Use of alhagi roots extract as new alternative source of nutrition Part II

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Use of alhagi roots extract as new alternative source of nutrition Part II

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Abstract: Background: It's well known that worldwide popular explosion especially in the Arab World and deficiency in sources of nutrition especially in difficult conditions like natural disaster and wars. To overcome this problem we must find an alternative sources of nutrition that deals with it in this study. Objective: This application a trial to prove alternative nutrition for human in difficult conditions especially in natural disaster and wars. Material and Methods: This study included many stages:First stage: water extraction from Alhagi roots (L J)plant and calculate their concentration. Second stage: determination the composition extract qualitatively like carbohydrate, protein, fats , minerals and vitamins by Biochemical methods. Third stage: Applied a specific dose extract to animal lab rodent (rats) though out three groups (fifteen rats were divided in to three groups, five rats for each group. 1st group (G1) feeds with extract only, 2nd group (G2) feeds with extract and ordinary rats food and 3rd group control group feeds with ordinary rats food only. All rats feeds for six weeks, and studies the psychological behaviors like general appearance, sensorimotor behavior immobility and its reflexes, locomotion, skilled movement, and species-specific behaviors. Fourth stage: hematological tests, random blood sugar, lipid profile, renal function test, liver function test and estimation of serum electrolytes. Final stage: histopathology tests for kidney, liver, spleen, pancreas, stomach and intestine.Results: We monitoring the amount of this extract (L J)that drinking by rats and found the rats in both groups satisfied this drinking and drink as increasing manners each week and weighting the rats weekly and found there are no significant differences in the mean of weights of rats among all groups and all rats were increase its weights progressively each week. All the physiological, biochemical, hematological and histological results showed within normal values with no any significant changes between controls and study groups.Conclusion: we can use (L J) extract as supplement and complement alternative food supply without any toxic effect.

Key words : Nutrition , Alternative foods , Alhagi, lovely Juice LJ

1. Introduction:

The world and specially Arab World are in continuing research about developing and improvement varieties of food production [(1)]. On the other hand war continuity and more precisely in third world, the degradation of economic world, monopoly system in the industrial countries and increasing poverty rates [(2-4)]. All these facts were the reason behind the brain storming, which is considered to be helpful

technique that comes with better sources of alternative foods[5]. The triangle description for any good nutrition is varieties, moderation, and balance. These factors represent the angles of food triangle and help any human to lower the risk of heart diseases, diabetic and overweight [7]. Alhagi is a genus of old world plants in the family Fabaceae. They are commonly called camel thorns or manna trees [8] . It has been used locally in folk medicine as a treatment for glandular tumors and nasal polyps [9]. It is used as a medicinal herb for gastro protective, diuretic, expectorant, laxative, antidiarrheal and antiseptic properties and in the treatment of rheumatism and hemorrhoids [10] . It has also been used as a sweetener additives [11], but it's not used as source of food till now.

The aim of this application was finding out the alternative foods which is rich with great power that is regarded to be the most helpful source for human beings to survive with lower cost.

2. Materials and Methods:

The study was programmed to be conducted on lab animal (rats) was done in animal house and laboratories of College of Medicine/ University of Babylon. Its case control study. This was conducted from July 2017 to January 2018 and conducted by many stapes as follow:

1. General procedure for Alhagi roots extract: The fresh sample of Alhagi roots were collected from Al-Mahaweel region in Babylon province during July 2017, cleaned , dried and stored at 10 ^{0} C. The roots sample were cutting to small pieces for grinding. Ten grams of powder were extracted with 250 ml distilled water under Soxhlet Extraction Technique (SET) for about 60 minutes. The extract concentrated under vacuum by using lympholizer (Freeze dryer) to produce about 2.5 % .
2. Biochemical tests for plant extract : Alhagi roots extract was examined biochemically to determine the contains of this extract qualitatively.

A. Determination of the total sugar soluble (SS) in mg/ gm of extract: estimation of carbohydrate in plant extract according to the procedure describe by, Yemm, E. and Willis, A.J. [12] The amount of total SS present in the extract calculated by using standard prepared from graded of glucose.

General procedure: about 10 ml of ethanol 80% was added to 100 mg of extract sample then centrifuged at 4000 rpm for 15 minutes. The supernatant was collected and re-extract the residue one more time with ethanol 80% and centrifuged at 4000 rpm for 15 minutes. Mixed the residue with supernatant. Concentrated the ethanol extract by lympholizer to dryness. Add 1 ml of de-ionize water to each test tube with mixing then 4 ml of Anthrone reagent add along the wall of test tube and mixed gently then heated on water bath at 70 C° for 20 minutes. Cooling the sample then directly measured the absorbance at about 590 nm.

B. Determination of the total protein (TP) : TP contents were determined by the Bradford method [13,14].

C. Determination of the minerals: the extract was determine by using Trichloroacetic acid Extraction method [15].

D. Determination of the vitamins :the extract was determine according to the M Angeline Christie Hannah, S Krishnakumari publication [16].

3. Labe animals: fifteen rats were divided in to three groups, five rats for each group. 1st group (G1) feeds with extract only, 2nd group (G2) feeds with extract and ordinary rats food and 3rd group (control group) feeds with ordinary rats food only. All rats feeds for six weeks.
4. Examination and investigation of rats: after 6 weeks duration done the following investigation for treated and control rats :

A. Weighting of all rats every week.
B. Studies the psychological behaviors: like general appearance, sensorimotor behavior, immobility and its reflexes, locomotion, skilled movement, and species-specific behaviors.

C. Hematological testes: Hemoglobin (g/d), packed cell volume (%), white blood cells count (x1000/µl), neutrophiles (%), lymphocytes (%), monocytes (%), esonophiles (%), basophiles (%) and platelets count (x1000/mm3).
D. Biochemical testes: Renal function testes [blood urea (mg/dl), serum creatinine (mg/dl) and serum protein (g/dl)]. Liver function testes [alanine aminotransferase (IU/L), aspartate aminotransferase (IU/L), total serum bilirubin (mg/dl) and serum alkaline phosphatase (IU/L). Lipid profile [serum cholesterol (mg/dl) and serum triglyceride (mg/dl). Other investigations [random blood sugar (mg/dl), serum sodium (mmol/l), serum potassium (mmol/l) and serum calcium (mg/dl)].
E. Histological examination: a histological examinations were done for kidney, liver, spleen, pancreas, stomach and intestine.
5. Statistical analysis: All values were expressed as means ± SD. The data were analyzed by using of SPSS (statistical package for social sciences) version 17 and Microsoft Excel computerized programs and taking $p < 0.05$ as the lowest limit of significance. ANOVA test was used to compare between all parameters among all groups and within groups.

3. Results:

1. Qualitative examination of Alhagi roots extract: all the qualitative Biochemical tests gave positive results except lipids test was negative.
2. The amount of Alhagi roots extract (2.5%) drinking (ml/day) :

During monitoring the amount of extract daily, the conclusion was that the amount of drinking by rats in both G1 and G2 increasing direct. The rats in both groups satisfied this drinking and drink as increasing manners each week apart from 3rd week drink. These amount as shows in table (1) and figure (1) respectively.

Table (1) : The Mean ± SD of the amount of Alhagi roots extract (2.5%) drinking (ml/day) by rats each week.

Groups	**Mean ± SD of the amount of drinking extract (2.5%) ml/day**					
	1st week	**2nd week**	**3rd week**	**4th week**	**5th week**	**6th week**
G1	31.7± 2.2	71.4± 3.7	51.4± 2.9	64± 7.1	85± 11.2	87± 9.9
G2	35± 3.9	59.3± 5.5	50.7± 4.7	65.7± 8	80± 9.1	92.5± 5.4
P-values	> 0.5					

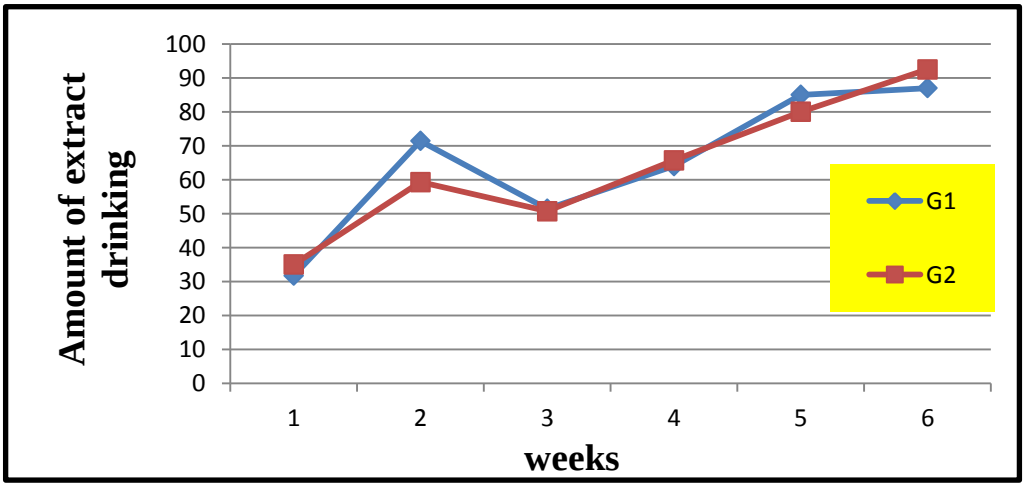

Figure (1): monitoring the amount of Alhagi extract that drinking by rats each week.

3. Weights of animals: weights the rats weekly and found there are no significant differences in the mean of weights of rats among all groups and all rats were increase its weights progressively each week. As shows in table (2) and figures (2).

Table 2: Represent the weight of animals weekly (five rat in each group)

Groups	Weights of rats (Mean ± SD) /gm					
	1st week	2nd week	3rd week	4th week	5th week	6th week
Control	176.6 ± 20.4	182 ± 18.3	188.8 ± 22.2	197.2 ± 19.9	202.4 ± 23.7	205.6 ± 20. 3
Group-1	151.2 ± 18.8	159.6 ± 17.3	169.4 ± 17.9	175 ± 19.5	186 ± 17.7	190.2 ± 18.6
Group- 2	192.8 ± 19.9	206.8 ± 20.1	214.2 ± 19	220.4 ± 21.8	225.8 ± 20.2	236.8± 21.5
P value	> 0.5					

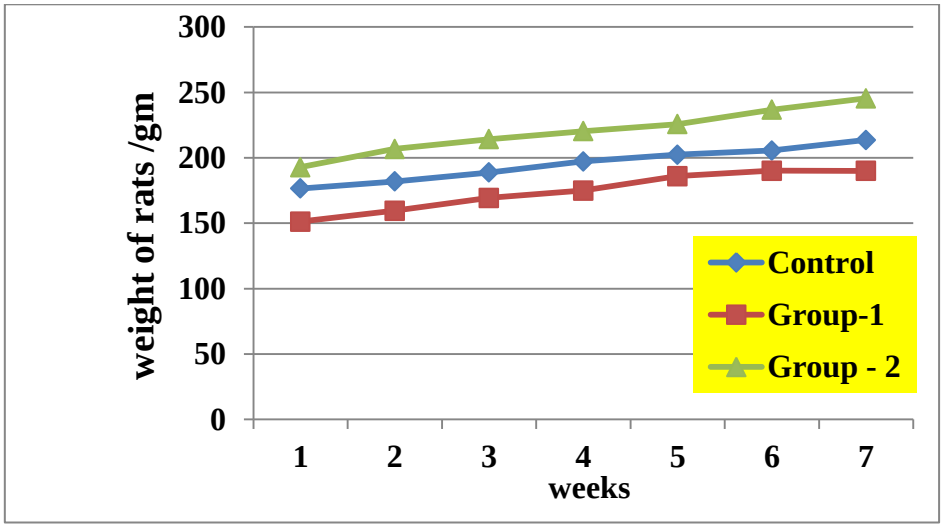

Figure (2): weight of rats weakly in all groups.

The psychological behaviors:

A. Vertical pole test : is used measure of motor coordination and balance. The animal is placed in the center of the pole.The time of latency to fall in group 1 was significantly increased as compared to both control group and group 2 ($P<0.05$). Also, the time of latency to fall in group 2 was significantly increased as compared to control group ($P<0.05$).These results indicate that extraction roots of L J plant increase the coordination and balance in the rats .

B.Hanging wire test : the test can measure the neuromuscular abnormalities by measuring motor strength of the animal.The time of latency to fall in group 2 was significantly increased as compared to both control group and group 3 ($P<0.05$). Also, the time of latency to fall in group 3 was significantly increased as compared to control group ($P<0.05$). Data shows that roots of LJ increase the strength of the muscles in the rats.

C.Forced swimming test: this test was proposed as a model to antidepressant activity.The total duration of immobility was recorded within the total test duration of five minutes. Each rat was used

only once. The swimming times in groups 2 and 3 were significantly decreased as compared to control group (P>0.05). These results indicate that roots of LJ have depressant effects in the rats.

D.Open field test : Rodents behaviors are changed when they are transferred into a new surroundings environment, and by using open field test under identical states situations any changes in the behavior such as anxiolytic or angiogenic activity can detected.There were no significant changes in the number of the squares crossed by rats, period of time spent in the central squared, and rearing in the groups 2 and 3 as compared to control group(P<0.05).While the number of grooming significantly increased in the groups 1 and 2 as compared to control group (P<0.05). These data show that there were no significant changes in the number of the squares crossed and rearing indicating that roots of Alhagi maurorum have no effects on spontaneous motor activity . The LJ extract increase their activity , this result indicate that roots of LJ have anxioginic effects on male rats.

4. Hematological and biochemical examination: by statistical analysis this study shows no significant differences in the hematological and biochemical parameters between and within groups and all values within normal range as shown in tables (3, 4, 5, 6 and 7).

Table (3): the hematological analysis for all rats in all groups

Parameters	Groups (Mean ± SD)			P-values
	Group-1 (N=5)	Group-2 (N=5)	Control (N=5)	
Hemoglobin (g/d)	14.7 ± 0.6	14.8 ± 0.3	15.5 ± 0.8	0.1
Packed cell volume (%)	45 ± 2	45.4 ± 0.8	47.6 ± 2.5	0.1
White blood cells (x1000/µl)	3 ± 0.3	2.7 ± 0.3	2.9 ± 0.08	0.6
Neutrophiles (%)	25.6 ± 2.1	26.6 ± 1.8	26 ± 1.8	0.7
Lymphocytes (%)	70.6 ± 2.4	68.2 ± 1.7	68.8 ± 2.	0.2
Monocytes (%)	2.4 ± 0.5	3.8 ± 1.3	3.2 ± 0.8	0.1
Esonophiles (%)	0.8 ± 0.4	1 ± 0.2	1.4 ± 0.5	0.4
Basophiles (%)	0.6 ± 0.5	0.40 ± 0.2	0.6 ± 0.4	0.8
Platelets (x1000/mm3)	6.4 ± 1	6.1 ± 1.2	7.3 ± 2.1	0.5

Table (4): The renal functions testes for all rats in all groups

Parameters	Groups (Mean ± SD)			P- Values
	Group-1 (N=5)	Group-2 (N=5)	Control (N=5)	
Blood urea (mg/dl)	45.8 ± 4.6	47.7 ± 5.6	41.8 ± 10.6	0.76
Serum creatinine (mg/dl)	0.34 ± 0.15	0.4 ± 0.14	0.26 ± 0.163	0.38

Table (5): The liver functions testes for all rats in all groups

Parameters	**Groups (Mean ± SD)**			**P-values**
	Group-1 (N=5)	Group-2 (N=5)	Control (N=5)	
Protein (g/dl)	5.9 ± 1.2	6.1 ± 0.8	5.5± 0.9	0.7
Alanine aminotransferase (IU/L)	33.9 ± 2.6	33.4 ± 3.6	36.9 ± 3.3	0.2
Aspartate aminotransferase (IU/L)	80.4 ± 7.6	86.8 ± 5	84 ± 10.9	0.5
Total serum bilirubin (mg/dl)	0.1 ± 0.01	0.06 ± 0.03	0.08 ± 0.06	0.7
Serum alkaline phosphatase (IU/L)	504.8 ± 60.6	508.8 ± 22.8	518.6 ± 15.2	0.7

Table (6): The lipid profile and random blood sugar for all rats in all groups

Parameters	Groups (Mean ± SD)			P- values
	Group-1 (N=5)	Group-2 (N=5)	Control (N=5)	
Serum cholesterol (mg/dl)	65.6 ± 6.1	65.40±7.232	74.00±3.000	0.06
Serum triglyceride (mg/dl)	47.8 ± 5.6	43.20±4.382	48.00±2.449	0.2
Random blood sugar (mg/dl)	79 ± 9.5	84.6 ± 11.6	72.6 ± 8.5	0.6

Table (7): The serum electrolytes for all rats in all groups

Parameters	Groups (Mean ± SD)			P-values
	Group-1 (N=5)	Group-2 (N=5)	Control (N=5)	
Serum sodium (mmol/l)	150.4 ± 4.1	148.8 ± 5.8	145.2 ± 4.6	0.3
Serum potassium (mmol/l)	4.1 ± 2.1	4.5 ± 0.6	4.7 ± 0.8	0.25
Serum calcium (mg/dl)	9.7 ± 0.3	10.2 ± 0.8	9.7 ± 0.2	0.22

5. **Histological examination**: after a histological examination of the viscera (kidney, liver, spleen, pancreas, stomach and intestine) for all rats in all groups we found no any pathological problems. As shown in pictures below.

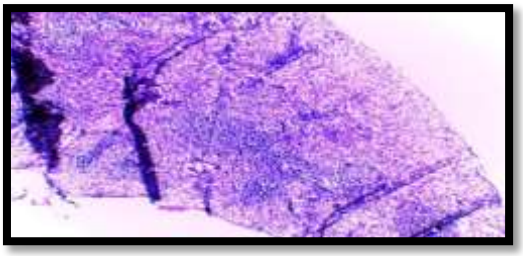

Figure 3: Spleen of rat in group-1

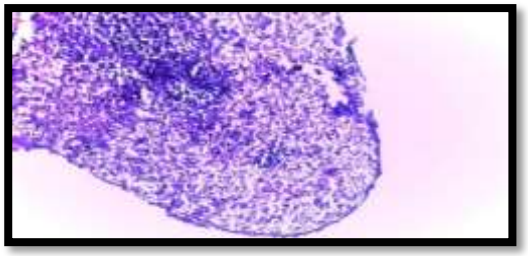

Figure (4): Spleen of rat in group-2

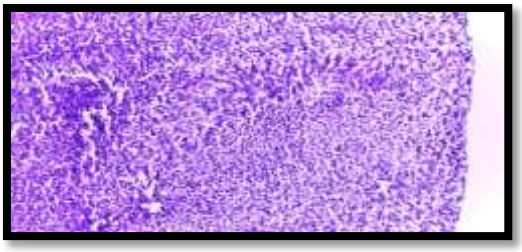

Figure (5): Spleen of rat in control group

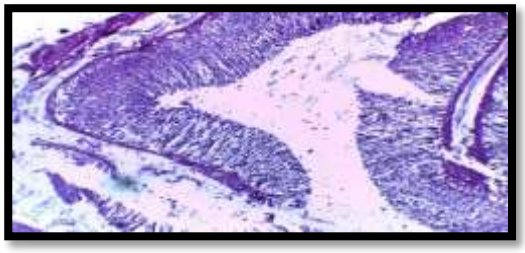

Figure (6): Stomach of rat in group-1

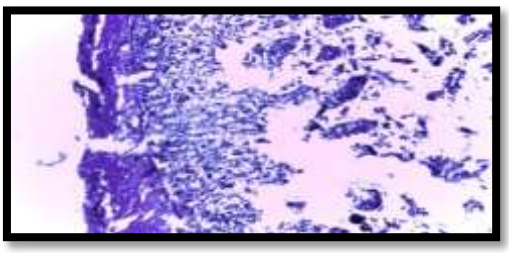

Figure (7): Stomach of rat in group-2

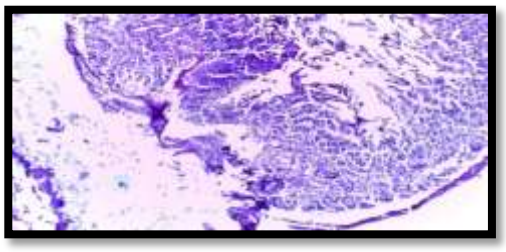

Figure (8): Stomach of rat in control

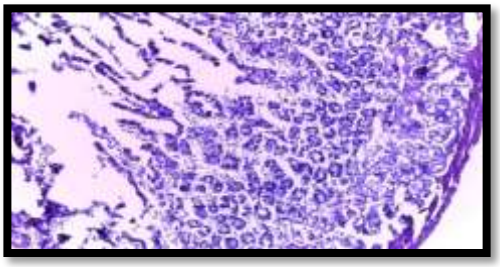

Figure (9): Stomach of rat in control group

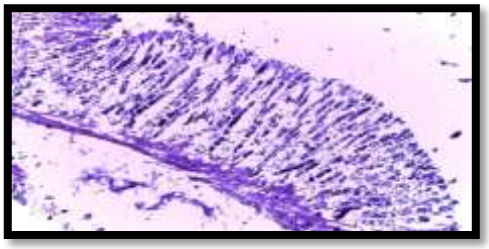

Figure (10): intestine of rat in group-1

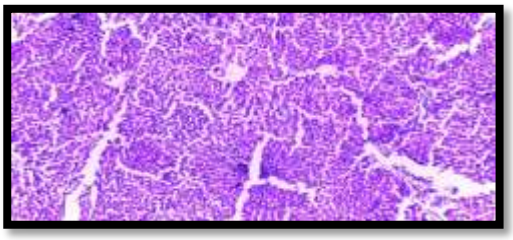

Figure (13): kidney of rat in group-1

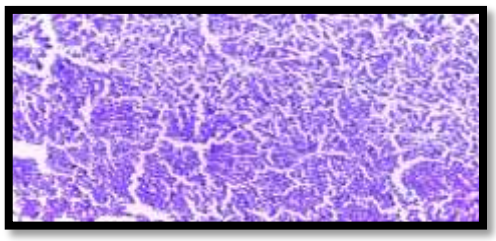

Figure (14): kidney of rat in group-1

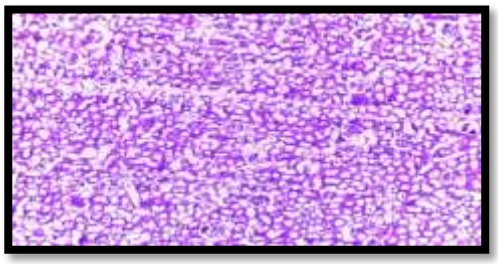

Figure (15): kidney of rat in group-2

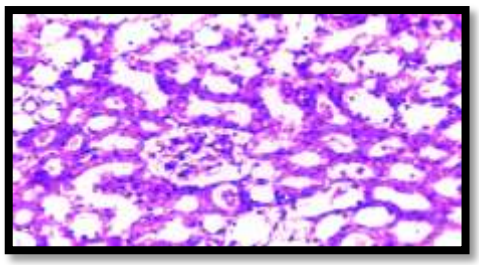

Figure (16): kidney of rat in control group

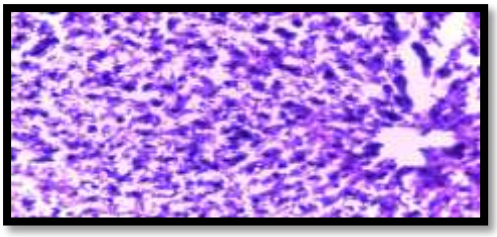

Figure (17): liver of rat in group-1

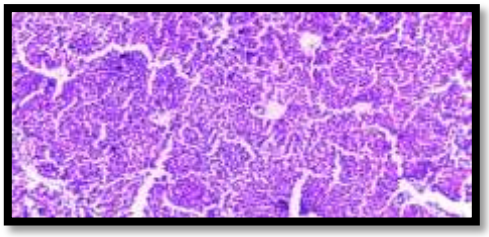

Figure (18): liver of rat in group-1

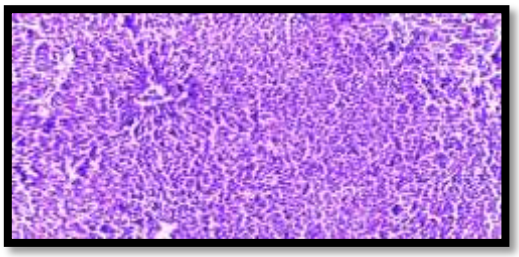

Figure (19): liver of rat in group-2

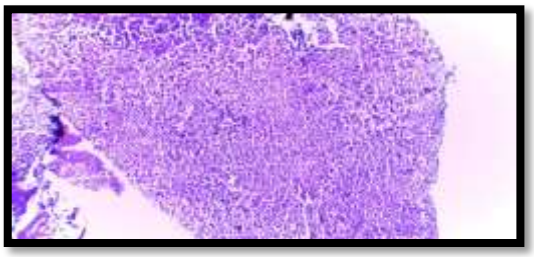

Figure (20): liver of rat in control group

4. Discussion:

A lot of publications about this kind of herbal plant .the roots extract that used previously as herbal therapy [17,18], but not yet uses as complementary or alternative food supplements. So this study dealt with this issue and the results of biochemical analysis illustrated that a roots extract contained good amount of carbohydrate, protein, minerals and vitamins that's important for maintenance life for a good time in stressful condition. On the other hand spectroscopic Analysis like FT.IR , 1H-NMR ,13C-NMR and Elemental analysis confirmed that plant have a lot of many bioorganic molecules like dimethoxylisoflavone, pratensein, tamarixetin, isoquercitrin, saliylic acid, vanillic acid, β-sitosterol and daucosterol[19]. The rats tolerate this extract and satisfied it. Also the animal gain weight without any psychological, physiological, hematological, biochemical and histological problems.

5. Conclusion and recommendation:

This study illustrated that the water root extract can uses as complementary or alternative food supplements (at least in rate till now) and recommended farther study to improve uses in human being.

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