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Effect of Ethanol Extract of Desmondium Adscendens Leaves on Some Oxidative Stress Indices in Phenylhydrazine Administered Wistar Rats.

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Abstract- Background: ***Medicinal plants, when regulated and used correctly can help to prevent and treat a variety of health conditions. We investigated the ameliorative potential of Desmondium adscendens leave (EDAL) extract on oxidative stress indices in Phenylhydrazine administered rats.***

Method: ***Twenty-five (25) male Wistar rats weighing 180-200g, were randomly assigned to 5 groups of five rats each. Group1(normal control), Group 2(PHZ) was challenged with phenylhydrazine (60mg/kg, B.W., intraperitoneal) without treatment. Group 3 received EDAL orally at 150mg/kg, B.W., Group 4 and 5 were challenged with phenylhydrazine (60mg/kg, B.W,) and treated orally with 150mg/kg and 300mg/kg of extract of Desmondium adscendens leaves (EDAL) respectively. All animals were allowed free access to food and water pre and post treatment for 14 days. At the end of the treatment period, the animals were euthanized, and blood samples collected via cardiac puncture for biochemical analysis.***

Result: ***Oxidative stress indices (SOD, CAT and GSH) revealed significant($P<0.05$) decrease, except MDA which increased in PHZ group compared to normal group and extract group. However, there was improvement in the above biochemical parameters in extract treated group, when compared with the PHZ group.***

Conclusion: ***Our findings suggest that the extract of Desmondium adscendens leaves has the capacity to ameliorates the Phenylhydrazine-induced oxidant-antioxidant imbalances in Wistar rats.***

Index Terms- ***Desmondium adscendens, Phenylhydrazine, Biochemical indices***

I. INTRODUCTION

Since time immemorial, man quest for survival and healthcare has led to the discovery of many plants of medicinal value before the advent of orthodox medicine. Although, its usage, particularly in Africa, was first seen to be archaic and incorrectly contested by foreign nations, beginning with colonial control in Africa, and then by conventional or orthodox medical practitioners (Sofowara, 1993; Josephine and Antoinette, 2019). However, in the recent time, biomedical research has proven and authenticate the effectiveness of numerous therapeutic plants for a range of conditions and illnesses, including management, prevention, and therapy.

Oxidative stress is a product of either increased ROS generation or impaired antioxidant defense system (Kıran et al., 2023; Aribo et al. 2024). Increased reactive oxygen species (ROS) generation and a corresponding decline in antioxidant capacity are linked to phenylhydrazine, leading to oxidative stress. Oxidative stress induces lipid peroxidation in tissues; the end product of which is malondialdehyde (MDA) (Bey et al., 2024). Superoxide dismutase (SOD) and glutathione are the two main ROS-scavenging enzymes in the body (Nsa et al., 2025). The first line of defence against free radicals is SOD. It catalyses the dismutation of superoxide radicals to H2O2 and molecular oxygen. GSH on the other hand

reduces hydroperoxides like ROOH and H2O2 (Gekpe et al., 2025).

Over the years, Phenylhydrazine (PHZ) has been used to experimentally induced oxidative stress in animal model. The effect of PHZ stem from its ability to distort oxidative balance via production of lipid peroxidation and reactive oxygen species of red blood cells (Berge, 2007; Amer et al. 2004). This compromised the integrity of the cell membrane resulting in rapid apoptosis (Sheth et al. 2021). Exposure to Phenylhydrazine causes damaging effects on tissues such as the spleen, kidney and liver (Nsa et al. 2025; El-Shafey et al. 2023).

Desmondium adscendens is one of the species in the Desmodium genus, typically found in tropical regions of Africa, South America, Asia, Australia, and Oceanis (Taylor, 2005). The plant is known with different names such as; Strong-back, beggarlice, pega pega and amer seco, hardstick, tick clover among others (Rastogi et al. 2011). It is one of the many therapeutic plants that have been utilised for a variety of purposes. Native inhabitants of the regions where it grows are thought to have utilised it for thousands of years to treat a range of medical conditions such as diarrheas, malaria, rheumatism, pneumonia, fever, epilepsy, asthma, jaundice, gastroduodenal ulcer, diabetes, sickle cell anaemia, hepatic diseases among others (Muanda et al. 2011; Kuldeep et al. 2015). Numerous scientific findings (Muanda et al. 2011; Seriki et al. 2019) documented therapeutic phytochemicals in Desmondium adscendens includes; alkaloids (indolialkaloids), flavonoids (vitexin and isovitexin), saponins (Dehydrosoyaponin), polyphenols, tannin, anthrocyanin and astragalin. Rastogi et al. (2011) listed that the main chemical found in amer seco are astragalin, beta- phenylethylamines, cosmosiin, cyaniding-3-0-sophoroside, dehydrosoyasaponins, hordenine, pelargonidin-3-0-rhaminoside, salsoline, soyasaponins, tectorigenin, tetrahydroionsquinolines and tyramine. These isolated compounds demonstrated a broad range of pharmacological actions both in-vitro and in-vivo, including anti-leishmanial, immunomodulatory, anti-asthmatics, anti-bacterials, anti- viral, cardioprotective, anti-inflammatory, hepatoprotective, relaxation of smooth muscles and anti-oxidant activities, anti-ulcer, vermicidal, (Rastogi et al. 2011; Ma et al. 2011). However, this plant has been extensively research, and its effect as a blood cleanser reported. There is paucity of report to buttress that claim. Therefore, the research on ethanolic extract of Desmondium adscendens leaves on some oxidative stress indices in Phenylhydrazine administered Wistar rats.

II. MATERIALS AND METHODS

Plant and Drugs collection: Fresh Desmondium adscendens leaves was obtained from a farrow land in Bayside, Calabar South, Cross River state and identified by a botanist in Department of Botany, University of Calabar, Nigeria.

Phenylhydrazine hydrochloride (Sigma-Adrich Chemical Company, St. louis, USA) was bought from BEZ pharmacy, Etagbor, Calabar.

Extract preparation: The extraction was done according to the method of Horablaga et al. (2023); the leaves of Desmondium adscendens were kept out of direct sunshine and dried for seven days in a spacious room. The dried leaves were grinded into a coarse powder. 113g of the powdered leaves were soaked in 800ml of ethanol solution in a container and a stirrer was used to stir and mixed it thoroughly and kept for 72hours.

White cotton cloth was used to filter the mixture, and Whatman no. 1 filter paper was used to further filter the filtrate. To make the filtrate dry, it was evaporated under room temperature, and 16g of extract was collected into plain sample bottles and tag; Ethanolic Extract of Desmondium adscendens leaves (EDAL). This was stored in refrigerator prior to its use.

Experimental animals: Twenty-five (25) male Wistar rats weighing 180-200g were used. They were allowed to acclimatize for two weeks under standard environmental condition and fed with rodent feed and water at will.

Experimental Design: Random sampling method was implored to assign the rats to five strata, and each group containing five animals were kept in wooden cages. The grouping was as follows; Group1(control)

receive only normal saline, Group2 (PHZ) received intraperitoneal injection (IP) of Phenylhydrazine(60mg/kg B.w) twice in 48hours. Group 3(EDAL only) received 100mg/kg B.w of ethanoic extract of Desmondium adscendens leaves only, while Group4 (PHZ+ EDALLD) and Group5 (PHZ+EDALHD) were administered PHZ as Group 2 and then treated with 100mg/kg B.w and 200mg/g B.w of ethanolic extract of Desmondium adscendens (EDAL) respectively (Udo et al. 2026). All treatment except Phenylhydrazine (IP) was given orally using oral cannula and 14 days was the treatment regimen.
All experimental procedures and animal handling were approved(343PHY301) by the University of Calabar's Faculty of Basic Medical Sciences' Animal Research Ethics Committee, Calabar, Nigeria. The experimental duration was chosen based on previous studies, while the dosage of Phenylhydrazine and EDAL were based on the studies of Archibong et al. (2025) and Ayoola et al. (2018) as used by Udo et al. (2026) respectively.

Collection of Blood samples: At the end of the administration, on day 15, after an overnight fast, the animals were put under anaesthesia in a room containing chloroform. Cardiac punctures were used to obtain blood samples, which were then placed in plain sample vials for biochemical analysis.
Biochemical analysis: For fifteen minutes, the blood samples were centrifuged at 700 rpm and serum was separated and used for the evaluation of the following biochemical parameters; Super oxide dismutase (SOD) was determined using Elisa Kits, Catalase (CAT), reduced Glutathione (GHS) and Malondialdehye (MDA) were determined following the method of Sinha (1972), Beutler et al., (1963) and Draper and Hadley, (1990) respectively.

Statistical analysis
A one-way analysis of variance (ANOVA) and a post hoc test were used to analyse the data.
The mean ± standard error of mean was used to present the results. $P<0.05$ values were regarded as statistically significant.

III. RESULT

Oxidative stress biomarkers in different experimental groups

The mean values of superoxide dismutase (SOD) for control, PHZ, EDAL only, PHZ+ EDALLD, PHZ+ EDALHD were 9.25±0.320, 4.41±0.270, 11.0±0.640, 7.41±0.270 and 8.21±0.310 respectively.

The mean values of catalase (CAT) for control, anemic control, EDAL only, PHZ+ EDALLD, PHZ+ EDALHD were 61.6±1.90, 31.2±1.60, 69.7±2.12, 48.3±2.32 and 56.5±1.73 respectively.

The mean values of glutathione (GSH) for control, PHZ, EDAL only, PHZ+ EDALLD, PHZ+ EDALHD were 161±1.46, 74.5±1.74, 177±0.870, 142±1.24 and 157±1.64 respectively.

The results of the effects of EDAL on oxidative stress biomarkers as presented in figure 1-4, show significant decrease($P<0.05$) in the mean serum level of SOD, CAT, GSH in PHZ compare to normal control. Treatment with EDAL in Group 4(PHZ+ EDALLD) show a significant increase($P<0.005$) in these parameters compare to PHZ, but significantly decrease compare to normal control. Also, Group 5 (PHZ+ EDALHD) show a significant increase($P<0.005$) compare to PHZ, and Group 4, however, there was no significant different compared to normal control.

The mean values of malondialdehyde (MDA) for control, PHZ, EDAL only, PHZ+ EDALLD, PHZ+ EDALHD were 32.5±0.540, 68.5±1.14, 29.5±1.74, 41.3±0.680 and 48.6±1.36 respectively.

There was significant increase($P<0.05$) in MDA in PHZ compared to normal control, however, treatment with EDAL in Group 4 and Group 5 show a significant decrease($P<0.005$) compared to PHZ. Similarly, administration of EDAL only shows a significant improvement($P<0.05$) in all parameters compared to normal control.

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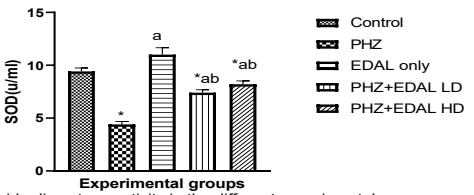

FIG. 1: Superoxide dismutase activity in the different experimental groups.
Values are expressed as mean +SEM, n = 5.
* = p<0.05 vs Control
a=p<0.05 vs PHZ
b=EDAL only

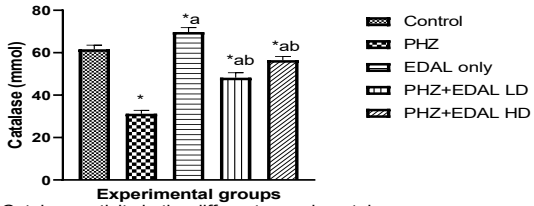

FIG. 2: Catalase activity in the different experimental groups.
Values are expressed as mean ± SEM, n = 5.
* = p<0.05 vs Control
a=p<0.05 vs PHZ
b=P<0.05 vs EDAL only

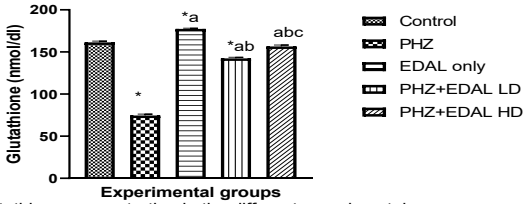

FIG. 3: Glutathione concentration in the different experimental groups.
Values are expressed as mean ± SEM, n = 5.
* = p<0.05 vs Control
a=p<0.05 vs PHZ
b=P<0.05 vs EDAL only
c=P<0.05 vs PHZ+EDAL LD

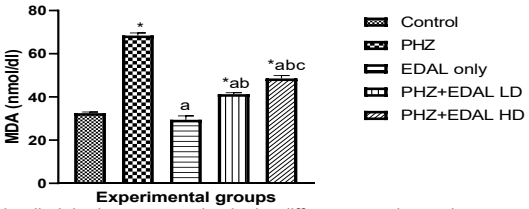

FIG. 4: Malondiadehyde concentration in the different experimental groups.
Values are expressed as mean ± SEM, n = 5.
* = p<0.05 vs Control
a=p<0.05 vs PHZ
b=P<0.05 vs EDAL only
c=P<0.05 vs PHZ+EDAL LD

IV. DISCUSSION

This study demonstrated the ability of the ethanolic extract of Desmondium adscendens leaves (EDAL) to reverse oxidant stress markers derangement in Phenylhydrazine-induced anaemic rats. Apart from nutritional, idiopathic factors and environmental concerns arising from chemicals and drugs-induced health conditions in both human and animals; Phenyhydrazine (PHZ) is one of such drugs, and has been used over the years to experimentally induced oxidative stress in animal models (WHO, 2000).

An essential antioxidant enzyme called superoxide dismutase (SOD) catalyses the conversion of superoxide radicals (O_2^-) into hydrogen peroxide and other compounds, shielding cells from oxidative stress. Alterations in SOD activity often reflect changes in the oxidative balance of tissues.

Catalase (CAT) is a important antioxidant enzyme that breaks down hydrogen peroxide (H_2O_2) into water and oxygen, thus preventing oxidative damage to cellular components. Alterations in CAT activity is widely utilized as an oxidative stress indicator.

Using reduced glutathione peroxidase (GPx) as a cofactor, glutathione (GSH), an antioxidant enzyme, detoxifies lipid peroxides and hydrogen peroxide ($H2O_2$). GSH is essential for preventing oxidative damage to cell membranes. Altered GSH activity reflects changes in oxidative stress and redox balance.

Earlier reports have it that Phenylhydrazine (PHZ) causes distortion in the oxidant balance through oxidative stress (Sheth et al. 2021; Berger, 2007). PHZ generate more ROS, increase lipid peroxidation and decrease antioxidant capacity leading to oxidative stress (Amer et al. 2004).

These reports are supported by our results evidenced in significant reduction of serum SOD, CAT, GSH and elevation of MDA in PHZ treated group compared to normal control. This signifies depletion of endogenous antioxidants pool which might altered oxidant-antioxidants ratio, predisposing oxidative stress (WHO, 2000).

The detoxify ability of the extract could also explain the normalization of the blood cells relative to anaemic control. EDAL reveals therapeutic potentials by increasing antioxidants levels, this could ameliorate PHZ induced oxidative stress by replenishing body's antioxidants capacity.

V. CONCLUSION

It is concluded that the ethanolic extract of Desmondium adscendens leaves (EDAL) has the ability to reversed oxidative stress markers distortion induced by Phenylhydrazine toxicity.

Ethical Consideration

The National Institutes of Health's 1985 publication on laboratory animal care was followed when handling the rats. The University of Calabar Ethics Committee (Nigeria) provided ethical approval (343PHY301) and rules for animal experimentation.

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