

CHAPTER V

Antimicrobial Property Study of Wild Edible Plants

Plants are important sources of natural antimicrobial compounds and antimicrobials of plant origin are more effective in the treatment of infectious diseases than the synthetic drugs which are often associated with several adverse effects on human health [1]. The healing properties of the plants are due to the presence of various bioactive compounds such as polyphenols, flavonoids, steroids, terpenoids, and alkaloids which have several modes of action on bacterial cells [2, 3]. In recent times, the researchers around the world have given much attention on plants for the development of new and alternative antimicrobial agents or antibiotics for controlling undesirable pathogens as the plant derived products are safe, easily degradable, and can be used without any adverse effects [4, 5].

In this study, antibacterial properties of methanol and aqueous extracts of seven plant species viz. *S. zeylanica*, *C. hirsuta*, *O. javanica*, *T. angustifolium*, *M. perpusilla*, *S. media* and *C. sinensis* were investigated against two strains of gram-positive bacteria (*Staphylococcus aureus* and *Bacillus cereus*) and two strains of gram-negative bacteria (*Escherichia coli* and *Proteus vulgaris*) and the results are reported.

V.1 Materials and Methods

V.1.1 Materials

Mueller-Hinton broth and Mueller-Hinton agar, resazurin were obtained from Hi-Media Laboratories, Mumbai, India. Bacterial strains were obtained from the Institute of Microbial Technology (IMTECH), Chandigarh, India. Filter paper diffusion disc was obtained from Hi-Media, Mumbai, India. Amoxicillin was obtained from Sigma Aldrich, Bangalore, India. Dimethyl sulfoxide (DMSO) and methanol was obtained from Merck, Mumbai, India.

V.1.2 Bacterial strains

The antibacterial property of plant extract was determined by using four bacterial strains causing food poisoning diseases. Two strains of gram-positive bacteria viz.

Staphylococcus aureus (MTCC-7443, MTCC = microbial type culture collection) and *Bacillus cereus* (MTCC-430) and two strains of gram-negative bacteria viz. *Escherichia coli* (MTCC-40) and *Proteus vulgaris* (MTCC-7299) were taken for this study.

V.1.3 Preparation of samples

The powdered samples of plants were prepared as per the procedure mentioned in the **Section II.2.3 (Page No. 73)**. The methanol and aqueous extracts of powdered samples were prepared as per the procedure described in the **Section IV.1.2 (Page No. 98)**.

V.1.4 Inoculums preparation

Each bacterial strain obtained was revived by culturing at 35°C in Mueller-Hinton broth overnight and again sub-cultured in the fresh Mueller-Hinton broth by incubating at the same temperature for overnight. The turbidity formed was adjusted to 0.5 optical densities at 570 nm by sterile saline water using UV-VIS spectrophotometer (Lambda 35, Perkin Elmer, USA) to attain viable cell count of 10^7 Colony-forming unit (CFU)/mL.

V.1.5 Determination of antibacterial activity of plant extracts

The antimicrobial activities of aqueous and methanol extracts of seven plant species were evaluated by disc diffusion method [6]. The aqueous extract (200 mg) and methanol extract (200 mg) was dissolved in 4 mL of distilled water and 4 mL of 10% (v/v) of DMSO, respectively and sterilized through membrane filter (0.22 µm). The solutions were then diluted to different concentrations (10, 20, 30 mg/mL). Thereafter, 5 µL of each concentration of each sample to be tested was loaded over 6 mm diameter of sterile filter paper disc and allowed to dry for a min. On the other hand, 10–15 mL of Mueller-Hinton agar medium was poured into sterilized Petri-dishes and allowed to solidify for 15 min followed by addition of 100 µL of bacterial culture on top of the plate, spread with sterile L-loop and allowed to dry for few min. The sterile filter paper disc loaded with different concentrations of samples was placed on the top of Mueller-Hinton agar plates and allowed to incubate for 24 h. *E. coli* and *S. aureus* were incubated at 37°C and *P. vulgaris* and *B. cereus* were incubated at 27°C. Filter paper disc loaded with 30 µg/mL of amoxicillin was taken as positive control. Filter paper

disc soaked in sterile distilled water and DMSO was taken as negative control. The inhibition zones were measured by Vernier calliper and the diameter of inhibitions were expressed in millimetre (mm).

V.1.6 Determination of minimum inhibitory concentration (MIC)

MIC is defined as the lowest concentration of the plant extract that inhibits the microbial growth after 24 h of incubation. MIC was determined in accordance with the protocol of Kitzberger *et al.* [7] using 96-well microtiter-plates methods by serial dilution of a factor 2. In brief, 200 μL of each sample (30 mg/mL) were placed in column no. 3 and then 100 μL was transferred to the column no. 4 and the volume was adjusted to 200 μL by adding DMSO (10%) and again 100 μL was transferred to the next column followed by adding DMSO and so on upto column no. 12. Similarly, for aqueous extract, sterile distilled water was taken for adjusting the volume. Finally 100 μL from column no. 12 was discarded. The column 1 was taken as positive control by adding an antibiotic amoxicillin and in column 2, only the culture medium was added which was taken as negative control. After that, 20 μL of microbial suspension was added in each of the well in the plate along with 10 μL of resazurin (0.015%) to observe cell's viability which is initially blue and turns pink when cell grows. The plates were incubated to grow the cells at their respective temperatures (37°C and 27°C) for 24 h. After incubation, the plates were observed by naked eye to observe the colour change from blue to pink and it was considered for MIC which is the last well in the plate before the colour changes to pink.

V.1.7 Determination of minimum bactericidal concentration (MBC)

Determination of MBC was performed following the method given by [7]. MBC is defined as the minimum amount of plant extract to completely kill the respective microorganism after 24 h of incubation on the freshly inoculated agar plates. Determination of MBC was executed by streaking method which was taken from the two lowest blue colour solution of MIC plates and it was again re-cultured on the sterile trypticase soy agar plates by incubating at 35°C for 24 h and observed for bacterial growth in respective concentration.

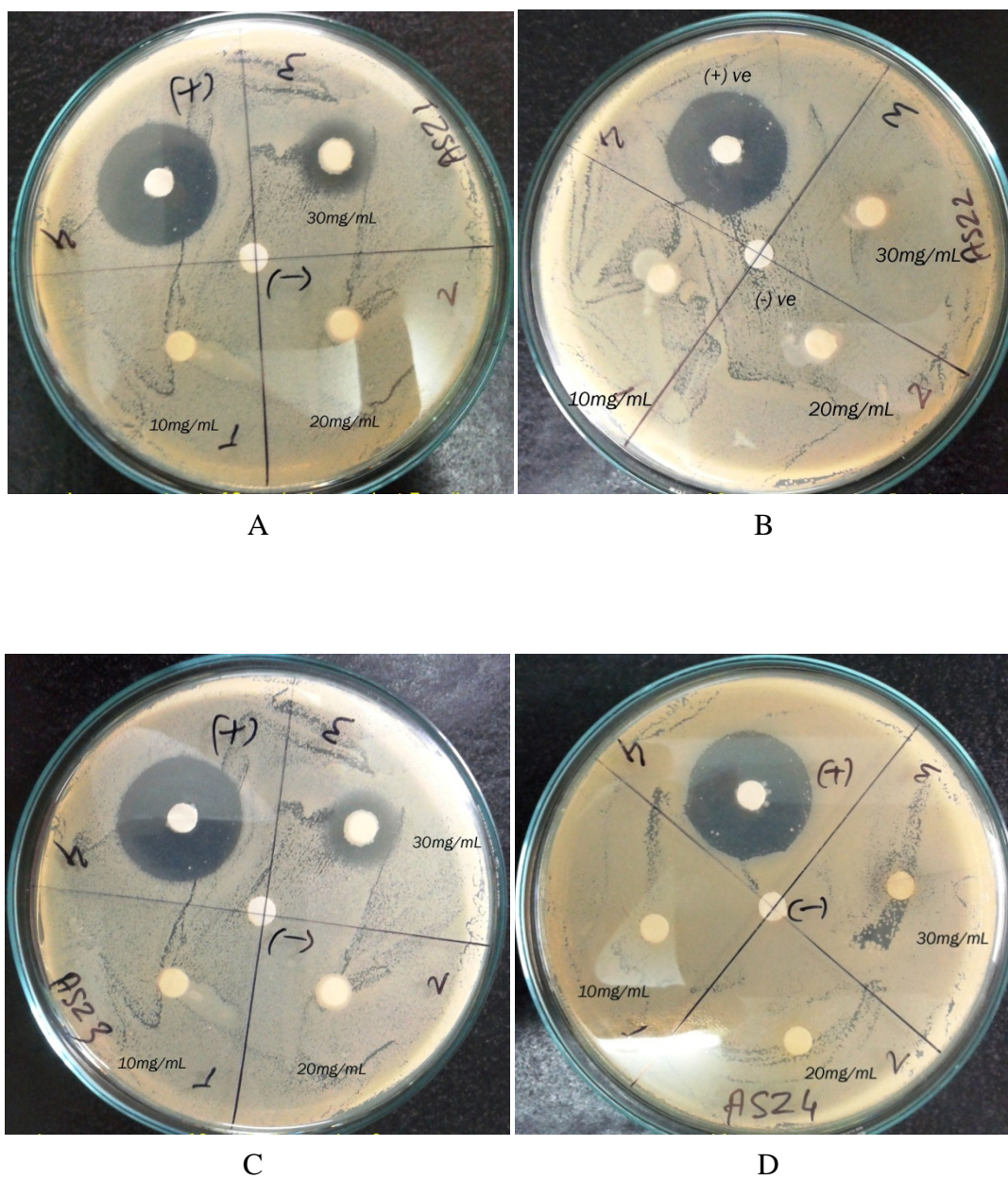


Fig.V.1a: Antibacterial activity of aqueous extract of *S. zeylanica* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

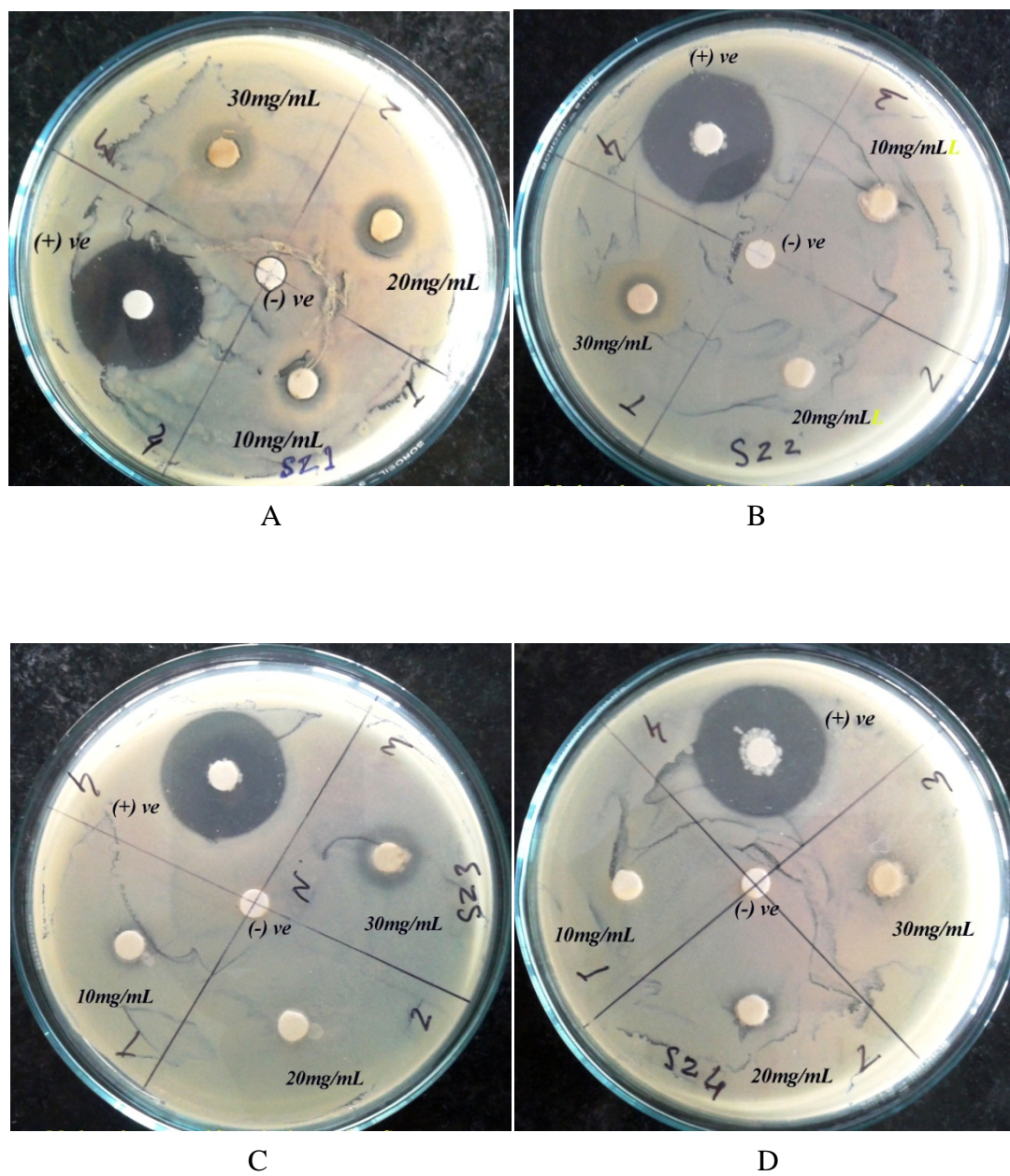


Fig.V.1b: Antibacterial activity of methanol extract of *S. zeylanica* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

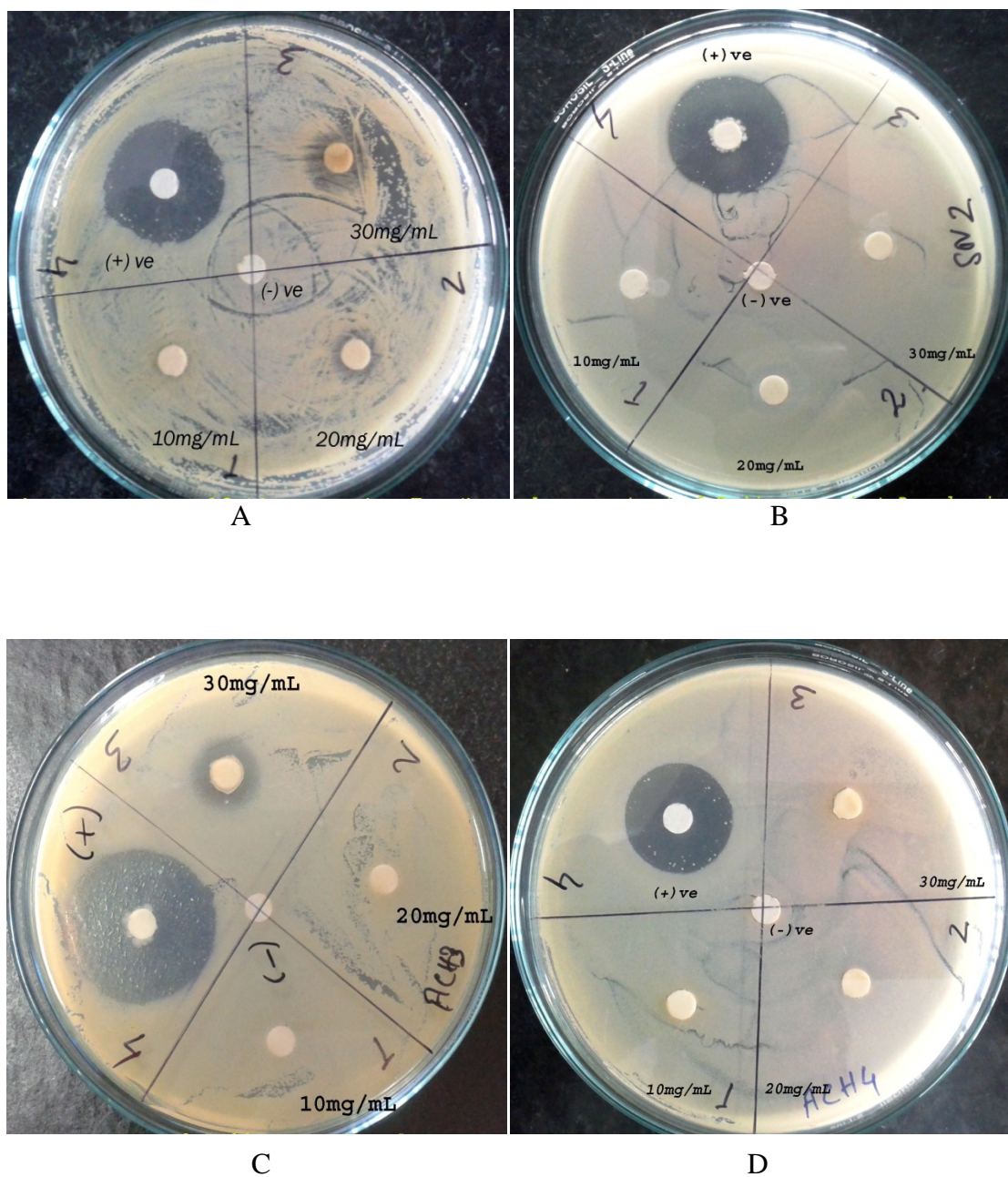


Fig.V.2a: Antibacterial activity of aqueous extract of *C. hirsuta* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

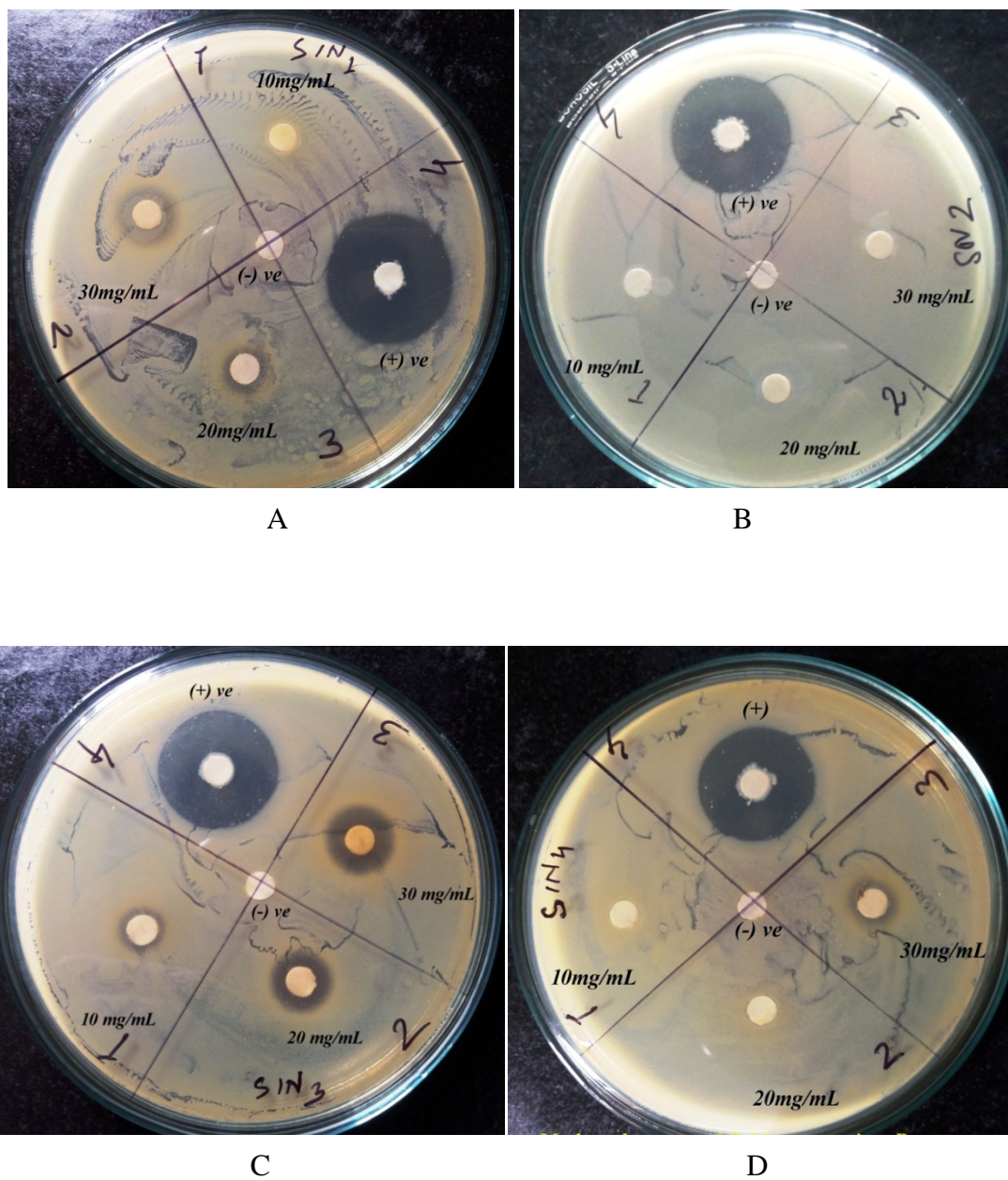


Fig.V.2b: Antibacterial activity of methanol extract of *C. hirsuta* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

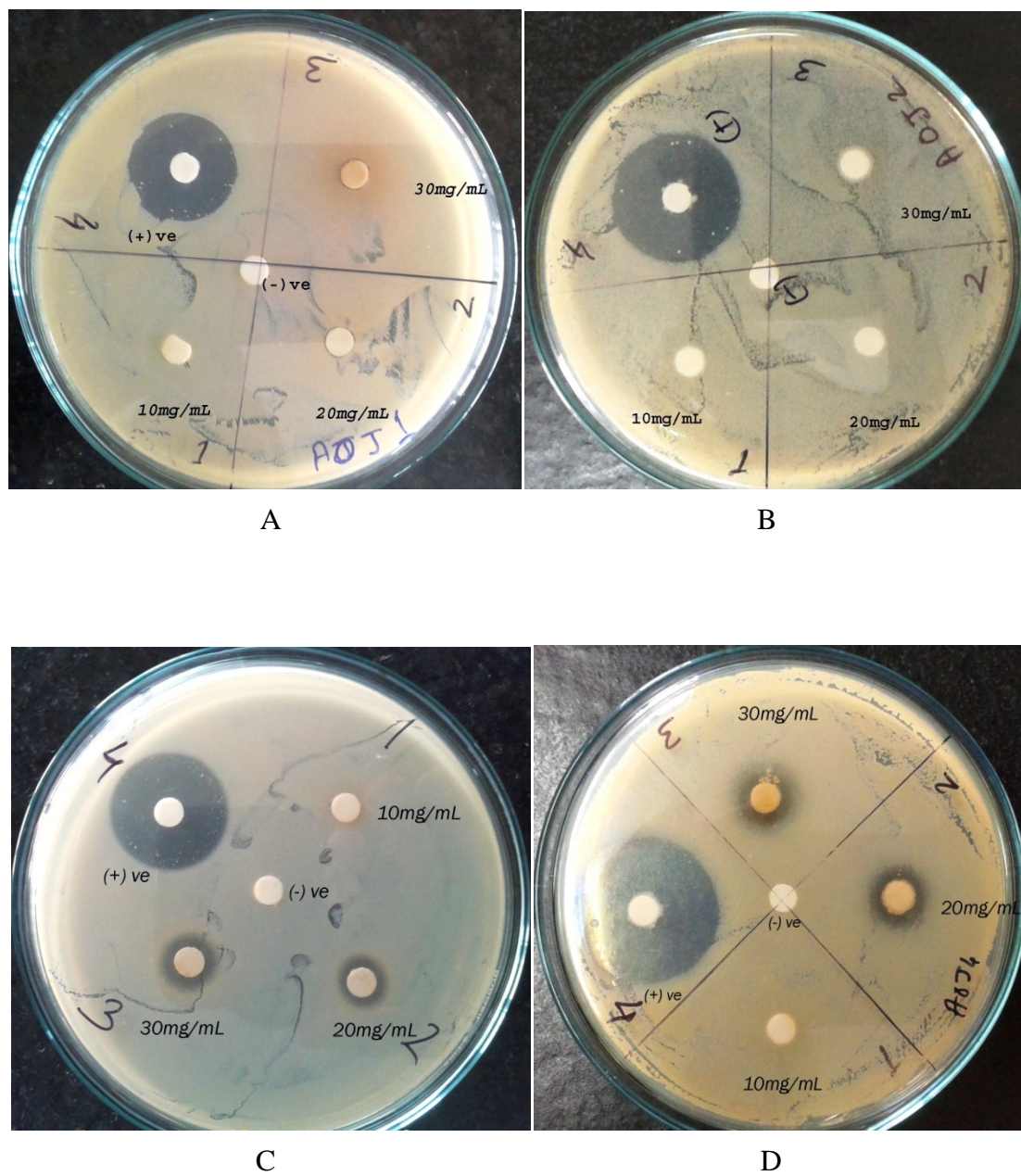


Fig.V.3a: Antibacterial activity of aqueous extract of *O. javanica* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

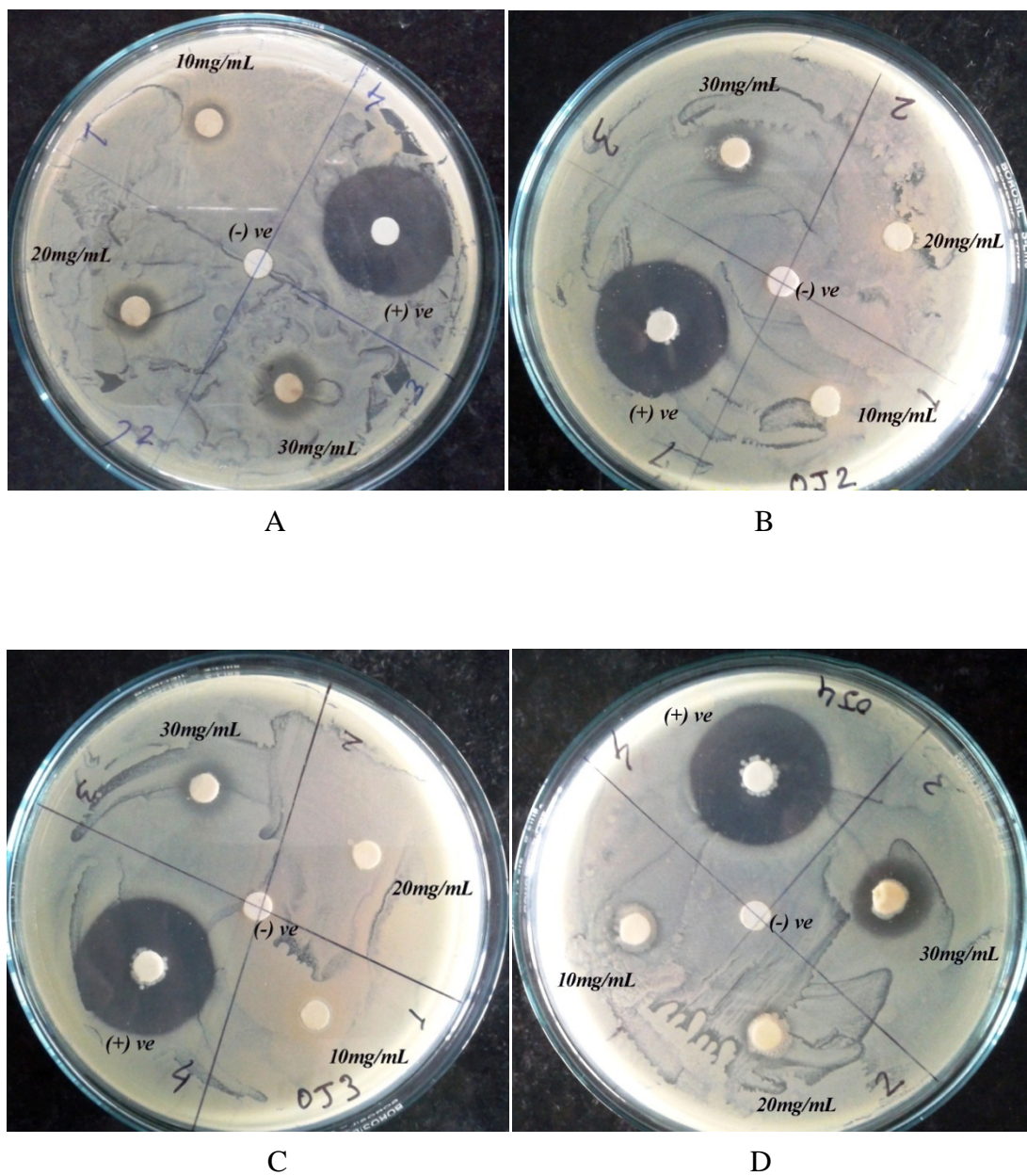


Fig.V.3b: Antibacterial activity of methanol extract of *O. javanica* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

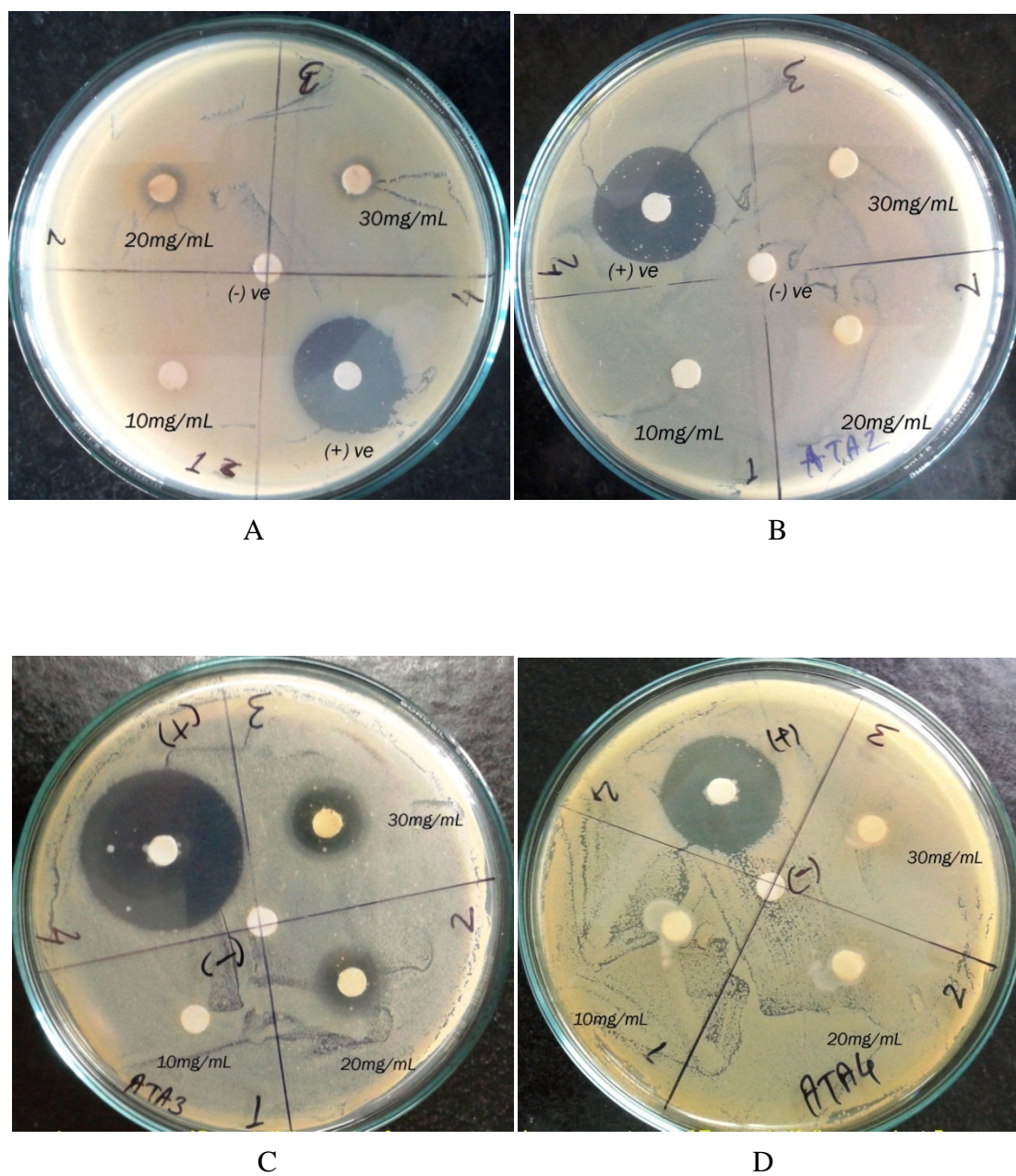


Fig.V.4a: Antibacterial activity of aqueous extract of *T. angustifolium* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

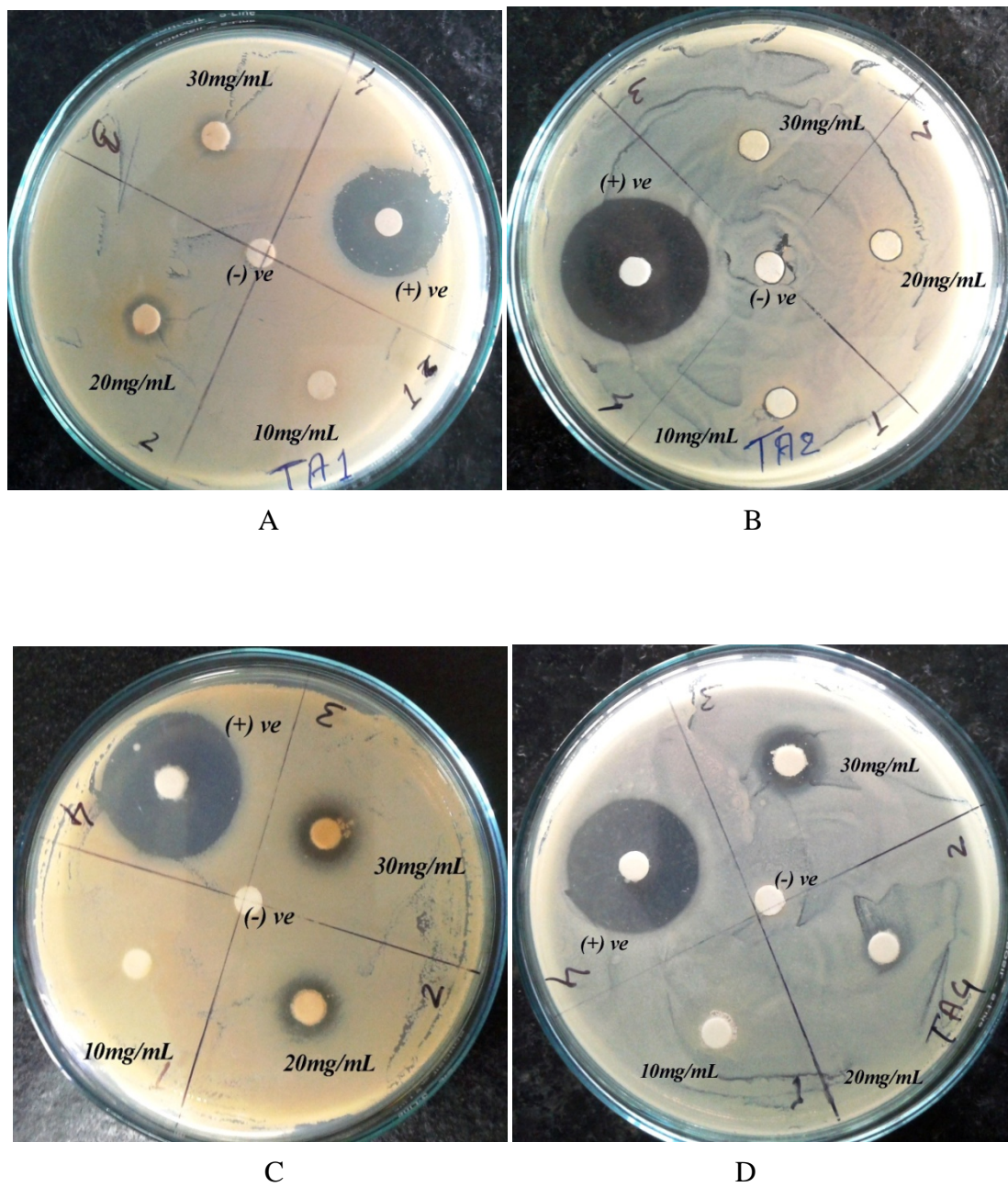


Fig.V.4b: Antibacterial activity of methanol extract of *T. angustifolium* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

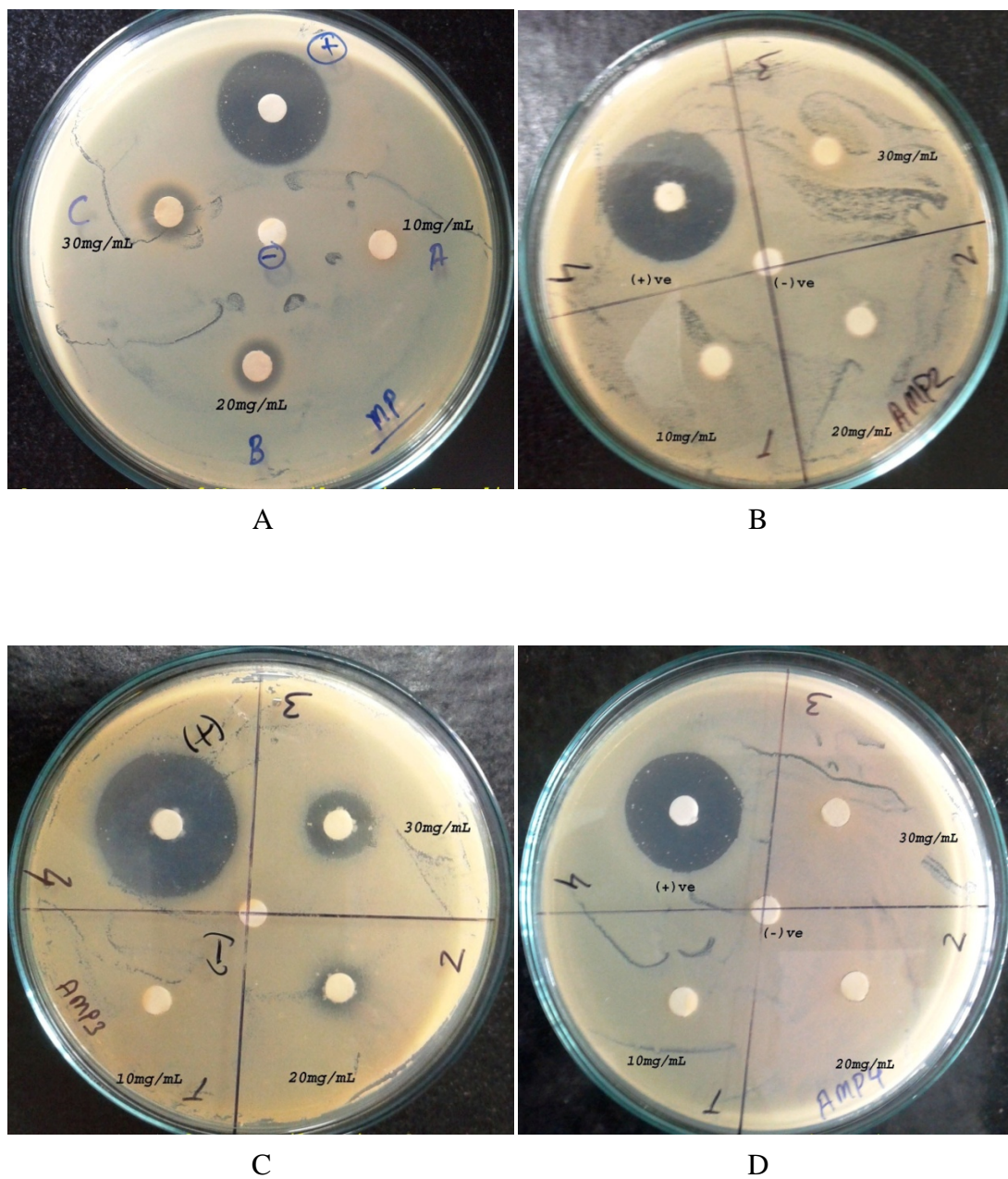


Fig.V.5a: Antibacterial activity of aqueous extract of *M. perpusilla* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

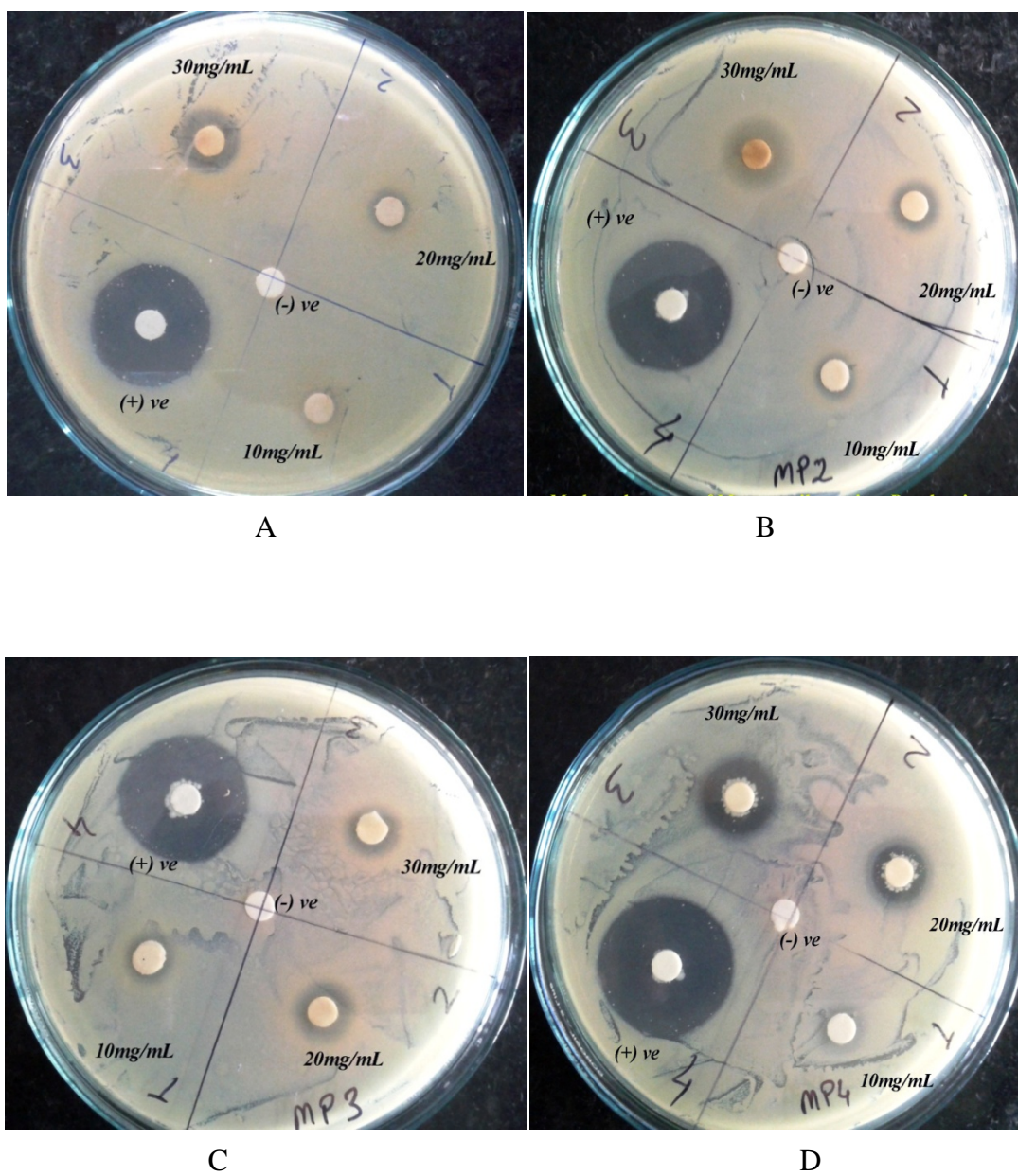


Fig.V.5b: Antibacterial activity of methanol extract of *M. perpusilla* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

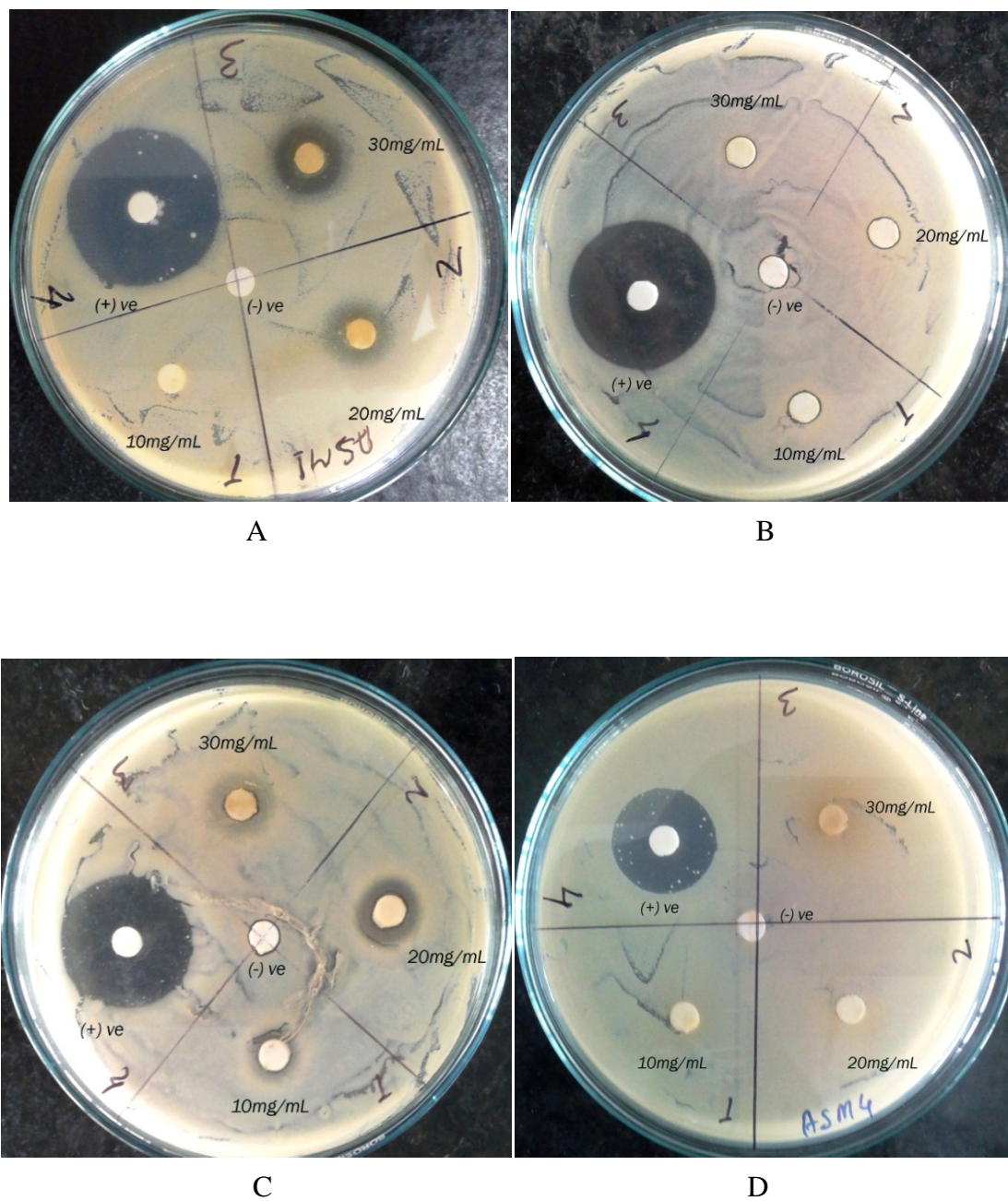


Fig.V.6a: Antibacterial activity of aqueous extract of *S. media* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

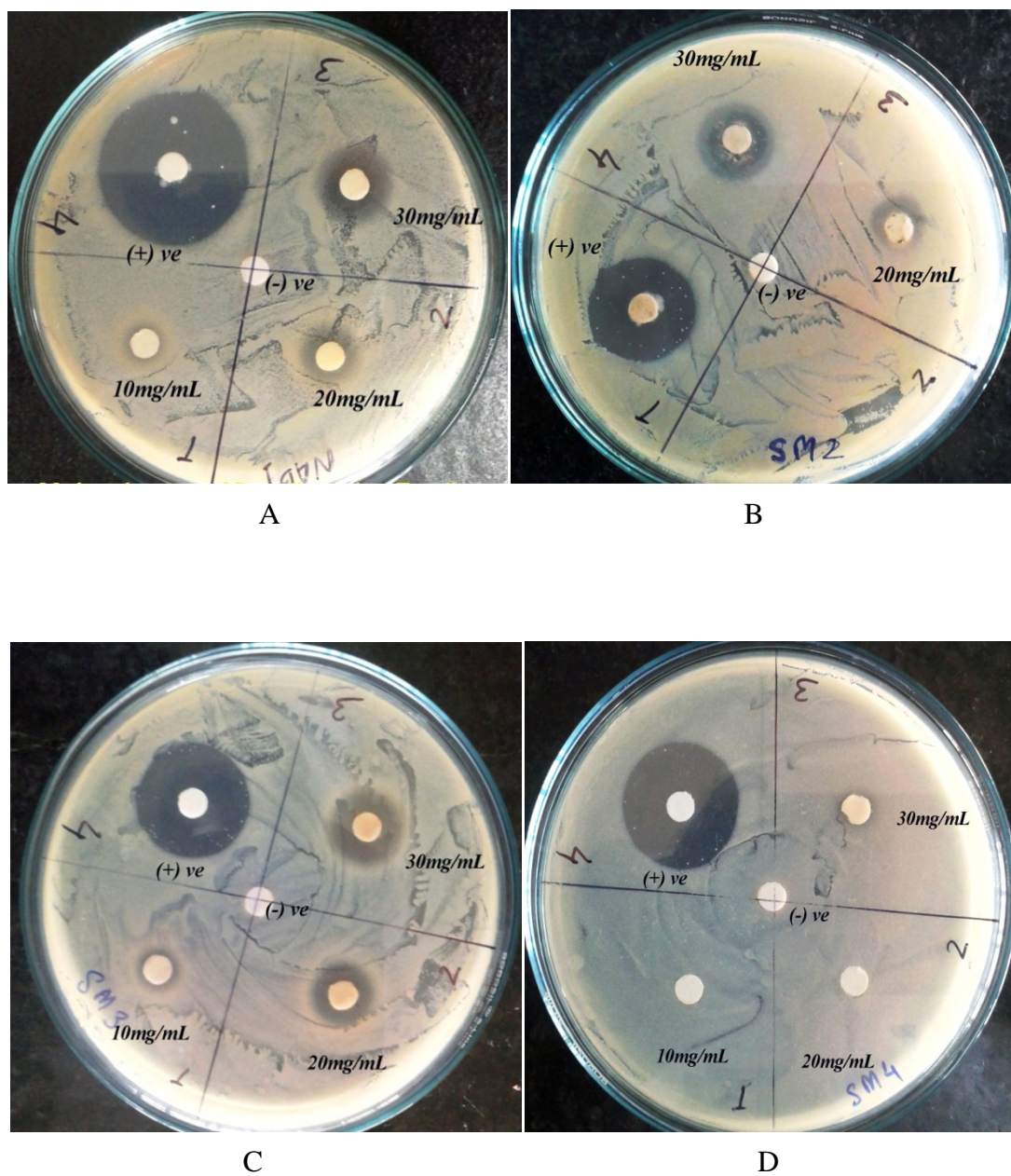


Fig.V.6b: Antibacterial activity of methanol extract of *S. media* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

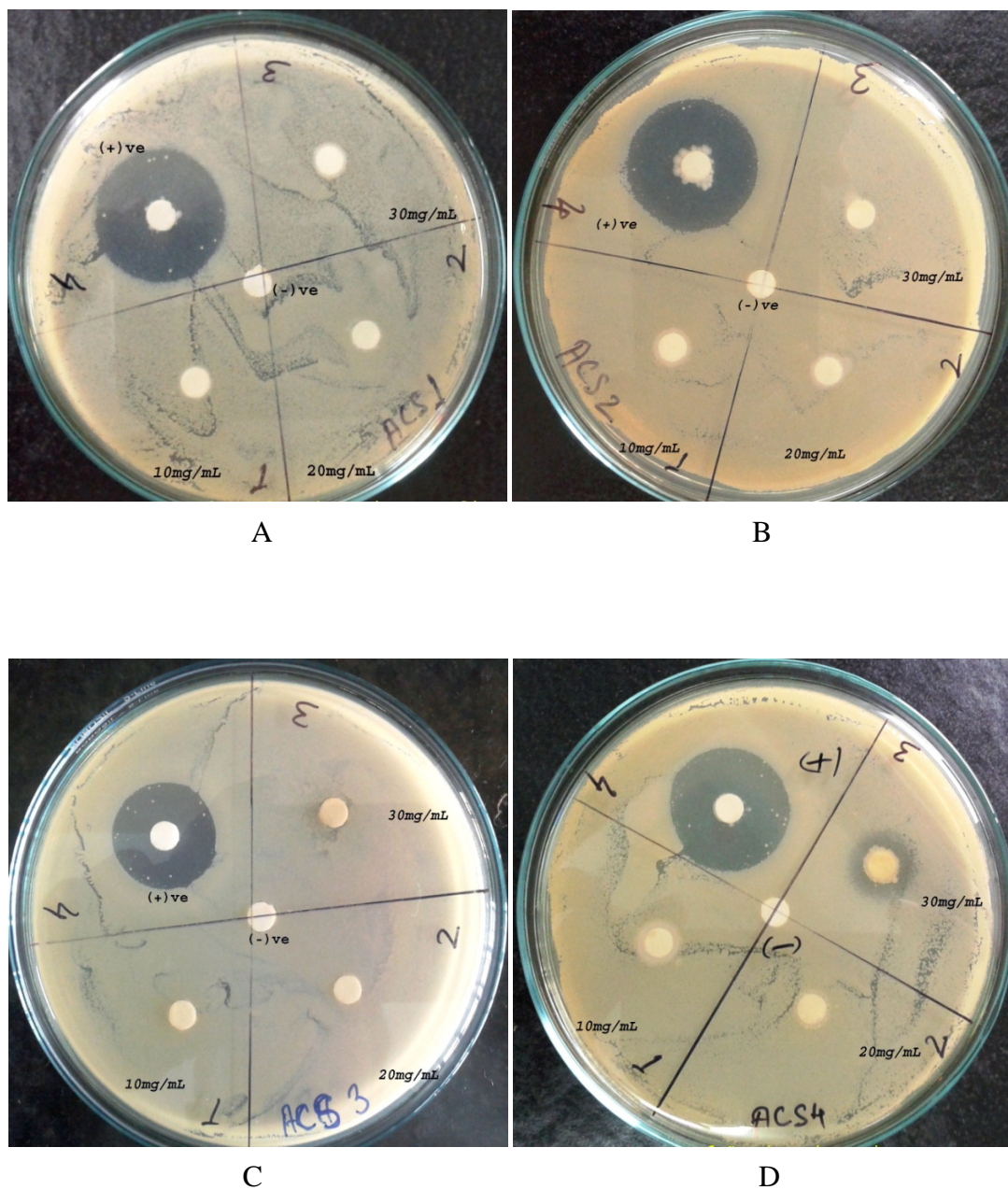


Fig.V.7a: Antibacterial activity of aqueous extract of *C. sinensis* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

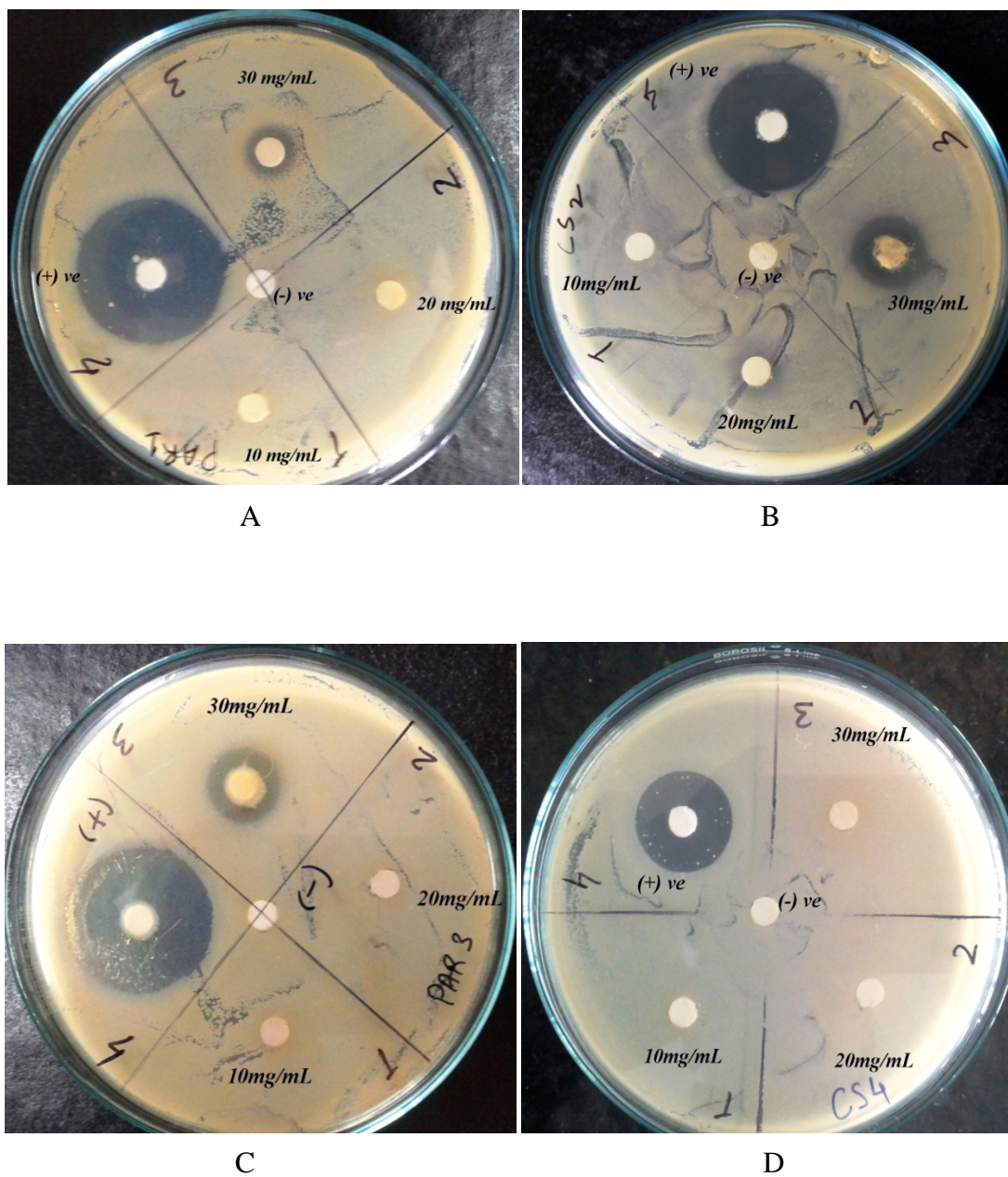


Fig.V.7b: Antibacterial activity of methanol extract of *C. sinensis* against A = *E. coli*; B = *P. vulgaris*; C = *S. aureus*; D = *B. cereus*; (+) = Positive control; (-) = Negative control.

V.2 Results and Discussion

In this study, the methanol and aqueous extracts of seven plant species viz. *S. zeylanica*, *C. hirsuta*, *O. javanica*, *T. angustifolium*, *M. perpusilla*, *S. media* and *C. sinensis* were investigated for antibacterial properties against two strains of gram-positive bacteria (*S. aureus* and *B. cereus*) and two strains of gram-negative bacteria (*E. coli* and *P. vulgaris*). The antibacterial property studies of the aqueous and methanol extracts against four microorganisms are shown in **Fig.V.1** to **Fig.V.7**. The antibacterial activities in terms of zone of inhibition along with MIC and MBC values of both the extracts of the plant species and the antibiotic against *E. coli*, *P. vulgaris*, *S. aureus* and *B. cereus* are summarized in **Table V.1**, **Table V.2**, **Table V.3** and **Table V.4**, respectively. It has been observed that the zone of inhibition was found increasing with increasing the concentration of sample extracts and consequently, the highest concentration (30 mg/mL) was taken for the study of the zone of inhibition.

It is seen from the **Table V.1** that the highest zone of inhibition (13 mm) was exhibited by the aqueous extracts of *S. zeylanica*, *C. hirsuta* and *S. media*, and the lowest zone of inhibition (9 mm) was exhibited by *T. angustifolium* at a concentration of 30 mg/mL against *E. coli*. On the other hand, the methanol extracts at the same concentration showed the highest zone of inhibition (16 mm) in *S. media* and the lowest zone (9 mm) was observed in *T. angustifolium* against *E. coli*. However, the aqueous extracts of *O. javanica* and *C. sinensis* were not showing any zone of inhibition in this concentration. Farjana *et al.* [8] studied antibacterial activities in aqueous and methanol extracts of some medicinal plants against *E. coli* and reported zone of inhibition (10 mm) which is comparable to the results of current study. The chloroform extract of a medicinal plant viz. *Artemisia dracuncululus* reported by Benli *et al.* [9] showed the zone of inhibition as 13 mm against *E. coli*. Similar result has also been reported in the methanol extract (9 to 15 mm) and water extract (8 to 10 mm) of a medicinal plant (*Ammi majus*) grown in Oman [10]. Al-Daihan *et al.* [11] also reported similar zone of inhibition (8 to 12 mm) in the methanol extracts of *Curcuma longa*, *Zingiber officinal*, *Pimpinella anisum* and *Commiphora molmol* against *Escherichia coli*. However, they also studied the aqueous extract of these plants and reported 7 to 11 mm inhibition zone against this bacterium which is also comparable to the present study.

Table V.1: Antibacterial activity of plant extracts and antibiotic against *E. coli*

Plants	Solvent extract	Zone of inhibition of sample extracts and antibiotic (mm)				MIC (mg/mL)	MBC (mg/mL)
		10 mg/mL	20 mg/mL	30 mg/mL	Amoxicillin 30 µg/mL		
Sz	Aq	0	0	13	24	<30	<30
	Me	9	13	13	24	7.5	<15
Ch	Aq	0	8	13	24	15	15
	Me	0	9	11	25	15	15
Oj	Aq	0	0	0	23	0	0
	Me	8	10	14	22	7.5	<15
Ta	Aq	0	8	9	24	15	<30
	Me	0	8	9	22	15	<30
Mp	Aq	0	9	11	23	15	15
	Me	0	8	14	28	15	15
Sm	Aq	0	10	13	28	15	<30
	Me	8	12	16	29	7.5	15
Cs	Aq	0	0	0	26	0	0
	Me	0	0	12	28	<30	<30

Aq = Aqueous, *Me* = Methanol, *Sz* = *S. zeylanica*, *Ch* = *C. hirsuta*, *Oj* = *O. javanica*, *Ta* = *T. angustifolium*, *Mp* = *M. perpusilla*, *Sm* = *S. media*, *Cs* = *C. sinensis*.

Table V.2: Antibacterial activity of plant extracts and antibiotic against *P. vulgaris*

Plants	Solvent extract	Zone of inhibition of sample extracts and antibiotic (mm)				MIC (mg/mL)	MBC (mg/mL)
		10 mg/mL	20 mg/mL	30 mg/mL	Amoxicillin 30 µg/mL		
Sz	Aq	0	0	0	21	0	0
	Me	0	0	9	25	<30	30
Ch	Aq	0	0	0	24	0	0
	Me	0	0	0	24	0	0
Oj	Aq	0	0	0	24	0	0
	Me	0	0	16	25	<30	<30
Ta	Aq	0	0	0	21	0	0
	Me	0	0	0	28	0	0
Mp	Aq	0	0	0	26	0	0
	Me	7	9	13	24	7.5	15
Sm	Aq	0	0	0	28	0	0
	Me	0	11	15	19	<15	15
Cs	Aq	0	0	0	24	0	0
	Me	0	9	13	25	15	<30

Aq = Aqueous, Me = Methanol, Sz = *S. zeylanica*, Ch = *C. hirsuta*, Oj = *O. javanica*, Ta = *T. angustifolium*, Mp = *M. perpusilla*, Sm = *S. media*, Cs = *C. sinensis*.

Table V.3: Antibacterial activity of plant extracts and antibiotic against *S. aureus*

Plants	Solvent extract	Zone of inhibition of sample extracts and				MIC (mg/mL)	MBC (mg/mL)
		antibiotic (mm)					
		10 mg/mL	20 mg/mL	30 mg/mL	Amoxicillin 30 µg/mL		
Sz	Aq	0	0	11	24	<30	<30
	Me	0	0	13	26	<30	<30
Ch	Aq	0	0	11	26	<30	<30
	Me	9	12	14	23	7.5	<15
Oj	Aq	0	9	11	22	15	15
	Me	0	0	10	26	<30	<30
Ta	Aq	0	10	13	30	15	15
	Me	0	11	12	24	<15	15
Mp	Aq	0	8	10	13	15	15
	Me	9	10	11	24	7.5	<15
Sm	Aq	8	10	11	26	7.5	15
	Me	8	12	14	21	7.5	<15
Cs	Aq	0	0	0	21	0	0
	Me	0	0	15	26	<30	<30

Aq = Aqueous, *Me* = Methanol, *Sz* = *S. zeylanica*, *Ch* = *C. hirsuta*, *Oj* = *O. javanica*, *Ta* = *T. angustifolium*, *Mp* = *M. perpusilla*, *Sm* = *S. media*, *Cs* = *C. sinensis*.

Table V.4: Antibacterial activity of plant extracts and antibiotic against *B. cereus*

Plants	Solvent extract	Zone of inhibition of sample extracts and antibiotic (mm)				MIC (mg/mL)	MBC (mg/mL)
		10	20	30	Amoxicillin		
		mg/mL	mg/mL	mg/mL	30 µg/mL		
Sz	Aq	0	0	0	24	0	0
	Me	0	8	9	25	15	<30
Ch	Aq	0	0	0	20	0	0
	Me	0	0	9	22	<30	<30
Oj	Aq	0	10	11	26	15	15
	Me	0	9	14	26	15	15
Ta	Aq	0	0	0	24	0	0
	Me	0	8	13	26	15	<30
Mp	Aq	0	0	0	23	0	0
	Me	9	12	15	26	7.5	<15
Sm	Aq	0	0	0	19	0	0
	Me	0	0	8	22	<30	<30
Cs	Aq	0	0	9	24	<30	<30
	Me	0	0	0	19	0	0

Aq = Aqueous, Me = Methanol, Sz = *S. zeylanica*, Ch = *C. hirsuta*, Oj = *O. javanica*, Ta = *T. angustifolium*, Mp = *M. perpusilla*, Sm = *S. media*, Cs = *C. sinensis*.

From the **Table V.2**, it was observed that the aqueous extracts of the plants showed no zone of inhibition against *P. vulgaris* even in 30 mg/mL concentration. Similarly, the methanol extracts of *C. hirsuta* and *T. angustifolium* were not showing any zone of inhibition against *P. vulgaris*. However, methanol extracts of *S. zeylanica*, *O. javanica*, *M. perpusilla*, *S. media* and *C. sinensis* exhibited zone of inhibition against *P. vulgaris* at 30 mg/mL that ranged from 9 mm (*S. zeylanica*) to 16 mm (*O. javanica*). In a study reported by Nair and Chanda [12], methanol and acetone extracts of *Psidium guajava* showed a zone of inhibition of 13.5 mm against *P. vulgaris*.

It is observed in **Table V.3** that the growth inhibition zones in aqueous extracts of plants against *S. aureus* ranged from 10 mm in *M. perpusilla* to 13 mm in *T. angustifolium* at 30 mg/mL concentration. However, in methanol extracts of plants

studied against *S. aureus*, the zone of inhibition varied from 10 mm in *O. javanica* to 15 mm in *C. sinensis*. In this study, all the sample extracts exhibited the capability to inhibit *S. aureus* except the aqueous extract of *C. sinensis* which did not show any activity even at 30 mg/mL concentration. A study of some medicinal plants reported by Al-Daihan *et al.* [11] indicated the similar zone of inhibition against *P. vulgaris*. Similarly, the chloroform extract of *Barringtonia acutangula* against *Pseudomonas aeruginosa* showed 13 mm zone of inhibition by the disc diffusion assay [13] which was also similar to the methanol extracts of *S. zeylanica*, *C. hirsuta*, *T. angustifolium* and *S. media* of the present study against *S. aureus*. In the antibacterial study of aqueous extracts of plants against *B. cereus* (**Table V.4**), only *C. sinensis* and *O. javanica* showed a zone of inhibition of 9 mm and 11 mm, respectively at the concentration of 30 mg/mL. The aqueous extracts of five other samples were not showing any zone of inhibition even at 30 mg/mL concentration. On the other hand, the highest zone of inhibition exhibited by the methanol extracts was found 15 mm in *M. perpusilla* and the lowest zone was shown by *S. media* (8 mm) at the same concentration. However, methanol extract of *C. sinensis* did not show any zone of inhibition against this bacterium. These results are also in agreement with the findings in the methanolic extracts of *Agathosma betulina* and *Lippia javanica* reported by Huffman *et al.* [14] and Moolla *et al.* [15]. The antibacterial study of *S. zeylanica* against food borne diseases such as *S. aureus*, *E. coli*, *P. vulgaris* and *Aspergillus niger* reported by Gowri *et al.* [16] showed smaller zone of inhibition (4.6 to 6 mm) at 50 mg/mL concentration in comparison to the present study.

The antibacterial potential of the plant extract is determined on the basis of minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC). MIC value of sample extract indicates the capacity to inhibit the growth of bacteria and MBC value of sample indicates the bactericidal capacity to kill or degrade the bacterial growth [7]. In this study, the MIC values of aqueous extracts of selected plants ranged from 15 to <30 mg/mL and that of methanol extracts varied from 7.5 to <30 mg/mL and the MBC values of aqueous extracts varied from 15 to <30 mg/mL and that of methanol extracts ranged from <15 to <30 mg/mL against *E. coli* (**Table V.1**). The result of present study showed less efficient in comparison to the methanol extract of clove against *E. coli*, *S. aureus*, and *P. aeruginosa* with MIC values ranging from 0.1 to 2.31 mg/mL [17]. Similarly, Dua *et al.* [18] showed MIC values ranging from 6.25 to 25 mg/mL against *E. coli*, *P. aeruginosa*, *S. aureus* and *B. pumilus* in methanol extract of spice cumin which is comparable to the results of current study. The MIC

value against *P. vulgaris* ranged from 7.5 to <30 mg/mL and MBC value ranged from 15 to 30 mg/mL in methanol extracts of the plants which are showing zone of inhibition (**Table V.2**).

Similarly, the MIC values were found in the range of 7.5 to <30 mg/mL in both methanol and aqueous extracts of selected plants against *S. aureus*. However, MBC values of this bacterium ranged from 15 to <30 mg/mL in aqueous extracts and from <15 to <30 mg/mL in methanol extracts (**Table V.3**). However, Gupta *et al.* [19] reported higher MIC values ranging from 25 to 125 mg/mL in the ethanol extracts of some medicinal plants studied against *S. aureus*, *P. aeruginosa* and *B. Subtilis*. The MIC values of methanol extracts against *B. cereus* ranged from 7.5 to <30 mg/mL and it ranged from 15 to <30 mg/mL in aqueous extract. While the MBC values of plants studied against this bacterium was found in the range of 15 to <30 mg/mL in aqueous extracts and <15 to <30 in methanol extract (**Table V.4**). The results of current study are also comparable with the results reported of Akinpelu *et al.* [20] in which they studied antimicrobial activity of aqueous and butanolic extracts of *Persea americana* against food poisoning bacteria *Bacillus cereus* and their study exhibited MIC values of 25 and 10 mg/mL, respectively and MBC values of 3.12 and 12.5 mg/mL, respectively.

The results of this study (**Table V.1 – Table V.4**) revealed that almost all the plant species are exhibiting antibacterial activities against the studied microorganisms. The methanol extracts of all the selected plants are showing more effective anti-microbial activities in comparison to the aqueous extracts. However, the plants showed comparatively smaller zone of inhibitions in comparison to the standard antibiotics. Among gram-positive bacteria, *S. aureus* was inhibited by all the plant extracts except aqueous extract of *C. sinensis* (**Table V.3**). On the contrary, *B. cereus* was inhibited only by some of the methanol and aqueous extracts (**Table V.4**). Thus, *S. aureus* was found to be the most susceptible and *B. cereus* was moderately susceptible bacteria. On the other hand, among the gram-negative bacteria, *E. coli* was inhibited by both the extracts except aqueous extract of *O. javanica* and *C. sinensis* which are showing resistant even at 30 mg/mL concentration. However, none of the aqueous extract could inhibit *P. vulgaris* even at 30 mg/mL concentration and thus *P. vulgaris* is showing resistant against the selected plants. Parekh *et al.* [21] reported in their study that the gram-negative bacteria showed more resistant in comparison to the gram-positive bacteria and these results are in accordance with the findings of the current study.

In the present study, the MIC values of some plant species reached up to 30 mg/mL which may be due to low concentrations of active compounds in the extracts. The

difference in MIC values of both aqueous and methanol extracts may be due to the variation in their bioactive compounds and solubility nature of the compounds. In current study, the methanol extracts of the selected plant species showed more antimicrobial activities which may be due to the presence of bioactive compounds which are readily soluble in methanol. It has been reported that methanol or ethanol extracts were generally more effective against microorganisms than the aqueous extracts because the bioactive compounds present in the plant species are more soluble in polar organic solvents such as ethanol and methanol than in water [22]. It has been reported by many researchers that antimicrobial properties of the plant extracts are due to the presence of numerous bioactive compounds such as phenolic compounds, alkaloids and terpenoids which interact with proteins and enzymes causing cell death by obstructing enzymes necessary for biosynthesis of amino acids in microbial cell [23, 24].

V.3 Conclusion

This study reveals that all the selected plant species showed different antimicrobial activities with the highest zone of inhibition (16 mm) in the methanol extracts of *S. media* against *E. coli* with MIC value of 7.5 mg/mL and MBC value of 15 mg/mL and in the methanol extracts of *O. javanica* against *P. vulgaris* with MIC and MBC values of <30 mg/mL. However, the methanol extract of *C. sinensis* showed the highest zone (15 mm) against *S. aureus* with MIC and MBC values of <30 mg/mL. Also the methanol extract of *M. perpusilla* showed the highest zone (15 mm) against *B. cereus* with MIC value of 7.5 mg/mL and MBC value of <15 mg/mL. In aqueous extract, the highest zone of inhibition (13 mm) was observed in *S. zeylanica*, *C. hirsuta*, and *S. media* at 30 mg/mL concentration against *E. coli*. The aqueous extract of *T. angustifolium* showed the highest zone of inhibition (13 mm) against *S. aureus*. However, no zone of inhibition was observed in the aqueous extracts of plants against *P. vulgaris*. Thus, the methanol extracts of all the selected plants are showing more effective antimicrobial activities in comparison to the aqueous extracts and this may be due to the microbial active compounds which are more soluble in methanol than water. Finally, consumption of these wild plants could be suggested for prevention of several diseases.

References

- [1] Silver L, Bostian K. Screening of natural products for antimicrobial agents. *Eur. J. Clin. Microbiol. Infect. Dis.* 1990; 9: 455–461.
- [2] Latha LS, Reddy PN. Antimicrobial, antidiarrhoeal and analysis of phytochemical constituents of *Sphaeranthus amaranthoides*. *Indian J. Sci. Technol.* 2009; 2(3): 45–48.
- [3] Khanahmadi M, Rezazadeh S, Taran M. *In vitro* antimicrobial and antioxidant properties of *Smyrniium cordifolium* Boiss. (Umbelliferae) extract. *Asian J. Plant Sci.* 2010; 9: 99–103.
- [4] Wannissorn B, Jarikasem S, Siriwangchai T, Thubthimthed S. Antibacterial properties of essential oils from Thai medicinal plants. *Fitoterapia* 2005; 76: 233–236.
- [5] Chika CO, Jude NO, Ifeanyi CO, Anyanwu NB. Antibacterial activities and toxicological potentials of crude ethanolic extracts of *Euphorbia hirta*. *J. Am. Sci.* 2007; 3(3): 11–16.
- [6] Jorgensen JH, Turnidge JD, Washington JA. Antibacterial susceptibility tests: Dilution and disc diffusion methods. *Manual of clinical Microbiology*. Washington DC: American Society for Microbiology; 1999. 7th edn. 1526.
- [7] Kitzberger CSG, Smamia J, Pedrosa RC, Ferreira SRS. Antioxidant and antimicrobial activities of shiitake (*Lentinula edodes*) extracts obtained by organic solvents and supercritical fluids. *J. Food Eng.* 2007; 80: 631–638.
- [8] Farjana A, Zerin N, Kabir MS. Antimicrobial activity of medicinal plant leaf extracts against pathogenic bacteria. *Asian Pac. J. Trop. Dis.* 2014; 4(2): 920–923.
- [9] Benli M, Kaya I, Yigit N. Screening antimicrobial activity of various extracts of *Artemisia dracuncululus* L. *Cell Biochem. Funct.* 2007; 25: 681–686.
- [10] Al-Hadhrami RMS, Hossain MA. Evaluation of antioxidant, antimicrobial and cytotoxic activities of seed crude extracts of *Ammi majus* grown in Oman. *Egyptian journal of basic and applied sciences* 2016; 3: 329–334.
- [11] Al-Daihan S, Al-Faham M, Al-Shawi N, Almayman R, Brnawi A, Zargar S, Bhat R. Antibacterial activity and phytochemical screening of some medicinal plants commonly used in Saudi Arabia against selected pathogenic microorganisms. *Journal of King Saud University – Science* 2013; 25: 115–120.
- [12] Nair R, Chanda S. In-vitro antimicrobial activity of *Psidium guajava* L. leaf extracts against clinically important pathogenic microbial strains. *Brazilian J. Microbiol.* 2007; 38: 452–458.
- [13] Sahoo S, Panda PK, Mishra SR, Parida RK, Ellaiah P, Dash SK. Antibacterial activity of *Barringtonia acutangula* against selected urinary tract pathogens. *Indian J. Pharm. Sci.* 2008; 70(5): 677–679.

- [14] Huffman A, Klepser ME, Ernst EJ, Keele D, Roling E, Viljoen AM. Susceptibility of opportunistic bacteria and yeasts to the volatile oils of *Artemisia afra*, *Lippia javanica*, *Lippia scaberrima*, *Myrothamnus flabellifolius* and *Osmitopsis asteriscoides*. *Journal of Infectious Disease Pharmacotherapy* 2002; 5:43–50.
- [15] Moolla A, Viljoen AM. ‘Buchu’ — *Agathosma betulina* and *Agathosma crenulata* (Rutaceae): A review. *J. Ethnopharmacol.* 2008; 119: 413–419.
- [16] Gowri J, Sahayaraj PA, Amaladasan M. Antimicrobial activity of the leaf, flower and stem extracts of *Sphenoclea zeylanica*. *Int. J. Appl. Sci. Biotechnol.* 2016; 4(3): 325–329.
- [17] Pandey A, Singh P. Antibacterial activity of *Syzygium aromaticum* (Clove) with metal ion effect against food borne pathogens. *Asian J. Plant Sci. Res.* 2011; 1(2): 69–80.
- [18] Dua A, Gaurav G, Balkar S, Mahajan R. Antimicrobial properties of methanolic extract of cumin (*Cuminum cyminum*) seeds. *Int. J. Res. Ayurveda Pharm.* 2013; 4(1): 104–107.
- [19] Gupta RN, Kartik V, Manoj P, Singh PS, Alka G. Antibacterial activities of ethanolic extracts of plants used in folk medicine. *Int. J. Res. Ayurveda Pharm.* 2010; 1(2): 529–535.
- [20] Akinpelu DA, Aiyegoro OA, Akinpelu OF, Okah AI. Stem bark extract and fraction of *Persea americana* (Mill) exhibits bactericidal activities against strains of *Bacillus cereus* associated with food poisoning. *Molecules* 2015; 20: 416–429.
- [21] Parekh J, Jadeja D, Chanda S. Efficacy of aqueous and methanol extracts of some medicinal plants for potential antibacterial activity. *Turkish J. Biol.* 2005; 29: 203–210.
- [22] Gonzalez-Guevara JL, Gonzalez-Lavaut JA, Pino-Rodriguez S, Garcia-Torres M, Carballo-Gonzalez MT, Echemendia-Arana OA, Molina-Torres J, Prieto-Gonzalez S. Phytochemical screening and in vitro antiherpetic activity of four *Erythroxylum* species. *Acta Farmaceut. Bonaer.* 2004; 23(4): 506–509.
- [23] Burt S. Essential oils: Their antibacterial properties and potential application in foods: A review. *Int. J. Food Microbiol.* 2004. 94: 223–253.
- [24] Gill AO, Holley RA. Disruption of *Escherichia coli*, *Listeria monocytogenes* and *Lactobacillus sakei* cellular membranes by plant oil aromatics. *Int. J. Food Microbiol* 2006. 108: 1–9.