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Research Article

Effect of alcoholic and nano-extract of red radish, *Raphanus sativua* on liver and kidney function in rats induced jaundiced

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Abstract
This study was conducted to investigate the effect of the alcoholic and nano-extract of red radish (*Raphanus sativua*) on the function of the liver and kidneys in rats that had experimentally induced jaundice. Forty-eight white rats were divided into six groups: the control group dosed with physiological solution (0.9% Nacl), the T1 in which jaundice was induced, T2, the group of animals that treated with 200mg/kg of plant extract, T3, the group that dosed the nanocomposite of the plant extract at a dose of 200mg/kg daily, T4, the group that jaundice was induced and dosed with the normal extract of the plant, T5, the group that jaundice was developed and the plant extract nanocomposite was dosed at a dose of 200mg/kg daily. After the 21-day experiment, the animals were sacrificed, and the measurements were performed on the liver and kidney. The results showed that the induced of jaundice in the T1 caused a significant (*P*<0.05) decrease in the average weight of the liver and kidney, while there was a significant increase Liverl enzyme (ALK, LST, AST, urea and creatinine concentrations). The results also showed that the treatment of rats with hyperbilirubinemia with radish leaf extract group (T2) led to a significant (P<0.05) increase in liver and kidney weight and a significant decrease in liver enzymes (ALK, ALT, and AST,) and urea and creatinine concentration, compared with the T1. In the group treated with the red radish leaf nano-extract, the results showed improvements in the studied parameters. The histopathological changes were as hemorrhage, necrosis, rupture of liver tissue, glomerular atrophy, necrosis, disintegration and some inflammatory cells in the kidney tissue. It was concluded that the red radish leaf nanocomposite effectively improves the negative changes associated with hyperbilirubinemia.

Keywords: Nano-extract, red radish, liver, kidney, Rats, jaundice.

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Introduction

In many societies, jaundice is the most common disease of neonates and adults, which remains a major cause of brain damage, physical and mental disability, and premature death among newborns, and neonatal morbidity and mortality is still high in developing countries (Jabbar et al. 2019). Jaundice is the most common liver disorder, which means yellow discoloration of the skin and mucous membranes due to the high level of bilirubin in the blood (Sharma et al. 2012). It is observed during the first week after birth in about 60% of infants and 80% of premature infants (Jardn & Woodgate 2012). Jaundice is mainly seen in neonates and is uncommon in adults. However, it indicates a serious condition when it is present in adults (Memon et al. 2016).

Neonatal jaundice appears on the third day, and it reaches its peak on the 5-7th day and disappears by the 14th day, known as physiological jaundice. It is considered a normal physiological condition that passes undiagnosed, but the problem is because of the high level of unconjugated bilirubin, which is a neurotoxic substance, that its accumulation in a child's brain leads to a serious condition called kernicterus. It may cross the blood-fibrous barrier, causing damage to the auditory nerve and the basal ganglia. Consequently, the child will develop cerebral palsy and mental retardation (AAP 2004). Also, the high levels of bilirubin in adults is a problem that can cause many physiological changes that would cause many damages to the health of the individual.

One of the medically important plants is radish, *Raphanus sativus* L, which belongs to the family Brassicaeae, and genus *Brassica*. This family includes more than 330 genera and approximately 3,700 species (Wolfaging 2009). Egypt is considered the origin of its cultivation (Jan & Bader 2012). It is widespread in Japan and Southeast Asia (Agarwel & Varma 2014), and grows in temperate regions, mountainous areas, and northern plains (Arauna et al. 2012).

The radish is a multi-use plant, as its different parts are used in many foods, therapeutic and industrial fields. It has high economic importance, as the leaves are used as a food material because it contains a high percentage of proteins, amino acids, carbohydrates, and minerals such as calcium, potassium, magnesium, copper, and iron (Ibrahim et al. 2016). Thus it is used to treat malnutrition diseases related to digestive pain and various stomach diseases. It is also used as a diuretic to treat various urinary system problems (Alqasoumi et al. 2008). The radish plant is a good source of vitamin C, so it has a role in detoxifying the liver, jaundice and treating gynecological disorders and high blood pressure (Agarwel & Varma 2014; Mohammed & Qasim 2021).

The development of nanotechnology in the twentieth century, which is considered one of the latest technologies, had an important role in overcoming many of the difficulties facing medicines in reaching their target, through improving absorption and solubility, increasing stability and therapeutic efficiency, as well as delivering medicines to the target sites as well as mitigating its side effect caused by medications (Ochekpe et al. 2009). The current study aimed to study the alcoholic and nano-extract of red radish on the function of the liver and kidneys in rats that had experimentally induced jaundice.

Materials and Methods

The plant leaves were used after being washed with water to remove dirt and dust stuck and left in the shade at room temperature to dry. The extraction was carried out in the Molecular Research Laboratory of the College of Education, University of Al-Qadisiyah. After grinding the leaves using an electric mill and the obtained fine powder was used for the extraction, according to Gebrehiwot et al. (2019). The modified method of Kolekar et al. (2011) was used to prepare the hybrid nano-drug by adding 50ml of alcoholic extract of radish leaves drop by drop to 50ml of zinc oxide solution. The results were confirmed by Infrared (FTIR), X-ray diffraction (XRD), atomic force microscopy (AFM) and scanning electron microscopy (SEM).

Experimental animals: Natural albino rats weighing 170-190g were obtained from the College of Veterinary Medicine, University of Al-Qadisiyah, ranging from 6-8 weeks. The animal used in the experiment (48) were divided into eight groups, i.e. each treatment with eight animals, as follows (1) control group: the normal saline physiological solution was used for the 21-day experiment, (2) T1: the group induced jaundice by Phenylhdrazine hydrochloride at a dose of 75mg/kg (Mejia et al. 2008) for two days, (3) T2: a group of animals that dosed the alcoholic extract of red radish leaves at a concentration of 200mg/kg of body weight (Anwar et al. 2018), (4) T3: the group of animals that dosed the alcoholic extract of nano-loaded red radish leaves at a dose of 200mg/kg of body weight for 21 days, (5) T4: the group of animals in which jaundice was induced and dosed with the usual extract of red radish leaves at a dose of 200mg/kg of body weight for 21 days, and (6) T5: The group of animals in which

Table 1. The effect of the alcoholic extract and the red radish leaf nanocomposite on the rate of the liver and kidneys in jaundice induced rats.

Parameters	Liver weight ratio	Average total weight ratio
C	5.73±0.56[b]	0.49±0.0.03[b]
T1	4.08±0.55[c]	0.36±0.04[b]
T2	6.76±0.2[a]	0.42±0.01[a]
T3	7.06±0.94[a]	0.44±0.02[a]
T4	4.99±0.29[b]	0.42±0.02[a]
T5	5.62±0.32[b]	0.46±0.04[a]
LSD	0.89	0.07

The numbers indicate the mean ± standard error, C: The control group that dosed the physiological saline solution for the duration of the experiment (21 days), T1: The first treatment represents the group of rats in which jaundice was induced by being dosed with phenylhydrazine, T2: The second treatment represents the group of rats that were dosed with alcoholic extract of red radish leaves for the duration of the experiment, T3: The third treatment represents the group of rats that were dosed with a nano-alcoholic extract of radish leaves for the duration of the experiment, T4: The fourth treatment represents the group of rats in which jaundice was induced and dosed with alcoholic extract of red radish leaves for the duration of the experiment.T5: The fifth treatment represents the group of rats in which jaundice was developed and dosed with red radish leaf nano extract for the duration of the experiment. The different letters indicate a significant difference between the groups (*P*<0.05).- Similar letters indicate that there are no significant differences between groups (*P*>0.05).

jaundice was induced and dosed with alcoholic extract of nano-loaded red radish leaves at a dose of 200mg/kg of body weight for 21 days.

Sacrifice animals and draw blood: At the end of the experiment, the animals were weighed and then sacrificed by anesthetizing with chloroform. The blood samples were drawn from all groups, and placed in sterile 10ml tubes free of anticoagulant to obtain serum. After that, serum was obtained by centrifugation at 2500rpm for 15min, then the serum was placed in special tubes and kept at -20°C for the measuring biochemical parameters.

Laboratory tests: The aminotransferase enzymes AST and ALT activity was measured by the colorimetric method based on Reitman & Franke (1957). The enzyme activity is measured according to Belfeld & Goldberg (1971), and functional tests of the kidney were done according to Henry (1974) and Pation & Crpuch (1977).

Histological preparations: Autopsy of the samples were done from the liver and kidneys in different groups and fixed in 10% formalin. Then, histological slides were prepared based on Presnell & Schreibman (1997).

Statistical analysis: The statistical analysis was done by SPSS V.25. The data treatments were compared using the one-way ANOVA test. The means of the trial groups were compared when the differences between them were significant using the Least Significant Difference (LSD) at the 0.05 level of significance.

Results and Discussion

Liver weight ratio: A significant decrease (P<0.05) in the mean percentage of liver weight in the T1 and T4 compared to other treatments was observed in agreement with the findings of Matsumoto et al. (2016) and Allahmoradi et al. (2019). The phenylhdrazine is a toxic substance that causes cell damage, and hepatotoxicity (Valenzuela et al. 1987) and also hyperbilirubinemia (Nawaz et al. 2016) and oxidative stress also lead to hepatotoxicity, which is formed due to the accumulation of iron entering liver cells as a result of high levels of ROS, causing damage to hepatic membranes lipids, proteins and DNA (Basu et al. 2018). Phenylhydrazine causes liver tissue damage and causes oxidative stress in the liver and elevated liver enzymes (lbrahim et al. 2010), and liver damage that may explain the low liver weight ratio in this study.

In T2 and T3, which dosed the alcoholic extract of the leaves of the regular and nano-radish plant, a significant increase in the average weight of the liver were observed, and these results agreed with the findings of the Kalantari et al. (2009), Sadeek (2011) and Dash et al. (2013). This can be explained by the

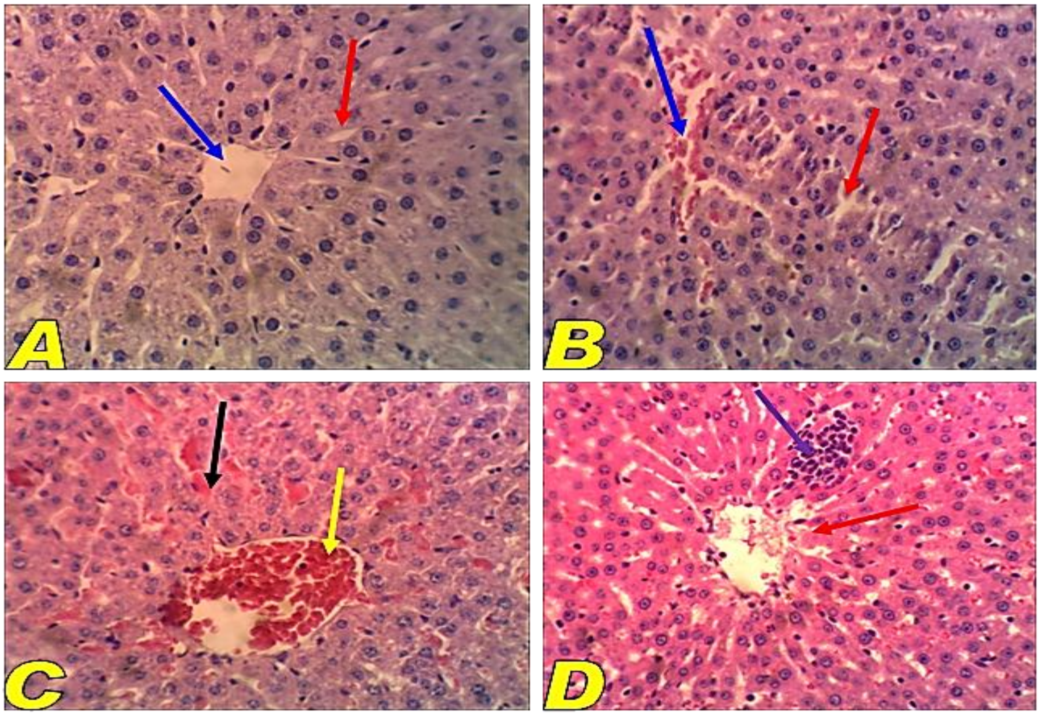

Fig.1. Cross sections of rat liver tissue: (A) represent the control group showing liver tissue is normal and retains the hexagonal geometric structure, including the blood sinusoids (red arrow) and the central vein (blue arrow), (B,C and D) a section of the liver tissue of a rat from the first treatment group (T1) showing hemorrhage (blue arrow) and disintegration and necrosis of the liver tissue (red arrow), and congestion in the central vein (yellow arrow) and blood sinuses (black arrow). Some inflammatory cells are collected (blue arrow). (H&E) (X40).

fact that the red radish extract prevented the depletion of the level of glutathione (Singh et al. 2009), and increased the production of an adequate amount of glutathione, which detoxifies phenylhydrazine through the interference of glutathione peroxides (GPX). This indicates that the radish extract works to stabilize the hepatocyte membrane, repair damaged tissue cells, and combat lipid peroxidation (Aniya & Anders,1985), and also radish contains glucosinolates and isothiocyanates that activate antioxidants and detoxify the liver (Scholl et al. 2011).

For aggregates of the nano-extracts, the results showed an increase in liver weight; these results were consistent with the finding of Yang et al. (2019). The red radish leaf nanocomposite improved liver function, rehabilitated the structure of the liver, protected it from inflammation and necrosis, and improved the concentration of enzymes. The liver effectively reduces the increase in ROS inside the liver, which is a potent activating factor in inhibiting liver fibrosis in rats and can deliver other therapeutic drugs within the different hepatocyte slits (Lin et al. 2016).

Kidney weight ratio: Significant decrease in kidney weight in the T1 compared to other treatments, which is caused by the toxic effect of phenylhydrazine, which is consistent with the study results of Anbara et al. (2018) and Zangeneh et al. (2019) which showed the toxic effect of phenylhydrazine. Hydrazine on the kidney tissue led to necrosis and degeneration, explains for the low weight of the kidney for this treatment, as well as the cause of deformation in the kidney tissue, congestion of glomeruli and blood vessels, and inflammatory infiltration of stem cells, which is a sign of acute tubular necrosis. Histological and functional impairment such as high urea and creatinine concentrations and electrolyte disturbances maybe the reason for the low weight ratio of the kidneys (Musyoka et al. 2016).

For other treatments that were dosed with the

Table 2. Shows the effect of alcoholic extract and the nanocomposite of red radish leaves on liver enzymes in rats with experimentally induced jaundice.

Parameters	ALK(IU/L)	AST (IU/L)	ALT (IU/L)
C	253±22.53[b]	151.33±15.88[c]	49.66±1.2[c]
T1	357±13[a]	243.66±51.89[a]	72.66±19.18[a]
T2	259.66±11.4[b]	156.29±83.81[c]	43.74±29.44[d]
T3	277.33±71.23[b]	158.66±24.18[c]	48±1.33[c]
T4	201.11±14.71[c]	195.5±0.57[b]	50.33±17.67[a]
T5	280.88±13.13[b]	191±37.5[b]	58.08±21.42[b]
LSD	52.77	6.76	2.21

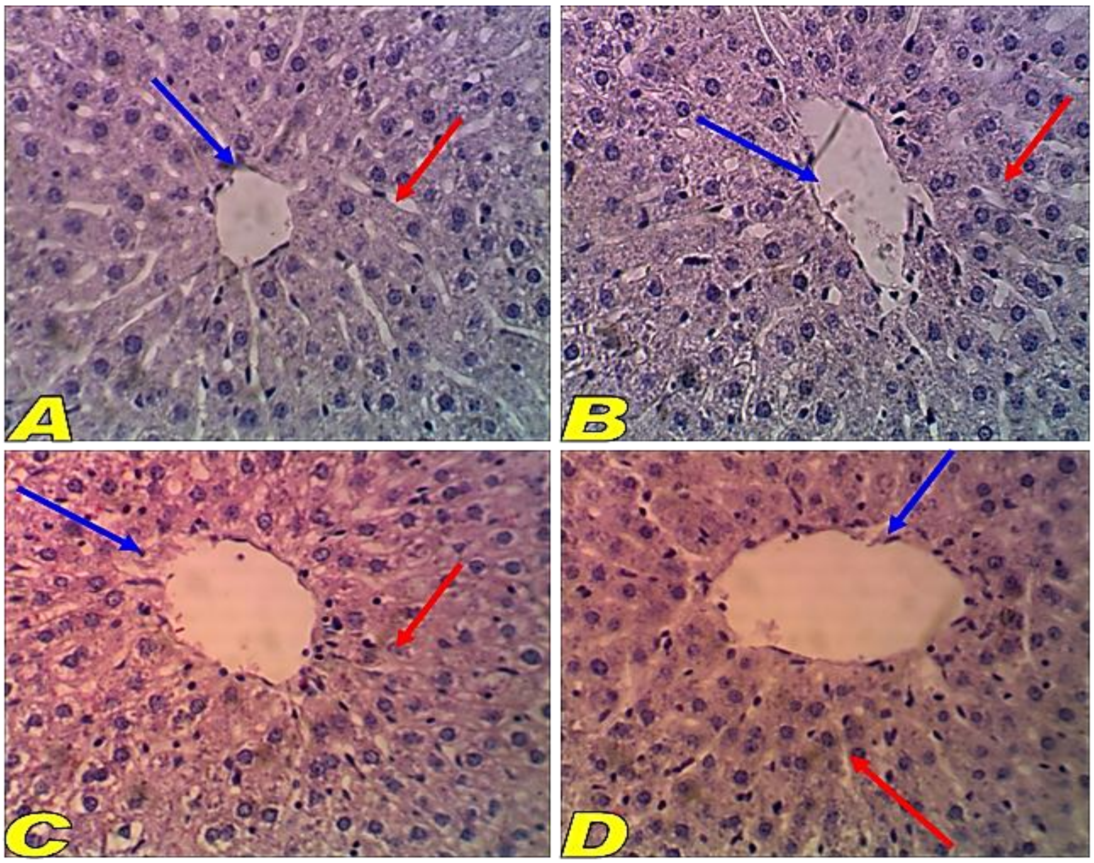

Fig.2. Cross sections of rat liver tissue: Images (A) represent the second treatment group (T2) liver tissue is normal and retains the hexagonal geometry, the blood sinusoids (red arrow) and the central vein (blue arrow). (B) liver tissue from the third treatment group (T3), showing normal liver tissue that retains the hexagonal geometry, between the blood pockets (red arrow) and central vein (blue arrow), (C) rat liver tissue from group The fourth treatment (T4) with normal liver tissue and retains its approximately hexagonal geometry with simple necrosis (blue arrow) and normal blood pockets (red arrow, (D) liver tissue from the fifth treatment group (T5) showing tissue The liver is normal and retains the hexagonal structure, between the blood sinusoids (red arrow) and the central vein (blue arrow) (H&E) (X40).

alcoholic extract of the leaves as the regular and nano radish, the results showed a significant increase ($P<0.05$) compared to T1, but these treatments revealed a convergence in the average weight of the kidney with the control and there was no significant ($P>0.05$). These results agreed with the findings of Aziz & Hassan (2020) and Ushakiran et al. (2017) showed that the radish extract reduces and prevents the formation of urinary stones. This mechanism is related to its diuretic effect and reduces urinary concentrations and thus prevents the formation of urinary stones (Aziz & Hassan (2020; Ushakiran et al. 2017). The radish may contribute and preventively against lipid peroxide in removing kidney damage.

Table 3. Shows the effect of the alcoholic extract and the nanocomposite of red radish leaves on kidney functions in rats that had experimentally induced jaundice.

Parameters	Serum creatinine concentration (gm/100ml)	The concentration of urea in the blood serum (gm/100ml)
C	$0.29{\pm}0.005^{b}$	$51.60{\pm}2.83^{b}$
T1	$1.365{\pm}0.005^{b}$	$56.36{\pm}0.08^{a}$
T2	$0.288{\pm}0.051^{b}$	$47.53{\pm}3.77^{c}$
T3	$0.286{\pm}0.001^{b}$	$48.23{\pm}5.45^{c}$
T4	$0.243{\pm}0.003^{b}$	$44.23{\pm}3.28^{d}$
T5	$0.265{\pm}0.011^{b}$	$0.96{\pm}0.08^{c}$
LSD	0.058	2.11

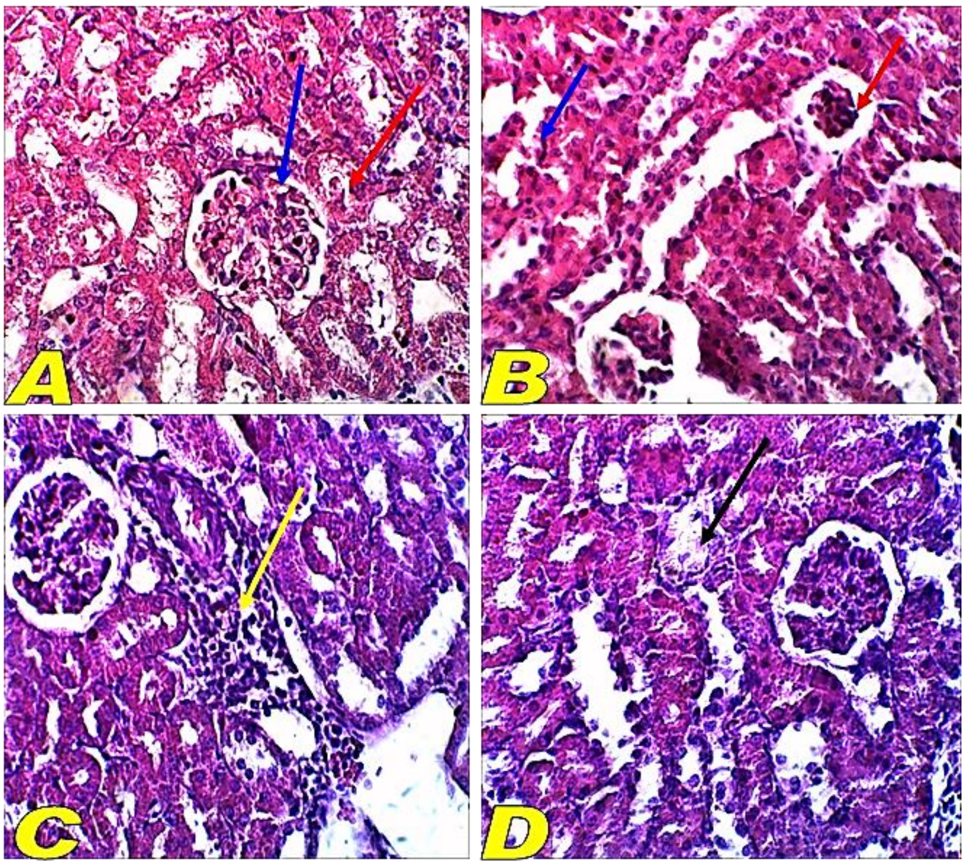

Fig.3. Cross section of the kidney tissue in rats: (A) control group showing the kidney tissue is normal and the glomerulus retains its normal structure (blue arrow) and the proximal tubules (red arrow). (B, C and D) kidney tissue from the first treatment group showing the disintegration of kidney tissue (blue arrow) and glomerular atrophy (red arrow), the accumulation of some inflammatory cells (yellow arrow) and necrosis in renal tubules (black arrow. (H&E) (X40).

Also, because the chemical components of radish leaves had a role, it is significant in protecting the kidneys and having an antioxidant activity (Mahjoubi-Samet et al. 2008).

In the treatments fed the nanocomposite, they showed an improvement in the weight of the kidney. This may be attributed to the role of the nanocarriers in enhancing the therapeutic index of the nanocomposite by increasing the bioavailability and biological efficacy while decreasing the side effects of phenylhydrazine (Zhang et al. 2010; Chavhan et al. 2013), and the enzyme activity was restored

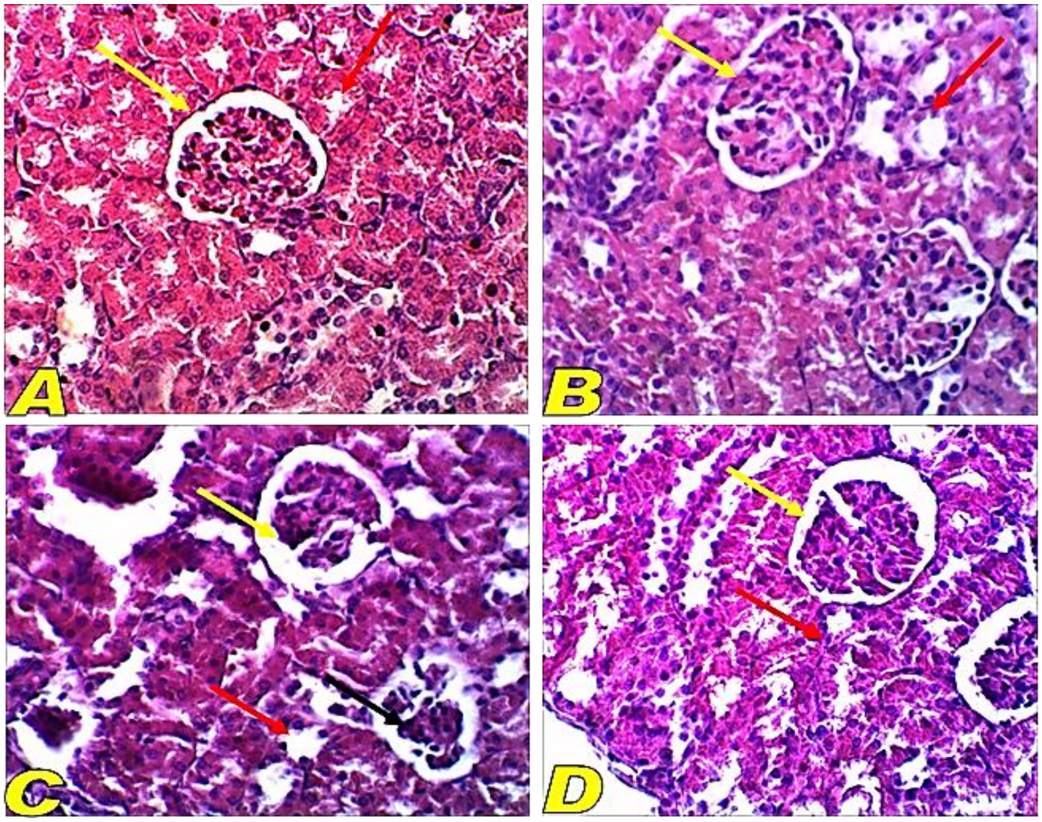

Fig.4. Cross section of rat kidney tissue: (A) the second treatment (T2) showing normal kidney tissue, normal glomerular (yellow arrow) and proximal tubules (red arrow). (B) the tissue of kidney from the third treatment (T3) showing normal kidney tissue, in which the glomeruli retain their normal shape (yellow arrow) and the proximal tubules (red arrow). (C) tissue from the fourth treatment (T4) showing atrophy of some glomeruli (black arrow) and slight necrosis of nearby tubules (red arrow). (D) liver tissue of a rat from the fifth treatment (T5) showing normal kidney tissue, the glomeruli (yellow arrow) and nearby tubules (blue arrow). (H&E) (X10).

through the action of nanoparticles on the regeneration of kidney and liver cells (Patrick-Iwuanyanwu et al. 2010).

Liver enzyme concentrations: Based on the results, a significant increase (P<0.05) in T1 about AST and ALTwas observed (Table 2). These results agreed with Allahmoradi et al. 2019) and Antai et al. (2009). The phenylhydrazine acts as a strong oxidant (Fibach & Rachmilewitz 2008), and the increase in these enzymes damage liver cells (John et al. 2012). When the cell membrane is broken in liver tissue, these enzymes, which are usually present in the cytosol, leak into the bloodstream (AL-Rawi & Maisaa 2007). The results can also be attributed to the increased production of free radicals, which leads to lipid peroxidation, causing cell damage, as a result of stimulating cytochrome P-450 in the liver to produce the highly reactive trichloromethyl free radical, which in the presence of oxygen generated by metabolic leakage from mitochondria, peroxidation of membrane lipids. Leading to a loss of integrity of cell membranes and damage to liver tissue with a subsequent increase in the level of ALT and AST enzymes (Arhan et al. 2009; Saki et al. 2011). Liver cells, leading to the destruction of these cells and the release of their contents, including ALT and AST enzymes, into the bloodstream.

In the T4 that dosed the alcoholic extract of the leaves of the regular radish, a significant decrease in the enzyme concentration rate was observed. These results agreed with the study of Chaturvedi (2008) and Vivarelli et al. (2016). Free radicals have demonstrated that polyphenols such as cinnapic acid and ferulic acid and flavonoids such as kaempferol present in radish leaf extract have antioxidant properties (Takaya et al. 2003), and this may be due to its hepatoprotective and anti-inflammatory effect (Beevi et al. 2010). The increase in the concentration of alkaline phosphatase enzyme ALP for the first treatment (T1) compared with the control and treatments agree with the study of Antai et al. (2009) and Zangeneh et al. (2019). Exposure to phenylhydrazine leads to significant damage to liver cells, and thus leakage of this enzyme from tissues to serum, due to oxidative damage to their membranes, which leads to membrane permeability (Appidi et al. 2009). In the T4 also a decrease was found in the concentration of the enzyme. The antioxidant property of phenols is due to the formation of the benzene ring, which is characterized by its ability to donate gamma electrons. The red radish has antioxidant properties containing higher levels of anthocyanins, dominated by pelargonidine derivative. This compound found in radish extract cleans the roots of phenols. 2,2-azino-bis(3-ethylbenzothiazoline-6-sulphonicacid) (ABTS+) and exercised the activity of removing free radicals, the chelation process hinders the formation of reactive oxygen species (Mazza 2018).

Histological changes in the liver: The results of examining the histological tissues of the liver in T1 showed histopathological changes as bleeding, necrosis and rupture, and congestion and accumulation of some inflammatory cells (Fig. 1B, C, D). This result agreed with the findings of Ashraf (2011) that indicated the appearance of bile duct obstruction, mild enlargement of Kupffer cells and a small number of hepatocytes showing a degenerative condition, and the reason could be that phenylhydrazine causes damage to liver tissue and causes oxidative stress in the liver and high enzymes (lBrahim et al. 2010)

The results T2 and T3 groups showed normal liver tissue compared to the control group (Fig. 2C, B) and these results agreed with the findings of Syed et al. (2014) and Youns et al. (2019) that showed an improvement in liver tissue after using the radish extract and repairing the damage caused by phenylhydrazine treatment. The reason can be attributed to the radish extract's ability to protect the liver. This effect is related to the antioxidant property present in the alcoholic extract of radish and that the radish extract works to protect the liver.

The improvement in the tissue sections of the T4 and T5 dosed with the regular and nano-extract of radish is inconsistent with the study of Abed-Al-Azeez et al. (2015). The use of radish extract reduces the hepatotoxicity caused by phenylhydrazine in albino rats by blocking lipid peroxidation that replenishes the levels of the non-protein sulfhydryl fraction (NP-SH) and enhances the detoxification system of the liver due to its sulfur and phenolic compounds that prevent the accumulation of free radicals (Rafatullah et al. 2008). The tissue sections dosed with the plant extract nano-loaded were less damaged than the other treatments affected by phenylhydrazine. It effectively improves the concentration of liver enzymes and reduces the increase in ROS inside the liver (Hassan et al. 2020; Kumar 2020; Bokov et al. 2022).

Histological changes in the kidney: The results of the histological study showed the histopathological changes represented by glomerular atrophy, necrosis, disintegration, and some inflammatory cells in the T1 (Fig. 3D, C, B). This result agreed with the study of Elaby & Ali (2018) and Henneh et al. (2021). The exposure of the kidney to a toxic substance leads to an increase in oxidative stress and thus an increase in ROS, which leads to functional impairment Glomerulosclerosis, tubular damage in the glomeruli, decreased renal filtration rate, and elevated urea and creatinine enzymes are evidence of nephrotoxicity (Hamman et al. 2016; El-Damaty et al. 2012). In addition, the results showed normal kidney tissue in the T2 and T3 (Fig. 4). The result agreed with the study of Sadeek et al. (2018). The group treated with radish extract showed a moderate gap in the epithelial lining of the renal tubules, and this can be attributed to the presence of a phenolic compound in the radish extract, especially folic acid, which is a strong antioxidant and is known for its positive effect on human health (Srinivasan et al. 2007). The T4 and T5 also showed an improvement in kidney tissue, which is consistent with the study of Al-Timimi et al. (2019) that showed the ability to resist oxidative stress, improve kidney tissue, and prevent nephrotoxicity caused by Gentamicin in laboratory animals.

Kidney function test: An increase in the kidney functions was observed in the T4 compared to (Table 3) other treatments that were consistent with studies of Kutshik et al. (2020) and Goorani et al. (2020). The phenylhydrazine The reason for the increase in reactive oxygen species and thus the increase of oxidative stress in the kidney tissues, which led to a change in kidney function that is associated with decreased blood circulation and the inability to excrete as a result of oxidative damage to the kidneys resulting from muscle cell damage (Debebe et al. 2017). Renal induced by phenylhydrazine resulted in impaired glomerular function, distension of Bowman's capsule, tubular damage to the kidneys, decreased glomerular filtration rate, lymphocytic infiltration of the renal cortex and cortical hemorrhage (Arthur et al. 2012).

The improvement in urea concentration in the T4 that was fed regular and nano radish leaf extract was consistent with the study of Al-Timimi et al. (2019). The radish containing vitamin C that protects against nephrotoxicity caused by Gentamicin and Vitamin C reduces cytoplasmic changes. Also, it decreased coagulation in renal tubules (Rehman et al. 2012), as it acts as a powerful antioxidant capable of absorbing reactive oxygen species and thus prevents kidney cell damage (Moreira et al. 2014). The extract of the leaves of the radish plant reduces and prevents the formation of kidney stones, as the mechanism is related to its effect as a diuretic and a decrease in urinary concentrations that form stones (Ushaliran et al. 2017).

For the creatinine concentration, all groups showed a significant decrease ($P<0.05$) compared to T1, but these treatments showed a convergence in creatinine concentration with the control and there was no significant difference ($P>0.05$) between them. This result was consistent with the study of Bojan et al. (2016). The radish extract has a protective effect on the kidneys because it contains a group of polyphenol compounds such as catechin, syringic acid, and cinnamic acid (Nowack et al. 2011), where this mechanism is associated with protecting the urinary system by reducing the activity of creatinine enzyme and increasing the activity of antioxidants SOD and CAT (Ozcan et al. 2007; Sindhu et al. 2015). In treatments fed the nanocomposite and the improvement occurred, it may be due to the nanocomposition. The extract of horseradish enhanced the drug targeting to the kidneys. It decreased the level of its enzymes (Petyaev 2015) by reducing the size of particles from the micro-scale to the nanometer scale and increasing the surface area (Patel et al. 2011), as the size of the designed particles has tremendous power in enhancing the availability district and the effectiveness of the nanocomposite in returning the concentrations of kidney enzymes to the normal level by changing the physicochemical properties of the nanocomposite that is less soluble in water (Chavhan et al. 2013).

References

Abed-Al-Azeez, L.A.; Ali, A.H. & Haba, M.K. 2015. Study the Protective Effect of Radish (*Raphanus sativus*) Seeds extract against harmful effects of sodium nitrite on some physiological and histological parameters in male rabbits. Iraqi Journal of Biotechnology 14(2): 59-78.

Agarwal, K. & Varma, R. 2014. Radical scavenging ability and biochemical screening of a common Asian vegetable-*Raphanus sativus* L. International Journal of Pharmaceutical Sciences Review and Research 27(1): 127-134.

American Academy of Pediatrics Subcommittee on Hyperbilirubinemia. 2004. Management of hyperbilirubinemia in the newborn infant 35 or more weeks of gestation. Pediatrics 114(1): 297-316.

Allahmoradi, M.; Alimohammadi, S. & Cheraghi, H. 2019. Protective effect of *Cynara scolymus* l. on blood biochemical parameters and liver histopathological changes in phenylhydrazine-induced hemolytic anemia in rats. Pharmaceutical and Biomedical Research 5(4): 53-62.

Alqasoumi, S.; Al-Yahya, M.; Al-Howiriny, T. & Rafatullah, S. 2008. Gastroprotective effect of radish *Raphanus sativus* L. on experimental gastric ulcer models in rats. Farmacia-Bucuresti 56(2): 204.

AL-Rawi, M. & Maisaa, M. 2007. Inhibitory Effect of Melatonin on Histological Changes Induced in Rat Liver by Thioacetamide Intoxication. Journal of Biological Sciences 7: 10-18

Al-Timimi, M.L.M.; Hassan, E.S.; Al-Nafakh, R.T.; Lteef Alfadhul, S.A. & Mohammad, A.R. 2019. Biochemical study of the protective effect of vitamin c and radish on gentamicin-induced nephrotoxicity in rats. Indian Journal of Forensic Medicine and Toxicology 13(4): 436-440.

Anbara, H.; Shahrooz, R.; Mozafari, A.A.; Malekinejad, H.; Morovvati, H.; Sheybani, M.T. & Saadati, S. 2018. Protective effects of ethyl pyruvate and vitamin E against phenylhydrazine-induced nephrotoxicity in mice. Yafteh 20(1): 52-67.

Aniya, Y. & Anders, M.W. 1985. Alteration of hepatic glutathione S-transferases and release into serum after treatment with bromobenzene, carbon tetrachloride, or N-nitrosodimethylamine. Biochemical Pharmacology 34(24): 4239-4244.

Antai, A.B.; Eyong, E.U.; Eteng, M.U.; Itam, E.H.; Eko, M.E. & Ita, S.O. 2009. Serum protein and enzyme levels in rats following administration of ethanolic leaf extract of *Ageratum conyzoides* (goat weed). Nigerian Journal of Physiological Sciences 24(2).

Appidi, J.R.; Yakubu, M.T.; Grierson, D.S. & Afolayan, A.J. 2009. Toxicological evaluation of aqueous extracts of *Hermannia incana* Cav. leaves in male Wistar rats. African Journal of Biotechnology 8(10): 23-29.

Arhan, M.; Öztürk, H.S.; Turhan, N.; Aytac, B.; Güven, M.C.; Olcay, E. & Durak, I. 2009. Hepatic oxidant/antioxidant status in cholesterol-fed rabbits: Effects of garlic extract. Hepatology Research 39(1): 70-77.

Arthur, F.K.; Woode, E.; Terlabi, E.O. & Larbie, C. 2012. Bilirubin lowering potential of Annona muricata (Linn.) in temporary jaundiced rats. Journal of Pharmacological and Toxicological Methods 7(2): 33-40

Aruna, G.; Yerragunt, V.G. & Raju, A.B. 2012. Photochemistry and pharmacology of *Raphanus* sativus.

Ashraf, N. 2011. Studies on Hummaz (*Oxalis corniculata* Linn.) for its hepatoprotective activity in rat models. Doctoral dissertation. 168 p.

Aziz, F.M. & Hassan, D.H. 2020. Radish juice promote kidney stone deposition in ethylene glycol-induced urolithiasis in rats. Cihan University-Erbil Scientific Journal 4(1): 57-61.

Basu, T.; Panja, S.; Shendge, A.K.; Das, A. & Mandal, N. 2018. A natural antioxidant, tannic acid mitigates iron-overload induced hepatotoxicity in Swiss albino mice through ROS regulation. Environmental Toxicology 33(5): 603-618.

Beevi, S.S.; Narasu, M.L. & Gowda, B.B. 2010. Polyphenolics profile, antioxidant and radical scavenging activity of leaves and stem of *Raphanus sativus* L. Plant Foods for Human Nutrition 65(1): 8-17.

Belfield, A. & Goldberg, D.M. 1971. Revised assay for serum phenyl phosphatase activity using 4-amino-antipyrine. Enzyme, 12: 561-573.

Bojan, M.S.; Rajappa, R.; Vijayakumar, D.R.K. & Gopalan, J. 2016. Protective effect of *Raphanus sativus* on D-galactosamine induced nephrotoxicity in rats. Nutrition Clinique et Métabolisme 30(1): 22-28.

Chaturvedi, P. 2008. Inhibitory response of *Raphanus sativus* on lipid peroxidation in albino rats. Evidence-Based Complementary and Alternative Medicine 5(1): 55-59.

Chavhan, S.; Joshi, G.; Petkar, K. & Sawant, K. 2013. Enhanced bioavailability and hypolipidemic activity of Simvastatin formulations by particle size engineering: Physicochemical aspects and in vivo investigations. Biochemical Engineering Journal 79: 221-229.

Debebe, M.; Afework, M.; Makonnen, E.; Debella, A.; Geleta, B. & Gemeda, N. 2017. Evaluations of biochemical, hematological and histopathological parameters of subchronic administration of ethanol extract of *Albizia gummifera* seed in Albino Wistar rat. Clinical Toxicology 7(1): 337.

Elaby, S.M. & Ali, J.B. 2018. The anti-anemic effect of dried beet green in phenylhydrazine treated rats.

Archives of Pharmaceutical Sciences Ain Shams University 2(2): 54-69.

El-Damaty, E.S.M.; Farrag, A.R.H.; Rowayshed, G. & Fahmy, H.M. 2012. Biochemical and histopathological effects of systemic pesticides on some functional organs of male Albino rats. Journal of Applied Sciences Research 5459-5469.

Fibach, E. & Rachmilewitz, E. 2008. The role of oxidative stress in hemolytic anemia. Current Molecular Medicine 8(7): 609-619.

Gebrehiwot, S.; Giday, M.; Erko, B. & Mekonnen, Y. 2019. In vivo antimalarial activity of Capp*aris tomento*sa Lam. in mice infected with *Plasmodium berghei*. Journal of Ayurvedic and Herbal Medicine 5(2): 44-48.

Goorani, S.; Shariatifar, N.; Seydi, N.; Zangeneh, A.; Moradi, R.; Tari, B. & Zangeneh, M.M. 2019. The aqueous extract of *Allium saralicum* RM Fritsch effectively treat induced anemia: Experimental study on Wistar rats. Oriental Pharmacy and Experimental Medicine 19(4): 403-413

Hamman, L.L.; Amaza D.S.; Zirahei, J.V.; Goji, A.D.T.; Mari, H. & Amali, F. 2016.Effect of aqueous extract of bitter leaf (*Vernonia amygdalina*) on phenyl-hydrazine induced kidney damage in albino rat. International Journal of Advanced Research 4(11): 39-47.

Hassan, S.A.; Hammam, O.; Hussein, S.A. & Aziz, W.M. 2020. The Conquering influence of the Nano extracts of pomegranate peels and pistachio leaves in amelioration of acute liver failure. BioRxiv 1-43.

Henry, R.J. 1974. Determination of serum creatinine. Clinical Chemistry: Principles and Techniques. 2nd ed. Harper and Row Publishers, New York. 552 p.

Ibrahim, M.N.; El-Messmary, M.F. & Elarfi, M.G. 2010. Synthesis of Spiro heterocyclic compounds. E-Journal of Chemistry 7(1): 55-58.

Ibrahim, L.F.; Elkhateeb, A.; Marzouk, M.M.; Hussein, S.R.; Abdel-Hameed, E.S.S. & Kassem, M.E. 2016. Flavonoid investigation, LC–ESIMS profile and cytotoxic activity of *Raphanus raphanistrum* L. (Brassicaceae). Journal of Chemical and Pharmaceutical Research 8(7): 786-793.

Jan, M. & Badar, A. 2012. Effect of crude extract of *Raphanus sativus* roots on isolated trachea of albino rat. Pakistan Journal of Physiology 8(1): 23-26.

Jabbar, M.Z. Hatam, A.A. & Hamad, H.A. 2019. Causes and Management of neonatal jaundice in almosawi pediatric hospital in Nasiryiah City: A retrospective study. University of Thi-Qar Journal of Medicine, 18(2), 120-127.

Jardine, L.A. & Woodgate, P. 2012. Neonatal jaundice. American Family Physician 85(8): 824-5.

John, O.R.; Yahaya, A.A. & Emmanuel, A. 2012. Aqueous ethanolic extract of *Mangifera indica* stem bark effect on the biochemical and haematological parameters of albino rats. Archives of Applied Science Research 4(4): 1618-1622.

Kalantari, H.; Kooshapur, H.; Rezaii, F. & Moosavi, M. 2009. Study of the protective effect of *Raphanus sativus* (radish) seed in liver toxicity induced by carbon tetrachloride in mice. Jundishapur Journal of Natural Pharmaceutical Products 4(1): 24-31.

Kolekar, T.V.; Yadav, H.M.; Bandgar, S.S. & Deshmukh, P.Y. 2011. Synthesis by sol–gel method and characterization of ZnO nanoparticles. Indian Streams Research Journal 1(1): 1-4.

Kumar, M. 2020. Synthesis, characterization of plant-mediated green nanoparticles and validation of medicinal impact on mammalian model. Ph.D. Thesis submitted to Ranchi University. 589 p.

Kutshik, R.J.; Jiyil, M.K.; Mafuyai, C.E. & Ichor, L. L. 2020. Anti-anaemic effect of *Cnidoscolus chayamansa* (Mc Vaugh) leaf extract on phenyl hydrazine-treated rats. Journal of Advances in Biology and Biotechnology 23-32.

Lin, T.T.; Gao, D.Y.; Liu, Y.C.; Sung, Y.C.; Wan, D.; Liu, J.Y. & Chen, Y. 2016. Development and characterization of sorafenib-loaded PLGA nanoparticles for the systemic treatment of liver fibrosis. Journal of Controlled Release 221: 62-70.

Mahjoubi-Samet, A.; Fetoui, H. & Zeghal, N. 2008. Nephrotoxicity induced by dimethoate in adult rats and their suckling pups. Pesticide Biochemistry and Physiology 91(2): 96-103.

Matsumoto, M.; Kano, H.; Suzuki, M.; Katagiri, T.; Umeda, Y. & Fukushima, S. 2016. Carcinogenicity and chronic toxicity of hydrazine monohydrate in rats and mice by two-year drinking water treatment. Regulatory Toxicology and Pharmacology 76: 63-73.

Mazza, G. 2018. Anthocyanins in fruits, vegetables, and grains. CRC Press. 215 p.

Mejia, G.B.; Sanz, C.R.; Avila, M.M.; Peraza, A.V.; Guzmán, D.C.; Olguín, H.J. & Cruz, E.G. 2008. Experimental hemolysis model to study bilirubin encephalopathy in rat brain. Journal of Neuroscience Methods 168(1): 35-41.

Memon, N.; Weinberger, B.I.; Hegyi, T. & Aleksunes, L.M. 2016. Inherited disorders of bilirubin clearance. Pediatric Research 79(3): 378-386.

Moreira, M.A.; Nascimento, M.A.; Bozzo, T.A.; Cintra, A.; da Silva, S.M.; Dalboni, M.A. & Higa, E.M. 2014. Ascorbic acid reduces gentamicin-induced nephrotoxicity in rats through the control of reactive oxygen species. Clinical Nutrition 33(2): 296-301.

Musyoka, T.M.; Dorothy, N.W.; Wycliffe, A.M.; Juma, K.K. & Nzioka, M.D. 2016. In vivo antianaemic effect and safety of aqueous extracts of *Erythrina abyssinica* and *Zanthoxylum usambarensis* in mice models. Journal of Hematology & Thromboembolic Diseases, pp. 2-10.

Nawaz, H.; Shad, M.A. & Iqbal, M.S. 2016. Optimization of phenylhydrazine induced hyperbilirubinemia in experimental rabbit. Experimental Animals 65(4): 363-372.

Nowack, R.; Flores-Suarez, F.; Birck, R.; Schmitt, W. & Benck, U. 2011. Herbal treatments of glomerulonephritis and chronic renal failure: Review and recommendations for research. Journal of Pharmacognosy and Phytotherapy 3(9): 124-136.

Ochekpe, N.A.; Olorunfemi, P.O. & Ngwuluka, N.C. 2009. Nanotechnology and drug delivery part 1: background and applications. Tropical Journal of Pharmaceutical Research 8(3).

Ozcan, A.; Atakisi, E.; Karapehlivan, M.; Atakisi, O. & Citil, M.E. 2007. Effect of L-carnitine on oxidative damage to liver, kidney and spleen induced by phenylhydrazine in mice. Journal of Applied Animal Research 32(1): 97-100.

Patel Anita, P.; Patel, J.K.; Patel Khushbu, S.; Deshmukh Aaishwarya, B. & Mishra Bharat, R. 2011. A review on drug nanocrystal a carrier free drug delivery. International Journal of Research in Ayurveda and Pharmacy 2(2): 448-458.

Patrick-Iwuanyanwu, K.C.; Onyeike, E.N. & Wegwu, M.O. 2010. Hepatoprotective effects of methanolic extract and fractions of African mistletoe *Tapinanthus bangwensis* (Engl. & K. Krause) from Nigeria. Excli Journal 9: 187.

Patton, C.J. & Crouch, S.R. 1977. Spectrophotometric and kinetics in- vestigation of the Berthelot reaction for the determination of ammonia. Analytical Chemistry 49: 464–469.

Petyaev, I.M. 2015. Improvement of hepatic bioavailability as a new step for the future of statin. Archives of Medical Science: AMS 11(2): 406.

Presnell, J.K.; Schreibman, M.P. & Humason, G.L. 1997. Huma son's animal tissue techniques. Johns Hopkins University Press. 512 p.

Rafatullah, S.; Al-Sheikh, A.; Alqasoumi, S.; Al-Yahya, M.; El-Tahir, K. & Galal, A. 2008. Protective effect of fresh radish juice (*Raphanus sativus* L.) against carbon tetrachloride induced hepatotoxicity. International Journal of Pharmacology 4(2): 130-134.

Rehman, K.; Akash, M.S.H.; Azhar, S.; Khan, S.A.; Abid, R.; Waseem, A. & Sherazi, T.A. 2012. A biochemical and histopathologic study showing protection and treatment of gentamicin-induced nephrotoxicity in rabbits using vitamin C. African Journal of Traditional, Complementary and Alternative Medicines 9(3): 360-365.

Sadeek, E.A. 2011. Protective effect of fresh juice from red beetroot (*Beta vulgaris* L.) and radish (*Raphanus sativus* L.) against carbon tetrachloride induced hepatotoxicity in rat models. African Journal of Biological Sciences 7(1): 69-84.

Sadeek, R.A.; Abd Elsabor, R.G. & Hassan, H.M. 2018. Potential protective effects of red radish (*Raphanus sativus*) in alleviation of health complications caused by ibuprofen 1 -10

Saki, F.; Hemmati, F. & Haghighat, M. 2011. Prevalence of Gilbert syndrome in parents of neonates with pathologic indirect hyperbilirubinemia. Annals of Saudi Medicine 31(2): 140-144.

Scholl, C.; Eshelman, B.D.; Barnes, D.M. & Hanlon, P.R. 2011. Raphasatin is a more potent inducer of the detoxification enzymes than its degradation products. Journal of Food Science 76(3): C504-C511.

Sharma, J.; Gairola, S.; Gaur, R.D. & Painuli, R.M. 2012. The treatment of jaundice with medicinal plants in indigenous communities of the Sub-Himalayan region of Uttarakhand, India. Journal of Ethnopharmacology 143(1): 262-291.

Sindhu, G.; Nishanthi, E. & Sharmila, R. 2015.

Nephroprotective effect of vanillic acid against cisplatin induced nephrotoxicity in wistar rats: a biochemical and molecular study. Environmental Toxicology and Pharmacology 39(1): 392-404.

Singh, D.; Singh, R.; Singh, P. & Gupta, R.S. 2009. Effects of embelin on lipid peroxidation and free radical scavenging activity against liver damage in rats. Basic and Clinical Pharmacology and Toxicology 105(4): 243-248.

Srinivasan, M.; Sudheer, A.R. & Menon, V.P. 2007. Ferulic acid: therapeutic potential through its antioxidant property. Journal of Clinical Biochemistry and Nutrition 40(2): 92-100.

Syed, S.N.; Rizvi, W.; Kumar, A.; Khan, A.A.; Moin, S. & Ahsan, A. 2014. In vitro antioxidant and in vivo hepatoprotective activity of leave extract of *Raphanus sativus* in rats using CCL4 model. African Journal of Traditional, Complementary and Alternative Medicines 11(3): 102-106.

Valenzuela, A.; Guerra, R. & Garrido, A. 1987. Silybin dihemisuccinate protects rat erythrocytes against phenylhydrazine-induced lipid peroxidation and hemolysis. Planta Medica 53(05): 402-405.

Vivarelli, F.; Canistro, D.; Sapone, A.; De Nicola, G.R.; Babot Marquillas, C.; Iori, R. & Paolini, M. 2016. *Raphanus sativus* cv. Sango sprout juice decreases diet-induced obesity in sprague dawley rats and ameliorates related disorders. PloS one 11(3): e0150913.

Zainab I.M. & Maytham T.Q. 2021. Hormonal profile of men during infertility. Biochemical and Cellular Archives 21: 2895-2898.

Wolfgang, K. 2009. Diplomarbeit. Jaranese medicinal plants used in rural communities. pp: 48-50.

Yang, H.Y.; Kim, K.S.; Lee, Y.H.; Park, J.H.; Kim, J.H.; Lee, S. Y. & Kim, H.S. 2019. Dendropanax morbifera ameliorates thioacetamide-induced hepatic fibrosis via TGF-β1/Smads pathways. International Journal of Biological Sciences 15(4): 800.

Olegovich Bokov, D.; Jalil, A.T.; Alsultany, F.H.; Mahmoud, M.Z.; Suksatan, W.; Chupradit, S. & Delir Kheirollahi Nezhad, P. 2022. Ir-decorated gallium nitride nanotubes as a chemical sensor for recognition of mesalamine drug: a DFT study. Molecular Simulation 1-10.

Younus, I.; Siddiq, A.; Baig, S.G.; Khan, S.S.; Ahmed, S. & Osama, M. 2019. Evaluation of pharmacological and toxic effects of ethanolic extract of radish pods in albino rabbits: A biochemical and histopathological study. Pakistan Journal of Pharmaceutical Sciences 32(3 (Suppl): 1275-1279.

Zangeneh, M.M.; Pooyanmehr, M. & Zangeneh, A. 2019. Biochemical, histopathological, and pharmacological evaluations of cutaneous wound healing properties of *Quercus brantii* ethanolic extract ointment in male rats. Comparative Clinical Pathology 28(5): 1483-1493.

Zhang, Z.; Bu, H.; Gao, Z.; Huang, Y.; Gao, F. & Li, Y. 2010. The characteristics and mechanism of simvastatin loaded lipid nanoparticles to increase oral bioavailability in rats. International Journal of Pharmaceutics 394(1-2): 147-153.