

## THE EFFECT OF MUSHROOM INTAKE ON AGING –FT-IR STUDIES ON LIVER

### SAMPLE

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### Abstract

For the betterment of human health, intake of many nutritional and medicinal plants is increased in our daily life. The consumption of mushroom leads to many health benefits but there are some adverse effects on taking in. The present work highlights the effect of mushroom intake using FT-IR studies on liver sample of albino rats (young, aged, aged treated with *Valvariella volvacea* mushroom). The animal treated with *Valvariella volvacea* mushroom reduces the intensity of total lipid level and enhances the total protein level.

### Introduction

Mushroom in recent times has become a contemporary business enterprise because of its high nutritional and medicinal values[1-6], and consequently high societal demand. There is therefore, need to maintain a constant supply of mushroom by cultivating rather than depend on seasonal forest supplies. Mushrooms are known to be among the largest of fungi that attracted the attention of naturalists before microscopes were invented. The growths of mushrooms are simple[7-11], highly useful and easily available in market. Because of high medicinal importance of these mushrooms, we have tested against albino rats. Using the FT-IR study leads the important role of mushroom in the field of liver damage.

Peter C.K. Cheung [12-19] has suggested the hypocholesterolemic effect of extracellular polysaccharide from the submerged fermentation of mushroom. Male Sprague-Dawley rats were fed two semisynthetic diets supplemented with 2% cholesterol and 1%  $\beta$ -glucan type extracellular polysaccharide isolated from two liquid cultures of straw mushroom (*Volvariella volvacea*) mycelium containing different carbon sources[20-26].

Cheung [27-30] has suggested the fruiting bodies and the mycelia of straw mushroom contain a large amount of cell-wall polysaccharides (25% and 50% dry weight, respectively) of which about two-thirds are the  $\beta$ -glucan type. Bohn and BeMiller [3] have been extensive studies on fungal polysaccharides[31-40], especially  $\beta$ -glucans as biological response modifiers. During the submerged fermentation of the straw mushroom mycelium [4], it was found that a soluble  $\beta$ -glucan type extracellular polysaccharide was present in the liquid medium and could be recovered by ethanol precipitation. Cheung et al., [5] have reported Methanol and water crude extracts from Shiitake mushroom (*Lentinus edodes*) and straw mushroom (*Volvariella volvacea*) were investigated for their antioxidant capacity in three

different assays, namely, the  $\beta$ -carotene and linoleic acid system[41-46], 1,1-diphenyl- 2-picrylhydrazyl (DPPH) radical scavenging activity, and inhibition of hemolysis of rat erythrocyte induced by peroxy radicals. Recently, water extracts of oyster Mushroom were reported to provoke contraction of nonvascular tracheal smooth muscle in vitro [47-50].

The pathogenic mould is the main cause of aspergillosis, an invasive systemic disease with high mortality rates. Besides asp-haemolysin, a plethora of other molecules isolated from *A. fumigatus* have been reported to be associated with its toxicity [7]. Mushrooms and entomopathogenic fungi are also exemplary sources of natural medicines with antidiabetic activity. With little scientific evidence, many investigators have endeavored to study the hypoglycemic effect from either the fruiting body or mycelia of various edible/medicinal fungi including *Tremella aurantia*, *Cordyceps sinensis*, and *Lentinus edodes* [8].

Cheung and Peter C.K. Cheung [9] have reported Methanol and water extracts, with antioxidant activity, from two edible mushrooms (*Lentinus edodes* and *Volvariella volvacea*) were subfractionated by liquid-liquid partition using organic solvents and by membrane ultrafiltration, respectively. The dichloromethane subfraction of the methanol extract of *V. volvacea* and the low molecular weight (LMW) subfraction of the water extract of *L. edodes* had the highest antioxidant activity against lipid peroxidation of rat brain homogenate with  $IC_{50}$  values of 0.109 and 1.05 mg/ml, respectively. The ethyl acetate subfraction of the methanol extract of *V. volvacea* was found to have comparable antioxidant activity ( $p > 0.05$ ) to caffeic acid against the oxidation of human low-density lipoprotein (LDL).

The quantification of psilocin and psilocybin in fungus material was carried out by high-performance liquid chromatography (HPLC) with ultra-violet (UV), native fluorescence (FL), and electrochemical (EC) detections [10]. The methods using gas chromatography (GC) and HPLC, coupled with mass spectrometry (MS), have recently been introduced to increase the sensitivity and selectivity of psilocin in such biological samples as plasma and urine [10]. Jayakumar et al., investigate the putative antioxidant activity of the oyster mushroom *Pleurotus ostreatus* on  $CCl_4$ -induced liver damage in male Wistar rats. Jayakumar et al., [12] investigate the putative antioxidant activity of the oyster mushroom, *Pleurotus ostreatus*, on lipid peroxidation and antioxidant status of major organs of aged (24 month old) rats when compared to young (4 month old) rats.

Sonoyama et al., reported that the plasma cholesterol concentration was significantly lower in rats fed sugar beet fiber than in those fed fiber-free or cholestyramine diets, and this difference was due mainly to a lower high-density lipoprotein (HDL) cholesterol concentration. Some Basidiomycotina mushrooms have the ability to lower the serum cholesterol concentration. It is reported that hiratake (*Pleurotus ostreatus*) lowers the serum cholesterol concentration in rats [14], and that the fruiting body of the straw mushroom (*Volvariella volvacea*) lowers elevated plasma cholesterol in hypercholesterolemic hamsters, whereas the mycelium does not [15]. These results suggest that the fruiting body of the straw mushroom could influence hepatic cholesterol synthesis and fecal excretion of cholesterol.

## Results and Discussion

### FT-IR Spectral Studies on Liver

FTIR spectra of liver of control and diet with *Valvariella volvacea* mushroom treated animals were recorded in the region  $4000\text{--}400\text{ cm}^{-1}$ . The bands occurring at  $3292\text{ cm}^{-1}$ ,  $2958\text{ cm}^{-1}$ ,  $2925\text{ cm}^{-1}$ ,  $2854\text{ cm}^{-1}$ ,  $1654\text{ cm}^{-1}$ ,  $1534\text{ cm}^{-1}$ ,  $1450\text{ cm}^{-1}$ ,  $1236\text{ cm}^{-1}$  and  $1080\text{ cm}^{-1}$  have been found to be strong and prominently present in all the spectra recorded in the present study. Fig. 1 represents the FTIR spectra recorded in the region  $4000\text{--}400\text{ cm}^{-1}$  for the liver samples of normal young (4 month) rats, while Fig. 2 corresponds to those recorded for aged old (24month) rats, Fig. 3 corresponds to those recorded for aged old rats with 5.0% *Valvariella volvacea* mushroom in 30 days.

Many of the bands in the region  $1800\text{--}1000\text{ cm}^{-1}$  are unresolved and to be closely scrutinized for data analysis to acquire a rich treasure of information of diagnostic value. Therefore, the present research work has been restricted only to the C-H stretching region ( $3000\text{--}2800\text{ cm}^{-1}$ ) representing the chromophoric groups mainly due to Lipids and Proteins. Assignment of these bands to various chromophoric groups and their probable origin has been listed out (Table 1).

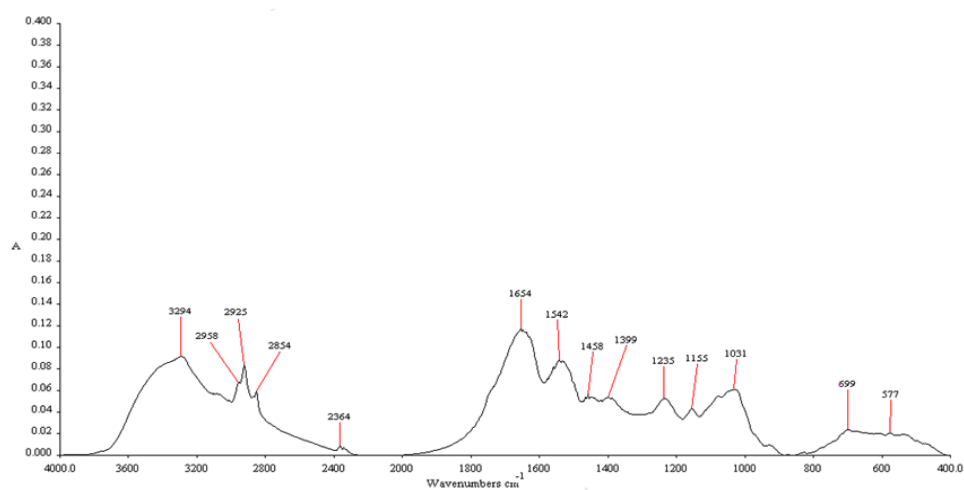


**Fig.1 FT-IR spectrum of liver of group A (Young) Albino rat**

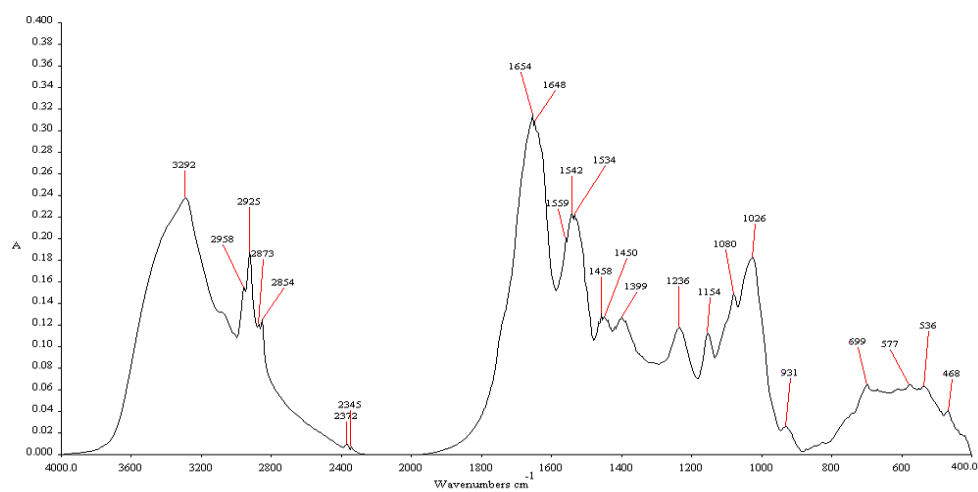


**Fig.2 FT-IR spectrum of liver of Group B (Aged) Albino rat**

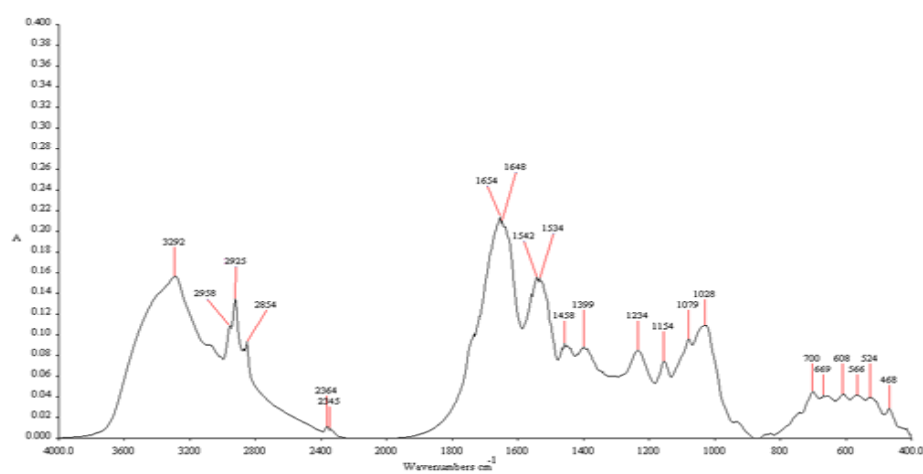


Fig.3 FT-IR spectrum of liver of Group C (Aged treated with *Valvariella volvacea*) Albino rat

**Table 1**  
**FTIR tentative assignment for three groups (young, aged, aged treated with *Valvariella* *volvacea*) of liver tissue samples**

| Wavenumber (cm <sup>-1</sup> ) |         |         | Tentative assignments                                                                                                                        |
|--------------------------------|---------|---------|----------------------------------------------------------------------------------------------------------------------------------------------|
| Group A                        | Group B | Group C |                                                                                                                                              |
| 3294                           | 3292    | 3292    | Mainly N-H stretching (amide A) of proteins with the little contribution from O-H stretching of polysaccharides and intermolecular H bonding |
| 2958                           | 2958    | 2958    | CH <sub>3</sub> asymmetric stretch: mainly lipids, with the little contribution from proteins, carbohydrates, nucleic acids                  |
| 2925                           | 2925    | 2925    | CH <sub>2</sub> asymmetric stretch: mainly lipids, with the little contribution from proteins, carbohydrates, nucleic acids                  |
| -                              | 2873    | -       | CH <sub>3</sub> symmetric stretch: mainly proteins, with the little contribution from lipids, carbohydrates, nucleic acids                   |
| 2854                           | 2854    | 2854    | CH <sub>2</sub> symmetric stretch: mainly lipids, with the little contribution from proteins, carbohydrates, nucleic acids                   |
| 1654                           | 1654    | 1654    | Amide I (protein C=O stretching): $\alpha$ -helices                                                                                          |
| 1542                           | 1542    | 1542    | Amide II (protein N-H bend, C-N stretch): $\alpha$ -helices                                                                                  |
| 1458                           | 1458    | 1458    | CH <sub>2</sub> Bending: mainly lipids, with the little contribution from proteins                                                           |
| 1399                           | 1399    | 1399    | COO <sup>-</sup> symmetric stretch: fatty acids and amino acids                                                                              |
| 1235                           | 1236    | 1234    | PO <sub>2</sub> <sup>-</sup> asymmetric stretch: mainly nucleic acids with the little contribution from phospholipids                        |
| 1155                           | 1154    | 1154    | CO-O-C asymmetric stretching: glycogen and nucleic acids                                                                                     |
| 1031                           | 1080    | 1079    | PO <sub>2</sub> <sup>-</sup> symmetric stretching: nucleic acids and phospholipids C-O stretch: glycogen                                     |

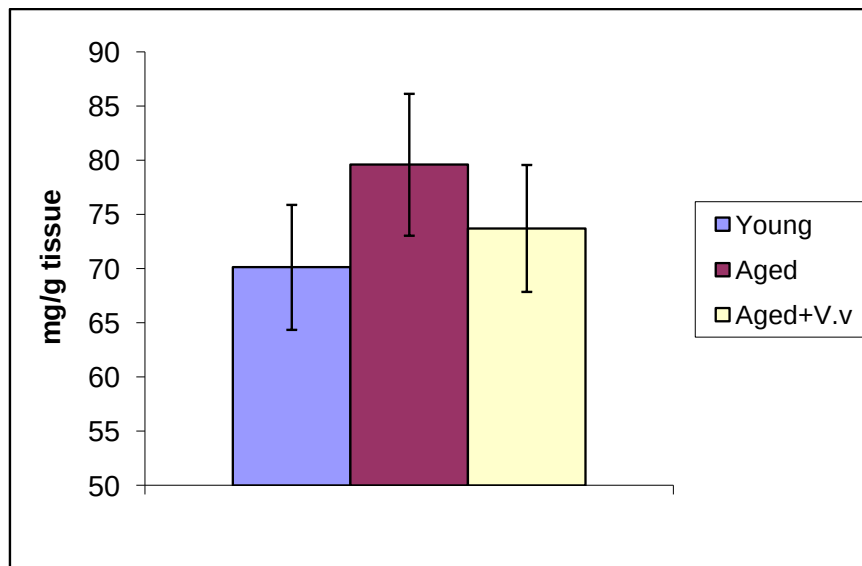


**Fig.4 Variation of *Volvariella volvacea* mushroom on total lipid in rat liver**

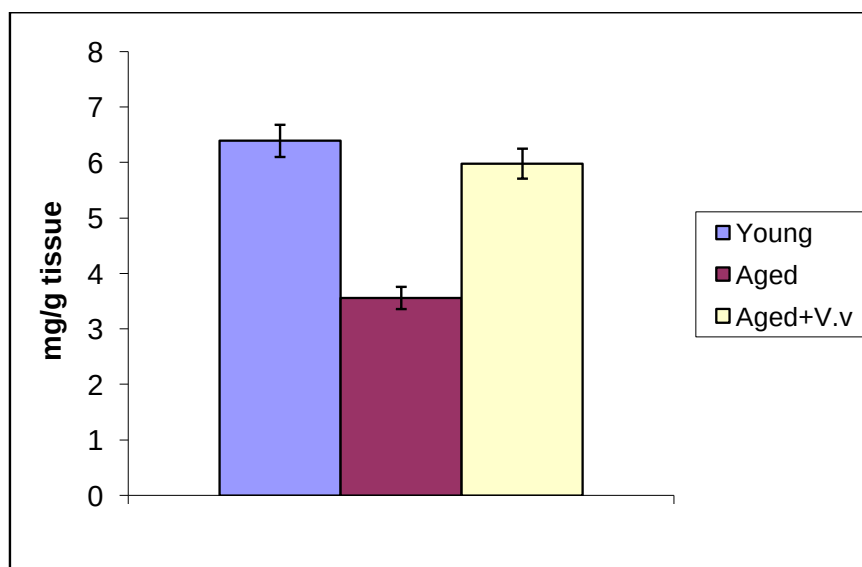


**Fig.5 Variation of *Volvariella volvacea* mushroom on total protein in rat liver**

antioxidant activity which is well correlated with their total phenolic content [17]. Phenolic compounds in plants are powerful free radical-scavengers which can inhibit lipid peroxidation by neutralizing peroxy radicals generated during the oxidation of lipids [18].

Shahidi and Amarowicz [19] have suggested that total lipid has been decreased and total protein has been increased in mushrooms. Since mushrooms (edible fungi) also possess phenolic compounds [20], it is interesting to investigate the antioxidant activity of mushroom in relation to their total phenolic contents which have been increased. In general, a correlation between higher antioxidant activity and larger amount of total phenolics was found in the mushroom extracts.

Mushrooms have long been appreciated for their flavour and texture. Now they are recognized as a nutritious food as well as an important source of biologically active compounds of medicinal value [21]. Mushrooms accumulate a variety of secondary metabolites, including phenolic compounds, polyketides, terpenes and steroids. Also, a mushroom phenolic compound has been found to be an excellent antioxidant and synergist that is not mutagenic [22].

Peter C.K. Cheung has suggested that male Sprague-Dawley rats were fed two semisynthetic diets supplemented with 2% cholesterol and 1%  $\beta$ -glucan type extracellular polysaccharide isolated from two liquid cultures of straw mushroom (*Volvariella volvacea*) mycelium containing different carbon sources. The experimental diets containing the two mycelial extracellular polysaccharides had induced a significant reduction in the levels of serum total cholesterol, LDL-cholesterol, and liver total cholesterol in the animals. There were no significant changes in the concentrations of serum triacylglycerol, HDL-cholesterol and liver total lipids [23].

Mushrooms have long constituted an integral part of the normal human diet; these are reported to contain relatively large amounts of vitamins A, C and  $\beta$ -carotene, all of which have protective effects because of their antioxidant properties. Mushrooms also contain many phenols, which are very efficient scavengers of peroxy radicals [24]. The extract of the mushroom, *Ganoderma lucidum*, has been reported to remove the hydroxyl radical, the main factor in the human ageing process [25]. *Pleurotus ostreatus* is reported to contain higher concentrations of cystine, methionine and aspartic acid than other edible mushrooms, such as *Agaricus bisporus* (brown), *A. bisporus* (white) and *Lentinus edodes* [26]. Lovastatin, a cholesterol-lowering drug derived from *Pleurotus* species, and its analogues are reported to be the best therapeutic agents for correcting hypercholesterolemia [27].

Ethyl acetate and methanol extracts of *P. florida* have been found to exhibit potent scavenging of hydroxyl radicals and inhibition of lipid peroxidation activities [28]. Lipid peroxidation, a process induced by free radicals, leads to oxidative deterioration of polyunsaturated lipids. Under normal physiological conditions, only low levels of lipid peroxides occur in body tissues. The excessive generation of free radicals leads to peroxidative changes that ultimately result in enhanced lipid peroxidation [29].

## Conclusion

From the results above administration of a *Valvariella volvacea* mushroom to protect the liver in aged wistar albino rats against oxidative stress by reducing the intensity of total lipid levels and increasing total protein levels. It remains to be seen whether a similar protective effect can be demonstrated in humans during the process of ageing. An extract of this mushroom can be conveniently incorporated in the diet as a nutritional supplement, therein augmenting the body's defences against oxidative stress.

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