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Protective appraisal of *Gynandropsis pentaphylla* leaves against hepatic injury in rats

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Abstract
To appraise the protective effect of methanol extract of *Gynandropsis pentaphylla* leaves in carbon tetrachloride (CCl_4)-induced hepatic injury in rats. The hepatic injury was induced in rats with the administration of a 1:1 ($^{v}/_{v}$) mixture of CCl_4 in olive oil at the dose level of 1ml/kg intra-peritoneally once a week. The methanol extract of *Gynandropsis pentaphylla* (50 and 100 mg/kg) and the standard drug Silymarin (25 mg/kg) were given orally from 1 to day 15. The extract of *Gynandropsis pentaphylla* produced significant ($p \leq 0.001$) dose-dependent protection on the altered hepatic-SOD (superoxide dismutase), CAT (catalase), GSH (glutathione reduced), GPx (glutathione peroxidase), LPO (lipid peroxidation) and cytochrome *P*-450 enzyme when compared with the toxic control. Serum was also analyzed for SGOT, SGPT (transaminases), ALP (alkaline phosphatase), GGT (gamma-glutamyl transpeptidase), LDH (lactate dehydrogenase), total bilirubin, and total protein levels. *Gynandropsis pentaphylla* extract produced a protective effect by decreasing the activities of serum enzymes, and total bilirubin along with increasing the total protein level when compared with toxic control. The results were compared with the standard drug silymarin. From the above results, it can be concluded that the observed hepatoprotective potential of *Gynandropsis pentaphylla* leaves might be due to the presence of tannins, alkaloids, terpenoids, steroids, total phenolics, and flavonoids in the extract and/or the purified compounds- Lupeol (I), β-sitosterol (II), α-amyrin (III), and β-amyrin (IV), which were isolated from the methanol extract of *Gynandropsis pentaphylla* leaves.

Keywords: *Gynandropsis pentaphylla*, Bioactive compounds, CCl_4, Hepatic injury markers

Introduction
Traditionally used herbal medicines play a key function inside the organ system of the human body by delivering definite physiological action due to the presence of bioactive components like tannins, alkaloids, terpenoids, steroids, total phenolics, flavonoids, etc. [1]. These bioactive components are supplementary biomolecules, which help to give beneficial effects to the human body against several diseases. Therefore, herbal medicines have been proposed as valuable substitutions for the anticipation and management of hepatic injury and its chronic complications based on experimental studies [2].
Gynandropsis pentaphylla (Capparidaceae), commonly known as Hurhur, is found as a weed, annual erect, grows up to 60cm in height, throughout the warmer parts of India. This herb is edible due to its nutritional value as the leaves are rich in vitamins A & C, iron, phosphorus, and calcium [3]. Therefore, it has several applications in the Indian traditional medical practice. This plant is also used in cough and as ant scorbutic, diaphoretic, and cobra bites [1, 3]. In previous studies, it has been reported as an antioxidant, antimalarial, anticancer, antimicrobial, and anti-fever by different scientific communities [3, 4]. Therefore, due to the bunch of its medicinal properties, the interest in phytochemistry began as early as 1906; so far more than 50 compounds have been isolated from the different parts of this herb such as saponins, glycosides, lectins, steroids, flavonoids, tannins, triterpenes, resins, phenolic compounds, glucosinates, and anthraquinones [1, 4]. According to those valuable phytotherapeutic studies of this herb, this paper describes the protective appraisal of *Gynandropsis pentaphylla* leaves against hepatic injury in rats.

2. Materials and Methods
2.1 Animals
Colony-bred healthy, adult male albino rats (Wistar strain) (*Rattus norvegicus*) weighing 150–160g, were used in the present study.

The rats were housed in polypropylene cages under controlled conditions of temperature (25±3 °C), humidity (55%-65%), and light (12h light/dark cycle). They were provided with a nutritionally adequate standard laboratory diet (Lipton, India Ltd., Bangalore, India) and tap water *ad libitum*.

2.2 Plant material

The plant material (*Gynandropsis pentaphylla* leaves) was collected from the NREC college campus and authenticated by Dr. Vishal Kaushik, In-charge of Herbarium, Department of Botany, NREC College, Khurja, District- Bulandshahr 203 131, U.P., India.

2.3. Extraction, isolation, and characterization of compounds

The plant material was shade dried, crushed to a rough powder, and treated with petroleum ether for defatting as well as to remove chlorophyll. The powder was packed into a soxhlet apparatus and subjected to hot continuous percolation using methanol as a solvent for 48 hours at 58–60 °C. The extract was concentrated under a vacuum, dried in a vacuum desiccator, and yielded 5.7% w/w as a dark greenish-brown solid mass. The solid mass was then powdered and washed with chloroform to remove the remaining content of chlorophyll present in the extract. Half of the extract was suspended in a proper volume of olive oil to prepare the desired concentration for oral administration to rats.

Rest of the methanol extract (30gm) was subjected to traditional column chromatography for fractionation with different solvents. For this purpose, a column (1.4 m × 5 cm) filled with 800g Si-gel (60–120 mesh) was used. A number of compounds were fractionated and isolated from the methanol extract by eluting the column with petroleum ether : benzene at different concentrations. The purity of fractions was checked by qualitative thin-layer chromatography and HPLC using different solvent systems. After ascertaining the purity of compounds, it was subjected to detailed spectral analysis (IR, 1H NMR, 13C NMR, and MS) to establish the structures of yielded compounds [data not shown]. As a result, the compounds- lupeol (I), β-sitosterol (II), α-amyrin (III), and β-amyrin (IV), were isolated, purified, and characterized [5, 6].

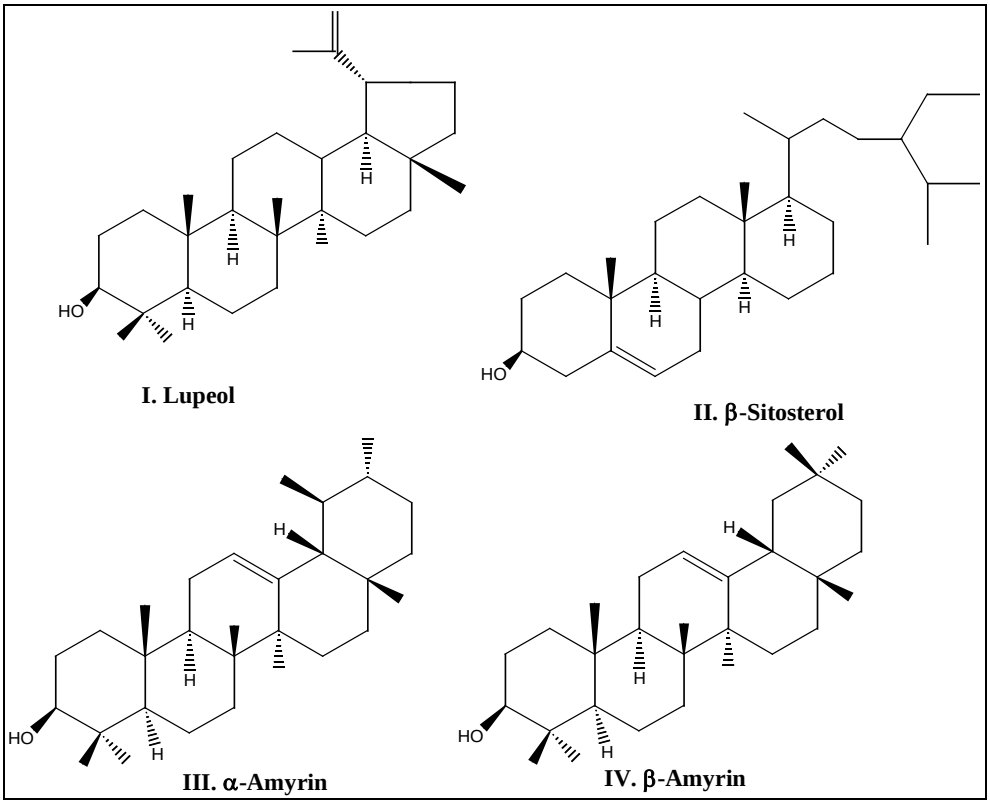

Fig 1: Chemical structure of isolated and purified compounds

2.4. Chemicals

All chemicals and reagents have been used of analytical grade and obtained from Sigma Chemical Company (St. Louis, MO, USA). The kits of SGOT (batch no. 61105), SGPT (batch no. 60805), and GGT (batch no. 34004) were purchased from Accurex Biomedical Pvt. Ltd., Mumbai, India, and LDH (lot no. 6854), ALP (lot no. 7093), total bilirubin (lot no. 6801) and total protein (lot no. 6808) were purchased from Span Diagnostics Ltd., Surat, India. The standard drug silymarin was purchased from Metro Scientific Chemicals, Jaipur, India.

2.5. Standard Drug

Silymarin was used as a standard drug during the designed experiment and purchased from MP Biomedicals, France and it was dissolved in olive oil for oral administration to rats at the dose level of 25 mg/kg body weight/day [7].

2.6. Ethical Aspects

The study was approved by the ethical committee (Protocol No: 1678/Go/a/12/CPCSEA/112) of the University Department of Zoology, Jaipur, India. Indian National Science Academy, New Delhi (INSA, 2000) guidelines were followed for the maintenance and use of the experimental animals.

2.7. Toxicological study

The extract was administered to all the test groups in graded doses ranging up to 800mg/kg body weight and the rats were observed for signs of toxicity and mortality for 15 days

afterward. The extract was found to be practically non-toxic when given orally to rats and its LD_{50} value was found to be higher than 800mg/kg body weight [data not shown]. The minimum dose levels *viz*. 50mg and 100mg/kg body weight were used for oral administration to rats during the experimentation [2].

2.8. Experimental design
After acclimatization of 15 days, the animals were divided into the following groups of 06 rats in each group:

Group I: Untreated rats were kept on a normal diet and served as control.

Group II: Rats were intoxicated with carbon tetrachloride (1 ml/kg body wt/once a week, IP with olive oil, 1:1) for 15 days.

Group III: Rats received 50mg/kg body wt/day, PO with olive oil of *Gynandropsis pentaphylla* extract and CCl_4 as group II, for 15 days.

Group IV: Rats received 100mg/kg body wt/day, PO with olive oil of *Gynandropsis pentaphylla* extract and CCl_4 as group II, for 15 days.

Group V: Rats received 25mg/kg body wt/day, PO with olive oil of silymarin and CCl_4 as group II, for 15 days.

2.9. Biochemical analysis
After 24 hours of the last dose delivery, all rats of each treated group were anesthetized with ether, and blood was collected by cardiac puncture. The serum was separated by centrifugation at 2500rpm at 37°C for 20 min and analyzed for SGOT, SGPT, ALP, GGT, LDH, total bilirubin, and total protein, using diagnostic kits. After the collection of blood, the liver was immediately excised, washed with cold saline, blotted, minced, and homogenized for SOD [8], CAT [9], GSH [10], GPx [11], LPO [12] determination. After that, a liver microsomal fraction was prepared [13] and the cytochrome *P*-450 enzyme content in this fraction was measured from a reduced carbon monoxide difference spectrum [14], respectively.

2.10. Statistical analysis
All the results were expressed as mean ± SEM and analyzed using the student's *t*-test. A probability value of $P \leq 0.05$ was considered significant.

3. Results
The results of the biochemical parameters revealed that the administration of CCl_4 to rats caused significant ($P \leq 0.001$) hepatic injury as evidenced by marker enzymes and antioxidant defense system through the liver and serum contents (Table I and II).

Table I depicts that the activities of hepatic antioxidants such as SOD, CAT, GSH, GPx, and enzyme cytochrome *P*-450 declined significantly ($P \leq 0.001$) along with significant ($P \leq 0.001$) elevation of LPO, upon CCl_4 administration alone to rats (group II) when compared with group I (vehicle control). These altered levels of hepatic tissue parameters were dose-dependently brought about towards normalization by *Gynandropsis pentaphylla* leaves extract against CCl_4-induced alterations in the hepatic antioxidants and enzyme levels. The degree of protection was observed statistically similar in both silymarin (Group V) and higher dose (100 mg/kg) of *Gynandropsis pentaphylla* leaves extract (group IV).

Carbon tetrachloride caused a significant ($P \leq 0.001$) elevation in serum enzymatic activities of SGOT, SGPT, ALP, LDH, GGT, and total bilirubin along with a concurrent decline in total protein levels in comparison to vehicle control (group I). In contrast, treatment with *Gynandropsis pentaphylla* leaves extract showed a dose-dependent restoring effect on CCl_4-induced alterations in serum levels of treated rats in groups III and IV (Table II). The elimination of hepatic oxidative stress by silymarin (group V) and a higher dose (100 mg/kg) of *G. pentaphylla* leaves extract (group IV) was significantly ($P \leq 0.001$) Similar (Table II).

Table I: Showing to appraise the protective effect of methanol extract of *Gynandropsis pentaphylla* leaves and silymarin against hepatic injury in rats through hepatic tissue's antioxidant defense and enzymatic levels

Treatment design	SOD (µ mole/mg protein)	CAT (µ mole H_2O_2 consumed / min/mg protein)	GSH (n mole/g tissue)	GPx (n mole NADPH consumed/min/ mg protein	LPO (n mole MDA/ mg protein	Cytochrome-*P*-450 (n mole/ mg protein)
Control (Vehicle-treated) Group I	12.28 ± 0.44	65.35 ± 2.88	5.22 ± 0.27	15.39 ± 0.26	1.92 ± 0.08	5.21 ± 0.18
CCl_4 (1 ml/kg b wt, IP, once a week with olive oil, 1:1) Group II	5.82 ± 0.16[a]	36.15 ± 1.96 [a]	2.32 ± 0.13 [a]	8.42 ± 0.19 [a]	5.12 ± 0.18 [a]	2.08 ± 0.14 [a]
CCl_4 + *G. pentaphylla* extract (50 mg/kg b wt/day, orally) Group III	7.75 ± 0.10 [a]	46.80 ± 1.68 [b]	3.72 ± 0.09 [a]	10.91 ± 0.17[a]	4.48 ± 0.13[c]	2.93 ± 0.08[a]
CCl_4 + *G. pentaphylla* extract (100 mg/kg b wt/day, orally) Group IV	9.85 ± 0.14 [a]	55.19 ± 2.92 [a]	4.43 ± 0.08 [a]	13.72 ± 0.21 [a]	2.98 ± 0.12 [a]	4.22 ± 0.13 [a]
CCl_4 + Silymarin (25 mg/kg b wt/day, orally) Group V	11.68 ± 0.32 [a]	61.21 ± 2.12 [a]	6.30 ± 0.34 [a]	14.82 ± 0.27 [a]	2.21 ± 0.10 [a]	4.89 ± 0.15 [a]

Levels of significance:
Data are mean ± SEM (n = 6)
a = $P \leq 0.001$ Group II compared with control (Group I)
a = $P \leq 0.001$; b = $P \leq 0.01$; c = $P \leq 0.05$ Group III compared with Group II
a = $P \leq 0.001$ Group IV and V compared with Group II

Table 2: Showing to appraise the protective effect of methanol extract of *Gynandropsis pentaphylla* leaves and silymarin against hepatic injury in rats through serum markers

Treatment design	SGOT (IU/L)	SGPT ((IU/L))	ALP (KAU)	GGT ((IU/L))	LDH (IU/L)	Total bilirubin (mg/100 ml)	Total protein (gm/dL)
Control (Vehicle-treated) Group I	128.21 ± 2.10	108.32 ± 2.87	21.33 ± 1.30	9.52 ± 0.93	84.15 ± 2.57	0.85 ± 0.06	6.21 ± 0.24
CCl_4 (1 ml/kg b wt, IP, once a week with olive oil, 1:1) Group II	206.14 ± 2.87 [a]	187.27 ± 3.09 [a]	35.10 ± 1.42 [a]	28.16 ± 1.41 [a]	142.22 ± 2.88 [a]	1.79 ± 0.10 [a]	3.14 ± 0.17 [a]
CCl_4 + *G. pentaphylla* extract (50 mg/kg b wt/day, orally) Group III	179.20 ± 3.56 [a]	138.21 ± 2.89 [b]	30.12 ± 0.92 [c]	21.42 ± 0.69 [a]	116.13 ± 1.21 [a]	1.35 ± 0.14 [c]	4.52 ± 0.16 [a]
CCl_4 + *G. pentaphylla* extract (100 mg/kg b wt/day, orally) Group IV	143.18 ± 2.33 [a]	129.10 ± 2.14 [a]	27.18 ± 0.47 [a]	16.39 ± 0.72 [a]	98.10 ± 1.19 [a]	1.06 ± 0.07 [a]	6.32 ± 0.23 [a]
CCl_4 + Silymarin (25 mg/kg b wt/day, orally) Group V	122.20 ± 2.88 [a]	114.19 ± 1.42 [a]	23.85 ± 0.66 [a]	12.30 ± 0.42 [a]	80.10 ± 1.52 [a]	0.92 ± 0.08 [a]	6.88 ± 0.23 [a]

Levels of significance:
Data are mean ± SEM (n = 6)
a = P≤ 0.001 Group II compared with control (Group I)
a = P≤ 0.001; b = P ≤ 0.01; c = P≤ 0.05 Group III compared with Group II
a = P≤ 0.001 Group IV and V compared with Group II

4. Discussion

Liver plays a vital role in regulating homeostasis along with detoxification and transformation of chemicals[15]. Although, it is in a way exposed to their harmful effects increasing its susceptibility to innumerable ailments like hepatitis, hepatic steatosis (fatty liver), fibrosis, cirrhosis, and alcoholic/drug-induced illnesses [16].

Carbon tetrachloride (CCl_4) is a well-known hepatotoxicant in human beings and animal models. It has been successfully used in hepatotoxicity studies as a model and to appraise hepatoprotective agents [2]. Because, it is now generally accepted that the oxidative injury in the liver by CCl_4 is the result of reductive dehalogenation, which is catalyzed by the *P*-450 enzyme and forms the highly reactive trichloromethyl free radical ($CCl_3^{\bullet}$). This then readily interacts with molecular oxygen to form the trichloromethyl peroxy radical ($CCl_3OO^{\bullet}$). After that, both radicals are capable of covalently binding to proteins or lipids or of abstracting a hydrogen atom from an unsaturated lipid, which initiates lipid peroxidation and hepatic injury. Therefore, the suppression of *P*-450 can result in a reduction in the level of the reactive metabolites, and correspondingly, less tissue injury. The metabolic activation of CCl_4 is believed to be mediated through P-450 2E1 [17]. The inhibitory effect of CCl_4 on cytochrome *P*-450 level was also compensated by *Gynandropsis pentaphylla* extract and silymarin through maintenance of its normal level. The role of *Gynandropsis pentaphylla* extract in the protection of CCl_4-mediated loss in cytochrome *P*-450 content may be considered an indication of improved protein synthesis in the hepatic cells [17, 18].

The LPO degradation of biomembranes is one of the major causes of oxidative injury of hepatic cells induced by CCl_4. Because, LPO is observed as an intricate biochemical reaction involving free radicals, oxygen, metal ions, and a host of other factors in the hepato-biological system. Therefore, LPO is the effort of intense activity in relation to its possible involvement in hepatic health and disease [19]. In our present study, the measurement of LPO in the hepatic tissue is an appropriate practice to monitor oxidative cell damage. Inhibition of elevated LPO has been observed in *Gynandropsis pentaphylla* extract and silymarin-treated groups due to its antioxidant and free radical scavenging activities through the re-establishment of biomembranes of hepatic parenchymal cells [2].

The generation of reactive oxygen species (ROS) leading to oxidative injury due to O_2^- is contained by dismutation with SOD, which converts the reactive O_2 to H_2O_2, if not scavenged by catalase caused LPO through an increase in the generation of hydroxyl radicals. Hence, a decrease in SOD and CAT levels in the hepatic tissue of CCl_4-treated rats may lead to an increased accumulation of reactive products resulting in the oxidative injury of hepatic cells [20]. GSH plays a critical role in important cellular functions, which include the maintenance of the thiol status of proteins, the destruction of H_2O_2, lipid peroxides, free radicals, translocation of amino acids across cell membranes, the detoxification of foreign compounds, and the biotransformation of drugs [19, 20]. The decreased level of GSH in CCl_4-treated rat liver tissue may be due to its use by the extreme number of free radicals. This depletion not only compromises cellular defenses against attack by reactive molecules but also has profound effects on normal hepatocellular function [21]. GPx is an isoenzyme, that acts as a free radical scavenger, and traps peroxyl radicals before they can initiate lipid peroxidation [22]. Therefore, the depleted level of GPx is supposed to be utilized in the hepatic cells. Oral administration of *Gynandropsis pentaphylla* leaves extract to rats showed significant elevation in the depleted levels of SOD, CAT, GSH, and GPx due to the antioxidant activity of *Gynandropsis pentaphylla* leaves extract through scavenged free radicals. Further, noteworthy protection against CCl_4-induced hepatic antioxidant abnormalities was achieved with the silymarin treatment.

In the assessment of hepatic injury by CCl_4, the determination of enzyme levels such as SGOT, SGPT, ALP, GGT, and LDH were increased remarkably in plasma by the release of these enzymes from hepatic parenchymal cells, which were indicating a considerable hepatocellular injury [2]. Oral treatment with *Gynandropsis pentaphylla* leaves extract and silymarin attenuated these increased enzyme activities produced by CCl_4 and a subsequent recovery towards normalization of these enzymes strongly suggests the possibility of *Gynandropsis pentaphylla* leaves extract being able to improve the condition of hepatocytes so as to cause accelerated regeneration of hepatic parenchymal cells, thus protecting against membrane fragility, decreasing the leakage of marker enzymes into the circulation, stabilization of serum-total bilirubin and total protein levels through the administration of extract is further a clear indication of the improvement of a functional status of the hepatic cells [7, 21].

5. Conclusion

Biochemical alterations observed in hepatic oxidative injury seem to be mainly due to an oxy-radical-mediated mechanism, involving lipid peroxidation, under conditions of reduced antioxidant levels that scavenge superoxide, hydrogen peroxide, and lipid peroxides. The results yet available are overwhelming, which suggests that *Gynandropsis pentaphylla* extract may be helpful in quenching free radicals and induction of an *in vivo* antioxidant defense system might be due to the presence of tannins, alkaloids, terpenoids, steroids, total phenolics, and flavonoids in the extract and/or the purified compounds-Lupeol, β-sitosterol, α-amyrin, and β-amyrin which are present in the extract.

6. Conflicts of Interest

The authors declare no conflict of interest.

7. Acknowledgments

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8. References

1. Rao MRK, Kumar SS. Preliminary phytochemical analysis and antioxidant properties of *Gynandropsis pentaphyla*. Der Pharmacia Lettre. 2015;7(12):20-24.
2. Singh D, Arya PV, Singh V, Arya D, Bhagour K, Gupta RS. Role of *Cressa cretica* on CCl_4- induced liver damage in experimental rats. Journal of Pharmacognosy and Phytochemistry. 2018;7(1):1368-1373.
3. Dodiya T. Pharmacognostic and preliminary phytochemical investigation of *Gynandropsis gynandra* Linn. International Journal of Ayurveda and Pharmaceutical Chemistry. 2019;11(2):177-185.
4. Lokesha R. The African spider plant (*Gynandropsis pentaphylla* – DC. Syn. *Cleome gynandra* Linn.) (Capparaceae): Phytochemistry, pharmacological and biotechnological properties - a review. International Journal of Pharmacognosy and Chinese Medicine. 2018;2(4):000144.
5. Janovik V, Boligon AA, Frohlich JK, Schwanz TG, Pozzebon TV, Alves SH, *et al*. Isolation and chromatographic analysis of bioactive triterpenoids from the bark extract of *Cariniana domestica* (Mart) Miers. Natural Product Research. 2012;26(1):66-71.
6. da Silva UP, Furlani GM, Demuner AJ, da Silva OLM, Varejão EVV. Allelopathic activity and chemical constituents of extracts from roots of *Euphorbia heterophylla* L. Natural Product Research. 2019;33(18):2681-2684.
7. Singh D, Arya PV, Aggarwal VP, Gupta RS. Evaluation of antioxidant and hepatoprotective activities of *Moringa oleifera* Lam. leaves in carbon tetrachloride-intoxicated rats. Antioxidants. 2014;3:569-591.
8. Marklund S, Marklund G. Involvement of the superoxide anion radical in the autoxidation of pyrogallol and a convenient assay for superoxide dismutase. European Journal of Biochemistry. 1974;47(3):469-474.
9. Aebi H. Catalase *in vitro*. In: Colowick, SP, Kaplan, NO, editors. Methods in enzymology, Academic Press: New York. 1984;105:121-126.
10. Moron MS, Depierre JW, Mannervick B. Levels of glutathione, glutathione reductase and glutathione-s-transferase activities in rat lung and liver. Biochimica et Biophysica Acta (BBA) - General Subjects. 1979;582(1):67-78.
11. Paglia DE, Valentine WN. Studies on the quantitative and qualitative characterization of erythrocyte glutathione peroxidase. The Journal of Laboratory and Clinical Medicine. 1967;70(1):158-169.
12. Ohkawa H, Ohishi N, Yagi K. Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. Analytical Biochemistry. 1979;95(2):351-358.
13. Schneider WC, Hogeboom GH. Intracellular distribution of enzymes. V. Further studies on the distribution of cytochrome c in rat liver homogenates. Journal of Biological Chemistry. 1950;183(1):123-128.
14. Omura T, Sato R. The carbon monoxide binding pigment of liver microsomes. Journal of Biological Chemistry. 1964;239(7):2370-2378.
15. Latief U, Ahmad R. Herbal remedies for liver fibrosis: A review on the mode of action of fifty herbs. Journal of Traditional and Complementary Medicine. 2018;8(3):352-360.
16. Ghosh N, Ghosh R, Mandal V, Mandal SC. Recent advances in herbal medicine for treatment of liver diseases. Pharmaceutical Biology. 2011;49(9):970-988.
17. Singh D, Arya PV, Sharma A, Dobhal MP, Gupta RS. Modulatory potential of α-amyrin against hepatic oxidative stress through antioxidant status in wistar albino rats. Journal of Ethnopharmacology. 2015;161:186-193.
18. Mandal PK, Anupam Bishayee A, Chatterjee M. Stimulation of tissue repair by *Mikania cordata* root extract in carbon tetrachloride-induced liver injury in mice. Phytotherapy Research. 1993;7(1):103-105.
19. Singh V, Singh D. Antioxidative stimulation by di-and tri-organotin (IV) oximates against hepatic peroxidative stress in experimental animals. International Journal of Chemical Studies. 2019;7(1):443-448.
20. Singh D, Arya PV, Sharma A, Aggarwal VP, Dobhal MP, Gupta RS. Antioxidant potential of plumieride against CCl_4-induced peroxidative damage in rats. Antioxidants. 2014;3:798-813.
21. Singh D, Arya PV, Koolwal N, Singh V, Saxena R, Sharma MC, *et al*. Protective role of *Bacopa monniera* L. against hepatic oxidative stress in wistar albino rats. Journal of Pharmacognosy and Phytochemistry. 2015;4(4):233-241.
22. Ray G, Husain SA. Oxidants, antioxidants, and carcinogenesis. Indian Journal of Experimental Biology. 2000;40(11):1213-1232.