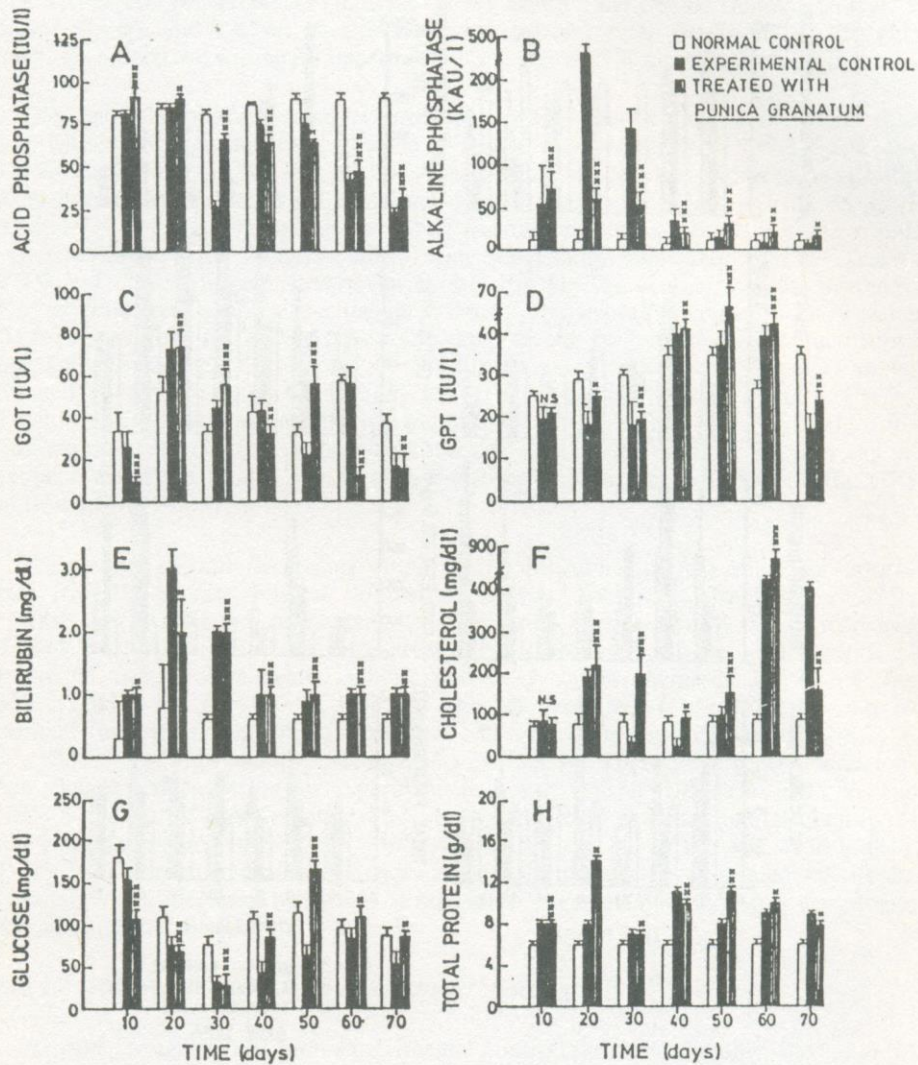


Fig. 3: Biochemical changes in rabbits (intraperitoneally and sub-cutaneously injected with 45000 protozoecles) after oral administration of *Punica granatum* flower and ovary part (2 g/day). Values given are mean  $\pm$  S.D. of 4 normal control, 3 experimental control and 6 treated rabbits for 70 days. The statistical significance has been determined by Student's 't' test and probability represented by stars: \* P < 0.05; \*\* P < 0.01; \*\*\* P < 0.001.



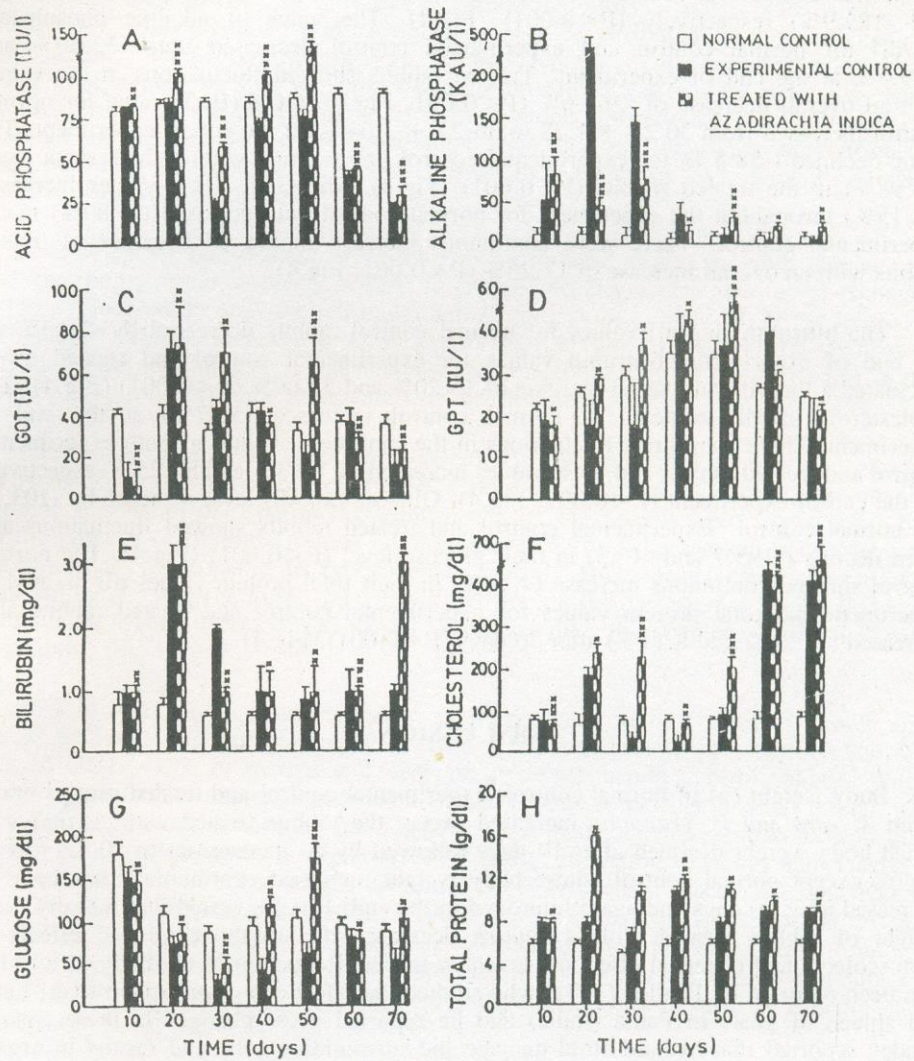


Fig. 4: Biochemical changes in rabbits (intraperitoneally and sub-cutaneously injected with 45000 protoscolices) after oral administration of *Azadirachta indica* fruit (2 g/day). Values given are mean  $\pm$  S.D. of 4 normal control, 3 experimental control and 6 treated rabbits for 70 days. The statistical significance has been determined by Student's 't' test and probability represented by stars: \* P < 0.05; \*\* P < 0.01; \*\*\* P < 0.001.

The acid phosphatase (AcP) (IU/dl) increase (9.65%) throughout the experiment for normal control but decreased in experimental control and treated rabbits (-245.8% and -183.9%) respectively ( $P < 0.001$ ) (Fig.4). The value of alkaline phosphatase (IU/dl) for normal control and experimental control decreased upto -32.60% and -689.7% at the end of experiment. Treated rabbits showed fluctuations in the values with an overall decrease of -261.6% ( $P < 0.001$ , Fig.4). GOT (IU/l) value for normal control increased from  $30.25 \pm 1.27$  to  $36.25 \pm 3.64$  till the end of experiment. The value declined (-58.5%) for experimental control group along with an increasing trend (14.98%) in the treated rabbits ( $P < 0.001$ ) (Fig.4). The GPT (IU/l) values increased (13.15%) throughout the experiment for normal control but decreased (-8.80%) in the experimental control. There were continuous increase in the GPT values of treated rabbits with an overall increase of 17.26% ( $P < 0.001$ , Fig.4).

The bilirubin (mg/dl) values for normal control rabbits decreased by -26.3% till the end of experiment. Bilirubin values for experimental control and treated group fluctuated a lot with an overall increase of 15.20% and 57.69% ( $P < 0.001$ ) (Fig.4). The cholesterol (mg/dl) contents in normal control increased (5.87%) at the end of experiment. There were great fluctuations in the cholesterol values of both experimental control and treated rabbits and these values increased by 80.3% and 88.25% respectively till the end of experiment ( $P < 0.001$ , Fig.4). Glucose (mg/dl) level decrease by -103.42 for normal control. Experimental control and treated rabbits showed fluctuations and faced decline (-195.7 and -46.3) in their glucose level ( $P < 0.001$ ) (Fig.4). The normal control showed continuous increase (4.14%) in their total protein values till the end of experiment and total protein values for experimental control and treated rabbits also increased (17.20% and 8.11%) after 70 days ( $P < 0.001$ ) (Fig.4).

## DISCUSSION

Body weight (g) of normal control, experimental control and treated groups orally given *A. vera* and *P. granatum* increased except the rabbits treated with *A. indica* in which body weight declined after 10 days followed by an increase up to 50 days in all groups except normal control whose body weight increased continuously but later on decreased after 60 days and again improved in the end. It is suggested that initially body weight of rabbits treated with *A. indica* decreased due to the combined effects of protoscoleces and chemical effect of *A. indica* leaves. Reduction in the body weight has also been reported by Pandey (1971) who studied the effect of hydatidosis in liver, lungs and spleen of goats in Patna (India) and he reported gross changes in these tissues. Pandey reported that hydatid fluid damage the surrounding cells and results in organs condemnation which lead to weight reduction. Economic losses due to hydatidosis through low quality and reduced yield of milk, meat and retarded growth have also been reported by Anonymous (1985) and Schwabe (1986). Protein deficiency and reduction of weight took place in animals treated with hydatid fluid have also been studied by Iqbal *et al.* (1989). Similar findings have earlier been reported by Anwar *et al.* (1997a) in rabbits experimentally infected with *E. granulosus* and treated with *Prosopis glandulosa* (leaves) and *Embelia ribes* (fruits).

In the present investigation, the body weight of treated rabbits improved either due to the development of resistance, or due to certain inhibitory effect of plants against the protoscolexes. The increased body weight can also be attributed to some of pregnant female rabbits. The decrease noted in the body weight of experimental control and treated groups may be due to some bacterial infections and finally there was again increasing trend in the body weight of rabbits which showed that plants *A. vera*, *P. granatum* and *A. indica* have developed some resistance and they have antagonized the effects produced by the incoming protoscolexes. Increase in the body weight of treated rabbits was a curing mechanism.

Two types of phosphatase are commonly estimated in serum. Acid phosphatase (ACP) that helps in the autolysis of cells after their death whereas alkaline phosphatase (AP) is a brush border enzyme rich in epithelial lining of bile canaliculi of the liver (Ali and Shakoori, 1993). Increase in ACP activity may be due to pre-necrotic changes in different tissues including liver and increase in AP activity may be due to lesions in the biliary epithelium, or lesions in the intestinal brush border.

The results of our present finding showed decreased acid and alkaline phosphatase activity in experimental control and treated rabbits as compared to normal ones. In the beginning the values of experimental control and treated groups were almost same but with the passage of time the treated rabbits recovered from disease and values became almost normal as compared to experimental control. This normal trend showed that plants *A. vera*, *P. granatum* and *A. indica* were effective against the toxicity of protoscolexes. The decrease in values may be due to affected lysosomal acid phosphatase.

GOT is found in high concentration in liver, heart, kidney, skeletal muscle and pancreas while GPT is much abundant in liver. Results of present investigation showed increased GOT, GPT in treated and decrease in experimental control rabbits. After herbal treatment these values improved in treated rabbits somewhat close to the normal values that may be due to certain inhibitory effects of plants against protoscolexes or due to pathological response of the hepatocytes instead of induction (Sanchez and Sanchez, 1971), or hemolysis of blood or gentamycin's effect or skeletal muscle damage (Khan, 1990). However, Frayha and Haddad (1980) reported that enzymes present in the protoscolexes could alter the permeability of cell membrane of hepatocyte due to which level of transaminase became high in blood serum or it may be due to leaching of enzyme in blood serum after destruction of pathological cells formed by toxicity and enzyme activity of protoscolexes.

Bilirubin level is an index applied for detecting (i) liver disease (hepatic jaundice), (ii) hemolytic anaemia, and (iii) degree of jaundice. The extent of increase in bilirubin level determines the type of jaundice (Khan, 1990). In the present findings the bilirubin level increased in rabbits treated with *A. vera* and *A. indica* as compared to experimental control and values of rabbits treated with *P. granatum* showed decline but were more nearer to normal control as compared to experimental control. Increased bilirubin level can be attributed to some damage in kidney and liver cells which ultimately resulted in enormous RBCs breakdown and disintegration of haemoglobin molecules and obstruction in biliary tract (Khan, 1990). Decreased bilirubin level showed that although

production of haemoglobin in this group increased but the chemicals of *Punica granatum* have inhibited the breakdown of haemoglobin, thereby keeping the bilirubin at normal level. Increased bilirubin by toxic effect of protoscolecis in rabbits have also been reported by Anwar *et al.* (1997a).

Cholesterol is a component of cell membrane and precursor of the steroid hormones. It mainly occurs in the liver and synthesized by virtually all tissues in humans (Guyton and Hall, 1996). In the present study although cholesterol content increased in all the groups, but the values of treated rabbits were more nearer to the normal control as compared to the experimental control which showed that treatment with plants was a curing mechanism against hydatidosis. In present work increased cholesterol level indicated that it was not used in biosynthesis of steroid hormone. The increased cholesterol content can also be attributed either to worm load retained in host system or metabolic disturbance and immunobiochemical reactions of the host, or hormonal metabolism that have disturbed in *de novo* synthesis of cholesterol in liver and intestine (Vardhani and Rao, 1995).

Glucose forms different compounds in the body and it is instant source of energy. In the present investigation glucose level decreased most of the time thereby resulting in hypoglycemia in all normal control, experimental control and treated groups, but the values of glucose in treated groups orally given *A. vera*, *P. granatum* and *A. indica* became close to normal control as compared to experimental control. It may be suggested that glucose level decreased because it might be forcefully catabolized by the aerobic and anaerobic enzymes present in the protoscolecis (Agosin *et al.*, 1957).

Anonymous (1985) and Schwabe (1986) observed the economic losses due to hydatidosis in domestic livestock through reduced quality and yield of milk, meat and retarded growth. Iqbal *et al.* (1989) observed that due to hydatidosis protein deficiency results in infected animals. Decreased protein values may be due to direct proteolytic effect of hydatid cyst fluid as it contained many lytic enzymes (Frayha and Haddad, 1980) or due to some interference in the process of synthesis in liver or due to some malfunctioning of kidney (Khan, 1990). However, increased protein level may be attributed to the formation of antibodies against the antigen present in the protoscolecis inoculated in the rabbits. In the present investigation total proteins increased in normal control, experimental control and treated rabbits orally given *A. vera*, *P. granatum* and *A. indica* respectively which means that treated rabbits develop resistance against protoscolecis. This means that plants were effective in improving the total protein in treated groups as compared to experimental control. This increase in total protein may be due to i, increased seepage of soluble protein including enzymes through the abnormal liver parenchymal cells, which affects the cell membrane permeability; ii, due to increased protein synthesis in the liver; iii, utilization of free amino acid contents for energy needs in the absence of glucose oxidation (Abdel-Salam *et al.*, 1982); or iv, as a result of some catabolic reactions protein synthesis yet arrested and amino acids which are absorbed are not incorporated and hence level of total protein increases. Another strong reason for the increased protein contents is the heavy inflow of proteins through the regularly inoculated protoscolecis.

In the present work, results of all biochemical parameters indicated that plants *Aloe vera*, *Punica granatum* and *Azadirachta indica* are equally effective against hydatidosis and showed the curing mechanism in rabbits infected with protoscoleces. This can provide basis for the treatment of hydatidosis which can secure man and his livestock. Among the plants tested in the present investigation, *A. indica* have also been reported as effective protoscolecidal by Anwar *et al.* (1997). Keeping in view their findings role of *A. indica* was further checked in curing hydatidosis. In the end it can be suggested that these plants were equally effective in curing hydatidosis and further investigations are needed to find out their active ingredient.

#### Acknowledgements

Financial assistance provided by Punjab University Research Committee 1997 is gratefully acknowledged.

#### REFERENCES

- AGOSIN, M., VON BRAND, T., RIVERA, C.F. AND MACMAHON, P., 1957. Studies on the metabolism of *Echinococcus granulosus*. I. General chemical composition and respiratory reactions. *Exp. Parasitol.*, **6**(1): 37-51.
- ABDEL-SALAM, E.B., ADAM, S.E.I. AND TARTOUR, G., 1982. The combined action of dieldrin and phosphamidon in goats. *Z. Veterinaerme*, **29**: 136-141.
- AKHTAR, M.S., 1986. Anthelmintic evolution of indigenous medicinal plants for veterinary usage. 2nd Progress Report of PARC Research Project Univ. of Agri., Faisalabad.
- AKHTAR, M.S., 1987. Anthelmintic evolution of indigenous medicinal plants for veterinary usage. 3rd Progress Report of the PARC Research Project Univ. of Agri., Faisalabad.
- ALI, S.S. AND SHAKOORI, A.R., 1993. Short-term toxicity of Endrin in Sprague Dawley rats. Biochemical and histological changes in liver. *Punjab Univ. J. Zool.*, **8**: 1-13.
- ANONYMOUS, 1985. Echinococcosis/hydatidosis surveillance, prevention and control: FAO/UNEP/WHO guidelines. FAO, U.N., pp.147.
- ANWAR, Z., NOSHABA, N. AND TANVEER, A., 1997. *In vitro* protoscolecidal property of some local plants. *Sci. Int. (Lahore)*, **9**(2): 197-200.
- ANWAR, Z., NOSHABA, N. AND TANVEER, A., 1997a. Control of hydatidosis through local plants. *Sci. Int. (Lahore)*, **9**(2): 201-204.
- CHI, P., ZHANG, Z., HASYET, M., LUIZ, F., TOLLEY, H.D. AND SCHANTZ, P.M., 1990. Cystic echinococcosis in the Xinjiang/Uygur Autonomous region, Peoples Republic of China: I. Demographic and epidemiologic data. *Trop. Med. Parasitol.*, **41**(21): 157-162.
- CHOPRA, R.N., NAYYER, S.L. AND CHOPRA, I.C., 1956. *Glossary of Indian Medicinal Plants*. Council of Scientific and Industrial Research, New Delhi, pp.195.
- DACIE, S.J. AND LEWIS, S.M., 1991. *Practical heamatology*. 7th edition. Churchill Livingstone, Edinburgh London, pp.100-131.

- DAVIS, A., PAWLOWSKI, Z.S. AND DIXON, H., 1986. Multicentre clinical trials of benzimidazole carbamates in human echinococcosis. *Bull. World Hlth. Org.*, **64**: 383-388.
- FAO (Food and Agriculture Organization). 1993. *Animal health year-book*, 1993. FAO-WHO-OIE. Rome: Italy.
- FRAYHA, G.J. AND HADDAD, R., 1980. Comparative chemical composition of protoscolecetes and hydatid cyst fluid of *Echinococcus granulosus*. *Int. J. Parasitol.*, **10**: 359.
- GUYTON, C. AND HALL, J.E., 1996. *Textbook of Medical Physiology*, ed. 9th. W.B. Saunders Company, U.S.A., pp.1-1148.
- HORTON, R.J., 1989. Chemotherapy of *Echinococcus* infection in man with albendazole. *Trans. Royal Soc. Trop. Med. Hyg.*, **83**: 97-102.
- IKRAM, M. AND HUSSAIN, S.F., 1978. Compendium of medicinal plants. Pakistan Council of Scientific and Industrial Research, Pakistan.
- IQBAL, Z., HAYAT, C.S., HAYAT, B. AND KHAN, M.N., 1986. Incidence of hydatidosis in Teddy goats slaughtered at Faisalabad abattoir. *Pakistan Vet. J.*, **6**(2): 70-72.
- IQBAL, Z., HAYAT, C.S., HAYAT, B. AND KHAN, M.N., 1989. Prevalence, organ distribution and economics of hydatidosis in meat animals at Faisalabad Abattoir. *Pakistan Vet. J.*, **9**: 70-74.
- JUNEJO, M.A., JUNEJO, A., MEMON, N., SIDDIQUI, S. AND PARADEEP, K., 1995. Hepatic hydatid cyst "Double line sign" on Ultrasound. *Pakistan Journal of Medical Research*, **34**(1): 254-256.
- KHAN, A.Z., 1990. *Implications of Clinical Chemistry*. In: *Diagnosis Lab. Systems*. Liaison Office, pp.12-14.
- KHAN, D. AND HASEEB, M.A., 1984. Hydatidosis of livestock in Pakistan. *Folia Parasitologica*, **31**(3): 288.
- MATOSSIAN, R.M., AWAR, G.N., RADWAN, H., CRAIG, P.S. AND MESHEFEDJIAN, G.A., 1992. Immune status during albendazole therapy for hydatidosis. *Ann. Trop. Med. Parasitol.*, **86**(1): 67-75.
- NADKARNI, A.K., 1954. *Indian Materia Medica*. 3rd Ed. (Revised and enlarged) Popular Book Depot, Bombay.
- OHGASHI, H., MINAMI, S., FUKUI, H., KOSHINIZU, K., MIZUTANI, F., SUGIURA AND TOMANA, T., 1982. Flavonols as growth inhibitors from the leaves of *P. persica*. *Agric. Biol. Chem.*, **46**(10): 2555-2562.
- OHNISHI, K., 1985. Isolation of larval *Echinococcus multilocularis* by injection of infected human hepatic tissue homogenate into the Chinese hamster. *Gen. Z. Parasit. Ked.*, **71**: 693-695.
- PAL, R.A. AND JAMIL, K., 1986. Incidence of hydatidosis in goats, sheep and cattle. *Pakistan Vet. J.*, **6**: 65-69.
- PANDEY, V.S., 1971. Biochemical observations on hydatid fluid. A preliminary report. *Indian Vet. J.*, **48**: 899-901.
- PANDEY, V.S., OUHELLI, H. AND MOUMAN, A., 1988. Epidemiology of hydatidosis/echinococcosis in Quarzazate the pre-Saharan region of Morocco. *Ann. Trop. Med. Parasitol.*, **82**(5): 461-470.

- SAID, M., 1969. *Hamdard Pharmacopeia of Eastern Medicine*. The Time Press Sadar, Karachi.
- SANCHEZ, F.A. AND SANCHEZ, A.C., 1971. Estudio do algunas propiedades fisicary y componentes quimicos del liquido Y pared germinative de diversas especies Y de diferente localization. *Revta. Iber. Parasitol.*, 31: 347-366.
- SANYAL, M., SHYAMAL, K.R. AND DATTA, P.C., 1985. Pharma cognostic evolution of *Embelia ribes* fruit. *J. Ecom. Taxon. Bot.*, 5(5): 1253-1256.
- SCHANTZ, P.M., 1984. Echinococcosis (hydatidosis). In: *Tropical and Geographical Medicine* (K.S. Warren and A.F. Mahmoud, eds.), pp.487-497. McGraw-Hill, New York.
- SCHWABE, C.W., 1986. Current status of hydatid diseases: A zoonosis of increasing importance. In: *The Biology of Echinococcus and Hydatid Disease* (ed. R.C.A. Thompson), pp.81-113.
- STALLBAUMER, M.F., CLARKSON, M.J., BAILEY, J.W. AND PRITCHARD, J.E., 1986. Epidemiology of hydatid disease in England and Wales. *J. of Hygiene*, 96(1): 121-128.
- VARDHANI, V.V. AND RAO, B.V.K., 1995. The relationship between serum cholesterol and parasitism in mice. *Pak. J. Zool.*, 27(4): 373-375.

(Received: March 19, 1998)