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PROTECTIVE POTENTIAL OF AQUEOUS LEAF EXTRACT OF *BRIDELIA FERRUGINEA* IN NORMAL ALBINO RATS AGAINST CARDIOTOXICITY AND ANTI-OXIDANT IMBALANCE DUE TO EXPOSURE TO PETROL FUMES

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ABSTRACT
The present study evaluated the protective potential of aqueous leaf extract of *Bridelia ferruginea* in normal albino rats against cardio-toxicity and anti-oxidant imbalance caused by exposure to petrol fumes. A total of 21 albino rats of Wistar strain and of average weight were used in this study. After exposure of two test groups to petrol fumes for 1 hour and 3 hours daily, they were treated with the aqueous leaf extract of *Bridelia ferruginea* (400mg/kg) and vitamin E (200mg/kg) respectively for 28 days. Blood samples were collected periodically from the submandibular vein for spectrophotometric determination of lactate dehydrogenase and creatine kinase activities. On the 28th day of the experiment, all the rats were sacrificed and their hearts obtained for histological studies and spectrophotometric assay of Catalase, superoxide dismutase and lipid peroxidation. Results obtained indicated that exposure to petrol fumes caused the activities of lactate dehydrogenase and creatine kinase to increase when compared to normal control($P<0.05$). Catalase and superoxide dismutase activities were considerably reduced while malondialdehyde level increased ($P<0.05$). Treatment with aqueous leaf extract of *Bridelia ferruginea* and vitamin E, however, reversed this imbalance ($P<0.05$). Histological examination showed adverse changes in the hearts of untreated rats. However, these changes were reversed by the administration of aqueous leaf extract of *Bridelia ferruginea*. The plant may therefore provide protection against cardio-toxicity caused by petrol fumes.

KEYWORDS: Petrol, toxicity, *Bridelia ferruginea*, lactate dehydrogenase, creatine kinase, antioxidants.

INTRODUCTION
Petrol or gasoline is a volatile liquid that consists of a complex mixture of aliphatic and aromatic hydrocarbons. It is commonly used as fuel for internal combustion engines. Some of its constituents are known to be highly toxic or carcinogenic to humans. Many of the toxicological effects associated with the exposure to petrol can be attributed to specific components of petrol, such as benzene, toluene, ethylbenzene and xylene (BTEX), which are also known as volatile organic compounds (VOCs). BTEX have been reported to be the most dangerous aromatic hydrocarbon components of petrol (Adami *et al.*,2006).

A number of studies have demonstrated that the occurrences of various health problems are closely associated with occupational exposures to VOCs. Uzma *et al*. (2008) found that exposure to benzene from petrol vapour caused haematotoxicity among petrol station workers. This study also found significantly diminished pulmonary function that was associated with duration of exposure to gasoline vapour. Furthermore, other studies have also suggested that there is a causal relationship between industrial exposures to benzene and the incidence of some types of leukaemia and aplastic anaemia (Uzma *et al.*, 2008).

Medicinal plants play an important role in health care in Africa and the world over (Prohp *et al*., 2008). *Bridelia ferruginea* (Euphorbiaceae) is a woody shrub that grows in the Savannah or rain forests of Africa (Njamen *et al.*, 2012) and is traditionally used to treat diabetes (Onunkwo *et al.*, 1996), arthritis (Olajide *et al.*, 2007) and boils (Talla *et al*., 2002).

The aqueous extract of the plant contains quinones, gallic and catechic tannins, alkaloids, sterols, polyterpenes, polyphenols, reducing compounds, flavonoids and saponosides (Batomayena *et al*., 2019).

Its common names in Nigeria are Kirni, Kimi (Hausa), Maren (Fulani), Iralodan (Yoruba), and Oha, Ede, Ola in Igbo. Studies have revealed that *Bridelia ferruginea* possesses medicinal properties such as ulcer-protection and anti-diarrheal by the stem bark (Akuodor *et al.*, 2012), type 2 diabetes (Bakoma *et al.*, 2013), purgative and vermifuge activities (Cimanga *et al.*, 1999), traditional gargle " Ogun efu" (Orafidiya *et al.*, 1990), antimicrobial activities against some micro-organisms known to cause enteric and secondary upper respiratory tract infections (Magistretti *et al.*, 1988) and anti-inflammatory activity (Olajide *et al.*, 1999).

Lactate dehydrogenase (LDH) is an enzyme found in nearly all living cells. LDH catalyzes the conversion of lactate to pyruvate and back, as it converts NAD+ to NADH and back. A dehydrogenase is an enzyme that transfers a hydride from one molecule to another. LDH is expressed extensively in body tissues, such as blood cells and heart muscle. Because it is released during tissue damage, it is a marker of common injuries and disease such as heart failure. This enzyme exits in different forms (Holmes, 2009).

Creatine kinase (CK), also known as creatine phosphokinase (CPK) or phosphocreatine kinase, is an enzyme expressed by various tissues and cell types. CK catalyzes the conversion of creatine and uses adenosine triphosphate (ATP) to create phosphocreatine (PCr) and adenosine diphosphate (ADP). This CK enzyme reaction is reversible and thus ATP can be generated from PCr and ADP. (Bong *et al*., 2008).

Antioxidants are molecules that fight free radicals in your body. Free radicals are compounds that can cause harm if their levels become too high in your body. They're linked to multiple illnesses, including diabetes, heart disease, and cancer. Your body has its own antioxidant defenses to keep free radicals in check. However, antioxidants are also found in food, especially in fruits, vegetables, and other plant-based, whole foods. Several vitamins, such as vitamins E and C, are effective antioxidants. Antioxidant preservatives also play a crucial role in food production by increasing shelf life. Antioxidant are enzymatic and non- enzymatic. Antioxidants can increase the shelf life of both natural and processed foods. Therefore, they're frequently used as food additives. For instance, vitamin C is often added to processed foods to act as a preservative. (Peter, 2018).

MATERIALS AND METHOD

Experimental Animals

21 male Wistar rats weighing between 150g- 200g were obtained from the animal house of the Department of Pharmacology, Faculty of Basic Clinical Sciences, Niger Delta University, Wilberforce Island, Bayelsa State, Nigeria. They were housed in standard plastic cages in the research laboratory of the department for two weeks to acclimatize. They were maintained under a 12-hour light/ dark cycle, controlled temperature (20-25^{0}C) and were fed standard chow and water ad libitum. All experimental procedures were carried out in accordance with the guidelines of the Research Ethical Committee of the Niger Delta University, Wilberforce Island, Bayelsa State, Nigeria.

Plant materials and preparation of extracts

Fresh leaves of *Bridelia ferruginea* plant leaves were collected from the compound of College of Health Sciences, Niger Delta University, Wiberforce Island, Bayelsa State, Nigeria and was identified by Prof Kola Ajibesin of the Department of Pharmacognosy, Faculty of Pharmacy, Niger Delta University, Wilberforce Island, Bayelsa State, Nigeria and was confirmed with the aid of google lens. The leaves were washed with cold water and dried under shade for two weeks. The dried leaves were then pulverized into fine powder using an electric blender. 500g of the pulverized leaves was soaked in 2 liter of distilled water in a glass jar and was allowed to stand for 72 hours with intermittent stirring. After 72 hours, it was filtered with 110 mm whatman filter paper and the filtrate was evaporated in constant temperature water bath at 60 ^{0}c. The extract was stored in sample bottles and refrigerated until time of use.

Petrol/Chemical Reagents

The petrol used for this study was procured from NNPC filling station Edepie, Yenagoa, Bayelsa State, Nigeria. Enzyme kits used to assay the activities of Lactate dehydrogenase and creatine kinase were procured from Randox^{TM} Laboratories Ltd, Crumlin, Co, Antrim, United Kingdom. All other chemicals used for antioxidant assay were purchased from Loba Chemie PVT LTD India.

Experimental Procedure

Experimental Design: After two weeks of acclimatization, a baseline values for plasma LDH and CK were assayed for each rat. The animals were then fasted overnight and grouped randomly.

Twenty-one (21) male albino rats of Wistar strain were divided into 7 groups of 3 rats each and treated as follows for 28 days:

Group 1 (Normal Control) was fed with normal chow and distilled water (*ad libitum)* throughout the experiment.

Group 2 (Positive control 1) was exposed to petrol fumes for one hour daily and was fed with normal chow and distilled water (*ad libitum*) throughout the experiment.

Group 3 (Positive control 2) was exposed to petrol fumes for three hours daily and was fed with normal chow and distilled water (*ad libitum*) throughout the experiment.

Group 4 (Test group 1) was exposed to petrol fumes for one hour daily and received aqueous stem bark extract of *Bridelia ferruginea* (400mg/kg body weight) twice daily for 28 days. They were also fed with normal chow and distilled water (*ad libitum)* throughout the experiment.

Group 5 (Test group 2) was exposed to petrol fumes for three hours daily and received aqueous stem bark extract

of *Bridelia ferruginea* (400mg/kg body weight) twice daily for 28 days. They were also fed with also normal chow and distilled water (*ad libitum)* throughout the experiment

Group 6 (Standard control 1) was exposed to petrol fumes for one hour daily and received vitamin E (200mg/kg), twice daily for 28 days. They were also fed with normal chow and distilled water (*ad libitum*) throughout the experiment.

Group 7 (Standard control 2) was exposed to petrol fumes for three hours daily and received vitamin E (200mg/kg), twice daily for 28 days. They were also fed with normal chow and distilled water (*ad libitum*) throughout the experiment.

The dose of the extract used was derived from a study previously conducted to determine the lethal dose (LD50) of the aqueous extract.

Exposure to petrol fumes

The animals were exposed to petrol fumes according to the method that was described by Uboh *et al.* (2005) and Owagboriaye *et al.* (2016) with slight modifications. A 1000ml plastic beaker containing 500 ml of petrol was placed in a fume chamber measuring 150 cm × 90 cm × 210 cm 1 hour before the commencement of exposure to ensure that the chamber is saturated with vapor. The animals were transferred into plastic baskets and were placed in the fume chamber to inhale the fumes evaporating from the can for 1 hour and 3 hours daily according to their groupings. Afterwards, the animals were transferred to a fume-free section of the laboratory. The exposure lasted periodic for 28 days.

Collection of samples

Blood samples were collected from the sub-mandibular vein of the rats after a mild chloroform anaesthsia into lithium heparin bottles for baseline and afterwards on days 0,1,7,14,21, and 28. The blood sample were centrifuged at 2000 rotation per minute for 10 minutes. The supernatant (Plasma) was collected for creatine kinase (CK) and lactate dehydrogenase (LDH) enzyme activities assay.

All the animals were sacrificed on the 28th day after anesthesia using chloroform and their hearts were harvested for antioxidants and histological studies.

Enzyme Assay

Creatine kinase (CK) and lactate dehydrogenase (LDH) enzyme activities were estimated following the instructions in the inserts of biochemical kits purchased from Randox Laboratories (Crumlin, Co, Antrim, United Kingdom).

Antioxidant Assay

The heart tissues of the rats were homogenized in 0.1 M Phosphate buffer (pH 7.4) to make 10% homogenate and the homogenate was centrifuged at 3000 x g revolution per minutes (rpm) for 15 minutes at 4^{o}C. The supernatant was collected and used for antioxidant assay. Superoxide dismutase (SOD) was assayed following the method described by Marklund and Marklund (1974), Catalase (CAT) activity was estimated according to the method of Aebi *et al*., (1974). Lipid peroxidation was estimated as malondialdehyde (MDA) according to the method described by Armstrong and Al-Awadi (1991) with slight modifications.

Histopathological Examination

Portions of the hearts were fixed in 10% formal-saline, dehydrated in graded alcohol, cleared by xylene, and embedded in paraffin wax. The tissues were then cut into 3- to 4-mm-thick sections by a microtome, fixed on the slides, and stained with Hematoxylin & Eosin. The slides were examined under a light microscope (Olympus CH; Olympus, Tokyo, Japan), and photomicrographs were taken at x400 magnifications.

Statistical Analysis

Results from this study were expressed as mean ± SD. The statistical comparisons among the groups were performed using one-way analysis of variance (ANOVA) (SPSS 10.0). Significant difference was analyzed at $P<0.05$ level.

RESULTS

The results of the present study are presented in the tables below.

Table 1: Mean plasma activities of Lactate dehydrogenase (U/L) in rats exposed to petrol fumes (1hour).

TEST DAY	NORMAL CONTROL	POSITIVE CONTROL 1	TEST GROUP1	STANDARD CONTROL1
BASELINE	155.67±2.4	152.64±2.09	153.87±1.89	151.23±1.2
DAY0	148.55±3.12	147.43±2.28	149.9±2.41	143.99±0.95
DAY1	154.1±2.21	154.01±1.14	153.1±2.55	152.75±1.80
DAY7	154.23±1.97	158.8±2.02	154.61±1.70	157.44±0.44
DAY14	155.75±3.01	179.31±0.75	168.44±0.63	165.5±1.32
DAY21	156.67±2.55	196.61±2.34	173.68±1.47	168.45±1.58
DAY28	155.65±1.52	204.88±2.93	178.9±1.81	177.41±4.33

Data are expressed as the mean ± SD. Means within the same row (in each parameter) carrying different superscripts (a, b, c, d) are significantly different ($p < 0.05$).

Results (Table 1) reveal that one-hour exposure to petrol fumes caused a significant ($p<0.05$) increase in the activity of Lactate Dehydrogenase (Positive Control 1) relative to the Normal control rats. Administration of aqueous leaf extract of *Bridelia ferruginea* (Test group 1) and Vitamin E (Standard group 1) significantly ($p<0.05$) reduced the elevated activities of Lactate Dehydrogenase compared to the Petrol fumes intoxicated group (Positive Control 1).

Table 2: Mean plasma activities of Lactate dehydrogenase (U/L) in rats exposed to petrol fumes (3 hours).

	NORMAL CONTROL	POSITIVE CONTROL 2	TEST GROUP 2	STANDARD CONTROL 2
BASELINE	155.67±2.4	163.21±10.1	158.32±10.24	153.81±10.50
DAY 0	148.55±3.12	160.3±20	154.9±10.07	153.6±10.9
DAY 1	154.1±2.21	165.3±19.9	161.1±20	154.6±20
DAY 7	154.23±1.97	172.99±10	165.61±19.8	157±20
DAY 14	155.75±3.01	201.3±10.1	172.88±20.11	160.5±19.8
DAY 21	156.67±2.55	228.7±11.2	183.98±9.01	165.34±19.91
DAY 28	155.65±1.52	299.52±0.21	194.6±2.1	170.66±20.12

Data are expressed as the mean ± SD. Means within the same row (in each parameter) carrying different superscripts (a, b, c, d) are significantly different ($p < 0.05$).

Results (Table 2) reveal that three-hours exposure to petrol fumes caused a significant ($p<0.05$) increase in the activity of Lactate Dehydrogenase (Positive Control 2) relative to the Normal control rats. Administration of aqueous leaf extract of *Bridelia ferruginea* (Test group 2) and Vitamin E (Standard group 2) significantly ($p<0.05$) reduced the elevated activities of Lactate Dehydrogenase compared to the Petrol fumes intoxicated group (Positive Control 2).

Table 3: Mean plasma activities of Creatine Kinase (U/L) in rats exposed to petrol fumes (1 hours).

	NORMAL CONTROL	POSITIVE CONTROL 1	TEST GROUP 1	STANDARD CONTROL 1
BASELINE	$14.14±1.98^{a}$	$14.97±1.0^{a}$	$14.47±1.04^{a}$	$14.59±0.09^{a}$
DAY 0	$14.11±2.92^{a}$	$13.67±0.55^{a}$	$13.01±1.0^{a}$	$13.92±0.92^{a}$
DAY 1	$15.99±5.43^{a}$	$15.19±0.66^{a}$	$14.54±1.16^{a}$	$14.57±0.58^{a}$
DAY 7	$15.13±1.98^{a}$	$32.88±2.50^{b}$	$27.66±1.60^{c}$	$25.98±0.33^{c}$
DAY 14	$15.98±1.01^{a}$	$48.66±1.3^{b}$	$39.87±1.32^{c}$	$41.67±0.92^{c}$
DAY 21	$15.08±3.61^{a}$	$58.45±1.78^{b}$	$47.87±1.74^{c}$	$45.82±1.18^{c}$
DAY 28	$15.10±2.01^{a}$	$66.32±1.71^{b}$	$59.86±1.01^{c}$	$54.99±1.05^{d}$

Data are expressed as the mean ± SD. Means within the same row (in each parameter) carrying different superscripts (a, b, c, d) are significantly different ($p < 0.05$).

Results (Table 3) reveals that one-hour exposure to petrol fumes caused a significant ($p<0.05$) increase in the activity of creatine kinase (Positive Control 1) relative to the Normal control. Administration of aqueous leaf extract of *Bridelia ferruginea* (Test group 1) and Vitamin E (Standard group 1) significantly ($p<0.05$) reduced the elevated activities of creatine kinase compared to the Petrol fumes intoxicated group (Positive control 1).

Table 4: Mean plasma activities of Creatine Kinase (U/L) in rats exposed to petrol fumes (3 hours).

	NORMAL CONTROL	POSITIVE CONTROL 2	TEST GROUP 2	STANDARD CONTROL 2
BASELINE	$14.14±1.98^{a}$	$14.19±2^{a}$	$15.19±8.23^{a}$	$15.22±2.91^{a}$
DAY 0	$14.11±2.92^{a}$	$14.88±1.09^{a}$	$14.1±7.32^{a}$	$15.01±6.20^{a}$
DAY 1	$15.99±5.43^{a}$	$16.02±6.2^{a}$	$15.68±1.91^{a}$	$16.27±2.08^{a}$
DAY 7	$15.13±1.98^{a}$	$37.44±3.23^{b}$	$29.95±4.82^{c}$	$31.55±0.18^{a}$
DAY 14	$15.98±1.01^{a}$	$45.87.±1.89^{b}$	$33.6±8.13^{c}$	$31.56±9.78^{c}$
DAY 21	$15.08±1.01^{a}$	$58.41±8.82^{b}$	$47.22±8.09^{c}$	$46.16±9.98^{c}$
DAY 28	$15.10±5.44^{a}$	$71.2±10^{b}$	$58.6±9.82^{c}$	$60.95±6.10^{c}$

Data are expressed as the mean ± SD. Means within the same row (in each parameter) carrying different superscripts (a, b, c, d) are significantly different ($p < 0.05$).

Results (Table 4) reveals that three-hours exposure to petrol fumes caused a significant ($p<0.05$) increase in the activity of creatine kinase (Positive Control 2) relative to the Normal control. Administration of aqueous leaf

extract of *Bridelia ferruginea* (Test group 2) and Vitamin E (Standard group 2) significantly ($p<0.05$) reduced the elevated activities of creatine kinase compared to the Petrol fumes intoxicated group (Positive control 2)

Table 5: Mean antioxidant activities on Petrol fumes induced cardiotoxicity in wistar rats (1 hours).

	NORMAL CONTROL 1	POSITIVE CONTROL 1	TEST GROUP 1	STANDARD CONTROL 1
CATALASE	3.86 ± 0.01^{a}	1.16 ± 0.14^{b}	2.81 ± 0.13^{c}	2.29 ± 0.09^{c}
SOD	4.87 ± 0.08^{a}	3.92 ± 0.4^{b}	4.77 ± 0.38^{a}	5.07 ± 0.47^{a}
MDA	10.8 ± 0.1^{a}	41 ± 6.73^{b}	29.6 ± 0.79^{c}	25 ± 0.75^{d}

Data are expressed as the mean ± SD. Means within the same row (in each parameter) carrying different superscripts (a, b, c, d,) are significantly different ($p < 0.05$).

Results (Table 5) reveals that one-hour daily exposure to petrol fumes caused a significant ($p<0.05$) decreae in the activities of SOD and catalase (Positive Control 1) relative to the Normal control. Administration of aqueous leaf extract of *Bridelia ferruginea* (Test group 1) and Vitamin E (Standard group 1) significantly ($p<0.05$) reduced the elevated activities of SOD and catalase compared to the Petrol fumes intoxicated group (Positive control 1). Exposure to petrol fumes also increased the level of tissue MDA compared to normal group and this was reduced by the administration of the 400 mg/kg extract (Test group 1) and 200 mg/kg vitamin E (Standard group1) compared to the Petrol fume intoxicated group (Positive control group 1).

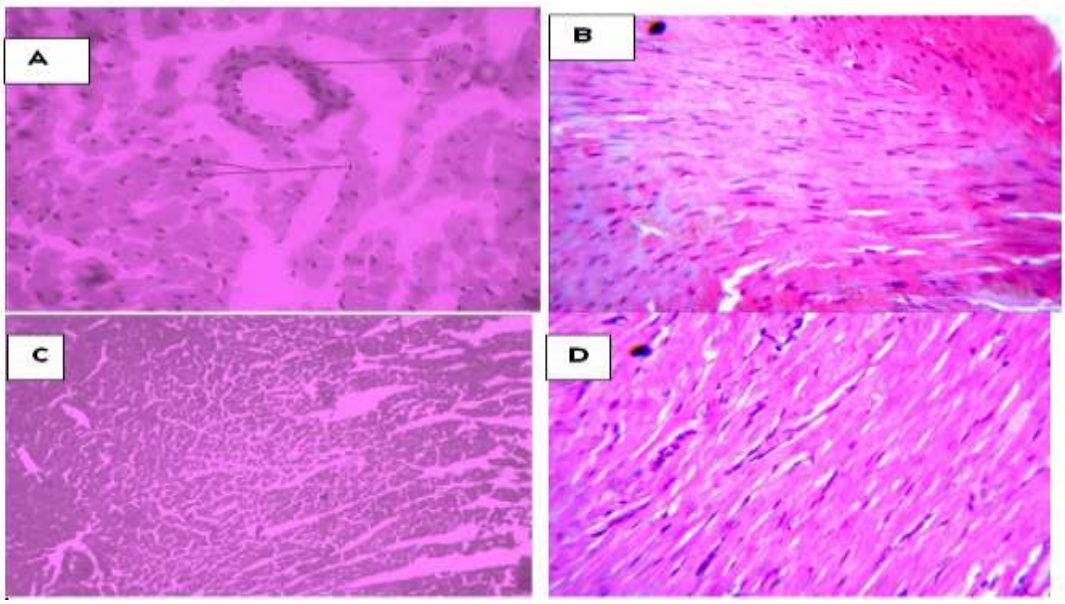
Table 6: Mean antioxidant activities on Petrol fumes induced cardiotoxicity in wistar rats (3 hours).

	NORMAL CONTROL 2	POSITIVE CONTROL 2	TEST GROUP 2	STANDARD CONTROL 2
CATALASE	3.66 ± 0.01^{a}	0.82 ± 0.09^{b}	1.71 ± 0.14^{c}	1.82 ± 0.99^{c}
SOD	4.87 ± 0.08^{a}	1.22 ± 0.99^{b}	3.19 ± 0.08^{c}	3.27 ± 0.02^{c}
MDA	10.8 ± 0.1^{a}	58 ± 1.46^{b}	30.6 ± 0.25^{c}	29.03 ± 8.27^{c}

Data are expressed as the mean ± SD. Means within the same row (in each parameter) carrying different superscripts (a, b, c, d,) are significantly different ($p < 0.05$).

Results (Table 5) reveals that three-hours exposure to petrol fumes caused a significant ($p<0.05$) decreae in the activities of SOD and catalase (Positive Control 2) relative to the Normal control. Administration of aqueous leaf extract of *Bridelia ferruginea* (Test group 2) and Vitamin E (Standard group 2) significantly ($p<0.05$) reduced the elevated activities of SOD and catalase compared to the Petrol fumes intoxicated group (Positive control 2). Exposure to petrol fumes also increased the level of tissue MDA compared to normal group and this was reduced by the administration of the 400 mg/kg extract (Test group 2) and 200 mg/kg vitamin E (Standard group1) compared to the Petrol fume intoxicated group (Positive control group2).

Histology



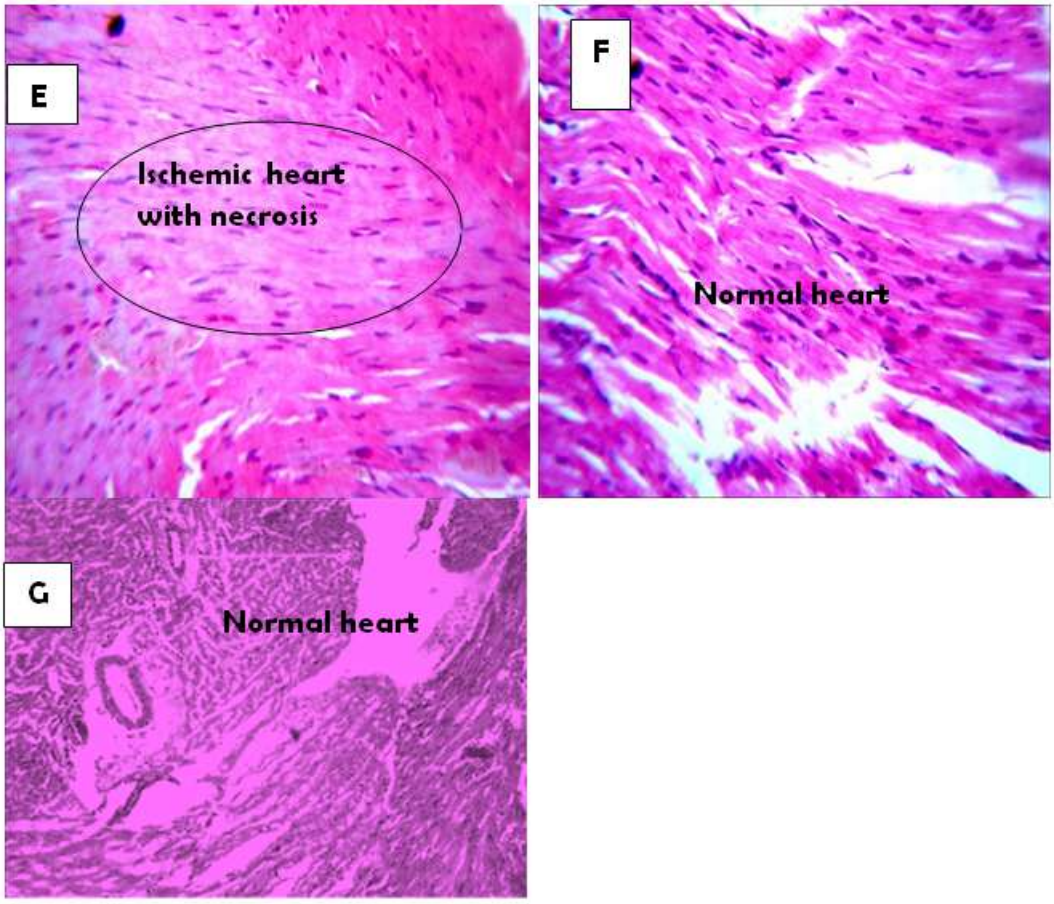


Figure 1: Representative photomicrograph of cardiac tissues exposed to petrol fumes.

Transverse section of the heart tissues stained with haematoxylin and eosin technique x 400 magnification.
NORMAL CONTROL, & SANDARD CONTROL 1 show smooth bundles of myocardiocytes with normocchromic oval shape nuclei and fenestrae, a, b and c respectively consistent with normal histology of the heart. POSITIVE CONTROL 1 shows areas of severe necrosis and artheromatous plaque consistent with abnormal heart tissue. Test 1 shows area of mild ischemic necrosis.
(A) Normal control: normal muscle cells and Intercalated disc consistent with normal histology of the heart.
(B) Positive control 1: Petrol fumes intoxicated animal (1 hours) shows severe areas of ischemic necrosis with anisionucleosis consistent with abnormal heart tissue.
(C) Test Group 1: Petrol fumes intoxicated animal (1 hours) with 400mg/kg aqueous leaf extract of *Bridelia ferruginea treatment* shows a well differentiated muscle fibre, which reflects the muscle cellsIntercalated disc.
(D) Standard Group 1: Petrol fumes intoxicated animal (1 hours) with 200mg/kg Vitamin E treatment shows smooth bundles of myocardiocytes with normochromic oval shape nuclei and fenestrae consistent with normal histology of the heart.
(E) Positive control 2: Petrol fumes intoxicated animal (3 hours) shows areas of severe ischemic necrosis with anisionucleosis consistent with abnormal heart tissue.
(F) Test Group 2: Petrol fumes intoxicated animal (3 hours) with 400mg/kg aqueous leaf extract of *Emilia Bridelia ferruginea* shows smooth bundles of myocardiocytes with normochromic oval shape nuclei and fenestrae consistent with normal histology of the heart.
(G) Standard Group 2: Petrol fumes intoxicated animal (3 hours) with 200mg/kg Vitamin E treatment shows smooth bundles of myocardiocytes with normochromic oval shape nuclei and fenestrae consistent with normal histology of the heart.

DISCUSSION

Petrol is a volatile liquid with a complex mixture of aliphatic and aromatic hydrocarbons. Commonly used as fuel for internal combustion engines, thinner, decorative agent, and industrial solvent. Some of its constituents are known to be highly toxic or carcinogenic to humans. Many of the toxicological effects associated with the exposure to gasoline can be attributed to specific components of gasoline, such as benzene, toluene, ethylene and xylene, which are also known as volatile organic compounds (VOCs). A number of studies have demonstrated that the occurrences of various health problems are closely associated with occupational exposures to VOCs (Uzma *et al*.,2008).

Plants have been used for millennia to cure a wide range of illnesses. Various plant parts such as the leaf, stem,

bark, and root are used. *Bridelia ferruginea* (Euphorbiaceae) is traditionally used to treat diabetes (Onunkwo *et al.,* 1996), arthritis (Olajide *et al.,* 2007) and boils (Talla *et al.,* 2002). The aqueous extract of the leaf contains antioxidant phytochemicals, quinones, gallic and catechic tannins, alkaloids, sterols, polyterpenes, polyphenols, reducing compounds, flavonoids and saponins.

Lactate Dehydrogenase (LDH) and Creatine Kinase are vital enzymes that are biomarkers of the heart due to tissue damage. Result from Present study shows that exposure to petrol fumes caused a significant increase ($p<0.05$) in the plasma activities of LDH and Creatine Kinase in the positive control group compared to the normal control group and this could be as a result of oxidative stress which may have led to leakage of the enzyme from the tissues into the blood as a result of necrosis of the tissues (Aulbach *et al*., 2017). However, treatment with aqueous leaf extract of *Bridellia ferruginea* (400mg/kg) and vitamin E (200mg/kg) significantly decreased the effect caused by petrol on the heart plasma activities of LDH and Creatine Kinase.

Oxidative stress is considered as an imbalance between pro- and antioxidant species, which results in molecular and cellular damage (Conti *et al*., 2016). Antioxidants are molecules that fight free radicals in the body thereby alleviating oxidative stress. Superoxide Dismutase (SOD) and catalase are vital enzymes of the antioxidant defense system. Results from this study shows that exposure to petrol fumes caused a significant decrease in the plasma activities of Superoxide Dismutase (SOD) and Catalase in the positive control group compared to the normal control group and this could be as a result of oxidative stress. Exposure to petrol fumes also significantly increased the level of Malondialdehyde, indicating a high level of lipid peroxidation in the heart. However treatment with aqueous leaf extract of *Bridelia ferruginea* (400mg/kg) and vitamin E (200mg/kg) significant ($p<0.05$) reversed the oxidative effects of petrol fumes. Plasma levels of SOD and Catalase were increased while MDA level was decreased compared to the positive control group. This result is in agreement with the work of (Olubukola *et al*., 2021).

Histological examination of myocardial tissues obtained from positive control group show area of severe ischemic necrosis and artheromatous plaque consistent with abnormal heart tissue compared to the normal control group which showed clear membrane integrity and smooth bundles of myocardiocytes with normocchromic oval shape nuclei and fenestrae consistent with normal histology of the heart. Treatment with aqueous extract of *Bridelia ferruginea* shows less ischemia and no artheromatous plaque compared to the positive control. This further confirms the membrane stabilizing effect of the extract. Treatment with vitamin E showed clear membrane integrity and smooth bundles of myocardiocytes with normocchromic oval shape nuclei and fenestrae consistent with normal histology of the heart. This indicates that vitamin E provides a complete protection from the cardiotoxic effect of exposure of the animals to petrol fumes.

CONCLUSION

In conclusion, the aqueous leaf extract of *Bridelia Ferruginea* exhibited the potential to prevent cardio-toxicity caused by petrol fumes. As clearly portrayed in the results obtained, it significantly reversed the distortion in the activities of LDH, CK,SOD, Catalase and lipid peroxidation due to exposure petrol fumes.

Conflict of Interest

There is no conflict of interest.

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