Research Article

Evaluation of Hepatoprotective and Antioxidant Activity of *Polygala javana* DC Whole Plant - CCl_4 Induced Hepatotoxicity in Rats

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ABSTRACT

The effect of ethanol extract of *Polygala javana* whole plant was evaluated in carbon tetrachloride induced hepatotoxicity in rats. Liver necrosis was produced by administrating single dose of carbon tetrachloride (CCl_4 2.5ml/kg body weight with normal saline) for 14 days. The liver damage was evidenced by elevated levels of serum glutamate oxaloacetate transaminase (SGOT), serum glutamate pyruvate transaminase (SGPT), alkaline phosphate (ALP), total, conjugated and unconjugated bilirubin reduced, the liver antioxidant such as superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and glutathione reductase (GRD). Ethanol extract of whole plant of *Polygala javana* pretreatment (100 and 200 mg/kg body weight) significantly (p<0.01; p< 0.05) reduced CCl_4 induced elevation of SGOT, SGPT, ALP, total, conjugated and unconjugated bilirubin. While the reduced concentration of SOD, CAT, GPx and GRD were reversed. Silymarin (100 mg/kg body weight) a known hepatoprotective drug showed similar results.

Keywords: *Polygala javana*, Hepatoprotective, ALP, Bilirubin, MAD.

INTRODUCTION

Liver diseases, especially viral hepatitis occurs predominantly in the developing world with an enormous impact on public health and economy[1]. Liver is a key organ regulating homeostatis within the body by various functions. Liver injury caused by toxic chemicals and certain drugs has been recognized as a toxicological problem. Hepatotoxicity is one of very common ailment resulting into serious debilities ranging from severe metabolic disorders to even mortality[2].

Herbs play a major role in the management of various liver disorders along with other system associated diseases. Plant drugs in Indian ayurvedic system of medicine and Chinese herbal medicine have long been used for liver and biliary diseases. Some plants have also been found to possess hepatoprotective activity and the underlying mechanism of action involves their antioxidant property[3-5].

Polygala javana belongs to Polygalaceae family. It is commonly known as “Palpiranthi”. Paste prepared from Fresh leaves is applied by Kanikkar tribal woman on the breast twice a day for 2-3 days to check lactation and to get relief from the pain developed while lactating[6].Biological activities such as anti-inflammatory, antidiabetic, anti-fertility and *in vivo* and *in vitro* antioxidant activities were reported[7-10]. However, no work has been reported on the hepatoprotective property of this plant. Keeping in view, the present study has been undertaken to investigate hepatoprotective activity and antioxidant role of the ethanol extract of *P. javana* on CCl_4 induced liver damage in rats.

Carbon tetrachloride (CCl_4) is one of the most commonly used hepatotoxins in the experimental study of liver diseases. The hepatotoxic effect of CCl_4 is largely due to its active metabolic, trichloro methyl radical[11]. The administration of CCl_4 in rats enhances hepatic protein oxidation and results in the accumulation of CCl_4 oxidized proteins in the liver[12]. The present study was conducted to evaluate the hepatoprotective effect of the extracts of whole plant of *P. javana*.

MATERIALS AND METHODS

Plant material

The well grown whole plant of *Polygala javana* DC was collected from Courtallam, Tirunelveli District, Tamil Nadu. The collected plants were

identified by the Botanical Survey of India, Coimbatore. A voucher specimen was retained in Ethnopharmacology unit, Research Department of Botany, V.O.Chidambaram College, Tuticorin for further reference.

Preparation of plant extracts for phytochemical Screening and Hepatoprotective Studies

The whole plant was dried under shade and then powdered with a mechanical grinder to obtain a coarse powder, which was then subjected to extraction in a Soxhlet apparatus using ethanol. The extract was subjected to qualitative test for the identification of various phytochemical constituents as per standard procedures[13-15].The ethanol extracts were concentrated in a rotary evaporator. The concentrated ethanol extract were used for hepatoprotective studies.

Animals

Normal healthy male Wistar albino rats (180-240g) were used for the present investigation. Animals were housed under standard environmental conditions at room temperature (25±2°C) and light and dark (12:12h). Rats were fed with standard pellet diet (Goldmohur brand, MS Hindustan lever Ltd., Mumbai, India) and water *ad libitum.*

Acute Toxicity Studies

Acute oral toxicity study was performed as per OECD-423 guidelines (acute toxic class method), albino rats (n=6) of either sex selected by random sampling were used for acute toxicity study [16]. The animals were kept fasting for overnight and provided only with water, after which the extracts were administered orally at 5mg/kg body weight by gastric intubations and observed for 14 days. If mortality was observed in two out of three animals, then the dose administered was assigned as toxic dose. If mortality was observed in one animal, then the same dose was repeated again to confirm the toxic dose. If mortality was not observed, the procedure was repeated for higher doses such as 50, 100 and 2000 mg/kg body weight.

Experimental Design

In the investigation, a total of 25 rats (CCl_4 hepatic toxicity induced rats and 5 normal rats) were taken and divided into five groups of 5 rats each.

Group I: Rats received normal saline was served as a normal control.

Group II: CCl_4 hepatic toxicity induced control: Rats received 2.5ml/kg body weight of CCl_4 for 14 days.

Group III: Liver injured rats received ethanol extract of whole plant of *P. javana* at the dose of 100mg/kg body weight for 14 days.

Group IV: Liver injured rats received ethanol extract of whole plant of *P. javana* at the dose of 200mg/kg body weight for 14 days.

Group V: Liver injured rats received standard drug silymarin at the dose of 100mg/kg body weight for 14 days.

Biochemical Analysis

The animals were sacrificed at the end of experimental period of 14 days by decapitation. Blood was collected, sera separated by centrifugation at 3000g for 10 minutes. Serum protein[17] and serum albumins was determined quantitatively by colorimetric method using bromocresol green. The total protein minus the albumin gives the globulin. Serum glutamate pyruvate transaminase (SGPT), serum glutamate oxaloacetate transaminase (SGOT), serum alkaline phosphatase (ALP), total, conjugated bilirubin, unconjugated bilirubin were determined as per the standard procedures[18, 19]. Liver homogenates (10%W/V) were prepared in ice cold 10mM tris buffer (pH7.4). Quantitative estimation of MDA formation was done by determining the concentration of thiobarbituric acid reactive substances (TBARS) in 10% liver homogenates by the method of Pal et al[20]. Antioxidants such as superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and glutathione reductase (GRD) were also assayed in liver homogenates as per the standard procedures[21, 22].

Statistical Analysis

The data were expressed as the mean ± S.E.M. The difference among the means has been analyzed by one-way ANOVA. $p<0.05$and $p<0.01$ were considered as statistical significance using SPSS Software.

RESULTS

The ethanol extract of whole plant of *Polygala javana* subjected for phytochemical study showed the presence of alkaloids, coumarin, glycosides, flavonoids, saponins, steroids, phenols, tannins and xanthoproteins. The ethanol extract did not show any sign and symptoms of toxicity and mortality upto 2000mg/kg dose. The effect of ethanol extract of *P. javana* on serum total protein, albumin, globulin, A/G ratio, serum transaminases, alkaline phosphatases in CCl_4 intoxicated rats are summarized in Table 1. There was a significant ($p< 0.01$) increase in serum GOT, GPT and ALP levels in CCl_4 intoxicated group (Group II) compared to the normal control

group (Group I). The total protein and albumin levels were significantly (p< 0.01) decreased to 6.18g/dl and 3.28g/dl in CCl_4 intoxicated rats from the levels of 7.98g/dl and 4.88g/dl respectively in normal group. Ethanol extract of *P. javana* whole plant at the dose of 100 and 200mg/Kg orally significantly decreased the elevated serum marker enzymes and reversed the altered total protein and albumin to almost normal level.

The effect of ethanol extract of *Polygala javana* on total, conjugated and unconjugated bilirubin is shown in Table 2. A significant elevation of total, conjugated and unconjugated bilirubin in the serum of CCl_4 intoxicated group (Group II) when compared to normal control (Group I). The ethanol extract of *Polygala javana* at the dose 100 and 200mg/kg reduced the levels of total, conjugated and unconjugated bilirubin (Group III and Group IV). The decreases in the concentration of total bilirubin, conjugated bilirubin and unconjugated bilirubin were found to be greater in standard silymarin (Group V) followed by Group IV and Group III (Table 2).

The effects of ethanol extract of *P. javana* on lipid peroxidation (LPO), Glutathione peroxidase (GPx), Glutathione reductase (GRD), Superoxide dismutase (SOD) and Catalase (CAT) activity is shown in Table 3. Lipid peroxidation level was significantly (p<0.01) increased and glutathione peroxidase, glutathione reductase, superoxide dismutase and catalase activity were significantly (p< 0.01) decreased in CCl_4 intoxicated rats when compared with those of the animals in normal control group. Rats treated with ethanol extract of *Polygala javana* at the doses of 100 and 200 mg/kg significantly decreased the elevated lipid peroxidation levels and restored the altered glutathione peroxidase, glutathione reductase, superoxide dismutase and catalase levels towards the normal levels in a dose dependent manner. The results are well comparable with silymarin (standard drug) treated group.

DISCUSSION

It is well established that CCl_4 induces hepatotoxicity by metabolic activation; therefore it selectively causes toxicity in liver cells maintaining semi-normal metabolic function. CCl_4 is bio-transformed by the cytochrome P_{450} system in the endoplasmic reticulum to produce trichloromethyl free radical (CCl_3). Trichloromethyl free radical then combined with cellular lipids and proteins in the presence of oxygen to form a trichloromethyl peroxyl radical, which may attack lipids on the membrane of endoplasmic reticulum faster than trichloromethyl free radical. Thus, trichloromethyl free radicals lead to elicit lipid peroxidation, the destruction of Ca^{2+} homeostasis and finally, results in cell death[23, 24]. These results in changes of structures of the endoplasmic reticulum and other membrane, loss of enzyme, metabolic enzyme activation, reduction of protein synthesis and loss of glucose-6-phosphate activation, leading to liver damage[25, 26]. Hepatotoxic compounds like CCl_4 are known to cause marked elevation in serum enzyme activities. In the present study, treatment with *P. javana* whole plant extract attenuated the increase in the activities of SGOT, SGPT and ALP produced by CCl_4 indicating that *Polygala javana* whole plant extract protects liver injury induced by CCl_4 towards normalization. Silymarin, a prototype hepatoprotective agent also showed similar changes.

Bilirubin is the main bile pigment that is form the breakdown of heme in the red blood cells. It is transported to the liver where it is secreted by the liver into the bile. Conjugation of bilirubin is a prerequisite for its excretion into the bile[27]. Malfunctioning of the liver was evidenced by the significant increase (p< 0.01) in the level of unconjugated bilirubin in the serum of the group treated with only CCl_4 when compared to normal control. Increase in the level of unconjugated bilirubin in the blood may result from a defect in the function of the liver to conjugate the bilirubin being produced. The significant reduction (p< 0.05) of unconjugated bilirubin level in the serum when CCl_4 was simultaneously administrated with the ethanol extract of *P. javana* when compared with the administration of CCl_4 alone indicates that the conjugating function of the liver was improved. The reduction of the unconjugated bilirubin level by the ethanol extract suggest that the extracts may activate the constitutive androstance receptor (CAR) which is a key regulator in bilirubin clearance in the liver[28]. The primary function of CAR is the bilirubin clearance pathway is to direct cooridnate response to elevated levels of bilirubin by increasing the hepatic expressive of each component of the pathway[29].

The ability of simultaneous administration of CCl_4 with ethanol extract of *P. javana* to significantly reduce (p< 0.01) the level of serum total bilirubin when compared with that of the CCl_4 treated group suggests the potential of the extract is clearing bilirubin from the serum when its level elevated. Since the results obtained for the serum total protein and albumin concentrations followed the same trend, it thus implicated the same mechanism by which the ethanol extract of *Polygala*

javana exerts its effect on these parameters. The administration of CCl_4 alone may adversely interfere with protein metabolism probably by inhibiting the synthesis of proteins. Administration of ethanol extract of *P. javana* whole plant reversed these changes may be by increasing protein synthesis. This indicates the hepatoprotective activity of *P. javana* whole plant against damage by CCl_4. Stimulation of protein synthesis has been advanced as a contributory hepatoprotective mechanism which accelerates regeneration of cells[30].

Lipid peroxidation has been postulated to the destructive process of liver injury due to CCl_4 administration. In the present study the elevations in the levels of end products of lipid peroxidation in the liver of rat treated with CCl_4 were observed. The increase in malondialdehyde (MDA) levels in liver suggests enhanced lipid peroxidation leading to tissue damage and failure of antioxidant defense mechanism to prevent formation of excessive free radicals. Treatment with ethanol extract of *P. javana* significantly reversed these changes. Hence it may be possible that the mechanism of hepatoprotection by *P. javana* extract is due to its antioxidant effect.

The enzyme antioxidant defense system is the nature protector against lipid peroxidation. SOD, CAT and GPx enzymes are important scavengers of superoxide ion and hydrogen peroxide. These enzymes prevent generation of hydroxyl radical and protect the cellular constituents from oxidative damage[31]. In the present study, it was observed that the ethanol extract of *P. javana* significantly ($p< 0.01$) increased the hepatic SOD activity in CCl_4 induced liver damage in rats. Ethanol extract of *P. javana* can reduce reactive free radicals that might lesson oxidative damage to the tissues and improve the activities of the hepatic antioxidant enzyme.

Catalase (CAT) is an enzymatic antioxidant widely distributed in all animal tissues and the highest activity is found in the red cells and in the liver. CAT decomposed hydrogen peroxide and protects the tissue from highly reactive hydroxyl radicals[32]. Therefore the reduction in the activity of these enzymes may result in a number of deleterious effects due to the accumulation of superoxide radicals and hydrogen peroxide. Administration of ethanol extract of *Polygala javana* increased the activities of CAT in CCl_4 induced liver damage in rats to prevent the accumulation of excessive free radicals and protected the liver from CCl_4 in toxication.

To prevent lipid peroxidation, it is very important to maintain the level of GSH, GSSG is reduced to GSH by GR, which is NADPH-dependent. It plays a role in maintaining adequate amounts of GSH. Accordingly, the reduction of GR results in decreasing GSH[33]. In CCl_4 intoxicated rats, the activity of GR is significantly ($p<0.05$) decreased. However, ethanol extract of *P. javana* with 100 and 200 mg/kg bodyweight brought the activity of GR towards of normalization.

In conclusion, the results of this study demonstrate that the ethanol extract of *P. javana* whole plant have a potent hepatoprotective action against CCl_4 induced hepatic damage in rats. The hepatoprotective and antioxidant potential of extract could have been brought about by various phytochemical principles i.e. flavonoids, alkaloids, phenolics, saponins and tannins present in *P. javana* whole plant. The enhanced levels of antioxidant enzymes and reduced amount of lipid peroxides are suggested to be the major mechanism of *P. javana* ethanol extract in prevents the development of liver damage induced by CCl_4.

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Table 1: Effect of whole plant extracts of *Polygala javana* on the protein, albumin, globulin concentration and enzyme activity of serum GOT, GPT, and ALP in the normal, liver damaged and drug treated rats

Groups	Parameters						
	T.Protein (mg/dl)	Albumin (g/dl)	Globulin (g/dl)	A/G Ratio	SGOT (U/L)	SGPT (U/L)	ALP (U/L)
I	7.98±0.81	4.88±0.34	3.1±0.11	1.5:1	19.56±1.36	26.16±0.93	143.29±5.32
II	6.18±0.34*	3.28±0.11*	2.9±0.23	1.1:1	40.11±1.21*	43.19±1.08*	196.11±6.84*
III	6.78±0.16	3.91±0.17	2.87±0.34	1.3:1	29.16±1.13*	24.61±1.16*a	157.31±7.15*
IV	7.56±0.21*a	4.08±0.12*a	3.48±0.16	1.1:1	21.31±1.73**a	27.19±1.31*a	155.16±5.27
V	7.48±0.11*	4.51±0.31*	2.97±0.16	1.5:1	21.33±1.19*a	27.06±1.33*a	146.55±6.94*a

Each Value is SEM ± 5 individual observations * P < 0.05; ** P<0.01 Compared normal control vs liver injured rats
A P < 0.05; aa P<0.01 Compared liver injured rats vs drug treated

Table 2: Effect of whole plant extracts of *Polygala javana* on the serum Total, conjugated and unconjugated bilirubin levels in the normal control, liver injured and drug treated rats

Groups	Parameters		
	Total Bilirubin (µmol/L)	Conjugated (µmol/L)	Unconjugated (µmol/L)
I	0.68±0.03	0.24±0.01	0.44±0.02
II	3.69±0.43**	1.49±0.03*	2.20±0.06**
III	2.23±0.38	0.43±0.01	1.80±0.11
IV	1.32±0.14**	0.31±0.03*	1.01±0.04*
V	0.88±0.01**a	0.20±0.01*a	0.68±0.3*aa

Each Value is SEM ± 5 individual observations * P < 0.05;
** P<0.01 Compared normal control vs liver injured rats
a- P < 0.05; aa - P<0.01 Compared liver injured rats vs drug treated

Table 3: Effect of whole plant extracts of *Polygala javana* on liver LPO,GPX,GRD, SOD and CAT in the normal control, liver injured and drug treated rats

Groups	Parameters				
	LPO (n mole of MDA/mg protien)	GPX (u/mg Protien)	GRD (u/mg)	SOD (u/mg)	CAT (u/mg)
I	0.82±0.03	13.54±1.23	8.56±0.59	10.45±0.19	9.56±0.82
II	3.16±0.54**	4.22±0.56**	3.05±0.28*	4.22±0.28**	2.87±0.70**
III	1.89±0.21*	9.68±0.86^{a}	5.53±0.19	5.23±0.83*	7.85±0.64^{aa}
IV	1.04±0.14^{a}	11.64±0.67^{aa}	8.12±0.38^{aa}	9.45±0.74^{aa}	11.34±0.58^{aa}
V	0.89±0.11^{aa}	12.56±0.97^{aa}	8.67±0.11^{aa}	11.71±0.12^{aa}	13.56±0.55^{aa}

Each Value is SEM ± 5 individual observations * P < 0.05 ;
** P<0.01 Compared normal control vs liver injured rats
A P < 0.05; aa P<0.01 Compared liver injured rats vs drug treated

REFERENCES

1. Mondal SK, Chakraborthy G, Gupta M and Mazumder UK. Hepatoprotective activity of Diospyrus malabarica bark in carbon tetrachloride intoxicated rats. European Bulletin of Drug Research. 2005; 13:25-30.
2. Patel RK, Patel MM, Patel MP, Kanzaria NR, Vaghela KR and Patel NJ. Hepatoprotective activity of Moringa oleifera Lam. fruit on isolated rat hepatocytes. Phcog mag. 2008; 4:118-123.
3. Gupta M, Mazumder UK and Sampathkumar R. Antioxidant and protective effects of Ervatamia coronarai Stapf. Leaves against CCl_4 – induced liver injury. European Bull Drug Res. 2004a; 12: 13-22.
4. Gupta M, Mazumder UK and Sivakumar R. Antioxidant and protective effects of Bauhinia

racemosa against paracetamol and CCl_4 – induced liver injury in rats. Iranian J Pharmacol Therapeutics. 2004b; 12: 20.
5. Kukongviriyapan V, Janyacharoen T and Kukonguviriyapan U. hepatoprotective and antioxidant activities of Tetracera loureiri. Phytother Res. 2003; 17:717-721.
6. Lalitha rani S, Kalpanadevi V, Tresinasoris P, Maruthupandian A and Mohan VR. Ethnomedicinal plants used by kannikar of Agasthiarmalai Biosphere Reserve Western ghats. J Ecobiotech. 2011;3: 16-25.
7. Alagammal M, Paulpriya K and Mohan VR. Evaluation of anti-inflammatory activity of ethanol extracts of Polygala javana DC whole plant. Int research journal of Pharmacy. 2012; 3: 212-213.
8. Alagammal M, Agnel Ruba A and Mohan VR. Antidiabetic and antihyperlipidaemic effect of P. javana DC on Alloxan induced diabetic rats. Int Research J Pharmacy. 2012; 3: 231-234.
9. Sakthidevi G, Alagammal M and Mohan VR. Anti-fertility efficacy of the plant Polygala javana DC on male albino rats. Pharma Science Monitor. 2012.
10. Sakthidevi G and Mohan VR. Comparative in vitro freeradical scavenging activity of Polygala javana, P. chinensis and P. rosmarinifolia Wight & Arn (Polygalaceae). J Cur Chem Pharm Sci. 2012; 2: 294-298.
11. Das KK, Das SN and Das Gupta S. The influence of ascorbic acid on nickel induced hepatic lipid peroxidation in rats J Basic Clin Physiol Pharmacol. 2001;12:187-195.
12. Premila Abraham P, WilFred G and Ramakrishna B. Decreased activity of hepatic alkaline protease in rats with carbon tetrachloride-induced liver cirrhosis. Indian J Exp Biol. 1999;37: 1243-1244.
13. Brinda P, Sasikala P and Purushothaman KK. Pharmacognostic studies on Merugan kizhangu. Bulletin in Medical Ethnobotanical Research. 1981;3:84-96.
14. Anonymous. Phytochemical investigation of certain medicinal plants used in Ayurveda. Central Council for Research in Ayurveda and Siddha, New Delhi: 1990.
15. Lala PK. Lab manuals of Pharmacognosy CSI Publishers and Distributers, Kolkata: 1993
16. OECD, (Organisation for Economic co-operation and Development). OECD guidelines for the testing of chemicals/Section 4: Health Effects Test No. 423; Acute oral Toxicity-Acute Toxic Class method. OECD. Paris: 2002.
17. Lowry OH, Rosenbrough NJ, Farr AL and Randall RJ. Protein measurement with the Folin's phenol reagent. Journal of Biological Chemistry. 1951;193:265-275.
18. Shanmugasundaram R, Kalpanadevi V, Tresina PS, Maruthupandian A and Mohan VR. Hepatoprotective activity of ethanol extracts of Clitoria ternatia L. and Cassia angustifolia Vahl. leaf against CCl_4 induced liver toxicity in rats. Int Research J Pharmacy. 2010;1:201-205.
19. Thangakrishnakumari S, Nishanthini A, Muthukumarasamy S and Mohan VR. Hepatoprotective and antioxidant activity of Canscora perfoliata Lam (Gentiaceae) against CCl_4 induced hepatotoxicity I rats. Int. J. Res. Ayur. Pharm. 2012;3: 822-826.
20. Pal A, Banerjee B, Banerjee T, Masih M and Pal K. Hepatoprotective activity of Chenopodium album Linn. plant against Paracetamol induced hepatic injury in rats. Int J Pharm Sci. 2011;3: 55-57.
21. Anitha M, Daffodil ED, Muthukumarasamy S and Mohan VR. Hepatoprotective and antioxidant activity of ethanol extract of Cynoglossum zeylanicum (Vahlex Hornem) Thurnb ex Lehm in CCl_4 treated rats. J Applied Pharmaceutical Science. 2012; 2: 099-103.
22. Suky TMG, Parthiban B, Kingston C, Tresina soris P and Mohn VR. Hepatoprotective and antioxidant activity effect of Balanites aegyptiaca (L) Del against CCl_4 induced heptotoxicity in rats. Int J Pharmaceutical science and Research. 2011; 2: 887-892.
23. Shanaz Banu, Arunachalam G, Jajaveera KN, Ashoka Babu VL and Vimal Kumar. Hepatoprotective activity of methanolic extract of Barleria Montana leaves in ethanol treated rats. Asian Pac J Trop Diseas. 2012; S748-S752.

24. Velayudam, Arul Amuthan and Ilavarasan. Hepatoprotective activity of Kadukkai Maathrai (ASiddha polyherbal formulation) against CCl_4 induced liver damage in rat. Research J Pharmaceutical Sciences. 2012; 1: 17-21.
25. Vipin Kumar, Pankaj K Modi and Saxena KK. Exploration of heptoprotective activity of aqueous extract of Tinospora cordifolia an experimental study. Asian J Pharmaceutical and Clinical research. 2012; 6: 87-91.
26. Sheeba KO, Wills PJ, Latha BK, Rajalekshmy R and Latha MS. Antioxidant and antihe patotoxic efficacy of methanolic extract of Elephantopus scaber Linn in Wistar rats. Asian Pac J Trop Disease. 2012; S904-S908
27. Iniaghe OM, Malomo S and Adebayo JO. Hepatoprotective effect of the aqueous extract of leaves of Acalypha racemosa in carbon tetrachloride treated rats. Int J Med Plants Res. 2008; 2; 301-305.
28. Moore D, Jun Z and Werdorg H. Induction of bilirubin clearance by xenobiotic receptor CAR. J Clinc Invest. 2004; 113: 137-143.
29. Gupta M, Mazumdar UK, Kumar RS, Sivakumar T, Gomathi P and Rajeshwar Y. Antioxidant defense system induced by a methanol extract of Caesapinia bonducella in rat liver. Pharmaceutical Biology. 2005; 43: 411-419.
30. Rajalakshmi G, Arul Jothi K, Venkatesan RS and Jegatheesan K. Hepatoprotective activity of Andrographis paniculata on paracetamol induced liver damage in rats. J Pharmacy Research. 2012; 5: 2983-2986.
31. Rajesh V, Perumal P, Rajesh G and Rajavel R. Hepatoprotective and in vivo antioxidant activity of methanol extract of Carica papaya Linn. Leaves on paracetamol induced liver damage in rats. J Pharmacy Research. 2012; 5: 3085-3091
32. Arun K and Balasubramanian U. Comparative study on hepatoprotective activity of Aegle marmelos and Eclipta alba against alcohol induced in albino rats. Int J Environmental Sciences. 2011; 2: 389-402.
33. Reckenge RO, Glende Jr, EA and Britton RS. Free radical damage and lipid peroxidation. In: Histotoxicology meeks, R.G., Harrison, S.D., Bull, R.J. (Eds.), CRC Press; Florida 1991:401-436.