Journal of Pharmacy and Pharmacology 10 (2022) 79-86
doi: 10.17265/2328-2150/2022.03.003

Remediation of Hepatotoxic Effect of Tamoxifen with Ginger in Rat Model

Vanditha. S[1], Princy Louis Palatty[1], Malini Eapen[2], Thennavan. A[3], Manjeshwar. S. Baliga[4] and Greeshma. C. Ravindran[5]

1. Department of Pharmacology, Amrita School of Medicine, Amrita Vishwa Vidyapeetham, Kochi, India

2. Department of Pathology, Amrita School of Medicine, Amrita Vishwa Vidyapeetham, Kochi, India

3. Veterinary Medicine Department, Amrita School of Medicine, Amrita Vishwa Vidyapeetham, Kochi, India

4. Mangalore Institute Of Oncology, Mangalore, India

5. Department of Biostatistics, Amrita School of Medicine, Amrita Vishwa Vidyapeetham, Kochi, India

Abstract: Background: Tamoxifen is a non-steroidal oestrogen receptor modulator. It has strong anti-estrogenic activity on oestrogen receptor 2 in the breast tissue. It is the treatment of choice in breast cancer cases following surgery and radiotherapy, it is the most potent weapon against breast cancer. All stages of hormone-dependent breast cancer are currently treated with tamoxifen. Long-term tamoxifen therapy may cause hepatotoxicity which would perclude the use of tamoxifen. Hence this study was done to determine whether ginger has a role in protecting liver toxicity caused by tamoxifen. **Methods:** Twenty female albino rats (SD) were divided into 4 groups. Each group formed of five rats: Group I: Serves as control group received normal rodent diet and water, Group Ⅱ: Received ginger powder (200 mg/kg) dissolved in normal saline, orally daily for 6 weeks, Group Ⅲ: received TAM at a dose of 20 mg/kg daily for 6 weeks orally Group Ⅳ: Received TAM (20 mg/kg) then after 2 hrs they are orally given Ginger powder (200 mg/kg) daily for 6 weeks. **Results**: Tamoxifen 20 mg/kg/day caused hepatotoxicity in rats. When ginger is added to tamoxifen there is no development of hepatotoxic features like: marked sinusoidal congestion in the lobules and lab parameters showed decrease in serum AST and ALT levels when compared to tamoxifen group levels. **Conclusion:** Ginger supplementation would reduce the incidence of hepatotoxicity & related adverse drug reactions among breast cancer patient on therapy.

Key words: Breast cancer, tamoxifen, ginger, raised liver enzyme levels, hepatotoxicity, hepatic sinusoidal congestion.

1. Introduction

Breast cancer is a chapter in the life of many across the globe and is the second leading cause of cancer-related deaths in women [1]. Almost 12% of women worldwide are affected with breast cancer [2]. Worldwide, an estimate of 19.3 million new cancer cases and almost 10.0 million cancer deaths occurred in 2020 [1]. Some of the common features seen in patients with breast cancer are: advancing age, female gender, obesity, sedentary life [3], exposure to ionizing radiation, early menarche, alcohol intake and hormone replacement therapy..

Corresponding author: Princy Louis Palatty, MD, research field: pharmacology. Email: drprincylouispalatty@gmail.com.

Tamoxifen, non-steroidal oestrogen receptor modulator, is the most effective (endocraine therapeutic agent) treatment currently used for all stages of oestrogen receptor positive breast cancers [4, 5]. The Food and drug administration in 1977, approved the use of Tamoxifen in women with advanced breast carcinoma. By means of its antiestrogenic effect on oestrogen receptor-2, in the breast tissue, it can be used as a formidable weapon against breast cancer following surgery and radiotherapy [6]. Usually the common adverse effects occur with tamoxifen are hot flashes, weight gain, gynaecological symptoms such as vaginal dryness, discharge, pruritus vulvae, depression and memory loss [7]. Long-term use of tamoxifen is associated with a high risk of developing fatty liver

disease or non-alcoholic steato hepatitis, cirrhosis, and rarely acute liver injury in breast cancer patients which is usually associated with obesity, diabetes, metabolic syndrome and age factors [8, 9]. In a study by Gao et al, it was observed that tamoxifen (6 mg/kg/day) given for two weeks daily as injections causes hepatic toxicity with increases in transaminase activity and histopathological features like swollen hepatocytes with pyknotic nuclei [10].

Ginger (family-Zingiberaceae), the rhizome of *Zingiber officinale* Roscoe plant, is indisputably one of the most extensively used spice and culinary agent worldwide [11]. Ginger has time-tested therapeutic properties which has been extensively exploited till date. Ginger has free radical scavenging property [12]. It has also been shown to have a wide range of bioactivities, from antibacterial to anticancer effects. It has anti-bacterial, anti-cancer, anti-viral, gastric protection, anti-diabetes, cardiac protection, chemoprophylaxis and immune regulation effects [13, 14]. It has analgesic and anti-inflammatory properties also [15]. The hepatoprotective role of ginger against toxicity induced by CCl4, paracetamol [16] and Piroxicam [17] has been proved in rat study. The alleviation of tamoxifen induced hepatoticity is not studied using ginger. Hence this study was performed to determine whether ginger has a protective effect against TAM induced hepatotoxicity.

2. Methods

2.1 Selection and Description of Participants

2.1.1 Study design

This study was a prospective experimental animal study.

2.1.2 Study duration

2 Years.

2.1.3 Selection process

20 healthy adult female Albino rats were included in the study; they were divided into four groups and each containing five animals.

2.1.4 Inclusion criteria

Healthy adult female Albino rats, 10-12 wk old, weighing 150-170 g

2.1.5 Exclusion criteria

Pregnant rats, rats with illness (infections), rats with obvious deformities were excluded from the study.

2.1.6 Experimental animals

The animals used in the study were procured from the Central Lab Animal Facility (CPCSEA REG NO: 527/02/a/CPCSEA, dated 21/01/2002, Renewel No. 527/PO/ReRcBi–S/ReRc- L/02/CPCSEA, dated 13 /7/2021, Amrita School of medicine, AIMS, Kochi) They were housed in polypropylene plastic cages with metallic cage cover and a paddy husk bed with adequate food and water. There are five animals in each cage. They were kept in animal house under 12:12 hours light and dark cycle at 25 ± 2 ^{o}C and humidity and other micro and macro environmental conditions as suggested by the CPCSEA. They were allowed to have normal rodent pellet diet, purchased from Amrut feeds, Krishna Valley Agrotech Bangalore, which was supplied under hygienic conditions and were given water.

The study got approval on the IAEC meeting held on 29/5/2020, (Ref.no.: IAEC/2020/1/1). Experiments are designed and conducted according to the CPCSEA guidelines.

2.2 Chemicals and Drugs

2.2.1 Tamoxifen

Tamoxifen (tamoxifen citrate) tablets, trade name Cytopam, manufactured by Cipla. It was dissolved in distilled water and was given orally to the experimental animals by gastric tube at a dose of 20 mg/kg body weight (equivalent to therapeutic dose for human, according to Paget & Barnes 1964) daily for 6 consecutive weeks.

2.2.2 Ginger powder

Ginger powder, manufactured by Vanitha Herbals, (ISO certified). It was dissolved in normal saline and given orally at a dose of 200 mg/kg/day, by gastric gavages for 6 consecutive weeks.

2.3 Conduct of the Study

Animals are divided into four groups, each containing five animals.

Group 1 (Control group): Animals served as control group and given standard rodent diet and water only.

Group 2 (Ginger group): Animals were given Ginger powder (200 mg/kg) daily for 6 weeks orally using 18 G gastric gavage needle.

Group 3 (TAM group): Animals were given Tamoxifen at a dose of 20 mg/kg daily for 6 weeks orally using 18G gastric gavage needle

Group 4 (TAM followed by Ginger group): Animals were given orally Tamoxifen (20 mg/kg) then after 2 hrs they were given orally Ginger powder (200 mg/kg) daily for 6 weeks, using 18G gastric gavage needle.

2.4 Sample Collection

Blood samples were collected from the animals on the first day, at 3 weeks and one day after the last drug dose (at 6 weeks) from lateral tail vein of rat for biochemical analysis. Before sample collection, the rats will be anesthetized using 3% isoflurane by keeping the rats in an anesthesia chamber for not less than1 minute.

At the end of the six weeks, the animals were euthanized by keeping the mina carbon dioxide inhalational chamber with a flow rate of 3 liters/minute for 5 minutes. The abdomen were opened and the liver specimen were collected in 10% formalin and send for histopathological examination to reveal the toxic effect of Tamoxifen and the protective effect of ginger on tamoxifen induced hepatotoxicity.

2.5 Measurements and Data Collection

Biochemical Parameters for Liver Function: Blood analysis was performed by estimating liver enzymes; ALT and AST. These markers are measured by colorimetric method through specific kits according to manufacturer instructions.

Determination of Total Cholesterol & Triglyceride Serum Concentration: These markers were measured by colorimetric method through specific kits according to manufacturer instructions.

Histopathological Examination of Liver Specimen: The abdomen was opened, the liver dissected and then fixed in 10% neutral buffered formalin. After fixation, specimen were dehydrated in ascending series of ethyl alcohol, cleared in 3 changes of Xylene and infiltrated in three changes of molten paraffin wax (melting point of 58-60 ^{o}C) and then embedded in molten paraffin. Sections of 5 microns thickness were cut by using rotary microtome. For histological examination, sections are stained with H &E stains.

2.6 Statistics

Statistical analysis was carried out using IBM SPSS 20. (SPSS Inc, Chicago, USA). Descriptive statistics of both groups were expressed as mean ± SD and median (Q1-Q3) for continuous variables and frequency and percentage for categorical variables. To test the statistical significance of the comparison of median difference of numerical variables between groups, Mann Whitney U test was applied. To test the statistical significance of the comparison of median difference of numerical variables among the groups, Kruskal Wallis test was applied. Multiple comparison tests were done by using Bonferroni test. All statistical tests were two-sided and conducted in an explorative manner on a significance level of $p < 0.05$.

3. Results

In our study, we have included twenty adult female Albino rats (Sprague dawley), 10-12 wk old weighing 150-170 g. Animals were divided into four groups, each group containing five animals. Group 1 (Control group): received standard rodent diet, Group 2 (Ginger group): received ginger powder (200 mg/kg), orally daily for 6 weeks. Group 3 (TAM group) received tamoxifen 20 mg/kg daily for 6 weeks orally, Group 4 (TAM followed by Ginger group) received tamoxifen (20 mg/kg) then after two hours they are given orally Ginger powder (200 mg/kg) daily for 6 weeks. Oral

administration of Tamoxifen 20 mg/kg/day for 6 weeks causes hepatotoxicity as demonstrated by the significant increase in serum ALT and AST levels when compared to control group (Table 2). But there is no significant changes shown in serum cholesterol and triglycerides levels.

These biochemical results are confirmed by the histopathological results of the liver tissue, which shows marked sinusoidal congestion in the lobules (Figs. 1 and 2). The portal tract also shows dilated & congested portal veins. While administration of ginger followed by tamoxifen caused significant decrease in serum ALT, AST, total cholesterol and triglyceride levels when compare to tamoxifen group at 6 weeks. Section from liver of a rat treated Tamoxifen followed by Ginger for six weeks also shows preserved Lobular architecture with minimal focal sinusoidal congestion (Fig. 3). From these observations, we can draw the conclusion that Ginger supplementation would reduce the hepatoxicity induced by tamoxifen.

Table 1 Effect of Ginger (200 mg/kg/day), TAM (20 mg/kg/day) either singly or in combination on liver enzymes, total cholesterol and triglycerides in all studied group (mean ± SD) and Median (Q1-Q3) at 3 weeks.

Parameter		Group			
		Control mg/kg/day	Ginger 200 mg/kg/day	TAM 20 mg/kg /day	TAM 20 mg/kg/day + Ginger 200 mg/kg/day
Alanine Transaminase (U/L)	Mean ± SD	47.50 ± 3.57	50.20 ± 5.10	63.940 ± 4.354	51.860 ± 5.094
	Median (Q1-Q3)	46.4 (45.5-48.9)	53.6 (46.0-54.0)	64.9 (63.4-66.0)	50.9 (50.2- 51.4)
Aspartate transaminase (U/L)	Mean ± SD	97.867 ± 8.168	103.420 ± 12.342	102.580 ± 6.704	100.92 ± 5.295
	Median (Q1-Q3)	98.5 (93.9-102.1)	107.9 (102.1-108.3)	103.700 (96.7-107.3)	102.5 (95.8-104.3)
Total cholesterol concentration (mg/dL)	Mean ± SD	58.300 ± 15.014	57.440 ± 6.354	44.380 ± 4.688	39.180 ± 3.995
	Median (Q1-Q3)	63.4 (52.4-66.7)	55.10 (52.8-63.8)	45.6 (40.6-45.70)	40.2 (36.3-41.5)
Triglyceride concentration (mg/dL)	Mean ± SD	51.600 ± 6.609	66.560 ± 8.843	60.120 ± 13.860	58.200 ± 11.307
	Median (Q1-Q3)	54.8 (49.4-55.4)	66.7 (61.7-72.5)	56.3 (48.4- 74.5)	65.1 (46.2- 66.9)

Table 1 showed that tamoxifen administration caused significant increase in serum ALT levels (p = 0.033) when compared to the control group at 3 weeks. While administration of ginger in tamoxifen intoxicated rats caused significant decrease in serum ALT when compared to TAM groups (p = 0.016) at 3 weeks

Table 2 Effect of Ginger (200 mg/kg/day), TAM (20 mg/kg/day) either singly or in combination on liver enzymes, total cholesterol and triglycerides in all studied group (mean ± SD) and Median (Q1-Q3) at 6 weeks.

Parameter		Group			
		Control mg/kg/day	Ginger 200 mg/kg/day	TAM 20 mg/kg /day	TAM 20 mg/kg/day + Ginger 200 mg/kg/day
Alanine Transaminase (U/L)	Mean ± SD	54.967 ± 5.613	46.220 ± 3.894	70.280 ± 7.056	47.700 ± 5.610
	Median (Q1-Q3)	54.0 (51.9-57.5)	46.0 (44.0-48.8)	70.7 (64.2-74.3)	46.5 (44.4-47.2)
Aspartate transaminase (U/L)	Mean ± SD	94.267 ± 1.617	107.920 ± 9.875	124.160 ± 3.715	98.640 ± 4.599
	Median (Q1-Q3)	94.0 (93.4 -95.0)	106.0 (104.6-114.0)	123.7 (122.4-124.5)	99.9 (94.4-100.2)
Total cholesterol concentration (mg/dL)	Mean ± SD	64.133 ± 14.500	60.280 ± 12.160	47.940 ± 6.071	38.280 ± 3.891
	Median (Q1-Q3)	72.0 (59.7-72.5)	62.6 (50.6-66.5)	49.1 (45.4- 51.8)	36.9 (36.3- 40.2)
Triglyceride concentration (mg/dL)	Mean ± SD	66.100 ± 10.860	68.780 ± 14.878	93.300 ± 21.719	59.480 ± 10.627
	Median (Q1-Q3)	69.3 (61.6-72.1)	72.4 (67.5-77.5)	100.40 (86.0-104.7)	65.1 (48.5- 67.3)

Table 2 showed that tamoxifen treatment caused significant increase in serum ALT (p = 0.025) and AST levels (p = 0.025) when compared to control group at 6 weeks. While administration of ginger followed by tamoxifen caused significant decrease in serum ALT (p = 0.009), AST (p = 0.009), total cholesterol (p = 0.028) and triglyceride levels (p = 0.047) when compare to tamoxifen at 6 weeks.

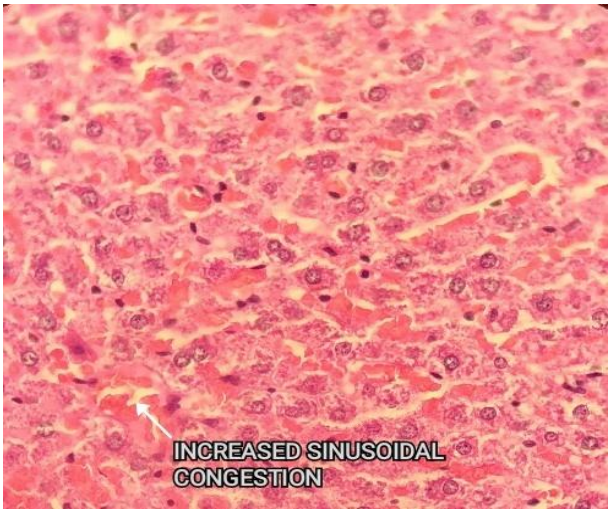

Fig. 1 Section from liver of a rat treated with tamoxifen for six weeks showing preserved Lobular architecture but there is marked sinusoidal congestion in the lobules.

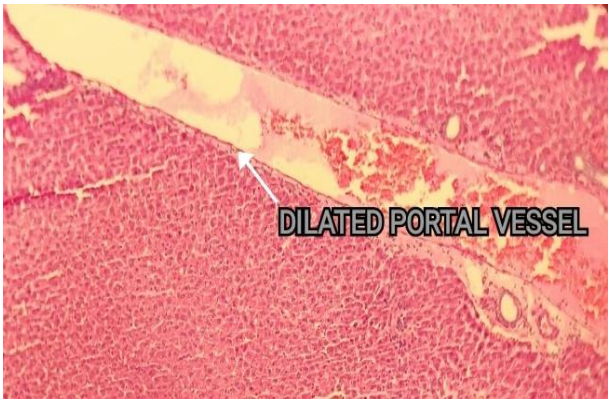

Fig. 2 Section from liver of a rat treated with tamoxifen for six weeks showing dilated and congested portal vessels.

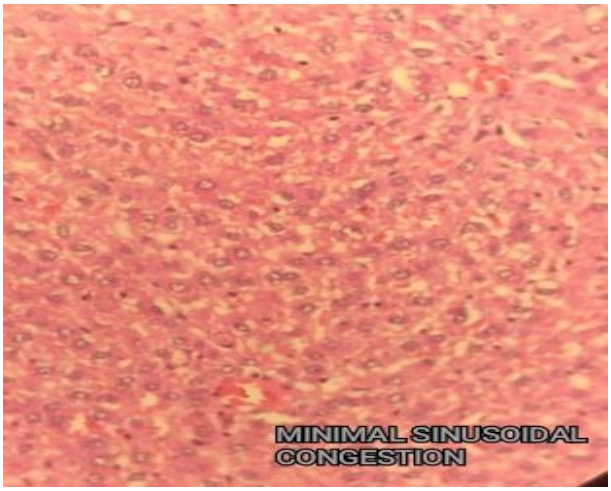

Fig. 3 Section from liver of a rat treated Tamoxifen followed by Ginger for six weeks showing preserved Lobular architecture with minimal focal sinusoidal congestion.

4. Discussion

In this study, it was observed that oral administration of Tamoxifen 20 mg/kg/day for six weeks caused statistically significant elevation in Serum concentration of ALT ($p = 0.025$) and AST levels ($p = 0.025$) without any substantial increases in serum concentration of cholesterol or triglyceride levels. Similar results were observed in the study done by Gao et al. The elevated liver enzymes could be caused by hepatic structural damage, which is due to the release of liver enzymes from the cytoplasm of the hepatocyte into the blood [10].

The liver section of female albino rats treated with Tamoxifen 20 mg/kg for six weeks showed intense hepatotoxic changes in the histopathology. Section from liver of rats treated with tamoxifen for six weeks shows dilated and congested portal vessels and the portal tract shows ectatic portal veins. The lobular architecture of the hepatic tissue is preserved, but there is marked sinusoidal congestion in the lobules (Vide Fig. 1). Similar results have been observed by Morsy et al [18] & Abd El- Mawla and Osman [19] in their study. Pressure of necrotic and inflammatory cells on portal vein tributaries [20], could cause hepatic sinusoids dilatation which may be a sign of portal hypertension [21]. In a previous study by Fang et al [22], divulged that DNA damage occurs in the early stages of tamoxifen therapy, which raises the likelihood of developing hepatocellular carcinoma in the long run.

The examination of animals treated with Tamoxifen 20 mg/kg/day followed by Ginger 200 mg/kg/day for 6 weeks shows that the deranged liver function are normalized as evidenced by lab parameters more over the histopathological situation is also beginning to revert back to normal. The serum levels of ALT & AST shows significant reduction with a p value of 0.009 when ginger is given along with tamoxifen. There is significant decrease in serum ALT levels ($p = 0.008$) & AST levels ($p = 0.010$) at 6 weeks among

Control group, TAM group and TAM + Ginger group. The study shows that, even at 3 weeks itself administration of ginger along with tamoxifen showed statistically significant decrease in serum ALT levels ($p = 0.016$).

The decrease in serum transaminases levels implies that hepatic damage induced by tamoxifen is ameliorated by ginger supplementation, by decreasing enzyme leakage from the cytoplasm. These outcomes are in accordance with previous study done by Hasan et al, by showing that there is significant decrease in serum ALT and AST levels in CCl4 induced liver toxicity with administration of ginger [23]. Ginger has also been shown to reduce the hepatotoxicity owing to cisplatin, according to previous researches. Antioxidant and anti-inflammatory properties are exhibited by most of constituents of ginger. Antioxidant property of ginger is mainly due to reduction in the formation of free radicals and the protective effect of ginger against DNA damage is due to gingerol which also has an oxygen radical scavenging property.

Moreover, there is significant reduction in level of serum cholesterol ($p = 0.028$) and triglycerides ($p = 0.047$) in rats those treated with tamoxifen along with ginger. Marked histological amelioration was observed in liver tissue of rat treated with tamoxifen along with ginger compared to tamoxifen group as most of the hepatocytes, appears to be normal with preserved lobular architecture with minimal focal sinusoidal congestion (Vide Fig. 3). These inferences are in line with previous study by Valko et al, which found that ginger can reduce lipid peroxidation and free radical production [24] thereby reducing the detrimental effect of free radicals. In their study, Masuda et al. found that gingerols (polyphenols) found in fresh ginger rhizome, has potent antioxidant properties [25]. Previous literature reviews covey that, ginger was found to deduce the histopathological alterations caused by cisplatin, paracetamol, and carbon tetrachloride [23, 26]. In their study, Galila et al validated these findings, noting that daily administration of 200 mg/kg body weight of crude ethanolic ginger extract in conjunction with CCl4 significantly reduced the hepatic fibrosis caused by CCl4 [27]. The study showed that the hepatoprotective activity of ginger is due to the presence of antioxidant compounds in ginger extract.

Thus we can draw the conclusion from this study that Ginger supplementation would reduce the incidence of hepatotoxicity & related adverse drug reactions among breast cancer patient on TAM therapy. The development of hepatotoxicity would perclude the use of tamoxifen. Addition of Ginger throughout therapy with tamoxifen would lead to the reduction in hepatotoxicity & symptoms of hepatic dysfunction. This is a transilational study which can be immediately advocated for ginger supplementation in tamoxifen treated patients.

5. Conclusions

Breast cancer is commonly treated with tamoxifen which is very effective drug that reduce mortality and has improved the quality of life. Tamoxifen is known to cause hepatotoxicity and consequent derangement of the liver function. Our quest for hepatoprotective agent that could be used concomitantly with tamoxifen, brought us to select the humble ginger as a probable solution. Hence, the protective effect of ginger was investigated on rats with tamoxifen induced hepatotoxicity. This study shows that administration of ginger significantly decreases the serum ALT, AST, cholesterol and triglyceride levels in tamoxifen induced rat models at 6 weeks. The present study shows that tamoxifen 20 mg/kg/day given for 6 weeks caused obvious hepatotoxicity as shown by lab parameters & histopathological results.

As tamoxifen therapy in breast cancer patients is recommended for 5 years, liver injury will be ascertain event. When Ginger is administered along with tamoxifen there is significant decrease in serum ALT and AST levels and it also ameliorates the

histopathological changes caused by tamoxifen. When findings in this animal study are extrapolated to the clinical scenario, it is expected that ginger reduces hepatic impairment and allows safe and effective continued use of tamoxifen affording breast cancer free period. Hence this is an immediate translation and the results will move from bench to bed.

References

[1] Sung, H., Ferlay, J., Siegel, R. L., et al. 2021. "Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries." *CA: A Cancer Journal for Clinicians* 71 (3): 209-249.

[2] McGuire, A., Brown, J. A., Malone, C., et al. 2015. ''Effects of Age on the Detection and Management of Breast Cancer." *Cancers (Basel)* 7 (2): 908-29.

[3] Colditz, G. A., et al. 2012. ''Family History and Risk of Breast Cancer: Nurses' Health Study." *Breast Cancer Res Treat* 133 (3): 1097-104.

[4] Desai, P. B., Nallani, S. C., Sane, R. S., et al. 2002. "Induction of Cytochrome P450 3A4 in Primary Human Hepatocytes and Activation of the Human Pregnane X Receptor by Tamoxifen and 4-hydroxytamoxifen." *Drug Metab Dispos* 30 (5): 608-12.

[5] Jordan, V. C. 2003. "Tamoxifen: A Most Unlikely Pioneering Medicine." *Nat Rev Drug Discov* 2 (3): 205-13.

[6] Jordan, V. C. 1988. "The Development of Tamoxifen for Breast Cancer Therapy: A Tribute to the Late Arthur L. Walpole." *Breast Cancer Res Treat* 11 (3): 197-209.

[7] Lorizio, W., Wu, A. H. B, Beattie, M. S., et al. 2012. "Clinical and Biomarker Predictors of Side Effects from Tamoxifen." *Breast Cancer Res Treat* 132 (3): 1107-18.

[8] Saphner, T., et al. 2009. "The Association of Nonalcoholic Steatohepatitis and Tamoxifen in Patients with Breast Cancer." *Cancer* 115 (14): 3189-95.

[9] Pan, H-J., Chang, H-T., and Lee, C-H. 2016. "Association between Tamoxifen Treatment and the Development of Different Stages of Nonalcoholic Fatty Liver Disease among Breast Cancer Patients." *J Formos Med Assoc* 115 (6): 411-7.

[10] Gao, F. F., Lv, J. W., Wang, Y., et al. 2016. "Tamoxifen Induces Hepa-totoxicity and Changes to Hepatocyte Morphology at the Early Stage of Endocrinotherapy in Mice." *Biomed Rep* 4 (1): 102-106.

[11] Baliga, M. S., Haniadka, R., Pereira, M. M., et al. 2012. "Radioprotective Effects of *Zingiber officinale* Roscoe (Ginger): Past, Present and Future." *Food Funct* 3 (7): 714-23.

[12] Maghsoudi, S., et al. 2011. "Preventive Effect of Ginger (*Zingiber officinale*) Pre-treatment on Renal Ischemia-reperfusion in Rats." *Eur Surg Res* 46 (1): 45-51.

[13] Baliga, M. S., Haniadka, R., Pereira, M. M., et al. 2011. "Update on the Chemopreventive Effects of Ginger and Its Phytochemicals." *Crit Rev Food Sci Nutr* 51 (6): 499-523.

[14] Ali, B. H., et al. 2008. "Some Phytochemical, Pharmacological and Toxicological Properties of Ginger (*Zingiber officinale* Roscoe): A review of Recent Research." *Food Chem Toxicol* 46 (2): 409-20.

[15] Jagetia, G. C., et al. 2003. "Influence of Ginger Rhizome (*Zingiber officinale* Rosc) on Survival, Glutathione and Lipid Peroxidation in Mice after Whole-body Exposure to Gamma Radiation." *Radiat Res* 160 (5): 584-92.

[16] Yemitan, O. K., and Izegb, M. C. 2006. "Protective Effects of *Zingiber officinale* (Zingiberaceae) against Carbon Tetrachloride and Acetaminophen-induced Hepatotoxicity in Rats." *Phytother Res* 20 (11): 997-1002.

[17] Badawi, M. S. 2019. "Histological Study of the Protective Role of Ginger on Piroxicam-induced Liver Toxicity in Mice." *J Chin Med Assoc* 82 (1): 11-18.

[18] Morsy, F. A., et al. 2010. "Histopathologic Study of the antiestrogenicnolvadex Induced Liver Damage in Rats and Vitamins Ameliorative Effect." *Nature and Science* 8 (5): 1-15.

[19] Abd El-Mawla, A, M, A, and Osman, H. E. H. 2012. "Role of Propolis in Improving the Histological and Ultrastructural Changes of Liver after Treatment with Tamoxifen." *Spatula DD* 2 (1): 35-42.

[20] Ferrell, l. 2000. "Liver Pathology: Cirrhosis, Hepatitis, and Primary Liver Tumors. Update and Diagnostic Problems." *Mod Pathol* 13 (6): 679-704.

[21] Vince, A. R., et al. 2014. "Hepatic Injury Correlates with Apoptosis, Regeneration, and Nitric Oxide Synthase Expression in Canine Chronic Liver Disease." *Vet Pathol* 51 (5): 932-45.

[22] Fang-Fang, G., Jia-Wei, L., Ying, F., et al. 2016. "Tamoxifen Induces Hepatotoxicity and Changes to Hepatocyte Morphology at the Early Stage of Endocrine Therapy in Mice." *Biomed Rep* 4 (1): 102-106.

[23] Hasan, I. H., El-Desouky, M. A., Hozayen, W. G., & Abd el Aziz, G. M. 2016. "Protective Effect of *Zingiber Officinale* against CCl4-Induced Liver Fibrosis Is Mediated through down regulating the TGF-β1/Smad3 and NF-κB/IκB Pathways." *Pharmacology* 97 (1-2): 1-9.

[24] Valko, M., et al. 2004. "Role of Oxygen Radicals in DNA Damage and Cancer Incidence." *Mol Cell Biochem* 266 (1-2): 37-56.

[25] Masuda, Y., et al. 2004. "Antioxidant Properties of

Gingerol Related Compounds from Ginger." *Biofactors* 21 (1-4): 293-6.

[26] Hamid, A., Lee, L. S., Karim, S. R., & Jufri, N. F. 2018. "Hepatoprotective Effects of Zerumbone against Paracetamol-Induced Acute Hepatotoxicity in Rats." *Malays J Med Sci* 25 (2): 64-71.

[27] Yacout, G. A., Elguindy, N. M., and El Azab, E. F. 2007. "Amelioratic Effect of *Zingiber Officinale* on Experimentally Induced Liver Fibrosis in Rats." *JMRI* 28 (2): 154-59.