

Research Article

***In Vitro* Determination of Antimicrobial and Hypoglycemic Activities of *Mikania cordata* (Asteraceae) Leaf Extracts**

Pavithra L. Jayatilake  and **Helani Munasinghe** 

Department of Botany, University of Sri Jayewardenepura, Nugegoda 10250, Sri Lanka

Correspondence should be addressed to Helani Munasinghe; helani@sci.sjp.ac.lk

Received 29 February 2020; Accepted 21 October 2020; Published 10 November 2020

Academic Editor: Robert J. Linhardt

Copyright © 2020 Pavithra L. Jayatilake and Helani Munasinghe. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Infectious diseases and diabetes mellitus are counted responsible for a substantial amount of mortality among the human population. The current study was performed to detect the antimicrobial activities and hypoglycemic potential of *Mikania cordata* (Asteraceae) leaves extracted into aqueous media and several organic solvents (ethyl acetate and methanol). The ethyl acetate extract of *Mikania cordata* (MEA) leaves was observed to possess significantly ($p \leq 0.05$) greater antimicrobial capabilities (susceptible against *Bacillus cereus* ATCC 11778 and *Staphylococcus aureus* ATCC 25923) when compared with that of the methanol (MME) and aqueous extracts (MDW) which were assessed based on Kirby-Bauer disk diffusion assay. The minimum inhibitory concentration of MEA (against *B. cereus*, *S. aureus*, and *Escherichia coli* ATCC 25922) and MME (against *B. cereus*, *S. aureus*, *E. coli*, and *Candida albicans* ATCC 10231) lies in a similar range of $1.13 > \text{MIC} > 0.56$ mg/ml. In the present study, a single compound (from MEA) of R_f value 0.64 was isolated by thin-layer chromatography (TLC) that was responsible for the zone of inhibition against *B. cereus* (20.3 ± 0.3 mm). The results of this study also depicted the antihyperglycemic properties of *M. cordata* leaves which followed the same trend as the commercial drug Metformin in a glucose concentration-independent manner when tested in a glucose uptake assay by yeast cells. Therefore, it is evident that *Mikania cordata* is a reservoir of useful bioactive compounds which with further research will be paving the path for drug commercialization. This is the first record of TLC-based isolation of antimicrobial compounds of *M. cordata* and analysis of the hypoglycemic properties of *M. cordata* leaves.

1. Introduction

A vast amount of commercially available antimicrobial therapeutics have rather become resistant towards the more evolved species of pathogenic microorganisms. The enhancement of drug resistance has posed a major threat in their treatment processes, thus indicating the need for more novel therapeutics. Inappropriate drug utilization and overdosing are found to be the main culprits of catastrophic drug resistance [1, 2]. *Mikania cordata* have traditionally long been exploited for its wound healing, antimicrobial, and anti-inflammatory properties. It is also widely used to treat eye sores, scorpion and snake bites, coughs, and various gastrointestinal infections [3]. Hence, it is evident that *M. cordata* is a reservoir of useful bioactive compounds that await further analysis. *M. cordata* is a fast-growing,

perennial vine with cordate leaves that are arranged in an opposite manner. Propagation is via seed dispersal through wind and by rooting at nodes which come in contact with the soil. Seeds are produced in large numbers, and the long-distance dispersal of the seeds is ensured by the pappus. The vine is dense growing and, hence, acts as a barrier for sunlight to reach the ground and other parts of vegetation. The traditional practice is to destroy the vine mesh at the onset of the flowering season because if left undisturbed, it could be a devastating weed [4].

Persistent hyperglycemia and impaired metabolism of carbohydrates, proteins, and fats characterize the systemic metabolic disease of diabetes mellitus. The most effective management of diabetes involves medications along with alterations to both diet and exercises hence, requiring a lifestyle change. The current medications consist of insulin

supplementation along with other oral hypoglycemic medications that can be grouped into several classes such as meglitinides (Repaglinide and nateglinide), biguanides (Metformin), Thiazolidinediones (rosiglitazone, pioglitazone), D-phenylalanine derivatives, meglitinides, α -glucosidase inhibitors, sulfonylureas (glyburide), and SGLT2 inhibitors (dapagliflozin), which are utilized to lower the blood glucose level to a safe range. The undesirable side effects (kidney damage, bloating, weight gain, dizziness, gastrointestinal abnormalities, palpitation, abdominal pain, etc.) could be named as the major setback of the long-term use of these medications [5–7]. Herbal medicine has been the basis of traditional medicine for centuries. A considerably large population of the world relies on plant-based natural medication for diabetes, cancer, microbial infections, etc. primarily due to the fact that they seem to develop fewer side effects. Naturally occurring therapeutic phytochemicals could be either primary or secondary metabolites of the plant. Hence, developing drugs based on phytotherapeutic compounds after careful analysis and screening procedures (for toxicity, etc.) could potentially provide credible alternatives. *Mikania cordata* is a resource that has not yet been explored exclusively for several medicinal properties but is known to be in use in traditional practice [8]. This is the first study detailing the *in vitro* antimicrobial activity of the methanol, ethyl acetate, and aqueous extracts of *Mikania cordata* leaves, thin layer chromatography- (TLC-) mediated separation of an antimicrobial compound, and hypoglycemic activity of *Mikania cordata* leaves.

2. Materials and Methods

2.1. Plant Material Collection. *Mikania cordata* plant parts were collected from Sri Jayewardenepura Kotte, Sri Lanka (6°54'8.218" N 79°54'15.152" E). Plant voucher herbarium specimen was prepared [9], and its authentication was performed by the National Herbarium of the Royal Botanical Gardens, Peradeniya, Sri Lanka.

2.2. Human Pathogenic Microorganisms Utilized. A representative set of standard cultures of human pathogens were used.

- (a) *Bacillus cereus* (ATCC 11778)
- (b) *Staphylococcus aureus* (ATCC 25923)
- (c) *Escherichia coli* (ATCC 25922)
- (d) *Pseudomonas aeruginosa* (ATCC 25853)
- (e) *Candida albicans* (ATCC 10231)
- (f) *Candida tropicalis* (ATCC 13803)
- (g) *Candida parapsilosis* (ATCC 22019)

2.3. Preparation of Aqueous and Organic Plant Extracts. A published protocol [10] was followed with few modifications. Fresh and uninfected *Mikania cordata* leaves were collected and washed with running tap water until dust and dirt were removed. Excess water was removed by patting the leaves on blotting papers. The leaves were then left in a 37°C

hot air oven for 6 days for complete drying until a constant weight was obtained. Next, a coarse powder of the dried leaves was prepared. A sequential extraction procedure was used to prepare the leaf extracts with analytical grade ethyl acetate (EA), methanol (ME), and distilled water (DW). Powdered leaf weighing 25 g was extracted into 75 ml of EA (1:3 w/v). The conical flask containing the mixture was placed on the shaker (150 rpm, room temperature) for three days followed by filtration through a double-layered muslin cloth and then through a Whatman No. 1 filter paper. This procedure was repeated twice with additional 75 ml aliquots of EA (conical flask was left on the shaker for 18 h at 150 rpm at room temperature). The three separate fractions were then pooled together and centrifuged (3000 rpm, 10 min), and the supernatants were collected. An angular rotary evaporator (Buchi R-124, Switzerland) (150 rpm at 38°C) was used to evaporate the supernatant to dryness. The residue was air-dried and it was used to obtain the ME and DW fractions using the same procedure that was applied to obtain *Mikania cordata* leaf ethyl acetate (MEA) fraction. A freeze dryer (FD-2.5 E, Israel) at –70°C was used for the aqueous fraction. The dried crude extracts were stored at –20°C in airtight containers until use. The following equation was used to calculate the yield percentage of EA, ME, and DW extracts:

$$\text{Percentage of yield} = \frac{\text{final weight of the dried plant extract}}{\text{initial dry weight of the plant leaves}} \times 100\% \quad (1)$$

2.4. In Vitro Antimicrobial Assay of Plant Leaf Fractions. Kirby-Bauer disk diffusion method was followed [11].

Filter paper disks of 6 mm diameter were prepared using Whatman No. 1 filter papers. The disks were impregnated with 10 μ l of EA fraction and dried, and the procedure was repeated twice so that the final disk was containing 30 μ l of EA plant leaf extract. The extraction solvent was used to transfer the crude extracts onto the filter paper disks.

Negative control was 30 μ l/disk of EA.

Chloramphenicol 30 μ g/disk was the positive control for *S. aureus* and *B. Cereus*.

Gentamycin 10 μ g/disk was the positive control for *P. aeruginosa* and *E. coli*.

Three MEA disks, a negative control disk, and a positive control disk were placed on each Mueller Hinton Agar (MHA) plate seeded with a pathogenic bacterium (turbidity of 0.5 McFarland standards). The experiment was triplicated. They were incubated at 37°C for 24 h.

A similar procedure was followed to *Mikania cordata* leaf methanol (MME) and leaf aqueous (MDW) fractions where the negative controls were methanol and sterile distilled water, respectively.

The mean diameter of zones of inhibition (ZOI) measured to the nearest millimeter were recorded following the overnight incubation.

The antifungal assay was carried out according to a similar procedure. The modification made was the use of 2% glucose (w/v) MHA (2% g-MHA) so that the medium is more suitable for fungal growth. Fluconazole (25 μ g/disk)

was used as the positive control for *C. albicans*, and ketoconazole (15 µg/disk) was used as the positive control for *C. parapsilosis* and *C. tropicalis*. The extraction solvents used to obtain the leaf crude extracts were used as negative controls.

2.5. In Vitro Determination of Minimum Inhibitory Concentration (MIC) of *M. cordata* Leaf Fractions against the Selected Human Pathogens. MME fraction was chosen to determine the MIC against *B. cereus*, *S. aureus*, *C. albicans*, and *E. coli*. MDW fraction was chosen to determine the MIC against *B. cereus*, *S. aureus*, *C. albicans*, and *E. coli*. MEA fraction was chosen to determine the MIC against *B. cereus*, *E. coli*, and *S. aureus*.

The broth microdilution method performed using sterile 96-well microdilution plates was used to determine the MIC ranges. The experiments were performed according to CLSI standards [12] and Eloff [13].

The weight of the MME was measured and it was dissolved in 1 ml of double strength Mueller Hinton Broth (x2 MHB) to prepare a working solution of 36 mg/ml.

A positive control was 0.56 mg/ml of Chloramphenicol in 100 µl of x2 MHB and 100 µl of the pathogenic bacterial suspension. Gentamycin was used for *E. coli* and Fluconazole for *C. albicans*.

Negative control was 100 µl of x2 MHB and 100 µl of pathogenic suspension.

Blank was 100 µl of x2 MHB and 100 µl of SDW.

Double strength of MHB supplemented with 2% glucose was used to prepare the crude extract dilutions for *Candida albicans*. The plates were prepared in triplicate. The microdilution plates were incubated at 37°C for 24 h.

A similar procedure was used to determine the MIC of the MDW and MEA against the listed set of test pathogenic microorganisms.

2.6. Thin Layer Chromatography (TLC) Indirect Bioautography for Detection of Antimicrobial Compound(s) in MEA. The MEA extract was chosen for TLC separation of antimicrobial compounds and indirect bioautography based on the results observed for its antimicrobial potential. The published protocol was followed on a few modifications on

the volumes utilized [14]. A volume of 10 µl of MEA was spotted on a precoated Kieselgel 60 F₂₅₄ TLC plate, and it was developed using a solvent system of EA and *n*-hexane (7:3 v/v). The TLC plate was removed when the solvent front reached an approximate level of 0.5 cm from the top margin of the TLC plate. The plate was dried at 40°C for three days. The visualization of the developed spots was done using iodine vapor and UV₃₁₂ (Vilber Lourmat, TFX-20M, France). An MHA plate was seeded with *B. cereus* (turbidity of 0.5 McFarland standards). The developed and dried TLC plate was overlaid on the seeded plate so that the silica-coated side was in direct contact with the agar surface. It was incubated at 4°C for 1 h to allow the metabolites to diffuse onto the agar. Then, the TLC plate was removed, and the plate was incubated further at 37°C for 24 h. The presence of compound/s with antimicrobial potential was identified by inhibition zone/s on the MHA plate. The *R_f* value was calculated using the following equation:

$$R_f = \frac{\text{distance travelled by the solute (cm)}}{\text{distance travelled by the solvent (cm)}} \quad (2)$$

2.7. Evaluation of Antidiabetic Activity of *Mikania cordata* Leaf Extracts. The assay was performed according to Cirillo [15]. One gram of commercial baker's yeast was washed with distilled water and was repeatedly centrifuged (3,000 x g, 5 min) until a clear supernatant fluid was obtained. A 10% (v/v) suspension was prepared by adding distilled water to the supernatant after discarding the pellets. Varying concentrations (1–5 mg/ml) of *M. cordata* leaves extracted into EA were prepared. They were supplemented with 1 ml of glucose solution (5, 10, 25 mM) and the mixtures in a 1:1 ratio and were incubated at 37°C for 10 minutes. The reaction was initiated by adding 100 µL of yeast suspension to each of the incubated mixtures of glucose solution and plant extract. They were vortexed and incubated at 37°C for 60 minutes. Incubation was followed by centrifugation of the tubes (2,500 x g, 5 min), and the glucose in the supernatant was estimated by measuring the absorbance at 540 nm using the UV spectrophotometer (CromTech CT-8200, China). The percentage increase in glucose uptake by the yeast cells was calculated using the following equation:

$$\text{increase in glucose uptake} = \frac{(\text{absorbance of control}) - (\text{absorbance of sample})}{(\text{absorbance of control})} \times 100\%. \quad (3)$$

The negative control contained all reagents except for plant extract. The positive control used was Metformin (1–5 mg/ml) which is a standard drug. All the experiments were triplicated.

2.8. Statistical Analysis. Minitab 17 was used to perform one-way ANOVA and pairwise Tukey tests. The results/data were considered significantly different given that $p \leq 0.05$.

3. Results and Discussion

The percentage of yield depicts the yield of extractable metabolites in aqueous and organic solvents. The highest percentage of yield was exhibited in the ME fraction of the leaves. A similar degree of extractability was depicted in the DW fraction. This could be due to the fact that the plant metabolites are dissolved in the cell sap where the solvent is water, and hence, the metabolites are more polar. This could

lead to higher extractability to be observed in polar solvents (water and methanol). The lowest but a considerable percentage of yield was shown by the EA fraction (Table 1).

The leaf materials should be dried well when extracting into organic solvents. This is to prevent the compounds from being partitioned between the organic solvents as well as in the inherent moisture of the sample [16]. The leaf materials were sequentially extracted into organic solvents with increasing polarity (the polarity increases in the order of ethyl acetate < methanol < water). This method ensures that the active metabolites of the plant leaves are extracted according to their polarities. For better extraction of metabolites from the leaf samples, a mixture of organic solvents could be used. The polarity and the size of the secondary compounds determine the extractability when organic solvents are used [17]. The extracts should be concentrated and stored without directly storing them while still in the solvent to avoid artifact formation and isomerization of different contents found in the extract.

Broad-spectrum antimicrobial activity was depicted by leaf methanol (MME), aqueous (MDW), and ethyl acetate (MEA) crude extracts. Both MME and MDW fractions exhibited antimicrobial activity against gram-positive *B. cereus*, *S. aureus*, gram-negative *E. coli*, and *C. albicans*, while the MEA exhibited antibacterial properties against gram-positive *B. cereus*, *S. aureus*, and gram-negative *E. coli* (Table 2).

The gram-positive *B. cereus*, *S. aureus*, and the gram-negative *E. coli* showed susceptibility to the ethyl acetate crude plant leaf fraction. However, the MEA fraction was observed to be highly effective against *B. cereus*. Plant leaf methanol fraction (MME) was susceptible to *E. coli* but showed intermediate antimicrobial activity against *B. cereus*, *S. aureus*, and *C. albicans*. The aqueous fraction of the plant leaves showed intermediate antimicrobial activity against all four pathogens (Table 2). The plant leaf MEA fraction showed comparatively a higher antimicrobial activity against the three bacterial pathogens than that shown by MME and MDW. Antibacterial activities against *B. cereus*, *E. coli*, and *S. aureus* have been observed in the ethanol extract of *M. cordata* leaves [18].

None of the plant fractions depicted antimicrobial activity against *P. aeruginosa*, *C. parapsilosis*, and *C. tropicalis*. The reason could be the lack of antimicrobial compounds that could cause an inhibition on the growth and activity of the abovementioned pathogens or the presence of them in extremely minute quantities that do not satisfy the minimum inhibitory concentrations needed by the pathogens to achieve effective inhibition. The bioactive compounds might have been denatured or volatilized when concentrating the fractions in the rotary evaporator and the freeze dryer at temperatures 40°C and -70°C for the organic fractions and the aqueous fractions, respectively. The decomposition of thermolabile bioactive compounds can be reduced by maintaining a water bath at a temperature below 40°C. Antimicrobial compounds have been reported to be present in volatile oils of many medicinal plants. These include monoterpenes, sesquiterpenes, low molecular weight aliphatic hydrocarbons, acids, alcohols, and aldehydes [19].

The solvent used in the preparation of the crude extract plays a major role in the degree of antimicrobial activity observed. Some organic solvents such as methanol and chloroform are toxic to living cells. Therefore, the ideal mode of extraction is into an aqueous medium. The natural solvent present in plant cells is water, and hence most of the bioactive components are water soluble. The method of extraction should be the same even though the solvent phase may be changed. This is to ensure reproducibility and also to assess the extraction efficiency [20].

The MHA is considered as the ideal medium for antibiotic susceptible tests owing to its higher degree of reproducibility, simple technicality, and inexpensiveness and is ideal for the growth of nonfastidious bacteria. The quality of the medium places a vital role in providing accurate and reproducible results. The concentration of free cations remaining in the medium after autoclaving is one major variable that determines the final quality of the medium. Excessive cations will reduce the size of ZOI while a low number of cations could result in unacceptably large ZOI. When MHA is supplemented with 2% of glucose, it facilitates fungal growth. The addition of methylene blue could enhance the edge of zones [21].

A similar range of MIC (1.13 > MIC > 0.56) was observed for the MME and MEA fractions, against all four pathogens. The MIC range observed for MDW was higher than that of MME and MEA for *B. cereus*, *S. aureus*, and *C. albicans* which suggests that the bioactive compounds that have been extracted into methanol and ethyl acetate fractions are comparatively stronger in activity than those extracted into an aqueous medium (Table 3). Similar studies of *M. cordata* have not been reported in the literature. However, the evaluation of antibacterial properties of crude EA extract of *Mikania micrantha* leaves has shown the exact MIC value of *S. aureus* to be 1.25 mg/ml [22]. The results obtained for the range of MIC suggest that *Mikania cordata* crude EA leaf extract depicts better antibacterial activity against *S. aureus* than that of *M. micrantha*.

The TLC-indirect bioautography carried out for the MEA indicated the detection of a single compound that owed for the single inhibition zone against *B. cereus*. The R_f value of the corresponding antibacterial compound was 0.64 and was not detected when the TLC plate was developed under UV₃₁₂. The compounds that were detected when the TLC plate was developed using UV₃₁₂ did not demonstrate any antimicrobial activities. The compounds visualized when viewed using UV₃₁₂ and iodine vapor are mentioned in Table 4. The analyses of antibacterial compounds of *Mikania cordata* leaf extracts using TLC have not yet been cited in the literature.

The results obtained indicated that MEA increased the glucose uptake by the yeast cells as its concentration was increased gradually. The trend observed for MEA was similar to that of the standard drug, Metformin, in a glucose dose-independent manner. The degree of enhancement of glucose uptake brought about by MEA against 10 mM and 25 mM glucose concentrations was closely similar to that exhibited by Metformin (Figures 1 and 2). The maximum increase in the glucose uptake was in the presence of 5 mg/ml of MEA at all glucose concentrations evaluated. The effect of glucose uptake by yeast cells was directly related to the concentration of glucose. Therefore, the maximum effect of MEA was observed

TABLE 1: Percentage of the yield of *M. cordata* leaf extracts in different solvents.

Initial dry weight of the plant leaves (g)	Final weight of the dried plant extract (g)			Percentage of yield (%)		
	Ethyl acetate crude fraction	Methanol crude fraction	Aqueous crude fraction	Ethyl acetate crude fraction	Methanol crude fraction	Aqueous crude fraction
25.000	1.632	1.943	1.914	6.528	7.772	7.656

TABLE 2: Effect of antimicrobial activity of *M. cordata* leaf aqueous (MDW), methanol (MME), and ethyl acetate (MEA) crude extracts (mean values sharing common letters in each row are not significantly different, $p \leq 0.05$).

Test pathogenic organism	Mean diameter of the ZOI \pm SD (mm) for plant crude extracts ($n = 3$)			Mean diameter of the ZOI \pm SD (mm) for positive control ($n = 3$)		
	MDW	MME	MEA	Chloramphenicol (30 μ g/disc)	Gentamycin (10 μ g/disc)	Fluconazole (25 μ g/disc)
<i>B. cereus</i>	14.3 \pm 0.6 ^C	14.7 \pm 0.6 ^C	20.3 \pm 0.3 ^B	22 \pm 0 ^A	—	—
<i>S. aureus</i>	13.0 \pm 0 ^D	15.3 \pm 0.6 ^C	17.7 \pm 0.6 ^B	28 \pm 0 ^A	—	—
<i>E. coli</i>	14.0 \pm 0 ^C	16.0 \pm 0.5 ^B	15.3 \pm 0.3 ^B	—	22 \pm 0 ^A	—
<i>C. albicans</i>	16.0 \pm 0 ^B	15.7 \pm 0.3 ^B	—	—	—	40 \pm 0 ^A

TABLE 3: Range of MIC values of the crude aqueous (MDW), methanol (MME), and ethyl acetate (MEA) leaf extracts of *M. cordata* leaves against the selected human pathogenic microorganisms by broth microdilution method.

Test pathogenic organism	Range of MIC (mg/ml)		
	MDW	MME	MEA
<i>B. cereus</i>	2.25 > MIC > 1.13	1.13 > MIC > 0.56	1.13 > MIC > 0.56
<i>S. aureus</i>	1.13 > MIC > 0.56	1.13 > MIC > 0.56	1.13 > MIC > 0.56
<i>E. coli</i>	2.25 > MIC > 1.13	1.13 > MIC > 0.56	1.13 > MIC > 0.56
<i>C. albicans</i>	2.25 > MIC > 1.13	1.13 > MIC > 0.56	—

TABLE 4: Thin-layer chromatography (TLC) analysis of the ethyl acetate fraction of the *Mikania cordata* leaves after developing the plate in ethyl acetate and n-hexane (7:3 v/v) solvent system.

Visualizing agent	Number of spot/s observed on the TLC plate	R_f value/s
Iodine vapor	1	0.64
UV ₃₁₂	2	0.55 and 0.80

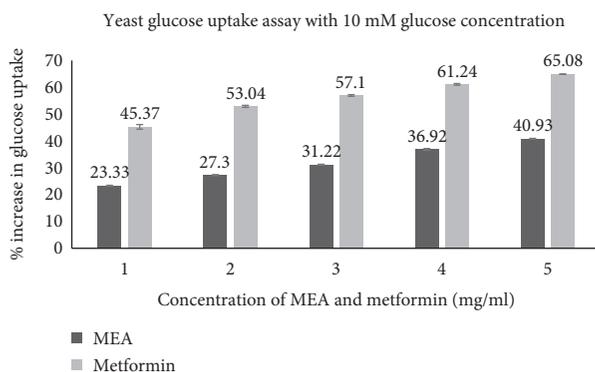


FIGURE 1: The effect of different concentrations (1.0–5.0 mg/ml) of ethyl acetate extract of *Mikania cordata* leaves (MEA) on the percentage increase in glucose uptake by yeast cells at 10 mM glucose concentration. Similar concentrations (1.0–5.0 mg/ml) of Metformin as the control. Values are expressed as mean \pm SD of triplicate data. The error bars represent \pm SE of triplicate data.

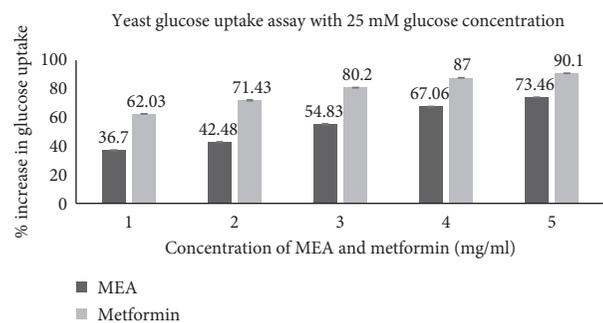


FIGURE 2: The effect of different concentrations (1.0–5.0 mg/ml) of ethyl acetate extract of *Mikania cordata* leaves (MEA) on the percentage increase in glucose uptake by yeast cells at 25 mM glucose concentration. Similar concentrations (1.0–5.0 mg/ml) of Metformin as the control. Values are expressed as mean \pm SD of triplicate data. The error bars represent \pm SE of triplicate data.

in the presence of 25 mM glucose concentration. The uptake of glucose by yeast cells brought about by MEA at 25 mM

concentration of glucose was 1.6 times higher than that at 10 mM glucose concentration and 9.0 times greater than that at 5 mM glucose concentration (Figure 3) at the lowest

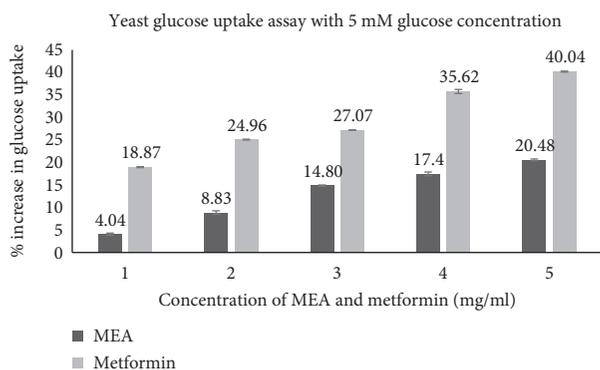


FIGURE 3: The effect of different concentrations (1.0–5.0 mg/ml) of ethyl acetate extract of *Mikania cordata* leaves (MEA) on the percentage increase in glucose uptake by yeast cells at 5 mM glucose concentration. Similar concentrations (1.0–5.0 mg/ml) of Metformin as the control. Values are expressed as mean \pm SD of triplicate data. The error bars represent \pm SE of triplicate data.

concentration of MEA. There is no available information on the antidiabetic effect of *M. cordata* to further compare and analyze the obtained results. However, the trend observed is similar to that obtained for *Cinnamomum zeylanicum* which is well known for its antidiabetic activity [23, 24].

The uptake of glucose by *Saccharomyces cerevisiae* is complex and occurs across a concentration gradient via facilitated diffusion. It is reported that the transport of sugars across the yeast cell membranes occurs via forms of membrane carriers which are stereospecific. The continuity of glucose uptake into the cell is mediated by the degree of removal of intracellular glucose [25]. The results obtained in this study conform to the fact that the *Mikania cordata* ethyl acetate leaf extract is capable of enhancing the uptake of glucose, which in turn suggests that it is capable of enhancing the utilization of glucose by yeast, hence, maintaining the blood glucose level at a normal.

The effect of the plant extract on its ability to extend the time taken to release glucose from starch could be assessed. The glucose dialysis retardation index (GDRI) could also be determined to evaluate the effect of the plant extract on α -amylase which determines the release of glucose from starch [26].

The yeast model is rather efficient and convenient to screen plant extracts for their antidiabetic activity before implementing time and resources for intensive research.

4. Conclusions

This is the first study of exploring the *in vitro* antidiabetic potential of *Mikania cordata* and the TLC-mediated separation of the antimicrobial compounds from the ethyl acetate extract of *M. cordata* leaves. The ethyl acetate extract of *Mikania cordata* (MEA) leaves was susceptible against *B. cereus* and *S. aureus* while the methanol extract (MME) and the aqueous extract were found to be effective against *B. cereus*, *S. aureus*, *E. coli*, and *C. albicans*. The greatest degree of inhibition was observed by MEA. The minimum inhibitory concentrations of MEA (against *B. cereus*,

S. aureus, and *E. coli*) and MME (against *B. cereus*, *S. aureus*, *E. coli*, and *C. albicans*) were in the range of $1.13 > \text{MIC} > 0.56$ mg/ml. An antimicrobial compound was separated from MEA based on TLC with an R_f value of 0.64 which was effective against *B. cereus*. The results of this study indicate the antidiabetic properties of *M. cordata* leaves which followed a similar pattern to Metformin. Therefore, it is evident that *Mikania cordata* harbors a vast amount of useful bioactive compounds that await exploration.

Data Availability

All data that support the conclusions of this study are described in the article.

Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

Acknowledgments

This study was supported by the Department of Botany (Faculty of Applied Sciences) and Department of Microbiology (Faculty of Medical Sciences), University of Sri Jayewardenepura, Sri Lanka. The authors acknowledge Dr. S.A Krishnarajah (Director General, Royal Botanical Gardens, Sri Lanka), Mr. K. Piyasena (Retired Director, Seed Certification & Plant Protection Centre, Ministry of Agricultural Development, Sri Lanka), and Dr. R.A.S.W Ranasinghe (Deputy Director, National Herbarium, Peradeniya, Sri Lanka) for the guidance and assistance rendered in plant authentication.

References

- [1] G. S. Tillotson and S. H. Zinner, "Burden of antimicrobial resistance in an era of decreasing susceptibility," *Expert Review of Anti-infective Therapy*, vol. 15, no. 7, pp. 663–676, 2017.
- [2] M. B. D. O. Chagas, I. Prazeres dos Santos, L. C. Nascimento da Silva et al., "Antimicrobial activity of cultivable endophytic fungi associated with *hancornia speciosa* gomes bark," *The Open Microbiology Journal*, vol. 11, no. 1, pp. 179–188, 2017.
- [3] R. K. Paul, A. Jabbar, and M. A. Rashid, "Antiulcer activity of *Mikania cordata*," *Fitoterapia*, vol. 71, no. 6, pp. 701–703, 2000.
- [4] V. P. De Almeida, A. A. Hirt, P. A. Raeski et al., "Comparative morphoanatomical analysis of *Mikania* species," *Revista Brasileira de Farmacognosia*, vol. 27, no. 1, pp. 9–19, 2017.
- [5] P. R. Rehani, H. Iftikhar, M. Nakajima, T. Tanaka, Z. Jabbar, and R. N. Rehani, "Safety and mode of action of diabetes medications in comparison with 5-aminolevulinic acid (5-ALA)," *Journal of Diabetes Research*, vol. 2019, Article ID 4267357, 10 pages, 2019.
- [6] Y.-W. Wang, S.-J. He, X. Feng et al., "Metformin: a review of its potential indications," *Drug Design, Development and Therapy*, vol. 11, pp. 2421–2429, 2017.
- [7] E. Otto-Buczkowska and N. Jainta, "Pharmacological treatment in diabetes mellitus type 1–insulin and what else?" *International Journal of Endocrinology and Metabolism*, vol. 16, no. 1, 2017.

- [8] A. K. Kiang, K. Y. Sim, and S. W. Yoong, "Constituents of *Mikania cordata* (burm. f.) B. L. Robinson (compositae)-II," *Phytochemistry*, vol. 7, no. 6, pp. 1035–1037, 1968.
- [9] K. Maden, "Plant collection and herbarium techniques," *Our Nature*, vol. 2, no. 1, pp. 53–57, 2004.
- [10] M. K. Pathmanathan, K. Uthayarasa, J. Jeyadevan, and E. C. Jayaseelan, "In vitro antibacterial activity and phytochemical analysis of some selected medicinal plants," *International Journal of Pharmaceutical & Biological Archives*, vol. 1, no. 3, pp. 291–299, 2010.
- [11] A. W. Bauer, W. M. M. Kirby, J. C. Sherris, and M. Turck, "Antibiotic susceptibility testing by a standardized single disk method," *American Journal of Clinical Pathology*, vol. 45, no. 4-ts, pp. 493–496, 1966.
- [12] CLSI, *Reference Method for Broth Dilution on Antifungal Susceptibility Testing of Yeast*, CLSI, Wayne, PA, USA, 2017, https://clsi.org/media/1897/m27ed4_sample.pdf, 4th edition.
- [13] J. Eloff, "A sensitive and quick microplate method to determine the minimal inhibitory concentration of plant extracts for bacteria," *Planta Medica*, vol. 64, no. 8, pp. 711–713, 1998.
- [14] D. L. Valle, J. J. M. Puzon, E. C. Cabrera, and W. L. Rivera, "Thin layer chromatography-bioautography and gas chromatography-mass spectrometry of antimicrobial leaf extracts from PhilippinePiper betleL. Against multidrug-resistant bacteria," *Evidence-Based Complementary and Alternative Medicine*, vol. 2016, Article ID 4976791, 7 pages, 2016.
- [15] V. P. Cirillo, "Mechanism of glucose transport across the yeast cell membrane," *Journal of Bacteriology*, vol. 84, no. 3, pp. 485–491, 1962.
- [16] W. P. Jones and A. D. Kinghorn, "Extraction of plant secondary metabolites," *Natural Products Isolation*, vol. 20, pp. 323–351, 2006.
- [17] E. C. Jeyaseelan, S. Jenothiny, M. Pathmanathan, and J. Jeyadevan, "Antibacterial activity of sequentially extracted organic solvent extracts of fruits, flowers and leaves of *Lawsonia inermis* L. from Jaffna," *Asian Pacific Journal of Tropical Biomedicine*, vol. 2, no. 10, pp. 798–802, 2012.
- [18] M. S. Ali, M. S. Islam, M. M. Rahman, M. R Islam, M. A Sayeed, and M. R Islam, "Antibacterial and cytotoxic activity of ethanol extract of *Mikania cordata* (burm.F.) B.L. Robinson leaves," *Journal of Basic and Clinical Pharmacy*, vol. 2, no. 2, pp. 103–107, 2011.
- [19] H. J. D. Dorman and S. G. Deans, "Antimicrobial agents from plants: antibacterial activity of plant volatile oils," *Journal of Applied Microbiology*, vol. 88, no. 2, pp. 308–316, 2000.
- [20] M. I. Ngaha njila, E. Mahdi, D. Massoma lembe, Z. NDE, and D. Nyonsue, "Review on extraction and isolation of plant secondary metabolites," in *Proceedings of the 7th International Conference on Agricultural, Chemical, Biological and Environmental Sciences*, pp. 67–72, Kuala Lumpur Malaysia, May 2017.
- [21] P. R. Murray and J. R. Zeiting, "Evaluation of Mueller-Hinton agar for disk diffusion susceptibility tests," *Journal of Clinical Microbiology*, vol. 18, no. 5, pp. 1269–1271, 1983.
- [22] A. Matawali, L. P. Chin, H. S. Eng, and G. J. Azlan, "Antibacterial and phytochemical investigations of *Mikania micrantha* H.B.K. (Asteraceae) from sabah, Malaysia," *Transactions on Science and Technology*, vol. 3, no. 2, pp. 244–250, 2016.
- [23] S. H. Kim, S. H. Hyun, and S. Y. Choung, "Anti-diabetic effect of cinnamon extract on blood glucose in db/db mice," *Journal of Ethnopharmacology*, vol. 104, no. 1-2, pp. 119–123, 2005.
- [24] T. Woldemariam and J. Van Winkle, "In Vitro hypoglycemic effect of *Salvia hispanica* using a yeast glucose uptake model," *Journal of Pharmaceutical Sciences and Pharmacology*, vol. 2, no. 2, pp. 119–122, 2015.
- [25] G. Rehman, M. Hamayun, A. Iqbal et al., "In vitro antidiabetic effects and antioxidant potential of *Cassia nemophila* pods," *BioMed Research International*, vol. 2018, no. 3, 6 pages, Article ID 1824790, 2018.
- [26] M. A. Bhutkar, S. D. Bhinge, D. S. Randive, and G. H. Wadkar, "Hypoglycemic effects of *Berberis aristata* and *Tamarindus indica* extracts in vitro," *Bulletin of Faculty of Pharmacy, Cairo University, Cairo University*, vol. 55, no. 1, pp. 91–94, 2017.