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# Research Article
# Effect of *Rosa canina* Distilled Water on Tamoxifen-treated Male Wistar Rats

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## Abstract

**Background and Objective:** In spite of therapeutic effect of tamoxifen on the breast cancer, it has some side effects on the liver including non-alcoholic fatty liver disease. In this study the effects of *Rosa canina* distilled water on the tamoxifen-induced fatty liver and oxidative stress status in male rats were investigated. **Materials and Methods:** Twenty four adult male Wistar rats were randomly divided into 4 groups of 6: 1st group: Untreated control rats (C), 2nd group (T): The rats received tamoxifen, 3rd group (T+R): Rats received tamoxifen and *Rosa canina* distilled water and 4th group (R): Rats received only *Rosa canina* distilled water. Tamoxifen at 1 mg $kg^{-1}$/day was injected subcutaneously for 7 days and the rats received orally *Rosa canina* distilled water at 1 mL/rat/daily for 14 days. At the end of the study, animals were studied for serum biochemical parameters (glucose, lipid profile, BUN, creatinine, uric acid, urea, ALT, AST, ALP, total protein, bilirubin, oxidative stress indices, sperm analysis and histology of the liver. The data were analyzed with SPSS software version 20 and expressed as Mean±SD. **Results:** *Rosa canina* distilled water improved liver enzyme and renal function indices which disturbed due to tamoxifen treatment. While tamoxifen enhanced lipid peroxidation, *Rosa canina* distilled water reduced it. In addition, tamoxifen reduced the mobility, morphology and viability of sperms, but the *Rosa canina* distilled water enhanced the sperm parameters. Histological results also confirmed the adverse effect of tamoxifen and the favorable impact of the *Rosa canina* distilled water on the liver structures of animals. **Conclusion:** *Rosa canina* distilled water could modulate tamoxifen-induced fatty liver as well as improving the sperm parameters.

Key words: *Rosa canina* distilled water, fatty liver, tamoxifen, oxidative stress, rat, cirrhosis, hepatic disorders

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

## INTRODUCTION

Non-alcoholic fatty liver disease (NAFLD) is the most common liver malfunction in the world. The NAFLD is a metabolic syndrome associated with insulin resistance[1]. The NAFLD is happened in the absence of alcohol consumption and it covers a big range of simple steatosis to liver disease such as non-alcoholic steatohepatitis, fibrosis, cirrhosis and hepatocellular carcinoma[2,3]. The incidence of NAFLD is 2-3 times higher than that of hepatitis B and C and alcohol-related liver disease[4]. Chronic lipid accumulation in the liver can lead to non-alcoholic steatohepatitis (NASH) that consequently results in hepatotoxicity and hepatic failure, such as cirrhosis[5]. One of the important functions of the liver is the storage and metabolism of fats in the body. The main factors leading to the fatty liver are associated with increased activity of lipogenesis pathways or disorder in metabolism of fatty acids and can be influenced by environmental stimuli or genetic factors resulting in inflammation and destruction of the liver cells and eventually causing cirrhosis[6-8]. The drugs that produce fatty liver can affect the synthesis or peroxidation of fatty acids[6].

Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) are the main liver enzymes that increase in NAFLD. Besides, an increased level of gamma-glutamyl transpeptidase (GGT) is an indicator of NAFLD and insulin resistance[9]. Oxidative stress and antioxidant defense is a network of enzymatic and non-enzymatic processes. Hepatotoxic diseases are also characterized by oxidative stress[10].

Tamoxifen citrate as a non-steroidal anti-estrogen drug, also known as Nolvadex is used to treat and prevent diseases such as breast cancer, infertility, gynecomastia and bipolar disorder. However, it has some side effects, including preventing osteoporosis, increasing the risk of uterine cancer and causing fatty liver disease (steatosis) and in the developed stage it can cause steatohepatitis, fibrosis, cirrhosis and hepatocellular carcinoma[11,12]. Several mechanisms have been suggested for tamoxifen in fatty liver induction such as promoting deficiency of beta-oxidation of fatty acids, increasing lipogenesis, fatty acid synthesis and triglyceride production[6,7].

*Rosa canina* growing in the forest, water wells and pits has antioxidant, lipoxygenase inhibitory and hypoglycemic effects. The fresh fruit of *Rosa canina* (Hypseus) contains vitamin C in the form of ascorbic acid and dehydroascorbic acid, pectin, tannins, citric acid and malic acid, flavonoids, yellow and red pigments, especially carotenoids, vanillin, essential oils and sugars[13-16]. The main fatty acids found in *Rosa canina* contain linoleic, oleic, linolenic, palmitic and stearic acids. In general, more than 90% of total oil in *Rosa canina* contains unsaturated fatty acids[13].

Although tamoxifen has long been the only choice for the treatment of hormone dependent breast cancer, there are adverse effects to the use of tamoxifen; among them are development of liver cancers, increasing blood clothing, retinopathy, corneal opacities and non-alcoholic fatty liver disease (NAFLD)[6]. Thus the objective of this study was to investigate the effects of the *Rosa canina* distilled water on liver histology, serum biochemical parameters, oxidative stress indexes and sperm parameters in the rats with tamoxifen-induced fatty liver.

## MATERIALS AND METHODS

**Location and duration of study:** The study was carried out in Hamadan University of medical sciences, West of Iran from April-November, 2018.

**Materials:** OH-Tamoxifen (Sigma, Malaysia) at a concentration of 1 mg $kg^{-1}$, was dissolved in sesame oil contains 1% benzyl alcohol[17]. *Rosa canina* distilled water was purchased from the local shop in Hamadan city.

**Animal study schedule:** Twenty-eight adult male Wistar rats weighing 200-250 g were collected from the Animal Center of Hamadan University of Medical Sciences. Animals were placed in special cages with appropriate bedding and free access to the standard plate and drinking water under standard conditions; temperature (23±2°C), humidity (60±5%) and light (12 h light and 12 h darkness). The animals were allowed to acclimatize for 1 week. Then, the rats were randomly divided into 4 groups of 6 rats as follows: Control rats (C) received 0/5 mL sesame oil+benzyl alcohol 1% by subcutaneous injection daily for 1 week, 2nd group (T) the rats received tamoxifen (1 mg $kg^{-1}$ dissolved in 0/5 mL sesame oil+benzyl alcohol 1% by subcutaneous injection daily for 1 week), 3rd group (T+R): Rats received tamoxifen (1 mg $kg^{-1}$ dissolved in 0/5 mL sesame oil+benzyl alcohol) by subcutaneous injection daily for 1 week and 1 mL *Rosa canina* distilled water by daily oral gavage for 2 weeks and group 4 (R): Rats received 1 mL *Rosa canina* distilled water by daily oral gavage for 2 weeks. The blood of rats' tails was collected and measured for glucose by glucometer (Bionim, Taiwan) at the collecting day, pretreatment and post-treatment with tamoxifen (TAM) and *Rosa canina* distilled water. At the end of the study, the rats were weighed, sacrificed and their

blood was collected in a heparinized tube. The serums were prepared by centrifuged at 1500 g for 10 min and frozen at -20°C until analyzed for biochemical parameters. The experimental schedule was managed according to the guidelines for the care and use of laboratory animals of the Payame Noor University, Tehran, IRAN (No. 6144622).

**Biochemical assays:** The serum biochemical parameters, fasting blood sugar (FBS), blood urea nitrogen (BUN), creatinine (Cre), uric acid (UA), total cholesterol (Cho), HDL-Cho, total protein, triglyceride (TG), alkaline phosphatase (ALP), alanine transaminase (ALT), aspartate transaminase (AST), bilirubin, were measured by Autoanalyzer (Mindray-BS 480, USA) using Pars Azmun kits (Iran). Serum LDL and VLDL levels were calculated by the following formula:

$$LDL = (TG/5+HDL)\text{-Total Cholesterol}$$

$$VLDL = TG\times 0.2$$

The serum urea was also obtained according to:

$$\text{Urea (mg dL}^{-1}) = \text{Bun (mg dL}^{-1})\times 2.14$$

**Determination of oxidative stress indexes:** Oxidative stress parameters, total antioxidant capacity (TAC), total oxidant status (TOS), catalase and lipid peroxidation as MDA were assayed according to the previous work by Heidarisasan *et al*.[18].

**Sperm analysis:** At the end of the study, the animals were anesthetized, the epididymis were removed and animal's sperms analyzed for the count, motility, viability and morphology. The sperm count was assessed using a hemocytometer. The morphology and motility of sperms were evaluated qualitatively and quantitatively using a light microscope (Zeiss 15447, Germany) with a magnification of 40. The vitality of the sperms was studied with eosin dye. Then, the percentage of live sperm to total sperm was determined. The reddish sperms were viewed as dead sperms.

**Histological assay of liver tissue:** At the time of sacrifice, the liver of the animals was removed, washed with physiologic serum, fixed in 10% neutral phosphate-buffered formalin solution and embedded in paraffin. The liver samples were dehydrated, cleared, paraffin embedded, sectioned at 5 µm thick, mounted on a glass slide and stained with hematoxylin and eosin (H and E). The liver sections were studied histologically by a pathologist using a microscope equipped with a digital camera (Leitz, dialax 20, Germany).

**Statistical analysis:** Data were analyzed with SPSS software Version 20 (SPSS, Chicago, IL, USA) and expressed as mean±standard deviation (SD). The Comparison between groups was conducted with one-way analysis of variance (ANOVA) followed by *post hoc* test, the least significant difference (LSD). Results were considered statistically significant at the probability (p) value of 0.05.

## RESULTS

**Effect of tamoxifen and *Rosa canina* distilled water on the animal's body weight and blood glucose:** According to Table 1, the weight of the rats increased significantly through the study, ($p<0.05$). Although the difference between the weight of the studied groups at starting day and pretreatment were significant ($p<0.05$), they were not significant at post treatment. As it was presented in Table 2, although there was no significant difference in blood glucose levels between the rats, the group that treated with tamoxifen showed significant increased blood glucose level ($p<0.05$) in post-treatment compared to pretreatment.

**Effect of tamoxifen and *Rosa canina* distilled water on the serum biochemical parameters:** Table 3 shows tamoxifen slightly decreased serum FBS compared to the control group (C). However, *Rosa canina* distilled water further decreased it. No significant changes in serum bilirubin and total protein levels were shown among the groups ($p>0.05$). Nevertheless, treatment with tamoxifen caused a slight increase in serum total protein of the animals.

According to results in Table 4, while tamoxifen slightly increased the serum BUN compared to the control group, *Rosa canina* distilled water decreased it when comparing to the group T. The changes were not meaningful. In the group T+R, treatment with *Rosa canina* distilled water slightly decreased serum creatinine in comparison to the groups C and T. Although tamoxifen decreased serum level of uric acid, *Rosa canina* distilled water further reduced it. *Rosa canina* distilled water also dropped the increased serum level of the urea due to tamoxifen.

**Effect of tamoxifen and *Rosa canina* distilled water on the serum levels of liver enzymes:** Regarding the results were presented in Table 5, although tamoxifen increased the serum levels of the liver enzyme comparing to the control group, treatment with *Rosa canina* distilled water decreased them compared to the groups C and T. The changes were not meaningful.

Table 1: Effect of treatment with TAM and *Rosa canina* on body weight (g) of animals

| | Groups | | | | |
|---|---|---|---|---|---|
| Time | C | T | T+R | R | p-value |
| Starting day | 217.50±10.40 | 217.00±2.19 | 212.0±0.00 | 232.50±1.56# | 0 |
| Pretreatment | 239.00±1.09* | 240.25±3.01* | 230.5±1.09*# | 243.00±7.12 | 0 |
| Post treatment | 259.33±20.36* | 254.83±19.34* | 244.0±20.67* | 257.83±23.97* | 0.597 |
| p-value | 0 | 0 | 0 | 0.03 | |

All values are expressed as mean±standard deviation, *Significant difference in column at p<0.05 as compared to starting day and pretreatment, #Significant difference in row at p<0.05 as compared to between in the studied groups

Table 2: Effect of treatment with TAM and *Rosa canina* on blood glucose (mg $dL^{-1}$) in rats

| | Groups | | | | |
|---|---|---|---|---|---|
| Time | C | T | T+R | R | p value |
| Starting day | 76.00±10.19 | 68.50±11.97 | 80.00±8.96 | 83.00±11.38 | 0.137 |
| Pretreatment | 80.20±16.39 | 69.00±8.00 | 81.50±16.79 | 74.50±9.05 | 0.36 |
| Post treatment | 79.33±10.23 | 82.00±7.48* | 80.33±11.41 | 81.17±7.46 | 0.982 |
| p value | 0.881 | 0.04 | 0.978 | 0.29 | |

All values are expressed as mean±standard deviation, *Significant difference in column at p<0.05 as compared to pretreatment

Table 3: Effect of treatment with TAM and *Rosa canina* on serum FBS, total bilirubin and total protein

| | Groups | | | | |
|---|---|---|---|---|---|
| Factors | C | T | T+R | R | p value |
| Glucose (mg $dL^{-1}$) | 178.33±48.38 | 166.60±25.51 | 138.17±21.89 | 166.17±24.10 | 0.204 |
| Bilirubin (mg $dL^{-1}$) | 0.05±0.00 | 0.05±0.01 | 0.05±0.00 | 0.04±0.00 | 0.337 |
| Total protein (g $dL^{-1}$) | 6.11±0.44 | 6.20±0.43 | 5.63±0.53 | 5.96±0.65 | 0.308 |

All values are expressed as mean±standard deviation, p-value means difference in a row

Table 4: Effect of treatment with TAM and *Rosa canina* on rats serum (mg $dL^{-1}$) BUN, Cre, UA and Urea

| | Group | | | | |
|---|---|---|---|---|---|
| Factors | C | T | T+R | R | p-value |
| BUN | 25.67±4.22 | 29.80±4.49 | 27.50±5.05 | 28.50±3.33 | 0.454 |
| Cre | 0.75±0.05 | 0.75±0.08 | 0.67±0.05 | 0.72±0.07 | 0.15 |
| UA | 3.25±1.74 | 2.18±0.80 | 1.80±0.39 | 2.33±0.42 | 0.121 |
| Urea | 54.92±9.04 | 63.77±9.61 | 58.85±10.80 | 60.99±7.12 | 0.454 |

All values are expressed as mean±standard deviation, p-value means difference in a row

Table 5: Effect of treatment with TAM and *Rosa canina* on liver enzymes (U $L^{-1}$) in serum of rats

| | Groups | | | | |
|---|---|---|---|---|---|
| Factors | C | T | C+T | R | p-value |
| ALP | 485.80±120.57 | 580.60±312.30 | 371.33±105.00 | 439.33±133.46 | 0.314 |
| ALT | 167.40±32.60 | 172.40±31.66 | 149.17±37.58 | 157.17±13.06 | 0.587 |
| AST | 61.20±8.01 | 62.40±18.09 | 58.50±15.85 | 71.17±15.74 | 0.515 |

All values are expressed as mean±standard deviation, p-value means difference in a row

**Effect of tamoxifen and *Rosa canina* distilled water on the serum lipid profile:** Effect of tamoxifen in decreasing serum levels of TG, Cho and LDL-Cho and increasing serum level of HDL-Cho was presented in Table 6. It was also shown that serum HDL-Cho in group T+R was significantly lower than groups T. Treatment with only *Rosa canina* distilled water did not significantly change the lipid profile.

**Effect of tamoxifen and *Rosa canina* distilled water on the serum oxidative stress parameters:** As it was presented in Table 7, although tamoxifen slightly decreased TAC, significantly increased MDA in the serum of the animals compared to the control group. *Rosa canina* distilled water significantly reduced the serum level of MDA in the groups T+R and R compared to the group T. There was no meaning difference in the level of serum catalase between the groups.

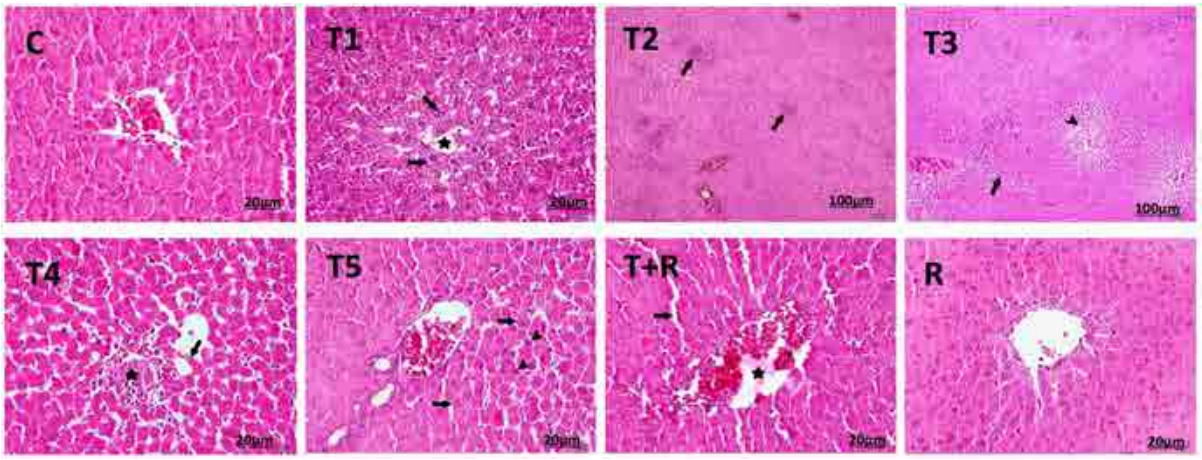


Fig. 1: Microscopic H and E view of the livers (A, B, E, F, G and H ×400, C, D ×100)

C: Control, T1-T5: Groups treated with tamoxifen, T+R: Group treated with tamoxifen and *Rosa canina* distilled water, R: Group treated with *Rosa canina* distilled water. Arrows (T1): Centrilobular hepatocytes with spherical shapes, Star: Around central venues, Arrows (T2): Formation of small granulomatous like masses, Arrows (T3): Mild hyperplasia of bile ductular epithelium at the portal triad, Arrowhead (T3): Dissociation of hepatic cords along with distention of sinusoids, Arrows (T4): Distorted microscopic architecture of the centrilobular area with the disintegration of hepatocytes, focal necrosis and inflammation surrounded by mononuclear inflammatory cells, Arrows (T5): Disarrangement of lobular architecture at portal triad with polygonal hepatocytes, Arrowhead (T5): Pyknotic nuclei representing primary stages of apoptosis

Table 6: Effect of treatment with TAM and *Rosa canina* on lipid profile (mg $dL^{-1}$) in serum of rats

| | Groups | | | | |
|---|---|---|---|---|---|
| Factors | C | T | T+R | R | p-value |
| TG | 52.67±13.86 | 50.00±11.35 | 54.50±10.42 | 56.00±13.34 | 0.87 |
| Cho | 64.50±12.02 | 56.80±5.93 | 51.67±2.87 | 54.50±8.55 | 0.073 |
| HDL-Cho | 22.83±1.169 | 25.00±4.00 | 20.00±1.67* | 22.00±2.89 | 0.035 |
| LDL-Cho | 31.13±11.87 | 21.80±4.33 | 20.77±2.27 | 21.30±5.93 | 0.067 |
| VLDL | 10.53±2.77 | 10.00±2.27 | 10.90±2.08 | 11.20±2.66 | 0.87 |

All values are expressed as mean±standard deviation, p-value means difference in a row, *Significant difference at p<0.05 as compared to T

Table 7: Effect of treatment with TAM and *Rosa canina* on oxidative stress parameters in serum of rats

| | Groups | | | | |
|---|---|---|---|---|---|
| Factors | C | T | T+R | R | p-value |
| TAC (mg $dL^{-1}$) | 0.63±0.07 | 0.58±0.08 | 0.50±0.06 | 0.60±0.06 | 0.052 |
| TOS (mg $dL^{-1}$) | 0.04±0.00 | 0.04±0.00 | 0.04±0.00 | 0.05±0.01 | 0.158 |
| MDA (mg $dL^{-1}$) | 386.16±182.86 | 772.83±62.29* | 244.76±115.92$^{\#}$ | 297.80±122.74$^{\#}$ | 0.000 |
| Catalase (U $mL^{-1}$) | 0.05±0.02 | 0.04±0.00 | 0.03±0.01 | 0.03±0.01 | 0.248 |

All values are expressed as mean±standard deviation, p-value means difference in a row, *Significant difference at p<0.05 as compared to C, R and T+R, $^{\#}$Significant difference at p<0.05 as compared to T

**Effect of tamoxifen and *Rosa canina* distilled water on the sperm parameters:** According to the results shown in Table 8, treatment with tamoxifen caused significantly decreased motility, viability and normal morphology and also non-significantly reduced count. Although treatment with only *Rosa canina* distilled water significantly increased motility and sperm count of the animals compared to the control group, it could not improve effectively the sperm parameters when co-administered in group T.

**Effect of tamoxifen and *Rosa canina* distilled water on the tissue structure of livers:** The Microscopic view of the animal's liver is shown in Fig. 1. The group treated with tamoxifen and *Rosa canina* distilled water (T+R) showed the hepatic architecture is rearranging with respect to the repositioning of hepatic cords. There was also minimal sinusoid distension remained around hepatic venues. Receiving *Rosa canina* distilled water had no effect on the liver histology.

Table 8: Effect of treatment with TAM and *Rosa canina* on sperm parameters (%) in rats

| | Groups | | | | |
|---|---|---|---|---|---|
| Factors | C | T | T+R | R | p-value |
| Motility | 45.20±5.97 | 35.00±5.00* | 30.83±7.36* | 51.33±2.94$^{\#}$ | 0.000 |
| Viability | 68.33±2.58 | 59.20±1.78** | 55.00±4.47* | 62.50±5.24$^{\#\#}$ | 0.000 |
| Morphology | 75.83±2.04 | 66.60±4.21** | 64.17±3.76** | 68.83±6.64 | 0.002 |
| Count | 20.33±1.75 | 18.40±2.51$^{\dagger}$ | 17.00±3.52$^{\dagger}$ | 36.33±3.14**$^{\dagger}$ | 0.000 |

All values are expressed as mean±standard deviation, p-value means difference in a row, *Significant difference at p<0.05 as compared to C, R, $^{\#}$Significant difference at p<0.05 as compared to T and T+R, **Significant difference at p<0.05 as compared to C, $^{\#\#}$Significant difference at p<0.05 as compared to T+R, $^{\dagger}$Significant difference at p<0.05 as compared to R

## DISCUSSION

Tamoxifen is used to treat breast cancer and is associated with the adverse side effects in cardiovascular system, bone turnover and liver metabolism. Toxicity and adverse effects of tamoxifen on hepatic cells have been proved by several studies by Li *et al.*[19] and Desai *et al.*[20]. Cytotoxicity of tamoxifen on primary human cells from breast cancer has been reported by Abbasalipourkabir *et al.*[21]. Tamoxifen induces triacylglycerol accumulation in the liver by increasing fatty acid and triglycerides synthesis[7,22]. The amount of visceral and liver adipose tissues in the patients treated with tamoxifen is higher than individuals that do not consume tamoxifen[23].

Besides, it is suggested that hepatocyte steatosis-induced tamoxifen is happened by increased fatty acid synthesis through enhanced gene expression of SREBP-1c and target genes (FAS, ACC and SCD)[24].

Tamoxifen can induce non-alcoholic steatohepatitis by increasing fatty acid synthesis via increased focal fatty sparing (FFS), as well as decreasing VLDL secretion from the liver[25]. In the rats, tamoxifen treatment can induce fatty liver by increasing fatty acid synthesis following activation of acetyl coenzyme A carboxylase (ACC), adenosine monophosphate and protein kinase (AMPK)[22].

Oxidative stress contributed to tamoxifen[26] is characterized by an increased level of malondialdehyde (MDA) as a lipid peroxidation marker. Significantly decreased level of MDA following distilled *Rosa canina* distilled water treatment indicating the protective effect of *Rosa canina* distilled water against lipid peroxidation and suggests the antioxidative effect of this plant. The antioxidative effect of *Rosa canina* distilled water is funded not only to vitamin C but also to phenolic compounds such as pro-anthocyanins and flavonoids[27]. Administration of 80% aqueous-alkali extract of fruit and seed of *Rosa-canina* significantly decreased body weight and visceral fat without affecting food intake in mice within 2 weeks[28]. The HESA-A (Marine-Herbal) composition used as medicine to treat non-alcoholic fatty liver has no side effects and complicated therapeutic effects compared with atorvastatin[29]. It was reported that green tea (*Camellia sinensis*) extract has an antioxidant effect and improves tamoxifen-induced liver injury in rats[30]. In addition, numerous researches evaluated the effect of plant extracts on liver function of animal models of non-alcoholic fatty liver disease[31,32]. Previous studies reported the hepatoprotective effects of other herbal extracts such as Silybum marianum and *Cichorium intybus*[33].

These results showed that *Rosa canina* distilled water has significant effects on the reduction of lipid accumulation in serum and TAC level as well as liver and kidney function improvement indicating the antioxidant effect. The results also revealed a rise in liver enzymes, ALT, AST and ALP, indicating the role of tamoxifen in fatty liver induction. Changes in lipid accumulation, including triglyceride, cholesterol, HDL, LDL and VLDL, also prove the effects of tamoxifen on liver function. These changes improved with *Rosa canina* distilled water treatment. Furthermore, the effect of *Rosa canina* distilled water on sperm analysis factors including sperm count, morphology, viability and motility was investigated. The results indicate a significant increase in sperm count following *Rosa canina* distilled water extract treatment. Besides, *Rosa canina* distilled water can reduce the increased level of total serum protein concentration.

In this study, the histopathological results showed that *Rosa canina* distilled water extract had no adverse effect on the liver histology that confirmed the obtained results. Centrilobular hepatocytes with spherical shapes (arrows in Fig. 1-T1) around central venule (star) showing mild microvesicular steatosis, also formation of small granulomatous like masses (arrows in Fig. 1-T2) which are micronodular regeneration of parenchymal hepatocellular loss (inset) and mild hyperplasia of bile ductular epithelium at the portal triad (arrow in Fig. 1-T3) and dissociation of hepatic cords along with distention of sinusoids (arrowhead in Fig. 1-T3), the distorted microscopic architecture of the centrilobular area with the disintegration of hepatocytes (arrow in Fig. 1-T4) and focal necrosis and inflammation surrounded by mononuclear inflammatory cells (star in Fig. 1-T4) and disarrangement of lobular architecture at portal triad with polygonal hepatocytes (arrows in Fig. 1-T5)

and pyknotic nuclei representing primary stages of apoptosis (arrowheads in Fig. 1-T5) exhibited the adverse effect of tamoxifen on the liver of the animals. The H and E staining of the liver structure of the animals treated with tamoxifen and *Rosa canina* distilled water showed that the hepatic architecture is rearranging with respect to the repositioning of hepatic cords. It can be concluded there was minimal sinusoid distension remained around hepatic venues as shown in the Fig. 1, T+R.

Because of short-term study, the histological results revealed an insignificant effect of tamoxifen on the hepatic histology suggesting that further studies are required to evaluate the toxic effect of tamoxifen of liver histology.

## CONCLUSION

The rats treated with tamoxifen showed a higher level of liver enzymes, lipid peroxidation and urea compared with the control group. Furthermore, decreased sperm motility and viability were shown due to tamoxifen treatment. The results of current study revealed that *Rosa canina* distilled water improved the disturbed liver enzyme, serum biochemical parameters, stress oxidative indexes and sperm analysis factors.

## SIGNIFICANT STATEMENT

This study discover the protective effect of *Rosa canina* distilled water that can be beneficial for toxicity-induced tamoxifen treated Wistar rats. This study will help the researcher to uncover the side effects of tamoxifen that many researchers were not able to explore. Thus a new theory on most powerful medicinal plants and herbs may be arrived at.

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