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Circular reuse of bio-resources: the role of *Pleurotus* spp. in the development of functional foods

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The basidiomycetes fungi belonging to the genus *Pleurotus* could make an important contribution to sustainable functional food design because they possess an elevated protein content with a valuable essential amino acid scoring pattern, a unique dietary fibre profile, mainly comprised of branched β-glucan, high levels of some vitamins of the B group, vitamin D, Fe, Zn, Cu, Se and some bioactive mycochemicals, while the Na and fat contents are low. Moreover, *Pleurotus* spp. can grow efficiently on various clean by-products of food processing, such as wheat straw, wheat stalk and spent beer grain, thus representing a sustainable food source. This review illustrates the compositional variability of *Pleurotus* spp. grown on various by-products, in order to clarify its potential ability to address the needs of populations with endemic nutritional deficiencies as well as those populations at risk or affected by some chronic diseases. The perspectives for *Pleurotus* applications in functional foods decisively depend on consumers' acceptability. Hence, the sensory properties of *Pleurotus* spp. are also clarified herein. Lastly, the three main strategies of functional food development using *Pleurotus* spp. are summarized, namely its use as a fortifying agent, high-cost protein replacer and prebiotic ingredient.

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Introduction

Global food production is facing an uphill task to address many relevant challenges nowadays, including growing populations, the effects of climatic change on agricultural production, the noticeable impact of the agro-food system on the environment and an imbalanced economic situation caused by the worldwide financial crisis.[1,2] Additionally, with the surge in the incidence of cardiovascular disease, type-2 diabetes and cancer, there is a need to develop new dietary strategies, and to develop foods that could potentially support disease prevention.[3]

In this context, the basidiomycetes fungi belonging to the genus *Pleurotus* can make a valuable contribution because they combine the ability to grow with a negligible use of bioresources and can support the production of value-added foods. Indeed, *Pleurotus* spp. are a fast growing fungi that can be obtained with limited capital investment and technical skill, both in temperate and in tropical regions. Moreover, *Pleurotus* spp. can use various by-products from the food industry as growth substrates, since they efficiently decompose lignocellulose-rich substrates due to their enzymatic complexes, including phenol oxidases and peroxidases.[4] Through this conversion, *Pleurotus* spp. yield a fungal biomass that represents a source of protein with good levels of essential amino acids, dietary fibre with unique structural features (branched β-glucans), vitamins, minerals and low-molecular weight bioactive compounds, also known as mycochemicals.[5] The nutritional value of *Pleurotus* spp. has long been recognized.[6–8] Moreover, *Pleurotus* spp. are becoming increasing attractive as sources for the development of new drugs and functional foods due to their potential antioxidant, antimicrobial, antiproliferative, immunomodulatory, anti-inflammatory and antihypertensive properties.[5]

Around 200 species of *Pleurotus* have been identified, but only a few have been used for food applications to date, namely *P. ostreatus*, *P. eryngii*, "*P. sajor-caju*" and *P. pulmonarius*. The name "*P. sajor-caju*" is considered improper because either it has been used for a tropical ecotype of *P. pulmonarius* or it has been incorrectly used for a species belonging to the genus *Lentinus*, which was later named as *Lentinus sajor-caju* (Fr.) Fries.[9] A number of studies have been performed to characterize *Pleurotus* spp. compositions. However, these produced some contradictory results regarding the identification and quantification of some of its components, which raises attention to the methodology applied.[10,11] There are also a growing number of studies on the use of this mushroom in new functional foods. The aim of this review is to summarize the existing literature information on the composition, nutritional value, health studies performed on humans, perception of sensory attributes and acceptability, and food applications of the most common species of *Pleurotus*, in order to evaluate the potential ability of this mushroom to address the needs of populations with endemic nutritional deficiencies and/or to act as a dietary supplement in the prevention of some diseases.

Composition and nutritional value

Dietary fibres and purified β-glucan fractions

The health effects of *Pleurotus* spp. are mainly due to its fibre fraction, which is comprised of glucans, chitin, mannoproteins, galactomannans, cellulose and polyglucuronic acids.[12] The total dietary fibre content found in various studies is in the range 10.58–56.99 g per 100 g of fruit body dry weight (d.w.) (Table 1).[7,8,13–21] The AOAC enzymatic gravimetric method is the most frequently used to determine dietary fibre contents in mushrooms. However, the presence in the residue of non-protein nitrogen (N) originating from chitin (which is generally not mentioned) impairs the calculation and could be one of the reasons for the different values found by various authors. Nevertheless, the effect of the growth substrate on the dietary fibre content seems to be important, whereby the use

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of wheat stalk and straw has resulted in high dietary fibre contents,[13–15] while olive by-products have yielded a lower dietary fibre content in the literature studies.[15]

Among the constituents of the dietary fibre of *Pleurotus* spp., β-glucans are the major components. These polysaccharides have a backbone of D-glucose-linked β-(1→3) with no branches or variable amounts of β-(1→6) branches.[12] The glucose chains of β-glucans are twisted and create a single or a triple helix stabilized by inter-chain hydrogen bonds.[22,23] The array of relative molecular weights of β-glucans is quite wide ranging, from tens to thousands of kilodaltons.[23] Regarding the amount of β-glucans, assays based on enzymatic hydrolysis with β-glucanase have yielded low values. Hence, it has been recommended to calculate the β-glucan content as the difference between the total glucans (by measuring glucose obtained through a controlled acid hydrolysis) and the α-glucans (by measuring glucose released from α-glucans through enzymatic hydrolysis with α-amylase and amyloglucosidase).[10] Using this latter approach, a study on the intraspecific variability among 16 strains of *P. ostreatus* mushrooms revealed that the total glucan content varied in the range 14–25 g per 100 g d.w., with β-glucans in the range of 10.9–22.9 g per 100 g d.w.[24] A higher β-glucan content, *i.e.* 32.3 g per 100 g d.w., was also observed in one strain of *P. ostreatus*.[10] Similarly, considering two strains of *P. eryngii*, the β-glucan content was found to vary between 23.85 and 37.1 g per 100 g d.w.[10,17] Beside the genetic factors, the growing conditions affect the β-glucan content: substrates with a high content of polyphenolic compounds induce an increased synthesis of β-(1→3) D-glucan synthetase in their fruiting bodies. Factors such as the C/N ratio, pH of the substrate and the incubation temperature are also important and species specific.[23] However, knowledge of the effects of the growing conditions on the β-glucan content is still scarce.

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Parameters such as the main chain structure, degree of branching and molecular weight affect the solubility of the β-glucans. Procedures to recover concentrated hot-water-soluble, alkali-soluble and insoluble β-glucan fractions from *Pleurotus* spp. have been proposed (Table 2, Fig. 1). Nevertheless, the structure–bioactivity relationship of *Pleurotus* β-glucans has not been clarified yet.[25] Karacsonyi *et al.*[26] purified an alkali-insoluble fraction obtained from one strain of *P. ostreatus* and found that it was composed of branched β-(1→3),β-(1→6)-D-glucans with trace branched β-(1→3),β-(1→4)-D-glucans. This fraction was referred to as pleuran and accounted for 4.6% of the fruit body d.w.[26] Carbonero *et al.*[27] obtained a highly purified β-glucan fraction from both one strain of *P. ostreatus* and one strain of *P. eryngii*, through freezing of the hot water soluble fraction followed by mild thawing at 4 °C. However, the recovery yields for this purified fraction were low, *i.e.* 2.7 and 2.5 g per 100 g of the fruit body d.w. for *P. ostreatus* and *P. eryngii*, respectively.[27] By another approach, Synytsya *et al.*[28] isolated and characterized both hot-water-soluble, alkali-soluble and insoluble-glucan rich fractions from four strains of *P. ostreatus* and one strain of *P. eryngii*. The hot-water-soluble fraction mainly contained branched β-(1→3),β-(1→6)-D-glucans (44.2–72.0 g per 100 g d.w. in *P. ostreatus* and 33.6 g per 100 g d.w. in *P. eryngii*) with proteins and traces of both heteropolysaccharides and starch; while the alkali-soluble fraction mainly contained linear α-(1→3)-D-glucans (45.9–71.2 g per 100 g d.w. in *P. ostreatus* and 55.4 g per 100 g d.w. in *P. eryngii*) with proteins and traces of both heteropolysaccharides and starch. The residue mainly contained branched β-(1→3),β-(1→6)-D-glucans (65.8–86.9 g per 100 g d.w. in *P. ostreatus* and 66.4 g per 100 g d.w. in *P. eryngii*) with starch, heteropolysaccharides and chitin. Considering a moisture content of 10% for the fruit body, the yields of the water-soluble and alkali-soluble fractions were ∼5% d.w., while that of the residue was ∼30% d.w. In *P. ostreatus*, removal of proteins from the hot-water-soluble and alkali-soluble fractions increased the glucan contents to 78.9–85.0 g per 100 g d.w. (deproteinized hot-water-soluble fraction) and 84.3–89.2 g per 100 g d.w. (deproteinized alkali-soluble fraction).[28]

Proteins

As the world's population increases rapidly and against the constraints of limited land, water and food resources, it is important to find efficient protein sources to meet human nutritional needs.[29] The protein content of foods is generally determined on the basis of total N content as evaluated by the Kjeldahl method, which is then multiplied by the conversion factor 6.25.[30] However, regarding edible mushrooms, many studies have indicated a probable digestibility of 60% to 70% for protein calculated as N × 6.25, due to their noteworthy amount of non-protein N in the form of glucosamine in their chitinous cell walls.[31] Hence, a conversion factor for N content

Table 1 Major components of the fruit bodies of *Pleurotus* spp. (g per 100 g d.w.) grown on different waste substrates or collected from the market

Pleurotus species and growth substrate[a]	Total dietary fibre[b]	Protein[b,c,d]	Fat[b]	Ash[b]	Ref.
P. ostreatus					
Unspecified	N.R.[e]	19.93 ± 0.20[c]	N.R.[e]	7.80 ± 0.74	6
Unspecified	N.R.[e]	34.73 ± 0.35[c]	N.R.[e]	8.49 ± 1.40	6
Unspecified	47.3 ± 0.7	18.59 ± 0.23[c]	4.16 ± 0.23	10.3 ± 0.1	7
Unspecified	30.0 ± 0.1	21.8 ± 0.1[d]	4.4 ± 0.1	8.0 ± 0.1	8
Unspecified	N.R.[e]	14.7 ± 0.04[c]	1.53 ± 0.25	5.69 ± 0.64	32
Blank paper	N.R.[e]	9.71 ± 0.02[c]	1.18 ± 0.01	15.9 ± 1.2	32
Printed paper	N.R.[e]	9.29 ± 0.08[c]	1.68 ± 0.49	10.5 ± 0.8	32
Spent beer grain + wheat bran	N.R.[e]	32.4 ± 0.1[d]	4.4 ± 0.1	7.3 ± 0.1	33
Spent beer grain + wheat bran	N.R.[e]	37.4 ± 0.1[d]	4.3 ± 0.1	6.7 ± 0.1	33
Maize straw	N.R.[e]	22.25 ± 0.51[d]	N.R.[e]	N.R.[e]	34
Pumpkin straw	N.R.[e]	21.24 ± 0.51[d]	N.R.[e]	N.R.[e]	34
Wheat stalk	30.25 ± 0.12	17.99 ± 0.65[d]	2.60 ± 0.22	4.78 ± 0.04	13
Wheat stalk	30.25 ± 0.06	17.10 ± 0.56[d]	2.59 ± 0.12	4.79 ± 0.03	14
Cotton stalk	29.80 ± 0.04	14.97 ± 0.76[d]	2.90 ± 0.10	4.60 ± 0.01	14
Soybean stalk	27.0 ± 0.06	22.15 ± 0.15[d]	2.45 ± 0.05	4.85 ± 0.03	14
Millet stalk	31.32 ± 0.12	14.77 ± 0.19[d]	3.15 ± 0.21	4.71 ± 0.04	14
Olive mill by-products	12.50 ± 5.44	19.74 ± 1.19[c]	2.72 ± 0.23	9.48 ± 1.93	15
Almond + walnut shells	13.00 ± 0.53	31.36 ± 0.57[c]	2.49 ± 0.25	9.86 ± 0.27	15
Beech sawdust	15.78 ± 0.61	16.06 ± 1.76[c]	3.46 ± 1.14	6.21 ± 0.12	15
Corn cobs	13.76 ± 4.13	15.41 ± 0.78[c]	3.37 ± 0.65	8.02 ± 0.49	15
Wheat straw	19.07 ± 2.33	14.64 ± 1.38[c]	2.56 ± 0.17	8.56 ± 0.89	15
Olive-press cake	13.68 ± 2.54	21.41 ± 2.34[c]	1.64 ± 0.35	6.98 ± 1.26	15
Pine needles	13.68 ± 0.16	22.74 ± 0.04[c]	2.44 ± 0.25	7.50 ± 0.67	15
P. eryngii					
Unspecified	N.R.[e]	22.89 ± 0.17[c]	N.R.[e]	10.55 ± 0.31	6
Unspecified	N.R.[e]	22.74 ± 0.11[c]	N.R.[e]	9.16 ± 0.26	6
Unspecified	25.9 ± 3.2	16.42 ± 0.75[c]	5.97 ± 0.01	9.0 ± 0.74	16
Unspecified	43.34 ± 1.04	N.R.[e]	N.R.[e]	N.R.[e]	17
Wheat stalk	28.45 ± 0.09	12.55 ± 0.98[d]	7.50 ± 0.08	4.89 ± 0.06	13
P. sajor-caju					
Unspecified	56.99 ± 0.01	22.41 ± 0.01[c]	2.30 ± 0.01	7.79 ± 0.01	18
Straw	13.3 ± 0.1	18.6 ± 0.1[d]	2.00	6.5 ± 0.1	19
Cotton waste	14.1 ± 0.1	21.2 ± 0.1[d]	1.70	6.7 ± 0.1	19
Cotton waste + straw	11.4 ± 0.1	21.3 ± 0.1[d]	2.00	6.6 ± 0.1	19
Cotton waste + tea leaves	14.5 ± 0.1	25.0 ± 0.1[d]	1.70	6.4 ± 0.1	19
Paddy straw	12.3 ± 0.26	29.03 ± 1.03[c]	0.9 ± 0.06	6.8 ± 0.48	20
Wheat stalk	30.67 ± 0.12	17.59 ± 1.07[d]	1.15 ± 0.18	5.84 ± 0.09	13
Bean straw	16.55 ± 0.01	16.30 ± 0.01[c]	3.26 ± 0.01	6.26 ± 0.01	21
Apple pomace	10.58 ± 0.01	24.44 ± 0.01[c]	3.84 ± 0.01	6.12 ± 0.01	21
Grape bagasse	19.60 ± 0.01	27.83 ± 0.01[c]	3.12 ± 0.01	7.05 ± 0.01	21
Wheat straw	N.R.[e]	29.36 ± 0.44[c]	2.07 ± 0.06	8.05 ± 0.13	35
P. pulmonarius					
Unspecified	N.R.[e]	30.48 ± 0.22[c]	N.R.[e]	8.35 ± 0.77	6
Straw + wheat bran	N.R.[e]	15.9 ± 2.5[d]	N.R.[e]	N.R.[e]	36

[a] Unspecified substrate means that data refer to wild or cultivated mushrooms collected from the market. [b] Data obtained by the AOAC procedure. [c] Data were expressed as N × 4.38. [d] Data were recalculated as N × 4.38. [e] N.R.: not reported.

equal to 4.38 (*i.e.* 0.7 × 6.25) was proposed. This latter conversion factor was used to fill out Table 1, to obtain a close approximation of protein content of four *Pleurotus* spp., which resulted in values varying from 9.29 to 37.4 g per 100 g of fruit bodies d.w. (Table 1).[6–8,13–16,18–21,32–36] Indeed, these mushrooms are considered as a good source of protein, especially for vegetarians. Overall on a dry basis, the protein content of *Pleurotus* spp. is higher than that of rice, *i.e.* 7.1–8.3 g per 100 g d.w.,[37] which is one of the major crops contributing to the human food supply.[38] Moreover, *Pleurotus* spp. has a good essential amino acid scoring pattern for human needs.[39] The species and strain, stage of maturation, part of the mushroom body, harvest location and most of all the composition of the substrate all have a significant effect on the protein content of *Pleurotus* fruit bodies.[31] Interestingly, the biomass of *Pleurotus* spp. rich in high quality protein can be obtained through the conversion of agro-wastes (Table 1). Regarding *P. ostreatus*, the lowest amount of crude protein (9.29 g per 100 g d.w.) was found in the fruit body grown on printed paper,[32] while the highest amount (37.4 g per 100 g d.w.) was achieved when spent beer grain added with wheat bran was used as the substrate.[33] For *P. eryngii* and *P. pulmonarius*, data on the effect of the substrate on their protein content are lacking, but it is noticeable that the protein content of these species grown on wheat stalk was found to be lower than that observed in wild and commercial mushrooms of the same species. The lowest protein content of *P. sajor-caju* (16.30 g per 100 g d.w.) was found on a bean straw medium,[21] while the highest protein

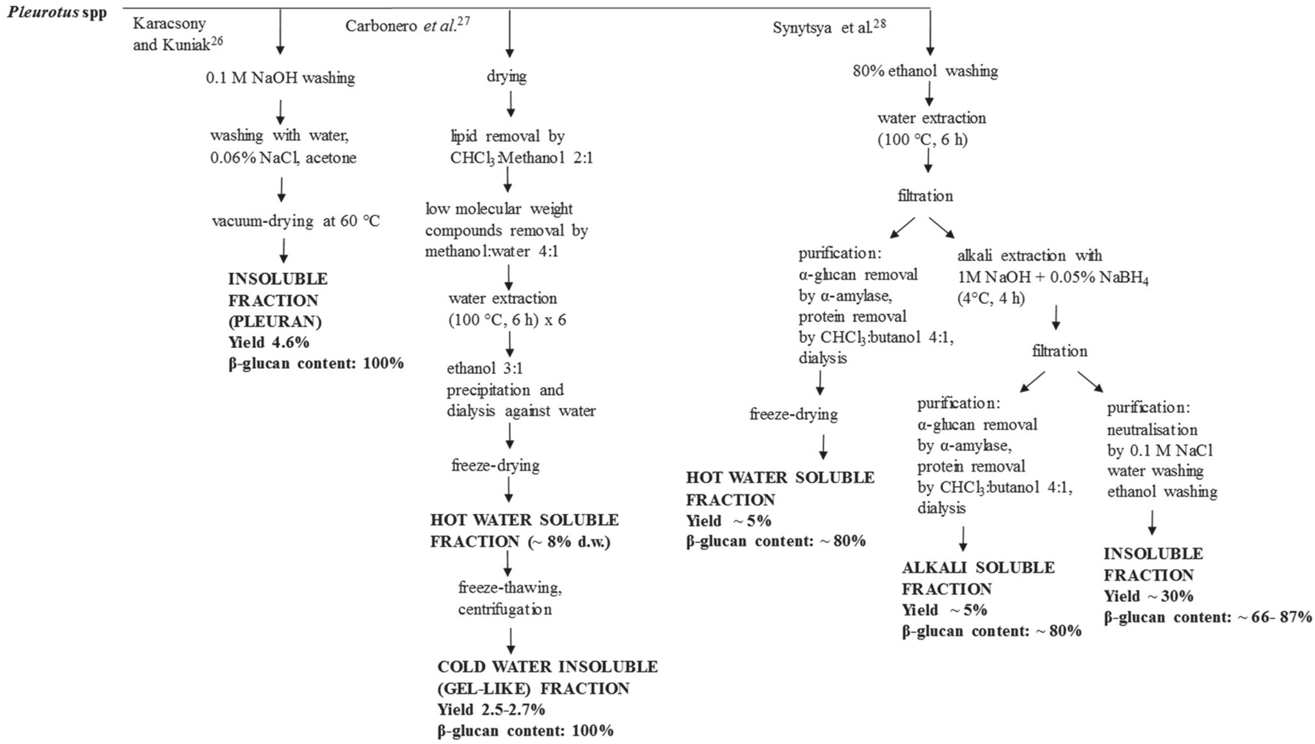

Fig. 1 Proposed processes to obtain concentrated and purified β-glucan rich fractions from *Pleurotus* spp. The yield, β-glucan content and composition of the fractions are shown in Table 2.

Table 2 β-Glucan content in the fruit body of *Pleurotus* spp. and in fractions (g per 100 g d.w.), fraction yield (g fraction per 100 g of fruit body d.w.) and composition

Pleurotus species and fruit body/fraction	β-Glucan content	Yield[a]	Composition	Ref.
P. ostreatus				
Fruit body	10.9 ± 0.01–32.3 ± 0.1			10 and 24
Alkali-insoluble fraction (pleuran)	100	4.6	Branched β-(1→3),β-(1→6)-D-glucans; branched β-(1→3),β-(1→4)-D-glucans (traces)	26
Hot-water-soluble fraction	100	2.7	Branched β-(1→3),β-(1→6)-D-glucans	27
Hot-water-soluble fraction	44.2 ± 0.1–72.0 ± 0.1	5[a]	Branched β-(1→3),β-(1→6)-D-glucans; heteropolysaccharides and starch (traces); proteins	28
Alkali-soluble fraction	45.9 ± 0.1–71.2 ± 0.1	5[a]	Linear α-(1→3)-D-glucans glucan; heteropolysaccharides and starch (traces); proteins	28
Insoluble fraction	65.8 ± 0.1–86.9 ± 0.1	30[a]	Branched β-(1→3),β-(1→6)-D-glucans; heteropolysaccharides and starch; chitin	28
Deproteinized hot-water-soluble fraction	78.9 ± 0.1–85.0 ± 0.1	N.R.[b]	Branched β-(1→3),β-(1→6)-D-glucans; heteropolysaccharides and starch (traces); proteins (traces)	28
Deproteinized alkali-soluble fraction	84.3 ± 0.1–89.2 ± 0.1	N.R.[b]	Branched 1,3-1,6-β-D-glucan; heteropolysaccharides and starch (traces); proteins (traces)	28
P. eryngii				
Fruit body	23.85 ± 1.60–37.1 ± 0.1			10 and 17
Hot-water-soluble fraction	100	2.5	Branched β-(1→3),β-(1→6)-D-glucans	27
Hot-water-soluble fraction	33.6 ± 0.1	5[a]	Branched β-(1→3),β-(1→6)-D-glucans; heteropolysaccharides and starch (traces); proteins	28
Alkali-soluble fraction	55.4 ± 0.1	5[a]	Linear α-(1→3)-D-glucans glucan; heteropolysaccharides and starch (traces); proteins	28
Insoluble fraction	66.4 ± 0.1	30[a]	Branched β-(1→3),β-(1→6)-D-glucans; heteropolysaccharides and starch; chitin	28

[a] The yield was transformed from a fresh basis to a dry basis considering a dry matter content of the fruit body of 10 g per 100 g f.w. [b] N.R.: not reported.

content (29.36 g per 100 g d.w.) was obtained when growing *P. sajor-caju* on a wheat straw substrate.[35]

It is noteworthy that the protein of *Pleurotus* spp. generally meets the essential amino acid scoring patterns recommended for children, adolescents and adults[29] (Table 3). Regarding wild *Pleurotus* mushrooms, some strains of *P. eryngii*, *P. ostreautus* and *P. sajor-caju* were found to meet the reference pattern for children and adults, while for *P. pulmonarius*, leucine and lysine contents were limited.[6] In general, the levels of histidine and threonine in *Pleurotus* spp. protein are also good with respect to those recommended for infants, but the other essential amino acids are limited for infants' requirements. The use of wheat stalk as a growth substrate for *P. ostreatus*, *P. eryngii* and *P. sajor-caju* has led to very good essential amino acid scoring patterns, with isoleucine, threonine, valine[13,35] and aromatic amino acids[13] also meeting the infants' requirements.

Besides proteins, mushrooms contain free amino acids, among which glutamic acid (Glu) is prevalent. The typical presence of this amino acid is one of the factors that allow these mushrooms to be used as a functional food or as a raw material for functional foods.[40] In fact, free Glu plays an important physiological role in the process of digestion, nutrient absorption and energy homeostasis *via* the gut–brain axis. These activities are mediated *via* several receptors in the oral cavity, where Glu is responsible for the "umami taste" (as described under the sensory attributes and perception paragraph). Moreover, Glu stimulates luminal gut glutamic acid sensors that are linked to the afferent branches of the vagus nerve, which in turn modulates a number of target areas in the brain, thus enhancing the secretion of digestive juices and insulin.[41] Only a limited number of *Pleurotus* species have been analysed for free Glu content. In some strains of *P. ostreatus* and *P. eryngii*, contents of the free form of this amino acid were found to be in the range 0.071–4.109 g per 100 g d.w.[40,42–46] The effects of the growth substrates on free Glu content have also been poorly investigated. In *P. eryngii*, sawdust was found to be beneficial for the free Glu content with respect to corncob.[45]

Fats

Pleurotus spp. are low in fat content. Previous reports have found that the fat content ranges between 1.18 and 4.4 g per 100 g d.w. in *P. ostreatus*, between 5.97 and 7.5 g per 100 g d.w. in *P. eryngii* and between 0.9 and 3.84 g per 100 g d.w. in *P. sajor-caju* (Table 1).[7,8,13–16,18–21,32–36] The fat fraction of mushrooms includes, in general, representative compounds of all classes of lipids, namely, free fatty acids, mono-, di- and triglycerides, sterols, sterol esters and phospholipids. Triglycerides are prevalent, while squalene, ergosterol (free and esterified) and ubiquinone have also been reported as minor components.[31] In an intraspecific study on 16 strains of wild *P. ostreauts*, polyunsaturated fatty acids were found to be the most prevalent, ranging between 58.84% and 80.63% of total fatty acids, while monounsaturated fatty acids were between 6.76% and 20.29% of total fatty acids and saturated fatty acids were between 8.77 and 17.07% of total fatty acids. Linoleic acid dominated in all samples (56.8–80.5%) followed

Table 3 Recommended essential amino acid scoring patterns for different age groups (mg g^{-1} protein) and essential amino acid content (mg g^{-1} protein) of the fruit bodies of *Pleurotus* spp. grown on different waste substrates or collected from the market

Groups	Recommended essential amino acid scoring pattern									Ref.
	HIS	ILE	LEU	LYS	SAA[a]	AAA[b]	THR	TRP	VAL	
Infant (birth to 6 months)	21	55	96	69	33	94	44	17	55	29
Child (6 months to 3 years)	20	32	66	57	27	52	31	8.5	43	29
Older child, adolescent, adult	16	30	61	48	23	41	25	6.6	40	29
Pleurotus species and growth substrate[c]	Amino acid content									
P. ostreatus										
Unspecified	43.3	53.2	76.5	69.9	41.9	99.6	61.6	14.8	55.3	6
Unspecified	44.1	24.2	38.9	37.5	16.5	45.9	29.7	6.6	26.1	6
Spent beer grain + wheat bran	32.0	41.8	66.3	59.1	19.6	73.5	44.1	12.4	54.2	33
Maize straw	60.6	52.5	68.2	87.2	N.R.[e]	N.R.[e]	N.R.[e]	N.R.[e]	49.8	34
Pumpkin straw	89.6	44.8	71.2	86.9	N.R.[e]	N.R.[e]	N.R.[e]	N.R.[e]	51.1	34
Wheat stalk	N.D.[d]	54.9	90.9	62.7	15.0	100.2	52.4	N.R.[e]	58.4	13
P. eryngii										
Unspecified	29.9	35.4	66.9	63.5	31.1	73.5	49.0	12.4	37.0	6
Wheat stalk	19.9	57.9	85.8	58.2	N.R.[e]	96.0	54.0	N.R.[e]	59.0	13
P. sajor-caju										
Unspecified	22	44	70	57	30	113	55	12	53	31
Wheat stalk	N.D.[d]	63.7	83.1	32.7	N.R.[e]	86.4	50.9	N.R.[e]	57.1	13
Wheat straw	34.7	43.6	69.5	53.5	41.2	N.R.[e]	55.9	N.R.[e]	85.5	35
P. pulmonarius										
Unspecified	29.5	43.4	31.6	28.5	26.2	51.7	64.2	11.3	53.1	6

[a] SAA (Sulphur Amino Acids): CYS + MET. [b] AAA (Aromatic Amino Acids): PHE + TYR. [c] Unspecified substrate means that data refer to wild or cultivated mushrooms collected from the market. [d] N.D.: not determined. [e] N.R.: not reported.

by oleic and palmitic acids.[24] A similar fatty acids profile was observed for the samples obtained with paper scraps as the substrate.[32] The fatty acid profile of one strain of *P. eryngii* was found to be different, with a profile of 25.79%, 49.05% and 25.17% saturated fatty acids, monounsaturated and polyunsaturated fatty acids with respect to total fatty acids, respectively.[47]

Organic acids and soluble sugars/polyols

The amounts of organic acids and sugars/polyol of *Pleurotus* spp. were found to vary in the ranges 3.0–9.8 g per 100 g d.w and 15.6–32.9 g per 100 g d.w., respectively.[32,45,47] The patterns of organic acids reported by various studies showed some differences: in one strain of *P. ostreatus*, citric acid, oxalic acid, fumaric acid and malic acid were observed.[48] The same pattern was found in a *P. erjngii* strain,[47] while in another *P. ostreatus* strain, quinic acid was detected instead of malic acid.[32] Among the sugars and polyols, trehalose, mannitol and glucose were found to be the most prevalent in the *Pleurotus* genus.[32,35,45,47] The amount of sugars and polyols in *Pleurotus* spp. greatly depends on the growth substrate. Using printed paper and blank paper as the substrate for *P. ostreatus*, the amounts of sugars/polyols were 9.45 and 17.6 g per 100 g d.w., respectively, while the amount in the control was 26.2 g per 100 g d.w.[32] In *P. eryngii*, a corncob substrate was beneficial to high contents of trehalose, soluble carbohydrates and polysaccharides, while sawdust produced the lowest content, being beneficial for protein.[45] In *P. sajor-caju*, the use of increasing amounts of detoxified mahua cake up to 20% in the growth substrate, led to a decrease in total sugars/polyol from 7.54 to 5.39 g per 100 g d.w.[35]

Minerals

Mushrooms are potential dietary sources of the minerals that are necessary for metabolic reactions, the transmission of nerve impulses, bone formation, regulation of water and for salt balance. In fact, all edible mushrooms can accumulate minerals in their fruit bodies.[49] The ash content of the fruit bodies of *Pleurotus* spp. ranges between 4.60 and 10.55 g per 100 g d.w.[6–8,13–21,32–36] (Table 1). The mineral levels are largely affected by the growth substrates since substrates high in a particular mineral produce mushrooms relatively high in the content of that mineral.[49] From the mineral analysis reports of different studies, it was revealed that the major minerals in *Pleurotus* spp. are P (496–1647.6 mg per 100 g d.w.), K (271–4054.3 mg per 100 g d.w.), Na (13–310 mg per 100 g d.w.), Ca (1–330 mg per 100 g d.w.), Mg (137–203.4 mg per 100 g d.w.), Mn (1.1–1.6 mg per 100 g d.w.), Fe (5.4–15.62 mg per 100 g d.w.), Zn (8.3–13.7 mg per 100 g d.w.) and Cu (0.84–2.5 mg per 100 g d.w.).[6,15,19,31,33,50,51] The cultivated species of *Pleurotus* were found to contain only low levels of the undesirable elements, such as Cd, Pb and Hg.[49]

The distinctive presence in *Pleurotus* spp. of a low Na content and high K content (second major mineral after P) is beneficial from a nutritional point of view. In fact, to reduce blood pressure, the risk of cardiovascular disease, stroke and coronary heart disease in adults (≥16 years of age), the recommended upper limit for Na dietary intake is <2000 mg per day, while a dietary intake of 3510 mg per day for K is suggested. These latter amounts adjusted based on the different energy requirements have also been recommended for children (2–15 years of age) to control blood pressure.[52,53] Interestingly, in both wild *Pleurotus* spp. and *Pleurotus* spp. grown on different waste substrates, the concentration range of Na in 100 g of dried fruits is notably lower than the recommended upper limit for Na daily dietary intake, while the concentration range of K in general meets the recommended K daily dietary intake (Table 4).

Comparing the Fe and Zn contents of *Pleurotus* spp. with their recommended daily dietary intake,[54] it is outstanding to note that the *Pleurotus* species are able to provide more than adequate quantities of these minerals (Table 4). However, the amount provided by foods is not always enough to meet nutritional requirements if the bioavailability is low. This latter depends on dietary sources due to the presence of inhibitors and promoters of absorption.[55,56] A diet containing at least small amounts of meat and fish is associated with good levels and bioavailability of Fe and Zn, while these minerals are found in low amounts and have low bioavailability in cereal- and tuber-based diets. Hence, a deficiency in these minerals is common in developing countries, where the diet is limited with respect to their content and bioavailability. Additionally, bioavailability is diminished in phytate-containing foods. Other reasons for Fe anaemia in many tropical countries are infestations with hookworms, which lead to intestinal blood losses. Patients who have gastric diseases and celiac subjects may also develop Fe deficiency because of impaired Fe absorption. The populations most at risk for Fe and Zn deficiency are infants, children, adolescents and women of childbearing age, especially pregnant women.[54] Interestingly, a previous study carried out with a mice animal model indicated that the bioavailability of Fe present in the fruit bodies of *P. sajor-caju* was high.[57] However, human studies are necessary to define the possible role of mushrooms in the prevention of Fe and Zn deficiencies.

P. ostreatus has also shown a great potential to improve the dietary intake of Se when grown on Se-enriched substrates. Se deficiency is endemic in regions where this mineral is poorly available from soil for staple crops, covering especially localities from northeast to southwest China and Siberia, where it the primary factor for the occurrence of Keshan and Kaschin-Beck diseases. Fluctuations in the Se status of many communities in northern Europe has also been observed, which reflect the intrinsically low Se content of glacial soil in this region. Non-endemic Se depletion is also common in subjects maintained on parenteral or enteral feeding for long periods. Additionally, the possibility that increased intakes of Se might protect against the development of cancer in humans has generated great interest, although the question of "whether Se protects against cancer" remains wide open.[54] *P. ostreatus* was able to absorb and accumulate Se from selenite added to coffee husks used as a growth substrate in the range

Table 4 Recommended dietary intakes of some minerals for different age groups, pregnancy and lactation (mg d^{-1}) and mineral content (mg per 100 g d.w.) of the fruit bodies of *Pleurotus* spp. grown on different waste substrates or collected from the market

Groups	Dietary reference intakes for minerals					Ref.
	K[a]	Na[a]	Se	Fe[b]	Zn[c]	
Infant (birth to 12 months)	N.R.[d]	N.R.[d]	0.006–0.010	9.3	2.8–4.1	52–54
Child (1 to 9 years)	>3510	<2000	0.017–0.021	5.8–8.9	4.1–5.6	52–54
Adolescent, adult, elderly	>3510	<2000	0.025–0.033	13.7–32.7	7.0–8.6	52–54
Pregnancy and lactation	>3510	<2000	0.028–0.042	15	5.5–10	52–54
Pleurotus species and growth substrate[e]	Mineral content					
P. ostreatus						
Unspecified	2682.3 ± 53.0	136 ± 0.4.6	N.R.[d]	N.R.[d]	N.R.[d]	6
Unspecified	3443.8 ± 109.0	25.2 ± 5.7	N.R.[d]	N.R.[d]	N.R.[d]	6
Unspecified	3730 ± 1	13 ± 1	0.015 ± 0.01	5.4 ± 0.1	8.3 ± 0.1	50
Spent beer grain and wheat bran	2171.4 ± 0.1	21.9 ± 0.1	N.R.[d]	7.1 ± 0.1	13.7 ± 0.1	33
Soybean straw	2320 ± 9	310 ± 4	N.R.[d]	14.35 ± 0.16	N.R.[d]	51
Paddy straw	2260 ± 9	290 ± 4	N.R.[d]	14.94 ± 0.16	N.R.[d]	51
Soybean straw and paddy straw	2100 ± 9	295 ± 4	N.R.[d]	15.62 ± 0.16	N.R.[d]	51
Soybean straw and wheat straw	2000 ± 9	260 ± 4	N.R.[d]	14.20 ± 0.16	N.R.[d]	51
Wheat straw and paddy straw	1900 ± 9	275 ± 4	N.R.[d]	13.13 ± 0.16	N.R.[d]	51
Wheat straw	2100 ± 9	305 ± 4	N.R.[d]	13.88 ± 0.16	N.R.[d]	51
Wheat straw	352 ± 4	104 ± 6	N.R.[d]	N.R.[d]	N.R.[d]	15
Almond and walnut shells	371 ± 9	89 ± 2	N.R.[d]	N.R.[d]	N.R.[d]	15
Corn cobs	325 ± 18	80 ± 2	N.R.[d]	N.R.[d]	N.R.[d]	15
Grape marc plus cotton gin trash	374 ± 18	91 ± 3	N.R.[d]	N.R.[d]	N.R.[d]	15
Olive mill by-products	510 ± 27	100 ± 10	N.R.[d]	N.R.[d]	N.R.[d]	15
Extracted olive-press cake	277 ± 7	76 ± 3	N.R.[d]	N.R.[d]	N.R.[d]	15
Date palm tree leaves	342 ± 5	96 ± 2	N.R.[d]	N.R.[d]	N.R.[d]	15
Pine needles	271 ± 1	81 ± 3	N.R.[d]	N.R.[d]	N.R.[d]	15
Coffee husk + 102 mg kg^{-1} Se			85.8 ± 0.1			58
P. eryngii						
Unspecified	3095.0 ± 40	50.4 ± 1.1	N.R.[d]	N.R.[d]	N.R.[d]	6
Unspecified	4054.3 ± 244.2	76.6 ± 2.8	N.R.[d]	N.R.[d]	N.R.[d]	6
P. sajor-caju						
Chopped rice straw	3260 ± 1	N.R.[a]	N.R.[d]	12.4 ± 0.1	12.9 ± 0.1	31
Straw	2400 ± 1	238 ± 1	N.R.[d]	11.5 ± 0.1	N.R.[d]	19
Cotton waste	2207 ± 1	158 ± 1	N.R.[d]	5.9 ± 0.1	N.R.[d]	19
Cotton waste and straw	2322 ± 1	172 ± 1	N.R.[d]	5.0 ± 0.1	N.R.[d]	19
Cotton waste and tea leaves	2130 ± 1	256 ± 1	N.R.[d]	5.6 ± 0.1	N.R.[d]	19
P. pulmonarius						
Unspecified	2818.9 ± 36.0	103.4 ± 2.1	N.R.[d]	N.R.[d]	N.R.[d]	6

[a] The recommended level of intake should be adjusted downward based on the energy requirements of children relative to those of adults. [b] Values refer to a moderate dietary Fe bioavailability (10%). [c] Values refer to a moderate dietary Zn bioavailability (30%). [d] N.R.: not reported. [e] Unspecified substrate means that data refer to a wild or cultivated mushrooms collected from the market.

of 3.2–100 mg of Se per kg. The lowest concentration of Se in the substrate (3.2 mg of Se per kg) resulted in mushrooms with 5.76 mg of Se per 100 g d.w., while the highest concentration used (100 mg of Se mg per kg) resulted in mushrooms with 85.8 mg of Se per 100 g d.w. Interestingly, for the enriched mushrooms, the Se bioavailability in rats was higher than that of sodium selenite.[58] However, human studies on the bioavailability of mineral microelements in mushrooms are lacking and hence preclude drawing general conclusions.

Vitamins and pro-vitamins

Wild and cultivated mushrooms from *Pleurotus* genus are good sources of some B group vitamins (Table 5).[14,19,31,50,59,60] In a few studies, the level of the B group vitamins was found to be affected by the ingredients used in the growth substrate.[14,19] However, information on the effect of the growth substrate on the vitamin content in *Pleurotus* spp. is lacking and hence general rules cannot be drawn. The levels of B group vitamins in *Pleurotus* spp. were found to vary between 0.02 and 1.96 mg per 100 g for B_1, 0.15 and 6.66 mg per 100 g for B_2 and 0.59 and 65 mg per 100 g for B_3.[14,19,31] Thiamine deficiency has been observed in developing country populations as well as in Japanese elderly and people with chronic alcoholism.[54] African and Asian children commonly demonstrate clinical signs of riboflavin deficiency during periods of the year when gastrointestinal infections are prevalent. However, the major cause of hyporiboflavinosis is an inadequate dietary intake as a result of a limited food supply. Niacin deficiency, which can causes pellagra disease, is also endemic in poorer areas of Africa, China and India. Interestingly, the content of niacin in mushrooms is higher than those generally found in vegetables. Information on vitamins B_5 and B_6 in *Pleurotus* spp. is limited. For vitamin

Table 5 Recommended dietary intakes of vitamins for different age groups, pregnancy and lactation (mg d^{-1}) and vitamin content (mg per 100 g d.w.) of the fruit bodies of *Pleurotus* spp. grown on different waste substrates or collected from the market

Groups	Recommended nutrient intakes							Ref.
	B_1	B_2	B_3[a]	B_5	B_6	B_9[b]	D_2[c]	
Infant (birth to 12 months)	0.2–0.3	0.3–0.4	2–4	1.7–1.8	0.1–0.3	0.08	0.005	54
Child (1 year to 9 years)	0.5–0.9	0.5–0.9	6–12	2–4	0.5–1.0	0.15–0.3	0.005	54
Adolescent, adult, elderly	1.1–1.2	1.0–1.3	14–16	5	1.2–1.7	0.4	0.005–0.015	54
Pregnancy and lactation	1.4–1.5	1.4–1.6	17–18	6.0–7.0	1.9–2.0	0.5–0.6	0.005	54
Pleurotus species and growth substrate[d]	Vitamin content							
P. ostreatus								
Unspecified	0.9 ± 0.1	2.5 ± 0.1	65 ± 1	N.R.[e]	N.R.[e]	0.64 ± 0.01	0.0003 ± 0.0001	50
Unspecified	0.30 ± 0.01	1.62 ± 0.08	9.98 ± 0.57	N.R.[e]	0.0701 ± 0.0012	N.R.[e]	N.R.[e]	59
Unspecified	N.R.[e]	N.R.[e]	N.R.[e]	N.R.[e]	N.R.[e]	N.R.[e]	0.083 ± 0.06	63
Millet stalk	0.14 ± 0.00	0.15 ± 0.00	0.93 ± 0.02	N.R.[e]	0.23 ± 0.01	N.R.[e]	N.R.[e]	14
Wheat stalk	0.12 ± 0.01	0.19 ± 0.02	0.67 ± 0.00	N.R.[e]	0.23 ± 0.01	N.R.[e]	N.R.[e]	14
Cotton stalk	0.25 ± 0.00	0.21 ± 0.03	1.43 ± 0.00	N.R.[e]	0.21 ± 0.02	N.R.[e]	N.R.[e]	14
Soybean stalk	0.07 ± 0.02	0.20 ± 0.00	0.59 ± 0.00	N.R.[e]	0.21 ± 0.00	N.R.[e]	N.R.[e]	14
Wheat straw	1.92 ± 0.01	3.3 ± 0.1	35.98 ± 0.01	N.R.[e]	N.R.[e]	N.R.[e]	N.R.[e]	60
Wheat straw	1.96 ± 0.01	3.7 ± 0.1	36.56 ± 0.01	N.R.[e]	N.R.[e]	N.R.[e]	N.R.[e]	60
P. sajor-caju								
Unspecified	1.75 ± 0.23	6.66 ± 1.22	60.0 ± 4.7	21.1 ± 3.1	N.R.[e]	1.278 ± 0.130	N.R.[e]	31
Straw	0.02 ± 0.01	1.36 ± 0.01	18.2 ± 0.1	N.R.[e]	N.R.[e]	N.R.[d]	N.R.[e]	19
Cotton waste	0.02 ± 0.01	1.32 ± 0.01	20.7 ± 0.1	N.R.[e]	N.R.[e]	N.R.[d]	N.R.[e]	19
Cotton waste and straw	0.03 ± 0.01	1.33 ± 0.01	21.3 ± 0.1	N.R.[e]	N.R.[e]	N.R.[d]	N.R.[e]	19
Cotton waste and tea leaves	0.06 ± 0.01	1.21 ± 0.01	20.6 ± 0.1	N.R.[e]	N.R.[e]	N.R.[d]	N.R.[e]	19

[a] Expressed as mg niacin equivalents (NE) per d. [b] Expressed as mg dietary folate equivalents (DFE) per d. [c] The vitamin D content reported for the different species of *Pleurotus* only refers to vitamin D_2. [d] Unspecified substrate means that data refer to wild or cultivated mushrooms collected from the market. [e] N.R.: not reported.

B_5, a level of 21.1 mg per 100 g d.w. was found in one strain of *P. sajor-caju.*[31] For vitamin B_6, a range of values between 0.0701 and 0.23 mg per 100 g was found in *P. ostreatus.*[14,59] A nutritional deficiency of vitamins B_5 or B_6 alone is uncommon because it usually occurs in association with a deficit in other B complex vitamins and other nutrients.[54]

Regarding the vitamin B_9, mushrooms contain moderately high amounts of this, and their contents are of the same magnitude as those generally found in vegetables like spinach. In addition, the bioavailability of mushroom folates appears to be as good as that for folic acid, unlike the bioavailability of folates from some vegetables, such as peas and spinach.[61] A high content of folates was found in one strain of *P. ostreatus* (0.64 mg per 100 g d.w.)[50] and in one strain of *P. sajor-caju* (1.278 mg per 100 g d.w.).[31] A deficiency of folate is common in people consuming a limited diet and in pregnant women, because pregnancy significantly increases the folate requirement, especially during periods of rapid foetal growth. During lactation, losses of folate in milk also increase the folate requirement.[54]

The content of vitamin B_{12} was only reported for *P. ostreatus* and found to be 0.6 μg per 100 g d.w.[50] However, among 38 common edible fungi analysed for vitamin B_{12} content, only 9 were found to contain this vitamin, where one of the best producers was *P. ostreatus*.[62] Hence, this mushroom could be a good B_{12} source for vegans, because otherwise it would normally enter the human food chain through incorporation in food of an animal origin. In mushrooms, the vitamin probably derives from surface microorganisms that can synthesize it.[50]

In addition to vitamins from the B group, the genus *Pleurotus* contains elevated amounts of the vitamin D_2 (ergocalciferol) precursor, *i.e.* ergosterol – a component of the fungal cell membrane. The ergosterol content in *P. ostreatus* varies from 290 to 754 mg per 100 g d.w.[63–65] The natural level of vitamin D_2 in *Pleurotus* spp. is generally low and highly variable: both undetectable levels,[64] and low levels, such as 0.3 μg per 100 g d.w.[50] and values in the range 0.083–0.156 mg per 100 g d.w., have been reported.[63] However, vitamin D_2 in mushrooms is converted from ergosterol through UV irradiation during growth, after harvest and after drying too. After the exposure of *P. ostreatus* powder with no detectable amount of vitamin D_2 to 2800–2900 mJ cm^{-2} UVB at 60–66 °C for 10 min, 11 mg per 100 g d.w. of vitamin D_2 was obtained.[64] Accordingly, treatment with 411 mJ cm^{-2} UVB at 20 °C for 10 min led to a vitamin D_2 formation of 4.07 mg per 100 g d.w.[65] The fact that the enrichment in vitamin D_2 may be performed after drying, greatly facilitates the potential use of UV technology in the processing of mushrooms in order to improve their nutritional value. It is estimated that about one billion people in the world have a vitamin D deficiency: infants, adolescents, elderly, pregnant and lactating women constitute the populations most at risk.[54] Moreover, mushrooms are a natural source of vitamin D for some consumer groups, including vegetarians and vegans, and people intolerant to lactose, since most of the products fortified with vitamin D include dairy products.

Another vitamin is vitamin C, where its content in *Pleurotus* spp. was reported to vary from 9.10 (*P. ostreatus*) to 111 mg per 100 g d.w. (*P. sajor-caju*).[59,60] The vitamin C recommended dietary intake varies from 25 (for infants) to 70 (for lactating women) mg per day and a deficiency in vitamin C is associated with malnutrition.[54] With regard to vitamin E precursors, the α-, β-, γ- and δ-tocopherols were found in the *Pleurotus* genus. *P. ostreatus* showed significant amounts of total tocopherols (while β-tocopherol was lacking) in the range 0.279–2.87 mg per 100 g d.w.[32,59] A total tocopherol content of 0.086 mg per 100 g d.w. (while δ-tocopherol was lacking) was found in one strain of *P. eryngii.*[47] In general, these amounts are low with respect to the vitamin E recommended intake, which is in the range 2.7–10 mg d^{-1}.[54] *P. ostreatus* was reported to contain 1.075 mg per 100 g d.w. of the vitamin A precursor, *i.e.* β-carotene, and 0.638 mg of lycopene per 100 g d.w.[59] However, in another study, the presence of carotenoids in *Pleurotus* spp. was denied.[66]

Ergothioneine, lovastatin and γ-aminobutiryc acid

Ergothioneine (EGT), lovastatin (also known as monacolin K, mevinolin or mevacor) and γ-aminobutyric acid (GABA) are secondary metabolites from fungal growth, occurring both in the mycelium and in the fruiting body, that are thought to be beneficial for human health.[67]

EGT is not synthesized by higher organisms. However, in humans, EGT has been shown to accumulate in various cells and tissues at high concentrations (100 μM to 2 mM), most abundantly in erythrocytes, bone marrow, liver, kidney, seminal fluid and the lens and cornea of the eyes. EGT is not currently considered an essential dietary component and there are no reports of symptoms due to its deficiency. A wide body of evidence suggests that EGT may function as a physiological antioxidant. The biological role of EGT is under investigation for its positive impact on the inflammatory process.[68] Among various fungi, the *Pleurotus* genus contains a considerably high amount of EGT, which is higher in the fruiting body than in the mycelium. The level of EGT in the fruiting body was found to be in the ranges of 94–259 mg per 100 g d.w. for *P. ostreatus* and 62.4–84.0 mg per 100 g d.w. for *P. eryngii.*[67,69,70]

Lovastatin is one of the natural statins (3-hydroxy-3-methyl-glutaryl coenzyme A reductase inhibitors), which inhibit the rate-limiting enzyme in the production of cholesterol and have been proven to reduce the risk of coronary heart disease. Contrary to EGT, the lovastatin content in the fruiting body of fungi is lower than that of the mycelium. In the fruiting body, its level was found to be in the ranges of 16.5–60.6 mg per 100 g d.w. for *P. ostreatus* and 12.0–15.2 mg per 100 g d.w. for *P. eryngii*.[67]

Several *in vivo* experiments have demonstrated the hypotensive effect of GABA. In a screening study with various *Pleurotus* strains, the level of GABA was found to be in the ranges of 14.3–280.8 mg per 100 g d.w. in *P. ostreatus*, 53.3–54.6 mg per 100 g d.w in *P. eryngii* and 165.4 mg per 100 g d.w. in *P. pulmonarius*.[6]

Phenolic compounds

The high total phenolic content of *Pleurotus* spp. is likely responsible for its ability to scavenge free radicals and other reactive oxygen species that are continuously being produced *in vivo*, as well as its ability to chelate Fe^{++} ions, which catalyze oxidative processes. These properties result in the prevention of cell death and tissue damage. Indeed, phenolic extracts from *Pleurotus* spp. possess antioxidant, anti-inflammatory and antimicrobial activities.[5,71] However, knowledge of the phenolic pattern of *Pleurotus* spp. is still lacking. HPLC-MS analysis of the methanolic extract for 16 strains of *P. ostreatus* revealed the presence of *p*-hydroxy-benzoic acid (n.d.–424.7 μg per 100 g d.w.), *p*-hydroxy-phenylacetic acid (10.3–120.9 μg per 100 g d.w.), 3-4-dihydroxy-phenylacetic acid (n.d.–35.4 μg per 100 g d.w.), protocatechuic acid (n.d.–32.3 μg per 100 g d.w.), syringic acid (n.d.–14.4 μg per 100 g d.w.), vanillic acid (n.d.–12.9 μg per 100 g d.w.), caffeic acid (0.5–5.4 μg per 100 g d.w.), cinnamic acid (n.d.–110 μg per 100 g d.w.), ferulic acid (n.d.–2.2 μg per 100 g d.w), vanillin (n.d.–30.2 μg per 100 g d.w.), tyrosol (n.d.–8.6 μg per 100 g d.w.) and resveratrol (5.4–95.8 μg per 100 g d.w.).[24] Phenolic acids can also be released from the polysaccharides of the cell wall after alkaline hydrolysis. Particularly in a strain of *P. ostreatus*, bound coumaric and ferulic acids were found at the levels of 556 and 90 μg per 100 g d.w., respectively.[72] The presence of phenolic acids in the *Pleurotus* genus was confirmed in other studies with *P. ostreatus*, *P. eryngii* and *P. sajor-caju*.[73–75]

Some authors have identified the presence of flavonoids in *Pleurotus* spp. However, this identification was not confirmed by MS studies and it has been considered misleading because edible mushrooms do not have the main enzymes involved in the flavonoids metabolic pathway. Additionally, mushrooms have been found unable to accumulate flavonoids present in the growth substrates.[11]

Human studies on *Pleurotus* health properties

Immunomodulatory properties and anti-allergic effects

The immunomodulatory activity of insoluble β-glucan of *Pleurotus* spp. (pleuran) is well documented and recognized (Table 6). The mechanism of its action in the organism is mediated through several receptors, especially the dectin-1 receptors, toll-like receptors, complement receptor 3, scavenger receptor and lactosylceramid. After the binding of β-glucan to its receptors, it stimulates the production of many cytokines or other mechanisms of immune and non-immune reactions.[76]

Since excessive and exhausting physical loads depress the immune system, the immunomodulatory activity of *Pleurotus* β-glucan has been studied in athletes. A *P. ostreatus* insoluble β-glucan supplement (Imunoglukan 1) was orally administered

Table 6 Health effects of the intake of *Pleurotus* spp. whole powder or pleuran (purified insoluble β-glucans) as documented by *in vivo* human studies with healthy or unhealthy subjects

Supplement[a]	Daily dose and trial period	Subjects	Main conclusion[b]	Ref.
P. ostreatus I-β-glucan	100 mg for 2 months	Athletes (n = 20)	Modulation of exercise-induced changes in natural killer cell activity	77
P. ostreatus I-β-glucan	100 mg in combination with 100 mg of vitamin C for 3 months	Athletes (n = 50)	Decrease in the incidence of upper respiratory tract infections symptoms and increase in the activity and number of natural killer cells	78
P. ostreatus I-β-glucan	10 mg in combination with 10 mg of vitamin C per 5 kg body weight for 6 months	Children with recurrent respiratory tract infections (n = 175)	Improvement of the humoral and cellular immunity and prevention of infectious respiratory diseases	76
P. eryngii powder	5–10 g for 2 days	Healthy human subjects (n = 12)	Enhancement of the innate and acquired immune responses	80
P. ostreatus powder	10 g for 6 weeks	Patients with dyslipidemia (n = 57)	Decrease in blood TG and TC levels	81
P. ostreatus powder	30 g for 21 days	Healthy human subjects (n = 20)	Decrease in TG, ox-LDL levels and TC levels	82
P. sajor-caju	Not reported dose for 3 months	Type 2 diabetic patients (n = 120)	Reduced fasting blood glucose, glycosylated haemoglobin as well as blood cholesterol levels	85
P. ostreatus powder	3 g for 3 months	Type 2 diabetic patients (n = 27)	Decrease in fasting plasma glucose level and reduction in the level of glycosylated haemoglobin	86
P. ostreatus powder	50 mg per kg of body weight for 1 month	Healthy human subjects (n = 22) type 2 diabetic patients (n = 28)	Decrease in fasting plasma glucose level and increased the serum insulin levels in diabetic patients	84

[a] I-β-glucan: purified insoluble β-glucan fraction (pleuran). [b] TG: triglycerides; TC: total cholesterol; ox-LDL: oxidised low density lipoproteins.

to athletes to investigate the effects on cellular immune response and respiratory tract infections. In a double-blind pilot study, 20 elite athletes were randomized into insoluble β-glucan (n = 9) or placebo (n = 11) groups. These groups consumed 100 mg of β-glucan (Imunoglukan®) or placebo supplements, respectively, once a day for 2 months. The study showed that insoluble β-glucan supplementation from *P. ostreatus* may play a role in modulating exercise-induced changes in natural killer cell activity in intensively training athletes.[77] In a second study, 50 healthy male (n = 26) and female (n = 24) top-level athletes were enrolled and randomized into a pleuran or placebo group. The experimental pleuran group consisted of athletes (n = 25) who were required to take 100 mg of β-glucan (Imunoglukan®) and 100 mg of vitamin C or a placebo (100 mg of vitamin C only) in the morning on an empty stomach for 3 months. The study confirmed that pleuran reduced the incidence of upper respiratory tract infections symptoms and increased the activity and number of natural killer cells.[78]

Additionally, the immunomodulatory properties of pleuran were studied in children with respiratory diseases. In this study, 175 children from 2 to 5 years of age with recurrent respiratory tract infections were enrolled and randomized into an active group, treated with 1 mL per 5 kg of Imunoglukan P4H® syrup (10 mg of pleuran and 10 mg of vitamin C in 1 mL of syrup) and a placebo group treated with vitamin C only, for 6 months. The results showed that in the active group, the humoral and cellular immunity improved and prevented infectious respiratory diseases.[76] Patients were also monitored for parameters for allergy against a standardized panel of inhalant and food allergens, and it was revealed that pleuran showed a potential suppressive effect on the markers of allergic inflammation in peripheral blood, especially in atopic subjects. This effect led the study researchers to conclude that pleuran could also be applied as a complementary adjuvant therapy in allergic patients.[79]

Besides pleuran, *P. eryngii* superfine powder administered at a daily dosage of 5 to 15 g to 12 healthy volunteers for 2 days enhanced their innate and acquired immune responses.[80]

Hypolipidemic effects

The hypolipidemic effects of *Pleurotus* spp. have been investigated but a precise identification of the molecules involved is still lacking. Statins, such as lovastatin, which has been found in *Pleurotus* spp.[67] and acts as an inhibitor of 3-hydroxy-3-methylglutaryl coenzyme A reductase, are likely to be involved.

P. ostreatus showed a significant hypocholesterolemic effect in a clinical study with 57 patients with dyslipidemia (32 women and 25 men with an average age of 43 years old). Subjects were fed lyophilized powder of *P. ostreatus* in an average daily dose of 10 g. After 6 weeks of mushroom feeding, the blood triglycerides (TG) and total cholesterol (TC) levels of the individuals decreased significantly.[81] Likewise, in a study with 20 healthy human subjects (9 male and 11 female aged 20–34 years old), treatment with 30 g of dried *P. ostreatus* or a tomato soup as a placebo on a daily basis for 21 days decreased the TG concentrations and oxidized low density lipoprotein (ox-LDL) levels significantly, and showed a significant tendency towards lowering the TC values in comparison with the control group.[82]

Hypoglycaemic effects

Pleurotus spp. intake has been proven to have hypoglycaemic effects and to be able to decrease the levels of the marker of hyperglycaemia damage (glycosylated haemoglobin). In previous studies, the hypoglycemic effect from either the fruiting body or mycelia of some edible/medicinal fungi have been investigated *in vitro* and in animal models. The water extract and especially, the water-soluble polysaccharide fraction have been found to have hypoglycaemic properties. However, the hypoglycaemic effects can be observed in the whole dehydrated powder without any purification step but the molecules involved have not been precisely identified.[83] The mechanism of hypoglycaemic activity of *Pleurotus* spp. is possibly through increasing the glucokinase activity and promoting insulin secretion, thereby increasing the utilization of glucose by peripheral tissues, inhibiting glycogen synthase kinase and promoting glycogen synthesis.[84]

A study was conducted with 120 type 2 diabetic patients (randomly divided into three groups, with 40 patients in a mushroom-fed group, and the remaining groups serving as controls). It was found that patients in the group fed the *P. sajor-caju* mushroom for 3 months showed significantly reduced fasting blood glucose levels and glycosylated haemoglobin as well as blood cholesterol. However, the exact amount of *P. sajor-caju* supplemented daily was not specified.[85]

P. ostreatus powder was supplemented to 27 hypertensive males with type 2 diabetes mellitus (age range: 32 to 68 years old) at a daily dose of 3 g for 3 months. Both systolic and diastolic blood pressure decreased significantly. It was also observed that *P. ostreatus* decreased fasting plasma glucose levels and reduced the level of glycosylated haemoglobin.[86] The hypoglycaemic effect of freeze-dried and powdered *P. ostreatus* was also investigated with 22 healthy human volunteers and 28 type 2 diabetic patients on diet control at a dose of 50 mg per kg per body weight, followed by a glucose load. The *P. ostreatus* powder showed a significant reduction in fasting and the postprandial serum glucose levels of healthy volunteers and reduced the postprandial serum glucose levels and increased the serum insulin levels of type 2 diabetic patients.[84] Additionally, the inclusion of 8% of *P. ostreatus* powder in biscuits was found to decrease postprandial glycaemic response in 11 healthy participants (four males and seven females with no histories of carbohydrate malabsorption).[18]

Anticancer effects

Clear clinical evidence of the anticancer activities of *Pleurotus* mushrooms is still not available, even though different types of extracts from *Pleurotus* mushrooms have been reported as potential anticancer agents in several tumour cell lines, most likely acting through distinct mechanisms.[5]

Sensory attributes and perception

Appearance. Mushrooms exist in nature under different dimensions and shapes but certainly, colour is the appearance indicator that has the greatest influence on consumer choice. Colour is also one of the descriptors most subjected to fluctuation during fresh mushrooms storage, preliminary processing and then the storage of finished products because of non-enzymatic or enzymatic darkening. In fact, although the visual appearance and cell fluid leakage of unblanched and blanched frozen *P. ostreatus* were found to be very stable over a 12-months storage period, the colour quality, as evaluated both instrumentally and by a sensory panel, decreased. In particular, storage for 12 months led to a progressive decrease in the intensity of white and cream colours and to an increase in ash and grey colours. However, white colour saturation increased in mushrooms pre-treated with aqueous solutions of anti-darkening substances, while at the same time decreasing the grey colour saturation.[87] Growth substrates or supplements can be also used to develop different colour intensities in mushrooms. In this context, the use of aromatic plant wastes was found to increase the intensity of the brown colour in *P. ostreatus*,[88] whereas the golden colour of *P. sajor-caju* was reported to improve by the addition of cracked corn to corn stover substrates.[89]

Taste. Mushrooms contain free amino acids that have been classified into four groups: sweet (alanine, glycine, threonine and serine); bitter (arginine, histidine, isoleucine, leucine, methionine, phenylalanine and valine), tasteless (lysine and tyrosine) and umami (aspartic acid, Asp and Glu). However, the peculiar taste of mushrooms is umami.[90] Besides Glu and Asp, the 5′-nucleotides were also identified as "umami ingredients".[46] The umami taste has been widely investigated in recent years.[40] Monosodium L-aspartate has low intensity values for the umami taste of less than 10% of monosodium glutamate (MSG), but both the Asp and Glu are classified as umami or MSG-like amino acids and the sum of these two amino acids is frequently adopted to describe the umami taste of mushrooms. 5′-Nucleotides (5′-AMP, 5′-adenosine monophosphate; 5′-CMP, 5′-cytosine monophosphate; 5′-GMP, 5′-guanosine monophosphate 5′-UMP, 5′-uridine monophosphate; 5′-XMP, 5′-xanthosine monophosphate) cannot activate umami-taste receptors on their own, but they can intensify the umami sensation caused by Glu by a factor up to eight times.[40] The presence of these "umami ingredients" and their quantities in mushrooms are influenced by many factors, including the species type, maturity stage, the part of the mushroom, substrate and storage time.[44] Umami ingredients are highly palatable since they are effective flavour enhancers of savoury foods but they remain ineffective on sweet, fruity or bland foods.[40,90] *Pleurotus* spp. are reported to be among the richest mushrooms in umami-tasting amino acids, with *P. ostreatus* showing the highest values of equivalent umami concentration (EUC) and umami taste among 17 edible mushrooms, as evaluated by a trained sensory panel.[46] Similarly, *P. eryngii* was found to be in the middle range of umami-taste perception among different edible fungi solutions, as evaluated by combined e-tongue analysis and a trained sensory panel.[91] However, only a limited number of species have been analysed.

Odour and flavour. Among volatile components, a series of eight carbon atom compounds (C8), such as 1-octen-3ol, 3-octanol, 1-octanol, 1-octen-3-one and 3-octanone, have been reported as the major contributors to the characteristic mushroom flavour.[92] These compounds could represent up to 90%

(w/w) of the volatile fraction from fresh bodies.[93] The content of C8 compounds varies among large fruiting bodies, small fruit bodies and the base. In this context the total amount of these compounds in *P. eryngii* was in the order: large fruiting bodies > small fruiting bodies > base.[95]

A detailed knowledge on *Pleurotus* spp.'s aromatic profile is lacking, because only a few strains have been characterized. The aroma components of different *Pleurotus* spp. were studied by combining gas chromatography and electronic nose and sensory analysis involving a trained panel of assessors.[95] This study confirmed that the main aroma constituents of *Pleurotus* spp. were C8 compounds, mainly 1-octen-3-ol, 3-octanol and 3-ocatonone. The highest amount of 1-octen-3-ol was measured in *P. ostreatus*, with an optical purity of (*R*)-(−)-1-octen-3-ol that accounted for 97.3%.[96] Previous research indicated that (*R*)-(−)-1-octen-3-ol has a mushroom-like odour, whereas (*S*)-(+)-1-octen-3-ol has a mouldy, grassy note.[97] In one strain of *P. eryngii*, the major volatile compound was found to be benzaldehyde, which confers a highly appreciated almond flavour.[94] In another strain of *P. eryngii*, methional (potato-like odour), 1-octen 3-ol (mushroom odour) and nonanal (described as sweet, citrus and green) were found to be the main aroma components.[92] Methional (potato-like odour) and 1-octen 3-ol (mushroom odour) were also found to be the main aroma component in *P. sajor-caju*.[98]

Texture. Although texture is an important parameter to establish mushroom quality, as well as fruit and vegetable quality, a very limited number of studies are available on *Pleurotus* spp. and most of those available adopted instrumental rather than sensory approaches to evaluate texture properties. The most common texture properties used to describe *Pleurotus* spp. are hardness, springiness, firmness, fibrousness and rubbery. Reduction of the springiness and firmness and an increase in the soft, spongy and fibrous texture in mushrooms are reported to be critical for consumer acceptance.[99] The softening of mushrooms or loss of firmness is related to protein and polysaccharide degradation, hyphae shrinkage, central vacuole disruption and expansion of the intercellular space at the pilei surface, which are phenomena occurring during post-harvest storage.[100] However, the texture is also influenced by the substrate used. The addition of cracked corn (50%) to corn substrates was found to modify the texture of *P. sajor-caju* by decreasing the perceived level of toughness, rubberiness and fibrousness.[89] In *P. sajor-caju*, the addition of corn gluten to both wheat straw and soybean hull substrates gave lower rubbery scores, while the use of sugar beet pulp as a substrate gave low rubbery scores both in the presence and in the absence of corn gluten.[101]

Food applications

Application of *Pleurotus* spp. as a fortifying agent

It is well known that cereal-based products are consumed throughout continents and civilizations, representing one of the most consumed foodstuffs; hence, studies have been carried out to improve their nutritive value and functional effect by the substitution of some ingredients with *Pleurotus* spp. powder or β-glucan-rich fractions (Table 7).

In bread, *Pleurotus* powder was added to replace 5–25% of the flour, with an aim to increase the protein and dietary fibre contents.[102] In a study by Ng *et al.*,[18] it was demonstrated that the addition of 8% of *Pleurotus* powder to biscuits increased dietary fibre content from 3.37% to 8.62% and decreased the *in vivo* glycaemic index. This effect was attributed to the mush-

Table 7 Nutritional effects of the use of dried *Pleurotus* spp. or β-glucan-rich fractions obtained from *Pleurotus* spp. as ingredients in model foods

Food	Ingredient[a]	Main results	Ref.
Bread	*P. pulmonarius* powder 5–25% of flour	Increase in protein and dietary fibre contents from 7.96 to 14.21 and from 0.51 to 2.48 g per 100 g f.w., respectively	102
Biscuits	*P. sajor-caju* powder 4–12% of flour	Increase in dietary fibre contents from 3.37 to 8.62 g per 100 g f.w. Decrease in the glycaemic index; upon 8% addition: glycaemic index from 57.2 to 49	18
Pasta	*P. eryngii* I-β-glucan fraction 2–6% of flour	Fortification with I-β-glucans at final levels of 0.79–2.4 g per 100 g of flour	17
Tapioca cracker	*P. sajor-caju* powder 5–20%	Increase in protein content from 0.47 to 3.88 g per 100 g f.w.	107
Instant drink	*P. eryngii* broth	Fortification with ergothioneine at final levels of 6.22–11.57 mg per g d.w. and γ-aminobutyric acid at final levels of 4.19–8.30 mg per g d.w.	108
Chicken patty	*P. sajor-caju* powder 25–50%	Decrease in fat content, use of a cost-effective protein source: upon 25% addition fat from 11.91 to 9.86 g per 100 g f.w. and protein from 14.79 to 13.52 g per 100 g f.w.	109
Beef patty	*P. sajor-caju* powder 25–50%	Use of a cost-effective protein source; upon 25% addition: fat from 13.38 to 12.07 g per 100 g f.w. and protein from 22.73 to 19.37 g per 100 g f.w.	110
Ready-to-eat paste	*P. sajor-caju* powder 4–20%	Decrease in fat content, use of a cost-effective protein source; upon 20% addition fat from 13.82 to 8.16 g per 100 g f.w. and protein from 7.12 to 11.67 g per 100 g f.w.	111
Milk	*P. ostreatus* HW-β-glucan 0.25–1%	Increase in the counts of *S. thermophilus* and *L. bulgaricus*	113
	P. eryngii HW-β-glucan 0.125–0.5%	Increase in the counts of *S. thermophilus*	114
Soymilk	*P. eryngii* HW-β-glucan 0.5%	Increase in the counts of *B. longum*	115

[a] I-β-glucan: insoluble β-glucan; HW-β-glucan: hot-water-soluble β-glucan.

Table 8 Sensory attributes elicited by the addition of dried *Pleurotus* spp. or β-glucan-rich fractions obtained from *Pleurotus* spp. as ingredients in model foods

Food	Ingredient[a]	Sensory attribute	Judges	Hedonic scale	Ref.
Bread	*P. pulmonarius* powder 5–25% of flour	Appearance, crust and crumb colour, texture, taste, chew ability, flavour and overall acceptability	20	9-Points (1 = extremely unacceptable; 9 = extremely acceptable)	102
	P. ostreatus powder 5–10% of flour	Crumb colour, crumb texture, aroma, taste and overall acceptability	20	9-Points (1 = dislike extremely; 9 = like extremely)	103
Biscuits	*P. sajor-caju* powder 2–15% of flour	Colour, texture, taste, odour and overall acceptability	10	9-Points (1 = excellent; 9 = very poor)	104
	P. sajor-caju powder 4–12% of flour	Aroma, colour, appearance, crispiness, flavour, overall acceptability	60	7-Points (1 = dislike the most; 7 = like the most)	18
	P. sajor-caju powder 2–6% of flour	Aroma, colour, appearance, crispiness, flavour and overall acceptability	60	7-Points (1 = dislike extremely; 7 = like extremely)	106
Pasta	*P. eryngii* I-β-glucan fraction 2–6% of flour	Colour, flavour, hardness, and overall acceptability	30	9-Points (1 = dislike extremely; 9 = like extremely)	17
Tapioca cracker	*P. sajor-caju* powder 5–20%	Colour, odour, crispness, taste, and overall acceptability	30	7-Points (1 = dislike extremely; 7 = like extremely)	107
Instant drink	*P. eryngii* broth	Colour, flavour and overall acceptability	50	7-Points (1 = dislike extremely; 7 = like extremely)	108
Chicken patty	*P. sajor-caju* powder 25–50%	Aroma, colour, springiness, juiciness, flavour and overall acceptability	60	7-Points (1 = dislike extremely; 7 = like extremely)	109
Beef patty	*P. sajor-caju* powder 25–50%	Colour, juiciness, elasticity, flavour and overall acceptability	60	7-Points (1 = dislike extremely; 7 = like extremely)	110
Ready-to-eat paste	*P. sajor-caju* powder 4–20%	Aroma, colour, viscosity, hotness, sourness, aftertaste and overall acceptability	50	7-Points (1 = dislike extremely; 7 = like extremely)	111

[a] I-β-glucan: insoluble β-glucan.

room fibre, which interfered with the starch granules by reducing the sizes and inducing uneven spherical shapes, resulting in reduced starch susceptibility to digestive enzymes.

The effects of the addition of *Pleurotus* spp. powder on the sensory properties of bread and biscuits were also investigated (Table 8). Okafor *et al.*[102] found that bread samples supplemented with over 15% (flour basis) of *P. pulmonarius* powder negatively affected the liking scores, maybe due to a poor loaf size, dark colour and a pronounced mushroom taste and flavour. Accordingly, Ndung'u *et al.*[103] found that wheat flour could be replaced with a low concentration (5%) of *P. ostreatus* powder to make fortified bread without adversely affecting the sensory acceptability. Indeed, the liking scores of the colour attribute decreased with increasing the mushroom content, due to the presence of dark coloured mushroom flour. Moreover, all the composite breads had a characteristic odour that could be responsible for the poor rating in aroma. Similarly, Prodhan *et al.*[104] found that biscuits without incorporation of the mushroom powder obtained the highest score for overall acceptability compared to the fortified samples. However, considering the three mentioned studies, it must be taken into account that the number of semi-trained panellists involved was not appropriate.[105] Concerning the supplementation of biscuits with different concentrations of *P. sajor-caju* powder, two studies have been conducted involving an adequate number of consumers.[18,106] Wan Rosli *et al.*[106] added lower concentrations of mushroom powder and observed no significant differences in overall acceptance among samples. Ng *et al.*[18] found that supplementation with *P. sajor-caju* powder up to 8% to biscuits could lead to a more desirable aroma, colour and flavour when compared with the biscuit without supplementation. Nevertheless, with higher amounts of *P. sajor-caju* powder, undesirable results were obtained, with decreasing liking scores due to the higher degree of firmness and the stronger aroma and flavour as well as the darker surface colour of the biscuits.

In pasta, insoluble dietary fibre separated from mushroom powder was added at levels of 2–6% of semolina to fortify the product with mushroom β-glucans (Table 7) (Kim *et al.*, 2016).[17] The results of sensory evaluation showed that common wheat pasta obtained the lowest liking scores, while the acceptability increased with the addition of the insoluble β-glucan fraction. In particular, the sample with 2% of the β-glucan-rich fractions added to replace wheat flour was significantly preferred compared to the sample without supplementation. However, an unsuitable number of judges was involved (Table 8).[105]

Yahya *et al.*[107] incorporated powdered *P. sajor-caju* in a popular snack food in Malaysia and other Asian countries. Usually, these snacks (fried crackers) are produced with tapioca flour and fresh seafood, whereas the authors used mushroom powder as an alternative protein source, which was also suitable for vegetarians (Table 7). The fortified snacks showed higher mean scores for all the sensory attributes and for overall acceptability compared to the sample without the addition, maybe due to odour and taste enhancement by *P. sajor-caju* powder. However, it was difficult to draw firm conclusions due to the small group of consumers and the scale used in the sensory evaluation (Table 8).[105]

Lin *et al.*[108] utilized the centrifuged broth from blanched *P. eryngii*, rich in taste-effective and bioactive components (which is a by-product of *Pleurotus* spp. processing), to develop a novel functional product as an instant drink. The centrifuged broth recovered consisted of 54.2–62.8% of the total weight of blanched mushrooms. The solids of the centrifuged broth contained free amino acids (15.20–34.23%), 5′-nucleotides (7.44–9.71%), sugars and polyols (33.55–34.97%) and substantial amounts of ergothioneine (5.49–9.90%) and γ-aminobutyric acid (1.23–6.90%). The indigestible dextrin Fibersol-2 was used as the carrier for the *Pleurotus* broth components (Table 7). Instant drinks (centrifuged broth mixed with Fibersol-2 at ratios of 1 : 3 and 1 : 5) dissolved in hot water were rated the highest in colour, flavour and overall acceptability, suggesting that the centrifuged broth could be developed as a functional food in the form of drink (Table 8).

Application of *Pleurotus* spp. as a high-cost protein replacer

Efforts have been also made to try to replace high-cost proteins in processed meat and poultry products. In this context, some authors investigated the ability of *P. sajor-caju*, which also permits maintaining the same protein content while decreasing production costs. To this aim, *P. sajor-caju* powder was added to chicken or beef patties at levels of 25–50% (Table 7).[109,110] Results of the sensory evaluation showed that the patties made with different levels of mushroom powder (25% and 50%) were accepted by the consumers, since the liking scores for all the sensory attributes (*e.g.* aroma, colour, elasticity, juiciness, flavour) and overall acceptability were not significantly different compared to the unfortified samples (Table 8). Saiful Bahri and Wan Rosli[111] investigated the effect of *P. sajor-caju* addition to replace coconut milk powder at a level of 4–20% on nutritional composition and the sensory acceptability of a Malaysia ready-to-eat paste (Table 7). The formulations had increased protein content and decreased fat content. Formulations with more than 40% of mushroom powder were accepted by the consumers (Table 8).

Application of *Pleurotus* spp. as a prebiotic ingredient

Like other dietary fibre components, β-glucans isolated from *Pleurotus* spp. can stimulate the growth of colon microorganisms (probiotics), *i.e.* act as prebiotics. The water- and alkali-soluble fractions of *P. ostreatus* and *P. eryngii* (separated as described in Fig. 1) showed potential prebiotic activity *in vitro* towards *Lactobacillus* spp., *Bifidobacterium* spp. and *Enterococcus faecium*. Indeed, these fractions supported the probiotic bacteria growth rate, biomass and short chain fatty acid production.[28] The insoluble fraction of *P. eryngii* was applied in a mice model system and resulted in an increased abundance of *Porphyromonadaceae, Rikenellaceae*, *Bacteroidaceae* and *Lactobacillaceae*.[112]

β-Glucan-rich fractions were also applied in model foods, including fermented milk and soymilk (Table 7). Pelaes Vital *et al.*[113] formulated a hot water extract obtained from *P. ostreatus* powder with milk (1 : 1), corresponding to a final mushroom powder concentration in the range of 0.25–1% before fermentation with *Streptocuccus thermophilus* and *Lactobacillus bulgaricus*. Li *et al.*[114] applied the hot-water-soluble fraction obtained from *P. eryngii* to milk at a level of 0.125–0.5% before fermentation with *S. thermophilus*. In both these studies, β-glucan-rich fractions increased the counts of probiotic bacteria at the time of production and during storage for 1 month at 4 °C. In one study the effect of β-glucan addition to fermented milk on the content of angiotensin-I-converting enzyme (ACE)–inhibitory peptides was also investigated. These latter are defined as bioactive peptides having demonstrated anti-hypertensive properties and are produced as metabolites of bacterial proteinase, which have been widely found in dairy products. The addition of 0.125% of the hot-water-soluble fraction of *P. eryngii* led to increased levels of ACE-inhibitory peptides. However, higher additions led to lower ACE-inhibitory activity, probably due to the increase in proteolytic activity.[114]

The hot-water-soluble β-glucan fraction obtained from *P. eryngii* and the whole *P. eryngii* powder were also added to soymilk at a level of 0.5% before fermentation with *Bifidobacterium longum.* This study revealed that the β-glucan-rich fraction had a higher bifidogenic effect compared with the whole *P. eryngii* powder.[115]

Moreover, the functional fermented milk and soymilk also showed different physical properties than the control product due to having a less dense microstructure, as revealed by texture analysis and scanning electron microscopy and/or confocal laser scanning microscopy. However, liking tests with consumers were not performed.[113–115]

Conclusive remarks and future work

The concepts proposed in this review are to explore the use of *Pleurotus* spp. as a sustainable food ingredient to address the needs of populations with endemic nutritional deficiencies as well as the needs of populations at risk or affected by some chronic diseases.

Even though there has been some progress in the reduction of large-scale nutritional deficiencies in the world, there are periodic reports of outbreaks of protein, vitamin and mineral deficiencies related to populations under various distress conditions. It is also worth considering that nutritional deficiencies could even be underestimated, given that many cases are not reported in the medical literature.[54] From the studies above summarized, it can be concluded that *Pleurotus* spp. grown on various food processing by-products can meet, to a considerable extent, the daily requirements of some essential amino acids, vitamins of the B group, vitamin D, Fe, Zn and Se. However, a better knowledge on the effects of the growth substrate and species on *Pleurotus* composition would lead to a more efficient design of its dietary applications.

There should also be a shift towards the use of sustainable sources to be used in the dietary prevention and management of the major chronic diseases. The human studies reported above have demonstrated potential immunomodulatory, hypolipidemic and hypoglycaemic effects of *Pleurotus* consumption. While the role of β-glucans as an anti-inflammatory agent has been well documented, the identification of possible healthy roles of other molecules that are bioactive *in vitro* is still lacking and deserves further investigation.

The so-far described food applications of *Pleurotus* powder or β-glucan-rich fractions isolated from *Pleurotus* spp. have mainly considered this mushroom as a source of proteins and β-glucans. However, to take advantage of the great potential of *Pleurotus* spp., a major focus on its micronutrients and bioactive compounds is needed. Moreover, the sensory properties of functional foods enriched with *Pleurotus* spp. play a pivotal role in food acceptance by consumers. In this context, sensory evaluation with a proper number of assessors could make a fundamental contribution to product optimization.

Conflicts of interest

There are no conflicts to declare.

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