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PHYTOCHEMICAL SCREENING AND BRINE SHRIMP LETHALITY TEST (BSLT) OF MANGIFERA INDICA LEAVES OF BONGAO, TAWI-TAWI, PHILIPPINES

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ABSTRACT

Phytochemical screening in terrestrial plants play a vital role in human health by providing protective benefits against various diseases and can be used as a potential ingredient for the progression of functional foods and pharmaceutical drugs while, brine shrimp lethality test is an essential method to examine the cytotoxic attributes of the species that will merit further investigation for various medicinal application. This study aimed to determine the bioactive compounds in *Mangifera indica leaves* and assess their toxicity through (BSLT). The result showed that *Mangifera indica leaves* possessed diverse range of bioactive compounds exhibiting positive results for alkaloids, flavonoids, phenols, tannins, saponins, triterpenoids, and cardiac glycosides however, steroids was not determined. The toxicity result indicates that the *Mangifera indica leaves* had LC_{50} value of 3090.29 ppm pointing it as non-toxic. The result synthesized suggests that *Mangifera indica* may have potential application in traditional medicinal used.

KEYWORDS: *Mangifera Indica, Phytochemical Screening, Toxicity, Philippines.*

INTRODUCTION

Mango *(Mangifera Indica)* also known as “manga” in the Philippines is considered the most prevalent horticultural crops in the tropical and subtropical region around the world (Khan *et. al.*, 2015). Mango *(Mangifera indica L.)* ascribed to the family Anacardiaceae has been adjudged as the vital traditionally significant and one of the most economically important tropical fruit crop globally world (Barreto, *et. al.*, 2008). It is an evergreen tree with a lot of traditional medicinal resources apart from its very famous fruits. The Philippines ranked seventh in mango exports, with USD 91M in both fresh and dried mango exports for 4% share global market (UN Comrade 2016). Locally, many studies had been conducted to continuously increase the industry’s profitability, but they are strongly focused on breeding, fruit production, and processing. There were only a few works exploring the potentials and utilization of the non-fruit parts of the mango tree such as the bark and leaves. Further studies proved their appropriate applications in food, pharmaceuticals, and cosmetics (Charrier *et al*. 2006; Masibo and He 2008; Morsi *et al.* 2010; Mohan *et al.* 2013). For generations, diverse cultures have used mango leaves medicinally, believing them to have anti-inflammatory, antioxidant, antimicrobial, and anti-diabetic effects. This traditional knowledge has spurred increased scientific research into the leaves' chemical makeup and their potential health benefits. Extracts of the Mango leaves have been utilized for traditional medicines to cure diabetes, bronchitis, diarrhea, asthma, kidney, scabies, respiratory problems, syphilis, and urinary disorders (Shah *et al.*, 2010; Kulkarni 2014). In traditional medicine, dried leaves of M. indica were considered useful in the treatment of respiratory infections and diabetes (Zhang et al., 2019). M. indica was enlisted in TRAMIL (Program of Applied Research to Popular Medicine in the Caribbean) as an active agent in treating fever, ulcers, gastritis, and diarrhea (Robineau & Saejarto, 1996).

The plants abundantly grown well in tropical in the area of Bongao Tawi-Tawi where the elderly often recognized

its medical properties and used its parts as herbal medicine. The study was conducted to determine comprehensive phytochemical screening of *Mangifera Indica leaves* to identify and quantify these bioactive compounds. Moreover, Brine shrimp lethality test was done to determine the toxicity of *Mangifera Indica* for future pharmaceutical drugs. The BSLT not only assesses toxicity but also indirectly reflects the pharmacological potential of the phytochemicals present. A higher lethality rate may indicate the presence of potent bioactive compounds, while a low lethality rate might suggest safety and endorsement for potential therapeutic applications. Overall, by integrating phytochemical analysis with toxicity assessment, we aim to provide broad perspective of health implications related to this plants.

MATERIALS AND METHODS

The Mango leaves that were used in this study were collected from the Terrestrial area of Pahut on September 2024. Officially, Pahut is a barangay in the Municipality of Bongao, Tawi-Tawi, Province, (Region 9), Philippines. Geographically, Pahut is located at 5° 1' 38" N 119° 44' 31" E. The Phytochemical screening in this study was determined using the different standard chemical tests of (Kaur & Arora, 2009). Brine Shrimp Lethality Test is a screening method for test material that uses Artemia salina Leach which is an animal that has high sensitivity, where shrimp larvae have thin skin, so that the death of a larva due to the cytotoxic effect of bioactive compounds can be analogous to the death of a cell in an organism. (Fenton & Longo, 2011). It was used to assess the toxicity of *Mangifera indica leaves* for the preliminary evaluation.

RESULT AND DISCUSSION

Phytochemical Screening

The results for the phytochemical screening are shown in Table 1.

Table 1: Phytochemical Screening of *Mangifera Indica leaves.*

Phytochemical	Screening *Mangifera indica*
Alkaloids	+
Flavonoids	+
Phenols	+
Saponins	+
Triterperoids	+
Steroids	-
Cardiac Glycosides	+

Where '+' indicates presence '-'indicates absence

Table 1 provides a comprehensive summary of the phytochemical composition of Mangifera indica. The findings indicated the existence of bioactive compounds including alkaloids, flavonoids, phenol, tannins, saponins, triterpenoids, and cardiac glycosides, whereas steroids were not detected. The findings were in alignment with the study conducted by Diso (2017), which similarly recognized the existence of secondary metabolite compounds in *Mangifera indica*.

According to is Alshammaa (2016). *Mangifera indica* an important source of many pharmacologically and medicinally important chemicals. It is a potential source of polyphenolic compounds with high antioxidant activity that protect the body against damage linked to oxidative stress.

The presence of alkaloids was ascertained via the Wagner test, which produced a positive outcome following the incorporation of 0.5 g of the sample into 2 ml of hydrochloric acid (2N). The emergence of a light brown-to-red precipitate signifies the existence of an alkaloid.

Alkaloids. Alkaloids were present in *Mangifera indica* leaf extract using Wagners suggesting potential medicinal uses. The significant alkaloid content in mango leaves contributes to the plant's therapeutic value. Alkaloids play an essential role in both human medicine and in an organism's natural defense. Alkaloids make up approximately 20% of the known secondary metabolites founds in plants.

In plants, alkaloids protect plants from predators and regulate their growth Chik, S. C *et al.,* (2013). Therapeutically, alkaloids are particularly well known as anaesthetics, cardio protective, and anti-inflammatory agents. Well-known alkaloids used in clinical settings include morphine, strychnine, quinine, ephedrine, and nicotine Kurek (2019).

To test the flavonoids two to three drops of sodium hydroxide, were introduced to 2 ml. of the extract. Initially, a deep yellow hue manifested, which, upon the addition of a few drops of dilute hydrochloric acid, gradually transitioned to a colorless state, signifying the presence of flavonoids.

Flavonoids. Flavonoids were detected as positive in *Mangifera Indica* similar to Okwu *et. al*. (2008) reported that the flavonoids was very high in the leaves (11.24mg). These quantification supported the findings that flavonoids are responsible for the antioxidant effects of the mango leaves.

The existence of Phenolic was ascertained through the application of Ferric Chloride Test. Mango leaves have been reported to have antioxidant activity mostly from phenolic compounds (Ali et al. 2020). **Phenols**. Phenols generally protect plants and human cells from oxidative damage (Ali et al. 2020).

The same procedure were conducted to Tannins through the application of Ferric Chloride Test indicating positive result. **Tannins**. Tannins (commonly referred to as tannic acid) are water-soluble polyphenols that are present in many plant foods. They have been reported to be

responsible for decreases in feed intake, growth rate, feed efficiency, net metabolizable energy, and protein digestibility in experimental animals (Chung, K. T *et. al*., Wong 1998).

Saponins. Plant Saponins help human to fight fungal infections, combat microbes and viruses, boost the effectiveness of certain vaccine and knock out ome kins of tumor cells particularly lung and blood cancers. Saponins natural tendency to ward off microbes makes them good candidates for treating fungal and yeast infections. These compounds served as natural antibiotics, helping the body to fight infections and microbial invasion D. E. Okwu and I. N. Emenike (2006).

Steroids. Steroids detected as negative in *Mangifera indica*. The study showed inconfirmity with the study of Doughari and Manzara on *In Vitro antibacterial activity of crude leaf extracts of M. indica*, the preliminary phytochemical analysis revealed the presence of tannins, glycosides, saponins, and phenols.

Keller-Kiliani test was tested to determine cardiac glycosides and indicates positive. Similar to the study of (Ghossh *et. al.,* 2022). Cardiac glycosides are used to treat congestive heart failure and cardiac arrhythmia. Therefore, mango leaf extract might be used for cardiac ailments.

Brine Shrimp Lethality Test

The results for the brine shrimp lethality test are shown in Table 2.

Table 2. Brine Shrimp Lethality Test of *Mangifera Indica leaves.*

Concentration	%Mortality LC_{50}
500	70
1000	90
1500	50 3090.29
2000	90
2500	20

Concurrently with Phytochemical Screening. The toxicity test exhibited through brine shrimp lethality test to assess the cytotoxicity of leaf extracts and determine its safety from *Mangifera Indica*. Brine Shrimp Lethality Test (BSLT) essential method which used Artemia Salina Leach Larvae for toxicity test and present Lethal Concentration 50 (LC_{50}) value.

Toxicity assessment of *Mangifera indica* was done using (LC_{50}). The probit analysis showed that LC_{50} value obtained from the extract of mango *Mangifera indica* was 3090.29 ppm in the toxic category which indicates a non-toxic classification. Plant methanol extract LC_{50} values below 1000 ppm were classified as cytotoxic, as specified by (Gupta *et al.*, in 1996). Similar to (Ahomadegbe *et. al.,* 2018) stated that the extracts of *Mangifera indica* showed positive results (lethality) on the Brine Shrimp larvae indicating that the test samples are biologically active. Toxicological studies was performed on mango leaf extract (*Mangifera indica*) containing 60% mangiferin (MLE). No evidence of genotoxicity was found in a bacterial reverse mutation test (Reddeman *et.al.,* 2019).

With LC_{50} value of 3090.29 ppm, the mango extract showed significantly low toxicity to seawater compared to the standard toxicity threshold (1000 µg/mL) for plant extracts. The results suggest the extract poses minimal risk to seawater life at the concentrations tested. The absence of toxicity in the Brine Shrimp Lethality Test further corroborates these findings, highlighting the potential of this extract as a candidate for further research in pharmacology and therapeutic applications.

CONCLUSION

The findings of this study showed that the bioactive compounds found in *Mangifera indica* leaves possesses a diverse range that may contribute to the plant's medicinal properties, supporting its use in traditional medicine and its potential role in contemporary therapeutic applications. Moreover, the LC_{50} result value of *Mangifera indica* obtained 3090.29 ppm pointing as non-toxic, suggesting its safe use in traditional and modern medicine. While this is encouraging, further research is crucial to assess its safety in other systems, explore its therapeutic potential, and understand its mechanisms of action before developing any mango-derived products.

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