



**INVESTIGATE THE BIOACTIVE SPECTRUM OF THREE VARIOUS
PLANT EXTRACTS AGAINST WOUND CAUSING DIABETIC
PATHOGENS**

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ABSTRACT

Medicinal plants are a rich source of antimicrobial agents and provide a safer and cost effective way of treating infections against the pathogenic microbes. The present study describes the phytochemical profile and antimicrobial activity of three different medicinal plants such as *Andrographis paniculata*, *Senna auriculata* and *Stevia rebaudiana* along with three various solvents such as Acetone,

Methanol. Ethanol, Chloroform and Control. Among the three plant, *S. auriculata* possessed widely seven phytochemical compounds such as Flavonoids, Tannins, Saponins, phenols, glycosides steroids and Coumarins and etc, In addition *A. paniculata* and *S. rebaudiana* extract showed only some phytochemical compounds such as four and five respectively.

Antibacterial Spectrum: Besides, the antibacterial spectrum study made on five various diabetic wound pathogenic organisms; initially minimum (3 ± 1.14 for *Pseudomonas*, 3 ± 0.67 for *E.coli* and 3 ± 1.03 for *Streptococcus*) to maximum (14 ± 3.03 for *E. coli*) zone of inhibition was noticed on Chloroform and Acetone extract. It was the significant effect of the antibacterial activity against the wound pathogen. The second Plant extracts of *S. auriculata* showed a significantly immensed activity against *Pseudomonas* in acetone extract (8 ± 0.45 mm) also the similar effect has been noticed on methanol extract against the both organisms of *Streptococcus* (8 ± 2.34) and *Klebsiella* (8 ± 2.05) respectively. Moreover, the third plant such as *S. rebaudiana* showed a significantly maximum activity against

Pseudomonas in acetone extract (18 ± 2.30 mm) it was one fold higher activity when compared with its control. Meanwhile, the highest concentration of 100 μ l also been given the least activity in chloroform extract (8 ± 1.36) noted against the **diabetic wound pathogenic** organism of *Pseudomonas*. **GC-MS Analysis:** The methanolic extract of *S. rebaudiana* plant was subjected to GC-MS analysis and the meticulous compounds elucidation such as the active principle, area of the peak, Concentration (%) and Retention Time (RT) are presented in present on table-5. Initially the two identical prevailing peak compounds were observed such as 1-Naphthalenepropanol alpha.-ethenylde1-and Naphthalenepentanol, decahydro-5-(hydr) and its representing abundance is similar range 525% but retention time was varied from 32.7 to 34.19. Moreover, other second peak compound was 3, 7, 11, 15-Tetramethyl-2-hexadecen-1-ol noted at 24.0 mints of retention time with 410% of abundance. From the present result clearly indicated that these three plant extracts are act as a excellent antibacterial activity against the **diabetic wound pathogenic** organism.

KEYWORDS: Andrographis paniculata, Senna auriculata and Stevia rebaudiana, diabetic wound pathogenic.

INTRODUCTION

Medicinal plants have been used for centuries as remedies for human diseases because they contain components of therapeutic value (Jamine *et al.*, 2007). About three quarter of the world's population relies on plants and their extracts for their health care. India represented by rich culture, traditions and natural biodiversity, offers a unique opportunity for drug discovery researchers (Jachak, and Saklani, 2007). The increasing failure of chemotherapeutics and antibiotic resistance exhibited by pathogenic microbial infectious agents has lead to the screening of several medicinal plants for their potential antimicrobial activity (Imran *et al.*, 2010).

The use of plant extracts to treat microbial infections is also reported in our ancient Ayurvedic Compendium 'Charak and Sushrat (Chatterjee and Prakash, 1994) and other several researchers have carried out screening of plant extract for antimicrobial screening of plant extract for antimicrobial activities and new effective drugs having natural or synthetic origin Plant extracts and their products are clinically safer than antibiotics (Kelmanson, 2000; Srinivasan, 2001). Another important experimental plant of *Senna auriculata* has the medicinal property as follows. The root is used in decoctions against fever, diabetes, diseases of urinary system and constipation. The leaves have laxative properties (Subhadradevi *et al.*,

2011). The dried flowers and flower buds are used as a substitute for tea in case of diabetes patients. It is also believed to improve the complexion in women. *A. paniculata* is frequently used for preventing and treating the common cold and flu (influenza) (Ivorra *et al.*, 1989). It is used for digestive complaints including diarrhea, constipation, intestinal gas, colic and stomach pain for liver conditions including an enlarged liver, jaundice and liver damaged due to medications for infectious diseases (Shahid, 2011). The leaves and seeds of the *C. roseus* contain vincamine, precursor to the chemical vinpocetine, which is used medicinally to naturally enhance memory in aging, minds. Both leaf and flower extracts were found to have antidiabetic activity.

The medicinal use of *S. rebaudiana* includes regulating blood sugar, preventing hypertension, treatment of skin disorder and prevention of tooth decay. *Diabetes mellitus* is the name given to the group of disorders characterized by absent or deficient insulin secretion or peripheral insulin resistance resulting in hypoglycemia. Impaired metabolism of a number of biomolecules such as glucose, lipids, proteins and glycoproteins has been reported (Dhawan *et al.*, 1996). For a variety of reasons, Indians have a larger than normal likelihood of developing diabetes mellitus. The disease promises to be one of the major health care burdens for this country in the coming decades. For reasons of genetics and lifestyles, Indians form the world's largest diabetic population (Meena *et al.*, 2006). There is no other information available from the previous literature. Since the present work has been designed the following objectives such as study the bioactive compounds from the three various extracts from the experimental plant(s), followed by isolate and characterized the diabetic pathogens from wound samples, Then assessed the antimicrobial spectrum of selected plant extracts against the diabetic pathogens. At last to find out the important bioactive phyto-compounds from the experimental plants by GCMS analysis

MATERIALS AND METHODS

Collection and Preparation of Selective plant Extracts

Five different plants selected for antimicrobial activity and phyto chemical screening they were named as *Stevia rebaudiana*, *Azadirachta indica*, *Andrographis paniculata*, *Vinca rosea*, *Senna auriculata*. The fresh and healthy leaves of the plants were collected from in and around Kaliakkavilai.

The selective medicinal plant extracts were prepared using acetone, ethanol, methanol, chloroform and distilled water. The plant selected were *Stevia rebaudiana*, *Andrographis*

paniculata, *Vincarosea*, *Senna auriculata*. The extracts were collected and stored for further experimental purpose.

Preparation of plant Extracts

The air dried finely ground plant parts were taken separately in air tight bottles and 10ml of different solvent, (ethanol, methanol, Acetone, Chloroform and Distilled water) were added and kept under dark. After two days, the contents were stirred and filtered using whatmann no: 1 filter paper. The filtrate was collected and stored in sterile glass beakers for further study.

Collection of sample: Wound sample were obtained from the diabetic patients of Grace Hospital Kollemcode.

Isolation of bacterial pathogens

i) Enumeration of Bacterial pathogens: The nutrient agar medium was used for the enumeration of total bacterial population in the sample. It was done by serial dilution and agar plating technique.

Identification of pathogens

a) Enrichment of sample: The samples that are collected for microbiological analysis were enriched by nutrient broth. To 5ml of nutrient broth a loopful of sample was added and incubated at 37°C for 24 hours. After incubation the culture were examined by the following methods and compared with Bergey's manual of Determinative Bacteriology Pathogens were identified by, Morphological identification, Microscopic identification, Biochemical test.

Morphological Identification and Biochemical test: Pathogens were morphologically identified by growth on nutrient agar plates and selective medium. Microscopically identification was done by Gram staining. Pathogens were identified by biochemical test such as Indole, Methyl red test, voges proskauer test, citrate, Triple sugar iron test, and urease test.

Phytochemical Analysis: The plant extracts were screened for the presence or absence of secondary metabolites such as alkaloids, flavonoids, tannis, saponins, steroidal compounds, phenolic compounds, glycosides, coumarin and anthraquinones evaluated by the procedure of Harbour, (2003).

Antimicrobial Activity assay: Antimicrobial activity of the plant extracts was studied against five pathogenic bacteria by the agar well diffusion method. Sterile Muller Hinton

agar plates were prepared bacterial strains. Inoculum was aseptically introduced onto the surface of sterile agar plates and sterilized cotton swabs were used for even distribution of the inoculum. Wells were prepared in the agar plates using a sterile cork borer. The leaf extracts to be tested were prepared various concentration of 10 μ l, 50 μ l and 100 μ l were added to the well. The plates were incubated at 37⁰C for 24 hours. After incubation, the size (diameter) of the inhibition zone was measured.

GC-MS Analysis: GC-MS (Agilent 6890 Series GC coupled to a GC Mate II mass spectrometer) analysis was done using a DB-5 capillary column (J&W Scientific, Inc.), 30 m length \times 0.25 mm i.d. \times 0.25 μ m film, run under the following GC temperature program: initial, 70 $^{\circ}$ C; held for 3.5 min; raised to 160 $^{\circ}$ C at 30 $^{\circ}$ C/min rate; raised to 270 $^{\circ}$ C at 70 $^{\circ}$ C/min rate; raised to 310 $^{\circ}$ C at 35 $^{\circ}$ C/min rate; and finally held at this temperature for 3 min. The injection port, GC interface, and ionization chamber were maintained at 260, 200, and 120 $^{\circ}$ C, respectively. The carrier gas was ultrahigh-purity helium at a 1 mL/min flow rate. The sample injection volume was 1 μ L. The MS detector was a magnetic sector; spectra were acquired in the positive, lower solution, selected-ion monitoring mode. AMPA derivative was observed at 7:23 min (m/z 571, 502, 446, 372), and glyphosate derivative was observed at 7:59 min (m/z 611, 584, 460). Extract of the plant samples were quantified from a calibration curve of derivatized standards.

The Kovats index system has been widely used in the analysis of food flavors, pesticides and essential oil analysis. Kovats retention index, (I) is defined and calculated by following equation (Douglas, 2000).

$$I = 100 N + 100 \log'R (N+n) - \log'R(N).$$

Where

$t'R (N)$ = adjusted retention time of n paraffin hydrocarbon of Carbon number eluting before solute-A.

$T'R (N+n)$ = adjusted retention time of n paraffin hydrocarbon of Carbon number (N+n) eluting after solute A.

$T'R (A)$ = adjusted retention time of solute-A.

The plant extracts (*stevia rebaudiana*) was subjected to GC-MS analysis for the separation of compounds present in it.

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RESULT

Phytochemical Analysis: Phytochemical screening of the plant extracts were done to analyse the constituents present in the plants (Table: 1). The compounds present in the extracts are flavonoids, alkaloid, tannin, saponin, coumarin, anthraquinone, phenols, steroids and glycosides. In addition, other two plants showed the

Table-1: Phytochemical Analysis for selected Plant extracts

S.No	Phytochemical Constituents	<i>Andrographis Paniculata</i>	<i>Senna auriculata</i>	<i>Stevia rebaudiana</i>
1.	Flavonoid	-	+	+
2.	Alkaloid	-	-	-
3.	Tannins	+	+	+
4.	Saponins	-	+	+
5.	Phenols	-	+	+
6.	Glycosides	+	+	-
7.	Steroids	+	+	-
8.	Coumarin	+	+	-
9.	Anthraquinone	-	-	+

The table-1 showed among the three plant, *S. auriculata* possessed widely seven phytochemical compounds such as Flavonoids, Tannins, Saponins, phenols, glycosides steroids and Coumarins and etc, In addition *A. paniculata* and *S. rebaudiana* extract showed only some phytochemical compounds such as four and five respectively.

Table-2: Antimicrobial activity of plant extracts *Andrographis paniculata* against diabetic wound pathogens.

S.No	Organisms	Concentration (µl)	Extracts(mm)				
			Acetone (mm)	Ethanol (mm)	Methanol (mm)	Chloroform (mm)	Dist. Water (mm)
1.	<i>E.coli</i>	100	10±1.25**	5±1.43	1±0.36*	9±1.08**	5±1.64
		50	6±1.06	3±0.67	9±1.65	5±1.07	3±1.11
2.	<i>Staphylococcus</i>	100	14±3.03**	-	9 ±1.52	11±1.24**	9±1.36
		50	10±2.03**	-	-	8±1.47**	5±1.30
3.	<i>Streptococcus</i>	100	7±2.50	5±1.3	8±1.65**	8±2.03**	13±3.42
		50	5±1.02	3±1.03	4±2.04	6±2.05	10±2.81**
4.	<i>Klebsiella</i>	100	7±2.57**	5±1.41	8±1.41	7±0.36**	11±0.65**
		50	6±1.64	3±0.57	5±0.67**	5±1.14	7±1.57
		100	9±2.03	11±1.03**	8±1.46	6±1.65	13±2.64**

5.	<i>Pseudomonas</i>	50	6±1.64	7±2.31	5±0.97	3±1.14*	6±1.64
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*- Significant at 5% level

** - Highly significant of 0.01% level.

^{ls}- Insignificant

Antimicrobial Activity of *A. paniculata* extract against diabetic wound pathogens.

100µl plant extract of *A. paniculata* showed the elevated zone of inhibition was noticed inhibition against *Staphylococcus* (acetone-14±3.0314mm). Similarly *E.coli* and *Staphylococcus* species are effectively noted against acetone and methanolic extract with similar measurement of zone of inhibition such as 10±2.03 and 10±2.03mm. Furthermore, more or less similar size of zone of inhibition was observed against the various experimental organisms like and *E.coli*, *Staphylococcus Klepsiella* and *Pseudomonas sp.*, whereas, minimum (3±1.14 for *Pseudomonas*, 3±0.67 for *E.coli* and 3±1.03 for *Streptococcus*) to maximum (14±3.03 for *E. coli*) zone of inhibition also been noticed with its respective solvent extracts. It was the significant effect of the antibacterial activity against the wound pathogen.

Table: 3 Antimicrobial activity of plant extract *S. auriculata* against diabetic wound pathogens

S.No	Organisms	Concentration (µl)	Extracts(mm)				
			Acetone	Ethanol	Methanol	Chloroform	Distilled Water
1.	<i>E.coli</i>	100	5±1.23	7±1.30	7±0.25	4±1.65*	6±1.41
		50	4±2.3	6±0.54**	5±0.64	3±0.11	5±0.65
2.	<i>Staphylococcus</i>	100	6±2.30 ^{ls}	5±1.47*	7±1.63	6±1.14*	8±.387**
		50	5±1.85	5±0.47*	6±0.54	5±1.32	7±1.47**
3.	<i>Streptococcus</i>	100	6±1.65	6±2.94	8±2.34**	6±0.86**	5±1.38
		50	-	-	-	-	4±1.85
4.	<i>Klebsiella</i>	100	7±2.07	6±1.01	8±2.05**	7±2.10	8±1.31
		50	5±1.31	5±1.11	6±0.54	6±1.30	7±3.61**
5.	<i>Pseudomonas</i>	100	8±0.45**	7±1.30**	7±2.04	6±1.11	5±.35
		50	6±2.04	6±0.56**	5±1.30	5±1.82	4±1.43

*- Significant at 5% level

** - Highly significant of 0.01% level

^{ls}- Insignificant

Antimicrobial Activity of *S. auriculata* extract against diabetic wound pathogens.

100µl plant extract of *S. auriculata* showed a significantly towering activity against *Pseudomonas* in acetone extract (8±0.45mm). In addition similarly further significant activity

also been noticed on methanol extract against the both organisms of *Streptococcus* (8 ± 2.34) and *Klebsiella* (8 ± 2.05) respectively. It was more or less similar response when compared to control. Though, least activity was observed in chloroform extract (3 ± 0.11) against *E.coli*. Surprisingly, streptococcus an organism doesn't give any other remarkable (null) effect on the tree experimental extract but in control expressed fairly optimum range of zone of inhibition. From the overall result showed in this table quietly moderate activity range between (4 ± 2.3 to 8 ± 0.45) was pronounced on the three solvent accompanied with four organisms except streptococcus 50 μ l concentration. Apart from the result clearly depicted Among the three plant that excellent activity was made by *A. paniculata* throughout all the five experimental organisms against diabetic wound.

Table -4: Antimicrobial Activity of Plant Extracts *S. rebaudiana* against diabetic wound.

S.No	Organism	Concentration (μ l)	Extracts(mm)				
			Acetone	Ethanol	Methanol	Chloroform	Distilled Water
1	<i>E. Coli</i>	100	7 ± 1.54	$10\pm 1.15^{**}$	10 ± 2.31	5 ± 0.64	8 ± 0.95
		50	$5\pm 1.64^*$	7 ± 3.31	9 ± 0.34	3 ± 0.11	6 ± 1.30
2	<i>Staphylococcus</i>	100	6 ± 1.56	8 ± 0.65	$12\pm 2.38^{**}$	7 ± 1.04	$10\pm 2.36^{**}$
		50	5 ± 1.06	7 ± 1.65	6 ± 1.54	5 ± 1.02	7 ± 1.05
3.	<i>Streptococcus</i>	100	$6\pm 0.75^*$	5 ± 1.03	$10\pm 0.56^{**}$	6 ± 1.64	7 ± 1.36
		50	-	-	-	-	$5\pm 0.64^{**}$
4	<i>Klebsiella</i>	100	$7\pm 0.57^{**}$	6 ± 0.36	7 ± 0.41	7 ± 2.10	$10\pm 1.0^{**}$
		50	-	-	-	-	5 ± 1.013^{ls}
5	<i>Pseudomonas</i>	100	$18\pm 2.30^{**}$	11 ± 1.47	$10\pm 1.88^{**}$	8 ± 1.36	6 ± 2.44
		50	5 ± 2.94^{ls}	7 ± 1.64	7 ± 2.04	6 ± 1.03	5 ± 0.15

*- Significant at 5% level

** - Highly significant of 0.01% level

^{ls}- Insignificant

Antimicrobial Activity of *S. rebaudiana* extract against diabetic wound pathogens

Plant extracts of *S. rebaudiana* showed a significantly maximum activity against *Pseudomonas* in acetone extract (18 ± 2.30 mm) it was one fold higher activity when compared with its control. Meanwhile, the highest concentration of 100 μ l also been given the least activity in chloroform extract (8 ± 1.36) noted against the organism of *Pseudomonas*. In addition illustrate the average zone 7 ± 1.647 of inhibition have been noticed against the following organism such as, 100 μ l concentration of acetone extract on *E.coli* then at 50 μ l concentration of ethanol extract against both organism of *Staphylococcus sp.*, also methanol

and chloroform extract on *Klebsiella* sp., as well as ethanol and methanol extract against the activity seen on *Pseudomonas* sp.,. Eventhough notably 50µl concentration has been given no effect on any other experimental organism with the treated extract of experimental plant of *S. rebaudiana*. Moreover, minimum to maximum range of zone of inhibition was noticed clearly on 3±0.11 chloroform extract on *E.coli* and *Pseudomonas* 18±2.30 on acetone extract at 100µl concentration respectively.

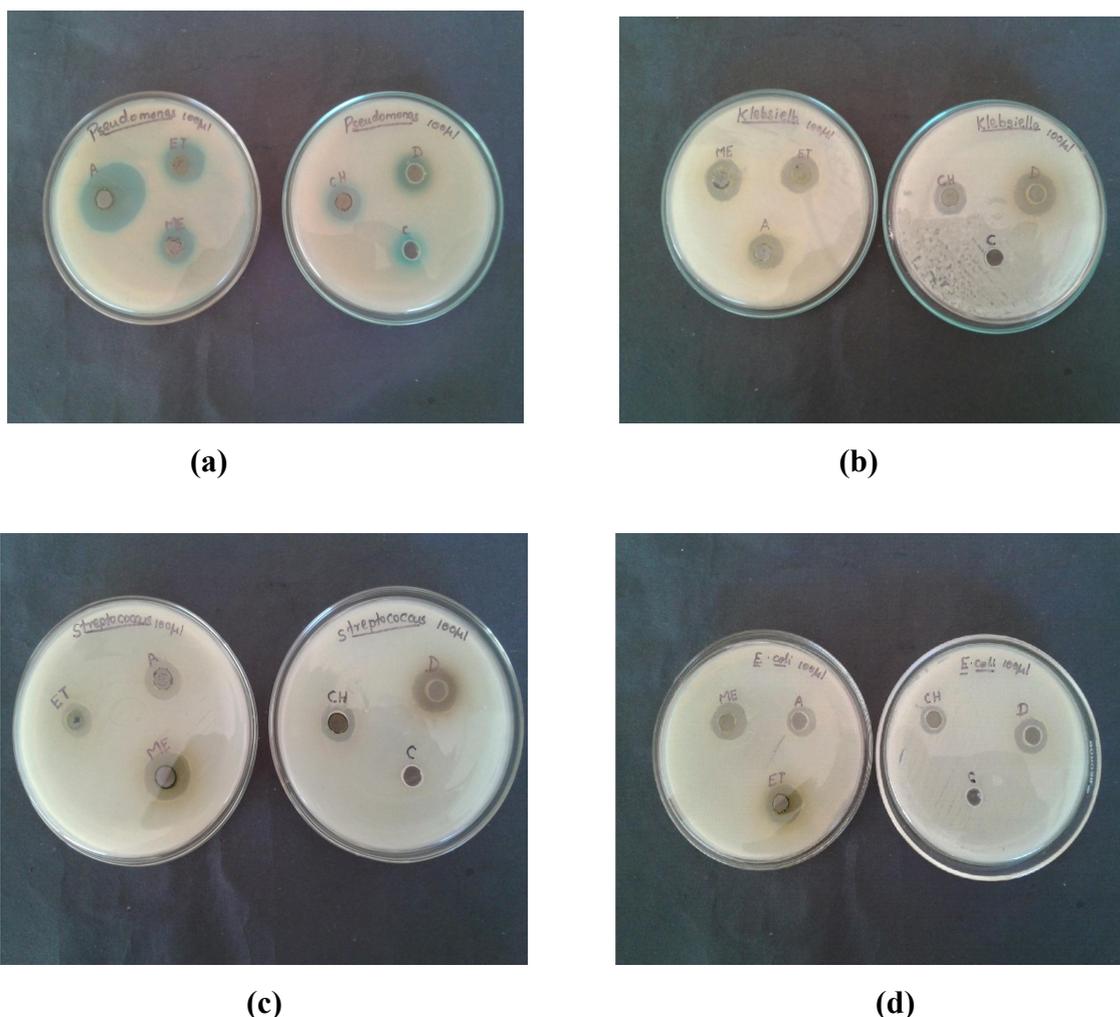


Figure- 1: Antimicrobial spectrum of *S. rebaudiana* Extracts Against (a) *Pseudomonas* sp., (b) *Klebsiella* sp., (c) *Streptococcus* sp., (d) *E.coli* sp.,

A-Acetone, ET-Ethanol, ME -Methanol, CH-Chloroform, D - Distilled water, C – Control.

Table-5: GCMS Bioactive compounds elucidation from more methanolic extract of *S. rebaudiana* plant

S.NO.	RETENTION TIME (RT)	COMPOUNDS SEPARATED (NAME OF THE ANALYTES)	ABUNDANCE (%)
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1.	15.09	1H-Cycloprop[e]azulen-7-ol, decahydro-1,	135
2.	17.18	Naphthalene, 1,2,3,4,4a,5,6,8a-octahydro	250
3.	22.85	2,7-Octanedione, 4,4-dimethyl-3-[2-(1-hy	115
4.	23.29	3-Eicosyne	120
5.	24.5	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	410
6.	25.97	Pentadecanoic acid, 13-methyl-, methyl e	93
7.	26.28	7,9-Di-tert-butyl-1-oxaspiro(4,5)deca-6,	90
8.	26.65	Benzenepropanoic acid, 3,5-bis(1,1-dimet	205
9.	29.88	9,12,15-Octadecatrienoic acid, methylene	145
10.	30.09	Tricyclo (7,16) triacontane, 1	165
11.	32.7	1-Naphthalenepropanol, .alpha.-ethenylde	525
12.	34.19	1-Naphthalenepentanol, decahydro-5- (hydr	525
13.	30.07	Farnesyl bromide	150

GC-MS Analysis: The methanolic extract of *S. rebaudiane* plant was subjected to GC-MS analysis and the particular compounds elucidation such as the active principle, area of the peak, Concentration (%) and Retention Time (RT) are presented in present on table-5. In addition, experimental plant fractionated corresponding peak and other relative compounds are shown figure-5. Initially the two identical prevailing peak compounds were observed such as 1-Naphthalenepropanol .alpha.-ethenylde1-and Naphthalenepentanol, decahydro-5- (hydr) and its representing abundance is similar range 525% but retention time was varied from 32.7 to 34.19. Moreover, other second peak compound was 3, 7, 11, 15-Tetramethyl-2-hexadecen-1-ol noted at 24.0 mints of retention time with 410% of abundance. For instance, other optimum level of the compound also been detected such as Naphthalene, 1, 2, 3, 4, 4a, 5, 6, 8a-octahydro (250%), Benzenepropanoic acid, 3,5-bis (1,1-dimet) (205%). From the present study this experimental plants methanolic fractions totally 13 compounds were gained. Among the thirteen compounds medium compounds also noticed the range between 115 to 165 % that are named as 2, 7-Octanedione, 4,4-dimethyl-3-[2-(1-hy), 3-Eicosyne (120); 1H-Cycloprop[e]azulen-7-ol, decahydro-1 (135%), 9,12,15-Octadecatrienoic acid, methylene (145%), Farnesyl bromide (150) and Tricyclo (7,16) triacontane, (165%). Furthermore, two least Pentadecanoic acid, 13-methyl-, methyl e and 7, 9-Di-tert-butyl-1-oxaspiro(4,5)deca-6, declined level of the bio-compounds are observed with its responsible abundance is 90 and 93% respectively.

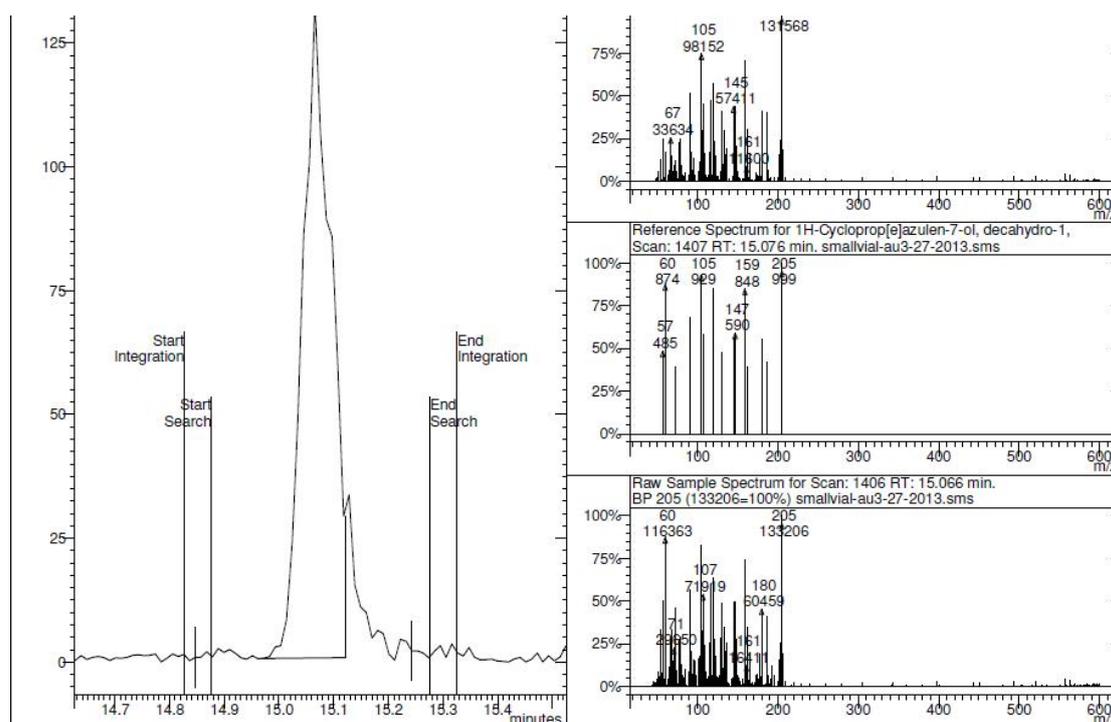


Figure-2: Chromatogram for methanolic extract of *S. rebaudiana* plant

DISCUSSION

The bacterial pathogens were isolated from the samples Anandi *et al.*, (2004) made a study on pathogens of diabetic patients accompanied with diabetic foot lesion. Organisms are predominant found which is also concordance with the findings by Shankar *et al.*, (2005); Bansalet *et al.*, (2007) Dhar *et al.*, (1968) made a study on 103 patients, 157 organisms were isolated. Among the bacterial isolates, gram negative comprised 76 % and positive accounted for 24%. In the present study five different plants were used (*A. paniculata*, *S. incarnosea*, *S.rebaudiana*). Herbal extracts and herbal formulations, used in the ayurvedic literature, have recently been reviewed and have against importance for the control of type-II diabetes depicted by Dhawan *et al.* (1998). They are being used directly or indirectly for the preparation of many modern drugs (Khan *et al.*,2009); Odhav *et al.*, 2010). Medicinal plants have been found useful in the cure of number of diseases including bacterial diseases. Phytochemical constituents of different plant extracts were analysed through a different phytochemical screening test and the compounds present in the plant extracts (Preethi *et al.*, 2011). Similar compounds also extracted such as flavonoid, alkaloid, tannins, anthraquinones, saponins, steroid, coumarins and glycosides also derived by Kiruba *et al.*, (2011). Similar results were noticed in *C. auriculata* leaf extracts (flavonoid, tannins, steroids, phenols, saponins, coumarin and glycosides) by Subhadradevi *et al.*, (2011). Phytochemical analyses for *Stevia rebaudiana* plant extract contain phytochemical

constituents such as alkaloids, steroids, glycosides, anthraquinones, flavanoid, tannins, and saponins and phenols Abdou *et al.* (2011) Phenols also present in the ethanolic extract (Jachak and Saklani, 2007; Preethi *et al.*, 2011).

Antimicrobial activity of plant extracts were done by well diffusion method and the results were tabulated in table 1 to 3. The highest zone was observed in *Stevia rebaudiana* plant extracts (methanol extract) showed high activity against all tested pathogens. Recently, Prabakaran and Pugalvendhan, (2012) reported that among the five types of extracts (acetone, chloroform, ethanol, hexane and petroleum ether) the highest inhibition zone was observed in ethanol extract and second most acetone extract followed by as chloroform, petroleum ether and hexane. Methanolic extract of *A. paniculata* leaf extract showed the significant activity against the tested pathogens (Rajesh *et al.*, 2012).

CONCLUSION

Diabetes mellitus is a chronic disorder and affects large segment of population and is a major health problem. In the present study five medicinal plants extracts were used to evaluate the antimicrobial activity assay. Among three plants selected, *Stevia rebaudiana* showed higher activity against the tested pathogens when compared to commercial antibiotics. Phytochemical analysis revealed that the plant extracts contain flavonoids, alkaloids, tannins, saponins, Glycosides, steroids, phenols, coumarin and anthraquinone. The immense activity of *Stevia rebaudiana* possibly will be the presence of the following compounds such as 1 H – Cycloprop [e] azulen -7- , Naphthalene, 1, 2, 3, 4, 4a, 2, 7 – octanedione, 4, 4 – dim 3 – Eicosyne, 3, 7, 11, 15 – Tetramethy 1 – 2- , 3, 7, 11, 15 – Tetramethy 1 – 2 -, Pentadecanoic acid, 13 – m, 7,9 – Di – tert – buty 1 – 1- oxas, Benzenepropanoic acid, 3, 9, 12, 15 – Octadecatrienoic, Tricyclo [20 . 8 . 0 . 0 (7,16)] 1 – Naphthalenepropanol, 1 – Naphthalenepentanol, d , Farnesy 1 bromide. Therefore the result of the present work indicates that the leaf extracts may be an ideal source of pharmaceutical and natural plant based product. Hence the present study proved that all the three herbal plants were used as a potential antimicrobial source intended for the development of new drugs particularly treatment of diabetes also helps in preventing diabetic complications of antidiabetic drugs.

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