CHAPTER ONE

1. Introduction

1.1 Fungi

Fungi are distinct group among the plants. The term fungi (syn: fungus) is commonly used for a group of achlorophyllous thallus. Fungi are eukaryotic and spore-bearing organisms surrounded by a well defined cell wall made up of chitin, with or without fungal cellulose, along with many other complex organic molecules (Sharma, 2001).

Fungi consists of great variety of shape and sizes ranging from unicellular like yeast, but most string their cells together in long, thread-like strands called hypha. Most fungi produce an extensive system of hyphae, which may be visible when growing thickly in a mass called mycelium (commonly referred to as mould). Mycelium can be of any size from tiny clusters to massive acre wide systems, which effectively form the feeding and growing body of the fungus (F:\Fungi classification-What are fungi.htm). The word "fungus" gives us an image of mushrooms and toadstools that are found growing in the woods and fields.

Fungi are not able to ingest their food like animals do, nor can they manufacture their own food the way green plants do. Instead, fungi feed by absorption of nutrients from the environment around them. They accomplish this by growing through and within the substrate on which they are feeding. Hyphae spread widely through soil, rotten wood, etc., feeding on organic remains by secreting enzymes to dissolve the organic matter, then reabsorbing the nutrients. They continue accumulating nutrients until the internal and external conditions are right for the production of fruiting (F:\Fungi classification-What are fungi.htm).

Most fungi are saprophytes, feeding on dead or decaying material. Together with bacteria, the fungi are the principal agents of decay and so through the decomposition of the organic matter helps to remove leaf litter and other debris that would otherwise accumulate on the ground. Nutrients absorbed by the fungus then become available for other organisms which may eat fungi. A very few fungi actively capture prey, such as *Arthrobotrys* which snares nematodes on which it feeds. Many fungi are parasitic, feeding on living organisms without killing them. Ergot, corn smut, Dutch elm disease, and ringworm are all disease caused by parasitic fungi (Alexopolus & Mims, 1993).

Generally two types of reproduction can be found in this group of plant i.e. sexual and asexual. The asexual reproduction is carried out by the production of special type of reproductive cells for example by spores, fragmentation, budding, etc (Alexopolus & Mims, 1993).

Mostly in the fungi, the sexual reproduction includes the union of the compatable nuclei. The process of sexual reproduction is completed in three distinct phases, i.e. the fusion of two protoplasts of two gametes (plasmogamy), fusion of the two nuclei of both the fusing gametes (karyogamy) to form a diploid zygotic nucleus, and the formation of four haploid spores by the process of reduction division (meiosis) (Sharma, 2001).

1.1.1 Mushroom

The word mykes, from which mycology is derived, actually meant mushroom to the ancient Greeks and thus, etymologically, mycology is the study of mushrooms. Mushrooms are fungi. Unlike green plants, mushrooms are heterotrophs. Not having chlorophyll, they cannot generate nutrients by photosynthesis, but take nutrients from outer sources. However mushroom can produce a wide range of enzymes that degrade the complex substrate on which they grow, following which they absorb the soluble substance for their nutrition. This absorptive nutrition is the characteristic of fungus. Mushroom mycelium, which normally inhibits the ground, has the tendency to grow in all directions from a central point forming a large invisible circular colony. When the time for sporulation arrives, the sporophores are produced at the periphery of the colony and thus form a ring. This ring is called a fairy ring because of an old superstition that mushroom growing in a circle represents the path of dancing fairies (Alexopolus & Mims, 1993). The body of the mushroom stores nutrients and other essential compounds, and when enough material is stored and the conditions are right they start to fruit- produce mushroom.

Mushroom breed by spores. Under the proper conditions, spores germinate into hyphae (collectively, mycelia). Mycelia are filamentous and generally unseen with the naked eye. Germinating hyphae form primary mycelia, and then secondary mycelia through plasmogamy (hyphal fusion). They accumulate nutrients from the substrate (soil for plants) colonize substrate. When stimulated by temperature, humidity, etc., the mycelial colony forms pins under certain conditions and grow to fruitbodies (fruit for plants). Young fruit bodies are called pins (buds for plants). Pins differentiate into a cap and stem forming fruitbodies. Under

the cap, spores are produced in the gills. Fruitbodies release spores in order to produce the next generation (Cho, 2004).

1.1.2 *Pleurotus species*

Pleurotus mushroom is generally referred to as 'Oyster Mushroom', has its origin from Greek word "pleuro" which means formed laterally in a side way position, referring to the lateral position of the stipe (stem) in relation to the pileus (cap) (Suman & Sharma, 2005). It grows naturally in the temperate and tropical forest on death and decaying wooden logs or sometimes on dying trunks of deciduous or coniferous woods. It can also grow on decaying organic matter.

Visually the basidiocarp or the fruit bodies of an oyster mushroom have three distinct parts: (1) a fleshy shell or spatula shaped cap (pileus), (2) a short or long lateral or central stalk called stipe and (3) long ridges and furrows underneath the pileus called gills or lamellae. The gills stretch from the edge of the cap down to the stalk and bear the spores. The spores are smooth, cylindrical, or allantoid. The spores are heterothallic and germinate very easily on any kind of mycological media and within 48-96 hr whitish thread like colonies develop. The mycelium of *Pleurotus* is pure white in colour. Basidiospores on germination form primary mycelium. Fusion between two compatible primary myceliums develops into secondary mycelium, which is having clamp connections and it is fertile. Primary mycelium is clampless and non fertile. The fruit bodies of the mushroom are distinctly shell, fan or spatula shaped with different shades of white, cream, grey, yellow, pink or light brown depending upon the species. The colour of the sporophores is extremely variable character influenced by the temperature, light intensity and nutrient of the substrate (Suman & Sharma, 2005).

Oyster mushrooms are one of the most popular edible mushrooms and belong to the genus *Pleurotus* and the family Pleurotaceae. Many of *Pleurotus* mushrooms are primary decomposers of hard wood trees and are found worldwide (Kong, 2004). This mushroom has basidia with four basidiospores and a tetrapolar mating system. Its hyphae have clamp connections and most members of the genus, excepting a small minority, have a monomitic hyphal system (Suman & Sharma, 2005).

Pleurotus is appereciated for its culinary properties and broad adaptability under varied agroclimatic conditions. (Suman & Sharma, 2005).

To date approximately 70 species of *Pleurotus* have been recorded and a new species are discovered more or less frequently although some of these are considered identical with previously recognized species (Kong, 2004). Presently about 25 species are commercially cultivated in different parts of the world which include, *P. ostreatus, P. florida, P. flabellatus, P. sajor caju, P. citrinopileatus, P. sapidus, P. cystidiosus, P. eryngii, P. fossulatus, P. opuntiae, P. cornucopiae, P. yuccae, P. platypus, P. djamore, P. tuber-regium, P. australis, P. purpureo-olivaceus, P. populinus, P. levis, P. columbinus, P. nembanaceus,* etc (Suman & Sharma, 2005).

The species of *Pleurotus* exhibits much diversity in their adaptability to varying agro-climatic conditions and this flexible nature provides much more cultivated species in this genus than any other cultivated mushroom genus. Broadly all the oyster mushroom species are grouped in two main types: the grey type (European strains which fruit at low temperature i.e. below 12°C) and the white type (those strain which fruit at temperature up to 30°C) (Suman & Sharma, 2005).

1.1.3 Classification

Class: Basidiomycetes
Sub-class:homobasidiomycetidae
Series: Hymenomycates
Order: Agaricales
Family: Pleurotaceae
Genus: *Pleurotus*

1.1.4 *Pleurotus florida*

Pleurotus florida is an excellent edible and highly nutritious mushroom. It is widespread in temperate, sub-tropical and tropical zones (Kong, 2004). The edible fruit bodies develop in large numbers as a group on fallen trees, logs of wood and wooden poles. The cap measurement may range from 1.5 to 7.5 cm diameter while the stipe is 0.5 cm to 2.5 cm long, annulus is absent and the spore is cream-white in colour (Jonathan, 2002). It is similar in appearance to and was considered as subspecies of *Pleurotus ostreatus.* Some modern

mycologists are inclined to regard it as another species with different colour and different temperature requirements (Kong, 2004).

At low temperature, the colour of the caps is light brown, but they turn pale with increasing temperature. It could be harvested in warmer temperatures as its fruiting temperature range is wider than other *Pleurotus* mushroom and it does not require fruiting induction (cold shock). Moreover, it shows the highest yield among the *Pleurotus* species (Kong, 2004).

1.1.5 History of mushroom cultivation

Mushroom has been used as delicacy for more than two thousand years. During the middle ages, the Greeks and Romans considered mushrooms as a special food and they were obtained only in autumn and spring. Because of mushroom poisoning, the wild form or "toadstools", became objects of fear and distrust. Most poisoning cases were characterized by extreme pain and suffering before death (Pathak *et al*. 2000).

The first intentional cultivation of mushrooms was done in China around 600 AD, with the cultivation of *Auricularia auricula* on wood logs (Chang & Miles, 1993). In Europe sporadic attempts on mushroom cultivation without any systematic process were done even during the Roman times around 900 AD. However, the practice of cultivation of the European forms of mushroom in a systematic manner originated in France in the later part of the 17^{th} century. An early account of mushroom growing was written by Bonnefons, published in 1650. He described that growing mushroom belonging to *Agaricus*, one should make a bed of horse manure, about four fingers thick. The bed would first heat up and when it would start cooling, it should be planted with bits of mushroom stems and other parts not used in the kitchen. After a short period, the mushroom would appear on such bed. Thus, the concepts for making the bed for mushroom cultivation appear to have originated (Sing, 2007).

The practice of casing the bed with soil appears to have been initiated by French Horticulturist, Jean de la Quintinye in 1690. Another important milestone in developments of mushroom culture was the idea of using the spawn, put forth by Tournefort in 1707. He asserted that seeds of mushrooms were in the manure and piece of mouldy manure introduced in the new bed, mushroom would appear earlier and in good numbers. However, pure culture bottle spawn, using sterilized horse manure was invented many years later in 1894. Further developments in spawn making led to a method of preparing the grain spawn. The grain

spawn, which is in use even today, was first innovated in the USA and patented by Sinden in 1932. Side by side with the developments in the spawn making techniques, other aspects of mushroom cultivation also continued to progress. One important milestone in the developments of commercial mushroom farming was mushroom cultivation in the green house and this practice was initiated in Sweden around 1754. Thus, with more experiments and research efforts in many parts of the world, mushroom-growing activities became more and more scientific and technologically advanced (Sing, 2007).

Cultivation of *Pleurotus* is considered as an agriculture enterprise that challenges the combined skills of both industrial and agricultural technology. A preliminary form of *Pleurotus* culture (without use of inoculum) existed in Europe more than one and half century ago. In this case, Lumber man used to carry logs/stumps in which Oyster mushroom mycelium was growing naturally and set these logs in cool damp place, which enable them to periodically collect oyster mushrooms from these logs. First successful experimental cultivation of *Pleurotus* was achieved in Germany by Falck in 1917. He inoculated tree stumps and wooden logs with mycelium of *P. ostreatus* and could harvest fresh Oyster mushroom (Suman & Sharma, 2005).

1.1.6 History of mushroom cultivation in Nepal

In Nepal, mushroom cultivation was initiated by the Division of Plant Pathology, Nepal Agricultural Research Council (NARC) in 1974. The growing technology for white button mushroom was developed during that early period and extended to general farmers starting in 1977. It utilized the synthetic media of paddy straw, which is harvested twice a year in Kathmandu (Manandhar, 2004).

Two mushroom species are the most popular in Nepal. They are button mushroom (*Agaricus bisporus*) and oyster mushroom (*Pleurotus ostreatus). P. sajor-caju* and *Pleurotus sp*. have also been cultivated in Nepal. In the last few years *shiitake (Lentinus edodes*) has also been cultivated on a very limited scale (Adhikary & Manandhar, 1993). The area of production and number of people engaged in the cultivation has not been commercially quite significant. Species of *Auricularia* has also been recommended for cultivation in Nepal. Though a few growers cultivate this mushroom on a very limited scale, it has yet to assume a commercial significance. *Ganoderma lucidum*, a medicinally very important mushroom is being

experimented in the Division of Plant Pathology, NARC. Although it has been cultivated experimentally by some growers in Nepal, no large farming of this species has been done so far. One or more private farms have started producing spawn of this mushroom (Singh, 2007).

The growing technology to grow oyster mushroom using chopped straw packets was introduced to the farmers in 1984, and since then mushroom cultivation has become more popular among farmers (Manandhar, 2004).

Mushroom consumption in Nepal has been increasing rapidly in recent years. Information provided by Kantipur Television on Magh 23, 2061 (Feb, 2005), based on interviews with growers in Balambu area in Kathmandu, revealed that the annual mushroom production and sale reached to the amount of five crores (NRs 50 million). In other words, mushroom production based on estimation of Balambu area alone was about 625 metric tons at the rate of NRs 80/kg of mushroom. The same day Nepal television revealed that in Morang and Saptari districts, mushroom production reached as much as 700kg/group/season. Therefore, mushroom production in Nepal reached about 700 metric tons in the year (Singh, 2007).

1.1.7 Significance of mushroom production

Mushroom has been part of human diet since time immemorial. Undoubtedly, mushrooms were man's earliest food and they were often considered as exotic and luxurious food reserved for the rich. Today mushrooms were food for the both the rich and poor. They can be grown anywhere as long as the condition for their growth and cultivation are provided (Quimio, 2004).

Pleurotus spp can be grown on cereal straw, sugarcane bagasse, maize cob, saw dust, banana pseudostem, ipil-ipil leaves, water hyacinth and others. *Pleurotus* mushroom can degrade any kind of agricultural or forest wastes which are containing lignin, cellulose and hemicellulose. This is a great advantage of solving the organic problem faced in agriculture, forestry and agrobased industries and to avoid pollution (Jana & Das, 2007).

Cellulosic wastes obviously increase pressure on environmental pollution creating health hazards and disposal problem. Research thrusts for waste recycling as an alternate energy

source, will not only thus relieve pressure on environmental pollution, but also will help in alleviating health hazards (Jana & Das, 2007). Mushroom cultivation is essentially a recycling process in which agricultural wastes are converted into protein rich biomass of mushroom.

Substrate preparation for oyster mushroom is very simple. Moreover, composting-the difficult preliminary step for button and straw mushroom is not required for oyster mushroom cultivation.

While availability of land may be a limiting factor in most crop production operation, it is of relatively little importance in mushroom culture. They can be grown under cover: inside mushroom houses, in basements, garages or other suitable areas within the houses. Mushroom cultivation does not require large space as in the case of other cash crop. In addition to the floor, air space can be utilized for higher production. They maybe located on any unused land or available space between trees or beside buildings or houses (Pathak *et al*., 2000). Even landless peasant can grow mushrooms as a valuable crop as long as they have the proper technology, the proper substrates, and the planting material called spawn.

Generally, the growing of oyster mushroom is less expensive than that of other cash crops. The major region for this is, it requires little space and inexpensive raw materials. It does not require fertilizers and other costly inputs. The mushroom growing required low cost material and technologies while offering a high and quick return on money invested. Therefore in many developing countries of the world, mushroom can mean cash for the poor and a new source of nutrition (Quimio, 2004). Oyster mushroom is economically efficient for the farmers of other cash crops, who do not have to buy the raw materials for substrate and can use low cost substrate for mushroom cultivation on seasonal basis.

Mushroom growing is the women friendly operation. There is need to diversify the income opportunities available to women, to raise their productivity and lighten their workload. Mushroom growing is one of the agricultural activity in which women can play a vital role without sacrificing their household responsibility. (Pathak *et al*., 2000)

The productivity of oyster mushroom is very high as compared to all other cultivated mushrooms. One can harvest minimum of about 600 to 700 kg of fresh oyster mushroom

from one ton of dry wheat or paddy straw in 45-60 days, while with the same quantity of straw only about 200-250 kg of white button mushrooms are obtained in 100-120 days. Yield of mushroom can further be increased by supplementing the substrate with suitable nitrogen source like viz soyabean and cotton meal or by introducing high yield cultures/strain (Suman & Sharma, 2005).

Among all the cultivated mushrooms *Pleurotus* has maximum number of commercially cultivated species suitable for round the year cultivation. Moreover, variation in shape, colour, texture, aroma are also available per consumers choice (Suman & Sharma, 2005).

An adequate food intake is one of the fundamental human requirements. In most of the developing countries the rate of food production is very low in compare to the rate of population growth. The upward shooting prices of protein rich commodities such as milk, meat, poultry and pulses in developing countries is clear indication that the Demand-Supply syndrome is building in favour of increasing demand vs. short supply (Kausar, 1988). No one can deny the fact that the mushroom, a novel source of protein offers a promising way of alleviating protein malnutrition in shorter time.

Unlike white button mushroom, the oyster fruit bodies can be easily dried and stored. Dried oyster mushroom can be instantly used after soaking in hot water for 5 to 10 minutes or it can be used in powder form for several preparations. Fresh mushroom has a shelf life of 24-48 hours even at room temperature. (Suman & Sharma, 2005)

1.1.8 Nutritional value of mushroom

Mushrooms are being used as food and medicine since times immemorial. The Buddha is believed to have eaten mushrooms before being transported to Nirvana. Prophet Muhammad declared mushroom as a gift of God with its water a superb eye-healer. The Aryanas used an intoxicating drink “soma” in religious rites. According to Wasson the “soma” in the Rig Veda refers to *Aminita muscaria* (Kausar, 1988).

Mushroom is a highly nutritional food. It may be considered as a pool of nutrients and also popularly called “vegetable meat”. It consists of protein, carbohydrate, fats, vitamins, minerals, fiber, etc.

Mushrooms are good source of high quality proteins and contain 20-35% protein (dry wt. basis) (Suman & Sharma, 2005). The protein found in mushroom is less than meat but higher than other foods. In an amount of crude protein mushroom rank below most animal meat but well above other food including milk which is an animal products. The protein content of mushroom is about twice that of asparagus and cabbage and four times and twelve times those of orange and apple respectively (Aryal, 2003). In view of general shortage of animal protein in the developing countries, the need to utilize vegetable protein, as an alternate source has been duly recognized (Altschul, 1965). Among unconventional sources of protein, higher fungi particularly mushrooms stand out as a distinct class.

Fat is almost free in mushroom. The fat content in different species of mushroom ranges from 1.1 to 8.3% of dry weight. At least 72% of total fatty acids were found to be in unsaturated form in mushroom. Unsaturated fatty acids are essential to our diet whereas saturated fatty acids, which are present in high amount animal fats, may be harmful to our health. The finding of a high proportion of unsaturated fatty acid in many mushrooms is a significant factor in regarding mushroom as a healthy food (Chang & Miles, 1993)

The carbohydrate also found in very less amount. The absence of starch in mushroom makes it an ideal food for diabetic patients and for person who wish to shed excess fat from their bodies (Bahl, 2000).

Mushroom are also good source of several vitamins including thiamine (Vit B1), riboflavin (Vit B2), biotin and ascorbic acid (Vit C), Vit A, B and D. Due to presence of Vit B, it helps to control the cholesterol (Aryal, 2003) .

Mushrooms are good source of minerals (P, K, Fe, Cu and Na). The mineral content particularly calcium and phosphorus are remarkably higher in mushroom than in many fresh fruits and vegetables (Suman & Sharma, 2005).

Thus presence of mushrooms in human diet can help in supplementing protein, vitamins and minerals in the diet of the vulnerable groups i.e. children, pregnant and lactating women and labourers. It is also considered as an ideal diet for heart patients in view of its low fat and high protein contents (Kausar, 1988).

Flegg and Maw (1976) concluded that digestibility of *Pleurotus florida* was higher (79.07%) than that of spinach protein (73%) lower than that of meat protein (99%).

1.1.9 Medicinal values of mushrooms

They have good medicinal properties like antibiotic activities, anticancer, hypolipidemic, hypocholestermic and anti-hypertension effects. Potassium/sodium ratio is very high in mushrooms which are desirable for the patients of hypertension (Suman & Sharma, 2005).

Mushrooms have very less calories and contain approximately 80 to 90 percent water. At the same time, they have low sodium, carbohydrate and fat content and high fiber content. This is the reason why mushrooms are considered good for those aiming for weight loss.

Mushrooms are an excellent source of potassium. In fact, it is said that there is more potassium in a mushroom than a banana. Since potassium helps lower blood pressure and diminished the risk of stroke, mushrooms are recommended to people suffering from hypertension.

Mushrooms are rich in copper, a mineral that has cardio-protective properties. A single serving of mushrooms is said to provide about 20 to 40 percent of the daily needs of copper.

Mushrooms are believed to help fight against cancer. They are an excellent source of selenium, an antioxidant that works with vitamin E to protect cells from the damaging effects of free radicals.

After a long term research and experiments done by large Japanese hospitals and research institutes, red mushroom, *Ganoderma lucidum* having positive effects been confirmed in every respect especially its hemocatharsis effect, i.e. improves blood circulation and cell quality. It allows the internal organs to function properly, improves a weak constitution, and enhances the immune system (www.redmushrooms-healthmanna.com).

Being rich in fiber, protein and Vitamin B, mushrooms help maintain a healthy metabolism.

It has been found that mushroom extract helps stop migraine headaches and is beneficial for people suffering from mental illnesses, like obsessive-compulsive disorder.

Oyster mushrooms are said to be useful in strengthening of veins and relaxation of the tendons (http://lifestyle.iloveindia.com/lounge/benefits-of-mushrooms-1337.html)

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1.2 Objectives of the study

1.2.1 General objective

To determine an effect of different substrate on the production of oyster mushroom (*Pleurotus florida*).

1.2.2 Specific objectives

1. To identify the low cost substrate.
2. To conserve environment by recycling agricultural and industrial wastes.
3. To identify the problem of mushroom growers.

1.3 Justification of the study

Nepal is an agricultural country. Being an agro-based country, the raw materials for the preparation of substrates are available in plenty, added with cheap labour. The raw materials for mushroom cultivation are agricultural waste and are usually available in at the farmers' door. There are different types of crop residue that may be used in mushroom production. But all the farmers are using only rice straw for *Pleurotus* cultivation. Most farmers in our country are unknown about it. So, idea should be given to the farmers about the use of other raw materials which are available in their local area.

Large quantity of agricultural and industrial waste materials are produced in our country which are mostly discarded, burned and neglected. In some areas these materials are used as fuel and soil conditioner, etc. Channeling of these wastes into efficient and productive system is a dire need.

Mushroom is nature’s re-cycler which converts our agricultural and industrial wastes into good quality protein rich vegetable. In the process of mushroom growing, environmental

pollution from such practices may be reduced. The useless by-product can be recycled to produce value added mushroom.

From dietary stand point mushroom are particular favourable foods. It can significantly reduce protein malnutrition.

Nepal also has large number of agro-climatic regions that can offer congenial climatic condition for mushroom cultivation. Mushroom is one of the income generating and fast maturing crops.

1.4 Limitation of the study

1. The study is the partial fulfillment of Master's Degree.
2. The study is carried out only for few months.
3. Different political hindrance during the period of experiment.

CHAPTER TWO

2. LITERATURE REVIEW

Kausar (1988) did the experiment on different *Pleurotus spp*. Cereal crop residues i.e. rice straw and rice husk were used as substrate for oyster mushroom cultivation. Experiment on *P. florida* and *P. sajor-caju* cultivation were carrried out from April to October and *P. sapidus* and *P. ostreatus* from November to March because required temperature and relative humidity could be easily maintained. 30°C was found to be optimum temperature for the growth of *P. florida*. Rice straw proved to be better substrate than rice husk because of 29-48% higher yield by different substrate mushroom.

Balaza (1981) observed that *Pleurotus florida* can be cultivated on cereal straws or maize cobs. The substrate could be enriched by some other agricultural wastes. Alfalfa flour, oat meal, rape straw or soya straw was mixed in different concentrations with wheat straw which was used as bedding materials. Supplementation of wheat straw increased the yield of mushroom.

Khan *et al*. (1981) studied the yield performance of four strains of oyster viz. *Pleurotus ostreatus* (strain 467), *P. florida* (strain 3526), *P sajor- caju* and *P. ostreatus* on paddy straw during winter. The temperature of growing room varied between 16^{o}C and 24°C during the course of the experiment. The results showed that *P. ostreatus* (strain 467) was the most productive strain followed by *P. florida* (strain NRRL 3526), *P. sajor caju* and *P. ostreatus.*

Khanna & Garcha (1981) studied the cultivation of *Pleurotus florida* NRRL 3526 on chopped paddy straw in fruit baskets during the period of mid December to March. The results indicated that a cumulative yield of 32 per cent (on fresh weight basis) was obtained in 104 days. The results of different spawning rates showed that higher spawning rate did not increase the yield proportionally.

Ponmurungan *et al*. (2007) studied the effect of different biowastes such as paddy straw, sorghum straw, sugarcane molasses, saw dust and paper waste on the growth and biochemical constituents of oyster mushroom (*Pleurotus florida*). Favourable conditions were created to attain the maximum yield of mushrooms. The result revealed that the mushroom growth was

better in paddy straw followed by sugarcane molasses and least in wood saw dust and paper waste.

Custodio (2004) tried coco lumber sawdust for cultivation of *Pleurotus ostreatus* by bag cultivation. According to them hardwoods like mahogany or nara contain much higher amount of structural carbohydrate than softwood trees like coconuts. This means that the hardwoods have more nutrients that can be used by mushroom than softwood.

Shah *et al*. (2004) carried out research experiment to investigate the cultivation of Oyster mushroom on different substrates during August to September year 2002. The substrates used for the cultivation of oyster mushroom where, Saw dust 50% + Wheat straw 50% Saw dust 75% + leaves 25%, Saw dust 100% Wheat straw 100%, Wheat straw 50% +Leaves 50% Leaves 100%. Sawdust produced highest yield, biological efficiency and number of fruiting bodies, recommended as a best substrate for Oyster mushroom cultivation.

Cho *et al*. (1981) reported that a mixture of cotton seed hulls and saw dust was a good substrate for the growth of *Pleurotus sajor-caju* in Australia. Supplementation of the substrate with wheat bran resulted in a significant increase in the yield of mushroom. The result indicated that light was inhibitory to mycelial growth. However, exposure to light after formation of pin heads was important to avoid abnormalities in the fruiting bodies.

Sivrikaya *et al*. (1998), some forest and agricultural wastes of Eastern Black Sea Region of Turkey were subjected to mushroom (*Pleurotus florida*) cultivation in this study. 1:1, 1:3 and 3:1 (w:w) mixture were prepared mainly by tree leaves wood wastes of timber workshops, cupola of nut trees (NC) and leaves (NL), corn stalks (CS), waste tree leaves (WTL) of tree factories, wheat straw (WS) and waste paper (WP). Results indicated that wood waste yield highest mushroom production . Other regional agricultural and forest wastes were also gave remarkable yield values. In *P. florida* cultivation, corn stalk and cupola and nut increased the yield values when used with wood wastes as mixtures. Presence of wood waste in the prepared mixtures with wheat straw and corn stalks improved the quality of properties of fruit body as cupola of nut resulted in smaller caps in diameter. Further studies are being continued in strain development of different *Pleurotus* species and increasing mushroom yield by many activators and additives for the region.

Baysal & Perker (2001) studied *Pleurotus ostreaus* mushroom's cultivation on waste paper and husk rice additionally. In the scope of study, waste of paper has been used as main substrate where as waste of husk rice is co-substrate. Results indicated that waste of rice increased important cultivation parameters of *P. ostreatus* mushroom's such as mycelia development and mushroom yield. Mixtures based on waste of paper and husk rice gave more yield than only waste of paper. The best mycelia development and yield was obtained mixtures of WP+HR (75+25

Das & Mukherjee (2007) studied *Pleurotus ostreatus* using dry weed plants like *Leonotis sp, Sida acuta, Parthenium argentatum, Ageratum conyzoideas, Cassia sophera, Tephrosia purpurea* and *Lantana camara. Leonotis sp.* was found to be the best substrate in fruit body production of *P. ostreatus* when it was mixed with rice straw (1:1, wet wt. / wet wt.) for mushroom cultivation. The fruiting time for *P. ostreatus* was also less on *Leonotis sp*. than on any other weed substrates tested in this investigation. *T. purpurea* was the least suited weed for oyster mushroom cultivation. The main problem of oyster mushroom cultivation on weed substrates was found to be low yield in the second flush that could be overcome blending weed plants with rice straw.

El-Kattan & Salama (1995) carried out the study to evaluate legume waste and/or gypsum as possible additives to rice straw for the cultivation of the oyster mushrooms *Pleurotus florida* and *Pleurotus ostreatus*. Both yield and quality of oyster mushroom have been improved by supplementing rice straw with legume waste. The highest beneficial effect on yield was achieved when rice straw was supplemented with 50% legume waste. The biological efficiency was 121.3 and 116.2 for *P. ostreatus* and *P. florida*, respectively. Lower mushroom yields were obtained from the two species when substrates composed of either lower or higher percentages of legume wastes had been used. The highest mushroom yield was obtained with substrates composed of equal amounts of rice straw and legume waste with 5% gypsum; these yields were, however lower than those obtained without gypsum addition. Clearly the gypsum addition adversely inhibited the favorable effect observed with legume waste on mushroom yield.

Donini *et al*. (2009) did the research to evaluate the cultivation of the strains of *Pleurotus ostreatus* in elephant grass substrate supplemented with different kinds of bran. The experiment consisted in the use elephant grass substrate supplemented with soy, wheat, rice

or corn bran in concentrations of 0, 10 or 20% poured in flasks that were inoculated with spawns of BF24, DF33 and HF19 strains of *P. ostreatus* and incubated at room temperature (20 - 28 ºC). After the complete colonization of the substrate, the flasks were transferred to a fructification chamber with temperature between 20 and 26 ºC and average damp of 75 – 90%. The BF24 strain was found to be the most productive one in relation to the others and the supplementation of the elephant grass with wheat bran in concentrations of 10 and 20% favors higher productiveness and biological efficiency for the BF24, DF33 and HF19 strains of *P. ostreatus.*

Murthy & Manonmani (2008) carried out the study to cultivate *Pleurotus florida* on Coffee industry wastes such as Coffee Cherry husk, Coffee parchment husk, Silver skin, Coffee spent wastes, dried leaves with and without supplementation of agricultural wastes such as wheat bran in different combination. Other physicochemical parameters were also optimized for maximum mushroom yield. *P. florida* is a promising mushroom in terms of its high productivity, simple means of bioconversion on cheap organic substrates such as coffee industry wastes and a short production cycle compared to many other cultivated mushrooms.

Aryal (2003) studied the effect of straw and the mixture of straw and saw dust in the cultivation of *Pleurotus sajor-caju*. Higher production was obtained from the mixture of rice straw and saw dust than only rice straw.

Jwanny *et al*. (1995) studied the technical feasibility of using agricultural wastes (mango and date industry wastes) as a substrate for the cultivation of *Pleurotus ostreatus* NRRL-0366. When comparing the biological efficiency of mushroom production, the highest yield of fruiting bodies was obtained using a mixture of date waste and rice straw at a ratio (1:1) (11.96%), followed by a mixture 3:1 (11.16%). The lowest one was the mixture 2:1 (9.19%). Fungus *Pleurotus ostreatus* NRRL-0366 can also be cultivated on mango waste supplemented with rice straw at a different ratio. The best one was the 1:1 mixture (10.18%), whereas the lowest was a mixture 3:1 (6.4%).

Yildiz *et al*. (2002), yield values, diameters and numbers of fruit bodies obtained from the cultivation of *Pleurotus ostreatus* mushrooms were determined and the effects of different substrate combinations on productivity were investigated. Wastes of some lignocellulosic materials such as leaves of hazelnut (LH, *Corylus avellana, C. maxima, C. colurna*), leaves of

tilia (LT, *Tilia rubra, T. phylatiphyllos* [*T. platyphyllos*]) and leaves of European aspen (LEA, *Populus tremula*), wheat straw (WS), sawdust (S) waste paper (WP) were used for producing *P. ostreatus*. The best main material and the best substrate combination for mushroom productivity were WS and WS+WP (50%+50%) respectively. Mixtures which involve WP generally produced higher yield values. Mixtures which contained bran (25%) increased the risk of contamination. The lowest yield and the smallest fruit body diameters values were obtained from LT (100%) and LEA+S (50%+50%). The greatest number of fruit body was obtained in the combination WS+LH+WP (30%+50%+20%). The largest diameter of fruit body was obtained from OT (100%), even though few fruit bodies were observed.

Ahmed *et al*. (2009) carried out the experiment on *Pleurotus florida* (Mont) Singer on different agro-wastes viz. soyabean straw, paddy straw, wheat straw and their combination in 1:1 proportion to determine the effect of these agro waste on yield, moisture content.

CHAPTER THREE

3. Materials and Methods

3.1 Materials

Different equipments and chemicals were used in performing the research work. All the materials used are listed in Annex A.

3.2 Site of experiment

The research activities of the present study were carried out in the laboratory of Central Department of Botany, Tribhuvan University and Bagbani Development Project, Kritipur.

A field study was conducted in March-April, 2007 in Chapagaon and Balambu villages to analyze the present practice of Oyster mushroom cultivation in Nepal. Information related to mushroom cultivation was obtained from the farmers growing mushroom. Twenty questionnaires were administered to the mushroom growers of local area, (annex D).

3.3. Methodology

3.3.1 Preparation of substrate

The preparation of substrate was based on the dry weight of each component before mixing. The substrate used in this study for the cultivation of Oyster mushroom was straw, saw dust, newspaper and rice husk. The rice straw was chopped into small pieces (2-3 inches long). The newspaper was shredded into small pieces manually. Rice husk and saw dust were used as such as substrate. First all the substrates were individually used. Then newspaper, saw dust and rice husks were combined with straw in the ratio 1:1 (w:w). Then the substrate newspaper was supplemented with rice bran in the ratio 1:9(w:w).

3.3.2 Watering of substrate material

Suitable amount of water should be maintained in the substrate during the whole process of cultivation. The substrates were soaked in the water for 24 hrs to moisten them thoroughly. The excess water was drained off. The "Palm method" was followed. It is used to check whether the substrate mixture has the proper moisture content. First the fistful of substrate is taken and squeezed tightly. If only the palm got wet and no drop of water released, the substrate mixture has the proper water content. If the moisture content is too high, the

substrate is more vulnerable to infection. If the moisture content is too low, the spawn could grow poorly and the harvest quality decline (Kwon & Kim, 2004).

3.3.3 Sterilization

The substrate was placed in the bag and autoclaved at 121°C at 15lbs pressure and allowed to cool overnight.

3.3.4 Bagging and spawning

Bagging and spawning was done simultaneously. The spawn of *Pleurotus florida* was obtained from NARC. The cooled substrate was packed in the plastic bag upto 4 inch deep and grain spawn was sprinkled layers by layers.

5% of *P. florida* was spread uniformly all along the periphery of the bag according to the dry weight of substrates The last layer of spawn was covered with very shallow layer of substrate. The bag was packed well without empty spaces and then the bag opening was tied very tightly using thread. The bags were perforated using scalpel for exhaust of gases.

3.3.5 Spawn run / Incubation

Then these bags were incubated under complete darkness. The bags were arranged on the floor with some spaces between them to prevent overheating. The room temperature during the incubation period was 27-29^{O}C

3.3.6 Fruiting and harvesting

Once the substrate has a colonization of mycelium, cross-slashes were cut to allow the mushrooms to grow out. After the bags turned white due to growth of mycelium, the pinheads appeared which turned into full sized mushroom later on. Although mycelium can grow properly even in total darkness, the primordial formation and fruit body growth requires certain amount of light. The colour of the cap is also related to the light intensity. Fruit bodies cannot grow normally without fresh air. Because most edible mushrooms are 90% water, humidity is critical during the fruiting stage (Kwon & Kim, 2004). So in the productive growth stage frequent watering should be done in order to raise the relative humidity. The bags were watered during cropping with the water sprayer. Harvesting should be done by pulling the mushrooms from the substrate. If they are cut, the cut surface remaining on the substrate is an ideal place for *Trichoderma* (green mold) to enter.

3.3.7 Data recorded

The experiment was laid out in a complete randomized design (CRD) with three replications (Kausar, 1988). The data was analyzed on various aspects. Completion of spawn running: The days required for full colonization of the substrate were recorded.

Appearance of pinheads: The data were recorded for the appearance of pinheads in different substrate.

Maturing of fruiting bodies: The days taken by pinheads to form the mature fruiting bodies in different substrate were recorded.

Harvesting dates and total yield of mushroom: The data were recorded for the harvesting of first, seconds and third crops. The weight of crops in different substrate was noted.

Number of fruiting bodies: The caps of Oyster mushroom is also counted in first, second and third harvest.

3.3.8 Biological efficiency

The mushroom yields for each trial was compared and the Biological efficiency was calculated as indicated below (Royce, 1985).

The formula used for the calculation of Biological Efficiency was

$$\text{Biological efficiency} = \frac{\text{Weight of fresh mushroom harvested}}{\text{Weight of dry substrate used}} \times 100$$

3.3.9 Coefficient of variation

Effort was made to analyze data statistically to observe the variability in weight and number of fruit bodies in different samples of each of the substrate. For computing coefficient of variation the following formula was used.

$$\text{Coefficient of variation} = \frac{\text{Standard deviation}}{\text{Mean}} \times 100$$

CHAPTER FOUR

4. RESULTS

The trial was undertaken in three 500gm samples each of 8 different types of substrate like rice straw, rice husk, paper, saw dust, mixture of rice straw+paper, mixture of rice straw+saw dust, mixture of rice straw+rice husk and mixture of paper+10% bran.

For colonization from inoculation the average days taken was 60.9/8 = 7.61 days, and for primordial formation 109.7/8 = 13.81 days. The highest time taken for colonization was in substrate rice husk and substrate sawdust (9.7 days) and shortest was in substrate

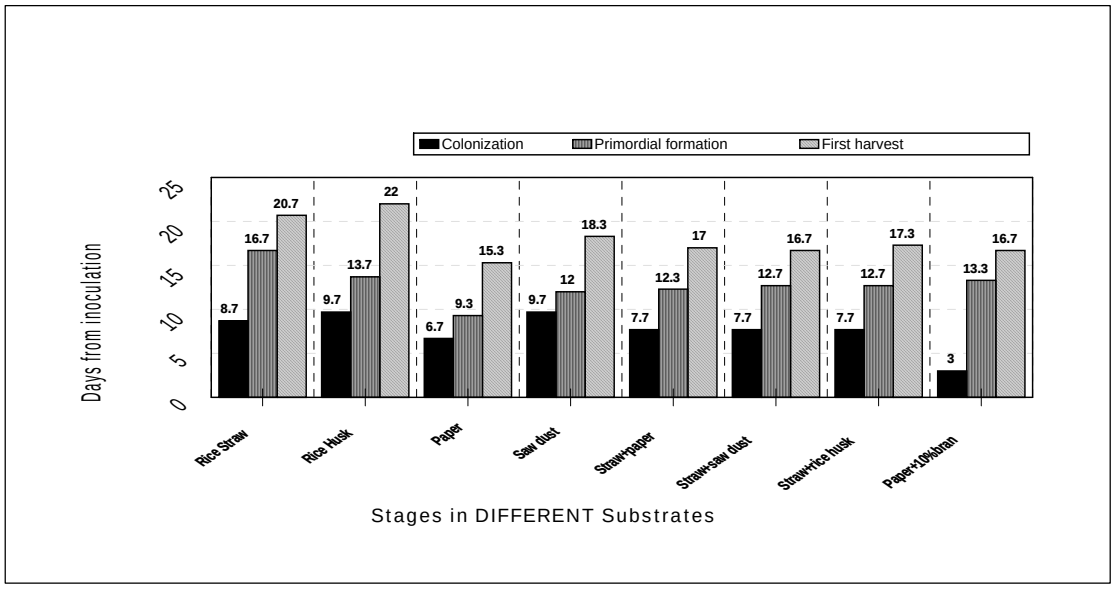

Figure 1. Average days from inoculation to colonization, primordial formation and first harvest in different substrates

paper+10%bran (3 days). For primordial formation the longest time taken was in substrate rice straw (16.7 days) and shortest in substrate paper (9.3 days).

The average time taken for the crop to be ready for harvesting was 144.0/8=18.0 days. The maximum average time taken for the crop to be ready for first harvest was 22 days in rice husk and shortest was 15.3 days in paper (Figure 1). In the substrate rice straw, rice husk, rice straw+paper, straw+saw dust and straw+ rice husk the harvest was done 3 times while in rest that is paper, sawdust and paper+10%bran, the harvest was undertaken only 2 times.

4.1 BY SAMPLE WISE

4.1.1 Yield of mushroom

4.1.1.1 In weight

The total yield of mushroom in all the substrate together was 5,881 gm average being 735.12 gm. Maximum production 1714gm (29% of total) was in substrate rice straw followed by substrate straw + rice husk with the production 974.7 gm (16.6%) and least was 74 gm (1.3 %) in sawdust. It seemed that straw is good to be used as substrate in production of mushroom. The production of mushroom in all the substrate containing straw is higher than those of substrate without rice straw. The substrates straw+paper, straw+sawdust and straw + rice husk gave 916 gm (15.6%), 906 gm (15.4%) and 975 gm (16.6%) respectively of the

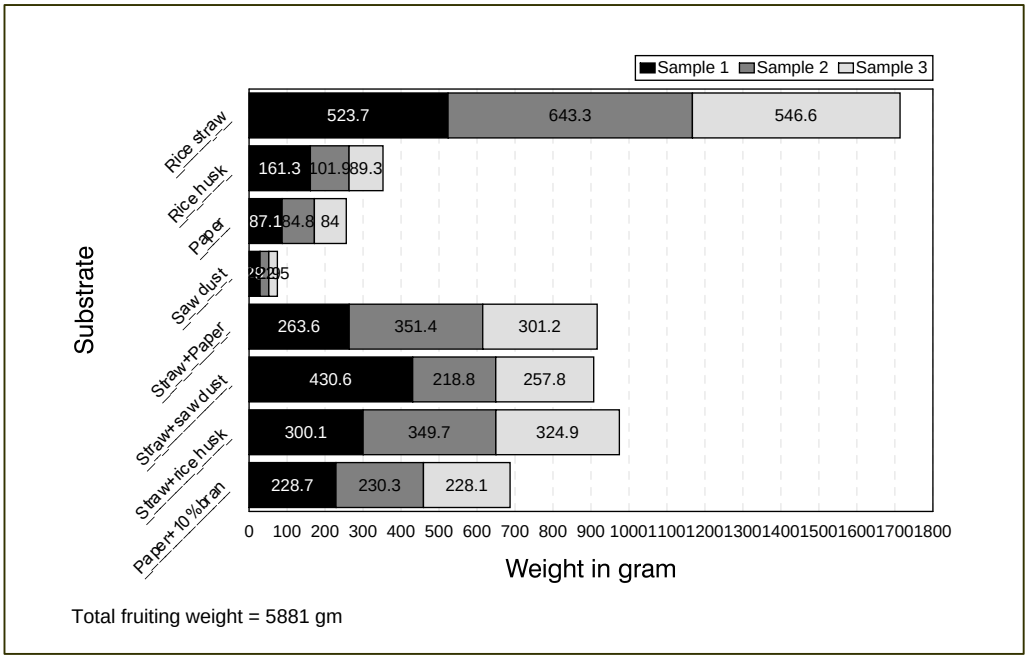

Figure 2. Cultivation of *Pleurotus florida*: Weight of mushroom in gm produced in different substrates

total production, where as in substrates without straw e.g. rice husk, paper, sawdust and paper+10%bran gave 352 gm (6.0%), 256 gm (4.4%), 74 gm (1.3 %) and 687 gm (11.7%) respectively (Figure 2).

4.1.1.2 In number and weight of fruit body

The total number of fruit body in all the 8 substrates was 2,173 average being 271.5. The highest number 422 (19.4%) was in substrate paper+10%bran followed by 403 (18.7 %) in straw+rice husk, 365 (16.8%) in rice straw. The least number of fruit body was 79 (3.6%) in saw dust.

The average weight of fruit bodies harvest is 2.7 gm. The maximum average weight of fruit body was 4.7 gm produced in substrate rice straw followed by straw+sawdust with 3.3 gm, straw+paper with 2.7 gm and straw+rice husk with 2.4 gm respectively.

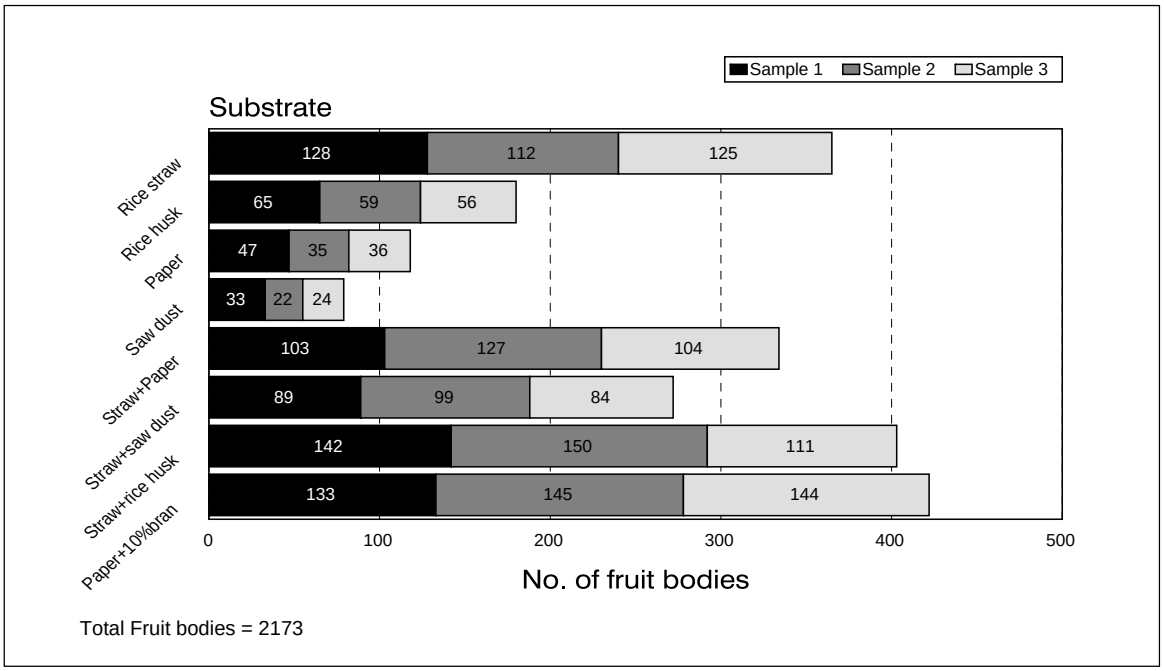

Figure 3. Cultivation of *Pleurotus florida*: Number of fruit bodies produced in 3 different samples of each 8 substrates

The fruit body produce with least average weight 0.9 gm was in sawdust. The maximum number of fruit body (422) with a very small average weight 1.6 gm was in substrate paper+10%bran i.e. only one third the average weight of fruit body produced in substrate rice straw (Figure 3).

Biological efficiency The average percentage of production in 1500 gm of dry substrate was 49%. The highest percentage production was 114.24% (1718 gm from 1500 gm of dry substrate), followed by 64.98% in substrate straw+rice husk. In all the substrates containing rice straw the production on average is always more than 60%, where as all the substrate without rice straw is less than 50%, least 4.96% in saw dust. If sample wise production is considered the highest percentage 128.7% is in second sample of substrate rice straw and least was third sample of saw dust. Among all the substrates, the highest production was in rice straw (114.24%). Although the next highest production was in first harvest of straw+saw dust (86.12%), the average production was only 60.58% i.e. less than that of straw+paper and straw+ rice husk (Figure 4).

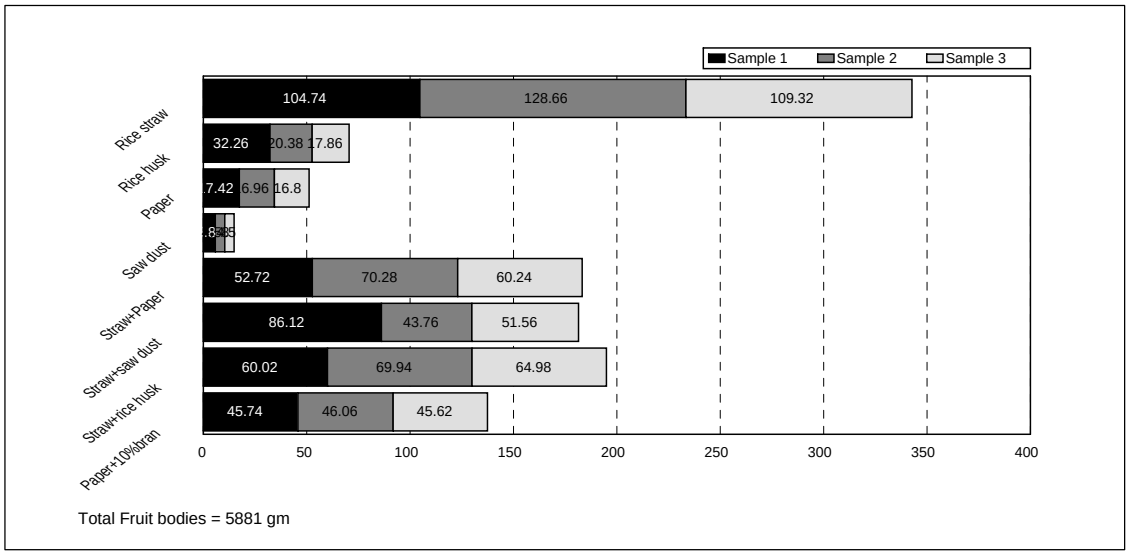

Figure 4. Biological efficiency (%) of mushroom produced in 500 gm of dry substrate

In analysis of variation in production of mushroom in weight, the maximum variation 37.3% was found in substrate straw+saw dust followed by rice husk with variation 32.8%. The minimum variation 0.5% was observed in substrate paper+10%bran followed by substrate paper with 1.9%. It should be noted that in both these substrates no production was there in

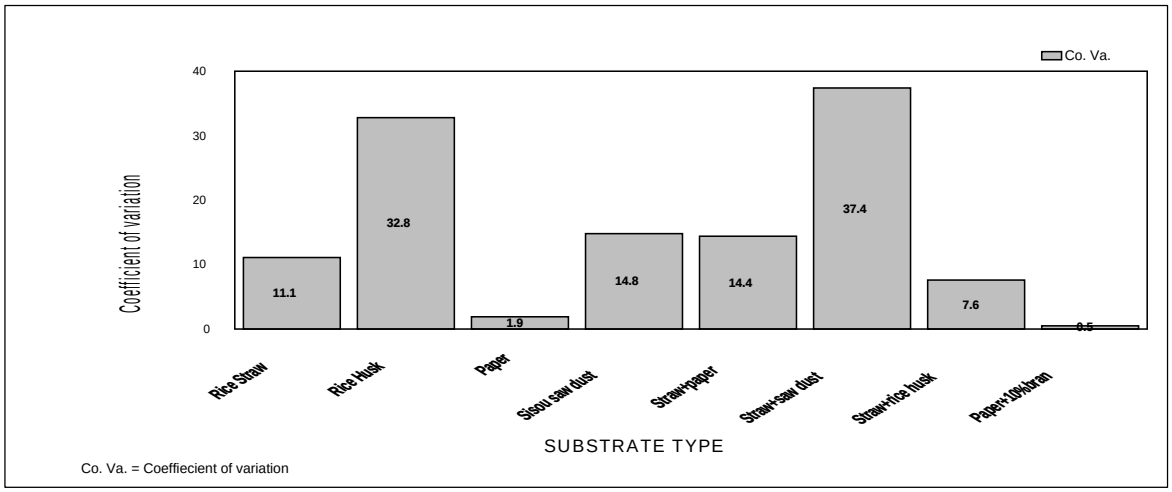

Figure 5. Coefficient of Variation: Production of fruit bodies in weight in Gm in different types of 500 gm of dry substrate.

third harvest. In substrate straw, variation was 11.1%, fourth from lowest variation (Figure 5). Variation study in production of number of fruit bodies in different samples of all the substrate showed maximum variation 22.3% in substrate saw dust followed by paper with 15.6% variation. The lowest variation was 4.7% in substrate paper+10%bran followed by 7% rice straw (Figure 6).

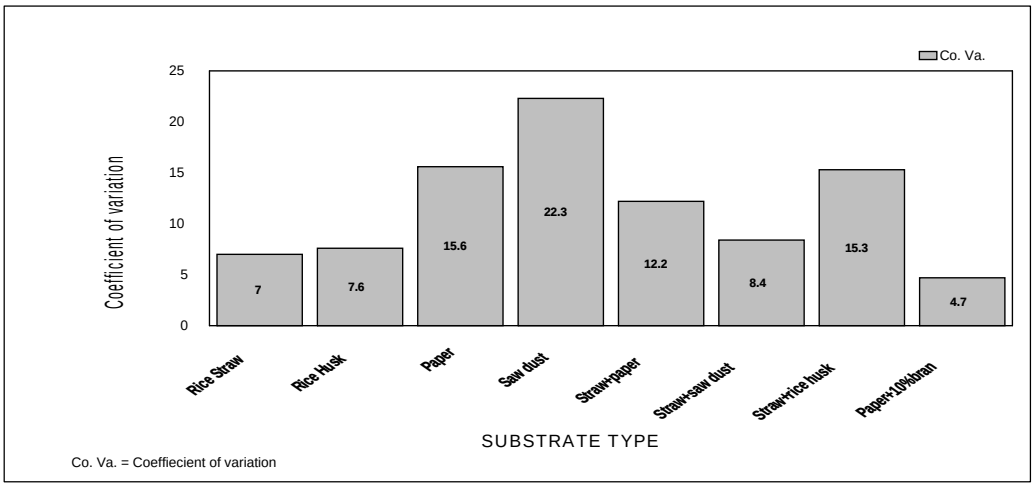

Figure 6. Coefficient of variation: Production of number of fruit bodies in different types of 500 gm of Substrate

4.2 BY HARVEST PERIOD

4.2.1 Yield in mushroom

Out of total production 5881 gm of fruitings, the yield in weight in first harvest period was 3846.6 gm (65.4 %), in second harvest was 1383.7 gm (23.5%) and in third harvest was 651.0 gm (11.1%). In all first, second and third harvest of all the substrates, the highest percentage 25.25%, 34.56% and 40.55% respectively was in substrate rice straw. Lowest production was in first harvest 1.03% in substrate saw dust and in second harvest 1.81% in substrate paper. In third harvest in three substrates namely paper, saw dust and paper+10%bran there was no yield at all. (Figure 7) .If substrate wise in different harvest period is considered the highest percentage of production was 92.5% in substrate paper+10%bran followed by substrate paper

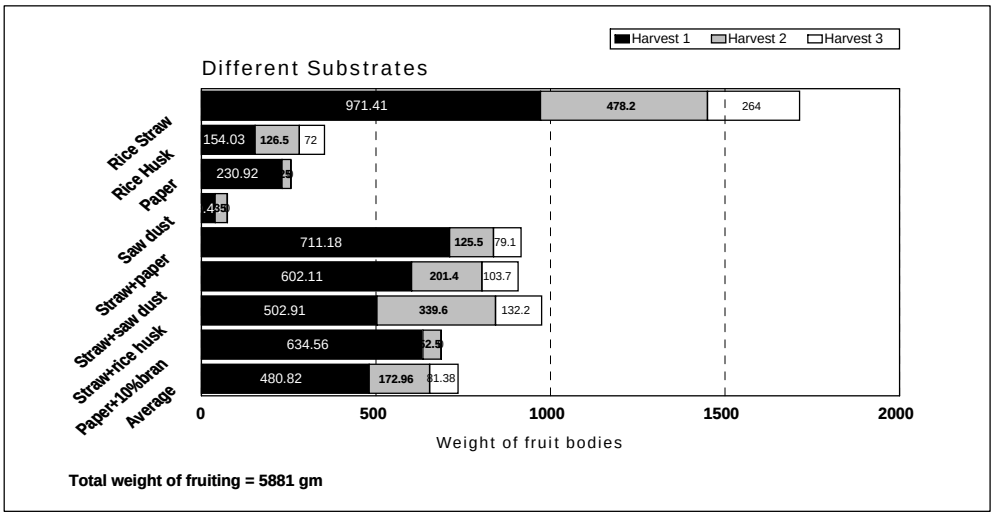

Figure 7. Weight of fruit bodies in different harvest periods and substrates

with 90.2%. In both of these substrates the yield was nil in third/last harvest period. In second harvest period the highest yield 47% was in substrate saw dust followed by rice husk (35.9%)

and straw+rice husk 34.8%. In third harvest period the highest yield, 20.4% was in substrate rice husk followed by rice straw (5.4%).

In the substrate rice straw, rice husk, rice straw+paper, straw+saw dust and straw+rice husk the harvest was done 3 times while in rest that is paper, sawdust and paper+10%bran the harvest was undertaken only 2 times. The maximum yield was obtained in first flush than in second and third.

Effort was made to analyze data on weight of the fruit bodies in different substrates in different harvest period. The average weight of the fruit body as already mentioned earlier is highest, 4.7 gm in substrate rice straw followed by 3.34 gm in substrate straw+saw dust. Lowest was 0.94 gm in substrate saw dust. Except in substrate rice straw and paper+10% bran, in all other substrates the average weight of fruit body is variable by more than 0.5 gm per fruit body in different harvest period. In substrate rice straw which gave heaviest fruit

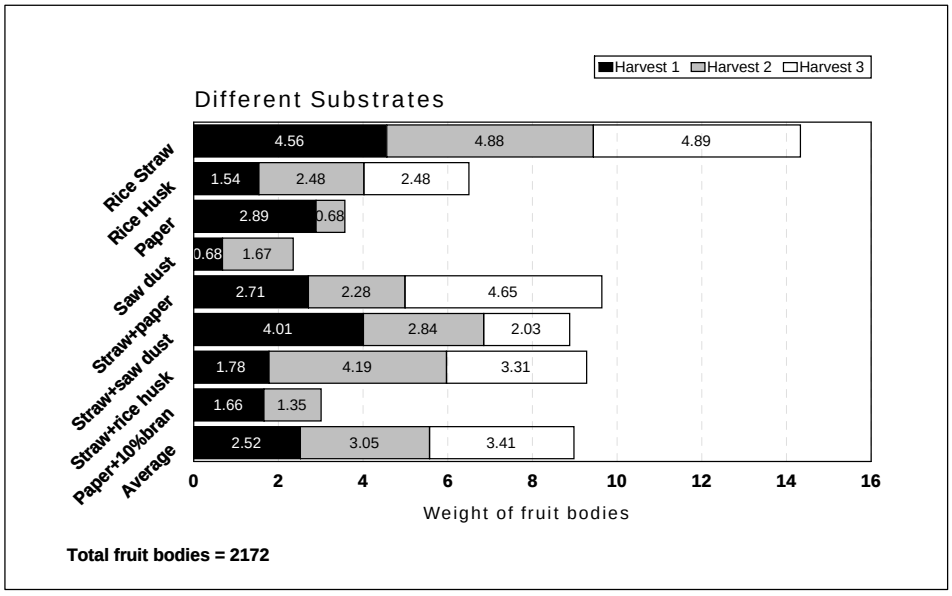

Figure 8. Average weight of fruit bodies in different harvest period and substrates

bodies, the difference in average weight of fruit body in different period is less than 0.33 gm only. In substrate paper+10%bran which produced maximum number of fruit bodies and which did not give fruit bodies in third harvest period, the difference in average weight of fruit body in different harvest period is less than 0.32 gm (Figure 8).

CHAPTER FIVE

5. **Discussion**

Nepal is basically an agricultural country where about 80% of the economically active population is involved in farming. A vast quantity of crop residues that are produced may be used in mushroom production. Nepal also has large number of agro-climatic regions that offer congenial climatic condition for mushroom cultivation.

Oyster mushroom is one example of edible mushrooms that can utilize lignocellulosic materials as substrate. This capability of the oyster mushroom is due to the presence of its lignocellulitic enzymes which help it convert cellulose and lignin into useful carbohydrates such as glucose that can be used as an energy source for the fungi. Any source that contains cellulose and lignin is a possible substrate for growing this fungus (Custodio, 2004).

This study was conducted to determine the efficiency of different agricultural wastes as substrate for growing mushrooms. The maximum yield was given by rice straw when used individually. This may be because the rice straw contained sufficient amount of necessary nutrients for fungal development. Best yield of different *Pleurotus spp* on wheat and paddy straw have been stated by many workers (Bansal & Perker, 2001).

The emergence of mushroom from the bed is purely based on the amount of cellulose present in the substrate (Sivaprakasam & Kandasamy, 1981). The cellulosic substance can be degraded very easily by growing mushroom, whereas non-cellulosic substances are not easily degraded. The degraded substances are used for their metabolic activities. The increase in yield of mushroom in paddy straw is due to easier way of getting of sugars from the cellulosic substances (Ponmurugan *et al*., 2007).

In all the other substrate i.e. paper, rice husk and sawdust the yield of mushroom is low when individually used. According to Kurtzman (2005), paper can be the possible substrate, maybe 100% cellulose, but in mixture that can be the advantage.

The substrate should hold water tightly because we want air and if the water is not held and flows, it will plug the air spaces and the growth will be limited. Soft material used in substrate often pack so tightly that although they hold water tightly, there is still no air

(Kurtzman, 2005) In sawdust and paper due to compactness of the material, which prevented easy aeration and in rice husk due to lack of capability to hold enough water the yield was not maximum.

Substrate containing two or more kinds of plant wastes are recommended. Mixtures generally will allow air spaces, but will pack quite tightly. Straw is among the best substrate for holding air (Kurtzman, 2005).

In this research also when paper, sawdust and rice husk are mixed with rice straw in 1:1 proportion the yield was increased considerably. Das & Mukherjee (2007) also found that when weed plants were mixed with rice straw in the ratio 1:1, there is increase in the yield than when used individually. The increase in yields in the combined substrate might be due to higher water retention capacity of the combined substrate (particularly for rice straw) than that of the individual substrate.

The time taken for the colonization is minimum in paper+10%bran mixture. According to Gabriel (2004) supplementation with additives doesn't increase productivity significantly, but does accelerate mycelial growth by increasing substrate temperature. But in this research, bran seem to contribute in giving more production of mushroom. If paper is considered, paper alone gave 256 gm (4.4 %), straw+paper 916 gm (15.6%) and paper+bran 687 gm (11.7%). The main substrate material alone sometimes cannot provide enough nitrogen required for optimal growth of mushrooms. Additives such as rice or wheat bran provide a nitrogen source (Choi, 2004). Amounts of supplements that should be added varies with the substrate chosen. Oei (2005) suggested a range of 5-10% wheat bran.

Nepalese farmers are growing mushroom in the thatched house or in a plastic tunnel. The plastic tunnels are made up of thick plastic sheets with bamboo support. For pasteurization the farmers are using drum and some are using clay pot (potasi).

Two kinds of mushroom – *Agaricus bisporus* and *Pleurotus spp* (*P. ostreatus and P. sajor-caju*) have been cultivated by local farmers. Chapagaon mushroom farmers prefered *Agaricus bisporus*. *Pleurotus spp* are more popular among most of the farmers in Balambu than *Agaricus* since it is easy to grow, less time consuming and labour cost is less, as there is no need for compost making.

The farmers are cultivating *P. ostreatus* from September to April in Kathmandu. After April it is considered off season for the cultivation of *P. ostreatus.* The farmers donot prefer *P. sajor-caju* which fruit at high temperature as this gives less yield than *P. ostreatus.* They prefer cultivating *P. ostreatus* all round the year. It does not need temperature control in the cold season. Difficulty encountered was providing the proper temperature in summer (off season) for *P. ostreatus* to fruit abudantly when temperature is high. Farmers are cultivating *P. ostreatus* in cooler area in the off season as they can sell it in higher price. According to respondents the contamination is more in the off-season. To prevent contamination they are using different types of pesticides which should not be encouraged. They are using different types of chemicals which are very harmful to our health.

All the farmers are growing mushroom using rice straw as substrate. Some of the farmers have tried to grow *Pleurotus* in locally available substrates like sawdust, mustard pod, and mustard stem. But due to less yields, they did not continue the production in these substrate.

Most of the growers are from the lower middle class to higher middle class family. According to most of the local farmers the mushroom cultivation has helped them to increase their livelihood and if the mushroom farming is done throughout the year, the earned money is sufficient for their living.

In some family it has provided family labour, thus providing all the members of the family with employment.

Some farmers are using the compost that remains after harvesting as manure in other vegetable cultivation.

The mushrooms are very perishable. So after harvesting it has to be used immediately. Farmers do not have the knowledge about the proper storage of the harvested mushroom. The proper storage and canning facilities needs to be developed. So farmers are forced to sell mushroom in less price in season when there is abundant harvest.

Five crops of oyster mushroom can be produced per year. Harvesting can be done three times from the business point of view. Harvesting interval will be 10 to 15 days.

Different types of diseases and pests like green mold, brown blotch, viral, larva of various insects causes serious problem in oyster mushroom cultivation and sometimes causes even crop failure. So precaution should be taken to prevent contamination and diseases. Among various diseases, the green mold disease is the most common.

To prevent contamination, farmers spread limestone (Calcium Carbonate) in the room where the mushroom bags are to be incubated.

Diseases are more frequent in the time of incubation. So in incubation period bags should be observed carefully for the sign of any disease. If there is appearance of any sign of disease in the bag that part should be rubbed with cotton dipped in chemicals like dorasalt, nuan or pure alcohol. If certain portion is affected, that portion should be immediately thrown away. If large part of the bag is affected, that bag should be immediately discarded. It is better to keep the bag in which the disease has been seen in the separate room as it may cause contamination to other packets.

CHAPTER SIX

6. Conclusion and Recommendations

6.1 Conclusion

Rice straw is the most appropriate substrate in the mushroom production. This study has also successfully demonstrated the possibility of using paper, rice husk and sawdust as substrate in mushroom production.

Although the amount of yield is lower than in rice straw, rice husk and paper can be used as the substrate when there is no any other substrate available. When paper, rice husk and sawdust are mixed with straw the yield was increased significantly. So, the pollution of these materials can be decreased by using in oyster mushroom cultivation. The minimum average weight was in sissoo sawdust and in paper+bran. Maybe mushroom from these substrate will not be favoured by growers and consumers due to its small cap and stalk.

Mushroom cultivation could possibly offer solution for poverty alleviation. Mushroom growing is labour intensive. As in our country the jobs are scare, mushroom growing can create jobs. Unlike other agronomic crops, the set up cost production are low. Fertilizers and machinery are not used.

Production of mushrooms might help to improve the malnutrition problem that continues to exit in the world. The mushroom cultivation technique can pave the way for the future food for human beings.

6.2 Recommendations

1. Further research should be carried out in above substrates for the commercial production.
2. The effect of different nitrogen supplement on these substrates has to be determined.
3. Proper training should be given to mushroom farmers. Government should financially and technically support them to grow mushrooms by themselves.

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Annex A

Materials used for the study

- Autoclave
- Electronic balance
- Simple balance
- Autoclavable disposable bag
- Hot air oven
- Scalpel
- Thread
- Spawn
- Rice straw
- Sawdust (Sissoo)
- Rice husk
- Newspaper
- Rice bran
- Plastic sheet and bag
- Sprit
- Scissors
- Field note book

Data record: Cultivation of *Pleurotus florida* in substrate: RICE STRAW

Substrate		Inoculation	Colonization		Primordial formation	
Sample no.	Wt. in gm	Date	Date	Days from inoculation	Date	Days from inoculation
1	**500**	18-Aug-08	25-Aug-08	7	2-Sep-08	15
2	**500**	18-Aug-08	26-Aug-08	8	4-Sep-08	17
3	**500**	8-Sep-08	19-Sep-08	11	26-Sep-08	18
Average	**500**			**8.67**		**16.67**

Sample 1						
Harvest no	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
1	7-Sep-08	20	69	295.47	523.65	164.55
2	17-Sep-08	30	38	140.50		
3	5-Oct-08	48	21	87.68		

Sample 2						
Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
1	7-Sep-08	20	70	385.60	643.28	214.43
2	17-Sep-08	30	31	199.40		
3	29-Sep-08	42	11	58.28		

Sample 3						
Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
1	30-Sep-08	22	74	290.34	546.63	182.21
2	10-Oct-08	32	29	138.30		
3	20-Oct-08	42	22	117.99		

Data record: Cultivation of *Pleurotus florida* in substrate: RICE HUSK

Substrate		Inoculation	Colonization		Primordial formation	
Sample no.	Wt. in gm	Date	Date	Days from inoculation	Date	Days from inoculation
1	**500**	27-Aug-08	7-Sep-08	11	11-Sep-08	15
2	**500**	27-Aug-08	5-Sep-08	9	9-Sep-08	13
3	**500**	27-Aug-08	5-Sep-08	9	9-Sep-08	13
Average	**500**			**9.67**		**13.67**

Sample 1

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	24-Sep-08	28	39	88.37	161.33	53.78
SECOND	30-Sep-08	34	19	48.08		
THIRD	19-Oct-08	53	7	24.88		

Sample 2

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	15-Sep-08	19	30	36.84	101.86	33.95
SECOND	29-Sep-08	33	15	35.06		
THIRD	15-Oct-08	49	14	29.96		

Sample 3

harvest No	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	15-Sep-08	19	31	28.82	89.27	29.76
SECOND	29-Sep-08	33	17	43.33		
THIRD	15-Oct-08	49	8	17.12		

Data record: Cultivation of *Pleurotus florida* in substrate: PAPER

Substrate		Inoculation	Colonization		Primordial formation	
Sample no.	Wt. in gm	Date	Date	Days from inoculation	Date	Days from inoculation
1	**500**	4-Aug-08	10-Aug-08	6	13-Aug-08	9
2	**500**	4-Aug-08	11-Aug-08	7	14-Aug-08	10
3	**500**	5-Aug-08	12-Aug-08	7	14-Aug-08	9
Average	**500**			**6.67**		**9.33**

Sample 1

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt in gm	Total production	Mean production
FIRST	19-Aug-08	15	29	78.12		
SECOND	23-Aug-08	19	17	9.00	87.12	43.56
THIRD						

Sample 2

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	20-Aug-08	16	25	76.80		
SECOND	24-Aug-08	20	10	8.00	84.8	42.4
THIRD						

Sample 3

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	20-Aug-08	15	26	76.00		
SECOND	25-Aug-08	20	10	8.00	84	42
THIRD						

Data record: Cultivation of *Pleurotus florida* in substrate: SAW DUST

Substrate		Inoculation	Colonization		Primordial formation	
Sample no.	Wt. in gm	Date	Date	Days from inoculation	Date	Days from inoculation
1	**500**	21-Aug-08	31-Aug-08	10	2-Sep-08	12
2	**500**	21-Aug-08	31-Aug-08	10	3-Sep-08	13
3	**500**	22-Aug-08	31-Aug-08	9	2-Sep-08	11
Average	**500**			**9.67**		**12**

Sample 1

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	8-Sep-08	18	25	16.60	29.03	14.52
SECOND	24-Sep-08	34	8	12.43		
THIRD						

Sample 2

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	10-Sep-08	20	13	8.82	22.87	11.44
SECOND	27-Sep-08	37	9	14.05		
THIRD						

Sample 3

Harvest no	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	8-Sep-08	17	20	14.02	22.52	11.56
SECOND	24-Sep-08	33	4	8.50		
THIRD						

Data record: Cultivation of *Pleurotus florida* in substrate: STRAW PLUS PAPER

Substrate		Inoculation	Colonization		Primordial formation	
Sample no.	Wt. in gm	Date	Date	Days from inoculation	Date	Days from inoculation
1	**500**	27-Aug-08	3-Sep-08	7	9-Sep-08	13
2	**500**	3-Sep-08	11-Sep-08	8	15-Sep-08	12
3	**500**	3-Sep-08	11-Sep-08	8	15-Sep-08	12
Average	**500**			**7.67**		**12.33**

Sample 1

Harvest no.	Date of Harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	15-Sep-08	19	88	207.72		
SECOND	3-Oct-08	37	9	29.63	263.57	87.86
THIRD	14-Oct-08	48	6	26.22		

Sample 2

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	19-Sep-08	16	88	253.13		
SECOND	15-Oct-08	42	31	64.54	351.36	117.12
THIRD	24-Oct-08	51	8	33.39		

Sample 3

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	19-Sep-08	16	86	250.33		
SECOND	15-Oct-08	42	15	31.37	301.2	100.4
THIRD	24-Oct-08	51	3	19.50		

Data record: Cultivation of *Pleurotus florida* in substrate: STRAW PLUS SAW DUST

Substrate		Inoculation	Colonization		Primordial formation	
Sample no.	Wt. in gm	Date	Date	Days from inoculation	Date	Days from inoculation
1	**500**	22-Aug-08	29-Aug-08	7	4-Sep-08	13
2	**500**	3-Sep-08	11-Sep-08	8	15-Sep-08	12
3	**500**	8-Sep-08	16-Sep-08	8	21-Sep-08	13
Average	**500**			**7.67**		**12.67**

Sample 1

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	8-Sep-08	17	45	275.52	541.52	180.51
SECOND	17-Sep-08	26	30	125.92		
THIRD	5-Oct-08	44	14	29.2		

Sample 2

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt in gm	Total production	Mean production
FIRST	19-Sep-08	16	56	133.13	218.81	72.94
SECOND	29-Sep-08	26	21	39.79		
THIRD	15-Oct-08	42	22	45.89		

Sample 3

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	25-Sep-08	17	49	193.46	257.76	85.92
SECOND	5-Oct-08	27	20	35.68		
THIRD	20-Oct-08	42	15	28.62		

Data record: Cultivation of *Pleurotus florida* in substrate: STRAW PLUS RICE HUSK

Substrate		Inoculation	Colonization		Primordial formation	
Sample no.	Wt. in gm	Date	Date	Days from inoculation	Date	Days from inoculation
1	**500**	27-Aug-08	4-Sep-08	8	9-Sep-08	13
2	**500**	3-Sep-08	11-Sep-08	8	15-Sep-08	12
3	**500**	8-Sep-08	15-Sep-08	7	21-Sep-08	13
Average	**500**			**7.67**		**12.67**

Sample 1

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	15-Sep-08	19	75	136.12	300.13	100.04
SECOND	25-Sep-08	29	52	114.43		
THIRD	15-Oct-08	49	15	49.58		

Sample 2

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	19-Sep-08	16	125	222.73	349.7	116.57
SECOND	29-Sep-08	26	13	87.3		
THIRD	20-Oct-08	47	12	39.67		

Sample 3

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	25-Sep-08	17	82	144.06	324.89	108.30
SECOND	5-Oct-08	27	16	137.86		
THIRD	20-Oct-08	42	13	42.97		

Data record: Cultivation of *Pleurotus florida* in substrate: PAPER PLUS 10% BRAN

Substrate		Inoculation	Colonization		Primordial formation	
Sample no.	Wt. in gm	Date	Date	Days from inoculation	Date	Days from inoculation
1	**500**	5-Aug-08	8-Aug-08	3	18-Aug-08	13
2	**500**	5-Aug-08	8-Aug-08	3	18-Aug-08	13
3	**500**	5-Aug-08	8-Aug-08	3	19-Aug-08	14
Average	**500**			**3**		**13.33**

Sample 1

Harvest no.	Date of Harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	21-Aug-08	16	122	211.74	321.74	160.87
SECOND	30-Aug-08	25	11	16.94		
THIRD						

Sample 2

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	21-Aug-08	16	132	213.27	230.29	115.15
SECOND	30-Aug-08	25	13	17.02		
THIRD						

Sample 3

Harvest no.	Date of harvest	Days from inoculation	No. of fruiting	Total wt. in gm	Total production	Mean production
FIRST	23-Aug-08	18	129	209.55	228.13	114.07
SECOND	2-Sep-08	28	15	18.58		
THIRD						

Annex C

Table Showing Number and weight of fruiting bodies in different harvest period in different samples of each different substrates

Cultivation of *Pleurotus florida*:
Number of sample and their size, date of inoculation, days of colonization, primordial formation, harvest from the date of inoculation, weight of fruiting in different harvest period in three different samples of each substrate.

Substrate				Days from inoculation		First harvest		Second harvest		Third harvest			
Name	Wt. in gm	Sample no.	Date of inoculation	Colonization	Primordial formation	Days from inoculation	Wt. in gm	Days from inoculation	Wt. in gm	Days from inoculation	Wt. in gm	Total weight in gm	Yield % in gm of substrate
Rice straw	500	1	18-Aug-08	7	15	20	295.5	30	140.5	48	87.7	523.7	104.7
	500	2	18-Aug-08	8	17	20	385.6	30	199.4	42	58.3	643.3	128.7
	500	3	8-Sep-08	11	18	22	290.3	32	138.3	42	118.0	546.6	109.3
Average	500			8.7	16.7	20.7	323.8	30.7	159.4	44.0	88.0	571.2	114.2
Rice husk	500	1	27-Aug-08	11	15	28	88.4	34	48.1	53	24.9	161.3	32.3
	500	2	27-Aug-08	9	13	19	36.8	33	35.1	49	30.0	101.9	20.4
	500	3	27-Aug-08	9	13	19	28.8	33	43.3	49	17.1	89.3	17.9
Average	500			9.7	13.7	22.0	51.3	33.3	42.2	50.3	24.0	117.5	23.5
Paper	500	1	4-Aug-08	6	9	15	78.1	19	9.0	No fruiting bodies		87.1	17.4
	500	2	4-Aug-08	7	10	16	76.8	20	8.0			84.8	17.0
	500	3	5-Aug-08	7	9	15	76.0	20	8.0			84.0	16.8
Average	500			6.7	9.3	15.3	77.0	19.7	8.3			85.3	17.1
Saw dust	500	1	21-Aug-08	10	12	18	16.6	34	12.4	No fruiting bodies		29.0	5.8
	500	2	21-Aug-08	10	13	20	8.8	37	14.1			22.9	4.6
	500	3	22-Aug-08	9	11	17	14.0	33	8.5			22.5	4.5
Average	500			9.7	12.0	18.3	13.1	34.7	11.7			24.8	5.0
Straw+ paper	500	1	27-Aug-08	7	13	19	207.7	37	29.6	48	26.2	263.6	52.7
	500	2	3-Sep-08	8	12	16	253.1	42	64.5	51	33.4	351.1	70.2
	500	3	3-Sep-08	8	12	16	250.3	42	31.4	51	19.5	301.2	60.2
Average	500			7.7	12.3	17.0	237.1	40.3	41.8	50.0	26.4	305.3	61.1
Straw+ saw dust	500	1	22-Aug-08	7	13	17	275.5	26	125.9	44	29.2	430.6	86.1
	500	2	3-Sep-08	8	12	16	133.1	26	39.8	42	45.9	218.8	43.8
	500	3	8-Sep-08	8	13	17	193.5	27	35.7	42	28.6	257.8	51.6
Average	500			7.7	12.7	16.7	200.7	26.3	67.1	42.7	34.6	302.4	60.5
Straw+ rice husk	500	1	27-Aug-08	8	13	19	136.1	29	114.4	49	49.6	300.1	60.0
	500	2	3-Sep-08	8	12	16	222.7	26	87.3	47	39.7	349.7	69.9
	500	3	8-Sep-08	7	13	17	144.1	27	137.9	42	43.0	324.9	65.0
Average	500			7.7	12.7	17.3	167.6	27.3	113.2	46.0	44.1	324.9	65.0
Paper+ 10%bran	500	1	5-Aug-08	3	13	16	211.7	25	16.9	No fruiting bodies		228.7	45.7
	500	2	5-Aug-08	3	13	16	213.3	25	17.0			230.3	46.1
	500	3	5-Aug-08	3	14	18	209.6	28	18.6			228.1	45.6
Average	500			3.0	13.3	16.7	211.5	26.0	17.5			229.0	45.8

Annex D

Questionnaire

Date:

Name:

Address:

Sex:

Age:

Economic status:

Since when did you started mushroom cultivation?

Do you have any training in mushroom cultivation?

Is the earned money sufficient for the family?

Has your living standard increased?

Where did you get the spawn?

How much spawn is used?

What substrate do you use for mushroom cultivation?

When do you cultivate the mushroom?

- all year round
- once in a year

When do you cultivate mushroom?

What are the most preferred species?

What are the difficulties faced during mushroom cultivation?

What are the precautions taken to prevent contamination?

What are the common diseases?

Do you have any recommendation to concerned authorities to enhance mushroom production?