Research Article

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Anti-inflammatory, antipyretic, analgesic, and antioxidant activities of *Haloxylon salicornicum* aqueous fraction

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Abstract: The medicinal plant *Haloxylon salicornicum* is utilized for therapeutic purposes. We previously reported the antioxidant potential of hexane fraction and methanol extracts of the same species. However, since these solvents could be clinically toxic, the current findings investigated the pharmacological effects of the water fraction. The pain relieving, antioxidant, anti-inflammatory, and antipyretic potential of *H. salicornicum* water extract (HEW) were studied at two concentrations (250 and 500 mg/kg) in rodents. The carrageenan stimulated rat paw edema assay was exercised to assess anti inflammatory potential in rats; yeast-stimulated hyperthermia was utilized to test antipyretic activity in mice; analgesic properties were assessed based on acetic acid-induced writhing, tail

flicking, and hot-plate test; and antioxidant potential was examined with the 2,2-diphenyl-1-picrylhydrazyl assay. We found that 500 mg/kg HEW inhibited edema by 44.03%. Yeast-induced hyperthermia in mice was reduced by 250 and 500 mg/kg HEW after 30, 60, and 120 min with significant level of ($P < 0.001$) compared to rectal temperature of yeast administered group. The high dose of HEW (500 mg/kg) improved the reaction time of mice in the hot-plate test from 6.66 ± 0.33 to 11.33 ± 0.49 s after 120 min. In the acetic acid-stimulated writhing test, 250 and 500 mg/kg HEW decreased writhing by 32.71% and 51.40%, respectively, after 20 min. HEW also showed antioxidant effects. These results demonstrate that HEW is bioactive and has therapeutic potential for treating a variety of ailments.

Keywords: *Haloxylon salicornicum*, Anti-inflammatory, Antipyretic, Analgesic, Saudi Arabia

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1 Introduction

Plant-based medicines are extensively utilized around the world; it is estimated that over three-quarters of the global inhabitants cannot meet the expense of pharmaceutical drugs and depend on plant-based conventional treatment for different diseases, which can be useful in the case of certain diseases that do not require prolonged treatment. In particular, rural communities in developing countries use herbal medicines based on their strong beliefs and limited access to allopathic therapies [1,2]. *Haloxylon salicornicum* is a shrub or under shrub of the Amaranthaceae family that is found in Pakistan, Bahrain, Iraq, Palestine, Iran, UAE, Kuwait, Egypt, Kingdom of Saudi Arabia, Jordan, Afghanistan and Qatar [3, 4]. Antiseptic, antidiabetic, and anti-inflammatory activities have been attributed to *H. salicornicum* [5]; glycosides, alkaloids, tannins, flavonoids, saponins and triterpenoids, are presumed

to be main source of its antioxidant, hepatoprotective, pain relieving and anti-inflammatory potential. The C_2H_5OH extract of this plant demonstrated significant antimicrobial potential against numerous fungi species including *Penicillium chrysogenum*, *Aspergillus fumigatus*, *Aspergillus flavus*, *Candida tropicalis*, and as well as bacterial stains such as *Staphylococcus aureus*, *Sarcina ventriculi*, *Candida albicans*, *Micrococcus luteus*, *Bacillus subtilis*, *Salmonella typhi* etc [6, 7]. We previously reported that both the methanol and hexane extracts (HEM and HEH, respectively) of *H. salicornicum* had hepatoprotective effects at concentrations of 250 and 500 mg/kg, whereas 250 μ g/ml HEH and HEM inhibited the formation of 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical by 35.65%, and 38.16% respectively [8]. Though, methanol and hexane are potentially toxic solvents and for this reason, water extracts of *H. salicornicum* would be preferable for medicinal use. To this end, the current study weigh upped the antipyretic, anti-inflammatory, analgesic, and antioxidant potential of the water fraction of *H. salicornicum* (HEW) in rodents.

2 Material and methods

2.1 Plant material

H. salicornicum was assembled from Wadi Hafr Al-Batin, Saudi Arabia on February 24, 2016. Taxonomic identification was performed with the assistance of Dr. Syed Riafyatullah (Plant taxonomist at King Saud University). The specimen was stored in the herbarium of the College of Pharmacy, King Saud University, Riyadh, Saudi Arabia (voucher number SY270).

2.2 Plant extraction preparation

Plant material was air-dried under shade, then crushed and soaked in methanol for seven days. It is then filtered and the methanol was evaporated *in vacuo* (Buchi, Flawil, Switzerland). The greenish methanol crude extract (33.3 g) was dissolved in distilled water and with the help of separating funnel it is partitioned between solvents with increasing order of polarity as *n*-hexane (7.1 ± 0.03 g), chloroform (3.4 ± 0.03 g), ethyl acetate (2.3 ± 0.03 g), *n*-butanol (10.6 ± 0.03 g), and water (9.9 ± 0.03 g).

2.3 Reagents and Animals

All reagents/chemicals utilized were of analytical grade. Wistar rats (170–210 g) and male albino mice (18–24 g, Swiss) were utilized in testing. The animals were retained at $22^\circ C \pm 2^\circ C$ in a room on a 12:12-h light/dark cycle with open entrance to water and food (standard pellet diet). The animals were kept and familiarized to the room conditions for at least 1 h prior to experimentations. Every animal was utilized just one time. Mice were utilized to assess the analgesic and antipyretic properties whereas the rats were utilized to investigate anti-inflammatory activity of the HEW.

2.4 Acute toxicity test

The toxicity of *H. salicornicum* (HEW) was evaluated in rats and mice at doses of 150, 350, 550, and 750 mg/kg. The animals were continuously monitored for sign of indications of mortality and toxicity [9].

2.5 Anti-inflammatory activity

Inflammation in rats paw was stimulated as previously described [10]. Briefly, 0.05 ml/kg of 1% of sodium salt of carrageenan was subcutaneously infused, with the help of subplantar aponeurosis, into the right hind paw of all experimental rats. Rats were distributed in 4 groups ($n = 6$ each). Rates were classified and treated as shown in Table-1. Plethysmometer (Apelex, Massy, France) was utilized to measured Paw volume. Anti-inflammatory activity was measured with formula: Anti-inflammatory potential (% inhibition) = $(1 - D/C) \times 100$

Where D is the % difference in paw volume after HEW injection and C is the % difference in paw volume in the control group.

2.6 Antipyretic activity

Hyperthermia was persuaded in mice by subcutaneous injection of 20 ml/kg body weight of a twenty percent brewer's yeast into the back near the nape of the neck [11], and then mice were classified and treated as shown in Table-2. The mice were fasted for the duration of the experiment, with open approach to water. After treatment body temperature of mice were noted at different interval of time i.e. 30, 60, and 120 min.

Table 1: The classification/treatment of rats Groups.

S. No	Groups (n=6)	Treatment	Given doses
1	Group I	Carrageenan	0.05 ml/kg
2	Group II	HEW (Orally)	250 mg/kg
3	Group III	HEW (Orally)	500 mg/kg
4	Group IV (Positive Control)	Phenylbutazone	100 mg/kg

Table 2: The classification/treatment of mice Groups.

S. No	Groups (n=6)	Treatment	Given doses
1	Group II	HEW (Orally)	250 mg/kg
2	Group II	HEW (Orally)	500 mg/kg
3	Group III (Positive Control)	indomethacin	4 mg/kg

2.7 Analgesic activity (hot plate test)

Response latency to pain caused by heat was assessed with the hot plate test [11]. A glass cylinder of twenty-four centimeter diameter was located on a hot plate with the temperature set to $55^{\circ}\text{C} \pm 1^{\circ}\text{C}$. The mice were separated into three groups ($n = 6$); two were given 250 and 500 mg/kg HEW and one received 4 mg/kg indomethacin by oral administration. Each mouse was set in the glass chamber on the hot plate and the time until a response was elicited (like claspings of forepaws/jumping) was noted before and 30, 60, and 120 min after treatment.

2.8 Analgesic activity (acetic acid-induced writhing test)

The pain relieving effect of HEW was assessed with the acetic acid induced writhing test [12]. Animals were orally treated with two doses (500, 250) mg/kg of HEW or four mg/kg of indomethacin, thirty min earlier to IP injection of 0.7% (v/v) acetic acid. The animals were scrutinized for writhing—characterized as “constriction and elongation of the abdomen” and “hind limbs extension” [13] for fifteen min.

2.9 Antinociceptive activity (tail flicking test)

Pain relieving effect of HEW at two doses of (500, 250) mg/kg in animals was assessed by means of a tail flick apparatus (Tail Flick Model DS 20 Sorrel; Apelex, Bagneux, France). Each animal was set in a restraining device two min earlier than treatment, and baseline response time to an intensity-controlled light beam focused on the distal one-third of the tail was recorded. HEW doses or standard drug was orally administered straight away subsequent to this step and 25 min later post-drug response time was considered. A 10-s cutoff time for the light beam was exercised in turn to prevent tissue damage [14].

3 GC-MS analysis of HEW

A Perkin Elmer model Clarus 600 T combined with single quadrapole mass spectrometer was utilized for Gas chromatography–mass spectrometry examination. Required time was twenty two minutes for one sample analysis. The temperature was kept primarily to 40°C (for 2 min), was raised to 150°C (for 2 min) 280°C (2 min), then increased further to 280°C at $15^{\circ}\text{C min}^{-1}$ (held for 2 min). The MS ion source temperature was 220°C and inlet line temperature at 240°C . The scan range was set at 40 to 600 mass ranges at 70 eV electron energy and the solvent delay of 4 min. At last, unidentified constituents were identified from online library

3.1 Antioxidant activity

The antioxidant activities of the HEW at different concentrations (100, 150, 200, and 250 $\mu\text{g/ml}$) were evaluated with the DPPH assay. Newly ready DPPH (purple-colored) solution changed into yellowish-colored after incubation for thirty minutes with test samples at room temperature, and the color change was recorded at 514 nm with a spectrophotometer (UVmini-1240; Shimadzu, Kyoto, Japan). The below equation was utilized to calculate antioxidant activity [8].

$$\text{Antioxidant activity (\%)} = (\text{AC} - \text{AS}) / (\text{AC}) \times 100$$

Where; AC= absorbance of control, AS= absorbance of sample

Table 3: Effect of HEW fraction on carrageenan-induced paw edema in albino rats.

Group (n = 6)	Dose (mg/kg)	Before carrageenan	3 h After carrageenan	Net	% Inhibition
Carrageenan		1.04 ± 0.02	1.59 ± 0.01	0.55 ± 0.01	
HEW	250	0.99 ± 0.03	1.45 ± 0.01	0.46 ± 0.02*	16.46
HEW	500	1.01 ± 0.04	1.352 ± 0.02	0.31 ± 0.01***	44.03
Phenylbutazone	100	1.03 ± 0.03	1.22 ± 0.02	0.19 ± 0.01***	65.86

Values represent mean ± SEM. *P<0.05, ***P<0.001 (analysis of variance followed by Dunnett's multiple comparisons test).
HEW, *Haloxylon salicornicum* water extract.

Table 4: Effect of HEW on yeast-induced hyperthermia in mice.

Treatment (n = 6)	Dose (mg/kg)	Baseline rectal temperature (°C)	Rectal temperature (°C) after administration of 20 ml/kg of 20% yeast	Post-drug rectal temperature (°C)		
				30 min	60 min	120 min
HEW	250	35.50 ± 0.12	38.88 ± 0.13***	38.01 ± 0.17**	37.76 ± 0.11***	37.66 ± 0.11***
HEW	500	35.63 ± 0.15	38.95 ± 0.08***	37.61 ± 0.11***	37.16 ± 0.08***	37.00 ± 0.05***
Indomethacin	4	35.78 ± 0.13	38.85 ± 0.16***	37.10 ± 0.09***	36.46 ± 0.16***	36.28 ± 0.12***

Values represent mean ± SEM. **P < 0.01, ***P < 0.001 (analysis of variance followed by Dunnett's multiple comparisons test).
HEW, *Haloxylon salicornicum* water extract.

3.2 Statistical analysis

Data are recorded as mean ± standard error of two replicates. Differences among groups were calculated by examination of variance chased by Dunnett's multiple comparisons test.

4 Results and Discussion

4.1 Acute toxicity

HEW at concentrations of 150, 350, 550, and 750 mg/kg had no adverse effects and did not cause mortality during the experimental time.

4.2 Anti-inflammatory potential

Carrageenan-stimulated paw edema persisted for 3 h post-injection (Table 3). Treatment with HEW significantly reduced volume of paw in a dose-dependent manner (33.23% and 65.86% reduction at 250 and 500 mg/kg, respectively, contrasted to the standard drug phenylbutazone.

4.3 Antipyretic activity

Yeast-induced hyperthermia in mice was decreased by 250 and 500 mg/kg HEW at 30, 60, and 120 minute subsequent to the treatment with significant level (P < 0.001). The baseline rectal temperature of 35.50°C–35.63°C increased to 38.88°C and 38.95°C after yeast injection, but returned to 37.00°C ± 0.05°C at 120 min after administration of 500 mg/kg HEW, which was comparable to the temperature achieved by the standard drug indomethacin (36.28°C ± 0.12°C) (Table 4).

4.4 Analgesic activity (hot-plate reaction time)

HEW showed significant, dose-dependent analgesic activity: a concentration of 500 mg/kg stretched the response time of mice from 6.66 ± 0.33 s to 11.33 ± 0.49 s at 120 min post-treatment (P < 0.001), which was comparable to the results obtained with indomethacin (13.66 ± 0.42 s) (Table 5).

Figure 1: GC-MS analysis of HEW fraction.

Table 5: Effect of HEW on hot plate reaction time in mice.

Treatment (n = 6)	Dose (mg/kg)	Pre-drug reaction time (s)	Post-drug reaction time (s)		
			30 min	60 min	120 min
HEW	250	7.16 ± 0.30	8.66 ± 0.42*	8.33 ± 0.33*	8.83 ± 0.30**
HEW	500	6.66 ± 0.33	10.00 ± 0.36***	10.83 ± 0.40***	11.33 ± 0.49***
Indomethacin	4	7.11 ± 0.36	11.16 ± 0.40***	13.83 ± 0.30***	13.66 ± 0.42***

Values represent mean ± SEM. *P < 0.05, **P < 0.01, ***P < 0.001 (analysis of variance).

HEW, *Haloxylon salicornicum* water extract.

Table 6: Effect of HEW on acetic acid-induced writhing in mice.

Treatment (n = 6)	Dose (mg/kg)	Writhing number in 20 min	% Inhibition
Acetic acid		35.66 ± 0.66	
HEW	250	24.00 ± 0.57***	32.71
HEW	500	17.33 ± 0.84***	51.40
Indomethacin	4	6.50 ± 0.42***	81.77

Values represent mean ± SEM. ***P < 0.001 (analysis of variance followed by Dunnett's multiple comparisons test).

HEW, *Haloxylon salicornicum* water extract.

4.5 Analgesic activity (acetic acid-induced writhing)

The quantity of writhings in mice at baseline was 35.66 ± 0.66. At 20 min after administration of 250 and 500 mg/

kg HEW, the number of writhings reduced to 24.00 ± 0.57 (32.71% inhibition) and 17.33 ± 0.84 (51.40% inhibition), respectively (Table 6). By comparison, indomethacin treatment resulted in 81.77% inhibition.

4.6 Antinociceptive activity (tail flick in mice)

HEW increased the time until tail flicking in a dose-reliant mode (P < 0.001) after 30, 60, and 120 min as compared to indomethacin (Table 7).

4.7 GC-MS Analysis of HEW

Different phyto-constituents in HEW fraction, analyzed with help of GC-MS analysis are given in Figure 1 and Table-8. Results showed that the highest constituents in

Table 7: Effect of HEW on tail flicking in mice.

Treatment (n = 6)	Dose (mg/kg)	Pre-drug reaction time (s)	Post-drug reaction time (s)		
			30 min	60 min	120 min
HEW	250	4.66 ± 0.33	5.16 ± 0.30*	5.83 ± 0.30*	6.00 ± 0.036*
HEW	500	4.83 ± 0.30	6.16 ± 0.30*	6.50 ± 0.34**	7.33 ± 0.33***
Indomethacin	4	3.83 ± 0.30	7.50 ± 0.22***	9.16 ± 0.030***	10.16 ± 0.0***

Values represent mean ± SEM. *P < 0.05, **P < 0.01, ***P < 0.001 (analysis of variance followed by Dunnett's multiple comparisons test). HEW, *Haloxylon salicornicum* water extract.

Table 8: Phyto-constituents analysis of HEW fraction.

S.No	Name	RT	Area %	Area
1	1-(2-Furanyl)-Ethanone	6.02	0.750	136662
2	N-Ethyl-N-[(1-Methyl)-Ethanamine	6.19	1.170	213633
3	2-Hydroxy-2-Cyclopenten-1-one	6.40	0.470	85649
4	5-Methyl-2-Furancarboxaldehyde	6.67	1.160	212539
5	5-Butoxy-2-pentene	6.81	55.510	10162445
6	1,2,3-PROPANETRIOL	7.55	6.600	1209012
7	2,3-Dihydro-3,5-dihydroxy-6-methyl-4h-pyran-4-one	8.87	14.640	2679449
8	Heptanal	9.20	1.040	189782
9	Nonanal	10.75	0.740	135371
10	D-Allose	13.56	10.080	1845674
11	Octanal	14.27	2.580	472923
12	N-Carboxyhyd-yl-pyrrolid-2-one	14.42	0.110	19675
13	9-Octadecenoic acid	14.90	0.600	109198
14	2-[1,2-Dihydroxyethyl]-9-[.Beta.-D-Ribofuranosyl]Hypoxanthine	14.93	2.450	448796
15	Octanal	15.10	0.200	36650
16	2-(Aminoxy)-Propanoic acid	15.48	0.500	91244
17	Oxalic acid	16.34	0.420	77600
18	4-Hydroxy-.Delta.-8-p-menthen-3-one	21.31	0.980	179514

HEW by area was 5-Butoxy-2-pentene (55.510%), Followed by 2,3-Dihydro-3,5-dihydroxy-6-methyl-4h-pyran-4-one (14.640%) and D-Allose (10.080%). Bioactive compound oxalic acid also found.

4.8 Antioxidant activity

HEW at concentrations of 100–250 µg/ml reduced DPPH formation, with the greatest inhibition (26.69%) observed at 200 µg/ml. It is given in Table 9.

H. salicornicum is known in folk medicine for its antiseptic and anti-inflammatory properties; traditional healers use the decoction to treat intestinal ulcers [15]. However, no studies to date have investigated the antipyretic, anti-inflammatory, analgesic, and antioxidant potentials.

Stigmasterol, Methylnicotinate, β -sitosterol, ursolic acid, phenethylamine, quercetin, esculetin, xanthoxol, β -amyrin, Lupeol, isooxyimperatorin, umbelliferone, scopolin, scopoletin, Kaempferol,

Table 9: Antioxidant activities of HEW, *H. salicornicum* water extract fraction.

	HEW concentration			
	100 (µg/ml)	150 (µg/ml)	200 (µg/ml)	250 (µg/ml)
Activity	26.37%	25.81%	26.69%	23.50%

N-methyltyramine, tyramine, oxedrine, halosaline, haloxine, aldtripiperidine, anabasine piperidine and Betaine chloride are different bio-active phytoconstituents reported from different extract and fractions of *H. salicornicum* [16, 17]

Literature revived showed that the acute phase of wound is because of main inflammatory mediators (bradykinin, serotonin, histamine and prostaglandin) are discharged in order to support vascular permeability and vasodilatation as well as edema [18]. The carrageenan-stimulated paw edema is measured as a representation of the acute stage of inflammation which is far and wide exercised for finding and exploration of active drug used for anti-inflammatory potential [19]. Particularly, this model is acknowledged to be insightful to cyclooxygenase inhibitors and has been utilized to weigh up the outcome of NSAIDs, which grips the inhibition of synthesis of PGs [20]. The primary stage is reasoned by the discharge of bradykinin, serotonin and histamine, and followed by the discharge of prostaglandins (PGs) at the third hour and remain for around six h subsequent to carrageenan injection [21]. The last stage emerges to be highly exciting in which the maximal vascular reaction arises with the migration of leukocyte to the area inflamed [22]. We found here that a dose level of 250 mg/kg b.w HEW suppressed inflammation (16.46%), reducing paw edema in rats. These findings suggesting anti-inflammatory potential of HEW by blocking the inflammatory mediators of the acute stage of inflammation. The very significant inhibitory effect (44.03% at dose level of 500 mg/kg) on carrageenan-stimulated paw edema at the third h recommends that these fractions may engage in jamming of the prostaglandins (PGs) synthesis. Fever stimulated by yeast is identified pathogenic fever. Its etiology consist of creation of prostaglandins, mostly PGE₂ emerges to be a concluding mechanism accountable for fever causing stimulated by numerous pyrogens [22]. Nearly all of the Non-steroidal anti-inflammatory drugs (NSAIDS) illustrate the antipyretic effect by blocking the prostaglandin synthesis. It is thus recommended that the antipyretic potential of HEW take places in a parallel manner as standard drug indomethacin. Two special analgesic assays were used with the aim of finding central analgesic (tail clip and hot plate) and peripheral effect

(acetic acid-induced writhing protocol). The acetic acid persuaded writhing is a visceral pain model that has been linked with discharge of bradykinins, cyclooxygenase, arachidonic acid and (PGE₂ and PGF_{2α}), have an important role in nociceptive mechanisms [23]. In our present study considerable pain relieving potential in the hot plate and the tail flik models expose that the analgesic activity of HEW is of the type generated by non-narcotic analgesics. Previous studies have reported that flavonoids, terpenoids, saponins, tannins, and alkaloids are present in *H. salicornicum* and that methanol and ethanol extracts of this plant have potent anti-inflammatory possessions even at a dose of 400 mg/kg [24]. Phytochemicals such as tannins, saponins, flavonoids, and alkaloids are bioactive compounds that have an extensive range of beneficial pharmacological effects like; antimicrobial, antihypertensive, antioxidant, anti-inflammatory, anticancer, and anti-diabetic activities, in addition to alleviating hypercholesterolemia [25–27]. A phytochemical analysis has shown that *H. salicornicum* contains the alkaloids methylisosalsoline, salsolidine, sosalsolidine, dehydrosalsolidine, N-methyltyramine, oxedrine, tyramine, carnegine, halosaline, smipine, haloxine, piperidine, tryptamine, scopolin, nicotine, haloxynine, hordenine, aldtripiperidine, anabasine, and dipterine as well as coumarins such as scopoletin, umbelliferone, xanthotoxol, isooxyimperatorin, and esculetin. Plants generally contain saponins, volatile oils, tannins, cardiac glycosides, sterols and anthraquinones [28]; phytochemicals such as tannins, saponins, and lignans are known for their antioxidant and other biological properties [29]. The isolation and characterization of bioactive compounds in *H. salicornicum* is essential for the improvement of novel drugs.

5 Conclusions

The water fraction of *H. salicornicum* demonstrates bioactivity including anti-inflammatory, antipyretic, analgesic, and antioxidant activities, without significant toxicity. Thus, HEW is safe for therapeutic applications and may be effective in the treatment of a variety of ailments.

Authors' contributions: RU performed experimental work. MSA and ASA supervised the work AAS composed the MS AAN, HMM collect the plant and ground it. SA revised the MS.

Ethics approval: Animals were treated and kept as per the recommendations of the “Guide for the Care and Use of Laboratory Animals”. The study permitted by ethical committee of College of Pharmacy King Saud University (Dated 06/01/2014, No. 05763-891).

Conflict of interests: The contributor's of this MS have declared no conflict of interests.

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References

- [1] Tariq A., Adnan M., Iqbal A., Sadia S., Fan Y., Nazar A., Mussarat S., Ahmad M., Olatunji O.A., Begum S., Mazari P., Ambreen B., Khan S.N., Ullah R., Khan A.L., Ethnopharmacology and toxicology of Pakistani medicinal plants used to treat gynecological complaints and sexually transmitted infections. *South African Journal of Botany*, 2018, 114, 132–149.
- [2] Aziz M.A., Adnan M., Khan A.H., Shahat A.A., Al-Said M.S., Ullah R., Traditional uses of medicinal plants practiced by the indigenous communities at Mohmand Agency, FATA, Pakistan. *Journal of Ethnobiology and Ethnomedicine*, 2018, 14, 1–16.
- [3] Phondani P.C., Bhatt A., Elsarrag E., Horr Y.A., Ethnobotanical magnitude towards sustainable utilization of wild foliage in Arabian Desert. *Journal of Traditional and Complementary Medicine*, 2016, 6(3), 209–218.
- [4] El-Wahab R.H., Al-Rashed A.R., Al-Hamad Y., Conservation condition of *Haloxylon salicornicum* (Moq.) Bunge ex Boiss. In degraded desert habitats of northern Kuwait. *Int. J. Curr. Microbiol. App. Sci*, 2014, 3(10), 310–325.
- [5] Ajabnoor M.A., Al-Yahya M.A., Tariq M., Jayyab A.A., Antidiabetic activity of *Hammada salicornicum*, *Fitoterapia*, 1984, 55, 107–109.
- [6] Awaad A.S., Zain M.E., Al Othman M.R., Al-Dosary S.K., Phytochemical analysis and antimicrobial activity of medicinal plants against pathogenic microorganisms. *Life Science Journal*, 2014, 11, 350–354.
- [7] Eman A.A., Phytochemical screening on different plant parts of some succulent plants of Egypt. *New York Science Journal*, 2011, 4, 15–18.
- [8] Ullah R., Alsaid M.S., Shahat A.A., Naser A.A., Al-Mishari A.A., Adnan M., Tariq A., Antioxidant and hepatoprotective effects of methanolic extracts of *Zilla spinosa* and *Hammada elegans* against carbon tetrachloride-induced hepatotoxicity in rats. *Open Chemistry*, 2018, 16, 133–140.
- [9] Organization for Economic Cooperation and Development (OECD). OECD Guidelines for Testing of Chemicals, Acute Oral Toxicity, No. 401, 1993.
- [10] Winter C.A., Risley E.A., Nuss G.W., Carrageenan-induced oedema in hind paw of the rats as an assay for anti-inflammatory drugs. *Proceedings of the Society for Experimental Biology and Medicine*, 1962, 111, 544–547.
- [11] Mothana R.A., Al-Said M.S., Al-Rehaily A.J., Thabet T.M., Awad N.A., Lalk M., Lindequist U., Anti-inflammatory, antinociceptive, antipyretic and antioxidant activities and phenolic constituents from *Loranthus regularis* Steud. ex Sprague. *Food Chemistry*, 2012, 130, 344–349.
- [12] Amor E.C., Quanco J.P., Perez G.G., Analgesic activity of extracts of *Kyllinga monocephala*. *Pharmaceutical Biology*, 2009, 47, 624–627.
- [13] Miranda H.F., Sierralta F., Pinardi G., An isobolographic analysis of the adrenergic modulation of diclofenac antinociception. *Anesthesia & Analgesia*, 2001, 93, 430–435.
- [14] Al-Rehaily A.J., Al-Said M.S., Al-Yahya M.A., Mossa J.S., Rafatullah S., Ethnopharmacological studies on allspice (*Pimenta dioica*) in laboratory animals. *Pharmaceutical Biology*, 2002, 40, 200–205.
- [15] Shafi M.S., Ashraf M.Y., Sarwar G., Wild medicinal plants of Cholistan area of Pakistan. *Pakistan Journal of Biological Sciences*, 2001, 4, 112–116.
- [16] Singh J.P., Rathore V.S., Roy M.M., Notes about *Haloxylon salicornicum* (Moq.) Bunge ex Boiss., a promising shrub for arid regions. *Genetic resources and crop evolution*, 2015, 62(3), 451–463.
- [17] Kaur P., Bains N.S., Extraction of flavonoids from in vitro and in vivo tissue culture of some important halophytes of western Rajasthan. *Int J Pharm Tech Res*, 2012, 4(3), 1167–1171.
- [18] Carlson R.P., Chang J., Lewis A.J., Modulation of mouse ear edema by cyclooxygenase and lipoxygenase inhibitors and other pharmacologic agents. *Agents and Actions*, 1985, 17(2), 197–204.
- [19] Winter C.A., Risley E.A., Nuss G.W., Carrageenin-induced edema in hind paw of the rat as an assay for anti-inflammatory drugs. *Proceedings of the Society for Experimental Biology and Medicine*, 1962, 11, 544–547.
- [20] Rosa M.D., Willoughby D.A., Screens for anti-inflammatory drugs, *Journal of Pharmacy and Pharmacology*, 1971, 23(4), 297–298.
- [21] Vinegar R., Schreiber W., Hugo R., Biphasic development of carrageenin edema in rats. *Journal of Pharmacology and Experimental Therapeutics*, 1969, 166(1), 96–103.
- [22] Tomazetti J., Avila D.S., Ferreira A.P.O., Martins J.S., Souza F.R., Royer C., Rubin M.A., Oliveira M.R., Bonacorso H.G., Martins M.A.P., Zanatta N., Baker yeast-induced fever in young rats: Characterization and validation of an animal model for antipyretics screening. *J Neurosci Methods*, 2005, 147, 29–35.
- [23] Baird-Lambert J., Jamieson D.D., Possible mediators of the writhing response induced by acetic acid or phenylbenzoquinone in mice. *Clin Exp Pharmacol Physiol*, 1983, 10(1), 15–20.
- [24] Wasfi I.A., Bashir A.K., Abdalla A.A., Banna N.R., Tanira M.O.M., Antiinflammatory activity of some medicinal plants of the United Arab Emirates. *International Journal of Pharmacognosy*, 1995, 33, 124–128.
- [25] Karimi E., Jaafar H.Z., Ahmad S., Phytochemical analysis and antimicrobial activities of methanolic extracts of leaf, stem and root from different varieties of *Labisa pumila* Benth. *Molecules*, 2011, 16, 4438–4450.

- [26] Dash M., Patra J.K., Panda P.P., Phytochemical and antimicrobial screening of extracts of *Aquilaria agallocha* Roxb. African Journal of Biotechnology, 2008, 7, 3531–3534.
- [27] Kim S.K., Van Ta. Q., Potential beneficial effects of marine algal sterols on human health Advances in Food and Nutrition Research, 2011, 64, 191–198.
- [28] Ghazanfar S.A., Handbook of Arabia Medicinal Plants. CRC Press: Boca Raton, 1994, pp. 83–86.
- [29] Ahmad M., Eram S., Hepatoprotective studies on *Haloxylon salicornicum*: a plant from Cholistan desert. Pakistan Journal of Pharmaceutical Sciences, 2011, 24, 377–382.