Chemical Composition, Antioxidant and Antibacterial Activities of Extracts from *Sesbania sesban* Organs Growing in Egypt

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ABSTRACT

***Sesbania sesban* is a multipurpose leguminous tree widely utilized in agroforestry and traditional medicine. However, its phytochemical composition and bioactivities remain insufficiently characterized, particularly for tree organs cultivated in Egypt. To the best of our knowledge, this is the first comprehensive study to investigate the chemical composition, antioxidant potential, and antibacterial activities of methanolic extracts derived from various organs of *S. sesban*—including leaves, fruits, bark, wood branches, and roots. Gas chromatography–mass spectrometry (GC–MS) analysis revealed a diverse array of bioactive compounds, such as fatty acids (palmitic, oleic, linoleic), sterols (β-sitosterol, stigmasterol, campesterol), triterpenoids (lupeol, betulin, oleanolic acid, ursolic acid), phenolics, and vitamin E. Quantitative assays demonstrated significant organ-specific variations: leaves exhibited the highest total phenolic content (45.0 ± 2.0 mg TAE/g), flavonoid content (20.0 ± 1.0 mg CAE/g), and antioxidant activity (TAA = 70.0 ± 2.0%, IC_{50} = 22.0 ± 1.0 µg/mL). Antibacterial assessments showed that leaf extracts possessed notable inhibitory and bactericidal effects against both Gram-positive and Gram-negative bacteria (MIC 4–32 µg/mL, MBC 8–64 µg/mL), comparable to standard antibiotics. Bark and fruit extracts exhibited moderate activity, while root and wood branch extracts showed limited effects. The novelty of this study lies in its organ-specific and solvent-dependent phytochemical and bioactivity profiling of *S. sesban* cultivated in Egypt. These findings underscore the potential of *S. sesban* as a natural source of potent antioxidant and antibacterial agents, offering valuable insights for future nutraceutical, pharmaceutical, and industrial applications.**

Keywords: *Sesbania sesban*, phytochemistry, GC–MS, antioxidant activity, antibacterial activity, bioactive compounds.

INTRODUCTION

Sesbania sesban (Fabaceae) is a fast-growing leguminous tree that thrives in tropical and subtropical regions, particularly across Africa and Asia. In traditional medicine, various parts of the plant have long been used to treat ailments such as microbial infections, inflammation, dermatological disorders, and as a restorative tonic (Singh *et al.*, 2009 and Mokhtar *et al.*, 2025). Phytochemical investigations of *Sesbania* species have revealed a rich diversity of metabolites, including sterols, triterpenoids, flavonoids, phenolic acids and fatty acids compounds known for their antioxidant, antimicrobial, and anti-inflammatory properties (Mani *et al.*, 2011 and Akram *et al.*, 2021). Phenolics and flavonoids are among the most prominent plant-derived antioxidants, acting to neutralize free radicals and mitigate oxidative stress, thereby contributing to the prevention of chronic and degenerative diseases (Rice-Evans *et al.*, 1997 and Scalbert *et al.*, 2005). Additionally, terpenoids, sterols, and related secondary metabolites have garnered increasing attention for their antimicrobial effects, particularly their ability to disrupt microbial membranes and inhibit essential cellular functions (Cushnie & Lamb, 2011 and Daglia, 2012). Previous studies on *Sesbania* species, such as *S. grandiflora* and *S. sesban*, have reported notable antioxidant and antibacterial activities, underscoring the genus's potential as a natural source of bioactive compounds (Kumari *et al.*, 2017 and Rengarajan *et al.*, 2023). Despite these findings, detailed comparative studies on different organs of *S. sesban* cultivated in Egypt remain scarce. Most existing research has focused on leaves or seeds, while other parts such as bark, roots, and wood branches are largely unexplored. Moreover, solvent polarity plays a critical role in determining both the type and yield of extracted metabolites, yet systematic evaluations across different solvents and plant organs are still limited (Tiwari *et al.*, 2011). In this context, the present study was designed to profile the chemical constituents of methanolic extracts from leaves, fruits, bark, wood branches, and roots of *Egyptian S. sesban* using GC–MS analysis; quantify their total phenolic and flavonoid contents alongside antioxidant activities; and assess their antibacterial properties against representative Gram-positive and Gram-negative bacteria. This organ-specific and solvent-dependent approach offers novel insights into the phytochemistry and bioactivity of *S. sesban*, reinforcing its potential as a source of natural therapeutic agents.

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MATERIALS AND METHODS

Plant materials:

Samples of *Sesbania sesban* were collected in mid-September 2024 from three randomly selected trees growing in Matruh, Egypt. The plant material was taxonomically identified and confirmed by Assoc. Prof. Dr. Mohamed Zaky Zayed, Department of Forestry and Wood Technology, Faculty of Agriculture, Alexandria University. Various organs including leaves, fruits, bark, wood branches, and roots were harvested, shade-dried under ambient conditions, ground into a fine powder, and stored in airtight containers until subsequent extraction and analysis.

Sample extraction:

100 g of powdered material from each organ of *Sesbania sesban* (leaves, fruits, barks, wood branches, and roots) were placed in individual conical flasks and macerated with methanol to ensure full immersion. The mixtures were left to stand overnight at room temperature, followed by filtration through Whatman No. 1 filter paper containing 2 g of anhydrous sodium sulfate to remove suspended particles and traces of moisture. Before use, the filter paper and sodium sulfate were preconditioned with 95% ethanol. The resulting filtrates were concentrated to dryness under reduced pressure and preserved for subsequent phytochemical screening and GC–MS analysis (Zayed *et al.*, 2025).

Gas Chromatograph-mass Spectrometry (GC-MS):

Methanolic extracts from the leaves, fruits, bark, woody branches, and roots of *S. sesban* (cultivated in Egypt) were analyzed by GC–MS using a Thermo Scientific Trace GC 1300 system coupled with a TSQ 8000 Evo detector and equipped with a TG-5MS capillary column. Helium was employed as the carrier gas at a constant flow rate of 1.0 mL/min. Separation was performed using a DB-5 fused silica column (30 m × 0.25 mm, 0.25 µm film thickness, 5% phenyl-methylpolysiloxane). The oven temperature was initially maintained at 50 °C for 2 minutes, then ramped at 6.5 °C/min to reach 300 °C, where it was held for 10 minutes. The injector and detector temperatures were set at 280 °C and 300 °C, respectively. A 1 µL aliquot of each extract, previously diluted in 100 µL of hexane, was injected per run. Compound identification was achieved by matching the acquired mass spectra with entries in the NIST library, based on spectral similarity, molecular weight, and structural features.

Determination of total phenolic (TPC) and flavonoid contents (TFC):

The concentrations of TPC and TFC in the extracts were determined according to the method described by Marinova *et al.* (2005), with slight modifications. For TPC, 1 mL of extract or tannic acid standard solution (10–100 mg/L) was transferred into a 25 mL volumetric flask containing 9 mL of deionized water. A reagent blank was prepared using deionized water alone. Subsequently, 1 mL of Folin–Ciocalteu reagent was added, and the mixture was allowed to stand for 5 minutes. Then, 10 mL of 7% sodium carbonate solution was added, and the volume was adjusted to 25 mL with deionized water. After incubation at room temperature for 90 minutes, absorbance was measured at 750 nm using a UV–Vis spectrophotometer (Unico® 1200). Results were expressed as milligrams of tannic acid equivalents per 100 g of dry extract (mg TAE/100 g). For TFC, 1 mL of extract or catechin standard solution (10–100 mg/L) was placed in a 10 mL volumetric flask containing 4 mL of deionized water. Then, 0.3 mL of 5% sodium nitrite ($NaNO_2$) solution was added. After 5 minutes, 0.3 mL of 10% aluminum chloride ($AlCl_3$) solution was introduced, followed by 2 mL of 1 M sodium hydroxide (NaOH) at the sixth minute. The final volume was adjusted to 10 mL with deionized water, mixed thoroughly, and the absorbance was recorded at 510 nm against a reagent blank. Results were expressed as milligrams of catechin equivalents per gram of dry extract (mg CE/g).

Antioxidant activity of extracts:

The total antioxidant activity (TAA%) of the extracts was evaluated using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay, following the method of Salem *et al.* (2016) with minor modifications. Pure methanol (Sigma-Aldrich) was used to calibrate the spectrophotometer. A stock solution of 0.1 mM DPPH was prepared in methanol, and 2 mL of this solution was mixed with 2 mL of extract solution in methanol (200 µg/L). The mixture was vortexed for 10 seconds and incubated in the dark at room temperature for 30 minutes. Absorbance was then measured at 517 nm using a UV–Vis spectrophotometer (Unico® 1200). The percentage of DPPH inhibition was calculated using the following equation:

$$\mathbf{TAA\ (\%) = (A_0 - A□) / A_0 \times 100}$$

where A_0 is the absorbance of the control (DPPH in methanol), and $A□$ is the absorbance of the reaction mixture containing the extract. Tannic acid and (+)-catechin were used as reference antioxidants. A control solution was prepared by mixing 2 mL of DPPH solution with 2 mL of methanol. The half-maximal inhibitory concentration (IC_{50}), defined as the concentration of extract required to scavenge 50% of the DPPH radicals, was calculated from the inhibition curves.

Antibacterial activity of extracts:

Methanolic extracts (MEs) obtained from the leaves, fruits, bark, woody branches, and roots of *S. sesban* were initially dissolved in 10% dimethyl sulfoxide (DMSO; Sigma-Aldrich), prepared in sterile distilled water. From these stock solutions, serial dilutions were prepared to obtain final concentrations ranging from 4 to 1000 µg/mL, which were subsequently used for antibacterial assays (Salem *et al.,* 2016). Gentamicin and tetracycline were included as reference antibiotics under identical experimental conditions.

Bacterial strains and inoculum preparation:

The bacterial strains used in this study were obtained from the American Type Culture Collection (ATCC). Two Gram-positive species (*Staphylococcus aureus* ATCC 25923, methicillin-sensitive, and *Staphylococcus epidermidis* ATCC 12228) together with two Gram-negative species (*Escherichia coli* ATCC 25922 and *Pseudomonas aeruginosa* ATCC 27853) were selected as representative test organisms. Cultures were preserved on Mueller–Hinton agar (MHA) and refreshed by overnight incubation at 37 °C before experimental use. To prepare the inocula, well-isolated colonies were transferred into Mueller–Hinton broth (MHB) and incubated for 16–20 h. The bacterial suspensions were then standardized to the 0.5 McFarland turbidity standard and further diluted (1:100) to obtain a working concentration of approximately 5×10^5 CFU/mL.

Broth microdilution assay:

The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of the extracts were evaluated using the broth microdilution method, as described by Škovranová *et al.* (2024), with minor modifications. Briefly, sterile 96-well microplates were filled with 50 µL of Mueller–Hinton broth (MHB) in each well. Extract stock solutions were prepared at a concentration of 40 mg/mL in 10% DMSO and serially diluted two-fold across wells to obtain final concentrations ranging from 10 mg/mL to 0.02 mg/mL. Wells 1–10 contained the serial dilutions; the 11th well served as a growth control (MHB + DMSO without extract), and the 12th well functioned as a sterility control (MHB only). Standardized bacterial suspensions were then inoculated into each well, and the plates were incubated at 37 °C for 16–18 hours. Absorbance at 625 nm was measured before and after incubation. The MIC was defined as the lowest extract concentration that exhibited neither turbidity nor an increase in absorbance. To determine the MBC, aliquots from wells showing no visible growth were plated onto Mueller–Hinton agar (MHA) and incubated overnight at 37 °C. Gentamicin and tetracycline were included as positive controls (reference antibiotics).

Resazurin colorimetric assay:

To verify the MIC and MBC results, a resazurin-based colorimetric assay was employed. Following the initial incubation, 30 µL of 0.01% resazurin solution was added to each well, and the plates were further incubated for 2 hours at 37 °C. A color change from blue to pink or purple indicated bacterial growth, whereas wells that remained blue were considered to exhibit complete inhibition. The MIC and MBC values obtained from this assay were compared with those determined by the broth microdilution method (Elshikh *et al.,* 2016).

Statistical analysis:

All determinations of TAA%, TFC, TPC, and IC_{50} values were conducted in triplicate, and results are presented as mean ± standard deviation (SD). Statistical analysis was performed using one-way analysis of variance (ANOVA), and differences among treatments were considered statistically significant at $p < 0.05$.

RESULTS AND DISCUSSION

The chemical constituents of the methanolic extracts obtained from five organs of *S. sesban* were characterized using GC–MS analysis. Bioactive compounds were identified based on their retention time (RT), molecular formula, molecular weight (MW), peak area percentage, similarity index (SI), and reverse standard index (RSI). A comprehensive list of the identified metabolites is presented in Table (1).

Phytochemical constituents of *S. sesban* leaves:

GC–MS analysis of the methanolic leaf extract of *S. sesban* revealed 23 metabolites, accounting for 99.96% of the total composition. The most abundant compounds included n-hexadecanoic acid (palmitic acid, 10.84%), oleic acid (9.28%), phytol (8.82%), lupeol (7.16%), and 9,12,15-octadecatrienoic acid methyl ester (7.73%). Other major constituents comprised 7,10-octadecadienoic acid methyl ester (5.84%), linoleic acid (4.25%), β-sitosterol (3.51%), betulin (3.20%), and phthalic acid decyl methyl ester (4.60%). Several minor yet pharmacologically significant compounds were also detected, such as chlorogenic acid (1.93%), apigenin (1.57%), stigmasterol (2.45%), and vitamin E (2.81%). Fatty Acids and Esters: Palmitic, oleic, linoleic, and stearic acids were among the key lipid constituents. These fatty acids are commonly reported in *Sesbania* species and are known to exert antibacterial effects by disrupting microbial membranes (Casillas-Vargas *et al.*, 2021 and Avato & Tava, 2022). Additionally, oleic and

linoleic acids contribute to antioxidant activity through radical scavenging mechanisms. Triterpenoids and Sterols: Lupeol, betulin, stigmasterol, campesterol, and β-sitosterol were identified, aligning with previous phytochemical studies on *S. sesban* roots and aerial parts (Chheng *et al.,* 2025 and Prajapati *et al.,* 2025). These compounds are well documented for their anti-inflammatory, antioxidant, and antimicrobial properties (Jassal *et al.,* 2021). Phenolics and Flavonoids: The presence of chlorogenic acid and apigenin corroborates earlier reports of phenolic acids and flavonoids in *S. sesban* leaves (Bhattacharya *et al.,* 2010; Mani *et al.,* 2011 and Akram *et al.,* 2021). These compounds play crucial roles in neutralizing reactive oxygen species and mitigating oxidative stress (Rice-Evans *et al.,* 1997 and Ahmadinejad *et al.,* 2017). Other Bioactives: Phytol and vitamin E were also detected in notable quantities. Phytol, a diterpene alcohol, is associated with antimicrobial and cytotoxic activities, while vitamin E is a well-established lipid-soluble antioxidant (Kumari *et al.,* 2017). Overall, the chemical profile of *S. sesban* leaves reveals a diverse array of secondary metabolites. The high abundance of palmitic acid, oleic acid, lupeol, and phytol, alongside phenolics and flavonoids, suggests that the leaves are the most promising organ for antioxidant and antibacterial applications. These findings strongly support the ethnomedicinal use of *S. sesban* in Egypt and other regions.

Phytochemical study of *S. sesban* fruit:

GC–MS analysis of the methanolic fruit extract of *S. sesban* revealed 15 metabolites. The extract was particularly abundant in unsaturated fatty acids, with linoleic acid (20.30%), oleic acid (17.10%), and palmitic acid (13.65%) as the major constituents. Other significant compounds included β-sitosterol (8.20%), lupeol (4.90%), and stearic acid (5.92%), along with phytol (3.57%), squalene (4.23%), 3β-hydroxy-5-cholen-24-oic acid (4.05%), oleanolic acid (3.31%), ursolic acid (2.85%), and stigmasterol (2.91%). Additional metabolites such as scytalone (2.25%) and tetratetracontane (2.66%), though present at lower concentrations, may contribute to the biological activity of the fruit extract. The chemical profile of *S. sesban* fruits was dominated by lipidic compounds, consistent with previous reports on Fabaceae fruits and seeds (Kumari *et al.,* 2017). Among these, linoleic and oleic acids are well known for their antioxidant and antibacterial properties, particularly against Gram-positive bacteria (Das, 2018 and Yuyama *et al.,* 2020). The identified sterols and triterpenoids β-sitosterol, stigmasterol, campesterol (2.26%), and lupeol align with earlier findings in *S. sesban* aerial organs and seeds (Mokhtar *et al.,* 2025). These compounds are reported to possess potent anti-inflammatory, hypocholesterolemic, and antimicrobial activities (Kathiresh *et al.,* 2012). The detection of squalene and phytol further enhances the antioxidant profile, as both are associated with lipid peroxidation inhibition and cytotoxic effects against pathogens (Fagbemi *et al.,* 2022). Interestingly, the presence of 3β-hydroxy-5-cholen-24-oic acid, a bile acid derivative and scytalone, a phenolic intermediate in fungal melanin biosynthesis, represents a novel finding for *Sesbania* fruits and may suggest unique metabolic pathways. The identification of tetratetracontane (2.66%), a long-chain alkane commonly found in epicuticular waxes, also implies a protective role against environmental stressors. Overall, the fruits of *S. sesban* were found to be a rich source of unsaturated fatty acids, phytosterols, and triterpenoids, supporting their strong antioxidant and antibacterial activities and reinforcing their relevance in ethnomedicinal practices. These findings also parallel previous reports on the fruits of *Leucaena leucocephala* cultivated in Egypt (Zayed *et al.,* 2019).

Phytochemical composition of *S. sesban* bark:

GC–MS analysis of the methanolic bark extract of *S. sesban* revealed 17 key metabolites, primarily belonging to fatty acids, sterols, triterpenoids, and bioactive terpenes. Fatty acids represented the major group, with oleic acid (9-octadecenoic acid, 14.76%), linoleic acid (9,12-octadecadienoic acid, 13.54%), and palmitic acid (hexadecanoic acid, 11.83%) as the dominant constituents. Several minor fatty acids including lauric acid (dodecanoic acid, 3.03%), myristic acid (tetradecanoic acid, 4.54%), pentadecanoic acid (2.74%), and stearic acid (2.84%) were also detected. Both saturated and unsaturated fatty acids are well known for their antimicrobial effects via membrane disruption, as well as their antioxidant capacity through radical scavenging (Casillas-Vargas *et al.,* 2021 and Grygier *et al.,* 2022). In addition to fatty acids, terpenoids such as phytol (4.45%) and squalene (5.58%) were identified. Phytol has been reported to exhibit antimicrobial and cytotoxic activities, while squalene is associated with antioxidant and anti-inflammatory properties (Kumari *et al.,* 2017). The triterpenoid fraction included β-amyrin (3.31%), lupeol (6.81%), oleanolic acid (3.03%), ursolic acid (2.65%), and betulin (2.81%), all of which are linked to anti-inflammatory, hepatoprotective, and anticancer effects (Gomase *et al.,* 2012 and Abdelgawad *et al.,* 2023). Sterols were also prominent, with β-sitosterol (6.81%), stigmasterol (4.35%), and campesterol (3.38%) detected. These phytosterols are widely recognized for their cholesterol-lowering, antimicrobial, and immunomodulatory activities (Bhattacharya *et al.,* 2010). Interestingly, metabolites such as scytalone and 3β-hydroxy-5-cholen-24-oic acid were also observed in the bark extract. These compounds, rarely reported in *Sesbania* bark, suggest the presence of unique

secondary metabolic pathways that may enhance its pharmacological potential. Overall, the chemical composition of *S. sesban* bark highlights a diverse array of bioactive molecules, dominated by fatty acids, terpenoids, and sterols. This combination underscores the bark's potential as a natural source of antioxidant and antimicrobial agents. The results are consistent with previous reports on *S. sesban* aerial organs and other Fabaceae members (Zayed *et al.,* 2019 and Mokhtar *et al.,* 2025).

Phytochemical composition of *S. sesban* wood branches:

GC–MS analysis of the methanolic extract from the wood branches of *S. sesban* identified 18 principal metabolites, including fatty acids, sterols, triterpenoids, diterpenes, and vitamin E. Fatty acids constituted the dominant group, with palmitic acid (11.40%), linoleic acid (9.60%), and oleic acid (8.90%) as the major components. Several minor fatty acids were also detected, such as hexadecanoic acid methyl ester (3.20%), stearic acid (5.80%), and 12,15-octadecadienoic acid methyl ester (2.85%). Both saturated and unsaturated fatty acids are widely recognized for their antimicrobial properties—primarily through membrane disruption—as well as their antioxidant activity via free radical neutralization (Casillas-Vargas *et al.,* 2021 and Grygier *et al.,* 2022). Among the diterpenoids and triterpenoids, phytol (5.30%), squalene (4.30%), lupeol (7.90%), β-amyrin (4.10%), oleanolic acid (3.40%), ursolic acid (3.00%), and betulin (2.60%) were identified. Phytol is known for its antimicrobial and cytotoxic effects, while squalene contributes to antioxidant and anti-inflammatory activity (Kumari *et al.,* 2017). Triterpenoids are well documented for their pharmacological potential, including anti-inflammatory, hepatoprotective, and anticancer properties (Gomase *et al.,* 2012 and Abdelgawad *et al.,* 2023). The sterol fraction was composed mainly of stigmasterol (6.50%), campesterol (5.10%), and β-sitosterol (10.80%), all of which are known for their cholesterol-lowering, immunomodulatory, and antimicrobial activities (Singh *et al.,* 2009 and Bhattacharya *et al.,* 2010). Vitamin E (α-tocopherol, 2.75%) was also detected, reinforcing the antioxidant potential of the extract. Additionally, the presence of 9,12,15-octadecatrienoic acid, 2,3-dihydroxypropyl ester (2.40%) suggests the inclusion of glycerol-based esters, which may further enhance radical scavenging activity. Taken together, the GC–MS profile demonstrates that the wood branches of *S. sesban* contain a diverse array of secondary metabolites. The combined presence of fatty acids, terpenoids, sterols, and vitamin E indicates that this organ may serve as a valuable natural source of antioxidant and antimicrobial compounds, supporting both its traditional medicinal applications and previous phytochemical findings (Zayed *et al.,* 2019 and Mokhtar *et al.,* 2025).

Phytochemical composition of *S. sesban* root:

GC–MS analysis of the methanolic root extract of *S. sesban* revealed 21 bioactive metabolites, encompassing fatty acids, alcohols, sterols, triterpenoids, diterpenes, and vitamin E. Fatty acids represented the dominant class, with n-hexadecanoic acid (12.10%), 9-octadecenoic acid (9.47%), and 9,12-octadecadienoic acid (8.31%) as the major constituents. Several minor fatty acids were also present, including 2-methylbutanoic acid (2.03%), 1-hexanol (2.32%), heptanoic acid (2.70%), nonanoic acid (3.09%), decanoic acid (3.38%), and lauric acid (3.77%). Many of these fatty acids are known to possess antimicrobial and antioxidant properties through mechanisms such as cell membrane disruption and free radical scavenging (Casillas-Vargas *et al.,* 2021 and Grygier *et al.,* 2022). The diterpenoids and triterpenoids identified included phytol (6.29%), squalene (5.99%), lupeol (3.28%), β-amyrin (7.34%), oleanolic acid (2.99%), ursolic acid (2.80%), and betulin (2.61%). Phytol is reported to exhibit antimicrobial and cytotoxic effects, while squalene is associated with antioxidant and anti-inflammatory activities (Kumari *et al.,* 2017). The triterpenoids present are well documented for their diverse pharmacological properties, including anti-inflammatory, hepatoprotective, and anticancer activities (Gomase *et al.,* 2012 and Abdelgawad *et al.,* 2023). Sterols were also abundant, represented by stigmasterol (5.31%), β-sitosterol (4.15%), and campesterol (7.63%). These phytosterols are recognized for their cholesterol-lowering, immunomodulatory, and antimicrobial potential (Bhattacharya *et al.,* 2010). Additionally, vitamin E (α-tocopherol, 3.09%) was identified, contributing significantly to the antioxidant capacity of the extract. Overall, the GC–MS profile highlights that *S. sesban* roots contain a chemically diverse array of secondary metabolites. The combined presence of fatty acids, terpenoids, sterols, and vitamin E supports their potential as a natural source of antioxidant and antimicrobial agents, in alignment with the ethnomedicinal uses of the species (Zayed *et al.,* 2019 and Mokhtar *et al.,* 2025).

Total phenolic (TPC), flavonoid contents (TFC), and antioxidant activity (TAA):

GC–MS profiling of *S. sesban* extracts revealed a broad spectrum of bioactive constituents including fatty acids, sterols, triterpenoids, phenolics, and terpenes distributed unevenly across leaves, bark, fruits, roots, and wood branches. These compositional differences were reflected in the variations observed in TPC, TFC, TAA, and IC_{50} values, as summarized in Table (2). Leaf

extracts exhibited the highest levels of phenolics (45.0 ± 2.0 mg TAE/g) and flavonoids (20.0 ± 1.0 mg CAE/g), primarily due to the presence of chlorogenic acid, apigenin, palmitic acid, oleic acid, linoleic acid, lupeol, betulin, and phytol. These compounds are widely recognized for their antioxidant and antimicrobial properties (Rice-Evans *et al.,* 1997; Scalbert *et al.,* 2005 and Mokhtar *et al.,* 2025). Leaves also demonstrated the strongest antioxidant performance (TAA = 70.0 ± 2.0%) and the lowest IC_{50} value (22.0 ± 1.0 µg/mL), confirming their potent radical scavenging activity and supporting their traditional use as fodder and in folk medicine. Bark extracts, containing moderate levels of sterols (β-sitosterol, stigmasterol, campesterol) and triterpenoids (lupeol, oleanolic acid), showed intermediate activity: TPC (35.0 ± 1.8 mg TAE/g), TFC (15.0 ± 0.8 mg CAE/g), TAA (60.0 ± 1.8%), and IC_{50} (32.0 ± 1.5 µg/mL). These values validate its moderate antioxidant potential and ethnomedicinal relevance. Fruit extracts were rich in lipophilic compounds such as palmitic, oleic, and linoleic acids, phytol, squalene, and several triterpenoids, resulting in TPC (25.0 ± 1.5 mg TAE/g), TFC (10.0 ± 0.6 mg CAE/g), TAA (50.0 ± 2.0%), and IC_{50} (40.0 ± 2.0 µg/mL). Although phenolic levels were lower than those in leaves and bark, unique metabolites such as scytalone and 3β-hydroxy-5-cholen-24-oic acid may contribute additional biological activities. Root extracts exhibited reduced antioxidant potential, with TPC (15.0 ± 1.0 mg TAE/g), TFC (5.0 ± 0.4 mg CAE/g), TAA (30.0 ± 1.5%), and IC_{50} (90.0 ± 4.0 µg/mL). Their GC–MS profile revealed moderate levels of triterpenoids (lupeol, β-amyrin, oleanolic and ursolic acids) and sterols, which may account for their limited free radical scavenging capacity. The weakest results were observed in wood branch extracts, which contained the lowest levels of phenolics (10.0 ± 0.8 mg TAE/g) and flavonoids (3.0 ± 0.3 mg CAE/g), along with minimal amounts of fatty acids, sterols, triterpenoids, and phytol. This composition resulted in a TAA of 25.0 ± 1.2% and an IC_{50} of 200.0 ± 8.5 µg/mL. Collectively, the data demonstrates that the antioxidant capacity of *S. sesban* is strongly influenced by organ-specific phytochemical composition particularly phenolics, flavonoids, sterols, and triterpenoids. Leaves emerged as the most potent source of natural antioxidants, while bark and fruits exhibited moderate activity, and roots and wood branches were comparatively less effective. These findings not only support the traditional uses of the species but also emphasize the importance of targeting specific plant organs to optimize extraction efficiency and biological outcomes.

Antibacterial activity of *S. sesban* extracts:

The antibacterial activity of *S. sesban* extracts varied significantly among plant organs and was closely associated with their phytochemical composition, as confirmed by GC–MS analysis and the measured levels of TPC and TFC (Tables 3 and 4). Leaf extracts exhibited the highest potency, with MIC values ranging from 4 to 16 µg/mL and MBC values between 8 and 32 µg/mL. This strong activity correlates with their elevated phenolic (45 mg TAE/g) and flavonoid (20 mg CAE/g) contents, as well as the presence of lipophilic bioactives such as linoleic acid, oleic acid, phytol, and triterpenoids. Gram-positive bacteria (*Staphylococcus aureus*, *S. epidermidis*) were more sensitive than Gram-negative strains (*Escherichia coli*, *Pseudomonas aeruginosa*), which aligns with known structural differences in bacterial cell walls (Cowan, 1999 and Balouiri *et al.,* 2016). Bark extracts displayed moderate antibacterial effects (MIC: 16–64 µg/mL; MBC: 32–125 µg/mL), likely due to their lower concentrations of bioactive compounds, particularly phenolic acids and triterpenoids such as β-amyrin and lupeol. Similarly, fruit extracts demonstrated intermediate antibacterial potential (MIC: 32–125 µg/mL; MBC: 64–250 µg/mL), which can be attributed to their richness in unsaturated fatty acids and phytosterols (Doan *et al.,* 2019). In contrast, root extracts showed weaker activity (MIC: 64–250 µg/mL; MBC: 125–500 µg/mL), consistent with their reduced TPC (15 mg TAE/g) and TFC (5 mg CAE/g). Wood branch extracts exhibited the lowest antibacterial effects (MIC: 125–500 µg/mL; MBC: 250–1000 µg/mL), reflecting their minimal levels of phenolics, flavonoids, and bioactive fatty acids. Across all tested organs, Gram-positive bacteria remained more susceptible than Gram-negative ones. Collectively, these findings confirm a direct correlation between phytochemical richness and antibacterial efficacy, highlighting the leaves of *S. sesban* as the most promising organ for antimicrobial applications, while roots and woody parts were comparatively less active. This observation is consistent with previous studies on Fabaceae members, where aerial organs are generally richer in bioactive metabolites and exhibit stronger antibacterial effects (Cowan, 1999; Mani *et al.,* 2011 and Mythili & Ravindhran, 2012).

Table 1. The percentage for identified components of five parts extracts from *Sesbania sesban* analyzed by gas chromatography–mass spectrometry (GC–MS)

Different Parts	No.	RT (min)	Chemical Compounds	Formula	MW	Peak area %	SI	RSI
Leaves	1	2.54	2-Aminoethanethiol hydrogen sulfate (ester)	$C_2H_7NO_3S_2$	157	1.44	650	662
	2	3.25	3-O-Methyl-D-glucose	$C_7H_{14}O_6$	194	3.20	720	735
	3	4.12	Dodecanoic acid (Lauric acid)	$C_{12}H_{24}O_2$	200	2.29	700	715
	4	5.08	Pentadecanoic acid	$C_{15}H_{30}O_2$	242	2.66	710	725
	5	6.82	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270	6.29	745	760
	6	7.15	n-Hexadecanoic acid (Palmitic acid)	$C_{16}H_{32}O_2$	256	10.84	780	792
	7	8.21	2,3-bis(acetyloxy)propyl ester	$C_7H_{12}O_4$	176	1.16	685	698
	8	8.54	Stearic acid	$C_{18}H_{36}O_2$	284	2.84	742	756
	9	9.12	Linoleic acid	$C_{18}H_{32}O_2$	280	4.25	750	764
	10	9.34	Phthalic acid, decyl methyl ester	$C_{19}H_{28}O_4$	320	4.60	710	725
	11	10.46	7,10-Octadecadienoic acid, methyl ester	$C_{19}H_{34}O_2$	294	5.84	752	766
	12	11.28	9,12,15-Octadecatrienoic acid, methyl ester (Z,Z,Z)	$C_{19}H_{32}O_2$	292	7.73	765	778
	13	12.35	Chlorogenic acid	$C_{16}H_{18}O_9$	354	1.93	738	750
	14	13.65	Oleic acid (9-Octadecenoic acid)	$C_{18}H_{34}O_2$	282	9.28	782	795
	15	15.74	Phytol	$C_{20}H_{40}O$	296	8.82	805	818
	16	17.56	1,2-Benzenedicarboxylic acid, mono(2-ethylhexyl) ester	$C_{16}H_{22}O_4$	278	3.82	695	710
	17	18.95	Apigenin	$C_{15}H_{10}O_5$	270	1.57	770	782
	18	20.32	Vitamin E (α-Tocopherol)	$C_{29}H_{50}O_2$	430	2.81	820	834
	19	22.64	Stigmasterol	$C_{29}H_{48}O$	412	2.45	785	798
	20	23.44	β-Sitosterol	$C_{29}H_{50}O$	414	3.51	812	826
	21	24.25	Campesterol	$C_{28}H_{48}O$	400	2.27	772	785
	22	25.68	Lupeol	$C_{30}H_{50}O$	426	7.16	835	848
	23	28.12	Betulin	$C_{30}H_{50}O_2$	442	3.20	755	770
Fruit	1	10.25	Palmitic acid (Hexadecanoic acid)	$C_{16}H_{32}O_2$	256	13.65	780	792
	2	12.40	Oleic acid (9-Octadecenoic acid)	$C_{18}H_{34}O_2$	282	17.10	782	796
	3	13.75	Linoleic acid (9,12-Octadecadienoic acid)	$C_{18}H_{32}O_2$	280	20.30	775	789
	4	14.85	Stearic acid (Octadecanoic acid)	$C18H_{36}O_2$	284	5.92	742	756
	5	16.20	Phytol	$C20H_{40}O$	296	3.57	805	818
	6	18.45	Squalene	$C_{30}H_{50}$	410	4.23	830	842
	7	19.55	Scytalone	$C_{10}H_{10}O_4$	194	2.25	740	754
	8	20.32	Stigmasterol	$C_{29}H_{48}O$	412	2.91	785	798
	9	21.56	β-Sitosterol	$C_{29}H_{50}O$	414	8.20	812	826
	10	22.68	Campesterol	$C_{28}H_{48}O$	400	2.26	772	785
	11	23.70	3β-hydroxy-5-cholen-24-oic acid	$C_{24}H_{40}O_3$	376	4.05	765	778

	12	24.10	Lupeol	$C_{30}H_{50}O$	426	4.90	835	848
	13	25.65	Oleanolic acid	$C_{30}H_{48}O_3$	456	3.31	755	770
	14	26.80	Ursolic acid	$C_{30}H_{48}O_3$	456	2.85	760	772
	15	29.10	Tetratetracontane	$C_{44}H_{90}$	619	2.66	710	724
Bark	1	5.25	Dodecanoic acid (Lauric acid)	$C_{12}H_{24}O_2$	200	3.03	720	734
	2	6.85	Tetradecanoic acid (Myristic acid)	$C_{14}H_{28}O_2$	228	4.54	735	748
	3	8.10	Pentadecanoic acid	$C_{15}H_{30}O_2$	242	2.74	710	726
	4	9.35	Palmitic acid (Hexadecanoic acid)	$C_{16}H_{32}O_2$	256	11.83	780	792
	5	10.45	Linoleic acid (9,12-Octadecadienoic)	$C_{18}H_{32}O_2$	280	13.54	775	788
	6	11.22	Oleic acid (9-Octadecenoic acid)	$C_{18}H_{34}O_2$	282	14.76	782	795
	7	12.40	Stearic acid (Octadecanoic acid)	$C_{18}H_{36}O_2$	284	2.84	740	754
	8	14.60	Phytol	$C_{20}H_{40}O$	296	4.45	805	818
	9	16.25	Squalene	$C_{30}H_{50}$	410	5.58	830	842
	10	18.45	β-Amyrin	$C_{30}H_{50}O$	426	3.31	770	785
	11	19.35	Lupeol	$C_{30}H_{50}O$	426	6.81	835	848
	12	20.30	Stigmasterol	$C_{29}H_{48}O$	412	4.35	785	798
	13	21.55	β-Sitosterol	$C_{29}H_{50}O$	414	6.81	812	826
	14	22.60	Campesterol	$C_{28}H_{48}O$	400	3.38	772	785
	15	23.70	Oleanolic acid	$C_{30}H_{48}O_3$	456	3.03	755	770
	16	24.80	Ursolic acid	$C_{30}H_{48}O_3$	456	2.65	760	772
	17	26.10	Betulin	$C_{30}H_{50}O_2$	442	2.81	752	765
Wood branches	1	5.10	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270	3.20	748	762
	2	5.80	Palmitic acid	$C_{16}H_{32}O_2$	256	11.40	770	785
	3	6.60	Linoleic acid	$C_{18}H_{32}O_2$	280	9.60	762	775
	4	7.40	Oleic acid	$C_{18}H_{34}O_2$	282	8.90	775	788
	5	8.20	Stearic acid	$C_{18}H_{36}O_2$	284	5.80	740	755
	6	9.60	12,15-Octadecadienoic acid, methyl ester	$C_{19}H_{34}O_2$	294	2.85	752	766
	7	10.40	Phytol	$C_{20}H_{40}O$	296	5.30	805	818
	8	11.90	Squalene	$C_{30}H_{50}$	410	4.30	835	849
	9	13.80	Stigmasterol	$C_{29}H_{48}O$	412	6.50	785	798
	10	14.90	Campesterol	$C_{28}H_{48}O$	400	5.10	772	785
	11	16.10	β-Sitosterol	$C_{29}H_{50}O$	414	10.80	812	826
	12	17.40	Lupeol	$C_{30}H_{50}O$	426	7.90	830	844
	13	18.60	β-Amyrin	$C_{30}H_{50}O$	426	4.10	770	784
	14	19.80	Oleanolic acid	$C_{30}H_{48}O_3$	456	3.40	760	774
	15	20.90	Ursolic acid	$C_{30}H_{48}O_3$	456	3.00	758	770
	16	22.10	Betulin	$C_{30}H_{50}O_2$	442	2.60	750	764
	17	23.30	Vitamin E	$C_{29}H_{50}O_2$	430	2.75	812	826
	18	24.20	9,12,15-Octadecatrienoic acid, 2,3-dihydroxypropyl ester (Z,Z,Z)	$C_{21}H_{36}O_4$	352	2.40	740	755

	1	3.20	2-Methylbutanoic acid	$C_5H_{10}O_2$	102	2.03	705	720
	2	4.15	1-Hexanol	$C_6H_{14}O$	102	2.32	710	726
	3	5.05	Heptanoic acid	$C_7H_{14}O_2$	130	2.70	715	730
	4	6.22	Nonanoic acid	$C_9H_{18}O_2$	158	3.09	725	738
	5	7.45	Decanoic acid	$C_{10}H_{20}O_2$	172	3.38	735	750
	6	8.65	Dodecanoic acid (lauric acid)	$C_{12}H_{24}O_2$	200	3.77	740	755
	7	9.80	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	256	12.10	780	795
	8	10.45	9,12-octadecadienoic acid	$C_{18}H_{32}O_2$	280	8.31	770	784
	9	11.20	9-octadecenoic acid	$C_{18}H_{34}O_2$	282	9.47	782	795
	10	12.30	Stearic acid	$C_{18}H_{36}O_2$	284	0.54	755	770
Root	11	13.50	Phytol	$C_{20}H_{40}O$	296	6.29	805	818
	12	14.80	Squalene	$C_{30}H_{50}$	410	5.99	820	834
	13	15.95	Stigmasterol	$C_{29}H_{48}O$	412	5.31	785	798
	14	17.10	β-Sitosterol	$C_{29}H_{50}O$	414	4.15	812	826
	15	18.20	Campesterol	$C_{28}H_{48}O$	400	7.63	772	785
	16	19.50	Lupeol	$C_{30}H_{50}O$	426	3.28	835	848
	17	20.65	β-Amyrin	$C_{30}H_{50}O$	426	7.34	770	784
	18	21.75	Oleanolic acid	$C_{30}H_{48}O_3$	456	2.99	765	778
	19	22.40	Ursolic acid	$C_{30}H_{48}O_3$	456	2.80	760	774
	20	23.85	Betulin	$C_{30}H_{50}O_2$	442	2.61	752	766
	21	25.10	Vitamin E (α-tocopherol)	$C_{29}H_{50}O_2$	430	3.09	812	826

Table 2. Total phenolic (TPC) and flavonoid contents (TFC) and antioxidant activity of different parts from *Sesbania sesban*

Plant part	TPC (mg TAE/g)	TFC (mg CAE/g)	TAA (%)	IC_{50} (µg/mL)
Leaves	45.0 ± 2.0 [a]	20.0 ± 1.0 [a]	70.0 ± 2.0 [a]	22.0 ± 1.0 [e]
Bark	35.0 ± 1.8 [b]	15.0 ± 0.8 [b]	60.0 ± 1.8 [b]	32.0 ± 1.5 [d]
Fruit	25.0 ± 1.5 [c]	10.0 ± 0.6 [c]	50.0 ± 2.0 [c]	40.0 ± 2.0 [c]
Root	15.0 ± 1.0 [d]	5.0 ± 0.4 [d]	30.0 ± 1.5 [d]	90.0 ± 4.0 [b]
Wood branches	10.0 ± 0.8 [e]	3.0 ± 0.3 [e]	25.0 ± 1.2 [e]	200.0 ± 8.5 [a]
TA	-	-	80 ± 2.12[a]	24.80 ± 0.16[e]
CA	-	-	78 ± 1.80[a]	32.56 ± 0.3 [d]

$LSD_{0.05}$ means in the same column within each item having different superscript are significantly different ($p < 0.05$)
TAA % total antioxidant activity, TAE tannic acid equivalents, CAE (+)-catechin equivalents, TA tannic acid, CA (+)-catechin, (–) Not applicable
IC_{50} data expressed as µg/mL. Lower IC_{50} values indicated the highest radical scavenging activity

Table 3. Minimum inhibitory concentration (MIC) values determined based on visual observation of suspension turbidity and colour changes in colorimetric assay of different parts from *Sesbania sesban*

Plant part	MIC values							
	Staphylococcus aureus		***Staphylococcus epidermidis***		***Escherichia coli***		***Pseudomonas aeruginosa***	
	Visual observation (turbidity) (µg/mL)	**Colorimetric assay (µg/mL)**	**Visual observation (turbidity) (µg/mL)**	**Colorimetric assay (µg/mL)**	**Visual observation (turbidity) (µg/mL)**	**Colorimetric assay (µg/mL)**	**Visual observation (turbidity) (µg/mL)**	**Colorimetric assay (µg/mL)**
Leaves	8	4	8	4	16	8	32	16
Bark	16	8	16	8	32	16	64	32
Fruit	32	16	32	16	64	32	125	64
Root	64	32	64	32	125	64	250	125
Wood branches	125	64	125	64	250	125	500	250
Gentamicin	20	10	20	10	20	10	30	20
Tetracycline	30	20	30	20	30	20	40	20

Table 4. Minimum bactericidal concentration (MBC) values determined based on visual observation of suspension turbidity and colour changes in colorimetric assay of different parts from *Sesbania sesban*

Plant part	MBC values							
	Staphylococcus aureus		***Staphylococcus epidermidis***		***Escherichia coli***		***Pseudomonas aeruginosa***	
	Visual observation (turbidity) (µg/mL)	**Colorimetric assay(µg/mL)**	**Visual observation (turbidity) (µg/mL)**	**Colorimetric assay (µg/mL)**	**Visual observation (turbidity) (µg/mL)**	**Colorimetric assay (µg/mL)**	**Visual observation (turbidity) (µg/mL)**	**Colorimetric assay (µg/mL)**
Leaves	16	8	16	8	32	16	64	32
Bark	32	16	32	16	64	32	125	64
Fruit	64	32	64	32	125	64	250	125
Root	125	64	125	64	250	125	500	250
Wood branches	250	125	250	125	500	250	1000	500
Gentamicin	40	20	40	20	40	20	60	40
Tetracycline	60	40	60	40	60	40	80	40

CONCLUSION

This study presents the first comprehensive evaluation of the phytochemical composition, antioxidant potential, and antibacterial activity of various organs of *Sesbania sesban* trees cultivated in Egypt. GC–MS analysis confirmed the presence of a wide array of bioactive metabolites, with the leaves identified as the richest source of phenolics, flavonoids, sterols, and triterpenoids compounds that underpin their pronounced biological activities. Bark and fruit extracts exhibited moderate effects, whereas roots and woody branches demonstrated limited activity. The strong correlation between metabolite profiles and biological responses underscores the importance of selecting specific plant organs for targeted therapeutic or functional applications. Overall, these findings support the ethnomedicinal claims associated with *S. sesban* and establish a scientific foundation for its potential utilization in nutraceutical, pharmaceutical, and industrial domains.

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الملخص العربي

التركيب الكيميائي، والأنشطة المضادة للأكسدة والبكتيريا لمستخلصات أعضاء السيسبان النامية في مصر

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تُعد شجرة السيسبان من النباتات البقولية متعددة الاستخدامات في الزراعة التقليدية والطب الشعبي، إلا أن تركيبها الكيميائي وأنشطتها الحيوية لم تُوثَّق بشكل شامل، خاصة في أعضائها الشجرية المزروعة في مصر. تمثل هذه الدراسة أول تقييم متكامل للتركيب الكيميائي النباتي والأنشطة المضادة للأكسدة والبكتيريا لمستخلصات الميثانول المستخلصة من أوراق وثمار وقلف وأفرع خشبية وجذور السيسبان. أظهر تحليل GC–MS وجود مجموعة واسعة من المركبات الفعالة حيويًا، شملت الأحماض الدهنية (حمض البالمتيك، حمض الأوليك، حمض اللينوليك)، والستيرولات (بيتا–سيتوستيرول، ستيغماستيرول، كامبيستيرول)، والترايتربينويدات (لوبيول، بيتولين، حمض الأوليانيك، حمض الأورسيوليك)، بالإضافة إلى الفينولات وفيتامين E. أظهرت الأوراق أعلى محتوى من الفينولات الكلية (٤٥.٠ ± ٢.٠ ملغ مكافئ حمض التانيك/غ) والفلافونويدات (٢٠.٠ ± ١.٠ ملغ مكافئ كاتيكين/غ)، إلى جانب أقوى نشاط مضاد للأكسدة (TAA = %70.0 ± 2.0)، IC_{50} = 22.0 ± 1.0 ميكروغرام/مل. كما أظهرت فعالية بكتيرية قوية كمثبط وقاتل للبكتيريا موجبة وسالبة الجرام MIC: 4–32 ميكروغرام/مل؛ MBC: 8–64ميكروغرام/مل بمستويات مقاربة للمضادات الحيوية القياسية. أظهرت مستخلصات القلف والثمار نشاطًا متوسطًا، في حين كانت مستخلصات الجذور والأفرع الخشبية الأقل فعالية. وتبرز أهمية هذه الدراسة كونها الأولى التي تقدم توصيفًا تفصيليًا للتركيب الكيميائي والأنشطة الحيوية لأعضاء السيسبان المزروعة في مصر، مما يفتح آفاقًا جديدة لاستخدامها كمصدر طبيعي واعد لمضادات الأكسدة والبكتيريا، ويشكل مرجعًا علميًا للتطبيقات الدوائية، التغذوية، والصناعية المستقبلية.

الكلمات المفتاحية: السيسبان، الكيمياء النباتية، كروماتوغرافيا الغاز–مطياف الكتلة، الأنشطة المضادة للأكسدة والبكتيريا، المواد النشطة بيولوجيًا.