



Antioxidative Biomolecular protective activity of *Napoleonaea vogelii* Against Cyclophosphamide-induced hepatotoxicity in Rats

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ABSTRACT

Cyclophosphamide (CYP) is an anticancer drug that undergoes extensive metabolic biotransformation into pro-oxidant metabolites that are implicated in severe undesirable side effects in multiple organs. *Napoleonaea vogelii* (NV), is a tropical flowering plant with phytochemicals that have been shown to possess medicinal properties utilized in the management of cancer, dermatosis and peptic ulcer. There is paucity of information on its potential against toxicological profile of alkylating cytotoxic agents such as cyclophosphamide. This study therefore investigated the mitigative influence of NV against CYP-induced liver injury in rat. The rats intraperitoneally received cyclophosphamide (2 mg/kg) or orally co-treated with *Napoleonaea vogelii* (400 mg/kg) daily for 14 days. Co-treatment with *Napoleonaea vogelii* buffered the alteration on hematological parameters caused by cyclophosphamide. *Napoleonaea vogelii* in treatment group also mitigated impaired liver function by significant reduction in the activities of transaminases and the alkaline phosphatase relative to cyclophosphamide-intoxicated rats. CYP also altered redox status through significant reduction in serum total thiol, hepatic activities of catalase, superoxide dismutase and reduced glutathione concentration with concomitant elevation in serum advanced oxidized protein products-AOPPs and hepatic malondialdehyde concentration relative to control. However, co-treatment with *Napoleonaea vogelii* re-established the serum and hepatic antioxidant status.

Moreover, the increase in serum levels of proinflammatory markers: TNF-alpha, NF-KB and MPO were observed in treated rats exposed to benzene as compared to control. Meanwhile, co-treatment with *Napoleonaea vogelii* profers abrogative influence on the surge elevation observed in TNF-alpha, NF-KB and MPO in treated rats when compared to the CYP-exposed rats. Overall, *Napoleonaea vogelii* protected against oxidative hepatic injury and hematological derangement caused by cyclophosphamide in rats.

Keywords: *Napoleonaea vogelii*, cyclophosphamide, hepatoprotective, oxidative stress, antioxidant enzymes

INTRODUCTION

Non-alcoholic fatty liver diseases caused by drugs and environmental toxicant are on increase across the globe. Drug-induced hepatotoxicity constitutes a major clinical problem that accounts for greater proportion of cases of acute liver failure worldwide (Björnsson and Björnsson, 2022). Cyclophosphamide is a widely used alkylating chemotherapeutic drug for the treatment of immunosuppression, nephrotic syndrome and cancers such as leukemia, prostate, breast, lymphoma, lung and ovarian cancers (Paul et al., 1991; Kirkland et al., 1976; Shanafelt et al., 2007). Its usage causes the additional off-target hepatotoxic effect in cancer patients worldwide. The resultant hepatotoxic adverse effect of cyclophosphamide has a great limiting influence on its purported therapeutic

utility (Rajput and Singh, 2025). As part of its mechanism of action, cyclophosphamide is bioactivated by cytochrome P450 enzyme into reactive metabolites that induced oxidative stress and lipid peroxidation which ultimately leads to hepatocellular injury (García-Martínez *et al.*, 2025). Therefore, the side effect of this potent anticancer drug necessitates the search for the adjuvant agent that can mitigate its therapy-induced liver damage while maintaining its chemotherapeutic efficacy.

Application of medicinal plants in phytomedicine seems to be a viable substitute for synthetic drugs in modern contemporary medicine as it is both effective and has mild or no adverse effects (Al-Asmari *et al.*, 2014). The World Health Organization (WHO) reported the reliance of greater percentage of populace across the globe on traditional medicine as the main primary health care (WHO, 2019). *Napoleonaea vogelii* (Lecythidaceae family) is popularly known as Napoleon’s tree in English and “Yere odo” in Yoruba language among Nigerians. The plant has gained a lot of interest among its family due to its wide range of medicinal uses. It is historically used for the treatment of gastrointestinal problems, malaria, and also for liver diseases (Olounlade *et al.*, 2021). Previous research has also shown that extracts of *Napoleonaea vogelii* have strong antioxidant activity, including potent free radical scavenging and metal chelating properties (Ikumawoyi *et al.*, 2017). The potency of phytoconstituents in medicinal plant to neutralize oxidative stress is a key factor in their hepatoprotective benefits (Farghali *et al.*, 2015). Despite its extensive use in traditional medicine for managing liver-related ailments, its hepatoprotective potential against cyclophosphamide-induced liver toxicity remains largely unexplored. This study aimed to bridge this gap by evaluating the protective effect of *Napoleonaea vogelii* against cyclophosphamide-induced hepatic damage in rats.

MATERIALS AND METHODS

Preparation of *Napoleonaea vogelii* Extract

Fresh *Napoleonaea vogelii* plants were collected in April, 2024 from swampy regions in Ajaowa axis area of Oyo state in Nigeria. The plant was identified and authenticated by plant taxonomist at Herbarium section of Department of Botany, University of Ibadan, Nigeria with University of Ibadan. The bark was carefully stripped, thoroughly rinsed with tap water to remove impurities, and sliced into smaller fragments. These pieces were then air-dried under shed before being finely ground into powder utilizing a mechanical grinder. The sample was subsequently subjected to cold maceration in 80% methanol: water to extract bioactive compounds. The extract was concentrated using rotary evaporator and lyophilized by freeze drier.

Chemicals and Reagents

The acid, 1-Chloro-2,4-dinitrocyclophosphamide (CDNB), glutathione (GSH), epinephrine, hydrogen peroxide and 5,5 -dithio-bis-2-nitrobenzoic acid (DTNB) were got from Sigma® Chemical Company (London, UK). The transaminase (aspartate and alanine aminotransferase) kits were product of Fortress Diagnostics Ltd., Antrim, UK. All chemicals and other reagents used for the study were analytically of standard grade and obtained from British Drug House, Poole, UK.

Animals for Experiment

Twenty-four white wistar rats that weighed between 160-180g were purchased from Temmy Farms, Ogbomosho, Oyo State, Nigeria. The animals were accustomed to laboratory environment before study begins and the animals fed with commercial rat diet. The designed work was conducted with the approval of the Faculty of Natural Sciences Ethical review of Ajayi Crowther University, Oyo with approval code: FNS/ERC/2024/009. Anaesthesia was not used in the study and the protocol followed the stipulate guideline for use and care of laboratory animals (NRC, 2011).

Experimental Design

This research study used simple randomization laboratory design. A week after adaptation, animals were divideded into four groups (n = 6). The extract and cyclophosphamide doses were administered daily via intragastric and intraperitoneal respectively for fourteen (14) days whereas in co-treatment, animals received extract treatment before cyclophosphamide administration as shown in Table 1 below.

Table 1: Experimental Design

Treatment categories	Period of treatment (14 days)
Control	Distilled water
CYP alone	2 mg/kg cyclophosphamide
CYP + NV	400 mg/kg <i>N. vogelii</i> + 2 mg/kg CYP
NV alone	400 mg/kg <i>N. vogelii</i>

Preparation of Blood and Liver specimens

Twenty-four hours following the final treatment, animals were given an intraperitoneal injection of 0.04% colchicine to enhance cellular analysis, with a waiting period of two hours before they were sacrificed by cervical dislocation. The blood was phlebotomized from animal through the retro orbitals into ethylenediaminetetraacetate (EDTA) bottles for hematological assessment and also into the lithium heparinised tubes for biochemical analyses. Plasma was prepared by centrifuging blood samples in lithium heparinised tubes for 5 min at 4000 rpm using

a bench centrifuge (Analytica, Athens, Greece). The clear supernatant was used for the estimation of biochemical assays. The liver excised from rat was blotted of blood stains, and rinsed in 1.15% KCl and homogenized in 4 volumes of ice-cold 0.01 M potassium phosphate buffer (pH 7.4). The homogenates were centrifuged at 12,500 g in a refrigerated centrifuge (Eppendorf, Stevenage, UK) for 15 min at 4 °C and the supernatants, termed as the post mitochondrial fractions (PMF), were used for antioxidant assays.

Hematological Assessment

Hematological analysis was performed on blood collected in EDTA bottle, assessing the hematocrit, hemoglobin, mean corpuscular, mean corpuscular volume, hemoglobin, white blood cells, monocytes, red blood cells, lymphocytes, granulocytes (Gran), and platelets (PLT) using blood autoanalyzer (HRJ-BIO (HRJ-H300)).

Determination of Protein Content in plasma and liver: Plasma and liver homogenate protein levels were quantified as outlined by Gornall *et al.* (1949) using Biuret procedure. Briefly 0.5 ml of the diluted sample was added to 1.5 ml of Biuret reagent and the mixture was incubated at room temperature for 30 minutes after which the absorbance was read at 540 nm using distilled water and biuret reagent as blank. Protein concentrations in the samples were derived by comparing them to a standard curve generated using bovine serum albumin.

Determination of biomarker of liver injury: The enzymatic activities of plasma alkaline phosphatase (ALP), aspartate aminotransferase (AST), alanine aminotransferase (ALT) and bilirubin content were determined using established methodologies. AST and ALT activities were assessed through the hydrazone formation method introduced by Reitman and Frankel (1957). ALP activity was determined by tracking p-nitrophenol production at 405 nm, following the approach described by Wright *et al.* (1972). Bilirubin contents were quantified employing dimethyl sulfoxide procedure of Kohashi *et al.* (1988), which involves reaction with diazotized sulfanilic acid and subsequent absorbance measurement at 550 nm.

Assessment of Plasma Oxidative Stress Levels: Plasma advanced oxidation protein product (AOPP) was assessed by modified method of Witko - Sarsat *et al.* (1996). A plasma sample of 100 µl was combined with 400 µl of phosphate-buffered saline, followed by the addition of potassium iodide (25 µl of 1.16 molar concentration). After two minutes, 50 µl acetic acid was introduced, and absorbance was instantly recorded at 340 nm. Blank solution contains phosphate buffer, potassium iodide, and acetic acid was used as a reference. The total thiol

concentration was spectrophotometrically evaluated by standardized principle of Ellman (1959).

Assay of Hepatic Redox Enzymes: Reduced glutathione was quantified via a interaction with 5,5'-dithiobis-(2-nitrobenzoic acid), absorbing at 412 nm, as elaborated by Jollow *et al.* (1974). The SOD activity was assessed with reference to inhibition of epinephrine from auto-oxidation at pH 10.2, following Misra and Fridovich method (1972). Catalase activity was evaluated through the reduction of dichromate to chromic acetate when hydrogen peroxide is present, measured at 570 nm, using Singha's method (1972) approach.

Assessment of extent of Peroxidation of Lipid: Hepatic tissular lipid peroxidation was evaluated by quantifying malondialdehyde (MDA) concentrations via thiobarbituric acid (TBA) assay, as elaborated by Varshney and Kale (1990). MDA component of liver reacted with TBA and forms a pink colored complex at 532 nm. MDA levels were derived using a standard curve, with results expressed in nmol/g of liver tissue.

Assessment of Inflammatory Cytokine and Enzymes: The concentrations of TNF- α , NF- κ B, and MPO in liver tissue were measured using ELISA and spectrophotometric techniques. TNF- α and NF- κ B levels were analyzed with sandwich-ELISA kits, where optical density (OD) values corresponded to their concentrations, recorded at 450 nm. MPO activity was determined using O-dianisidine in the presence of hydrogen peroxide, with OD readings taken at 470 nm. The assays followed manufacturers' guidelines and Kim *et al.* (2012) methodologies, with TNF- α and NF- κ B concentrations calculated from standard curves, while MPO activity was expressed through OD values.

Statistical analysis

Data was expressed as means $\pm$ standard deviation (SD) which was then subjected to the one-way analysis of variance with the use of Systat software complemented with Duncan's multiple range test. The statistically significant was considered on the basis of $p < 0.05$.

RESULTS

Hepatoprotective effects of *Napoleonaea vogelii* treatment on the activities of plasma on liver function markers

Table 2 presents the results of liver function assessments in rats subjected to cyclophosphamide exposure and subsequently treated with *Napoleonaea vogelii*. Cyclophosphamide administration led to a significantly elevation in activities of ALT, ALP, AST and bilirubin

level. However, concurrent treatment with *Napoleonaea vogelii* effectively reversed these alterations, restoring liver

function markers to values comparable to the control group.

Table 2: Hepatoprotective Effects of *Napoleonaea vogelii* extract on some liver function parameters in rats.

Groups	ALT (U/L)	AST (U/L)	ALP (nmol/min/mgprotein)	Bilirubin (μmol/L)
Control	31.48 ± 3.83	46.50 ± 4.90	19.99 ± 2.65	3.34 ± 2.40
CYP	40.29 ± 2.25*	62.05 ± 4.45*	31.94 ± 2.47*	5.64 ± 1.51*
CYP + NV	32.17 ± 3.64 [#]	50.80 ± 5.62 [#]	22.68 ± 1.53 [#]	4.25 ± 0.29 [#]
NV	29.20 ± 4.25	44.47 ± 4.95	20.89 ± 2.81	3.42 ± 1.03

*significant different (p < 0.05) to control group; [#] significant different (p < 0.05) to CYP-only group

Action of *Napoleonaea vogelii* on alteration in haematological parameters in cyclophosphamide-induced toxicity in wistar rats

Table 3 shows that *Napoleonaea vogelii* treatment mitigates benzene-induced hematological alterations, with

enhanced PCV, hemoglobin, and RBC counts comparable to control group values.

Table 3: Effects of *Napoleonaea vogelii* on alteration in Haematological parameters in Cyclophosphamide-induced toxicity in Wistar rats

Groups	WBC (10 ⁹ /L)	Lym (10 ⁹ /L)	Gran (10 ⁹ /L)	RBC (10 ¹² /L)	HGB (g/dL)	HCT (%)	PLT (10 ⁹ /L)	MPV (fL)
CTRL	7.42 ± 1.32	3.75 ± 6.41	0.27 ± 0.28	7.93 ± 0.35	16.24 ± 0.38	46.5 ± 1.12	732.6 ± 228.8	7.2 ± 0.70
CYP	4.02 ± 0.79	3.55 ± 0.77	0.29 ± 0.38	7.29 ± 0.57	14.9 ± 1.05	44.24 ± 2.11	912.6 ± 117.46	7.38 ± 0.36
CYP+NV	4.83 ± 0.55	4.04 ± 0.57	0.21 ± 0.08	7.91 ± 0.10	15.76 ± 0.28	46.04 ± 1.14	930.2 ± 135.62	6.84 ± 0.27
NV	5.82 ± 1.27	4.36 ± 1.38	0.32 ± 0.30	7.74 ± 0.55	15.84 ± 1.15	47.18 ± 2.20	831.8 ± 85.72	7.04 ± 0.15

The values are expressed as mean ± SD. HCT- Haematocrit; RBC – Red Blood Cells; WBC - White Blood Cell Count; HGB– Haemoglobin; Gran-Granulocyte; MPV - Mean Platelet Volume, PLT- Platelet;

Protective effect of *Napoleonaea vogelii* on Cyclophosphamide Induced Toxicity of Hepatic Enzymatic Antioxidants in Rats Figure 1 illustrates the results of antioxidant status assays conducted on Wistar rats subjected to cyclophosphamide exposure and treated with *Napoleonaea vogelii*. Cyclophosphamide administration led to a marked reduction in superoxide dismutase, catalase

and glutathione (GSH) levels within liver tissue, indicating oxidative stress. However, simultaneous treatment with *Napoleonaea vogelii* significantly restored these antioxidant markers, highlighting its potential protective role in counteracting cyclophosphamide-induced oxidative damage.

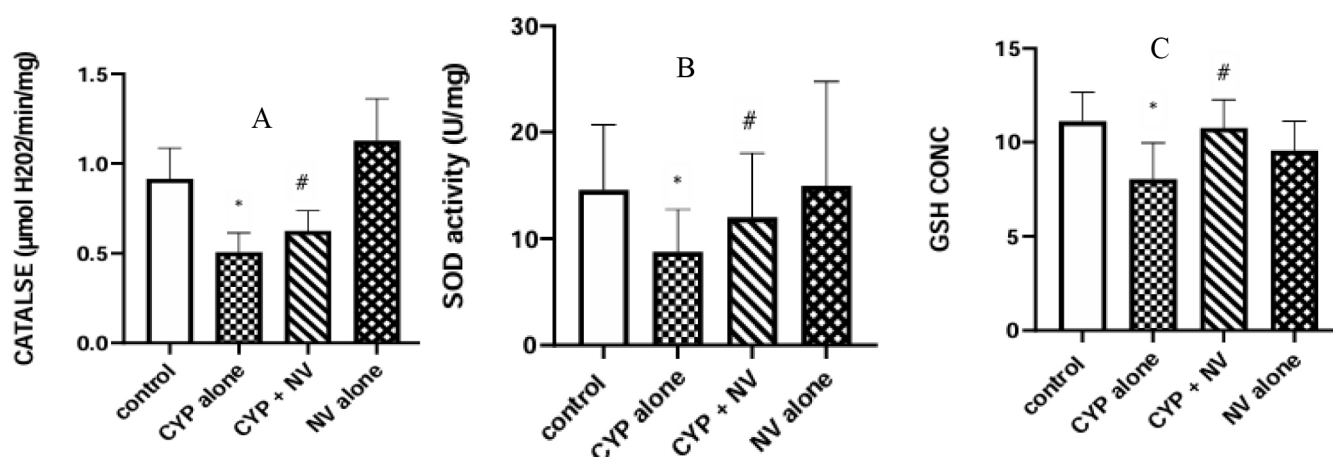


Figure 1: Effect of *Napoleonaea vogelii* on cyclophosphamide-induced toxicity on hepatic redox status in rats. * (p < 0.05) versus control; [#] (p < 0.05) against cyclophosphamide A - catalase, B-superoxide dismutase, C- reduced glutathione.

Anti-inflammatory effect of *Napoleonaea vogelii* on Cyclophosphamide-Induced toxicity on Inflammatory markers in Wistar Rats

Figure 2 shows inflammatory biomarker levels in Wistar rats exposed to cyclophosphamide and treated with *Napoleonaea vogelii*. Cyclophosphamide significantly increased plasma levels of Nuclear Factor Kappa β

(NFK- β), Tumor Necrosis Factor- α (TNF- α), and the myeloperoxidase activity. Co-administration of *Napoleonaea vogelii* with cyclophosphamide significantly reduced these inflammatory markers, suggesting a protective effect against cyclophosphamide-induced inflammation.

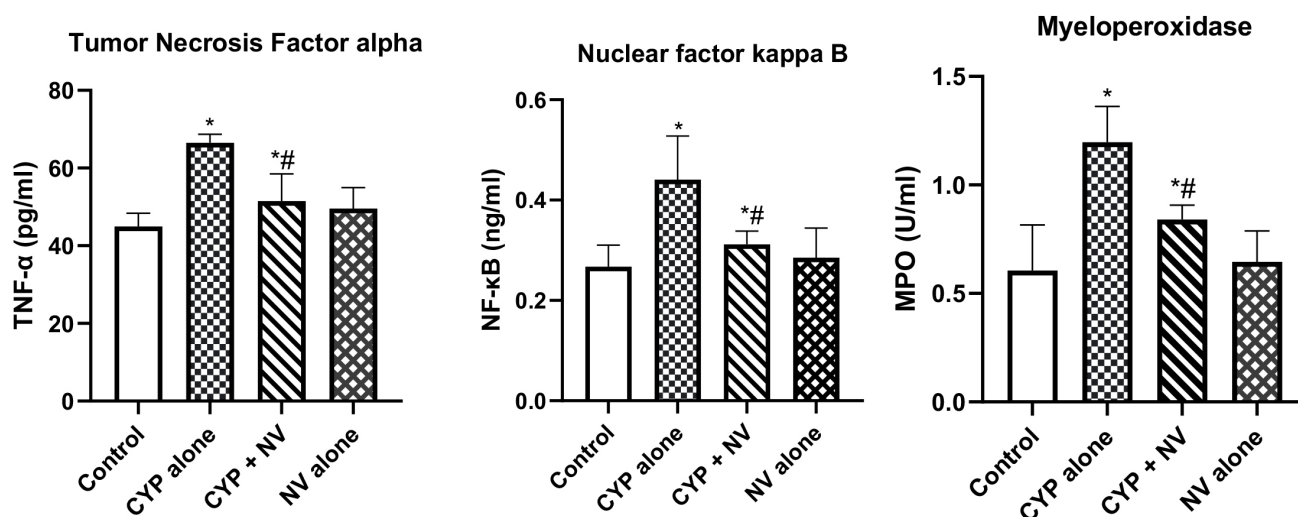


Figure 2: Effects of *Napoleonaea vogelii* on inflammatory biomarkers in cyclophosphamide-induced toxicity in Wistar rat. * ($p < 0.05$) versus control; # ($p < 0.05$) versus cyclophosphamide.

Effect of *Napoleonaea vogelii* on Oxidative Stress Markers in Cyclophosphamide-Induced Toxicity in Rats

Figure 3 shows oxidative stress assays in Wistar rats exposed to cyclophosphamide and treated with *Napoleonaea vogelii*. Cyclophosphamide significantly

increased advanced oxidation protein products (AOPP), Malondialdehyde (MDA) and decreased total thiol levels in plasma. Co-administration of *Napoleonaea vogelii* with cyclophosphamide significantly restored these oxidative stress markers, suggesting that its protective effect against cyclophosphamide oxidative damage.

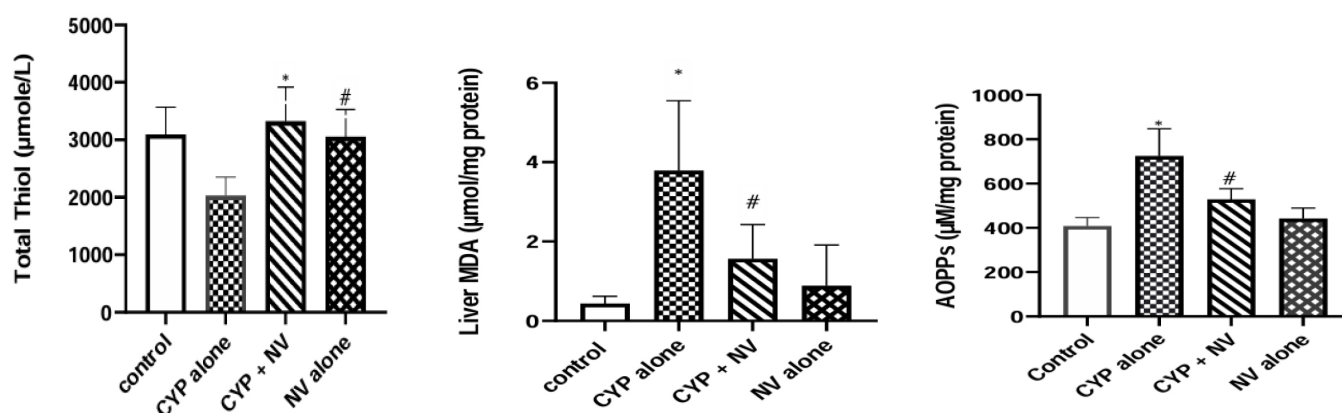


Figure 3: The effect of *Napoleonaea vogelii* on plasma oxidative stress markers in cyclophosphamide-induced toxicity in Wistar rats. * ($p < 0.05$) versus control; # ($p < 0.05$) against cyclophosphamide

DISCUSSION

Cyclophosphamide (CP), an inactive cytostatic alkylating agent, undergoes metabolic activation in the liver via microsomal enzymes, producing phosphamide mustard and acrolein (De Jonge et al., 2005). This bioactivation process is accompanied by the generation of reactive oxygen species that can affect both healthy and cancerous cells, leading to a reduction in antioxidant defenses (Chakraborty *et al.*, 2016). This study examined the ability of *Napoleonaea vogelii* to protect the liver of Wistar rats from cyclophosphamide-induced damage by analyzing various biochemical markers, as well as enzymatic and non-enzymatic antioxidants.

Previous study has shown that animals exposed to cyclophosphamide develop liver tissue damage and necrosis, as indicated by increased lipid peroxidation and elevated levels of liver-specific enzymes such as AST and ALT, along with decreased albumin (Ola and Odunola, 2023a). Activities of ALT and AST in serum are widely used indicators of liver injury (McGill, 2016) in which their surge increase in the bloodstream is linked to leakage from damaged liver cells (Kalas et al., 2021). ALP, an enzyme associated with the cell membrane, is another marker of liver damage (Poupon, 2015). Higher plasma ALP levels are linked to cholestasis, hepatobiliary injury, red blood cell breakdown, or disruptions in bilirubin metabolism (Youssef and Wu, 2024). Findings from this study show that cyclophosphamide significantly increased the activities of these enzymes in the treated group compared to the control group as presented in Table 3.1. The rise in enzyme activity in the toxicant group likely resulted from tissue damage caused by cyclophosphamide metabolism in the liver. However, co-treatment with *Napoleonaea vogelii* reduced the activities of these enzymes compared to the cyclophosphamide-only group, indicating its protective effect against cyclophosphamide-induced liver damage.

The influence of antioxidant enzymes and molecule in intervening in oxidative stress-mediated liver damage and remodeling of the liver vasculature has been reviewed (Banerjee et al., 2023). SOD plays a vital role in neutralizing superoxide radicals and protecting cells from oxidative stress due to its dismutation catalysis (Ozougwu, 2016). Catalase is also a key antioxidant enzyme that helps to protect cells from the harmful effects of lipid peroxidation (Ighodaro and Akinloye, 2018). In this study, catalase and SOD activities were significantly decreased in the cyclophosphamide-only group compared to the control group (Figure 3.1). This may likely be due to excessive free radical production overwhelming the antioxidant defense system (Pisoschi and Pop, 2015). However, the administration of *Napoleonaea vogelii* led to a noticeable increase in catalase and SOD activities in the treated group

compared to the cyclophosphamide group (Figure 3.1). The differences in SOD and catalase activities across groups highlight the interaction between oxidative stressors such as cyclophosphamide and natural antioxidants found in *Napoleonaea vogelii*. These findings support the potential use of plant-based treatments to counteract drug-induced oxidative liver damage.

Tumor Necrosis Factor alpha (TNF- α) is a key pro-inflammatory cytokine that plays a central role in systemic inflammation and acute phase reactions (Silva, 2019). It is primarily produced by activated macrophages but can also be secreted by lymphocytes, natural killer cells, and neurons. During inflammation, TNF- α triggers acute phase responses, leading to increased C-reactive protein levels, causing vasodilation and heightened vascular permeability (Urschel and Cicha, 2015). However, excessive TNF- α production can lead to various inflammatory diseases as it is a key regulator of immune and inflammatory responses (Silva, 2019). TNF- α can influence the NF- κ B pathway and promote its activation (Chen et al., 1999). Previous findings showed that oxidative stress-related NF- κ B activation is implicated in inflammation and fibrosis (Jokelainen et al., 2001). The TNF- α is involved in activation of NF- κ B which in turn can upregulate myeloperoxidase expression and therefore, forming a complex network of inflammatory signaling (Ribeiro et al., 2015). This study examined TNF- α , NF- κ B and myeloperoxidase activity in Wistar rats that were administered cyclophosphamide with or without *Napoleonaea vogelii*. The result of this work demonstrates the implication of the anticancer drug cyclophosphamide in triggering an inflammatory response, evidenced by elevated levels of TNF- α , NF- κ B and myeloperoxidase activity. Previous studies have shown that cyclophosphamide induces oxidative stress and triggers inflammatory pathways (Ibrahim et al., 2024). In contrast, NF- κ B, TNF- α , and myeloperoxidase activity were significantly reduced in the co-treatment group ($p < 0.05$), suggesting that *Napoleonaea vogelii* may have anti-inflammatory properties. This observation is consistent with the traditional use of *Napoleonaea vogelii* for managing inflammatory conditions (Eze and Unachukwu, 2015).

Lipid peroxidation caused by free radicals has been associated with membrane structure alterations and cellular dysfunctions such as mutations and cell death (Varshney and Kale, 1990). Lipid peroxidation is initiated when free radicals attack polyunsaturated fatty acid, causing the formation of various compounds like lipid peroxides, malondialdehyde (MDA) and reactive carbonyl species (Valko et al., 2006). The fluidity of membrane is maintained by polyunsaturated fatty acids within phospholipids distributed on both sides of lipid bilayer (Mukerjee et al., 2021). Excessive lipid peroxidation can

lead to membrane disruption and cellular instability. A rise in MDA levels is considered a marker of peroxidation of lipid (Yekti et al., 2018). In the present study, the animals administered with cyclophosphamide reflected significantly higher MDA levels compared to control, suggesting that cyclophosphamide induces oxidative stress via lipid peroxidation. However, results from this study indicate that the elevated MDA levels in cyclophosphamide-treated rats was relatively and significantly reduced in the co-treated group. This suggests that *Napoleonaea vogelii* mitigates the oxidative damage occurrence of peroxidation in lipid component of biological structure. Advanced oxidation protein products-AOPPs are cross-linked protein compounds that serve as indicators of oxidative protein damage (Conti et al., 2019). Findings from this study revealed a significant increase in hepatic AOPP levels in the cyclophosphamide-treated group as presented in Figure 3.3., aligning with previous research (Sherif, 2018). This suggests that cyclophosphamide treatment induces oxidative stress, leading to protein damage in the affected animals. However, co-administration of *Napoleonaea vogelii* reduced plasma AOPP levels, indicating its protective role against oxidative damage to protein.

Total thiol levels, particularly thiol groups found in proteins, are major antioxidants in plasma and contribute to cellular defense (Oliveira and Laurindo, 2018). Most surfydryl groups are present in ovalbumin, which functions as one of the primary reducing components in body fluids. Previous studies have found that lower levels of protein thiols negatively correlate with AOPP concentrations (Alagozlu et al., 2013). Cyclophosphamide is known to generate reactive species and induce oxidative stress, that led to the depletion of cellular thiols such as reduced glutathione (GSH) (Korkmaz et al., 2007). The GSH is a key intracellular non-enzymatic antioxidant that helps detoxify both internal and external toxic substances while also regenerating vitamins C and E into their active forms (Moussa et al., 2019). GSH is a cofactor for several enzymes, such as glutathione-S-transferase and glutathione peroxidase (GPx) and plays a role in neutralizing free radicals within cells (Averill-Bates, 2023). The decline in total thiols and GSH observed in cyclophosphamide-treated group compared to the control group suggests emergence of oxidative stress due to compromise of antioxidant defenses. Earlier study has reported the connection between cyclophosphamide-induced toxicity and cellular depletion of GSH (Liu et al., 2022). However, treatment with *Napoleonaea vogelii* buffered up the total thiol and GSH levels in co-treatment group. The protective effect of *Napoleonaea vogelii* may be linked to its high content of antioxidant compounds, which aid in preserving or regenerating thiol-containing protein and molecules (Oluseun et al., 2023).

In conclusion, *Napoleonaea vogelii* protected against oxidative modification of protein and lipid biomolecules as reflected on its influence on advanced oxidized protein products and formation of malondialdehyde content in cyclophosphamide exposed rats. This protective potential may due to its antioxidant capacity through buffering of redox status in rats.

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