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اسطنبول - تركيا

Physicochemical, phytochemical profiling and Biological activities of leaves extract of Bardy (*Typha domingensis* Pers.) from Al-Chibayish marshes in southern Iraq

Esam J. Al-Kalifawi[1], Yasamine J. Al-Azzawi[1], Khaled kat Al-Fartousi1[2]and Hind Mezher Musa[3]
[1]Biology Department, College of Education for Pure Science Ibn -Al- Haitham, University of Baghdad,
[2] Biology Department, [3]Department of Pathological analysis, College of Science, University of Dhi Qar Iraq
[1]aesam365@Yahoo.com

Abstract: This research was carried out in Al-Chibayish marshes in southern Iraq to support the inclusion of Iraqi marshes in the UNESCO's World Heritage List. This study was conducted to investigate Physicochemical, phytochemical profiling and Biological activities of the leaves extract of Bardy (*Typha domingensis* Pers.) collected from Al-Chibayish marshes in southern Iraq. Phytochemical screening of crude extracts of the leaves of the *Typha domingensis* Pers. reveals the presence of alkaloids, tannin, steroids, phenol, saponins, flavonoids in aqueous and methanolic extracts whereas carbohydrates, tannins, oils and fats were present in Petroleum ether and chloroform extract. In addition to this chloroform extract also contains flavonoids and phenols.

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The results showed that the leaves extract of Bardy (*Typha domingensis* Pers.) Had antimicrobial properties against and Gram-negative and Gram positive bacteria. The minimum inhibitory concentration (MIC) of the different extracts ranged from 0.78 mg/ml to 12.5 mg/ml. Haemostatic activity of leaves extract of Bardy (*Typha domingensis* Pers.) in Albino Rats was investigate. The results obtained indicate that methanolic leaf extract of *Typha domingensis* Pers. significantly decreased ($p < 0.05$) the bleeding time, prothrombin time and clotting time respectively in a dose dependent manner. In contrast, plasma fibrinogen concentration significantly increased ($p < 0.05$). The study concluded that methanolic leaf extract of Bardy (*Typha domingensis* Pers.) possesses antimicrobial and haemostatic activities.

Keywords: Physicochemical, Phytochemical, Bardy, Typha domingensis Pers., antimicrobial, haemostatic.

Introduction

Typha is a cosmopolitan genus of about eleven species of monocotyledonous plants in the family, Typhaceae, which has a distribution largely in the marshes in southern Iraq. The common name of the family is 'cattails', which defines the characteristic inflorescence of the genus. Cattails are familiar wetland plants in the world distributed in wet soil, marshes, swamps, and shallow fresh and brackish waters [1]. Typha species are monoecious and unisexual plants having wind-pollinated flowers, which develop in dense spikes. Male flowers form a narrow spike at the top of the stem, while the tiny female flowers form a dense spike just below the male spike [2]. In Turkish folk medicine, female flowers inflorescence of Typha species, without discriminating the species, are used externally to stop bleeding [3]. As well as for burns and wound healing [4]. Similar utilizations were also reported in the traditional medicines worldwide. For example, pollens of Typha species (Pollen Typhae) was attributed to possess the function of hemostasis and removing stasis, and was often prescribed in the treatment of bleedings of various origins such as nose bleeds, haematemesis, haematuria, uterine bleeding and dysmenorrhoea as well as haemostatic, desiccant and vulnerary in external injuries in Traditional Chinese Medicine [5, 6, 7, 8]. Phytochemical studies have found that Pollen Typhae mainly contains sterols, terpenoids, flavones and long chain hydrocarbons. In the pharmacological studies various activities were also attributed to pollens, i.e., cyclic adenosine monophospate (cAMP) inducing activity, cholesterol lowering effect, immunosuppressive and anticoagulation [5, 6, 8]. In spite of worldwide traditional recognition of Typha species in the management of wounds and burns, a reference survey revealed that wound healing potential of any of the species has not been evaluated so far. On the other hand, only the female flowers inflorescence has been used for wound management in Turkish folk medicine, however in Traditional Chinese Medicine documents only the pollens (Pollen Typhae) of the plant were defined as the efficient part. In The Prophetic Medicine, all parts of this plant such as rhizomes, stem, leaves, flowers, and seeds are used in diseases treatment. The Typha domingensis pers. also known as Cattail, is a member of the typhaceae family. These plants are herbaceous, rhizomatous perennial plants with long, slender green stalks topped with brown, fluffy, sausage-shaped flowering heads 15-40 dm tall. The spike is bright yellow-to-orange-brown. The basal leaves are 6-18 mm wide when fresh, 5-15 mm wide when dry. Typhaceae family is common in the warm temperature and tropical regions of the world always found in or near water, in marshes, ponds, lakes and depressions. Typha domingensis used externally for burns and wound healing, leaves are diuretic [9, 10]. The family Typhaceae have one genus and 10 to 15 species. The characteristic inflorescence gives the family the common name 'cat tails'. Individuals are tall and can reproduce clonally, by submerged rhizomes, forming dense stands. The leaves are diuretic and the pollen is astringent, desiccant, diuretic, haemostatic and vulnerary [11]. Pollen is used in the treatment of nose bleeds, haematemesis, haematuria, uterine bleeding, dysmenorrhoea, postpartum abdominal pain and

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gastralgia, scrofula and abscesses. It is also contraindicated for pregnant women [12]. The rootstock is astringent and diuretic [13].

He mentioned a lot about the use of Bardy in the treatment of many diseases in folk medicine and the Prophetic medicine. In Hadith 4414 in Sahih Muslim [14] mentioned the use of ashes of Bardy in stopping the bleeding of the Prophet Muhammad (may peace be upon him). It has been narrated on the authority of Abd-ul-'Aziz b. Abu Hazim, who learnt from his father (Abu Hazim). The latter heard it from Sahl b. Sa'd who was asked about the injury which the Messenger of Allah (may peace be upon him) got on the day of the Battle of Uhud. He said: The face of the Messenger of Allah (may peace be upon him) was injured, his front teeth were damaged and his helmet was crushed. Fatima, the daughter of the Messenger of Allah (may peace be upon him), was washing the blood (from his head), and 'Ali bin Abu Talib was pouring water on it from a shield. When Fatima saw that the bleeding had increased on account of (pouring) water (on the wound), she took a piece of Bardy mat and burnt it until it was reduced to ashes. She put the ashes on the wound and the bleeding stopped.

The peoples of Al-Ahwar (marshes) in the south of Iraq are uses ʻBardyʼ, the common name of Typhaceae family as folk medicin, the leaves are diuretic, haemostatic, and it is used in the treatment of bleeding, antiinflamintary and anticlotting. The pollen (Al-Kheret) is astringent, desiccant, diuretic, haemostatic and vulnerary. It is used in the treatment of nose bleeds, haematemesis, haematuria, uterine bleeding, dysmenorrhoea, postpartum abdominal pain and gastralgia, scrofula and abscesses. It is contraindicated for pregnant women. The pollen (Al-Kheret) and stem (Al-Akeed) are increased the male fertility therefore called Viagra of Al- Ahwar. The seed down is haemostatic. The rootstock is astringent and diuretic.

The aim of the present study was to investigate Physicochemical, phytochemical profiling and Biological activities of the different leaves extracts of Bardy (Typha domingensis Pers.) collected from Al-Chibayish marshes in southern Iraq.

Materials and Methods

2.1. Collection of plant materials and extraction

Aerial part (leaves) of Typha domingensis Pers. was collected in and around the Al-Chibayish marshes in southern Iraq figure 1, in the month of January and the plant was duly identified and authenticated in the Herbarium of the Department of Biology, College of Education for Pure Science Ibn -Al- Haitham, Baghdad University, Baghdad, Iraq.

The collected leaves were washed with running tap water and allowed to air dry. The plant materials were dried in shade for two to four weeks. Precaution was taken to avoid direct sun light otherwise it will destroy the active compounds of plant leaves. After drying, the plant leaves were grinded finely and stored in airtight container. The air dried leaf powders (50g) were successively extracted by soxhlet extraction with solvents of increasing polarity. The solvents include petroleum ether (60-80°C), chloroform, methanol and distilled water. The extracts were dried and stored in a sterile container for further use.

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Figure 1. Collection of Bardy form the Al-Chibayish marshes in southern Iraq.

2.2. Physicochemical analysis

The finely powdered leaves of Typha domingensis Pers. was subjected to various physicochemical studies for determination of ash value like total ash, acid insoluble ash and water soluble ash [15]. Extractive values like water soluble, methanol soluble,
chloroform soluble and petroleum ether soluble were determined.

2.3. Phytochemical screening of crude extracts

The phytochemical components of the Typha domingensis Pers. leaves were screened for using the standard method described by Harbone [16]. The components analyzed are alkaloids, proteins, glycosides tannin, steroids, phenol, saponins, flavonoids, carbohydrates, oils and fats.

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2.4. Collection of pathogens microbes

The pathogenic isolates collected from microbiology diagnosis laboratory, Al-Numan hospital, Baghdad, Iraq. Which included Escherichia coli, Proteus mirabilis, Klebsiella pneumonia, Streptococcus sp., Enterobacter cloacae, Bacillus cereus, Pseudomonas aeruginosa, Staphylococcus aureus and pathogenic yeasts Candida albicans, Candida glabrata, Candida parapsilosis and Candida tropicalis. These isolates was identified by standard biochemical tests and used API 20 C, API 20 E, API 50 CHB, API Staph kit and Vitek 2 compact system [17].

2.5. Standardization of microorganisms

200 ml of overnight cultures of each microorganisms was dispensed into 20 ml of sterilized nutrient broth and incubated at 37°C for 4-6 h to standardize the culture to 106 CFU/ml. A loopful of the standard cultures was used for the antimicrobial assay [18].

2.6. Screening for antibacterial activity

Antibacterial activities of all different extracts of Typha domingensis Pers. were determined by standard agar well diffusion assay [19]. MullereHinton Agar (MHA) plates were seeded with 18 h old culture of the isolates. Different extracts were dissolved in Dimethyl sulfoxide (DMSO) and made the final concentration of 50 mg/ml, from this 50 µl of different extracts were added into the sterile 6 mm diameter well. 1% Tween 80 and sterilized distilled water were used as negative controls while Ciprofloxacin antibiotic disc (5 µg, Oxoid) and fluconazole antifungal disc (25µg, Oxoid) were used as positive control and DMSO as negative control. A loopful each of the standardized culture of test organisms was streaked on the solidified medium and incubated for 24 h at 37°C. Antibacterial activity was assayed by measuring the diameter of the zone of inhibition formed around the well using standard (Hi-Media) scale. The experiment done in triplicate and the average values were calculated for antibacterial activity.

2.7. Determination of minimum inhibitory concentration

Minimum inhibitory concentration (MIC) was defined as the lowest concentration where no visible turbidity was observed in the test tubes. The concentrations were determined by the method described by Vollekova [20] with minor modification was employed. The MIC was determined for the microorganisms that showed maximum sensitivity to the test extracts.

In this method the broth dilution technique was used, where the leaf extract was prepared to the highest concentration of 25 mg/ml (stock concentration). By adding sterile distilled water serially diluted (two fold dilutions) using the nutrient broth and it is later inoculated with 0.2 ml standardized suspension of the test organisms. After 18 h of incubation at 37°C, the test tubes were observed for turbidity .The lowest concentration of the tube that did not show any visible growth can be considered as the minimum inhibitory concentration.

2.8 Animal model

Twenty wistar strain albino rats of both sexes weighing between 100 – 170 g were used. The rats were divided into four (4) groups of five (5) animals each. The rats were purchased from the College of Medical Sciences' animal house of the University of Nigeria, Nsukka. The rats were housed in wire mesh cages under standard conditions (temperature, 25 - 29°C, 12 h light and 12 h dark cycle) and allowed to acclimatize for 3 weeks. The rats were fed with standard pellets diet and water given ad libtum.

Experimental procedure

This study was carried out on 4 groups of rat; each group contained 5 rats and was placed in a different cage for proper identification. The control group was given (10 mg/kg) normal saline while test groups were given different concentrations (250, 500 and 750 mg/kg) of methanolic leaf extract of Typha domingensis Pers. intraperitoneally. The treatment regimens lasted for 2 weeks. All procedures

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involving the use of animals in this study complied to the guiding principles for research involving animals as recommended by the declaration of Helsinki and the Guiding principles in the care and use of animals [21]. Group 1 (Control), in this group, 5 rats were used. Each rat received 10 mg/kg of normal saline intraperitoneally for a period of 2 weeks. Group 2 (low dose), also, 5 rats were used in this group. Each rat received a low dose of methanolic leaf extract of Typha domingensis Pers. intraperitoneally (250 mg/kg) for a period of 2 weeks. Group 3 (medium dose), there were 5 rats used in this group. Each rat received a medium dose of methanolic leaf extract of Typha domingensis Pers. intraperitoneally (500 mg/kg) for a period of 2 weeks. Group 4 (high dose), there were 5 rats used in this group. Each rat received a high dose of methanolic leaf extract of Typha domingensis Pers. intraperitoneally (750 mg/kg) for a period of 2 weeks. At the end of treatment, the rats were anaesthetised with chloroform and blood samples were collected by cardiac puncture into sample vials containing sodium citrate in ratio 1:9 with the blood with aid of a 5 ml syringe. Only blood samples used for determination of clotting time were collected in anticoagulant-free vials.

Sample analysis

Determination of bleeding time

This was determined using a modified Duke method [22]. A skin puncture was made quickly using disposable lancet and the stopwatch was started as soon as bleeding started. The puncture was dabbed with filter paper every 15 s until the paper no longer stained red with blood. Bleeding time was then taken as the time when the blood stopped flowing from the puncture.

Determination of prothrombin time

Blood was collected into sample vials containing 3.2% sodium citrate (as specified in the prothrombin time (PT) test kit used) in the ratio 1:9 with the blood sample. The blood was then centrifuged at 1000 g for 15 min to obtain platelet poor plasma. Thromboplastin PT-S was placed in a water bath at 37°C; and 0.1 ml of test plasma
was also put into a test tube and placed in the water bath to prewarm to 37°C. A 0.2 ml of warmed thromboplastin PT-S was then forcibly added to the test plasma and the stopwatch was started. The tube was tilted repeated until a clot was formed and the time taken for clot to form was noted. This was repeated for all the blood samples (five in each group). Precaution was taken to perform test within 3 h of blood collection since the labile factor deteriorates quickly at room temperature.

Determination of clotting time

Blood was taken directly from the heart to avoid contamination with tissue thromboplastin (0.8 ml from each rat). A 0.2 ml of blood was then delivered into four glass test tubes that had previously been warmed and maintained at 37°C and the tubes immediately placed in a 37°C water bath to mimic the temperature of the internal environment. The stopwatch was started immediately the blood was delivered into the glass test tubes and the tubes were continually tilted at 40 s intervals (until blood in them stopped flowing when tilted at an angle of 90°), starting with the first, to see and note the time when the blood clotted. The clotting time was taken as the average of the times blood clotted in the four tubes.

Determination of fibrinogen concentration

Plasma fibrinogen concentration was determined as defined by the clot weight method of Ingram [23] though with modifications to accommodate the procedure for use of thrombin time (TT) test kit as given by the manufacturers of the kit. Blood was collected with the aid of plastic disposable syringes into sample vials containing 3.2% sodium citrate in the ratio 1:9 with blood. Blood plasma was obtained by centrifuging blood in a stopped vial at 1000 g for 10 min. 0.2 ml of the test plasma was put into a test tube and incubated in a water bath for 3 min at 37°C. 0.2 ml of thrombin time-reagent was added to test plasma, mixed and the clot formed harvested with a wooden applicator stick. The

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resulting clot was transferred into a tube containing acetone to dry and harden for about 10 min; the acetone was decanted and the clot placed on a filter paper for the remaining acetone to evaporate. The clot was then recovered and weighed. The process of fibrinogen concentration determination was completed within 3 h of blood collection. Thus, fibrinogen concentration of citrated plasma in mg/dl equals clot weight (mg) divided by plasma volume (dl).

Statistical analysis

All data were presented as mean ± SEM. The one way ANOVA was used to analyze the data, followed by a post-hoc test (LSD). The results were considered significant at p values of less than 0.05 [24].

Result

It is estimated that total ash value in leaves is 10.77%, acid insoluble ash and water soluble ash shows the value 4.86% and 3.18% respectively. The extractive value of methanol is more followed by aqueous, chloroform and petroleum ether with 20.17%, 6.88%, 4.55% and 2.15% respectively. Phytochemical screening of crude extracts of the aerial part of the Typha domingensis pers. reveals the presence of alkaloids, tannin, steroids, phenol, saponins, flavonoids in aqueous and methanolic extracts whereas carbohydrates, tannins, oils and fats were present in Petroleum ether and chloroform extract. In addition to this chloroform extract also contains flavonoids and phenols Table1.

Table 1: The phytochemical Analysis of *Typha domingensis* Pers. leaves extracts.

Phytochemicals	Aqueous Extract	Methanol Extract	Chloroform Extract	Petroleum ether extract
Alkaloids	+	+	-	-
Phenoles	+	+	+	-
Flavonoids	+	+	+	-
Tannins	+	+	+	+
Saponins	+	+	-	-
Steroids	+	+	-	-
Carbohydrates	-	-	+	+
Oils	-	-	+	+
Fats	-	-	+	+

The antimicrobial activity of different extracts against the test organisms with varying zones of inhibition ranging from 7 to 23 mm, Table 2 and figure 2 has revealed the antimicrobial potency of this plant. Methanolic extract showed highest zone of inhibition against *E. coli*, *Enterobacter cloacae*, *Candida albicans* and *Candida tropicalis* it was 23 mm followed by *Klebsiella pneumonia*, *Bacillus cereus*, *Candida glabrata* and *Candida parapsilosis* the inhibition zone was 21mm. the inhibition zone was 20 mm for *Proteus mirabilis*, *Streptococcus sp*., *Pseudomonas aeruginosa* and *Staphylococcus aureus*. The aqueous extract showed greater inhibition zone against *E. coli*, *Bacillus cereus*, *Staphylococcus aureus* and *Candida albicans* it was 15 mm, followed by *Proteus mirabilis*, *Klebsiella pneumonia*, *Streptococcus sp*. and *Candida tropicalis* the inhibition zone was 14 mm. whereas the lowest effect of aqueous extract was against *Enterobacter cloacae*, *Candida glabrata* and *Candida parapsilosis* it was 12 mm. Chloroform extract shows moderate inhibitory effect on these microbes. The highest effect was against *Klebsiella pneumonia*, *Candida albicans* and *Candida tropicalis* the inhibition zone was 12 mm. the lowest effect was against *Proteus mirabilis*, *Bacillus cereus* and *Candida parapsilosis* the inhibition zone was 7 mm. The Petroleum ether extract did not have any effect against the tested microbes. The result of positive control was highest effect of ciprofloxacin

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against bacteria and Fluconazole against pathogenic yeasts. The result of negative control show the Dimethyl sulfoxide (DMSO) has no effect against the tested microbes.

Table 2. Effect of different extracts of *Typha domingensis* Pers. leaves extracts on pathogenic bacterial and yeasts.

Pathogenic microbes	Inhibition zone mm*					
	Extracts				+ve con.	-ve con.
	AE	ME	CE	PE	AN1, AF1	DMSO
Escherichia coli	15	23	8	6	25	0
Proteus mirabilis	14	20	7	0	24	0
Klebsiella pneumonia	14	21	12	0	23	0
Streptococcus sp.	14	20	0	0	22	0
Enterobacter cloacae	12	23	6	0	22	0
Bacillus cereus	15	21	7	6	24	0
Pseudomonas aeruginosa	13	20	8	6	23	0
Staphylococcus aureus	15	20	8	0	22	0
Candida albicans	15	23	12	0	22	0
Candida glabrata	12	21	10	0	21	0
Candida parapsilosis	12	21	7	0	21	0
Candida tropicalis	14	23	12	0	22	0

AE=aqueous extract, ME=methanol extract, CE=chloroform extract, PE = Petroleum ether extract, mm*=millimetres, AN1=Ciprofloxacin CIF-5 µg /disc, AF= Fluconazole, FCA-25µg/ disc. DMSO= Dimethyl sulfoxide.

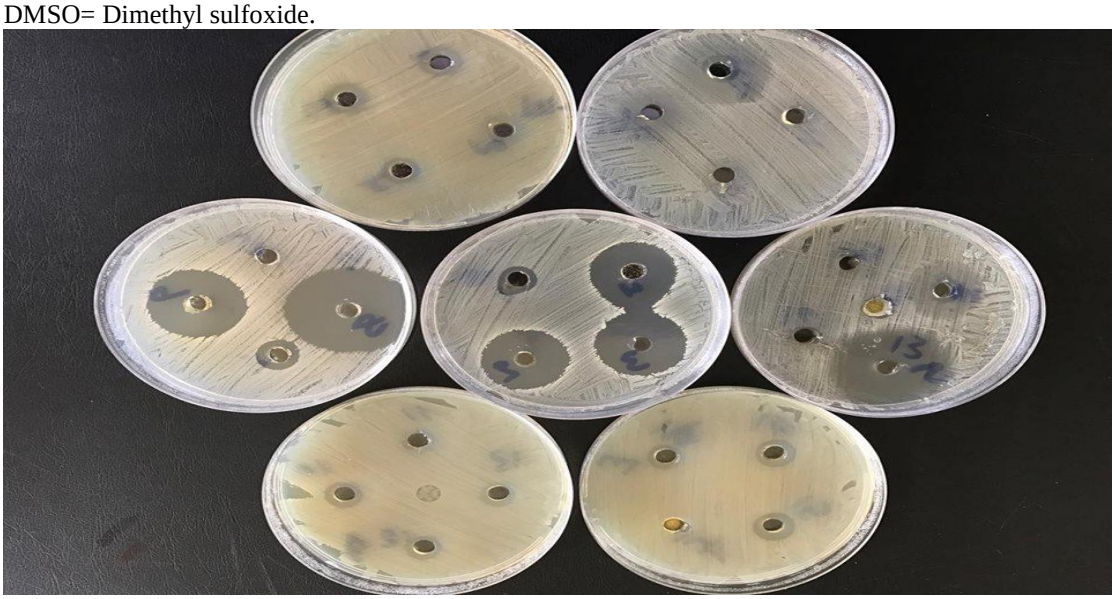

Figure 2. The antimicrobial activity of different extracts against the test organisms.

The results of (MIC) of *Typha domingensis* Pers. Leaves extracts on different microbes isolates shows methanol extract of *Typha domingensis* pers. exhibited the highest antimicrobial activity against

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bacteria and yeasts. Followed by aqueous extract while the chloroform extract was less effective. The MIC vales of methanol extract was 0.78mg/ml for *E. coli*, *E. cloacae*, *P.aeruginosa* and *C. albicans*. The MIC vales of aqueous extract was 1.5mg/ml for *E. coli*, P. *mirabilis*, *Streptococcus. sp*., *B. cereus*, *S. aureus* and *C. tropicalis*. The MIC vales of chloroform extract was 12.5mg/ml for all isolates except for *C. albicans* was 6.25mg/ml as shown in table 3.

Table 3. Minimum inhibitory concentration (MIC) of *Typha domingensis* Pers. Leaves extracts on different microbes isolates.

Microbes	Concentration (mg/ml)									
	EXT	0.098	0.195	0.39	0.78	1.563	3.125	6.25	12.50	25.00
E. coli	CE	+	+	+	+	+	+	+	*	-
	ME	+	+	+	*	-	-	-	-	-
	AE	+	+	+	+	*	-	-	-	-
P. mirabilis	CE	+	+	+	+	+	+	+	*	-
	ME	+	+	+	+	*	-	-	-	-
	AE	+	+	+	+	*	-	-	-	-
K. pneumonia	CE	+	+	+	+	+	+	*	-	-
	ME	+	+	+	+	*	-	-	-	-
	AE	+	+	+	+	*	-	-	-	-
Stre. sp.	CE	+	+	+	+	+	+	+	+	*
	ME	+	+	+	+	*	-	-	-	-
	AE	+	+	+	+	*	-	-	-	-
E. cloacae	CE	+	+	+	+	+	+	+	*	-
	ME	+	+	+	*	-	-	-	-	-
	AE	+	+	+	+	+	+	*	-	-
B. cereus	CE	+	+	+	+	+	+	+	*	-
	ME	+	+	+	+	*	-	-	-	-
	AE	+	+	+	+	*	-	-	-	-
P.aeruginosa	CE	+	+	+	+	+	+	+	*	-
	ME	+	+	+	*	-	-	-	-	-
	AE	+	+	+	+	+	*	-	-	-
S. aureus	CE	+	+	+	+	+	+	+	*	-
	ME	+	+	+	+	*	-	-	-	-
	AE	+	+	+	+	*	-	-	-	-
C. albicans	CE	+	+	+	+	+	+	*	-	-
	ME	+	+	+	*	-	-	-	-	-
	AE	+	+	+	+	+	*	-	-	-
C. glabrata	CE	+	+	+	+	+	+	+	*	-
	ME	+	+	+	+	*	-	-	-	-
	AE	+	+	+	+	+	+	*	-	-
C. parapsilosis	CE	+	+	+	+	+	+	+	*	-
	ME	+	+	+	+	*	-	-	-	-

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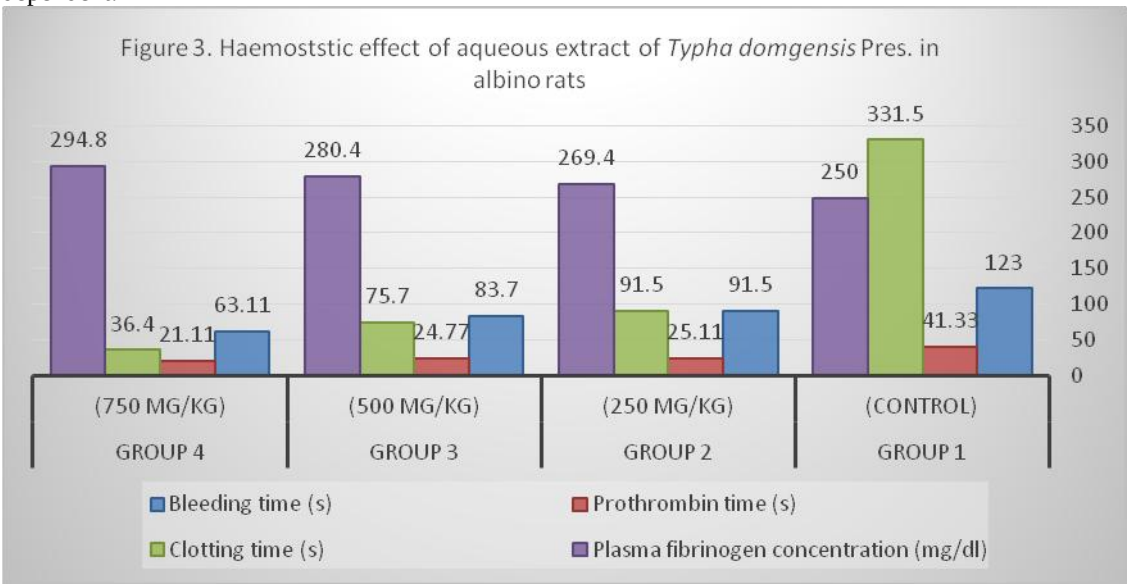
	AE	+	+	+	+	+	+	*	-	-
C. tropicalis	CE	+	+	+	+	+	+	*	-	-
	ME	+	+	+	*	-	-	-	-	-
	AE	+	+	+	+	*	-	-	-	-

CE = chloroform extract, ME = methanol extract, AE = aqueous extract + indicates turbidity is observed, - indicates turbidity is not observed, * represents the MIC value.

The extract decreased bleeding time in rats of the experimental group in comparison with the control group. The mean bleeding time in control group was 123.0 ± 16.0 s while those of group 2, 3 and 4 were 91.5 ± 7.5, 83.7 ± 17.5 and 63.11 ± 77 s respectively table 4. The analysis showed that the decrease was not significant in group 2 at $p > 0.05$ but significant in group 3 and 4 when compared independently with the control. The effect was most significant in the group administered the highest dose of the extract, group 4. The mean prothrombin time in the control group was 41.33 ± 5.5 s while those of group 2, 3 and 4 were 25.11 ± 37, 24.77 ± 4.4and 21.11 ± 3.6 s respectively as shown in Table 4. There was significant decrease ($p < 0.05$) of prothrombin time in the experimental groups compared with the control group. The decrease was dose dependent with most significant decrease in group 4. The mean clotting time in the control group was 331.5 ± 17.7 s while those of group 2, 3 and 4 were 91.5 ± 13.3, 75.7 ± 11.3and 36.4 ± 7.7 s respectively as shown in Table 4. There was also significant decrease ($p < 0.05$) of clotting time in the experimental groups compared with the control group. The decrease was also dose dependent. The mean plasma fibrinogen concentration in the control group was 250.0 ± 10.6 mg/dl while those of group 2, 3 and 4 were 269.4 ± 10.4, 280.4 ± 12.7 and 294.8 ± 7.0 mg/dl respectively as shown in figure 3. There was significant increase ($p < 0.05$) of plasma fibrinogen concentration in the experimental groups compared with the control group. The increase was dose dependent.



Discussion

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Al-Chibayish marshes (Ahwar) is located in the district of Al-Chibayish in the province of Dhi Qar. Dictator Saddam Hussein drained the Mesopotamian marshes in the 1990s, turning 95 percent of wetlands the size of Massachusetts to desert and all plants died. Many animals and birds were lived or visited the marshes in certain seasons disappeared. After the invasion of Iraq by the US army forces in 2003 and the removal of Saddam's regime, the water gradually began to return to Al-Chibayish marsh and returned to life again. However, unfortunately, due to the weakness and corruption of the Iraqi government and parliament. In addition, the irrigation projects in neighboring countries, especially the construction of dams; the marshes are threatened with drought again.

In 2016, the marshes of Iraq were inclusion in the UNESCO's World Heritage List. To support this project, this research was carried out.

Since the Bardy plant (Typhaceae family) is one of the most important plants in the marshes (Ahwar) after reeds, which is used mainly as feed for cattle, especially buffaloes. It is also the stem of the Bardy (Al-Akeed) used by the inhabitants of the marshes as a food. In addition, from the pollen grains of this plant made sweet called (Al-kheret). It is used to treat many diseases such as colitis and treatment of wounds and stop bleeding in addition to being used as a sexual enhancer for men.

Plants are important source of potentially bioactive constituents for the development of new chemotherapeutic agents. It has been well documented that the antimicrobial compound are abundantly present in medicinal plants [25]. The percentage of yield of methanol extract was more than that of the petroleum ether, chloroform and aqueous. The polar solvent was able to extract more of the extractives than non-polar solvents (petroleum ether, chloroform). Phytochemical constituents such as tannins, flavonoids, alkaloids, phenols and several other aromatic compounds are secondary metabolites of plants that serve as defence mechanisms against predation by many microorganisms, insects and herbivores [26]. Few researchers reported that several phytochemicals present in the plant extract exhibits antibacterial activity [27, 28]. The antimicrobial activities of all the three extracts tested, methanol extract significantly inhibited the growth of the organisms with 23 mm zones of inhibition. The result of this work however agrees with the findings of several studies [29, 30, 31]. In which showed that the methanolic extract of *Typha spp*. was active against Gram negative and Gram-positive bacteria. It is therefore conceivable that this extract can be used against *Escherichia coli*, *Proteus mirabilis*, *Klebsiella pneumonia*, *Streptococcus sp.*, *Enterobacter cloacae*, *Bacillus cereus*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*. The result of the present study shows also the antimicrobial activity of *Typha domingensis* Pers. extracts against pathogenic yeasts. The antibacterial activity of the methanol and aqueous extracts of *Typha domingensis* Pers. may be due to the presence of secondary metabolites like alkaloids, tannin, steroids, phenol, saponins, flavonoids compounds, which are previously reported for their antimicrobial property [29]. In the present study, the positive control in the experiment of the effect of different extracts of Typha domingensis Pers. leaves on pathogenic bacterial was ciprofloxacin, because ciprofloxacin is a new broad-spectrum antibiotic of the fluoroquinolone class. It is active against both Gram-positive and Gram-negative bacteria for these reasons was used in this study. Resistance to traditional antibiotics is an increasing problem. The introduction of ciprofloxacin (CIP) has been welcome, because it is safe and effective against many organisms (including drug-resistant ones) [32, 33, 34]. Fluconazole was used for yeasts because Fluconazole generally is active in vitro against *Candida albicans*, many of the non-albicans Candida species, and *C. neoformans* but not against *Candida krusei* or *Aspergillus species* [35, 36]. The results of the minimum inhibitory concentration showed that the methanolic and aqueous extracts of *Typha domingensis* Pers. have potent bactericidal properties against the tested organisms. The inhibitory effects of the extracts are most likely due to the presence secondary metabolites. The results obtained indicated the existence of antimicrobial compounds in the crude methanolic extracts of *Typha domingensis* Pers.

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This study was carried out to evaluate the potentials of *Typha domingensis* Pers. on the haemostatic mechanism, with primary interest on how it affects bleeding time, prothrombin time, clotting time and plasma fibrinogen concentration. The methanolic leaf extract of *Typha domingensis* Pers. exhibited haemostatic activities by decreasing bleeding, prothrombin and clotting times and also by increasing plasma fibrinogen concentration. These indices are measure of blood coagulation. While clotting time measure the intrinsic pathway, the prothrombin time measures the extrinsic pathway of blood coagulation. Fibrinogen concentration is critical to the formation of stable fibrin clot [22]. The reported decrease in bleeding time in this study is consistent with the findings of [37] when aqueous leaf extract of the plant was used. The results showed significant decrease in prothrombin time. Since prothrombin is a screening test for the extrinsic clotting system, that is, factor VII and can also detect deficiencies of factor V, X, prothrombin and fibrinogen [38], it follows then that the decrease in prothrombin time by the extract may be because of increase in the concentration of prothrombin or one of the other extrinsic clotting factors. These results agree with the reported findings of [39]. They reported decrease in (PT) and partial thromboplastin time (PTT) by *Fagara xanthoxyloides*, but contrasted with the observed effect of *Wobenzyme darages*, which increased PT in albino rats as reported by [40].

Clotting time test is a qualitative measurement of factors involved in the intrinsic pathway [22, 41]. Therefore, deficiency in the factors of the intrinsic pathway (I, II, V, VIII, IX, X, XI, and XII) will affect the result. From the results obtained, there was significant decrease in clotting time, reflecting that there was an increase in one or more of the clotting factors involved in the intrinsic pathway. These results correlate with the report by [42] on the haemostatic activities of the leaf extract of *Typha domingensis* Pers. which arrested bleeding from fresh wounds by reducing both bleeding and clotting times.

Plasma fibrinogen concentration was increased significantly by methanolic leaf extract of *Typha domingensis* Pers. Fibrinogen is synthesized in the liver and its synthesis is not dependent on the presence of vitamin K. Thrombin acts upon fibrinogen to remove four low-molecular weight peptides from each molecule, forming a molecule of fibrin monomer which can polymerize with other fibrin monomer molecules, thus forming fibrin [43]. Therefore, increase in fibrinogen concentration thus facilitates the rate of fibrin polymer formation which ultimately leads to more effective clot formation.

Composition of *Typha domingensis* Pers. includes flavonoids, alkaloids, essential oils and tannins, many of which are biologically active [44]. Tannins have been implicated in the haemostatic activity of plants where they arrest bleeding from damaged or injured vessels by precipitating proteins to form vascular plugs [42]. In addition, *Typha domingensis* Pers. plays an essential role in the synthesis of vitamin K by healing gastrointestinal disorders [45, 46, 47]. Vitamin K formed contributes to normal formation of prothrombin as well as a few other clotting factors which are responsible for the positive haemostatic effect.

In conclusion, methanolic leaf extract of *Typha domingensis* Pers. effectively inhibits the growth of pathogenic microbes and decreased bleeding, prothrombin, and clotting times and increased plasma fibrinogen concentration. Thus, indicating a positive haemostatic effect. This further provides a rationale for the use of the leaves of *Typha domingensis* Pers. in the wound management in traditional medical practice.

Conclusion

In conclusion, the methanolic leaf extract of *Typha domingensis* Pers. possessed antimicrobial and haemostatic properties as evidenced by inhibits the growth of pathogenic bacteria and yeasts. In addition, significant reduction on bleeding and clotting time in rats.

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