CONTRACT REPORT

THE USE OF COMPOSTED WOODCHIP AS A SUBSTRATE FOR GROWING VARIETIES OF EDIBLE MUSHROOMS

UNDERTAKEN FOR

Organic Centre Wales
University of Wales, Aberystwyth
under the
Farming Connect Scheme

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Period of Investigation:	May , 2004 – May, 2005
Date of issue of report:	January 2006

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AUTHENTICATION

I declare that this work was done under my supervision according to the procedures described herein and that this report represents a true and accurate record of the results obtained.
.... David Frost , Study Director
Date

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Introduction

The world market for edible mycorrhizal mushrooms has experienced considerable growth over the past two decades with annual production estimated to exceed 14 billion USD. The market continues to grow due to interest in the nutritional and health benefits of edible mushrooms. The size of the market for speciality mushrooms has also increased and the overall market trend has been towards fresh rather than canned product. Mushrooms are extensively used throughout the catering industry, with high demand for fresh supplies in tourist areas especially during peak holiday periods.

The demand for organic mushrooms is also growing, but increased production may be limited because of the standards that require all straw and manure used in substrates to come from an organic source. Because of the shortage of organic straw, and its high price, organic growers need to examine alternative substrates.

Despite the size of the market for mushrooms, according to the International Society for Mushroom Science, research has been limited and their potential use in waste management has yet to be fully explored. There are however a number of different cultural methods in commercial use, with a wide variety of species produced on different substrates. The choice of species depends on what growth media are available and on market considerations. Alternatives to the common mushroom, *Agaricus bisporous,* which is usually grown on composted cereal straw and animal manure, include Oyster mushroom, *Pleurotus spp*, which is produced on various growth media, and Shiitake *Lentinus edodes,* which is grown on logs, sawdust or woodchip.

Following on from the WDA funded project, *Woodchips as an alternative bedding material for livestock systems and the potential to produce an added value product from the composting of the resultant manure*, the aim of this project was to evaluate woodchip compost as a substrate to produce edible mushrooms.

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0 Literature review

0.1 Optimal growing conditions for exotic mushrooms.

The paragraphs below elaborate on the optimal growing conditions for a number of exotic mushrooms.

0.1.1 Optimal growing condition for Portobello (brown variety of Agricus bisporus)

Agricus blazei cultivation

According to Chen (2003) much of the cultivation techniques for Agricus Blazei has been adapted from growing A. bisporus with the major differences:

- higher temperature needed
- light needed for fruiting

There are a number of recommended compost formulations. Chen (2003), Huang (1997) and Oei (1996) mention a few. Compost is commonly fermented outdoors (during which the temperature should not exceed 60C). The compost should be moved indoors when the temperature drops to 50'C. The fermented compost is generally pasteurized at this phase. Pasteurization should occur at a temperature of 55~60'C for 2 days. Closing the doors and windows and steaming the room can be beneficial.

When the temperature drops to 25~35'C the substrate can be inoculated. This is done by applying spawn, divided into small quantities (egg-size), to the mushroom bed, 20 cm apart at a depth of approximately 10 cm.

When mycelium has grown throughout the substrate in the beds, trays or synthetic bags soil casing, adjusted to pH 7.0, can be applied (This is approximately 20 days after inoculation). Table X gives an overview of the optimal growth parameters in the different phases as recommended by a number of authors.

Table 1. management of growth parameters

	Inoculation	Primordia initiation (40 days after spawning)	Fruiting body development [1]	Reference
Temperature	23-27 C 10-37C optimum range	22-25C (20-28C) Optimum range	20-33C	Huang, 1997 natural day/night variation
Humidity		Plastic cover 2 days 90-95% R.H. under		
Temperature	22-26C		22-25C	Iwade & Mizuno, 1997
Humidity	60-75% R.H.		70-85% R.H.	
Soil casing	60-75% moisture			

[1] (approx. 10 days); production continues for 3-4 months with 4-5 flushes

Temperature	18-30 C		23-28C (day-night)	Okubo et al. 1991 better quality
Temperature	21-27 C (70-80F)	21-24C (70-75F)	24-27 C (75-80F)	Stamets, 2000
Humidity	90-100% R.H.	80-90% R.H.	75-85% R.H.	
Light	N/A	Minimal 100-200 F.C.	Minimal 100-200 F.C. (foot candle)	
CO_2	>5,000 ppm	400-800 ppm 5-7 air exchange per hr.	<2,000 ppm 5-7 air exchange per hr.	

Source: Chen, 2003

0.1.2 Optimal growing condition for oyster mushroom (Pleurotus ostreatus)

There are a number of methods to produce oyster mushrooms. Below, two methods are outlined. One for production in bags and one for production on shelves.

Production in bags

The substrate used for cultivation of oyster mushrooms commonly includes sawdust and a carbohydrate source. This substrate is placed in a polypropylene bag. It should be sterilized, cooled and inoculated. The inoculated substrate should be placed in a dark location.After the mycelium has grown throughout the substrate, openings are cut through the bag where fruiting bodies will develop. Optimal temperature for fruiting is 15-20 C. (source: cultivation of edible mushrooms http://www.botany.hawaii.edu/facultry/wong/BOT135/Lect18.htm.

Production on shelves

The method described below is an elaborate process in which the fermentation occurs in three steps (pre-fermentation, pasteurization and post-fermentation). Kyung Wha Choi (2004) mentions that shelf cultivation requires an pre-fermentation of the substrate material outdoor for 2-3 days, an pasteurization at 60-65C for 8-10 hours (at least 6 hours) and an post-fermentation at 45-55C for 3-4 days. However, not all oyster mushroom producers use this elaborate method for fermentation.

After fermentation, the substrate can be mixed with spawn (inoculum) indoors. Then the substrate is placed onto shelves. The moisture content should be round 70%. When the temperature has dropped to 20-25C, about 60-70% of the spawn can be inoculated and mixed with the substrate on the shelves. Substrate can be spread on the shelves and then the remaining 30-4-% of the spawn can be applied onto the substrate surface (Some authors mention mixing of substrate and spawn with machines before application). The substrate then should be mulched with a plastic sheet (or other casing layer) with very small holes for ventilation. Mushroom mycelia will develop for 17-23 days while covered with a plastic sheet. Temperature should be maintained at 20-22C during this first stage and then gradually be increased up to 25C.

To start the fruiting process, light has to be added, a cold shock has to be performed, and a high relatively humidity should be maintained. The light level should be between 80-120 lux 3 to 4 days before plastic is removed and the temperature should be lowered to 15-18 C. This optimal temperature may range between 10 and 24 C depending on species and strains. The Relative Humidity should be kept between 85 and 95% (Kyung Wha Choi, 2004).

0.1.3 Optimal growing condition for parasol mushrooms (Lepiota procera)

No literature available

0.1.4 Optimal growing condition for King Stropharia (Stropharia rugossa annulata)

King Stopharia is best grown on straw, compost, sawdust, mulch or woodchip. A 83% saturated substrate is optimal and the optimal daily temperature is 7-13 C. Contact with soil is said to be beneficial (either from underneath or on top of the growing medium and soil rich in compost will also give good yields). King Stropharia can be poduced by applying spawn to prepared plots outdoors but can also be cultivated in bags. The fruiting generally starts 12-20 weeks after preparation. Once the King Stropharia is established, small sections of colonised mulch can dug out and transplanted to new plots. http://www.14u.co.nz/st.htm.

0.2 Market potential of exotic mushrooms

A small market survey was undertaken as part of this report (in Wales). As part of the survey a number of vegetable box schemes and retailers were contacted and asked a number of questions relating to supply and demand of organic (exotic) mushrooms. The survey showed that retailers tend to source from wholesalers. The table below shows the range of prices paid to the producers.

Table 2. market prices for organic mushrooms

Mushroom species	
Oyster	£5.4- £12.0
Portobello	£ 3.61
Chestnut	£2.80- £3.56
Shitake	£10- £12
Flat	£4.39
White	£2.40- £3.56

Source: market telephone survey

The small market survey suggests that there is a market for local/ welsh organic mushrooms. Interviewees showed interest to source locally. However, cost of production should be calculated to show whether the costs are reasonable compared with the marketprice received.

1 Outline ADAS Pwllpeiran Mushroom Trial 2004

1.1 Objectives

To evaluate resulting compost from beech and spruce woodchip used as bedding for either cattle or sheep as a medium in the cultivation of culinary/medical mushrooms under two growing systems.

1. Intensive indoor system under environmentally controlled conditions.
2. Extensive outdoor system.

1.2. Materials

1.2.1 Test Composts

During the winter of 2003 silage fed cattle and sheep were housed on beech or spruce woodchip for an eight-week period. The four resulting beddings were stored in individual heaps indoors to compost for 6 months during early 2004. At the end of the composting process, temperature variations within each compost were minimal (Fig 1) and almost 100% pathogen kill had been achieved.

Figure 1: Analysis of Woodchip Compost

	Sheep hardwood	**Sheep softwood**	**Cattle hardwood**	**Cattle softwood**	**[2]Alternative compost[3]**
PH	8.8	8.5	7.4	8	8.1
Conductivity (∝S@20°C)	380	275	92	57	Not measured every time; in 2004: 11.5 ms/cm
Density (g/l)	480	501	456	442	Phase 1: 450 Phase 3: 350
Phosphorus (mg/l)	132	128	23	15	Not measured
Potassium (mg/l)	655	478	115	78	Not measured
Magnesium (mg/l)	10	12	2	1	?
Mineral Nitrogen (mg/l)	0	0	0	0	
Nitrate as N (mg/l)	< 6	< 6	< 6	< 6	0
Ammonia as N (mg/l)	< 1	< 1	< 1	< 1	Phase 1: 0.65% in dry matter (=2.9 g/l)
Calcium (mg/l)	22	22	6	5	?
Sodium (mg/l	88	55	34	16	?

[2] These numbers are retrived from an detailed analysis of the compost commonly used by mushroom producers in the Netherlands. The compost is made of (horse manure (o.b.v. straw), chicken manure (liquid or o.b.v. sawdust), straw (wheat), ghypsum (free of heavy metals)
Humidity percentages the compost: (indicative) are the following Phase 1: 75 % (fresh compost) Phase 2: 69 % (inoculated compost) Phase 3: 65 % (mycellium grown through compost)

[3] The substrate used for growing exotic mushrooms is based on woodchip; there are no analyses of this substrate available www.cnc.nl

Chloride (mg/l)	193	41	47	20	?
Sulphur (mg/l)	15.30	9.18	4.21	3.7	?
Boron (mg/l)	0.31	0.14	0.24	0.15	?
Copper (mg/l)	< 0.15	< 0.15	< 0.15	< 0.15	Phase 3: 15mgCu/kg in dry matter = 5.3 mg/l
Manganese (mg/l)	0.3	0.2	< 0.1	< 0.1	?
Zinc (mg/l///0	0.24	0.19	0.12	0.15	?
Iron (mg/l)	1.20	1.26	0.66	0.50	?

Source: pers. Comm. With W. Arts of CNC

1.2.2 Mushroom species

Four mushroom species were identified to be tested: -

a) Oyster (Pleurotus ostreatus)
b) Parasol (Lepiota procera)
c) King Stropharia (Stropharia rugossa annulata)
d) Agricus Blazei

The species identified were documented as growing on wood based substrates and would have a marketable value. The first two are well recognised as culinary mushrooms, King Stropharia to a lesser extent. Agricus Blazei, although not a culinary mushroom, is widely used in the increasing homeopathy market as an extract.

1.3. Methods

1.3.1 Inoculum production

1. Auto-claveable plastic bags containing organic wheat grain were sterilised at 135^{o}C (15psi) for 1 hour and left to cool under clean-room conditions for 30 minutes.
2. Segments of colonised agar of each culture were cut with a sterile scalpel within a laminar flow cupboard and placed into a labelled bag of grain.
3. The plastic bags were heat sealed and stored at 21^{o}C under natural lighting conditions until covered with mycelium.

1.3.2 Outdoor beds

During early March 32 plots arranged in a single randomised block were dug, each measuring 1 metre square and to a depth of 0.5 metre design (Appendix I). Each hole was separated by 0.5 metres guard area. Each hole was filled with around 60cm of compost and allowed to settle and weather.

% Moisture content at initial fill		
Cattle Hardwood		67.2
Cattle Softwood	73.8	
Sheep Hardwood	72.4	
Sheep Softwood	69.5	

Plots were inoculated with King Stropharia, Parasol and Agricus Blazei during mid July. Due to problems with the initial strains of Oyster cultures, plots were not inoculated until October. Each plot was inoculated by digging a 15cm depression in the surface of the compost from each corner to the other to form a X. One and half bags of grain were evenly spread along the X and covered with compost removed to form the depression. After inoculation half the plots were covered with mypex (Appendix I) and removed at the end of October.

1.3.3 Indoor Trays

Plastic trays measuring 60cm (L) x 40cm (W) x 15cm (D) were selected due to their similarity of equipment used in the button mushroom industry. During mid August trays were prepared for indoor fruiting by placing a thin layer of unsterilised substrate to the bottom of the tray (2.5cm) and then adding a layer of grain spawn, thinly distributed across the bed of chip. This layering process was repeated three times in each tray and topped with a thicker layer of chip - to 6cm. Trays were placed into a growing room at fluctuating temperatures between 12-20C and humidity set to fluctuate between 75-95%.

1.4. Assessments

1.4.1 Outdoor beds

1. Temperatures were recorded hourly at a depth of 25cm and the moisture content were assessed fortnightly in two additional holes dug alongside the inoculated plots.
2. Weekly visual monitoring of surface mycelium and fruiting bodies.

1.4.2 Indoor trays

3. Weekly visual monitoring of mycelial growth and fruiting bodies by Hummungus fungus staff.
4. Regular visual monitoring of surface mycelium and fruiting bodies by ADAS staff.

1.5. Results

1.5.1 Outdoor Beds

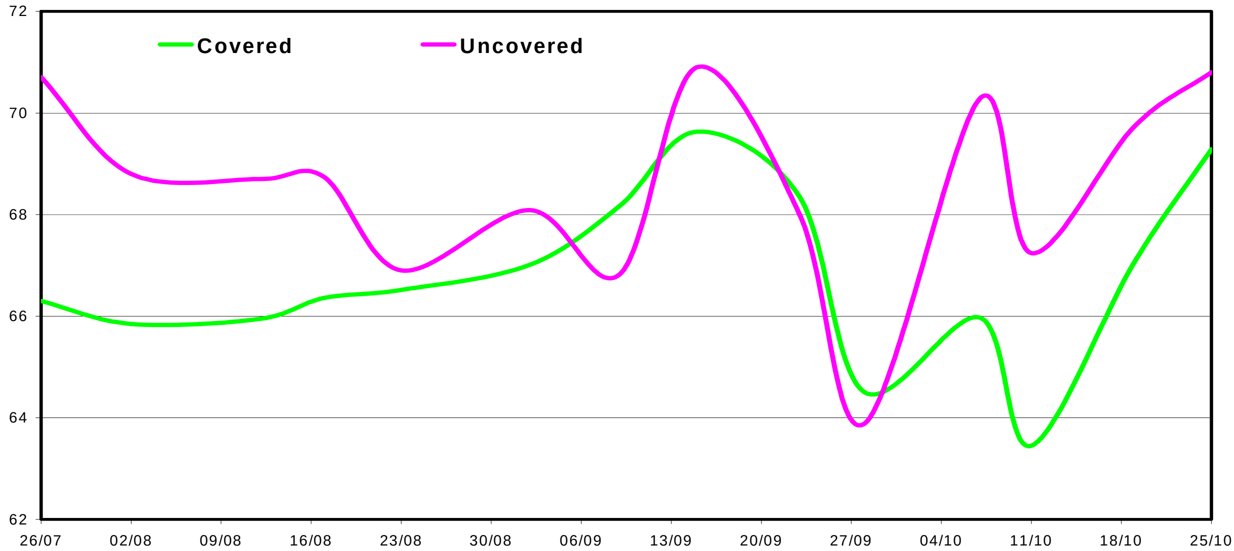
1. Although mycelium seen on the surface of King stropharia, Parasol and Agaricus blazei. No mushrooms grew
2. Oyster mushrooms fruited on cattle softwood, sheep hardwood and softwood, but not on cattle hardwood.
3. Yields of Oyster mushrooms were greater in plots covered with Mypex.
4. Fruiting occurred on the dates shown below:

Compost Type	**Covered**	**Uncovered**
Cattle softwood	Early Feb Early April Early May	Early Feb Early May
Sheep hardwood	Mid Feb Early April Mid April	Mid March
Sheep softwood	Mid Feb Mid April	Mid Feb Mid April

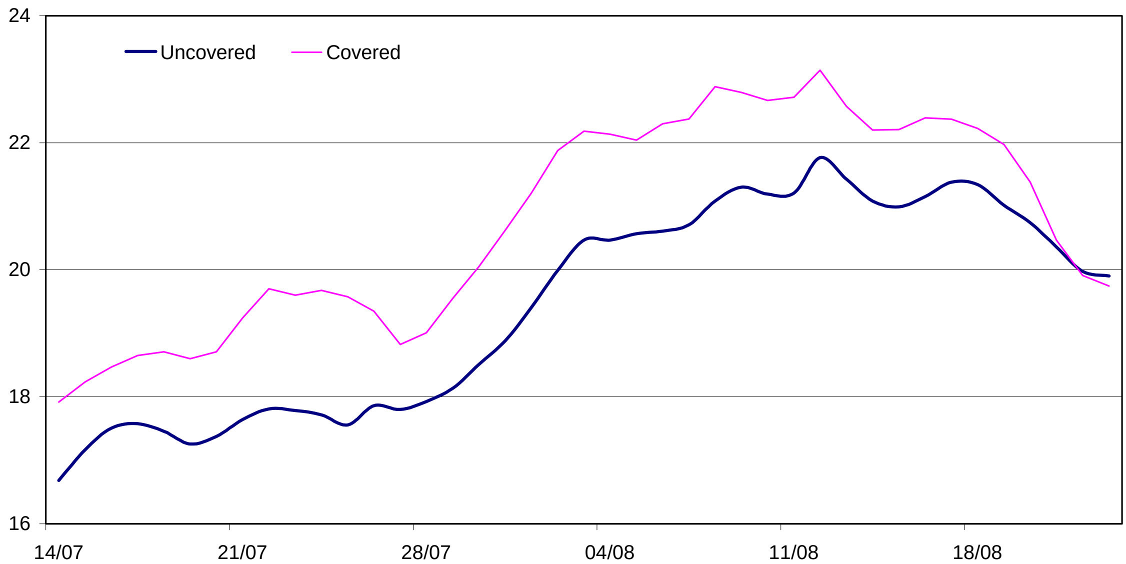
1.5.1.2 Total yield of mushroom (grams)

Compost Type	**Mushroom Species**	**Covered**	**Uncovered**
Cattle hardwood	Oyster	0.0	0.0
Cattle softwood	Oyster	5080.9	4664.2
Sheep hardwood	Oyster	7592.4	1393.4
Sheep softwood	Oyster	3516.2	2174.8
Cattle hardwood	Parasol	0.0	0.0
Cattle softwood	Parasol	0.0	0.0
Sheep hardwood	Parasol	0.0	0.0
Sheep softwood	Parasol	0.0	0.0
Cattle hardwood	King Stropharia	0.0	0.0
Cattle softwood	King Stropharia	0.0	0.0
Sheep hardwood	King Stropharia	0.0	0.0
Sheep softwood	King Stropharia	0.0	0.0
Cattle hardwood	Agricus Blazei	0.0	0.0
Cattle softwood	Agricus Blazei	0.0	0.0
Sheep hardwood	Agricus Blazei	0.0	0.0
Sheep softwood	Agricus Blazei	0.0	0.0

% Moisture of Compost at 15cm



25cm compost temperatures



1.5.2 Indoor trays

Regular assessments showed that mycelial growth began to develop during week 2 to 3 and slowly increased through the substrates until week 9. During week 9-10 the mycelium went from a bright white colour to a creamy white/yellow. Assessments carried out during week 11 showed a decline in the amount of mycelial growth, further assessments during week 14 again showed a further decline in mycelial growth and the first trial was ended.
Each substrate was assessed for a second time during the end of December; a hand-watering strategy was developed and introduced to prevent the substrate on the top layer from drying out so rapidly. Mycelial growth again began to develop during week 2/3 and increased through the substrates. Oyster mushrooms fruited on cattle soft and hardwood as well as sheep softwood during week 7. Mycelial growth increased on all the other substrates up until week 11 when mycelial growth again began to decline. No other fruiting had occurred by week 15 at which time mycelial growth had declined dramatically and the second trial was ended.

Fig.1 Sheep manure heaps temperature /c halfway down heap 0.9m depth

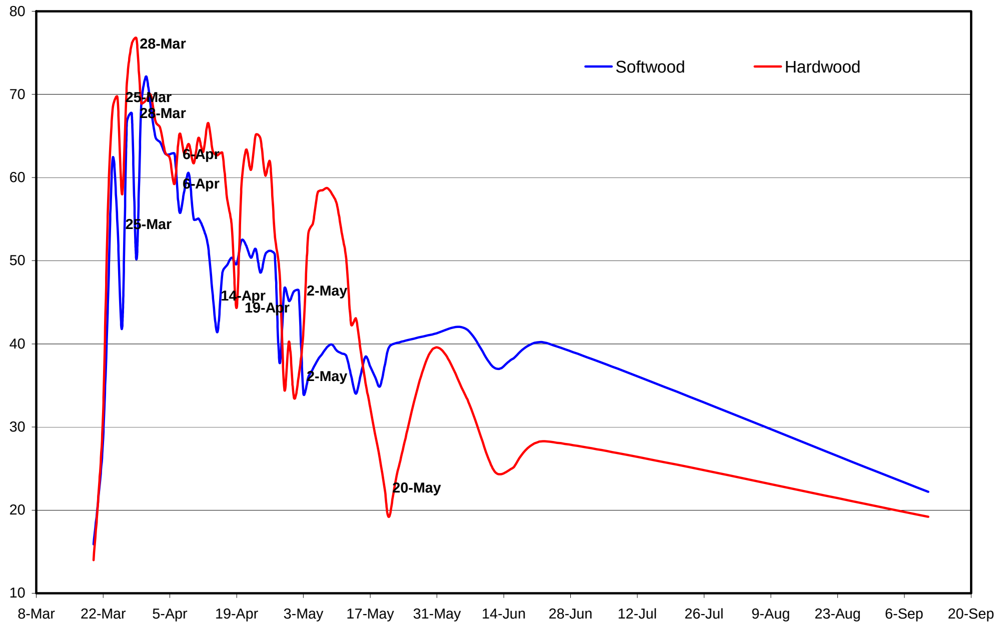
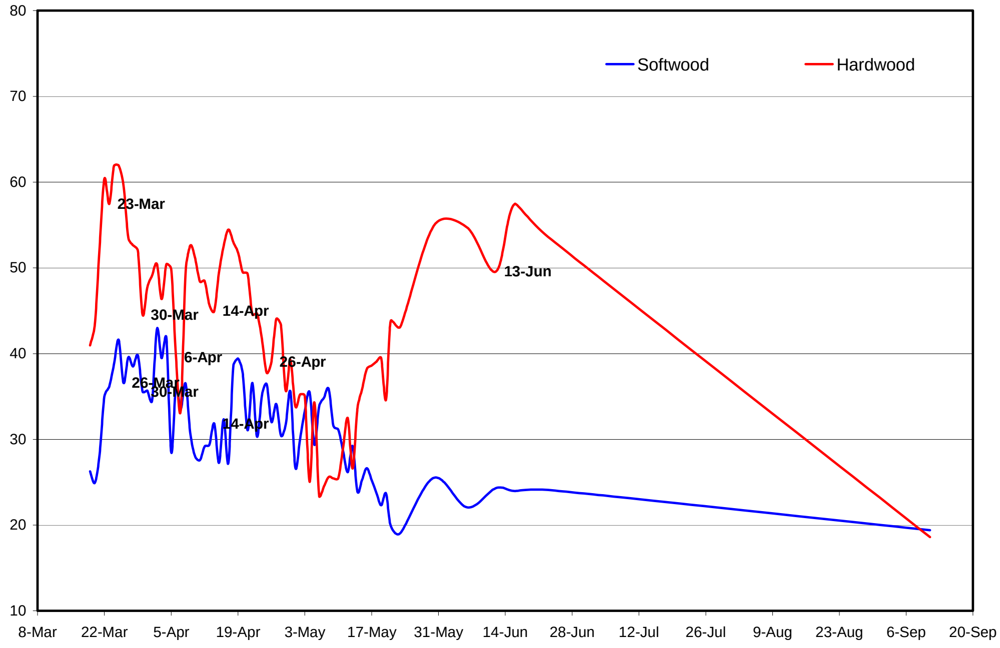


Fig.2 Cattle manure heaps temperature /c halfway down heap 0.9m depth



1.6. Conclusions and recommendations

Both indoor and outdoor systems resulted in mycelium growth, but fruiting bodies were only produced by Oyster mushrooms. Woodchip compost therefore appears to have potential as a growing medium but more development is necessary to make this a commercial proposition.

The specific issues arising from the trials that need further investigation are:

Composition of substrate – the trials demonstrated that mycelial growth occurs on composted woodchip, but the failure to produce fruiting bodies indicated that more investigations are needed to establish the necessary substrates for satisfactory mushroom production. Alternative substrates also need investigation.

Outdoor Beds - In the outdoor trial weather conditions – particularly high rainfall could be responsible for the lack of fruiting. Raised beds rather than filled pit beds could be a preferable option in high rainfall situations.

Indoor trays - casing layer. The decline of mycelial growth (apart from Oyster) before fruiting could indicate the need to case the indoor woodchip beds, for example with peat, as in traditional mushroom production on straw-based substrates.

Mushroom varieties –there are further varieties of 'exotic' mushrooms, many naturally occurring in Wales & the UK that could also be trialed for commercial potential using woodchip and other alternative to straw substrates.

1.6.1 Recommendations

- Continued evaluation of indoor controlled systems versus outdoor growing systems
- Trials with raised beds in outdoor mushroom production
- Trials using drilled logs in indoor and outdoor production
- Trials of alternative substrates – e.g. miscanthus, bark peelings
- Trials of alternative mushroom species

1.7 References

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http://www.botany.hawaii.edu/facultry/wong/BOT135/Lect18.htm.

APPENDIX I: Plot Layout

Sheep Hardwood	Oyster	Uncovered
Sheep Hardwood	Oyster	Covered
Sheep Softwood	Oyster	Uncovered
Sheep Softwood	Oyster	Covered
Cattle Softwood	Parasol	Uncovered
Cattle Softwood	Parasol	Covered
Sheep Softwood	Parasol	Uncovered
Sheep Softwood	Parasol	Covered
Sheep Hardwood	Agaricus blazei	Covered
Sheep Hardwood	Agaricus blazei	Uncovered
Sheep Softwood	King stropharia	Covered
Sheep Softwood	King stropharia	Uncovered
Cattle Softwood	King stropharia	Uncovered
Cattle Softwood	King stropharia	Covered
Cattle Softwood	Agaricus blazei	Uncovered
Cattle Softwood	Agaricus blazei	Covered
Cattle Hardwood	Parasol	Uncovered
Cattle Hardwood	Parasol	Covered
Cattle Hardwood	Agaricus blazei	Covered
Cattle Hardwood	Agaricus blazei	Uncovered
Cattle Softwood	Oyster	Covered
Cattle Softwood	Oyster	Uncovered
Sheep Hardwood	King stropharia	Covered
Sheep Hardwood	King stropharia	Uncovered
Sheep Softwood	Agaricus blazei	Uncovered
Sheep Softwood	Agaricus blazei	Covered
Sheep Hardwood	Parasol	Uncovered
Sheep Hardwood	Parasol	Covered
Cattle Hardwood	Oyster	Uncovered
Cattle Hardwood	Oyster	Covered
Cattle Hardwood	King stropharia	Covered
Cattle Hardwood	King stropharia	Uncovered
Cattle Softwood	**Guard plot**	Covered
Cattle Softwood	**Guard plot**	Uncovered

APPENDIX II

Mycelium Assessment 11 November 2004

Mushroom type	Compost type	Covered/ Uncovered	Mycelium on surface
Agaricus blazei	Cattle hardwood	Covered	Yes
		Uncovered	Yes
	Cattle softwood	Covered	Yes
		Uncovered	No
	Sheep hardwood	Covered	No
		Uncovered	No
	Sheep softwood	Covered	No
		Uncovered	No
King stropharia	Cattle hardwood	Covered	Yes
		Uncovered	No
	Cattle softwood	Covered	No
		Uncovered	Yes
	Sheep hardwood	Covered	No
		Uncovered	No
	Sheep softwood	Covered	No
		Uncovered	No
Oyster	Cattle hardwood	Covered	No
		Uncovered	No
	Cattle softwood	Covered	No
		Uncovered	No
	Sheep hardwood	Covered	No
		Uncovered	No
	Sheep softwood	Covered	No
		Uncovered	No
Parasol	Cattle hardwood	Covered	No
		Uncovered	Yes
	Cattle softwood	Covered	No
		Uncovered	No

	Sheep hardwood	Covered	Yes
		Uncovered	No
	Sheep softwood	Covered	No
		Uncovered	No

APPENDIX II

Mycelium Assessment 3 February 2005

Mushroom type	Compost type	Covered/ Uncovered	Mycelium on surface
Agaricus blazei	Cattle hardwood	Covered	No
		Uncovered	No
	Cattle softwood	Covered	No
		Uncovered	No
	Sheep hardwood	Covered	No
		Uncovered	No
	Sheep softwood	Covered	No
		Uncovered	No
King stropharia	Cattle hardwood	Covered	Yes
		Uncovered	No
	Cattle softwood	Covered	No
		Uncovered	Yes
	Sheep hardwood	Covered	No
		Uncovered	Yes
	Sheep softwood	Covered	No
		Uncovered	No
Oyster	Cattle hardwood	Covered	No
		Uncovered	No
	Cattle softwood	Covered	No
		Uncovered	No
	Sheep hardwood	Covered	No
		Uncovered	No
	Sheep softwood	Covered	No
		Uncovered	No
Parasol	Cattle hardwood	Covered	Yes
		Uncovered	Yes
	Cattle softwood	Covered	No
		Uncovered	No
	Sheep	Covered	No

hardwood		
	Uncovered	No
Sheep	Covered	No
softwood		
	Uncovered	No

APPENDIX III:

12 Mushroom yields (grams)

Compost type	Data	Mushroom type	Covered	Uncovered
Cattle hardwood	3/2	Oyster	0	0
	7/2	Oyster	0	0
	8/2	Oyster	0	0
	10/2	Oyster	0	0
	21/3	Oyster	0	0
	3/4	Oyster	0	0
	15/4	Oyster	0	0
	3/5	Oyster	0	0
Cattle softwood	3/2	Oyster	1549	3477
	7/2	Oyster	0	0
	8/2	Oyster	0	0
	10/2	Oyster	0	0
	21/3	Oyster	0	0
	3/4	Oyster	1233	0
	15/4	Oyster	0	0
	3/5	Oyster	2299	1187
Sheep hardwood	3/2	Oyster	0	0
	7/2	Oyster	0	0
	8/2	Oyster	0	0
	10/2	Oyster	2736	0
	21/3	Oyster	0	1393
	3/4	Oyster	4556	0
	15/4	Oyster	301	0
	3/5	Oyster	0	0
Sheep softwood	3/2	Oyster	114	209
	7/2	Oyster	1076	622
	8/2	Oyster	253	108
	10/2	Oyster	0	0
	21/3	Oyster	0	0
	3/4	Oyster	0	0
	15/4	Oyster	2074	1236
	3/5	Oyster	0	0

APPENDIX IIII: Mean Soil temperatures Pwlllpeiran

Mean monthly temperature and rainfall at pwllpeiran may 2004-may2005

Month	Temp Mean Max	Temp Mean Min	Rainfall total (mm)	Soil Temp Mean 10cm	Soil Temp Mean 20cm	Soil Temp Mean 50cm
May 04	14.37	5.77	97.3	11.71	11.69	11.22
June 04	16.85	9.39	98.4	14.26	14.33	13.89
July 04	16.52	10.02	142.6	14.75	14.76	14.58
August 04	18.91	12.03	179.3	15.74	16.01	16.08
Sept 04	16.15	9.84	283.5	12.99	13.76	13.16
Oct 04	11.96	6.36	333.3	9.35	10.04	11.81
Nov 04	9.39	4.85	154.5	7.66	8.24	9.78
Dec 04	7.87	2.74	209.0	5.46	6.03	7.64
Jan 05	7.79	2.80	170.5	5.11	5.58	6.99
Feb 05	5.89	.782	178.0	3.96	4.58	6.17
Mar 05	9.3	2.8	87.8	5.4	5.6	6.3
April 05	11.2	3.9	129.3	7.2	8.2	8.7
May 05	13.9	5.5	71.8	11.1	12.1	11.3