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Original Article

Toxicities of selected medicinal plants and floras of lower phyla

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ABSTRACT

The aim of this study was to evaluate the toxic effects associated with the administration of aqueous extracts (AE) of *Calliandra portoricensis* (CP), *Dracaena arborea* (DA), *Duranta repens* (DR), *Polytrichum juniperinum* (PJ), *Parmelia caperata* (PC), and *Nostartium officinale* (NO) on Wistar rats. LD₅₀ for each plant was obtained prior to administration. Seven groups of six rats each were orally gavaged for 28 days as follows; group 1–7 received normal rat pellets and saline, in addition, group 2 received 20 mg/kg b.w CP, group 3 & 4 respectively received 8 mg/kg b.w DA and DR, group 5 & 6 respectively received 4 mg/kg b.w PJ and PC, and group 7 received 100 mg/kg b.w NO. Liver enzymes; ALP, ALT, AST and GGT were significantly ($p < 0.05$) elevated by CP, DR, PJ and PC extracts. All the extracts caused significant alterations of the total protein, albumin and globulin levels. The urea levels were deranged by all the extracts while CP, PJ, PC, and NO extracts caused no significant effects on the creatinine levels. Both DR and NO deranged the serum electrolytes; Na, K, Cl, and HCO₃. Results for the lipid profile showed that all extracts significantly altered the phosphatidate phosphohyrolase and LDL levels while no significant effects were observed in the VLDL, TG, TC, HDL, cardiac risk ratio, arterogenic coefficients, and arterogenic index of plasma, of NO treated rats. For hematological parameters DR, PJ, and PC significantly deranged the RBC, HGB, MCHC, MCV, and MCH concentrations while the neutrophils, eosinophils and basophils were significantly altered on administration of all the extracts. No significant effects were observed on the platelets and plateletcrit level in rats gavaged with CP, whereas the MPV, PDW, and PCT concentrations were deranged by DR extracts. CP and NO caused no alterations in the MDA, GSH, and GST levels whereas the SOD, GPx, and xanthine oxidase levels were significantly deranged by all the plant extracts. Only NO treatment produced catalase, glutathione reductase, and xanthine dehydrogenase levels equivalent to the control group. This study has shown various degrees of deleterious effects on biochemical parameters associated with the consumption of these plants, thus raising serious concerns over their continuous applications as local medicaments.

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1. Introduction

The preference of plants over synthetic chemicals for treatment of diseases has resulted to extensive worldwide investigations into their applicability as medicaments.¹ However, this subjective interest in elucidating the therapeutic potentials of plants has consequently reduced the number of plants evaluated for their deleterious effects.² In other words, investigations related to plant efficacies greatly outnumber reports on toxicities.^{3,4} Thus, there is need to extend the elucidation of phytotherapies to include both long and short term toxic effects. *Calliandra portoricensis* also called

powder puff is an evergreen shrub of the Mimosaceae family that bears small bipinnate leaves and white globose flowers.^{5,6} It is applied in Nigerian tradomedical system as an arbotifacient,⁷ to expel worms,⁸ and according to Orishadipe et al.,⁹ possesses antiskickling and anticancer properties. Also, Aqil et al.,¹⁰ have reported its efficacy against convulsion, fever, diarrhea, malaria, and rheumatism, hence, the plant is regarded as a pharmacologically important herb. *Dracena arborea*, also referred to as the Dragon Tree, is an abundantly distributed member of the plant family; Agavaceae.^{11,12} Local inhabitants know the plants as Igede in Igbo, and Peregum in Yoruba.¹² In Nigeria, the plant is used as an analgesic, to relieve high blood pressure, for postpartum revitalization, and also applied as an arrow poison.¹² *Duranta repens* (Golden Dewdrop) is an ornamental shrub belonging to the Verbenaceae family, and widely distributed in various parts of Nigeria.¹³ The

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plant is toxic to grazing livestock.¹⁴ Castro et al.,¹⁵ reported the application of the plant as an antimalarial agent, while Ahmad et al.,¹⁶ showed its efficacy in the treatment of abscess. *Polytrichum juniperium* is a perennial evergreen moss species distributed all over the world.¹⁷ The leaves are classified as bryophytes with tissues capable of photosynthesizing at very low temperatures.¹⁸ Ethnobotanical efficacies of *P. juniperinum* include treatment of pneumonia, renal diseases, and hemorrhage.¹⁹ Also, Krzaczkowski et al.,²⁰ and Savaroglu et al.,²¹ have reported its fungicidal, bacteriostatic and cytotoxic properties. *Flavoparmelia caperata* is a lichen belonging to the Parmeliaceae family. When dry, it has a greenish yellow upper cortex with smooth rounded lobes at the thallus. Lichens generally produce taxonomically significant secondary metabolites with so much biological effects.²² On several occasions, lichens have been applied for medicinal purposes such as for pain relief, antitumor, antioxidant and antipyretic effects.²³ In addition, lichens have also shown toxicities to herbivores, thus serving as a feeding deterrent.²⁴ *Nasturtium officinale* (Water Cress) is a medicinal dicotyledonous plant classified under the family of Brassicaceae that grows along water banks.²⁵ Its therapeutic effects have been related to high amount of constituent glucosinolates.²⁶ Water cress has been used as a chemopreventive agent,²⁷ to enhance antioxidant capacity of cells,²⁸ and to relieve inflammations.²⁹ However, notwithstanding the evidence that all these above mentioned plants have been used in one way or another for the amelioration of health issues, a pertinent question is that while these plants are reportedly effective for the treatment of a particular ailment, what are their effects on vital organs like the liver, kidney, heart and blood cells? Do their consumptions bring about oxidative stress? In a bid to provide experimental views to these questions, this study was carried out to investigate the effects of *C. portoricensis*, *D. arborea*, *D. repens*, *P. juniperinum*, *P. caperata*, and *N. officinale* on Wistar rats.

2. Methodology

2.1. Sample collection and identification

Fresh leaves of *C. portoricensis*, *D. arborea*, *D. repens*, *P. juniperinum*, *P. caperata*, and *N. officinale* were obtained from a commercial garden owned by a botanist at Okigwe road, Owerri, Imo State Nigeria, and authenticated at the Department of Plant Science and Biotechnology, Imo State University, Owerri, by the botanist.

2.1.1. Sample preparation

The leaves were thoroughly washed and dried under room temperature. The dried samples were ground into finer particles and soaked in 500 ml of water for 24 h and filtered afterwards. The residues were continuously subjected to soxhlet extraction for three more times. The extracts obtained, were placed in a rotary evaporator under reduced pressure, to concentrate the extracts. This extraction process was carried out every 7 days to obtain fresh extracts administered to the experimental animals.

2.2. Experimental design

2.2.1. Determination of LD₅₀

Experimental handling of animals was in accordance with international guidelines on animal care and uses (NIH, 1985).

The LD₅₀ of the six plant leaves were obtained following the method of Lorke.³⁰ A total of one hundred and six albino mice were divided into six groups representing each of the extracts. The animals in each group were marked differently (3 each) representing those used for phase 1 and 2 of the applied doses. The LD₅₀ of the extracts was thus calculated through the geometric mean of the

highest dose with no observed mortality and lowest dose with 100% mortality. The LD₅₀ obtained were as follows; 100 mg/kg for *C. portoricensis*, 25 mg/kg for *D. arborea*, and *D. repens*, 10 mg/kg for *P. juniperinum* and *P. caperata*, and 400 mg/kg for *N. officinale*.

2.3. Administration of extracts

A total of fifty six (56) healthy male Wistar rats weighing 130–140 g were used. After acclimatization for seven days, they were divided into seven groups of six rats each and intestinally gavaged as follows;

Group 1–7: standard rat pellets and saline, *ad libitum*.

Group 1 (control group): received exclusively standard rat pellets and saline, *ad libitum*

Group 2: received 20 mg/kg AE of *C. portoricensis*.

Group 3 & 4: received 8 mg/kg AE of *D. arborea* and *D. repens* respectively.

Group 5 & 6: received 4 mg/kg AE of *P. juniperinum* and *P. caperata* respectively.

Group 7: received 100 mg/kg AE of *N. officinale*.

On the 28th day of administration of the extracts, the animals were sacrificed by cervical decapitation under mild anesthesia of ethyl ether. Both blood (collected by cardiac puncture) and sera was collected and prepared for different analysis to be carried out.

2.4. Liver function tests

Serum levels of the liver function markers; alkaline phosphatase (ALP), alanine transaminase (ALT), and aspartate transaminase (AST) were determined by following the instructions on the Randox reagent kit using 2, 4-dinitrophenylhydrazine as substrate.³¹

Following the method of Szasz,³² the gamma-glutamyl transpeptidase (GGT) activity was determined spectrophotometrically at 405 nm using the C-system γ-GT kit and γ-L-glutamyl-p-nitroanilide as substrate. The total bilirubin (T.bil) and conjugated bilirubin (C.bil) were colorimetrically determined using the method of Jendrassik and Grof,³³ while instructions on commercial kits (Laborlab, Guarulhos, SP, Brazil) were followed for determination of total proteins and serum albumin. Globulin was estimated by subtracting the albumin contents from total protein.

2.5. Kidney function tests

Mind ray test kits (Mindray Medical International Limited, China) were used for serum urea determination following the Urea-glutamyl Dehydrogenase -UV Berthelot method described by Weatherburn,³⁴ while creatinine and blood urea nitrogen (BUN) were determined using enzymatic colorimetric assay. Serum levels of the electrolytes were analyzed using audicom full auto electrolyte analyzer following the method of Ali et al.³⁵

2.6. Measurement of PAP activity and lipid profile

Precisely, 0.9% ice-cold saline was used to perfuse the inferior vena cava of the liver of the animals in order to remove inorganic phosphate and blood. With the aid of a homogenizer, an ice-cold buffer of pH 7.4 made up of EDTA (0.1 nM), sucrose (0.25 M), and PMSF (1mM) was used to homogenize the perfused liver, at 8×10^3 rpm for 6 min at 4 °C. The resulting liver homogenate was centrifuged at 4.5×10^2 rpm for 10 min at 4 °C, after which the supernatant was applied for the determination of PAP activity. The activity of phosphatidate phosphohydrolase (PAP) was measured spectrophotometrically as described by Yanagita et al.³⁶ with slight modifications. Briefly, an assay buffer made up of magnesium chloride (1.25 mM), phosphatidate (1 mM), Tris-HCl (50 nM at pH 7.4),

and 100 µg of the supernatant was incubated at 37 °C for 15 min. The reaction was controlled by adding 10% trichloroacetic acid (0.5 M). After 20 min, the phosphate molybdate color indicated the release of inorganic phosphate (Pi) and determined at 820 nm. The PAP activity was thus expressed as nanomoles of Pi/min/mg of protein.

Low density lipoproteins cholesterol (LDL-C), and very low density lipoprotein cholesterol (VLDL-C) were calculated by the method of Friedewald.³⁷ Total cholesterol (TC), triglycerides (TG), and high density lipoprotein cholesterol (HDL-C) were enzymatically determined using Sigma-Aldrich assay kits with BT-3000 auto analyzer (England).

The cardiac risk ratio, atherogenic coefficient, and atherogenic index of plasma were calculated as described by Ikewuchi et al.,³⁸ using the formula below;

$$\text{Cardiac Risk Ratio} = \text{Total cholesterol}/\text{HDL cholesterol}$$

$$\text{Atherogenic Coefficient} = \text{Cardiac risk ratio} - 1$$

$$\text{Atherogenic Index of Plasma} = \log \frac{\text{Triglyceride}}{\text{HDL cholesterol}}$$

2.7. Hematology

The red blood cells (RBC) and its indices (hemoglobin (HGB), mean corpuscular hemoglobin concentration (MCHC), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and hematocrit), white blood cells (WBC) count and its differentials (monocytes, neutrophils, eosinophils, basophils, and lymphocytes) and platelet count (PLT) and its differentials (mean platelet volume (MPV) and the platelet distribution width (PDW), and plateletcrit (PCT)) were determined using a BC-2600 model of haematology autoanalyzer (Bio-medical Electronics, UK).

2.8. In vivo measurement of oxidative stress indicators

Serum levels of malondialdehyde were measured as described by Ohkawa et al.,³⁹ through thiobarbituric acid reactive substance (TBARS) assay.

The superoxide dismutase (SOD) serum levels were determined through the inhibition of pyrogallol autoxidation.⁴⁰

Catalase was measured as the rate of decomposition of H₂O₂ at 240 nm for 30 sec interval for 5 min following the method described by Aebi.⁴¹

Glutathione reductase (GR) was estimated spectrophotometrically at 340 nm as reduction in absorption of NADPH following the description of Krohne-Ehrich et al.⁴²

The glutathione peroxidase (GPx) activity in whole blood was measured following the description of Paglia and Valentine,⁴³ using Randox GSH-px kit (RANSEL).

The determination of serum levels of glutathione-S-transferase was according to the description of Habig et al.⁴⁴ by spectrophotometrically measuring at 340 nm the rate of conjugation of reduced glutathione and 1-chloro, 2,4-dinitrobenzene (CDNB).

Determination of glutathione (GSH as acid-soluble sulfhydryl) levels was done by the method of Sedlak and Lindsay,⁴⁵ by monitoring its reaction with DTNB (Ellman's reagent) at 412 nm after 15 min.

Serum xanthine oxidase levels were spectrophotometrically measured at 295 nm following the method of Greenlee and Handler,⁴⁶ with slight modifications by using substrate containing xanthine standard (0.1 mM) in phosphate buffer (50 nM).

Serum xanthine dehydrogenase (XDH) levels were estimated by measuring the reduction of NAD⁺ to NADH according the method of Strittmatter.⁴⁷

2.9. Statistical analysis

The data obtained was expressed as mean ± SD of triplicate determinations. The data were analyzed using one way analysis of variance (ANOVA) by their least standard deviations (LSD). p values < 0.05 were considered as significant.

3. Results and discussion

Table 1 shows the effects of administration of *C. portoricensis*, *D. arborea*, *D. repens*, *P. juniperinum*, *P. caperata*, and *N. officinale*, on the hepatic dysfunction indicators of Wistar rats. The result showed that only the animals administered with water cress plant produced ALP, ALT, AST, GGT, T.Bil, and C. Bil levels comparable to the control, whereas other administered plant extracts significantly elevated the above mentioned liver function markers, with *D. arborea*, *P. caperata*, *C. portoricensis* extremely disrupting the ALP activities. Similar to the effects of water cress on ALT and AST activities, *D. arborea* produced significantly comparable results, while *P. caperata* showed no observable derangements of total bilirubin levels. The results of Table 1 further indicated that all the administered plants disrupted the GGT concentrations except for water cress and *D. arborea* extracts, while the total protein, albumin, and globulin levels were significantly altered by all the plants used in this study. Since tissue damage is associated with elevated levels of the enzymes in circulation, the selective alterations of the liver function indicators shown in this study by these plants necessitate an in-depth analysis of their bioactive components in order to affirm the cause(s) of the alterations. *P. caperata* and *P. juniperinum* though have been claimed in folklore to provide some medicinal properties, no report has been presented on their bioactive components to corroborate such claims. Water cress is particularly rich in phenolic acids and flavonoids, which could be responsible for the hepatoprotective properties,⁴⁸ while the antinutrients and other toxic phytochemicals present in *C. portoricensis*,⁴⁹ and *D. repens*¹³ are possibly responsible for their hepatotoxic effects shown in this study. Both ALP and AST are less sensitive hepatic damage markers than GGT and ALT, hence, the result of Table 1 implies that the functional status of the liver has been possibly compromised on administration of *C. portoricensis*, *D. repens*, *P. juniperinum*, and *P. caperata*. Further, the result showed elevated levels of total bilirubin on administration of *C. portoricensis*, *D. arborea* and *D. repens* possibly caused by an increased heme metabolism, however, the ability of the liver to conjugate bilirubin in these cases seemed appreciable. The consequent alterations in the total protein, albumin, and globulin levels on administration of these extracts provides further experimental evidence to complications in the liver's synthetic function.

The effects of administration of *C. portoricensis*, *D. arborea*, *D. repens*, *P. juniperinum*, *P. caperata*, and *N. officinale* on the kidney were shown in Table 2. The result revealed significant alterations in urea levels on administration of the extracts except for those animals that received *C. portoricensis*. Further, only *D. arborea* and *D. repens* produced deranged creatinine levels among the administered extracts while the blood urea nitrogen levels of animals in all the groups in Table 2, remained significantly equivalent to the control. The results also showed significant elevated sodium levels on administration of *C. portoricensis*, *D. repens*, and *N. officinale* but produced no significant effects on the zinc levels, while only the administration of *C. portoricensis* and *P. juniperinum* extracts had no significant effects on the potassium levels. The anions (Cl and HCO₃) were both significantly deranged by *C. portoricensis*, *D. repens*, *P. caperata*, and *N. officinale*. Urea and creatinine are both considered as reliable indicators of kidney functional status, and alterations in their levels suggest renal impairment.^{50,51} The urea

Table 1
Hepatic effects of some medicinal plants and floras of lower phyla.

Grp	ALP (U/L)	ALT (U/L)	AST (U/L)	GGT (U/L)	T. Bil (μmol/L)	C. Bil (μmol/L)	T. Pro (g/dl)	Alb (g/dl)	Glob (g/dl)
1	43.16 ± 1.76 ^a	48.16 ± 1.10 ^a	26.31 ± 0.75 ^a	3.78 ± 0.04 ^a	1.24 ± 0.09 ^a	0.61 ± 0.02 ^a	8.63 ± 0.23 ^a	5.70 ± 0.22 ^a	2.93 ± 0.17 ^a
2	57.61 ± 2.09 ^b	52.28 ± 1.72 ^b	39.36 ± 2.70 ^b	4.23 ± 0.16 ^{be}	1.51 ± 0.09 ^b	0.76 ± 0.04 ^b	6.84 ± 0.16 ^b	5.04 ± 0.16 ^b	1.80 ± 0.05 ^b
3	63.51 ± 2.89 ^c	48.31 ± 1.85 ^a	27.63 ± 2.26 ^a	3.48 ± 0.25 ^a	1.83 ± 0.07 ^c	0.86 ± 0.04 ^c	5.72 ± 0.07 ^c	4.42 ± 0.07 ^c	1.30 ± 0.01 ^c
4	56.07 ± 1.55 ^b	62.06 ± 1.65 ^c	38.99 ± 1.02 ^b	4.05 ± 0.05 ^b	1.70 ± 0.08 ^c	1.16 ± 0.07 ^d	6.63 ± 0.11 ^{bd}	4.72 ± 0.09 ^d	1.90 ± 0.06 ^b
5	50.96 ± 1.72 ^d	55.43 ± 1.17 ^d	34.55 ± 0.64 ^c	4.87 ± 0.15 ^d	1.44 ± 0.05 ^b	0.52 ± 0.03 ^e	6.14 ± 0.06 ^e	5.10 ± 0.10 ^b	1.04 ± 0.05 ^d
6	58.56 ± 0.87 ^b	55.91 ± 0.86 ^d	38.10 ± 0.85 ^b	4.44 ± 0.10 ^e	1.21 ± 0.09 ^a	0.36 ± 0.03 ^f	6.48 ± 0.07 ^d	4.25 ± 0.14 ^c	2.11 ± 0.16 ^e
7	42.47 ± 1.66 ^a	48.04 ± 1.58 ^a	26.26 ± 1.46 ^a	3.91 ± 0.09 ^a	1.24 ± 0.04 ^a	0.64 ± 0.04 ^a	8.30 ± 0.16 ^f	4.96 ± 0.16 ^b	3.34 ± 0.16 ^f

Values represent mean ± S.D of triplicate determinations. Values bearing dissimilar superscript letters down the column are significantly different at p < 0.05.

Table 2
Renal effects of administration of some medicinal plants and floras of lower phyla.

Grp	UREA (mg/dl)	CRT (mg/dl)	BUN (mg/dl)	Na (mmol/l)	K (mmol/l)	Zn (mmol/l)	Cl (mmol/l)	HCO ₃ (mmol/l)
1	39.49 ± 1.29 ^a	0.47 ± 0.02 ^a	21.94 ± 1.65 ^a	110.16 ± 3.49 ^a	8.89 ± 0.07 ^a	14.31 ± 0.37 ^a	95.27 ± 4.02 ^{ac}	24.72 ± 1.28 ^{ac}
2	37.19 ± 1.24 ^a	0.43 ± 0.02 ^{ac}	23.59 ± 1.84 ^a	130.91 ± 3.45 ^b	10.98 ± 0.11 ^a	14.20 ± 0.18 ^a	90.56 ± 2.85 ^b	19.19 ± 0.27 ^b
3	44.94 ± 2.89 ^c	0.52 ± 0.03 ^b	22.63 ± 2.49 ^a	114.19 ± 3.37 ^{ac}	12.30 ± 0.69 ^b	16.87 ± 0.23 ^b	94.81 ± 1.24 ^{ac}	23.00 ± 0.36 ^a
4	40.97 ± 2.08 ^d	0.41 ± 0.01 ^c	22.03 ± 1.97 ^a	118.77 ± 3.39 ^{cd}	8.42 ± 0.07 ^c	14.21 ± 0.25 ^a	97.86 ± 3.85 ^{cd}	18.70 ± 0.29 ^b
5	46.05 ± 1.17 ^c	0.43 ± 0.03 ^{ac}	23.40 ± 1.57 ^a	104.40 ± 4.49 ^a	8.78 ± 0.16 ^a	11.58 ± 0.32 ^c	91.14 ± 1.21 ^a	23.42 ± 1.60 ^a
6	46.02 ± 1.57 ^c	0.49 ± 0.02 ^a	22.40 ± 1.59 ^a	104.31 ± 2.77 ^a	11.75 ± 0.62 ^b	16.48 ± 0.36 ^b	84.39 ± 1.81 ^d	19.42 ± 0.97 ^b
7	53.27 ± 1.35 ^e	0.50 ± 0.02 ^a	23.87 ± 2.43 ^a	125.09 ± 6.05 ^{bd}	13.36 ± 0.57 ^d	14.24 ± 0.31 ^a	102.59 ± 5.18 ^e	25.15 ± 0.70 ^c

Values represent mean ± S.D of triplicate determinations. Values bearing dissimilar superscript letters down the column are significantly different at p < 0.05.

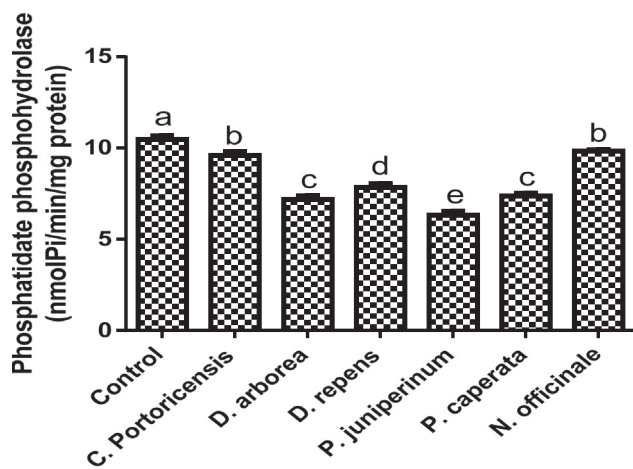


Fig. 1. Phosphatidate phosphohydrolase levels after administration of medicinal plants and floras of lower phyla.

result in Table 2 further implies that apart from *C. portoricensis*, administration of other extracts could have led to the impairment of the glomerular filtration leading to retained urea levels. Creatinine, a breakdown product of the muscles is not subject to any reabsorption, hence its elevations indicates dysfunctional glomerula filtration. Thus, the creatinine levels specifically confirms *D. repens* and *D. arborea* as toxic to the kidney. The high levels of serum Na, on administration of *C. portoricensis*, *D. repens*, and *N. officinale* could be testament to higher concentration of this element in the plants which might also be the case for potassium and zinc contents of *D. arborea*, and *P. caperata*. However, their Na/K ratios suggests overstressed kidneys of animals administered *D. repens*, and possibly weakened kidneys of animals on administration of *D. arborea*, *P. caperata*, and *N. officinale*; thus susceptibility to hypertension and hypotension respectively.

Figs. 1–9 summarizes the effect of *C. portoricensis*, *D. arborea*, *D. repens*, *P. juniperinum*, *P. caperata*, and *N. officinale* on lipid profile parameters and phosphatidate phosphohydrolase (PAP) levels of Wistar rats. As shown in Fig. 1, all the plant extracts significantly

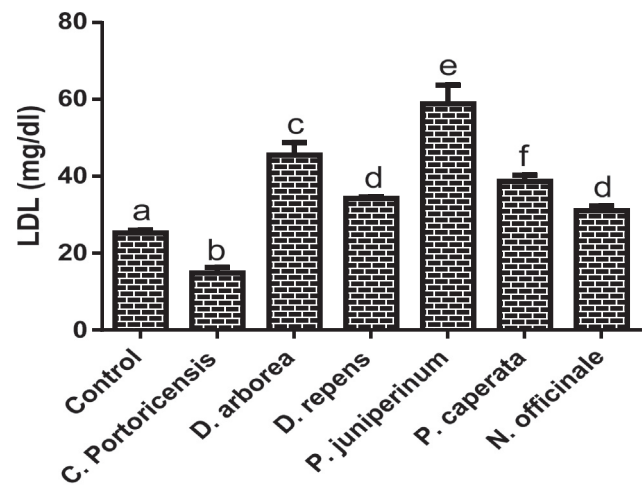


Fig. 2. LDL levels after administration of medicinal plants and floras of lower phyla.

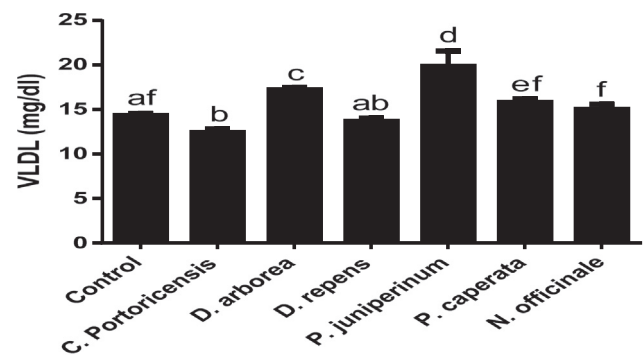


Fig. 3. VLDL levels after administration of medicinal plants and floras of lower phyla.

lowered the PAP levels when compared to the control, with *P. juniperinum* the most toxic, followed by *D. arborea* and *P. caperata*. Result for the LDL levels (Fig. 2) indicated significant elevations on

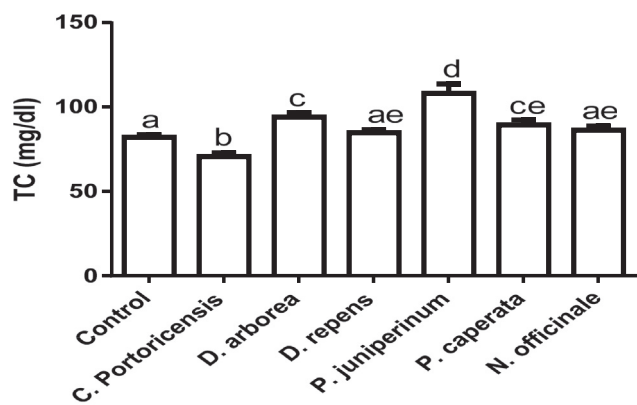


Fig. 4. TC levels after administration of medicinal plants and floras of lower phyla.

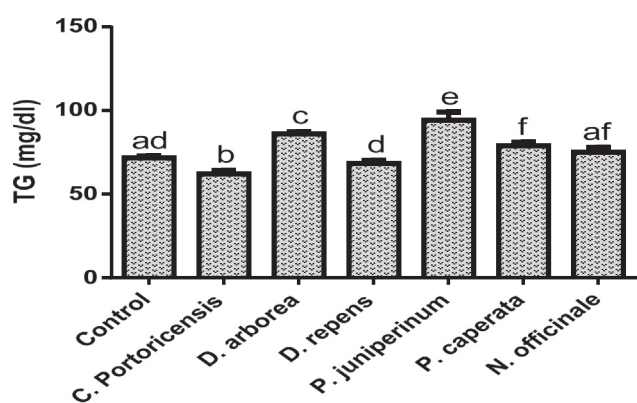


Fig. 5. TG levels after administration of medicinal plants and floras of lower phyla.

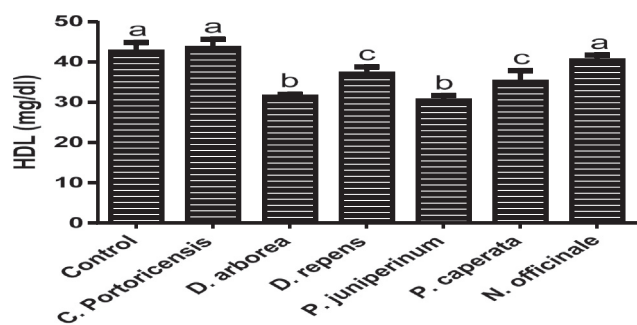


Fig. 6. HDL levels after administration of medicinal plants and floras of lower phyla.

administration of all the extracts except for *C. portoricensis* while animals administered *D. repens* and *N. officinale* produced VLDL, TC, and TG levels (Figs. 3–5 respectively) comparable to the control group. The result presented in Fig. 6 indicated that administration of *C. portoricensis* and *N. officinale* produced equivalent HDL levels to that of the control group. Figs. 7–9 clearly showed that among the extracts administered, *P. juniperinum* followed by *D. arborea*, produced the highest cardiac risk ratio, atherogenic coefficient, and atherogenic index of plasma, while *C. portoricensis* produced the least. Elevations in LDL, TG, and TC levels are regarded as indicators of cardiovascular diseases and according to Ikewuchi et al.,³⁸ are most commonly associated with obesity, hypertension, and insulin resistance. This suggests that consumption of *D. arborea* and *P. juniperinum* increases the susceptibility to cardiovascular diseases. Similarly, high TC and VLDL are also indicators of cardiovascular

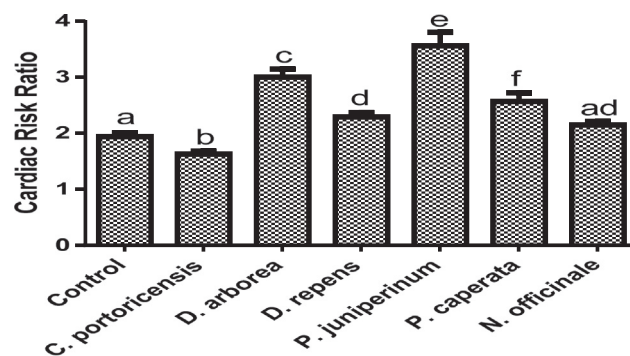


Fig. 7. Effects of administration of medicinal plants and floras of lower phyla on cardiac risk ratio.

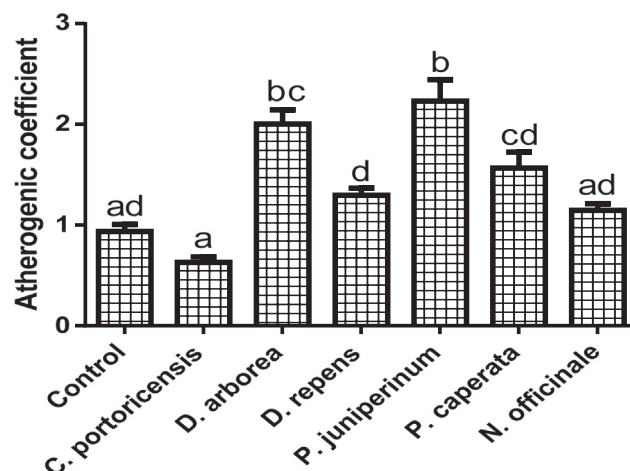


Fig. 8. Effects of administration of medicinal plants and floras of lower phyla on atherogenic coefficient.

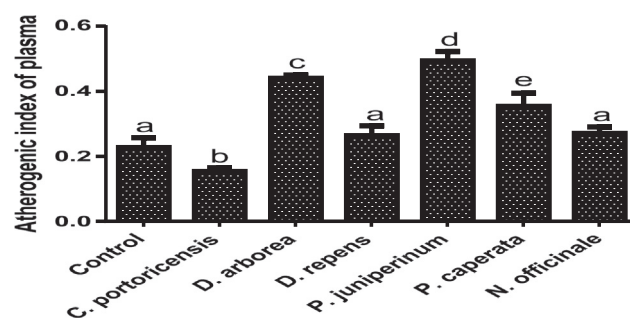


Fig. 9. Effects of administration of medicinal plants and floras of lower phyla on atherogenic index of plasma.

diseases and therefore provide further experimental evidence of the susceptibility on consumption of *D. arborea* and *P. juniperinum* which could have resulted from the significant derangement of the phosphatidate phosphohydrolase enzyme. HDL has been associated with numerous atheroprotective activities that include anti-inflammatory, antioxidant, and reverse cholesterol transport.⁵² Hence, *C. portoricensis* and *N. officinale* as indicated in Fig. 6, could possess significant atheroprotective properties, and may have occurred as a result of presence of myristicin and limonene.^{53–55} Cardiac risk ratio, also known as Castelli index is regarded as a very specific and sensitive parameter for the assessment of

Table 3
Effects of administration of some medicinal plants and floras of lower phyla on red blood cells indices.

Grp	RBC ($\times 10^6/\mu\text{L}$)	HGB (g/dl)	MCHC (g/dl)	MCV (fL)	MCH (pg)	HCT (%)
1	8.43 \pm 0.24 ^a	12.44 \pm 0.15 ^a	27.63 \pm 0.55 ^a	61.41 \pm 1.02 ^a	18.68 \pm 0.51 ^a	42.77 \pm 1.99 ^a
2	8.53 \pm 0.27 ^a	10.17 \pm 0.28 ^b	20.48 \pm 0.40 ^b	52.16 \pm 1.04 ^b	12.48 \pm 0.42 ^b	36.08 \pm 0.85 ^b
3	8.66 \pm 0.34 ^a	12.50 \pm 0.35 ^a	21.38 \pm 1.19 ^b	53.28 \pm 1.00 ^b	17.36 \pm 0.44 ^c	41.32 \pm 2.37 ^a
4	7.48 \pm 0.15 ^c	9.72 \pm 0.18 ^b	22.47 \pm 0.36 ^c	51.57 \pm 1.88 ^b	17.33 \pm 0.39 ^c	42.66 \pm 1.89 ^a
5	6.56 \pm 0.16 ^d	9.01 \pm 0.12 ^d	30.68 \pm 0.40 ^d	45.28 \pm 1.31 ^c	14.12 \pm 0.24 ^d	33.88 \pm 1.91 ^b
6	8.03 \pm 0.21 ^e	10.69 \pm 0.14 ^c	26.39 \pm 0.22 ^e	53.09 \pm 1.35 ^b	15.49 \pm 0.40 ^c	39.72 \pm 0.92 ^a
7	8.79 \pm 0.10 ^a	12.27 \pm 0.48 ^a	28.57 \pm 0.38 ^a	57.16 \pm 1.06 ^d	19.18 \pm 0.71 ^a	41.54 \pm 2.23 ^a

Values represent mean \pm S.D of triplicate determinations. Values bearing dissimilar superscript letters down the column are significantly different at $p < 0.05$.

Table 4
Effects of administration of some medicinal plants and floras of lower phyla on white blood cells differentials.

Grp	WBC ($\times 10^3/\mu\text{L}$)	Monocytes (%)	Neutrophils (%)	Eosinophils (%)	Basophils (%)	Lymph (%)
1	9.48 \pm 0.39 ^a	13.38 \pm 0.41 ^a	22.34 \pm 0.53 ^a	4.64 \pm 0.11 ^a	7.13 \pm 0.31 ^a	50.05 \pm 1.54 ^a
2	8.04 \pm 0.17 ^b	16.25 \pm 0.44 ^b	18.57 \pm 0.32 ^b	7.44 \pm 0.27 ^b	13.35 \pm 0.35 ^b	42.67 \pm 1.04 ^b
3	8.85 \pm 0.09 ^c	11.91 \pm 0.34 ^c	26.05 \pm 0.74 ^c	3.55 \pm 0.28 ^c	4.44 \pm 0.43 ^c	53.62 \pm 1.26 ^c
4	8.01 \pm 0.28 ^b	13.38 \pm 0.33 ^a	17.59 \pm 0.25 ^b	3.13 \pm 0.17 ^d	7.66 \pm 0.31 ^d	57.37 \pm 0.81 ^d
5	6.82 \pm 0.18 ^e	9.89 \pm 0.23 ^d	28.22 \pm 0.70 ^d	4.91 \pm 0.22 ^e	8.90 \pm 0.17 ^e	41.85 \pm 0.87 ^b
6	9.60 \pm 0.41 ^a	12.42 \pm 0.28 ^b	20.42 \pm 0.51 ^c	5.58 \pm 0.26 ^f	8.75 \pm 0.29 ^e	52.57 \pm 1.34 ^c
7	9.28 \pm 0.32 ^a	11.33 \pm 0.23 ^b	23.93 \pm 0.77 ^f	5.92 \pm 0.19 ^g	8.64 \pm 0.32 ^e	52.12 \pm 1.83 ^c

Values represent mean \pm S.D of triplicate determinations. Values bearing dissimilar superscript letters down the column are significantly different at $p < 0.05$.

Table 5
Effects of administration of some medicinal plants and floras of lower phyla on platelet functional indices.

Groups	PLT ($10^3/\mu\text{L}$)	MPV (fL)	PDW	PCT (%)
1	271.76 \pm 3.58 ^a	6.69 \pm 0.18 ^{ae}	12.63 \pm 0.35 ^a	0.66 \pm 0.03 ^a
2	265.64 \pm 4.55 ^a	5.30 \pm 0.25 ^b	13.49 \pm 0.52 ^b	0.61 \pm 0.04 ^a
3	258.92 \pm 3.34 ^b	6.89 \pm 0.22 ^{ad}	11.66 \pm 0.33 ^c	0.75 \pm 0.03 ^b
4	223.69 \pm 3.05 ^c	6.71 \pm 0.19 ^{ad}	11.61 \pm 0.28 ^c	0.67 \pm 0.02 ^a
5	207.38 \pm 2.51 ^d	6.97 \pm 0.17 ^c	8.64 \pm 0.25 ^d	0.44 \pm 0.03 ^c
6	247.32 \pm 2.25 ^e	5.88 \pm 0.14 ^d	12.50 \pm 0.41 ^a	0.54 \pm 0.03 ^d
7	281.52 \pm 4.55 ^f	6.34 \pm 0.15 ^e	12.67 \pm 0.25 ^a	0.70 \pm 0.04 ^{ab}

Values represent mean \pm S.D of triplicate determinations. Values bearing dissimilar superscript letters down the column are significantly different at $p < 0.05$.

vascular risk,⁵⁶ and the higher the ratio, the higher the risk, implying that in this study, administration of *P. juniperum* and *D. arborea* portends the greatest risks for cardiovascular diseases. The atherogenic coefficient is also a useful cardiovascular disease risk index that considers the ratio of atherogenic and atheroprotective cholesterol used in routine clinical practices. Further, according to Dobiášová and Frohlich,⁵⁷ the atherogenic index of plasma positively correlates the esterification rate of HDL with an inverse of size of LDL which is useful in depicting the complex interplay of lipoprotein metabolism and for the prediction of plasma atherogenicity. Ikewuchi et al.,³⁸ observed that the lower the atherogenic coefficient, the lower the cardiovascular risk, and vice versa, whereas, Dobiášová,⁵⁸ reported a cut-off point of 0.5 as an atherogenic risk indicator. This further supports the earlier claims in this study that administration of *C. portoricensis* and *N. officinale* poses the least atherogenic risk while consumption of *P. juniperum* and *D. arborea* presents much higher susceptibility to cardiovascular diseases.

The effects of administering *C. portoricensis*, *D. arborea*, *D. repens*, *P. juniperinum*, *P. caperata*, and *N. officinale* on RBC indices were presented in Table 3. No significant difference was recorded between the RBC levels of rats administered *C. portoricensis*, *D. arborea* and *N. officinale*, and that of the control group, while administration of *D. repens*, *P. juniperinum*, *P. caperata* significantly lowered the RBC. Similarly, administration of *D. arborea* and *N. officinale* produced a comparable HGB content when compared to the control. Further, the result showed that all the plant extracts significantly altered

the MCHC, MCV, and MCH concentrations, except for *N. officinale* in MCHC and MCV. Also, the HCT levels were altered on administration of *D. arborea* and *P. juniperinum*. Assessing the status of RBC indices is of paramount importance in evaluating the health effects of plant extracts and their products.⁵⁹ In this study, the significant decrease in RBC and HGB levels by *D. repens*, *P. juniperinum*, *P. caperata* strongly suggests hematotoxicity that could lead to anaemia,¹³ and from the results of the MCV, implies microcytic anaemia. Further, low levels of MCHC and MCH on administration of *C. portoricensis*, *D. arborea*, *D. repens*, *P. juniperinum*, *P. caperata* are indicative of either sequestration of iron or untimely breakdown of the red blood cells. Further, the HCT levels of the animals after consumption of the plant extracts shows that only the animals that consumed *C. portoricensis* and *P. juniperinum* could have blood cells with compromised oxygen carrying capacities.⁶⁰

Table 4 summarizes the effects of the administration of *C. portoricensis*, *D. arborea*, *D. repens*, *P. juniperinum*, *P. caperata*, and *N. officinale* on WBC differentials. The result showed significant reductions in the white blood cells on administration of all the plant extracts except for *P. caperata*, and *N. officinale*. The components of the WBC; neutrophils, eosinophils, basophils, and lymphocytes, were significantly altered, except for the administration of *D. repens* that produced comparable monocytes with the control. From the result of Table 4, the prolonged consumption of *C. portoricensis* may cause monocytosis possibly resulting from stress,⁶¹ whereas the immune-suppression observed from the monocyte levels on administration of other extracts are suggestive of possible

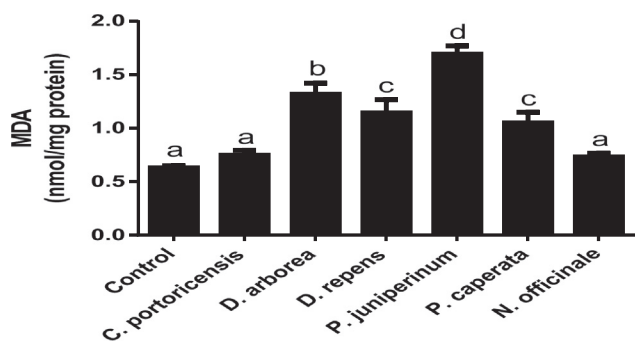


Fig. 10. MDA levels of rats administered selected medicinal plants and floras of lower phyla.

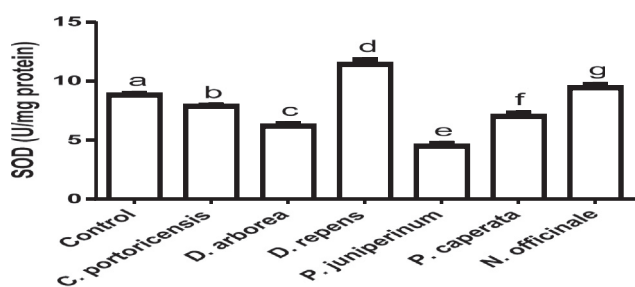


Fig. 11. SOD levels of rats administered selected medicinal plants and floras of lower phyla.

monocytopenia. Since inflammations are the primary causes of neutrophilia, it could be possible that the hematotoxicity associated with the administration of *D. repens* and *P. juniperum* may have also caused inflammations, whereas the decreased neutrophils as obtained from administration of other extract indicates possible aplastic anaemia.^{62,63} Furthermore the altered levels of eosinophils, lymphocytes and basophils on administration of these extracts further indicates their potentials to compromise ability of the system to elicit inflammatory responses, fight viral and opportunistic infections, and for the development of mammary glands.^{64,65}

The platelet functional indices after administration of *C. portoricensis*, *D. arborea*, *D. repens*, *P. juniperinum*, *P. caperata*, and *N. officinale* were shown in Table 5. The administration of *C. portoricensis* caused no significant change in the PLT levels, while the administration of other extracts significantly altered the PLT concentration. The result further showed no alterations in the MPV levels on administration of *D. repens*, and *D. arborea* which was the same case for PDW after the administration of *P. caperata*, and *N. officinale*. For the PCT, a significant increase was observed on administration of *D. arborea*, while *P. caperata*, and *N. officinale* significantly lowered the PCT levels. *N. officinale* possibly contains thrombopoietin releasing compounds, having significantly elevated PLT levels in this study.⁶⁶ High MPV is indicative of destruction of platelets or high and untimely release of platelets into circulation,⁶⁷ which might have been the result of administering *P. juniperinum*, whereas low levels of MPV as in the case of *C. portoricensis* and *P. caperata* suggests possibly the onset of aplastic anaemia and chronic vascular diseases.⁶⁸ Furthermore, from the result of Table 5, administration of especially *P. caperata* may impair blood clotting, having produced significantly lower PDW⁶⁹ while the PCT levels of the animals administered *P. juniperinum* and *P. caperata* further lends credence to the earlier claims of platelet reduction and possibly impaired blood clotting, in this study.

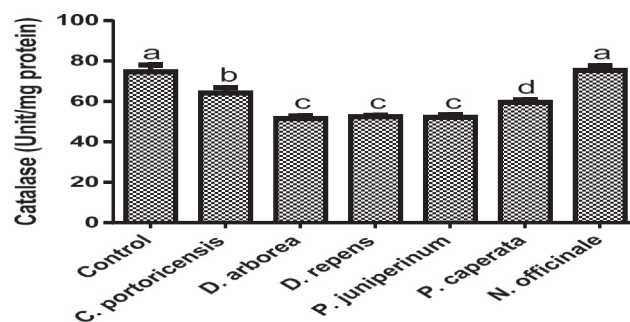


Fig. 12. Catalase levels of rats administered selected medicinal plants and floras of lower phyla.

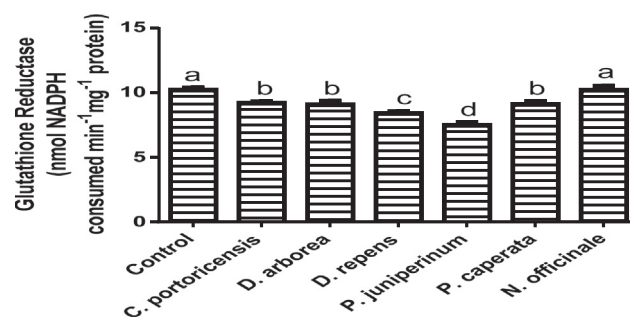


Fig. 13. Glutathione reductase levels of rats administered selected medicinal plants and floras of lower phyla.

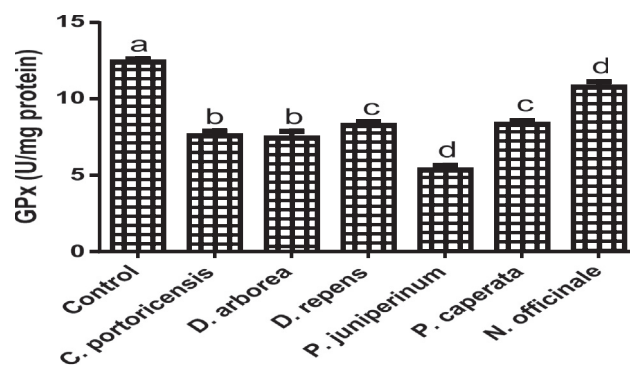


Fig. 14. Glutathione peroxidase levels of rats administered selected medicinal plants and floras of lower phyla.

Derangement in endogenous antioxidants and some markers of oxidative stress after administration of *C. portoricensis*, *D. arborea*, *D. repens*, *P. juniperinum*, *P. caperata*, and *N. officinale* were shown in Figs. 10–18. *C. portoricensis* and *N. officinale* caused no alterations in the MDA (Fig. 10) and glutathione (Fig. 16) levels whereas the SOD (Fig. 11), GPx (Fig. 14), and xanthine oxidase (Fig. 17) levels were significantly deranged by all the plant extracts. The result further indicated that only *N. officinale* produced catalase (Fig. 12), glutathione reductase (Fig. 13), and xanthine dehydrogenase (Fig. 18) levels that were equivalent to those of the control group. In addition, *C. portoricensis*, *D. arborea*, and *N. officinale*, caused no alterations on the GST levels (Fig. 15). Amadi et al.,⁷⁰ posited that elevated MDA levels are suggestive of depleted antioxidant enzyme systems, implying that *C. portoricensis*, *D. arborea*, *D. repens*, *P. juniperinum*, and *P. caperata* may have induced oxidative

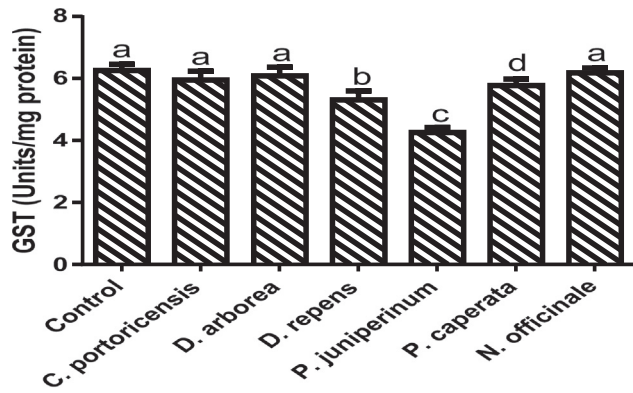


Fig. 15. Glutathione S transferase levels of rats administered selected medicinal plants and floras of lower phyla.

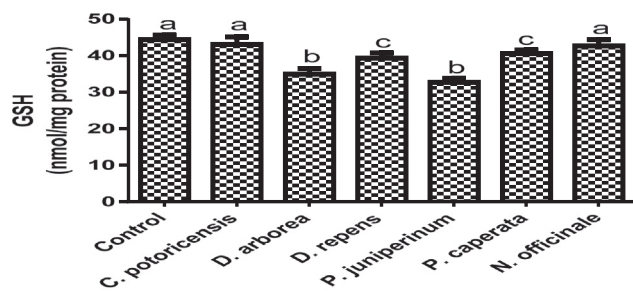


Fig. 16. Effects of administration of selected medicinal plants and floras of lower phyla on glutathione.

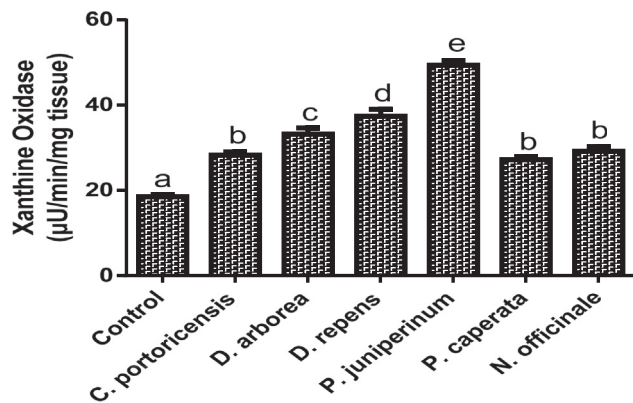


Fig. 17. Effects of administration of selected medicinal plants and floras of lower phyla on xanthine oxidase.

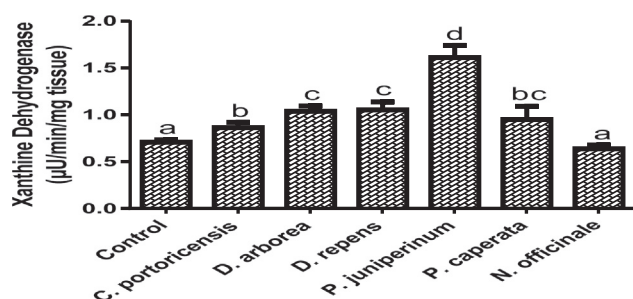


Fig. 18. Effects of administration of selected medicinal plants and floras of lower phyla on xanthine dehydrogenase.

stress in the experimental animals. Further, in agreement with the observations of Ogunka-Nnoka et al.,⁷¹ and Ogunka-Nnoka et al.,⁷² the reductions in levels of SOD, CAT, GSH, and GPx indicated the presence of toxic substances in the plant extracts, which may have suppressed their activities. Maintenance of intracellular levels of GST and GR is of paramount importance for removal of xenobiotics from the system. Glutathione reductase maintains the reduced state of glutathione, a reducing agent,⁷³ while glutathione-S-transferase catalyzes the conjugation of xenobiotics to glutathione which facilitates their detoxification.⁷⁴ The implication of this in this study is that the administration of especially *D. repens*, and *P. juniperinum* portends the possibility of compromising the efficiency of the system to detoxify xenobiotics. Both xanthine oxidase and dehydrogenase are interconvertible according to Ichida et al.,⁷⁵ and generates reactive oxygen species.⁷⁶ As shown in Figs. 17 and 18, the consumption of these plants extracts especially *D. repens* and *P. juniperinum* poses high risks of generating reactive oxygen species by elevating both xanthine oxidase and dehydrogenase.

4. Conclusion

This study has shown various degrees of toxicities associated with the consumption of these plants on the liver, kidney, blood cells, lipid profile and oxidative stress indicators. Though in some parameters evaluated, some of the extracts produced comparable results to the control animals, all extracts significantly altered at least one or more of each organ function indices and evaluated biochemical parameters. Hence, this raises serious concerns over the continued utilization of the extracts for treatment of various diseases sparsely reported in literature.

Conflict of interest

None declared.

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