In Vitro And *In Vivo* Evaluation Of The Antidiabetic Effect Of Different Extracts Of *Nepeta Cataria* In Streptozotocin Induced Diabetic Rats

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ABSTRACT: Both *in vitro* and *in vivo* studies were performed in the present research to investigate the pharmacological effects of successive *Nepeta cataria* extracts on some biochemical parameters in Streptozotocin diabetic rats compared to the currently used drug, Glicalized. These involved some biochemical parameters such as glucose, insulin, carbohydrate hydrolyzing enzymes; α –amylase, α- glucosidase, β- galactosidase, liver steoteosis; total cholesterol, HDL-cholesterol, LDL–cholesterol, triglycerides, total lipid, liver function enzymes; alanin aminotransferase(ALT), aspatrate aminotransferase (AST), alkaline phosphates (ALP) and total protein, oxidative stress markers; NO and DPPH. In addition histopathological investigations were performed. The results obtained revealed that *in vitro* analysis, different successive extracts of *Nepeta cataria* exhibited inhibitory effect on oxidative stress indices (NO and DPPH) and carbohydrate hydrolyzing enzymes (α-amylase, α-glucosidase and β-galactosidase) in linear relationships to some extent with concentration of inhibitors (dose dependant). Total ethanol (70%), petroleum ether and chloroform extracts showed respectively the most potent reducing power, while ethyl acetate and ethanol soxhlet appeared moderate or low reducing activity. In addition the *in vivo* anti-glycemic, antioxidant, antilipidemic effects of chloroform, petroleum ether as well as crude ethanol extracts in comparison with gliclazide as reference antidiabetic drug showed, these extracts have significant beneficial glycemic control, scavenging free radicals, normalized liver function, inhibited lipid synthesis associated with diabetic complication, as well as they have principle role in treatment and normalized liver and pancreas architecture. Hence, it could be concluded that *Nepeta cataria* extracts may be applied clinically for reducing complications against diabetes mellitus together with the ideal anti- diabetic drug glicalized. [Journal of American Science. 2010;6(10):364-386]. (ISSN: 1545-1003).

Keywords: Diabetes mellitus; *Nepeta cataria;* oxidative stress; liver function; carbohydrate hydrolyzing enzymes

INTRODUCTION

Diabetes mellitus (DM) is a serious health problem being the third greatest cause of death all over the world, and if not treated, it is responsible for many complications affecting various organs in the body. Diabetes mellitus is a disease results from abnormality of carbohydrate metabolism and characterized by absolute (type I) or relative (type II) deficiencies in insulin secretion or receptor insensitivity to endogenous insulin, resulting in hyperglycemia **(EL-Hilaly etal., 2007)**.

Hyperglycemia resulting from unregulated glucose level is widely recognized as the causal link between diabetes and diabetic complications. It was found that hyperglycemia cause tissue damage by mechanisms involving repeated changes in cellular metabolism. One of the key metabolic pathways as being major contributors to hyperglycemia induced cell damage, is the non enzymatic reaction between excess glucose and several proteins (as hemoglobin and albumin) to form advanced glycosylated end product (AGE). Production of AGE interferes with cell integrity by modifying protein function or by inducing receptors mediated production of reactive oxygen species (ROS)(**Thornalley , 2002**). Hyperglycemia-evoked oxidative stress plays a crucial role in the development of diabetic complications, including nephropathy, neuropathy, retinopathy and hepatopathy, which are considered to result from augmented reactive oxygen species generation, which probably results from both an excessive generation of reactive oxygen species and decreased antioxidant defenses**(Tepa *et al*., 2007).**

Hypoglycemic plants are still prevalent in developing countries , where they have been used to treat diabetes for many centuries More than 1200 species of plants have been used empirically for their alleged hypoglycemic activity This fact is attributed to the high cost and the lack of availability of current therapies for the majority of patients in developing countries. Nevertheless, many medicinal plants claimed effective by folk medicine require scientific investigation to ascertain their effectiveness, toxicity and then provide alternative drugs and therapeutic strategies(Marles and Farnsworth , 1994). There are more than 200 compounds from plant sources that have been reported to show blood glucose lowering effect. The wide variety of chemicals classes indicates that a variety of mechanisms of action are likely to be

involved in lowering blood glucose levels **(Kim *et al* ., 2008)**

Nepata cataria L.(family , Limiaceae; order Lamiales) , comprises about 400 spesies, most of which found in the Eastern Mediterranean, Southern Asia and China, is commonly known as Catnip or Catmint because of its irresistible action on cats. Due to lemony mint flavor it finds the ways in the herbal teas as well as in cooking. Medicinally, the plants is used in gastrointestinal and respiratory hyperactive disorders such, as colic, diarrhea, cough, asthma and bronchosis **(Miceli *et al.*, 2005)**. A limited number of studies exists on its biological activities include antibacterial **(Kalpoutzakis *et al.*, 2001)**, antifungal **(Nostro *et al.*, 2001)** and analgesic **(Aydin *et al.*, 1998)**. Various compounds have been identified by different groups of workers in the essential oil of *Nepeta cataria* the main constituents so far identified, include β-caryophyllene, caryophyllene oxide, 1,8 –cineol, citronellol , geraniol , elemol , nerol **(Mortuza –Semmani and Saeedi ,2004 ; Schultz *et al.*, 2004 ; Sajjadi , 2005)**.Also, urosolic acid, β–sisoterol, campesterol, α -amyrin, β-amyrin , and sitosterol β-glucopyranoside have been reported previously **(Miceli *et al* ., 2005).** In addition the plant also contained neptalactones and alkaloids, such as actinidine and iridomyecine **(Kalpoutzakis *et al*., 2001)**.

So the present study was designed to evaluate *in vitro* the inhibitory effect of different successive extracts of *Nepeta cataria* on oxidative stress markers(DPPH and NO) and carbohydrate hydrolyzing enzymes (α- amylase , α –glucosidase and β-galactosidase) . In addition the research is extended *in vivo* to demonstrate the hypoglycemic efficiency of petroleum ether, chloroform as well as 70% ethyl alcohol extracts of *Nepeta cataria* [compared with antidiabetic Gliclazide (diamicron) reference drug] in Streptozotocin induced diabetes mellitus in rats through measuring glucose, insulin levels, carbohydrate hydrolyzing enzymes, (α-amylase, α–glucosidase and β- galactosidase), free radical scavenger effects(NO) liver function enzymes (ALT, AST, ALP and total protein), lipid profile (total cholesterol , HDL-cholesterol , LDL –cholesterol , triglycerides and total lipid). Moreover histological examination of liver and pancreas was performed.

MATERIALS AND METHODS

Chemicals:

All chemicals in the present study were of analytical grade, product of Sigma, Merck and Aldrich. All kits were the products of Biosystems (Spain) , Sigma Chemical Company (USA), Biodiagnostic (Egypt).

Plant materials:

Seeds of catnip (*Nepeta cataria* L.) was obtained from company of Jelitto staudensamen, Schwarmstedt, Germany). The seeds of the plant were cultivated in the experimental farm of the Cultivation and Production of Aromatic Plants Department of the National Research Center, Giza, during two successive seasons of 2006 and 2007.

The seeds of catnip were sown in nursery on 15th of October in the two seasons. Two months later after sowing, the seedlings were transplanted in 8 cm pots in medium of 1:1:1 loam, sand and peat moss (by volume). The seedlings were planted in the field on 15th of March in hills 25 cm apart on rows 60 cm in-between.

The flowering aerial parts of *Nepeta cataria* was collected from the plants cultivated in the experimental farm of the Cultivation and Production of Aromatic Plants Department of the National Research Center, Giza, during two successive seasons of 2006 and 2007, and raised from seeds obtained from company of (Jelitto staudensamen , Schwarmstedt, Germany)

2. Phytochemical study

2.1. Phytochemical screening

The powdered air-dried aerial parts of *Nepeta cataria* was screened for carbohydrates and/or glycosides, flavonoids, tannins, saponins, coumarins, unsaturated sterols and/or triterpenes and alkaloids, applying chemical tests and thin layer chromatography **(Wagner *et al*., 1983)**.

2.2. Preparation of extracts and fractions

The powdered air-dried aerial parts of *Nepeta cataria* (720 g) was extracted with petroleum ether (60-80), chloroform, ethyl acetate and ethanol, in succession, to afford 35, 28, 4.5 and 29 g of extracts, respectively.

1- *in vitro* antioxidant study:

i-The antioxidant scavenging activity of serial concentrations of different plant extracts (10:1000µg/ml) on DPPH free radical was performed according to the method of **Chen, *et al*.,(2007).** The non reacted radical form of $DPPH^{\cdot}$ absorbed in the visible range, and the spectroscopic method is based on the measurement of color intensity at 518 nm against blank solution. The decrease in optical density of $DPPH^{\cdot}$ was calculated in relation to control as follows:

$$\% \text{ Inhibition} = \frac{A_{control} - A_{sample}}{A_{control}} \times 100$$

ii- Determination of NO- free radical scavenging activity: The NO-scavenging activity of extracts was determined according to the method of **Sreejayan and Rao (1997).** The absorbance of the chromophore formed during diazotization of nitrite

with sulphanilamide and its subsequent coupling with N-(1) naphthyl ethylene diamide was read at 546nm**.**

iii- Determination of α-Amylase inhibitory activity: α- Amylase was determined according to the method of **Ali et al., (2006)** which is the modified method of **Bernfeld (1955).** α-Amylase activity was determined by measuring absorbance at 540 nm.

iv- Determination of β-galactosidase inhibitory activity: β-galactosidase inhibitory activity was measured by the method of **Sanchez and Hardisson (1979)**, where the resulting colour of O-nitrophenolate ions was measured at 420 nm. Standard curve was done using different concentrations of O.nitrophenol.

v- Inhibition assay for α-glucosidase activity: α-glucosidase inhibitory activity was determined according to the method of **Kim *et al*., (2005)**, where reducing activity was estimated by measuring release of *p*-nitrophenol from *p*-nitrophenyl α-D-glucopyranoside at 400 nm. Standard curve was done using different concentrations of p.nitrophenol.

II- **Determination of hypoglycemic activity of *Nepeta cataria in vivo*:**

Animals:

98 male Wister albino rats (120-150g.) were obtained from animal house of National Research Centre, Dokki, Giza, Egypt. Rats were fed on a standard diet and free access to tap water. They were kept for one week to be acclimatized to the environmental conditions.

Toxicity studies:

Four main groups of male Wister albino rats were selected to study the acute toxicity of all plant extracts under investigation. Each main group was subdivided to 4 subgroups (6 rats each). All groups received one oral dose of 100, 300, 500 and 1000mg of plant extract/kg body weight. After 24 hours, there were no died animals; representing the safety action of all extracts.

Doses:

All plant extracts were administrated orally with a dose of 50 mg/kg body weight for 30 consecutive days. The dose was selected according the toxicity study, which were safety for all doses used. The selected dose was also confirmed by (**Miceli *et al.* 2005**) who used a dose of 50mg/kg body weight of the methanol extract of *Nepeta sibthorpii Bentham (*Lamiacae) leaves and flowers. **Rabbani et al. (2007)** who used a ranged dose between 25 and 400mg/kg body weight of the *hydro alcoholic* extract of *Nepeta persica* given at a dose 50 mg/kg body weight. Dose of the plant extract seemed to be the optimal dose.

Experimental design:

98 male albino rats were selected for this study and divided to fourteen groups (seven rats in each group) as follows:

Group 1**:** normal healthy control rats.

Groups 2-4: normal healthy rats orally administrated with different plant extracts (50 mg/kg body weight daily for 30 days , each rat received 7.5 mg /0.5 ml bidistilled water 15% w/v) .

Groups 5-7: considered as diabetic groups; where type 1 diabetes was induced by streptozotocin, each rat was injected intraperitoneally with a single dose of streptozotocin (65mg/Kg body weight dissolved in 0.01M citrate buffer immediately before use, each rat was received 9.75 mg /0.5ml citrate buffer 19.5 w/v) (**Vats *et al.*, 2004).** After injection, animals had free access to food and water and were given 5% glucose solution to drink overnight to counter hypoglycemic shock **(Bhandari, et al., 2005).** After 2 days of STZ injection fasting blood samples were obtained and fasting blood sugar is determined (>300 mg/dl). Hyperglycemic rats were used for the experiment and classified as follows:

Group5: Diabetic +ve control group sacrificed after 2 days of STZ injection

Group 6: Diabetic +ve control group sacrificed after 10 days of STZ injection

Group 7: Diabetic +ve control group sacrificed after 40 days of STZ injection and considered as recovery group.

Group 8: Diabetic animals treated with ***petroleum ether extract*** 50g/kg body weight for30 days, each rat received 7.5 mg /0.5 ml bidistilled water 15% w/v)

Group 9: Diabetic animals treated with ***chloroform extract*** (the same as group 8).

Group 10: Diabetic animals treated with ***70% ethanol extract.*** (the same as group 9).

Group11: Diabetic animals treated with glicalized 10mg /kg body weight (each rat received 1.5 mg/0.5 bidstilled water 3% w/v) gliclazide (diamicron) and considered as reference drug.

Group 12-14: healthy control group sacrificed after 2, 10 and 40 days and considered as control groups for body weight , liver weight and liver weight / body weight ratio of diabetic groups sacrificed after2,10and 40 days respectively.

Sample preparations:

Serum sample: Each animal was weighed, Blood collected by puncture the sub-tongual vein in clean and dry test tube, left 10 minutes to clot and centrifuged at 3000 r.p.m for serum separation. The separated serum was stored at -80^{o}C for further determinations of lipid profile, liver function tests, oxidative stress marker carbohydrate metabolizing enzymes and serum total protein.

Tissue sample: liver tissue was weighed and homogenized in cold 0.9 N NaCl (1:20 w/v), centrifuged at 3000 r.p.m for 10 minutes, separated the supernatant and stored at -80^{o}C for further determinations. For determination of NO in liver tissue

the extraction method was carried out using 1g tissue and homogenized in 10 ml 10% trichloroacetic acid (to give 10% homogenate 1%w/v).Then the sample was Centrifuge at 3,000 rpm for 10 min. and the supernatant was separated and stored .

Blood biochemical analysis:

i-Determination of blood glucose:-

Glucose was determined in serum by colorimetric assay according to ***Goldstein et al.,* (1994*).***

ii- Human Insulin Enzyme Immunoassay :

Insulin was determined by Insulin quantitative test kit according the method of **Sacks (1994)** based on a solid phase enzyme-linked immunosorbent assay. The colour is changed to yellow and measured spectrophotometrically at 450 nm. The concentration of Insulin is directly proportional to the colour intensity of the test sample.

iii- **Determination of total cholesterol in rat serum:**

Cholesterol was determined by the method of **Stein (1986)** The cholesterol is determined after enzymatic hydrolysis and oxidation. The quinoneimine is formed from hydrogen peroxide and 4-aminoantipyrine in the presence of phenol and peroxidase. Free and esterified cholesterol in the sample originates by the means of the coupled reactions, a coloured complex that can be measured at 500nm spectrophotometrically.

iv- Determination of cholesterol - HDL in rat serum:

Cholesterol- HDL was determined by the method of **Stein (1986)**.

Phosphotungstic acid and magnesium ions selectively precipitating all lipoproteins except the HDL – cholesterol present in the supernatant can be determined by the same method used for total cholesterol. Low density lipoprotein (LDL) in the sample is precipitated with phosphotungstate and magnesium ions. The supernatant contains high density lipoprotein (HDL). The HDL-cholesterol is then measured spectrophotometrically at 500 nm by the means of the coupled reactions.

v- **Determination of cholesterol- LDL in rat serum:**

Cholesterol- LDL was determined by the method of **Assmann et al. (2004)**.

Low density lipoprotein (LDL) in the sample precipitate with polyvinylsulphate their concentration is calculated from the difference between the serum total cholesterol and cholesterol in the supernatant after centrifugation. The cholesterol is then measured spectrophotometrically at 500 nm by the means of the coupled reactions.

vi- Determination of triglyceride:

Triglyceride was measured by the method of **Wahelfed (1974)** Triglycerides in the sample originates by the means of the cou

vii- **Determination of total lipids in rat serum:**

Total lipids was measured by the method of **Zollner and Kirsch (1962)** .

Lipids react with sulphuric, phosphoric acids & vanillin to form pink colored complex which measured calorimetrically at 545 nm.

viii- **Determination of alkaline phosphatase in rat serum:**

Alakaline phosphatase was measured by the method of **Belfield and Goldberg (1971).**

The liberated phenol is measured colorimetrically at 520 nm in the presence of 4- aminophenazone and potassium.

x- **Determination of aspartate aminotransferase (AST) and alanine aminotransferase in rat serum:**

AST and ALT were measured by the method of **Reitma**

Determination of AST or ALT activities were based on the keto acids pyruvate or oxaloacetate formed which is measured colorimetrically at 505 nm in its derivative form, 2,4-dinitrophenylhydrazone .

xi- **Determination of total proteins:**

Total proteins was assayed in rat serum by colorimetric assay according to the method of **Bradford, (1976**).Total proteins react with Bradford reagent to give a blue complex which is measured colorimetrically at wavelength 595 nm. Standard curve of protein was assessed using serial concentration of bovine serum albumin.

xii-Determination of nitric oxide (NO)in liver tissue homogenates:-

NO was determined in liver tissue homogentes according to **Moshage *et al*., (1995)**.

Promega´s Griess Reagent system is based on the chemical reaction between sulfanilamide and N-1-naphthylethylenediamine dihydrochloride (NED) under acidic condition to give colored azo colored – compound which can be measured colormetrically at 520-550nm .

xiii- Determination of α- amylase in liver tissue homogenates :

α- amylase activity was performed in liver tissue homogenate according to the method of **Caraway, (1959)**. The test is based on the hydrolysis of starch by **α-** amylase and the blue –black complex that forms when iodine reacts with starch. The amount of starch remains at the end of the incubation period is shown by produces a blue black color. The amylase activity is measured by the differences in absorbance of the starch –iodine complex at 660nm.

α-Glucosidase and β-galactosidase were performed in liver tissue homogenates by the same methods as *in vitro*, also total protein content was

estimated in liver homogenates by the same method as in serum.

III- Histopathology :

Liver and pancreas specimens were fixed in 10% formalin, processed to paraffin blocks, sectioned (4 μm thick) and stained with Hematoxyline and Eosin stain. They were examined using light microscopy for demonstration of pancreatic and haptic pathological changes including degeneration of β-cell of langerhans, atrophy, cell destruction, necrosis and the efficiency of *Nepeta cataria* extracts to ameliorate these pathological features.

Statistical analysis :

Data were analyzed by comparing values for different treatment groups with the values for individual controls. Results are expressed as mean ± S.D. The significant differences among values were analyzed using analysis of variance (one-way Anova) coupled with post-Hoc least significance difference (LSD) Anova at $p \leq 0.05$ using Co-stat program.

RESULTS

The present results demonstrated *in vitro* the inhibitory effect of different successive extracts, petroleum ether, chloroform, ethyl acetate and ethanol as well as 70% ethanol extract of *Nepeta cataria* , in addition the research is extended to evaluate e the *in vivo* biochemical effects, mechanism(s) of the hypoglycemic actions of the most potent extracts of *Nepeta cataria* and to investigate its possible hepatoprotective effects against liver disorders induced by reactive oxygen species associated with diabetic complications in diabetic rat. The investigated parameters included blood glucose, insulin (to investigate diabetic condition), total cholesterol, HDL-cholesterol, LDL-cholesterol, triglycerides, total protein, total lipid content as indices of liver steatosis, alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP) and total protein content as markers of liver functions, as well as hepatic nitric oxide (marker of lipid peroxidation) in addition hepatic α - amaylase, α-glucosidase and β- galactosidase indices for carbohydrate hydrolyzing enzymes .

Table (1) and Fig (1) show the DPPH free radical scavenging effects of different successive extracts of *Nepeta cataria.* All the tested extracts show appreciable free radical scavenging activities. 70% ethanol extract has the strongest radical scavenging activity at different concentrations compared to other successive extracts followed by petroleum ether and chloroform. However ethanol extract shows the lowest radical scavenging activity. A dose –response relationship is found in the DPPH radical scavenging activity, the activity increased as the concentration of extract increased for each individual extract. Total ethanol extract is able to reduce the stable radical, DPPH to the yellow –colored 2,2-diphenyl-1- picryl hydrazyl to give significant inhibitory percent 47.7±0.84, 60.53 ±0.50, 66.45± 0.29, 69.52 ± 52 and 72.19±0.69 % at concentration of inhibitor 10, 50, 100, 500 and 1000 μg/ml respectively. While petroleum ether extract shows reducing inhibitory percent of DPPH amounted 26.31 ± 0.50 %, 26.00±1.38%, 34.04 ± 0.94 % , 37.39±1.10 % and 40.17±0.9% at the same concentrations of inhibitors respectively . On the other hand, chloroform shows significant reducing activity of 16.39±4.63, 22.38 ±1.23, 32.54±1.42 and 35.94±1.42 and 38.48±0.70 %, respectively. In addition, ethyl acetate and ethanol extracts exhibit the lowest reducing activity power when compared to other extracts.

Table (2) and Fig (2) illustrate NO free radicals scavenging capacities of the different successive extracts of *Nepeta cataria* .The expression of antioxidant activity is thought to be concomitant with the development of reductons as these species are known to be free radical chain terminators. Therefore it was considered important to determine the reductive capacity of the different successive *Nepeta cataria* extracts as these may indicate their potential as antioxidants. The most reducing capacities is also considered for 70% ethanol extract which is capable of significantly reducing activity of NO by 37.60±1.68%, 39.16±9.13% ,52.13 ± 6.06%, 56.58±3.46% and 53.85±5.12 % with various concentrations of inhibitors (10-1000 μg/ml) and more or less did so in a linear concentration –dependent fashion. The reducing power of petroleum ether extract which has been used as one of antioxidant capability indicators is shown to be came later total ethanol extract and exhibits significant reducing activity of 15.26± 6.94, 24.94± 2.89, 33.30 ±2.65, 36.29 ±5.18 and 44.64 ± 4.29 %. From the percentage scavenging values, it can be seen that chloroform, ethyl acetate and ethanol as successive extracts demonstrate a moderate scavenging effect with linear relationship in a dose dependent manner and record significant maximum reducing percent of 41.24±3.27, 38.17±3.01 and 36.14±2.93 % at concentration of inhibitors 1000 μg/ml respectively.

Table (3) and Fig (3) show five successive extracts of Nepeta *cataria* that are tested to evaluate their effect on α -amylase carbohydrate hydrolyzing enzyme activity. Among them total ethanol extract as well as successive petroleum ether, chloroform and ethyl acetate extracts show interesting and comparatively potent α-amylase inhibitory activity which may be potentially useful in control of obesity and diabetes. The inhibition of α-amylase by chloroform and ethyl acetate extracts appear to be also a dose dependent and are found to have the highest significant reducing activity 44.59± 1.55 and 43.64 ±

1.79 % at concentration of inhibitor 1000µg/ml. The extent of α-amylase inhibition activity appears with crude ethanol extract since the inhibiting effect is most dramatic at 10 and 50 µg/ml inhibitors to give insignificant reducing activity 92.00±1.21 and 94.35±2.69 % respectively. While inhibitory activity recorded 88.40±7.29 % at 1000 µg /ml extract. Petroleum ether is shown to follow directly 70% ethanol extract as a second potent inhibitory effect on α -amylase by recorded significant inhibitory percent of 18.26± 3.97, 24 .55 ±4.03, 32.56±2.07, 37.14±0.89 and 37.97±1.86% in a dose dependent manner at 10,50.500 and 1000 ug/ml inhibitors respectively. Considering ethanol as a successive extract, shows the lowest inhibitory activity so it is demonstrated as a weaker α-amylase –inhibitory activity, in spite of it appears significant reducing power at concentration of inhibitor 1000µg/ml (38.13±0.64).

Table (4) and Fig (4) manipulated the inhibitory effect of different successive extracts of *Nepeta cataria* on α- glucosidase carbohydrate hydrolyzing enzyme activity. Remarkable inhibitory effect of total ethanol, petroleum ether, and chloroform is obtained greater than ethyl acetate or ethanol as successive extracts. The inhibition mode of the different extracts against α-glucosidas is similar, gives significant highest percent of reducing activity at 100 and 1000 µg / ml inhibitors reached to 40.44± 4.34, 40.55± 3.08, 32.22 ±3.96 , 28.95± 1.44 and 44.21 ±2.87% for petroleum ether, chloroform, ethyl acetate, ethanol as successive extracts and 70% ethanol extracts at 100µg/ml, respectively .While at concentration of inhibitors 1000µg /ml, the reducing activity amounted 39.32± 2.60, 38.11 ±2.70, 29.42± 0.62, 27.65± 2.37 and 42.50± 2.24% for the same previous reported extracts respectively. However, the inhibition percent appears to some extent be significant correlated with increase in concentration of inhibitors.

Table (5) and Fig (5) show β-galactosidase inhibitory activity against five extracts of *Nepeta cataria*. 70% ethanol, petroleum ether and chloroform extracts a gain provided additional support for the previous finding by having the strongest reducing activity at various concentration of inhibitors. Since total ethanol extract at 500 and 1000µg/ ml gives significant highest inhibitory percent amounted 67.82± 3.94 and 66.86 ± 3.79%, respectively followed by petroleum ether(62.63 ± 1.89 and 62.12± 4.37%, respectively) and chloroform (54.13 ± 2.44 and 55.08 ± 5.11 %, respectively). It can be noticed that ethyl acetate and ethanol successive extracts have comparable insignificant inhibitory activity of 45.89± 4.91, 43.77 ± 4.5, 46.58 ± 8.22 and 42.21± 5.28 % at 500 and 1000 µg/ml inhibitors respectively, and they are considered much weaker β-galactosidase inhibiting activity. From the table of inhibition we can deduce that, significant increase in reducing activity with increase in concentrations of the individual extracts (linear relationship) and at low doses the reducing activity shows insignificant change.

Table (6) and Fig (6) demonstrated the blood glucose and insulin levels in serum of control, normal - treated, diabetic and diabetic – treated groups. It is obvious that, there is no significant change between normal control and different control –treated groups either in blood glucose or in insulin levels except normal control –treated with 70% ethanol extract which exhibits significant reduction in blood glucose level amounted 94.50±16.05 mg/dl at P≤ 0.05 with percentage reduction -13.69%. Concerning diabetic groups, significant increase is noticed in blood glucose levels which is concomitant with significant reduction in insulin levels at day 2, 10 and 40 post injection of STZ drug record 373.00±2.94, 363.00±2.94 and 364.25±3.77 mg/dL with percentage increase +240.64 ,+231.51 and +232.62% for glucose, respectively and 0.59±0.01, 0.34±0.05 and 0.35±0.01 µIU/ml with percentage reduction reached to -88.97, -93.64 and -93.45%, for insulin, respectively. Significant amelioration is noticed in blood glucose and insulin levels in all diabetic – treated groups recorded the most pronounced effect for 70% ethanol extract (-5.94% for glucose and -14.21%.for insulin) followed by petroleum ether extract (+17.12 and -22.8%, respectively) then glicalized as a reference drug (+20.55and -27.66% respectively) and finally chloroform extract (+21.46 and -28.41%, respectively).

Table(7) and Fig (7) show the levels of lipid profile, total cholesterol, HDL- cholesterol, LDL-cholesterol, triglycerides and total lipid in control, normal - treated, diabetic and diabetic – treated groups. It can be easily noticed that, there is no significant change in total cholesterol, HDL- cholesterol , LDL-cholesterol and total lipid between different control – treated groups as compared to untreated control one. While triglycerides show significant reduction in crude ethanol extract treated – control group reached to 93.32±22.07 mg/dl with percentage reduction amounted -15.84% as compared to normal control group. With regard to diabetic groups, significant elevation in lipid profile is observed 2 days post STZ injection as compared to normal control group record 202.50±4.5, 81.62 ±3.35, 163.22±4.57, 211.75 ±8.22 and 1687.19 ±202.10 mg/dl with percent of elevation +54.17, +21.75 , +94.75, +90.97 and +72.05% for total cholesterol, HDL-cholesterol, LDL– cholesterol, triglycerides and total lipid, respectively. On the other hand, nearly simultaneously elevated levels in lipid profile is recorded 10 and 40 days post STZ injection with percentage of increase +82.66 , +35.37 ,+151.46, +177.57 and +84.12% for 10 day and+82.54,+35.37,

+151.46, +178.45 and +84.45% for 40 day, respectively. All diabetic – treated groups(G8- G11) show significant enhancement in lipid profile level as compared to the normal control group, since in petroleum ether –treated diabetic group, total cholesterol record 145.99± 10.97mg/dl with percentage increase amounted +11.15%.While in both total ethanol extract and chloroform, total cholesterol reach to 150.81±6.46 and 158.15±8.20 mg /dl, respectively with percent of +14.82 and +20.41%, respectively. With respect to gliclazide as reference drug, total cholesterol amounted 72.75±0.96 mg/dl with percent of reduction - 44.61% as compared to normal control group. Concerning HDL –cholesterol nearly similar results is obtained for different extracts-treated diabetic groups and gliclazide as reference drug, where insignificant change is observed either compared to each other or to normal control group. In addition, significant improvement is noticed in LDL-cholesterol level post treatment of diabetic rats with different extracts where insignificant change is observed as compared to each other and significant increase as compared to normal control group amounted 107.89±14.17, 118.02±11.50 and 111.25±2.15 mg /dl with percentage of elevation + 28.73,+40.82 and +32.74% for petroleum ether ,chloroform and 70% ethanol extracts, respectively. Controversy gliclazide -treated diabetic group exhibits insignificant change as compared to normal control and significant decrease as compared to other extracts. Remarkable, significant enhancement is noticed in triglycerides level post different types of treatments, where insignificant change is recorded as compared to normal control except for petroleum ether-treated diabetic rats, significant increase is noticed amounted 125.74± 10.83 mg/dl (+13.40%) as compared to normal control group. Total lipid reveals an enhanced significant mean value of 1373.17±41.65, 1414.58±195.92 , 1325±99.71 and 1237.50±33.04 for petroleum ether , chloroform , 7 ethanol extracts and gliclazide drug as compared to normal control(980.62±30.71) with percent +40.03, +44.25.,+35.12 and +26.20%, respectively.

Table (8) and Fig (8) demonstrate the level of liver function enzymes AST, ALT, ALP and total protein in serum of control, normal treated, diabetic and diabetic treated groups. It is obvious that insignificant change is recorded in AST level in serum of normal control treated either with chloroform or crude ethanol extracts. On the contrary significant inhibition is noticed in AST activity in normal control treated with petroleum ether extract amounted 1.97±0.25 umole /mg protein /min (-12.83 %.). With respect to ALT and Total protein, insignificant change is observed in their level in different control- treated groups as compared to untreated control one. Concerning ALP, significant inhibition is noticed in all control–treated groups recorded 2.94±0.40, 2.74±0.25 and 2.78±0.26 umole/mg protein/min with percentage decrease amounted -17.99 ,-13.27 and -19.17% for chloroform, 70% ethanol and petroleum ether extracts respectively as compared to the normal control group. With regard to diabetic condition, significant increase in all enzyme activities is noticed at day 2,10 and 40 post STZ injection reached to 2.73±0.24, 3.23±0.23, 3.37±0.11 µmole / mg protein/min for AST with percentage increase +20.80,+42.92 and +49.12%, respectively. While , ALT recorded 1.97±0.09, 2.44 ±0.17 and 2.48±0.08 µmole/ mg protein /min with percentage increase +23.13,+52.50 and +55.00%, respectively. ALP shows a value of 4.51± 0.36, 5.55±0.60 and 5.61±0.13 µmole/mg protein/min with percentage of elevation +33.04, +63.72 and +65.49%, respectively. Total protein content shows insignificant change at day 2 and 10 post STZ injection, while marked significant reduction at day 40 amounted 84.3 ±1.74 mg / ml (-22.12%). The curative effect of petroleum ether, chloroform, 70% ethanol extracts and gliclazide drug on diabetic rats can easily be noticed through the normalization of all enzymes tested returned more or less to the level of normal control, where an insignificant change is observed. While total protein content still recorded significant reduction post petroleum ether, chloroform and total ethanol extracts treatment. (In spite of its normalization with gliclazide drug) amounted 83.25±5.37, 89.25±4.92 and 83.50±5.06 mg/ml. with percentage decrease -23.09, -17.55 and -22.86%, respectively.

Table (9) and Fig (9) show the manipulation of NO in liver tissue homogenates of control, normal treated, diabetic and diabetic–treated groups. It can be easily noticed that NO level is insignificantly affected post various extracts- treated normal control rats as compared to untreated control one. In response to diabetic state, NO shows significant increase in its level of a value 62.96±2.32, 72.55±1.87 and 72.55 ±2.00 µg/g tissue for NO with percentage increase +42.86,+64.62 and +64.62 %.at day 2, 10 and 40 post STZ injection, respectively. The level of NO is significantly improved as a result of different treatments, shows the best pronounced effect for 70% ethanol extract and petroleum ether, where insignificant change is recorded either as compared to normal control or diabetic– gliclazide treated groups. In spite of, significant elevation in NO level is noticed in diabetic –chloroform treated group amounted 51.30 ±2.69µg /g tissue with percent +16.41 %.

Table (10) and Fig (10) recorded the level of carbohydrate metabolic enzymes α - amylase, β-galactosidase and α -glucosidase in liver tissue homogenates of the different studied groups. Careful inspection of the data would reveal that α - amylase

and β- galactosidase show insignificant change in different control- treated groups as compared to the normal untreated control one. While α-glucosidase shows significant increase as a result of treatment with both total ethanol (0.342±0.04 µmole/mg protein/min) and petroleum ether extracts (0.370±0.06 µmole/mg protein/min) with percentage increase amounted +25.74 and +36.03 %, respectively. On the other hand, total protein content reveals a significant reduction with a value of 39.50±8.22, 36.25±10.30 and 41.25±8.54 mg/g tissue in response to treatment of normal control rats with petroleum ether, chloroform and 70% ethanol extracts with percent -21.0, -27.5 and -17.5 %, respectively. It can be deduced that carbohydrate metabolizing enzymes are strongly affected with diabetic condition show significant inhibition at day 2, 10 and 40 post STZ injection reached to 20.07 ±1.40, 17.79 ±0.68 and 18.32 ±0.69 µmole/ mg protein/min for α-amylase with percent of change -38.27, - 45.28 and -43.65%, respectively. While, β-galactosidase enzyme activity recorded 0.052±0.02, 0.037±0.01 and 0.050±0.01 µmol/mg protein / min with percent -68.86, - 77.84 and -70.06 %, respectively. In addition, α-glucosidase shows a value of 0.192± 0.01, 0.187±0.01 and 0.190± 0.01 µmole/ mg protein /min with percent of change -29.41, -31.25 and -30.15 % respectively. On the contrary, total protein content show insignificant change at the different duration post STZ injection.

Treatment of the diabetic rats with the different extracts of *Nepeta cataria* produce obvious enhancement in all carbohydrate hydrolyzing enzymes tested. This can be easily seen through normalization of α-amylase level to show insignificant change as a result of treatment diabetic rats with crude ethanol extract and as compared to either normal or diabetic glicalized treated groups (reference drug). While α-amylase shows significant increase post treatments with petroleum ether (52.90±6.22 µmole /mg protein/min) and chloroform extracts (40.94±2.2 µmole/mg protein/min) with percentage increase +62.72 and +25.93 % respectively. The curative effect of the different extracts can be also seen through improvement in β- galactosidase level that is returned to its normal value as compared to both normal control and reference drug. In addition α -glucosidase activity shows insignificant change post chloroform treatment, while it recorded significant increase post petroleum ether, 70% ethanol extracts and gliclazide drug(0.335±0.02, 0.357±0.02 and 0.372.±0.03 µmole/ mg protein / min, respectively) with percentage increase +23.16, +31.25 and +36.76 %, respectively as compared to normal control. Concerning total protein content, insignificant change is observed in all diabetic –treated groups except in petroleum ether extract treated- diabetic rats, significant reduction is obtained amounted 35.00 ±8.16 mg /g tissue used with percentage -30 % as compared to normal control.

Table (11) and Fig (11) illustrate body weight, liver weight and liver weight / body weight ratio in control, different normal –treated, diabetic and diabetic – treated groups. It is obvious that body weight of normal control rats recorded significant increase (181.00 ±2.58 gm) with concomitant increase in liver weight (6.90±0.297) at day 40 post experiment as compared to body weight and liver weight at zero time with percentage increase amounted +19.47 and +16.50 %, respectively. While, body weight and liver weight show insignificant change at other durations (2 and 10 days). Moreover normal liver weight/body weight ratio shows insignificant change at different durations as compared to zero time. On the other hand, body weight and liver weight /body weight ratio of control rats treated - chloroform, total ethanol and petroleum ether show insignificant change either as compared to normal control at 40 day post start of experiment or as compared to each other. Liver weight of normal control 70% ethanol extract treated rats –exhibits significant increase amounted 8.50 ±0.37 g with percentage reached to + 23.14 %.While it shows insignificant change in other treatments. It is clearly noticed from the present study that diabetic condition is always associated with a significant reduction in body weight. This result is ascertained through the degradable remarkable significant reduction in body weight at day 2, 10 and 40 post STZ – induce diabetes amounted 142.25±4.34, 112.75 ±8.77 and 98.50±5.68 g with percentage of reduction -5.79, - 28.86 and -45.58%, respectively. On the contrary, liver weight and liver weight / body weight ratio manipulated significant increase of 7.67 ± 0.87, 9.56±1.86 and 8.25±0.42 g with percent +26.96, +53.49 and +22.06 % for liver weight and 0.0525±0.005, 0.085±0.013 and 0.088±0.005 for liver weight / body weight ratio with percentage increase +31.25, +112.50 and +118.75 % at day 2, 10 and 40 post STZ injection, respectively. Treatment of diabetic rats with different extracts produced obvious improvement in body weight, liver weight and their ratios, while chloroform extract treated diabetic rats shows significant increase in body weight amounted 123.50 ±6.02 with percent increase +25.38 % as compared to untreated diabetic group at day 40 (recovery group), but with respect to normal control rats at day 40, significant reduction is recorded (-31.77%). Nearly the same results are achieved for 70% ethanol extract treated– diabetic rats. In addition, diabetic rats –treated petroleum ether extract and gliclazide drug show significant increase in body weight amounted 131.00±4.24 and 153.25 ± 2.36 g with percent +32.99 and +55.58 %, respectively as compared to diabetic - untreated one while as compared to normal control, significant reduction is

noticed (-27.62 and -15.33 %, respectively). Concerning liver weight, insignificant change is observed in chloroform extract treated- diabetic rats and gliclazide drug, while significant reduction is recorded in petroleum ether and total ethanol extracts amounted 5.00±0.47 and 5.26±0.50 g with percentage -27.56 and -23.83%, respectively as compared to normal control rats (40 days). Liver weight /body weight ratio exhibit insignificant change in all diabetic –treated groups except chloroform extract which shows significant increase as compared to normal control group at day 40 (0.055±0.02) with percentage of increase +37.5%. In addition significant reduction is observed in liver weight and liver weight / body weight ratio in all diabetic –treated groups as compared to diabetic untreated one(40 days).

III. Histological studies on pancreas and liver:

Normal architecture in liver and pancreas of treated –control groups at the cellular level as compared to the normal un-treated one. Gradual cellular changes include imperfection, reduction in β-cells counts, number of islet and atrophic changes are appears in pancreas of Streptozotocin - treated groups at different durations (figs 5-7 1st sheet) .On the other hand, sections of diabetic liver at day 2,10 and 40 show degeneration of hepatocytes, necrosis and congestion of central vein (5-7 2nd sheets).

Different successive extracts of *Nepeta cataria* and gliclazide drug (8-11) appear to regulate diabetes at the cellular level resulting in restoration of near normal architecture pancreatic islets of langerhans and hepatocytes in the Streptozocin treated -groups, this suggests a possible regeneration or repair of the cells in diabetic - treated groups.

DISCUSSION

Reactive oxygen species (ROS), superoxide anion, hydrogen peroxide and hydroxyl, nitric oxide and peroxynitrite radicals, play an important role in oxidative stress related to the pathogenesis of various important diseases. In healthy individuals, the production of free radicals is balanced by the antioxidative defense system **(Shyur *et al.*, 2005)**. Moreover knowledge and application of such potential antioxidant activities in reducing oxidative stresses ***in vitro*** has prompted many investigators to search for potent and cost –effective antioxidants from various plant sources **(Cesquini *et al*., 2003)** .

Scavenging activity for free radicals of 1,1 –diphenyl -2- picrylhydrazyl (DPPH) has been widely used ***in vitro*** to evaluate the antioxidant activity of natural products from plant and microbial sources. Crude ethanol extracts and successive extracts (petroleum ether, chloroform, ethyl acetate, ethanol as successive) were prepared for investigation of their antioxidant activities .The dosage of extract is expressed in µg of dry weight of the extract (compound) per ml of the assay mixture.

The present results indicated that crude extract (70% total ethanol extract) exhibited the strongest DPPH free radical inhibitory effect at a dose response relationship compared to other successive extracts followed by petroleum ether and chloroform. However, ethanol as successive showed the lowest scavenging activity at concentration of inhibitor 10, 50, 100, 500 and 1000 µg/ml. In a good agreement with the present results **Shyur *et al.*, (2005)** found *Prunella vulgaris* plant species (Labiatae) extracts dose dependently and significantly inhibited DPPH free radical activity. Moreover and in close agreement with the present data **Souri *et al.*, (2008)** attributed the potent antioxidant and DPPH radical scavenging activities of *Mentha spicata* (Labiatae) to wide variety of antioxidant constituents such as phenolics, ascorbate, carotenoide and terpenoides. **Tepe et al., (2007)** examined the antioxidant activity of essential oil and various extracts (hexan , dichlorometane and methanol sub-fractions) of *Nepeta flavida* (Labiatae) and found, among the extracts, the strongest activity was exhibited by polar sub-fraction of the methanol extract, that ascertained correlation between the polarity and antioxidant activity. In addition, the amount of total phenolics was highest in the polar and non polar sub-fractions. Particularly, a positive correlation was observed between the total phenolics content and the antioxidant activity of the extracts .The amount of phenolic compounds were less in hexan and dichloromethane extracts than in the others. However **Dasmalchi *et al.*, (2007)** used successive extracts of Moldavian balm (*Dracocephalum moldavica L. Labiatae*), petroleum ether, dichloromethane, acetonitrile, ethyle acetate, methanol, n- butanol and water. Results of the DPPH free radical scavenging activity suggested that components within the extracts (hydroxylated cinnamic acids, their derivative and flavonoids), except for the petrol and dichloromethane extracts are capable of scavenging free radicals via electron –or hydrogen –donating mechanisms and thus should be able to prevent the initiation of deleterious free radical mediated chain reactions in susceptible matrices as biological membranes. In accordance to the present findings **Tsai *et al.*, (2007)** reported that Labiatae family including rosemary , oregano, marjoram and thyme, revealed significant DPPH inhibitory activity and phenolic compounds were considered as the main contributors responsible for the free radical scavenging activity **(Tsai *et al.*, 2005).**

Nitric oxide synthase (NOS) catalyzes the production of nitric oxide (NO). Inducible nitric oxide synthase (iNOS) is expressed by vascular endothelial cells and smooth muscle cells in response to cytokines, unlike the two other types of NOS, which are

constitutive. NO produced by iNOS is implicated in inflammatory diseases **(Brinker *et al.*, 2007).**

The present results revealed that all successive extracts of *Nepeta cataria* were capable of suppressing NO activity with the most reducing abilities for crude ethanol extract. Petroleum ether was found to be came later after total ethanol extract (70%). Chloroform, ethyl acetate and ethanol extracts demonstrated a moderate reducing power at the different concentrations of inhibitors (10-1000ug/ml). **Tsai *et al.*, (2007)** reported that food and phytochemicals exerts NO –supressing activity via three different pathways: the blocking of iNOS expression, inactivation of iNOS catalytic function and the scavenging NO. While, NO suppressing effect primarily through regulation of cellular iNOS expression. In concomitant with the present results **Kim *et al.*, (1998)** found that Labiatae family exhibited strong suppressing activity upon NO production and thus provided further convincing evidence to illustrate, at least partially, the relative benefits of this family as anti -inflammation , anticancer or antioxidant. It seems that the extracts do not only exert NO-suppressing effect through direct scavenging of NO radicals but also through inhibition of NOS catalytic activity and /or suppression of iNOS expression **(Sheu *et al.*, 2001).** The complexity of the extract composition one could speculate that the relative contribution of the polyphenolic compounds or the triterpenes on the suppressing activity of NO production may be the result of the synergistic/ antagonist effects between different classes of compounds **(Diouf *et al.*, 2009)**. It has been found that the presence of a catechol group in a flvonoides is essential required for excellent NO-scavenging ability and that gallic acid linked to flavan -3-ol also plays an important role in the NO scavenging of catechines (Nakagawa and Yokozawa ,2002). Furthermore, the rather striking NO –suppressing activity of rosemary (Labiatae) has been documented according to one of its constituents, namely carnosol a compound which inhibits iNOS by blocking the activation of tumor necrosis factor β **(Manosroi *et al.*, 2006)**. Cinnamic aldehyde , a compound found to be present in, specifically, the essential oil cinnamon as well as several flavonoides have been reported to decrease the transcriptional activity of iNOS gene **(Diouf *et al.*, 2009).** Moreover , **(Dudhgaonkar *et al.*,2009)** demonstrated the suppression of the oxidative stress and inflammatory response related to diabetes through the inhibition of tumor necrosis factor β- (TNF- β) signaling. Natural triterpenes such as ginsenosid Rh1, triterpene isolated from *Panax ginseng,* inhibited iNOS expression and the activation of NF- β. In addition oleanolic acid glycoside isolated from Ampelopsis radix, markedly suppressed the activity of NF-β and production of NO.

One therapeutic approach for treating diabetes is to decrease the post –prandial hyperglycemia .This is done by retarding the absorption of glucose through the inhibition of the carbohydrate hydrolyzing enzymes α-amylase, α -glucosidase and β –galactosidase in the digestive tract. Inhibition of these enzymes delay carbohydrate digestion and prolong overall carbohydrate digestion time, causing a reduction in the rate of glucose absorption and consequently blunting the post prandial plasma glucose rise **(Rhabasa –Lhoret and Chiasson ,2004).** Many natural resources have been investigated with respect to the antidiabetic and suppression of glucose. The fact that α –glucosidase and α-amylase showed different inhibition kinetics seemed to be due to structural differences related to the origin of the enzymes **(Kim *et al.*, 2005)**. In this aspects successive as well as crude ethanol extracts of *Nepeta cataria* were used *in vitro* to demonstrate their efficiency on carbohydrate hydrolyzing enzymes. The present results revealed that crude ethanol extract exhibited the highest reducing activity of amylase at concentration of inhibitor 10 an 50 ug /ml (92.00±1.21 and 94.35±2.69%, respectively) while petroleum ether showed reducing activity of 18.26± 3.97 and 24.55± 4.03%, respectively at the same concentration of inhibitor, then chloroform, ethyl acetate and ethanol as successive . While at 1000 µg /ml, crude extract demonstrated reducing activity of 88.40± 7.29% compared to 44.59± 1.55, 43.64±1.79 and 37.97±1.86 % for chloroform, ethyl acetate and petroleum ether, respectively. Considering ethanol as successive extract, the lowest reducing activity was noticed. (**Kim et al., 2005)** demonstrated that the triterpenoid acids showed a significant inhibition effect on α-amylase. Moreover urosolic acid, pentacyclic triterpenoids and oleanolic acid derivatives exhibited the strongest α- amaylase suppressing activity and are responsible for a major part of the activity of the total hexan extract of many antidiabetic plants. (**Manosroi *et al.*, 2006)** and **(Brinker *et al.*, 2007)** attributed the anti-dibetic, anti-inflammatory anti-tumor and anti-proliferative effect of many Labiatae species, to their essential oil which are composed of mono, sesquiterpenes, phenolic compounds and flavonoides such as cinnamic acid, caffeic acid and rosmarinic acid. In a parallel results **(McCue and Shetty 2004)** illustrated that the amylase activity was inhibited in the presence of herbal extracts containing rosmarinic acid (RA-*Labiatae*) and the extent of amylase inhibition correlated with increased concentration of RA. The anti- amylase inhibitory activity may be due to the ability of phenolic compounds to interact with and / or inhibit proteins enzymes **(Rohn *et al.*, 2002)**. The same authors added that phenolic substances that are able to form quinines (such as caffeic acid, chlorogenic acid, gallic acid, etc) are more reactive than those phenolics

that cannot form quinines and suggested that semi-quinones formed may react with amino acid side chains and free thiol groups on the enzyme.

The present results showed remarkable suppressing effect of petroleum ether, chloroform and crude extracts greater than chloroform or ethanol as successive on carbohydrate hydrolyzing enzyme α-glucosidase at concentration of inhibitors 100 and 1000 µg /ml and these extracts demonstrated significant dose dependent enzyme inhibitory activities. In addition, the inhibitory activity of the various successive and crude extracts of *Nepeta cateria* on carbohydrate β-galactosidase hydrolyzing enzyme indicated that crude ethanol, petroleum ether and chloroform extracts possessed, respectively the most powerful reducing activity at the different concentration of inhibitors. While, ethyl acetate and ethanol extracts exhibited the lowest reducing effect. Linear relationship was demonstrated between reducing activity and concentration of inhibitors.

Although inflammation is important in preventing diseases, there are numerous autoimmune disorders that involved deleterious inflammatory response, including rheumatoid arthritis, psoriasis and Type 1 diabetes. It was found that triptolide, diterpenes, sesquiterpenes and triterpenes inhibited the transcription of proinflammatory genes by blocking the transactivation of NF-β so these compounds acts as antiiflammatory and autoimmune conditions **(Brinker *et al.*, 2007)**. On the other hand, (**Ali *et al.*, 2006)** demonstrated several compounds isolated from many antidibetic plants such as flavanone glycoside, thiosugar kotalanol and luteolin (flavonoid) and proved to have inhibitory effect on α-glucosidase. In close agreement, **(Tepa *et al.*, 2007)** and **(Dudhgaonkar *et al.*, 2009)** returned the antidiabetic or antiiflammatory effects of Nepeta species to a biatene diterpenes, triterpenes and iridoid glycosides. However, (**Bhandari *et al.*, 2008)** attributed the antidiabetic effect through inhibition of digestive enzyme α-glucosidase to the potential enzyme inhibitors -3- O-galloylepicatechin and -3-O- galloylcatechin extracted from *Nepalese herb Pakhanbhed*(Nepeta species).

The present research is also extended to evaluate the ***in vivo*** possible improved effect of the most potent successive *Nepeta cataria* extracts (petroleum ether and chloroform as well as 70% crude ethanol extracts) against liver disorders associated with diabetic complications in STZ diabetic rats compared to the currently available antidiabetic gliclazide (diamicron drug), especially with respect to liver function enzymes(AST, ALT, ALP and total protein) , lipid profile (total cholesterol, HDL cholesterol, LDL-cholesterol, triglycerides and total lipid), oxidative stress marker (NO), hepatic carbohydrate hydrolyzing enzymes (α-amylase, α – glucosidase and β-galactosidase). The histological architectures of liver and pancreas were also studied. Diabetes mellitus (DM) is a chronic disease caused by inherited or acquired deficiency in insulin secretion(IDDM) or by decreased responsiveness of the organs to secreted insulin(Non IDDM), resulting in increased blood glucose level, This in turn can damage many of the body's systems, including blood vessels, nerves and causes oxidative tissue damage **(Matsui *et al* ., 2007)**.

The potential role of the medicinal plants as hypoglycemic agents has been reviewed by several authors, supported by the ethnobotanical surveys and traditional medicines of different cultures **(Yeh *et al.*, 2003; Biesalski , 2004 ; Li *et al* ., 2004)**.

The current study has demonstrated insignificant change between normal control and different normal –treated groups (G1-G5) either in blood glucose or insulin levels except for normal rats treated with 70% ethanol extract, that recorded significant decrease in blood glucose level (13.69%). Injection of rats with Streptozotocin (STZ) (G6-G8) induced significant elevation in fasting blood glucose with concomitant reduction in insulin levels at day 2 ,10 and 40 post STZ treatment as compared to control untreated group(G1) (from109.5± 6.65 mg/dl to 373.00±2.94, 363.00±2.94 and 364.25±3.77 mg /dl, respectively for glucose and 5.35µIU/ml to 0.59 ±0.01, 0.34±0.05 and 0.35±0.01 µIU/ml for insulin, respectively).

The present histological examinations at the cellular level, revealed atrophy, necrosis and degenerative changes in both hepatocytes and β-cells of pancreas, indicating establishment of diabetic state (Figs,5-7 liver and pancreas). **(Holemans *et al.*, 1997)** that, Streptozotocin induced beta cells destruction by necrosis. STZ is an antibiotic and anticancer agent which is widely used for inducing diabetes (Type 1 IDDM) in a variety of animals. It interferes with cellular metabolic oxidative mechanisms **(Bagri *et al.*, 2008)**. It selectively induces degenerative alterations and necrosis of pancreatic β-cells resulting in insulin deficiency and impairment in glucose oxidation **(DeCarvalho *et al.*, 2005)**. In the present study the change in the shape of pancreas cells can be attributed to partial damage of Streptozotocin to some beta cells. **(Ikebukuro *et al.*, 2002)** have reported that use of lower dose of Streptozotocin produced an incomplete destruction of pancreatic beta cells even though rats became permanently diabetic. In accordance to the present study (Figs 5-7 liver and pancreas), **(Mitra *et al.*,1996)** earlier reported that, the diabetic liver showed degeneration and congestation two hours after injection of STZ, hyperglycemia is observed with a concomitant drop in blood insulin level. The changes in blood glucose and insulin concentrations reflect abnormalities

in beta cell functions. The fluctuation in the blood sugar might also be attributed to the sensitivity to STZ that varies with species, strain, sex and nutritional state and there are batch differences in activity **(Mir *et al.*, 2008).** In a good agreement with the present results, **(Akbarzadeh *et al.*, 2007)** confirmed the destruction of islet cells in pancreatic biopsy of diabetic rats due to the effect of Streptozotocin and added that 60 mg /kg dose of STZ ensured induction of diabetes in rats and hyperglycemia, hypoinsulinemia, polyphagia, polyuria and polydipsia were seen in adult rats within 3 days of STZ treatment and the amounts of the relevant factors were almost stable , which indicates irreversible destruction of langerhans islets cells. Previous studies have reported that Streptozotocin enters the beta cells via a glucose transporter and causes alkylation of DNA. DNA damage induces activation of poly ADP ribosylation, a process that is more important for the diabetogenecity of Streptozotocin than DNA damage itself. Poly ADP- ribosylation leads to depletion of cellular NAD and ATP. Enhanced ATP dephosphorylation after Streptozotocin treatment supplies a substrate for xanthine oxidase resulting in the formation of super oxide radicals. Consequantly, hydrogen peroxide and hydroxyl radicals are generated. Furthermore Streptozotocin liberates toxic amounts of nitric oxide that inhibits aconitase activity and participates in DNA damage **(Fiordaliso *et al.*, 2000)**.

In diabetes state, degradation of liver glycogen and gluconeogensis are increased while glucose utilization is inhibited. Glucose -6-phosphatase increases in the liver, facilitating glucose release into the blood. The opposing enzyme which phosphorlyates glucose, i.e hexokinase, is unaffected by insulin while glucokinase is decreased in diabetes. As a result, the liver continues to produce glucose even with severe hyperglycemia. Under these circumstances the normal liver would shut off and deposit glycogen **(Shelila and james, 1993).**

The present results revealed significant amelioration in blood glucose and insulin levels post treatment of diabetic rats with the crude ethanol, petroleum ether and chloroform extracts of *Nepeta cataria* with percent of improvement amounted 239.68, 215.29 and 211.19 % for glucose and 79.25, 70.65 and 65.05% for insulin, respectively. The current investigation also showed that treatment of diabetic rats with gliclazide (reference drug) modulated the alterations in blood glucose and insulin within its normal levels (212.10 and 65.79%, respectively). In addition, the histological examination showed improvement in hepatocytes and pancreas β- cells (Figs 8-11 liver and pancreas). In line with the present study (**Vats *et al.*, 2004)** found that the crude ethanol extract of *Ocimum sanctum*(Labiatae) leaves significantly improve β–cells function and enhances insulin secretion leading to lowering blood glucose level. These authors added that the antihyperglycemic effect of *Ocimum sanctum* is at least partially dependent upon insulin release from the pancreas and significantly increased the activity of three key enzymes involved in carbohydrate metabolism, namely phosphofructokinase, glucokinase and hexokinase (PFK, GK, HK) towards normal levels. In addition it increases glycogen in muscle and liver by stimulating glycogen synthase, suggesting that the antihyperglycemic action is the result of an increased glucose utilization at the level of skeletal muscle as well as liver. The authors added that inhibition of disaccharide enzymes sucrose and maltase seems to be one of the factors which explain the hypoglycemic action of many antidiabetic plant extracts. Immune-stimulation also might be one of the mechanisms contributing towards the protective actions of *Ocimum sanctum* **(Sembulingam *et al.*, 2005)**. In addition, (**Zheng *et al.*, 2007)** studied the hypoglycemic effect of the aqueous ethanol extract of *Prunella vulgaris L.* (Labiatae) in STZ diabetic mice and attributed the hypoglycemic effect to increase the insulin sensitivity of target tissues and the antihyperglycemic effect of insulin is enhanced and prolonged, which could result from increased tissue metabolism or from suppressed levels of non- esterified fatty acids (NEFA). Excess plasma NEFA can inhibit insulin – stimulated glucose utilization in muscle and promote hepatic production of glucose .Whereas, reduction of plasma NEFA concentration improves glucose utilization and enhances the suppression of hepatic glucose production by insulin. Stimulation of glucose uptake by peripheral tissues and inhibition of endogenous glucose production may be involved in hypoglycemic mechanisms of Labiatae family. Some constituents in the *Prunella vulgaris L.* has been identified such as phenolic acids (rosmarinic, caffeic), triterpenoids (methyl oleanolate, methyl ursolate, methyl maslinate), flavonoides (quercetin, campherol, rutin), tannis and polysaccharide . The antihyperglycemic activity of *Prunella vulgaris L.* (Labiatae) may be due to any one or more of the constituents in the extracts. At this basis, The hypoglycemic action of the crude ethanol, petroleum and chloroform extracts of *Nepeta cateria* may be insulin –mediated by mechanisms in common with gliclazide. Another explanation for the hypoglycemic action of *Nepeta cataria* extracts is based on the presence of antioxidants such as flavonoides which may prevent the progressive impairment of pancreatic beta-cell function due to oxidative stress and may thus reduce the occurrence of type 2 diabetes **(Li, *et al.*, 2004; Bhandari *et al.*, 2008).** *Nepeta cataria* was found to possess relatively high percentage of essential oil (nerol, geraniol and citral as well as ursolic acid), and polyphenols

(favonoids, phenolic acid) and the antihyperglycemic, antisplasmodic and myorelaxant effects may be related to these constituents **(Gilani *et al.*, 2009)**.

With respect to lipid profile, the current results demonstrated insignificant change in total cholesterol, HDL-cholesterol, LDL-cholesterol and total lipid in different normal- treated groups, with significant reduction in triglycerides in normal - crude ethanol treated group. Meanwhile, significant elevation in lipid profile was noticed at day 2, 10 and 40 post STZ injections. In addition to abnormal glucose metabolism, DM often involves abnormal lipid metabolism which is considered an additional metabolic disorder, in diabetic complications. The same results were achieved by (**Sethi *et al.*, 2004)** who found significant elevation in lipid profile in serum of diabetic rats. In a good agreement with the present data, some authors revealed that hyperglycemia produced marked increased level of serum triglycerides, total –cholesterol and LDL- cholesterol (LDL-C), while in contrast to the present data HDL-cholesterol (HDL-C) showed reduced concentration in diabetic rats **(Abou –Seif and Youssef ,2004 ; Jurgonski *et al.*, 2008)**. Levinthal and **(Tavill, 1999)** reported that, hepatic fat accumulation is a well –recognized complication of DM. The most common clinical presentation in DM is hepatomegaly. This hyperlipidemia associated with DM may be attributed to insulin deficiency **(Morel and Chisolm, 1989)** and elevated cortisol level, which have an important role in the process of fat accumulation **(Hristova and Aloe, 2006).** Under normal circumstances insulin activates lipoprotein lipase which hydrolyzes triglycerides. Insulin deficiency results in failure to activate the enzyme, thereby causing hyper-triglyceridemia **(Shirwaikar *et al.*, 2004).** On the other hand, in insulin deficiency, the plasma free fatty acids concentration is elevated as a result of increased free fatty acids outflow from fat depots, where balance of the free fatty acids estrification, triglycerides lipolysis is displaced in favour of lipolysis **(Shirwaikar *et al.*, 2004)**. Also elevated cortisol promotes liberation of free fatty acids from adipose tissue into blood stream by inducing and maintaining synthesis of the hormone sensitive lipase, thus increasing free fatty acids level which contribute to cardio-vascular risk **(Lundberg, 2005).**

The elevation in cardioprotective HDL-C means increase of cholesterol afflux from the tissues, the first step in reverse cholesterol transport from the peripheral tissues to the liver. The antioxidant and antiatherogenic activities of HDL-C are enhanced when its circulating level is increased. LDL- C particles become small and dense which undergo oxidative modification, thus leading to a diabetic complication **(Kalousova *et al.*, 2002)**. In addition, **(Mir *et al.*, 2008)** reported hypercholesterolemia, hypertriglyceridemia associated with DM and explained these increments at the basis of Streptozotocin induced diabetes. There is excess of fatty acids in the serum , which promotes conversion of excess fatty acids into phospholipids and cholesterol in liver. These two substances along with excess triglycerides formed at the same time in liver may be discharged into blood in the form of lipoproteins.

In parallel results **(Mir *et al.*, 2008)** found high concentration of total lipid in serum of diabetic rabbits and attributed this elevation mainly to increased mobilization of free fatty acids from the peripheral fat depots, since insulin inhibits the hormone sensitive lipase.

Significant amelioration in lipid indices was observed after treatment of diabetic rats with the test extracts with percentage of amelioration amounting to 71.51, 62.13 , 67.75 %, respectively for total cholesterol(as compared to 127.15% in gliclazide-treated diabetic group). HDL-cholesterol recorded enhanced level of 41.02, 39.35, 43.00 %, respectively as compared to 44.00% for gliclazide. The improved level of LDL- cholesterol reached to 122.73, 110.64, 118.72 %, respectively (153.74 %for gliclazide). Triglycerides showed normalized level of 165.05, 169.18, 176.77% respectively and 166.17 % for gliclazide. Total lipid recorded percent of improvement amounted to 40.19, 44.42, 49.33%, respectively (58.25% for gliclazide). In concomitant with the present results **El-Hilaly and Lyoussi 2002); Brinker *et a*l.,2007)** found relative high percentage of essential oil with nerol, geraniol and citrol as well as ursolic acid, polyphenols (flavonoids , phenolic acid) and steroids in *Nepeta cataria* (Lbiatae) which may related to hypolipidemic and cholesterol –lowering effect. The mechanism(s) of hypolipidemic effect of many medicinal plants such as *Ajuga iva (L.) Schreber L.* (Liabiatae) was mediated through insulin –enhancing lipolytic activity by inhibition of hormone –sensitive lipase **(Al- Shamaony *et al.*, 1994)** or lipogenic enzymes **(Pari and Venkates, 2004)** , and / or activation of lipoprotein lipase **(Ahmed *et al.*, 2001)**. In addition, the hypolidimic lowering effect may be related to several active constituents extracted such as diglycerides , ecdysones , ecdysterones , iridoides , phenylcarboxylic acids , steroid compounds which is considered as anti-inflammatory agents **(Brinker *et al.*, 2007)** , thus the mechanism(s) of action of such family (Labiatae) as antihyperlipidemia may involve insulin –like effect **(Khushbaktova *et al*., 2001).**

However **(Sethi *et al.*, 2004)** found that, leaves of *Ocimum sanctum* (Labiatae) significantly reduce lipid profile in serum and tissue in normal and diabetic rats through inhibition of oxygen free radical incorporated in pathogenesis of diabetes and

enhancement of cellular enzymatic (SOD) and non enzymatic antioxidants (GSH).

The present results demonstrated, insignificant change in AST level in serum of normal treated rats either with chloroform or total ethanol extracts, while significant inhibition was noticed with petroleum ether extract. Serum total protein content and ALT showed insignificant change in different normal –treated groups. Considering ALP, significant inhibition was noticed in all normal treated groups. With respect to different diabetic- groups, high serum levels of these enzymes at day 2, 10 and 40 post STZ treatment was recorded which are associated with inflammation and /or injury to liver cells , a condition known as hepatocellular liver injury and apopotosis Fiordaliso *et al.*(2000). Histological examination of diabetic rat liver showed congestion, destruction of cells by necrosis and hepatocytes degeneration. In parallel with the present work, Hickman *et al.* (2008) revealed significant increased activities of serum enzymes relative to their normal levels. Supporting our findings, it has been found that hyperglycemia resulted in hepatolysis reflected by histopathological investigation and increased blood serum aminotransferase as one of the consequences of diabetic complication. The increment of such serum markers may be due to the leakage of these enzymes from the liver cytosol into the blood stream as a result of hepatomegaly (fatty liver) **(Muhammad *et al.*, 2008)**.

The present results revealed also, insignificant change in serum total protein content at day 2, 10 post STZ injection and significant reduction at day 40. In concomitant with the present results (**Otsuki and Williams, 1982)** found significant reduction in serum total protein concentrations in diabetic rats and this may be due to reduction in the three major phases in protein secretion, intracellular transport and discharge. Also, **(Alderson *et al.*, 2004)** demonstrated a significant increase of total protein excretion, albuminuria, glucosuria and urinary urea levels indicating impaired renal function. The reduction in serum total protein content in the present results may be related to reduction in albumin which is the most abundant blood plasma protein (70%) produced in liver. Non –enzymatic glycation of albumin was found the potential to alter its biological structure and function **(Mendez *et al.*, 2005).** It is mainly due to the formation of a Schiff base between amino – group of lysine (and sometimes arginine) residues and excess glucose molecules in blood to form glycoalbumin. Elevated glycoalbumin was observed in diabetes mellitus accompanied by decrease in albumin and this is confirmed with early studies suggested that the level of glycosylated albumin may indeed be a sensitive indicator of moderate hyperglycemia and of early glucose intolerance **(Miwa *et al.*, 2005)**. Hypoalbuminemia is one of the factors responsible for the onset of ascites related to liver fibrosis **(Horie *et al.*, 1998)**.

Significant improvement in liver function enzyme markers was noticed in treatment of diabetic rats with petroleum ether , chloroform as well as crude ethanol extracts with percent of amelioration amounted to 42.48, 39.38, 42.92%, respectively for AST as compared to 37.17% for gliclazide .While ALT recorded 55.63, 47.5, 55.63 %, respectively (58.13 % for reference drug). ALP showed ameliorated level amounted to 94.39, 101.18, 91.15% respectively as well as 100.88% for gliclazide drug. It was shown that administration of successive extracts and crude ethanol one to diabetic rats reflect an improvement of cellular damage as determined in the current research by histopathological examination of liver and pancreas (Figs 8-11) and as shown by normalization of altered liver enzymes in response to diabetic complications. Our results are consistent with previous studies that administration of some antioxidants (as zinc , selenium , vitamin C and E) to diabetic rats , normalized the elevated activities of liver function enzymes AST, ALT, ALP induced in response to diabetes mellitus **(Abdel Mageed, 2005).** The mechanism of hepatoprotective ability of extracts may be attributed to numerous bioactive compoundas such as terpenoids, flavonoids, sterols, essential oil, alkaloids and polysaccharides. Most of them (especially falvonoids, triterpenoids such as ursolic acid) showed a mechanism to improve the function of liver and pancreas cells and hence normalization of liver enzymes **(El Hilaly and Lyoussi , 2002; Li *et al.*, 2004 ; Zheng *et al*. 2007 ; Gilani *et al*., 2009)**.

Significant decrease was observed in serum total protein content in different–diabetic -treated groups as compared to both normal control and gliclazide -treated diabetic group .In contrast, (**Otsuki and Williams 1982); Sethi *et al.*, 2004)** demonstrated enhanced level of serum proteins post treatment of diabetic rats with aqueous extract of *Ocimum sanctum* (Labiatae) and attributed this effect to insulin –like factors contained in the extract , since insulin is reported to increase protein synthesis. In addition, the total thiols in Labiatae family play a vital role in the structure , activity and transport function of proteins , membranes and enzymes.

With respect to oxidative stress marker NO, insignificant change was observed in normal control treated group as compared to untreated one, while a significant increase is noticed in various diabetic groups. It was reported that NO over production has been linked to a variety of clinical inflammatory diseases **(Kim *et al*., 2002)**. Experimental studies suggested that NO may be responsible for increased liver injury **(Ma *et al*., 1995)**. The direct toxicity of NO

is enhanced by reacting with superoxide radical to give powerful secondary toxic oxidizing species, such as peroxynitrite (ONOO) which is capable of oxidizing cellular structure and causes lipid peroxidation **(Sayed Ahmed *et al.*, 2001)**, a process leading to membrane damage and considered the proximal cause of cell death . Lipids peroxidation can damage protein , lipid , carbohydrates and nucleic acids . Also it has been found that lipid peroxidation is one of the risk factor of protein glycation. The present results indicated significant elevation in NO in liver of diabetic rats. This increment may be due to oxidative stress which is considered as one of the necessary causative factors that link diabetes with the pathogenic complications of several tissues **(Anwar and Meki , 2003)** .

Significant improvement in NO level post various treatments with percent amounted 146.15, 176.92 , 169.23% for chloroform, petroleum and crude ethanol extracts respectively and 169.23% for gliclazide . In a good agreement with the present research , **(Sethi *et al*. 2004)** ; (**Vats *et al*., 2004)** found that treatment of diabetic rats either with aqueous or ethanolic extracts of *Ocimum sanctum* (OS -Labiatae) significantly increased activity of two antioxidant enzymes in liver namely , superoxide dismutase (SOD) and catalase . The protective effect of the plant extracts can be brought about directly by scavenging free radicals or indirectly by elevating glutathione levels (GSH). GSH protects the cell against oxidative stress by reacting with peroxiodes and hydroperoxides . SOD detoxifies superoxide radicals and converts them to H_2O_2 which is further converted to H_2O by catalase. Thus, the antihyperglycemic activity of OS supplemented with its adaptogenic and antioxidant activity will be an ideal multi-prolonged treatment for managing diabetes as it will target the stress, catabolism and glycemic effects associated with disease. Moreover, **(Sembulingam *et al*. 2005)** ascertained the reduction in the NO level by the component ursolic acid separated from OS. Essential oil in many species in the Labiatae family are composed of mono –and – sesquiterpenes in addition to phenolic compound and favonoids such as cinnamic acid , caffeic acid , sinapic acid , ferulic acid and rosmarinic acid have also been reported as antioxidants , free radical scavengers and metal chelators **(Manosroi *et al*.,2006)**. In this context , **Tepa *et al*., 2007)** proved that *Nepeta flavida(Nepeta species)* essential oils have various biological effects , including antioxidant activity due to the presence of 1,8-cineole , phenolic compounds especially terpenoids and phenolic acids. Furthermore, **(Souri *et al*., 2008)** found that, different antioxidant and radical scavenging activity of several array of medicinal plants may partly be due to wide variety of antioxidant constituents such as phenolics, ascorbate and carotenoids .

Concerning carbohydrate hydrolyzing enzymes , ά- amylase , ά- glucosidase , β-galactosidase as well as liver total protein content ,insignificant change was observed in ά- amylase, β-galactosidase post different normal -treated groups as compared to untreated one . While significant increase in ά- glucosidase post treatment of normal rats with petroleum ether and total ethanol extracts. Liver total protein content exhibited significant reduction in all normal –treated groups. Significant inhibition of all carbohydrate hydrolyzing enzymes was noticed in diabetic rats at the different durations post STZ treatment. However total protein content showed insignificant change. **(Messer and Dean 1975)** reported that liver and serum amylases are immunological identity and both are very similar to parotid gland and their differences from pancreatic amylase strengthens previous suggestion that liver is the main source of serum amylase and , further , eliminates the possibility of the pancreas being a source . In the same context, **(Terada and Nakanuma 1995)** found pancreatic enzymes in bile ducts and hepatocytes due to common cell lineage. The results of present study in agreement with the previous reports that indicated a decreased in pancreatic amylase activity in diabetic rats **(Otsuki and Williams, 1982).** Moreover, the pancreatic content of ribonuclease is also significantly reduced in diabetic acini's. The fall in amylase content either in pancreas or in liver is may be due to increased secretion or intracellular degradation in vivo and a decreased rate of synthesis. In addition the reduction in amylase content is paralleled by a change in specific messenger RNA content suggesting that insulin regulates the synthesis of amylase at the level of transcription **(Otsuki and Williams, 1982). Kim *et al*., 1990)** has reported that STZ induced diabetes resulted in reductions in glandular contents of DNA, RNA and amylase protein . However, the changes in amylase protein and its mRNA levels did not exactly parallel each other during diabetogenesis or subsequent insulin treatment. One reason for this discrepancy might be related to fluctuations in glands contents of amylase protein due to variations in the secretary activity of the glands in diabetic rats. The possibilities exists that the lower level of parotid amylase was related to an elevated rate of secretion due to increased mastication associated with hyperphagia in diabetic rats **(Anderson, 1983).** However, it is unlikely that the level of a secretary protein (amylase) mRNA will be reduced in glands with an increased secretary activity. The increase in secretary activity is likely to affect the glandular levels of all secretary proteins equally. Furthermore, **(Roy *et al*., 2005)** found a decrease in amylase level in liver, parotid glands and pancreas during STZ induced diabetes and this is due to a decrease in the gene expression of amylase RNA.

With regard to α-glucosidase and β-glactosidase in liver of diabetic rat, significant inhibition was demonstrated that is parallel with the results of **(Otsuki and Williams, 1982)** who noticed reduced maximal amounts of digestive enzymes released from acini of diabetic rats and explained this inhibition reduced secretary capacity, alterations in nutritional or other hormones states, a decrease of secretagogues or a combination of these factors.

Significant improvement in all carbohydrate hydrolyzing enzymes post chloroform, petroleum ether as well as crude ethanol extracts amounted 69.58, 106.37, 48.60%, respectively for amylase (as compared to 59.30 % for gliclazide); 33.82, 53.31, 61.39 % respectively for α- glucosidase comparing to 66.91% in gliclazide- treated diabetic group and 88.02, 68.88, 59.88% respectively for β-galactosidase (77.84% for gliclazide). The enhanced levels of carbohydrate hydrolyzing enzymes α-amylase, α-glucosidase and β-galactosidase may be related to the phenolic compounds inhibited the disaccharide enzyme activities as mucosal sucrase and maltase .The inhibition of glycolytic activity of brush border enzymes by polyphenolic compounds seems to be one of the factors which explain the discussed hypoglycemic action of *Nepeta cataria* **(Jurgonski *et al*., 2008).** The improvement in the level of carbohydrate metabolizing enzymes can be also explained at the basis of *Nepeta cataria* extracts contained fluctuated level of flavonoids . Flavonoids , like antioxidants may prevent the progressive impairment of pancreatic beta cell function due to oxidative stress and may thus reduce the occurrence of type 2 diabetes **(Bhandari *et al*., 2008).**

The present results demonstrated insignificant change in normal -treated body weight, liver weight (except for liver weight treated with crude ethanol extract which recorded significant increase as compared to normal control at day 40), and liver weight/ body weight ratio post treatment with chloroform ,petroleum ether and 70% ethanol extracts. Gradual significant reduction is noticed in body weight with concomitant increase in liver weight and liver weight / body weight ratio at day 2, 10 and 40 post STZ injection .In concomitant with the present results several authors **(Vats *et al*., 2004; Akbarzadeh *et al*. ,2007;Mir *et al*. ,2008)** reported that diabetes state is usually accompanied by weight loss and increase in liver weight were seen in adult rats within three days of streptozotocin induction. The literature regarding the effect of diabetes on liver weight is contradictory as some workers have shown an increase in hepatic weight in animals (Chen and **Ianuzzo , 1982 ;Sadique *et al*., 1987)** as well as human **(Van Lancker , 1976)** while others have reported no change **(Gupta *et al.*, 1999)**. Exact reasons of hepatic hypertrophy are not known, however fat deposition has been proposed to be the cause. The pattern of increase in liver weight /body weight ratio was manifested by the reduction in body weight and increase in liver weight of diabetic rats **(Vats *et a*l., 2004).**

Significant amelioration in body weight, liver weight and liver weight /body weight ratio post treatment of diabetic rats with chloroform , petroleum ether and 70% ethanol extracts as well as gliclazide as reference drug amounted 13.81, 13.95 , 17.96, 30.25%, respectively for body weight 20.54, 49.62, 45.89, 30.79% respectively for liver weight and 81.25, 112.5, 112.5 , 118.75% respectively for liver weight /body weight ratio .The enhancement in body weight may be attributed to anabolic action of ecdysones and ecdysterones found in Lbiatae family (EL Hilaly and Lyoussi, 2002). In concomitant with the present results **(Vats *et al*., 2004)** and Sembulingam *et al*.(2005) reported that *Ocimum sanctum* treated rats showed higher and significant gain in body weight in comparison to diabetic controls but was lower than in the normal controls. These authors added that ethanol extracts of *Ocimum sanctum* prevents increase in organ weight due to the protective action of urosolic acid concerned with free radical inhibition. Based on this findings, the ameliorative effect of *Nepeta cataria* may be due to various investigated phytochemicals compounds that can ameliorate physiological response to stress **(Gilani *et al*.,2009).**

In conclusion, the present study demonstrated in *vitro analysis* that, different successive extracts of *Nepeta cataria* exhibited inhibitory effect on oxidative stress indices (NO and DPPH) and carbohydrate hydrolyzing enzymes (α-amlase, α-glucosidase and β-galactosidase) in linear relationships to some extent with concentration of inhibitors (dose dependant) .Total ethanol, petroleum ether and chloroform extracts showed respectively the most potent reducing power , while ethyl acetate and ethanol soxhlet appeared moderate or low reducing activity. The present research was extended also to evaluate the *in vivo* anti-glycemic, antioxidant, antilipidemic effects of chloroform, petroleum ether and crude ethanol extracts in comparison with gliclazide as reference antidiabetic drug. The present data revealed that these extracts have significant beneficial glycemic control, scavenging free radical, normalized liver function, inhibited lipid synthesis associated with diabetic complication, as well as they have principle role in treatment and amelioration liver damage at the cellular level caused by hyperglycemia. Thus, the safely promising therapeutic dose used in the current study, can be effective in treatment and enhanced liver tissue from the damage induced by diabetes and may candidate as natural antidiabetic drugs.

Table (1) Percent of DPPH Inhibition in different successive extracts and 70% ethanol extract of *Nepeta cataria* plant

Groups / Concentrations	Petroleum ether extract	Chloroform extract	Ethyl acetate extract	Ethanol extract	70% Ethanol extract
10 µg/mL	26.31 ± 0.50 d	16.39± 4.63 d	11.75 ± 5.57 d	10.85 ± 5.42 c	47.7 ± 0.84 e
50 µg/mL	26.00 ± 1.38 d	22.38 ± 1.23 c	12.08 ±1.60 d	10.77 ± 2.09 c	60.53 ± 0.50 d
100 µg/mL	34.04 ± 0.94 c	32.54 ± 2.14 b	18.47± 0.19 c	18.09± 1.13 b	66.45 ± 0.29 c
500 µg/mL	37.39± 1.10 b	35.94 ± 1.42 ab	26.01 ± 2.84 b	22.81 ± 1.81 b	69.52 ± 0.97 b
1000 µg/mL	40.17 ± 0.9 a	38.48 ± 0.70 a	33.29± 2.21 a	30.64 ± 0.60 a	72.19 ± 0.69 a
LSD 5 %	1.83	4.46	5.55	5.06	1.26

DPPH is expressed in µg /ml; Data are mean ±SD of 3 replicates.
Statistical analysis is carried out using one way analysis of variance (ANOVA) using CoStat computer program.
Unshared superscript letters between treatments are significance values at P<0.001

Table (2) Inhibition percent of nitric oxide (NO) in different successive extracts and 70% ethanol extract of *Nepeta cataria* plant

Groups / Concentrations	Petroleum ether extract	Chloroform extract	Ethyl acetate extract	Ethanol extract	70% Ethanol extract
10 µg/mL	15.26 ± 6.94 d	13.50 ± 5.53 d	12.38 ± 6.22 d	10.30 ± 4.29 c	37.60 ± 1.68 e
50 µg/mL	24.94 ± 2.89 c	21.41 ± 4.42 c	18.57 ± 6.11 cd	16.92 ± 3.25 c	39.16 ± 9.31 d
100 µg/mL	33.30 ± 2.65 bc	27.80 ± 4.32 bc	26.76 ± 3.20 bc	21.95 ± 2.86 b	52.13 ± 6.06 c
500 µg/mL	36.29 ± 5.18 ab	30.32± 2.74 b	27.32 ± 2.52 b	23.59 ± 1.01 b	56.58 ± 3.46 b
1000 µg/mL	44.64 ± 4.29 a	41.24 ± 3.27 a	38.17 ± 3.01 a	36.14 ± 2.93 a	53.85 ± 5.12 a
LSD 5%	8.49	7.59	5.55	5.06	1.28

Nitric oxide (NO) is expressed in µg /ml; Data are mean ±SD of 3 replicates.
Statistical analysis is carried out using one way analysis of variance (ANOVA) using CoStat computer program.
Unshared superscript letters between treatments are significance values at P<0.001

Table (3) Percent of α-amylase inhibition in different successive extracts and 70% ethanol extract of *Nepeta cataria* plant

Groups / Concentrations	Petroleum ether extract	Chloroform extract	Ethyl acetate extract	Ethanol extract	70% Ethanol extract
10 µg/mL	18.26 ± 3.97 d	15.44 ± 2.68 e	12.17± 3.38 e	7.41 ± 2.61 e	92.00 ± 1.21
50 µg/mL	24.55± 4.03 c	21.72 ± 3.20 d	20.19 ± 0.57 d	18.75 ± 2.04 d	94.35 ± 2.69
100 µg/mL	32.56 ± 2.07 b	27.38 ± 1.73 c	24.57 ± 2.18 c	24.27 ± 3.07 c	85.22 ± 4.92
500 µg/mL	37.14 ± 0.89 ab	33.75 ± 2.31 b	31.21 ± 1.02 b	29.05± 1.47 b	89.38 ± 8.22
1000 µg/mL	37.97 ± 1.86 a	44.59 ±1.55 a	43.64 ± 1.79 a	38.13± 0.64 a	88.40 ± 7.29
LSD 5%	5.18	4.32	3.71	3.9	NS

α-amylase is expressed in µg /ml; Data are mean ±SD of 3 replicates.
Statistical analysis is carried out using one way analysis of variance (ANOVA) using CoStat computer program.
Unshared superscript letters between treatments are significance values at P<0.001

Table (4) Percent of α-glucosidase inhibition in different successive extracts and 70% ethanol extract of *Nepeta cataria* plant

Groups / Concentrations	Petroleum ether extract	Chloroform extract	Ethyl acetate extract	Ethanol extract	70% Ethanol extract
10 µg/mL	25.45± 4.04 c	23.47± 4.56 c	9.16 ± 2.99 d	8.06± 3.51 c	28.05± 1.63 c
50 µg/mL	33.91 ± 3.15 b	33.49± 1.76 b	23.60 ± 1.51 c	21.08± 4.68 b	37.80 ± 2.55 b
100 µg/mL	40.44± 4.34 a	40.55± 3.08 a	32.22 ± 3.96 a	28.95 ±1.44 a	44.21± 2.87 a
500 µg/mL	37.42± 1.14 ab	36.01 ±1..78 ab	27.13± 3.24 bc	27.24± 3.45 a	41.19± 1.59 ab
1000 µg/mL	39.32± 2.60 ab	38.11 ± 2.70 ab	29.42± 0.62 ab	27.65± 2.37 a	42.5± 2.24 a
LSD 5%	5.93	5.93	5.01	5.97	4.14

β- galactosidase is expressed in µg /ml; Data are mean ±SD of 3 replicates.
Statistical analysis is carried out using one way analysis of variance (ANOVA) using CoStat computer program.
Unshared superscript letters between treatments are significance values at P<0.001

Table (5) Percent of β- galactosidase inhibition in different successive extracts and 70% ethanol extract of *Nepeta cataria* plant

Groups / Concentrations	Petroleum ether extract	Chloroform extract	Ethyl acetate extract	Ethanol extract	70% Ethanol extract
10 µg/mL	23.09 ± 8.33 b	17.24 ± 6.39 c	5.47± 5.73 c	11.45 ±11.54 b	51.35± 5.24 b
50 µg/mL	34.78 ±11.03 b	35.35± 9.72 b	33.19 ± 18.04 ab	31.58 ± 22.03 ab	39.62± 8.53 bc
100 µg/mL	36.49 ±8.35 b	33.53 ± 11.38 b	24.12 ±16.46 bc	27.37 ±13.18 ab	34.49± 11.82 c
500 µg/mL	62.63 ± 1.89 a	54.13 ± 2.44 a	45.89± 4.91 a	46.58± 8.22 a	67.82 ±3.94 a
1000 µg/mL	62.12± 4.37 a	55.08± 5.11 a	43.77± 4.57 ab	42.21± 5.28 a	66.86± 3.79 a
LSD 5%	13.7	14.02	21.13	24.24	13.26

β- galactosidase is expressed in µg /ml; Data are mean ±SD of 3 replicates.
Statistical analysis is carried out using one way analysis of variance (ANOVA) using CoStat computer program.
Unshared superscript letters between treatments are significance values at P<0.001

Table (6): Evaleuation of various successive extracts and 70% ethanol extract of *Nepeta cataria* on blood glucose and insulin levels in control, normal treated, diabetic and diabetic-treated groups

Treatments	Glucose (mg/dL)	Insulin (uIU/ml)

Parameters		
Normal control	109.5 ±6.65 de	5.35± 0.62 a
Normal treated chloroform extract	105.25± 7.36 de	5.21 ±1.07 ab
Normal treated 70% ethanol extract	94.50 ±16.05 f	5.07 ±1.47 ab
Normaltreated petroleum ether extract	113.75 ±1.25 d	5.03 ±1.66 abc
Diabetes after 2 days	373.00± 2.94 a	0.59± 0.01 e
Diabetes after 10 days	363.00± 2.94 b	0.34± 0.05 e
Diabetes after 40 days	364.25± 3.77 ab	0.35 ±0.01 e
Diabetes treated chloroform extract	133.00± 5.35 c	3.83 ±0.59 d
Diabetes treated 70% ethanol extract	103.00± 2.44 ef	4.59± 0.59 abcd
Diabetes treated petroleum ether extract	128.25± 2.87 c	4.13± 0.57 bcd
Diabetes treated gliclazide (Ref.Drug)	132.00± 2.44 c	3.87± 0.05cd
LSD 5%	9.08	1.19

Table (7): Evaluation of different successive extracts as well as 70% ethanol extract of *Nepeta cataria* on lipid profile in serum of control , normal treated, diabetic and diabetic-treated groups.

Treatments / Parameters	T- cholesterol	HDL- Cho	LDL- Cho	Tri glycride	Total lipid
Normal control	131.34± 6.26 e	67.04±3.00 cd	83.81± 2.27 d	110.88±7.36 de	980.62± 30.71 d
Normal treated chloroform extract	126.72± 6.16 e	65.45 ±3.89 cd	81.99± 5.27 d	103.61± 7.55 ef	1033.50± 23.70 d
Normal treated 70% ethanol extract	128.79± 7.54 e	69.00 ±4.54 c	80.01± 7.29 d	93.32 ±22.07 f	931.94± 145.94 d
Normal treated petroleum ether extract	125.50± 6.40 e	65.00 ±4.16 cd	83.30± 6.08 d	114.06 ±11.09 cde	1049.00± 11.04 d
Diabetes after 2 days	202.50± 4.51 b	81.62± 3.35 b	163.22± 4.57 b	211.75± 8.22 b	1687.19 ±202.10 a
Diabetes after 10 days	239.91 ±10.89 a	90.75 ±1.89 a	210.72 ±9.96 a	307.77± 8.60 a	1805.55 ±103.93 a
Diabetes after 40 days	239.75 ±4.11 a	90.75±0.95 a	210.75± 4.98 a	308.75 ±1.25 a	1808.75± 8.54 a
Diabetes treated chloroform extract	158.15± 8.20 c	64.37±4.53 cd	118.02± 11.50 c	121.16± 4.65 cd	1414.58 ±195.92 b
Diabetes treated 70% ethanol extract	150.81± 6.46 cd	61.92 ±6.46 d	111.25± 2.15 c	112.72± 5.57 cde	1325.00 ±99.71 bc
Diabetes treated petroleum ether extract	145.99± 10.97 d	63.25 ±7.36 cd	107.89± 14.17 c	125.74±10.83 c	1373.17± 41.65 bc
Diabetes treated gliclazide (Ref.Drug)	72.75 ±0.96 f	61.25± 0.95c	81.90± 1.68 d	124.50± 4.20 cd	1237.50± 33.04 c
LSD 5%	10.27	6.06	10.7	14.07	153.84

Lipid profile (total chloesterol,HDL-cholesterol,LDL-cholesterol,Triglycride and Total Lipid) are expressed in mg/dL.
Data are mean ±SD of 7 rats in each group.
Statistical analysis is carried out using one way analysis of variance (ANOVA) using CoStat computer program.
Unshared superscript letters between treatments are significance values at P<0.001

Table (8) Evaluation of different successive extracts and 70% ethanol extract of *Nepeta cataria* on Liver function enzymes in serum of control, normal -treated , diabetic and diabetic-treated groups

Treatments / Parameters	AST	ALT	ALP	Total Protein
Normal control	2.26 ±0.02 c	1.60± 0.09 cd	3.39± 0.13 c	108.25± 5.67 ab
Normal treated chloroform extract	2.48 ±0.08 bc	1.72± 0.14 c	2.94± 0.40 d	112± 1.41 a
Normal treated 70% ethanol extract	2.50 ±0.07 bc	1.65 ±0.03 cd	2.74± 0.25 de	109± 6.21 ab
Normal treated petroleum ether extract	1.97± 0.25 d	1.64± 0.19 cd	2.78± 0.26 de	102± 7.70 b
Diabetes after 2 days	2.73± 0.24 b	1.97 ±0.09 b	4.51± 0.36 b	104.5± 8.42 ab
Diabetes after 10 days	3.23 ±0.23 a	2.44 ±0.17 a	5.55 ±0.60 a	108.75 ± 6.29 ab
Diabetes after 40 days	3.37 ±0.11 a	2.48 ±0.08 a	5.61 ±0.13 a	84.3± 1.74 c
Diabetes treated chloroform extract	2.48 ±0.36 bc	1.72± 0.04 c	2.41 ±0.08 ef	89.25± 4.92 c
Diabetes treated 70% ethanol extract	2.40± 0.27 c	1.66± 0.04 cd	2.52 ±0.31 def	83.5± 5.06 c
Diabetes treated petroleum ether extract	2.41± 0.13 c	1.59±0.11 cd	2.18± 0.17 f	83.25± 5.37 c
Diabetes treated gliclazide (Ref.Drug)	2.53 ±0.03 bc	1.55±0.04 d	2.19 ±0.08f	101.25 ± 5.05 b
LSD 5%	0.28	0.16	0.42	8.11

AST,ALT and ALP are expressed in µ mole/mg protein/min
Total protein is expressed in mg /ml
Data are mean ±SD of 7 rats in each group.
Statistical analysis is carried out using one way analysis of variance (ANOVA) using CoStat computer program.
Unshared superscript letters between treatments are significance values at P<0.001

Table (9) Evaluation of different successive extracts as well as 70% ethanol extract of *Nepeta cataria* on nitric oxide free radical in liver of control, normal-trated, diabetic and diabetic-treated groups

Treatments / Parameters	NO
Normal control	44.07 ±1.37 d
Normal treated chloroform extract	43.32 ±0.70 d
Normal treated 70% ethanol extract	44.03± 0.96 d
Normal treated petroleum ether extract	43.45 ±1.25 d
Diabetes after 2 days	62.96 ±2.32 b
Diabetes after 10 days	72.55 ±1.87 a

Diabetes after 40 days	72.55 ±2.00 a
Diabetes treated chloroform extract	51.30± 2.69 c
Diabetes treated 70% ethanol extract	43.92± 1.47 d
Diabetes treated petroleum ether extract	43.12 ±0.83 d
Diabetes treated gliclazide (Ref.Drug)	44.75 ±1.70 d
LSD 5%	2.4

Nitric oxide (NO) and is expressed in µg/g tissue; Data are mean ±SD of 7 rats in each group.
Statistical analysis is carried out using one way analysis of variance (ANOVA) using CoStat computer program.
Unshared superscript letters between treatments are significance values at P<0.001

Table (10) Evaluation of different successive extracts and 70% ethanol extract of *Nepeta cataria* on carbohydrate hydrolyzing enzymes in liver of control, normal treated, diabetic and diabetic-treated groups.

Parameters \ Treatments	α- amylase	β- glactosidase	α- glucosidase	Total protein
Normal control	32.51 ±4.49 de	0.167± 0.02 bc	0.272± 0.02 c	50.00±10.80 abc
Normal treated chloroform extract	32.36± 4.06 e	0.167± 0.03 bc	0.305± 0.01 bc	36.25± 10.30 d
Normal treated 70% ethanol extract	38.08± 5.43 bc	0.142± 0.03 c	0.342± 0.04 ab	41.25± 8.54 bcd
Normal treated petroleum ether extract	35.83 ±2.20 bcde	0.167± 0.01 bc	0.370± 0.06 a	39.50 ±822 cd
Diabetes after 2 days	20.07± 1.44 f	0.052± 0.02 d	0.192± 0.01 d	55.00 ±9.13 a
Diabetes after 10 days	17.79 ±0.68 f	0.037± 0.01 d	0.187± 0.01 d	48.75± 11.08 abc
Diabetes after 40 days	18.32 ±0.69 f	0.050± 0.01 d	0.190± 0.01 d	56.50± 2.38 a
Diabetes treated chloroform extract	40.94± 2.29 b	0.197± 0.02 a	0.282± 0.04 c	51.25±8.54 abc
Diabetes treated 70% ethanol extract	34.12 ±4.18 cde	0.150± 0.02 c	0.357± 0.02 a	52.50 ±6.45 ab
Diabetes treated petroleum ether extract	52.90 ±6.22 a	0.165± 0.01 bc	0.335± 0.02 ab	35.00± 8.16 d
Diabetes treated diamicron (Ref drug)	37.60 ±2.06 bcd	0.180± 0.01 ab	0.372± 0.03 a	53.92± 1.78 a
LSD 5%	5.13	0.029	0.044	11.95

Enzymes (α-amylase,β- glactosidase and α- glucosidase) are expressed in µmole/mg protein /min
Total protein is expressed in mg /g tissue; Data are mean ±SD of 7 rats in each group.
Statistical analysis is carried out using one way analysis of variance (ANOVA) using CoStat computer program.
Unshared superscript letters between treatments are significance values at P<0.001

Table (11):Evaluation of different successive extracts and 70% ethanol extract of *Nepeta cateria* on body weight, liver weight and liver weight/body weight ratio in control, normal treated, diabetic and diabetic-treated groups

Parameters \ Treatments	Body weigh	Liver weight	Liver weight/ body weight
Normal control zero time	151.50 ± 1.29 b	5.925 ± 0.25 def	0.04± 0 c
Normal control after 2 days	151.00 ± 5.47 b	6.0375± 0.149 def	0.04 ± 0 c
Normal control after10-days	158.50 ± 3.10 b	6.225 ± 0.22 def	0.04 ± 0 c
Normal control after 40-days	181.00± 2.58 a	6.9025± 0.297 cd	0.04 ± 0.008 c
Normal treated chloroform extract	186.5.00± 10.96 a	7.81± 0.64 bc	0.04 ± 0 c
Normal treated 70% ethanol extract	187.75 ± 3.5 a	8.50± 0.37 ab	0.0475± 0.005 bc
Normal treated petroleum ether extract	184.00± 4.69 a	7.7925± 0.80 bc	0.04 ± 0 c
Diabetes after 2 days	142.25± 4.34 c	7.665± 0.87 bc	0.0525± 0.005 b
Diabetes after 10-days	112.75± 8.77 e	9.555 ± 1.86 a	0.085 ± 0.0129 a
Diabetes after 40-days	98.50± 5.68 f	8.425± 0.419 ab	0.0875± 0.005 a
Diabetes treated chloroform extract	123.50± 6.02 d	7.0075± 2.248 cd	0.055± 0.017 b
Diabetes treated 70% ethanol extract	123.75 ± 3.68 d	5.2575± 0.50 ef	0.0425± 0.005 c
Diabetes treated petroleum ether extract	131.00± 4.24 d	5.00± 0.467 f	0.0425± 0.005 c
Diabetes treated gliclazide (Ref.Drug)	153.25± 2.36 b	6.30± 0.29 de	0.04± 0 c
LSD 5%	7.66	1.29	0.009

Body weight, liver weight and liver weight/body weight ratio are expressed in g
Data are mean ±SD of 7 rats in each group; Statistical analysis is carried out using one way analysis of variance (ANOVA) using CoStat computer program.
Unshared superscript letters between treatments are significance values at P<0.001

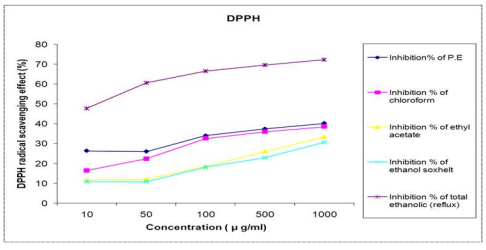

Fig (1) Inhibition Percent of DPPH in different successive extracts and 70% ethanol extract of *Nepeta cataria in vitro*

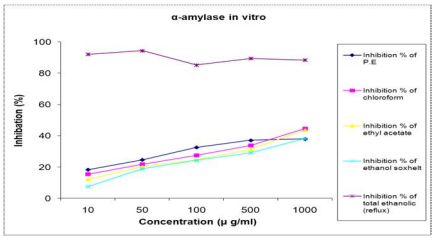

Fig (2) Inhibition percent of nitric oxide (NO) in different successive extracts and 70% ethanol extract of *Nepeta cataria*

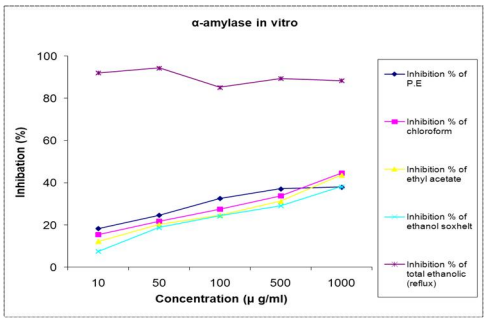

Fig (3): Inhibition Percent of α-amylase in different successive extracts and 70% ethanol extract of *Nepeta cataria*

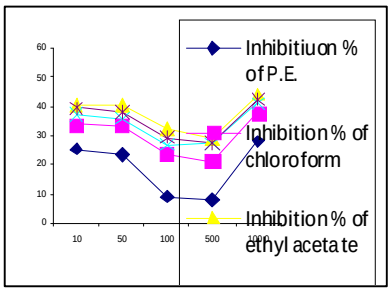

Fig (4) Inhibition Percent of α-glucosidase in different successive extracts and 70% ethanol extract of *Nepeta cataria*

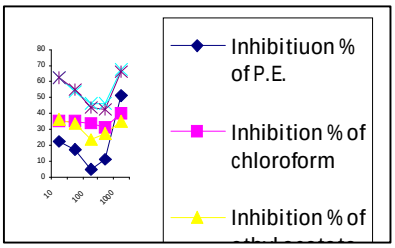

Fig (5) Inhibition Percent of β-galactosidase in different successive extracts and 70% ethanol extract of *Nepeta cataria*

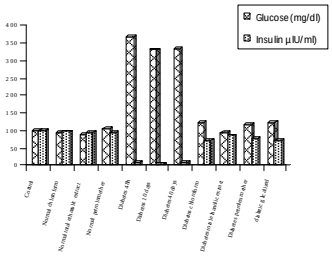

Fig. (6): Effect of *Nepeta cataria* extracts on blood glucose and insulin levels in normal control and various treated groups.

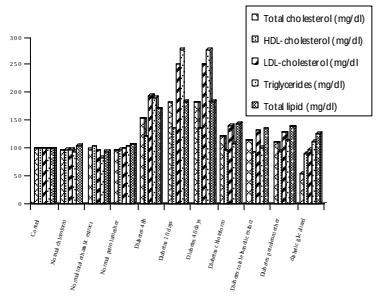

Fig. (7): Effect of *Nepeta cataria* extracts on blood lipid profile in normal control and various treated groups.

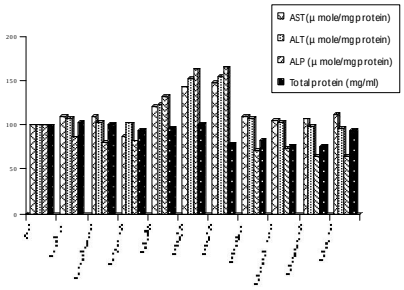

Fig. (8): Effect of *Nepeta cataria* extracts on liver function enzymes in normal control and various treated groups.

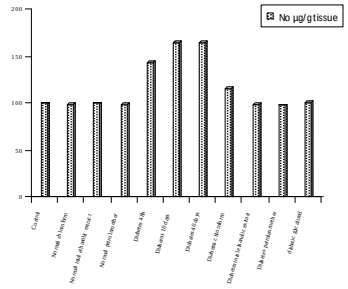

Fig. (9): Effect of *Nepeta cataria* extracts on nitric oxide (NO) free radical in liver of normal control and various treated groups.

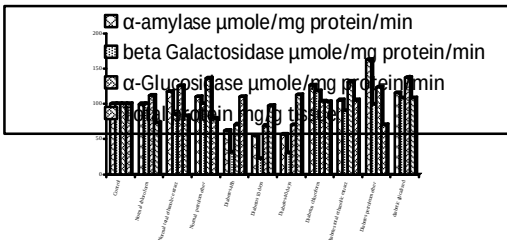

Fig. (10): Effect of *Nepeta cataria* extracts on carbohydrate metabolizing enzymes control and various treated groups.

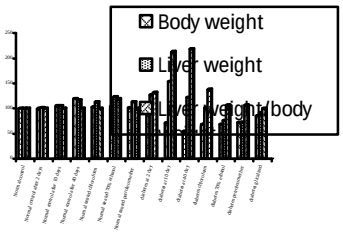

Fig. (11): Effect of *Nepeta cataria* extracts on body weight, liver weight and liver weight/body weight ratio in different control treated, diabetic and diabetic treated groups.

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