Comparative evaluation of biochemical profiling of 4', 4"- (4, 5, 6, 7-TETRAHYDRO- [1, 2, 3-] SELENADIAZOLO [4, 5e] PYRIDINE-4, 6-DIYL) BIS (BENZENE-1, 3-DIOL) and dipyrone on female rat's

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Comparative evaluation of biochemical profiling of 4',4"- (4,5,6,7-TETRAHYDRO- [1,2,3-] SELENADIAZOLO [4,5e] PYRIDINE-4,6-DIYL) BIS (BENZENE-1,3-DIOL) and dipyrone on female rat's

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Abstract: The current study has been carried out at the department of pharmaceutical chemistry, College of pharmacy. Novel synthetically selenium-containing compounds have potential therapeutic effects towards several diseases, such as: cancer microbial infections and neurodegenerative diseases. Therefore, the present study accentuated mainly on two significant items. A novel selenadiazole derivative i.e, 4',4"- (4,5,6,7-TETRAHYDRO- [1,2,3-] SELENADIAZOLO [4,5*e*] PYRIDINE-4,6-DIYL) BIS (BENZENE-1,3-DIOL) (T) and Dipyrone (Di) were used to detretmine their Biochemical effects on female rats. Biochemical test including; liver function tests; Renal functions tests; in addition lipid profile. Invivo study conducted using four groups, one as control (DW) and three treated groups (T, Di, and T&Di). The rats received 50mg/kg body weight (BW) of one of test treatments T and/or Di dissolved in 2 milliliter of distilled water and control group received same volume of distilled water for 30 days. Blood sample were collected directly from the rats heart under chloroform effect. The results indicated that Liver function test showed following results; Aspartate aminotransferase levels(AST) measurement it was cleared that only (T&Di) group (87.52 U/L ±12.20) was increased significantly than both DW(57.23 U/L ±10.43) and T(57.62 U/L ±16.54) groups. Alanine transaminase (ALT) concentration measurements showed only (T&Di) group (70.11 U/L ±13.09) value increased significantly than (DW), (T), (Di) groups. Alkaline Phosphatase (ALP) value of Di group (128.24 U/L ±27.9) highly elevated than in DW (66.68 U/L±15.29) and other test groups. Total protein (TP) concentrations of (Di) (4.97g/dL±1.02), (T) (10.87 g/dL ±3.25) and (T&Di) (5.05 g/dL ±0.76) groups highly reduced than (DW) group (14.80 g/dL ±1.98) level. Lipid profile test results show significant increase of Cholesterol (TC) level of (T) group (533.8mg/dL±52.5) than both DW (335.8mg/dL±27.01) and (T&Di) (390.3mg/dL±25.8) groups. Triglyceride (TG) serum levels only (T) group (100.1 mg/dL ±9.1) showed a significant reduction of TG value than in (Di) group (221.0572mg/dL ±39.8). Levels of HDL of (T) treated group (337.9 mg/dL±26.6) significant increased than all groups. VLDL levels results showed only Di group (43.4mg/dL±4.3) increased significantly than DW group; however there was significant decreased of T group VLDL level (20.03mg/dL±1.8) compare with Di (43.4mg/dL±4.3) and T&Di (35.9mg/dL±3.7) levels. Renal function data reveals significant reduction in blood urea levels of (T) (5.471 mg/dL ±3.745) and (T&Di) (10.633±5.431). Serum uric acid values showed significant decline of (T) group (2.601±0.743) than DW group (5.515±2.046). Also, the results of the present study illustrate only Di treated group (2.33±0.209) had essential increased of Creatinine values than all other study groups. The study concluded that synthesized novel selenadiazole derivative, and Dipyrone have mild effects on liver, kidney, and lipid profile. However, the companion of both drugs has some of undesirable effects.

1 Introduction

Vital physiological role of selenium (Se) is due to the potent antioxidant activity selenoproteins. Some of selenoproteins are well known functions such as thioredoxin reductase (TrxR) , iodothyronine deiodinases, and glutathione peroxidase , the functions of other proteins are not known up till now include selenoproteins T ,X ,Y,[1]. In spite of the rare knowledge of the particular biochemical functions, Several attempts have been made to show that inadequate Se, principally Se-proteins, are related with frequent human diseases including malignancy, diabetes mellitus, cardio-vascular, and immune system conditions[2]. However, much uncertainty still exists about the relation between serum selenium levels and reproductive effects. Selenaheterocyclic compounds primarily have bivalent selenium, such as diselenides, and cyclic selenides. Numeral of synthesized compounds designed to have a potent protective effect. Many of the synthetically selenium-containing compounds used as pharmaceutical drugs [3]. Therefore a significant rising interest on research design of selenadiazole derivatives as drugs is presently perceived in the medicinal organic chemistry field. Various studies concentrated on development of more stable and easily purified organo-selenium compounds. Novel compounds have a potential therapeutic effects towards several diseases, for example: cancer ,microbial infections, and neurodegenerative diseases[4]

Dipyrone (metamizole or novalgin) (Di) used in both human as well as veterinary medicine. It is a Pyrazolone derivative, first it produced in 1920 by Hoechst AG, a German company, and in 1922 Di has been global use. In several countries Di withdraw due to serious complication of agranulocytosis, while in others like Germany it obtainable as prescription drug and over the counter in India, Spain, Russia[5]. Di decreases endogenous glutathione levels and inhibits GPx action in dose dependent manner[6].

In general “liver function tests” a term indicates biochemical tests used for liver and biliary tract diseases detection. Usually those tests used to check liver function include: the alanine transaminase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), albumin, and bilirubin tests. The ALT and AST tests measure enzymes that released from the liver in response to damage or disease[7]. Albumin and bilirubin tests measure just how the liver forms albumin, a protein, and how it disposes of bilirubin, a waste product of the blood[8]. Aminotransferases levels also vary with age, sex, race, and body mass index Levels are found to be higher in obese patients and lower in dialysis patients[9].

Lipids have essential functions in all biological aspects of the cell. They are construction constituents, also lipids involved in hormonal and metabolism pathways. Lipids are transported in the blood as lipoproteins. These lipoproteins have a main role in dietary lipids absorption and transport from the liver to peripheral tissues, and vice versa. Another but to a less extant role is transporting of toxic foreign hydrophobic compounds, like bacterial endotoxin[10]. Plasma lipids chemical structures of four main structures include cholesterol, cholesterol ester, triglyceride and phospholipids. The two main lipids in plasma are Cholesterol (free and esterified forms), and triglycerides.

Blood tests measured the extent of various forms of lipids-fats and fatty substances in blood flow. A complete cholesterol test -is also called a lipid panel- or lipid profile; Total cholesterol, triglycerides, HDL cholesterol, and LDL cholesterol [11]. Triglycerides a subgroup of lipids, as well involve molecules like fatty acids, and their derivatives such as tri-glycerides, di-glycerides, mono-glycerides, and phospholipids, as well as other sterol-containing metabolites[12].

Renal function valuation is a basic component of analyzing and introducing prevention and treatment of chronic renal illness. The aims of renal function test are diagnosis of early renal disorder when there is no signs of illness, determining renal disease development, expected renal replacement therapy, support in proper dosing of medication. Also valuation of kidney functions should be done preceding cancer chemotherapy and all drugs with nephrotoxic effects. Decreasing in renal function need to a dose monitoring or even administration of alternative medications[13]. Serum tests evaluate the levels of specific proteins that excreted through renal system, which best detect glomerular filtration rate abnormalities, like serum creatinine and blood urea nitrogen(urea, BUN, or urea nitrogen) [14].

2 Material and Methods

Healthy female rats aged between 30-32 days, and weighted (210g± 21.6). The rats procured from veterinary medicine college/university of Basrah. Then the rats were kept in polypropylene cages lined with sawdust. In each cage 3-4 rats, the rats were provided usual rat pellet diet and tap water. At the beginning the rats were adapted to laboratory circumstance, natural day and light (12 hours day and 12 hours night. Room temperature 21±4°C[15]. Body weight of all rats was measured. Then the cages were labeled, and separated as groups.

2.1 Experimental design

Forty female rats divided in to four groups, 10 females in each group. The rat's groups received 50mg/kg BW of each either selenadiazole derivative (T), Dipyrone (Di) and both (T &Di) dissolved in 2mL of Distilled water (DW) for30 days. Control group received only 2mL of DW. A novel selenadiazole (T) was synthesized by the authors in pharmacetical chemistry deparetment, college of Pharmacy/ Basrah University. The compound was identified in Al al-Bayt University, Al-Mafraq, Jordan. While Dipyrone was provided from Shaanxi pioneer Biotech.,China. All Compounds are administered orally using a mouth gavage, for 30 days.

2.2 Biochemical examination

2.2.1 Liver function test

Total Bilirubin, Aspartate aminotransferase, Alanine transaminase, Alkaline Phosphatase, and Total protein. All measured via colorimetric determination using JOURILABS kit.

2.2.2 Lipids profile

Triglycerides, total Cholesterol, and High density lipoprotein levels were measured using specific colorimetric JOURILABS kit. LDL Cholesterol was calculated by Friedwald et al formula:

LDL-Cholesterol (mg/dL) =Total Cholesterol-(HDL-Cholesterol +T G/5)[16].

Very Low Density Lipoprotein (VLDL) = Triglycerides/5 [17].

2.2.3 Renal function test

Serum uric acid, serum urea, and serum creatinine were measured using specific colorimetric JOURILABS kit.

2.3 Statistical analysis

Results are documented as mean values and standard deviations (mean±SD). Differences among groups were evaluated using One-way analysis of variance (ANOVA). (p=0.05)[18].

3 Results

3.1 Liver function test

3.1.1 Aspartate aminotransferase (AST) level assessment

ASP results revealed that significant increase in serum AST levels of (T&Di) treated group was (87.52 U/L ±12.20) which was greater than the serum AST levels ($p<0.05$) of (DW) group (57.23 U/L ±10.43) and (T) group (57.62 U/L ±16.54), but non-significant change with (Di) group (64.60 U/L ±23.75). As its illustrated in Figure (1) and table (1)

3.1.2 Alanine transaminase (ALT) activity measurement

It is apparent from the table (1), and figure (1) that serum ALT levels of the study groups; T group (53.56 U/L ±11.16), Di group (47.86 U/L ±12.86) were non-significant alteration than concentration of DW group (38.75 U/L ±11.26). While (T&Di) group ALT value (70.11 U/L ±13.09), was significantly ($p<0.05$) greater than DW group level.

3.1.3 Alkaline Phosphatase (ALP)

Data in table (1), and figure (1) reveal that female rats of T group (60.42 U/L±12.03), and T&Di group (73.57 U/L±16.08) ALP concentration altered non-significantly to than that in DW group (66.68 U/L±15.29). While Di group rats exerted a significant increase ($p<0.05$) in ALP concentration to (128.24 U/L ±27.9) compared with DW, T, and T&Di groups.

3.1.4 Total Protein estimation (TP)

The results obtained from the preliminary analysis of TP levels significantly decreased in all treated groups in comparison with DW group. The results of the treated groups were correlated to the DW group (14.80 g/dL ±1.98); TP of (T) group (10.87 g/dL ±3.25), (Di) group (4.97g/dL±1.02) and (T&Di) group (5.05 g/dL ±0.76), which was significantly decrease ($p<0.05$). However, the ANOVA (one way) showed that there was not statistically significant between Di, and T&Di group TP levels. The results of the correlational analysis are summarized in Table (1), and in Figure(2).

3.1.5 Total Bilirubin (TB)

The data in Figure (2) and table (1) Indicate that no significant changes in TB concentrations Di (3.29mg/dl ±0.78), T (2.93mg/dl ±0.37), and T&Di (3.44mg/dl ±0.95) than TB concentration of control group was (2.8 mg/dl 9±0.87).

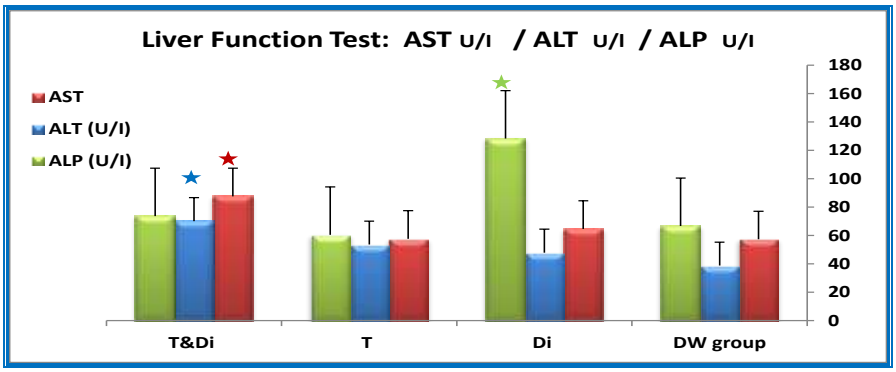

Figure 1: illustrates the effects of (T), or /and Di on AST, ALT, ALP.

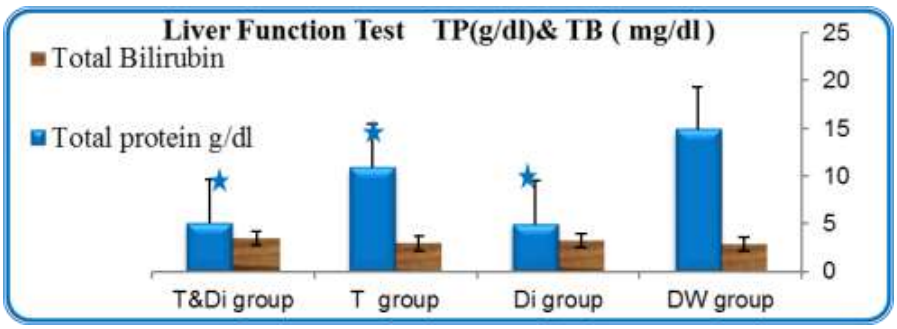

Figure 2: illustrated the effects of T, and/or Di treatment on TP, TB serum concentrations in female rats.

Table 1: illustrates Liver function Tests of groups of the experiments (mean±SD). Means bearing different capital letters vertically differs significantly at 0.05 level ($p<0.050$)

groups	Parameters				
	TB (mg/d)	**AST (U/L)**	**ALT (U/L)**	**ALP (U/L)**	**TP (g/dL)**
DW	2.89±0.87 A	57.23±10.43 A	38.75±11.26 A	66.68± 15.29A	14.80± 1.98 A
Di	3.29±0.78 A	64.60±23.75 A	47.86±12.86 A	128.24±27.9 B	4.97±1.02 B
T	2.93±0.37 A	57.6180±16.54 A	53.56±11.16 A	60.42±12.03 A	10.87±3.25 C
T&Di	3.44±0.95 A	87.52±12.20AB	70.11±13.09 B	73.57±16.08 A	5.05±0.76 B
LSD	NS	29.90	16.60	54.70	3.91

3.2 Lipid profile Tests

3.2.1 Measurement of Cholesterol (TC) concentration

Effect of (T), (Di), (T&Di) administration on serum cholesterol of female rat is shown in Table (2) and figure (3). The result showed (T) group was important increase ($p<0.05$) of TC level (533.8mg/dL±52.5) than in both (DW) group and (T&Di) group (390.3mg/dL±25.8). While non-significant alteration of (Di) group (403.08mg/dL±18.8) and T&Di (390.3mg/dL±25.8) group respectively, compare with DW group (335.8mg/dL±27.01).

3.2.2 Triglyceride (TG) concentration measurement

Also, the data in Figure (3) and table (2) apparent that no-significant alteration in TG level of (T) group (100.1 mg/dL ±9.1), Di (221.0572 mg/dL ±39.8), and T&Di (179.7 mg/dL ±18.3) groups, when compared to DW group (141.9 mg/dL±12.8).On the other hand, (T) group that shows significant decrease ($p<0.05$) than Di group TG level.

3.2.3 High density lipoprotein (HDL) estimation

Further evaluation of lipid profile detected that the daily administration of T compound significantly enhanced rat plasma level of HDL (337.9mg/dL±26.6) when compared with other studied groups; DW group (140.9 mg/dL ±12.8); T&Di group (218.6 mg/dL ±22.05) , and Di group (214.7 mg/dL ±8.4). The data illustrated in figure (4), and table (2)

3.2.4 Low density lipoprotein (LDL)

Analysis of the computed results From the data in Figure (4) and table(2), it is apparent that show the following findings: Non-significantly change of (T), (Di) and (T&Di) groups of LDL levels (185.9±56.7), (144.9±16.5) and (142.3±56.6) respectively in comparison with non-treated(DW) group (162.3±31.2).

3.2.5 (VLDL) estimation

The most striking result to emerge from the data is that statistically significant elevation in VLDL level associated with Di (43.4mg/dL±4.3), while non-significant alteration of T&Di (35.9mg/dL±3.7), and T value (20.03mg/dL±1.8) than DW group level (28.4 mg/dL±3.4). Furthermore, (T) Group was significant declined ($p<0.05$) to (20.03mg/dL±1.8) than in Di and T&Di groups, as in Figure (5), and table (2).

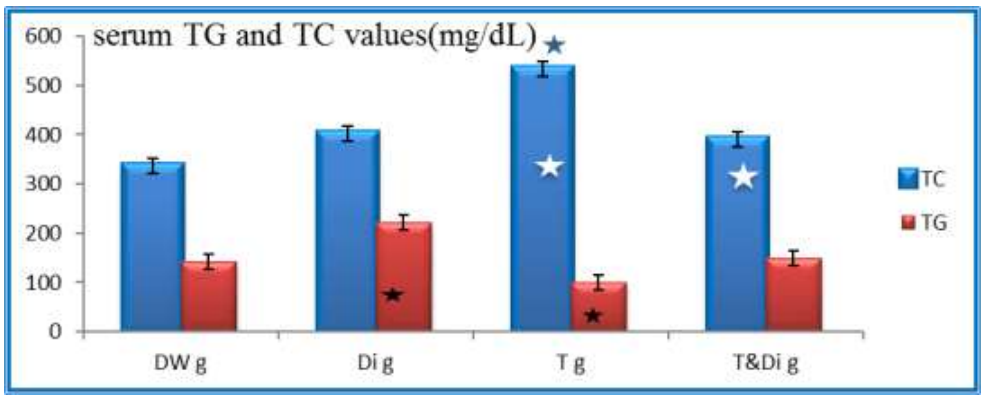

Figure 3: reveals effects of T, and/or Di on serum TC and TG of female rats treated groups

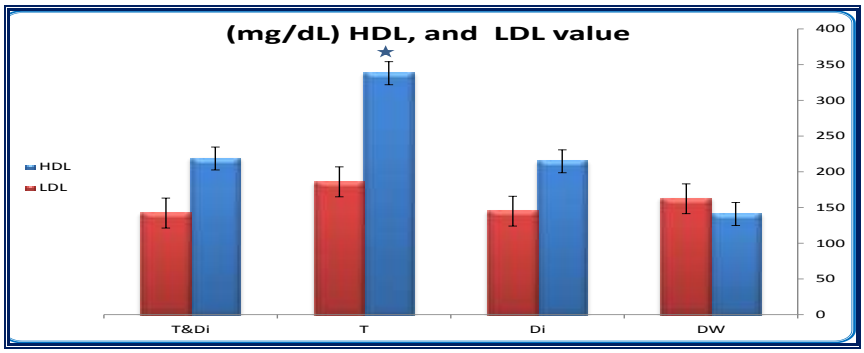

Figure 4: The histogram indicates that effects of T, and/or Di on HDL, LDL levels

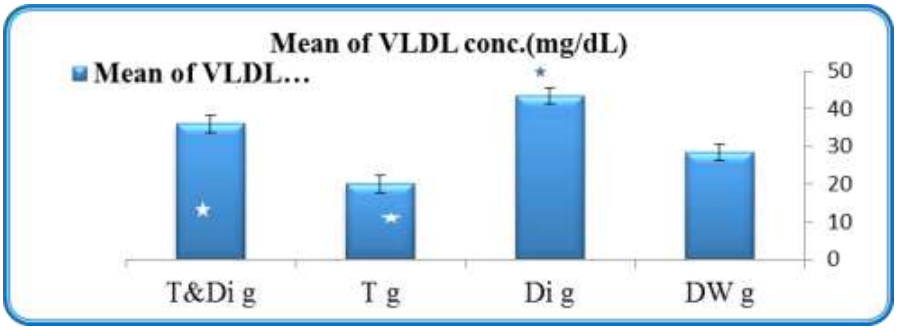

Figure 5: - Reveals important alteration in VLDL values after T, and/or Di treatment

Table (2):- The effects of synthesized (T), and/ or Di on Lipid profile.

groups	Parameters				
	TC mg/dL	TG mg/dL	HDL mg/dL	LDL mg/dL	VLDL mg/dL
DW	335.8±27.01 A	141.9±12.8 A	140.9±12.8 A	162.3±31.2 A	28.4±3.4 A
Di	403.08±18.8 A	221.1±39.8 A	214.7±8.4 AB	144.9±16.5 A	43.4±4.3 B
T	533.8±52.5AB	100.1±9.1 AB	337.9±26.6 C	185.9±56.7 A	20.03±1.8 AC
T&Di	390.3±25.8A	179.7±18.3 A	218.6±22.0A	142.3±56.6 A	35.9±3.7 AB
LSD	143.53	120.93	119.3	NS	15.05

Means bearing different capital letters vertically differs significantly at 0.05 level ($p<0.050$)

3.3 Renal function test

3.3.1 Urea level estimation (U)

Analysis of the data in table (3) and figure (6); reveals that serum urea concentration of T (5.471±3.745), Di (6.645±1.883) and T&Di (10.633±5.431) groups showed significant decreased ($p<0.05$) than in DW group (16.632mg/dL±5.59). Also, serum Urea level of T&Di group greater than T group

3.3.2 Uric acid measurement (UA)

The table(3) illustrate there is a clear trend of increasing levels of UA in circulation in both Di(5.74 mg/dL ±1.20) and T&Di(5.61 mg/dL ±0.71) groups which statistically non- significant($p <0.05$) compared to DW(3.52mg/dL±1.76). ANOVA (one way) showed that UA level in DW group and Tg (3.22mg/dL ±0.68) was statistically significant.

3.3.3 Serum Creatinine SCr concentration calculation

From data in figure (6) and Table (3), illustrate that there was a remarkably increased ($p<0.05$) in the SCr concentration in Di group (2.337 mg/dL±0.209), when compared with SCr level in DW group (0.868 mg/dL±0.144), T (0.907mg/dL±0.216), and T&Di(0.883 mg/dL±0.709) groups.

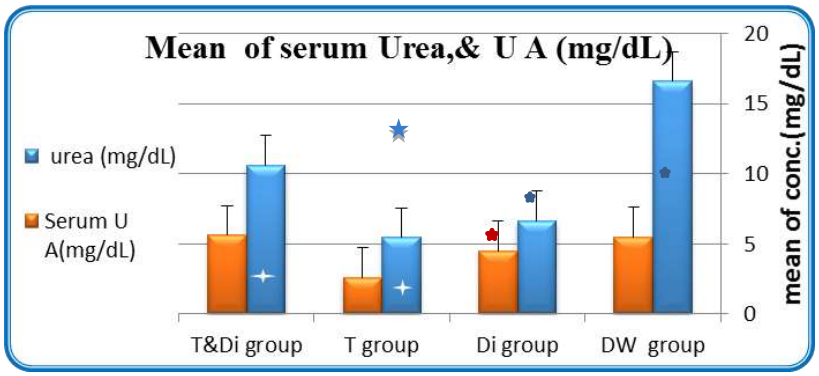

Figure 6: Illustrate serum urea, and UA values in treated groups, and DW group

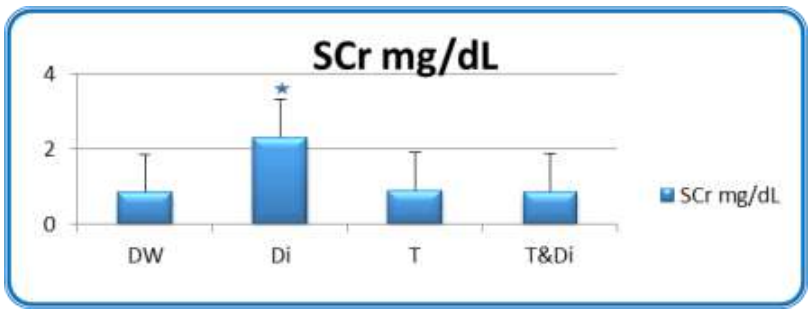

Figure 7: effects of T and/or Di on female rats SCr level

Table3: the effects of T, and/ or Di on Renal function of female rats.

Treatment groups	Parameters		
	Creatinine (mg/dL)	Blood urea(mg/dL)	Uric acid (mg/dL)
DW	0.8674±0.144 A	16.632±5.59 A	5.515±2.046 A
Di	2.33±0.209 B	6.645±1.883 B	4.521±0.346 A
T	0.907±0.216 A	5.471±3.745 B	2.601±0.743 B
T&Di	0.883±0.709 A	10.633±5.431 BC	5.635±0.71 A
LSD	1.43	5.162	2.914

Means bearing different capital letters vertically differs significantly at 0.05 level ($p<0.050$)

4 Discussion

4.1 Liver function Tests

The liver plays a major role in the detoxification and metabolism of exogenous chemicals which are toxic to the general health. Liver dysfunction may be revealed by the alteration in liver biomarker enzymes including AST and ALT. In fact, the damage in hepatocytes leads to changes in their membrane permeability, resulting in the escape of enzymes from cells[19]. In common AST, ALP Tests that interpret liver injury, they have been named, “liver injury tests”. In contrast, standard tests bilirubin, albumin, and prothrombin time are valuable in evaluating liver function. AST, ALT and ALP are sensitive biomarkers directly associated in the extent of liver damage. Their serum level rise could

possibly be referred to the release of these enzymes into the blood circulation from the cytoplasm. Aminotransferase enzymes AST and ALT elevated in hepatocellular injury, while ALP level increased in cholesteric injury[20].

The findings of the current research revealed that AST level of (T&Di) group was significantly greater than the serum AST levels of the DW group. In contrast result is found by [21]compensation of selenium with HgCl2 injection reduced AST level compare to group that received only HgCl2, in the present study T&Di group AST value decline than T group. Furthermore [22] reported that pretreatment with Diphenyl methyl selenocyanate re-established the actions of AST and ALT towards normal. Diphenyl methyl selenocyanate presented better protective activity against damage hepatocytes than control treatment group. The results of this investigation show that rats of (T&Di) group had the most elevated AST level. But, to the less extent Di group; however all treated groups with the normal reference range of rats serum liver enzymes[23].In contrast with the present results, previous study had demonstrated that Selenium inclined to increase plasma transaminases and ALP [24].

ALT is mostly accumulated in hepatocyte, but also found in serum, muscle, prostate, adipose tissue, and brain in very low concentrations, for example in serum the ALT activity is less in about 3000 time than in hepatocyte. Serum ALT increased mainly in hepatocyte injury[25]. The results of this investigation show that ALT of T&Di was significantly ($p<0.05$) higher than concentration of (DW), (T) and (Di) groups. In comparison with the reference ranges of rats liver test as listed in table (4); all treated groups are elevated ALT concentration rather than in DW group. The finding is consistent with findings of recent studies by A number of researchers such as [26], showed that no significant variances existed, between Se alone treated and control groups.

Table 4: reference levels of rat liver function tests adapted from [23]

Liver enzymes	Reference level/ IU/L
AST	50-150
ALT	10-40
ALP	30-130

Nevertheless selenium reduced ALT levels to nearly normal concentration when administered a long with hepatotoxic materials Ag nanoparticles. Also it was later shown by [24]those serum enzyme activities of ALT, AST and ALP levels of Se treated group has no statistically significant differences comparable to control. But Se- chlorpyrifos(CPF) treated group ALT level reduced significantly from chlorpyrifos treated group. Despite prior evidence a study by [27] reported that liver enzyme evaluation showed an increase in ALT level in high-fat diet group and Selenium doses at (0.25, 0.5, 1 mg) groups compared to control group.

A recent research has presented that a high AST: ALT ratio exceeded other non-histological indicators of cirrhosis in Primary Sclerosing Cholangitis, but still only attains in significant sensitivity of 65-79%. As with other liver diseases, there are suggestions that an AST: ALT ratio of >1 indicates the development of cirrhosis[28]. In the present study elevation of ALT value might be due to long term treatment. Medications for long-term should be carefully studied when faced an unexplained elevation ALT concentration, to confirm drug-ALT concentration the drug administration must be ceased [25]. However Elevated ALT levels up to 300 U/L in human are nonspecific. Noticeable increments of ALT values larger than 500 U/L detected most often in patients with ischemic liver injury, viral hepatitis, and toxin-induced liver injures[29]

ALP creates mainly from the bile duct epithelia in liver and osteoblasts, ALP may also be present in intestine, placenta, kidney and leucocytes. ALP rise due to physiological role, for example increase ALP in the third trimester of pregnancy, a result of an influx of placental ALP, or due to pathological causes like biliary obstruction[8].Based on the results of the current investigation, ALP concentration of T and

T&Di groups altered non-significantly than that in non-treated group. Di group ALP concentration were significantly increased ($p<0.05$) than DW, T and T&Di groups. Increased level of ALP enzyme activity resulted from enhanced functional activity of the tissues caused by the drug. Such increase can constitute hazard to cell life that depend on phosphate esters for their vital process. Decreased serum ALP this suggests that (T) may induce ALP synthesis in the tissues[30]. Still, the non-significant alteration in the serum alkaline phosphatase activities is a sign that there was no leakage of the enzyme into the serum[23]. Effects of selenium from the present study agrees relatively well with that from [24]Selenium tended to improve serum transaminases and ALP as established by author and Based on present observations, Se may supply a protection for long term medications against hepatotoxicity induced by toxins. From the subacute toxicity it can be assumed that the increased level of alkaline phosphatase, AST and ALT level may be responsible for the tissues damages in the liver and kidney. Despite to the prior evidence by rats that were given selenium alone showed an insignificant change in ALP serum level, while rats that were given either selenium or garlic with HgCl2 injection exhibited an increase in ALP activity by 9.1% and 16.2%, respectively, as compared with the HgCl2 group[21]. Both selenium and vitamin E act to restore the hepatic damage induced by bisphenol A (Bisphenol A)[31], that's agree with our results that of T and T&Di groups ALP level was less than in Di group.

The results obtained from the preliminary analysis of TP levels significantly decreased in all treated groups in comparison with DW group total protein concentration. Nevertheless, few studies are to be found providing detailed about effects of selenium containing compounds on the total protein concentration. A study by[32] concluded that decrease in total protein values refer to cellular damage and necrosis which lead to proteins leakage from the tissues to the blood, mainly occurred with liver, and/or renal problems, also occurred when there was reduced rate of protein synthesis [33] Also revealed the decrease in serum total protein content in T group could be attributed to the formation of Selenium containing proteins. Contrary to our results [34] reported that the selenium supplementation as sodium selenite or Se nanoparticles in the basal diet had no effect on serum glucose, serum TP and serum albumin. Furthermore,[35] reported that Different forms of selenium had no significant effect on the amount of total serum protein. The reduction in TP could be due to reduced protein synthesis. Also Metals are known to produce hepatic hypertrophy changes leading to the destruction and necrosis of the cells which may cause escape of proteins from the tissues to the blood

It was found that total bilirubin TB levels were not significantly altered by T, Di, and T&Di treatment. This might be explained by the results of histopathological experiment. Elevation in total bilirubin, direct and indirect bilirubin concentration related with bilirubin metabolic disorders, and fatty degeneration of the liver. All test groups in the present study were not produced degenerative liver change. The finding is consistent with findings of recent study by [31] concluded that Se and Vit. E supplementation has no change in both total and direct bilirubin compared to non-treated control group. Despite prior evidence [36] have demonstrated that Zinc –Selenium tea decrease bilirubin concentration in high sucrose high fat obese rats, they concluded that Zinc –Selenium tea has protective effects on liver function in experimental rats, the effects of which are better than normal green tea.

4.2 Lipid profile

Based on the results of TC in the present study, it can be seen that cholesterol value significant increase ($p<0.05$) in TC value T group than DW group, and T&Di group levels. Di and T&Di groups revealed non-significant change with DW group. Research finding by [37],also points towards increase total cholesterol from 13% to 27% in group fed with seleno-methionine containing diet. A study by [38]also found that treatment with organo-selenium compound increase level of total cholesterol. However, a number of studies showed that significant differences do exist, albeit findings are somewhat contradictory. More recently [27] investigated that plasma cholesterol value decreased in groups with maximum concentration of selenium diet in comparison with high fat diet group, this result agree with decline of TC in T&Di group than T group. The mean values of serum total cholesterol were significantly ($p<0.001$) lowered in all the Se supplemented Wistar rats compared to control group. Though, they were within the normal range (40-130 mg/dL) in rats. Also, [34] provided Se supplementation remarkably decrease serum total cholesterol levels in rats with the high Se nanoparticles supplemented groups. However, they were within the normal reference range in rats. In contrast, [39]

concluded that serum Di level of patients received the drug, there was observed a negative correlation between the concentrations of Di in the blood and triglycerides, and cholesterol concentration.

The results of present study a one-way ANOVA analysis revealed that TG value of T, Di and T&Di groups statistically non-important alteration compared to DW. However there was an important reduction of T group TG values than that in Di group.

Previous studies of selenadiazole compounds have not dealt with lipid profile specially TG. A recent study by [31]found that Selenium and Vit E supplement reduced TG. In addition [40] reported that increased serum TG and reduced body fat in the low selenium group suggest that lipolysis was increased by serum T_3 as has been reported by others Increased serum triacylglycerol driven by thyroid hormone has also been reported in selenium-deficient rabbits fed a high fat diet. The above findings contradict the study by [41]found that plasma TG values increased significantly diabetic group with Se and Vit E supplement compared with the normal control group.

HDL value measurement, in the present study the, administration of T compound significantly enhanced rat serum level of HDL than HDL value of DW group. Di and T&Di showed not important increased in serum HDL values. The results were also showed that T group had the more great ($p<0.05$) HDL level than of Di, and T&Di groups. Consistent with our findings recent studies by [27], the administration of selenium compound improve the HDL value in selenium receiving group compare with high fat receiving diets group. In addition Amraoui,et al 2018 [31] concluded that Supplementation of selenium and vitamin E significantly enhanced LDL, and HDL levels in all the experimental groups compared with the control groups. Furthermore [37] selenomethione used in feed increased HDL level in the blood" from 4% to 16%". Despite prior evidence pervious study by [41] suggested that no significant changed in serum levels of LDL, HDL and TC were observed following vitamin E and selenium administration for four weeks in the treated diabetic group compared with the normal control group. To the best of our knowledge, there were no controlled studies reported the effects of Di drug on lipid profile

Non -Important alteration in LDL value of T, Di, and T&Di groups than in DW group LDL level. The results of [42] study indicated that differences in Se levels between Chronic renal failure patients and healthy individuals on baseline study and after three months Se supplementation were not affected LDL values, however there was numerical increased in LDL, which agreed with our findings in current investigation. However, a number of studies show that significant differences do exist, albeit findings are somewhat contradictory like results found by [31] significant elevation in LDL. Another study by [27] report a significant increase in LDL serum level in the groups treated with average and maximum doses of selenium compared to the group with high-fat diet.

In contrast, the study by [43] were Found differences suggesting that reduction of LDL plasma concertation in group with Se supplement due to increase in activity of LDL receptors. Also [41] conclude that LDL levels decline in the treated rats with Vit E and Se is due to direct antioxidants effects on oxidation of lipids and lipoproteins

The present study indicates only Di group statistically significant increased than DW group VLDL value. However VLDL level of T and T&Di groups were numerically non- essential differences than DW; but there was a significant difference in VLDL of T group than in T&Di and Di groups. VLDL carries triglycerides (fat) to various body tissues for energy or storage purposes. It works in close concert with HDL and LDL, along with various enzymes[34]. Because there is no direct method for VLDL estimation, their levels calculation depends on TG values. However, far too little attention has been paid to VLDL assessment, due to direct proportionate with TG levels little attention has been paid to VLDL. Furthermore, to the best of our knowledge, no report has been found so far estimate level of serum VLDL relating to different Se levels. Nevertheless, there have been no controlled studies which compare effects of Di in serum Lipids. To the best of our knowledge we are the first who's reported such results.

4.3 Renal function test

This study indicates that U level of (T), (Di) and (T&Di) groups showed significant decreased than DW group. Production of urea related directly to high protein diets, medication for example, corticosteroids or

diseased conditions like trauma, heart failure, infections, and acute renal failure[13]. Urea and Cr are indicators of the renal structure and when the renal structure becomes damaged, the levels of these enzymes increased[44]. The present finding also supports by[44] Study which concluded that rats on Se treatment ischemia-reperfusion group revealed reduction in serum urea decreased than in Ischemia-reperfusion group. Our findings contradict the study examined the effect of Se on serum biochemical parameters, the results indicated that urea is not significantly altered with Se treatment[38].

The finding of present study revealed that UA concentration of (T) group was decline significantly than in DW group. Among the acceptable explanations for this finding is that, the reduction in serum UA value related to high HDL level. It was found that HDL value has negative association to UA level [45]. Also UA production directly related to TG synthesis in the liver; because TG synthesis is associated with the de novo synthesis of purine, which accelerating UA production [45]. Yet, very few studies have examined serum UA correlated to concentrations of Se in plasma. The research study by[46] found concentration of plasma Se levels in Hemodialysis Patients were negatively related with uric acid levels (inflammation biomarker; $p < 0.01$). In contrast to the results of the current study a study [39] reported that serum Di level has negative correlation with serum UA, Creatinine, TG, and TC

Serum creatinine (SCr) has been the most thoroughly investigated glomerular filtration marker. Creatinine is produced by skeletal muscle breakdown and is contained in cooked meat. The most remarkable result to arise from the data is that no significant changes occur in SCr level in T&Di and T groups, with significant increase in Di group compare to DW group.

Result obtained by[21] found that rats given selenium supplementation has no significant effects on SCr level, and significantly reduced SCr concentration in rat group received selenium with HgCl2, which is in good agreement with the results of the present study. Similar results reported by [44] Study which concluded that normal rats on Se treatment group revealed non- important alteration in serum SCr than control group, while in Se-treated Ischemia-reperfusion showed reduction than in Ischemia-reperfusion group. In contrast [39] reported that serum dipyrone level has negative correlation with serum UA, S Cr, TG, and TC. A study [47] was concluded that no study has examined the effect of Di on renal function in patients with a declined in ECV (effective circulating volume).

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