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Impact of different ultraviolet radiation on the ability to accumulate vitamin D2 and the composition of different mushroom strains

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ABSTRACT

Vitamin D deficiency is one of the most common nutritional and medical condition affecting more than 50 per cent of the world's population. There are very few foods having vitamin D such as fatty fish, liver, eggs and some fortified foods. Mushrooms exposed to UV radiation are an excellent source of dietary vitamin D2. The study was conducted to see the Vitamin D accumulation potential of different strains of button, oyster, milky, shiitake, paddy straw and lion's mane mushrooms upon UV light treatment for their potential use as vitamin D supplement. Four different UV treatments have been evaluated along with effect of sun light in this study. Post-harvest UV-B (209-310 nm) treatment for a period of two hours has resulted in significant increase of Vitamin D2 content (up to 300%) in all the tested strains of mushrooms with a maximum in *Lentinula edodes* (shiitake) and *Hericium erinaceous* (Lion's Mane) mushroom.

Keywords: Mushrooms, vitamin D, UV radiation, proximate composition

Role of vitamin D is very important in maintaining calcium and phosphate balance in the body, which impacts on bone health and muscle function. Severe vitamin D deficiency causes impaired bone mineralization, resulting in rickets in children and osteomalacia in adults (Jones, 2015). In older people, vitamin D deficiency is a predictor of falls, and low vitamin D status is associated with muscle weakness and impaired balance (Girgis *et al*., 2013). In addition to these well-established effects, vitamin D status might impact on risk of a range of diseases such as cardiovascular disease, respiratory disease in children, neurodegenerative diseases, both type 1 and type 2 diabetes, types of cancers, brain disorders such as autism and schizophrenia, and reduce immune function (Ford *et al.,* 2014; Gaksch *et al.,* 2017; Koduah *et al*., 2017), although current evidence for non-skeletal beneûts is inconclusive (Theodoratou *et al*., 2014). Dr Michael Holick (Boston University Professor of Medicine) claims that "Vitamin D deficiency is probably the most common nutritional and medical condition in the world, affecting more than 50 per cent of the world's population"(Holick *et al*., 2011). Unfortunately, sourcing adequate vitamin D from diet has been difficult as there is no known source except from exposure to sunlight (Jones, 2015). Today's lifestyles are such that many people spend much of the day indoors and sensibly, to reduce the risk of

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developing skin cancer, they cover up and use sunscreen when they go outside. Consequently, many people do not get sufficient exposure to sunlight. Therefore, a dietary source of vitamin D is needed to ensure adequate status for many people. Different agencies recommended dietary intake of vitamin D ranging between 5 to 20 µg/day (200-800 IU) for different countries such as Australia and New Zealand, USA, European Food Safety Authority, Canada and United Kingdom. As per one study, average 90% of the population has insufficient intake of Vitamin D in India(Gupta and Gupta, 2014). Very few foods contain significant amounts of vitamin D. Vitamin D is found in small quantities in a few foods such as fatty fish, Liver, eggs and fortified foods such as margarine and low-fat milk.

Mushrooms exposed to sunlight or UV radiation are an excellent source of dietary vitamin D2 (Katherine and Rasor, 2013; Szabó *et al*., 2012). The mushroom contains fats in the form of ergosterol (Pro-vitamin D2), gets converted to pre-vitamin D2 on UV exposure and rapidly isomerizes to ergocalciferol (Vitamin D2) in a similar manner that pre-vitamin D3 isomerizes to vitamin D3 (Cholecalciferol) in human skin(Cardwell *et al*., 2018). Mushrooms also contains pro-vitamin D4 (22, 23-dihydroergosterol), which gets converted to vitamin D4 (22-dihydroergocalciferol) on UV exposure. Presence of both Vitamin D2 and D4 makes mushroom a rich source of vitamin D (Phillips *et al*., 2012). However, the physiological functions of Vitamin D4 are not well studied. Excess exposure or pulsed UV irradiation dose results in the production of lumisterol-2 and tachysterol-2 (Keegan *et al*., 2013; Krings and Berger, 2014). The effect of these photoproducts on human health is still unknown (Kalaras *et al*., 2012).

Different workers have reported different effective wavelengths for maximum production of vitamin D2 in mushrooms such as UV-B (280–315 nm) (Jasinghe and Perera, 2006) and UV-C (<280 nm) radiations (Guan *et al*., 2016; Jeng-Leun Mau *et al*., 1998; Koyyalamudi *et al*., 2009; Teichmann *et al*., 2007) while some has used UV-A (315–400 nm) also (Jasinghe *et al*., 2006). Duration of irradiation was reported as a critical factor in vitamin D accumulation potential such as 90 min exposure to UV-B radiation was reported as optimal conditions for generating vitamin D (Wu and Ahn, 2014). Also, different mushroom has been reported to accumulate different quantity of Vitamin D for example oyster mushrooms generated more than twice the vitamin D2 than shiitake (Jasinghe and Perera, 2006). Varying Vitamin D accumulation potentials was also reported by various workers (Huang *et al*., 2015; P Urbain *et al*., 2011).

Seeing the status of vitamin D in human populations and its health impact, the study was planned to assess the vitamin D accumulation potential of different mushroom types, optimum frequency of ultra violet light and its duration. Although many studies have been undertaken on the Vitamin D accumulation by mushrooms after UV irradiation but only a few was made to see vitamin D accumulation potential of different mushroom strains and the changes in nutritional composition of mushroom after UV exposure. Thus, the study was also undertaken to see the effect of UV exposure on vitamin D accumulation of different mushroom strains and proximate composition of mushrooms.

MATERIALS AND METHODS

Germplasm used

A total 5 strains of *Agaricus bisporus*, one of *Lentinula edodes*, two of *Volvariella volvacea*, one each of *Calocybe indica* and *Hericium erinaceous,* and five *Pleurotus* species were taken for the study. All the cultures of different mushrooms were procured from ICAR-Directorate of Mushroom Research Culture bank. Names and accession numbers of the cultures are given in Table.1.

Table 1. Strains of different mushroom species used in the study along with their accession numbers

Mushroom	Species	Accession numbers	Mushroom	Strains/species	Accession numbers
Button mushroom	*Agaricus bisporus*	DMRA-146	Milky mushroom	*Calocybe indica*	DMRO-334
		DMRA-156	Lion's maine	*Hericium erinaceous*	DMRX-780
		DMRA-097	Oyster mushroom	*P. sajor-caju*	DMRP-112
		DMRA-147		*P. florida*	DMRP-136
		DMRA-119		*P. cornucopie*	DMRP-116
Shiitake mushroom	*Lentinula edodes*	DMRO-327		*P. djamore*	DMRP-205
Paddy straw	*Volvariella volvacea*	DMRO-463		*P. membranaceous*	DMRP-189
mushroom		DMRO-885			

Cultivation of mushrooms and UV treatment of fruit bodies

All the mushrooms strains/species were cultivated as per the standard practice. A total of 100 g mushroom was taken, cleaned and subjected to UV treatment. 8-Watt UV lamps were used in the study and kept at a constant distance of 30 cm from the samples to be exposed. A total of five treatments were given to fruiting bodies for a fixed period of time i.e. 120 minutes followed drying at a constant temperature of 50°C. The Treatments are defined in Table 2.

Duration of UV treatment

One strain/species of each mushroom type was selected with maximum vitamin D accumulation potential for further studies. Fruit bodies of the selected strains/species were subjected to the optimum UV irradiation identified in the previous experiment and the duration of the irradiation were varied from 60 minute to 180 minutes followed drying at a constant temperature of 50°C and kept for analysis.

Vitamin D extraction and estimation

Vitamin D extraction was done using Deep eutectic solvent (DES) following a previously established protocol by (Patil *et al*., 2018). Estimation was done following a procedure by (Kumar and Rajput, 2011) with minor changes.

Preparation of Deep eutectic solvent (DES) - Choline chloride + glycerol (1:3) were mixed on a magnetic stirrer at 200 rpm, for 2 h, at 50°C, until a clear and homogeneous liquid was formed.

Standard preparation — 25 mg ergocalciferol (vitamin D2) (HiMedia chemicals) was weighed accurately and taken in 25 ml volumetric flask with n-hexane. The solution was diluted so as to get different concentration. Absorbance of each dilution

Table 2. Treatments used in the study

T-1-1	UV 254 (Whole fruit body)	T-3-2	UV 365 (Sliced fruit body)
T-1-2	UV 254 (Sliced fruit body)	T-4-1	UV-290 (Whole fruit body)
T-2-1	UV 310 (Whole fruit body)	T-4-2	UV-290 (Sliced fruit body)
T-2-2	UV 310 (Sliced fruit body)	T-5	Control
T-3-1	UV 365 (Whole fruit body)		

was recorded at 264.5 nm against reagent blank and a standard graph was plotted taking concentration on X axis and absorbance on Y axis.

Extraction from samples – One-gram lyophilized sample was extracted with 20 mL of DES for 30 min in ultrasonic bath at 65Hz. The extract was centrifuged at 8000 rpm for 10 min at 25 °C to remove the debris and to collect the supernatant. Supernatant was added with 25 ml acetonitrile and mixed together vigorously in a separating funnel. The formation of two immiscible layers of DES and acetonitrile led to easier separation of solute from DES to the acetonitrile layer (organic layer). The acetonitrile layer was separated.

Purification of Vitamin D2 (Ergocalciferol) - A volume of 25 mL of boiling hot water was added dropwise along the walls of the beaker containing 25 mL of acetonitrile extract. The solution was stirred on a magnetic stirrer at 300 rpm until the solution attained room temperature (25 °C). Now 25 mL of n-hexane was added to the resulting aqueous layer, and the mixture was stirred at 200 rpm for 5 min again. Resulting hexane layer was separated using a separating funnel (250 mL). Hexane layer was washed thrice with water and then treated with 1 g of anhydrous calcium carbonate to remove the excess water. 10 mL of 50% KOH solution in acetonitrile was added and refluxed at 50°C for 1 h to facilitate saponification. The solution was further filtered using Whatman filter paper (11 ìm pore size) to remove the salt. Further, the filtrate was dried by evaporating n-hexane under reduced pressure on a rotary evaporator. The dried extract was dilute with 5 mL of acetonitrile.

Estimation of Ergocalciferol - The solution was filtered through 0.2 ìm syringe filter and absorption was measured using UV-Vis spectrophotometer (Lamda 25, Perkin Elmer) at 264.5 nm. The absorbance is plotted against a standard curve of pure ergocalciferol and converted to vitamin D using a regression formula Y = 0.013X.

Protein extraction and estimation

Protein was extracted following (Pandey and Budhathoki, 2007). One-gram mushroom powder was dissolved in 50 ml phosphate buffer (50mM, pH 7.6) and homogenized properly. The homogenate was centrifuged at 8000 rpm for 10-15 min and supernatant was collected and stored at 4ºC. The supernatant was then estimated by Bradford method (Bradford, 1976). After extraction, 30µl of different mushroom samples were taken out in separate tubes and were mixed with 70µl of distilled water separately. In all of these separate sample tubes 2.9 ml of Coosmassic Brilliant Blue solution was then added and mixed thoroughly. All these tubes were incubated for 5 minutes at room temperature and absorbance at 595 nm was recorded against the reagent blank. A standard curve of Absorbance 595 nm versus Concentration (µg) of protein was plotted using BSA protein as standard.

Fat extraction and estimation

One gram of powdered sample was extracted with petroleum ether by using Soxhlet extractor for four hours. The extract was evaporated to dryness in a weighed flask using a vacuum evaporator. The weighed flask was dried in the oven at 80ºC for two hours, allowed to cool and reweighed. The difference between the initial and final weights was regarded as the lipid content of the sample (Parent and Thoen, 1977).

Carbohydrate extraction and estimation

One gram of powdered sample was extracted with 80% ethanol by using Soxhlet extractor for six hours. Total sugar content was estimated by phenol sulphuric acid method (DuBois *et al*., 1956). Suitable diluted sugar solution (1ml) is pipette directly into

bottom of a test tube. Phenol reagent (1 ml) is added in same way followed by 5 ml concentrated sulphuric acid pipetted directly onto sugar solution with fast flow pipette. The solution is mixed immediately and allowed to cool before absorbance is read at 490nm. The total carbohydrate was estimated using sugar standard curve.

Determination of total ash

Total ash was assayed following (Manzi *et al*., 2001). One gram of the sample was weighed accurately into a crucible. The crucible was placed on a clay pipe triangle and heated first over a low flame till all the material was completely charred, followed by heating in a muffle furnace for about 5~6 hours 550 °C. It was then cooled in desiccators and weighed. To ensure completion of ashing, the crucible was then heated in the muffle furnace for 1 hour, cooled and weighed. This was repeated till two consecutive weights were the same and the ash was almost white or grayish white in color.

Statistical analysis

All the experiments were replicated three times to reduce the experimental error. The data shown in the table is statistical mean of the three replications. Statistical analysis was done on completely randomized design (CRD) using one-way ANOVA to calculate standard error and the critical difference (CD). The data presented in the table contains standard error as ± and the critical difference (CD at 0.05).

RESULTS

Fruit bodies of different strains were irradiated to UV radiation of four different wavelengths i.e. 254, 290, 310 and 365 nm. UV irradiations were applied on whole and sliced (5 mm thickness) fruit bodies for a fixed duration of 120 minutes to observe the maximum vitamin D accumulation potential of the strain/mushroom type/species. The results indicated that different mushroom strains/species behaved

Table 3. Increase in Vitamin D content of different mushroom strains/species upon varying wavelength of UV irradiation

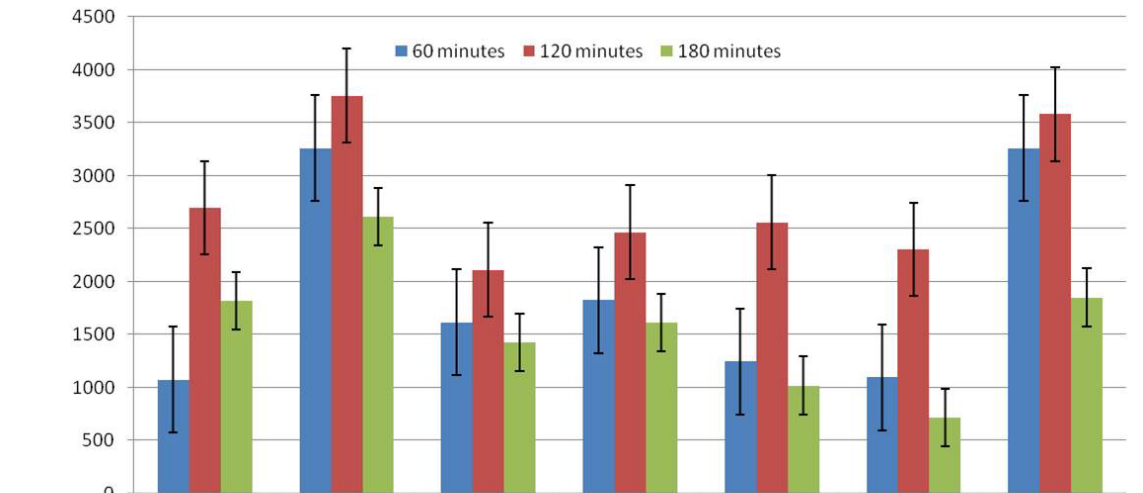
Mushroom strains	Vitamin D2 on UV irradiation (µg/g dry weight)										Control	CD (0.05)
	254 nm		290 nm		310 nm		365 nm		Sun light			
	A	B	A	B	A	B	A	B	A	B		
A. bisporus												
DMRA-146	60.1	31.2	64.6	35.0	55.3	24.8	32.5	24.5	47.6	29.9	18.1	5.7
DMRA-156	26.7	19.7	29.2	25.0	27.5	23.2	20.7	16.7	22.8	13.3	10.6	2.2
DMRA-097	41.3	9.5	51.5	29.6	29.9	25.3	22.8	13.3	35.3	14.1	15.5	4.7
DMRA-147	58.1	26.2	64.5	35.0	15.7	12.0	30.2	18.5	60.1	31.2	18.1	6.9
DMRA-119	42.5	22.6	54.7	30.7	30.9	23.4	31.5	17.9	38.6	31.6	17.1	4.3
L. edodes	57.3	52.8	94.5	62.6	91.4	56.6	44.4	40.9	72.8	47.1	45.1	6.7
V. volvacea												
DMRO-463	34.5	28.5	54.4	45.2	27.0	50.6	45.9	34.2	43.7	42.8	29.3	3.4
DMRO-885	42.1	35.7	55.8	48.4	52.1	49.2	40.2	31.6	42.4	40.7	25.1	3.3
C. indica	80.5	47.0	58.9	39.1	50.3	25.5	50.7	40.4	69.9	56.9	19.0	6.5
H. erinaceous	71.4	65.5	84.1	65.7	92.1	83.9	66.8	48.5	82.6	70.1	51.1	5.0
P. sajor-caju	40.9	25.0	72.3	20.4	38.9	29.5	48.9	26.6	43.9	42.7	19.0	5.6
P. florida	29.9	27.0	38.7	31.5	34.7	32.1	27.9	10.1	32.6	27.2	31.5	2.6
P. cornucopie	33.1	30.8	52.6	51.2	39.3	26.7	17.7	18.2	17.4	18.4	32.1	4.7
P. djamore	62.0	36.5	67.3	30.4	41.1	30.1	55.8	43.3	69.4	60.5	16.8	6.3
P. membranaceous	61.3	19.3	42.1	22.6	28.5	27.0	27.6	30.1	30.7	35.8	34.3	4.1

A – Whole fruit body; B – sliced fruit body

differently with different UV irradiation. It could be observed from the results that all the strains/species have shown significant increase in Vitamin D concentration on UV irradiation irrespective of the wavelength used. Maximum increase in vitamin D concentration in all the treatments was observed upon UV irradiation by 290 nm wavelength, which falls under UV-B range. It was also noticed that UV-C range (254 nm) also had at par results with UV-B range (290 nm) (Table 3). During the study, we have used whole and sliced mushroom for irradiation and significantly higher increase in vitamin D content was recorded in whole fruit body of mushroom in comparison to sliced fruit body. Out of all the strains/ mushroom tested, *H. erinaceous* and *L. edodes* showed the maximum vitamin D accumulation potential. On the basis of higher vitamin D content upon UV irradiation, DMRA-146 strain of *A. bisporus*, DMRO-327 of *L. edodes*, DMRO-885 of *V. volvacea*, DMRO-334 of *C. indica*, DMRX-780 of *H. erinaceous*, DMRP-205 of *P. djamore* and DMRP-112 of *P. sajor caju* were selected for further experimentation.

To optimize the duration of the exposure, one strain/species of each mushroom type was selected with maximum vitamin D accumulation potential. The whole fruiting body of the selected strains/species was subjected to 290 nm UV irradiation identified in the previous experiment and the duration of the irradiation was varied from 60, 120 and 180 minutes. After UV exposure, the fruit bodies were dried and analyzed for Vitamin D content. The results indicated both one- and two-hours treatments resulted in increase in Vitamin D content. However, maximum increase was observed in 2 h exposure whereas more than 2 h exposure resulted in decrease in vitamin D content (Fig 1). During the experiment also, *H. erinaceous* and *L. edodes* have shown the maximum Vitamin D content. Since all the mushroom have shown maximum vitamin D accumulation upon 2 h UV exposure, we have taken 2 h UV irradiated fruit bodies for analysis of protein, carbohydrate, fat and ash content against non-UV irradiated fruit bodies.

Although many studies have been undertaken on the Vitamin D accumulation by mushrooms after UV



	A. bisporus (DMRA-146)	L. edodes (DMRO-327)	V. volvacea (DMRO-885)	C. indica (DMRO-334)	P. djamore (DMRP-205)	P. sajor caju (DMRP-112)	H. erinaceous (DMRX-780)
60 minutes	1074.45	3260.12	1618.26	1826.24	1246.99	1097.81	3260.62
120 minutes	2694.89	3756.14	2112.64	2466.2	2560.62	2303.65	3580.7
180 minutes	1818.98	2612.23	1428.24	1616.25	1021.9	721.17	1852.28

Fig. 1. Effect of varied time interval of UV treatment (290 nm) on vitamin D content (IU) of selected mushroom

Table 4. Effect of UV treatment (290 nm) on proximate composition of selected mushrooms

Mushroom species	Proximate composition on fresh weight basis (Mean ± SE)							
	Protein (g/100 g)		Carbohydrate (g/100 g)		Fats (g/100 g)		Total Ash (g/100 g)	
	Untreated	Treated	Untreated	Treated	Untreated	Treated	Untreated	Treated
A. bisporus (DMRA-146)	3.26±0.23	3.33±0.23	3.96±0.82	4.12±0.82	0.43±0.10	0.24±0.10	4.46±0.12	4.21±0.13
L. edodes (DMRO-327)	1.53±0.21	1.62±0.21	6.94±1.62	7.62±1.62	0.48±0.13	0.23±0.13	3.33±0.13	3.06±0.14
V. volvacea (DMRO-885)	3.28±0.13	3.15±0.13	13.84±2.52	14.56±2.52	0.56±0.09	0.38±0.09	4.55±0.12	4.31±0.12
C. indica (DMRO-334)	1.78±0.22	1.65±0.22	18.10±2.12	19.23±2.12	0.90±0.14	0.63±0.14	2.56±0.13	2.30±0.13
P. djamore (DMRP-205)	2.24±0.25	2.02±0.25	4.12±0.92	5.48±0.92	0.91±0.09	0.72±0.09	2.8±0.21	3.21±0.21
P. sajor caju (DMRP-112)	2.65±0.19	2.75±0.19	9.36±1.26	9.96±1.26	0.81±0.03	0.55±0.03	4.5±0.20	4.11±0.20
H. erinaceous (DMRX-780)	3.25±0.15	3.14±0.15	2.05±1.24	3.36±1.24	1.20±0.20	0.80±0.20	3.95±0.18	4.30±0.18

irradiation but only a few studies have been made on the comparative vitamin D accumulation potential of different mushroom strains and changes in nutritional composition of mushroom after UV exposure. The study undertaken to see the effect of UV exposure on the proximate composition of different mushrooms revealed that there were no significant changes in the protein, carbohydrate and ash content of the mushrooms but it could be observed that fat content has been significantly reduced after the UV irradiation. It was also observed from the study that the mushroom accumulated more vitamin D upon UV irradiation showed more reduction in fat content in most of the cases (Table 4).

DISCUSSION

Human beings have the potential to maintain adequate vitamin D status through the regular exposure of bare skin to sunlight. However, modern lifestyles are such that many people spend much of the day indoors and sensibly, to reduce the risk of developing skin cancer, they cover up and use sunscreen when they go outside. Consequently, many people do not get sufficient exposure to sunlight. Therefore, a dietary source of vitamin D is needed to ensure adequate status of 25-hydroxyvitamin D (25(OH)D) for many people. Very few foods contain significant amounts of vitamin D such as fatty fish, liver, eggs and fortified foods such as margarine and low-fat milk (if fortified). It is estimated that 1 billion people worldwide are vitamin D deûcient (25(OH)D concentrations ≤50 nmol/L), with prevalence of excess of 50% being commonly reported in population-based studies.

Fungi and yeast contain dietary form of vitamin D i.e. D2 while Vitamin D3 is generally found in animal body. Lesser amounts of vitamin D3 and D4 are also found in fungi (Keegan *et al*., 2013; Phillips *et al*., 2012; Paul Urbain *et al*., 2016). During the past decade, the bio-efficacy of vitamin D2 and D3 has been questioned (Houghton and Vieth, 2006), and numerous studies were made to compare the effect of the two forms in human body. The studies were mostly based upon rise in plasma 25-OH-D levels

after vitamin D3 or vitamin D2 supplementation. Many studies have suggested that vitamin D2 is less effective in raising 25-OH-D levels compared to vitamin D3 (Leventis and Kiely, 2009; Romagnoli *et al*., 2008; Trang *et al*., 1998) whereas other studies could not validate these results and observed similar rises in 25-OH-D levels with both forms (Rapuri *et al*., 2004; Thacher *et al*., 2010). However, levels of plasma 25-OH-D are reported to be nonfunctional marker for determination of biological activity of vitamin D status. Despite their structural differences, in qualitative terms vitamins D2 and D3 exhibit identical biological responses in the body for 1á,25-(OH)2D33 (Haussler *et al*., 2011; Jones *et al*., 1998). Moreover, most of the steps involved in the metabolism and actions of vitamin D2 and D3 are proved to be identical through biochemical evidences. Also, speciûc signal transduction systems for vitamin D3, also reported to respond to physiological doses of vitamin D2 equally well (Roborgh and de Man, 1960; Romagnoli *et al*., 2008).

Mushrooms exposed to sunlight or UV radiation are an excellent source of dietary vitamin D2 (Katherine and Rasor, 2013; Szabó *et al*., 2012). Mushroom can carry out conversion of ergosterol in to Vitamin D2 even if they are exposed to normal sunlight as shown by researchers (Katherine and Rasor, 2013; Kristensen *et al*., 2012; Simon *et al*., 2011; Urbain and Jakobsen, 2015). During the present study, we have got the similar results and sunlight exposure of the mushroom has resulted in significant increase in vitamin D content of mushrooms. Unfortunately, mushrooms begin to shrivel and brown if they are left for extended periods in the sun. This brings down the market values of the product. However, when the mushrooms are exposed to ultraviolet light, it generates signiûcant amount of vitamin D and retains the good looks also (Jasinghe *et al*., 2006; Jeng-Leun Mau *et al*., 1998; Kalaras *et al*., 2012; Kamweru and Tindibale, 2016; Ko *et al*., 2008; Koyyalamudi *et al*., 2009, 2011; Simon *et al*., 2011; Urbain *et al*., 2016; Wittig *et al*., 2013). The present study suggested that the maximum accumulation of Vitamin D in different mushrooms takes place at a UV irradiation of 290 nm wavelength and this increase of vitamin D is recorded up to 300% in comparison to control. Earlier reports also suggested that mushrooms produce maximum vitamin D upon irradiation with ultraviolet B (between wavelengths of 290 nm to 315 nm) (Jasinghe *et al*., 2006). Some earlier reports suggested that sun-exposed sliced mushrooms produced more vitamin D than whole mushrooms from the same amount of UV radiation exposure (Katherine and Rasor, 2013; Urbain *et al*., 2016). In contrast, our studies on the whole and sliced mushroom showed that the increase of Vitamin D content was more in whole mushrooms.

In the present study, varied duration of UV exposure was used to see the effect on Vitamin D content of different mushroom and it was observed that the vitamin D content of mushroom increased up to 120 min UV exposure and then declined. Different researchers have reported the effect of varied duration of UV irradiation on the vitamin D content of mushrooms (Urbain and Jakobsen, 2015). They reported the increase of vitamin D content till 60 min exposure of sunlight. Other workers have also reported the increased vitamin D in sliced oyster mushrooms exposed for 90 min to UV-B lamp radiation (Jasinghe *et al*., 2006; Jasinghe and Perera, 2006; Krings and Berger, 2014a; Wittig *et al*., 2013; Wu and Ahn, 2014). An unpublished Australian study reported that sun exposure of small button mushrooms up to 120 minutes increased Vitamin D content in whole button mushrooms (personal communication, J. Ekman, Applied Horticultural Research, 12 August 2013).

Slawiñska *et al*., (2017) studied vitamin concentration in UV irradiated mushroom stored at

4°C for up to 10 days. During storage at 4°C, the amount of vitamin D2 was gradually decreased in *P. ostreatus* and *L. edodes*, whereas in *A. bisporus* vitamin D2 gradually increased until the sixth day, then decreased. The change in vitamin D concentration of mushrooms was also studied by (Bernaœ and Jaworska, 2017) and reported largest amount of vitamin D2 and ergosterol in canned mushrooms, whereas the smallest amount was retained in dried mushrooms after 12 months of storage. Moreover, cryogenically frozen mushroom showed higher levels of vitamin D2, whereas ergosterol levels were higher using air-blast freezing.

In the present study, we tried to see the effects of UV treatment on vitamin D status of different strains of different mushroom and on proteins, sugars, fats and other nutrient levels of mushrooms. The results obtained during the study showed maximum increase of vitamin D levels in shiitake (*Lentinula edodes*) and Lion's Mane (*Hericium erinaceous*) mushroom and but no significant changes in the protein, carbohydrate and ash content of the mushrooms could be observed. During the study, we could observe that fat content has been significantly reduced after the UV irradiation. This may be due to the conversion of ergosterol in to vitamin D2 (ergocalciferol).Banlangsawan and Sanoamuang (2016) also reported no significant change on proximate composition of mushroom if the mushroom fruit bodies are irradiated with UV-B or UV-C.

CONCLUSION

During the present study, it was observed that all the test species/strains of mushroom produced maximum vitamin-D2 when exposed to a UV irradiation in the range of UV-290 to UV-310 with a maximum increase of vitamin D levels in shiitake (*Lentinula edodes*) and Lion's Mane (*Hericium erinaceous*) mushroom. The vitamin D2 produced in mushroom can very well take care of vitamin D deficiency in human being as the physiological functions of vitamin-D2 and D3 are more or less similar and vitamin D2 can also be metabolized in human body in the similar process as Vitamin D3. Moreover, 120 min UV irradiation was proved to produce maximum amount of Vitamin D in mushroom. The UV irradiation are also reported to improve the shelf life in button mushroom. The study conducted on the effect of UV irradiation on proximate composition of mushroom showed that there is no significant deleterious effect on the nutritional properties of mushroom, hence UV irradiation (UV-290-UV310) after harvest for a period of 2 h can not only enhance the vitamin D status of mushroom but also will be helpful for enhancing the shelf life of mushroom.

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