ISSN NO. 2320-5407

Journal homepage: http://www.journalijar.com

INTERNATIONAL JOURNAL OF ADVANCED RESEARCH

RESEARCH ARTICLE

Taxonomy, physicochemical evaluation and chemical investigation of *Ganoderma applanatum* and *G. brownii*

Ranjeet Singh[1], Avneet Pal Singh[1], Gurpaul Singh Dhingra[1], Richa Shri[2]*

1.Department of Botany, Punjabi University, Patiala, India

2.Department of Pharmaceutical Sciences and Drug Research, Punjabi University, Patiala, India

Manuscript Info

Manuscript History:

Received: 12 March 2014
Final Accepted: 15 April 2014
Published Online: May 2014

Key words:
Basidiocarps, chemical composition, *Ganoderma*, polypore mushrooms, pharmacognostic study, Uttarakhand

Corresponding Author

Richa Shri

Abstract

Ganoderma species are used extensively in traditional medicine however quality standards have not been studied for all species. This investigation includes pharmacognostic study i.e. macroscopy, microscopy, evaluation of physicochemical parameters and phytochemical screening of basidiocarps of *Ganoderma applanatum* and *G. brownii* in order to establish the standards. These standardization profiles will help in authentication, determination of purity and chemical composition of these mushrooms.

.

INTRODUCTION

The genus *Ganoderma* P. Karst., includes poroid wood degrading macrofungi with cosmopolitan distribution mainly in tropical and sub-tropical forests. These mushrooms normally grow as parasites or saprophytes on living trees or sometimes associated with logs of wood and tree stumps. Species of *Ganoderma* are known to cause different types of rots by decomposing lignin as well as cellulose and other related polysaccharides in both angiosperm and gymnosperm hosts. Root rot and stem rot caused by different *Ganoderma* species result in the decline of numerous forest tree species (Bakshi, 1976; Foroutan and Jafary, 2007; Ko, 2009).

Fruiting bodies of some species of *Ganoderma* have been used in traditional medicine in China and other Asian countries for more than 2,000 years (Galor et al., 2004) as a valuable crude drug and health food that promotes health and longevity, lowers the risk of cancer, boosts the immune system, used in the treatment of debility and weakness, insomnia, diabetes, etc. A variety of products in the form of coffee, powder, dietary supplements, spore products, drinks, syrups, toothpastes, soaps and lotions are being prepared from *Ganoderma* fruiting bodies and have been commercialized as effective food and drug supplements for health benefits (Chang and Buswell, 1999; Lai et al., 2004). The commercial utilization of *Ganoderma* powder in dietary supplement products depends on its functional properties which should be determined before its incorporation in food and drug formulations. Some species of *Ganoderma* have been reported to contain different chemical constituents such as polysaccharides, proteins, amino acids, fatty acids, terpenoids, steroids, alkaloids, phenolic compounds, etc. (Boh et al., 2007; Mizuno, 1995; Paterson, 2006) with potential nutritional and therapeutic values. These bioactive constituents are reported to be responsible for the anti-inflammatory, anti-tumor, anti-oxidant, immunomodulatory, anti-diabetic, anti-viral, anti-bacterial, anti-fungal properties of the mushroom (Paterson, 2006; Kao et al., 2013), and has generated a lot of interest in the comprehensive study of these mushrooms.

Literature surveys revealed that the systematic evaluation including pharmacognostic study of these mushrooms is still lacking. Before any crude drug can be included in an herbal pharmacopeia, pharmacognostic

parameters and standards must be established (Chumbhale and Upasani, 2012). With this backdrop, it becomes extremely important to make an effort towards developing standards of the mushroom samples to be used as medicine. Hence, the present study was to evaluate the pharmacognostic parameters and chemical analysis of *G. applanatum* and *G. brownii* collected from Uttarakhand, India, with a view to establish standards for their identity, quality, purity and chemical composition.

Materials and Methods

Collection of mushroom samples

The fruiting bodies of *G. applanatum* and *G. brownii* were collected from different localities of Uttarakhand, India, during the monsoon months (July-September) of the year 2011. Field notes concerning host, name of locality, etc. were recorded for the collected specimens as shown in Table 1. Voucher specimens of *G. applanatum* (PUN 6563) and *G. brownii* (PUN 6564) have been deposited at the herbarium of the Department of Botany, Punjabi University, Patiala (PUN).

Macroscopic and microscopic evaluation

The specimens were identified on the basis of macroscopic and microscopic characters (Dhingra, 2005; Singh et al., 2014). For light microscopy, details of structures such as hyphae, basidiospores and pilear crust were observed by making crush mounts and free-hand sections in water and 5% KOH solution and staining in cotton blue (1% in lactophenol), Congo red (1% in distilled water), Phloxine (1% in distilled water) and Melzer's reagent. Line diagrams of different structures were made using camera lucida. For scanning electron microscopy (SEM) of basidiospores, portion of the hymenial surface was mounted on brass stubs using carbon tape, coated with a mixture of palladium and gold using a JEOL JEC-1600 coater at 20 mA for 15 sec under vacuum, and was observed using JEOL JSM-6510 LV SEM.

Chemicals

The chemicals used for extraction and qualitative chemical screening were of AR grade (Loba, Merck, India and SD Fine Chemical Ltd., India).

Physicochemical evaluation

The fresh basidiocarps were used to study foreign matter and moisture content (Indian Pharmacopoeia, 2007; WHO, 1998). The crude powder was subjected to the study of various physico-chemical parameters such as ash values, extractive values (Indian Pharmacopoeia, 2007; WHO, 1998); absorption properties, emulsion properties, foaming properties (Aremu et al., 2007); dispersibility (Kulkarni et al., 1991) and bulk density (Oladele and Aina, 2007). Each parameter was determined in triplicate.

Preparation of extracts

Dried powdered fruiting bodies (100 g) were subjected to successive Soxhlet extraction for 8 h using solvents in increasing order of their polarity viz. petroleum ether, chloroform, methanol, and water. Before each extraction, the powder was dried and then extracted with the next solvent. The scheme of preparation of extracts is shown in Figure 1. The extracts were concentrated, dried and weighed. The percentage yield and organoleptic parameters of each extract were recorded.

Chemical screening

The chemical examination of the extracts was performed by the standard methods (Harborne, 1973; Evans, 2003; Kokate, 2004).

Statistical analysis

The results of physicochemical evaluation have been expressed as mean ± standard error of the mean (SEM) and was analysed using the Student's t-distribution test of significance. Values were declared significant at $p < 0.05$.

Results and Discussion

The therapeutic use of herbal medicine is gaining considerable momentum in the world during the past decade. Hence, quality control standards for various medicinal natural products are essential to measure their authenticity, quality and purity. The quality of herbal drugs is the sum of all factors which contribute directly or indirectly to the safety, effectiveness and acceptability of the product.

The preliminary steps for establishing the quality control profile of any herbal drug is the macroscopic and microscopic evaluation. These are the simplest and reliable means for correct identification of source materials (Jafari et al., 2013). The combination of data obtained using both light and scanning electron microscopes is important for generating a more precise identification and confirmation of different species of *Ganoderma*. SEM is the most adequate technique to study the basidiospore surface.

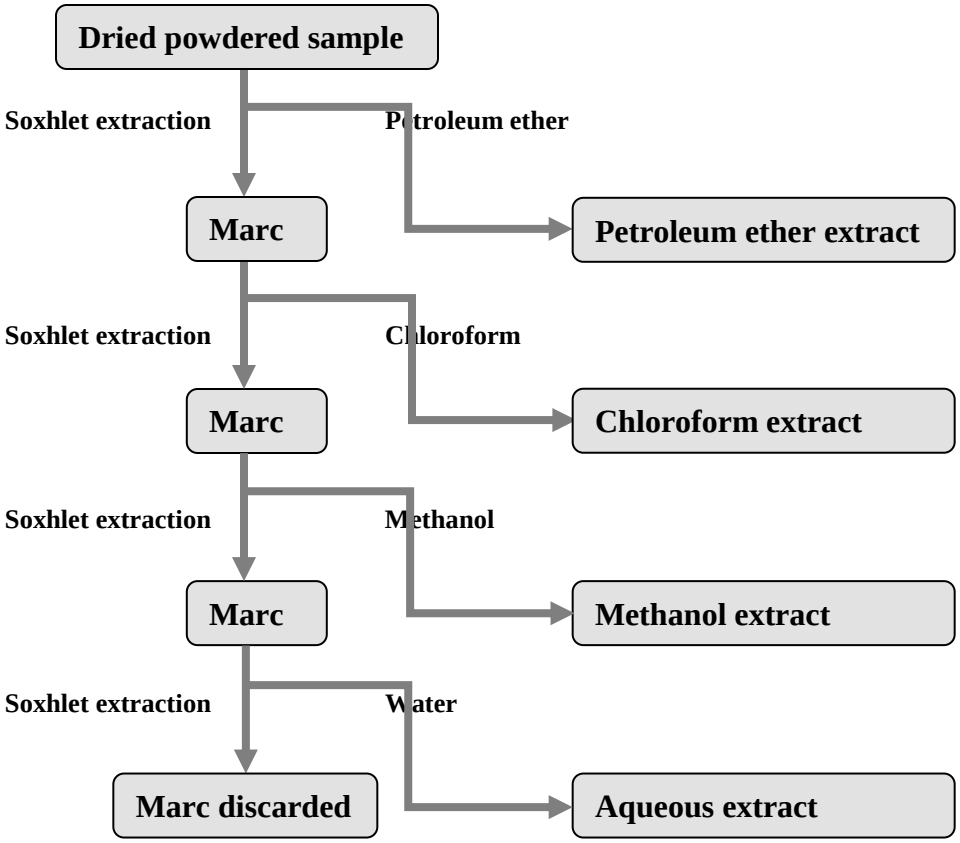

Fig 1 Scheme of preparation of extracts

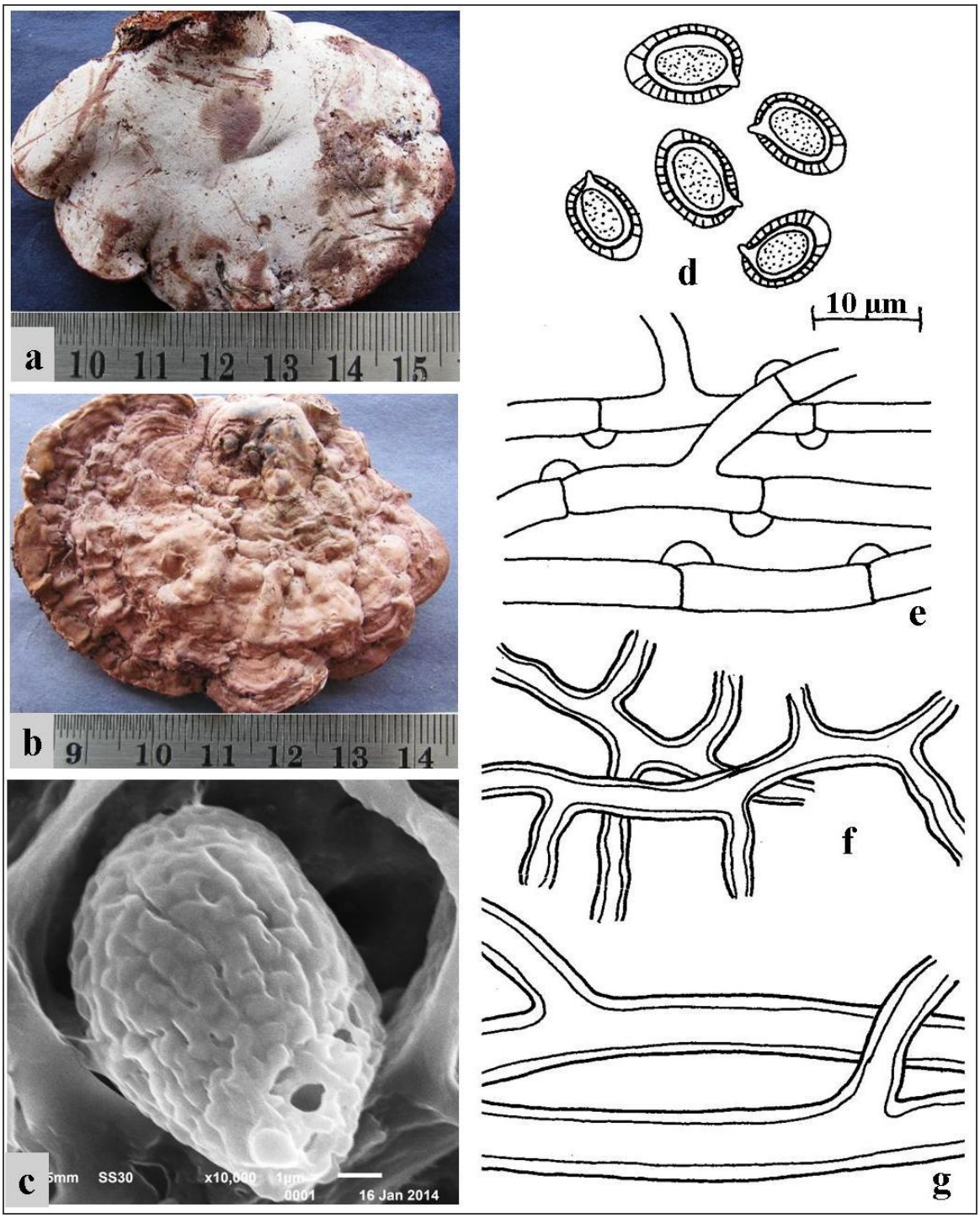

Fig 2 ***Ganoderma applanatum***. a- basidiocarp showing hymenial surface, b- basidiocarp showing abhymenial surface, c- SEM of basidiospore, d- basidiospores, e- generative hyphae, f- binding hyphae, g- skeleto-binding hyphae.

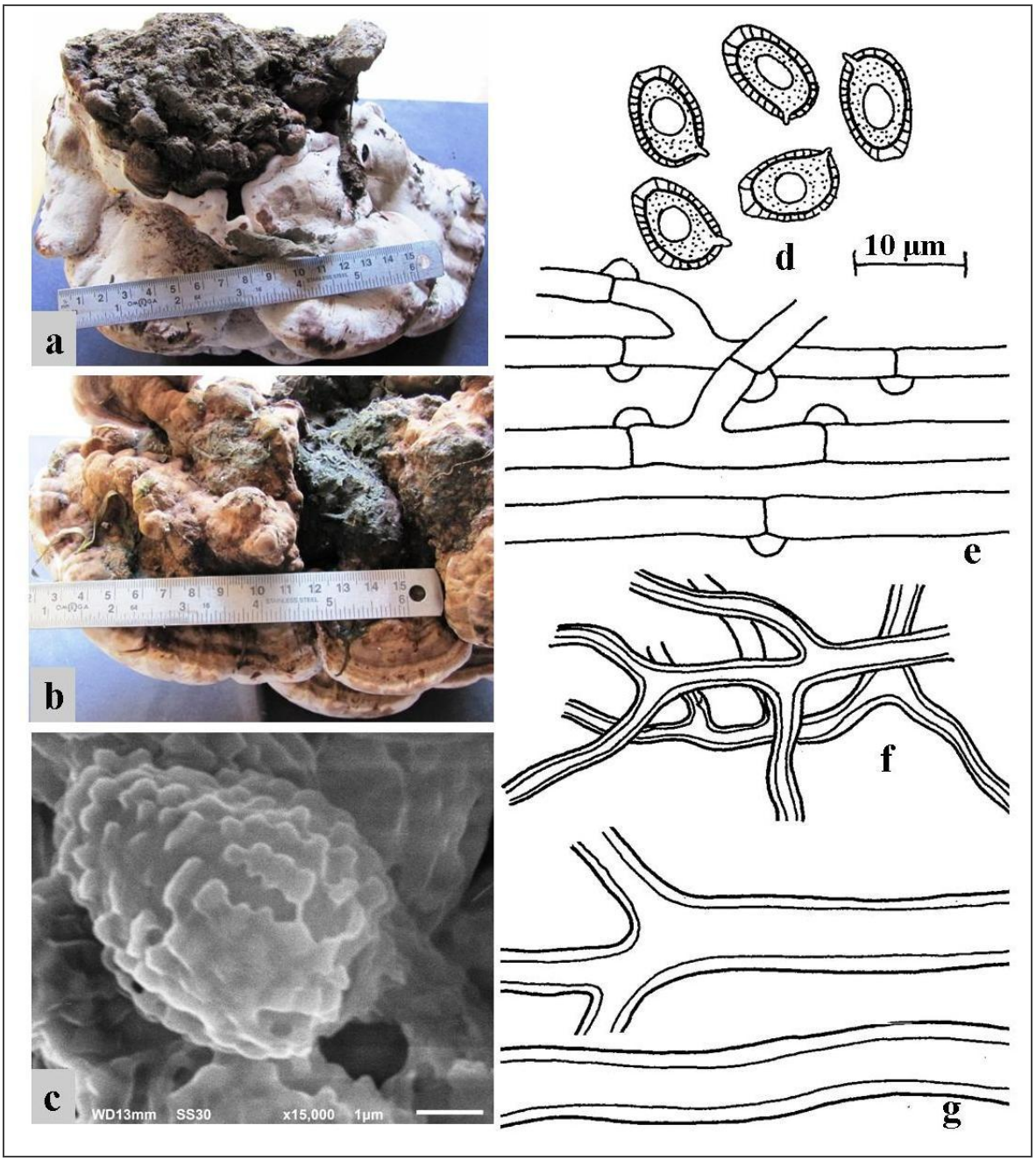

Fig 3 *Ganoderma brownii*. a- basidiocarp showing hymenial surface, b- basidiocarp showing abhymenial surface, c- SEM of basidiospore, d- basidiospores, e- generative hyphae, f- binding hyphae, g- skeleto-binding hyphae.

Table 1 Studied species and their origin (host and locality)

Species	Herbarium no.	Host	Locality
Ganoderma applanatum	PUN 6563	*Mangifera indica*	FRI, Dehradun
Ganoderma brownii	PUN 6564	*Gravillea robusta*	Tilwara, Rudraprayag

Table 2 Physicochemical analysis of *Ganoderma applanatum* and *G. brownii*

S. No.	Parameter	*G. applanatum* (Mean±SEM, n=3)	*G. brownii* (Mean±SEM, n=3)
1.	Foreign matter (% w/w)	0.061 ± 0.011^{A}	0.097 ± 0.016^{B}
2.	Moisture content (% w/w)	13.280 ± 1.833^{A}	14.940 ± 1.657^{A}
3.	Ash values (% w/w)		

	Total ash	8.022 ± 0.964 [A]	6.141 ± 0.736 [A]
	Acid insoluble ash	2.066 ± 0.205 [A]	1.302 ± 0.336 [A]
	Water soluble ash	5.353 ± 0.652 [A]	4.890 ± 0.219 [A]
4.	Extractive values (% w/w)		
	Alcohol soluble extractives	2.257 ± 0.023 [A]	3.455 ± 0.373 [A]
	Water soluble extractives	5.285 ± 0.157 [A]	8.354 ± 0.304 [B]
5.	Absorption properties (mlg^{-1})		
	Oil absorption capacity	1.233 ± 0.033 [A]	1.033 ± 0.088 [A]
	Water absorption capacity	1.267 ± 0.033 [A]	1.100 ± 0.058 [A]
6.	Emulsion properties (mlg^{-1})		
	Emulsifying capacity	14.680 ± 0.222 [A]	11.430 ± 0.095 [B]
	Emulsifying stability	6.878 ± 0.441 [A]	5.324 ± 0.019 [A]
7.	Foaming properties (%)		
	Foaming capacity	32.970 ± 0.722 [A]	37.070 ± 1.017 [B]
	Foaming stability	22.700 ± 1.758 [A]	27.330 ± 0.751 [B]
8.	Bulk Density (%)	0.380 ± 0.012 [A]	0.343 ± 0.015 [B]
9.	Dispersibility (%)	49.670 ± 1.335 [A]	60.900 ± 0.781 [B]

[a] SEM- standard error of means

[b] Values in the same row with the different superscript letter are significantly different ($p < 0.05$).

Table 3 Percentage yield and organoleptic properties of extracts of *Ganoderma applanatum*

Parameter	Petroleum ether extract	Chloroform extract	Methanol extract	Aqueous extract
Yield	1.52	1.02	2.21	4.04
Colour	golden brown to brown	dark brown	black	shining black
Odour	faint	characteristic faint	characteristic faint	very faint
Consistency	sticky semi solid	semi solid	soft solid	solid

[a] Yield–% w/w, dry weight basis

Table 4 Percentage yield and organoleptic properties of extracts of *Ganoderma brownii*

Parameter	Petroleum ether extract	Chloroform extract	Methanol extract	Aqueous extract
Yield	1.86	1.50	2.65	4.24
Colour	chocolate dark brown	greyish brown	dark brown	dark brown
Odour	faint	faint	faint	same as of fruiting body
Consistency	sticky	soft solid	solid	solid

[a] Yield–% w/w, dry weight basis

Table 5 Chemical screening of extracts of *Ganoderma applanatum*

S. No.	Chemical constituent	Petroleum ether extract	Chloroform extract	Methanol extract	Aqueous extract
1.	Carbohydrates	--	--	++	++
	Reducing Sugars	--	--	++	++
2.	Proteins	--	--	++	--
	Amino acids	--	--	++	--
3.	Lipids	++	++	--	--
	Steroids	++	++	--	--
	Terpenoids	++	--	--	--
4.	Alkaloids	--	--	++	--

5.	Phenolic compounds	--	--	++	++
	Tannins	--	--	--	--
	Flavonoids	--	--	++	++
6.	Saponins	--	--	--	++
7.	Glycosides				
	Anthraquinone glycosides	--	--	++	++
	Cardiac glycosides	--	--	--	--
	Cyanogenetic glycosides	--	--	--	--
8.	Mucilages	--	--	--	--

[a] ++ present [b] -- absent

Table 6 Chemical screening of extracts of *Ganoderma brownii*

S. No.	Chemical constituent	Petroleum ether extract	Chloroform extract	Methanol extract	Aqueous extract
1.	Carbohydrates	--	++	++	++
	Reducing Sugars	--	--	++	--
2.	Proteins	--	--	++	--
	Amino acids	--	--	++	--
3.	Lipids	++	++	--	--
	Steroids	++	++	--	--
	Terpenoids	++	--	--	--
4.	Alkaloids	--	--	++	--
5.	Phenolic compounds	--	--	++	++
	Tannins	--	--	--	--
	Flavonoids	--	--	++	--
6.	Saponins	--	--	--	++
7.	Glycosides				
	Anthraquinone glycosides	--	--	++	++
	Cardiac glycosides	--	--	--	--
	Cyanogenetic glycosides	--	--	--	--
8.	Mucilages	--	--	--	--

[a] ++ present [b] -- absent

Ganoderma applanatum (Pers.) Pat., Hyménomyc. Eur. (Paris): 143 (1887)

Syn: *Boletus applanatus* Pers., Observ. mycol. (Lipsiae) 2: 2 (1800).

It is macroscopically characterized by nonlaccate abhymenial surface and sessile basidiocarps, azonate context with black crustaceous layer, pilear crust easily broken when depressed with nail, and microscopically by trimitic hyphal system and broadly ovoid basidiospores. The basidiospore surface appears corrugated due to the presence of alternating folds and grooves (Figure 2a-g).

Ganoderma brownii (Murrill) Gilb., Mycologia 53(5): 505 (1962)

Syn.: *Elfvingia brownii* Murrill, Western Polypores 5: 29 (1915).

It is macroscopically characterized by nonlaccate abhymenial surface and sessile basidiocarps, concentrically zonate context without black crustaceous layer, hard pilear crust and microscopically by trimitic hyphal system and ellipsoid to ovoid basidiospores. The basidiospore surface shows evenly spaced depressions of grooves and ridges (Figure 3a-g).

Physicochemical analysis

Results of different physicochemical parameters of the dried, powdered basidiocarps are shown in Table 2. Our results show a high level of ash values, absorption properties, emulsion properties, and bulk density in case of *G. applanatum*, while *G. brownii* has higher values of foreign matter, moisture content, extractive values, foaming properties and dispersibility.

Physicochemical parameters like foreign matter, moisture content, ash content and extractive values are used to determine quality and purity of the crude drug (WHO, 1998). A drug containing appreciable quantities of foreign

matter may produce a critical impact on health. Therefore, the parameter must not be neglected. The legal limits for foreign matter as per standards should not be more than 2% w/w (Soni et al., 2011). Moisture content of drugs could be at minimal level to discourage microbial growth and undesirable enzymatic activity, which accelerates spoilage. The percentage moisture content values of the samples are comparatively more than the values for *G. lucidum* (10.54% w/w) reported previously (Usman et al., 2012). Ash content of a drug indicates presence of various impurities like carbonates, oxalates and silicates present along with the drug. The water soluble ash is used to determine the amount of inorganic compounds present in herbal drugs. Acid insoluble ash gives an idea about the amount of silica present and indicates contamination with earthy material. The results of the present studies show that the ash content values are comparatively higher than those reported for *G. lucidum* (5.93%, w/w) previously (Usman et al., 2012). Extractive values are useful to evaluate the chemical constituents present in the crude drug and also help in the estimation of specific constituents soluble in a particular solvent. Water soluble extractives were higher as compared to alcohol soluble extractives, which showed that *G. applanatum* and *G. brownii* had more water soluble polar constituents.

For efficient utilization and consumer acceptance of mushroom flours, it is desirable to study their absorption, emulsion and foaming properties. Quality attributes of developed food and drug products are generally affected by these functional properties of the powder (Kinsella, 1987). These properties affect the sensory characteristics of foods and drugs, play important roles in the physical behavior of foods and drugs or their ingredients during preparation, processing and storage. These properties provide a guide for the effective utilization of *Ganoderma* powder in various food applications and pharmaceutical preparations.

Absorption properties represents the ability of powder to associate with water or oil, which is a useful indication of whether powder or isolates can be incorporated into aqueous or oily food and drug formulations (Udensi and Okoronkwo, 2006). Water absorption capacity is important for moistness of the products to improve handling characteristics. Imbibition of water is an important trait in food products such as sausages, custards and doughs. Water absorption of powder is dependent mainly on the amount and nature of the hydrophilic constituents and to some extent on pH and nature of the powder (Gordon, 1993). *G. brownii* have contained more hydrophilic constituents than *G. applanatum*, which gave rise to higher water absorption capacity. Oil absorption capacity is attributed mainly to the physical entrapment of oils. It is an indication of the rate at which proteins bind to fats in food and drug formulations (Omimawo and Akubor, 2012). Oil absorption plays an important role in flavour retention, improvement of palatability and extension of shelf life where fat absorption is desired. Emulsion properties describe the ability of the powder to emulsify oil. Proteins can emulsify and stabilize the emulsion by decreasing surface tension of the oil droplet and providing electrostatic repulsion on the surface of the oil droplet (Sikorski, 2002), while some polysaccharides can help to stabilize the emulsion by increasing the viscosity of the system (Dickinson, 1994). Emulsions play an important role in pharmaceutical preparations such as cosmetics, pastes, etc. Emulsions have also been used for treating skin diseases and lacerations and for drug delivery, etc. (Khan et al., 2011). Foaming properties describes the ability of a powder to form foam. The foamability is related to the amount of solubilized protein (Narayana and Rao, 1982) and polar and non-polar lipids in a sample (Nwokolo, 1985). Saponins are also involved in the process of foam formation. Foams are used to improve texture, consistency and appearance of food and drug products (Akubor and Eze, 2012). Foaming properties are important in the preparation of shampoos, liquid detergents, toothpastes and beverages (Chen et al., 2010). Bulk density is a measure of heaviness of a powder sample, which is inversely proportional to the particle size (Omimawo and Akubor, 2012). The differences in the particle size may be the cause of variations in bulk density of powders. Bulk density is an indication of porosity of a powdered sample which influences the relative volume of the packaging material required which could be useful in material handling and application in wet processing in food and pharmaceutical industries (Kinsella, 1987). The dispersibility of powder in water indicates its reconstitutability (Kulkarni et al., 1991).

Chemical screening

The percentage yield and organoleptic parameters of the various extracts of *G. applanatum* and *G. brownii* are reported in Table 3 and 4. The results of chemical screening of the prepared extracts are given in Tables 5 and 6. Chemical screening of various prepared extracts revealed the presence of carbohydrates, proteins, amino acids, lipids, steroids, terpenoids, glycosides, phenolic compounds, alkaloids, and saponins, while tannins and mucilages were not detected in either of the species studied. Chemical analysis of different extracts revealed some differences in the constituents of the two species studied. Petroleum ether extract of both the species showed the presence of lipids, steroids and terpenoids. Chemical examination of chloroform extract of *G. applanatum* revealed the presence of lipids and steroids whereas carbohydrates, lipids and steroids were found to be present in chloroform extract of *G. brownii*. Carbohydrates, proteins, amino acids, alkaloids, phenolic compounds, flavonoids and glycosides were detected in methanolic extract of both the species. Aqueous extract of *G. applanatum* showed the presence of

carbohydrates, reducing sugars, phenolic compounds, flavonoids, saponins and glycosides whereas aqueous extract of *G. brownii* showed the negative results for the presence of reducing sugars and flavonoids.
Chemical evaluation is the criteria to estimate the major class of chemical constituents present in the crude drug. The results of chemical screening are in accordance with previous reports on mushrooms (Kadiri and Fasidi, 1992; Joseph et al., 2009; Ogbe et al., 2009; Asuquo and Etim, 2011; Ede et al., 2012). The presence of these primary and secondary metabolites attributes to the high nutritional and medicinal values of *G. applanatum* and *G. brownii*.

Conclusions

The aggregate data from the present study provides standards of the intact basidiocarps. These standards help to identify the genuine species and purity in the intact fruit body forms as well as powders available commercially. These contribute directly or indirectly to the safety, effectiveness and acceptability of the product, which can be used either as good source of nutraceutically important constituents or for evaluation of processed flour for several food and drug preparation applications. The preliminary chemical tests are helpful in finding the chemical constituents that may have medicinal properties and can be utilized for the treatment of various diseases. The results of the present study may be used in future for authentication and quality control of the different species of *Ganoderma*.

Acknowledgements

The authors are grateful to the University Grants Commission (UGC), New Delhi, India, for awarding a senior research fellowship to Ranjeet Singh. The authors also express their sincere thanks to the Department of Botany, Department of Pharmaceutical Sciences and Drug Research, Punjabi University, Patiala, for providing the infrastructure and support to carry out the research work. The authors also extend sincere thanks to the Sophisticated Instrument Centre, Punjabi University, Patiala, for providing SEM facility.

References

Aremu, M.O., Olaofe, O. and Akintayo, E.T. (2007): Functional properties of some Nigerian varieties of legume seed flour and flour concentration effect on foaming and gelation properties. J. Food Technol., 5:109–115.
Akubor, P.I. and Eze, J.I. (2012): Quality evaluation and cake making potential of sun and oven dried carrot fruit. Int. J. Biosci., 2:19–27.
Asuquo, J.E. and Etim, E.E. (2011): Phytochemical and antinutrients evaluation of *Oxyporus populinus*. J. Emerg. Trends Eng. Appl. Sci., 2:817–820.
Bakshi, B.K. (1976): Forest Pathology: Principles and Practice in Forestry. Dehra Dun, India.
Boh, B., Berovic, M., Zhang, J. and Bin, L.Z. (2007): *Ganoderma lucidum* and its pharmaceutically active compounds. Biotechnol. Annu. Rev., 13:266–301.
Chang, S.T. and Buswell, J.A. (1999): *Ganoderma lucidum* (Curt.: Fr.) P. Karst. (Aphyllophoromycetideae)- a mushrooming medicinal mushroom. Int. J. Med. Mushrooms, 1:139–146.
Chen, Y.F., Yang, C.H., Chang, M.S., Ciou, Y.P. and Huang, Y.C. (2010): Foam properties and detergent abilities of the saponins from Camellia oleifera. Int. J. Mol. Sci. 11:4417–4425.
Chumbhale, D.S. and Upasani, C.D. (2012): Pharmacognostic standardization of stems of *Thespesia lampas* (Cav.) Dalz & Gibs. Asian Pac. J. Trop. Biomed., 2:357–363.
Dickinson, E. (1994): Protein-stabilized emulsions. J. Food Eng., 22:59–74.
Dhingra, G.S. (2005): Genus Phlebia Fr. in the Eastern Himalaya. Indian J. Bot. Sci. 84:111–117.
Ede, S.O., Olaniru, E., Oteminyn, S., Aguiyi, J.C. and Ekwere, E.O. (2012): Analgesic and anti inflammatory activities of the ethanolic extract of the mushroom *Ganoderma applanatum*. Int. J. Res. Rev. Appl. Sci., 13:349–352.
Evans, W.C. (2003): Trease and Evans Pharmacognosy. Saunders, London.
Foroutan A. and Jafary, N. (2007): Diversity of heart and root rot fungi on park and roadside trees in Maharashtra, India. J. Appl. Sci. Environ. Manage., 11:55–58.
Galor, S.W., Tomlinson, B. and Benzie, I.F.F. (2004): Ganoderma lucidum (‘Lingzhi’), a Chinese medicinal mushroom: biomarker responses in a controlled human supplementation study. Brit. J. Nutr., 91:263–269.
Gordon, M.S. (1993): Simulation tools for the thermal processing of foods. Food Technol. Info., 8: 100–102.
Harborne, J.B. (1973): Phytochemical Methods. Chapman and Hall, London.
Indian Pharmacopoeia Commission (2007): Indian Pharmacopoeia. Vol. 1. Government of India, New Delhi.
Jafari, S., Saeidnia, S., Ardekani, M.R.S., Hadjiakhoondi, A. and Khanavi, M. (2013): Micromorphological and preliminary phytochemical studies of *Azadirachta indica* and *Melia azedarach*. Turk. J. Bot., 37:690–697.

Joseph, S., Sabulal, B., George, V., Smina, T. and Janardhanan, K.K. (2009): Antioxidative and antiinflammatory activities of the chloroform extract of *Ganoderma lucidum* found in South India. Sci. Pharm., 77:111–121.
Kadiri, M. and Fasidi, I.O. (1992): Secondary plant products in some Nigerian mushrooms. Niger J. Bot., 5:187–192.
Kao, C.H.J., Jesuthasan, A.C., Bishop, K.S., Glucinam, M.P. and Ferguson, L.R. (2013): Anti-cancer activities of Ganoderma lucidum: active ingredients and pathways. Funct. Foods Health Dis., 3:48–65.
Khan, B.A., Akhtar, N., Khan, H.M.S., Waseem, K., Mahmood, T., Rasul, A., Iqbal, M. and Khan, H. (2011): Basics of pharmaceutical emulsions: a review. Afr. J. Pharm. Pharmacol., 5:2715–2725.
Kinsella, J.E. (1987): Functional properties of protein: Possible relationship between structure and function in foods. Food Chem., 7:275–288.
Ko, W.H. (2009): Nature of slow and quick decline of macadamia trees. Bot. Stud., 50:1−10.
Kokate, C.K. (2004): Practical Pharmacognosy. Vallabh Prakashan, New Delhi.
Kulkarni, D.K., Kulkarni, D.N. and Ingle, U.M. (1991): Sorghum malt-based weaning food formulations. Preparation, functional properties and nutritive value. Food Nutr. Bull., 13:14–16.
Lai, T., Gao, Y. and Zhou, S.F. (2004): Global marketing of medicinal Ling Zhi mushroom Ganoderma *lucidum* (W.Curt.:Fr.) Lloyd (Aphyllophoromycetideae) products and safety concerns. Int. J. Med. Mushrooms, 6:189–194.
Mizuno, T. (1995): Reishi, *Ganoderma lucidum* and *Ganoderma tsugae*: bioactive substances and medicinal effects. Food Rev. Int., 11:151–166.
Narayana, K. and Rao, N.M.S. (1982): Functional properties of raw and heat processed winged bean (Psophocarpus tetragonolobus) flour. J. Food Sci., 47:1534–1538.
Nwokolo, E. (1985): Nutritional quality of the seeds of the African breadfruit (*Treculia africana* Decne). Trop. Sci., 27:39–47.
Ogbe, A.O., Ditse, U., Echeonwu, I., Ajodoh, K., Atawodi, S.E. and Abdu, P.A. (2009): Potential of a wild medicinal mushroom, Ganoderma sp., as feed supplement in chicken diet: effect on performance and health of pullets. Int. J. Poultry Sci., 8:1052–1057.
Oladele, A.K. and Aina, J.O. (2007): Chemical composition and functional properties of flour produced from two varieties of tigernet (*Cyperus esculentus*). Afr. J. Biotechnol., 6: 2473–2476.
Omimawo, I.A. and Akubor, P.I. (2012): Food Chemistry (Integrated Approach with Biochemcial background). Agbowo, Ibadan, Nigeria.
Paterson, R.R.M. (2006): *Ganoderma*–A therapeutic fungal biofactory. Phytochemistry, 67:1985–2001.
Singh, R., Dhingra, G.S. and Shri, R. (2014): A comparative study of taxonomy, physicochemical parameters, and chemical constituents of *Ganoderma lucidum* and *G. philippii* from Uttarakhand, India. Turk. J. Bot., 38:186–196.
Sikorski, Z.E. (2002): Proteins. In: Sikorski ZE (eds), Chemical and functional properties of food components, CRC, Florida, pp 133–178.
Soni, N., Lal, V.K., Agrawal, S. and Verma, H. (2011): Pharmacognostical and phyto physico-chemical profile of Curculigo orchioides (Gaertn). Adv. Res. Pharm. Biol., 1:130–138.
Udensi, E.A. and Okoronkwo, K.A. (2006): Effects of fermentation and germination on the physicochemical properties of Mucuna cochinchinensis protein isolate. Afr. J. Biotechnol., 5:896–900.
Usman, S.B., Kyari, S.U., Abdulrahman, F.I., Ogbe, A.O., Ahmad, G.Y., Ibrahim, U.I., Sakuma, A.M. (2012): Proximate composition, phytochemical and elemental analysis of some organic solvent extract of the wild mushroom–*Ganoderma lucidum*. J. Nat. Sci. Res., 2:24–35.
WHO, (1998): Quality Control Methods for Medicinal Plant Materials. World Health Organization, Geneva