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ORIGINAL ARTICLE

Chemical composition and potential bioactivities of essential oil from *Quercus mongolica* bark

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Abstract The chemical composition and potential bioactivities of essential oil from *Quercus mongolica* bark (EOQMB) were researched for value-added utilization processing by-product. The results of gas chromatography-mass spectrometry (GC–MS) analysis showed that 30 components accounting for 98.42% were identified in EOQMB, with pentadecanoic acid the most abundant compound accounting for 34.90%, which was further confirmed by the Fourier transform infrared observation. EOQMB exerts antioxidant activities, and the IC_{50} values for scavenging DPPH radical, ABTS radical, and hydroxyl radical were 8.48, 0.77, and 3.54 mg/mL, respectively. The effects of EOQMB on prolonging activated partial thromboplastin time and thrombin time and on decreasing fibrinogen are similar to those of heparin, and the promising anticoagulant activities of EOQMB could be largely contributed by pentadecanoic acid. Herein, the present study uncovered that the waste *Q. mongolica* bark can serve as a new potential material in pharmaceutical products.

1. Introduction

Quercus mongolica Fisch. ex Turcz (*Q. mongolica*), a member of the genus *Quercus*, is mainly distributed in Japan, South Korea, the Russian Far East, the Korean Peninsula, and northern and northeastern China (Zeng et al., 2016). *Q. mongolica* wood is used to make furniture. *Q. mongolica* acorns are utilized for the production of starch (Ning et al., 2018). *Q. mongolica* leaves are employed for the breeding of *Antherea pernyi* (Yang et al., 2010). Given the high nutritional value of *A. pernyi*, the demand for *Q. mongolica* in northern China is increasing, the planting area of *Q. mongolica* is increasing, and the amounts of

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side shoots pruned and cut in rotation are increasing annually. With the continuous degradation of *Q. mongolica* species (Kim et al., 2011), developing the valuable biological activities of its different parts is particularly important. A variety of chemical compositions have been extracted with ethanol and water from *Q. mongolica* leaves and showed antibacterial activity (Wang et al., 2016). Furan, essential phenol, and other ingredients are found in its heartwood, which has the effect of treating scald and trauma (Niu et al., 2021), *Q. mongolica* bark was extracted with absolute ethanol, and HPLC analysis showed that the bark contains dandelion ketone, ursolic acid ethyl ester, and other compounds (Wang et al., 2014). The bark of *Q. mongolica* was traditionally used as folk medicine to treat many diseases, including bacterial dysentery, acute gastroenteritis, dyspepsia in children, jaundice, bronchitis, hemorrhoids, burns, wounds, mild diarrhea, and minor oral mucosal inflammation (Deryabin and Tolmacheva, 2015). Although *Q. mongolica* bark is medicinally valuable, most of it is discarded as waste, and only the essential oil in *Q. mongolica* bark is rarely reported.

Pine needles as agricultural waste are steam-distilled to produce large quantities of pine needle oil, which is an important ingredient in pharmaceuticals, food and spices, and cosmetics (Antonella et al., 2017). Some essential oils can be used in the therapy of cardiovascular diseases (CDs) and are highly correlated with antioxidant and anticoagulant activities (Félix-Silva et al., 2014). At present, the mortality rate of CDs is higher than that of malignant tumors, accounting for >40% of disease-related deaths. Blood coagulation can induce thrombosis, which accelerates the progression of CDs (Lippi et al., 2011). Therefore, natural antioxidants play an important role in thrombosis (Saeidi et al., 2018). Anticoagulants have been widely used in the treatment of disseminated intravascular coagulation and the thrombosis of various diseases (Aboonabi and Singh, 2016; Wang et al., 2010). At present, heparin is limited by the supply and demand of pigs, and can cause a hemorrhage tendency (Dore et al., 2013). Some scholars have studied and developed the antithrombotic role of essential oils with reduced side effects and discernible effects (Xia et al., 2017).

In this study, we determined and analyzed the chemical composition of essential oil from *Q. mongolica* bark (EOQMB) by gas chromatography–mass spectrometry (GC–MS) and fourier transform infrared spectroscopy (FT-IR) by hydrodistillation. The potential biological activities, including antioxidant activities with DPPH radical, ABTS radical, and hydroxyl radical scavenging ability, and anticoagulant activities with activated partial thromboplastin time (APTT), thrombin time (TT), prothrombin time (PT), and fibrinogen (FIB), were explored to provide a scientific evidence for rationally improving the waste utilization of agricultural resources and developing its novel medicinal value.

2. Materials and methods

2.1. Plant material

Q. mongolica bark was collected from the scraped skin of large side shoots pruned on March 2018 at the Jilin Provincial Sericulture Institute, Jilin City, Jilin Province, China. Specimens were preserved in the Engineering Research Center for Agricultural Resources and Comprehensive Utilization of Jilin Province under the herbarium code QMB0601.

Fresh *Q. mongolica* bark powder (100 g) was placed on a 1000 mL two-necked flask equipped with a Clevenger-type apparatus and a thermometer. The power was added with 500 mL of distilled water, soaked for 2 h, and heated for 5 h. The mixture of essential oil and water was cooled to room temperature, dried with anhydrous Na_2SO_4, collected, and hermetically stored at 0 °C for GC–MS analysis.

2.2. Chemical composition of EOQMB

2.2.1. GC–MS analysis

A GCMS-QP2010 instrument with Rxi-5sil column was used to measure the chemical composition of EOQMB. The carrier gas was highly purified helium, and the flow rate was 1.0 mL/min. The collision energy for MS detection was 70 eV, and data were recorded within 40–450 amu. The vaporizer temperature and ion-source temperature were adjusted to 280 °C and 230 °C, respectively. Each chemical composition was identified by comparing the retention index obtained from a database (NIST05) with the retention indexes calculated on the basis of n-alkanes (C_8–C_{40}) and reported literature (Waterman, 2005).

An essential oil sample dissolved in ether (60 mg/mL) was injected automatically into a vaporizer at 250 °C with a split ratio of 1: 30, and the conditions of the programmed temperature were as follows: Start at 60 °C and maintain for 6 min; increase from 60 °C to 300 °C at 3 °C/min; maintain at 300 °C for 10 min.

2.2.2. FT-IR analysis

The FT-IR spectra of EOQMB and its main component (pentadecanoic acid) were determined by using an FTIR-650 spectrometer. Pentadecanoic acid and KBr were ground at a ratio of 1: 100 and then pressed into tablet. The obtained volatile oil was evenly spread on a KBr window, and the blank spectrum was scanned. The KBr film carrying the sample was detected for 20 times between 4000 cm^{-1} to 400 cm^{-1} by using an FT-IR spectrophotometer. Measurements were repeated every 3 min (Nie et al., 2018).

2.3. Antioxidant activity of EOQMB

The antioxidant activities of EOQMB were evaluated by detecting the scavenging ability of EOQMB on DPPH radical, ABTS radical, and hydroxyl radical. These methods are available for water-soluble and lipid-soluble antioxidants.

2.3.1. DPPH radical assay

Essential oil samples (2 mL) with different concentration gradients (0.5–20 mg/mL) and 2 mL of 0.5 mmol/L DPPH solution were mixed and shook thoroughly. Vc and ethanol were listed as a positive control and a negative control, respectively, and the absorbance was observed at 517 nm (Wang et al., 2010).

2.3.2. ABTS radical assay

The ABTS reserve liquid was prepared with 5 mL of 7 mmol/L ABTS liquid and 88 μL of 140 mmol/L $K_2S_2O_8$ liquid, and the diluted solution of ABTS radical was a solution diluted with 50 times ethanol. Diluted solution (2 mL) was added into 2 mL of essential oil sample (0.1–2.0 mg/mL). The absorbance was assayed at 734 nm (Hou et al., 2012).

2.3.3. Hydroxyl radical assay

Phenanthroline solution (1 mL, 0.75 mmol/L) was added into 2 mL of PBS (pH 7.40) and 1 mL of essential oil (1.0–10 mg/mL) in a test tube. Subsequently, 1 mL of 0.75 mmoL ferrous sulfate was added to 1 mL of 0.12% H_2O_2, the mixture was thermo-

statically incubated at 37 °C for 1 h, and the absorbance was measured at 536 nm. Distilled water rather than H_2O_2 was used as the positive control, and 1 mL of distilled water rather than volatile oil was the negative control (Amir et al., 2011).

2.4. Assay of anticoagulant action

The anticoagulant activities of EOQMB and pentadecanoic acid were assayed through coagulation tests in a CL-2000BV coagulometer (Jiangsu Sinnowa Medical Technology Co., Ltd, China) with heparin (positive control) and 0.90% NaCl (negative control) as a reference. APTT, TT, PT, and FIB content were used to evaluate the anticoagulant activity (Wang et al., 2013; Yin et al., 2017).

Plasma pretreatment was conducted as follows: Sodium citrate (0.109 mol/L) was precisely mixed with fresh and healthy human plasma at a volume ratio of 1: 9 and jiggled several times. The mixture was centrifuged for 20 min at 3000 r/min, and the supernatant of platelet-poor plasma was collected, packed and sealed in plastic tubes, and maintained in frozen storage. The plasma preheating was conducted at 37 °C before the experiment. All the following experiments were completed at 37 °C within 2 h.

The contents of four parameters were measured by using assay kits (Jiangsu Sinnowa Medical Technology Co., Ltd). EOQMB was dissolved in 20% dimethyl sulfoxide at five different concentrations (1.87, 3.75, 7.5, 15, and 30 mg/mL). The clotting time was recorded.

2.4.1. APTT assay

In the APTT assays, 20 μL of EOQMB solution with the five abovementioned concentrations were incubated with 80 μL of plasma for 60 s. The mixtures were added with 100 μL of APTT reagent for 1 min, followed by 25 mmol/L $CaCl_2$ (100 μL).

2.4.2. PT assay

In the PT assays, 20 μL of EOQMB solution with the five abovementioned concentrations were incubated with 80 μL of plasma for 30 s. The mixtures were then added with 200 μL of PT reagent.

2.4.3. TT assay

In the TT assays, 20 μL of EOQMB solution with the five abovementioned concentrations were incubated with 80 μL of plasma for 30 s. The mixtures were then added with 0.1 mL of preheated TT reagent.

2.4.4. FIB assay

In the FIB assays, a standard curve was first drawn. The fixed-value plasma of redissolved FIB was diluted in 20 mmol/L imidazole diluent at different proportions of 1: 5, 1: 10, 1: 15, 1: 20, and 1: 30. The fixed-value plasma of diluent at different concentrations was obtained at 200 μL, preheated at 37 °C for 30 s, and mixed with 100 μL of FIB thrombin to determine the clotting time. The plasma concentrations (mg/dL) with different solutions of FIB were used as the abscissa, and the corresponding clotting time (Antonella et al., 2017) was used as the ordinate. The standard curve was constructed in a double logarithmic coordinate system.

Different samples (20 μL, 1.87–30 mg/mL) were mixed with 80 μL of plasma. The mixtures were then diluted with 0.9 mL of 20 mmol/L imidazole and incubated at 37 °C for 30 s. The above diluted fixed-value plasma (0.1 mL) was added with 0.05 mL of the redissolved FIB thrombin. Subsequently, the content of FIB was computed by using the equation in accordance with the clotting time of the plasma to be tested.

2.5. Statistical analysis

The experiments were repeated in triplicate, and the results were analyzed on SPSS software. $P < 0.05$ and $P < 0.01$ indicated significant results and highly significant results, respectively.

3. Results and discussion

3.1. Essential oil characterization

The extraction yield of EOQMB was 0.08% ± 0.03%. EOQMB had an aromatic odor and pale-yellow to light-brown color. The essential oil was rich in fatty acids because it turned from liquid into solid when it was cooled to room temperature (Hao et al., 2018). GC–MS analysis successfully identified 30 compounds. These compounds accounted for 98.42% of the total composition. The qualitative and quantitative analyses of EOQMB are listed in Table 1. EOQMB is rich in acids (63.37%), esters (15.22%), aldehydes (8.53%), and alcohols (6.24%). The main constituents are pentadecanoic acid (34.90%), linoleic acid (19.80%), ethyl linoleate (4.87%), oleic acid (4.62%), octadecanal (4.62%), 2-pentylfuran (4.00%), phytol (2.85%), 1-octadecanol (2.25%), and hexadeca-1,4-lactone (2.24%).

In the FT-IR spectrum of the functional groups, the characteristic absorption peaks in all chemical components were overlapped. In accordance with the IR spectra in Fig. 1, EOQMB and pentadecanoic acid had mostly similar absorption peaks of functional groups, and their IR spectra were consistent.

The characteristic peaks at 2922, 2850, and 1464 cm^{-1} showed asymmetrical, symmetric stretching vibration peaks, and in-plane scissoring vibration peak of —CH_2, respectively. The absorption peaks at 2955, 1431, and 1380 cm^{-1} proved the presence of asymmetric stretching vibration peaks and bending vibration of —CH_3. The peak at 1703 cm^{-1} was caused by the stretching vibration of C═O among the carboxyl group (Hosseini et al., 2013). The peaks at 1412 and 1254 cm^{-1} were the bending vibrations of O—H and asymmetric stretching vibrations of C—O (δ—CH_2—COOH) (Lu and Deng, 1989). The strong broad peaks around 922 cm^{-1} were the rocking vibration of —OH (carboxylic acid). The adsorption bands at 1318, 1298, 1275, 1230, 1203, and 1188 cm^{-1} were the characteristic peaks of —$(CH_2)_n$— in-plane rocking vibrations, and the peak at 717 cm^{-1} confirmed the long chains of $n \geq 4$. The above IR analysis proves that the peak shape of pentadecanoic acid is complete, and the peak position is correct.

In the IR spectra, the functional groups of EOQMB and pentadecanoic acid were consistent. The IR analysis results further demonstrated that the main components of EOQMB were consistent with those of GC–MS.

Table 1 Chemical profile of the EOQMB.

No.	RT	Compound	Molecular formula	RIa	RIb	RIc	References	Chemical classification	Content/ %
1.	9.425	2-pentylfuran	$C_9H_{14}O$	1040	988.26	956	(Xia et al., 2017)	Ester	4.00
2.	12.429	*trans*-2-Nonenal	$C_9H_{16}O$	1112	1057.97	1112	(Wang et al., 2015)	Aldehyde	0.52
3.	24.059	(E)-2,(E)-4-decadienal	$C_{10}H_{16}O$	1220	1278.22	1314	(Czigle et al., 2006)	Aldehyde	0.56
4.	43.416	phytone	$C_{18}H_{36}O$	1754	1822.75	1790	(El-Shamy et al., 2012)	Ketone	0.69
5.	46.56	valeric acid,undec-2-enyl ester	$C_{16}H_{30}O_2$	1787	1819.43	–	–	Ester	0.84
6.	49.044	(Z)-7-hexadecenal	$C_{16}H_{30}O$	1808	1883.56	2157	(Zhao et al., 2016)	Aldehyde	1.66
7.	47.204	(Z)-11-hexadecenal	$C_{16}H_{30}O$	1808	1836.06	1812	(Uehara et al., 2015)	Aldehyde	1.17
8.	48.473	pentadecanoic acid	$C_{15}H_{30}O_2$	1869	1854.43	1869	(Rahman et al., 2016)	Acid	34.90
9.	47.015	11-hexadecyn-1-ol	$C_{16}H_{30}O$	1872	1831.18	–	–	Alcohol	1.14
10.	45.995	methyl palmitate	$C_{17}H_{34}O_2$	1878	1801.80	1825	(Sharma et al., 2010)	Ester	0.70
11.	45.685	hexahydrofarnesyl acetone	$C_{18}H_{30}O$	1902	1893.75	1905	(Zheng et al., 2006)	Ketone	1.00
12.	50.652	2-hexyl-decanoic acid	$C_{16}H_{32}O_2$	1904	1925.06	–	–	Acid	0.45
13.	52.025	palmitic acid	$C_{16}H_{32}O_2$	1968	1960.51	1966	(Sharma et al., 2010)	Acid	1.97
14.	51.155	hexadeca-1,4-lactone	$C_{16}H_{30}O_2$	1980	1938.05	–	–	Ketone	2.24
15.	54.619	octadecanal	$C_{18}H_{36}O$	1999	1991.46	2030	(Pino and Quijano, 2012)	Aldehyde	4.62
16.	46.845	hexanoic acid, 6-tridecyl ester	$C_{19}H_{38}O_2$	2013	2024.26	–	–	Ester	0.67
17.	51.336	phytol	$C_{20}H_{40}O$	2045	2168.37	2045	(Rahman et al., 2016)	Alcohol	2.85
18.	50.747	1-octadecanol	$C_{18}H_{38}O$	2053	2096.43	2070	(Boussaada et al., 2012)	Alcohol	2.25
19.	50.969	methyl linoleate	$C_{19}H_{34}O_2$	2093	2132.40	2088	(Myazawa et al., 2005)	Ester	0.89
20.	49.277	stearic acid	$C_{18}H_{36}O_2$	2167	2084.43	2170	(Khan et al., 2003)	Acid	0.98
21.	47.717	oleic acid	$C_{18}H_{34}O_2$	2175	2079.86	2161	(Mehdi et al., 2010)	Acid	4.62
22.	53.166	linoleic acid	$C_{18}H_{32}O_2$	2183	2115.54	2109	(Xiang et al., 2017)	Acid	19.80
23.	52.147	ethyl linoleate	$C_{20}H_{36}O_2$	2193	2193.59	2531	(Zhao et al., 2011)	Ester	4.87
24.	56.057	8,11-Icosadienoic acid methyl ester	$C_{22}H_{38}O_2$	2292	2307.28	–	–	Ester	0.70
25.	55.837	7,10,13-Icosatrienoic acid methyl ester	$C_{21}H_{36}O_2$	2300	2300.69	–	–	Ester	0.77
26.	57.934	hexanoic acid,4-hexadecyl ester	$C_{22}H_{44}O_2$	2311	2370.72	–	–	Ester	1.31
27.	56.393	*cis*-9-tricosene	$C_{23}H_{46}$	2315	2323.51	2269	(Dhief et al., 2011)	Alkene	0.69
28.	56.659	tetracosane	$C_{24}H_{50}$	2407	2329.22	2300	(Xiang et al., 2017)	Alkane	0.44
29.	59.201	bis(2-ethylhexyl) adipate	$C_{22}H_{42}O_4$	2414	2410.35	–	–	Ester	0.47
30.	45.196	tetracosanoic acid	$C_{25}H_{50}O_2$	2763	2735.33	2827	–	Acid	0.65
		Total	–	–	–	–	–	–	98.42

RI[a]:refers to the retention index identified by Database NIST 05;
RI[b]:refers to the retention index calculated from the retention time relative to that of C_8–C_{40} *n*-alkanes;
RIc:refers to the retention index searched from the literature;
EOQMB refers to the essential oils from the *Quercus mongolica* bark.

3.2. Antioxidant activity

As shown in Fig. 2A, the ability of EOQMB to scavenge DPPH radical increased with concentration within a certain range. The IC_{50} value of EOQMB for DPPH radical was 8.48 mg/mL, and that of Vc for DPPH radical was 0.01 mg/mL.

In Fig. 2B, the ABTS radical scavenging ability of EOQMB increased with concentration within a certain range. The IC_{50} value (0.77 mg/mL) of EOQMB was second only to that of Vc (0.05 mg/mL).

The hydroxyl radical scavenging was another experiment used to evaluate the antioxidant activity. As shown in Fig. 2C, the ability of EOQMB to scavenge hydroxyl radicals increased with concentration within a certain range. The IC_{50} value (3.54 mg/mL) of essential oil was larger than the IC_{50} value (0.03 mg/mL) of Vc.

The experimental results showed that EOQMB had an antioxidant effect on DPPH radical, ABTS radical, and hydroxyl radical, and had the strongest effect on ABTS radical. Different methods for determining antioxidant activity are available, and each method depends on lipophilic/hydrophilic balance and oxidant/antioxidant models used. Multiple detection is highly desirable when performing antioxidant testing on essential oils (Ray et al., 2018). The whole antioxidant effect can be related to fatty acids in EOQMB. Pentadecanoic acid (Sharma et al., 2016), linoleic acid (Santos et al., 2017), oleic acid (Xiang et al., 2017), and palmitic acid (Lv et al., 2014)

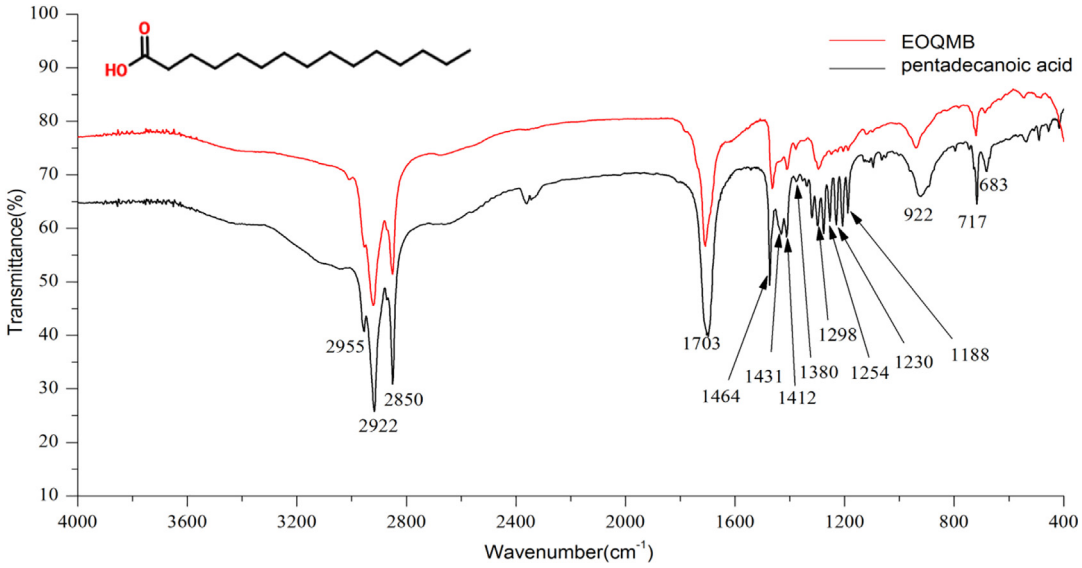

Fig. 1 The infrared spectrum of EOQMB and pentadecanoic acid.

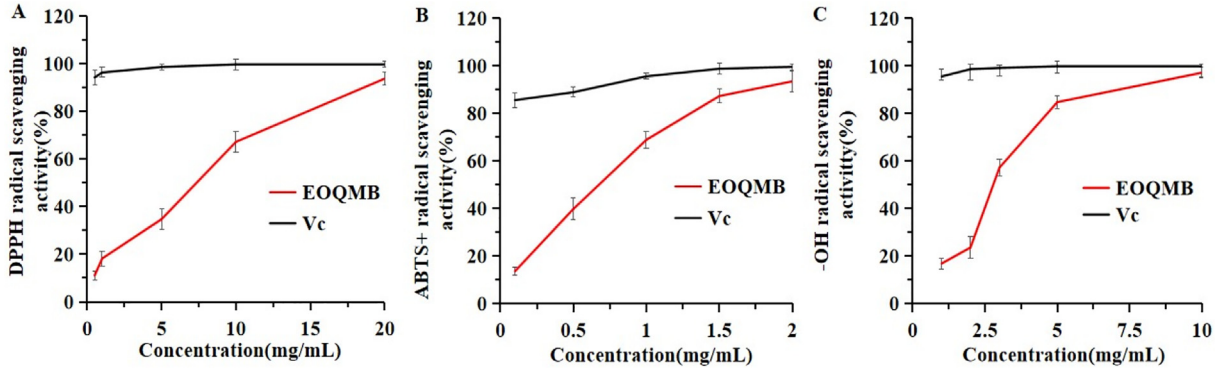

Fig. 2 Radical scavenging activity (%) of *Q. mongolica* bark essential oil; A, DPPH: Diphenyl picryl hydrazinyl radical; B, ABTS+: 2,2-azinobis 3-ethylben-zothiazoline-6-sulfonic acid; C,OH–: Hydroxyl radical.

have considerable antioxidant activity. Given that the GC–MS analysis showed that >60% of EOQMB components were fatty acids, the antioxidant activity of EOQMB may be due to fatty acids.

As shown in Fig. 3, the oxidative stress produced by vascular endothelial cells accelerated the release of tissue factor and interleukin-6, thereby promoting fibrin formation and initiating coagulation (Aizawa et al., 2015). Antioxidants inhibit the release of reactive oxygen species to protect vascular endothelial cells from oxidative damage (Fre, 1999).

3.3. Anticoagulant action of EOQMB

As shown in Fig. 3, coagulation is a series of enzymatic reactions that are activated by its former related factor and ultimately produces thrombin and fibrin. The results of the anticoagulation experiments of EOQMB and pentadecanoic acid are shown in Fig. 4.

As illustrated by the results of APTT assay in Fig. 4, anticoagulant activity was positively correlated with sample concentration. The APTTs of EOQMB and pentadecanoic acid groups were longer than the APTT of heparin at 3.75 mg/mL concentration ($P < 0.01$ and $P < 0.05$). Compared with the 0.9% NaCl group, EOQMB prolonged the time by threefold and showed excellent activity among the three samples at 30 mg/mL. The effect of EOQMB on APTT was attributed to pentadecanoic acid as the main component of EOQMB, which considerably prolonged APTT at 30 mg/mL and was twice that of the 0.9% NaCl group. The samples were potent anticoagulants. The mechanism of APTT prolongation may be attributed to the inhibition of endogenous factors, such as VIII, IX, XI, XII, and/or common pathways (Wang et al., 2010).

PT mainly reflects the status of an exogenous coagulation system and prolongs when congenital coagulation factors III, V, VII, and X are absent (Wei et al., 2014). Compared with the NaCl group, the PTs of EOQMB and pentadecanoic acid

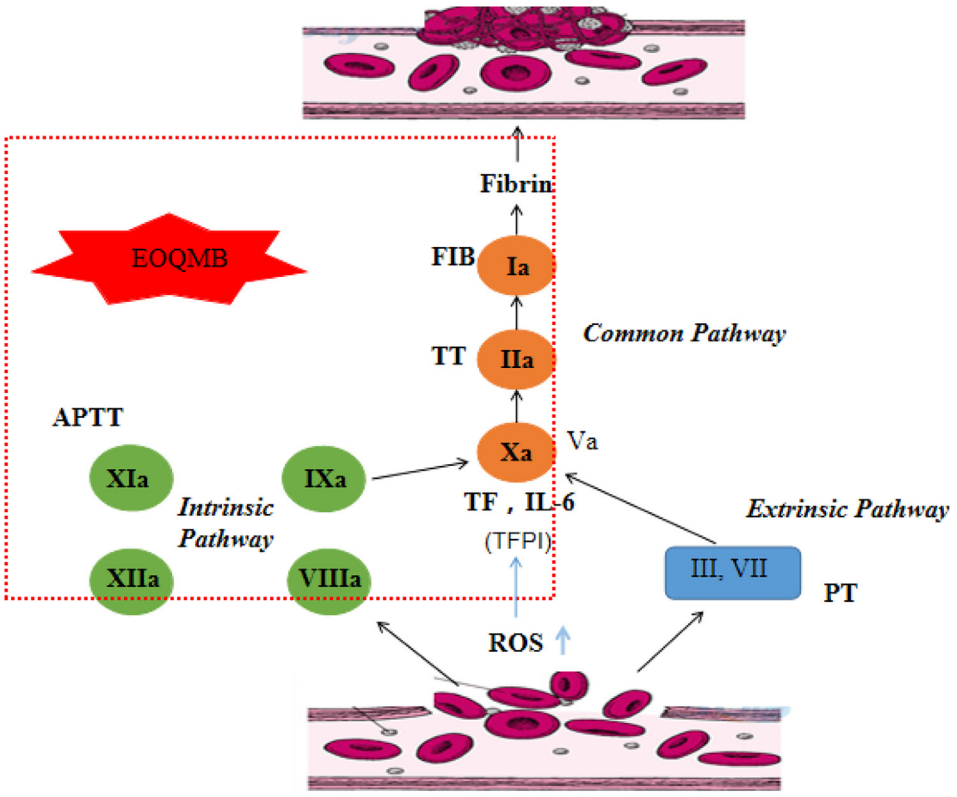

Fig. 3 Schematic diagram of anticoagulation mechanism.

showed significantly differences only when the concentration was higher than 15 mg/mL, and heparin concentration showed significant differences within the range of 3.75–30 mg/mL ($P < 0.01$). The PTs of EOQMB and pentadecanoic acid significantly differed from that of heparin within the concentration range of 1.87–30 mg/mL ($P < 0.05$ or $P < 0.01$), indicating that the inhibitions of EOQMB and pentadecanoic acid on the external coagulation pathway were weak. The fibrin clot formation activated by extrinsic pathway is considered to be a response of tissue injury (Qi et al., 2012). Several side effects, such as uncontrolled bleeding symptoms, are associated with the inhibition of coagulation by the extrinsic pathway (Pawlaczyk-Graja, 2018). Thus, the weak anticoagulation effect of EOQMB on the PT group may be considered an advantage to reduce the side effects.

The TT assay showed that the anticoagulant activities of EOQMB, pentadecanoic acid, and heparin increased with concentration. Compared with the 0.90% NaCl group, the TTs began to remarkably prolong when the concentrations of the above three samples were 3.75 mg/mL, and the results were statistically significant ($P < 0.05$, $P < 0.01$, and $P < 0.05$). The TTs of EOQMB and pentadecanoic acid were longer than the TT of heparin at 1.87 mg/mL but were statistically insignificant ($P > 0.05$). The above effects suggested that pentadecanoic acid might become an important anticoagulant ingredient in EOQMB. The TT of EOQMB at 30 mg/mL concentration was 92.32 s, and no significant difference was observed on the TT between EOQMB and heparin ($P > 0.05$, Fig. 4). TT mainly reflected the results of the time when FIB was changed to fibrin by thrombin (Wei et al., 2014), and the increments in TT suggested either impaired fibrin polymerization or thrombin inhibition (Qiu et al., 2017). The results indicated that the activities of EOQMB and pentadecanoic acid on common coagulation pathway were obvious at every concentration.

FIB mainly reflects the FIB content and plays a vital physiological action in coagulation. The standard curve of FIB was: $\lg T = -0.821 \lg C + 3.3316$, $R^2 = 0.9865$; the FIB value of EOQMB group at 30 mg/mL was one-tenth of that of the 0.90% NaCl group. Compared with the 0.90% NaCl group, the FIB values of EOQMB, pentadecanoic acid, and heparin group significantly decreased ($P < 0.01$ or $P < 0.05$). The results showed a dose-dependent manner within the concentration ranging from 1.87 mg/mL to 30 mg/mL. The FIB contents of EOQMB, pentadecanoic acid, and heparin at 30 mg/mL were 27.89 ± 6.45, 23.39 ± 8.45, and 31.96 ± 6.49 mg/dL, respectively, and the anticoagulant activities of EOQMB and pentadecanoic acid were better than the anticoagulant activity of heparin, but no statistical differences was observed ($P > 0.05$). Accordingly, the experimental results showed that EOQMB and pentadecanoic acid had a certain effect on the anticoagulation and fibrinolytic systems.

Given that the above effects of EOQMB on APTT, TT, and FIB are similar to those of heparin, EOQMB and its main component pentadecanoic acid have anticoagulant activity. Pentadecanoic acid and linoleic acid with other components in essential oil may have a synergistic anticoagulant effect (Benjamin et al., 2015; Jung-Hee et al., 2019) because a variety

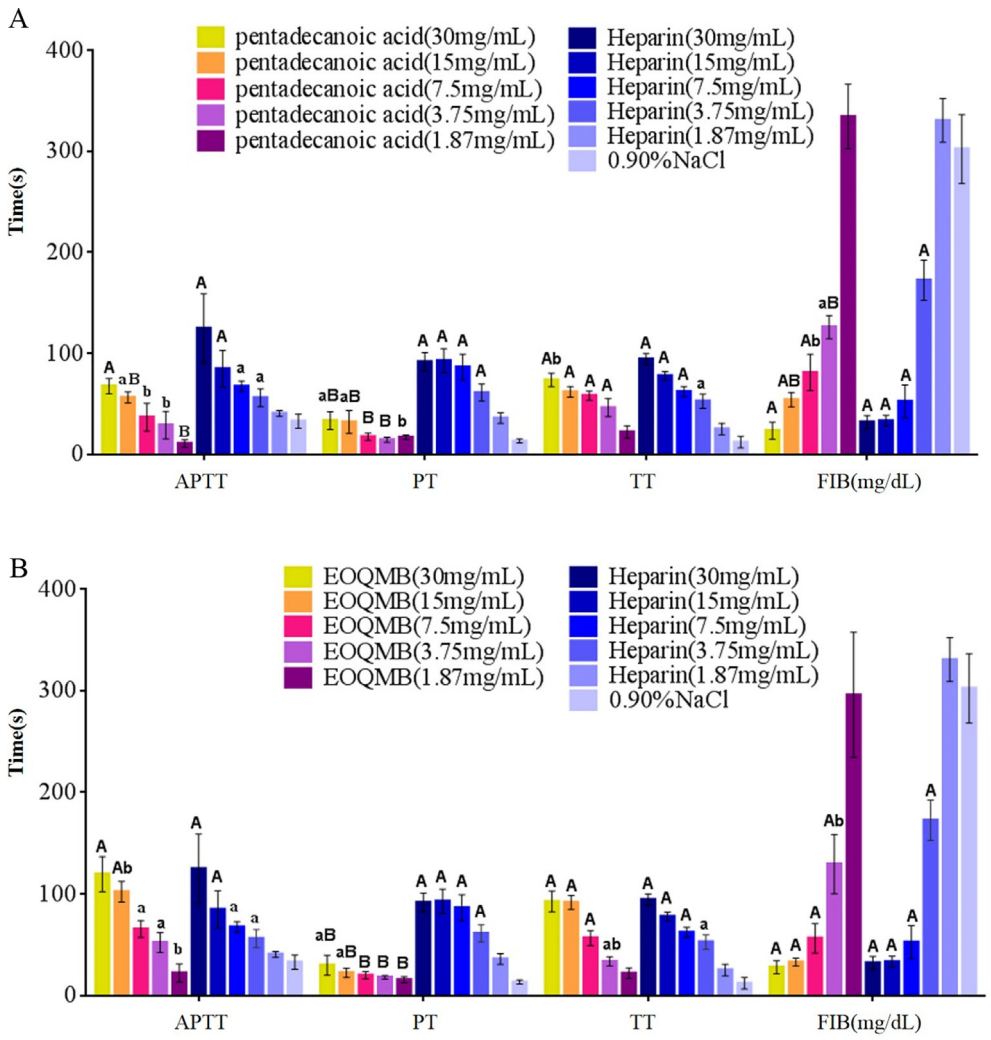

Fig. 4 Effects of EOQMB(Zeng et al.) and pentadecanoic acid(Deryabin and Tolmacheva) on plasma coagulation time measured by APTT, TT, PT and, FIB in vitro; ^{A} $P < 0.01$,or,^{a} $P < 0.05$ vs. Control group;^{B} $P < 0.01$,or, ^{b} $P < 0.05$ vs. Same concentration Heparin.

of fatty acids play an active role in human health (Itakura et al., 2011). The research results preliminarily demonstrated that pentadecanoic acid and EOQMB may inhibit the intrinsic pathways and/or common pathway or inhibit the conversion of FIB into fibrin or both.

4. Conclusions

The extraction yield of EOQMB was 0.08% ± 0.03%. Thirty components accounted were analyzed by GC–MS, and pentadecanoic acid was one of the main components accounting for 34.90%, which was further confirmed by the FT-IR observation. The IC_{50} values of EOQMB with DPPH radical, ABTS radical, and hydroxyl radical scavenging ability showed that it has good antioxidant activity but not as good as Vc. The effects of EOQMB on APTT, TT, and FIB are similar to those of heparin in vitro, and the fatty acids played a chief role in anticoagulant activity. This study proves the structure–activity relationship of EOQMB and its main components in the direction of antioxidation and anticoagulation.These findings indicate the EOQMB may act as a potential natural anticoagulant agent and provide a new idea for the medicinal development of *Q. mongolica* bark.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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