

CHEMICAL COMPOSITION OF LATVIAN WILD EDIBLE MUSHROOM *CANTHARELLUS CIBARIUS*

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Abstract

The aim of this study was to investigate the chemical composition of widely used wild edible mushroom *Cantharellus cibarius*. Mushrooms *Cantharellus cibarius* were collected in Jelgava region (Latvia) in late summer 2011. Mushrooms were freeze-dried and used to determine chemical composition.

Ash amount was determined by incineration at 550 °C; protein content was determined by Lowry method; total content of phenolic compounds was determined by Folin-Ciocalteu assay. The concentrations of β -carotene and lycopene were determined spectrophotometrically in methanol extract. Protein content in dry matter of *Cantharellus cibarius* was 190 mg g⁻¹. Electrical conductivity (2550 μ s cm⁻¹), titratable acidity (0.238 mmol of NaOH per g of dry matter) and formol number (0.163 mmol NaOH per g of mushrooms dry matter) were measured in water extract. The total content of phenolic compounds was 5.09 mg of gallic acid equivalents per 1 g of mushrooms dry matter. The content of β -carotene was 4 times higher than the content of lycopene for *Cantharellus cibarius*, but 4.6 times less than for *Boletus edulis f. beticola*. Using gas chromatography-mass spectrometry (GC-MS) volatile compounds for the first time were determined in both fresh and freeze-dried samples of mushroom *Cantharellus cibarius* and the dominant compound was found to be oct-1-en-3-ol. Results of the chemical composition of Latvian mushroom *Cantharellus cibarius* were compared to previously published chemical composition of *Cantharellus cibarius* collected in other European regions and to our previous results of the chemical composition of Latvian mushroom *Boletus edulis*. Although the amount of substances tested is slightly higher in mushroom *Boletus edulis*, Latvian mushroom *Cantharellus cibarius* is a rich source of biocompounds and mineral substances.

Keywords: chemical composition, mushroom *Cantharellus cibarius*.

Introduction

Wild edible mushrooms have been widely used for centuries as a source of food and food-flavoring material in many countries. Mushrooms are valued due to their aroma and flavour (Guedes de Pinho et al., 2008a), rich content of carbohydrates and fibres (Mattila et al., 2000), vitamins and minerals, proteins and unsaturated fatty acids (Ribeiro et al., 2009). Edible mushrooms are often regarded as a food with antimicrobial, cholesterol-lowering, anticancer and antioxidant properties (Ribeiro et al., 2006; Barros et al., 2007a; Aryantha et al., 2010). These mushrooms also have prophylactic properties against coronary heart disease and hypertension (Matilla et al., 2000).

Various studies have been carried out on the chemical composition of the European edible wild mushrooms from several countries like Finland (Mattila et al., 2002), Poland (Bernas, Jaworska, 2010), Spain (Diez, Alvarez, 2001), Portugal (Barros et al., 2006, 2007a, 2007b, 2008), Italy (Manzi et al., 2001, 2004), Macedonia (Bauer-Petrovska, 2001), Greece (Ouzouni et al., 2007), Turkey (Ayaz et al., 2011) and Croatia (Beluhan, Ranogajec, 2011).

Carbohydrates and crude proteins are the two main components, thus *Cantharellus cibarius* contains crude protein 53.7%, carbohydrates 31.9% and lipids 2.9% of the dry matter (Barros et al., 2008). Dry matter of mushrooms usually is in the range of 60–140 g kg⁻¹ (Kalač, 2009). Relatively low content of dry matter and lipids result in the low energy value of mushrooms, that for *Cantharellus cibarius* is 118 kJ 100 g⁻¹ of fresh mushrooms (Barros et al., 2008). The analysis of the obtained profiles of fatty acids showed that oleic,

linoleic and, to a lesser extent, palmitic and stearic acids were the main fatty acids in the studied *Cantharellus cibarius* (Ribeiro et al., 2009). The profile of organic acids consisted of citric, malic, fumaric, shikimic and ascorbic acids (Valentao et al., 2005). The content of free amino acids in mushrooms is low, only about 1% of the dry matter. The major amino acids found in *Cantharellus cibarius* were glutaminic acid, lysine, alanine, and threonine (Beluhan, Ranogajec, 2011; Surinrut et al., 1987).

Phenolic compounds have usually attracted attention due to their antioxidant properties. When the individual profile of phenolic compounds in edible mushrooms was studied, *Boletus edulis* was found to present the highest total content of phenolic compounds (Palacios et al., 2011) and total content of flavonoids (Palacios et al., 2011; Robaszkiewicz et al., 2010). *Cantharellus cibarius* was found to contain catechin, pyrogallol, myricetin, phenolic acids such as caffeic, ferulic, gallic, p-hydroxybenzoic, gentisic, homogentisic and protocatechuic acids. Content of caffeic acid and catechin in *Cantharellus cibarius* was higher than in other studied species, including *Boletus edulis*. Although the total content of phenolic compounds is lower, the total antioxidant activity in *Cantharellus cibarius* is higher than in any other mushroom studied, indicating that the increased content of caffeic acid and catechin is responsible for greater antioxidative power (Palacios et al., 2011). Valentao et al. (2005) have identified also the presence of 3-, 4- and 5-O-caffeoylquinic acids and rutin in *Cantharellus cibarius*. Mushrooms are also characterized by a high level of well assimilable mineral constituents. Potassium, magnesium, and phosphorus containing compounds are

the most abundant in *Cantharellus cibarius* (Falandysz et al., 2012; Konuk et al., 2006). Portuguese scientists have investigated some volatile components in *Cantharellus cibarius* (Guedes de Pinho et al., 2008b), but no information is available on the changes in the content of volatile compounds after freeze-drying.

The aim of the present study was to investigate the chemical composition of widely used wild edible mushroom *Cantharellus cibarius* collected in Latvia.

Materials and Methods

Samples of *Cantharellus cibarius* were collected in Jelgava region (Latvia) in late summer 2011. After collection, mushrooms were freeze-dried (Christ Freeze Dryer Alpha 1-2 LD plus, Germany). All dried mushroom samples were grounded in a blender and then stored in air-tight bags at the room temperature until analysis.

Results of analysis were standardized by dry matter of samples. The ash content of mushrooms was determined by incineration at 550 °C (Manjunathan, Kaviyarasan, 2011). The protein amount in mushroom dry matter was determined by Lowry procedure (Lowry et al., 1951) using albumin as a standard.

Water extract was prepared as follows: 1 g of powdered mushrooms was boiled in 50 mL of water for 30 min. The mixture was centrifuged (3000 g, room temperature for 10 min) and supernatant portioned and kept frozen at -23 °C until analysis and used for determination of total phenolic content, formol number, titratable acidity, electrical conductivity.

The total content of phenolic compounds in water extract was determined by Folin-Ciocalteu assay. Gallic acid (0–0.75 mg mL⁻¹) was used as a standard to produce the standard curve. The absorbance of the reaction mixture was measured at 765 nm using UV/Vis spectrophotometer Jenway UV 6405. The total content of phenolic compounds was expressed as milligrams of gallic acid equivalents (GAE) per gram of mushroom dry matter (Barros et al., 2007b).

In water extract titratable acidity was determined by potentiometric titration as described previously (Tanner, 1987) and calculated as mmol of NaOH per 1 g of mushroom dry matter.

The formol number was determined in water extract by potentiometric titration as described previously (Tanner, 1987). Formol number was calculated as mmol of NaOH per 1 g of mushroom dry matter.

Electrical conductivity in water extract was determined to characterise the total content of mineral substances using electrode TetraCon 325 connected to conductometer inoLab pH/Cond 720 (WTW, Germany).

Mushroom powder sample (1 g) was extracted with 50 mL of methanol at 25 °C for 24 h (Ribeiro et al., 2006; Barros et al., 2007a) and used for analysis of β -carotene and lycopene.

The content of the β -carotene and lycopene was determined spectrophotometrically. Obtained methanol

extract was evaporated and 100 mg of the remaining dry matter were stirred with 10 mL of acetone-hexane mixture and filtered. The absorbance was measured at 453, 505 and 663 nm. The content of β -carotene (mg 100 mL⁻¹) and lycopene (mg 100 mL⁻¹) was calculated according to the following equations (Barros et al., 2007b):

$$\text{Lyc} = -0.0458A_{663} + 0.372A_{505} - 0.0806A_{453}, \quad (1)$$

where Lyc – lycopene content, mg 100 mL⁻¹;

A_{663} – absorbance at 663 nm;

A_{505} – absorbance at 505 nm;

A_{453} – absorbance at 453 nm.

$$\beta\text{-carotene} = 0.216A_{663} - 0.304A_{505} + 0.452A_{453}, \quad (2)$$

where β -carotene – β -carotene content, mg 100 mL⁻¹.

The results were expressed as micrograms of carotenoid per gram of dry matter.

Volatiles from mushrooms were extracted using solid phase microextraction (SPME). 0.5 g of freeze dried mushrooms was weighed into a vial. Extraction and injection was performed manually. A 85 μ m carboxen/polydimethylsiloxane (Car/PDMS) fiber (Supelco Inc., Bellefonte, PA, USA) was used for headspace SPME sampling. SPME parameters were: equilibration time 10 min, extraction temperature 40 \pm 1 °C, extraction duration 30 min, desorption 15 min, 250 °C. For the analysis of the volatile compounds a Perkin Elmer Clarus 500 GC/MS equipped with a capillary column Elite-Wax ETR (60 m \times 0.25 mm i.d.; DF 0.25 μ m) was used. Operating conditions were: injector temperature 250 °C; oven temperature start at 40 °C, hold 7 min, programmed from 40 to 160 °C at 6 °C min⁻¹, hold 10 min, and from 160 to 210 °C at 15 °C min⁻¹, hold 15 min; carrier gas (He) 2 mL min⁻¹; split 1 : 2; ionization EI 70 eV; acquisition parameters in full scan mode: scanned m/z 50–300. Identification of compounds was achieved by comparing the mass spectra present in the NIST98 MS Library Database.

The results are presented as the mean \pm standard deviation of three measurements.

Results and Discussion

The content of dry matter of *Cantharellus cibarius* was 9.5 \pm 0.5% and was similar to our previous results of mushroom *Boletus edulis* (Kuka, Čakste, 2011). The content of dry matter of *Cantharellus cibarius* from Croatia was 14.2 \pm 0.2% (Beluhan, Ranogajec, 2011), while in study by Bernáš et al. (2006) the content of dry matter was found to be 7–12 g per 100 g of fresh matter and similar results were obtained in our study.

Ash amount was determined to characterize the content of mineral substances. Ash amount of *Cantharellus cibarius* was 7.76 \pm 0.02% of mushroom dry matter, more than 25% higher than for *Boletus edulis*. Similar tendency was observed when ash content was compared in *Cantharellus cibarius* (8.8 \pm 0.05 g 100 g⁻¹) and *Boletus edulis* (5.3 \pm 0.87 g 100 g⁻¹) mushrooms collected in Croatia (Beluhan, Ranogajec, 2011).

Electrical conductivity in water extract of *Cantharellus cibarius* was $2550 \pm 8 \mu\text{S cm}^{-1}$. The obtained result also indicates that the amount of strong electrolytes in *Cantharellus cibarius* is high. Titratable acidity, that characterises the total amount of acids, is shown in Figure 1. Titratable acidity for *Cantharellus cibarius* was similar to the *Boletus edulis f. pinicola* and *Boletus edulis f. beticola* (0.26 and 0.22 mmol NaOH g^{-1} respectively (Kuka, Cakste, 2011).

The formol number, which characterises the concentration of free amino acids, is shown in Figure 1. The concentration of free amino acids was almost negligible in *Cantharellus cibarius*, while the formol number for *Boletus edulis f. beticola* was 4.6 times higher (Kuka, Cakste, 2011).

The protein amount in mushroom *Cantharellus cibarius* was $190 \pm 2 \text{ mg g}^{-1}$, 1.7 times and 2.8 times lower than for *Boletus edulis f. beticola* and *Boletus edulis f. pinicola*, respectively (Kuka, Cakste, 2011). Content of protein in *Cantharellus cibarius* collected in Latvia was approximately 20%, while in several other studies the amount of protein was shown to vary from 10% (99 g kg^{-1}) (Danell, Eaker, 1992) to 53.7% of dry matter of *Cantharellus cibarius* (Kalač, 2009).

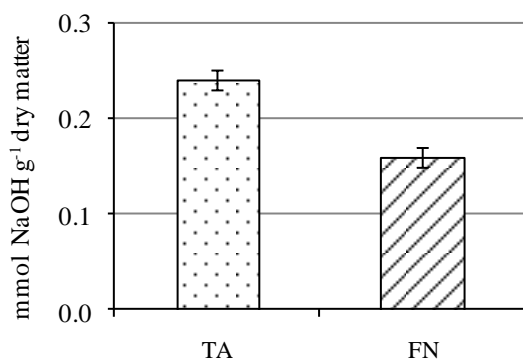


Figure 1. Titratable acidity (TA) and formol number (FN) expressed of *Cantharellus cibarius* dry matter

The content of total phenolic compounds was determined in water extract of *Cantharellus cibarius* and characterised using gallic acid equivalents (GAE) per 1 g of mushroom dry matter and was found to be $5.09 \pm 0.01 \text{ mg GAE g}^{-1}$ of mushroom dry matter. That is 2.2 times lower than for *Boletus edulis f. beticola* and 2.5 times lower than for *Boletus edulis f. pinicola* (Kuka, Cakste, 2011). *Cantharellus cibarius* from Poland and Portugal was shown to contain phenolic compounds $2.39 \pm 0.23 \text{ mg GAE g}^{-1}$ (Robaszekiewicz et al., 2010) and $1.75 \pm 0.50 \text{ mg GAE g}^{-1}$ (Barros et al., 2008) of dry matter, respectively, while *Cantharellus cibarius* from Latvia contained approximately 2-fold more phenolic compounds calculated as mg GAE g^{-1} .

The content of β -carotene and lycopene was determined using methanol extract of mushroom dry matter (Figure 2). The content of β -carotene was

4 times higher than the content of lycopene for *Cantharellus cibarius*, but 4.6 times less than for *Boletus edulis f. beticola* (Kuka, Cakste, 2011). *Cantharellus cibarius* collected in Portugal contained $5.77 \pm 0.42 \mu\text{g g}^{-1}$ of β -carotene and $1.95 \pm 0.28 \mu\text{g g}^{-1}$ of lycopene (Barros et al., 2008) and comparable amounts of β -carotene and lycopene were also found in mushrooms collected in Latvia.

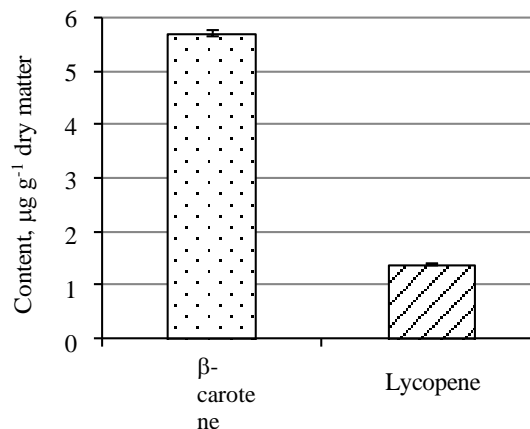


Figure 2. The content of β -carotene and lycopene of mushroom *Cantharellus cibarius* dry matter

Using gas chromatography-mass spectrometry (GC-MS) volatile compounds were identified in both fresh and freeze-dried mushrooms *Cantharellus cibarius*. Oct-1-en-3-ol, (Z)-oct-2-en-1-ol, hexanal, hept-1-en-3-one, ethyldeca-2,4-dienoate, (E)-oct-2-enal, octan-3-one were identified in fresh mushroom (Figure 3). The dominant compound was found to be oct-1-en-3-ol. The study of Du et al. (2010) similarly to the current research revealed that most abundant aroma compounds are alcohols, aldehydes, and ketones. Among other important compounds were identified terpenes, eight carbon compounds and their derivatives. According to the research of Cheng et al. (2012) oct-1-en-3-ol gives a unique earthy taste and sweetness to mushroom aroma. As show previous studies alcohols and ketones, mainly C8 aliphatic compounds, like octan-3-ol, oct-2-en-1-ol, octan-1-ol and octan-3-one were the main compounds responsible for characteristic mushroom-like aroma emitted by some of mushrooms (Malheiro et al., 2013). In the current study great losses of volatile compounds were found in freeze-dried mushrooms. The total sum of peak areas in fresh *Cantharellus cibarius* mushroom was 166.08×10^6 , which decreased as a result of drying till 93.8×10^6 . Content of oct-1-en-3-ol was decreased more than 5 times suggesting significant change of mushroom-like aroma. The loss of the noted alcohol could be due to its high volatility. (Z)-oct-2-en-1-ol, hept-1-en-3-one, ethyldeca-2,4-dienoate, (E)-oct-2-enal and octan-3-one were not identified in freeze-dried samples.

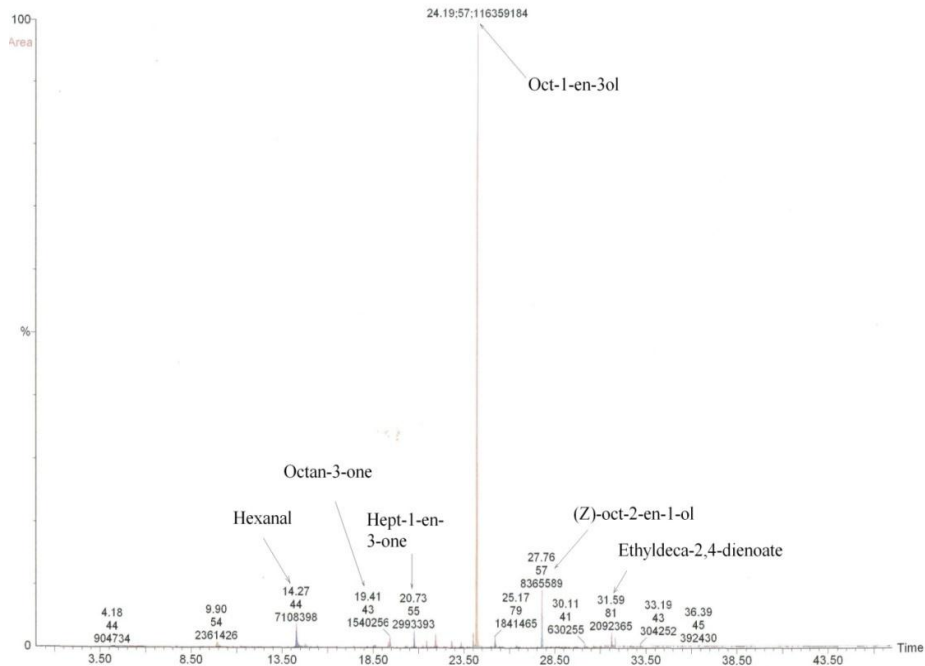


Figure 3. Chromatogram of volatile compounds of fresh mushroom *Cantharellus cibarius*

Conclusions

Although the amount of substances tested is slightly higher in mushroom *Boletus edulis*, mushroom *Cantharellus cibarius* is a rich source of biocompounds and mineral substances.

Freeze-drying significantly reduces the amount of volatile compounds and could affect flavour of mushrooms.

Together, Latvian *Cantharellus cibarius* have similar dry matter, ash and protein content, amount of β -carotene and lycopene, but two fold higher amount of phenolic compounds than *Cantharellus cibarius* collected in other regions of Europe.

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