

Antimycotic and antibacterial activities of stroma extracts of *Cordyceps militaris*

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Cordyceps militaris (L.) Fr., known as caterpillar fungus, is extremely renowned for its high medicinal value and diverse array of bioactivities, such as antimicrobial, anti-inflammatory, antioxidant, antitumor or immunomodulatory. The growing resistance of pathogenic micro-organisms to conventional antibiotics highlights the necessity for alternative antimicrobial agents. The objective of the research was to evaluate the antimicrobial efficacy of *in vitro* cultured stromata of *C. militaris*. Soxhlet extraction technique was utilized for the preparation of various extracts from stroma of *C. militaris* and screening against various fungal and bacterial pathogens. The antimicrobial efficacy of the extracts was evaluated by agar well diffusion and disc diffusion methods, and minimum inhibitory concentration (MIC) was also determined. The results indicated that all the extracts of *C. militaris* stroma showed significant inhibitory effects on tested pathogens. However, the methanolic extract exhibited the highest inhibition zones ranging from 9.86 to 15.42 mm for fungal pathogens and 10.31 to 16.27 mm for pathogenic bacteria and MIC values from 1.07 to 3.73 mm and 1.07 to 2.66 mm for fungi and bacteria, respectively. These findings indicate that *C. militaris* can be a potential alternative for natural antimicrobial agents and therefore, necessitate further investigation for its possible biocontrol activity and therapeutic applications.

Keywords : *Cordyceps militaris*, medicinal value, stroma, extracts, antimicrobial activity

INTRODUCTION

The discovery and development of antibiotics is perhaps the greatest scientific accomplishment of all in the last seventy years. Antibiotics have more than one mechanism of action, typically interfering with basic metabolic processes or structural components of microorganisms (Fuchs and Kuchenbecker 2004). However, the extensive and long-term use of antibiotics in medical practice has been responsible for the development of multidrug-resistant microbial strains. At the same time, growing public interest in natural and integrated therapy has fuelled demand for traditional and alternative medicine systems (Jin *et al.*, 2018).

Medicinal fungi are valuable sources of various biomolecules that are beneficial not only nutritionally (Kalaè, 2009), but also therapeutically (Borchers *et al.* 2004; Poucheret *et al.* 2006). The

genus *Cordyceps* has been valued traditionally for its medicinal and therapeutic properties. The bioactive compounds in *Cordyceps* species found to have potential health benefits are widely used in various medical applications (Paterson and Russel, 2008). Of all medicinally known species of *Cordyceps*, *C. militaris* (L.) Fr. is of greatest importance (Kobayasi, 1941). *Cordyceps militaris*, an entomopathogenic fungus (parasitic fungus growing on insects or insect larvae) has been employed extensively in East Asian countries as a folk tonic food and crude medication (Ying *et al.* 1987). It is reported to contain a number of bioactive substances, including adenosine, cordycepic acid, polysaccharide, cordycepin, ergosterol, etc. (Chen *et al.* 2012; Cohen *et al.* 2014; Chan *et al.* 2015). The main bioactive compounds are adenosine and cordycepin (Li *et al.* 2020) that have been reported to exhibit antibacterial (Eiamthawornet *et al.* 2022), antifungal (Reis *et al.* 2013), anti-inflammatory (Won and Park, 2005), antioxidant (Awang *et al.* 2021), anti-cancer (Rao

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et al. 2010) anti-diabetic and immunomodulatory (Lee *et al.* 2020) properties. As a potential reservoir of bioactive compounds, *C. militaris* has not only enhanced the current natural therapies but has also propelled the green pharmacy revolution in Korea, Japan, China and other Asian countries (Sung, 2007; Shrestha *et al.* 2012; Tuli *et al.* 2014). Therefore, in view of the high medicinal and therapeutic properties of *Cordyceps militaris*, the present investigation was focused on assessing the efficacy of various extracts of *C. militaris* against various pathogenic fungi and bacteria.

MATERIALS AND METHODS

Source of biological material

The fresh stromata of *C. militaris* were collected during the summer months (June to August) from Gool area (33°37'89" N and 75°10'34" E) of District Ramban, Jammu and Kashmir, India. The small bits (3-5 mm) of freshly collected *C. militaris* stroma were transferred aseptically to PDA (Potato dextrose agar) and incubated for 10 days at 25±2°C. The pure cultures obtained were maintained at 4°C. Subsequently, the mycelial discs of pure culture were transferred to nutrient broth and kept in shaking incubator (170 rpm) at 25±2°C for 8 days for the production of liquid inoculum. For cultivation, the substrate (brown rice along with nutritional broth) was inoculated with the liquid inoculum of *C. militaris* and further incubated at 20°C with relative humidity (RH) of 85-90%. After around 50 days of inoculation, the stromata were harvested, sun-dried and kept at 25°C for further study.

Preparation of extracts for antimicrobial activity

Extracts of shade dried stromata of *C. militaris* were prepared by taking powdered sample in a Soxhlet extractor (80°C) (De Castro and Garc ya-Ayuso, 1998). Different solvents used in ascending order of polarity, included ethanol, methanol and aqueous. Following that, a rotary evaporator was used to concentrate the extracts at low pressure. The concentrated extracts were then stored at 4°C for further research.

Test organisms

Four bacterial species and four fungal species (Table 1) were employed as test micro-organisms

to evaluate the antimicrobial activity of sample extracts (ethanol, methanol and aqueous) of *C. militaris*.

Preparation of culture media

To evaluate antibacterial and antifungal activities of sample extracts (ethanol, methanol and aqueous) of *C. militaris*, separate culture media and broths were used. For antibacterial activity, Mueller Hinton Agar (MHA) and Mueller Hinton broth were used. Likewise, PDA (Potato Dextrose agar) medium and PD (Potato Dextrose) broth were used for evaluation of antifungal activity.

Assay of antifungal activity

The antifungal activity of sample extracts (ethanol, methanol and aqueous) was determined by agar well diffusion method described by Wiegand *et al.* (2008). In this method, spore suspensions of one-week old PDA grown fungal cultures of *Aspergillus niger*, *Fusarium oxysporum*, *Penicillium chrysogenum* and *Mucor mucedo* were used to evaluate antifungal activity. In culture tubes, an inoculum comprising 20 µL of each fungal organism was mixed with 25 µL of PDA medium. Then the mixture was stirred and poured into 90 mm Petri plates. The medium was allowed to solidify and then a sterilized cork borer was used (5 mm dia) to make wells. The wells were filled with varying concentrations of sample extracts of *C. militaris* viz., 15mg/mL (C₁), 30mg/mL (C₂) and 60 mg/mL (C₃). 5% DMSO was used as negative control and Nystatin disc (50 mcg/disc) was used as positive control. The plates were labelled, sealed and incubated for 48 hrs at 25±2°C.

Determination of antibacterial activity

The antibacterial activity of sample extracts (ethanol, methanol and aqueous) of *C. militaris* was determined by disc diffusion method described by Valgas *et al.* (2007). The bacterial species were kept in 0.85% saline solution to maintain their viability. MHA medium was prepared, sterilised, poured in pre-sterilised Petri-plates and left to solidify. Using a sterilized cotton swab, the freshly prepared bacterial suspension was inoculated onto Petri-plates containing

solidified growth medium. Subsequently, sterile filter paper discs of 6 mm in diameter (Himedia) of different concentrations of sample extracts of *C. militaris*, viz., C₁ (15 mg/mL), C₂ (30 mg/mL) and C₃ (60 mg/mL) were placed on the inoculated agar plates. 5% DMSO and Gentamycin (50 mcg/disc) discs were used as negative and positive control, respectively. The plates were labelled, sealed and incubated for 24 hrs at 37±2°C.

Determination of MIC (minimum inhibitory concentration) value

MIC is the lowest concentration of test sample at which the growth of fungal or bacterial organism is completely inhibited. The sample extracts of *C. militaris*, which showed significant antimicrobial activity were assessed for their respective MIC values. In accordance with Wiegand *et al.* (2008), the microdilution method was used on 96-well biolog plates to determine MIC values. 50 µL of MH broth for test bacteria and PD broth for test fungus were added separately to each well of the 96-well biolog plates. A series of two-fold dilutions of extracts (0.1–51.2 mg/mL) of *C. militaris* were made and a 50 µL aliquot was added from 1st to 10th rows. 11th row contained positive control and 12th row contained negative control. The positive controls for the bacterial and fungal MIC assays were Gentamycin (50 µg/mL) and Nystatin (50 µg/mL), respectively. The negative control in both assays was double-distilled water. After that, an aliquot of 50 µL of the microbial suspension was added to each well of the 96-well biolog plates and incubated at 37±2°C for 24 hrs (for bacteria) and 25±2°C for 48 hrs (for fungi). To determine MIC for tested bacteria, TTC (2,3,5-Triphenyltetrazolium chloride) was employed as an indicator of bacterial cell survival. After adding TTC, the colour of wells changed from colourless to various shades of pink, indicating the presence of live bacterial cells. The lowest concentration of the extracts in the well of a column at which no colour change was seen was the minimum inhibitory concentration (MIC), and this reaction was manually visualised. For determining MIC against tested fungi, no colorimetric reagent was employed. Instead, formation of mycelial mat was used as an indicator of fungal growth. Similarly, MIC of the extract was least in the well of a column where no fungal growth was observed.

Statistical analysis

Experimentation was done in triplicates. Statistical comparison was performed between the data using SPSS 16.0 software, mean value and standard deviation were computed. Duncan's multiple range test (DMRT) was employed for comparing all of the treatments along with the comparisons between individual means at $P \leq 0.05$. Outcomes were mentioned in the forms of mean value and standard deviation (SD).

RESULTS AND DISCUSSION

Antifungal activity

The evaluation of antifungal activity of varying concentrations of different extracts (ethanolic, methanolic and aqueous) of *C. militaris* against *Aspergillus niger*, *Fusarium oxysporum*, *Penicillium chrysogenum* and *Mucor mucedo* was accessed by measuring the zone of inhibition (mm). It was revealed from the results (Table 2) that all the extracts exhibited concentration-dependent antifungal activity, with higher zones of inhibition observed at higher concentration. The highest concentration (60 mg/mL) of methanolic extract exhibited greater inhibition zones against *Fusarium oxysporum* (15.42±0.45 mm), followed by *Mucor mucedo* (14.48±0.49 mm), *Penicillium chrysogenum* (14.19±0.26 mm) and *Aspergillus niger* (13.47±0.35 mm) (Fig. 3 & 11). The ethanolic extract showed moderate results (Fig. 2 & 10) against the tested pathogens, with diameter of inhibition zones ranging from 8.35 to 12.81. Nevertheless, the aqueous extract exhibited the least antifungal efficacy against all the tested pathogenic fungi, with inhibition zones ranging from 7.10 to 10.84 mm (Fig. 1 & 9).

Antibacterial activity

In the present investigation, the antibacterial efficacy of *C. militaris* extracts (ethanolic, methanolic and aqueous) against *Escherichia coli*, *Staphylococcus aureus*, *Proteus vulgaris* and *Klebsiella pneumoniae* was determined by measuring the diameter of inhibition zones (mm). The antibacterial activity of all the extracts was concentration-dependent, with larger zones of inhibition seen at higher concentrations (Table 3).

The results revealed that the methanolic extract showed the largest zone of inhibition against *Escherichia coli* (16.27 ± 0.40 mm) at highest concentration (60 mg/mL), followed by *Staphylococcus aureus* (15.46 ± 0.57 mm), *Klebsiella pneumoniae* (15.19 ± 0.23 mm) and *Proteus vulgaris* (14.26 ± 0.49 mm) (Fig. 6 & 14). The diameter of inhibition zones of ethanolic extract ranged from 9.00 to 13.71 mm (Fig. 5 & 13), indicating intermediate performance against the tested bacterial pathogens. With inhibition zones ranging from 7.80 to 11.65 mm (Fig. 4 & 12), the aqueous extract, on the other hand, demonstrated least antibacterial efficacy against all the tested pathogenic bacteria.

Minimum inhibitory concentration of *C. militaris* extracts

In the present investigation, the ethanolic, methanolic, and aqueous extracts of *C. militaris* were tested against several bacterial and fungal pathogens to determine their minimum inhibitory concentration (MIC) values. It was clearly revealed from the results (Tables 4 & 5; Fig. 7 & 8) that methanolic extract exhibited the lowest MIC values against both bacterial and fungal pathogens. For bacteria, the methanolic extract showed MIC of 1.07 ± 0.38 mg/mL against *Escherichia coli*, 1.33 ± 0.37 mg/mL against *Staphylococcus aureus*, 2.66 ± 0.75 mg/mL against *Proteus vulgaris* and 1.60 ± 0.00 mg/mL against *Klebsiella pneumoniae* (Fig. 16), while as for fungi, the said extract exhibited the MIC values of 3.73 ± 1.99 , 1.07 ± 0.38 , 1.60 ± 0.00 and 1.85 ± 0.99 mg/mL for *Aspergillus niger*, *Fusarium oxysporum*, *Penicillium chrysogenum* and *Mucor mucedo*, respectively (Fig. 15). Likewise, ethanolic extract showed remarkably low MIC (minimum inhibitory concentration) values, ranging from 1.60 to 5.33 mg/mL for bacteria and 2.13 to 7.45 mg/mL for fungi. In contrast, aqueous extract exhibited comparatively least MIC values against tested bacterial and fungal pathogens.

In the present investigation, it was observed that *C. militaris* extracts showed significant antimicrobial activity, varying with type of solvent and extract concentration. Among all the extracts, the maximum antimicrobial activity was observed in methanolic extract, followed by ethanolic

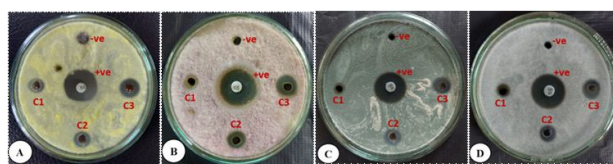


Fig. 1. Antifungal activity of aqueous extract of *C. militaris* against A. *Aspergillus niger*; B. *Fusarium oxysporum*; C. *Penicillium chrysogenum*; D. *Mucor mucedo*

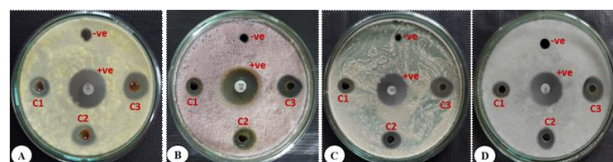


Fig. 2. Antifungal activity of ethanolic extract of *C. militaris* against A. *Aspergillus niger*; B. *Fusarium oxysporum*; C. *Penicillium chrysogenum*; D. *Mucor mucedo*

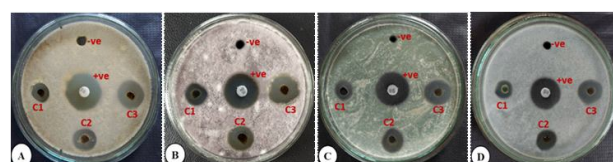


Fig. 3. Antifungal activity of methanolic extract of *C. militaris* against A. *Aspergillus niger*; B. *Fusarium oxysporum*; C. *Penicillium chrysogenum*; D. *Mucor mucedo*

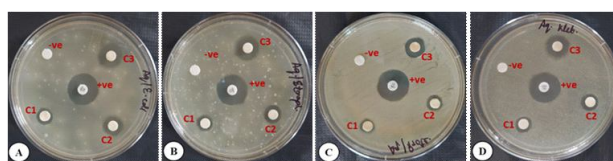


Fig. 4. Antibacterial activity of aqueous extract of *C. militaris* against A. *Escherichia coli*; B. *Staphylococcus aureus*; C. *Proteus vulgaris*; D. *Klebsiella pneumoniae*

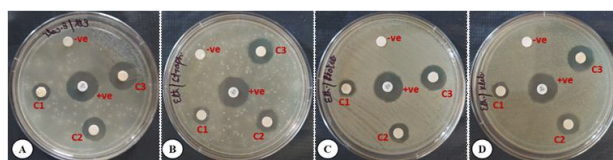


Fig. 5 : Antibacterial activity of ethanolic extract of *C. militaris* against A. *Escherichia coli*; B. *Staphylococcus aureus*; C. *Proteus vulgaris*; D. *Klebsiella pneumoniae*

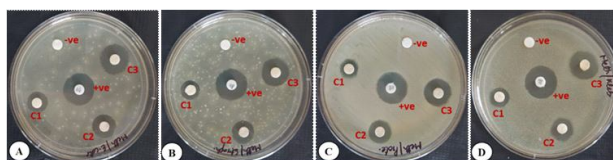


Fig. 6. Antibacterial activity of methanolic extract of *C. militaris* against A. *Escherichia coli*; B. *Staphylococcus aureus*; C. *Proteus vulgaris*; D. *Klebsiella pneumoniae*

extract, while, aqueous extract exhibited the least antimicrobial efficacy. The results indicates that methanol is effective solvent for the extracting antimicrobial bioactive compounds from *C. militaris*. This can be attributed to the presence

Table 1: List of species employed as test micro-organisms.

Bacteria/Fungi	Source
<i>Escherichia coli</i> (MTCC-407)	CSIR-IMTECH, Chandigarh, India
<i>Klebsiella pneumoniae</i> (MTCC-19)	CSIR-IMTECH, Chandigarh, India
<i>Proteus vulgaris</i> (MTCC-426)	CSIR-IMTECH, Chandigarh, India
<i>Staphylococcus aureus</i> (MTCC-96)	CSIR-IMTECH, Chandigarh, India
<i>Aspergillus niger</i>	Plant Pathology and Mycology, University of Kashmir, Srinagar, India.
<i>Fusarium oxysporum</i>	Plant Pathology and Mycology, University of Kashmir, Srinagar, India.
<i>Penicillium chrysogenum</i>	Plant Pathology and Mycology, University of Kashmir, Srinagar, India.
<i>Mucor mucedo</i>	Plant Pathology and Mycology, University of Kashmir, Srinagar, India.

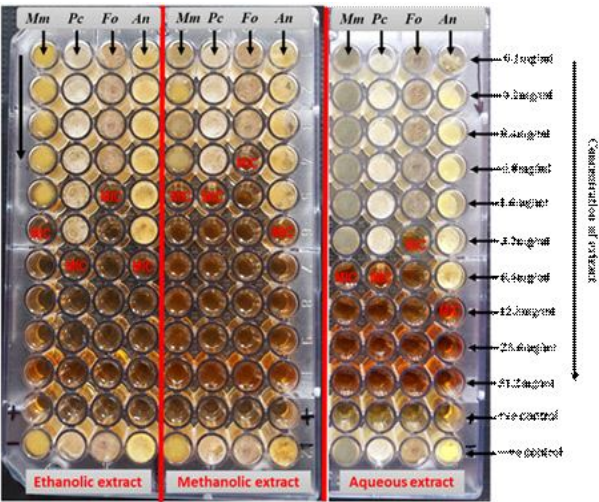


Fig. 7. MIC values of *C. militaris* extracts against *Aspergillus niger* (An), *Fusarium oxysporum*(Fo), *Penicillium chrysogenum* (Pc) and *Mucor mucedo* (Mm)

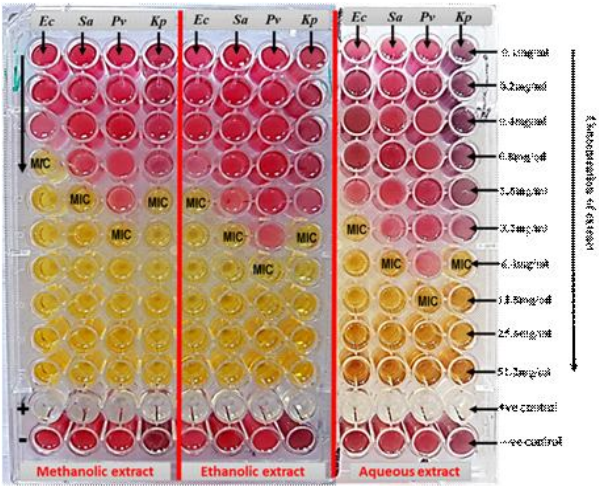


Fig. 8. MIC values of *C. militaris* extracts against *Escherichia coli* (Ec), *Staphylococcus aureus* (Sa), *Proteus vulgaris* (Pv), and *Klebsiella pneumoniae* (Kp)

of non-polar bioactive metabolites, such as, cordycepin and ergosterol which are known to show antimicrobial activity (Cunningham *et al.* 1950). Furthermore, all the extracts exhibited antimicrobial activity in dose-dependent manner, which is in agreement with the earlier studies. Park *et al.* (2009) purified and characterized a

novel protease from *C. militaris* and named it as *C. militaris* protein (CMP) which exhibited strong antifungal activity against the fungus, *Fusarium oxysporum*. Wong *et al.* (2011), isolated an antifungal peptide, cordymin from *C. militaris*, that exhibited antifungal activity against a number of pathogenic fungi, such as *Rhizoctonia solani*,

Table 2: Effect of *C. militaris* extracts on test pathogenic fungi

<i>C. militaris</i> extract	Test fungus	Conc.	*Diameter of inhibition zone (mm)			
			15 mg/mL	30 mg/mL	60 mg/mL	Control
Aqueous	<i>Aspergillus niger</i>		7.10±0.70 ^e	7.89±0.35 ^f	9.45±0.21 ^g	16.80±0.50 ^c
	<i>Fusarium oxysporum</i>		8.37±0.21 ^d	9.20±0.40 ^d	10.84±0.50 ^f	18.04±0.46 ^a
	<i>Penicillium chrysogenum</i>		7.24±0.56 ^e	8.03±0.34 ^e	9.60±0.45 ^g	17.30±0.60 ^b
	<i>Mucor mucedo</i>		7.57±0.40 ^e	8.40±0.60 ^e	9.95±0.75 ^g	17.54±0.21 ^b
Ethanol	<i>Aspergillus niger</i>		8.35±0.80 ^d	9.59±0.55 ^d	11.30±0.40 ^e	16.36±0.32 ^c
	<i>Fusarium oxysporum</i>		9.65±0.64 ^c	10.90±0.32 ^c	12.81±0.31 ^d	18.65±0.26 ^a
	<i>Penicillium chrysogenum</i>		8.46±0.23 ^d	9.68±0.49 ^d	11.53±0.60 ^e	17.00±0.71 ^b
	<i>Mucor mucedo</i>		8.77±0.70 ^d	9.92±0.40 ^d	11.79±0.87 ^e	17.25±0.92 ^b
Methanol	<i>Aspergillus niger</i>		9.86±0.42 ^c	11.32±0.50 ^b	13.47±0.35 ^c	16.75±0.20 ^c
	<i>Fusarium oxysporum</i>		11.40±0.55 ^a	12.85±0.71 ^a	15.42±0.45 ^a	18.30±0.31 ^a
	<i>Penicillium chrysogenum</i>		10.11±0.60 ^b	11.56±0.36 ^b	14.19±0.26 ^b	17.15±0.87 ^b
	<i>Mucor mucedo</i>		10.45±0.32 ^b	11.92±0.92 ^b	14.48±0.49 ^b	17.00±0.40 ^b

*Values are shown as the mean of three replicates.

Values with different superscripts indicate statistically significance difference ($P \leq 0.01$) between the values

Mycosphaerella arachidicola and *Bipolaris maydis*. Reis *et al.* (2013) reported that the methanolic extract of *C. militaris* exhibited strong antibacterial and antifungal activities against *Bacillus cereus* and *Aspergillus* species, respectively. In the study conducted by Dong *et al.* (2014), the methanolic extract from *C. militaris* stroma showed broad antibacterial activity against all tested fungi and bacteria, while methanolic extract from fermented mycelia demonstrated selective antimicrobial activity. According to Joshi *et al.* (2019), methanolic extract of *C. militaris* showed antimicrobial efficacy, with minimum inhibitory concentrations (MICs), ranging from 0.625 to 5 mg/mL against *Escherichia coli* and 1.25 to 10 mg/mL against *Staphylococcus aureus* as found in our study. The antibacterial potential of *C. militaris* extracts was assessed by Aanchal *et al.* (2024), who reported that both aqueous and

methanolic extracts were effective against both Gram-negative (*Escherichia coli*) and Gram-positive (*Bacillus subtilis*) bacteria. Kaewkod *et al.* (2024) also reported that the extracts of *C. militaris* and their bioactive compounds showed strong antibacterial effects against enteric pathogenic bacteria, preventing their adhesion and invasion on Caco-2 colon cells. Recently, like in our study, Afzal *et al.* (2025) found that the methanolic extract of *C. militaris* showed strong antimicrobial activity against *Escherichia coli*, *Klebsiella pneumoniae*, *Staphylococcus aureus* and *Streptococcus viridans*.

DECLARATION

Conflict of Interest. Authors declare no conflicts of interest.

Table 3 : Effect of *C. militaris* extracts on test pathogenic bacteria

<i>C. militaris</i> extract	Conc.	*Diameter of inhibition zone (mm)			
		15 mg/mL	30 mg/mL	60 mg/mL	Control
	Test bacterium				
Aqueous	<i>Escherichia coli</i>	9.13±0.40 ^d	10.00±0.17 ^d	11.65±0.21 ^f	18.90±0.15 ^a
	<i>Staphylococcus aureus</i>	8.55±0.38 ^e	9.32±0.55 ^e	10.87±0.45 ^g	17.34±0.35 ^b
	<i>Proteus vulgaris</i>	7.80±0.21 ^f	8.65±0.32 ^f	10.11±0.36 ^g	16.50±0.32 ^c
	<i>Klebsiella pneumoniae</i>	8.40±0.56 ^e	9.25±0.26 ^e	10.69±0.50 ^g	17.20±0.41 ^b
Ethanol	<i>Escherichia coli</i>	10.40±0.45 ^c	11.69±0.20 ^c	13.71±0.60 ^d	18.55±0.37 ^a
	<i>Staphylococcus aureus</i>	9.77±0.21 ^d	10.96±0.66 ^d	12.90±0.41 ^e	17.70±0.70 ^b
	<i>Proteus vulgaris</i>	9.00±0.50 ^d	10.18±0.35 ^d	12.06±0.32 ^e	16.36±0.34 ^c
	<i>Klebsiella pneumoniae</i>	9.62±0.17 ^d	10.84±0.21 ^d	12.78±0.35 ^e	17.64±0.40 ^b
Methanol	<i>Escherichia coli</i>	12.00±0.52 ^a	13.62±0.31 ^a	16.27±0.40 ^a	18.00±0.17 ^a
	<i>Staphylococcus aureus</i>	11.43±0.40 ^b	12.98±0.35 ^b	15.46±0.57 ^b	17.56±0.26 ^b
	<i>Proteus vulgaris</i>	10.31±0.21 ^c	11.80±0.44 ^c	14.26±0.49 ^c	16.79±0.46 ^c
	<i>Klebsiella pneumoniae</i>	11.04±0.34 ^b	12.57±0.17 ^b	15.19±0.23 ^b	17.85±0.21 ^b

*Values are shown as the mean of three replicates.

Values with different superscripts indicate statistically significance difference ($P \leq 0.01$) between the values

Table 4 : Minimum inhibitory concentration (MIC) against test fungal pathogens

<i>C. militaris</i> extract	*MIC (mg/mL) values			
	Test fungal pathogens			
	<i>Aspergillus niger</i>	<i>Fusarium oxysporum</i>	<i>Penicillium chrysogenum</i>	<i>Mucor mucedo</i>
Aqueous	10.67±3.01 ^a	3.20±0.00 ^a	8.53±3.02 ^a	6.40±0.00 ^a
Ethanol	7.45±3.99 ^b	2.13±0.75 ^b	5.33±1.50 ^b	3.20±0.00 ^b
Methanol	3.73±1.99 ^c	1.07±0.38 ^c	1.60±0.00 ^c	1.85±0.99 ^c

*Values are shown as the mean of three replicates.

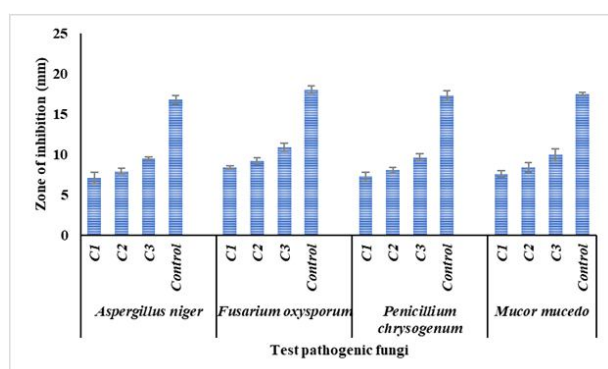
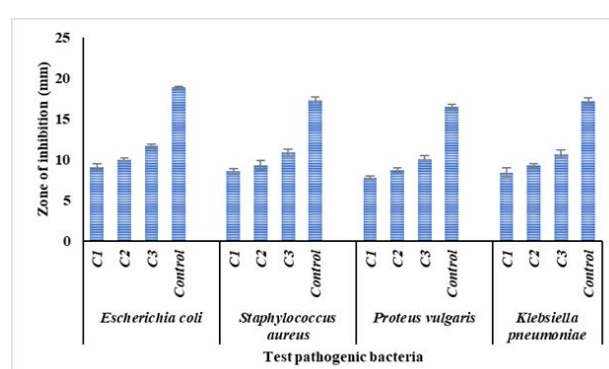
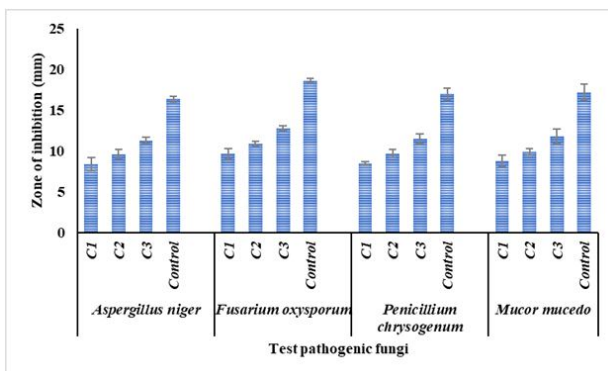
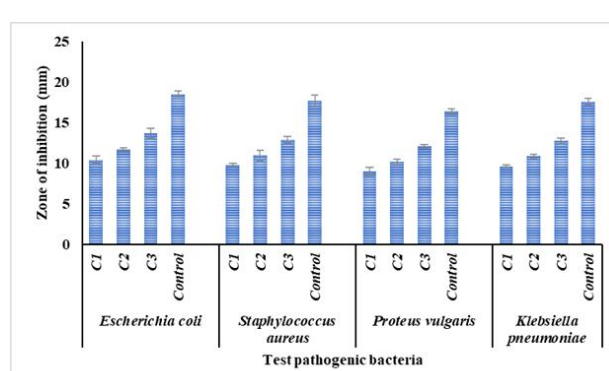
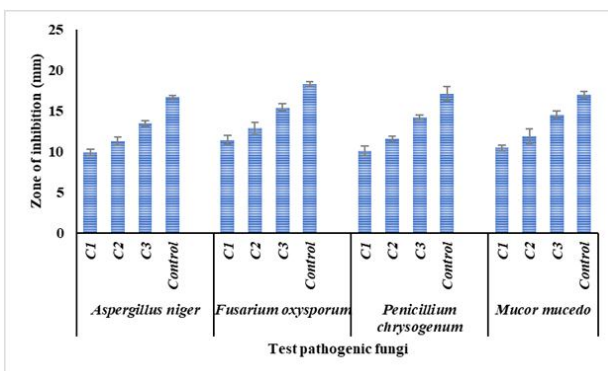
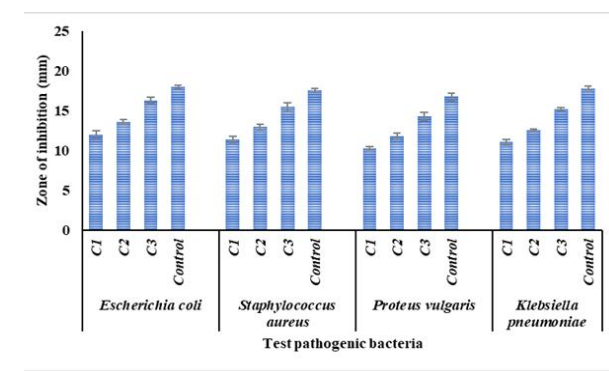
Values with different superscripts indicate statistically significance difference ($P \leq 0.01$) between the values

Table 5: Minimum inhibitory concentration (MIC) against test bacterial pathogens

<i>C. militaris</i> extract	*MIC (mg/mL) values			
	Test bacterial pathogens			
	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>	<i>Proteus vulgaris</i>	<i>Klebsiella pneumoniae</i>
Aqueous	2.66±0.75 ^a	6.4±0.00 ^a	10.66±3.01 ^a	8.53±3.01 ^a
Ethanol	1.6±0.00 ^b	3.73±1.99 ^b	5.33±1.50 ^b	4.26±1.51 ^b
Methanol	1.07±0.38 ^b	1.33±0.37 ^c	2.66±0.75 ^c	1.60±0.00 ^c

*Values are shown as the mean of three replicates.

Values with different superscripts indicate statistically significance difference ($P \leq 0.01$) between the values

**Fig. 9.** Antifungal activity of aqueous extract of *C. militaris* against some pathogenic fungi**Fig.12** Antibacterial activity of aqueous extract of *C. militaris* against some pathogenic bacterial strains**Fig. 10.** Antifungal activity of ethanolic extract of *C. militaris* against some pathogenic fungi**Fig. 13.** Antibacterial activity of ethanolic extract of *C. militaris* against some pathogenic bacterial strains**Fig. 11.** Antifungal activity of methanolic extract of *C. militaris* against some pathogenic fungi**Fig. 14.** Antibacterial activity of methanolic extract of *C. militaris* against some pathogenic bacterial strains

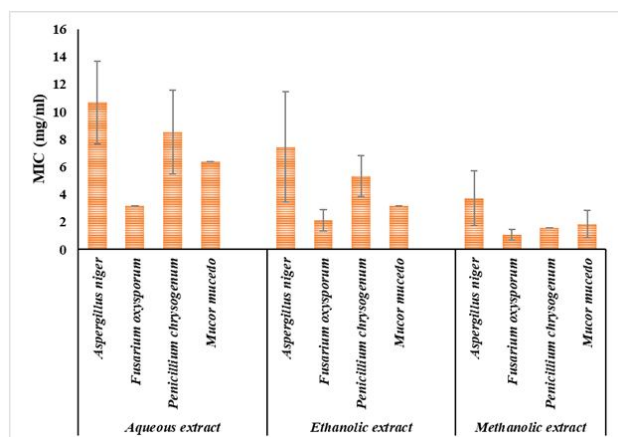


Fig. 15. MIC values of *C. militaris* extracts against test fungal pathogens.

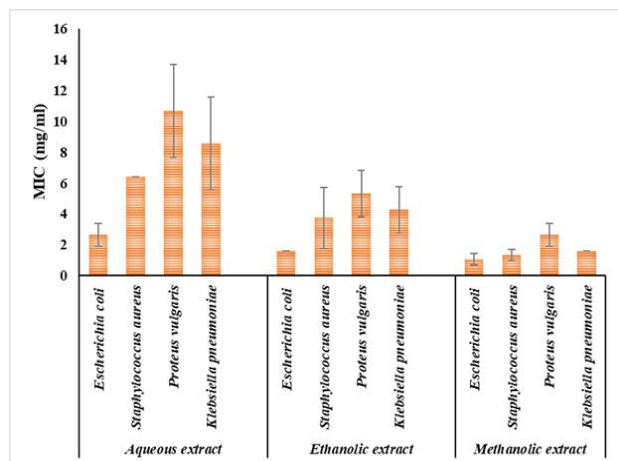


Fig. 16. MIC values of *C. militaris* extracts against test bacterial pathogens.

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