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COMPARATIVE ANTIFUNGAL ACTIVITY of FLAVONOID EXTRACTS FROM *Senna siamea* AND *Cnidoscolus chayamansa* AGAINST BANANA CROWN FUNGI

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Abstract

This study compared the antifungal efficacy of flavonoid extracts from *Senna siamea* and *Cnidoscolus chayamansa* against fungi isolated from banana crown using Potato Dextrose Agar (PDA) medium. Leaf extracts were prepared using ethanol maceration (1:10 w/v) for 24–48 h under dark conditions. Antifungal activity was evaluated by measuring radial fungal growth for 7 days at room temperature. The mean fungal growth diameter in the *S. siamea* treatment was 1.526 cm, slightly lower than in the *C. chayamansa* treatment 1.571 cm, The extracts from *Cnidoscolus chayamansa* and *Senna siamea* significantly inhibited fungal dispersion compared with the control group ($p < 0.05$). However, non statistically significant difference was observed between the extracts derived from the two plant specie

Keywords**:** *Cnidoscolus chayamansa, Senna siamea, flavonoid, Potato dextrose agar (PDA)*

INTRODUCTION

Fungal contamination in food poses potential health risks to consumers [8] A comprehensive large-scale survey in Northern and Northeastern Brazil demonstrated that banana crown rot is caused by a complex of at least six phylogeneticallydistinct *Lasiodiplodia* species, highlighting the diversity and pathogenicity of fungi associated with this disease [5] Consequently, inhibiting fungal growth in fruits and vegetables is essential for food preservation, household consumption, and export quality. In Thailand, bananas represent an economically important crop, with three major types commonly cultivated for commercial purposes: Golden Banana, Cavendish Banana, and cultivated banana. Most banana production is consumed domestically due to the fruit's high perishability. Environmental factors such as high humidity and elevated temperatures, along with biochemical

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Jurnal Pendidikan Matematika dan IPA
Vol. 17, No. 2 (2026) h. 323-330

processes that promote fungal growth and rapid ripening, contribute to postharvest losses. Banana ripening is accelerated by polyphenol oxidase activity in the presence of oxygen, leading to browning, tissue degradation, and increased susceptibility to mold growth. Additionally, storage in humid conditions or proximity to ethylene-producing fruits further hastens ripening and fungal development.

Currently, synthetic chemicals are widely used to delay spoilage and extend shelf life; however, chemical residues may induce allergic reactions and pose long-term health risks, including cancer and damage to the liver, kidneys, and nervous system when consumed excessively or continuously. Therefore, the application of natural extracts as antifungal agents represents a safer alternative, reducing chemical contamination while offering potential long-term health benefits.

The numerous plant extracts have demonstrated effective antifungal properties, particularly herbal extracts and essential oils such as oregano and cinnamon oils[1] ,[12] These extracts significantly inhibit spore germination and hyphal growth of Penicillium digitatum, a major causative agent of fruit mold. Concentrated cinnamon and oregano extracts show high efficacy in suppressing fungal development, while grape seed extract has also exhibited antifungal activity under laboratory and practical storage conditions. Owing to its antioxidant content and consumer safety, grape seed extract has potential applications across various industries, including food packaging for shelf-life extension.

In Thailand, several indigenous plants possess antifungal properties, notably Cassia leaf *(Senna siamea*) and Chaya leaf (C*nidoscolus chayamansa*). Extracts from *Senna siamea* leaves have been reported to inhibit the growth of fungi and bacteria, particularly banana rot pathogens such as *Fusarium sp.* [9] and *Lasiodiplodia theobromae*, which accelerate postharvest deterioration. High concentrations of Senna leaf extract effectively suppress spore germination and slow the growth of fungi causing anthracnose in golden bananas. When applied appropriately, such natural extracts may provide a sustainable and safe strategy for controlling postharvest spoilage in bananas and other fruits without reliance on synthetic chemicals.

Objective

To compare the antifungal Lasiodiplodia sp. efficacy of flavonoid extracts from Cassia leaf (Senna siamea) and Chaya leaf (Cnidoscolus chayamansa).

Hypothesis

The flavonoid extract from *Senna siamea* exhibits greater antifungal activity than extract from C*nidoscolus chayamansa.*

METHOD

Preparation of Plant Materials and Extraction of Flavonoid

Fresh leaves of Cassia leaf (*Senna siamea*) and Chaya leaf (*Cnidoscolus chayamansa*) were collected and used as plant materials in this study. The leaves were washed thoroughly with tap water followed by distilled water to remove dirt and contaminants, then wiped dry with

Jurnal Pendidikan Matematika dan IPA
Vol. 17, No. 2 (2026) h. 323-330

clean tissue paper.The cleaned leaves were sliced into small pieces and dried in a hot air oven at 60 °C until brittle. The dried samples were ground into a fine powder using a laboratory blender.

Anthraquinone extraction was performed using three independent biological samples for each plant species (n = 3). For each biological sample, three technical replicates were prepared.

Fifty grams of powdered leaves were soaked in ethanol at a 1:10 (w/v) ratio (50 g powder in 500 mL ethanol). The mixtures were kept in a dark and cool place at room temperature for 24–48 h with occasional shaking. After extraction, the solutions were filtered through Whatman No. 1 filter paper and concentrated under reduced pressure until approximately 50% of the original volume was obtained. The crude extracts were stored in amber glass bottles and kept at 4 °C until further use. Prior to antifungal testing, the extracts were diluted with sterile distilled water to obtain final concentrations of 25, 50, and 100 mg/mL.

Preparation of Culture Medium

Potato dextrose agar (PDA) was prepared by washing and peeling fresh potatoes, followed by cutting them into small cubes. Two hundred grams of potato were boiled in 1 L of distilled water until softened. The potato broth was separated from the solid residues.

To the broth, 20 g of glucose and 16 g of agar were added and dissolved completely. The medium was sterilized by autoclaving at 121 °C for 15 min.Sterile Petri dishes were prepared by marking two perpendicular lines across the bottom to divide the plate into four quadrants. The center point was designated for fungal inoculation, and one point in the center of each quadrant was marked for extract placement. The sterilized medium was poured into the Petri dishes and allowed to solidify.

A total of 16 Petri dishes were prepared: 5 dishes for *Senna siamea* extract 5 dishes for *Cnidoscolus chayamansa* extract 3 dishes for negative control (solvent only) and 3 dishes for fungal growth control.

Fungal Inoculation and Identification

The fungal pathogen used in this study was isolated from infected banana fruit and identified as *Lasiodiplodia* sp. based on colony morphology and microscopic characteristics. A 5-mm diameter mycelial plug was taken from an actively growing culture and placed at the center of each PDA plate. The plates were incubated at 25 °C for 1–2 days to allow initial fungal growth before extract application.

Preparation of Extract Discs and Antifungal Assay

The antifungal activity of anthraquinone extracts was evaluated using the disc diffusion method. Sterile filter paper discs (6 mm diameter) were prepared. Each disc was impregnated with 20 µL of extract, resulting in final doses of 0.5, 1.0, and 2.0 mg per disc for extract concentrations of 25, 50, and 100 mg/mL, respectively. The discs were air-dried under sterile conditions to remove excess solvent. Control discs were prepared using sterile distilled water or ethanol as negative controls. The impregnated discs were placed on the marked positions in each quadrant of the Petri dishes, approximately 2 cm from the

Jurnal Pendidikan Matematika dan IPA
Vol. 17, No. 2 (2026) h. 323-330

fungal inoculation point. All plates were incubated at 25 °C for 7 days, and fungal growth was observed daily.

Measurement of Antifungal Activity

Antifungal activity was assessed by measuring the diameter of the inhibition zone (mm) surrounding each disc. Measurements were taken in two perpendicular directions using a digital caliper, and the mean value was calculated. Results were expressed as mean ± standard deviation (SD).

Experimental Design and Statistical Analysis

The experiment was conducted using a completely randomized design (CRD) with: 3 biological replicates 3 technical replicates per treatment Differences between treated and control groups were analyzed using Student's t-test, which was considered appropriate due to the relatively small differences observed among treatments. Statistical significance was set at $p < 0.05$.

RESULT AND DISCUSSION

The Results found that the extracts from the leaves of Cassia leaf (*Senna siamea*) [2] were more effective in slowing down the growth of fungi than the extracts from the leaves of Chaya leaf (*Cnidoscolus chayamansa*), Which was known from the observation of the growth of the fungi over 7 days. It was found that the average growth distance of the fungi on the culture medium of the Cassia leaf (*Senna siamea*) leaf extract experiment set grew at 1.526 centimeters, which was lower than the average growth distance on the culture medium of the Chaya leaf extract experiment set, which grew at 1.571 centimeters. As shown in the table1.

Table 1. the average distribution of fungi (centimeter) over 7 days.

Treatment	**n**	**Mean**	***SD***
Controlled group	7	3.18	0.39
Cnidoscolus chayamansa	7	1.571	0.42
Senna siamea	7	1.526	0.41

Table2. Statistical test results (Independent samples t-test)

Comparison	**t**	**df**	**p**
Control vs *C. chayamansa*	7.37	12	0.001*
Control vs *S. siamea*	7.73	12	0.001*
C. chayamansa vs *S. siamea*	0.21	12	0.84

*statistic significant .05

From the table 2 The extracts from *Cnidoscolus chayamansa* and *Senna siamea* significantly inhibited fungal dispersion compared with the control group ($p < .05$). However, non statistically significant difference was observed between the extracts derived from the two plant specie. The results of this study demonstrate that both *Cnidoscolus chayamansa* and *Senna siamea* leaf extracts significantly inhibited fungal dispersion on banana crown compared with the control. The present study demonstrated that leaf

Jurnal Pendidikan Matematika dan IPA
Vol. 17, No. 2 (2026) h. 323-330

extracts from *Cnidoscolus chayamansa* and *Senna siamea* significantly inhibited fungal dispersion on banana crowns when compared with the control treatment. The control group exhibited extensive fungal growth, with a mean radial expansion of 3.18 ± 0.39 cm after seven days of incubation, whereas fungal growth in media supplemented with *C. chayamansa* and *S. siamea* extracts was markedly reduced to 1.571 ± 0.42 cm and 1.526 ± 0.41 cm, respectively. Statistical analysis using an independent samples t-test confirmed that both plant extracts significantly suppressed fungal growth relative to the control ($p < 0.05$). However, no statistically significant difference was observed between the two plant extracts ($p = 0.84$), indicating that both species exhibited comparable antifungal activity under the experimental conditions.

Despite the absence of statistical significance between the extracts, *S. siamea* showed a slightly greater inhibitory effect on fungal growth than *C. chayamansa,* as reflected by the lower mean fungal growth distance. This trend suggests a potential difference in antifungal potency that may be attributed to variations in phytochemical composition. The assessment method, based on measuring radial fungal dispersion from the inoculation point on Potato Dextrose Agar (PDA) after a seven-day incubation period, is widely accepted as a reliable indicator of antifungal efficacy in in vitro studies.

These findings are consistent with previous international research demonstrating the antifungal potential of plant-derived extracts against banana pathogens. [3] reported that medicinal plant extracts significantly inhibited the growth of major banana crown rot pathogens, including *Lasiodiplodia theobromae* and *Colletotrichum musae*, in both in vitro assays and postharvest banana fruit experiments. Similarly, [7] found that leaf extracts from various plant species effectively reduced spore germination and mycelial extension of *Mycosphaerella fijiensis*, the causal agent of Black Sigatoka disease in banana. These studies support the current results, reinforcing the role of plant extracts as natural antifungal agents for managing banana-associated fungal diseases.

The antifungal activity observed in this study is likely related to the presence of bioactive secondary metabolites, particularly flavonoids and phenolic compounds. Flavonoids are well documented for their broad biological activities, including antibacterial, antifungal, antiviral, and antioxidant properties Differences in antifungal efficacy between *S. siamea* and *C. chayamansa* may be explained by variations in the type and concentration of flavonoids present in each species. Previous studies have reported that *S. siamea* contains higher concentrations of flavonoids such as kaempferol and apigenin, which are known to possess strong antifungal properties [11]. Kaempferol, in particular, has been shown to disrupt fungal cell wall integrity and inhibit mycelial growth, leading to reduced fungal proliferation [6] This mechanism may account for the slightly enhanced inhibitory effect observed in the *S. siamea* treatment. Similar findings have been reported in other plant-based antifungal studies. [4] demonstrated that ethanolic plant

Jurnal Pendidikan Matematika dan IPA
Vol. 17, No. 2 (2026) h. 323-330

extracts rich in phenolic compounds, including kaempferol, significantly inhibited postharvest fungal pathogens such as Penicillium expansum and Botrytis cinerea by interfering with fungal membrane function and metabolic processes.

Furthermore, the antifungal effectiveness of *S. siamea* observed in this study aligns with reports on other flavonoid-rich plant extracts used against phytopathogenic fungi. [10] reported that extracts from Brazilian savanna plants exhibited strong antifungal activity due to their high phenolic and flavonoid content, suggesting their potential as environmentally friendly alternatives to synthetic fungicides. Comparable results have also been reported for extracts of Thunbergia laurifolia and galangal, which were shown to inhibit mycelial growth and spore germination of plant-pathogenic fungi, thereby reducing fungal reproduction.

Overall, the results of this study, together with supporting evidence from international literature, indicate that both *C. chayamansa* and *S. siamea* leaf extracts possess significant antifungal activity against fungi associated with banana crowns. Although both extracts were effective, *S. siamea* exhibited a slightly greater inhibitory effect, likely due to its richer flavonoid composition. These findings highlight the potential application of plant-derived extracts as natural, eco-friendly alternatives for controlling fungal diseases in bananas and other agricultural crops.

CONCLUSION

The results demonstrate clear differences in fungal growth among the experimental treatments over a 7-day period. As shown in Table 1, the controlled group exhibited the highest average fungal distribution (Mean = 3.18 cm, SD = 0.39). In contrast, fungal growth was markedly reduced in the treatments containing *Cnidoscolus chayamansa* (Mean = 1.57 cm, SD = 0.42) and *Senna siamea* (Mean = 1.53 cm, SD = 0.41).

The independent samples t-test results (Table 2) indicate that fungal growth in the control group was significantly greater than that observed in both the C. chayamansa treatment ($t = 7.37$, $df = 12$, $p < 0.05$) and the *S. siamea* treatment ($t = 7.73$, $df = 12$, $p < 0.05$). However, no statistically significant difference was detected between the *C. chayamansa* and *S. siamea* treatments ($t = 0.21$, $df = 12$, $p = 0.84$).

Overall, these findings suggest that both *C. chayamansa* and *S. siamea* effectively inhibited fungal growth when compared with the control, with no significant difference in antifungal efficacy between the two plant treatments at the 0.05 significance level.

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Jurnal Pendidikan Matematika dan IPA
Vol. 17, No. 2 (2026) h. 323-330

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Jurnal Pendidikan Matematika dan IPA
Vol. 17, No. 2 (2026) h. 323-330

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