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**Research article**

# Pharmacological and therapeutic value of bamboo mushroom *Phallus indusiatus* (Agaricomycetes)

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**ARTICLE INFO**

Received 29/03/2022; accepted 27/05/2022

<http://doi.org/10.6092/issn.2531-7342/14666>

**Abstract**

Various wild mushrooms have been appreciated as a source of food and medicines from centuries. Although wild mushrooms were formerly utilised on a modest scale, people now favour mushrooms as a source of nutritious or healthy food. Pharmaceutical scientists have discovered many therapeutic ingredients from these mushrooms. Since few decades researchers have confirmed the therapeutic implications of such mushrooms and similar under-utilized food. *Phallus indusiatus* Vent. is one of such mushrooms which well accepted by researchers as a source of bioactive compounds. It is synonymously known as *Dictyophora indusiata* (Vent.) Desv. It is also possible to evaluate the therapeutic roles of different bioactive compounds extracted from *P. indusiatus*. Most of them have great healthy benefits such as antioxidant and antimicrobial activity, immunomodulatory, organoprotective and anticancer effects. Due to the outstanding healthy benefits and therapeutic value various molecules from *P. indusiatus* have been studied in recent years.

**Keywords**

*Phallus indusiatus*, *Dictyophora indusiata*, mushroom, fungi, bioactive compounds, polysaccharides

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**Introduction**

*Phallus indusiatus* Vent. [syn. *Dictyophora indusiata* (Vent.) Desv.] is a cosmopolitan saprobic agaricomycetous fungus from order Phallales commonly known as stinkhorn fungus. Many species from genus *Phallus* are widely recognized as edible with medicinal properties. *Phallus indusiatus* is also acknowledged as medicinal in many regions of the world including southern Asia, Africa, Australia, and the Americas (Guzman et al., 1990). The species belonging to *Phallus* are usually short-lived and early development of their fruiting bodies (egg phase) is an oval or round-shaped structures which later form mature basidioma, differing in colour and pattern (Tuno, 1983; Gogoi and Parkash, 2014). This mushroom has several common names such as Queen of mushrooms, Stinkhorn fungus, Veiled lady mushroom, Bamboo mushroom etc. According to recent literature data, the *P. indusiatus* is considered as the valid taxonomic name although it is more popular as *D. indusiata* (Elkhateeb et al., 2020). As a saprobic fungus it grows well in rotted woody trunk or organically rich soil. Macroscopically it has three diagnostic features: a conical cap, a stalk and an indusium net-like white veil that hangs from the head/cap down to cover the leg/stalk (Habtemariam, 2019). The species is cosmopolitan in distribution and abundant in grassland habitat (Sitinjak, 2016). It is concluded



that the species can stimulate the growth of grass species and protect soil layers from degradation (Sitinjak, 2016). According to Kreisel (1996), the form and surface configuration of the receptacle, the colour of the receptacle and volva, the size of the basidiomata, and the presence or absence of the indusium are the most essential morphological criteria for the taxonomy of different *Phallus* species.

This fungus has been consumed by ethnic groups of many countries including China and other Asian countries. Along with advancements in planting techniques and the selection of superior cultivated varieties, the farming and consumption of *P. indusiatus* are growing at an alarming rate (Wang et al., 2021). However, this fungus is not only appreciated as a food, but also as an important repository of many bioactive compounds with therapeutic value. According to ethnomycological data, consumption of *P. indusiatus* has been first recorded in China since 618 AD (Zidan and Neameh, 2014). In traditional system, it is known to treat different nutritional bioactivities like benefits to eyes, cardioprotective, tumour suppressive effect, etc. (Elkhateeb et al., 2020). Research data revealed that the fungus possess antioxidant, organoprotective, anticancer, immuno-modulatory, anti-diabetic, anti-obesity, and other therapeutic effects. As the quality of the fruiting body degrades quickly due to ageing and autolysis, it must be consumed or processed within a few hours after maturity (Wang et al., 2021). In this review authors have tried to present the up-to-date information available on the therapeutic attributes of *P. indusiatus*. This review would also provide new perspectives on the research directions towards pharmacologically active natural products and their commercial utilization.

### **Bioactive compounds and therapeutic application**

The fruiting body of *P. indusiatus* is treasured for its appealing appearance, delectable taste, and high nutritional value, as well as its bio-active compounds with beneficial medicinal properties (Fig. 1). It is known to possess a wide range of bioactive compounds that impart vast nutritional and medicinal value to human. It exhibits antioxidative, anti-inflammatory, anticancer, immunomodulatory, antibacterial, hepatoprotective, and antidiabetic activities, as well as it contains several vitamins. Polysaccharide, flavone, vitamins, and unsaturated fatty acids are the primary active components of this fungus. It has gained a lot of attention in the pharmaceutical industry due to the therapeutic effects, such as anti-stress, anti-tumour, immunomodulatory agent. Many extensive studies on bioactive compounds of *P. indusiatus* have been conducted using animal model. Ker et al. (2011) revealed up to 47% carbohydrate composition of the dry weight, 38% is soluble polysaccharides, while crude protein and crude fibre content account for 6% and 29%, respectively. It is widely established that mushrooms contain polysaccharides that are potent immunomodulatory agents. Zidan and Neameh (2014) isolated 10 different kinds of monosaccharides such as rhamnose, fucose, ribose, arabinose, xylose, allose, talose, mannose, galactose, glucose and myco-inositol. Hara et al. (1982, 1983) isolated two soluble glucans, characteristically having  $\beta$ -(1 $\rightarrow$ 6) branches linked to  $\beta$ -(1 $\rightarrow$ 3)-D-glucosamine frames. Most of the researchers considered that the pharmacological and therapeutic activities of *P. indusiatus* are assigned to the polysaccharides (Wang, 2018). Lin (2003) isolated a polysaccharide (Di-S2P) with a homologous MW870 kDa consisting of monosaccharide D-glucose : D-galactose : D-mannose : xylose in a molar ratio 1.62 : 1.87 : 100 : 0.93 (Wang and Shiao, 2000). Among the most important polysaccharides is 1,3- $\beta$ -glucan which is well recognized for many therapeutic implications. Wang and Shiao (2000) also isolated a water-soluble triple helical glucan (PD3) having MW510 kDa. Ke and Lin (2001) obtained a glycoprotein DIGP-2, having the molar ratio D-galactose : D-glucose : D-mannose (0.78 : 2.12 : 1) (Ouyang et al., 1998). It consists of seven essential amino acids, 12 precious metallic ions and rather high content of vitamin E,  $\beta$ -carotene, thiamine, riboflavin, nicotinic acid, l-ascorbic acid, calcium and phosphate (Mau et al., 2001; Ma and Zhang, 2004; Bai and Fang, 2008). Ishiyama et al. (1999) isolated five monoterpene alcohols from the fruiting body of the mushroom in which one was identified as 3,7-dimethyl-1,6-octadiene-3,4-diol and structures

of remaining four were determined by means of spectral and chemical method and represented as 4-[(3R,4S)-3-hydroxy-3,7-dimethylocta-1,6-dienyl(Z)-9-octadecenoate; 4-[(3R,4S)-3-hydroxy-3,7-dimethyl-octa-1,6-dienyl-(9Z,12Z)-9,12-octadecenoate; 3,7-dimethyl-1,6-octadiene-3,4,5-triol; bis[6-(3,4,7 trihydroxy-dimethyloctenyl) ether (Sharma et al., 2004). *Phallus indusiatus* also possess a notable amount of furfural compound namely, 5-(hydroxymethyl) -2-furfural which have tyrosinase inhibitory activity and due to this compound, it is now added to the list of beneficial food like honey, fruit juices, milk, vinegar and beverages (Sharma et al., 2004; Habtemariam, 2019). Xu et al. (2015) isolated and purified a polysaccharide, DiPS-3, from the fruiting body of this mushroom and confirmed its rheological properties and also suggested that DiPS-3 could be used as a kind of natural food additives and polysaccharide gel in food industry (Xu et al., 2015). Sharma et al. (2004) reported anti-tyrosinase activity of the mushroom and its active component was isolated and characterized as 5-(Hydroxymethyl)-2-furfural (HMF) by spectroscopic analysis (Sharma et al., 2004). Using this mushroom tyrosinase, they observed that methanolic extract of the fruiting bodies showed significant inhibition of L-DOPA oxidation.

### Antioxidant activity

Antioxidant activity of *P. indusiatus* is well established by many researchers. A study reported that aqueous extract from *P. indusiatus* retains antioxidant properties like scavenging free radicals and superoxide anions (Oyetayo et al., 2009). Its stipe and pileus contain high antioxidants substances which are able to prevent oxidation processes and reduce the risk of chronic diseases (Boonsrirattana, 2020). Mau et al. (2002) conducted a series of experiments and concluded that *P. indusiatus* has excellent reducing power and scavenging activity on DPPH radicals. Li et al. (2012) also reported that aqueous extracts of the mushroom possessed the highest hydroxyl radicals scavenging capacity and ability to inhibit lipid peroxidation. Another valuable study reported that aqueous extract of the fungi at a concentration of 2 mg mL<sup>-1</sup> exhibited high scavenging activities for DPPH radicals (97.35%), hydroxyl radicals (52.28%) and superoxide anions (48.64%) as well as strong reducing power of 1.22 but a very weak ferrous ion chelating effect of 18.56% (Oyetayo et al., 2009). Ker et al. (2011) obtained six soluble polysaccharides from the mushroom, of which the one with the smallest weight exhibited the most potent scavenging effect against hydroxyl radicals and superoxide anion. Wang et al. (2015) invented a novel triple helical polysaccharide DiPS-3 and showed that it exhibited a variety of radical scavenging activities. Zhang et al. (2016) evaluated the capacity of DiPS against paraquat-induced oxidative toxicity in *Caenorhabditis elegans* and have found that DiPS is capable of reducing reactive oxygen species (ROS) and Malondialdehyde (MDA) levels and increasing superoxide dismutase (SOD) activity.

### Immunomodulatory effect

Polysaccharides derived from *P. indusiatus* has notable anti-inflammatory activity. Mushroom-covered mucus has a concentrated volva that has high hyaluronic acid (HA) and allantoin which has anti-inflammatory effect to reduce skin irritation, add moisture, and regenerate damaged skin cells. Many researchers reported that biologically active polysaccharides of *P. indusiatus* have capacity for the enhancement of immune responses. Hara et al. (1991) isolated five homogenous and one conjugated polysaccharide from the 70% aqueous ethanol extract and hot water extract of this mushroom confirmed their immunomodulatory effects like mitogenic and colony stimulating factor (CSF) inducing effects. According to this research two homologous polysaccharides fractions (T-3-Ad and T-4-N) and a conjugated polysaccharide fraction (T-2-A) exhibited mitogenic and colony stimulating factor (CSF) inducing activities in dose dependent manner, whereas T-3-M', T-5-N and T-2-HN were not effective at any dose. Another study considered that these polysaccharides act as

immunopotentiators as in many cases these polysaccharides promote Nitric oxide (NO), tumour necrosis factor  $\alpha$  (TNF- $\alpha$ ) and Interleukin 6 (IL-6) secretion and develop immune response to treat immunodeficiency and chronic infections (Liao et al., 2015). The study of Guo et al. (2005) concluded that extract of the mushroom has radiation protective effect and according to the study the thymus and pancreatic index, CD4<sup>+</sup>, CD16, CD57 and interleukin 2 were all improved, while CD8<sup>+</sup> was decreased when the extract was applied to the organism affected by radiation. Further findings have shown that on entering into the gastrointestinal tract DiPS are likely to first exert an immunomodulatory effect on the intestinal mucosal immune system, and then influence the overall immune system through a series of pathways, including controlling macrophages, regulatory T cells and secretion of IgA and various cytokines (Schepetkin et al., 2006; Huang et al., 2017). In a study, it was showed that DiPS had a good effect on the haemolysis antibody level in a dose dependent manner (Hua et al., 2012). Various researchers confirmed that mushroom polysaccharides are known to stimulate the immune system responses of natural killer cells, T-cells, B-cells and macrophages, but works of Hua et al. (2012) suggested that mushroom polysaccharides like DiPS could improve the non-specific immune function and lymphocyte function. Hua et al. (2012) isolated two polysaccharides from the mushroom which were later named as DIP-I and DIP-II D and further findings suggested that DIP-I could improve the weight of thymus organ of mice and phagocytosis of monocyte in a dose-dependent manner, whereas DIP-II could improve the weight of spleen and thymus of mice, restoring a delayed-type hypersensitivity reaction to dinitrofluorobenzene (DNFB), improving the Natural killer (NK) cell activity and the proliferation of splenocytes at high dose.

#### **Anti-obesity and antidiabetic effect**

Many mushrooms and their products are well recognized as anti-diabetic and anti-obesity food. Due to this reasons researcher are now set their goal to evaluate the anti-obesity and anti-diabetic potential of the mushroom *P. indusiatus*. Though it is believed in China and Japan that consumption of *P. indusiatus* reduces chance of diabetes and obesity but the scientific study in this aspect is just beginning to take shape. There is only a limited data about this topic. Some comprehensive experiments showed that 45 days oral consumption of the water extractible polysaccharides of the fungi were able to reduce high fat induced obesity in a mice model (Wasser et al., 2002; Wang et al., 2019a). Such activities also provide organoprotective effect particularly where oxidative stress is implicated. Another study also employed the high-fat-induced obesity and hyperlipidaemia model to assess the potential anti-diabetic effects of that polysaccharides obtained by enzyme assisted or alkali extraction methods (Wang et al., 2019b). They also have determined a direct glucose lowering effects under oral glucose tolerance test (OGTT) while the alteration in the serum adiponectin level and the increases in insulin and leptin were normalized. The acidic and alkali extracted polysaccharides have also been shown to increase Superoxide Dismutase (SOD) and Glutathione Peroxidase (GPx) in the D-galactose-induced senescence in rats (Hua et al., 2012).

#### **Hepatoprotective effect**

Different studies revealed that *D. indusiata* has significant hepatoprotective effects. In a study it was proved the anti-hyperlipidemic and hepatoprotective properties of alkali- and enzyme-extractable *P. indusiatus* polysaccharides (Al-DPS and En-DPS) on the hyperlipidemic mice (Wang et al., 2019b). In high fat-influenced mice model having severe oxidative damage oral administration of water extractible DiPS could abolish the increased Lipid peroxidation (LPO) and simultaneously elevating SOD, GPx, Catalase (CAT) and total antioxidant (T-AOC) activities in the liver and kidney tissues (Wang et al., 2019b; Wang et al., 2019c). DiPS has some interventional effect on liver damage. A study concluded that DiPS has a protective effect on the liver of rat with arsenic poisoning as



it can reduce the arsenic content in blood, urine and liver (Wang et al., 2018). In a study it was found that *P. indusiatus* polysaccharides significantly inhibited liver transplantation tumours in mice and effectively controlled the growth of ascites tumour in mice (Zong et al., 2013). Recently some workers proved that the polysaccharides inhibit abnormal cellular growth in human liver (Zhong et al., 2013; Hu et al., 2020).

### **Neuroprotective effect**

Mushrooms contain a wide range of bioactive metabolites with positive impacts on human cognitive function, and this is very plausible. Since ancient time *P. indusiatus* is recognized as beneficial product for nervous system. Several neuroprotective compounds were isolated from *P. indusiatus*. Kawagishi et al. (1997) isolated two bioactive molecules Dictyophorine A and Dictyophorine B and also confirmed their stimulating effect on Nerve Growth Factors (NGF). These are actually two derivatives of an eudesmane type sesquiterpenes compound teucrenone and have significant influence on astroglial cells to enhance the synthesis of NGF. The compounds are special as they consist of three isoprene units and it was shown that the quantity of NGF secretion into the medium in the presence of 3.3mM of dictyophorines-A was four times higher than that of the control without treatment (Kawagishi et al., 1997; Phan et al., 2017). Lee et al. (2002) isolated three neuroprotective compounds from methanol extract of the mushroom which were named as dictyoquinazol A, dictyoquinazol B and dictyoquinazol C. Further research proved that such dictyoquinazols are unique in nature and they also showed that such compounds protected primary cultured mouse cortical neurons from glutamate and NMDA induced excitotoxicity in a dose dependent manner. Another neuroprotective compound of sesquiterpenes class has been identified from dried fruiting body of the fungus albaflavenone (Huang et al., 2011). Zhang et al. (2016) conducted an experiment to demonstrate neuroprotective effect of DiPS on neurodegenerative *Caenorhabditis elegans* models and showed that it could increase survival rate and reduce stress level under paraquat-induced oxidative conditions. It was also determined by the researchers that DiPS was also able to decrease ROS and MDA level and increase SOD activity in *C. elegans* model and restore the functional parameters of mitochondria like membrane potential, ATP content. The mushroom may promotes neurite outgrowth in the brain by increasing NGF synthesis, imitating NGF responsiveness, or shielding neurons from neurotoxicant-induced cell death. Through the underlying processes of the mushroom's neurotropic chemicals, it may act as a preventative measure against the development of Alzheimer's disease.

### **Anti-tumour activity**

Many of the pharmaceutical scientist believed that *P. indusiatus* and allied species contain some anti-carcinogenic and anti-mutagenic substances. A study found that both methanol and ethyl acetate extracts of *Phallus* sp. have significant anti-proliferative action against breast cancer cell lines MCF-7 (Ray et al., 2020). There is no doubt that many fungal polysaccharides are potent anticancer agent. Hara et al. (1982) isolated some polysaccharides from these mushrooms and concluded that few of them exhibited potent anticancer bioactivity. Ukai et al. (1982) confirmed that T-2-HN, T-4-N and T-5-N could inhibit tumor in a dose dependent manner. Polysaccharides of *P. indusiatus* appear to display potential as anti-cancerous agent through two separate mechanism of actions direct antitumor effect and immunostimulation (Habtemariam, 2019). Liao et al. (2015) successfully chelated DiPS with  $ZnCl_2$  and proved the anti-proliferative activity of DiPS complex on a group of human cancer cell lines through induction of apoptosis. Their further findings suggested that the apoptosis by the complex was associated with the condensation of nuclei, fragmentation of DNA, arrest of S-phase during cell cycle and mitochondrial dysfunction. A group of researchers isolated a novel polysaccharide ZSP4 from this mushroom and evaluated its effect on prostate cancer and have found that ZSP4 proliferate

immune cells, reverse the immune-suppressive functions of prostate cancer associated fibroblast (CAFs) and inhibit the growth of tumour by affecting micro-environment of tumor (Han et al., 2017). Apart from these other findings indicated that a polysaccharide of the mushroom having molecular weight  $6.52 \times 10^4$  Da, may act as a chemopreventive and chemotherapeutic agent in osteosarcoma cell by reducing cell viability (Zhong et al., 2013). According to this research due to the polysaccharide treatment, osteosarcoma s180 cell significantly increase caspase-3, which in turn execute apoptosis of the cell. It has been also confirmed that *P. indusiatus* polysaccharides have potential anti-tumour effect on human hepatocellular carcinoma cells (HCC) and it was found that these polysaccharides inhibited HCC-LM3 cell proliferation in a time- and dose-dependent manner and blocked the cell cycle in the G2/M phase by regulating the apoptosis related genes (Hu et al., 2020). According to them, these polysaccharides up-regulate the expression of the genes for Bax and caspase-3 and inhibit Bcl-2/Bax heterodimer formation. Daba et al. (2020) conducted an experiment to evaluate the metabolic profiles of n-hexane extract of the mushroom and found that few medium chain fatty acids like caproic acid show mild anticancer and anti-proliferation activities against HCT116 human colon cancer cell lines. Due to presence of caproic acid and similar medium chain fatty acids, n-hexane extract of fruiting bodies could potentially be used to prevent and/or treat human colorectal, skin and breast cancer cells (Narayanan et al., 2015). Many investigations have shown that the methanol extract of this species is highly efficient at quenching free radicals, making it a very promising option not only as a safe alternative anticancer medication but also as an excellent chemotherapy antioxidant adjuvant.

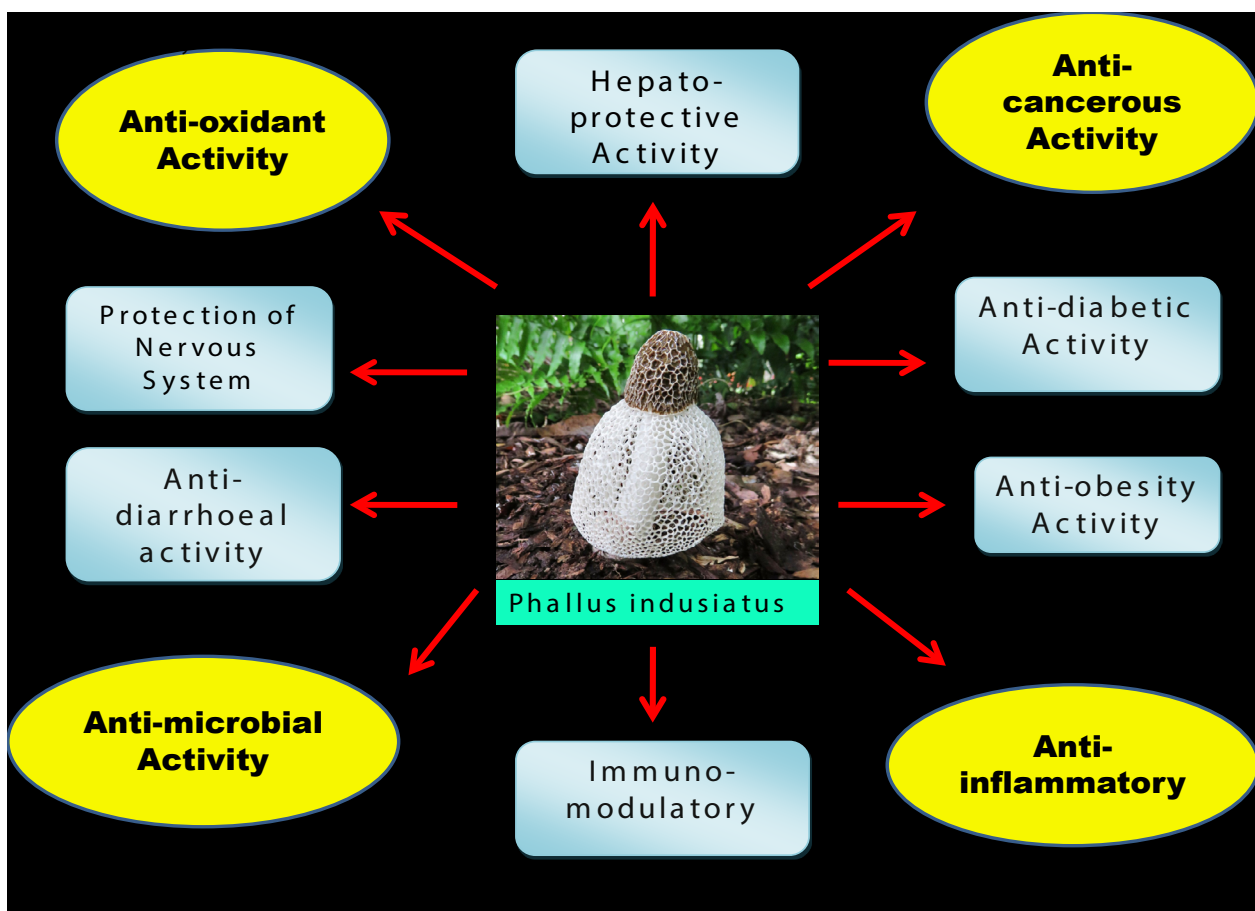
### **Anti-microbial activity**

Evaluation of the antioxidant and antibacterial properties of mushroom extracts could be extremely useful in the development of novel antioxidant and antimicrobial product and functional food additives in our diets to prevent oxidative and pathogenic damage. As a result, numerous researchers conducted in vitro studies to determine the antioxidant and antimicrobial activity of chloroform, 70% ethanol, and hot water extracts of *P. indusiatus* fruiting bodies. Antimicrobial property of methanol, ethyl acetate, ethanol, acetone and volatile oil extracts from *P. indusiatus* have been reported by many traditional and modern researchers (Tan et al., 1999; Mao et al., 2002). Gram positive and Gram-negative bacteria were both susceptible to antimicrobial effects of the fungal extracts, but microzymes and moulds were not (Hao et al., 2008). Huang et al. (2011) isolated a sesquiterpene antibiotic from the fungi, namely albaflavenone, and described its properties and bioactivity. They claim that this compound has an earthy or camphor-like odour and has potent antibacterial properties. Additionally, it was established that fungus has a significantly stronger inhibitory effect than bacteria (Wang et al., 2018). Using the crude polysaccharide (DIP) from the mushroom, researchers were able to reverse the antibiotic-induced dysbiosis and modulate the gut microbiota ecology by reducing pathogenic bacteria such as *Enterococcus*, *Bacteroides* and *Proteobacteria* (Wang et al., 2018). Both the crude and purified polysaccharides have inhibitory effect on bacteria but the former one has significant bacteriostasis and later one has not so obvious bacteriostatic activity (Kanwal et al., 2018). Oyetayo et al. (2009) performed an experiment to evaluate the antimicrobial activity of water extract of the species on some microbes and found that the extract had significant zone inhibition activity (Table 1).

<http://doi.org/10.6092/issn.2531-7342/14666>

**Table 1.** Antimicrobial effect of water extracts (200 mg mL<sup>-1</sup>) of *P. indusiatus* against selected test microorganisms (adapted from Oyetayo et al., 2009).

| Microorganism                  | Type of microbes | Inhibition zone radius (mm) |
|--------------------------------|------------------|-----------------------------|
| <i>Escherichia coli</i>        | Bacteria         | 2.00 ± 0.50                 |
| <i>Alcaligenes faecalis</i>    | Bacteria         | 7.15 ± 0.30                 |
| <i>Salmonella typhimurium</i>  | Bacteria         | 2.75 ± 0.25                 |
| <i>Shigella dysenteriae</i>    | Bacteria         | 4.00 ± 0.50                 |
| <i>Pseudomonas aeruginosa</i>  | Bacteria         | 3.75 ± 0.25                 |
| <i>Bacillus subtilis</i>       | Bacteria         | 3.00 ± 0.25                 |
| <i>Staphylococcus aureus</i>   | Bacteria         | 4.15 ± 0.25                 |
| <i>Bacillus cereus</i>         | Bacteria         | 2.35 ± 0.30                 |
| <i>Aspergillus niger</i>       | Fungi            | No inhibition               |
| <i>Aspergillus flavus</i>      | Fungi            | 1.25 ± 0.30                 |
| <i>Candida albicans</i>        | Fungi            | 5.50 ± 0.50                 |
| <i>Cryptococcus neoformans</i> | Yeast            | 3.65 ± 0.60                 |



**Fig. 1** – Therapeutic properties of *Phallus indusiatus*.

## Conclusion

The well-known edible medicinal fungus *P. indusiatus* is famous for its attractive colour and appearance. However, its natural resources are underutilized. Since last three decades study of medicinal properties and bioactive compounds have been increased to a great extent. It was showed that the edible part of *P. indusiatus* has several therapeutic implications like antioxidant activities, immunological activities, anti-diabetic, hepatoprotective, neuroprotective, anti-fatigue, anti-tumour and anti-cancer effects. Polysaccharides are regarded as the most important bioingredients of it. They are responsible for antioxidant, anti-tumour and immunoenhancing effects. Recent research concluded that some other compounds of this mushroom are also beneficial to human health. Due to health promoting effect and extraordinary therapeutic value, this mushroom has attracted people's attention and now it has been consumed regularly in many countries. This review has concluded that compounds protein, carbohydrates, vitamins and dietary fibre extracted from this species have superior health benefits and are also useful as complementary medicine to treat many ailments. Further research is also needed to confirm their activities mentioned in ancient traditional document as well as their safe application on human bodies. The further advanced study and research could improve and enhance their application in broader and more appropriate range. Large-scale clinical research in the population will confirm organoprotective capabilities of the mushroom.

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<http://doi.org/10.6092/issn.2531-7342/14666>

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