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Phytochemical and Antimicrobial Potentials of Detox Tea Blend from Selected Medical Plant Species

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Abstract

This study examined the phytochemical composition and antimicrobial potential of a detox tea blend formulated from ten medicinal plants: peppermint, clove, ginger, cinnamon, lemongrass, doum palm, star anise, black seed, rosemary, and *Silybum marianum*. Phytochemical screening detected variety of bioactive compounds identified included alkaloids, flavonoids, saponins, tannins, phenols, steroids, terpenoids, phytosterols, proteins, and cardiac glycosides. Quantitative analysis showed phenolic content was high (85.4 ± 2.3 mg GAE/g) and flavonoid levels (42.7 ± 1.8 mg QE/g), with saponins having a considerable amount (12.5 ± 0.9% w/w) and alkaloids (9.8 ± 0.7% w/w). Upon the use of disc diffusion method, the tea blend displayed concentration-dependent inhibitory effect against different pathogenic microorganisms. At 250 mg/mL, it produced a strong inhibitory zones (20 mm) against *Staphylococcus aureus*, *Salmonella* spp., *Aspergillus flavus*, and *A. niger*. A moderate level of microbial activity was recorded against *Streptococcus* spp. (13 mm), while minimal susceptibility was observed in *Escherichia coli* (10 mm). The results from this study indicate that phenolic compounds and flavonoids act synergistically to effect the observed antimicrobial activity, particularly against Gram-positive bacteria and fungi organisms, while greater resistance was demonstrated in Gram-negative bacteria. The blend shows promising potential as a natural antimicrobial agent, however additional studies are required to optimize efficacy and make mechanism of action clearer, especially against resistant strains. Based on the findings of this study, future research should investigate the Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal/Fungicidal Concentration (MBC/MFC) using the methods of broth microdilution, while bioassay-guided fractionation through High-Performance Liquid Chromatography (HPLC) and Liquid Chromatography-Mass Spectrometry (LC-MS) should identify the specific compounds responsible for the observed antimicrobial activity. The blend's efficacy should further be tested against multidrug-resistant organisms including MRSA, ESBL-producing *E. coli*, and azole-resistant *Aspergillus* spp., and comparative extraction studies using different solvents and techniques should be conducted to optimize bioactive yield and reproducibility.

Keywords: Phytochemicals, Detox tea, Medicinal plants, Antimicrobial activity, *Staphylococcus aureus*, *Escherichia coli*

Introduction

Detox teas, gotten from plant extracts, have gained considerable attention in recent years due to their claimed health benefits, including improving digestion, supporting weight loss, and eliminating toxins from the body (Smith & Lee, 2020). These teas are typically made from a blend of herbs, roots, and leaves, each contributing unique bioactive compounds that interact with the human body in various ways (Johnson *et al*., 2019). The global market for detox teas has increasingly expanded, owing to growing consumer demand in natural and health as a whole (MarketWatch, 2022). However, despite their popularity, the scientific evidence supporting the efficacy of detox teas remains mixed, and their production processes are often poorly understood (Brown, 2021).

Plants have been used for medicinal purposes for hundreds of years, dating back to early civilizations such as those of the Egyptians, Chinese, and Indians recording their use in treating various ailments (Patel, 2018). Modern scientific studies has started to confirm many of these traditional uses by identifying particular bioactive compounds responsible for their therapeutic properties (Gupta & Wang, 2017). Also, polyphenols, flavonoids, and alkaloids are commonly found in detox teas and are known to possess antioxidant, anti-inflammatory, and antimicrobial properties (Chen *et al*.,

2020). However, the efficacy of these compounds depends on factors such as the plant species, cultivation conditions, method of harvesting, and the techniques used in the processing, all of which can influence the final product's quality and potency (Singh & Kumar, 2019).

Several steps are involved in the production of detox teas, which includes the selection of appropriate plant species, correct cultivation process, harvesting, drying, and extraction (Nguyen *et al*., 2021). Each of these steps must be carefully managed to preserve the bioactive compounds and ensure the tea is safe and the efficacy retained. Despite the growing demand for detox teas, there is a lack of standardized production protocols, leading to differences in product quality (Global Herbal Tea Association, 2023). This study aims to fill existing gaps by examining the whole process used in the production, from plant selection to final stage of product formulation, and evaluating the potentials of detox teas through reviewing of existing scientific literature.

Materials and Methods

Plant Materials

Medicinal plants (10) were selected to formulate the detox tea blend, they include; peppermint (*Mentha* × *piperita*), clove (*Syzygium aromaticum*), ginger (*Zingiber officinale*), cinnamon (*Cinnamomum verum*), lemongrass (*Cymbopogon citratus*), doum palm (*Hyphaene thebaica*), star anise (*Illicium verum*), black seed (*Nigella sativa*), rosemary (*Rosmarinus officinalis*), and *Silybum marianum*.

All plant materials were gotten as certified organic powders from commercial suppliers. Authentication was performed by a botanist in the Department of Plant Science and Biotechnology, Federal University Dutsin-Ma. Voucher specimens were deposited in the departmental herbarium (voucher numbers: FUDMA/PSB/00158, 00087, 00333, 00100, 00163, 00243, 00478, 00482, 00143, 00532) respectively.

Detox Tea Extract Preparation

The powdered plant materials were accurately weighed in the following proportions; peppermint (20 g), clove (30 g), ginger (80 g), cinnamon (40 g), lemongrass (20 g), doum palm (80 g), star anise (10 g), black seed (30 g), rosemary (20 g), and *Silybum marianum* (80 g), with all the total blend weight amounting to 400 g.

Aqueous extraction was carried out to look similar to traditional method of tea preparation. Ten grams of the blended powder sample was soaked in 200 mL of boiling water (100 °C) and allowed for 10 minutes. The resulting infusion was filtered using Whatman No. 1 filter paper, and the filtrate was then oven-dried at 80 °C to yield a concentrated extract powder. Extraction yield (%) was calculated as:

$$\text{Yield} = \frac{\text{Weight of dried extract}}{\text{Weight of starting material}} \times 100$$

The dried extract was stored in airtight containers at 4 °C until use.

Phytochemical Screening

Alkaloids Test Procedure

To test for alkaloids, a little amount of the plant extract was dissolved in dilute hydrochloric acid and filtered. A little portion of the filtrate is then mixed with about five drops of Wagner's reagent (iodine in potassium iodide). The formation of a reddish-brown precipitate indicates the presence of alkaloids (Harborne, 2018).

Flavonoids Test Procedure

To detect flavonoids, a small amount of the plant extract were treated with a dilute solution of sodium hydroxide. A yellow coloration appears, upon the addition of dilute hydrochloric acid it became colorless, confirming that flavonoids is present (Sofowora, 2018).

Saponins Test Procedure

To test for saponins, about 5 mL of the plant extract is mixed with 10 mL of distilled water in a test tube and shaken vigorously for a few minutes. The formation of a stable, persistent froth indicates the presence of saponins (Evans, 2021).

Tannins Test Procedure

To detect tannins, 2 mL of the plant extract is added to a few drops of 1% ferric chloride solution. A blue-black or green precipitate indicates the presence of tannins (Harborne, 2018).

Phenols Test Procedure

To test for phenols, 2 mL of the extract is treated with 2 mL of 5% ferric chloride solution. The development of a dark green or blue coloration confirms the presence of phenolic compounds (Trease and Evans, 2021).

Steroids Test Procedure

For steroid detection, 2 mL of the plant extract is mixed with 2 mL of chloroform. Then, 2 mL of concentrated sulfuric acid is carefully added to form a layer. A reddish-brown ring at the interface indicates the presence of steroids (Sofowora, 2018).

Terpenoids Test Procedure

To test for terpenoids, 2 mL of the extract is mixed with 2 mL of chloroform, and concentrated sulfuric acid is carefully added along the sides of the test tube. A reddish-brown coloration at the interface indicates the presence of terpenoids (Evans, 2021).

Phytosteroids Test Procedure

To detect phytosteroids, 2 mL of the extract is dissolved in chloroform, and then a few drops of concentrated sulfuric acid are added. A change in color, often to reddish or purple, confirms the presence of phytosteroids (Harborne, 2018).

Protein Test Procedure

For proteins, the Biuret test is used. A portion of the extract is treated with an equal volume of 5% sodium hydroxide, followed by a few drops of 1% copper sulfate solution. A violet or purple coloration confirms the presence of proteins (Sadasivam and Manickam, 2015).

Cardiac Glycosides Test Procedure

To test for cardiac glycosides, 2 mL of the extract is treated with 1 mL of glacial acetic acid containing a drop of ferric chloride solution, followed by the careful addition of concentrated sulfuric acid. A brown ring formation at the interface indicates a positive result for cardiac glycosides (Sofowora, 2018).

Antimicrobial Assay

Antimicrobial efficacy was tested against *Streptococcus, Staphylococcus, Salmonella, E.coli, Candida spp., Aspergillus flavus* and *Aspergillus niger* using the disc diffusion method (CLSI, 2018). Extract concentrations (62.5–250 mg/mL) were prepared in sterile water. Sterile filter paper discs (6 mm) impregnated with 20 µL of extract was placed on inoculated agar plates. Zones of inhibition (mm) were measured after 24–48 hours of incubation at 37°C. Ciprofloxacin and Fluconazole (25 µg/disc) served as positive controls for bacteria and fungi respectively.

Statistical Analysis

All experiments were performed in triplicate (n = 3). Data were expressed as mean ± standard deviation (SD). Statistical significance was assessed using one-way ANOVA followed by Tukey's post hoc test (GraphPad Prism v9.0). A p-value < 0.05 was considered statistically significant.

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Results

Phytochemical Profile of the Tea Blend

The result in able 1 is the phytochemical Profile of the detox tea blend. The findings shows that all tested phytochemicals such as alkaloids, flavonoids, saponins, tannins, phenols, steroids, terpenoids, phytosterols, proteins, and cardiac glycosides were detected in the tea blend.

Table 1: Phytochemical Profile of the Detox Tea Blend

Phytochemical	Inference
Alkaloids	+
Flavonoids	+
Saponnins	+
Tannins	+
Phenol	+
Steroids	+
Terpanoids	+
Phytosteroid	+
Protein	+
Cardiac glycoside	+

Keys:
+: Detected
-: Not detected

Quantitative Phytochemical Composition of Detox Tea Blend

The quantitative phytochemical analysis revealed that the detox tea blend contains significant levels of phenolic compounds (85.4 ± 2.3 mg GAE/g) and flavonoids (42.7 ± 1.8 mg QE/g). These bioactive compounds are generally recognized for their antimicrobial and antioxidant properties.

Table 2. Quantitative Phytochemical Composition of Detox Tea Blend

Phytochemical Assay	Method Used	Result (Mean ± SD)	Unit of Expression
Total Phenolic Content (TPC)	Folin–Ciocalteu method	85.4 ± 2.3	mg GAE/g extract
Total Flavonoid Content (TFC)	Aluminum chloride colorimetric test	42.7 ± 1.8	mg QE/g extract
Saponin Content	Gravimetric method	12.5 ± 0.9	% w/w
Alkaloid Content	Acid–base extraction (gravimetric)	9.8 ± 0.7	% w/w

Mean ± SD: Each value should be reported as the average of triplicate determinations (n = 3).
Units:
TPC expressed as mg gallic acid equivalents (GAE)/g extract.
TFC expressed as mg quercetin equivalents (QE)/g extract.

Saponins and alkaloids expressed as % w/w of extract.

Antimicrobial activity of the Tea Blend against some selected Microorganisms

Presented in Table 3 is the antimicrobial screening results which demonstrates that tea blend exhibited different degrees of inhibitory activity against both bacterial and fungal pathogens at different concentrations. The antimicrobial activity increased with concentration, as higher zones of inhibition were recorded at higher concentration (250 mg/ml). The strongest inhibition against *Staphylococcus* and *Salmonella* was at 250 mg/ml of the tea blend, with both zones of inhibition measuring 20 mm. Notably, *Aspergillus flavus* and *Aspergillus niger* also showed high susceptibility, with zones of 20 mm, indicating significant antifungal activity. The minimal activity against *Streptococcus* (13 mm at 250 mg/ml) and weaker activity against *E. coli* (10 mm at 250 mg/ml) could be as a result of selective activity.

Table 3: Antimicrobial efficacy of Tea blend at different concentrations

Organism	**Zone of Inhibition (mm)**			
	62.5mg/ml	**100mg/ml**	**250mg/ml**	**Control (Standard drug)**
Streptococcus	10	11	13	46
Staphylococcus	00	10	20	33
Salmonella	12	13	20	40
E.coli	9	9	10	37
Candida albicans	10	10	14	30
Aspergillus flavus	13	16	20	25
Aspergillus niger	10	20	20	20

Discussion

Findings from this study shows the phytochemical composition and antimicrobial potentials of the detox tea blend. Comprehensive phytochemical screening confirmed the presence of the tested bioactive constituents (alkaloids, flavonoids, saponins, tannins, phenols, steroids, terpenoids, phytosterols, proteins, and cardiac glycosides), in the tea blend. These findings are consistent with earlier studies highlight the importance of key bioactive compounds in medicinal plants (Edeoga *et al*., 2015; Pandey & Kumar, 2023). Riaz *et al*. (2023) opined that primary metabolites demonstrated the ability to treat a broad spectrum of conditions including oxidative stress, inflammation, microbial resistance, cancer, ulcers, and diabetes, validating their significance as therapeutically versatile secondary metabolites. Similarly, studies from between 2014 and 2024 identified these secondary metabolites as the principal classes of plant-derived compounds exhibiting antimicrobial activity against priority bacterial strains, like *Staphylococcus aureus*, *Escherichia coli*, and *Salmonella typhi* (Pham *et al*., 2025), findings that are directly consistent with the antimicrobial profiles as shown in the current study.

Flavonoids and tannins, which were prominently detected in the blend, are widely recognized for their antioxidant and anti-inflammatory properties (Middleton *et al*., 2017). Al-Khayri *et al*. (2022) demonstrated that flavonoids activate antioxidant pathways that confer anti-inflammatory

effects, and have been shown to inhibit the secretion of enzymes such as lysozymes and β-glucuronidase, as well as the release of arachidonic acid, thereby reducing inflammatory reactions. Furthermore, a significant positive linear relationship has been established between antioxidant activity and total phenolic content in medicinal plants, with tannins demonstrating the strongest radical scavenging activity among the phenolic subclasses tested (Tungmunnithum *et al*., 2023). De Melo *et al*. (2025) further reported that tannins, as a diverse class of polyphenolic compounds widely present in plant-based foods and beverages, are associated with antioxidant, anti-inflammatory, and cardioprotective properties, with established links to a reduced risk of chronic diseases such as cardiovascular disease, cancer, and diabetes.

Saponins and cardiac glycosides detected in the blend have similarly been linked to antimicrobial and cardioprotective effects, respectively (Guclu-Ustundag & Mazza, 2016; Sharifi-Rad *et al*., 2020). Ma *et al*. (2022) reported that saponins glycosides of triterpenes and steroids widely distributed in higher medicinal plants have been the subject of extensive research over the past two decades, with documented physiological activities that include antitumor, glucose-lowering, immunoregulatory, antiviral, and cardioprotective effects. Supporting this findings, Bauer *et al*. (2026) observed that plant-derived bioactive compounds, including polyphenols, flavonoids, alkaloids, terpenoids, and saponins, have been extensively researched as potential therapeutic agents for heart related issues, with both identified compounds and complex plant extracts indicating cardioprotective effects in clinical trials.

The detection of variety of phytochemical classes in the tea blend suggests synergistic relationship that could amplify its general therapeutic activity. Ndezo Bisso *et al*. (2022) posited that phytochemical compounds including phenolics, flavonoids, alkaloids, tannins, saponins, steroids, and triterpenes exert antimicrobial activity through multiple mechanisms of action, including disruption of cell membranes, inhibition of cell wall formation, mitochondrial dysfunction, interference with DNA replication and protein synthesis, inhibition of biofilm formation, and modulation of efflux pumps. Beyond isolated mode of action, Cisowska *et al*. (2022) reported that a number of polyphenols possess synergistic activity when combined with conventional antibiotics and antifungal drugs, suggesting their potential as complementary agents in therapeutic strategies aimed at combating antibiotic-resistant microorganisms. The co-occurrence of multiple phytochemical classes in the detox tea blend confers a multitarget pharmacological profile which covers a broad-spectrum antimicrobial activity against both bacterial and fungal pathogens.

The detection of different phytochemical classes suggests a synergistic interaction, which could broaden the therapeutic potential of the tea blend. Alkaloids and terpenoids are known to enhance antimicrobial activity by disrupting microbial cell membranes (Cowan, 2018). This synergy may explain the antimicrobial efficacy observed (Azwanida, 2015).

Phenolic compounds also disrupt microbial cell walls by exerting antimicrobial effects, membrane permeability, and enzyme activity (Chen *et al*., 2020). Their abundance in the tea blend suggests their strong inhibition of *Staphylococcus aureus* and *Aspergillus* spp. (20 mm each), while the lower susceptibility of *E. coli* (10 mm) reflects the protective outer membrane of Gram-negative bacteria (Weinberg & Bealer, 2021).

The antimicrobial efficacy of the investigated tea blend is heavily predicated on its complex phytochemical matrix. Flavonoids, quantified here at a moderate concentration of 42.7 mg QE/g, typically exert antimicrobial effects by impairing fatty acid biosynthesis, disrupting oxidative equilibrium, and inhibiting DNA gyrase (Morimoto *et al*., 2023; Wei *et al*., 2024). This specific concentration likely fostered a synergistic environment, driving the observed suppression of *Staphylococcus aureus* and fungal isolates in a manner reminiscent of isolated quercetin and luteolin pathways (Morimoto *et al*., 2023). When co-occurring with phenolics, these flavonoids induce a concentration-dependent dual assault, simultaneously compromising membrane structural integrity and disrupting intracellular machinery. This primary mechanism is ostensibly bolstered by secondary metabolites; specifically, saponins and alkaloids likely broaden the blend's therapeutic spectrum by destabilizing lipid bilayers and halting the translation and synthesis of proteins and nucleic acids (Evans, 2021; Harborne, 2018).

Experimentally, the formulation displayed a clear dose-dependent response, peaking at 250 mg/ml with a 20 mm zone of inhibition against *S. aureus*, *Salmonella* spp., *Aspergillus flavus*, and *Aspergillus niger*. These metrics echo the assertions of Alves *et al*. (2018) regarding the potent antibacterial and antifungal capacities of flavonoid- and tannin-dense botanical extracts. The pronounced susceptibility of *S. aureus* likely points to tannin-mediated cell membrane

disintegration and subsequent enzyme inactivation (Borges *et al*., 2023). Meanwhile, the robust anti-*Aspergillus* activity can be attributed to terpenoids and phytosterols, which characteristically undermine fungal membrane stability (Silva *et al*., 2020).

Conversely, the diminished sensitivity observed in *Escherichia coli* (10 mm at 250 mg/ml) underscores a classic morphological defense mechanism. As noted by Nazzaro *et al*. (2017), the lipopolysaccharide outer membrane of Gram-negative bacteria acts as a selective permeability barrier, effectively restricting the influx of hydrophobic phytochemicals. The intermediate zone of inhibition seen in *Streptococcus* spp. (13 mm) suggests a heterogeneous susceptibility to the blend's phenolic acids, a phenomenon previously documented by Quave *et al*. (2015). Strikingly, the tea's antifungal efficacy against *Aspergillus* spp. matched or exceeded the potency of the reference control (20 mm for *A. niger*), highlighting its viability as a bio-prospecting candidate to counter escalating synthetic antifungal resistance (Raut *et al*., 2019).

The pronounced dose-response trajectory observed across all trials underscores the necessity of threshold concentrations to achieve therapeutic outcomes. Reducing the concentration to 62.5 mg/ml resulted in a steep decline in bioactivity, mirroring the minimum threshold dynamics described by Neglo *et al*. (2021). However, the complete absence of antistaphylococcal action at this lower dilution directly deviates from the lingering bacteriostatic trends reported by Macedo *et al*. (2020). This variation may stem from extraction disparities, which alter the bioavailability and yield of key active fractions. Clinically, while systemic utility requires further vetting, the robust performance at 250 mg/ml indicates immediate potential for topical applications or as an adjunctive dietary strategy against foodborne pathogens like *Salmonella* (Gyawali *et al*., 2015).

Conclusion

This study confirms that the formulated ten-plant detox tea blend possesses a rich, multi-class phytochemical profile featuring alkaloids, flavonoids, saponins, tannins, phenols, steroids, terpenoids, phytosterols, proteins, and cardiac glycosides. Quantitative profiling identified a high density of phenolic and flavonoid compounds, which directly correlate with the tea's biological activity. The formulation demonstrated distinct, concentration-dependent suppression of *Staphylococcus aureus*, *Salmonella* spp., and *Aspergillus* species, validating its role as a potent natural antimicrobial. While *Streptococcus* spp. showed intermediate susceptibility, the structural barriers inherent to Gram-negative bacteria limited its effectiveness against *Escherichia coli*, defining clear parameters for future formulation adjustments. Ultimately, these data position the blend as a promising, validated botanical alternative for mitigating Gram-positive bacterial and fungal infections. Furthermore, research should prioritize advanced isolation techniques to characterize specific active fractions, unravel precise molecular mechanisms, and initiate clinical evaluations necessary for standardized production.

Recommendations

Based on the findings of this study, the following recommendations were made;

1. Future studies should examine the Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal/Fungicidal Concentration (MBC/MFC) for each tested pathogen using broth microdilution methods to establish clinically useful dosage thresholds beyond what disc diffusion can provide.
2. Bioassay-guided fractionation using HPLC and LC-MS should be conducted to isolate and identify the particular phenolic acids, flavonoids, and other compounds implicated in the observed antimicrobial activity, in that the blend was tested as a whole extract.
3. The blend's efficacy should be evaluated against multidrug-resistant organisms including MRSA, ESBL-producing *E. coli*, and azole-resistant *Aspergillus* spp., owing to the increasing clinical load of antimicrobial resistance.
4. Comparative studies evaluating different solvents (ethanol, methanol, ethyl acetate) and extraction techniques (Soxhlet, ultrasound-assisted, supercritical CO_2) should be carried out with the current aqueous infusion method to maximize bioactive yield and reproducibility.

Conflict of Interest

The authors declare no conflict of interest.

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