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Mushroom-enriched millet pasta: exploring nutritional value, antioxidant activity, and sensory acceptability

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ABSTRACT

The growing demand for functional and nutritious foods has prompted the development of alternative food products that can be easily incorporated into daily diets. This study investigates the impact of mushroom supplementation on the nutritional profile of millet pasta, focusing on protein, fibre, vitamin D and antioxidant content. Three types of mushrooms i.e. oyster, button, and shiitake were incorporated at varying concentrations (0%, 5%, 10%, and 15%) into millet pasta formulations. Results showed a significant increase in protein content, with button mushrooms providing the highest enrichment (13.86 g/100 g). Dietary fibre content also increased substantially, particularly with oyster mushrooms (8.45 g/100 g), while carbohydrate and fat content remained largely unaffected. Vitamin D levels were significantly enhanced, with button mushrooms showing the highest content (6373 IU). Antioxidant activity, as measured by DPPH scavenging, was highest in shiitake mushrooms. Sensory evaluation revealed that while all mushroom types improved the nutritional value, oyster and button mushrooms were the most acceptable in terms of colour, taste, and texture, with shiitake mushrooms receiving lower acceptability due to their strong flavour and aroma. These findings highlight the potential of mushroom-fortified millet pasta as a nutritious, functional food product with enhanced health benefits.

Keywords: Mushrooms, millets, pasta, nutritional value, antioxidant activity, sensory evaluation, functional foods

The demand for healthier, nutrient-dense food alternatives has led to an increasing interest in functional foods that offer enhanced nutritional profiles. Pasta, a staple in many diets, typically contains high levels of carbohydrates and low amounts of essential nutrients like protein, fibre, and micronutrients. Millet, a nutritious pseudo-cereal known for their rich protein, dietary fibre, gluten-free nature, and abundant micronutrients (vitamin and mineral), serve as an ideal base for fortification while promoting overall health and wellness (Kumar *et al.*, 2024). Sorghum, one of the most popular millets in India is an excellent source of complex carbohydrates, iron, zinc, B vitamins, polyphenols, anthocyanins, and antioxidants (Kulamarva *et al.*, 2009). However, the addition of functional ingredients can further improve the nutritional value of millet pasta. Mushrooms, particularly button, oyster, and shiitake mushrooms, are rich in proteins, fibres, essential minerals (iron, phosphorus, copper, potassium and selenium), vitamins (notably Vitamin D and B), and antioxidants (Dawadi *et al.*, 2022). This study aims to explore the effect of mushroom supplementation on the nutritional profile of millet pasta, focusing on protein content, fibre,

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vitamin D, antioxidant activity, and sensory properties. By incorporating different types of mushrooms at varying concentrations, the research seeks to identify the most effective supplementation strategies for enhancing the nutritional quality and consumer acceptability of millet pasta.

MATERIALS AND METHODS

Raw Materials

Fresh button, oyster, and shiitake mushrooms were procured from the ICAR-Directorate of Mushroom Research, Solan. These mushrooms were washed, sliced, dried at 50°C for 20 hours, and ground into fine powders. Sorghum millet semolina used as the base for the pasta was procured from ICAR-Institute of Millets Research, Hyderabad.

Preparation of Mushroom-Millet Pasta

The pasta was prepared by fortifying sorghum millet semolina with varying percentages (0%, 5%, 10%, and 15%) of button, oyster, and shiitake mushroom powders. The formulations were processed using a cold extrusion method to achieve consistent texture and form (Table 1).

Proximate composition and energy value

All the product samples were analyzed for moisture, crude fat, ash and crude fiber content by following standard AOAC (2010) protocols. Protein content of the products was analyzed using Micro Kjeldahl method (Kelplus Classic Dx, Pelican Equipment's). Total carbohydrate content was calculated by difference method (Total carbohydrate = 100- [Moisture + Protein + Fat + Ash content]). Energy values of the products were obtained by multiplying protein, carbohydrates and fat content by the factor 4, 4 and 9, respectively.

Vitamin D assay

For determination of Vitamin D, product samples were first dried at 50°C for 12 hours and ground to a fine powder. One gram of this powdered sample was extracted with 25 ml of methanol/dichloromethane (75:25 v/v) solution at room temperature followed by centrifugation at 10,000 rpm for 5 min. This extraction process was repeated three times and the total extract was evaporated to dryness in a rotavapour at 40°C. The residue obtained after drying was dissolved in 2 ml of methanol and filtered through a 0.45 ìm nylon syringe filter. The absorption of filtrate was taken at 264 nm through a UV-VIS spectrophotometer. 25 mg vitamin D_2 working standard was weighed accurately and taken into a 25 ml volumetric flask containing solution mixture (methanol/ dichloromethane, 75:25 v/v). A standard curve was plotted from 100 to 1000 ìg concentration at A_{264} (Barreira *et al.*, 2014).

Antioxidant activity

Antioxidant activity of product samples was determined by 2, 2- diphenyl-2-picryl-hydrazyl (DPPH) inhibition method. For the extraction, 1 g of sample

Table 1. Preparation of mushroom pasta and vermicelli formulations using cold extrusion process

Mushroom powder (%)	Sorghum (Jowar) semolina (g)	Wheat semolina (g)	Mushroom powder (g)	Water (ml)
0	900	600	0	540
5	855	570	75	540
10	810	540	150	560
15	765	510	225	560

*Batch size- 1.5 kg

was added to 10 ml methanol and centrifuged at 4000 rpm. Supernatant was collected and to 0.1 ml of the supernatant was added to 3 ml of DPPH solution (25 µg/ml methanol) followed by incubation for 30 min at 25-30°C in water bath and the absorbance at 517 nm was measured using spectrophotometer (Perkin Elmer UV/VIS spectrometer Lambda 25, Germany). A blank was also run along with the samples. This assay is based on antioxidants reducing DPPH radicals in methanol, which results in a decrease in absorbance at 517 nm. The reduction in absorbance with the addition of sample was used to calculate antioxidant activity (Kang and Saltveit, 2002).

Sensory Evaluation

The sensory quality of the pasta was evaluated by a panel of 16 trained judges using a nine-point hedonic scale. The evaluation criteria included colour, appearance, texture, flavour, mouthfeel, and overall acceptability. Nine-point hedonic scale is as follows; like extremely - 9, like very much - 8, like moderately "7, like slightly - 6, neither like nor dislike - 5, dislike slightly 4, dislike moderately - 3, dislike very much - 2, dislike extremely – 1.

Statistical analysis

The data obtained in the present study were subjected to one factor statistical analysis of variance (ANOVA) technique using completely randomized design (CRD). The critical difference value at 5 per cent levels of significance was calculated and used for making comparison among different formulations. Statistical ranks based on significant difference by Duncan's multiple range tests are given in the form of superscript over corresponding data values.

RESULTS AND DISCUSSION

Effect of mushroom supplementation on nutritional profile of millet pasta

Protein content

The incorporation of mushroom powders significantly enhanced the protein content of millet pasta across all treatments (Tables 2, 3, and 4). Oyster mushroom supplementation increased protein content from 10.49 g/100 g in the control to 11.47 g/100 g at 15% supplementation, showing a gradual rise with increasing concentration. Similarly, button mushroom supplementation showed a marked improvement in protein content, reaching 13.86 g/100 g at 15% addition, which was the highest among the three mushroom types. Shiitake mushroom supplementation also significantly elevated protein content to 11.43 g/100 g at 15%. The observed increase in protein content can be attributed to the rich protein profile of mushrooms (Wang and Zhao, 2023), making them an effective protein-enhancing ingredient in millet-based formulations. The differences in protein content were statistically significant across all mushroom types, as indicated by the critical difference

Table 2. Effect of Oyster mushroom supplementation on nutritional profile of millet pasta

Oyster mushroom powder (%)	Protein (g/100g)	Ash (g/100g)	Fat (g/100g)	Fibre (g/100g)	Carbohydrate (g/100g)	Energy (g/100g)	Vit D (IU)	DPPH (mg AEAC/100g)
0	10.49^{d}	1.48	4.02	3.84^{d}	84.01	414.19	0.00^{d}	175.39^{d}
5	10.82^{c}	1.48	3.95	5.38^{c}	83.77	413.88	1537^{c}	212.92^{c}
10	11.14^{b}	1.46	3.88	6.91^{b}	83.52	413.57	3765^{b}	239.28^{b}
15	11.47^{a}	1.46	3.81	8.45^{a}	83.28	413.26	5373^{a}	257.37^{a}
CD (0.05%)	0.161	NS	NS	0.080	NS	NS	6.315	11.375

(CD) values at the 0.05% level. Among the three mushrooms, button mushroom powder exhibited the greatest increase, suggesting its superior protein enrichment potential.

Ash content

Ash content, which reflects the mineral composition of the pasta, showed notable improvements with button and shiitake mushroom supplementation but remained statistically non-significant in the case of oyster mushrooms (Table 2). Button mushroom powder increased ash content from 1.48 g/100 g in the control to 2.21 g/100 g at 15% supplementation, while shiitake mushroom supplementation enhanced it to 2.12 g/100 g. The results indicate that button and shiitake mushrooms provide additional mineral content, enhancing the nutritional value of millet pasta (Kalac, 2013; Li *et al.*, 2018, Sinha *et al.*, 2021).

Fat content

The fat content of millet pasta remained largely unaffected by mushroom supplementation across all treatments, with no statistically significant differences observed. This suggests that the inclusion of mushroom powders does not alter the lipid composition of the pasta, maintaining its low-fat nature. Such results are consistent with previous findings, where mushrooms have been reported to contain minimal fat (Ramle *et al.*, 2021).

Fibre content

Dietary fibre, an essential component for improving gut health, increased significantly with mushroom supplementation. In the case of oyster mushrooms, fibre content rose from 3.84 g/100 g in the control to 8.45 g/100 g at 15% supplementation (Table 2). A similar trend was observed for button and shiitake mushrooms, where fibre content reached 7.43 g/100 g and 5.62 g/100 g, respectively, at the highest level of supplementation (Tables 3 and 4). The substantial increase in fibre content can be attributed to the high dietary fibre present in mushrooms, particularly in their cell walls. This enhancement adds functional value to millet pasta, making it a healthier alternative for dietary fibre enrichment (Cheung, 2013).

Carbohydrate content

A gradual decrease in carbohydrate content was observed with increasing levels of mushroom supplementation. For oyster mushroom, carbohydrate content declined from 84.01 g/100 g in the control to 83.28 g/100 g at 15% supplementation. A more pronounced reduction was seen in button mushroom treatments, where carbohydrates decreased significantly to 79.94 g/100 g at 15%. Shiitake mushroom supplementation resulted in a slight decrease to 82.44 g/100 g. The observed reduction in carbohydrate content could be attributed to the dilution effect, as mushrooms primarily contribute

Table 3. Effect of Button mushroom supplementation on nutritional profile of millet pasta

Button mushroom powder (%)	Protein (g/100g)	Ash (g/100g)	Fat (g/100g)	Fibre (g/100g)	Carbohydrate (g/100g)	Energy (g/100g)	Vit D (IU)	DPPH (mg AEAC/100g)
0	10.49^{d}	1.48^{d}	4.00	3.84^{d}	84.01^{a}	414.19	0^{d}	175.39^{d}
5	11.61^{c}	1.72^{c}	4.02	5.04^{c}	82.66^{ab}	413.17	1937^{c}	192.02^{c}
10	12.74^{b}	1.96^{b}	4.00	6.23^{b}	81.30^{bc}	412.15	4258^{b}	199.29^{b}
15	13.86^{a}	2.21^{a}	3.99	7.43^{a}	79.94^{c}	411.12	6373^{a}	207.33^{a}
CD (0.05%)	0.478	0.109	NS	0.087	1.614	NS	13.512	5.004

protein, fibre, and ash, thereby reducing the proportion of carbohydrates in the final product (Kakon *et al.*, 2012).

Energy content

The energy content of millet pasta showed minimal variation with mushroom supplementation across all treatments. Oyster, button, and shiitake mushrooms resulted in slight but non-significant decreases in energy values. This consistency suggests that the energy density of millet pasta is maintained even with the inclusion of mushroom powders, which is beneficial for consumer acceptance.

Vitamin D content

A significant enhancement in vitamin D content was observed with mushroom supplementation. Control samples contained no detectable vitamin D, while supplementation with 15% oyster, button, and shiitake mushroom powders resulted in 5373 IU, 6373 IU, and 4101 IU, respectively. Button mushrooms exhibited the highest vitamin D content, likely due to their ability to synthesize vitamin D under ultraviolet light exposure (Singh *et al.*, 2023). This improvement highlights the potential of mushroom-fortified pasta as a natural source of vitamin D, addressing deficiencies prevalent in Indian population (Aparna *et al.*, 2018).

Antioxidant Activity (DPPH)

The antioxidant activity, measured as DPPH scavenging activity, improved significantly with mushroom supplementation. Oyster mushroom supplementation increased DPPH activity from 175.39 mg AEAC/100 g in the control to 257.37 mg AEAC/100 g at 15% addition (Table 2). Button and shiitake mushrooms showed similar trends, with the highest activities recorded at 15% supplementation (207.33 mg AEAC/100 g and 263.06 mg AEAC/100 g, respectively). Shiitake mushrooms demonstrated the most substantial improvement in antioxidant activity, likely due to their higher concentration of bioactive compounds such as phenolics and polysaccharides (Kumar *et al.*, 2021). A similar increase in the antioxidant activity of pasta with the incorporation of mushroom powder has been reported in an earlier study by Lu (2018).

Sensory evaluation of mushroom-supplemented millet pasta

The results of the sensory evaluation for millet pasta supplemented with oyster, button, and shiitake mushrooms are presented in Figures 1, 2, and 3, respectively.

Colour and appearance

Sensory scores for colour and appearance decreased with increasing levels of mushroom supplementation. For oyster mushroom-supplemented pasta, scores dropped from 8.0 (control) to 7.4 at 15% supplementation. Button and shiitake mushroom treatments followed a similar trend, with button mushroom pasta showing a reduction from 8.0 to 7.14

Table 4. Effect of Shiitake mushroom supplementation on nutritional profile of millet pasta

Shiitake mushroom powder (%)	Protein (g/100g)	Ash (g/100g)	Fat (g/100g)	Fibre (g/100g)	Carbohydrate (g/100g)	Energy (g/100g)	Vit D (IU)	DPPH (mg AEAC/100g)
0	10.49[d]	1.48[d]	4.02	3.84[d]	84.01	414.19	0[d]	175.39[d]
5	10.80[c]	1.69[c]	4.02	4.43[c]	83.49	413.34	1183[c]	239.66[c]
10	11.11[b]	1.90[b]	4.02	5.03[b]	82.96	412.48	2362[b]	251.78[b]
15	11.43[a]	2.12[a]	4.01	5.62[a]	82.44	411.62	4101[a]	263.06[a]
CD (0.05%)	0.120	0.065	NS	0.097	NS	NS	22.535	6.918

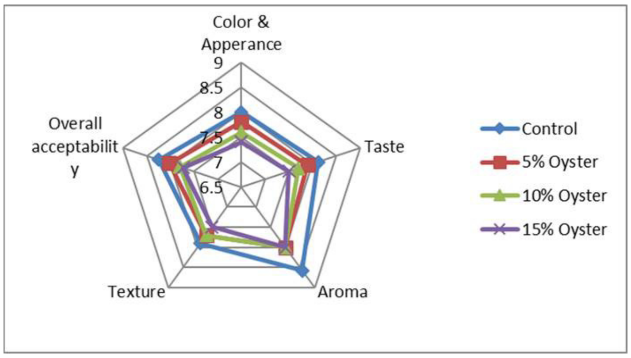

Fig. 1. Sensory evaluation of oyster mushroom supplemented millet pasta

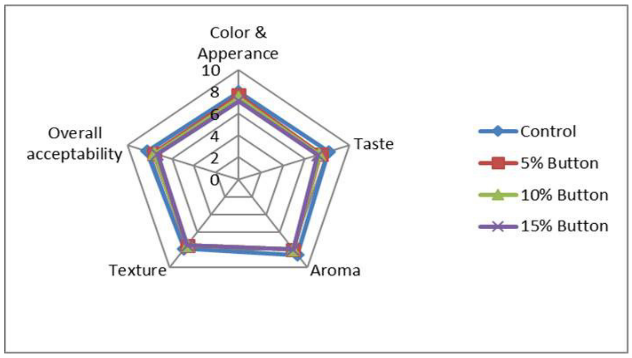

Fig. 2. Sensory evaluation of button mushroom supplemented millet pasta

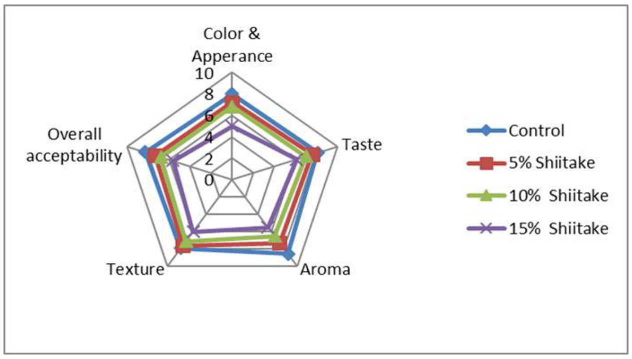

Fig. 3. Sensory evaluation of shiitake mushroom supplemented millet pasta

Fig. 4. Different formulations of shiitake mushroom fortified millet pasta

and shiitake pasta from 8.0 to 5.0. The darker colour imparted by mushroom powders may explain the lower acceptability, as lighter colour pasta is generally preferred by consumers. A similar reduction in sensory scores for colour and appearance has been observed in multigrain bread incorporated with oyster mushroom powder, as reported by Srivastava *et al.* (2024).

Taste

Taste scores also declined with increased mushroom supplementation. In oyster mushroom treatments, scores reduced from 8.12 (control) to 7.5 at 15%. A similar decline was observed for button mushroom (8.12 to 7.15) and shiitake mushroom pasta (8.12 to 6.1). The earthy and slightly umami flavour of mushrooms, especially at higher concentrations, may have impacted taste preferences. Likewise, Proserpio *et al.* (2019) observed a decrease in the taste preference for vegetable soup with higher concentrations of *Pleurotus ostreatus* powder.

Aroma

Aroma was better retained in oyster and button mushroom-supplemented pasta, with scores remaining consistent at 8.0 even at 15% supplementation. In contrast, shiitake mushroom treatments exhibited a significant reduction in aroma acceptability, dropping from 8.58 in the control to 5.5 at 15%. This reduction may be attributed to the strong, distinct aroma of shiitake mushrooms, which, despite being popular for their unique flavour properties, undergo significant changes during hot-air drying that increase total volatile content, reduce C8 and straight-chain sulphur compounds, and enhance cyclic sulphur compounds, thereby influencing aroma perception (Tian *et al.*, 2016; Wu and Wang, 2000).

Texture

Texture scores declined slightly across all mushroom-supplemented pasta samples. Oyster mushroom supplementation reduced texture scores from 7.9 (control) to 7.5 at 15%, while button and shiitake mushrooms led to scores of 7.5 and 6.0, respectively, at 15%. The incorporation of mushroom powders may have influenced pasta firmness and mouthfeel, aligning with findings by Lu *et al.* (2016), who reported that adding shiitake mushroom powder produced pasta with the highest firmness and tensile strength.

Overall Acceptability

Overall acceptability decreased with increasing levels of mushroom supplementation. Oyster mushroom-supplemented pasta showed a reduction from 8.25 (control) to 7.73 at 15%. Button mushroom pasta exhibited a similar trend, reducing to 7.33, while shiitake mushroom pasta showed the most pronounced decrease to 5.63. These results suggest that while mushroom supplementation enhances nutritional value, higher levels may negatively impact sensory attributes. These findings are consistent with a study on wheat and oat flour blends, where 5% mushroom powder resulted in the highest sensory scores, while higher concentrations led to a decline in sensory acceptability (Farzana *et al.*, 2019).

Thus, the sensory evaluation revealed that oyster and button mushroom fortification up to 15% was well-received by consumers, with slight declines in acceptability at higher levels for shiitake mushroom powder. Shiitake's strong flavour made it less desirable in higher concentrations, suggesting an optimal inclusion of 10%. In the comparative analysis of sensory attributes among the three mushroom types, oyster mushrooms exhibited the highest sensory acceptability, with minimal reductions across all attributes. Button mushrooms showed moderate declines, particularly in terms of color and taste, which affected their overall appeal. In contrast, shiitake mushrooms had the lowest acceptability scores, primarily due to their strong aroma and noticeable color changes, which were less favored by sensory evaluators.

CONCLUSION

The development of mushroom and millet-based pasta has proven to be an effective approach to creating nutritious and functional foods. The incorporation of button, oyster, and shiitake mushrooms significantly enhanced the protein, fibre, vitamin D, and antioxidant content of the pasta. Sensory evaluation confirmed the acceptability of the mushroom-supplemented formulations, with oyster and button mushrooms being the most suitable for fortification. Comparative analysis revealed that while all three mushroom types improved the nutritional profile of the pasta, distinct differences in their contributions were observed: button mushrooms provided the highest protein content, oyster mushrooms contributed the most dietary fibre, shiitake mushrooms demonstrated superior antioxidant activity, and button mushrooms also exhibited the highest vitamin D content. These findings highlight the potential of mushroom-millet-based pasta as a healthy, nutrient-dense alternative to traditional pasta. The variations among mushroom types reflect their unique biochemical compositions, positioning them as valuable functional food ingredients.

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