

# Evaluation of the antioxidant activity of fruit extracts of indigenous medicinal plant, *Zizyphus xylopyrus* (Retz.) Willd

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## Abstract

**Aim:** This study aims to investigate the antioxidant activity of fruit extracts of an indigenous medicinal plant, *Zizyphus xylopyrus* (Retz.) Willd. **Materials and Methods:** The coarsely powdered fruit of *Z. xylopyrus* Willd. was successfully extracted first with petroleum ether and chloroform, followed by ethyl acetate and at last ethanol. The total flavonoids content (TFC), total phenolic content (TPC), and total tannin content (TTC) were estimated using quercetin and tannic acid equivalents as standard. The antioxidant activity of different extracts was determined by different standard *in vitro* methods including. **Results:** Among all extracts, ethyl acetate extract (ZXEAE) possessed potent antioxidant activity, namely hydroxyl radical (OH<sup>•</sup>) scavenging assay (IC<sub>50</sub> 128.1 ± 0.63 µg/mL), 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical (DPPH<sup>•</sup>) scavenging assay (IC<sub>50</sub> 101.2 ± 1.75 µg/mL), nitric oxide radical (NO<sup>•</sup>) scavenging assay (IC<sub>50</sub> 122.5 ± 2.04 µg/mL), superoxide radical (O<sub>2</sub><sup>•-</sup>) scavenging activity assay (IC<sub>50</sub> 64.1 ± 1.24 µg/mL), and inhibition of lipid peroxidation (98.6 ± 2.05 µg/mL). Study results suggested that antioxidant activity of ZXEAE might be due to presence of polyphenols, namely TFC (31.23 ± 0.39 quercetin equivalent [QE] mg/g extract), TPC (198.14 ± 0.69 gallic acid equivalent [GAE] mg/g extract), and TTC (18.38 ± 1.79 GAE mg/g extract). **Conclusion:** The present study demonstrated the presence of flavonoids in *Z. xylopyrus* fruits showing marked antioxidant activity, which can be used for establishing the hepatoprotective potential of *Z. xylopyrus*.

**Key words:** Antioxidant, flavonoids, free radicals, *Zizyphus xylopyrus*

## INTRODUCTION

The liver is a vital organ estimated to have over 500 functions, responsible for the synthesis of a variety of proteins such as albumin, α1-antitrypsin, very low-density lipoprotein, coagulation factors, insulin-like growth factor, and thrombopoietin. Lipophilic substances (e.g., drugs) are metabolized by hepatocyte enzymes in the liver to either less/non-toxic substances (detoxification) or more toxic substances (toxification). Failure of these metabolic functions represents the basic pathophysiology of all forms of liver disease. Hepatocytes represent 60% of liver cells and are responsible for most of its synthetic and metabolic activities.<sup>[1,2]</sup>

Herbal plants have high percentage of phenolic compounds, which act as antioxidant compounds. Antioxidant compounds have

redox properties which show its action by neutralizing the free radicals and decompose peroxides.<sup>[3,4]</sup> The inadequacy of orthodox drugs in the management of liver disease is apparent in the continuing premature deaths and disabilities resulting from these diseases. Traditional medicine has over time offered, in the form of medicinal plants, an alternative to this malady. Quite a number of plants are used in altered parts of the world for the management of liver diseases. Among such plants, *Zizyphus xylopyrus* (Retz.) Willd. (family: Rhamnaceae) as folk medicine with distinct medicinal values have created

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curiosity among researchers worldwide for their various important biological activities. *Z. xylopyrus* possesses a number of bioactivities.<sup>[5-7]</sup> The study aims to execute the biological active constituents existing in different extracts of *Z. xylopyrus* fruits showing potent antioxidant activity, responsible for the hepatoprotection in contradiction of oxidative stress persuades liver damage.

## MATERIALS AND METHODS

### Chemicals and Reagents

Vitamin C, quercetin, gallic acid, thiobarbituric acid (TBA), trichloroacetic acid, sodium nitroprusside, nitro blue tetrazolium (NBT), hydroxylamine hydrochloride, streptomycin, and penicillin were purchased from Sigma-Aldrich, Mumbai. The carbon tetrachloride, 1-naphthylethylenediamine dihydrochloride, sulfanilamide, orthophosphoric acid, and Folin–Ciocalteu reagents were purchased from SD Fines Chemicals Pvt., Ltd., Mumbai. 2,2-diphenyl-1-picrylhydrazyl (DPPH) and ethylenediaminetetraacetic acid (EDTA) were purchased from Bioworld, Mumbai, and Hi-Media, Bengaluru, respectively. All other solvents and chemicals of analytical grade used in this study were obtained from local supplier.

### Collection and Authentication of Plant Material

The fruits of *Z. xylopyrus* (Retz.) Willd. (ZX) collected from the plants grown in Sagar, Madhya Pradesh, India, were validated by Dr. P. K. Tiwari, Department of Botany, Dr. H. S. Gour Central University, Sagar, Madhya Pradesh, India. The specimen of *Z. xylopyrus* (Retz.) Willd. with Bot/H/02/49/03 Herbarium number was deposited in botany department of university for upcoming reference. The plant constituents were initially washed by water followed by drying in shade and then coarsely powdered from grinder.

## EXPERIMENTAL ANIMALS

For the present investigation, Swiss albino rats of either sex with weight of 180–240 g were acquired from the College of Veterinary Sciences and Animal Husbandry, Mhow, Madhya Pradesh, India. The animals were made familiarize for the duration of 2 weeks from the beginning of the experiment and housed in standard laboratory surroundings, that is, 45–65% of relative humidity, 25 ± 2°C of temperature, and 12 h dark and 12 h light cycle. Animals were fed by standard laboratory animal feed and water *ad libitum* during the experiment. The protocol for experimental work on animals was approved by the Institution Animal Ethical Committee (IAECNo.1546/PO/E/S/11/CPCSEA).

### Extraction of Plant Materials

In a Soxhlet apparatus, the coarse powder of fruits was closely packed and successive extraction was carried out first with petroleum ether at 40–60°C and then with chloroform, followed by ethyl acetate and at last ethanol. Maceration for 48 h in distilled water was carried out with the leftover marc and then filtered. From thimble of the apparatus, some drops of extractive were collected and evaporated. The absence of residue confirmed completion of the extraction process. This ensures that no solvents remain; the marc was dried in air before using next solvent for extraction. Rotary evaporator set at 40 ± 2°C was used for vacuum drying the collected filtrates. The different ZX extracts used in the present study are - petroleum ether extract (ZXPEE), chloroform extract (ZXCHE), ethyl acetate extract (ZXEAE), ethanolic extract (ZXEE), and aqueous extract (ZXAAQE).

## DETERMINATION OF PHYTOCONSTITUENTS

- Preliminary phytochemical screening  
The preliminary phytochemical analysis outcomes of ZX extracts determined that ZXEAE contained tannins and flavonoids. ZXEE comprises alkaloids, carbohydrate, tannins, and flavonoids; ZXCHE contains phytosterols and triterpenoids, whereas ZXAAQE consists of glycoside and carbohydrate. Amino acid and proteins were absent in entire extracts although fats and fixed oil were present only in ZXPEE of ZX extract.
- Total flavonoid content (TFC)  
The reaction between aluminum trichloride (AlCl<sub>3</sub>) and flavonoids produces a red-colored compound, their intensity is measured to determine the TFC.<sup>[8]</sup>
- Total phenolic content (TPC) and Total tannin content (TTC)  
Precipitation of tannins using gelatin was carried out for the identification of TTC, while Folin–Ciocalteu method was utilized for the identification of TPC.<sup>[9]</sup>

### Determination of Antioxidant Activity

- Reducing power assay  
Antioxidants are potent reductants. Their presence would result in Fe<sup>3+</sup> to Fe<sup>2+</sup> reduction due to electron donation. This serves as an indication for antioxidant activity. Chromophore absorbance measured at wavelength of 700 nm to determine the reducing power of extracts.<sup>[10]</sup>
- DPPH radical scavenging assay  
This scavenging assay is used to understand the extracts' antiradical activities and to determine the presence of flavonoids and phenolics.<sup>[11]</sup> Reaction between DPPH° radical and proton donor leads to the formation of complex of yellow color. The intensity of this color is used to determine the DPPH° radical scavenging

activity.<sup>[8]</sup> The DPPH° radical scavenging activity was determined by Equation 1.

$$\text{Scavenging activity (\%)} = \left( \frac{A_o - A_t}{A_o} \right) \times 100 \quad (1)$$

Where,  $A_o$ =Control absorbance and  $A_t$ =Sample absorbance

- **OH° scavenging assay**  
OH° can be produced using Fenton reaction in occurrence of the reduced transition metals ( $\text{Fe}^{2+}$ ) and  $\text{H}_2\text{O}_2$ , known as the best reactive system amid all dioxygen reduced forms and thought to begin *in vivo* the cell injury.<sup>[12]</sup>
- **Nitric oxide ( $\text{NO}^\circ$ ) radical scavenging assay**  
Griess reagent has the ability to inhibit  $\text{NO}^\circ$  radical. The radical generated as of sodium nitroprusside solution in the phosphate buffer saline. The ability of Griess reagent to inhibit  $\text{NO}^\circ$  radical was analyzed at 546 nm.<sup>[13]</sup> The  $\text{NO}^\circ$  radical scavenging activity was determined by Eq. 1
- **Superoxide ( $\text{O}_2^-$ ) radical scavenging assay**  
NBT reagent method was used for the estimation of  $\text{O}_2^-$  radical scavenging action of extracts. In the presence of NBT, autooxidation of hydroxylamine hydrochloride results in the formation of  $\text{O}_2^-$ . The radical is then reduced to nitrite. In the presence of EDTA, the nitrite ion produces a colored complex, intensity measured at 560 nm.<sup>[14]</sup> The  $\text{O}_2^-$  radical scavenging ability was determined by Eq. 1.
- **Inhibition of lipid peroxidation (LPO) in rat liver homogenate**  
When malondialdehyde, secondary end product of  $\text{Fe}^{2+}$ -induced polyunsaturated fatty acids oxidation, reacts from two molecules of 2-TBA, it leads to the formation of pinkish-red-colored chromogen. The intensity of this chromogen is measured to determine LPO inhibition activity of the extracts. A decrease in LPO is indicated by a decrease in absorbance.<sup>[15,16]</sup> Percent inhibition of LPO was determined by Eq. 1.

## Statistical Analysis

The results of the studies were reported as mean  $\pm$  SEM ( $n = 3$ ). Total variation in a set of data was estimated by one-way analysis of variance, followed by Dunnett's test for determining statistical significance. The values  $< 0.05$  were considered as statistically significant.

## RESULTS AND DISCUSSION

### Determination of Phytoconstituents by Preliminary Phytochemical Screening

The preliminary phytochemical study outcome of extracts of ZX presented that ZXEAE comprises tannins and flavonoids;

ZXEE has carbohydrate, glycosides, tannins, flavonoids, and alkaloids; ZXCHE comprises triterpenoids and phytosterols although ZXAQE has glycoside and carbohydrate. Amino acids and proteins are lacking in all extracts, whereas fixed oil and fats were present only in ZXPEE. Table 1 represents the results of preliminary phytochemical screening.

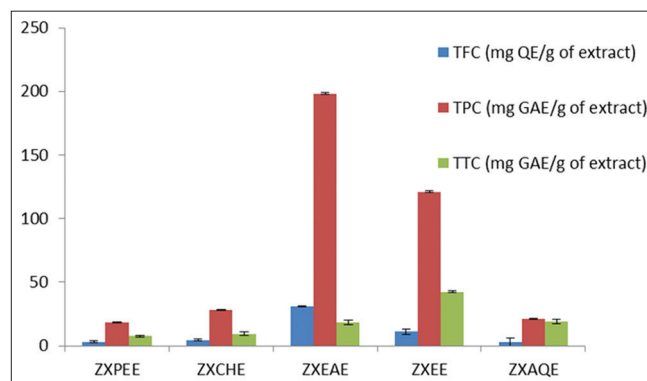
### Estimation of TPC, TFC, and TTC

TPC in different extracts of ZX was estimated through measure the blue color intensity formed at  $\lambda_{\text{max}}$  765 nm, whereas extracts TTC was estimated by deducting phenolics non-tannin from TPC. Their outcome was stated in comparison to gallic acid equivalent (GAE), mg of GAE/g of extract GAE determined by regression equation  $y = 0.0588x + 0.0025$  and  $r^2 = 0.9994$ , where x is absorbance and y is GAE. Different ZX extracts have different TPC, TFC, and TTC levels [Figure 1]. The maximum TPC was obtained in ZXEAE ( $198.14 \pm 0.69$  mg GAE/g of extract), followed by ZXEE ( $121.12 \pm 0.74$ ), ZXCHE ( $28.24 \pm 0.28$ ), ZXAQE ( $21.24 \pm 0.35$ ), and ZXPEE ( $18.51 \pm 0.31$ ). The maximum TTC was obtained in ZXEE ( $42.52 \pm 0.98$  mg GAE/g of extract), followed by ZXAQE ( $19.12 \pm 1.76$ ), ZXEAE ( $18.38 \pm 1.79$ ), ZXCHE ( $9.51 \pm 1.39$ ), and ZXPEE ( $7.42 \pm 0.89$  mg GAE/g of extract). TFC in the plant extracts studied by determining red color intensity developed due to reaction between  $\text{AlCl}_3$  and flavonoids at 420 nm  $\lambda_{\text{max}}$ . TFC was stated in comparison to the quercetin equivalent mg of QE/g of extract, estimated by means of regression equation  $y = 0.0199x + 0.0046$ ,  $r^2 = 0.9952$ , where y is QE and x is absorbance. Maximum flavonoid content was obtained in ZXEAE ( $31.23 \pm 0.39$  mg QE/g of extract) followed by ZXEE ( $11.07 \pm 1.97$ ), ZXCHE ( $4.52 \pm 0.65$ ), ZXPEE ( $3.18 \pm 0.94$ ), and ZXAQE ( $3.10 \pm 0.99$  mg QE/g of extract).

## ESTIMATION OF ANTIOXIDANT ACTIVITY

### Ferric Reducing Antioxidant Power Assay

In this study, the reducing ability of various extracts of ZX observed at various concentrations (20–200  $\mu\text{g/mL}$ )



**Figure 1:** Quantity of total phenolic content, total tannin content, and total flavonoids content extant in ZX extracts

**Table 1:** Phytochemical screening of different ZX extracts

Tests	ZXPEE	ZXCHE	ZXEAE	ZXEE	ZXAQE
Tests for carbohydrate					
Benedict's test	-	-	-	-	-
Molisch's test	-	-	-	+	+
Fehling's test	-	-	-	-	+
Tests for alkaloids					
Dragendorff's test	-	-	-	+	-
Mayer's test	-	-	-	+	-
Hager's test	-	-	-	+	-
Wagner's test	-	-	-	+	-
Tests for glycosides					
Keller–Killiani test	-	-	+	-	-
Legal's test	-	-	-	+	-
Foam test	-	-	-	-	+
Modified Borntrager's test	-	-	-	-	-
Tests for flavonoids					
Shinoda's test	-	-	+	+	-
Alkaline reagent test	-	-	+	+	-
Tests for triterpenoids and phytosterols					
Liebermann's test	-	+	-	-	+
Salkowski test	-	-	-	-	+
Liebermann's–Burchard test	+	+	-	-	+
Tests for amino acids and protein					
Ninhydrin test	-	-	-	-	-
Millon's test	-	-	-	-	-
Tests for tannins					
Lead acetate test	-	-	+	+	-
Gelatin test	-	-	+	+	-
Ferric chloride test	-	-	+	+	-
Test for fat and fixed oils					
Saponification test	+	-	-	-	-
Filter paper test	+	-	-	-	-

ZXPEE: Petroleum ether extract, ZXCHE: Chloroform extract, ZXEAE: Ethyl acetate extract, ZXEE: Ethanolic extract, ZXAQE: Aqueous extract, where (+) represents the presence and (-) represents the absence

[Figure 2]. The increase in absorbance further shows reducing power of extract, as a major indicator of antioxidant action potential.<sup>[17]</sup> The numerous extracts reducing power of ZX were obtained in given order - Vitamin C > ZXEAE > ZXEE > ZXPEE > ZXCHE > ZXAQE.

## OH° SCAVENGING ASSAY

Various extracts exhibited different and scavenging dose-dependent activity in tested limit of concentration. Further than the tested extracts, highest scavenging action detected with ZXEAE (Figure 3). IC<sub>50</sub> of Vitamin C was obtained as 8.9 ± 0.44 µg/mL. The manner of scavenging action (IC<sub>50</sub>) of Vitamin

C and extracts was resulted as follows: 8.9 ± 0.44 µg/mL of Vitamin C > 119.5 ± 0.51 µg/mL ZXEE > 128.1 ± 0.63 µg/mL ZXEAE > 197.4 ± 0.59 µg/mL ZXCHE > 243.6 ± 0.72 µg/mL ZXAQE > 315.2 ± 0.49 µg/mL ZXPEE. IC<sub>50</sub> of ZXEAE found 14.39 times lesser in contrast to Vitamin C. At 200 µg/mL of concentration, ZXEE and ZXEAE displayed 49.3% and 48.5% OH° radical scavenging action, respectively.

## DPPH RADICAL SCAVENGING ASSAY

DPPH is free radical (stable) with extreme absorbance at wavelength of 517 nm. It freely undergoes scavenging through an antioxidant and converted to 1,

1-diphenyl-2-picrylhydrazine. The discoloration degree shows the potential of scavenging activity of antioxidant substance.<sup>[12]</sup> It had been broadly used to examine the compounds ability like hydrogen donors or free-radical scavengers and further in evaluation of activity of antioxidant present in plant extracts. In this study, capacity of DPPH° radical scavenging activity of various extracts was observed at various concentrations (10–200 µg/mL). Extracts concentration was essential to reduce the DPPH initial concentration to 50% (IC<sub>50</sub>) below definite experimental situation was measured and outcome is expressed as mean ± SEM. Lesser IC<sub>50</sub> value shows more antioxidant action.

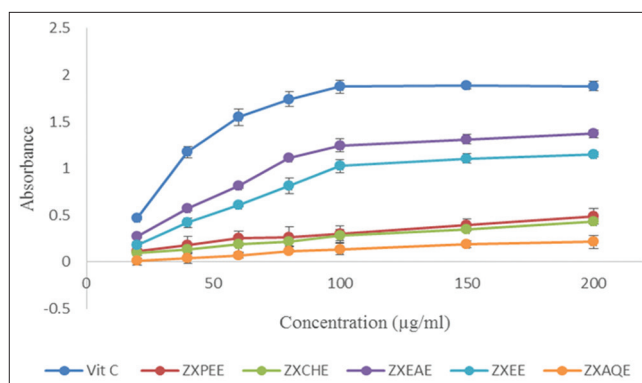
Several extracts exhibited different and scavenging activity-dependent dose in tested concentration range. Rest of the tested extracts, highest scavenging action was perceived by ZXEEAE [Figure 4]. IC<sub>50</sub> of Vitamin C obtained to be 8.4 ± 0.41 µg/mL. Therefore, command of scavenging activities (IC<sub>50</sub>) of plant extracts and Vitamin C resulted as Vitamin C (8.4 µg/mL) > ZXEEAE (101.2 ± 1.75 µg/mL) > ZXEE (118.2 ± 1.27 µg/mL) > ZXAQE (248.1 ± 2.02 µg/mL) > ZXCHE (287.1 ± 1.51 µg/mL) > ZXPEE (332.2 ± 2.06 µg/mL). IC<sub>50</sub> of ZXEEAE was obtained 12.05 times lesser as compared to Vitamin C. At 200 µg/mL concentration, ZXEE and ZXEEAE indicated 56.1% and 68.3% DPPH radical scavenging property, respectively.

### NO° radical scavenging assay

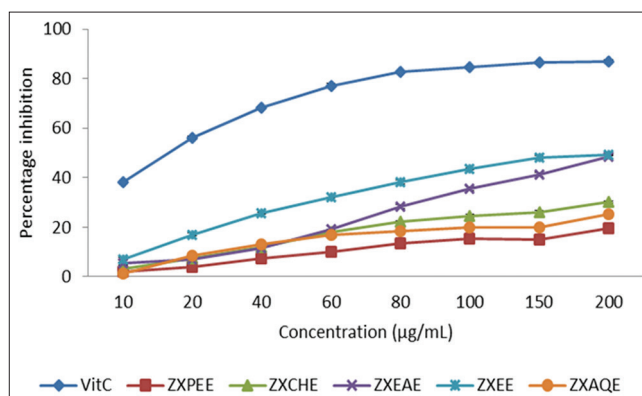
The percentage of radical scavenging ability of the standard Vitamin C and ZX extracts contrary to radical of NO° was improved in a dose (10–200 µg/mL)-dependent mode. Outside the ZX tested extracts, highest scavenging property was detected with ZXEEAE [Figure 5]. IC<sub>50</sub> of Vitamin C was observed as 22.1 ± 0.51 µg/mL and the manner of scavenging action (IC<sub>50</sub>) of Vitamin C and extracts was resulted as follows: 22.1 ± 0.51 µg/mL of Vitamin C > 122.5 ± 2.04 µg/mL of ZXEEAE > 138.3 ± 0.79 µg/mL of ZXEE > 263.3 ± 1.29 µg/mL of ZXAQE > 398.3 ± 1.63 µg/mL of ZXCHE > 418.3 ± 1.84 µg/mL of ZXPEE. IC<sub>50</sub> of ZXEEAE was found as 5.54 times lesser than Vitamin C further at 200 µg/mL of concentration, the ZXEE and ZXEEAE indicated 51.2% and 57.1% of NO° radical scavenging action, respectively.

### O<sub>2</sub>° radical activity assay

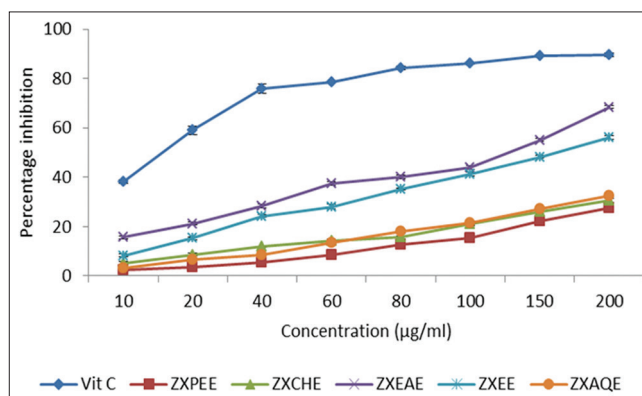
Various ZX extracts showed scavenging activity dose dependent on O<sub>2</sub>° radical. Outside the tested extracts, highest scavenging action was detected with ZXEEAE [Figure 6]. IC<sub>50</sub> of Vitamin C was resulted as 29.1 ± 0.39 µg/mL. The scavenging activity (IC<sub>50</sub>) of Vitamin C and extracts was as follows: 29.1 ± 0.39 µg/mL of Vitamin C > 64.1 ± 1.24 µg/mL of ZXEEAE > 107.1 ± 2.09 µg/mL of ZXEE > 257.2 ± 2.41 µg/mL of ZXAQE > 333.5 ± 1.96 µg/mL of ZXCHE > 351.2 ± 2.55 µg/mL of ZXPEE. ZXEEAE IC<sub>50</sub> is 2.20 times lesser as compared with Vitamin C. At 200 µg/mL



**Figure 2:** Reducing power of several extracts of ZX. The values are represented as mean ± SEM (n = 3)



**Figure 3:** Percent inhibition by ZX extracts in hydroxyl radical scavenging assay. The values are represented as mean ± SEM (n = 3)

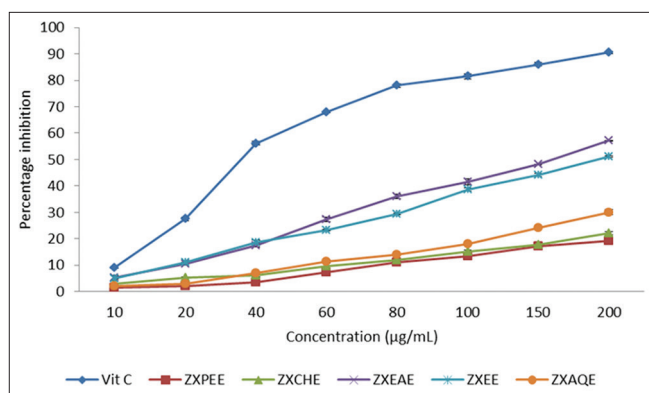


**Figure 4:** Percent inhibition by ZX extracts in 2,2-diphenyl-1-picrylhydrazyl radical scavenging assay. The values are represented as mean ± SEM (n=3)

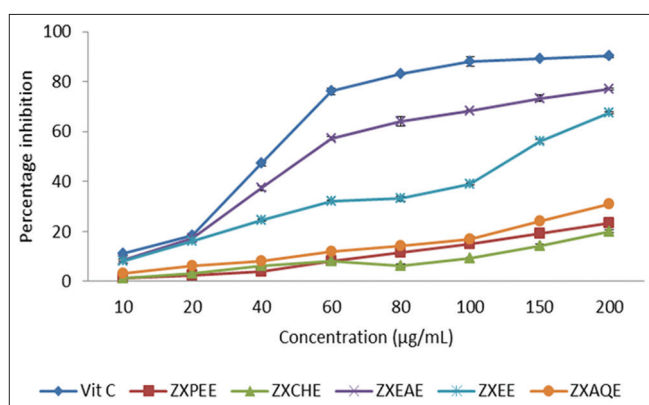
concentration, ZXEE and ZXEEAE presented 67.5% and 77.1% O<sub>2</sub>° radical scavenging activity, respectively.

### Inhibition of LPO Assay

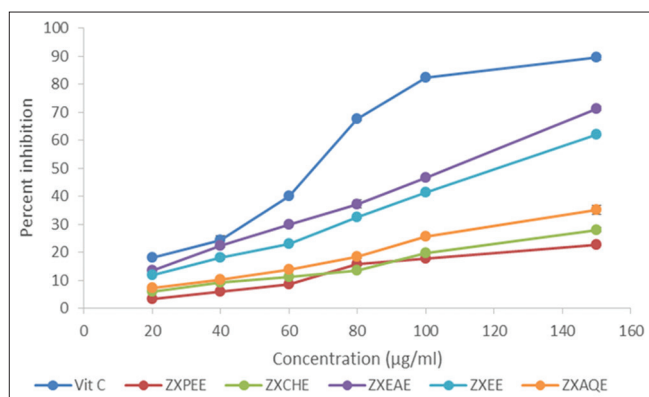
The capacity of LPO inhibition of various extracts of ZX was inspected at numerous concentrations as 20–150 µg/mL, the value of IC<sub>50</sub> of different extracts under definite experimental



**Figure 5:** Percent inhibition by ZX extracts in nitric oxide radical scavenging assay. The values are represented as mean  $\pm$  SEM ( $n=3$ )



**Figure 6:** Percent inhibition by ZX extracts in superoxide radical scavenging assay. The values are represented as mean  $\pm$  SEM ( $n=3$ )



**Figure 7:** Percent inhibition by ZX extracts in inhibition of lipid peroxidation assay. The values are represented as mean  $\pm$  SEM ( $n=3$ ).

situation was measured and conclusion was stated as mean  $\pm$  SEM. Further than tested ZX extracts, highest scavenging action was directed with ZXAE subsequently ZXEE [Figure 7].  $IC_{50}$  of Vitamin C was obtain as  $59.5 \pm 0.75$   $\mu$ g/mL and manner of scavenging potential ( $IC_{50}$ ) of Vitamin C and extracts was found as follows:  $59.5 \pm 0.75$   $\mu$ g/mL of Vitamin C >  $98.6 \pm 2.05$   $\mu$ g/mL of ZXAE >  $124.8 \pm 2.46$   $\mu$ g/mL of

ZXEE >  $304.7 \pm 3.05$   $\mu$ g/mL of ZXAQE >  $380.7 \pm 1.85$   $\mu$ g/mL of ZXCHE >  $395.6 \pm 2.96$   $\mu$ g/mL of ZXPEE.  $IC_{50}$  of ZXAE is 1.55 times lesser as compared to Vitamin C. At 150  $\mu$ g/mL of concentration, the ZXEE and ZXAE showed 62.2% and 71.5% reticence of LPO action, respectively.

## CONCLUSION

Different studies reflected that the extracts of *Z. xylopyrus* fruits showed antioxidant activities. These effects may be due to phytoconstituents present in the extracts including flavonoids. Outcomes also proposed that antioxidant activity of *Z. xylopyrus* fruits can be utilized for hepatoprotective activity.

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## REFERENCES

- Gordon-Weeks AN, Snaith A, Petrinic T, Friend PJ, Burls A, Silva MA, *et al.* Systematic review of outcome of downstaging hepatocellular cancer before liver transplantation in patients outside the Milan criteria. *Br J Surg* 2011;98:1201-8.
- McLaughlin JK, Lipworth L. Epidemiologic aspects of renal cell cancer. *Semin Oncol* 2000;27:115-23.
- Osawa T, Uritani I, Garcia VV. Novel Natural Antioxidants for Utilization in Food and Biological Systems, Post Harvest Biochemistry of Plant Food-Materials in the Tropics. Japan: Japan Scientific Societies Press; 1994. p. 241-51.
- Kahkonen MP, Hopia AI, Vuorela HJ, Rauha JP, Pihlaja K, Kujala TS, *et al.* Antioxidant activity of plant extracts containing phenolic compounds. *J Agric Food Chem* 1999;47:3954-62.
- Mansoori MH, Kaur C, Gupta MK, Vyas M, Mishra V. *Zizyphus xylopyrus* (Retz.) Wild: A plant of enormous biomedical potential. *Int J Green Pharm* 2018;12:S322-29.
- Sameera NS, Mandakini BP. Investigations into the antibacterial activity of *Zizyphus mauritiana* lam and *Zizyphus xylopyra* (Retz.) willd. *Int Food Res J* 2015;22:849-53.
- Singhal U, Kumar S. Antidiarrhoeal activity of fruit extracts of *Zizyphus xylopyrus* (Retz.) willd in rats. *Int J Pharm Erudition* 2011;1:57-61.
- Olajuyigbe OO, Afolayan AJ. Phenolic content and antioxidant property of the bark extracts of *Zizyphus mucronata* willd. Subsp. *Mucronata* willd. *BMC*

- Complement Altern Med 2011;11:130.
9. Chouhan HS, Singh SK. Phytochemical analysis, antioxidant and anti-inflammatory activities of *Phyllanthus simplex*. J Ethnopharmacol 2011;137:1337-44.
10. Sunil C, Ignacimuthu S. *In vitro* and *in vivo* antioxidant activity of *Symplocos cochinchinensis* S. Moore leaves containing phenolic compounds. Food Chem Toxicol 2011;49:1604-9.
11. Gul MZ, Ahmad F, Kondapi AK, Qureshi IA, Ghazi IA. Antioxidant and antiproliferative activities of *Abrus precatorius* leaf extracts-an *in vitro* study. BMC Complement Altern Med 2013;13:53.
12. Wang H, Gao XD, Zhou GC, Cai L, Yao WB. *In vitro* and *in vivo* antioxidant activity of aqueous extract from *Choerospondias axillaris* fruit. Food Chem 2008;106:888-95.
13. Shirwaikar A, Somashekar AP. Anti-inflammatory and free radical scavenging studies of *Aristyolochia bracteolata* Lam. India J Pharm Sci 2003;65:67-9.
14. Shukla S, Mehta A, John J, Singh S, Mehta P, Vyas SP. Antioxidant activity and total phenolic content of ethanolic extract of *Caesalpinia bonducella* seeds. Food Chem Toxicol 2009;47:1848-51.
15. Singh N, Rajini PS. Free radical scavenging activity of an aqueous extract of potato peel. Food Chem 2004;85:611-6.
16. Yen GC, Hsieh CL. Antioxidant activity of extracts from du-zhong (*Eucommia urmoides*) towards various peroxidation models *in vitro*. J Agric Food Chem 1998;46:3952-7.
17. Loganayagi N, Perumal S, Sellamuthu M. Antioxidant activity and free radical scavenging capacity of phenolic extracts from *Helicteres isora* L and *Ceiba pentandra* L. J Food Sci Tech 2013;50:687-95.

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