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Evaluation of *Tussilago farfara* L. Smoke by GC/MS: A Phytochemical Approach to a Traditional Medicine

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ABSTRACT

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Keywords: Tussilago farfara Smoke GC/MS Phenol Trapping Bioautography The smoke produced from natural substances such as medicinal plants is used in various cultures for different purposes. The use of medicinal fumes has been reported in nearly 50 countries. Among medicinal plants, Tussilago farfara L. known as coltsfoot has been introduced in Canon the famous book of Avicenna a Persian polymath, for chronic dry cough and various pulmonary diseases and shortness of breath. T. farfara is distributed in wet mountainous regions of Iran. For this study, the leaves and flowers of T. farfara were collected from Chalous Road in Iran. The smoke from the burning of T. farfara organs was prepared by homemade glassware trapping the smoke in methanol and then methanol was evaporated. In general, five grams of materials were burned and the smoke was dissolved and trapped in 100 ml of methanol. The trapped and dried materials from the smoke of extracts were filtered and injected into the GC/MS for analysis and identification of its constituents. 51 compounds representing 91.1 and 92.3 percent of smoke extracts of T. farfara were identified in leaf and flower. Also, 57 compounds were detected in the sample of EL and EF with 96.8 % and 97.7 %. The percentage of phenolic compounds that were identified in all extracts of smoke were SL and SF with 52.1 and 46.5, respectively. Phenol, Hydroquinone, P-Cresol and O-Cresol were the major compounds in the smoke extracts. Smoke leaves and flowers of T. farfara were selected to test the antimicrobial to continue. This study examined the bactericidal effect of smoke flowers. Fractions of effective constituents with the help of hexane-ethyl acetate with the method of thin layer chromatography (TLC) were isolated. The results of this experiment showed that a fraction (8:2) of hexane-ethyl acetate inhibited the bacteria Staphylococcus aureus. But Escherichia coli was not inhibited.

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1. Introduction

The demand for medicinal uses of plant-derived smoke outnumbers all other applications (Pennacchio *et al.*, 2010). The smoke produced from natural substances is used in various cultures and regions for different purposes including incense, medicine and food maintenance (Fabricant and Farnsworth, 2001). Therefore, different matters are recognized to be used to produce smoke for several utilizations (Pennacchio *et al.*, 2010). Human beings have used the smoke of medicinal plants to lead a healthy life long ago and in some cultures of the world, the smoke has been used in religious festivals and ceremonies (Mohagheghzadeh *et al.*, 2006). Plant-derived smoke at high temperatures

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exhibits fast pharmacological activity when inhaled (Nautiyal et al., 2007). Some medicinal plants are used in the form of smoke (Danial et al., 2018). Smoke mainly contains compositions that have antibacterial, anti-inflammatory antifungal, and antioxidant properties and some of them are used for the treatment of neuralgia, rheumatism, capillary bleeding, and skin disorders (Shafiee and Moravej-Salehi, 2015). Researchers are looking for natural medications that have fewer side effects and the Iranian traditional medicine is a valuable reference in this respect (Shafiee and Moravej-Salehi, 2015). The antimicrobial activity of smoke is attributed to the presence of compounds like phenols, carbonyls and organic acids (Holley and

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Patel, 2005). Many studies have presented that bacteria have increased resistance. There is an increasingly growing need for the development of new strategies for conventional antibiotic therapy (Shalayel et al., 2017). The use of medicinal smoke in the therapy of many diseases such as microbial diseases and infections in Iran, Turkey and Malaysia has long been popular (Danial et al., 2018). Roche et al. (1997) reported that smoke can protect seeds and seedlings against microbial corrosion. Kulkarni et al. (2011) indicated that smoke contains aromatic hydrocarbons, chlorine compounds and aldehydes that can reduce the infestation of endophytic fungi, as these smoke compounds show similar chemical structures to those of known fungicidal compounds. Another study result showed smoke generated by burning wood and medicinal herbs eliminated some of the bacteria that are harmful to horticultural plants (Nautiyal et al., 2007). Among the many plants whose smoke is used, Tussilago farfara L. known as coltsfoot has been introduced in Canon the famous book of Avicenna a Persian polymath, for chronic dry cough and various pulmonary diseases and shortness of breath (Mahdizadeh et al., 2015). In fact, the leaves of T. farfara are smoked like tobacco, as a domestic remedy for asthma (Qureshi et al., 2007). Coltsfoot (Tussilago farfara L.) from the Asteraceae family is a perennial plant (Ferrer et al., 2016). The flower buds of coltsfoot have a long history in the Chinese Pharmacopoeia for the treatment of cough, phlegm and asthmatic disorders (Qu et al., 2018). T. farfara is distributed in wet mountainous regions of the world and can also be found in Tehran, Azerbaijan and the Northern provinces of Iran (Norani et al., 2019). Antimicrobial resistance is a worldwide health problem associated with increased disease and mortality, while the factors associated with it are well known, unfortunately, the root causes of it continue to be declined (Shalayel et al., 2017). There are few works on the volatile combustion products of incense and their derivatives (Staub et al., 2011). In this study, we aim for the characterization and comparative analysis of the main volatile organic compounds (VOCs) present in the smoke of T. farfara organs.

2. Materials and methods

2.1. Plant materials and experimental conditions

In this study, the roots, leaves, barks and flowers of *T. farfara* were collected from Pol-e Zangholeh of Iran (51° 20' 17" N, 36° 48' 11" E). *T. farfara* samples were cleaned and then air-dried in the shade and powdered using a milling machine and kept in a cool dry place until ready to burn. Also, Green tea and black tea (*Camellia sinensis* (L.) Kuntze) were purchased from the Fuman farms in the north of Iran. Cloves (*Syzygium aromaticum* L.), and rosemary (*Rosmarinus officinalis* L.) were bought from the grocery store. The abbreviations of all samples are shown in Table 1.

Table 1. The smoke samples with their abbreviations.			
Abbreviation	Description		
ER	Extract of root of T. farfara		
EL	Extract of leaf of T. farfara		
EB	Extract of bark of T. farfara		
EF	Extract of flower of T. farfara		
SR	Smoke of root of T. farfara		
SL	Smoke of leaf of T. farfara		
SB	Smoke of bark of T. farfara		
SF	Smoke of flower of T. farfara		

2.2. Smoke collection and preparation of methanolic extract

The smoke of roots, leaves, barks and flowers of *T*. *farfara* was separately prepared by homemade glassware trapping the smoke in methanol (Fig. 1). In general, three grams of samples were burned and the smoke was dissolved and trapped in 100 ml of methanol. Extracts of root, leaf, bark and flower of *T*. *farfara* along with several well-known antioxidant plants including green tea, clove, black tea and rosemary were prepared by sonication of three g of dried plant material for 30 min in 50 mL of methanol. All extracts were filtered through Whatman no.1 filter paper and then concentrated at 40 °C using a rotary evaporator. The extracts were dried and stored at 4 °C until analysis.



Figure 1. Schematic representation of the collection the smoke of roots, leaves, barks and flowers of *T. farfara*.

2.3. GC–MS analysis and Identification of the smokes extract

GC-mass spectrometry (GC-MS) analysis was carried out by a Thermoquest-Finnigan gas chromatograph equipped with a fused silica capillary HP-5 column (60 m \times 0.25 mm i.d.; film thickness 0.25 µm) coupled with a trace mass spectrometer. Helium was used as the carrier gas at a flow rate of 1. mL/minute in a split ratio of 2: 00. Ionization voltage was 70 eV. Ion source and interface temperatures were 200° and 320°C, respectively. Identification was confirmed by comparison of each component's mass spectra with that of the internal mass spectra library of the main library, Wiley 7.0 and Adams and further identification was based on the comparison of peak retention indices by using a homologous series of nalkanes (C8 to C24) recorded under the same operating conditions and the published data (Adams, 2007).

2.4. Assessment of antioxidant activity against DPPH

The radical scavenging activity of all extracts against DPPH (2,2-diphenyl-2 picrylhydrazyl hydrate) was determined according to the previously described method of (Bozin *et al.*, 2007), using the IC₅₀ to compare the antioxidant properties. The absorbance was recorded at 517 nm with an ELISA reader (Epoch, BioTek instrument). The radical scavenging capacity (RSC) was calculated using the formula: In%=[(Ab-As)/ Ab] ×100, where In is DPPH inhibition, Ab is the absorbance of the blank, and As is the absorbance of the sample extract. Butylated hydroxytoluene (BHT) was used as a positive control. IC₅₀ is the concentration of the sample when the inhibition percentage is 50%.

2.5. Determination of total phenolic compounds

The total phenolic content compound was determined using the Folin- Ciocalteu method (Slinkard and Singleton, 1977). A calibration curve was prepared using a series of methanolic gallic acid solutions (10, 30, 100, 250, 500, 1000 μ g/ml), combined with 0.1 ml Folin–Ciocalteu reagent and after 3 min, 0.3 ml sodium carbonate (7.5%). The absorbance of the mixture was measured at 765 nm using a spectrophotometer (Smart spec plus, BIORAD). The experimental extracts (0.01 g/ml) of smoke and the other antioxidant plants were each combined with the same reagents and absorption was measured after 2 h to assess phenolic compounds, with

three technical replications. Gallic acid was used as the standard for a calibration curve, and the results were expressed as mg of the gallic acid equivalent dry weight of extract (mg GAE/g DW).

2.6. Bioautography of the Antimicrobial Compounds

The methanolic flowers and leaves smoke extracts were chromatographically analyzed using the thin layer chromatography (TLC) method. The samples were spotted on 2×10 cm² silica gel plates using spotting tubes about 1-2 cm above the bottom of the plates. Then, they were placed in a chromatography tank with selected solvents of different ratios of hexane and ethyl acetate enough to wet the lower edge of the plate, before the spotting was performed. The plates were left in the solvent for some time, during that, the solvent moved across the plate from bottom to top. The plates were removed from the tank, allowed to dry, and then visualized under ultraviolet irradiations at 254 and 366 nm by spraying with Ninhydrin. The plates with more spots were used in the bioautography test. The silica gel plates were seeded with Staphylococcus aureus and Escherichia coli and incubated for 20 hours at 37°C. The clear zones due to growth inhibition of the microorganisms indicated the location of antimicrobial compounds on the TLC plates.

2.7. Statistical analysis

Parametric data were analyzed according to the analysis of variance based on a completely randomized design with three replications, using SAS Statistical Package Program version 9.0 and SPSS software version 20. The means were compared with the Least Significant Difference (LSD) test at a 5% probability level.

3. Results

3.1. Plant extraction yield

The yields of all extractions are shown in Fig. 2. The highest extraction yields were obtained in EL (Extract of the leaf) with 11.4 % w/w. The sample of SR (Smoke extract of the root) had the lowest extraction yield with 4.1 % w/w.

3.2. Chemical composition of extracts

In total 51 compounds representing 91.1 and 92.3 percent of smoke extracts of *T. farfara* were identified in the leaf and flower (Table 2). Also, 57 compounds

were detected in the sample of EL and EF with 96.8 % and 97.7 % (Table 3). 2-Ethylhexyl hexanoate was the main component in the EL (20.0 %) and EF (25.3 %). Other important compounds identified in the extracts included Hydroquinone, Catechol, Neophytadiene, 9, 12, 15-Octadecatrienoic acid and N-Hexadecanoic acid. The results showed that most of the compounds identified in smoke are phenolic compounds. The phenolic compounds identified in SL and SF were 52.1 % and 46.5 %, respectively. Phenol, Hydroquinone, Catechol, P-Cresol and hydroquinone with 19 %, 10.5 %, 10.3 % and 5.7 % respectively were the major compounds in the smoke of leaf (Fig. 3). As a main compound, Phenol (19.8 %), O-Cresol (5.3 %), Hydroquinone (4.8 %) and Phenol, 4-ethyl (4.1 %) were identified in flower smoke (Fig. 4). The other important components identified in all smokes include Phenol, 4-ethyl, 3-Methyl-1,2-cyclopentanedione, n-Hexadecanoic acid, Phenol, 2,5-dimethyl, Phenol, 2methoxy-3-(2-propenyl) and Neophytadiene.



Figure 2. The yields of all extractions from *T. farfara* root, leaf, bark and flower.

3.3. Total phenolic contents (TPC)

Results in Fig. 5-A showed the highest TPC in EL (extract of the leaf) with 341 mg GAE/g DW. The bark extract (EB) had the lowest TPC with 119 mg GAE/g DW. In comparison with green tea, cloves, black tea and rosemary; interestingly the phenolic content of this sample was higher than rosemary but lower than the green tea, cloves and black tea.

3.4. Antioxidant activity (AA)

The results of the comparison of antioxidant activity have been demonstrated in Fig. 5-B. In the DPPH assay, the highest AA was observed in EL (extract of the leaf) with IC₅₀ 82 µg/ml. The lowest activity was found in SR (Smoke extract of the root) with IC₅₀ 164.3 µg/ml. In general, in comparison with well-known antioxidant plants such as clove, black tea, green tea and rosemary, the smoke extracts of *T. farfara* showed comparable antioxidant activity.

 Table 2. Chemical composition (%) of different organs smoke of *Tussilago farfara* L.

No	RT	Components	SL%	SF%	RIc
1	8.1	3-Methyl-2-cyclopenten-1-one	0.1		973
2	8.4	Phenol	19.0	19.8	992
3	9.5	3-Methyl-1,2-cyclopentanedione	3.3	3.7	1043
4	10.1	O-Cresol	0.7	5.3	1068
5	10.5	P-Cresol	5.7		1093
6	10.8	Guaiacol	1.0	2.6	1096
7	10.9	2(3H)-Furanone, dihydro-4-hydroxy	1.5	3.9	-
8	11.5	Maltol	3.8	1.0	1129
9	11.8	3-Ethylphenol		1.1	1142
10	12.0	Phenol, 2,5-dimethyl	2.7	2.4	1150
11	12.2	Phenol, 4-ethyl	1.6	4.1	1165
12	12.7	(S)-(+)-2',3'-Dideoxyribonolactone	1.7		-
13	12.8	Creosol		0.5	1192
14	12.8	Butanoic acid, 1-methylhexyl ester	0.9		1197
15	13.0	Catechol	10.3	1.2	1219
16	13.1	1,5-Dioxonane, 2-ethoxy-9-methyl	2.7		-
17	13.3	Benzofuran, 2,3-dihydro	1.7	0.9	1223
18	14.3	Hydroquinone	10.5	4.8	1241
19	15.3	Phenol. 2.6-dimethoxy		2.5	1256
20	15.5	Phenol. 2-methoxy-3-(2-propenyl)	0.6	2.2	1263
21	15.9	1H-Indole, 3-methyl	1.0		1264
22	16.0	Skatole		0.7	1296
23	16.8	6-Undecylamine	0.8	017	1354
24	17.3	1-Dodecene	1.7		1390
25	17.4	Tetradecane	0.6	0.7	1396
26	17.8	Benzene 1.2.3-trimethoxy-5-methyl	2.9	17	-
20	17.0	M-Dioxan-4-ol 2 6-diethyl-5-	2.9	1.,	
27	18.0	methyl- acetate	0.8	2.9	1450
28	187	Hexadecane	04	16	1496
20	10.7	Quinic acid	2.5	1.0	1530
30	19.1	Tetradecanol	2.5	0.6	1575
31	19.0	Hentadecane	0.6	0.0	1596
32	20.6	Tetradecanoic acid	0.0	1.6	1659
32	20.0	Octadecane	17	0.6	1695
34	21.0	Neophytadiene	3.8	0.0	1810
35	21.5	Phthalic acid diisobutyl ester	5.0	0.0	1774
36	$\frac{21.7}{22.2}$	Hentadecanenitrile	0.0	0.4	1800
50	22.2	Hevadecanoic acid 15 methyl	0.9	0.0	1000
37	22.4	methyl ester		1.0	1822
20	22.7	N Havadaaanaja aajd	16	15	1960
20	22.7	Dibutyl phthelete	1.0	4.5	1860
39	22.9	Dibutyi pilulalate	1.0	0.4	1009
40	23.0		1.0	0.0	1001
41	24.0	1-Octadecanol	0.0	0.8	1989
42	25.0	Hexadecanamide	0.0	0.7	2080
43	26.0	Heneicosane	0.0	0.7	2195
44	26.3	(Z)-13-Octadecenal	0.8	0.7	2225
45	27.8	Iricosane		0.7	2300
46	28.3	Phthalic acid, bis (2-ethylhexyl)	0.7	7.3	2531
	•••	ester			• • • • •
47	29.4	<i>N</i> -Hexacosane		0.9	2600
48	29.8	1,3-Benzenedicarboxylic acid, bis		0.6	2704
		(2-ethylhexyl) ester			
49	30.4	Squalene	0.3	1.5	2780
50	30.8	<i>N</i> -Octacosane	0.6	0.7	2800
_		Benzenepropanoic acid, 3,5-bis(1,1-			
51	38.5	dimethylethyl)-4-hydroxy-,		4.5	3800
		octadecyl ester			
Tot	al con	npounds	91.1%	92.3%	

of 7	Tussila	ago farfara L.			
No	RT	Components	EL%	EF%	RIc
1	7.1	Trimethylene oxide	2.9		543
2	7.12	Propylene oxide		4.6	477
3	7.15	3-Chloropropanoic acid		1.6	-
		Heptadecafluorononanoic			
4	7.19	acid dodecyl ester		1.7	-
5	7 21	A Amino 2 fluoro N methylbenzamide		13	
5	7.21	n Dodoconovi chlorido	22	1.5	-
7	7.25	<i>n</i> -Dodecanoyi chioride	2.5		-
/	7.28		1.9		-
8	7.42	Isovaleric acid	1.0		-
9	8.01	Butyl(dimethyl)propoxysilane	2.2	1.4	-
10	8.1	3-Chloro-1,2-propanediol	2.9		-
11	8.12	Pyrido[3,4-d]pyrimidin-4(3H)-one		1.6	-
12	8 22	Succinic acid, 3-oxobut-2-yl		1.0	_
12	0.22	pentadecyl ester		1.0	
13	8.24	Benzene, (3,3-dimethyl-4-pentenyl)-	1.3		-
14	8.3	(1,4-Dioxan-2-yloxy)(trimethyl)silane	1.4		-
1.5	0.2	2,4-Dihydroxy-2,5-dimethyl-3(2H)-		2.6	077
15	8.3	furanone		3.6	9//
16	8.4	Phenol	2.9	3.4	992
17	8.54	gamma-Butyrolactone	2.1	2.1	_
18	87	1-Aminocyclopropanecarboxylic acid	2.1	2.1	_
10	0.7	1 4 Dioxin 2.3 dihydro 5.6 dimethyl	2.2	16	
20	9.7	1 Bonzul 4 piporidono	1.0	1.0	-
20	9.0	1 Amin acual annon an acut anulia acid	1.0	15	-
21	10.85	1-Anniocyclopropanecarboxylic acid		1.5	-
22	11.85	4H-Pyran-4-one, 2,3-dihydro-3,5-		2.5	1150
		dihydroxy-6-methyl-			
23	12.44	(5R)-5-Methoxypyrrolidin-2-one		1.2	-
24	12.88	Catechol	1.0	1.0	1219
25	13.57	Glyceryl diacetate	1.2	0.9	1230
26	14.01	Hydroquinone	11.7	1.6	1241
27	14.18	Ethyl 3-hydroxypentanoate		1.3	-
28	14.45	Tetradecane		1.1	1396
29	16.6	2-Hydroxy-4-methylbenzaldehyde	2.3	2.7	-
30	17.23	Pentadecene	1.1		1486
31	18.6	Spathulenol		1.6	1578
32	18.7	Carvonhyllene oxide		14	1594
33	19.0	2-Ethylbeyyl beyanoate	20.0	25.3	-
24	20.14	2.2 Dihudrovyovalahovonona	20.0	23.5	-
54	20.14	2,5-Dillydroxycyclonexallone		1.0	-
35	20.9	Acetic acid, 2-(2,2,0-trimethyl-7-0xa-	1.1		-
26	01.46	bicyclo[4.1.0]hept-1-yl)-propenyl ester	<i>c</i> 0		1010
36	21.46	Neophytadiene	6.3		1810
37	21.93	Phytol acetate	2.2		-
38	22.74	N-Hexadecanoic acid	3.9	3.9	1860
39	24.21	Methyl linolenate	1.7		-
40	24.33	Phytol	4.5		1881
41	24.5	9,12-Octadecadienoic acid		1.9	-
42	24.6	9,12,15-Octadecatrienoic acid	8.4	2.5	-
45	26.0	Henicosane		6.4	2100
		3-Furanacetic acid, tetrahydro-2-oxo-			
46	27.5	methyl ester		1.2	-
17	27 74	n Triaccana		5 17	2200
4/	21.14	<i>n</i> -mcosale		3.47	2300
48	28.23		1.0	1.0	-
49	28.3	Bis(2-ethylhexyl) phthalate	1.2		2499
50	28.55	4-Aminoquinoline	2.9		
51	29.2	1,3,5-Cycloheptatriene, 7,7-dimethyl-		1.2	
52	29.33	<i>n</i> -Octadecane		3.8	
53	30.39	Squalene		1.4	2780
54	34.18	Campesterol	1.7		-
55	34.48	Stigmasterol	1.5		-
56	35.14	beta -Sitosterol		1.0	-
57	38 30	Myristyl myristate		0.9	_
<u> </u>	ol and	nounda	06 90/	07.70/	
1 O t	ai com	DOULIUS	70.8%	71.1%	





Figure 3. Gas chromatography-mass spectrometry (GC-MS) chromatogram of leaves's smoke of *T. farfara*.







Figure 5. Comparison of total phenolic compounds (A) and antioxidant activity (B) in all samples with green tea, cloves, black tea and rosemary.

3.5. Correlation between AA and TPC

The correlation between AA and TPC in smoke extracts is presented in Table 4. The results showed the

significant, respectively.

strongest positive correlation with total AA and TPC (r=0.75, $p\le0.01$), (Fig. 6).

Table 4. Simple correlation among antioxidant, total phenolic content and total flavonoid of smoke extracts.

	Total Phenol	Antioxidant activity
Total Phenol	1.000	0.75**
Antioxidant activity	0.75^{**}	1.000
n=30. ** and ns sign	ificant at 1%,	level of probability and non-



Figure 6. Correlation among antioxidant and total phenolic content in all samples.

3.6. Antimicrobial activity of smoke extracts

Fig. 7 refers to the flower's smoke extract which has 1,2,3,4 and 5 fractions on the plate. Each spot was 20 mg of the sample that dissolved in 200 μ l of solvent. For staining, 20 microliters are laid out for each spot. It was observed that *S. aureus* growth was inhibited around samples 2, 3 and 4 (Table 5, Fig. 8-A) While the growth of *E. coli* has not been inhibited (Fig. 8-B).



Figure 7. TLC separation of the flower and leaf smokes of *T. farfara* in the present study.

Table 5. Antibacterial activity against human pathogens of smoke extracts against *Staphylococcus aureus* and *Escherichia coli*

Samplas	Main compounds	Staphylococcus Escherichia		
Samples		aureus	coli	
SL	Phenol, Catechol, Hydroquinone	-	-	
SF	Phenol, O-Cresol, Hydroquinone	+	-	



Figure 8. Detection of the antibacterial activity of smoke of *T. farfara* by direct bioautography against *Staphylococcus aureus* (A) and *Escherichia coli* (B).

4. Discussion

The results of extract yields showed a considerable difference in all samples. The differences in the extract yields among these samples might be related to the different availability of extractable components, resulting from the varied chemical composition of plants (Sultana *et al.*, 2009).

A comparison of volatile compounds of the flower, bark and leaves of *T. farfara* between our results in this study and previous studies (Norani *et al.*, 2019; Judzentiene and Budiene, 2011) revealed no similarity between the chemical compositions of the smoke and those of the essential oils. The essential oils were dominated by monoterpene hydrocarbons, oxygenated

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monoterpenes, sesquiterpenes, and oxygenated sesquiterpenes, whereas the smoke extracts were marked by phenol derivatives. Catechol is a natural polyphenolic compound that occurs widely in fruits, teas, vegetables, tobacco and some of those used in traditional Chinese medicines (Mahdavi et al., 2018). Catechol has been widely studied because of its biological importance such as antivirus and antioxidant activities and effects on several enzymes (Lin et al., 2009). Generally, phenols are well-known as antimicrobial agents and some of these components have been used as disinfectants or as preservatives in cosmetic and food products (Kubo et al., 1995). In a previous study, the smoke of other matters was analyzed by GC-mass and several substances were identified such as dichloromethane, limonene, acetic acid, etc., some antibacterial agents; i.e., phenol, cresol, licochalcone A, carvacrol, etc., some anti-neoplasms such as prospidium chloride (Sweetman, 2009), and monoterpenes, diterpenes, sesquiterpenes, antioxidants, etc. (Joharchi et al., 2020).

As a radical scavenging investigation (DPPH) on smoke, Soares et al. (2016) reported that has shown activity by detaching the DPPH free radical with IC₅₀ of 244 ug/mL. The antioxidant activity of smoke depends mostly on its composition, especially in phenolic compounds (Soares *et al.*, 2016).

The antimicrobial effects of smoke may be pertinent to their phenolic and polar compounds (Danial *et al.*, 2018; Fouladi Fard and Farajinia, 2016). Lignin can be converted into phenolic compounds in the form of liquid smoke (Chen, 2014). Phenolic compounds have been associated with positive effects on cardiometabolic health, cognition, type II diabetes, obesity, neuroinflammation and others as well as safe and represent a new strategy to for treating skin (Mahdavi *et al.*, 2018).

These results were in agreement with Sim and Nyam (2019), Kho et al. (2019) and Sim et al. (2019) in which a high correlation was observed between antioxidant capacities and phenolic content of kenaf leave tea and *Hibiscus cannabinus* L. leaves. Phenolic compounds are very important natural antioxidants in various plants, which exhibit antioxidant activity through radical scavenging (Norani *et al.*, 2019).

The result of biological activity may be due to the presence of phenols or phenolic compounds in the smoke extract, which is better known as antimicrobial agents (Cetin-Karaca and Newman, 2015). In addition, different strains of tuberculosis and non-tuberculous strains such as *M. kyorinense* and *M. kansasii* were inhibited by hydroquinone (Jyoti *et al.*, 2016). Also, some reports have demonstrated that hydroquinone possesses antioxidant properties (Yamaguchi *et al.*, 2006).

Beneficial bioactivities such as the antibacterial activity of phenolic compounds are related to their chemical structures, particularly the presence of an aromatic structure and hydroxyl groups able to neutralize free radicals and other reactive oxygen species (Lima *et al.*, 2019). The published researches are not definite about the differences in the mechanisms of function of phenolic compounds on Gram-positive and Gram-negative bacteria (Sanhueza *et al.*, 2017).

5. Conclusion

In conclusion, *T. farfara* smoke has relatively high antioxidant potency and total phenolic compounds. Also, a positive correlation between the total phenolic content and antioxidant activity of all smoke extracts was established. The phenolic compounds display a wide perspective of biological factors such as antimicrobial and anticancer activities, as well as protective effects against neurodegenerative diseases. The pleiotropy bioactivities of phenolic compounds are limited by their bioavailability. To deal with this challenge, novel delivery systems like nanoencapsulation an encouraging delivery systems for the effective forwarding and release of phenolic compounds to desired targets.

Abbreviation

AA= antioxidant activity, TPC= Total phenolic contents, DPPH= 2,2-diphenyl-2 picrylhydrazyl hydrate, TLC= thin layer chromatography

Conflict of Interests

The authors have to declare their conflict of interest.

Ethics approval and consent to participate

No human or animals were used in the present research.

Consent for publications

All authors read and approved the final manuscript for publication.

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Availability of data and material

All the data are embedded in the manuscript.

Authors' contributions

The first author [M. N.]: performance of the research project and writing the article

The second author [A. C.]: cooperation in the implementation of the research project

The third author [A. A.]: statistical analysis of the data The fourth author [M. A.]: implementation of the research project

Informed Consent

The authors declare not to use any patients in this research.

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