Mushroom Research, Production, and Technology Transfer at Haramaya University

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Introduction

- Malnutrition is one of the major factors responsible for high mortality and morbidity in Ethiopia
- Animal protein is beyond the reach of low income group, which
 forms a large proportion of our population. Therefore,
 mushrooms, when grown with low cost technology at farmers
 level, will be an excellent source of protein to the rural poor.
- The great value in promoting the cultivation of mushrooms lies in their ability to grow on cheap carbohydrate materials and to transform various waste materials, which are inedible by man into a highly valued food protein for direct human consumption. This is extremely important in the rural areas where there is enormous quantity of wastes that have been found to be ideal as growing substrates for mushrooms.
- The spent compost, the substrate left over after mushroom harvesting, can be converted to stock feed and plant fertilizer as soil conditioner

Table1: Analysis of some commonly used mushrooms (Percent on fresh weight basis)

Mushroom <i>Agaricus</i>	Moisture	Ash	Protein	Fat	Crude Fibre	Carbo- hydrate V	Energy /alue (K cal)
bisporus Pleurotus	89.5	1.26	3.94	0.19	0.09	6.28	34.4
flabellatus Volvariella	90.9	0.97	2.78	8.65	1.08	5.33	24.4
diplasia	90.4	1.10	3.90	0.25	1.67	5.51	29.2

On dry matter basis, protein content of mushrooms ranges between 21-30 percent. Accordingly they are rich in proteins as compared to cereals, pulses, fruits (apple 0.3%, banana 0.8%, grape 1%) and vegetables (cabbage 1.4%, carrot 1%, potato 1.8% and tomato 1%).

Table 2: Comparison of amino acid composition of edible mushrooms

Amino acids	FAO reference protein	Content mg/g of corrected protein nitrogen in mushrooms			
		P.ostreatus	A.bisporus	V.diplasia	
Isoleucine	250	366	491	267	
Leucine	440	580	312	610	
Lysine	340	527	384	287	
Cystine+Methionine	220	197	285	126	
Phenylalanine+Tyrosi	ne 380	626	580	422	
Threonin	250	366	375	290	
Tryptophan	60	143	98	87	
Valine	310	420	607	326	

Introduction (contd.)

- Mushroom technology can go to rural poor and marginal farmers in a short period of time and bring about significant impact. It can be full time occupation to a sizeable section of poor and marginalized farmers including women and unemployed youth once they are trained. At the same time, it need not to be a main business for many other farms e.g. farmers of cereal and horticultural plantations can also run mushroom cultivation as a sideline business. In this way, they can get from their farm wastes high quality protein food for themselves, or they might sell them in local markets and get some money for other foods.
- Presently there should not be overemphasis on its large-scale development for export over domestic consumption, but there is need to recognize the significance and over all benefits of this small scale business to the local people. Because of the fact that the people of the developing countries like Ethiopia eat too little protein, the significant role of mushroom cultivation in these areas becomes very obvious.

Introduction (Contd.)

- Mushroom cultivation involves a number of different operations
 - Preparation of pure culture
 - Spawn substrate and preparation of spawn
 - Compost materials and composting
 - Seeding and spawn running
 - Mushroom harvesting and processing
 - the spent compost, the substrate left over after mushroom harvesting, can be converted to stock feed, plant fertilizer and soil conditioner.

Introduction (Contd.)

- The main handicap with the farmers is the hygienic conditions required for spawn production and the lack of training in mushroom production technology.
- Haramaya University, through mushroom project, aims to give strong support to mushroom business in rural areas by providing good spawn and training in mushroom cultivation to rural poor, unemployed youth and marginal farmer population, particularly to women who have, many a times, more than fifty percent participation in farming in Ethiopia.

Introduction (Contd.)

Activities undertaken by HU on production of mushrooms for demonstration purposes are as under:

1. In laboratory under strict hygienic conditions

Spore/Tissue culture → Stock culture → Spawn substrate → Spawn

2. HU with participating and trainee farmers are involved in:

Prepared compost → Pasteuriztion → Spawning → Spawn running → Harvesting

The success of mushroom production depends on how it is organized and managed. However, under the current situation prevailing in Ethiopia with regard to the mushroom cultivation, the best option is to supply spawn through HU Spawn Laboratory to small and marginal farmers who would then grow the crop in their own substrate (Compost) or the compost supplied by HU. The ultimate aim of the HU Mushroom Research, Production and Training Laboratory is to encourage and give incentive to farmers, unemployed youth and other economically backward families for setting units in villages

Objectives

The activities were therefore taken up to meet following objectives:

- Selection of edible mushroom species
- Identification and selection of suitable media for spawn making,
- Selection of organic substrates for growing mushrooms and their processing
- The improvement of bed caring and casing materials for button mushrooms

Objectives (Contd.)

- Production of mushroom spawn (Seed) for distribution among farmers and mushroom growers
- Small scale production of Button (Agaricus bisporus), Oyster (Pleurotus sp.) for research, local sale, demonstration and training purposes.
- Production of substrate (readymade compost) and it's supply to mushroom growers

Objectives

- Training of farmers (both male and female), unemployed youth, small scale agriculture entrepreneurs on mushroom growing systems; selection of growing containers; spawning and spawn-run; supplementation before casing; casing; controlling temperature, relative humidity and carbon dioxide concentration; watering of bags/beds; harvesting; care after picking and cooking-out of crop rooms.
- Specialized training in spawn production is imparted to the interested farmers and entrepreneurs. The participants will be trained in methods of raising pure culture, preparation of master/stock culture, multiplication of spawn from stock culture and quality control measures to be followed for getting a good spawn.

Isolation and selection of exotic and indigenous edible mushroom species

 During the last 4 years, extensive surveys of various locations in Western Harraghe region conducted to collect indigenous mushroom species from farmers fields and forest areas. The locations were: Ades forest areas, Asbetefari, Bedessa (Odda Robba), Chelenko Lolla, Chiro (Sorro area), Darolebu (Mechara), Dobba (Oda Jalela PA), Habro (Nursery site, Bako PA, Sukai PA, Hajie Kutte and Melkabelo PA), Hirna (AU Research Station and Kirekfies PA), Kerssa (Toma), Kunni (Arbarekete PA, Bereda PA and Welenbo, Sororo), Kobo, Kara Ude, Lange (Yabeta, Lencha), Wacho (Bereda).

Isolation and survey (Contd.)

- Throughout the region people, even small children, were harvesting edible mushroom species from farms or forest.
- Mushrooms appear in first few weeks of rainy season in shaded areas of forest or farms where there was enough organic matter and picked up for home consumption.
- Farmers want to learn technology that will enable them to cultivate mushrooms throughout the year.
- The mushrooms are picked when they were just appearing out of the soil as older heads become fibrous and unfit to eat.
- Only one species of edible mushrooms, with some variation in the size of mushroom head, was prevalent in the entire region.
- Two commercial strains of Agaricus bisporus (Button mushroom) and four strains of Pleurotus sp (Oyster mushroom) were obtained from Mushroom Research and Training Centre, G. B. Pant University of Agriculture and Technology, Pantnagar, India and commercial spawn produced in Thailand and Germany. The spawn prepared from these species were used for various activities under the project.

Starting materials for growing mushrooms and preparation of substrate

- Mushrooms being heterotrophic, their nutrition from substrates is popularly known as 'compost'. Traditionally compost is made from horse manure. However one of the major activity at HU has been the selection and use alternative organic materials such as wheat, maize, barley, teff, bean and sorghum straws; discarded chat leaves; saw dust; lantana, eucalyptus, acacia and parthenium leaves and dried water hyacinth for growing *Pleurotus* species.
- Similarly wheat, sorghum, maize and barley straws were evaluated for preparation of compost for Agaricus species.

Button Mushroom (Agaricus bisporus)

Composting for mushroom, depending on the type of straw, took 25-35days to complete. During this period, the compost was turned over 8-10 times. Following composition was used for making compost:

Wheat/Sorghum/Maize/Barley straw	100 kg
Ammonium sulphate or Calcium ammonium nitrate	3 kg
Super phosphate	1 kg
Muriate of potash	1 kg
Urea	1 kg
Gypsum	10 kg
Wheat bran	5 kg

Compost Preparation

Day 0

To the straw (75% moisture), wheat bran, Calcium ammonium nitrate, urea, super phosphate and muriate of potash were mixed. With the help of wooden planks, a pile of the straw measuring 1.0 W X 1.0 H X 1.5 L m was made. The planks were removed after making the pile. The temperature of the pile reached 70-75°C within 24 hours. Water was sprayed on the outer surface of the pile once or twice a day.

Day 6

On the sixth day, the outer 15 cm of straw from all around the pile was removed and put in one place. Similarly the remaining inner portion of straw was collected in another place. Now with the help of wooden planks, compost pile was made again by keeping the straw from outer portion inside and inner portion outside the pile.

Composting (Contd.)

Day 10

Inner and outer portions of the pile were separated. The outer portion of the pile was moistened with water and the pile was made by putting the outer portion inside and the inner portion outside.

Day 13

Outer and inner portions were separated and the pile was made after mixing gypsum in both the portions.

Day 16, 19 and 22

Outer and inner portions were turned over as before, and the pile was made.

Day 25

Compost made with wheat or barley straw became ammonia free on this day and was ready for seeding (Spawning). Piles containing maize and sorghum straw were once more turned over on 28th and 30th day respectively to make them ammonia

Oyster Mushroom (Pleurotus species)

- Straw shredded to 4-5 cm was taken in a water storage container, and formalin was mixed to make 1.5% solution. The shredded straw was filled in gunny bags, and soaked in formalin solution for 18 hrs.
- Then the bags were allowed to drain the excess solution.
- One kg of dry straw of wheat or barley usually gave about 4 kg of wet straw for seeding.
- Organic wastes such as lantana, eucalyptus, parthenium and dried water hyacinth leaves were also tested for growing Pleurotus sp.
- Lantana, parthenium and eucalyptus, probably, due to presence of certain antifungal agents did not encourage growth of mushroom mycelium.
- Water hyacinth imbibed lot of water during soaking which gave a pulp like mass resulting in low oxygen exchange and soon the mycelium died due to anaerobic conditions.

Selection of suitable media for spawn making

- Spawn production: Spawn is a medium impregnated with mushroom mycelium and serves as the 'seed' or 'inoculum' for mushroom cultivation.
- Preparation of master/stock culture: Pure culture was inoculated into a suitable substrate. Wheat, maize, teff and sorghum were examined for this purpose.
- Ten kg grains were boiled in 15 I water for 10-15 minutes and then allowed to soak for another 20-25 minutes without heating. Water was then drained off.
- Next day 120 g gypsum and 30 g lime were mixed in 9 kg of boiled grain to prevent clumping and adjust the pH to 6.5-6.7. The grains were filled to 2/3rd capacity autoclavable glass bottles.
- After plugging, bottles were sterilized at 1kg/cm² steam pressure for ½ hr. Autoclaved bottles were shaken to avoid clumping and then allowed to cool.
- Bottles were inoculated with bits of agar medium colonized with pure culture and incubated at 25°C for 7 days. Bottles were then again shaken to mix the mycelium with grains. Two weeks after inoculation, bottles were ready as stock for further multiplication of spawn.

Multiplication of spawn

- Substrate remained the same and spawn was also prepared employing the same technique as for preparation of stock culture.
- After inoculation and incubation at 25°C the bottles were ready for use in 2 weeks. Incubation time was shortened by shaking the bottles or bags during growth of the mycelium.
- It was observed that incubation time was shorter by 3-4 days with wheat as compared to sorghum. During incubation, bottles were shaken at least twice to distribute and spread the mycelium evenly.

Activity 4: Selection of casing materials for button mushroom production

Casing Soil: Casing is the covering of a suitable material placed on the compost surface to induce the fruit body formation of button mushrooms. A variety of materials like soil, spent compost, farm yard manure, vermi-compost, burnt rice husk could be used as casing material. Suitability of these materials and their combinations will depend on their water holding capacity, pH, nutritional level and texture.

Materials of following composition were evaluated for suitability as casing soil:

- 1. Animal manure (2yrs old) + Garden soil (3:1)
- 2. Animal manure (2yrs old) + Garden soil (2:1)
- 3. Animal manure (2yrs old) + Vermi compost (1:1)
- 4. Animal manure (2yrs old) + Vermi compost (2:1)

Casing material (Contd.)

Pasteurization of casing soil

On a pile of covering soil 4% solution of formalin was mixed and then the pile was covered with a polythene sheet.

The material was turned everyday for 3-4 days so that the casing soil became free from the odour of formalin.

Casing soil could also be treated with steam at 65°C for 6-8 days, however the method was not found suitable from the point of view of farmers.

Use of casing soil

A 4 cm layer of different casing soil combinations were spread over different types of seeded composts for button mushroom The compost was sprayed with a 2% solution of formalin after application. Water was sprayed over the casing soil once or twice a day.

Standardization of techniques for production of edible mushroom species

Button mushroom:

The compost using wheat/sorghum/maize/barley straw was spread on a clean cemented floor/polythene sheet was inoculated with 500 g (@ 0.5%) of spawn per 100 kg of compost.

The compost mixed with spawn was filled in polythene bags @ 10 kg per bag. The bags were closed after seeding and were kept 10-15 cm apart on wooden racks in the crop room.

Water was sprayed over the bags and also on the floor and walls of the crop room at least twice a day.

Six to seven days after seeding, threadlike growth of mushroom mycelium was seen which covered the whole surface of compost in 15-22 days.

The compost prepared with wheat recorded the fastest growth followed by barley, maize and sorghum respectively.

Button mushroom production (Contd.)

- At this stage, the compost surface was covered with casing soil
 Mushroom heads started coming after 18-24 days of casing.
- The mushrooms grown on wheat or barley were ready for harvest in 18-20 days whereas those grown on sorghum or maize were harvested between 21-24 days.
- The mushrooms were harvested by twisting between the fingers. One hundred kg of compost of wheat straw yielded 14 kg mushrooms, however, composts made of barley, maize and sorghum straw gave 13.5, 13 and 12 kg mushrooms per 100 kg compost.
- Various casing materials did not differ in terms of yield on different composting materials.

Production technology for oyster mushrooms

Oyster Mushroom (Pleurotus sajor-caju)

- The substrate using wheat/ barley/ sorghum/ maize straw for growing oyster mushroom was prepared as per procedure mentioned before
- The wet straw was spread on floor and mixed with spawn @ 2%
- The spawn mixed with straw was filled in polythene bags @ 4-6 kg per bag. Eight to ten holes of approximately 1cm radius were made all around the bag.
- The seeded bags were kept 15-20 cm apart on wooden racks in the crop room.
- The walls and floor were sprayed with water to keep the temp. around 25°C.
- In a period of 20-25 days, the mushroom mycelium covered whole straw in the bag. There was no difference among various straws in terms of rate of growth of mycelium.

Production technology for oyster mushrooms (Contd.)

- When mycelium had fully covered the straw, the bags were cut, and the mycelium covered straw was kept again on the racks as before.
- The piles were sprayed with water twice a day. After 4-5 days pin heads appeared and in next 4-6 days oyster shaped mushrooms were ready for harvesting which were plucked by twisting between the thumb and fingers.
- With the help of a knife, the lower portion of mycelium covered straw was removed after first harvest. Mushrooms were allowed to grow again on the remaining straw.
- Mushrooms were harvested for second time from the same material after 10 days and then third time after another 6 days. Thus 1 kg of dry straw yielded 300-400 g fresh mushrooms. There was no difference among different straws in terms of yield of oyster mushrooms.

Training in spawn and mushroom production

S. No.	Place/Organization	Date	No. Persons
1.	Project Staff	2004-05	4
2.	Farmers sponsored by Urban Agriculture Department, Dire Dawa	Apr. – May, 2005	60
3.	Technical Staff, Agricultural Research Centre, Melkasa	AprMay, 2006	1
4.	Technical Staff, Agricultural Research Centre, Holetta	May- June, 2006	2
5.	Unemployed Youth from Addis Ababa	JunJuly., 2006	4
6.	Regional Agricultural Research Office, Barka Woreda, Addis Ababa	Sept., 2006	3
7.	Unemloyed Youth (Male and Female) sponsored by Urban Agriculture Department, Addis Ababa	Nov., 2006 and May, 2007	11
8.	Existing mushroom growers in Addis Ababa	Jan. and April, 2007	3
9.	Graduating students of HU	Jun., 2006 and Dec., 2007	15
10.	Staff and Framers sponsored by NGOs	2007-2008	42
	TOTAL		145

Training in composting, spawn and mushroom production

- 1. Training of technical and field staff in composting and spawn production and spawn run:
 - Two Tech. and two Field Asst. of HU and 3 from Melkassa and Holetta are fully trained in all the aspects of mushroom production technology.
- 2. Training of farmers, unemployed youth in mushroom production technology at Dire Dawa: A 3 weeks training program in mushroom production technology for 60 persons farmers, unemployed youth and women was organized in April-May, 05 in collaboration with Dire Dawa Dev. and Adm. Council. Participants were trained in mushroom growing systems; selection of growing containers; spawning and spawn-run; watering of beds; harvesting; care after picking and cooking-out of crop rooms.

Training Programs (contd.)

- Urban Agriculture Departments of Dire Dawa and Addis Ababa regularly sponsor unemployed youth and interested growers for mushroom training.
- Some of the existing mushroom growers in and around Addis Ababa were trained in mushroom cultivation, who are now regular customers of spawn produced at HU lab. Earlier they were importing spawn from Germany and Thailand.
- Practical exercises on mushroom production technology are included in UG course on 'Agricultural Microbiology'
- Passing out UG students (15) were trained and 2 of them have established their mushroom unit in Addis Ababa and supplying fresh mushrooms to supermarkets.

Training Programs (contd.)

- One post graduate student trained in the HU Mushroom laboratory has won a project from Min. of Ag. to establish a button mushroom production unit in Addis Ababa/Bahir Dar.
- The University is shortly going to sign a MOU with BioDiFood Association, an NGO based in Addis Ababa, for large scale production and training programs among farmers
- Specialized training in spawn production was given to the technical staff (5 nos.) of the Dire Dawa Ag. Office and an NGO at Sandafa. The participants were trained in methods of raising pure culture, preparation of master/stock culture, multiplication of spawn from stock culture and quality control measures to be followed for getting a good spawn.
- So far 145 persons have been trained. However, it is now a continuing activity of the Lab.

Popularization of Mushroom Technology

- Participation in Trade Fairs
- The Laboratory participated in International Trade Exhibition and Fair (AGRIfex Ethiopia 2005) organized by Ethiopian Chamber of Commerce in Addis Ababa from June 2-6, 2005.
- On behalf of HU, an exhibition stall was put up showing live samples of mushroom fruit bodies; posters and photographs of the indigenous edible mushroom species, and mushroom production process in the form of live samples and posters and extension bulletins.
- More than thousand people visited the stall and requested for training program in Addis Ababa.
- During last two years the laboratory has also participated in 4 trade fairs organized by Dire Dawa and Harar Development and Administrative Councils

Popularization of Mushroom Technology

- It is observed that currently there is enough demand of mushrooms in Addis Ababa, Nazerath, Bahir Dar and other tourist towns and locations.
- Due to unavailability of spawn and training in low cost production technology, the farmers, small and large scale entrepreneurs were unable to enter the business in an effective and regular basis.
- Haramaya University, therefore has taken the responsibility of producing good quality spawn for supply to growers and conduct training programs in Addis Ababa and other suitable locations for the benefit of interested growers.

Popularization of Mushroom Technology

Preparation of extension literature

- An extension bulletin on production technology of button and oyster mushrooms was prepared in English, Amharic and Ooromiya was prepared
- In order to disseminate the technology to larger farmer community, a video CD of the whole production process is being produced.
- The CDs will be used while conducting on farm demonstration trials on mushrooms at various locations in the country.
- The CDs would also be available on sale to interested mushroom growers.

Spawn Production

- The spawn production of *Pleurotus* sp. is now a regular activity of the project since last one years
- The HU administration has approved the sale of spawn at ETB 30 and mushroom at ETB 40 per kg.
- Large number of farmers, mostly in and around Addis Ababa are regular customers
- So far more than 250 kg. of spawn has been sold to mushroom growers in last 6 months
- The farmers have reported a greater success of fruiting body formation from spawn grown in maize compared to that produced on sorghum or wheat.
- Therefore demand of spawn on maize is increasing

Post Graduate Research on Mushrooms

Following MSC. Research projects on mushroom production technology were successfully completed and defended in last two years:

- Effect of organic substrates and their pasteurization methods on yield and quality of Oyster mushroom (*Pleurotus sajor-caju*) (The substrates were Wheat straw, shredded maize stalk, tef straw, chat leaves, saw dust and bean straw and pasteurization methods were Cold water, hot water and formalin treatment)
- Effect of spawn substrates and their inoculation rates on yield and quality of oyster mushroom (*Pleurotus florida*)
 (Grain substrates were wheat, maize, sorghum and teff at inoculation rates of 1, 2, and 3 %)

Future strategies for mushrooms

Cropping systems

- Marginal growers cropping system: It can be a seasonal activity for resource poor farmers using their own farm waste to generate additional income. The mushroom houses could be made up of bamboo, straw, tarpaulin and plastered by cattle dung plus soil and husk and have racks of any suitable wooden sticks like sorghum stems. The produce could be sold in nearby local market or collected by peasant associations and sold to city-markets.
- 2. Small growers cropping systems: A large number of growers having access to unused space in their buildings or having unused old buildings can convert them to mushroom crop rooms. Depending on resources, construct one or more insulated crop rooms of approx.15x10x10 m size and take 1-2 or more crops in a year based on prevailing temperature at a location. Wooden trays, plastic bags or shelves could be used for growing mushrooms by such farmers.
- Big growers cropping systems: These farmers would be growing mushrooms on commercial basis for local and export market. The growing units would be equipped with spawn laboratory, pasteurization chambers, cropping rooms with controlled conditions and inbuilt forced air system, canning unit etc.

Other Future Strategies

- The location of marginal/small farms should be in the periphery of big townships and cities so that the farmers could market their fresh produce easily. The government, particularly the departments of urban agriculture, should give incentives to farmers, unemployed youth and economically backward families for setting up mushroom production units in villages close to such locations.
- The regional and central government departments should also come forward to establish large mushroom production units with high production capacity (1-4 tons/day) in technical collaboration with research institutes or universities engaged in mushroom research. These large units should be export oriented and should be able to grow mushrooms throughout the year under controlled conditions. These units can also purchase the produce of marginal/small farms located around them.
- There is also the possibility of exploring the international collaboration on mushroom research, which will help the spread of existing knowledge and technologies.

Other Future Strategies

- In Ethiopia a small number of mushroom farms are operating as independent units using own substrate, inoculating, growing, producing and marketing the crop. Even importing the spawn either from European or South-East Asian countries. None is producing their own spawn.
- Therefore, lack of locally available quality spawn is one of the biggest bottle-neck in the popularization of this technology among the farmers of the country. The first priority should be to develop facilities for production of quality spawn in sufficient amount by organizations at region/district level.

Other Future Strategies

- Relatively less educated and resource poor farmers, who are in large number, could be encouraged and trained to produce the crop while other aspects of production such as spawn multiplication and supply may be dealt with small number of research institutes, large mushroom growing units/companies or trained operators.
- Marketing of the produce may be a problem for some growers, particularly the small scale farmers. More and more farmers will be encouraged to go into mushroom business if they are relieved from the task of marketing. Therefore, it is suggested that marketing of mushrooms should be taken up by a central organization at the region/district/ peasant association level. A proper marketing strategy is, therefore, of paramount importance to avoid initial years of competition from other vegetables, coupled with conservative eating habits of people.