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Hematological and ultrastructural toxicity of first-line anti-tuberculosis drugs and the ameliorative effects of *Berberis aristata* in rats

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Abstract

Background Drug toxicity with respect to adverse drug reactions (ADRs) poses a substantial barrier to meeting global healthcare demands, impacting both the recovery of patients and the economic aspects of healthcare systems. Studies have consistently demonstrated the potential toxicity of the combination of anti-tubercular (anti-TB) drugs, such as isoniazid, rifampicin, and pyrazinamide.

Methodology In the present study, Wistar rats have been divided into four groups, namely the GI—control group, GII—the drugs group (combination of anti-TB drugs, i.e., isoniazid + rifampicin + pyrazinamide), GIII—*B. aristata* group, and GIV—treatment groups (anti-TB drugs + *B. aristata*) orally for 28 days.

Results The groups exposed to the combination of anti-TB drugs exhibited a significant decrease in hemoglobin concentration and total leukocyte count. In contrast, supplementation with *B. aristata* resulted in a significant restoration of physiological alterations compared to the drugs group. Scanning electron microscopy revealed significant alterations in the topography of erythrocytes, including acanthocytes, echinocytes, and stomatocytes, in the drugs group, which were observed to be mitigated in the *B. aristata* group.

Conclusion These findings underscore both the impact of anti-TB drugs and *B. aristata* as a supplement to counter the negative impact.

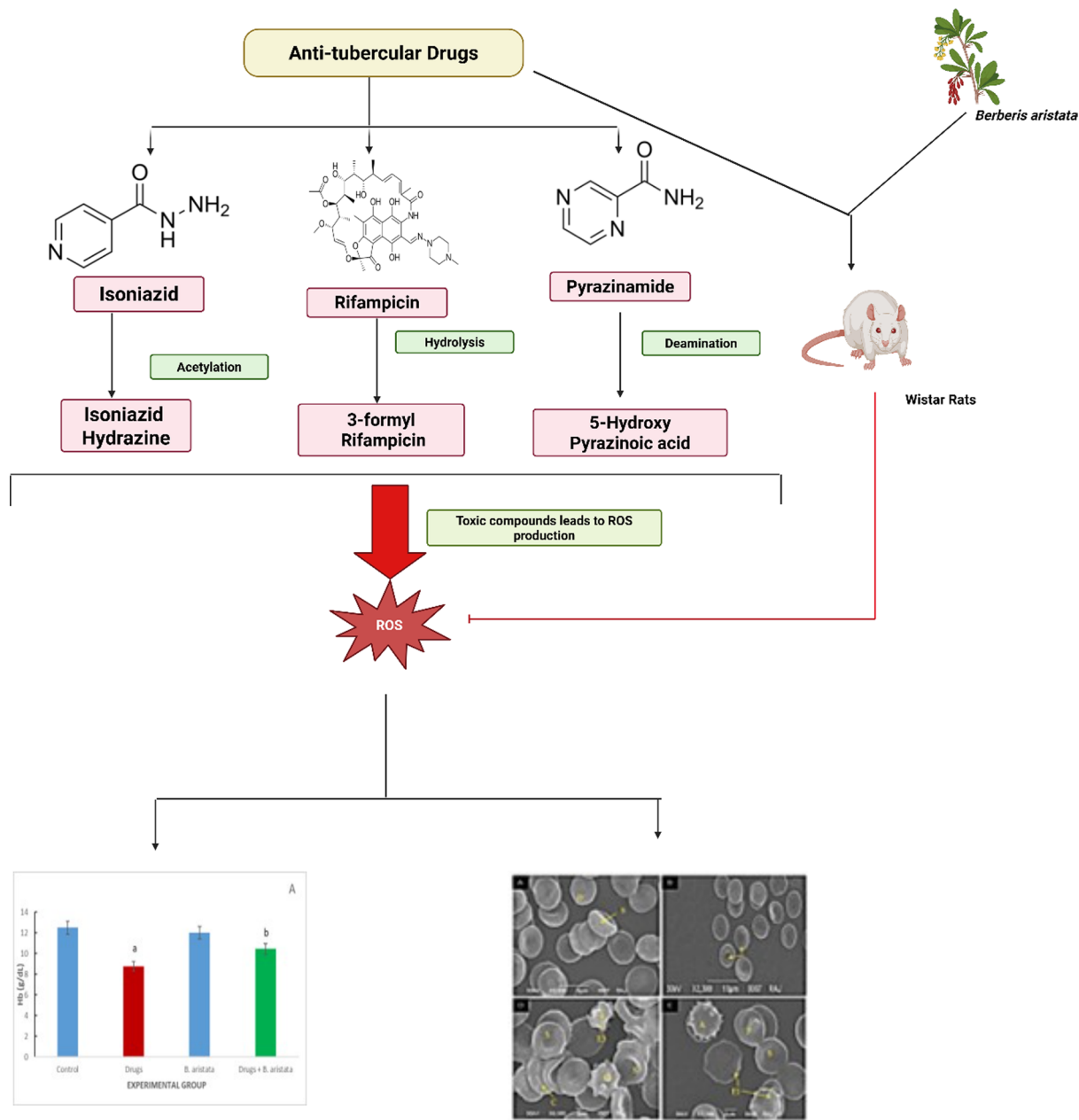
Keywords Anti-TB drugs, Blood, Hematological alterations, Toxicity

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Graphical Abstract



Introduction

Tuberculosis (TB) is caused by the bacterium *Mycobacterium tuberculosis*. It is considered a dreadful disease. According to the Global TB report, around 10.6 million cases were reported globally. The mortality rate reached 1.3 million in 2022 (WHO, 2007). India leads

with 27% of the reported cases (Global Tuberculosis Report, 2023).

TB is a chronic airborne disease, but it is preventable and curable upon treatment with combination chemotherapy. Combinational chemotherapy includes the administration of four first-line drugs: pyrazinamide

(PZA), isoniazid (INH), rifampicin (RIF), and streptomycin (SM) or ethambutol (EMB) (Konstantinos, 2010). In case of the development of resistance against first-line anti-TB drugs, second-line treatments, including capreomycin, kanamycin, cycloserine, and ethionamide, are prescribed for about 6 to 8 months. These drugs induce hematological alterations and can impact blood components, including red and white blood cells, platelets, and the coagulation system (Sweeney & Cummings, 2020). The metabolism of these drugs often leads to the formation of reactive metabolites, whose detoxification is challenging or decreases cellular defenses and can precipitate adverse drug reactions (ADRs) (Guengerich & MacDonald, 2018).

ADRs to anti-TB medications are common and often need extensive medical management. This can extend the duration and cost of treatment. ADRs may cause significant morbidity and even mortality. In some cases, they result in therapy discontinuation (Fei et al., 2018). Individuals with HIV and those born in Asia are particularly at risk due to immune and vitamin deficiency conditions (Yee et al., 2008). Life-threatening ADRs from anti-TB drugs include arrhythmias, hepatotoxicity, acute renal failure, lactic acidosis from antiretroviral therapy, hyperuricemia, and gastrointestinal bleeding (Granowitz & Brown, 2009; Shi et al., 2024). Previous studies in our laboratory demonstrated that combinations of these anti-tubercular drugs induce toxicity in the liver, kidney, and spleen. They also cause genotoxicity in Wistar rats when given for 28 days, as shown by various biochemical, hematological, and histopathological analyses (Sharma & Sharma, 2017; Sharma et al., 2018).

Berberis aristata (De Candolle), belonging to the family Berberidaceae, is known anti-haemorrhoidal, antidiabetic, lipid-lowering, antimicrobial, anti-inflammatory, and antioxidant properties (Marchelak et al., 2025; Sharma & Sharma, 2015; Sharma et al., 2019). These comprise phytoconstituent as isoquinoline alkaloids such as berberine, jatrorrhizine, aramine; flavonoids and phenolics such as quercetin, rutin; and tannins, glycosides, and terpenoids (Marchelak et al., 2025; Potdar et al., 2012).

Owing to the virtue of its phytoconstituents, these were hypothesized to alleviate the impact of drugs and subsequently ADRs (Abdel-Daim et al., 2018). Thus, the present study attempted to evaluate the different hematological alterations induced in Wistar rats due to the co-administration of first-line anti-TB drugs—INH, RIF, PZA, and *B. aristata* in the rat model.

Material and methods

Experimental animal model

Adult male Wistar albino rats ($n=24$) with an average weight of 150–200 g and an age of around 6–8 weeks were acquired from the Central Animal House, Panjab University, Chandigarh, India. The rats were housed in cages under sterile circumstances and permitted to acclimatize for 2 weeks to a new environment. The rats were maintained at a controlled temperature (25 ± 2 °C) under a 12-h light–dark cycle and relative humidity of 40–70%. Rats were fed with standard rodent pellets (Ashirwad Industries, Punjab, Hindustan Lever, India) and tap water ad libitum. The experiments were performed following the guidelines of the Institutional Animal Ethics Committee of Panjab University, Chandigarh (PU/IAEC/S/14/147).

Chemicals used

The chemicals used were sourced from HIMEDIA Ltd., CDH, and Chromous Biotech, Bangalore, India. Pure salts of first-line anti-TB drugs, namely isoniazid (CAS number-54–85-3), rifampicin (CAS number-13,292–46-1), and pyrazinamide (CAS number-98–96-4), were obtained from Lupin Pharmaceuticals Ltd., India. A quality assurance certificate of each drug was obtained from Lupin Pharmaceuticals Ltd.

Plant extract preparation

Berberis aristata was collected from the Kangra region of Himachal Pradesh, India. The plant was identified by the taxonomist in the Department of Botany, Panjab University, Chandigarh, with voucher no. 5197. Collected plants were shade-dried, pulverized, and processed for Soxhlet (ethanolic) extract. Ethanol was used for extracting *B. aristata* due to its high efficiency in solubilizing alkaloids like berberine and its safety in pharmacological applications. The HPLC protocol for qualitative and quantitative analysis of the extract was standardized by adapting and optimizing methods reported by Pasrija et al. (2010), Muralidharan et al. (2012), and Shigwan et al. (2013).

Preparation of dosages and their extrapolation to human equivalent dose

Dosages of the three first-line anti-TB drugs were determined according to the guidelines of the Food and Drug Administration (Centre for Drug Evaluation and Research, 2005). FDA has suggested that the extrapolation of animal dose to human dose can be correctly performed by normalization to body surface area (BSA), which is generally represented in mg/m^2 (Sharma et al., 2019). The human dose equivalent is calculated using the formula given below.

$$\text{HED (mg per kg b.wt.)} = \text{Animal doses} \\ (\text{mg per kg b.wt.}) \times \frac{\text{Animal } K_m}{\text{Human } K_m}$$

where

HED is the human equivalent dose; BSA is the body surface area; and K_m factor is the weight (kg)/BSA (m^2). For the change of human dose to rat dose, conversion factor of 6.17 was used (Shin et al., 2010) (Table 1).

The dose of *B. aristata* extract (300 mg/kg b.wt.) was selected based on previous in vivo studies (e.g., Sharma et al., 2019) where similar doses showed significant pharmacological effects without toxicity. Additionally, in-house acute toxicity studies confirmed an $\text{LD}_{50} > 5000$ mg/kg, classifying the extract as practically nontoxic according to OECD guidelines.

Experimental group

Wistar rats aged 6–8 weeks were randomly divided into four different groups of $n=6$ rats. GI (control group) was given saline; GII (Drugs group) was given anti-TB drugs combination, namely INH (30.85 mg/kg b.wt.) + RIF (61.7 mg/kg b.wt.) + PZA (132.65 mg/kg b.wt.); GIII (positive control) was supplemented with *B. aristata* (300 mg/kg b.wt.); and GIV (Treatment groups) was co-administered with anti-tubercular drugs and *B. aristata* (300 mg/kg b.wt.) orally for 28 days.

On the 29th day, blood samples from the jugular vein of rats were collected in anticoagulant tubes for the determination of total hemoglobin (Hb), total leukocyte count, differential leukocyte count, as well as for scanning electron microscopy studies.

Hematological studies

The hematological parameters studied were total leukocyte count (TLC), hemoglobin (Hb), and differential leukocyte count (DLC) in all the groups.

Hemoglobin (Hb) estimation

Sahli hemoglobinometer used to estimate Hb in blood. The tube of the haemoglobinometer was rinsed with distilled water and then with absolute alcohol. After air

drying, N/10 HCl was added in the tube with a dropper up to the 2-ml mark. Next, blood was sucked into the pipette up to 0.2-ml mark and added into the N/10 HCl of the hemoglobinometer tube and mixed with a stirrer. The sample was kept for 5–10 min, and after that, distilled water was added drop by drop. The point where the color of the mixture was similar to the color of the standard tube was noted as the endpoint value of Hb in g/dL.

Total leukocyte count (TLC)

The Neubauer hemocytometer chamber was used to calculate TLC. Blood was taken into the white bead pipette up to the 0.5-ml mark. It was mixed with Turk's fluid and was kept undisturbed. The coverslip was placed on the Neubauer slide, and blood from the pipette entered through the narrow opening on the slide under the coverslip after passing over 1–2 drops. The number of WBCs in four squares was calculated as below:

Say the number of WBC's in squares 1,3,7,9... is $a + b + c + d + \dots = y$.

Area of each square = 1 mm^2 , height = $1/10 \text{ mm}^3$

Therefore, volume = $1/10 \times 1 = 1/10 \text{ mm}^3$

$1/10 \text{ mm}^3$ contains = $y/4$ WBC's

1 mm^3 contains = $y/4 \times 10$ WBC's

As the solution is diluted 20 times,

Therefore, 1 mm^3 contains = $y/4 \times 10 \times 20 = z$ WBC's.

Differential leukocyte count (DLC)

A thin blood smear was air-dried, fixed in methanol, and stained with Giemsa. After this, the spreads were washed in running water, air-dried, and viewed under a microscope at magnification (100X) utilizing immersion oil. The percentage of WBCs was calculated as:

$$\frac{\text{Number of mononuclear/polymorphonuclear cells}}{\text{Total number of cells}} \times 100$$

Scanning electron microscopy of RBC

Phenotypic alterations in erythrocytes were analyzed through SEM (Kavita & Mittal, 2014). For this, blood samples were obtained from the tails of rats using an EDTA-coated microcentrifuge tube; 5 μl of blood was

Table 1 The drug dose given to the animals

	Drug	Dose equivalent in human (adult)-HED	Conversion factor in rat	Dose equivalent in rat (HED $\times$ CONV. FAC.)
1	Isoniazid (INH)	5 mg/kg b.wt	6.17	30.85 mg/kg b.wt
2	Rifampicin (RIF)	10 mg/kg b.wt		61.70 mg/kg b.wt
3	Pyrazinamide (PZA)	18.2–25.0 mg/kg b.wt. (21.6 mg/kg b.wt.)		132.65 mg/kg b.wt

added to 0.5 ml of gluteraldehyde–paraformaldehyde fixative and fixed for 2 h. Then the sample was centrifuged at 1000 rpm for 10 min, the supernatant was discarded, and the pellet was washed with phosphate buffer. A 2–3 μ l of erythrocytes was added over metallic tape stuck to the aluminum stub and dried under lamp light until RBCs were settled. Sputter coating with gold was done using the coating instrument JFC 1600 and viewed under the JSM JEOL 6100 scanning electron microscope (RSIC, CIL, Panjab University, Chandigarh).

The percentage of RBCs morphological alteration was determined by considering random sections with a total of 100 cells (Sinha & Jindal, 2019).

Statistical analysis

SPSS version 21 was utilized for all the analyses. The normality of the data was assessed using the Kolmogorov–Smirnov and Shapiro–Wilk test, which suggested that the data distribution was normal. All values were expressed as mean $\pm$ standard deviation (SD) of six animals per group. One-way ANOVA statistical test was used to determine whether different sets of data are significantly different from each other. Tukey post hoc test was used to make pair-wise comparisons. Values with $p \leq 0.05$ were considered statistically significant.

Results

Phytochemical analysis of *B. aristata*

HPLC of *B. aristata* displayed a retention time of 16.387 min with a regression equation of $y = 954.54x - 99,410$ ($R^2 = 0.994$) for berberine (active ingredient) (Figs. 1 and 2). Similar observations have been recorded by Pasrija et al. (2010), Muralidharan et al. (2012), and Shigwan et al. (2013).

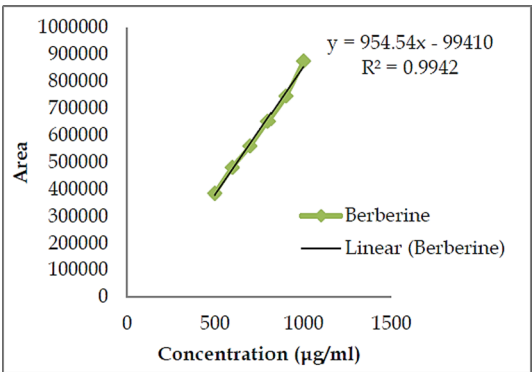


Fig. 2 Calibration curve of berberine in the mobile phase: potassium hydrogen phosphate (pH 2.5) and acetonitrile in the ratio of 80:20 (%v/v) by reverse phase high-performance liquid chromatography (RP-HPLC) method

Estimation of hemoglobin concentration

The hemoglobin concentration was observed to be 12.50 g/dL in the control (GI) group. The group administered with first-line anti-TB drugs (GII) showed a significant decrease in hemoglobin concentration compared to the control (GI) and *B. aristata* (GIII) group (Fig. 3A). A significant restoration of about 10% was observed in GIV compared to the drugs (GII) group (Fig. 3A).

Total leukocyte count (TLC)

The total leukocyte count was observed to be $24.5 \times 10^3 \text{ mm}^{-3}$ in the control (GI) group. There was a significant decrease in the total leukocytes in the drugs (GII) group compared to the GI and GIII groups while about 4.89% improvement was observed in GIV compared to the drugs (GII) group (Fig. 3B).

Differential leukocyte count (DLC)

Nonsignificant change was observed in neutrophil levels in the drugs (GII) group compared to the GI and GIII

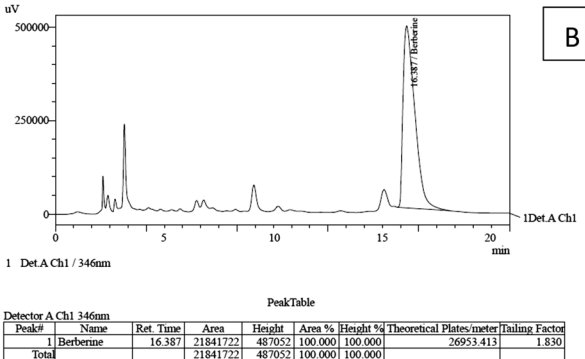
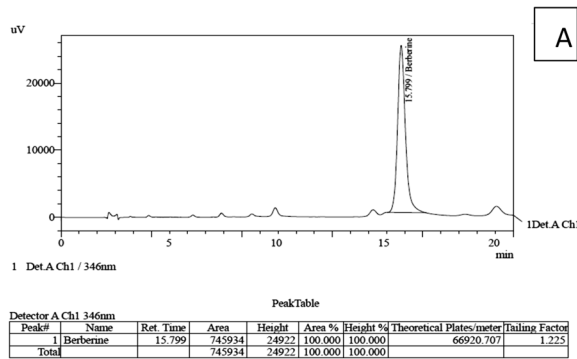


Fig. 1 Chromatogram of standard berberine (A) and *B. aristata* extract sample (B) in the mobile phase: potassium hydrogen phosphate (pH 2.5) and acetonitrile in the ratio of 80:20 (%v/v)

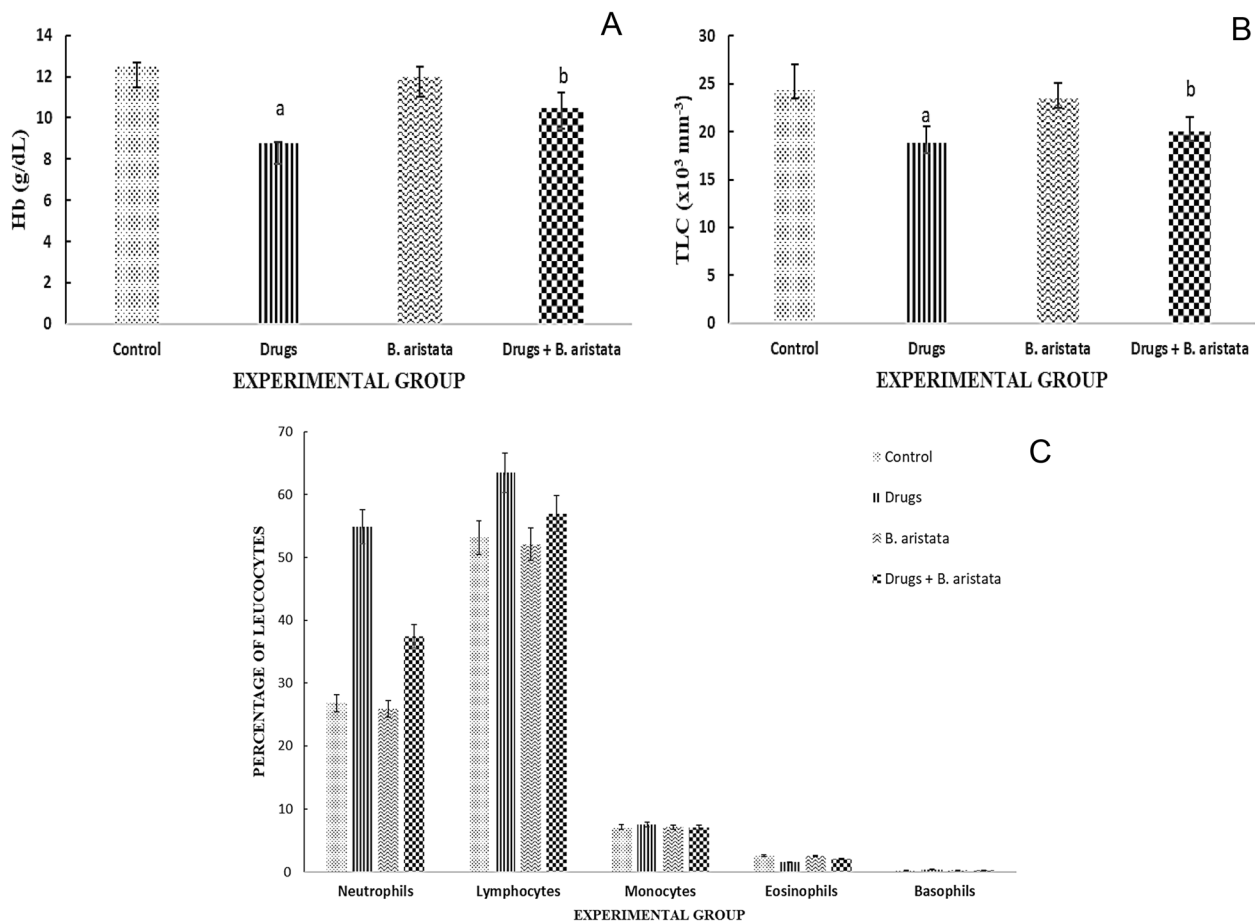


Fig. 3 Variation in hematological parameters of treatment groups. **A** Hemoglobin (g/dL), **B** TLC (g/dL), and **C** DLC (g/dL) levels in control and treatment groups. Significant difference at ($P < 0.05$) where alphabet 'a' represents with regard to the control and *B. aristata* alone and 'b' with respect to the drugs group

groups (Fig. 3C). The lymphocyte, monocyte, and basophil levels increased by 19.3%, 5.7%, and 44.8% respectively, compared to the GI and GIII groups. In contrast, the eosinophils were reduced by 39.84% in the drugs (GII) group compared with the GI and GIII (Fig. 3C). In the treatment (GIV) group, the level of neutrophil, lymphocyte, monocyte, and basophil decreased by 31.78%, 10.23%, 6.37%, and 23.80% respectively, whereas eosinophils increased by 31.21% compared to the drugs (GII) group (Fig. 3C).

Scanning electron microscopy of RBC

Erythrocytes from the control and positive control (GI and GIII) rats exhibited a typical appearance of disk-shaped cells (known as discocytes). A few exhibited cup-like cells termed stomatocytes (Fig. 4) (Kavita & Mittal, 2014). In contrast, erythrocytes from the drugs-treated rats showed significant morphological alterations (29.23%) and a high frequency of anisocytosis, i.e.,

variation in size and shape of the cell (Fig. 5). Most cells transformed into stomatocytes (9.23%) and echinocytes (7.69%), i.e., cells with irregular and spiky projections at varied development stages: stage I: echinocytes with irregular and jagged edges; stage II: cells displayed spicules protruding from the surface and maintaining a somewhat flat shape; and stage III: echinocytes with evenly distributed spicules across the entire cell surface, making them appear round but with a spiky texture. Additionally, acanthocytes with several spicules (or spikes) of varying lengths scattered unevenly across the cell surface, and knobby ends were observed (Kavita & Mittal, 2014). Furthermore, some of these cells displayed focal or peripheral bulges, known as blebbing and clubbing of cells, which indicates membrane instability. Blebbing of erythrocytes was evidenced in all the experimental groups. Herbal extract supplementation of *B. aristata* (300 mg/kg b.wt.) (GIV) revealed restored (12.5%) structure compared to the drugs (GII) group

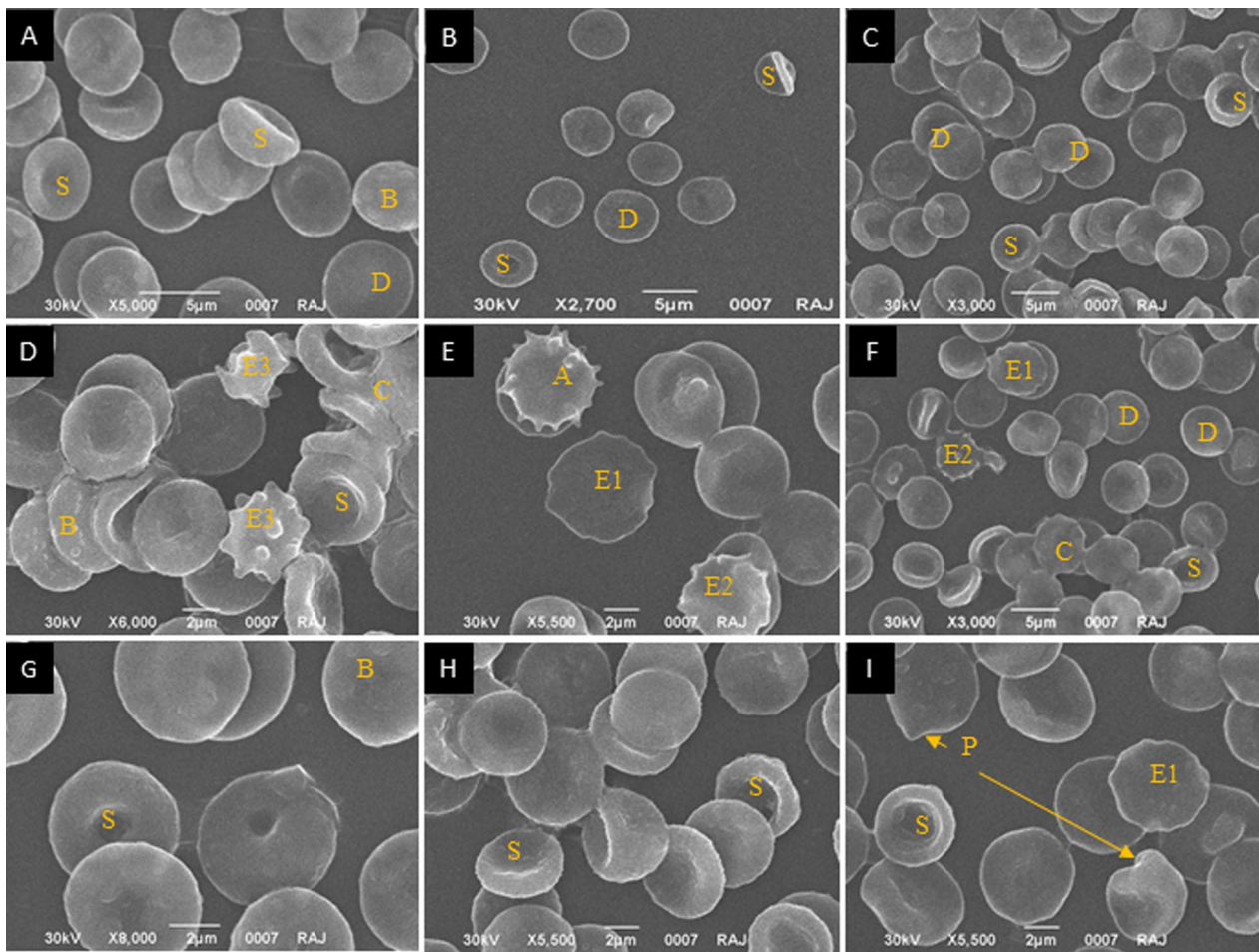


Fig. 4 Scanning electron micrograph of the rat erythrocytes upon exposure to first-line anti-tubercular drugs and *B. aristata* for 28 days. Control (A, B); *B. aristata* (C), drugs group (D, E, and F), and drugs + *B. aristata* (G, H, I). A-Acanthocyte, C-clubbing of erythrocytes, D-discocyte, E1-stage 1 of echinocyte, E2-stage 2 of echinocyte, E3-stage 3 of echinocyte, P-protuberance, S-stomatocyte

(Fig. 4), and quantitative analysis suggested significant amelioration through reduction in RBCs abnormalities.

Discussion

TB is a disease of antiquity caused by bacteria of the *Mycobacterium tuberculosis* complex, which principally affects the lungs and may spread to the kidneys, lymph nodes, and brain. The first-line drugs used to treat and cure this disease are isoniazid, rifampicin, and pyrazinamide. Although these drugs are effective, they can result in a variety of ADRs, which primarily affect organs involved in drug metabolism and excretion (Schaberg et al., 1996). The toxicity of first-line anti-TB drugs is due to the high production of ROS by their reactive metabolites, such as hydrazine from isoniazid, pyrazinoic acid from pyrazinamide, and deacetyl rifampicin from rifampicin (Sharma & Sharma, 2015; Song et al., 2022). The abundance of ROS in the body results in oxidative stress that damages the DNA, RNA, proteins, and other

biomolecules, ultimately leading to tissue autophagy. Previous studies conducted in our laboratory on various organs, including the liver and kidney (Sharma & Sharma, 2017) and testes (Sharma et al., 2018), have indicated tissue damage from the ROS generated during metabolism of anti-tubercular drugs.

Studies have indicated hematological alterations in patients upon treatment with the anti-TB drugs (Kassa et al., 2016a, 2016b; Reta et al., 2023a, 2023b). Congruent with this, hematological studies have shown that treatment of rats with anti-TB drugs caused a significant reduction in RBC count (Fig. 3), which may be accredited to the destruction of mature RBCs (Baynes et al., 1986; Berkowitz, 1991). Further, these drugs have been observed to inhibit erythropoietin release from the kidneys, leading to decrease in the rate of erythropoiesis (Mintzer et al., 2009). A significant decrease in Hb content signifies a decreased oxygen-carrying capacity of the blood and the amount of oxygen delivered to the tissues,

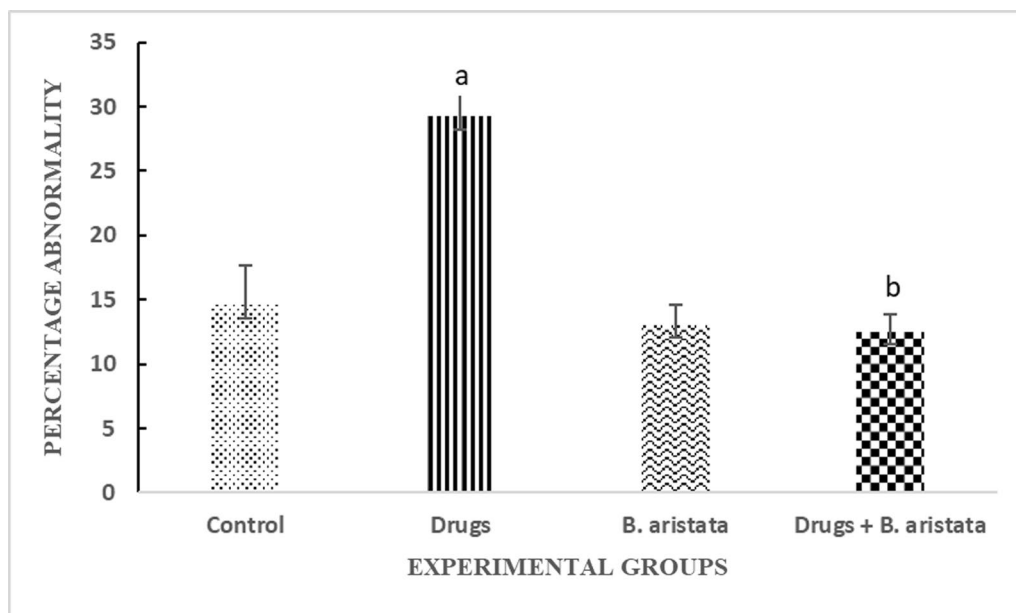


Fig. 5 Percentage abnormality in the rat erythrocytes upon exposure to first-line anti-tubercular drugs and *B. aristata* for 28 days. Significant difference at ($P < 0.05$) where alphabet 'a' represents with regard to the control and positive control *B. aristata* and 'b' with respect to the drugs group

thus contributing to erythrocytic abnormalities and cell death (Karandeniya et al., 2022; Reta et al., 2023a, 2023b; Yakar et al., 2016).

SEM of erythrocytes revealed a biconcave, disk-like shape of RBCs—discocytes, constituting the typical shape of healthy RBCs in rats. It is crucial for their function in the transportation of gases (Karandeniya et al., 2022; Rojas-Maya et al., 2016). Exposure to drugs resulted in cellular abnormalities that may be attributed to oxidative stress and lipid peroxidation of the cell membrane (Marganwar & Dhurvey, 2021). Stage 1 of echinocyte (E1) is identified with irregularly shaped flat RBCs. Subsequent stage 2 of echinocyte is flattened RBCs with a few spikes on the cell surface, while stage 3 of echinocyte, commonly referred to as burr cells, is spherical RBCs with a large number of spicules or pointed projections dispersed equally over their surface. These have lowered the cell surface-to-volume ratio which severely impacts their function (Lu & Shevkoplyas, 2020). These echinocytes are associated with RBC dehydration, increased pH, and decreased ATP/energy crisis (Lu & Shevkoplyas, 2020). Erythrocytes with protuberances are cells with multiple processes, perforations, and crypt-like excavations. The cells showing focal and peripheral projections exhibited signs of damage, such as sequestration of cell components associated with pathological conditions (Lu & Shevkoplyas, 2020). Stomatocytes are cup-like cells with a characteristic convex–concave form and a pallor in the center. Such structures are observed due to

an increased water content in cells and the imbalance of cation content and flux (Lu & Shevkoplyas, 2020). Stomatocytes have also been reported in cases of alcoholism, drug exposure, liver illness, and genetic disorders (Lu & Shevkoplyas, 2020). Structures like acanthocytes are deformed RBCs with disorganized and uneven spikes, revealing irregular membrane surfaces. Iron deficiency, hemolytic anemia, and massive anemia may also account for the condition (Darras et al., 2021; Sharma et al., 2018). Further, these changes are attributed to peroxidation of the lipids in the RBC membrane due to toxic metabolites of drugs and loosening of the cytoskeleton (Reta et al., 2023a, 2023b; Sharma & Sharma, 2015). Another reason for RBC cell membrane scrambling and cell shrinkage is the simulation of suicidal death or eryptosis brought about by rifampicin through the formation of ceramide by unregulated Ca^{2+} entry (Abed et al., 2012; Lopez-Canizales et al., 2021).

Supplementation of *B. aristata* (GIV) with berberine as an active ingredient significantly restored the levels of Hb and TLC in the rat and RBC morphology. Amelioration of hematological parameters thought to achieve by reducing oxidative damage to red blood cells (RBCs) and maintaining a healthy hemoglobin level and white blood cells (WBCs). Due to its antioxidant and anti-inflammatory effects, the extract can help mitigate hemolysis, anemia, and leukopenia induced by drug toxicity. It also supports the bone marrow's hematopoietic function, contributing to the regeneration of blood cells and preventing

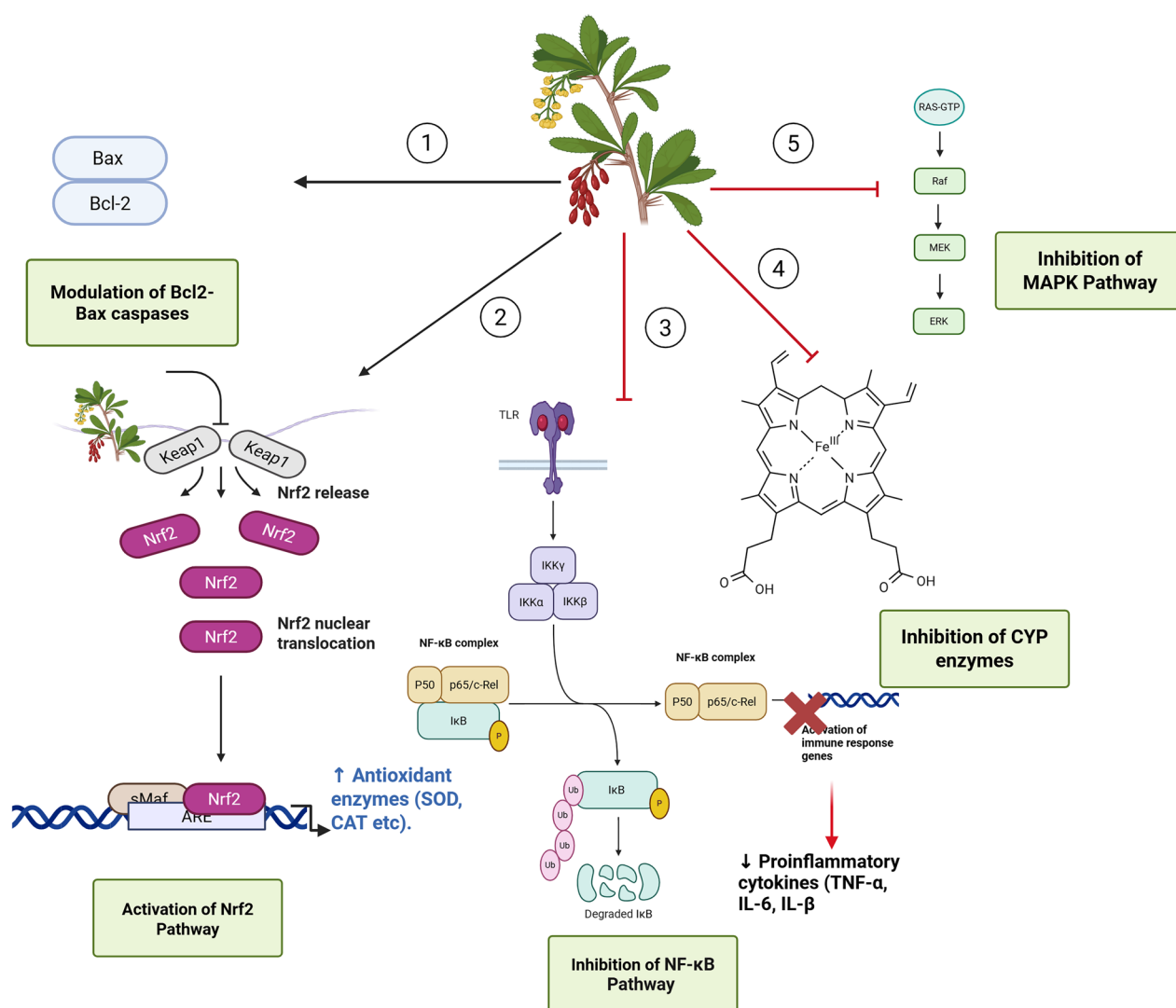


Fig. 6 Prospective protective mechanism of *B. aristata*

the hematological alterations typically seen during anti-tubercular drug therapy (Sharma & Sharma, 2017). *B. aristata* acts through its active compound berberine, which may offer protection against anti-TB drug-induced toxicity by targeting multiple molecular mechanisms (Fig. 6). First, berberine acts as a potent antioxidant, scavenging reactive oxygen species (ROS) and activating the Nrf2/ARE pathway, which upregulates antioxidant enzymes like superoxide dismutase (SOD) and catalase (CAT), thus mitigating oxidative stress and protecting cells from damage (Sharma et al., 2019). Additionally, berberine's anti-inflammatory effects, mediated through the inhibition of NF-κB and MAPK pathways, reduce the secretion of pro-inflammatory cytokines (TNF-α,

IL-6, IL-1β) that contribute to organ damage, particularly in the liver and kidneys (Gao et al., 2022). Furthermore, berberine regulates apoptosis by modulating the expression of Bcl-2 family proteins and inhibiting caspases, preventing excessive cell death in tissues affected by anti-TB drugs (Ozturk et al., 2021). It also modulates cytochrome P450 (CYP) enzymes, potentially altering the metabolism of toxic drug metabolites, thus reducing liver and kidney toxicity (Mangwani et al., 2020). By activating AMPK, berberine promotes cellular energy balance and autophagy, supporting tissue repair and reducing the adverse effects of anti-TB drugs on cellular metabolism (Raqib & Sarker, 2024). Through these combined actions, antioxidant, anti-inflammatory, anti-apoptotic, metabolic

regulation, and organ-specific protection, *B. aristata* extract may provide comprehensive protection against the toxicity induced by anti-TB drugs, safeguarding vital organs such as the liver, kidneys, spleen, and hematological health.

Conclusion

Significant hematological alterations were observed in the drugs group, including a decline in hemoglobin (Hb) and TLC. The scanning electron microscopy (SEM) study confirmed and extended the ultrastructural findings in RBCs, showing cell shrinkage and membrane scrambling induced by anti-tubercular drugs. Cells possess protective mechanisms against such stresses, including apoptosis regulation, cell cycle control, DNA repair, and detoxification. Reactive oxygen species (ROS) are pivotal in triggering various cellular cascades, such as the activation of tyrosine kinases and the mitogen-activated protein kinase (MAPK) pathway, followed by the initiation of transcription factor subsets. In response to oxidative stress, antioxidants are activated to scavenge ROS and regulate oxidative stress pathways. The protection of hematological health from anti-tubercular drug-induced toxicity is essential, as it can lead to severe conditions such as anemia, leukopenia, and thrombocytopenia, undermining both patient health and the effectiveness of tuberculosis treatment. The supplementation of *B. aristata* extract, rich in bioactive phytoconstituents like berberine, exhibited a significant alleviating effect against the toxicity caused by these drugs, thereby mitigating ADRs. The extract's antioxidant, anti-inflammatory, and anti-apoptotic properties provide a comprehensive protective mechanism against hematological and organ-specific damage. Further studies are warranted to fully explore the therapeutic potential of *B. aristata* in combating the harmful effects of anti-tubercular drug therapy and improving patient outcomes.

Abbreviations

ADR	Adverse drug reactions (ADRs)
AMPK	AMP-activated protein kinase
ANOVA	Analysis of variance
CYP	Cytochrome
DLC	Differential leukocyte count
FDA	Food and Drug Administration
HED	Human equivalent dose
HPLC	High-performance liquid chromatography
IAEC	Institutional Animal Ethics Committee
IL	Interleukin
INH	Isoniazid
MAPK	Mitogen-activated protein kinase
PZA	Pyrazinamide
RIF	Rifampicin
CAT	Catalase
ROS	Reactive oxygen species
SD	Standard deviation
SEM	Scanning electron microscopy
SOD	Superoxide dismutase
TB	Tuberculosis
TLC	Total leukocyte count

TNF	Tumor necrosis factor
WBC	White blood cells

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Author contributions

Radhika Sharma and Vijay Lakshmi Sharma designed the research work. Radhika Sharma and Seema Rani performed the research work. Radhika Sharma and Reshma Sinha wrote and revised the manuscript. Yogita Thakur created images and Vijay Lakshmi Sharma supervised the project.

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Data availability

The data presented in this study are available from the Radhika Sharma upon reasonable request.

Declarations

Ethical approval and consent to participate

The experiments were performed following the guidelines of the Institutional Animal Ethics Committee of Panjab University, Chandigarh (PU/IAEC/S/14/147).

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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