



## Preliminary Phytochemical Profiles and Antioxidant Activities of Methanolic, Chloroform and Hexane Extracts of *Alpinia galangal* Rhizome and *Morshella esculenta* Mycelia

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**ABSTRACT:** *Alpinia galangal* and *Morshella esculenta* are ethnomedicinal plants due to their radical scavenging and gastronomic qualities. This prompted us to carry out this study about the antioxidant activity of plants. Extraction was carried out using solvents by maceration followed by phytochemical screening. Purification of crude was done by silica gel column chromatography. Total phenolic and flavonoid content (TPC) and (TFC) of crude and fractions were determined. The antioxidant activities were assessed by scavenging assays. TPC of methanolic extract of *A. galangal* was found highest with the value of 49.787±0.063 GAE mg/g and showed higher TFC with the value of 38.013±2.116 QCE mg/g. The highest H<sub>2</sub>O<sub>2</sub> activity was shown by fraction MEC5 (83.363%). The highest ABTS radical scavenging activity was shown by AGC (99.68%). However, our study faced different challenges such as differences between antioxidant activities, H<sub>2</sub>O<sub>2</sub> activity etc. In conclusion, three different solvent extracts of *A. galangal* and *M. esculenta* revealed the presence or absence of phytochemicals tested and varying degrees of in-vitro antioxidant activity.

**Keywords:** *Alpinia galangal*, *Morshella esculenta*, phytochemicals, antioxidant, hydrogen peroxide.

### INTRODUCTION

India is one of the largest producers of phytotherapeutics and acknowledged as the botanical garden of the world (Ranjan *et al.*, 2012). An antioxidant can be widely defined as any substance that defers or inhibits free radical-induced oxidative stress to a target molecule (Meena *et al.*, 2010). Phenolic compounds are stable radical intermediates, a marked source of natural antioxidants which are known to be produced in huge amounts by different plant parts such as the bark of stems, seeds and roots and leaves in grating environments with strong light intensities which majorly protects the plants against pathogenic agents (Fraikue, 2018). The molecular structure of flavonoids has been known to be responsible for their strong antioxidant activity at very low concentrations (Pawar *et al.*, 2022). *A. galangal* Wild. (Fam: Zingiberaceae) is an aromatic perennial rhizomatous herb used since Ayurveda in the name of sugandhamula, rasna, kulanjan and greater galangal and the genus is the most marked and ubiquitous in the Zingiberaceae family that provides many useful products for food, condiments, dyes, perfume and aesthetics. It is extensively found in western ghats, Malabar coasts and Gujarat in India and produced by other Asian countries Thailand, China and Malaysia (Tushar *et al.*, 2010; Unnisa and Parveen 2011; Avasthi *et al.*, 2015).

The rhizome of *A. galangal* has therapeutic value and is extensively used as an antioxidant, antiaging, gastroprotective, anti-fungal, anti-helminthic, anti-diuretic, anti-diabetic, anti-dementia, aphrodisiac, antimicrobial and antitumor (Chouni and Paul 2018; Oonmetta-aree *et al.*, 2006; Eram *et al.*, 2019); Chudiwal *et al.*, 2010; Chompoo *et al.*, 2012). *A. galangal* rhizome extracts in different solvents have been identified earlier as potent in-vitro antioxidants reservoirs by studying their TPC, TFC and antioxidant activity using ABTS, Ferric Antioxidant Power (FRAP) and 2,2-diphenyl-1-picrylhydrazyl (DPPH) methods by different extraction methods (Malik *et al.*, 2016; Phucho *et al.*, 2017; Devi and Devi 2018). Our study is the first study to report TFC and antioxidant activity using ABTS and H<sub>2</sub>O<sub>2</sub> methods using hexane solvent by maceration whereas total phenol, flavonoid content and antioxidant activity using H<sub>2</sub>O<sub>2</sub> method using chloroform solvent in rhizome extracts of *A. galangal*. *M. esculenta* (Pezizales, Ascomycota) is one of the costliest and predominantly found edible morel species in the North-West Himalayas mainly, Kashmir and is renowned for their gastronomic quality due to their aroma, extraordinary flavour and meaty texture, indigenous and nutritional health benefits as a functional food due to their anti-oxidative, anti-inflammatory, and immunostimulatory properties, and high levels of protein, fibre and minerals (Raman *et al.*, 2018; Badshah *et al.*, 2021; Thakur *et al.*, 2021;

Cheung *et al.*, 2003; Kalač, 2013). Morels are garnered in the wild majorly in India, China, Turkey, the USA and Mexico where they grow abundantly (Winder 2006; Singh *et al.*, 2022). Earlier, commercial cultivation of morels was confined to Israel, the USA and China. Recently, commercial mushroom cultivation has started in India wherein states such as Himachal Pradesh, Punjab, Uttarakhand, Haryana, Uttar Pradesh, Tamil Nadu, Maharashtra, Karnataka, and Telangana are the major mushroom-growing states at the trade level. Very recently, Morshella was cultivated artificially by the Indian Council of Agriculture Research-run-Directorate of Mushroom Research (DMR) which is a major milestone in the Indian history of mushroom production. Previous studies reported TPC, TFC and antioxidant activities of *M. esculenta* using ABTS, DPPH, reducing power ability etc. in different solvents but are limited (Mau *et al.*, 2004; Nitha *et al.*, 2010; Akyüz *et al.*, 2019). Our study first reports TPC, TFC and antioxidant activities of *M. esculenta* using ABTS and H<sub>2</sub>O<sub>2</sub> methods in hexane and chloroform extracts. The present study evaluates the quantitative phytochemicals, TPC, TFC and in-vitro antioxidant activity of methanolic, chloroform and hexane extracts of *A. galangal* and *M. esculenta*; so that their uninvestigated antioxidant potential can further be exploited.

## MATERIAL AND METHODS

**Sample collection.** The investigation was carried out in the research institute, Allele Life Sciences (P) Ltd. of Uttar Pradesh, India. *A. galangal* rhizome was collected from Karnataka (India) and *M. esculenta* mycelia were collected from Uttarakhand (India).

**Preparation of extracts.** Samples were thoroughly cleaned with distilled water to remove the dust particles present in them and air dried in the shade to remove moisture. Following it, the dried samples were grounded into a coarse powder using a mortar and pestle. For methanolic extraction, 10 g powder of both plant samples was mixed with 40 ml solvent whereas for hexane and chloroform extraction 5 g powder of both plants was mixed with 20 ml solvent to maintain a minimum concentration of 250 mg/ml followed by maceration with occasional shaking for 5 days at room temperature with varying polarities such as methanol, hexane and chloroform and homogenized separately for each extract. The extracts were filtered through Whatman No. 1 filter paper and evaporated using rotavapor at low temperature and pressure by a rotary evaporator to obtain the crude extracts. All the extracts were preserved at 4°C for further analysis.

**Determination of extractive value.** The obtained crude extracts were concentrated to complete dryness by keeping filtrate for complete evaporation of the solvent. The percentage extractive values were calculated by using the following formula and recorded (Pawar and Jadhav 2016):

$$\text{Extractive value(\%)} = \frac{\text{Weight of dried extract}}{\text{Weight of plant material}} \times 100 \quad (1)$$

**Morphological characterization of rhizome extracts of *A. galangal*.** The colour and consistency of rhizome extracts of *A. galangal* in each solvent were also recorded by observations.

**Phytochemical screening.** Crude extracts of *A. galangal* and *M. esculenta* in hexane, chloroform and methanol were subjected to different phytochemical tests for the detection of bioactive chemical constituents such as alkaloids, phenolics, tannins, flavonoids, carbohydrates, saponins and proteins using standard procedures (Angle and Vernekar 2022; Talnia and Trehan 2022).

**Total phenolic content.** TPC of *A. galangal* and *M. esculenta* crude extracts in each solvent were determined by the FC method through some modifications (Samyudurai and Saradha 2016; Sheela and Cheenickal 2017). FC reagent was freshly prepared (diluted 1:1 with de-ionized water) before use. Plant extracts (0.025 mL) were mixed with FC reagent (2.5 ml) and then allowed to stand for 5 min at room temperature to allow a complete reaction with the reagent. The reaction mixture was further neutralized with sodium carbonate solution (2 ml, 7.5%, w/v). It was kept in the dark at room temperature for 30 min with intermittent shaking for blue colour development. After 30 min, the absorbance was read at 765 nm UV-visible spectrophotometer (Thermo Spectronic) against a blank solution containing all the reagents except the FC reagent which is replaced by an equal volume of distilled water. All the measurements were carried out in triplicates and averaged. Methanolic solution of gallic acid served as standard (1 mg/ml) phenolic compound (0.2, 1, 2, 3, 4, and 5 µg/mL) for plotting the calibration curve. TPC in the extracts was expressed as Gallic acid equivalent (GAE) (mg of Gallic acid equivalent/g of dry weight sample) and was calculated by the formula (Barku *et al.*, 2013):

$$T = \frac{C \times V}{M} \quad (2)$$

T= Total content of phenolic/g of plant extract, in GAE;  
C= Concentration of Gallic acid established from the regression curve, microgram/ml (µg/ml);

V= Volume of methanolic plant extract; M=Weight of methanolic plant extract, g.

**Total flavonoid content.** The TFC of *A. galangal* and *M. esculenta* crude extracts in each solvent was determined through some modifications (Kavitha *et al.*, 2018). Plants extracts (0.5 ml) were separately heterogeneously mixed with methanol (1.5 ml), potassium acetate (0.1 ml, 1M), aluminium chloride (0.1 ml, 10% w/v), and distilled water (2.8 ml). Furthermore, the mixture was incubated for 30 min at room temperature. After 30 min, using a UV-visible spectrophotometer (Thermo Spectronic) the absorbance was measured at 415 nm against a blank solution containing all the reagents except aluminium chloride which is replaced by an equal volume of distilled water. All the measurements were carried out in triplicates and averaged. Methanolic solution of quercetin from 20 to 100 µg/mL served as a standard (1 mg/ml) flavonoid compound to obtain the calibration curve. TFC was

expressed as quercetin equivalent (QCE) (mg of quercetin equivalent/g of dry weight sample) and was calculated by the formula mentioned earlier for TPC [Eq. 2]

T= Total content of flavonoid /g of plant extract, in QCE;

C= Concentration of quercetin established from the regression curve, ( $\mu\text{g/ml}$ );

V=Volume of methanolic plant extract; M= Weight of methanolic plant extract, g.

**ABTS radical scavenging assay.** The radical scavenging activity of *A. galangal* and *M. esculenta* crude extracts in each solvent were assessed by ABTS (Rangasamy *et al.*, 2019) through some modifications. ABTS aqueous stock solution (7 mM) and potassium persulfate stock solution (2.45 mM) were freshly prepared. Equal volumes of the stock solutions were mixed and allowed to react in the dark at room temperature for 12-16 h to obtain ABTS<sup>+</sup> free radical. After 12-16 h, ABTS<sup>+</sup> working solution was diluted with methanol to attain an absorbance of  $0.70 \pm 0.02$  at 734 nm. Briefly, each methanolic extract (10  $\mu\text{l}$ ) was mixed with a working solution of ABTS<sup>+</sup> (2 ml) and absorbance was taken 1 min after initial mixing using a UV-visible spectrophotometer (Thermo Spectronic). All the measurements were carried out in triplicates and averaged. The total antioxidant capacity (TAC) was calculated as per cent inhibition of ABTS radical using the following equation,

$$\text{Percentage inhibition of ABTS (\%)} = \left( \frac{C_{\text{abs}} - S_{\text{abs}}}{C_{\text{abs}}} \right) \times 100 \quad (3)$$

where  $C_{\text{abs}}$  is the control absorbance (ABTS radical + methanol);  $S_{\text{abs}}$  is the sample absorbance (ABTS radical + sample extract).

**H<sub>2</sub>O<sub>2</sub> radical scavenging assay.** The H<sub>2</sub>O<sub>2</sub> scavenging capacity *A. galangal* and *M. esculenta* crude extracts in each solvent were evaluated through some modifications (Oktay *et al.*, 2003). A solution of H<sub>2</sub>O<sub>2</sub> (2m mol/L) was prepared in a standard phosphate buffer solution (1X, pH 7.4). H<sub>2</sub>O<sub>2</sub> concentration was determined spectrophotometrically from absorption at 230 nm. Crude extracts in distilled water were added to a hydrogen peroxide solution (0.6 ml). The absorbance of H<sub>2</sub>O<sub>2</sub> at 230 nm was noted after 2 min against a blank solution of phosphate buffer without H<sub>2</sub>O<sub>2</sub>. The amount of H<sub>2</sub>O<sub>2</sub> radical scavenged by each extract was calculated as per cent inhibition of H<sub>2</sub>O<sub>2</sub> radical using the following equation,

$$\text{Percentage inhibition of H}_2\text{O}_2 (\%) = \left( \frac{C_{\text{abs}} - S_{\text{abs}}}{C_{\text{abs}}} \right) \times 100 \quad (4)$$

where  $C_{\text{abs}}$  is the control absorbance (Ascorbic acid +H<sub>2</sub>O<sub>2</sub> radical);

$S_{\text{abs}}$  is the sample absorbance (H<sub>2</sub>O<sub>2</sub> radical+ sample extract).

**Column chromatography of *A. galangal* and *M. esculenta* crude extracts.** Each crude extract in each extraction solvent of both the plant samples was subjected to silica gel column chromatography for the separation, identification and purification of phytoconstituents through some modifications (Gini and Jothi 2018). A vertical glass column (length- 60 cm, diameter-1.5 cm) made of borosilicate material was used for the fractionation. The column was rinsed well with methanol and was completely dried before packing. A small piece of cotton wool was attached at the column bottom using a glass rod. Sea sand (about 60-120 particlesin size) was transferred on top of the cotton wool at a height of 1cm. The particles were rinsed down using the solvent. Hexane was introduced into the column up to the 3/4<sup>th</sup> level by closing the stopcock. 15 g of silica gel (100-200 mesh size) was used as the packing material. Silica slurry was prepared with hexane and poured from the top of the column around 2/3<sup>rd</sup> of the column with continuous draining of the solvent to ensure proper packing of the column. A small amount of sea sand was added above the silica slurry about 1cm height and the sea sand particles were also rinsed down by the solvent. 5 ml of extracts was introduced at the top of the sand gradually with the help of a funnel. The solvent level was maintained at 4 cm height from the above sample, preventing drying of the glass column. The isocratic elution method was followed to separate fractions from extracts by using solvents from low polarity to high polarity (i.e., hexane to methanol). The flow rate was adjusted to 60 drops/min. The fractions collected were tested for two phytochemicals namely, phenols and flavonoids and selected fractions were tested further for TPC, TFC, ABTS and H<sub>2</sub>O<sub>2</sub> antioxidant assays.

## RESULTS AND DISCUSSION

**Extractive value of plant materials.** The percentage (%) methanol soluble extractive value, chloroform soluble extractive value and hexane soluble extractive value of *A. galangal* extract was found to be 60, 45, and 34 respectively, while *M. esculenta* extract 65, 47, and 32 respectively.

**Morphological characterization of rhizome extracts of *A. galangal*.** Morphological characterization of rhizome extracts of *A. galangal* revealed a brown to brownish yellow colour of extracts with semisolid consistency (Table 1).

**Phytochemical screening of crude extracts.** The phytochemical study of *A. galangal* revealed the presence of alkaloids, tannins, flavonoids, phenolics, carbohydrates, proteins and saponins, while the phytochemical study of *M. esculenta* revealed the presence of alkaloids, tannins, flavonoids, phenolics and proteins except, carbohydrates and saponins (Table 2).

**Table 1: Morphological characterization of rhizome extracts of *A. galangal*.**

Extracts	Colour	Consistency
Methanol	Brownish-yellow	Semi-solid
Chloroform	Dark brown	Semi-solid
Hexane	Brown	Semi-solid

**Table 2: Phytochemical tests of *A. galangal* and *M. esculenta* in different solvents.**

Phytochemicals	Tests	Extracts	<i>A. galangal</i>	<i>M. esculenta</i>
Alkaloids	Wagner's	Methanol	++	++
		Chloroform	+++	+++
		Hexane	+++	+++
	Dragendorff's	Methanol	++	+
		Chloroform	+++	+++
		Hexane	+++	+++
Phenols	Ferric Chloride	Methanol	+	+
		Chloroform	-	-
		Hexane	-	-
Tannins	Ferric Chloride	Methanol	+	-
		Chloroform	+	+
		Hexane	+	+
Flavonoids	Alkaline Reagent	Methanol	+	+
		Chloroform	+	+
		Hexane	+	+
Carbohydrates	Benedict's Reagent	Methanol	-	-
		Chloroform	-	-
		Hexane	-	-
	Barford's Reagent	Methanol	-	-
		Chloroform	-	-
		Hexane	-	-
	Fehling's Reagent	Methanol	+	-
		Chloroform	-	-
		Hexane	-	-
Saponin	Foam	Methanol	++	-
		Chloroform	-	-
		Hexane	-	-
Proteins	Biuret	Methanol	-	-
		Chloroform	-	-
		Hexane	-	-
	Xanthoproteic	Methanol	+++	-
		Chloroform	-	-
		Hexane	-	-
	Ninhydrin	Methanol	-	-
		Chloroform	+++	+++
		Hexane	-	-

+: present in little concentration, ++: present in modest concentration, +++: present in high concentration, -: absent

**Total phenolic content.** TPC of crude extracts and selected fractions were calculated using the following linear regression equation obtained from the standard plot  $y = 0.4152x + 0.446$ ,  $R^2 = 0.9906$ , where  $y$  is the absorbance and  $x$  is the quantity of gallic acid in  $\mu\text{g}$  (Fig. 1). TPC of the methanolic extract of *A. galangal* and *M. esculenta* were  $49.787 \pm 0.063$  and  $6.180 \pm 0.083$ , GAE mg/g respectively. TPC of the hexane and chloroform extracts of *A. galangal* and *M. esculenta* were  $1.805 \pm 0.008$ ,  $2.17 \pm 0.003$ ,  $0.845 \pm 0.003$  and  $0.514 \pm 0.003$ , GAE mg/g respectively.

**Total flavonoid content.** TFC of crude extracts and selected fractions were calculated using the following linear regression equation obtained from the standard plot  $y = 0.0184x + 0.118$ ,  $R^2 = 0.9944$ , where  $y$  is the absorbance and  $x$  is the quantity of quercetin in  $\mu\text{g}$  (Fig. 2). TFC of methanolic, chloroform and hexane extracts of *A. galangal* and *M. esculenta* were  $38.013 \pm 2.116$ ,  $0.141 \pm 7.5E-05$ ,  $0.360 \pm 0.000$ ,  $1.573 \pm 0.557$ ,  $0.108 \pm 0.000$  and  $0.336 \pm 0.000$ , QCE mg/g respectively.

**Column chromatography of crude extracts.** 10 fractions (8ml) of each solvent crude extract of *A. galangal* and *M. esculenta* was collected using solvents from low polarity to high polarity (i.e., hexane to methanol).

**Phytochemical screening of column fractions.** The column fractions of *A. galangal* and *M. esculenta* were

subjected to qualitative phenolic and flavonoid tests and only those fractions were selected which gave positive results. The selected fractions were: *A. galangal* methanolic extracts (AGM1, AGM2, AGM3, AGM4, AGM5) showed positive results for flavonoids and phenols, *A. galangal* hexane extract (AGH1) showed positive for flavonoids, *A. galangal* chloroform extract (AGC1) showed positive for flavonoids, *M. esculenta* methanolic fractions (MEM1, MEM2, MEM3, MEM4, MEM5) tested positive for both phenols and flavonoids, *M. esculenta* hexane extract (MEH1, MEH2) tested positive for flavonoids and *M. esculenta* chloroform fractions (MEC5 and MEC6) tested positive for flavonoids and phenols.

**Total phenolic content of fractions.** TPC of AGM1, AGM2, AGM3, AGM4, AGM5, MEM1, MEM2, MEM3, MEM4, MEM5 and MEC6 were  $0.442 \pm 0.001$ ,  $0.189 \pm 0.000$ ,  $0.350 \pm 0.001$ ,  $0.193 \pm 0.039$ ,  $0.300 \pm 0.001$ ,  $0.1375 \pm 0.001$ ,  $0.324 \pm 0.115$ ,  $0.185 \pm 0.003$ ,  $0.151 \pm 0.001$ ,  $0.153 \pm 0.002$ , and  $3.620 \pm 0.001$ , GAE mg/g respectively.

**Total flavonoid content of fractions.** TFC of AGM1, AGM2, AGM3, AGM4, AGM5, AGC1, AGH1, MEM1, MEM2, MEM3, MEM4, MEM5, MEH1, MEH2 and MEC5 were  $0.075 \pm 5.23E-05$ ,  $0.127 \pm 6.03E-05$ ,  $0.050 \pm 0.000$ ,  $0.050 \pm 5.23E-05$ ,  $0.062 \pm 3.02E-05$ ,  $0.097 \pm 5.23E-05$ ,  $0.051 \pm 0.000$ ,  $0.041 \pm 3.02E-05$ ,  $0.043 \pm 6.03E-05$ ,  $0.042 \pm 7.98E-05$ ,  $0.042 \pm 0.000$ ,

0.042±0.000, 0.051±3.02E-05, 0.120±0.000, and 0.096±3.02E-05, QCE mg/g respectively.

**ABTS assay of crude extracts and fractions.**

ABTS% radical scavenging activity of methanolic, chloroform and hexane extracts of *A. galangal* (AGM, AGC and AGH) and *M. esculenta* (MEM, MEC and MEH) were 85.289±0.196, 99.686±0.127, 4.960±0.149, 33.775±0.200, 2.872±0.146, and 3.863±1.585 respectively. ABTS% radical scavenging activity of fractions: AGM1, AGM2, AGM3, AGM4, AGM5, AGC1, AGH1, MEM1, MEM2, MEM3, MEM4, MEM5, MEH1, MEH2, MEC5 and MEC6 were 95.869±0.303, 81.599±0.176, 97.908±0.299, 74.280±0.491, 59.905±0.102, 37.963±0.135, 23.811±0.213, 2.717±0.143, 1.881±0.125, 11.604±0.233, 9.984±0.330, 0.575±0.322, 65.274±0.241, 14.830±0.331, 29.086±0.192, and 15.091±0.080 respectively.

**H<sub>2</sub>O<sub>2</sub> assay of crude extracts and fractions.**

H<sub>2</sub>O<sub>2</sub>% radical scavenging activity of methanolic, chloroform and hexane extracts of *A. galangal* and *M. esculenta* were 69.904±1.713, 3.915±3.141, 9.542±3.782, 44.852±2.669, 67.595±2.370, and 17.664±2.910 respectively. H<sub>2</sub>O<sub>2</sub>% scavenging activity of fractions: AGM1, AGM2, AGM3, AGM4, AGM5, AGC1, AGH1, MEM1, MEM2, MEM3, MEM4, MEM5, MEH1, MEH2, MEC5 and MEC6 were 79.676±3.341, 74.231±2.403, 72.714±4.805, 81.975±2.049, 76.530±4.882, 42.164±2.002, 24.195±3.148, 42.110±3.718, 42.404±2.489, 58.800±2.499, 43.258±4.430, 56.078±4.327, 42.164±2.002, 70.360±2.617, 83.363±0.507 and 82.030±1.454 respectively.

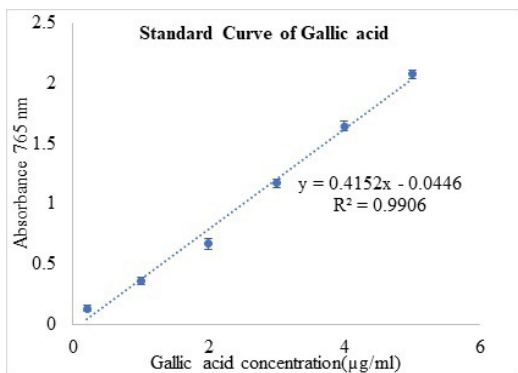


Fig. 1. Gallic acid regression curve.

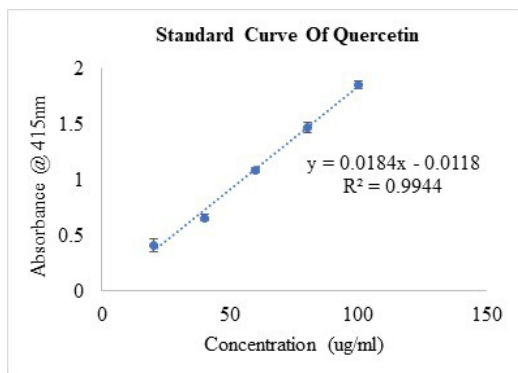


Fig. 2. Quercetin regression curve.

Based on our results which were quite consistent with the results of previous studies, phenols and flavonoids were found to be the most common phytochemicals in both the plants except, chloroform and hexane extracts which showed negative results for phenols. In our study, methanolic extracts gave the highest number of phytochemical constituents for both plant samples. Silica gel column chromatography was employed for the separation, identification and purification of phytoconstituents and only those fractions were selected for further quantitative estimations and antioxidant assays, which showed positive results for either qualitative phenol, flavonoid tests or both. Quantitative phenolic estimation of hexane and chloroform extracts of *A. galangal* was not performed because preliminary phytochemical results for phenols showed negative results which were in agreement with the results of a previous study which reported the absence of phenols in *A. galangal* hexane extract (Sani *et al.*, 2019). While the presence of phenolic content in the methanolic extract of *M. esculenta* showed lower TPC with the value of 6.180±0.083 GAE mg/g, as compared to the methanolic extract of *A. galangal* which can be supported by the fact that the composition of phenolic compounds of mushrooms generally depends on genetic, environmental and other factors. Previous studies reported that TPC in methanolic extract of *M. esculenta* was found to be 2.02±0.26 µg/ml whereas 238.52±0.021 GAE mg/g phenolic content have also been reported which indicated that our result is quite consistent with results reported by previous studies (Thakur and Lakhnpal 2014). Other studies reported that polyphenol constituents of *M. esculenta* were found markedly higher than other wild mushrooms such as species of Ramaria, Lycoperdon, Clavaria, Marasmius, Pleurotus, and Russula (Türkoğlu *et al.*, 2007; Ramesh and Pattar 2010). Recently, thermal processing methods have been reported to modify the TPC of *M. esculenta* due to an increase in the free phenols content by releasing bound phenols by heat treatment (Avrămiuc, 2018). The methanolic extract of *A. galangal* showed higher TFC with a value of 38.013±2.116 as compared to the methanolic extract of *M. esculenta* (1.573±0.557) QCE mg/g. Several flavonoids have been reported from *A. galangal* rhizomes such as galangin and galangin 3-methyl ether (Buckingham *et al.*, 2015). The presence of phenolic compounds (phenolic acids and flavonoids) as the major antioxidants and the presence of tocopherols, ascorbic acid and carotenoids in limited quantities in mushrooms was also reported by another study (Ferreira *et al.*, 2009). Each of the different plant extracts and selected fractions were screened for radical scavenging activity using H<sub>2</sub>O<sub>2</sub> and ABTS scavenging assays. There was a difference in our results, which was due to the difference in the type of free radicals and the sensitivity (Shalaby and Shanab 2013). H<sub>2</sub>O<sub>2</sub> being not so reactive in nature is toxic because penetrates biological membranes and gives rise to hydroxyl radicals in the calls (Gülçin *et al.*, 2010). H<sub>2</sub>O<sub>2</sub> scavenging activities might be attributed to antioxidant components which acted as good electron donors, thus facilitating the conversion of H<sub>2</sub>O<sub>2</sub> to water (Pavithra

and Vadivukkarasi 2015). The antioxidative power of *M. esculenta* might be attributed to its reducing nature which stabilized the oxidants and made them neutral at the cellular level, hence alleviating the chances of oxidative stress and their related disorders (Wagay *et al.*, 2019). The extract yields and antioxidant activities of the plants are strongly dependent on the nature of extracting solvent, the presence of different antioxidant compounds of varied chemical characteristics and polarities that might or might not be soluble in a particular solvent (Sultana *et al.*, 2009).

## CONCLUSIONS

From the above study, it can be concluded that three different solvent extracts of methanol, chloroform and hexane of *A. galangal* rhizome and *M. esculenta* mycelia showed the presence and absence of phytochemicals tested and varying degrees of in-vitro ABTS and H<sub>2</sub>O<sub>2</sub> antioxidant activities. Ethnomedicinal plants *A. galangal* and *M. esculenta* possessed antioxidant potential due to the presence of principal antioxidants such as phenols and flavonoids. Methanolic extracts of *A. galangal* rhizome rose out to be the profound hub for antioxidant activities because of their high TPC, TFC and ABTS radical scavenging potential. *M. esculenta* mycelia might be a promising herb due to its increased H<sub>2</sub>O<sub>2</sub> radical scavenging activity. We are blessed with a plethora of magical natural ingredients from nature, it solely depends upon us to scientifically explore them for societal benefits.

## FUTURE SCOPE

Despite the promising antioxidant activities exhibited by *A. galangal* and *M. esculenta*, it is still necessary to have many research studies for a better understanding of antioxidant mechanisms under different environmental conditions. Therefore, research work in future might be undertaken to study the benefits of these medicinal plants on the antioxidant defence system of the human body.

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**Conflict of Interest.** None.

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