Launching Mushroom Biotechnology In Goa By Dr. Nandkumar M. Kamat, Asst. Professor, Department of Botany, Goa University (first published in BuildArena-Goa)

With the arrival of monsoons, the mushroom season is here. M/s Zuari Foods and Farms pvt. ltd. managed by food technologist Dr. Sangam Kurade has set up a modern Button mushroom plant at Bhatapal Canacona. Mushroom biotechnology has entered Goa with this factory. With a capacity of 600 MT per year, these cultivated mushrooms are marketed all round the year. They have captured Pune, Mumbai, Bangalore and Chennai markets. There is great potential in Goa for bioindustries based on mushroom technology. I have been working since 1986 on edible mushrooms of Goa. This gave me an opportunity to learn about the local knowledge of wild mushroom diversity and practice the technology of a few cultivated species. Edible mushrooms are very popular in Goa but there is poor knowledge among the consumers about these species, their ecological and biological aspects. During my 25 years of research I have recorded about 100 different species of edible mushrooms in Goa. 35 of these species are collected by the locals from the wild for consumption and only 12-14 species produce a marketable crop which is sold from July to September. Local species have interesting local names-Olmi or alami, Roenichim, toshali, chochyali, khut or khuti, shiti, shitol, shiringar olmi, shendari, kuski, dukor, surya olmi, tel alami, fuge, bhuifod are some of the common local names. But, scientifically, what are these mushrooms?. The word mushroom is thought to be derived from the French 'mousseron' (muceron), 'moussee' or 'moss'. The Greeks used the word "mykes' for mushroom generally, and the science of fungi or mycology, etymologically is the study of mushrooms. In simple words, mushrooms are larger fungi, or "plants without chlorphyll'. Mushrooms are fully vegetarian items. Only in the natural biopolymers Chitin and Chitosan which occur in their cell walls, the mushroom share their chemical properties with the shrimps and crabs. Fungi, unlike the plants can not produce their own food using the sunlight as source of energy. The mushroom fungus exists in a thin, white filamentous stage. Only when it grows and produces a visible, big fruiting structure, we recognise it as mushroom. Just as plants produce seeds within the fruits, the mushrooms produce microscopic seeds called 'spores'. A single specimen can produce about 2-4 billion spores. Not all of these germinate. Those which survive, under wet conditions germinate and give rise to a filamentous stage which is recognised by biotechnologoists as powerful source of biomolecules like enzymes. These filaments are known as mycelium. Each mycelium is made up of small tubular cells, called hypha. The secret of developing commercial technology for cultivating mushrooms depends on understanding the growth of these hyphae, as the mushroom species completes its' life cycle from a spore. A lot of research has been carried out in 20 th century, in UK, USA, Holland, France, Japan to domesticate new species of mushrooms. In India, such research began only after 1950.

Mushrooms are useful fungi:-

Over 100,000 fungi are known in the world. Of these more than 3000 are mushrooms. But all are not edible and exploitable. The edibility of mushrooms was discovered thousands of years ago. Mostly by the pre-historic nomadic humans. A common man can not distinguish an edible mushroom from a look-alike poisonous variety. So, inexperienced people should not venture out to collect mushrooms. For serious students and experts computerised programs, with pictorial keys like **Matchmaker** (Kendrick, 2002, http://www.mycolog.com) have been developed based on major morphological characters for identification of about 1200 mushroom taxa.

Love or hate mushrooms?

An american anthroplogist, Wasson has divided the humans into 'mycophobes' (those who hate mushrooms) and 'mycophiles' (mushroom lovers). Mycophagy means eating fungi. Ethnomycophagy deals with tradition of certain cultures to consume fungi, mostly mushrooms. Each country has its' own ethnomycophagic tradition. There are myths and legends about mushrooms. But that could be a separate topic.

Mushroom habitats:-

Habitat means a house, a fixed location. In nature mushrooms grow wild in almost all types of soils on decaying organic matter, wooden stumps etc. Fruiting habitats are divided according to the substrates on which fruitbodies develop such as:. (1)Humicolous (humus inhibiting) e.g. *Agaricus campestris*, (2)Lignicolous (wood inhibiting) e.g. *Pleurotus ostreatus*. (3) Coprophilous (dung inhibiting) fungi e.g. *Coprinus sp.* (4) fungicolous (fungus inhibiting) parasitic or saprophytic eg. *Boletus* sp. etc. People who traditionally collect mushroom crops from the wild identify their habitats correctly. Such collection trips are known as 'forays' and these are immensely popular in the western world. In Afro –Asian countries however, mostly the tribals gather and market the wild edible mushrooms.

Edible mushrooms:-

Like the discovery of the edible plants, the knowledge of edible mushrooms dates back universally to the pre-historic phase of human cultural history. More than 2000 worldwide species of wild edible mushrooms which are gathered from their natural habitats prove the antiquity of the ethnomycophagic traditions.

How to identify edible mushrooms?

There are no fixed criteria to identify edibility of the wild mushrooms merely from the knowledge of its' habit, habitat or morphology or through simple chemical tests. But there are very general criteria which help to distinguish the edible species from the non-edible ones. The criteria includes the following aspects or features:-

- 1. All the established, non-toxic and fleshy species are edible-generally this knowledge is derived from the time- tested local ethnomycophagic traditions.
- 2. Organoleptic properties (texture, smell, taste etc.). Tough and leathery species are always excluded.
- 3. Idiotypic preferences. The allergy inducing species are excluded.
- 4. All those species, when consumed in a quantity of 100-200 g either by frying or baking do not cause health disorders.

Mushrooms as choicest food:-

Mushrooms form very delicious and choicest food stuffs. The very high nutritive value of edible mushrooms rank these above all the vegetables and most of the legumes and they are second only to meat .mushrooms provide a rich addition to the diet in the form of proteins, carbohydrates, valuable salts and vitamins. Experiments have proved that mushrooms are well suited to supplement diets which lack proteins and in sense have rightly been called "vegetable meat". As an illustrative example some of the benefits of *Pleurotus ostreatus* (Oyster mushroom) are given in the Box 1.

Box 1 Beneficial aspects of Oyster mushrooms

- 1. Oyster mushrooms contain 16 % proteins, 54 % carbohydrates, 12% fiber, 4.5 % lipids and 8 % minerals.
 - 2. Oyster mushrooms have antitumour, anti-cancer properties and lower VLDL and cholesterol and hence are recommended for patients with hypertension and cardio-vascular problems.
- 3. Oyster mushrooms have 19 mg of bio-assimilable Iron per 100 gms dry weight. A diet of oyster mushrooms boosts blood Haemoglobin.
- 4. In China, Oyster mushrooms in dry powdered form are used to trat lumbago. numbness and tendon discomfort.

Mushrooms are nutritious:-

Mushrooms have a high protein content (19-35% on a dry weight basis) of good quality (all essential amino acids for man including lysine and methionine, which are present in plants in very small amounts). This can be compared to 7.3% in rice, 13.2 % in wheat, 39.1 % in Soybean and 25.2 % in milk. Considering the Protein-calorie mal-nutritional problems in underdeveloped countries, mushroom cultivation on commercial scale has been identified as a promising area to guarantee food security in the 21 st century. Mushrooms can be compared more favourably with other crops in terms of yield per unit area. For example cereals can give an annual yield 3000 to 6000 kg/ha. but mushrooms may give up to 2 million kg/ha. Furthermore mushrooms have a high proportion of unsaturated fatty acids, are a good source of vitamins such as riboflavin and nicotinic acid and a good source of pantothenic acid as also appreciable amounts of thiamine, folic acid, and ascorbic acid. Mushrooms contain fibre and minerals and are low in calories, sodium, fat and cholesterol. In addition their nucleic acid content is not high to limit their daily use as a vegetable.

Medicinal mushrooms:-

Mushrooms are also known for their medicinal properties. These properties are being aggresively explored by major pharmaceutical companies. Hundreds of patents on bioactive compounds from mushrooms have been granted. Mushrooms have notable place in folklore throughout the world, and traditions of many cultures, past and present, describe medical effects of variously employed fungi. The most commonly cited, potential edible application of a higher edible fungus appears to be calvacin, a possible antitumour agent isolated from *Calvatia gigantea*. Some of the other medicinal properties of mushrooms are as follows:

- 1). Antibiotic activities- Some of the wood fungi are known to have effective antibacterial effects. e.g. *Polyporus*. Gregory and other scientists screened more than 7000 cultures of mushrooms for antitumor activity in rodent tumor systems. Fungi such as *Flammulinav velutipes* have also been known to have such properties. Other mushroom species having this property are *Clitocybe*, *Daedalea*, *Marasmius*, *Pleurotus* sp.etc.
- **2)Antifungal effects-** species like *Lentinus edodes*, Coprinus comatus and *Oudemansiella mucida* etc. are known to have relatively high rate of occurrence of antifungal antibiosis.
- 3) Antitumour effects-the mushroom species having antitumour effects are the major and current antibiotic screening activity. The species like *Ganoderma lucidum* and *G. applanatum* have been used from ancient time as 'mushroom tea' because these have antitumour activity. Some edible species like *Lentinus, Morchella, Boletus, Pleurotus ostreatus, Auricularia auricula-judae* are known to have strong antitumour properties. The reports also suggests that this sources also displayed hypocholesterolemic activities. Mushroom derived chemically compounds associated with antitumour activity and their sources include polysaccharides like lentinan from *Lentinus edodes* flammulin from *Flammulina Velutipes* and a triterpenoid from *Poria obliqua*, obliquol.

Mushroom cultivation:-

Mushroom cultivation technology is coming of age with the development of powerful biotechnological tools. Theoretically, it is possible to cultivate all the mushroom species. But the basic biology has to be first worked out in a research laboratory. Then artificial cultivation is attempted on trial scale. Depending on the mushroom species it could take years or decades to develop a new technology. Although, about 2000 wild edible mushroom species from 30 genera are regarded as prime edible mushrooms, only 80 of these are grown experimentally. Of these 40 are cultivated economically and around 20 cultivated commercially. Only 5 to 6 species are produced on commercial scale (Chang & Miles, 1993). These are (1) Agaricus bisporus (2) Pleurotus ostreatus (3) Volvariella volvacea (4) Lentinus edodes (5) Morchella sp. (6). Flammunila velutips. However species like Auricularia polytricha, Pholiota nameko, Russula sp. etc are cultivated in south eastern countries and are highly priced in those countries. Japan, Korea, China, Taiwan, etc. are the leading countries in the production of mushrooms. Total mushroom production world-wide has increased more than 10-fold in the period of 25 years from about 350,000 t in 1965 to about 4,300,000 t in 1991. The bulk of this increase has occurred during the eighties. A considerable shift has occurred in the composite of genera that constitute the mushroom supply. During the 1979 production year, the button mushroom, Agaricus bisporus, accounted for over 70% of the world's supply. By 1991, only 37% of world production was A. bisporus. Mainland China is the major producer (2,200,000 t or about 50% of the total) of edible mushrooms. The country wise details of production are given in Table 1

Table 1. World production of cultivated edible mushrooms in 1986 and 1991 (Chang 1993).

	Fresh wt (x 1,000 t)				
Species	1986		1991		Increase (%)
Agaricus bisporus	1,215	(55.8%)	1,590	(37.2%)	30.9
Pleurotus spp.	169	(7.8%)	917	(21.5%)	442.6
Lentinula edodes	320	(14.7%)	526	(12.3%)	64.4
Auricularia spp.	119	(5.5%)	465	(10.9%)	290.8
Volvariella volvacea	178	(8.2%)	253	(5.9%)	42.1
Flammulina velutipes	100	(4.6%)	187	(4.4%)	87.0
Tremella fuciformis	40	(1.8%)	140	(3.3%)	250.0
Hericium erinaceus			66	(1.5%)	
Pholiota nameko	25	(1.1%)	40	(0.9%)	60.0
Hypsizygus marmoreus			32	(0.7%)	
Grifola frondosa			8	(0.2%)	
Others			49	(1.2%)	
Total	2,176	(100.0%)	4273	(100.0%)	96.4

The technology of domestication of the wild edible mushroom species has evolved from very crude initial attempts made by the Chinese in 600 A.D. and over the period of past 200 years it has advanced to become a separate area of fungal biotechnology. Mushroom biologist Dr. Chang (1993) considers mushroom cultivation a "complicated business" involving a number of operations including the selection of an acceptable fruiting culture. Commercial cultivation involves preparation of spawn and compost, inoculation of the compost, crop care, harvesting, preservation of the mushroom and marketing. There are a large number of popular books/reviews which give detail account of these technologies (Chang and Hayes, 1978; Wood and Smith, 1987; Haque, 1989; Stametes, 1994; Bahl, 1994; Singh, 1996; Pathak et.al., 2000) and the readers are requested to consult these. Besides the well known Button mushrooms, there is a growing market for specialty mushrooms. A review article by Royse (1996) highlights their growing importance. A few of these species are mentioned below.

Auricularia auricula

Commonly known as wood ear, *Auricularia auricula* is the first recorded cultivated mushroom . *Auricularia* spp. production now represents about 11% of the total cultivated mushroom supply world-wide. *Auricularia auricula* and *A. polytricha* commonly are produced on a synthetic medium consisting of sawdust, cotton seed hulls, bran, and other cereal grains or on natural logs of broad-leaf trees. For cultivation on natural logs, members of the oak family (Fagaceae) are preferred, but many other species of both hard and softwoods may be used.

Ganoderma lucidum

Known as reishi or mannentake to the Japanese and Ling Zhi to the Chinese, *G. lucidum* is renown for its medicinal properties. Reishi often is associated with health and recuperation, longevity, wisdom, and happiness. It is believed that certain triterpenes and polysaccharides may account for the multiple activities of Reshi. Most cultivation of *G*.

lucidum is on supplemented sawdust contained in heat-resistant polypropylene bottles or bags.

Lentinus edodes

The cultivation of *L. edodes* (shiitake) first began in China about AD 1100. It is believed that shiitake cultivation techniques developed in China were introduced to the Japanese by Chinese growers. Various species of trees have been used for the cultivation of shiitake. One of the primary species used in one area of Japan in past years was the shii tree--thus the derivation of the name shii-take (Singer 1961). Most production today, however, is on various species of oak

Pleurotus ostreatus

Oyster mushroom production has increased at rapid rate world-wide during the last few years . From 1986 to 1991, oyster mushroom production increased from 169,000 t to 917,000 t (442% increase).

Volvariella volvacea

The straw mushroom derives its name from the substrate on which it originally was grown. Cultivation of *Volvariella* was believed to have begun in China as early as 1822. Many agricultural by-products and waste materials have been used to produce the straw mushroom. These include paddy straw, water hyacinth, oil palm bunch, oil palm pericarp waste, banana leaves and sawdust, cotton waste, and sugarcane waste. *Volvariella* is well suited for cultivation in the tropics because of its requirement for higher production temperatures. In addition, the mushroom can be grown on non-pasteurized substratemore desirable for low input agricultural practices.

Mushroom cultivation in India:-

Over 1500 species of mushrooms have been recorded in India . Of these the literature shows that about 300 are edible. As compared to the western countries, the commercial cultivation of mushrooms began very late in India. For more details refer to Dey (2000) and Pathak et. al. (2000) and the survey of agriculture 2002 (Hindu).

Mushroom cultivation in Goa

Since people have been collecting and consuming wild mushrooms in Goa, there were no attempts to introduce the technology of artificial cultivation. Some people attempted button mushroom cultivation in 1970s and then found it uneconomical. After 1985, the directorate of agriculture conducted demonstration camps for popularizing the technology of Oyster mushroom and Paddy straw mushroom cultivation. Oyster mushrooms were found suitable for Goa. The agriculture department launched a centrally sponsored scheme to conduct free mushroom training workshops. There was tremendous response. A modern mushroom spawn laboratory was set up in 1996 at Ela-farm to produce the oyster mushroom spawn. From 1997, the Goa state horticultural corporation took over the mushroom training camps. So, far more than 2000 people have been trained by both agriculture department and the horticulture corporation. As a resource person, I was involved in these training programs. Despite the best intentions of the government, only a few people came forward to produce and market the oyster mushrooms. The demand for

mushrooms from the hotel industry is about 500 MT per year. It goes up during the tourist season. But local mushroom cultivators have not properly understood the demand-supply equations and the economies of scale which needs to maintained for good profit making. The central and state government has declared amany incentives for mushroom producers. The Ministry of food processing , NABARD and APEDA have schemes for the mushroom growers. Except Dr. Sangam Kurade, who began systematically by setting up a Rs. 3 crores worth, air-conditioned plant in 1996, nobody has ventured in this field where tremendous opportunity exists for processing and exports.

Danger to local mushroom wealth:-

My field work and marketing surveys found that overexploitation of Goa's popular wild mushroom species which grow on ant-hills or termite hills, had eroded their diversity. The government had imposed a ban on their collection in 1993. There are 25 species of this mushroom in Goa. It is world's largest gene pool of termitophilic species to be found in any single state. The Chinese scientists have recently found important brain stimulating compounds in these species. The scientists at Indian Institute of Chemical biology have found useful enzymes in these species. My research has shown that these mushrooms can be cultivated with some difficulty, but more research is required. For any future research in biotechnology, we need to conserve and protect our mushroom biresources growing in their wild natural habitats. If these resources are overexploited or eroded, then India would lose her biological wealth. So, the ban imposed by the government of Goa for sanctuary areas is timely and appropriate.

Promoting mushroom biotechnology in Goa:-

Oyster mushrooms have great future in Goa. These can be grown almost for 10 months of year on simple raw materials like paddy straw. These mushrooms can be processed for value addition. Oyster mushroom cultivation could become a cottage industry. Women's groups, farmers' co-operatives could establish oyster mushroom cultivation units. At Goa university we have been working on cultivation of new species. We have got success in cultivation of wood ear mushroom or Auricularia species, on wooden logs. This species has big market in south-east Asia. We are attempting the cultivation of world's most popular medicinal mushroom Ganoderma species of Reishi. The Botany department which has a modern mycological laboratory has signed a MOU with M/s Zuari foods and farms ltd. for technical collaboration, R & D. We have successfully prepared and supplied the button mushroom spawn to them on trial scale. These efforts would be continued in future. Considering the craze for fast-food, Chinese food and the growth of tourism and hospitality industry in Goa, mushroom cultivation, processing and marketing holds great potential. As an integral part of state governments' effort to popularize Biotechnology in Goa, mushroom cultivation could become a money-spinner in future. V

alue-addition by processing:-

There is more value if the mushrooms are processed. Mushroom soup powders, pickles, wafers and noodles are very popular in Goa. This market can be developed. The growers need to learn the basic techniques of processing mushrooms first. An outline is given below.

Mushrooms respire fast. They lack protective covering and being rich in water

and soft in texture are highly perishable at ambient temperatures. The post-harvest technology of mushrooms focusses on the nullification of ill-effects such as decrease in freshness, water loss, browning, off-flavour development etc (Pathak et.al.,2000). Some methods of mushroom preservation include the following:-

- 1. Deep freezing
- 2. In 20% salt solution
- 3. In vinegar
- 4. In sodium or Potassium metabisulphite solution
- 5. Sun drying
- 6. Dehydration using an oven or drier
- 7. Freeze-drying
- 8. Frying and salting
- 9. Canning and bottling
- 10. Pickling

Of these freeze drying, canning and bottling methods are expensive and are used only by the large processing units whereas other methods are inexpensive and suitable for small quantities of mushrooms. More information could be obtained from Central Food Technological research institute-CFTRI, Mysore.

There is tremendous amount of information on Internet regarding various aspects of mushrooms and mushroom technology. A list of essential publications is given below for the readers interested knowing more on the subject.

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- 4. http://www.fungi.com website of Paul Stametes's Fungi Perfecti company.
- 5. http://www.faostat.org for statistics on world mushroom trade.