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Phytochemical analysis and biological activities of three wild *Mesembryanthemum* species growing in heterogeneous habitats

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ABSTRACT

The objective of this study was to analyze the phytochemicals and to determine the antioxidant, antibacterial and allelopathic potential of three wild *Mesembryanthemum* species (*M. crystallinum* L., *M. forsskaolii* Hochst. Ex Boiss and *M. nodiflorum* L.). The phytochemical composition of the methanolic extract of studied species revealed the considerable quantities that might be responsible for their powerful antioxidant activity. The IC₅₀ values were 386.51, 592.97, and 752.23 μg/ml for *M. nodiflorum*, *M. crystallinum* and *M. forsskaolii* extracts respectively. The antibacterial activity index was calculated for each extract in comparison with the standard antibiotics. *M. nodiflorum* showed higher potency than ampicillin and penicillin G against against *Klebsiella pneumoniae*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Bacillus subtilis*. The allelopathic potential showed that the studied *Mesembryanthemum* species expressed a significant phytotoxic activity against *Chenopodium murale* weed in a dose dependent manner. *M. nodiflorum* sample showed most phytotoxic effect among the studied species.

KEYWORDS: *Mesembryanthemum* species, antioxidants, antimicrobials, activity index, phytotoxic activity

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INTRODUCTION

There is a worldwide increasing interest in using the traditional foods and ingredients from natural resources (Muthukrishnan *et al.*, 2018). The Egyptian desert is blessed with plenty of wild plants that significantly affect the daily life of Bedouins in urban areas (Zahrán & El-Amier, 2013). The Egyptian flora plays a key role in maintaining the region's environmental sustainability (McClanahan, 1998; Zaki *et al.*, 2018). In addition to their role in stabilizing slope and improving soils, plants are rich in chemical constituents and are used in insecticidal, herbicidal, folk medicine and other industrial applications (Hosseinzadeh *et al.*, 2015; Naboulsi *et al.*, 2018). The knowledge about the phytoconstituents of plants leads to a better understanding of their possible medicinal and agro-industrial uses (Yakhin *et al.*, 2017). The biological activity of plants might be ascribed to the presence of secondary metabolites like polyphenols, flavonoids, lignins, alkaloids, terpenoids, carotenoids, vitamins, etc. (Vinson *et al.*, 2005; Zaki *et al.*, 2016; Zaki *et al.*, 2017).

Family Aizoaceae is a widely distributed family in Africa that comprises around 127 genera and 1860 species distributed in tropical and sub-tropical areas. They are also cultivated as ornamental ground covers (Mabberley, 1997; Klak *et al.*, 2004). In Egypt, it is represented by 9 species that are succulents ranged from pebble-like leaf succulents to small succulent shrubs. They are characterized by distinctive seed capsules. In Egyptian flora, three species of *Mesembryanthemum* (*M. crystallinum* L., *M. forsskaolii* Hochst. ex Boiss and *M. nodiflorum* L.) were recorded and is distributed in Mediterranean coastal strip, Desert and Oases (Boulos, 1999). Members of family Aizoaceae are known to have diverse biological activities including antihyperlipidemic, antipyretic, diuretic, antioxidant, anticancer, larvicidal, analgesic, anti-rheumatic, anticholera, emetic, laxative, anti-inflammatory and antimicrobial (Ibtissem *et al.*, 2010; Mohammed *et al.*, 2012; Ibtissem *et al.*, 2012; Doudach *et al.*, 2013; Moawad *et al.*, 2016).

The biological activities of genus *Mesembryanthemum* like antioxidant, antimicrobial, anticarcinogenic and antiviral

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activity was reported (Rood, 1994; van Wyk, 2008; Falleh *et al.*, 2011; Gawad *et al.*, 2018). Moreover, this genus has recorded to possess several ethnomedicinal uses such as treatment of liver diseases, ocular infection, alexiteric, analgesic, laxative and diabetes (Mustafa *et al.*, 1995; Bouftira *et al.*, 2009; Al-Faris *et al.*, 2010; Falleh *et al.*, 2011). The phytochemical screening of *Mesembryanthemum* genus concluded the presence of carbohydrates, protein, antioxidant enzymes, tannins, triterpenes, alkaloids and flavonoids van (der Watt and Pretorius, 2001; Bouftira *et al.*, 2008; Doudach *et al.*, 2013). Therefore, our study aimed to determine the phytochemical constituents present in the studied *Mesembryanthemum* species collected from two different habitats in Egypt, to evaluate the antioxidant activity, the antimicrobial potential against several pathogenic bacterial strains and the allelopathic effect of the plants extracts against the noxious weed *Chenopodium murale* as potential green eco-friendly bioherbicide.

MATERIALS AND METHODS

Preparation of Plant Material

The aerial parts of *M. crystallinum* and *M. nodiflorum* were gathered during flowering stage from sand flats in north Nile Delta of Egypt (31°29'28.08"N 31°23'46.90"E), while *M. forsskaolii* was collected from Wadi Hagul, North Eastern Desert, Egypt (29°53'27.31"N 32°13'22.58"E). The studied species were identified according to Täckholm (1974) and Boulos (1999) (Table 1). The collected plants were dried in shade at room temperature for 21 days, grinded into fine powder and kept in a polyethylene container for further use.

Phytochemical Analysis

Qualitative analysis

Phytochemical screening for the presence of alkaloids, saponins, tannins, phenolics, anthraquinone, steroid, flavonoids, glucosides and terpenoids in the studied species was done using the standard methods described by Sofowora (1996), Williamson *et al.* (1996), Harborne (1998), Evans (2000), Banso and Ngbede (2006) and Rasineni *et al.* (2008).

Quantitative analysis

The total phenolics were measured according to the method adopted by Chlopicka *et al.* (2012), the total flavonoids were estimated according to Stankovic *et al.* (2011), the total tannins were measured according to van Buren and Robinson (1969) and the alkaloids were measured according to the assay adopted by Joshi *et al.* (2013).

Antioxidant Activities

DPPH radical scavenging capacity estimation

The radical scavenging activity of *Mesembryanthemum* species extracts was measured using 2,2-diphenyl-1-picryl-hydrazil (DPPH) radical according to the assay adopted by Miguel (2010). Briefly, 2 milliliters of 150 µM DPPH was added to 2 milliliters of plant extracts using concentrations from 100 to 1000 mg/l and kept in dark at 40 °C for 30 min. The absorbance was measured at 520 nm using Milton Roy Spectronic 21D UV-Visible Spectrophotometer (USA), all samples were analyzed in triplicate, and the IC₅₀ values were estimated using exponential curve.

$$\% \text{ of DPPH scavenging} = [1 - (A_{\text{sample}} / A_{\text{control}})] \times 100$$

Antibacterial Potential

Tested bacteria

The antimicrobial potential of *Mesembryanthemum* species extracts were estimated eight pathogenic bacteria including five Gram-negative bacteria (*Klebsiella pneumoniae* (ATCC10031), *Listeria monocytogenes* (ATCC19116), *Escherichia coli* (ATCC10536), *Salmonella typhi* (ATCC25566) and *Pseudomonas aeruginosa* (ATCC9027)) and three Gram-positive bacteria (*Streptococcus epidermis* (EMCC1353^t), *Staphylococcus aureus* (ATCC6538) and *Bacillus subtilis* (DMS1088)).

Disc diffusion assay

Sterilized filter paper discs were immersed in the studied extracts then loaded over the plates seeded with the tested strains and incubated at 37 °C for 18-24 hours. The zone of growth inhibition was estimated and subtracted from the diameter of the filter paper discs (6 mm) (Cappuccino & Sherman, 2008). The Activity Index (AI) was used as a parameter for measuring the antibacterial potential of the studied extracts in comparison with standard antibiotics (Shekhawat & Vijayvergia, 2010).

$$\text{Activity Index (AI)} = \text{Inhibition zone of samples} / \text{Inhibition zone of standard}$$

Allelopathic Activity

Chenopodium murale seeds were gathered from cultivated fields in Mansoura city, Egypt. Uniform and ripened seeds were sterilized by immersing in NaOCl (0.3%) for three minutes then washed by bi-distilled sterilized water several times. The sterilized seeds were dried and kept for future use.

Table 1: Description of the studied *Mesembryanthemum* species

Botanical name	Common name	Duration	Chorotype	Habitat
<i>M. crystallinum</i> L.	Ghasoul, Iceplant	Annual	ME+ER-SR	Sd, Sm, Dr, La
<i>M. forsskaolii</i> Hochst. exBoiss.	Hamad	Annual	SA-SI	Sm
<i>M. nodiflorum</i> L.	Ghasoul	Annual	ME+SA-SI+ER-SR	Sd, Sm, Rw, Dr, La

ME: Mediterranean; ER-SR: Euro-Siberian; SA-SI: Saharo-Sindian; Sd: Sand dunes; Sm: Salt marshes; Dr: Drains; La: Lake; Rw: Railways

The phytotoxic activity was measured using glass Petri dishes with bottom covered with Whatman No. 1 filter paper, on each of them 20 sterilized seeds were loaded. 4 ml of each extract was added in concentrations of 2.5, 5, 10, 20 and 40 g/l, then the dishes were incubated in growth chamber at 25-27 °C (Rice, 1972; Abd El-Gawad *et al.*, 2015). Three replications for each treatment were measured. The percent of germination and the inhibition of germination of shoot and root growth was calculated as follows: Inhibition % = 100×(Length of control – Length of treatment)/Length of control.

RESULTS AND DISCUSSION

Phytochemical Constituents

Nowadays, plants are considered as important sources of bioactive molecules that are beneficial in various fields and could be used as drugs, food supplements, antimicrobials, allelochemicals, etc. The phytochemical screening of the dried aerial parts of the studied *Mesembryanthemum* species revealed the presence of all of the estimated phytoconstituents except anthraquinones in *M. crystallinum* and *M. nodiflorum* species while *M. forsskaolii* showed absence of anthraquinone and glycosides and presence of the other phytoconstituents in traces as recorded in Table 2. The ethanolic extract obtained from each of the studied plants was quantitatively assayed for phenolics, flavonoids, tannins and alkaloids. Ethanol was chosen due to its efficiency of extraction for biologically active compounds like phenolics (Raks *et al.*, 2018; Krakowska-Sieprawska *et al.*, 2020). In the present study, there were variable levels of alkaloids, phenolics, flavonoids and tannins in *Mesembryanthemum* species. *M. nodiflorum* expressed the highest content of tannins, alkaloids and phenolics (36.14, 9.22 and 21.36 mg/g dried plant, respectively) followed by *M. crystallinum* (31.64, 6.21 and 19.55 mg/g dried plant, respectively) while the lowest content was in *M. forsskaolii* (22.87, 3.88 and 16.22 mg/g dried plant, respectively). The highest content of flavonoids recorded in *M. crystallinum* (11.41 mg/g dried plant) as illustrated in Figure 1. Our data were comparable with earlier studies on common desert plants (Hariprasad & Ramakrishna, 2011; Alzuaibr *et al.*, 2020; El-Amier & Al-hadithy, 2020), but were not in agreement of Bohnert and Cushman (2000) who depicted that *M. crystallinum* tissues contain negligible amounts of phenolic and flavonoid compounds.

Antioxidant Activity

Halophytic plants have the ability to overcome and scavenge harmful reactive oxygen and reactive nitrogen species produced under stress of increased salinity, since they possess a powerful antioxidant system (Qiu-Fang *et al.*, 2005; Kapoor *et al.*, 2019). The antioxidant properties of *Mesembryanthemum* species have been evaluated by measuring their DPPH radical scavenging activity using the ethanolic extracts of the studied species. All organic extracts of *Mesembryanthemum* species exhibited an antioxidant activity in a dose dependent manner, which was comparable with ascorbic acid as reference standard (Table 3). IC₅₀ values were calculated and recorded in Table 3. The IC₅₀

value of *M. nodiflorum*, *M. crystallinum* and *M. forsskaolii* extracts were 386.51, 592.97, and 752.23 µg ml⁻¹, respectively and that of ascorbic acid was 112.31 µg ml⁻¹. These results suggest

Table 2: Qualitative phytochemical screening of the studied *Mesembryanthemum* species

Phytochemical screened	<i>Mesembryanthemum</i> species		
	<i>M. crystallinum</i>	<i>M. nodiflorum</i>	<i>M. forsskaolii</i>
Alkaloid	+	+++	++
Saponins	+	+	+
Tannins	++	++	+
Flavonoids	++	+++	+
Phenolics	++	+++	++
Glycosides	++	+	-
Anthraquinone	-	-	-
Steroids	+	++	+
Terpenoids	+	++	+

(+): presence; (++): considerable presence; (+++): abundance; (-): absence

Table 3: The antioxidant scavenging activity of the studied *Mesembryanthemum* species

Studied species	Concentrations (µg/ml)	% of DPPH scavenging	IC ₅₀ (µg/ml)
<i>M. crystallinum</i>	50	19.02±0.51	592.97
	100	25.45±0.68	
	200	32.48±0.87	
	400	42.17±1.13	
	600	52.21±1.39	
	800	55.84±1.49	
<i>M. nodiflorum</i>	50	17.36±0.46	386.51
	100	33.47±0.89	
	200	41.23±1.10	
	400	55.38±1.48	
	600	67.49±1.80	
	800	73.37±1.96	
<i>M. forsskaolii</i>	50	7.42±0.20	752.23
	100	12.50±0.33	
	200	25.29±0.67	
	400	33.59±0.90	
	600	41.55±1.11	
	800	50.43±1.35	
Ascorbic acid			112.31

Values are means ± standard error (n=3). IC₅₀: the antioxidant concentration capable of diminishing 50% of the used DPPH radical

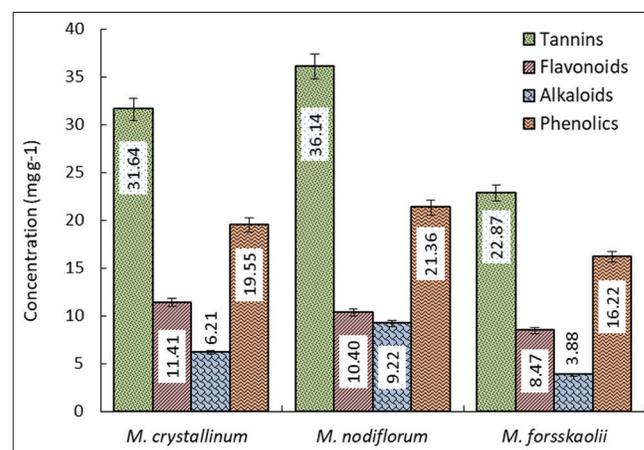


Figure 1: The active secondary constituents in the studied *Mesembryanthemum* species

that ethanol extracts of *Mesembryanthemum* species has an obvious effect on scavenging of DPPH radical. *M. nodiflorum* expressed the highest antioxidant activity followed by *M. crystallinum* while *M. forsskaolii* was the lowest. These results could be attributed to the levels of phytoconstituents that are responsible for the antioxidant activity. Antioxidant enzymes and compounds like ascorbic acid, phenolics, flavonoids and glutathione provide protection for living organisms against free radical damage (Rice-Evans *et al.*, 1996; Chen *et al.*, 2020). The antioxidant activity of the studied *Mesembryanthemum* species might be attributed to their high content of phenolics, flavonoids, tannins, alkaloids, triterpenes and antioxidant enzymes (van der Watt & Pretorius, 2001; Bouftira *et al.*, 2008; Doudach *et al.*, 2013; Atzori *et al.*, 2017) that have antioxidant, antimicrobial, anticarcinogenic (Falleh *et al.*, 2013) and ethnomedicinal uses (Bouftira *et al.*, 2009; Al-Faris *et al.*, 2010). The estimated antioxidant activity of *M. crystallinum*, *M. nodiflorum* and *M. forsskaolii* were higher than those reported by Ibtissem *et al.* (2012), Doudach *et al.* (2013) and Moawad *et al.* (2016), respectively and were comparable with those of Salem *et al.* (2016), Alzuaibr *et al.* (2020) and El-Amier and Al-hadithy (2020) on some similar xerophytes.

Antibacterial Activity

The antibacterial activity of crude extracts of *Mesembryanthemum* species was estimated in vitro using 8 different pathogenic bacterial strains. The studied extracts exhibited broad antibacterial spectrum (Table 4).

The results in Figure 2 showed that the ethanolic extract of *Mesembryanthemum* species exhibited broad antimicrobial spectrum against most of the tested bacterial strains where each of *M. nodiflorum* and *M. crystallinum* inhibited 62.5% while *M. forsskaolii* inhibited only 50% of the screened bacterial strains as illustrated in Figure 2. The ethanolic extract of *M. nodiflorum* expressed the highest zones of inhibition against the tested gram positive bacteria *S. aureus* and *B. subtilis* (21.2 ± 0.87 and 19.8 ± 0.74 mm, respectively) and gram negative bacteria *P. aeruginosa*, *E. coli* and *K. pneumoniae* (25.3 ± 0.94 , 21.7 ± 0.92 and 19.3 ± 0.82 mm, respectively), while *S. aureus* was the most sensitive bacteria in case of *M. crystallinum* extract (21.4 ± 0.91 mm). *M. forsskaolii* extract exhibited no potential against

any of the tested pathogens. *S. typhi* and *S. epidermis* were the most resistant bacteria among the tested strains to the *Mesembryanthemum* species extracts.

The phytochemical constituents like tannins, triterpenes, alkaloids, phenolics and flavonoids might be the motive of the activity against the examined bacterial strains (van der Watt & Pretorius, 2001; Bouftira *et al.*, 2008; Doudach *et al.*, 2013) and this agrees with the results obtained as *M. nodiflorum* content of these chemicals was the highest among the three studied *Mesembryanthemum* species, meanwhile it expressed the broadest antibacterial spectrum with significant inhibitory activity.

It is obvious from the results that that gram-negative bacteria are more sensitive to three plant extracts than gram-positive bacteria and these results agrees with the previously reported by Kaneria *et al.* (2009), El-Amier *et al.* (2014), Kumar *et al.* (2016). Manandhar *et al.* (2019) reported that the nature of the cell wall of Gram-negative bacteria make them more susceptible to different compounds than Gram positive bacteria. The antimicrobial activity of plants seems to depend on the efficacy of the extraction, the solvent used and the metabolic activity of

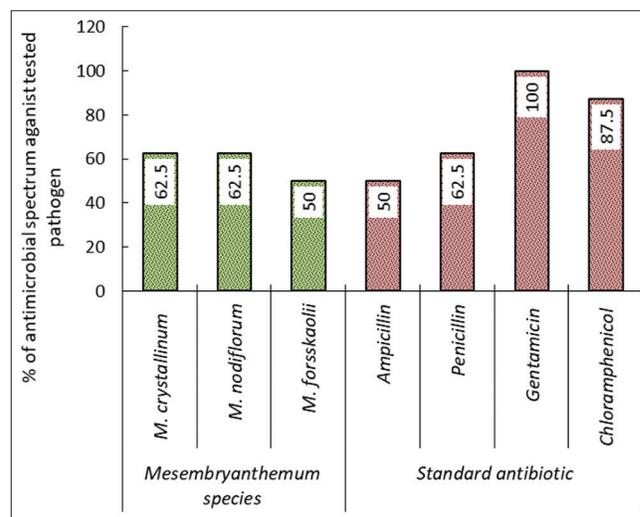


Figure 2: % of antimicrobial spectrum of *Mesembryanthemum* species extracts and standard antibiotics against the studied pathogenic bacteria

Table 4: The antibacterial potential of *Mesembryanthemum* species extracts against the tested pathogens

Tested microorganisms	Zone of inhibition for the extracts			DMSO*	Zone of inhibition for the Standard antibiotic				LSD _{0.05}
	<i>M. crystallinum</i>	<i>M. nodiflorum</i>	<i>M. forsskaolii</i>		Ampicillin	Penicillin	Gentamicin	Chloramphenicol	
Gram negative bacteria									
<i>Klebsiella pneumoniae</i>	12.3±0.52	19.3±0.82	11.4±0.46	n.a	16±0.68	n.a	21±0.89	32±0.85	1.043***
<i>Listeria monocytogenes</i>	11.2±0.48	n.a	n.a	n.a	n.a	7±0.30	22±0.93	n.a	0.423***
<i>Escherichia coli</i>	n.a	21.7±0.92	n.a	n.a	10±0.42	15±0.64	20±0.85	22±0.77	0.503***
<i>Salmonella typhi</i>	n.a	n.a	n.a	n.a	n.a	11±0.47	24±0.91	21±0.68	0.160***
<i>Pseudomonas aeruginosa</i>	13.5±0.57	25.3±0.94	7.2±0.31	n.a	9±0.38	7±0.30	18±0.76	24±0.73	3.853***
Gram positive bacteria									
<i>Streptococcus epidermis</i>	n.a	n.a	n.a	n.a	n.a	n.a	30±0.58	29±0.57	0.288***
<i>Staphylococcus aureus</i>	21.4±0.91	21.2±0.87	5.1±0.22	n.a	12±0.51	17±0.72	26±0.84	27±0.92	3.748***
<i>Bacillus subtilis</i>	10.3±0.44	19.8±0.74	6.2±0.26	n.a	n.a	n.a	9±0.24	18±0.74	1.629***

Disc diameter 6mm was subtracted from each obtained value. Values are calculated as means ± standard error of triplicates. n.a: Not active, values of significant variation at $p < 0.05$

the tested microbes (Cowan, 1999; Kaneria *et al.*, 2009; Kumar *et al.*, 2016; Manandhar *et al.*, 2019).

Moreover, the results revealed that the antibacterial performance of ethanolic extracts of *M. nodiflorum* and *M. crystallinum* was better than the antibiotics ampicillin and penicillin as presented in Table 4, thus it could be suggested that ethanolic extracts of these species could be used as good alternative for antibiotics especially for antibiotic resistant bacteria (Table 4).

Activity Index (AI)

The significance of using the prepared extracts in comparison with the standard antibiotics (ampicillin, penicillin, gentamicin and chloramphenicol) against the tested pathogenic bacterial strains was estimated using the activity index (Table 5). The activity index varied in gram positive from 0.09 to 3.61 and in gram-negative bacteria from 0.2 – 2.20. The maximum activity index values were observed against *Pseudomonas aeruginosa* and *Escherichia coli* (3.61 and 2.17, respectively) while the lowest activity index value was for *Escherichia coli* (0.09). Ampicillin and penicillin expressed moderate and low effects against the tested pathogenic strains. Activity index values above one expressed higher potential of herbal extracts while those below one expressed higher antibiotics potential against

tested pathogenic strain (Shekhawat & Vijayvergia, 2010). The obtained results affirmed the strength of using the studied extracts in comparison with the tested antibiotics except chloramphenicol.

M. nodiflorum showed higher potency than ampicillin and penicillin G against *K. pneumoniae*, *E. coli*, *P. aeruginosa*, *S. aureus* and *B. subtilis*. It was also more potent than gentamicin and chloramphenicol against *P. aeruginosa* and *B. subtilis*. On the other hand, *M. monocytogenes* showed higher potency against *L. monocytogenes*, *P. aeruginosa*, *S. aureus* and *B. subtilis* than ampicillin and penicillin G. These results indicated the probability of using such these potent extracts against antibiotic resistance bacteria like *P. aeruginosa*, *S. aureus* and *K. pneumoniae*.

Allelopathic Activity

The ethanolic extracts of *Mesembryanthemum* species significantly exhibited allelopathic effect against *C. murale* weed in a concentration dependent manner (Figure 3). *M. nodiflorum* showed most phytotoxic effect among the three *Mesembryanthemum* samples. At the highest concentration (40 g mL⁻¹), the ethanolic extracts of *M. nodiflorum*, *M. crystallinum* and *M. forsskaolii* inhibited the germination of

Table 5: Activity index analysis of *Mesembryanthemum* species extracts against human pathogenic bacteria

Pathogens	Extracts of <i>Mesembryanthemum</i> species			Antibiotics (IZ)
	<i>M. crystallinum</i>	<i>M. nodiflorum</i>	<i>M. forsskaolii</i>	
<i>Klebsiella pneumoniae</i>	0.77	1.21	0.71	Ampicillin (16)
	E>A	E>A	E>A	Penicillin G (0)
	0.59	0.92	0.54	Gentamicin (21)
	0.38	0.6	0.36	Chloramphenicol (32)
<i>Listeria monocytogenes</i>	E>A	-	-	Ampicillin (0)
	1.6	-	-	Penicillin G (7)
	0.49	-	-	Gentamicin (22)
	E>A	-	-	Chloramphenicol (0)
<i>Escherichia coli</i>	-	2.17	-	Ampicillin (10)
	-	1.45	-	Penicillin G (15)
	-	0.99	-	Gentamicin (20)
	-	0.09	-	Chloramphenicol (22)
<i>Salmonella typhi</i>	-	-	-	Ampicillin (0)
	-	-	-	Penicillin G (11)
	-	-	-	Gentamicin (24)
	-	-	-	Chloramphenicol (21)
<i>Pseudomonas aeruginosa</i>	1.5	2.81	0.8	Ampicillin (9)
	1.93	3.61	1.03	Penicillin G (7)
	0.75	1.41	0.4	Gentamicin (18)
	0.56	1.05	0.3	Chloramphenicol (24)
<i>Streptococcus epidermis</i>	-	-	-	Ampicillin (0)
	-	-	-	Penicillin G (0)
	-	-	-	Gentamicin (30)
	-	-	-	Chloramphenicol (29)
<i>Staphylococcus aureus</i>	1.78	1.77	0.43	Ampicillin (12)
	1.26	1.25	0.3	Penicillin G (17)
	0.82	0.82	0.2	Gentamicin (26)
	0.79	0.79	0.19	Chloramphenicol (27)
<i>Bacillus subtilis</i>	E>A	E>A	E>A	Ampicillin (0)
	E>A	E>A	E>A	Penicillin G (0)
	1.14	2.2	0.69	Gentamicin (9)
	0.57	1.1	0.34	Chloramphenicol (18)

E: extract; A: antibiotics; E > A and > 1 values indicate extracts have higher effect against bacterial pathogens in comparison with antibiotics; E < A and < 1 values indicate antibiotics have higher effect against bacterial pathogens in comparison with extracts, IZ= inhibition zone

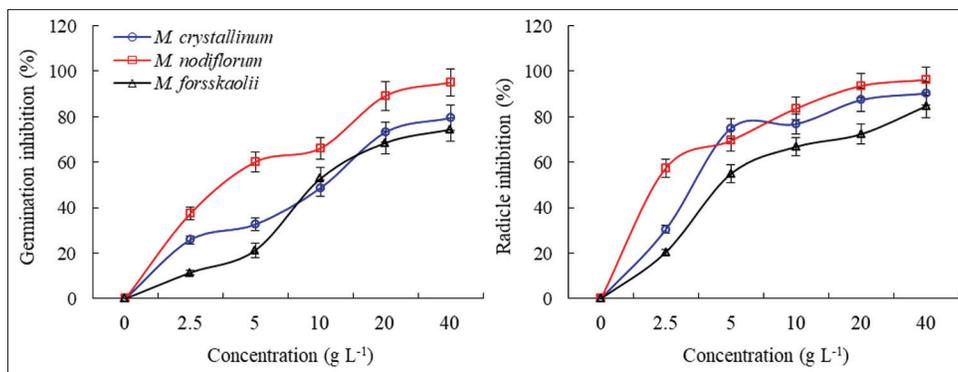


Figure 3: Allelopathic activity of the studied *Mesembryanthemum* species extracts on germination and radicle growth of *Chenopodium murale*.

C. murale by 95.11%, 79.48% and 74.41% with respect to control, while the radicle growth was inhibited by 96.17%, 90.22% and 84.66%, respectively as illustrated in Figures 3.

Although, no significant variation observed between the coastal (*M. nodiflorum* and *M. crystallinum*) and desert (*M. forsskaolii*) samples, the growth of root revealed significant variation (Figure 3). These variations could be attributed to the habitat effects. The allelochemicals may have phytotoxicity or stimulation depend on the nature and concentration of the compound, abiotic and biotic soil factors, the assay species, and physical/climatic factors (Abd-ElGawad *et al.*, 2020). Similar results were reported by Abd-ElGawad *et al.* (2019), Salem *et al.* (2016) and Alzuaibr *et al.* (2020) on some xerohalophytes plants.

From these results, the allelopathic effect of *Mesembryanthemum* species could be attributed to the bioactive components that act in a synergistic manner or to compounds which regulate one another such as flavonoids, phenolics, tannins and alkaloids. The allelopathic activity of the coastal (*M. nodiflorum* and *M. crystallinum*) may be due to the high content of carbohydrates, protein, antioxidants enzymes, tannins, triterpenes, alkaloids and flavonoids (van der Watt & Pretorius, 2001; Bouftira *et al.*, 2008; Doudach *et al.*, 2013). Phenolic compounds are a class of the most important and common plant allelochemicals in the agro-ecosystem that could diffuse into the rhizosphere soil and inhibit germination and growth of the plants (Inderjit, 1996; Li *et al.*, 2010).

On the other hand, the allelopathic activity of the inland (*M. forsskaolii*) could be ascribed to the presence of bioactive allelochemicals like tannins, Rutin, Apigenin, Apigenin-7-O-glucoside, Kaempferol-3-O-glucoside, Isorhamnetin-3-O- β -glucopyranoside (Moawad *et al.*, 2016), aliphatic compounds, triterpenoids, alkaloids, beta-sitosterol, campesterol (Bilel *et al.*, 2020). *M. forsskaolii* found to be rich in flavonols, tannins and phenolics (Lee *et al.*, 2011; Abdel-Farid *et al.*, 2016; Bilel *et al.*, 2020). Therefore, the reduction in the seedlings growth in this study might be related to their reduction in cell division that alters the ultrastructure of the cells and leads to alteration in the ions uptake, water-balance, phytohormonal balance, respiration and inactivate many enzymes (Li *et al.*, 2010; Fahmy *et al.*, 2012).

In reality, weeds (*Chenopodium murale*, *C. album*, *Phalaris minor*, *Amaranthus hybridus*, etc.) are more than just a nuisance; weeds have severe economic impacts and threaten the global food and natural ecosystem. Weeds compete with crops for moisture, nutrients, sunlight, and space (Capinera, 2014). *Chenopodium murale* is recorded as nuisance weed that compete with various crops such as barley (Al-Johani *et al.*, 2012), wheat (Majeed *et al.*, 2012), and chickpea (Batish *et al.*, 2007). In this context, the present results revealed that the ethanol extract from coastal samples showed higher phytotoxic activity against tested weed *C. murale*. Therefore, ethanol extract still considered as new promising resource and eco-friendly bioherbicide against harmful weeds like *C. murale*.

CONCLUSION

In conclusion, the studied *Mesembryanthemum* species could be used as sustainable sources of antioxidants and antimicrobials that could be added to foods for preservation, to fodders for raising the immunity and for protection of feeded animals against diseases. These results indicated the probability of using such these potent extracts against antibiotic resistance bacteria like *P. aeruginosa*, *S. aureus* and *K. pneumoniae*. They could be used as source of phytotoxic extracts that possess a significant allelopathic activity against *C. murale* weed that impact threats on valuable economic crops.

AUTHOR'S CONTRIBUTION

All authors contributed equality in carrying out the research study and the development of this paper.

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