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Potential Antioxidants in Bio-oil Obtained by Pyrolysis of Yew (*Taxus baccata* L.) Tree Bark

Potencijalni antioksidansi u bioulju dobivenom pirolizom kore drva tise (*Taxus baccata* L.)

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ABSTRACT • *Bio-oil (BO), ethanol extract (EtE), and superheated ethanol extract (SHEtE) from the bark of* Taxus baccata *L. tree trunk fallen due to strong wind or landslide were obtained from pyrolysis of the crushed sample in the furnace at a temperature of 500 °C for 30 min, ordinary solvent extraction of air-dried powder sample for 24 hours using a 250 mL laboratory-scale Soxhlet extractor, and a small laboratory-scale reactor with a 50 ml cylindrical stainless steel vessel and maximum working temperatures of (300 ± 3) °C for superheated solvent extraction, respectively. The BO and extracts were analyzed for their antioxidant activities using the DPPH method. The presence of organic compounds in the BO and extracts was predicted by analyzing the functional groups using Fourier transform infrared (FTIR) spectroscopy. Among the BO and extracts from yew bark, the most active fraction was analyzed for its chemical composition using gas chromatography-mass spectrometry (GC/MS). BO was proven to have a higher antioxidant capacity (58.76 %) compared to the standard ascorbic acid (45.14 %) at a concentration of 1000 ppm. The FTIR analysis suggested the presence of active compounds in BO samples. The quantification of the BO and extracts showed that yew bark BO was richer in phenolic compounds than the extracts. The major components identified in the BO by GC/MS analysis were guaiacol (11.65 %), followed by phenol (8.48 %), β-D-Glucopyranose, 1,6-anhydro- (6.54 %), furfuryl alcohol (5.64 %), 3,4-dihydro-2H-pyran (5.52 %), p-cresol (4.50 %), 4-methylguaiacol (4.24 %), 3-methylcyclopent-2-enone (3.45 %), 2-hydroxy-3-methylcyclopent-2-en-1-one (3.29 %), and catechol (3.23 %).*

KEYWORDS: Taxus baccata *L.; bark bio-oil; chemical composition; antioxidant activity; GC/MS and FTIR*

SAŽETAK • *U radu su opisana istraživanja antioksidativne aktivnosti bioulja (BO), etanolnog ekstrakta (EtE) i pregrijanoga etanolnog ekstrakta (SHEtE) dobivenih iz kore debla* Taxus baccata *L., srušenoga zbog jakog vjetra ili klizišta. Bioulje je dobiveno pirolizom usitnjenog uzorka u peći, na temperaturi od 500 °C tijekom 30 minuta. Etanolni ekstrakt dobiven je iz uzorka praha kore osušene na zraku i ekstrahiranoga otapalom tijekom 24 sata uz pomoć laboratorijskog Soxhlet ekstraktora od 250 mL. Pregrijani etanolni ekstrakt dobiven je primjenom maloga laboratorijskog reaktora s cilindričnom posudom od nehrđajućeg čelika volumena 50 mL i maksimalnim radnim temperaturama od (300 ± 3) °C za ekstrakciju pregrijanim otapalom. Antioksidativna aktivnost bioulja i ekstraka-*

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ta analizirana je DPPH metodom. Prisutnost organskih spojeva u bioulju i ekstraktima utvrđena je analizom funkcionalnih skupina primjenom Fourierove infracrvene spektroskopije (FTIR). Kemijski sastav najaktivnije frakcije dobivene od bioulja i ekstrakata iz kore drva tise analiziran je uz pomoć plinske kromatografije s masenom spektrometrijom (GC/MS). Dokazano je da bioulje pri koncentraciji od 1000 ppm ima veći antioksidativni kapacitet (58,76 %) od standardne askorbinske kiseline (45,14 %). FTIR analizom potvrđeno je postojanje aktivnih spojeva u uzorcima bioulja. Kvantifikacija bioulja i ekstrakata iz kore drva tise pokazala je da je bioulje bilo bogatije fenolnim spojevima od ekstrakata. Glavne komponente identificirane GC/MS analizom u bioulju bile su gvajakol (11,65 %), fenol (8,48 %), β-D-glukopiranoza, 1,6-anhidro-(6,54 %), furfurilni alkohol (5,64 %), 3,4-dihidro-2H-piran (5,52 %), p-krezol (4,50 %), 4-metilgvajakol (4,24 %), 3-metilciklopent-2-enon (3,45 %), 2-hidroksi-3-metilciklopent-2-en-1-on (3,29 %) i katehol (3,23 %).

KLJUČNE RIJEČI: *Taxus baccata L.; bioulje iz kore; kemijski sastav; antioksidativna aktivnost; GC/MS; FTIR*

1 INTRODUCTION
1. UVOD

A living system and organisms can produce a broad spectrum of reactive oxygen/nitrogen/chlorine species (RO/N/CS) and free radicals that play an essential role in regulating numerous physiological and metabolic functions of the body, where their reactivity and diffusibility are varied (Halliwell, 2003; Rajamanikandan *et al*., 2011). Hence, the excessive generation of RO/N/CS in living organisms can defeat these antioxidant defense mechanisms, give rise to oxidative stress (Mandal *et al*., 2011), cause damage to different biomolecules such as lipids, proteins, carbohydrates, and nucleic acids, and induce several types of degenerative diseases, for example, cancer, aging, atherosclerosis, cardiovascular disorders, neurodegenerative disorders, and inflammation (Finkel and Holbrook, 2000; Valko *et al*., 2007; Thitilertdecha *et al*., 2008). Antioxidant defense mechanisms of organisms directly scavenge or prevent the generation of RO/N/CS to protect living systems against their adverse effects (Rajamanikandan *et al*., 2011).

Forming complex organic compounds to restrain the accessibility of easily utilizable substrates, i.e., sugar and starches, is a defense strategy for trees to prevent their susceptibility to degradation (Scott Reading *et al*., 2003). The bark of trees works as a protective barrier against extreme weather conditions, insect pests, termites, and fungi, and it also has a significant role in wood fires (Hosseinashrafi *et al*., 2023). The softwood bark is composed of cellulose (18-38 %), hemicellulose (15-33 %), and lignin (30-60 %) (Díez *et al*., 2020).

Bio-oils from the bark have been used as protective substances obtained by pyrolytic methods (Singh and Singh, 2012). Phenols were major products in the bio-oil distillate from softwood lignin obtained by pyrolysis (Larson *et al*., 2017). Pyrolysis is an efficient technology in which lignin is converted into valuable chemicals, enhancing its valorization. Pyrolysis can yield phenolic compounds and/or aromatic hydrocarbons (AHs), demonstrating potential applications as biochemical intermediates and bio-fuel additives (Lu and Gu, 2022). When using bark as a source of biopesticide production, it must be taken in consideration that the bio-reactivity of bark extracts from different sources can differ (Singh and Singh, 2012).

Also, plants and their products have been shown to employ antioxidant activity against RO/N/CS, allowing their practical use as food preservatives, dietary supplements, and food flavoring agents (Huang *et al*., 2005; Lee *et al*., 2011).

Taxus baccata L. (European yew) is a member of the family Taxaceae, a native evergreen non-resinous gymnosperm that can grow up to 20-28 m and widely distributed in Europe and Asia (Thomas and Polwart, 2003). *Taxus baccata* L. is one of the four conifers indigenous to the Hyrcanian forests of Iran. This rare, native, old, and endangered tree species has recently been scrutinized due to its addition to the red list of threatened species (Alavi *et al*., 2019; Rezaei Karmozdi *et al*., 2022).

Sadeghi Aliabadi *et al*. (2009) examined different solvent-based extraction systems from the needles of yew. Their results indicated that the extract obtained with 100 % acetone contains the highest amount of taxol and other taxanes as well as alcohols, phenolic acids, simple sugars, disaccharides (sugar), free fatty acids, and resins found in the yew tree bark as the main compounds.

To the best of the authors' knowledge, the chemical composition of the bark of the yew (*Taxus baccata* L.) BO has yet to be reported. However, the chemical composition of the extract of this part has already been reported. Moreover, this novel approach investigated the antioxidant activity of its BO, EtE, and SHEtE and compared them with standard ascorbic acid (AA). This study investigated the antioxidant activity of the bark bio-oil of yew, which has not been reported before.

2 MATERIALS AND METHODS
2. MATERIJALI I METODE

2.1 Plant material
2.1. Biljni materijal

In the present study, three wooden discs with a thickness of 0.5 meters were prepared from the breast height of three yew (*Taxus baccata* L.) tree trunks fallen due to strong wind or landslide. The fallen trunks were collected from Afratakhteh forests in Golestan province, Iran, in October 2017 (autumn). The bark of prepared discs was separated and poured into a glass jam jar with lids and then kept in a refrigerator at 4 ºC until pyrolysis and extraction. The plant material with voucher specimen 4657 was identified by Sayed Khosrow HossinAshrafi (Assistant Professor) and deposited to the Herbarium in the College of Agricultural and Natural Resources, Karaj Branch, Islamic Azad University, Karaj, Iran.

In this work, for the extraction of valuable compounds from the bark of the yew tree, three methods have been employed: pyrolysis, solvent extraction using a Soxhlet extractor, and superheated solvent extraction.

Prior to pyrolysis and extraction operations, yew bark samples were oven-dried at (103±2) ºC for 1 hour. The samples had a mass loss of 20 % due to the oven-dry procedure.

2.2 Experimental setup for pyrolysis
2.2. Eksperimentalna oprema za pirolizu

A small laboratory-scale electrical furnace was used to pyrolyze *Taxus baccata* dry bark. A schematic diagram of the furnace used in this study is shown in Figure 1.

This apparatus used a tubular ceramic core (5 cm ID and 30 cm length) equipped with an electrical heater (500 W) with a controlling temperature range of 100 to 600 ± 5 °C. The batch experiment was conducted in a closed system at the constant N_2 flow rate of 50 cm^3/min. For conducting pyrolysis experiments, about 20 grams of the crushed sample were placed in the furnace, with both ends sealed by mineral wool. The samples were then pyrolyzed in the furnace at a temperature of 500 °C for 30 min. During the process, nitrogen gas was used as the carrier gas at a flow rate of 50 cm^3/min.

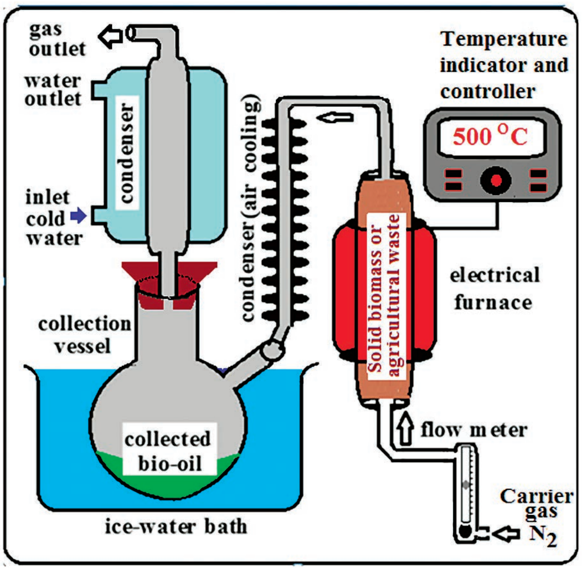

Figure 1 A schematic diagram of a small laboratory-scale electrical furnace (Bagi and Baseri, 2021)
Slika 1. Shematski dijagram male laboratorijske električne peći (Bagi i Baseri, 2021.)

During the pyrolysis process, the used carrier gas carried out the lighter products, and in the condensers, they were condensed into the liquid BO. However, the uncondensed gases were discharged into the atmosphere. The condensed BO was collected in the collection vessel at about 0 °C.

At the end of the experiment, the system was discharged, and the collected BO was weighed and stored in the closed vessel in the refrigerator until their compositions were characterized. Moreover, the solid residue was weighed, and the yields of the products (weight percentages) were calculated using Eq 1 (Bagi and Baseri, 2021; Raeisian *et al*., 2025).

$$Yield\ of\ product\,(\%) = \frac{M_2}{M_1} \times 100 \qquad (1)$$

Where M_2 and M_1 denote the weight of BO and the weight of biomass (dry feed), respectively.

2.3 Solvent extraction and superheated solvent extraction
2.3. Ekstrakcija otapalom i ekstrakcija pregrijanim otapalom

High-purity ethanol (99 %) was selected as the extraction solvent in the ordinary and superheated solvent extraction experiments. For the ordinary solvent extraction, 10 g of air-dried powder sample was placed into a thimble and was extracted for 24 hours using a 250 mL laboratory-scale Soxhlet extractor. After the extraction process, the solvent was removed under reduced pressure in a rotary evaporator, and the remaining extracts were weighed and stored in the closed flask in the refrigerator (Jayawardena and Smith, 2010; Vibala *et al*., 2020; Smith, 2022; Yu *et al*., 2023).

A small laboratory-scale reactor was used for superheated solvent extraction to extract *Taxus baccata* air-dried bark powder. A schematic diagram of the used reactor is shown in Figure 2.

In this system, a 50 mL cylindrical stainless steel vessel (18 mm ID and 200 mm length) is used as the extraction vessel, and the electrical heaters (500 W) with maximum working temperatures of (300 ± 3) °C to heat the vessel. The pressure of the vessel is increased by adding N_2 gas from the gas cylinder up to 4 MPa, and it is monitored by pressure indicators.

For a typical experiment, 5 g of air-dried powder sample is charged in the vessel, and 15 mL of solvent is added to the system. Then, the reactor is closed, and the air is discharged by purging of N_2 gas in the reactor. The pressure of the reactor increases to 7 bar by adding N_2 gas, which is controlled by a pressure control valve. Temperature of the system is adjusted to 110 (± 1 %) °C. To pre-

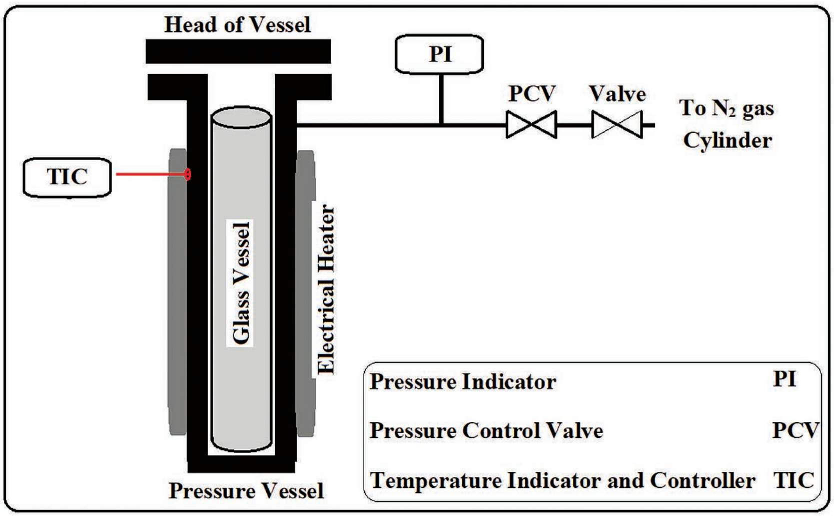

Figure 2 A schematic diagram of reactor used for superheated solvent extraction process (Mohseni and Baseri, 2022)
Slika 2. Shematski dijagram reaktora upotrijebljenog za proces ekstrakcije pregrijanim otapalom (Mohseni i Baseri, 2022.)

vent solvent evaporation, the system pressure is adjusted to values higher than the equilibrium vapor pressure of the extraction solvent in the system temperature.

At the end of the experiment, the reactor is cooled to room temperature, and the pressure is decreased by the constant rate of 15 bar/hour. Then, the reactor is opened, and the solid and liquid phases are separated by centrifuging (*RPM*=4000 and *t*=5 min). To calculate the total yield of extract, the remaining solid phase is dried at 100 °C for 1 hour and weighed.

Finally, the solvent is separated by a rotary evaporator under the reduced pressure, and the remaining extract is weighed and stored in the closed flask.

2.4 FTIR analysis bark bio-oil and extracts
2.4. FTIR analiza bioulja i ekstrakata iz kore drva tise

The functional groups of the yew bark BO and extracts were studied by Fourier transform infrared spectroscopic analysis with a Unicam 4600 FTIR spectrometer (Mattson, USA) over a scanning wave range of 400 to 4000 cm^{-1} (Wu *et al*., 2009; Packialakshmi and Naziya, 2014).

2.5 Antioxidant activity of yew bark bio-oil and extracts
2.5. Antioksidativna aktivnost bioulja i ekstrakata iz kore drva tise

The antioxidant potential of the yew bark BO and extracts was evaluated using a 1,1-Dipheny-2-picrylhydrazyl (DPPH) assay, according to the method described in the literature (Halliwell, 1997; Kim *et al*., 2002; Emami *et al*., 2010).

In short, 2.5 mL of a 0.1 mM DPPH solution in 70 % methanol (stock solution) was added to different concentrations (100 µL) of the BO, extracts, and ascorbic acid (AA) as the positive control. Serial dilutions were made from the stock solution and the working solution of the tested bark BO and solvent extracts to obtain concentrations of 1000, 500, 250, 125, 62.5, 31.25, 15.62, and 7.8 ppm. The preparation process of the working solution was conducted according to a previously reported method (Hosseinihashemi and Aghajani, 2017; Barzegari *et al*., 2023).

The control samples (DPPH solution) were prepared using an equal volume of solution without any test compounds or referenced standards, serving as the negative control (Pillai *et al*., 2019; Alam *et al*., 2021). Pure methanol (Sigma-Aldrich, Darmstadt, Germany) was used to prepare the control sample. Before analysis, the UV scanning spectrophotometer device (JENWAY 6320D, Standford, UK) was calibrated and adjusted using 70 % methanol. The reaction mixture was mixed for 10 s and left to stand at room temperature in a dark place for 30 min. The absorbance was measured at 517 nm, using a UV scanning spectrophotometer (Hosseinihashemi *et al*., 2017).

2.6 GC/MS analysis of yew bark bio-oil
2.6. GC/MS analiza bioulja iz kore drva tise

The yew bark BO was analyzed by GC/MS using a GC Agilent 7890A and MS Agilent 5975C mass spectrometer detector (Agilent Technologies, Palo Alto, CA, USA) equipped with an HP-5MS cross-linked capillary column (30 m long and 0.25 mm internal diameter, 0.25 µm film thickness).

100 µL of yew bark BO sample was dissolved separately with 900 µL of hexane. Samples (1 µL) were injected in the split mode (50:1) with a flow rate of 1 mL/min. The column temperature program was conducted as follows: injector temperature of 260 °C; transfer line of 270 °C; oven temperature program of 60 °C for 4 min, 3 °C/min to 100 °C for 2 min, then 4 °C/min to 250 °C for 5 min; carrier gas was He with a flow rate of 1 mL/min. The intrinsic energy that hits the

sample in the MS system was 70 eV. The total chromatographic run time was about 52 min.

The identification of yew bark BO components was carried out by comparing their mass spectra with those available on a NIST database mass spectral library, with spectra from literature (Joulain and Konig, 1988; Adams, 2001), as well as retention index and literature retention indices values calculated for each detected BO composition.

2.7 Statistical analysis
2.7. Statistička analiza

The results are given in mean values with their standard deviations. Statistical analysis was performed using the SPSS program, version 24.0 (International Business Machines (IBM) Corp., Armonk, NY, USA). Univariate analysis of variance (ANOVA) was conducted to determine the significance of differences between analytical results at $p<0.05$ significance level.

3 RESULTS AND DISCUSSION
3. REZULTATI I RASPRAVA

3.1 FTIR of yew bark bio-oil and extracts
3.1. FTIR bioulja i ekstrakata iz kore drva tise

Figure 3 depicts the FTIR spectrums of the components extracted with the ethanol, superheated ethanol, and the BO produced by the pyrolysis process. As can be seen, the spectrums of samples extracted by ethanol and superheated ethanol are relatively the same, with a slight increase in the intensity of some peaks. However, significant changes are observed in the BO produced by the pyrolysis process.

The main chemical groups in the ethanol extracts and the produced BO are as follows: the broad peak at about 3400 cm^{-1} represented the O-H stretch and H-bonded groups that may suggest the presence of phenols, alcohols, carboxylic acids, or water molecules. As can be seen, this peak strongly appeared in three samples studied.

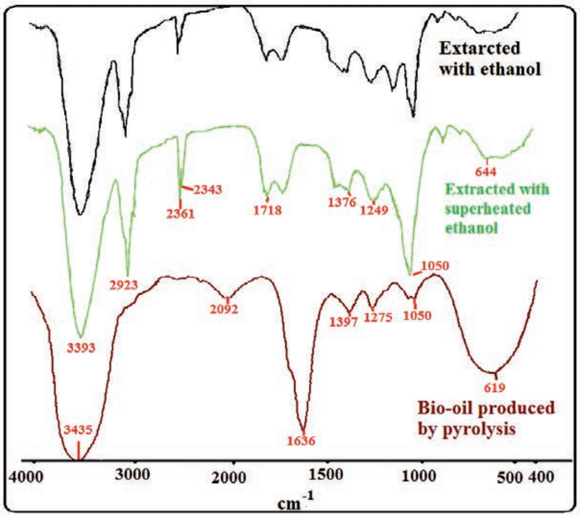

Figure 3 FTIR spectrums of components extracted with EtE, SHEtE, and BO produced by pyrolysis process
Slika 3. FTIR spektri ekstrakata EyE, SHEtE i bioulja proizvedenoga procesom pirolize

The absorption peak at about 2923 cm^{-1} was related to the C-H (-CH$_3$) stretching vibration of cycloalkanes and aliphatic hydrocarbons (Zhou *et al*., 2022). However, the binding vibration of –CH$_3$ groups was observed in about 1375 cm^{-1}.

The peaks that appeared in 1718 and 1450 cm^{-1} were caused by carbonyl groups (C=O) in carboxylic acids, ketones, or aldehydes (Zhou *et al*., 2022). In addition, the –C-O stretching and vibration of the –CH skeleton were depicted between 1267 and 1023 cm^{-1}. For the infrared spectra of BO, the broad peak bound at 1634 cm^{-1} may confirm the presence of aromatic functional groups or show the presence of amide I components in the produced BO (Kalisz *et al*., 2021). The broad peak in about 500 to 800 cm^{-1} can be assigned to benzene rings or vibration absorption of alkenes (Li *et al*., 2013).

3.2 Antioxidant activity of yew bark bio-oil and extracts
3.2. Antioksidativna aktivnost bioulja i ekstrakata iz kore drva tise

The yield of EtE and SHEtE from the bark of *T. baccata* was 10.3 % and 12.4 %, respectively. Pyrolysis of *T. baccata* gave dark BO with a yield of 17.41 mL/100 g, based on the dry weight of the bark.

The antioxidant activity of BO and extracts from the bark of *T. baccata* at eight different concentrations of 1000, 500, 250, 125, 62.5, 31.25, 15.62, and 7.8 ppm was measured in the present study by using the DPPH method and compared with AA as the reference standard (Table 1 and Figure 4). Statistically, substantial differences were observed among the treatments, including BO, EtE, SHEtE, AA, and their respective concentrations.

The data in Table 1 and Figure 4 show a higher antioxidant activity when tested at higher concentrations. The BO from yew bark exhibited excellent antioxidant activity overall compared to other extracts. The lowest antioxidant activity (-0.1 %) was observed in the EtE at the concentration of 7.8 ppm, which was lower than the value of AA (4.7 %) at the same concentration. The highest antioxidant activity was found in bark BO (58.7 %) at 1000 ppm, which was higher than the value of AA (45.1 %) at a similar concentration. The same trends were observed between the reference (AA) and BO after 250 ppm. Still, the antioxidant activity of the EtE and SHEt extracts has increased very slightly or remained constant with increasing concentration. There was a significant decrease in the concentration of DPPH radical as the concentration of BO increased due to its ability to scavenge free radicals

Table 1 Percentage of antioxidant activity (Mean ± SD) as affected by concentration of bark BO and extracts of *T. baccata* compared with AA
Tablica 1. Postotak antioksidativne aktivnosti (srednja vrijednost ± SD) u odnosu prema koncentraciji bioulja i ekstrakata iz kore drva *T. baccata* u usporedbi s AA

Treatment AOA, % *Tretman AOA, %*	**Concentration** / *Koncentracija*							
	1000	500	250	125	62.5	31.25	15.78	7.8
BO	58.76 ± 3.05 m	43.53 ± 1.69 l	12.98 ± 0.51 i	5.44 ± 0.19 bcd	0.90 ± 1.07 a	0.59 ± 0.92 a	0.07 ± 0.79 a	0.21 ± 1.32 a
EtE	13.14 ± 0.44 i	12.78 ± 0.91 i	8.96 ± 2.55 efg	2.65 ± 0.74 abc	2.43 ± 2.20 abc	1.32 ± 0.46 a	1.13 ± 0.97 a	-0.10 ± 1.62 a
SHEtE	18.86 ± 0.95 j	13.20 ± 4.76 i	12.19 ± 2.42 hi	7.72 ± 1.49 defg	1.19 ± 3.51 a	0.52 ± 0.83 a	0.66 ± 1.25 a	1.92 ± 1.21 ab
AA	45.14 ± 4.95 l	33.95 ± 0.49 k	9.86 ± 1.74 gh	9.23 ± 1.01 fg	6.68 ± 2.31 def	5.04 ± 0.37 cd	6.13 ± 1.40 de	4.69 ± 1.73 bcd

*Different letters in columns and rows indicate a statistical difference ($p<0.05$) among the treatment groups
**Različita slova u stupcima i redcima označavaju statističku razliku (p<0,05) među ispitivanim skupinama.*

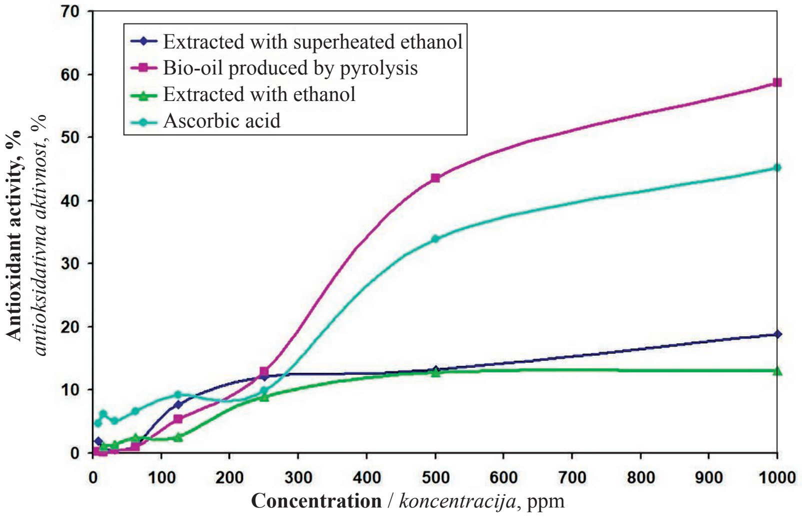

Figure 4 Antioxidant activity of components extracted with the EtE, SHEtE, and BO produced by pyrolysis process
Slika 4. Antioksidativna aktivnost ekstrakata EyE, SHEtE i bioulja proizvedenoga procesom pirolize

(Patra *et al*., 2015). The scavenging activity of BO is attributed to its ability to donate hydrogen ions or electrons to DPPH to neutralize free radicals (Rajamanikandan *et al*., 2011).

3.3 Chemical composition of yew bark bio-oil

3.3. Kemijski sastav bioulja iz kore drva tise

GC-MS analysis showed the chemical composition of volatile compounds in yew bark BO. The chemical profiles of yew bark BO have not been previously reported. Figure 5 shows the chromatogram of the BO of *T. baccata* bark obtained in October 2017 (autumn) at Afratakhteh forest station and it is analyzed in the HP-5MS column.

The identified constituents are presented in Table 2, where all compounds are listed in order of their elution from the HP-5 column. In total, 44 compounds comprising 98.75 % of the volatile compounds in the BO were selected and identified (Table 2) in this study.

The components of BO are mainly comprised of aldehydes, alcohols, acids, ether, phenols, and phenol derivatives. Among them, the top-seven most abundant compounds are guaiacol (11.65 %), phenol (8.48 %), β-D-Glucopyranose, 1,6-anhydro- (6.54 %), furfuryl alcohol (5.64 %), 3,4-dihydroxyphenol-*2H*-pyran (5.52 %), *p*-cresol (4.50 %) and 4-methylguaiacol (4.24 %).

In GC/MS analysis of the first harvest of biomass samples from *T. canadensis* shrubs, some compounds were identified such as phenol, 4-vinylphenol, 4-hydroxybenzaldehyde, 3,5-dimethoxyphenol, guaiacol, isoeugenol, and an anhydrosugar as some of the primary components of the pyrolysis oil based on published spectra (Cass *et al*., 2001). Jean *et al*. (1993) had previously identified 3,5-dimethoxyphenol as a major aglycone in *T. canadensis*; thus, its presence was ex-

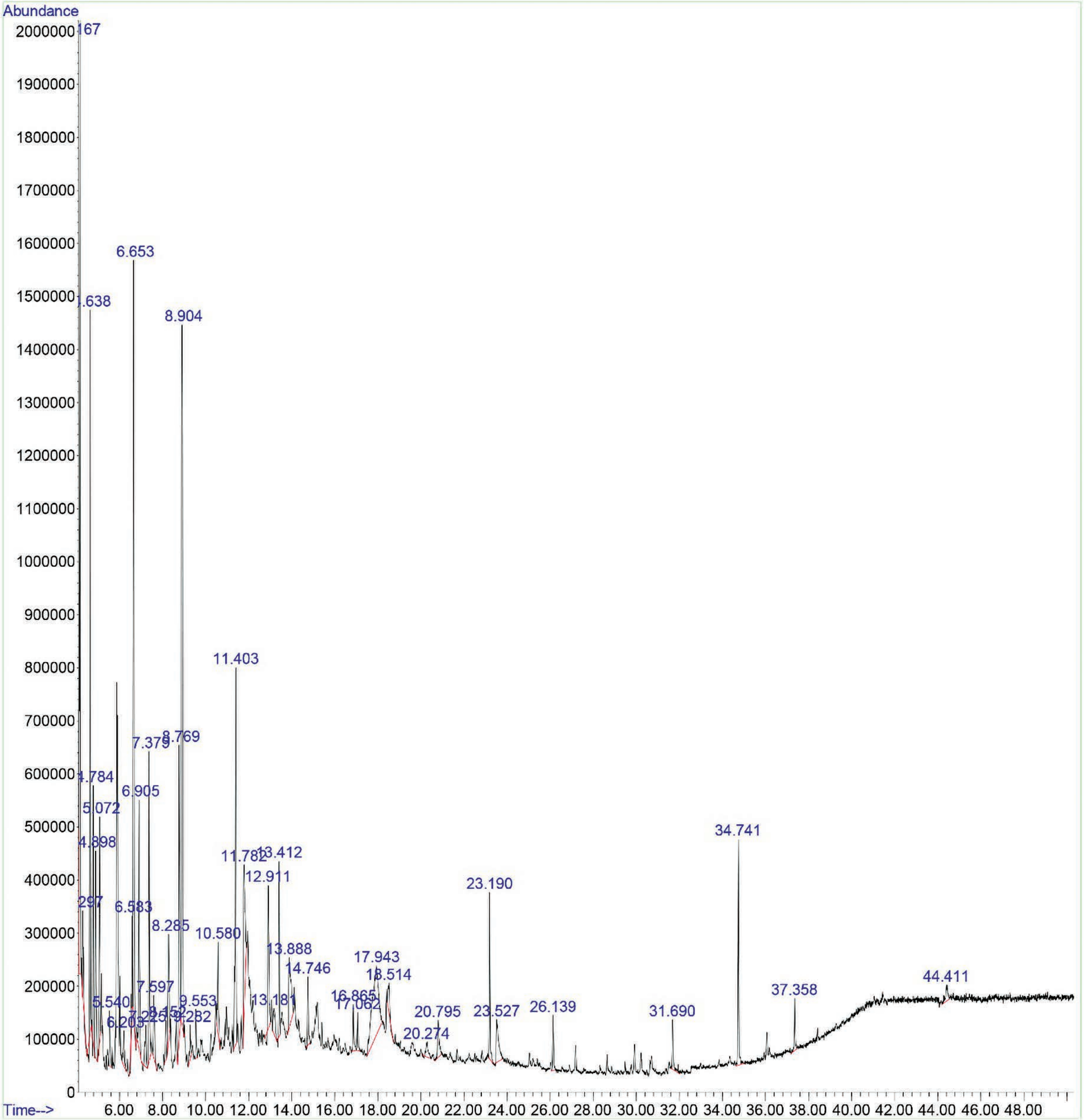

Figure 5 Chromatogram of BO from *T. baccata* bark harvested in October 2017 (autumn). Components: furfuryl alcohol (4.165 min), 3,4-dihydro-*2H*-pyran (4.637 min), phenol (6.650 min), *p*-cresol (8.767 min), guaiacol (8.902 min), and 4-methylguaiacol (11.403 min), *β*-D-Glucopyranose, 1,6-anhydro- (17.941 min)
Slika 5. Kromatogram bioulja iz kore *T. baccata*, srušene u listopadu 2017. (jesen). Sastojci: furfurilni alkohol (4,165 min), 3,4-dihidro-2H-piran (4,637 min), fenol (6,650 min), p-krezol (8,767 min), gvajakol (8,902 min) i 4-metilgvajakol (11,403 min), *β*-D-glukopiranoza, 1,6-anhidro- (17,941 min)

pected. Eugenol has also been identified in the essential oil and in the aglycone component of *T. canadensis* (Jean *et al*., 1993).

Among the identified compounds, some components such as phenol, *o*-cresol, and *p*-cresol in 2.19 %, 2.79 %, and (6.02 %) amounts were found in the bio-oil of Japanese red pine sawdust, respectively (Patra *et al*., 2015). Guaiacol, syringol, and 4-methylguaiacol in 3.60 %, 14.86 %, and 3.08 % amounts as well as in 1.26 %, 4.21 %, and 1.32 % amounts were also identified in the bio-oil from pyrolysis of undecayed and decayed *Fagus orientalis* wood, respectively (Hosseinihashemi *et al*., 2017).

According to the researchers' and our findings, the types of compounds in the essential oil and bio-oil of the yew plant are different from each other, but the class of some compounds may be the same (Radulović *et al*., 2011; Jian-chun *et al*., 2012; Yasar, 2013; Stefanović *et al*., 2016; Zhao *et al*., 2016; Wei and Yin, 2019; Huong *et al*., 2020; Benlembarek *et al*., 2021).

T. baccata of Algeria essential oil showed a moderate antioxidant activity (Benlembarek *et al*., 2021). In Algeria, Bekhouche *et al*. (2021) showed that the methanolic extract of *T. baccata* needles exhibits a greater antioxidant activity than the synthetic antioxidant BHT. The essential oil from the stems of *T. cuspi-*

Table 2 Bio-oil composition of *T. baccata* bark harvested in October 2017 (autumn)
Tablica 2. Sastav bioulja iz kore *T. baccata*, stabla srušena u listopadu 2017. (jesen)

No.	Compound / *Spoj*	*RT*, min	Area, %	KIL_{exp}	KI_{lab}
1	Acetoxypropanone	4.134	1.46	856	839
2	Furfuryl alcohol	4.165	5.64	861	866
3	3-Methylpyridine	4.3	0.88	882	-
4	3,4-Dihydro-*2H*-pyran	4.637	5.52	912	705
5	2-Methylcyclopent-2-enone	4.782	1.96	920	893
6	2-Acetylfuran	4.896	2.15	927	902
7	Diisoamylether	5.073	1.97	936	-
8	2,4-Dimethylpyridine	5.54	0.69	959	933
9	5-methylfurfural	5.872	2.86	974	934
10	3-Methylcyclopent-2-enone	5.908	3.45	976	935
11	4-oxo-5-methoxy-2-penten-5-olide	6.028	0.50	981	-
12	1-Methyl-2-furoate	6.204	0.58	988	956
13	2-(1-Methylheptyl)-cyclopentanone	6.583	0.80	1005	1128
14	Phenol	6.65	8.48	1008	1002
15	Mepivacaine metabolite	6.904	2.94	1020	-
16	2-Hexene, 2-methyl-	7.226	0.73	1035	-
17	2-Hydroxy-3-methylcyclopent-2-en-1-one	7.382	3.29	1042	1015
18	2,3-Dimethylcyclopent-2-enone	7.6	1.08	1051	1018
19	*o*-Cresol	8.285	1.86	1079	1058
20	*p*-Cresol	8.767	4.50	1097	1090
21	Guaiacol	8.902	11.65	1103	1092
22	Maltol	9.281	0.53	1120	1093
23	3-Ethyl-2-hydroxycyclopent-2-en-1-one	9.556	0.58	1133	1100
24	Piperidine	10.578	0.80	1176	-
25	1,4:3,6-Dianhydro-*α*-d-glucopyranose	11.336	0.95	1206	-
26	4-Methylguaiacol	11.403	4.24	1209	-
27	Catechol	11.782	3.23	1227	-
28	3-Methoxycatechol	12.913	2.11	1276	1239
29	3-Methylcatechol	13.183	0.76	1287	1263
30	2-Methoxyphenethyl alcohol	13.411	1.73	1296	1309
31	4-Methylcatechol	13.888	2.22	1319	1246
32	Syringol	14.744	0.91	1358	1347
33	Dehydroacetic acid	16.867	0.67	1459	-
34	Isoeugenol	17.064	0.48	1469	1436
35	*β*-D-Glucopyranose, 1,6-anhydro-	17.941	6.54	1511	1491
36	2,3,5-Trimethoxytoluene	18.511	0.59	1540	1538
37	Propanamide, N-acetyl-	20.276	0.49	1630	-
38	Homovanillic acid	20.794	0.88	1658	1657
39	3,5-Diethoxycarbonyl-2,6-dimethylpyridine	23.192	1.88	1790	-
40	4-(1-Hydroxyethyl)benzaldehyde	23.529	1.42	1807	-
41	3,5-Diethoxycarbo	26.139	0.72	1945	-
42	2-ethylhexyl-3-(4-methoxyphenyl)acrylate	31.691	0.79	2341	-
43	Di(ethylhexyl) phthalate	34.741	2.72	2573	2509
44	Hexamethyl-cyclotrisiloxane	44.413	0.52	3356	-

RT – Mean retention time; KI_{exp} and KI_{lit} – experimental and literature Kovats indices, respectively, on HP-5MS column in reference to *n*-alkanes
RT– srednje vrijeme zadržavanja; KI_{exp} *i* KI_{lit} *– eksperimentalni i literaturni Kovatovi indeksi, redom na HP-5MS koloni u odnosu prema n-alkanima*

data has shown a strong antioxidant capacity against the DPPH radical (Bajpai and Baek, 2016). In India, the study of the antioxidant activity of *T. wallichiana* raw extracts from the leaves showed a very remarkable antioxidant activity (Ahmad *et al*., 2015).

Derivatives of methoxyphenol have shown a wide range of therapeutic benefits, primarily based on their antioxidant properties (Koleva *et al*., 2018). Volatile compounds of medicinal importance, such as furan, furan derivatives, catechol, and cresol, were found in the BO. Furan, as the basic skeleton of some compounds, has cardiovascular properties; it is also widely used for its antiviral, anti-inflammatory, antibacterial, antifungal, antitumor, and antihyperglycemic characteristics, as well as in the treatment of ventricular and atrial fibrillation (Hoyle and Roberts; 1973; Meotti *et al*., 2003; Verma *et al*., 2011). Cresols are used in the preparation of antioxidants and as abortives, diaphoretics, and emmenagogue drugs (Hoyle and Roberts, 1973; Meotti *et al*., 2003; Kim *et al*., 2011; Syazana *et al*., 2011).

p-Cresol (4-methylphenol) was found in high concentration (4.5 %) in yew bark BO. It can be transformed using a Bronsted acid functionalized ionic liquid in the tert-butylation process to produce other valuable antioxidants and diphenol antioxidants with comparatively low toxicity (Fiege *et al*., 2000). According to Elavarasan *et al*. (2010), those conversion products may find application in the food, pharmaceutical, and cosmetic sectors.

4 CONCLUSIONS
4. ZAKLJUČAK

In this research, the antioxidant capacity of extracts and the chemical composition and antioxidant capacity of bio-oil from the bark of *T. baccata* were studied. The antioxidant activity of bio-oil from the bark of *T. baccata* indicated the high potential of this tree for nutrition and pharmaceutical purposes. Considering the findings of this research, the bio-oil extracted from *T. baccata* bark possessed an impressive capacity for scavenging DPPH free radicals compared to ascorbic acid. Regarding bio-oil chemical composition, the principal constituents shown in the bio-oil from bark included guaiacol, phenol, furfuryl alcohol, 3,4-dihydro-*2H*-pyran, *p*-cresol, and 4-methylguaiacol.

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