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Effect of *Launaea Taraxacifolia* Aqueous Leaves Extracts on ICAM-1 and VCAM-1 Gene Expressions in Benzene-Induced Haematotoxicity in Albino Wistar Rats

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ABSTRACT: *Launaea taraxacifolia*, is a plant used in traditional medicines, due to its antioxidant and anti-inflammatory properties. There is a need to study its ability to mitigate the harmful effects of benzene-induced haematotoxicity which potentially involves modulation of Intercellular Adhesion Molecules 1 (ICAM-1) and Vascular Cell Adhesion Molecules 1 (VCAM-1) gene expressions. Therefore, the aim of this study was to determine the effects of aqueous leaf extracts of *Launaea taraxacifolia* on ICAM-1 and VCAM-1 gene expression in benzene induced haematotoxicity in albino Wistar rats using standard methods. Data obtained was analyzed by the Statistical Package for Social Science (SPSS) software. The mRNA expression of ICAM-1 was significantly lower in groups B and C when compared to group A ($p<0.05$); While the mRNA expression of VCAM-1 in groups C, D, E and F was significantly lower when compared to groups A and B ($p<0.05$). This study concludes that benzene exposure induced a reduction in ICAM-1 mRNA expression, while VCAM-1 mRNA expression significantly increased. Co-administration of benzene and varying *Launaea taraxacifolia* concentrations reversed the effects on ICAM-1 and VCAM-1 expression caused by benzene.

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Keywords: *Launaea taraxacifolia*; Intercellular Adhesion Molecules-1 gene; Vascular Cell Adhesion Molecules-1 gene; Haematological parameters; Benzene.

Launaea taraxacifolia, commonly known as African lettuce, is a plant that is traditionally used in African folk medicine for its potential therapeutic properties. It can be found mostly in the western, central, and eastern parts of the continent, in nations like Nigeria, Cameroon, Uganda, Tanzania, and Kenya (Dansi *et al*., 2012). The plant thrives in grasslands, savannahs, and disturbed places like roadsides and wastelands. It's a perennial herb of the Asteraceae family, which includes daisies and sunflowers (Sakpere and Aremu, 2008). *Launaea taraxacifolia* has been reported to possess antioxidant, anti-inflammatory, and hepato-protective effects, among others (Adebisi, 2004). The leaves of L. *taraxacifolia* have been found to contain minerals, proteins, flavonoids, fatty acids, and vitamins (Adinortey *et al*., 2012). Phytochemical studies of *L. taraxacifolia* indicated that the plant contains chemical classes recognized to have phyto-therapeutic significance for humans, such as phenolic glycosides, flavonoids, saponins, and terpenoids (Olugbenga *et al*., 2015). Intercellular bond particle 1 (ICAM-1) is a trans-membrane protein in the immunoglobulin superfamily communicated on the outer layer of different cell populaces and up regulated by provocative boosts. It intervene cell glue connections by restricting to the β2 integrins macrophage antigen 1 and leukocyte capability related antigen 1, as well as different ligands (Hopkins *et al*., 2004). It plays significant parts in the resistant framework, remembering for leukocyte grip to the

endothelium and trans-endothelial relocation, and at the immunological neural connection shaped among lymphocytes and antigen-introducing cells. Vascular cell bond protein (VCAM-1) is a glycoprotein that is prevalently communicated in endothelial cells. Under elevated degrees of aggravation and persistent circumstances in certain sicknesses, VCAM-1 additionally is communicated on the outer layer of different cells, including tissue macrophages, dendritic cells, bone marrow fibroblasts, myoblasts, oocytes, and disease cells (Sharma *et al*., 2017). Benzene is a generally utilized modern compound and natural poison that presents critical wellbeing dangers to people and creatures. Delayed openness to benzene has been related with different unfriendly impacts, including haematotoxicity, which influences the blood-framing organs and prompts irregularities in platelet creation and capability. Long haul inward breath openness of benzene has been displayed to cause haematotoxicity and an expanded rate of intense myelogenous leukemia in people (Farris *et al*., 1997). Constant openness to benzene can influence the bone marrow and lead to the concealment of the insusceptible framework, possibly affecting the articulation and capability of particles like ICAMs and VCAMs. Intercellular adhesion molecule 1 (ICAM-1) and Vascular cell adhesion protein (VCAM-1) are cell adhesion molecules that play a crucial role in the inflammatory process. Elevated levels of ICAM-1 and VCAM-1 have been observed in various inflammatory diseases, including atherosclerosis, rheumatoid arthritis, and certain types of cancer. Given that benzene exposure can induce inflammation and oxidative stress, it is plausible that benzene-induced toxicity may also lead to a dysregulation of ICAM-1 and VCAM-1. There is growing interest in the potential of plant-derived compounds to mitigate the adverse effects of environmental toxins, including benzene. However, the specific effects of *Launaea taraxacifolia* leaf extracts on the expression of ICAM-1 and VCAM-1 in the context of benzene-induced toxicity have not been extensively investigated which therefore necessitates this study. The aim of this study therefore is to determine the effect of *Launaea taraxacifolia* leaf extracts on Intercellular adhesion molecule 1 (ICAM-1) and Vascular cell adhesion protein 1 (VCAM-1) gene expressions in benzene-induced haematotoxicity in albino Wistar rats.

MATERIALS AND METHODS

*Study Population***:** In this study, animal (rats) model was used. A total of sixty (60) adult male albino Wistar strain were purchased from the animal holdings of the Department of Anatomy, University of Benin, Benin City, Nigeria. The rats were housed at the animal housing wing of the Department of Anatomy, University of Benin.

Identification of the Launaea taraxacifolia Leaves: Launaea taraxacifolia leaves were harvested from Faculty of Agriculture in the University of Benin, Nigeria. The leaves were then identified and authenticated by Dr. A. O Akinnibosun of the Department of Plant Biology and Biotechnology, Faculty of Life Sciences, University of Benin, Benin City (Obazelu *et al*., 2021).

Preparation of Plant Extract: About 3.65kg leaves were pulverized (after drying) by a commercial blender and (665g of powder obtained), soaked in distilled water using 1g of powder to 5ml of distilled water and allowed to stand at room temperature for 72 hours at room temperature. The extract was filtered using Whatman's (Nitro cellulose 45; 0.45µm pore size) filter paper and the filtrates were concentrated to dryness at 100ºC in a water bath. Thereafter, it was put in an airtight container and refrigerated until use (Obazelu *et al*., 2021).

Animal Care: Animals were housed in a cross ventilated room in the animal holdings of the department of anatomy, University of Benin, Benin City. Animals were exposed to 12 hours dark and light cycles with access to feed and water *ad libitum*. The rats were acclimatized for a period of two (2) weeks before commencement of the experiment (Obazelu *et al*., 2021).

Ethical Consideration: Ethical approval was obtained from Research Ethics Committee on animal subjects from Edo State Ministry of Health, Benin City (Ref Number: HA/737/23/B/200600143 issued on 30th June, 2023).

Preparation of Benzene and Cyclophosphamide Drug Solution: Benzene Solution: Benzene solution was made by mixing benzene (Manufactured by LOBA Chemie laboratory reagents and fine chemicals limited, Batch Number: L246641711), distilled water v/v and 2-propanol in the ratio of 1:5:5. That is, 1 part of benzene was mixed with 5 part of distilled water v/v and 5 parts of 2-propanol. 0.2ml of this benzene solution was administered to each animal in the various test groups with an average weight of 150g every 48 hours for 28 days (Akanni *et al*., 2014).

*Cyclophosphamide Drug Solution: C*yclophosphamide drug solution was made by mixing 500mg of the powdered drug in 25ml of distilled water. 0.3ml of this drug solution was administered orally to each animal in group C of an average weight

of 150g every 48 hours for 28 days (Akanni *et al*., 2014).

Grouping of Animals: Sixty (60) Mature Wistar rats weighing 150-200g were randomly selected and divided into six groups (n = 10 per group). The groups were the Group A, Group B, Group C, Group D, Group E and Group F.
Group A: This was the control group. Animals in this group received only standardized feed (Manufactured by KARMA AGRIC FEEDS AND FOOD LIMITED, Oyo State) and clean water *ad libitum.*
Group B: This group received only benzene intraperitoneally.
Group C: Animals in this group were administered benzene solution and treated with the standard drug solution (cyclophosphamide) intraperitoneally.
Group D: Animals in this group were administered benzene solution intraperitoneally and treated with low dose of *Launaea taraxacifolia* leaves extract orally.
Group E: Animals in this group were administered benzene solution intraperitoneally and treated with a higher dose of *Launaea taraxacifolia* leaves extract orally.
Group F: Animals in this group were administered benzene solution intraperitoneally and treated with the highest dose of *Launaea taraxacifolia* leaves extract orally.

*Dosage of Cyclophosphamide Administered (Akanni et al., 2014): Administered Doses of Launaea taraxacifolia Extract***:** Group A (control) received only standardized feed and clean water *ad libitum*. Group B (benzene treated group) were administered 0.2ml of benzene solution intraperitoneally every 48 hours for 28 days. Group C (cyclophosphamide drug solution treated group) were administered 0.2ml of benzene solution intraperitoneally every 48 hours for 28 days and treated with 0.3ml of 6mg/ml of cyclophosphamide 48 hourly for 28 days. Group D were administered with 0.2ml of benzene solution intraperitoneally every 48 hours for 28 days and treated with 0.15ml of 100mg/kg body weight of *Launaea taraxacifolia* leaves extract orally using a gavage tube every 24 hours for 28 days. Group E were administered with 0.2ml of benzene solution intraperitoneally every 48 hours for 28 days and treated with 0.3ml of 200mg/kg body weight of *Launaea taraxacifolia* leaves extract orally using a gavage tube every 24 hours for 28 days. Group F were administered with 0.2ml of benzene solution intraperitoneally every 48 hours for 28 days and treated with 0.6ml of 400mg/kg body weight of *Launaea taraxacifolia* leaves extract orally using a gavage tube every 24 hours for 28 days.

Sacrifice of Animals and Collection of Samples: At the end of the experimental period, the animals were grossly observed for general physical characteristics. A midline incision was made through the ventral wall of the rats after anaesthetizing (using chloroform) and cervical dislocation. Five milliliters (5ml) of blood were collected from each rat using a sterile syringe and placed in an Ethylene Diamine Tetra-acetic Acid (EDTA) container for full blood count analysis. Bone marrow samples were also obtained from the rats by opening the femur longitudinally and exposing the marrow cavity. A sterile forceps was used to obtain the bone marrow from the cavity and placed in an Eppendorf container containing Trizol for molecular analysis.

Haematological Profile Analysis: The full blood count parameters were analysed immediately after sample collection using the automated five parts ERMA Haematology Auto analyser PCE-210N (Diamond Diagnostic; Holliston, USA). Calibration and standardization of the equipment, processing and analysis of the samples were done strictly according to the manufacturer's instructions.

Detection Principle of Haematology Autoanalyser: The instrument counts and sizes the cells. It detects and measures changes in electrical resistance when a particle (such as a cell) passes through a gem aperture sensor. Sample was diluted in a conductive liquid. Each time a blood cell will pass through the aperture a resistant signal will be generated because blood cells are bad conductors. When cell goes through the aperture, the resistance increases with increase in cell volume. According to the Ohm formulary: U=RI (U =Voltage I =Current R =Resistance). If I is invariable, U is increased as cell volume increases. Treat by magnifying circuit, the voltage signal is amplified; background noise is removed, and receives the signal to analysis. WBC and RBC/PLT are analysed by two different circuits. The MPU analyses and calculates the cells, then gives the histograms. The count of PLT adopts an advanced liquid, electron and soft system, which can settle the repetitive count of the cells. If RBC enters the analysis area, they will have similar pulses with PLT.

Procedure: The whole blood was properly mixed and inserted into the probe. Then 20 µL of the blood was aspirated into the instrument. The analysis was immediately done and the results displayed on the screen after about 1-2 minutes, which was printed by the printer.

ICAM-1 and VCAM-1 mRNA Assay: Isolation of Total RNA: Total RNA was isolated from tissue samples

with Quick-RNA MiniPrep™ Kit (Zymo Research). The DNA contaminant was removed following DNAse I (NEB, Cat: M0303S) treatment. The RNA was quantified at 260 nm and the purity confirmed at 260 nm and 280 nm using A&E Spectrophotometer (A&E Lab. UK).

cDNA Conversion: One (1) µg of DNA-free RNA was converted to cDNA by reverse transcriptase reaction with the aid of cDNA synthesis kit based on ProtoScript II first-strand technology (New England BioLabs) in a condition of 3-step reaction: 65 °C for 5 min, 42 °C for 1 h, and 80 °C for 5 min (Elekofehinti *et al*., 2020).

PCR Amplification and Agarose Gel Electrophoresis: Polymerase chain reaction (PCR) for the amplification of ICAM-1 and VCAM-1 gene was carried out with OneTaqR2X Master Mix (NEB) using the following primers (Inqaba Biotec, Hatfield, South Africa). PCR amplification was performed in a total of 25µl volume reaction mixture containing cDNA, primer (forward and reverse SEE BELOW) and Ready Mix Taq PCR master mix. Under the following condition: Initial denaturation at 95ºC for 5 min, followed by 30 cycles of amplification (denaturation at 95ºC for 30 seconds, annealing for 30 seconds and extension at 72ºC for 60 seconds) and ending with final extension at 72ºC for 10 min. The amplicons were resolved on 1.0% agarose gel. The Glyceraldehyde-3-Phosphate Dehydrogenase (GAPDH) gene was used to normalize the relative level of expression of each gene, and quantification of band intensity was done using "image J" software (Elekofehinti *et al*., 2020).

Primer Sequences
ICAM-1: Forward: AGA AGG ACT GCT TGG GGA A
Reverse: CCT CTG GCG GTA ATA GGT G
VCAM-1: Forward: CTG ACC TGC TCA AGT GAT GG
Reverse: GTG TCT CCC TCT TTG ACG CT
GAPDH: CTCCCTGGAGAAGAGCTATGA
AGGAAGGAAGGCTGGAAGA

Statistical Analysis: Data obtained from this research was presented and analyzed using statistical package for social sciences (SPSS) version 21.0 (IBM Inc. USA). Analysis of variance (ANOVA) was used to compare treatment groups of continuous variables. Tukey HSD *post hoc* was applied where a significant difference was observed in the ANOVA. Bar chart was used to represent the mRNA gene expression patterns. A p value of ≤0.05 was considered statistically significant.

RESULTS AND DISCUSSION

Table 1 shows the comparison of Mean±SD of white blood cell parameters of six groups namely; groups A, B, C, D, E and F, representing control, benzene group, cyclophosphamide group, benzene + 100mg/kg *Launaea taraxacifolia*, benzene + 200mg/kg *Launaea taraxacifolia* and benzene + 400mg/kg *Launaea taraxacifolia* respectively. Total white blood cell count (TWBC µL) was significantly higher in group A (18.4±1.159) when compared to group C (10.89±0.4298), D (14.61±0.3318), E (14.56±0.3145) and F (13.05±1.372) ($p<0.05$). Lymphocyte count (%) was significantly lower in group C (59.6±3.655) when compared to groups A (85.15±0.708), B (86.3±1.463), D (89.19±0.96), E (87.79±1.331) and F (90.69±0.49) ($p<0.05$). Monocyte count (%) was significantly higher in group C (20.79±2.95) when compared to groups A (5.66±0.12), B (6.21±0.65), D (3.02±0.31), E (4.33±0.3) and F (4.27±0.27) ($p<0.05$). Neutrophil count (%) was significantly higher in group C (11.12±0.87) when compared to groups A (6.01±0.54), B (4.6±0.51), D (5.71±0.38), E (5.2±1.062) and F (3.47±0.17) ($p<0.05$). Basophil count (%) was significantly higher in group C (8.24±0.71) when compared to groups A (2.87±0.19), B (2.48±0.57), D (1.67±0.42), E (2.51±0.59) and F (1.28±0.22) ($p<0.05$). Table 2 shows the comparison of Mean±SD of platelets parameters of six groups namely; groups A, B, C, D, E and F, representing control, benzene group, cyclophosphamide group, benzene + 100mg/kg *Launaea taraxacifolia*, benzene + 200mg/kg *Launaea taraxacifolia* and benzene + 400mg/kg *Launaea taraxacifolia* respectively. Platelet count was significantly lower in group A (489.8±22.7) when compared to group C (753.7±37.27) ($p<0.05$). Group B (605.8±41.23) had a significantly lower platelet count when compared to group C (753.7±37.27) but significantly higher when compared to groups D (479.8±14.58), and F (475.4±23.06) ($p<0.05$). Platelet count of group C (753.7±37.27) was significantly higher than groups D (479.8±14.58), E (524.8±11.53) and F (475.4±23.06) ($p<0.05$). Mean platelet volume (MPV) was significantly higher in groups A (6.41±0.02), B (6.31±0.10) and C (6.26±0.016) when compared to group F (5.75±0.197) ($p<0.05$). PCT was significantly lower in group A (0.315±0.015) when compared to group C (0.467±0.024) ($p<0.05$). PCT was significantly lower in group B (0.387±0.03) when compared to group C (753.7±37.27), but higher when compared to groups D (0.299±0.004), E (0.305±0.005) and F (0.252±0.014) ($p<0.05$). PLCR was significantly higher in group A (4.81±0.33) when compared to group E (3.28±0.071) ($p<0.05$).

Table 1. Mean Comparison of White Blood Cell Parameters of Studied Groups

Parameters	Group A (Control) (n=10)	Group B (Benzene only) (n=10)	Group C (Benzene+ Cyclophosphamide) (n=10)	Group D (Benzene+ 100mg/kg) (n=10)	Group E (Benzene+ 200mg/kg) (n=10)	Group F (Benzene+ 400mg/kg) (n=10)	F value	p value
TWBC ($\times10^6$/µL)	18.4 ±1.159^{cdef}	18.59 ±0.4769^{cdef}	10.89 ±0.4298^{abde}	14.61 ±0.3318^{abc}	14.56 ±0.3145^{abc}	13.05 ±1.372^{ab}	14.18	<0.0001
Lymphocyte (%)	85.15 ±0.708^{c}	86.3 ±1.463^{c}	59.6 ±3.655^{abdef}	89.19 ±0.96^{c}	87.79 ±1.331^{c}	90.69 ±0.49^{c}	43.26	<0.0001
Monocyte count (%)	5.66 ±0.12^{c}	6.21 ±0.65^{c}	20.79 ±2.95^{abdef}	3.02 ±0.31^{c}	4.33 ±0.3^{c}	4.27 ±0.27^{c}	28.26	<0.0001
Neutrophil count (%)	6.01 ±0.54^{c}	4.6 ±0.51^{c}	11.12 ±0.87^{abdef}	5.71 ±0.38^{c}	5.2 ±1.062^{c}	3.47 ±0.17^{c}	16.03	<0.0001
Eosinophil count (%)	0.31 ±0.069	0.41 ±0.04	0.25 ±0.07	0.41 ±0.08	0.17 ±0.01	0.29 ±0.02	2.472	0.0435
Basophil count (%)	2.87 ±0.19^{c}	2.48 ±0.57^{c}	8.24 ±0.71^{abdef}	1.67 ±0.42^{c}	2.51 ±0.59^{c}	1.28 ±0.22^{c}	26.56	<0.0001

Key: p≤0.05- Significant; p ≥ 0.05- Not significant. a represents significance with control, b represents significance with benzene group, c represents significance with benzene + cyclophosphamide group, d represents significance with benzene + 100mg/kg, e represents significance with benzene + 200mg/kg, f represents significance with benzene + 400mg/kg.
TWBC: Total White Blood Cell

Table 2. Mean Comparison of Platelet Parameters of Studied Groups

Parameters	Group A (Control) (n=10)	Group B (Benzene only) (n=10)	Group C (Benzene+ Cyclophosphamide) (n=10)	Group D (Benzene+ 100mg/kg) (n=10)	Group E (Benzene+ 200mg/kg) (n=10)	Group F (Benzene+ 400mg/kg) (n=10)	F value	p value
PLT (10^3/µL)	489.8 ±22.7^{bc}	605.8 ±41.23^{acdf}	753.7 ±37.27^{abdef}	479.8 ±14.58^{bc}	524.8 ±11.53^{c}	475.4 ±23.06^{bc}	15.86	<0.0001
MPV (µM^3)	6.41 ±0.02^{f}	6.31 ±0.10^{f}	6.26 ±0.016^{f}	6.17 ±0.097	6.02 ±0.041	5.75 ±0.197^{abc}	5.387	0.0004
PCT (%)	0.315 ±0.015^{c}	0.387 ±0.03^{cdef}	0.467 ±0.024^{abdef}	0.299 ±0.004^{bc}	0.305 ±0.005^{bc}	0.252 ±0.014^{bc}	16.28	<0.0001
PDW (%)	28.46 ±5.90	32.87 ±2.21	24.73 ±0.50	32.65 ±1.82	27.68 ±1.75	29.3 ±1.90	1.156	0.343
PLCR (%)	4.81 ±0.33^{e}	3.94 ±0.275	3.91 ±0.093	4.16 ±0.41	3.28 ±0.071^{a}	3.72 ±0.266	3.497	0.0082

Key: p≤0.05- Significant; p ≥ 0.05- Not significant. a represents significance with control, b represents significance with benzene group, c represents significance with benzene + cyclophosphamide group, d represents significance with benzene + 100mg/kg, e represents significance with benzene + 200mg/kg, f represents significance with benzene + 400mg/kg. PLT: Platelets, MPV: Mean Platelet Volume, PCT: Plateletcrit, PDW: Platelet distribution width PLCR: Platelet Larger Cell Ratio

Figure 1 shows the expression of genes as represented by gel electrophoresis picture and internal control (Glycealdehyde-3-Phosphate Dehydrogenase {GADPH}) of mRNA expression of ICAM-1 of groups A, B, C, D, E and F, representing control, benzene group, cyclophosphamide group, benzene + 100mg/kg *Launaea taraxacifolia*, benzene + 200mg/kg *Launaea taraxacifolia* and benzene + 400mg/kg *Launaea taraxacifolia* respectively, represented on different bars on the bar chart. There was a significant decrease in the expression of ICAM-1 in groups B and C when compared to the expression in group A ($p<0.05$). Expression of ICAM-1 in groups D, E and F significantly increased when compared to groups B and C ($p<0.05$). Higher concentration of extract showed higher expression of ICAM-1 expression. Figure 2 shows the expression of genes as represented by gel electrophoresis picture and internal control (Glycealdehyde-3-Phosphate Dehydrogenase {GADPH}) of mRNA expression of VCAM-1 of groups A, B, C, D, E and F, representing control, benzene group, cyclophosphamide group, benzene + 100mg/kg *Launaea taraxacifolia*, benzene + 200mg/kg *Launaea taraxacifolia* and benzene + 400mg/kg *Launaea taraxacifolia* respectively, represented on different bars on the bar chart. There was a significant increase in the expression of VCAM-1 in group B when compared to the expression in group A ($p<0.05$). Expression of VCAM-1 in groups C, D, E and F decreased significantly when compared to groups A and B ($p<0.05$). Benzene is a widely used industrial chemical and a prevalent component of air pollutants and its exposure has been associated with various health concerns, including haematotoxicity, which disturbs the balance of haematopoietic processes within the body (Wang *et al*., 2012). In the pursuit of addressing the harmful effects of benzene-induced haematotoxicity, researchers have been exploring various ways to uncover potential therapeutic interventions. Among these, medicinal plants have emerged as promising candidates, owing to their rich reservoir of bioactive compounds with potential health benefits (Shakya, 2016).

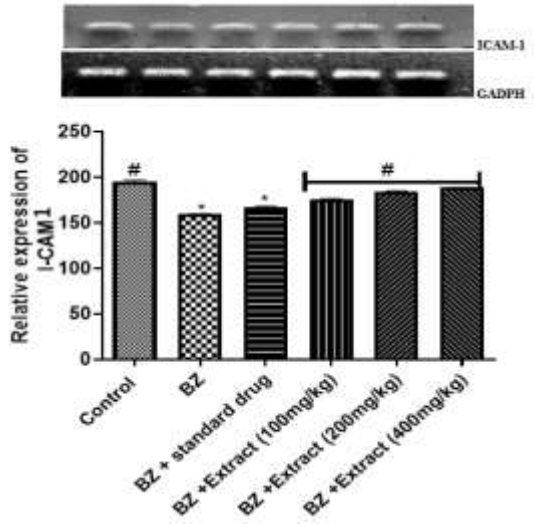

Fig. 1: mRNA Expression of ICAM-1 of the Studied Groups. * Represents statistical difference to control. # Represents statistical difference to benzene induced group at p<0.05. Key: BZ=Benzene, GADPH=Glyceraldehyde-3-Phosphate Dehydrogenase

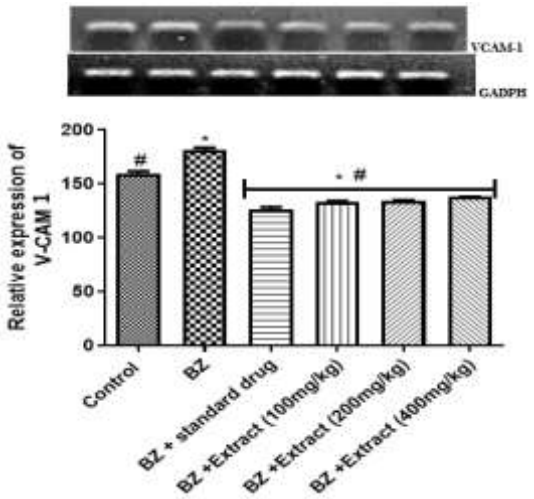

Fig. 2: mRNA Expression of VCAM-1 of the Studied Groups. * Represents statistical difference to control. # Represents statistical difference to benzene induced group at p<0.05. Key: BZ=Benzene, GADPH=Glyceraldehyde-3-Phosphate Dehydrogenase.

An important part of benzene-induced haematotoxicity is the altered expression of key genes associated with the regulation of adhesion molecules. Intercellular Adhesion Molecule-1 (ICAM-1) and Vascular Cell Adhesion Molecule-1 (VCAM-1) are vital in generating immune responses and cellular interactions within the vascular system. Any changes in their gene expressions can disrupt the equilibrium of immune responses and the vascular system (Fotis *et al*., 2012). By exploring the impact of *Launaea taraxacifolia* on these adhesion molecules, valuable insights can be gained into the mechanisms underlying its purported protective effects against the haematotoxicity induced by benzene exposure. The assessment and understanding of overall health heavily rely on the cellular constituents of blood. Blood comprises diverse cellular components, namely red blood cells, white blood cells, and platelets. Each of these elements performs unique roles, and alterations in their quantities, shapes, or functions can offer significant information about the well-being of an organism (Azuonwu *et al*., 2017). In this study, benzene administration did not lead to significant changes in the total white blood cell count, as well as lymphocytes, monocytes, neutrophils, eosinophils, and basophils. This lack of change may be attributed to the timing and duration of the benzene exposure. Haematotoxicity is often characterized by alterations in the production, differentiation, and lifespan of blood cells. The lack of significant changes in the WBC count and its subtypes could suggest that the exposure duration or dose of benzene used in the study might not have been sufficient to induce detectable haematotoxic effects. This result is in contrast with that of Ola *et al*., (2022) who reported an increase in total white blood cell (WBC) count after benzene administration in albino Wistar rats. Cyclophosphamide however, caused a reduction in total white blood cell count and lymphocytes which is consistent with its expected immunosuppressive effects. This reduction of white blood cell counts and lymphocyte by cyclophosphamide is in line with a study by Ukpo *et al*., (2013). In this study, the co-administration of different doses of *Launaea taraxacifolia* and benzene had an impact on the overall count of white blood cells. Administration of 100mg/kg, 200mg/kg, and 400mg/kg of *Launaea taraxacifolia* alongside benzene, led to a decrease in the total white blood cell count. These findings align with the results from Aboderin *et al*., (2017), who similarly noted a decreased total white blood cell count following *Launaea taraxacifolia* administration. This decline in white blood cell count might be attributed to the anticancer properties of *Launaea taraxacifolia*, as substances with such properties are known to potentially inhibit the bone marrow's ability to generate white blood cells, as indicated by (Thomford *et al*., 2015). This study revealed that benzene and cyclophosphamide administration led to a significant increase in platelet count. This increase might be a compensatory mechanism triggered by benzene-induced haematotoxicity. Also, the activation of platelet production could be linked to the body's response to inflammation and tissue damage caused by both benzene and cyclophosphamide. This increase in platelet count is however in contrast to the study of Wang *et al*., (2016) who noted a decrease in platelet count in rats after benzene administration. Similarly, the increase in platelet count after cyclophosphamide

administration is in contrast to the study of Ukpo *et al*., (2019) who noted a decrease in platelet count in rats after cyclophosphamide administration. The administration of 100mg/kg, 200mg/kg, and 400mg/kg doses of *Launaea taraxacifolia* with benzene did not lead to any significant change to platelet count. It's possible that the doses used in the study, ranging from 100mg/kg to 400mg/kg, might not have been sufficient to elicit significant changes in platelet count when combined with benzene exposure. Dose-response relationships are often nonlinear, and different biological effects can be observed at varying concentrations. ICAM-1 (Intercellular Adhesion Molecule-1) is a gene that encodes a cell surface protein involved in mediating cell adhesion and inflammation (Roebuck and Finnegan, 1999). In this study, it was observed that benzene administration led to a reduction in the expression of the ICAM-1 gene. Decreased expression of ICAM-1 might indicate a potential suppression of immune responses or reduced ability of cells to adhere and communicate with each other (Yusuf *et al*., 2022). However, the subsequent administration of different doses of *Launaea taraxacifolia* with benzene appeared to counteract the benzene-induced reduction in ICAM-1 gene expression. The increase in ICAM-1 gene expression suggests that *Launaea taraxacifolia*, at these specific doses, might possess properties that modulate immune responses or cellular adhesion mechanisms. This counteractive effect could be due to various bioactive compounds present in *Launaea taraxacifolia*, which could have protective or regulatory effects on cellular processes, including gene expression (Bello *et al*., 2018). VCAM-1 (Vascular Cell Adhesion Molecule-1) is a gene responsible for encoding a cell surface protein involved in mediating the adhesion of immune cells to blood vessel walls (Pae *et al*., 2006). An increase in VCAM-1 gene expression is often linked to inflammatory responses and the recruitment of immune cells to sites of inflammation (Mabon *et al*., 2000). In this study, benzene administration led to an increase in the expression of the VCAM-1 gene. However, the subsequent administration of *Launaea taraxacifolia* at various doses appeared to counteract the heightened VCAM-1 gene expression induced by benzene exposure. This reduction in VCAM-1 gene expression could indicate that *Launaea taraxacifolia*, at these specific doses, possesses properties that modulate inflammatory processes or immune cell adhesion mechanisms. The bioactive compounds present in *Launaea taraxacifolia* might exert protective or regulatory effects on these cellular processes, leading to the observed decrease in VCAM-1 gene expression.

Conclusion: Data from this study has shown that benzene administration led to a reduction in the expression of the ICAM-1 gene. However, subsequent administration of different doses of *Launaea taraxacifolia* appeared to counteract the benzene-induced reduction in ICAM-1 gene expression. Benzene administration also led to an increase in the expression of the VCAM-1 gene. However, the subsequent administration of *Launaea taraxacifolia* at various doses appeared to counteract the heightened VCAM-1 gene expression induced by benzene exposure.

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