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Original Article

Effect of Curcumin and *Dioscorea alata* L. On Aniline Induced Spleen Toxicity in Rats

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ABSTRACT

Background: In this investigation, the consequences of standardized ethanolic extracts of curcumin and *Dioscorea alata* L. To assess their therapeutic potential individually and together in spleen poisoning caused by aniline in rats, focusing on hematological, physiological parameters, serum parameters, antioxidant parameter, and histopathological changes over a 30-day period. **Materials and Methods:** The investigation included six Wistar rats of both sexes per group, weighing between 200-270 g. For 30 days, Water with 100 ppm of AH was administered to rats. To cause splenic damage. The treatment groups were given aniline with Curcumin and *Disocorea alata* alone and in combination (75 mg/kg/day, orally). Physiological parameters, Hematological parameters, tissue oxidative parameters (GSH, LPO, NO) and serum parameters total protein, total iron were assessed to evaluate treatment effects. **Results:** Significant Progressions were observed in rats treated with combined Curcumin and *Dioscorea alata* therapy, these included decrease in body weight of AH-treated rats. Improvements in overall amounts of iron and protein, noteworthy decreases in oxidative stress, significant increases in organ weight of spleen and liver, Hematological analysis showed significant decrease in WBC count, Erythrocyte count and hemoglobin showed significant increase, the spleen always showed notable histological alterations, decreased severity of hemorrhages and aggregation of RBCs in the red and white pulp of spleen tissue. In addition to showing hepatomegaly toxicity, these findings demonstrate a strong link between the combined potential of curcumin and *Dioscorea alata* in recovering aniline exposure-related splenic lesions and erythrocyte destruction. **Conclusion:** The study shows the remarkable combined efficacy of curcumin and *Dioscorea alata* in managing spleen toxicity resulting from exposure to aniline hydrochloride.

Keywords: Curcumin, *Dioscorea alata*, Spleen toxicity, oxidative stress, hematological parameters.

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INTRODUCTION

The chemical industry has made substantial use of aniline, a poisonous aromatic amine, to produce pigments and resinscompounds.[1] Aniline exposure over an extended period of time causes fibrosis, hyperpigmentation, hyperplasia, splenomegaly, and elevated erythropoietic activity.[2-4] After ingesting or skin contact, aniline exposure can cause rapid (within 1-3 hr) manifestations of clinical symptoms.[3]

Previous research has demonstrated that exposure to aniline triggers oxidative and nitrosative stress as a result of excess iron and the start of lipid peroxidation. Excessive generation of radicals without bounds can harm nucleic acids and proteins, changing the structure and function of the spleen.[5] The hazardous aromatic amine aniline is produced in excess of 900 million pounds per year in the United States and is widely employed in industrial operations. The main harmful consequence of aniline causes production of Methemoglobin (MetHb), which reduces the blood's ability to transport oxygen.[6-8]

Aniline byproducts oxidize hemoglobin, leading to the production of MetHb. In rats, aniline-induced methemoglobinemia is mostly caused by phenylhydroxylamine.[9] *In vivo* aniline exposure causes increased dosages of splenic cancers, hemolysis, hemolyticanemia, and spleen damage.[5]

Curcuma longa contains curcumin, it is an essential part of the spice turmeric and is well-known for a variety of biological activities, including the presence of antioxidants.[10]

In cases where oxidative stress is a factor in a disease, natural compounds with antioxidant properties are becoming more and more important. The Dioscorea genus of vegetable includes *D. alata* L. (DA), also referred to as white, water, or winged yam. *Dwarf alfalfa* is primarily grown for its huge, highly carbohydrate-rich

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edible meat roots. The yams have sapogenins such diasogenin in addition to steroid saponins. Dioscorea species have long been utilized to cure ailments like rheumatism, diabetes, diarrhoea, and skin problems. They are also used as a tonic for conditions pertaining to the stomach, lungs, kidneys, and spleen.[11] Rats were exposed to aniline, which caused spleen poisoning. Curcumin and *Dioscorea alata* were chosen for this investigation because of their strong antioxidant activity and historical applications.

Curcumin has shown promising anti-inflammatory, antioxidant, and anticancer effects, but the exact ways it works in the body are not fully clear. Studies suggest that it affects several key pathways, such as NF-κB, MAPK, PI3K/Akt, and JAK/STAT, which are involved in inflammation, cell growth, and survival.

Because curcumin works on many targets and has low bioavailability, its effects can differ across diseases and tissues. Therefore, more research using advanced tools like omics technologies and CRISPR is needed to better understand how it works. These studies can help identify curcumin's main targets and improve its effectiveness in future therapies.

Dioscorea alata (water yam) is known for its health benefits, including antioxidant, anti-inflammatory, and antidiabetic effects. These benefits are linked to compounds like flavonoids, phenolics, anthocyanins, and diosgenin. However, the exact ways these compounds work in the body are not fully understood.

More studies are needed to explore how these bioactive compounds affect key pathways such as NF-κB, AMPK, and insulin signaling. Advanced tools like cell studies, animal models, and omics technologies can help identify how *D. alata* works at the molecular level. This research will support its use in medicine and functional foods.

MATERIALS AND METHODS

Materials

Chemicals

Standardized extracts of *Dioscorea alata* and curcumin were purchased with an analytical certificate from Kashipra Biotech Pvt. Ltd., in Indore, India and aniline hydrochloride (AH) from Otto Chemie Pvt. Ltd., Mumbai. All substances utilized in the investigation were all of analytical grade.

Standardization of Curcumin and *Dioscorea alata* L.

Standardize curcumin obtain form *Curcuma longa* (turmeric) with Greater than or equals to 95% curcuminoids (curcumin, desmethoxycurcumin, bisdemethoxycurcumin) with the help of HPLC, UV-vis, MS/NMR techniques

Standardize *Dioscorea alata* obtain from Tuber of *Dioscorea alata* (purple yam) which contain Anthocyanins (e.g., cyanidin, peonidin) - antioxidants/pigments Diosgenin - steroidal saponins TPC - overall antioxidant content. It can obtain with the help of HPLC/UV-Vis/LC-MS/MS.

Characterization of Curcumin and *Dioscorea alata* L.

Curcumin is a natural polyphenolic compound derived from the rhizomes of *Curcuma longa* (turmeric). It is the principal bioactive component responsible for turmeric's yellow colour and many of its medicinal properties. Chemically, curcumin is a diarylheptanoid with the formula C12H20O6.

Dioscorea alata L. Commonly known as water yam or purple yam, is a high-yielding, tuberous crop widely cultivated in tropical and subtropical regions. It is characterized by large, starchy tubers that vary in flesh color from white to deep purple. The plant has winged vines, heart-shaped leaves, and is typically dioecious.

Animals

Wistar rats (200-270 g) were used for induction of aniline toxicity, animals were procured from LacsmiBioform Pvt. Ltd., Pune, under the rats were kept in ordinary polypropylene cages at regulated temperatures 22°C±2°C, humidity at 55±5%, and 12 hr of darkness and light. They were fed a typical diet and had unrestricted access to water, adhering strictly to CCSEA 1277.

Methods

Experimental protocol

The animals that had fasted for the entire night were split up into the following six groups (n=6):

Group I: 1% CMC (1 mL/kg, p.o.).

Group II: AH (100ppm in d.w., for 30 days).

Group III: AH (100ppm in d.w., for 30 days.) + Curcumin (75 mg/kg/p.o) for 30 days.

Group IV: AH (100ppm in d.w., for 30 days.) + DA (75 mg/kg/p.o) for 30 days.

Group V: AH (100ppm in d.w., for 30 days.) with curcumin and DA (75 mg/kg/p. o) for 30 days.

Computational studies

Rodents' oral toxicity can be predicted using the web server ProTox. Toxic fragment identification is incorporated into the prediction approach, which is based on the comparison of substances with known median lethal doses (LD_{50}).[12]

Biochemical evaluation

Comprehensive measures such as total body weight, weight of the liver and spleen, amount of Feed and water consumption were monitoredduring the experiment and ultimately assessed.

Upon completion of the therapy, a glass capillary was used to draw blood from the retroorbital plexus. As the residual blood

was utilized to create the serumfor hematological investigation. The liver and spleen,were promptly taken out, blotted and weighed. Using a hematologyanalyzer (Beckman Coulter, USA), hematological parameters, such as platelet count, WBC count, differential cell count, hematocrit (HCT), hemoglobin, Packed Cell Volume (PCV), Mean Corpuscular Volume (MCV), hemoglobin content, and erythrocyte countwere measured.[13] Serum samples were utilized to calculate the protein and iron contents.[14]

Evaluation of tissue characteristics

Tissue homogenization

The liver and spleen were promptly placed in ice-cold Tris HCl buffered saline (pH 7.4) after the animals were put to sleep. The samples were weighed on a balance after being blotted clear of tissue fluids and blood. The organs were swiftly blotted on filter paper after being cross-sliced into thin slices using a surgical blade and hung in a cooled 0.25 M sucrose solution. After that, the tissues were chopped into small pieces and homogenized to a 10% w/v concentration in cooled tris hydrochloride buffer (10 mM, pH 7.4). In order to liberate soluble proteins, prolonged homogenization under hypotonic circumstances was intended to cause as much disruption to the cell's structure as feasible. A Remi C-24 high speed cooling centrifuge was used to centrifuge the homogenate for 15 min at 10,000 rpm and 0°C. Reduced glutathione was utilized to measure antioxidant activity, and the clear supernatant was used to measure lipid peroxidation.[15]

Assessment of lipid peroxidation

After combining 2.0 mL of freshly made 10% w/v Trichloroacetic Acid (TCA) with 2 mL of tissue homogenate supernatant, the mixture was incubated for 15 min in an ice bath. After incubation, 2.0 mL of the clear supernatant and 2.0 mL of freshly made Thiobarbituric Acid (TBA) were mixed after the precipitate was separated by centrifugation at 2,000 rpm for 10 min. The resultant combination was immediately cooled in an ice bath for 5 min after being cooked for 10 min in a boiling water bath. At 532 nm, the produced color was assessed in relation to a blank for the reagent. Standard malondialdehyde (MDA) solutions were made from 1,1,3,3-tetraethoxypropane at various concentrations (0-23 nM) and treated in a similar manner to produce a standard curve. According to Slater and Sawyer,[16] the findings are presented as nM of MDA per milligram of tissue.

Assessment of GSH content

20% TCA and equal quantities of tissue homogenate supernatant were mixed together. Centrifugation was used to separate the precipitate, and 0.25 mL of the supernatant was mixed with 2 mL of DTNB reagent. Phosphate buffer was used to get the total volume down to 3 mL. At 412 nm, the color developed was quantified in relation to a blank for the reagent. Using standard glutathione purchased from Sigma Chemicals in St. Louis, Missouri, the United States, a standard curve was created. The Lowry technique with Folin's phenol reagent was used to assess the protein level of the spleen. Micrograms of reduced glutathione (GSH) per milligram of protein is the unit of measurement for this amount of GSH.[17]

Evaluation of the levels of nitric oxide (NO)

After adding 1 mL of Griess reagent to 1 mL of tissue homogenate, the mixture was incubated at 37°C for 15 min. A Griess reagent blank was used to read the absorbance at 540 nm. The standard was a solution of sodium nitrite. The resulting standard curves were used to determine the quantity of nitrite present in the samples.[18]

Necropsy

The rats were euthanized by decapitation, and their spleens underwent microscopic histopathological inspection after being removed and stored in 10% neutral buffered formalin. Hematoxylin and eosin was used to stain the tissues after they had been sectioned, embedded in paraffin, and seen under a microscope.[13]

Statistical analysis

One-way ANOVA followed by the Bonferroni *post hoc* test for intergroup comparisons. Results are presented as Mean±SE. A noteworthy level of $p>0.05$ denoted no significant distinction, while $p<0.05$ and $p<0.01$ indicated significant and highly significant differences, respectively.

RESULTS

Computational study

According to Figure 1, LD_{50} value of 250 mg/kg, the ProTox results also demonstrated that aniline had good oral absorption. Aniline has a 200-300 mg/kg experimental LD_{50} value in rats. According to the globally standardized method of classification of chemical labelling based on LD_{50}, aniline is categorized as a toxicity class 1 substance.

Effects of Curcumin and DA both independently and in conjunction on body weight, organ weight, feed consumption and water intake

When compared to the control group, the AH-treated group's body weight, water intake, and feed consumption were found to have changed somewhat by the end of the thirty-day period, as seen in (Table 1, Figure 2). When compared to the rats treated with AH, the rats given a 30-day chronic administration of Curcumin and DA (75 mg/kg, po) both alone and in combination showed a substantial increase in body weight, water intake, and feed consumption.

The liver and spleen weights were noted in Table 1 on the thirty-first day of the assessment. Compared to the rats in the

control group, the animals treated with AH had larger liver and spleen weights. In comparison to the groups treated with aniline, the weights of the spleen and liver were recovered after 30 days of chronic therapy with Curcumin and DA, both alone and in combination (75 mg/kg).

Effect of Curcumin and DA alone and in combination on hematology parameters

Aniline-treated rats displayed a notable rise in WBC count and reduction in RBC level when contrast to normal animals. Rats treated with aniline also had substantially lower haemoglobin levels. In contrast to the rats given aniline, the rats receiving chronic treatment for 30 days with 75 mg/kg of both Curcumin and DA alone and in combination show notable improvements in haemoglobin levels, WBC count, and in RBC count shown in (Table 2, Figure 3). Among other parameters, PCV, MCV, MCH content, HCT, platelets Analysis revealed no appreciable difference from rats treated with AH.

Effect of Curcumin and DA both independently and in conjunctionon LPO, GSH and NO levels

The levels of endogenous antioxidants, such as LPO and GSH, were measured in spleen tissue homogenates, along with NO levels (Table 3, Figure 4). In the aniline-treated rats, LPO levels were significantly increased in both spleen tissue compared to the control group indicating high oxidative stress due to toxicity, while GSH amounts were markedly reduced, while NO levels were also markedly raised. Chronic treatment for 30 days with Curcumin and DA, either individually or in combination (75 mg/kg), resulted in a noteworthy rise in GSH and NO levels and a noteworthy drop in LPO levels in comparison to the aniline-treated group.

Histopathological Examination

As shown in Figure 5, normal histomorphological features of the red pulp and white pulp are evident in normal control rats. Histological only the spleen experienced alterations as a result of AH therapy at all-time points, the spleen in the AH-treated rats exhibited notable histopathological changes. These included moderate pathological changes in the splenic parenchyma, with multifocal areas of hemorrhages and aggregation of RBCs in both the red and white pulp. There was also distortion of the red and white pulp, with a loss of demarcation due to the mixing of RBCs and lymphocytes in the splenic parenchyma. Curcumin and DA, either individually or in combination (75 mg/kg), shows less multifocal areas of hemorrhages and aggregation of RBCs in the red and white pulp. ascompareto AH treated animal.

Table 1: Impact of Curcumin and DA both independently and in conjunction on Body weight, Spleen weight (g) and Liver weight (g).

Group	Body weight(g)	Feed Consumption (g)	Water intake (mL)	Spleen weight (g)	Liver weight (g)
NC	260±2.40	17.66±0.60	39 ±0.57	0.60±0.01	6.25±0.35
DC	220±4.41$^{###}$	11.83±0.16$^{###}$	16±2.72$^{###}$	1.34±0.20$^{###}$	10.95±0.11$^{###}$
CU	245±6.65***	14.5±0.50***	21±0.57**	0.94±0.05***	5.67±0.10***
DA	251±8.14***	15.66±0.60***	23±2.08***	0.62±0.05***	6.77±0.13***
CU+DA	259±4.48$^{***, aa}$	15.19±0.28***	24±2.84***	0.70±0.01$^{***, aa}$	5.93±0.20$^{***, bbb}$

[NC- Normal control, DC- Disease control, CU-Curcumin, DA-*Dioscorea alata*, CU + DA- Curcumin + *Dioscorea alata*. Data presented as Mean±SD, (n=6). One Way ANOVA followed by Bonferroni test for multiple comparison. $^{###}p$<0.001 when contrast to normal control. $^{***}p$<0.001, $^{**}p$<0.01 when contrast to disease control, ^{aa}p<0.01 when in contrast to CU group. $^{###}p$<0.001 when contrast to normal control, $^{***}p$<0.001 when contrastto disease control, ^{aa}p<0.01 when contrast to CU group, ^{bbb}p<0.001 when contrast to DA group].

Table 2: Impact of Curcumin and DA both independently and in conjunction on hematology.

Parameters	NC	DC	CU	DA	CU+DA
Hb(g/L)	14.13±0.48	9.86±0.52$^{##}$	13.0±0.20***	13.53±0.17***	14.13±0.24$^{***, aaa}$
RBCs(10^6/µL)	9.4±0.11	6.8±0.24$^{###}$	8.8±0.24***	9.3±0.41***	9.4±0.23$^{***, a}$
WBCs(x10^3/µl)	8.4±0.30	14.8±0.30$^{###}$	10.4±0.35***	8.09±0.46***	9.87±0.57$^{***, bbb}$
PLT(x10^6/mL)	552.0±243.3	597.2±265.2	478.7±265.2	458.2±165.7	554.4±285.5
HCT (%)	40.7±1.80	41.95±1.80	40.3±1.80	42.5±1.80	47.5±1.80
MCV (fl)	61.1±1.38	53.90±1.38	56.8±1.38	58.2±1.38	53.4±1.38
MCH (pg)	19.2±64	18.66±94	19.0±79	18.3±89	17.5±84
Lymphocytes (%)	43.05±4.14	42.07±2.76	41.03±4.1	40.05±3.14	44.2±2.63
Neutrophils (%)	65.30±4.58	52.60±3.19	55.13±2.1	53.60±5.19	52.60±3.17
Monocytes (%)	4.30±0.26	4.50±0.26	4.56±0.54	4.10±0.76	5.30±0.66

[NC- Normal control, DC- Disease control, CU-Curcumin, DA-*Dioscorea alata*, CU + DA- Curcumin + *Dioscorea alata*. Data presented as Mean±SD, (n=6). One Way ANOVA followed by Bonferroni test for multiple comparison. $^{###}p$<0.001 when compared to normal control, $^{***}p$<0.001 when compared to disease control, ^{aaa}p<0.001 and ^{a}p<0.5 when compared to CU group, ^{bbb}p<0.001 when compared to DA group].

Table 3: Curcumin and DA alone and in combination their effects on the LPO, GSH, and NO levels in spleen tissue.

Group	Spleen Tissue		
	LPO Level (MDA/mg)	GSH Level (mg/g protein)	NO Level(nmol/L)
NC	27.05±0.14	79.10±7.76	51.47±40.46
DC	77.13±1.26$^{###}$	44.57±6.38$^{###}$	119.1±5.57$^{###}$
CU	41.33±0.24***	50±3.97***	71.79±4.79**
DA	26.09±12.5***	80±14.81***	50.02±8.26***
CU+DA	51.89±13.5$^{***, bbb}$	71.06±0.74***	47.9±4.70***

[NC- Normal control, DC- Disease control, CU-Curcumin, DA-*Dioscorea alata*, CU + DA- Curcumin + *Dioscorea alata*. Data presented as mean± SD, (n=6). One Way ANOVA followed by Bonferroni test for multiple comparison. a) $^{###}p<0.001$ significantly different when compared to normal control, $^{***}p<0.001$ significantly different when compared to disease control, $^{bbb}p<0.001$ significantly different when compared to DA group].

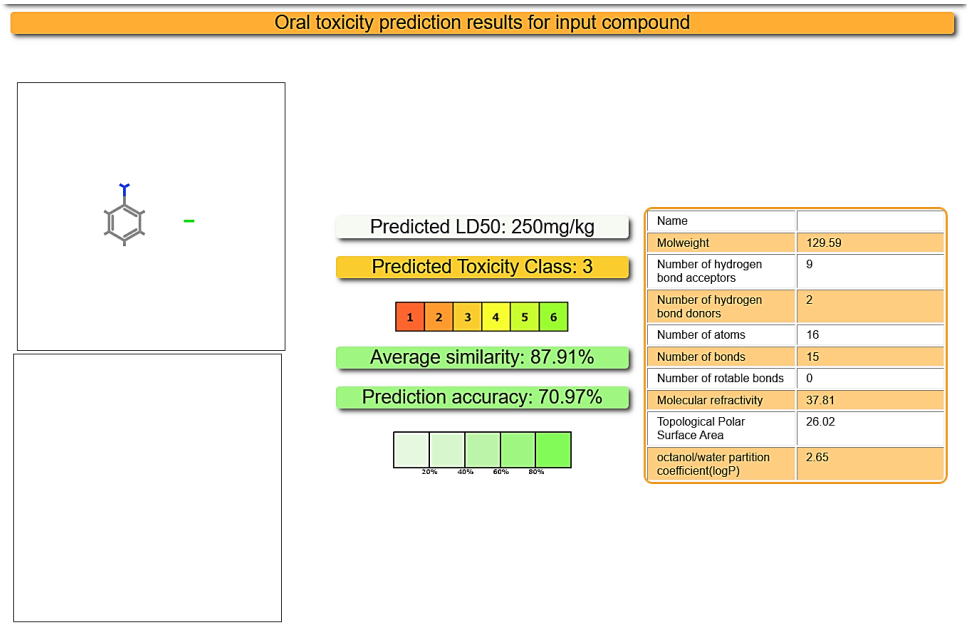

Figure 1: Prediction of aniline-induced oral toxicity (ProTox).

DISCUSSION

The spleen and erythrocytes have been linked to aniline-induced toxicity in a various investigation.[5,8] These investigations, however, did not look into the frequency or connection between splenic lesions and AH-related alterations in blood chemistry. The results of this investigation show that the spleen and erythrocytes are the main organs that aniline toxicity affects. The evolution of splenic lesions appears to be dependent on the period of exposure, and they suggest that the compound's initial contact with erythrocytes and the spleen's scavenging of them might be important elements in their growth. Rats that are exposed to aniline and its substitutes develop a specific toxicity of the spleen. Research has revealed that rats exposed to aniline had much higher total iron content and oxidative stress.[2,19] There is a correlation between the extent of splenotoxicity and erythrocyte destruction in rats exposed to AH. In this study, Splenic toxicity resulted from a long-term consumption of AH (100 ppm) through drinking water. The assessment of hemoglobin levels verified the toxicity to the spleen, with a significant decrease observed by the 30th day. Additionally, notable drops in body weight, feed intake, and water intake in rats given aniline treatment were noted, likely due to aniline's toxicity reducing food consumption, directly correlating to the decreased body weight and water intake. The study's primary discovery was the rise in liver and spleen weights (splenomegaly) in rats given AH. In this study, the treatments reversed the effects of AH therapy on body weight, feed consumption, and water intake. The changes in these broad parameters show that curcumin and DA, when taken separately and together, are effective in preventing AH toxicity. The excessive deposition of erythrocytes modified with phenylhydroxylamine (PHA) was found to be the cause of changes in hemoglobin, RBC, and WBC levels.[20] Overproduction of oxidative and nitrosative stress may be the cause of the change in WBC count. These blood parameter alterations are consistent with previous studies on aniline and its derivative.[5] Treatment with Curcumin and DA individually and in combination significantly improved hemoglobin levels and RBC and WBC counts, likely due to powerful anti-oxidant and anti-free radical capabilities. Rats given AH had a much higher iron burden and lower protein content in this investigation. As a modulator of AH-induced splenotoxicity, iron is essential.[2,3] Treatment with aniline causes a large amount of iron to accumulate, which in turn catalyses

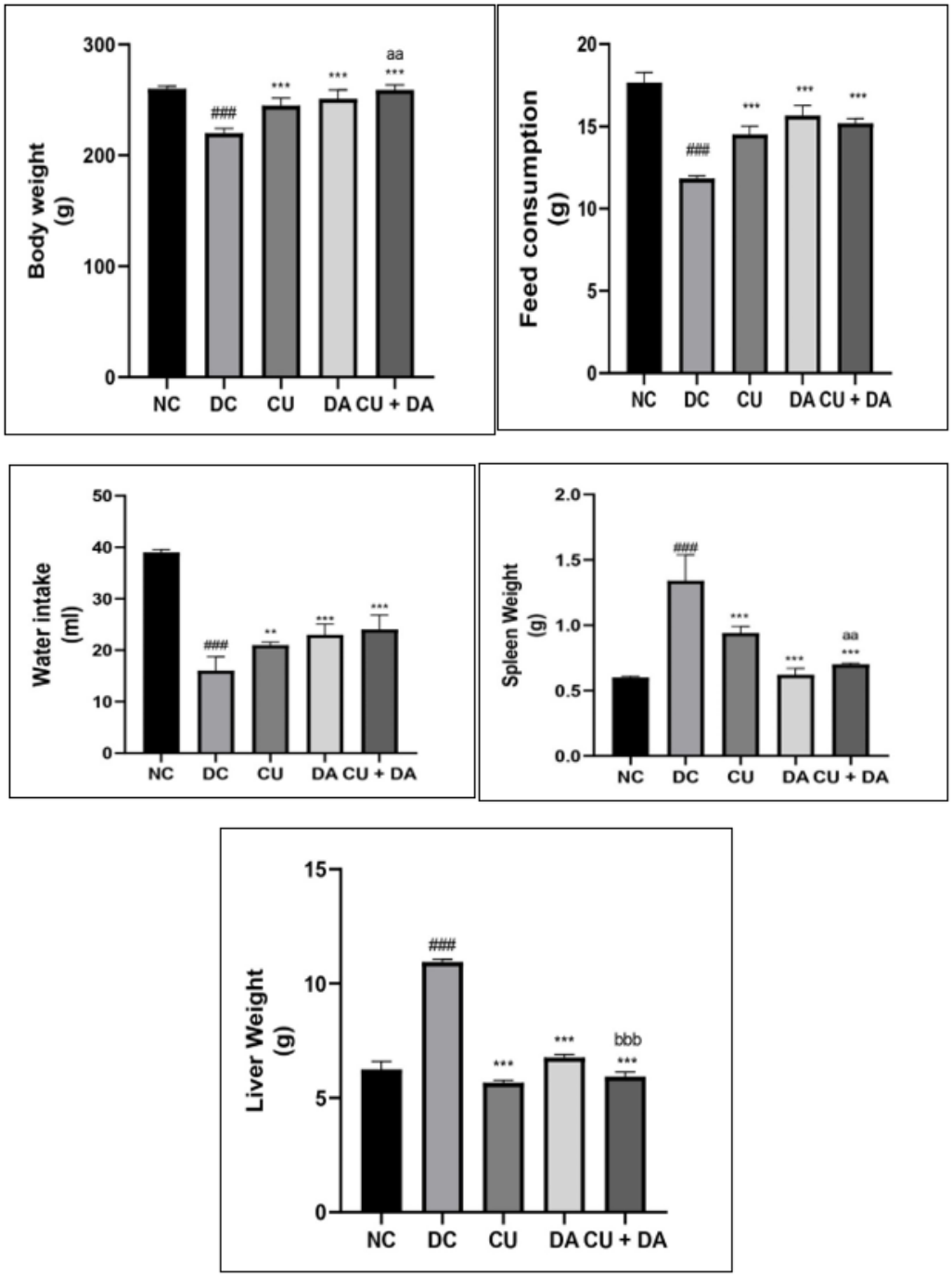

Figure 2: Impact of Curcumin and DA alone and in combination on Body weight, feed consumption,water intake, Spleen weight (g) and Liver weight (g).

the creation of excessive ROS, which in turn damages proteins, nucleic acids, and lipids, ultimately resulting in malfunction of the cells.[20] The spleen experiences oxidative stress due to LPO and iron overload as a result of this exposure. The two most important early metabolic processes in AH-induced splenic damage are LPO and protein oxidation. This investigation assessed oxidative stress indicators. The spleen and liver of the AH-induced group had significantly lower GSH levels and significantly higher LPO and NO levels. Due to the fact that aniline causes both protein and lipid oxidation, oxidative stress is a major factor in AH-induced splenic damage.[20] This implies that aniline-induced splenic damage involves oxidative stress. AH-treatment led to noticeably greater amounts of LPO and reduced levels of GSH, suggesting that lipid peroxidation changes the function of proteins and contributes to splenic toxicity. Reactive oxygen species production brought on by iron accumulation in the spleen may harm cellular constituents. Curcumin and DA, both alone and in combination, exhibit diverse biological activities. Treatment with these compounds attenuated splenic toxicity

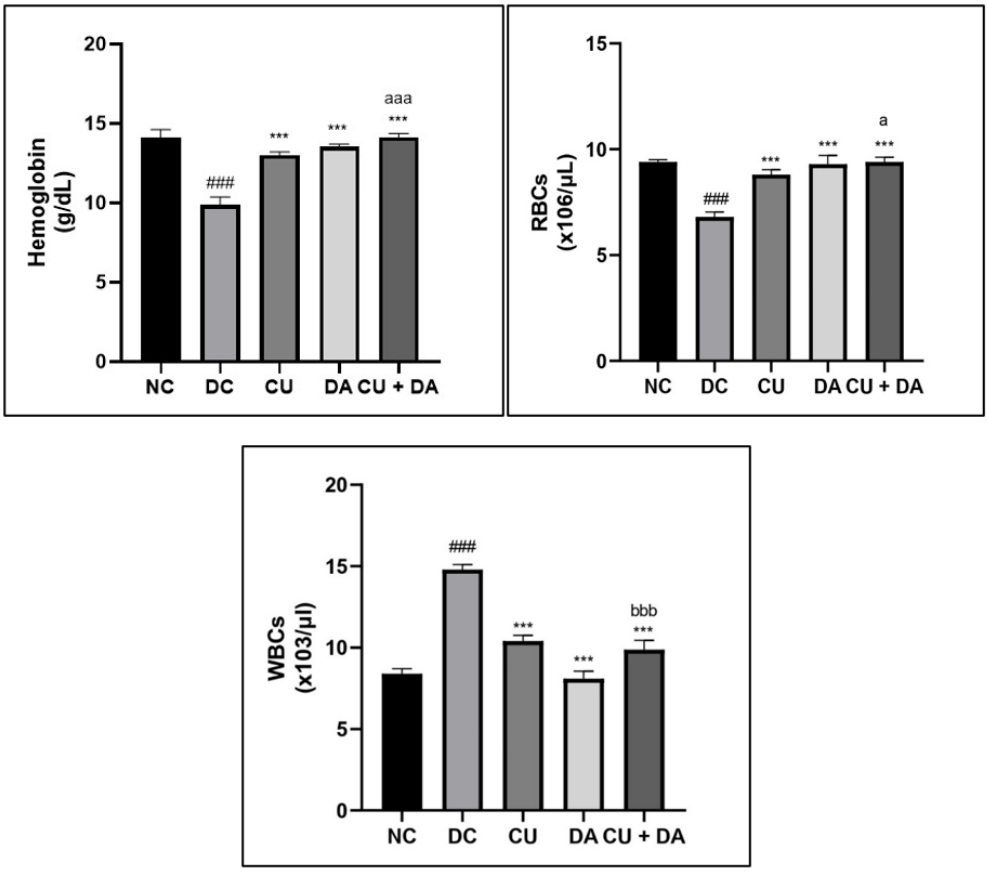

Figure 3: Impact of Curcumin and DA both independently and in conjunction on Hb,RBC,WBC.

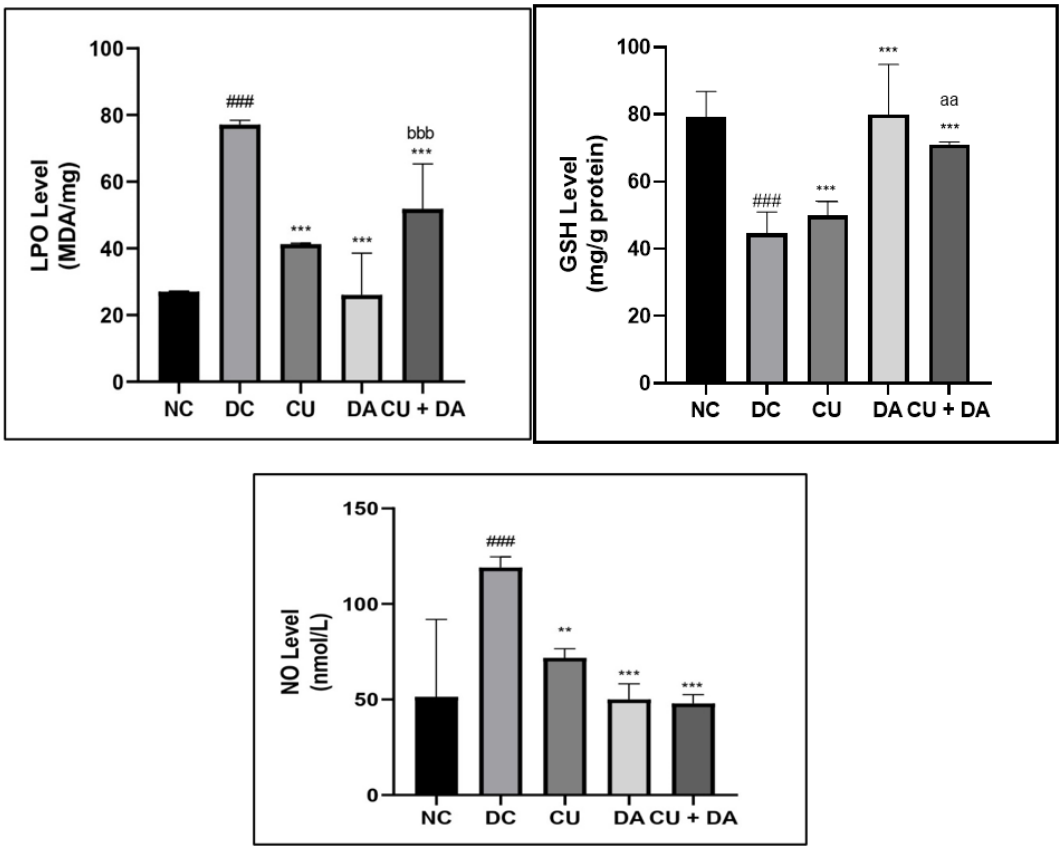

Figure 4: Result of Curcumin and DA both independently and in conjunction on Spleen tissue LPO, GSH and NO level.

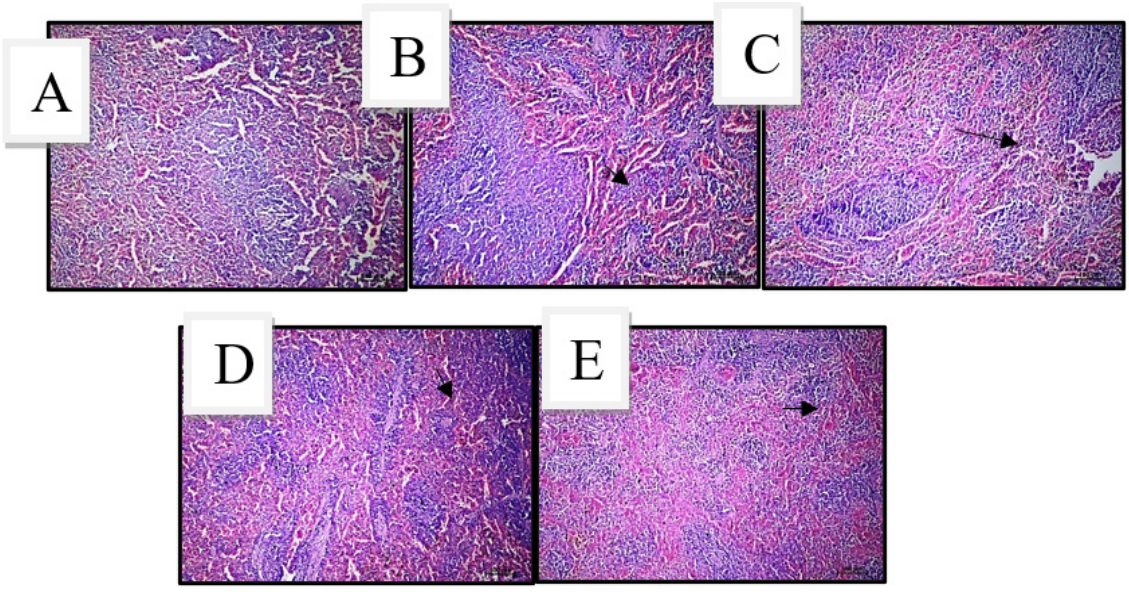

Figure 5: The control image showed the fine histomorphological characteristics of white and red pulp. Moderate pathological changes of splenic parenchyma with multifocal areas of hemorrhages in RBC. Mild pathological changes of splenic parenchyma with multifocal areas of hemorrhages and aggregation of RBCs in the red and white pulp was observed on treated groups.

caused by aniline, likely due to their ability to inhibit ROS and the strong free radical scavenging they do.[21,22]

Curcumin and *Dioscorea alata* (water yam) possess multiple bioactive compounds with potential applications in human health. Curcumin has shown strong anti-inflammatory, antioxidant, anticancer, and neuroprotective effects, making it useful in managing chronic conditions such as arthritis, cancer, and neurodegenerative diseases. *Dioscorea alata* contains phenolics, flavonoids, and diosgenin, which contribute to its antioxidant, antidiabetic, and hormone-regulating properties. Both can serve as complementary agents in existing therapies and may reduce drug side effects. Advances in formulation, like nanoparticles and functional foods, are improving their bioavailability and supporting their potential use as natural therapeutics.

Curcumin has low bioavailability, which limits its absorption and effectiveness. Advanced formulations like nanoparticles and liposomes are being developed to overcome this. *D. alata* lacks detailed clinical data, and the exact molecular mechanisms of its effects in humans are still unclear. More human trials are needed. Standardized doses, safety profiles, and potential interactions with medications remain areas requiring further research for both.

CONCLUSION

This project work was aimed to evaluate effect of AH in test subjects. These findings suggest that curcumin and *Dioscorea alata* L. offer significant effects against aniline-induced spleen toxicity in rats. Their ability to reduce oxidative stress, enhance antioxidant defensemechanisms, improvehematology action and suppress inflammation highlights their therapeutic potential in reducing the adverse effects of aniline toxicity. Curcumin and *Dioscorea alata* L. could be considered as effective natural agents for protecting the spleen from chemical-induced damage, potentially contributing to the development of new treatments for industrial toxic exposure. Further research may explore their mechanisms of action and potential applications in human health. Research has shown that prolonged exposure to AH raises the production of free radicals and splenic toxicity in test animals. The design of experiment was made for evaluation of antioxidant and Curcumin and *DA* alone and in combination therapeutic strategy for managing spleen toxicity in experimental wistar rats. Curcumin and *Dioscorea alata* have shown promising health benefits, including anti-inflammatory, antioxidant, and disease-fighting effects. Curcumin may help with conditions like cancer, diabetes, and brain disorders, while *D. alata* may support blood sugar control and overall health.

However, curcumin's low absorption and the lack of detailed studies on *D. alata* limit their current use. Future research should focus on improving delivery methods, understanding how they work in the body, and confirming their effects in human studies.

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ABBREVIATIONS

NO: Nitric oxide; **LPO:** Lipid peroxidation; **GSH:** Glutathione; **MDA:** Malondialdehyde; **AH:** Aniline hydrochloride; **CU:** Curcumin **DA:** *Dioscorea alata*.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ETHICAL APPROVAL AND CONSENT TO PARTICIPATE

Rats were procured from LacsmiBiofarms Pvt. Ltd., Pune (Registration No: CPCSEA Reg No.1277). Approval was taken from the Institutional Animal Ethics Committee of KBHSS trusts institute of pharmacy Malegaon (IAEC registration no. 1566/PO/Re/S/11/CPCSEA).

SUMMARY

This study evaluated the effects of curcumin and *Dioscorea alata* L. (DA) on aniline-induced spleen toxicity in rats. Findings indicate their significant ability to reduce oxidative stress, enhance antioxidant defenses, improve hematological parameters, and suppress inflammation. These natural agents show promise in protecting the spleen from chemical-induced damage, with potential applications in treating industrial toxic exposure. Further research is needed to understand their mechanisms and explore human health applications.

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