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ORIGINAL ARTICLE

Chemical composition and nutritive value of *Pleurotus citrinopileatus* var *cornucopiae*, *P. eryngii*, *P. salmoneo stramineus*, *Pholiota nameko and Hericium erinaceus*

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Abstract The chemical composition and nutritive value of five mushrooms species, some less studied such as *Pleurotus citrinopileatus* var. *cornucopiae*, *P. salmoneo stramineus or Pholiota nameko*, were determined. Protein, sugar and fat contents ranged between 16.2 to 26.6, 52.7 to 64.9 and 2.3 to 3.5 g/100$g_{dry\ mushroom}$, respectively. Highest total phenolic content was observed for *P. citrinopileatus* var. *cornucopiae* with 1140 μg cathecol equiv./$g_{dry\ mushroom}$. Higher content in mono and polyunsaturated fatty acids (FA) than saturated FA characterized mushrooms FA profile with high linoleic acid concentration (>30 gFA/100g_{fat}). In addition, these mushrooms may be considered good sources of K, Mg, P, highlighting K (2627–3736 mg/$Kg_{dry\ mushroom}$) as the most predominant, and of Zn, Cu and Fe; some contributing over 15 % of their recommended daily intakes. β-glucans, α-glucans and evidence of glucan-protein complexes were identified by FTIR-ATR. The reported values emphasize the nutritional potential of the five species to be consumed in a healthy diet.

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Keywords Edible mushrooms · Elemental and proximate composition · Fatty acids · FTIR-ATR characterization · Nutritional value

Introduction

Edible mushrooms have been appreciated ever since early times for their sensory characteristics and culinary suitability. Furthermore, they are well recognized for their nutritional and health benefits (Sabaratnam et al. 2011). Over the last 10 years edible mushrooms have been receiving an increased attention from researchers. Mushrooms are a great source of proteins (20–25 %), polysaccharides (37-48 %), fibers (13–24 %), vitamins and minerals (Sabaratnam et al. 2011; Alam et al. 2008) and of some secondary metabolites, including phenolic compounds, polyketides, terpenes and steroids (Cheung et al. 2003). In addition, they are low in calories (Sabaratnam et al. 2011) and in fat content (4–5 %) (Alam et al. 2008) which represents an advantage for the currently more balanced diets. From the health perspective mushrooms have been recognized for their antifungal, antibacterial, antioxidant, antimicrobial and antiviral properties, and have also been envisaged in functional foods development (Wani et al. 2010). They have been used in several therapeutical applications, including antitumor, immune-modulation and anti-diabetic treatments (Cheng et al. 2012) and are reported to have a preventive role in some cardiovascular diseases (Guillamón et al. 2010), anti-inflammatory and analgesic properties (Smiderle et al. 2008).

There are several hundreds of wild species of edible mushrooms yet only about 20 species are cultivated and used more extensively as food (Kalač 2013) and only 10 species are cultivated on an industrial scale (Reis et al. 2012a). The most commonly eaten species are *Agaricus bisporus* (Paris

mushroom) and *Lentinula edodes* (Shiitake mushroom). The 'Shiitake mushroom' is as common in Asian countries as *Agaricus bisporus* is in the western world (Ghorai et al. 2009). Although cultivated mushrooms species has grown recently, there is still a lack of knowledge on their chemical composition which is essential to assess their nutritional value as well as their potential for functional ingredients provision. Therefore, the main objective of the present study was to determine the chemical composition of five cultivated edible mushrooms, some less characterized such as *Pleurotus citrinopileatus* var. *cornucopiae* or *Pleurotus salmoneo stramineus, Pholiota nameko* and *Hericium erinaceus*. Information on the proximal composition, fatty acids (FA) profile, elemental composition and main chemical compounds identified by FTIR-ATR of the five cultivated mushrooms is presented and discussed as well as their nutritive value and possible role in a healthy diet. To our knowledge this is the first study describing the FA composition of the edible *P. salmoneo stramineus, P. citrinopileatus* var *cornucopiae*, *Ph. nameko* and *H. erinaceus* and therefore this work brings valuable data about the nutritional evaluation of such mushrooms species.

Materials and methods

Mushrooms species and cultivation conditions

Dried specimens of mushrooms were supplied by Bioinvitro, Biotecnologia, Lda. (Gandra, Portugal): 3 specimens of order Agaricales and family Pleurotaceae, *Pleurotus citrinopileatus* var. *cornucopiae, Pleurotus eryngii* and *Pleurotus salmoneo stramineus*; 1 specimen of order Agaricales and family Strophariaceae, *Pholiota nameko*; and 1 specimen of order Russulales and family Hericiaceae, *Hericium erinaceus.* The classification of mushrooms was based on MycoKey™ (Petersen and Læssøe 2013). Mushrooms were cultivated in filter bags with sterilized organic substrate. Table 1 summarizes the cultivation conditions (Bioinvitro information) for each species of mushroom. After fructification and growth, entire clean mushrooms were dried in a ventilated drier through 24 h between 40 and 60 °C. The dried mushrooms were subsequently milled to less than 1.0 mm particle size.

Proximate composition

Moisture, organic matter and ash were determined according to AOAC (1990) methods. Protein content was determined by the Kjeldahl method adapted from US ISO 5983–1 (2009) using 4.38 as converting factor to protein (Kulshreshtha et al. 2013). Total fat was determined by Soxhlet extraction procedure. Total sugar was determined indirectly by calculation, subtracting protein and fat content from total organic content. Total poliphenolic content were extracted from 0.1 g of dry mushroom in 10 mL of ethyl acetate after 30 min of sonication (Ultrasonik, Germany). The extract was filtered with anhydrous sodium sulfate (Sigma) and brought to dryness with a rotary evaporator (Heidolph, Germany). The residue was re-dissolved in 5 ml of milliQ water and phenolic content of 2 mL was determined by colorimetric method of Folin-Ciocalteu (Mulinacci et al. 2001), using cathecol (0 to 75 mg/L) as standard and expressed as μg cathecol equivalent per g of dry mushroom.

Fatty acid analysis

For the analysis of total fatty acid (FA) composition, 100 mg of sample were accurately weighed and prepared according to Sánchez-Ávila et al. (2009). For quantitation purposes 100 μL of methyl tricosanoate (1.28 mg/mL) were added to samples prior to derivatization. FAME were analyzed in a gas chromatograph HP6890A (Hewlett-Packard, USA), equipped with a flame-ionization detector (GLC-FID) and a BPX70 capillary column (50mx0.32mmx0.25 μm; SGE, France) according to the conditions described by Vingering and Ledoux (2009) Supelco 37 fatty acid methyl ester (FAME) mix (Sigma-Aldrich, USA) and butterfat CRM-164 from Fedelco Inc. (Madrid, Spain) were used for identification of fatty acids. GLC-Nestlé36 (Elysian, USA) was assayed for calculation of response factors and detection and quantification limits (LOD: 0.15 μg/mL; LOQ: 0.46 μg/mL).

Analysis of elements

The acid digestion was based on microwave-assisted digestion proposed by Speedwave MW-3+ (Berghof, Germany) for dried plant samples with some modifications for determination of Mo, B, Zn, P, Cd, Co, Ni, Mn, Fe, Mg, Ca, Cu, Na, Al and K in dried samples of mushrooms. A sample with up to 0.2 g of each dry powdered mushroom was placed in the digestion vessel and 5 mL of concentrated nitric acid were added. The vessels were capped and placed in a microwave pressure digestor Speedwave MWS-3+ (Berghof, Germany) and subjected to microwave radiation at 20 bar according to the following program: room temperature was raised first to 130 °C at 22 °C/min and 30 % of irradiation power, then to 160 °C at 6 °C/min and 40 % of irradiation power, for 5 min, and to 170 °C at 5 °C/min and 50 % of irradiation power, for 5 min. The cooling process consisted of decreasing temperature first to 100 °C for 4 min and then to room temperature. After cooling, acid digests were made up to 20 mL with Milli-Q water. Three replicates were performed for each sample as well as blanks.

Mineral content determination was performed using an inductively coupled plasma (ICP) optical emission spectrometer model Optima™ 7000 DualView ICP-OES (PerkinElmer, USA) with radial plasma configuration. Standard plasma conditions were used namely 1300 W for radio-frequency power, 1.5 mL/min pump rate, and 15.0, 0.2 and 0.8 L/min for

Table 1 Summary of the growth conditions for each of the cultivated mushroom species

Mushroom	Organic substrate[a]	Water content[b]	Incubation	Fructification
Pleurotus citrinopileatus var. cornucopiae	Sawdust spruce, 38 % Wood shavings, 5 % Fibers (wood, straw), 20 % Crushed grain corn, 18 % Wheat bran, 13 % Crushed oil seed cake, 6 %	68 %	20–30 days 22 °C	5–10 days 18–20 °C
Pleurotus salmoneo stramineus				5–10 days 20–22 °C
Pleurotus eryngii	Sawdust spruce, 20 % Wood shavings, 5 % Fibers (wood, straw), 30 % Sugar beet pulp, 5 % Crushed grain corn, 14 % Wheat bran, 14 % Crushed oil seed cake, 12 %	67 %	20–30 days 20–22 °C	13–18 days 15–22 °C
Hericium erinaceus	Sawdust beech, 70 % Fibers (wood, straw), 5 % Crushed grain corn, 10 % Wheat bran, 12 % Crushed oil seed cake, 3 %	64 %	30–40 days 20–22 °C	6–10 days 16–20 °C
Pholiota nameko	Sawdust beech, 76 % Fibers (wood, straw), 5 % Crushed grain corn, 9 % Wheat bran, 7 % Crushed oil seed cake, 3 %	65 %	30–40 days 20–22 °C	15–21 days 16–20 °C

[a] Composition based on percentage of dry matter of each organic component

[b] Water content based on fresh weight

plasma, auxiliary and nebulizer gas flow, respectively. Detection wavelengths are depicted in Table 2.

A multi-element standard (Inorganic Ventures) containing up to 10 mg/L of Mo, 2,5 mg/L of B, 15 mg/L of Zn, 750 mg/L of P, 0.2 mg/L of Co and Ni, 1 mg/L of Cd and Cu, 15 mg/L of Mn and Fe, 1000 mg/L of Mg, 3000 mg/L of Ca, 50 mg/L of Na, 5 mg/L of Al and 2000 mg/L of K was used for the preparation of standard solutions in 2 % HNO_3. Successive dilutions of the stock reference solution (100, 50 and 10 times) were prepared and used for calibration models and the concentration of each element was determined by direct interpolation in the standard curve within its linear dynamic range. The limits of detection

Table 2 Analysis of IPE 120 (*Agaricus bisporus*) certified reference material

Element	Wavelengths detection	Limits of detection (mg/L)	Certified value (g/kg)	Determined value (g/kg)	% Recovery
K	769.896	3.75	43.6±2.36	40.8±1.03	94±2
Mg	279.955	0.18	1.13±0.086	1.02±0.021	91±2
P	214.914	0.40	9.82±0.631	9.49±0.284	97±3
Ca	317.933	2.06	0.393±0.1059[a]	0.329±0.0364	84±9
Na	330.237	1.17	–	–	–
			(mg/g)	(mg/g)	
B	208.889	0.0009	8.49±1.276	7.74±0.254	91±3
Cu	324.752	3.32	17.4±1.39	16.7±0.25	96±1
Mn	257.610	0.014	5.30±1.175	4.55±0.447	86±8
Zn	213.857	0.010	42.2±4.19	39.6±1.10	94±3
Fe	259.939	0.16	31.4±8.09[a]	32.8±4.56	105±14
Cd	226.502	0.002	–	–	–
Co	228.616	0.002	–	–	–
Ni	231.604	0.003	–	–	–
Al	394.401	0.037	–	–	–

[a] Indicative values by IPE 120

(LODs –Table 2) were calculated using y=yB+3SB, where SB is the standard deviation (SD) of the blank signal estimated as sy/x, the residual SD taken from the calibration line, and yB is the blank signal estimated from the intercept, also taken from the calibration line (Miller and Miller 2005).

The accuracy of the method (microwave acid digestion and ICP-OES analysis) was assessed by analysis of certified reference material IPE 120 (*Agaricus bisporus*; WEPAL, Holland). Five replicates of reference material were subject to microwave digestion and analyzed three times by ICP-OES. Recovery ranged between 84 and 105 % (Table 2).

FTIR-ATR analysis

Samples of milled, dried mushroom material were analyzed by Fourier Transform Infrared Spectroscopy with attenuated total reflectance (FTIR-ATR). The FTIR spectra were recorded on a Bruker Tensor 27 spectrometer (Bruker Scientific Instruments, USA), using a Golden Gate single reflection diamond ATR system (Specac Lda, USA). All spectra resulted from the average of two counts, with 128 scans each and a resolution of 2 cm^{-1}.

Statistical analysis

One-way analysis of variance (ANOVA) was carried out with SigmaStat™ (Systat Software, USA), to assess differences between mushroom species in terms of proximate or elemental composition at a significance level of P=0.05. The Holm-Sidak method was used for pairwise comparisons at a significance level of P=0.05. Fatty acid data in turn, were analyzed using the IBM SPSS Statistics v22 for Mac. Normality and homogeneity were examined and One way ANOVA with the Bonferroni test for post-hoc analyses was applied to evaluate statistical differences between mushroom species ($p<0.05$).

Results and discussion

Proximate composition of cultivated mushroom species

Mushrooms are known for being rich in protein and polysaccharides and poor in fat content, which from a nutritional point of view is of particular interest especially for low calorie diets with low fat content. The composition reported for the five species under evaluation (Table 3) was no different from this trend, yet among them, between the different families and within the same family, significantly different compositions were found, in particular, in what concerns protein and organic matter contents. Differences in total fat content were less relevant and only *P. salmoneo stramineus* presented a significantly different ($p<0.05$) value; in fact the lowest fat content among the five species studied. Within the same family, i.e., Pleurotaceae family, *P. eryngii* was characterized by the lowest total protein content (16.2 $g/100g_{dry\ mushroom}$) followed by the highest content in total sugar (64.9 $g/100g_{dry\ mushroom}$) and in total fat (3.4 $g/100g_{dry\ mushroom}$) whereas *P. salmoneo stramineus,* presented the highest protein content (26.6 $g/100g_{dry\ mushroom}$) but the lowest content in total sugar (52.7 $g/100g_{dry\ mushroom}$) and in total fat (2.3 $g/100g_{dry\ mushroom}$). *Pleurotus citrinopileatus* var. *cornucopiae* had a similar proximate composition to *P. salmoneo stramineus* differing to *P. eryngii* (Table 3). A similar proximate composition characterized *Ph. nameko* and *H. erinaceus* with 16.8–19.2, 59.6–61.2 and 2.9–3.2 $g/100g_{dry\ mushroom}$ of total protein, total sugar and total fat content, respectively.

It is known that mushroom proximate composition may be affected by several factors, such as species, development stage, maturity of fruiting body, mushroom sampled part, level of nitrogen available (Colak et al. 2009) as well as by substrate composition and cultivation procedures (La Guardia et al. 2005). Our results demonstrate such interspecies variability and corroborate the importance of cultivation optimization and standardization to increase yield and nutritional value. Protein content in the five similarly cultivated mushrooms ranged between 16.2 and 26.6 $g/100g_{dry\ mushroom}$ for *Pleurotus* spp. and 17–19 $g/100g_{dry\ mushroom}$ for *Ph. nameko* and *H. erinaceus*, respectively (Table 3). Lower protein contents (8.5–19.7 % dried mushroom) was reported by Akyüz and Kirbağ (2010) who studied the effect of various agro-wastes (wheat straw, wheat strawcotton stalk and rice bran) on the nutritive value of cultivated *P. eryngii* var. *ferulae*. La Guardia et al. (2005) reported a proximate composition with higher levels of protein (26.6 $g/100\ g_{dry\ mushroom}$), similar to those reported for *P. salmoneo stramineus* herein, for *P. eryngii* var. *eryngii* cultivated in wheat straw and sugar beet based substrate. Reported values of protein content for *P. citrinopileatus* are lower than those reported herein and ranged between 9.2 and 17.2 $g/100g_{dry\ mushroom}$ when cultivated on substrate based on the sludge of handmade paper and cardboard industrial waste used alone or in combination with wheat straw (Kulshreshtha et al. 2013). A value of 15.6 $g/100g_{dry\ mushroom}$ for total protein was reported by Guo et al. (2007) for cultivated *P. djamor* (former name of *P. salmoneo stramineus*) purchased in a local supermarket in Guangzhou city, China. According to Khan and Tania (2012) *Pleurotus* species are recognized as good source of protein with values ranging from 11 to 42 $g/100g_{dry\ mushroom}$, fitting well the results reported for the 3 species included in this study.

Low total fat contents (2.3–3.5 $g/100g_{dry\ mushroom}$) and high total sugar contents (52.7–64.9 $g/100g_{dry\ mushroom}$) characterize, in general, the cultivated mushrooms analyzed similarly to those found in the literature for cultivated *P. eryngii* var. ferulae Akyüz and Kirbağ (2010). Lower total fat content was reported for *P. citrinopileatus* cultivated in different substrate (0.13 to 0.46 $g/\ 100g_{dry\ mushroom}$) (Kulshreshtha et al.

Table 3 Proximate composition of mushrooms and their percentage contribution in nutrients intake

Parameter	*Pleurotus citrinopileatus var. cornucopiae*	*Pleurotus salmoneo stramineus*	*Pleurotus eryngii*	*Hericium erinaceus*	*Pholiota nameko*
% Moisture ($g/100g_{dry\ mushrooms}$)	10.69±0.05 c	11.10±0.01 d	9.47±0.08 a	10.11±0.5 b	11.91±0.01 e
% Total Protein ($g/100g_{dry\ mushrooms}$)	23.8±0.1 d	26.6±0.2 e	16.2±0.3 a	18.8±0.1 c	16.92±0.03 b
% Total sugars[1] ($g/100g_{dry\ mushrooms}$)	54.7	52.7	64.9	61.3	59.6
% Total Fat ($g/100g_{dry\ mushrooms}$)	3.5±0.1 b	2.3±0.1 a	3.4±0.1 b	2.9±0.1 b	3.2±0.1 b
Total phenolic content (mg cathecol equiv./$g_{dry\ mushrooms}$)	1140±90 c	464±12 a	709±36 b	753±4 b	675±44 b
% Organic matter ($g/100g_{dry\ mushrooms}$)	82.03±0.07 c	81.63±0.02 b	84.54±0.01 e	82.98±0.06 d	79.68±0.06 a
% Ash ($g/100g_{dry\ mushrooms}$)	7.29±0.12 c	7.27±0.01	5.99±0.08 a	6.91±0.01 b	8.40±0.06 d

		Pleurotus citrinopileatus var. cornucopiae		*Pleurotus salmoneo stramineus*		*Pleurotus eryngii*		*Hericium erinaceus*		*Pholiota nameko*	
Nutrients	g/day	$g/8.4g_{portion}$	% RDI[3]	$g/8.4g_{portion}$	% RDI[3]	$g/8.4g_{portion}$	% RDI[3]	$g/8.4g_{portion}$	% RDI[3]	$g/8.4g_{portion}$	% RDI[3]
Protein	50	2.00	4.0	2.23	4.5	1.36	2.7	1.58	3.8	1.42	3.4
Total sugars	270	4.59	1.7	4.43	1.6	5.45	2.0	5.15	2.3	5.01	2.2
Fat	70	0.29	0.4	0.19	0.3	0.29	0.4	0.24	0.4	0.27	0.5
	mg/day[2]	$mg/8.4g_{portion}$	% RDI[3]	$mg/8.4g_{portion}$	% RDI[3]	$mg/8.4g_{portion}$	% RDI[3]	$mg/8.4g_{portion}$	% RDI[3]	$mg/8.4g_{portion}$	% RDI[3]
Calcium	800	1.42	0.2	1.42	0.2	1.52	0.2	1.46	0.2	1.90	0.2
Potassium	2000	240.85	12.0	220.78	11.0	220.70	11.0	242.05	12.1	313.89	15.7
Magnesium	375	12.65	3.4	11.41	3.0	11.20	3.0	7.97	2.1	13.21	3.5
Phosphorus	700	70.15	10.0	88.97	12.7	82.21	11.7	64.03	9.2	91.50	13.1
Iron	14	0.78	5.6	1.96	14.0	0.35	2.5	0.59	4.2	2.33	16.7
Zinc	10	0.88	8.8	1.30	13.0	0.72	7.2	0.55	5.5	1.02	10.2
Copper	1	0.10	9.6	0.21	20.8	0.07	7.0	0.16	16.0	0.23	23.0
Manganese	2	0.06	3.1	0.08	4.2	0.08	4.0	0.13	6.7	0.15	7.40

a–f, in a raw: Different letters indicate significant differences ($p<0.05$) between species

[a] % Total Sugars (%)=Organic matter (%) – Total Protein (%) - Total Fat (%)

[b] WHO, 2004

[c] Based on a serving size of 84 g fresh mushroom (O'Neil et al. 2013) equivalent to 8.4 g portion of dry mushroom

2013) and for *P.djamor* (1.65 g/ 100$g_{dry\ mushroom}$) (Guo et al. 2007). In turn, La Guardia et al. (2005) reported much higher values for fat content (8.6 g/100$g_{dry\ mushroom}$) for *P. eryngii* var. *eryngii* cultivated in wheat straw and sugar beet based substrate; other reported fat values for *Pleurotus* species ranged between 0.5 and 8 g/100$g_{dry\ mushroom}$ (Khan and Tania 2012). The characteristic low fat content found in mushrooms coupled to its unsaturated nature provided by the predominating linoleic and oleic free fatty acids (Kalač 2013) is of great dietary importance.

Mushrooms are a good source of carbohydrates. Values ranging between 51.4 and 59.9 g/100$g_{dry\ mushroom}$ were reported by La Guardia et al. (2005) and by Guo et al. (2007) for *P. eryngii* var. *eryngii* and for *P. djamor*, respectively. According to Kalač (2013) the carbohydrate content constitutes about one-half of mushroom dry matter being present mainly as polysaccharides and glycoproteins making them suitable for incorporation into low-calorie diets; 36–60 g/100$g_{dry\ mushroom}$ of carbohydrates have been reported for *Pleurotus* species (Khan and Tania 2012) which are comparable to the data in Table 3.

Comparison between proximate composition data for cultivated *Ph. nameko* and *H. erinaceus* and published data was not possible because to the best of our knowledge the only existing characterization studies targeted wild-growing *H. erinaceus*. Mau et al. (2001) reported carbohydrate, moisture, crude protein, crude fat and ash content values of 57.0, 4.3, 22.3, 3.5 and 9.4 % of air dried weight for wild-growing *H. erinaceus*, respectively.

Edible mushrooms have been reported as a source of phenolic compounds with antioxidant properties (Preeti et al. 2012). According to Preeti et al. (2012) natural phenolic compounds are produced and accumulated with end products ranging from simple molecules (phenolic acids) to highly polymerised compounds (tannins). Among the 5 edible mushroom species tested, the Pleurotaceae family revealed great variability englobing the highest (1140 μg cathecol equiv./$g_{dry\ mushroom}$ found in *P. citrinopileatus* var. *cornucopiae*) and the lowest (464 μg cathecol equiv./$g_{dry\ mushroom}$ found in *P. salmoneo stramineus*) total phenolic contents. This trend somewhat contrasts with those reported by Mishra et al. (2013) who determined the total phenolic content in mushroom mycelium of the same 3 species, i.e., *Pleurotus eryngii, P. djamor* and of *P. citrinopileatus* but grown in malt extract; higher values were observed for mycelium of *P. eryngii*, followed by *P. djamor* and *P. citrinopileatus*, respectively.

Apparent higher phenolic content was reported by Yildirim et al. (2012) for wild-grown edible *P. eryngii* (29 to 32 mg of gallic acid equiv./$g_{dry\ mushroom}$) collected from different regions of Tunceli (Turkey). However direct comparison will not be appropriate given their different origin: cultivated *vs* wild-growing mushrooms. In terms of cultivated *Pleurotus* sp., Reis et al. (2012b) reported 7.14 mg of gallic acid equiv./$g_{methanolic\ extract}$ from *P. eryngii* mushrooms obtained in local supermarkets (Bragança, Northeast Portugal).

Ash content was quite variable and statistically different among the different species, ranging from 5.99 to 8.40 g/100$g_{dry\ mushroom}$ in *P. eryngii* and in *Ph. nameko* (Table 3). No statistical differences were observed between *Pleurotus spp.* for ash content.

Fatty acids profile of cultivated mushrooms

From the analysis of data in Table 4, it was found that total FA concentration ranged from 22.75 to 14.12 μg/$mg_{dry\ mushroom}$ in *P. eryngii* and in *P. salmoneo stramineus* ($p<0.05$). All samples showed concentrations of linoleic acid (C18:2 c9 c12) above 30 g FA/100g_{fat}, indicating that this is main FA in the composition for *P. salmoneo stramineus* and *P. citrinopileatus* var *cornucopiae* (69.09–78.33 g FA/100g_{fat}). The other mushroom species had significantly lower values especially in *H. erinaceus* (38.69 g FA/100g_{fat}). The FA composition of the mushroom species was characterized by much higher content of mono (MUFA) and polyunsaturated FA (PUFA) than of saturated FA (SFA). The SFA distribution was characterized by palmitic (C16) and stearic acids (C18). Higher content was observed in *H. erinaceus* and in *P. citrinopileatus* var *cornucopiae* (16.52–18.60 g FA/100g_{fat}) whereas values between 10.3 and 12.5 g FA/100g_{fat} were observed for the other mushroom species. For C18 the highest amount for this FA was observed in *H. erinaceus* (6.15 g FA/100g_{fat}; $p<0.05$) which was 3.3 to 3.7 times higher compared to the other mushroom species (1.64–1.85 g FA/100g_{fat}).

Interestingly, in *P. salmoneo stramineus* and *P. citrinopileatus* var *cornucopiae* the concentration of linoleic acid was higher than 65 % but the concentration of oleic acid (C18:1 c9) was the lowest (3.7–5.5 g FA/100g_{fat}). For the other mushroom species this ratio was lower and more variable (Table 4).

The FA composition of edible mushrooms can comprise from butyric acid (C4) to docosahexanoic acid (C22:6) depending on the mushroom species but in general C16, C18:1 c9 and C18:2 c9 c12 are among the main FA (Ergönül et al. 2013). Previous studies reporting the FA composition of wild *P. eryngii* or obtained from local supermarkets in Portugal, showed that SFA content were 25.8–17.4 %, MUFA were 49–13.1 % and PUFA were 69.4–25.2 % (Reis et al. 2012a, 2014). Such variation were associated to the values of palmitic (14.9–12.8 %), oleic (47.5–12.3 %) and linoleic acids (68.8–24.7 %). However the detailed profile was solely focused on the mentioned FA as well as on the stearic and linolenic acids.

To our knowledge this is the first study describing the FA composition of the edible *P. salmoneo stramineus, P. citrinopileatus* var *cornucopiae*, *Ph. nameko* and *H. erinaceus* and therefore this work brings valuable data about the nutritional evaluation of such mushrooms species.

Table 4 Fatty acid composition (g FA/100g_{fat}) and total content (μg FA/$mg_{dry\ mushroom}$) of cultivated edible mushroom species

	Pleurotus citrinopileatus var. cornucopiae		*Pleurotus salmoneo stramineus*		*Pleurotus eryngii*		*Hericium erinaceus*		*Pholiota nameko*	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
C14	0.17 b	0.02	0.18 b	0.01	0.26 a	0.01	0.24 ab	0.01	0.16 b	0.02
C15	2.31 a	0.02	0.82 d	0.07	1.27 b	0.02	1.44 c	0.03	1.88 a	0.05
C16	16.52 b	0.07	12.45 c	0.04	12.49 c	0.03	18.60 a	0.02	10.29 b	0.66
C16:1 c7	<LOD c	—	<LOD c	—	0.10 b	0.02	0.17 a	0.01	0.06 b	0.01
C16:1 c9	0.22 b	0.02	0.23 b	0.01	0.26 b	0.04	0.39 a	0.01	0.22 b	0.02
C17i	0.20 b	0.04	0.36 a	0.02	0.14 b	0.01	0.11 b	0.02	0.19 b	0.02
C16:2 c9 t12	<LOD c	—	<LOD c	—	0.04 b	0.01	0.38 a	<0.01	<LOD c	—
C17	0.32 c	<0.01	0.58 a	0.05	0.17 b	<0.01	0.49 a	0.02	0.21 bc	0.02
C17:1 c9	<LOD b	—	<LOD b	—	<LOD b	—	0.08 a	0.01	<LOD b	—
C17:1 c10	<LOD d	—	0.12 b	0.04	0.07 c	<0.01	0.14 b	<0.01	0.15 a	0.02
C18	1.79 b	0.01	1.85 b	0.02	1.66 c	0.01	6.15 a	0.04	1.64 c	0.03
C18:1 t4+t5	<LOD c	—	0.20 a	0.01	0.09 b	<0.01	0.09 b	0.02	0.14 b	0.01
C18:1 t9	<LOD c	—	<LOD c	—	0.05 b	0.01	0.07 b	0.01	0.13 a	0.03
C18:1 c9	5.47 c	0.05	3.71 c	0.01	27.13 a	0.01	27.36 a	0.09	17.82 b	0.01
C18:1 c11	0.63 d	0.04	0.30 e	0.01	0.82 c	0.01	4.58 a	0.12	3.51 b	0.03
C18:2 c9 t12	0.09 a	<0.01	0.11 a	0.02	0.13 a	0.01	0.09 a	0.01	0.12 a	0.02
C18:2 t9 c12	<LOD b	—	0.07 a	0.01	<LOD b	—	0.04 a	<0.01	0.06 a	<0.01
C18:2 c9 c12	69.09 b	0.19	78.33 a	0.10	53.90 d	0.05	38.69 e	0.15	62.22 c	0.05
C18:3 c9 c12 c15	<LOD b	—	<LOD b	—	0.09 a	<0.01	0.05 a	<0.01	0.08 a	0.04
C20	<LOD c	—	<LOD c	—	0.11 b	0.01	0.11 b	0.02	0.21 a	0.01
C20:1 c11	<LOD c	—	<LOD c	—	0.13 a	0.02	0.05 b	0.01	0.11 a	0.03
C20:3 c11 c14 c17	2.74 a	0.09	0.42 b	0.01	0.43 b	0.01	0.24 c	0.01	0.08 d	<0.01
C24	0.46 a	0.01	0.28 b	0.04	0.27 b	0.01	0.42 ab	0.01	0.35 b	<0.01
C24:1 c15	<LOD b	—	<LOD b	—	0.39 a	0.02	<LOD b	—	0.37 a	0.02
SFA	21.76 b	0.01	16.51 c	0.06	16.36 c	0.02	27.56 a	0.02	14.93 d	0.04
MUFA	6.32 d	0.11	4.56 e	0.07	29.04 b	0.06	32.94 a	0.01	22.51 c	0.04
PUFA	71.93 b	0.10	78.93 a	0.13	54.59 d	0.08	39.50 e	0.02	62.56 c	0.01
μg FA/$mg_{dry\ mushroom}$	14.14 c	0.32	14.12 c	0.50	22.75 a	0.57	19.37 b	0.36	17.20 b	0.62

Data expressed as mean (Mean; n=3) and standard deviation (SD). c/t: cis/trans double bond. SFA/MUFA/PUFA: total of saturated/monounsaturated/polyunsaturated fatty acids

a–e: in a row, significant differences among mushroom species

Elemental composition of cultivated mushroom

Cultivated mushrooms have been reported as a good source of minerals, and may contain calcium(Ca), magnesium (Mg), sodium (Na), potassium (K), phosphorus (P), copper (Cu), iron (Fe), manganese (Mn), and zinc (Zn) (Cheung 2008). In this study, the species analyzed revealed to be good sources of macroelements such as K, Mg, and P, where K stood out as the most predominant macroelement with values ranging between 26,274 and 37,368 mg/$Kg_{dry\ mushroom}$ followed by P (7623–10,892 mg/$Kg_{dry\ mushroom}$) and Mg (949–1572 mg/$Kg_{dry\ mushroom}$). Their combined percentage was about 98–99 % where K represented 6–76 %, followed by P with 20–27 % and Mg with 2.5–3.9 %. Higher contents, but not statistically different ($p>0.05$), of K, P and Mg were observed for both *P. eryngii* and *P. salmoneo stramineus*. Higher content of K was observed in *Ph. nameko* ($p<0.05$) in comparison to *H. erinaceus*. Potassium and Mg were also the predominant elements in other cultivated *Pleurotus* spp. For example, Lee et al. (2009) reported 26,273 and 1233 mg/$Kg_{dry\ mushroom}$ for K and Mg for *P. eryngii* cultivated in complex substrate based on pine sawdust, corncob and rice bran with beet pulp (sawdust and beet pulp are two common organic substrates also used to cultivate *P. eryngii* -Table 1). Akyüz and Kirbağ (2010) reported 14.3–18.8 mg/$g_{dry\ mushroom}$ for K for *P. eryngii* var. *ferulae*. Phosphorus content was not reported in both studies. Guo et al. (2007) also reported abundance of these macroelements in *P. djamor*: 1.21, 7.57 and 12.3 mg/$g_{dry\ mushroom}$ for Mg, P and K. According to Kalač (2013) K is highly accumulated in mushroom fruit bodies, reaching 20–40-fold higher values than in the substrate.

Among the remaining macroelements, Ca was found at rather low levels with values ranging from 170 to 226 mg/$Kg_{dry\ mushroom}$ and sodium (Na) was not detected above limit of detection. According to Cashman (2002), Ca is the specific nutrient most important for preventing and treating osteoporosis. This finding coupled to the non-detection of Na may be considered as good advantages from a nutritional aspect.

Statistically higher content of Ca was observed in *Ph. nameko* ($p<0.05$) in comparison to other mushrooms tested, but the content of Ca was not statistically different among the three cultivated *Pleurotus* spp. and *H. erinaceus*. Variable content of Ca and Na were reported in the literature for cultivated *Pleurotus* spp. For example higher content of Ca (120–700 mg/$kg_{dry\ mushroom}$) in comparison to Na (100–307 mg/$kg_{dry\ mushroom}$) were reported in *P. eryngii* var. *ferulae* (Akyüz and Kirbağ 2010) but the opposite was observed in other cultivated *P. eryngii:* 8 and 147 mg/100$g_{mushroom}$ reported by La Guardia et al. (2005) and 162.5 and 253.6 mg/$kg_{dry\ mushroom}$ reported by Lee et al. (2009) for Ca and Na, respectively. According to Lee et al. (2009) their reported Ca content in the mushroom fruit bodies was very low despite high concentrations in the substrate. The authors suggest that either Ca was present in the substrate in a less bioavailable form or mushrooms do not have efficient Ca uptake channels.

The most predominant observed microelements were Zn and Fe in the 5 cultivated mushroom species analyzed (Fig. 1). According to Khan and Tania (2012) Fe and Zn are the most abundant elements among the trace minerals in mushrooms. In particular, Fe content was high in *P. salmoneo stramineus* and in *Ph. nameko* (233 and 278 mg/$Kg_{dry\ mushroom}$) whereas Zn was high in *P. salmoneo stramineus, P. citrinopileatus* var. *cornucopiae* and in *Ph. nameko* (155, 104 and 121 mg/$Kg_{dry\ mushroom}$). The reported values were, in general, higher than those reported by Gençcelep et al. (2009) for various edible mushrooms collected from the Erzurum region of Turkey. Lee et al. (2009) reported 39.0 mg/$Kg_{dry\ mushroom}$ for Fe and 52.2 mg/$Kg_{dry\ mushroom}$ for Zn for cultivated *P. eryngii* whereas Akyüz and Kirbağ (2010) reported 519–620 mg/$kg_{dry\ mushroom}$ for Fe, 40.5–102.5 mg/$kg_{dry\ mushroom}$ for Zn for *P. eryngii* var. *ferulae.* Iron is known to be essential for cellular energy and metabolism (Jankowska et al. 2013) and its deficiency is associated to anaemia which affect adversely patients with chronic heart disease (Comín-Colet et al. 2013). Zinc is present in all organs, tissues, fluids, and secretions participating in all major biochemical pathways playing multiple roles in the perpetuation of genetic material (Brown et al. 2004). Zinc deficiency may be associated to adverse outcomes of pregnancy (King 2000), sickle cell disease (Prasad 2002) or metabolic syndrome and diabetes (Miao et al. 2013). Among the remaining trace elements it is worthwhile referring Cu contents that were clearly higher in *P. salmoneo stramineus* and in *Ph. nameko* (24.7 and 27.3 mg/$Kg_{dry\ mushroom}$) than in the other cultivated species analyzed. According to Khan and Tania (2012) the inclusion of *Pleurotus* spp. mushrooms in the diet could help to minimize Fe, Zn, Cu and other micronutrients deficiencies, but their bioavailability still have to be tested in animal and human studies since contradictory results still do persist in literature.

In general, the five cultivated mushrooms studied were of important nutritional value not only due to their proximate composition but especially due to their elemental composition. Taking current trends into account, studied mushrooms offer a wide array of nutrients, at concentrations that may meet with nutritional requirements, particularly in what concerns elements and, in some cases, may even allow for application of nutritional claims. Data on daily intake of mushrooms are unavailable for the Portuguese population and according to O'Neil et al. (2013) mushroom intake data are sparse. Considering a serving size of 84 g of fresh mushroom, reported by O'Neil et al. (2013) based on FDA food labelling information (2013) the possible contribution of analyzed mushrooms to daily nutritional requirements was calculated (Table 3). Whereas such daily intake contributes with a small fraction to protein, carbohydrate and fat requirements, in terms of microelements some of the mushrooms contribute over 15 % (minimum requirement for nutritional claim) of the recommended daily intakes (RDIs). Variable contribution

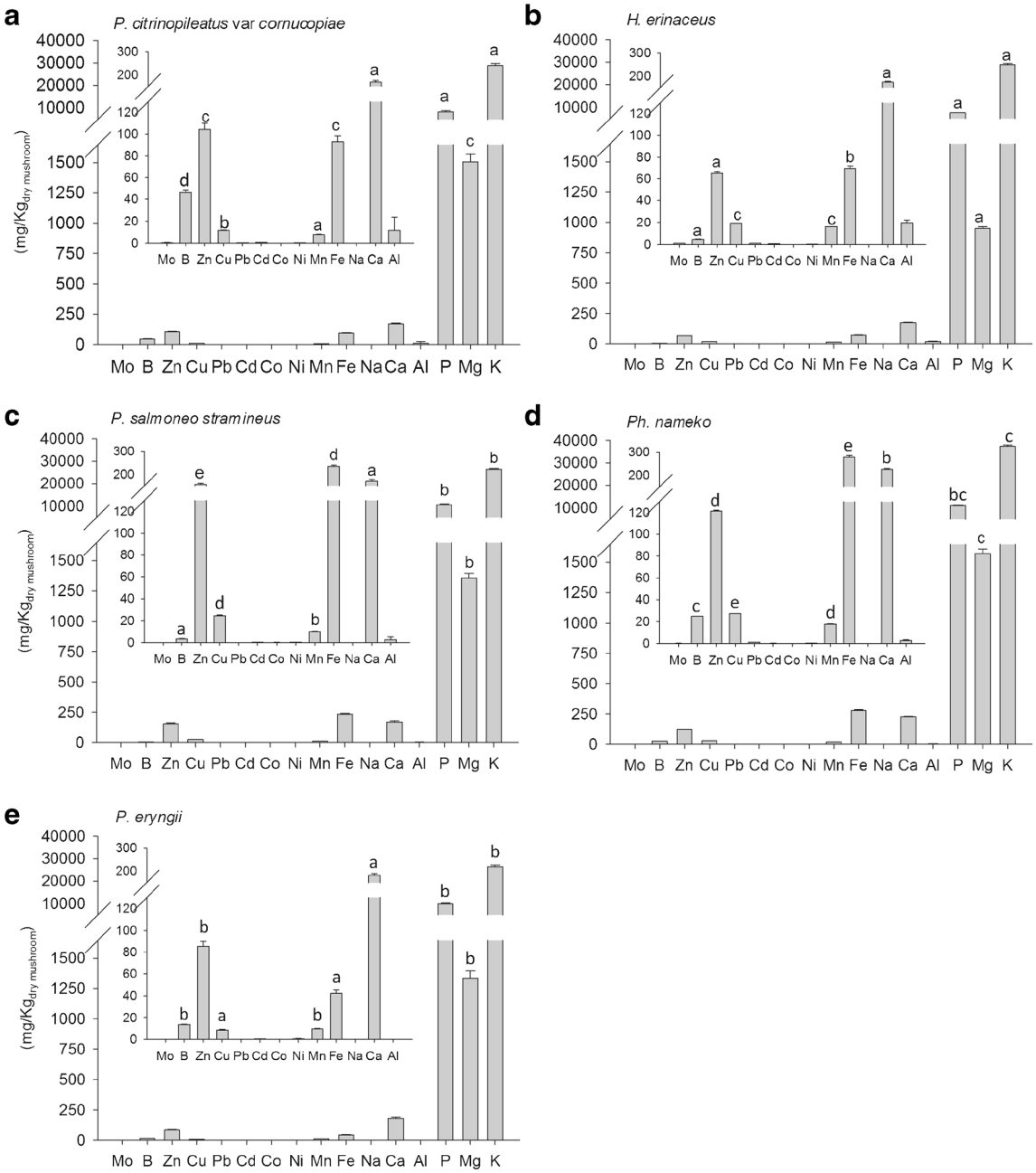

Fig. 1 Elements content (mg/kg$_{dry\ mushroom}$) in the different cultivated mushroom species: **a** *P. citrinopileatus* var. *cornucopiae;* **b** *P. salmoneo stramineus*; **c** *P. eryngii;* **d** *H. erinaceus*; **e** *Ph. nameko*. For each element, *different letters* indicate significant differences ($p<0.05$) between mushroom species

to the RDI for the elements is observable in Table 3. In terms of macro-elements all species can be considered good contributors to RDI of K, especially *Ph. nameko* with values as high as 15.7 %. In terms of P, values between 9.2 and 13.1 % RDI are reported with higher values found in *Ph. nameko*. Lower Ca and Mg suppliers characterized the cultivated species. In terms of microelements, interesting values of contribution to Fe, Cu and Zn RDI was observed in *P. salmoneo* and *Ph. nameko* with values ranging between 10 and 23 %. Lower values were, in general, found for Mn.

According to O'Neil et al. (2013) mushroom consumption is associated with a better nutrient profile and higher diet quality. For example higher intakes of Cu and K are reported by these authors for mushroom consumers than non-consumers. The recommendations by O'Neil et al. (2013) go further and state that mushroom consumption should be encouraged by health professionals. In a meta-analysis on mushroom intake and its relation to the reduction of breast cancer, Li et al. (2014) concluded that greater edible mushroom consumption may be associated with a lower risk of breast cancer.

FTIR-ATR characterization

Mushrooms are rich in polysaccharides followed by protein content with low fat content (Table 3) yet a more specific characterization of these macrostructures is a further asset to establish structure-function relationships. Several authors (Zhao et al. 2006a, b) describe the chemical characteristic features of specific regions of FTIR spectra for mushrooms truffles (Tuberaceae) and for different *Amanita* species, according to:

i) 4000–1800 cm^{-1} with a prominent broad band centered around 3300 cm^{-1}, that could be assigned to O-H and C-H stretching vibrations and two sharper bands around 2900–2880 cm^{-1} assigned to CH_2 and CH_3 stretching of fatty acids from the cell wall;

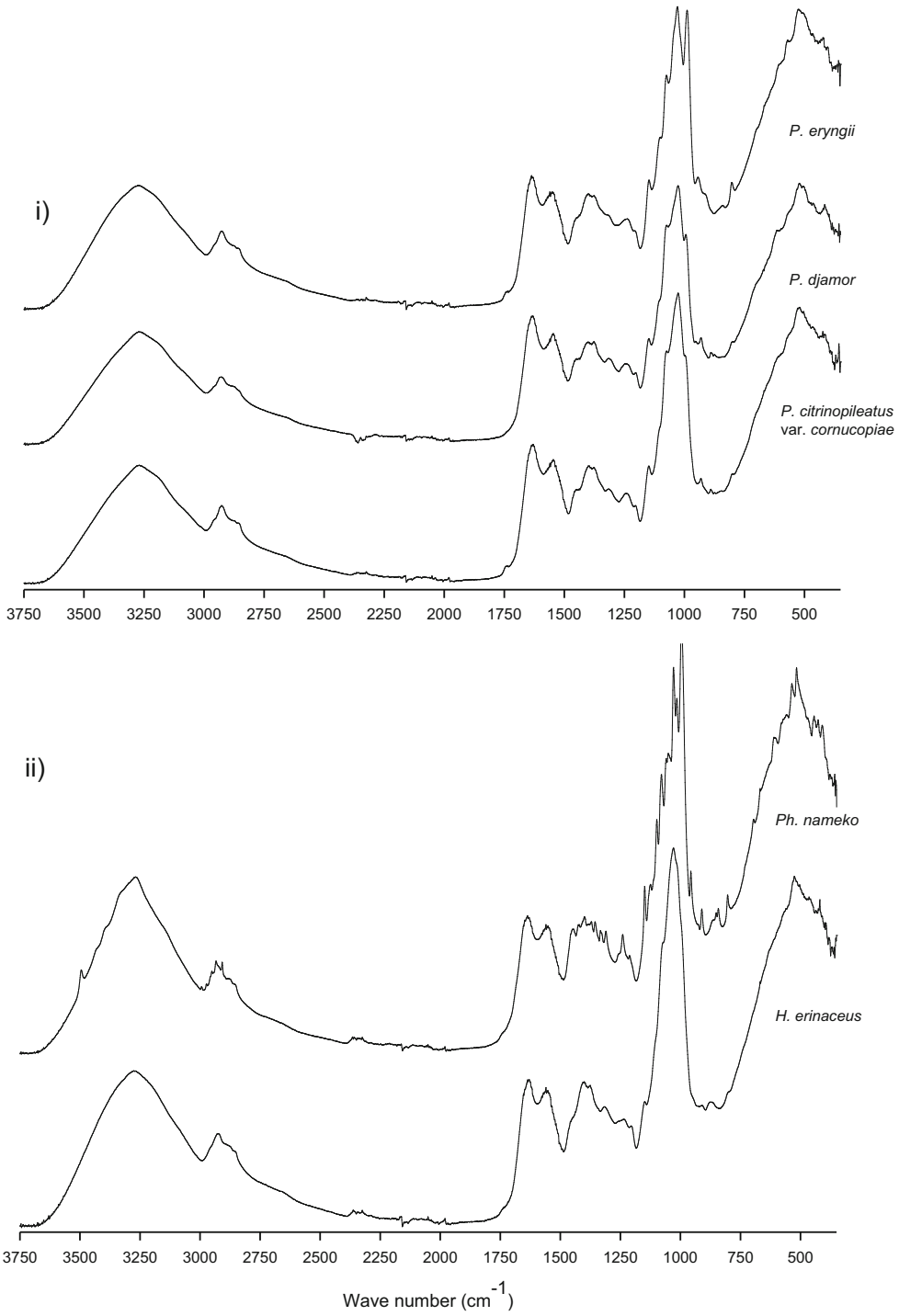

Fig. 2 FTIR-ATR spectra of the five cultivated mushroom species: **i** *P. eryngii*, *P. salmoneo stramineus* and *P. citrinopileatus* var. *cornucopiae;* **ii** *Ph. nameko and H. erinaceus*

ii) 1800–1500 cm^{-1} with two major bands around 1650 and 1560 cm^{-1} assigned to amide I and amide II of proteins; a band around 1740 cm^{-1} that could correspond to carbonyl stretching vibration of alkyl-esters indicating the presence of oil;

iii) 1500–750 cm^{-1} region associated with vibrations of proteins, lipids but also polysaccharides - 1077 and 1042 cm^{-1} have been assigned to C-O stretching of polysaccharides;

iv) 950–750 cm^{-1} region that has been associated with identification of anomeric configuration of polysaccharides - 890 cm^{-1} band has been assigned to β-glycosides and 860–810 cm^{-1} for α-glycosides. This information is equally supported by Mohaček-Grošev et al. (2001) studies who used vibrational spectroscopy to characterize several wild growing mushroom species.

In Fig. 2, the 4 specific regions are observable for the 5 mushroom species. At first sight there seems to be little qualitative difference between spectra of the 3 cultivated *Pleurotus* species (Fig. 2i); apparent differences are more evident in *Ph. nameko* spectrum especially in the 1500–750 cm^{-1} region which is associated with vibrations of proteins, lipids but also polysaccharides (Fig. 2ii). Evidence of presence of proteins (due to 1650 and 1560 cm^{-1} bands), fatty acids (due to two sharper bands around 2900–2880 cm^{-1}) and polysaccharides (bands in 1500–750 cm^{-1} region) are easily perceived in the 5 cultivated mushroom species spectra taking into account the band assignments by Zhao et al. (2006b) and Mohaček-Grošev et al. (2001) According to Liu et al. (2006) the region between 750 and 1200 cm^{-1} could serve as fingerprints to discriminate mushrooms whereas according to Mohaček-Grošev et al. (2001) the spectral region between 1200 and 1000 cm^{-1} could serve as an indicator of mushroom genus.

In Figs. 3 and 4, the 1500–750 cm^{-1} region is amplified enabling a more detailed observation of the bands present in

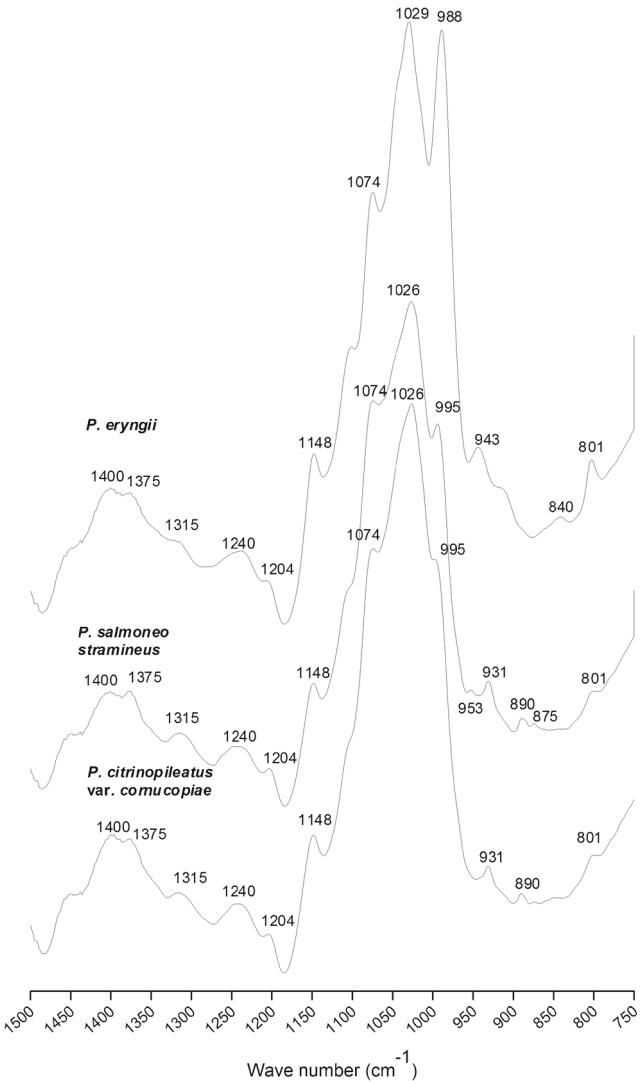

Fig. 3 FTIR-ATR spectra of the three *Pleurotus* sp. (*P. eryngii*, *P. salmoneo stramineus* and *P. citrinopileatus* var. *cornucopiae*) mushrooms between 1500 and 750 cm^{-1}

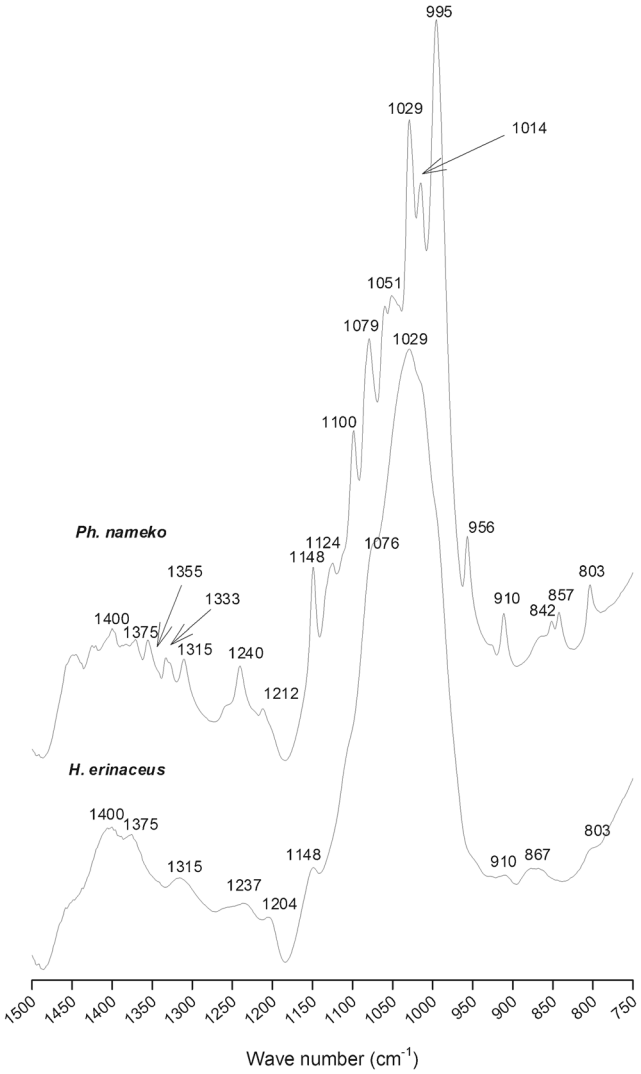

Fig. 4 FTIR-ATR spectra of *Ph. nameko and H. erinaceus* between 1500 and 750 cm^{-1}

this region. Characteristic bands found in the 5 spectra are 1400, 1375, 1315, 1237–1240, 1204–1212, 1148, 1074–1079, 1026–1029, 995–988, 890–910 and 801–803 cm^{-1}. The majority coincide with main observed bands in the 5 Amanitas species and the 5 truffles studied (Zhao et al. 2006a, b). According to Gonzaga et al. (2005) 1028 cm^{-1} is assigned to C-O stretching, 1074 cm^{-1} to anomeric C_1H group vibration and 1165 to C-O-C stretching of glycosidic structures. Protein patterns have been associated with characteristic absorption at 1654, 1544, 1409 and 1242 cm^{-1}; a similar pattern was found in the 5 cultivated mushroom species studied which according to Gonzaga et al. (2005) may be evidence of the presence of a glucan-protein complex.

Although bands between 950 and 750 are very weak they are considered important for the identification of anomeric configuration of polysaccharides (Mohaček-Grošev et al. 2001; Barbosa et al. 2003). According to Zhao et al. (2006a, b) β-glucan and chitosan standards with β-glycosidic linkage presented bands at 889 and 897 cm^{-1}, respectively, whereas α-glycosidic linkage typical in a standard starch presented a characteristic band at 858 cm^{-1}. According to Barbosa et al. (2003) bands at 890 and 1370 $cm^{-1}$are typical of (1→3)-β-glucans and of β-glucans, respectively. In *P. citrinopileatus* var. *cornucopiae* and *P. salmoneo stramineus* spectra (Fig. 3) as well as in *Ph. nameko* and *H. erinaceus* spectra (Fig. 4), two weak bands at 890–910 cm^{-1} and 801–803 cm^{-1} were observable; such could indicate that both α- and β-glycosidic linkages exist in these mushroom species. In addition, the detection of the band at 1375 cm-^{1} in the five cultivated mushroom species suggests the existence of β-glucans. According to Wasser (2002) mushroom polysaccharides are present mostly as glucans with different types of glycosidic linkages, such as (1→3), (1→6)-β-glucans and (1→3)-α-glucans but also as heteroglucans; in this case, side chains contain glucuronic acid, xylose, galactose, mannose, arabinose, or ribose as a main component or in different combinations.

Possible discriminant bands between the 3 cultivated *Pleurotus* species are the bands at 943 and 840 cm^{-1} only present in *P. eryngii*, 953 and 875 cm^{-1} only present in *P. salmoneo stramineus*, whereas the band at 931 cm^{-1} is visible in *P. salmoneo stramineus* and *P. citrinopileatus* var. *cornucopiae* (Fig. 3). In turn, possible discriminant bands for *Ph. nameko* are observable in the 1500–1000 cm^{-1} region such as 1124, 1100, 1014 cm^{-1} and in the 950–750 cm^{-1} region such as 956, 842 and 851 cm^{-1}. The *Ph. nameko* spectrum is characterized by more and diverse distinct bands than the other 4 cultivated mushroom species including in the 1500–1200 cm^{-1} region. A broad weak band around 867 cm^{-1} appears to be discriminative for *H. erinaceus* (Fig. 4).

Zhao et al. (2006a, b) suggested several absorption ratios to discriminate between mushroom species. In accordance, the absorption ratios $A_{1029/1148}$, $A_{1029/1074}$ were calculated for each cultivated mushroom species. The ratio $A_{1029/1148}$ did not reveal to be discriminant (1.29–1.10), which is not the case for the $A_{1029/1074}$ ratio where clear differences were detected; calculated values were 1.24, 1.18, 0.41, 0.41 and 0.38 for *P. eryngii*, *P. salmoneo stramineus*, *P. citrinopileatus* var. *cornucopiae, H. erinaceus* and *Ph. nameko*, respectively.

Conclusions

The chemical composition of five cultivated edible mushrooms in terms of proximate composition, fatty acids profile and elemental composition showed significant differences among the different species, emphasizing the nutritional potential of the five different cultivated edible mushrooms to be consumed in a healthy diet. High contents in proteins and polysaccharides associated with low content of fat, which profile is characterized by higher concentration in mono and polyunsaturated FA than in saturated FA, being also interesting sources of phenolic compounds as well as of some macro and micronutrients highlights its potential as healthy food. According to FTIR-ATR spectra, the presence of β-glucans, α-glucans and glucan-protein complexes are among main representative polysaccharides in the five species. The presence of these polysaccharides further upholds the interest in exploring these mushrooms for applications in health-related fields, for example, drug or nutraceutical delivery approaches.

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Conflicts of interest The authors declare that there are no conflicts of interest.

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