

Project title: Understanding Mushroom Nutrition:
Project aimed at improving yield,
substrate efficiency and utilisation and
flavour

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[The results and conclusions in this report are based on an investigation conducted over a one-year period. The conditions under which the experiments were carried out and the results have been reported in detail and with accuracy. However, because of the biological nature of the work it must be borne in mind that different circumstances and conditions could produce different results. Therefore, care must be taken with interpretation of the results, especially if they are used as the basis for commercial product recommendations.]

AUTHENTICATION

We declare that this work was done under our supervision according to the procedures described herein and that the report represents a true and accurate record of the results obtained.

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9 July 2014

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9 July 2014

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GROWER SUMMARY

Headlines

- Addition of protein-based supplements (ProMycel Gold, Champfood E, MCSubstradd and Natural Gold (a lipid-protein blend) to phase 3 composts increased mushroom yields significantly for strain A15.
- Some of the protein-based supplements increased mushroom cap density but none of the supplements affected the dry matter content or the colour of the mushrooms.

Background

The nutrition from compost that is available for mushroom growth is a key factor for the success of the mushroom industry. At the moment we do not have precise knowledge of what nutrition is optimal for the mushroom growth, yield and quality, whether and how nutritional supplements perform on phase 3 compost, and a relevant and precise test for mushroom compost quality. This project aims to evaluate the effects of four different types of compost supplement applied to two different phase 3 composts on crop yield, flushing pattern and quality. This project will also develop our understanding on mushroom nutrition using the mushroom to report on its own biology in response to different supplements by employing microarray technology. This may provide knowledge of how supplements stimulate higher yields and how further improvements can be developed. The four supplement types under evaluation are available as commercial products:

- Protein-based (largely from soy) – three products tested (ProMycel Gold, Champfood E, MCSubstradd)
- A blend of lipid and protein-based product (Natural Gold)
- A carboxylic acid-based product (MycroNutrient)
- Mineral micronutrients, a mix of calcium, magnesium, sulphur, boron, copper, iron, manganese, molybdenum and zinc salts (Micromax)

This report is of the first year of the project and provides the results of a proving trial of nutrient supplement use with a brown strain of mushrooms and a full trial of the effects of supplementation on the white strain A15. A further full trial is taking place in the second year of the project. Therefore this report provides interim results and conclusions.

Summary

Supplements for phase 3 composts are used extensively in the UK and European mushroom industry; however we are not aware of a detailed scientific study to compare supplements and to examine whether and how supplements on phase 3 compost are effective in improving mushroom yield and quality. In this two year study we are examining the effects of four different supplement types (protein-based, lipid-protein blend, carboxylic acids and mineral nutrients) on compost temperatures, mushroom yield and quality. During the first year of the project, two crop experiments were carried out: a proving trial was carried out on phase 3 compost spawned with a brown strain, and a major crop experiment during which we measured effects of supplements on two phase 3 compost types (based on straw and chicken manure or based on straw, horse bedding/manure and chicken manure) on A15 mushroom yields and quality.

Initial conclusions are based only on the first year's work and so care must be taken to avoid over-interpretation. From the data so far we can conclude:

- Significant yield improvements were recorded when protein-based supplements (ProMycel Gold, Champfood E, MCSustradd) or a lipid-protein-blend (Natural Gold) were added to phase 3 composts. The highest yield for the straw-based compost was with supplementation with Natural Gold and for the horse manure-based compost the highest yield was with Champfood E supplementation. Micromax and MycroNutrient supplements had no effect on mushroom yields for both compost types. However it should be noted that MycroNutrient is marketed as a casing supplement and this use was not tested in this project
- Averaged over two flushes, the protein-based supplements, ProMycel Gold, and Champfood E, increased the density of the mushroom caps on both types of compost. Mushrooms grown with MCSustradd were not tested for density in both flushes, and the effect in the first flush was not significant. Natural Gold increased mushroom density on horse manure-based compost but not on straw-based compost. The ability to increase the density of mushrooms by application of supplements is an important result as density correlates with mushroom cap texture (an important quality attribute) and increased density should lead to improved picking rates (higher density means more weight picked per mushroom or per hour)

- For the horse manure-based compost, the improvement in yield by protein-based supplements was largely in the first flush, not at all in the second flush and again in the third flush. For the straw-based compost, the improvement in yield by protein-based supplements was observed in the first and second flushes but not the third flush
- Supplementation did not change the dry matter content of the mushrooms (dry weight/fresh weight ratios). However, the horse manure-based compost produced mushrooms with higher dry matter than the straw-based compost. Further experimentation will determine whether this is a general trend or may represent batch-to-batch variability
- There were no major effects of supplementation on mushroom colour
- Some differences in yield and mushroom quality were observed between the two compost types
- Only one of the supplements used, the protein-based MCSustradd, resulted in a significant yield increase in the brown strain Heirloom

Supplementation of phase 3 composts by protein-based supplements increases mushroom yield significantly. This work has also shown that the degree of yield improvement depends in part on the characteristics of the compost. Growers are advised to note and relate yield to the brand of supplement used. When we make our final report for this project with data from trial 3, we will be in a better position of surety to make clear recommendations.

Brown strains are sometimes grown in unsupplemented compost. The initial results from this work indicate that browns can also benefit from protein supplementation.

Financial Benefits

Clearly there are financial benefits for increased yield of mushrooms by the supplementation of phase 3 compost with protein-based products. These are of course offset by the cost of supplement. Supplement prices can vary as soy is a world-traded commodity.

In addition, the increase in mushroom cap density by protein-based supplements has positive financial benefits as cap density determines cap texture (an important quality attribute) and a higher cap density leads to the potential for improved picking rates (higher density means more weight picked per mushroom or per hour).

Action Points

This report covers the initial year of this project; as such, it is too early to recommend any action points for growers.

SCIENCE SECTION

Introduction

Understanding mushroom nutrition is one of the key cornerstones of the mushroom industry as it determines yield, flushing pattern and quality. Knowledge on mushroom nutrition is limited and the mechanisms of action of supplements (how they work) are poorly understood. In addition the measurement of compost quality (ash, dry matter, pH, nitrogen, etc) is perhaps arbitrary, and could be described as a list of what *can* be measured. This project aims to provide practical and strategic knowledge on the effects of nutrient supplements and the nutritional requirements of the mushroom.

The mushroom, *Agaricus bisporus*, is described as a secondary degrading fungus i.e. it has evolved to grow on partially degraded plant material after primary degradation by other fungi and bacteria. *Agaricus* does not grow well on non-composted plant material, and it is easily out-competed by other fungi. The purpose of the composting process is to convert plant material to a more selective form for *Agaricus* growth. This partial degradation has a number of functions

- (i) to open up the straw structure and make it available to breakdown by *Agaricus* enzymes,
- (ii) to make the straw more wettable,
- (iii) to convert the substrates to forms which favour *Agaricus* growth i.e. (a) humic compounds (the brown 'sticky' material) produced during composting which inhibit growth in many other fungi and (b) nitrogen is locked-up or sequestered as humic-protein and in bacteria. *Agaricus* has the machinery (enzymes) to grow and thrive in the presence of humic compounds.

However, while composting makes the substrate more selective for *Agaricus* growth, it does not necessarily follow that the substrate is optimal for growth. Compost contains all of the components for *Agaricus* nutrition but their concentrations depend on the chemical composition of the starting materials.

Nitrogen is a limiting component for mushroom yield. Nitrogen can be increased in compost by addition of nitrogen-rich chemicals e.g. urea, addition of protein-rich supplements like soy, and compost formulation with increased amounts of poultry manure. A range of

nutrient supplements are available for the mushroom industry. The claims made for these products are very much dependent on the substrate to which they are added. If the compost is of poor quality or lacking in certain nutrients then addition of nutrient supplements will stimulate yields but it does not necessarily follow that addition of supplements to good, well balanced composts will also stimulate yield increases. Nutritional supplements are available in different formulations and pre-treatments. The major highlighted components are given below.

1. Protein supplement is the most widely known of the nitrogen supplements. The protein is based largely on formaldehyde treated soya meal, although other protein sources (such as keratin-based) are used by some manufacturers. Soy supplementation of phase 2 composts increases yield significantly for many types of compost, but the level of yield increase is dependent on the quality of the compost, growing system and rate and method of application of the soy.
2. Lipids have been shown to have hormonal action on mycelial growth and mushroom production (Parker, 1993, Schisler and Patton, 1970). These chemicals are available in a mixture (together with protein and other chemicals) as Natural Gold and ProMycel Gold.
3. Carboxylic acids have been claimed to increase mushroom yields possibly by a hormonal effect by stimulating lignin breakdown (Parker, 2009). Carboxylic acids supplement is available for commercial use as MycroNutrient.
4. The addition of minerals and micronutrients to Phase II compost has been claimed to improve yields (Royse and Beelman, 2008). However another report questioned whether micronutrients are a production-limiting factor (Desrummaux, Calus and Sedeyn, 2000). Micromax ® is a mixture of elements: Calcium (12%), Magnesium (3%), Sulphur (12%), Boron (0.1%), Copper (1%), Iron (17%), Manganese (2.5%), Molybdenum (0.05%), Zinc (1%) and inert ingredients (57.35%).
5. There are a number of other chemicals and products with claims of yield improvement, but many of these have not been commercially successful presumably due to their lack of clear effects and/or cost. These include: cellulose, *Scytalidium thermophilum* inoculum and enzymes.

Many of these products are so different chemically that it is likely their mechanisms of action are also different. This leaves open the possibility for further increases in yield by the

combination of supplements. This project is aimed at giving clear direction and precision to this approach.

The first part of the project proposes to examine the effect of supplements on cropping performance. The second part has a different approach: to use the mushroom's own genes to report on what it 'sees' and its enzymes activities, and then to look for applications of practical use.

The aims are to understand how the mushroom obtains nutrition from compost and how the use of supplements can be maximised. Furthermore as the detailed chemical analysis of the compost substrate is technically challenging due to the complexity of the polymers, the approach of this project may lead to new measures or indicators of compost quality and yield potential.

The project consists of three components:

1. Review of the current use of supplements in phase 3 compost in the UK and European mushroom industries.
2. The effects of different supplements on mushroom production Three experiments to examine the effects of nutritional supplements on phase 3 compost by measuring mushroom yield, flushing pattern and quality:
 - a. Oct-Nov 2013 – a proving trial to test out experimental procedures and a new test cropping facility and to assess four supplement types on one phase 3 compost spawned with a brown strain
 - b. Jan/Feb 2014 – a trial with four supplement types and two phase 3 composts, and measuring mushroom yield and quality (as determined by cap colour, cap density (texture) and dry weight/fresh weight ratio). This experiment was analysed before starting the third trial to establish which supplements gave statistically strong results ahead of sampling and analysis of the effects of supplements on mushroom gene expression
 - c. May/June 2014 – a trial with four supplement types and two composts, measuring mushroom yield and quality as determined by cap colour, cap density (texture), dry weight/fresh weight ratio and flavour analysis

3. Gene expression analysis using Microarray technology to measure the expression level of every gene (a good proxy for the enzyme levels). Compost was collected from the experimental trays of the May/June 2014 trial at mid-flush (first and second) for gene expression analysis to understand *why* certain supplements are able to influence yield. This component of the project will develop our understanding of mushroom nutrition using the mushroom to report on its own biology in response to different supplements. This will provide knowledge of how supplements stimulate higher yields and how further improvements can be developed. Also the microarray analyses may lead to improved compost analysis techniques.

This annual report describes:

- Progress from the review of supplement usage in the UK and European mushroom industry (usage appears to be dynamic and changing and so we report on what the situation is at the present time)
- Results from the proving trial Oct/Nov 2013 and the trial Jan/Feb 2014. The May/June 2014 trial is taking place while this report is being written and so the results are not yet complete or analysed

Materials and Methods

The review of nutritional supplement usage

The review of nutritional supplement usage was conducted by talking to growers in the UK, Ireland, The Netherlands, Belgium and Poland, at the Mushroom Days exhibition (May 2013), HDC Mushroom Panel meetings, visits to farms and during conversations at the MushTV project meetings. We also conducted specific consultations with supplement suppliers (Table 1, below).

Table 1: Supplement suppliers and contact names

Supplement suppliers	Contacts
Amycel	John Kidder, John Clay, Hubert Hay
Champfood	Eric de Nooij
Lambert	Scott McIntyre
MCSustradd Havens	Bart de Leeuw
NutriGain	Stuart Whitehall, Frank Parker

The mushroom growth trials

The mushroom cropping trials were conducted at Moreton Pinkney Mushroom Farm, Moreton Pinkney, Northamptonshire, UK. For all three trials, three replicate trays were used for each supplement x compost treatment, and six replicate trays for unsupplemented controls. One tray of each supplemented treatment or two trays of unsupplemented controls were arranged in three areas (blocks) in the growing room.

The first trial (Oct/Nov 2013) used phase 3 compost spawned with the brown strain 'Heirloom' growing on a straw-based compost. This compost was supplemented as indicated in Table 2 and compared with non-supplemented phase 3 compost. For comparison, two trays of A15 growing on a horse manure-based phase 3 compost were also grown and the yields determined. These A15 data are shown in Figure 2 but have not been subject to statistical analysis due to the low replication.

Table 2: Supplements and their rate of addition used in Trial 1 (Oct/Nov 2013)

Supplement Type	Product Name	Rate of Supplementation (w/w)
Protein	Champfood E*	1.5%
Protein	MCSustradd Soya	1.5%
Protein	ProMycel Gold *	1.5%
Lipid/protein blend	Natural Gold	1.5%
Carboxylic acid-based	MycroNutrient	0.5%
Carboxylic acid-based	MycroNutrient	0.25%
Mineral-based nutrient	Micromax	0.15%
Mineral-based nutrient	Micromax	0.23%

* At the time of writing the manufacturer's respective web-sites describe ProMycel Gold as "A balanced nutritional content of lipids, carbohydrates, micronutrients, and multiple protein sources" and Champfood E to contain vitamins, minerals and trace elements.

For the second trial (Jan/Feb 2014), two phase 3 compost types were used: a straw-based compost and a horse manure-based compost. The spawn used for both composts was A15 (Sylvan Inc.). The phase 3 composts were filled into growing trays at 9 kg/tray, either as non-supplemented controls or mixed with supplement at the following rates (Table 3):

Table 3: Supplements and their rate of addition used in Trial 2 (Jan/Feb2014)

Supplement Type	Product Name	Rate of Supplementation (w/w)
Protein	Champfood E	1.5%
Protein	MCSustradd Soya	1.5%
Protein	ProMycel Gold	1.5%
Lipid/protein blend	Natural Gold	1.5%
Carboxylic acid-based	MycroNutrient	0.25%
Mineral-based nutrient	Micromax	0.15%

The rates chosen for the experiments were based on commercial recommendations and reported rates. In the case of Natural Gold a 1.5% rate was chosen to be consistent (rather than the recommended 1.2%) for comparison with the protein-based supplements. Due to the low inclusion rate of the MycroNutrient and Micromax supplements, these were first suspended in water at 450 and 270 g/L respectively. The water was added to the compost at a rate of 5.6 ml/kg, with the suspended supplement. Water was also added to all the other treatments during mixing at the same inclusion rate.

The composts were cased with commercial McArdle casing, cased with phase 3 compost and mushrooms were grown as per commercial conditions. Temperature probes were inserted into the composts to monitor temperature changes after supplementation and during mushroom growth.

In trial one (Oct/Nov 2013) the mushrooms were harvested over three flushes and the harvest weights (yields) were measured. The results were analysed by ANOVA (Excel).

In trial two (Jan/Feb 2014) the mushrooms were harvested over three flushes and the harvest weights were measured. Mushroom quality was determined as:

1. Mushroom cap colour using a Minolta meter which measures overall whiteness (parameter 'L') and colour (parameter 'a' for red to green, and parameter 'b' for yellow to blue). Three replicate mushrooms were examined from flushes one and two from each tray. Each mushroom was measured on the top and on two opposite sides.
2. Mushroom cap density (from the non-supplemented compost, and ProMycel Gold, Natural Gold and Champfood E treatments) by measuring the weight of a 10 mm cube of tissue. Mushroom density is closely correlated with texture or firmness (McGarry and Burton, 1994). Three replicate mushrooms were examined from flushes one and two from each tray.
3. Mushroom dry weight/fresh weight ratio: the fresh weight of approx. 150 g of mushrooms was accurately weighed from each tray and for each flush. The mushrooms were then oven dried and the dry weights determined.

All samples of composts used in the experiments were analysed for moisture, nitrogen, ammonium-N, and ash contents, pH and electrical conductivity. The moisture content of the composts was also determined at the end of cropping. The supplements were also analysed for moisture and ash contents. These values will be presented in the final report.

Data from trial 2 were analysed by Analysis of Variance (ANOVA) using Genstat. There were three replicate trays per treatment/compost type combination and six replicate trays for the non-supplemented controls. To account for any within house variation, the replicate trays were distributed in the mushroom house in replicate blocks.

Results

The review of nutritional supplement usage

Numerous conversations have been held with mushroom growers, compost manufacturers and supplement suppliers concerning usage of nutritional supplements. The situation is dynamic with growers and composters prepared to change or look at different supplements, the partial replacement of soy with keratin-based products and the recent interest in liquid casing supplements.

The key findings of these conversations are:

- >90% of phase 3 compost in Europe is supplemented because it is believed that supplement use has a beneficial effect on quality and production of mushrooms.
 - Less supplementation during summer
 - Some Heirloom growers have reduced supplement usage
 - Protein supplement most commonly used
 - Rate: 1.4 – 1.5% for protein supplements, 1.2% for Natural Gold
 - Protein from soy and other protein products:
 - soy
 - soy + various formulations (alternative protein sources and minerals)
 - NutriGain products (lipids and carboxylic acids) in commercial use
 - Cracked maize (Poland)
- There is considerable interest in the potential of liquid supplement for compost and/or casing

A list of available supplements is given in the table (Table 4) below:

Table 4: List of available supplements. The ingredients and the proportion of protein in the supplements can vary with time. The products and their compositions are accurate at the time of writing (the rate of application is given for phase 2 (Ph 2) and phase 3 (Ph 3) composts).

Company	Product	Ingredient	Protein %	Moisture %	Rate % w/w
Lambert	T6	feathermeal+ polysaccharide	44	Unknown	0.8 (Ph 2)
Lambert	T7	feathermeal+ polysaccharide	66	Unknown	0.8 (Ph 2) <0.8 (Ph 3)
Lambert	S44	soya	44	Unknown	Unknown
Amycel	ProMycel Gold	soya + feathermeal	54	7	1.5 (Ph 3)
Amycel	PROCO 50 (a new product)	rape meal + soya + trace elements	50	11	1.5 (Ph 3)
MCSustradd (Havens)	“Protein-cocktail”	feathermeal, soya, veg. proteins, rape meal	44	12	≤1.5 (Ph 3)
MCSustradd (Havens)	Soybean based	soya	?	Unknown	≤1.5 (Ph 3)
Champfood	Champfood C	soya, animal protein, rape	48	Unknown	≤1.5 (Ph 3)
Champfood	Champfood	soya, animal protein, rape meal, minerals	48	Unknown	≤1.5 (Ph 3)

Company	Product	Ingredient	Protein %	Moisture %	Rate % w/w
	E and EXC				
NutriGain	MycroNutrient	citric acid, propionic acid	0	Unknown	casing application
NutriGain	Natural Gold	Protein/lipids blend	0	Unknown	0.8 – 1.4 (Ph3)
Everris	Micromax	Ca (12%), Mg (3%), S (12%), B (0.1%), Cu (1%), Fe (17%), Mn (2.5%), Mo (0.05%), Zn (1%)	0		

Mushroom growth trials – the effects of nutritional supplements on mushroom yield and quality

Trial One (October/November 2013):

Compost temperatures

Compost temperatures of the different treatments were measured and shown in Figure 1.

Supplementation with Micromax or MycroNutrient did not change the compost temperatures compared with the non-supplemented control. However supplementation with the protein-based supplements (ProMycel Gold, Champfood E, MCSustradd and Natural Gold) did cause a rise in compost temperature compared with the non-supplemented control.

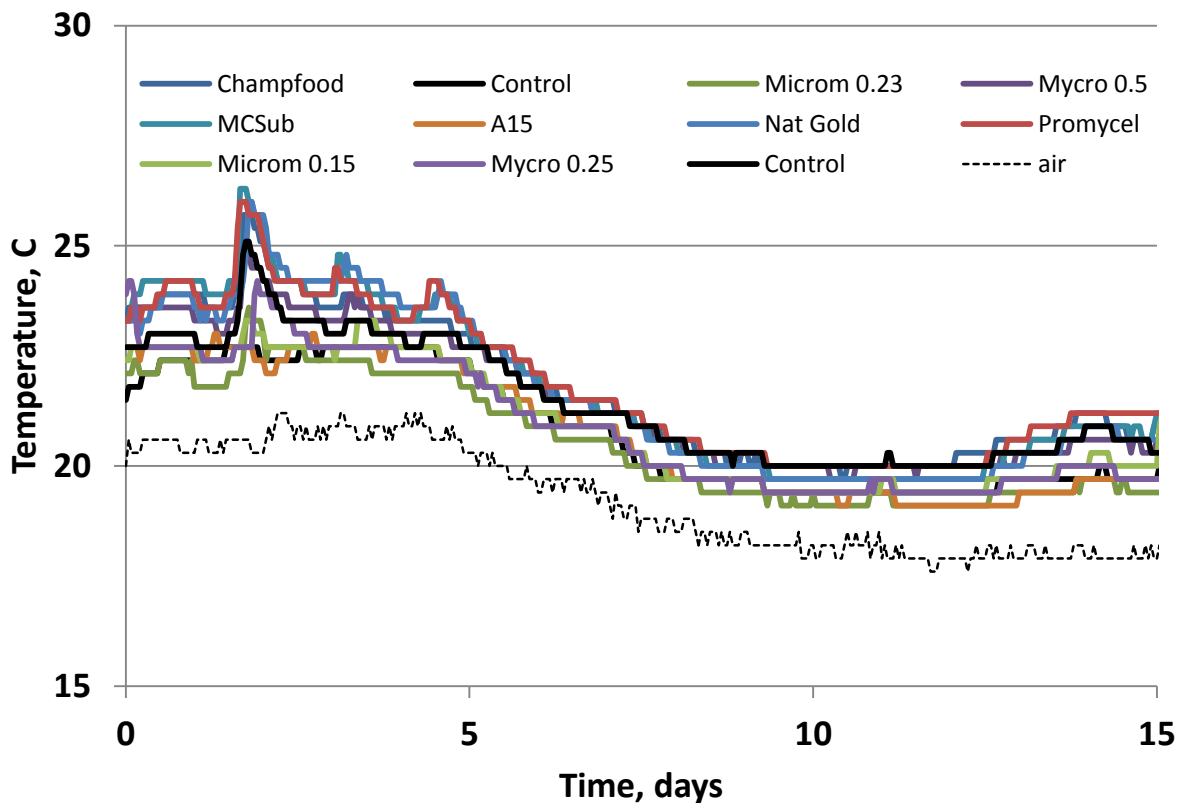


Figure 1: The effects of supplementation on compost temperatures

Effects of supplements on mushroom yield

The effects of supplements on mushroom yield over three flushes for Heirloom strain is shown below in Figure 2.

Non-supplemented phase 3 compost spawned with Heirloom strain produced a yield equivalent to 271 kg/tonne. The supplementation with MCSubstradd soya increased the yield significantly to 368 kg/tonne. Supplementation with ProMycel Gold, Champfood E, Natural Gold, MycroNutrient and Micromax had no significant effect on yield.

By way of comparison, strain A15 (non-supplemented) produced a yield of 321 kg/tonne.

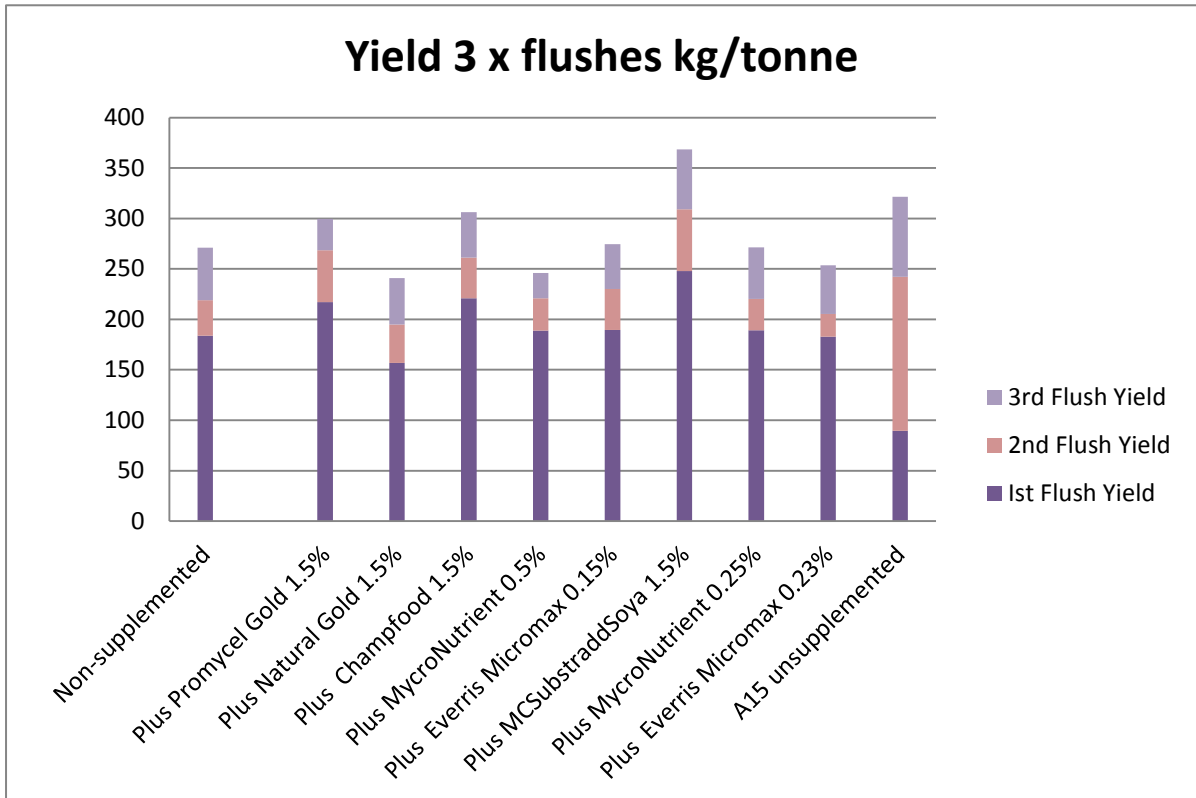


Figure 2: The effects of supplements on three flushes of mushroom yield of Heirloom strain from Trial One

The main purpose of this proving trial was to test out experimental procedures and a new test cropping facility. However the work does suggest that one of the protein-based supplements (MCSubstradd) can significantly benefit yield of a brown strain, Heirloom. This will be of interest to growers of brown strains and deserves more detailed attention in the future.

Dry weight/fresh weight ratios

The dry weight/fresh weight ratios of the mushrooms were determined and are shown in Figure 3. There was no significant difference between the treatments.

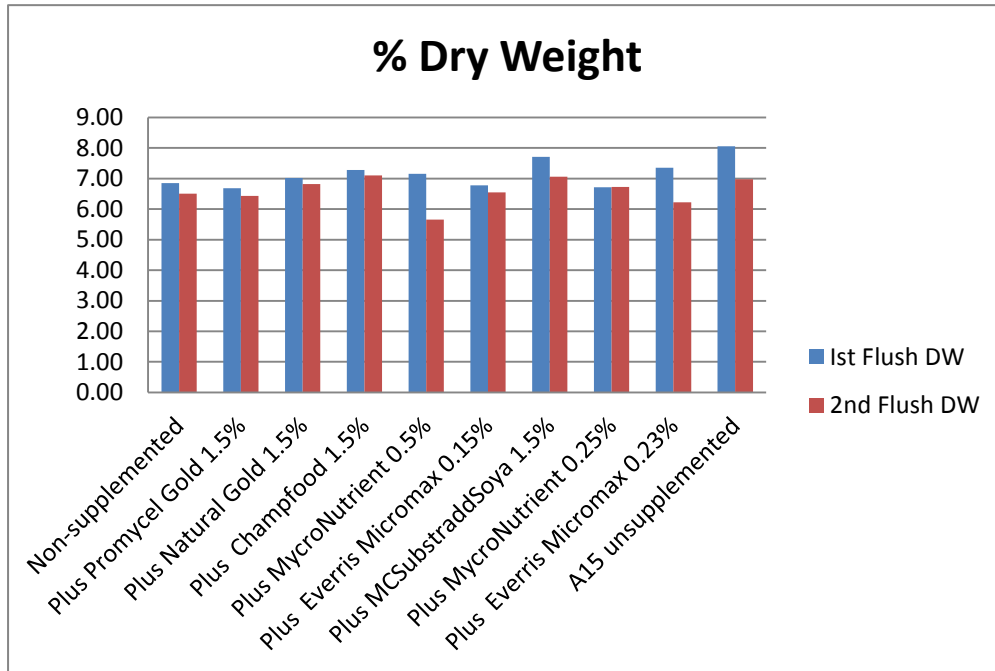


Figure 3: The dry weight/fresh weight ratios of Heirloom mushrooms from Trial One

Trial Two (January/February 2014):

Effect of Supplements on Compost Temperatures

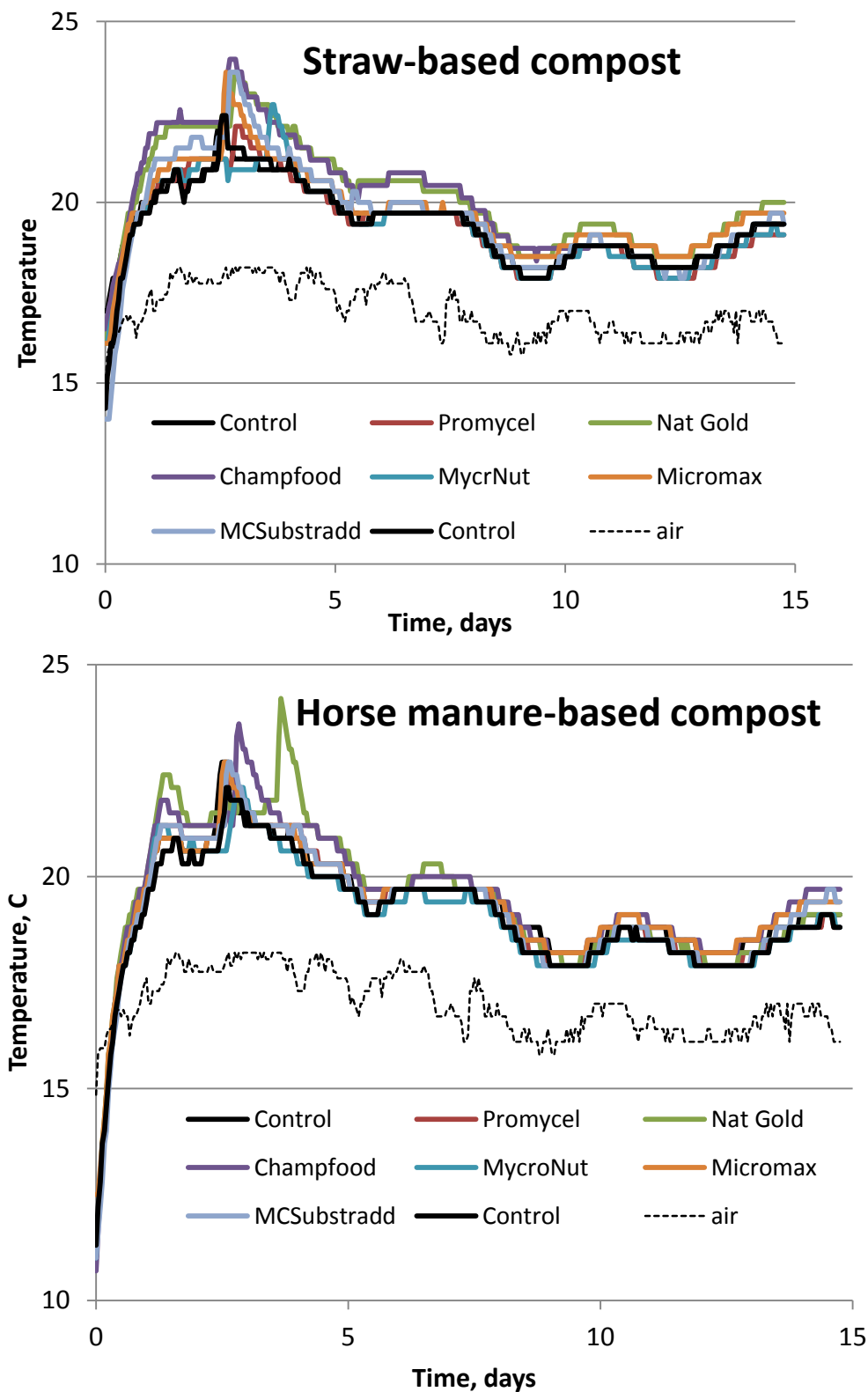


Figure 4: Effects of supplements on compost temperatures

In both composts, the Champfood E and Natural Gold resulted in the highest compost temperatures, up to 3 °C higher than the unsupplemented control treatment. The MCSustradd Soya supplement increased compost temperature by 1-2 °C above the control treatment.

Effect of Supplements on Mushroom Yields

Yields between the two types of compost were found to be statistically different, the horse manure-based compost produced a greater yield of mushrooms than the straw-based compost. However it should be noted that this represents two batches of compost and therefore it is not a thorough study of compost types but reflects a batch to batch variation.

The protein-based supplements (ProMycel Gold, Champfood E, MCSustradd Soya and Natural Gold) all improved mushroom yield (5% statistically significant) (Figure 5 below). The highest yield for the straw-based compost was with supplementation with Natural Gold and for the horse manure-based compost the highest yield was with Champfood E supplementation. Micromax and MycroNutrient had no effect on mushroom yields for both compost types.

It should be noted that MycroNutrient is marketed as a casing supplement which was not tested in this project. The MycroNutrient added to compost treatment was included to be scientifically consistent i.e. supplements added to compost. It is planned that in the future we will examine casing supplements more thoroughly.

Interesting flush effects were recorded. For the horse manure-based compost, the improvement in yield by protein-based supplements was largely in the first flush, not at all in the second flush but improved again in the third flush (at the 5% significance level). For the straw-based compost, the improvement in yield by protein-based supplements was observed in the first and second flushes but not the third flush.

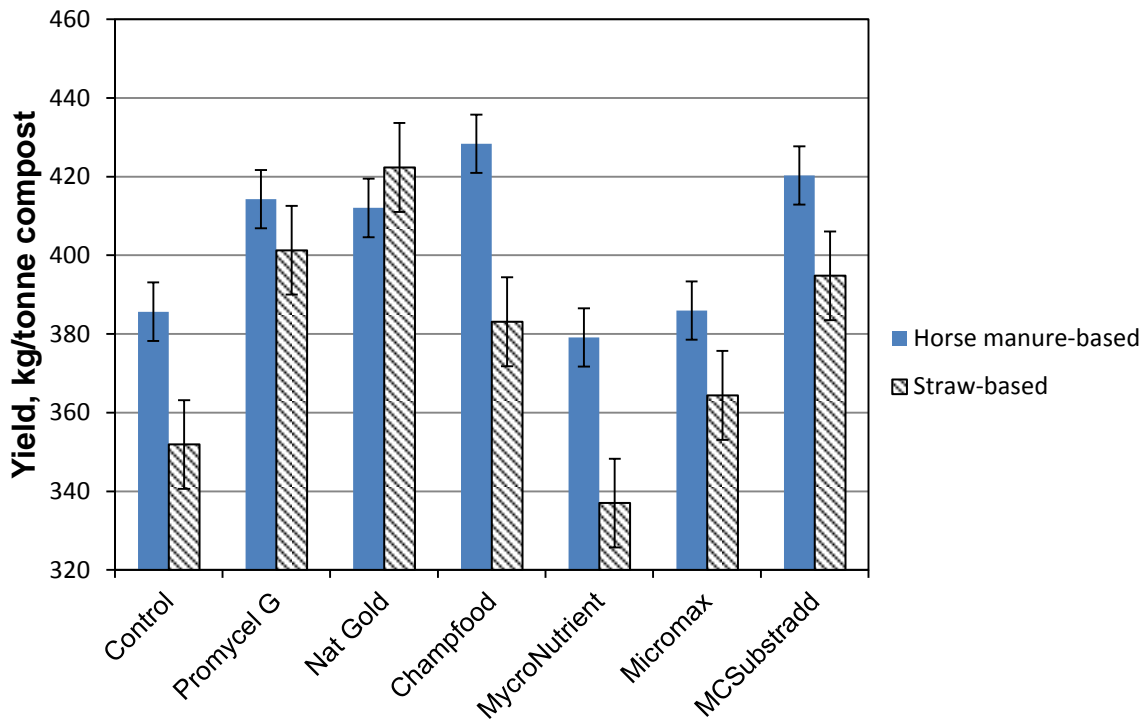


Figure 5: The effects of supplements on mushroom yield (strain A15) on the two compost types: Horse-manure-based and straw-based, from Trial Two. Error bars show SED

Effect of Supplements on Mushroom Cap Density (Texture)

ProMycel Gold and Champfood E protein-based supplements were found to increase the density of the mushroom caps (Table 5). This increase was statistically significant (5% level) when averaged over two flushes. Mushrooms grown with MCSustradd were only tested in the first flush and the effect on density was not significant. Natural Gold increased mushroom cap density on horse manure-based compost but had no effect on density using straw-based compost. Mushrooms grown on the horse manure-based compost produced denser mushrooms than those grown on the straw-based compost. Flush 1 mushrooms were denser than flush 2 mushrooms.

This result is an important one for the industry as it demonstrates the ability to improve an important quality attribute (i.e. mushroom cap texture) and the potential to improve picking rates (higher density means more weight picked per mushroom or per hour).

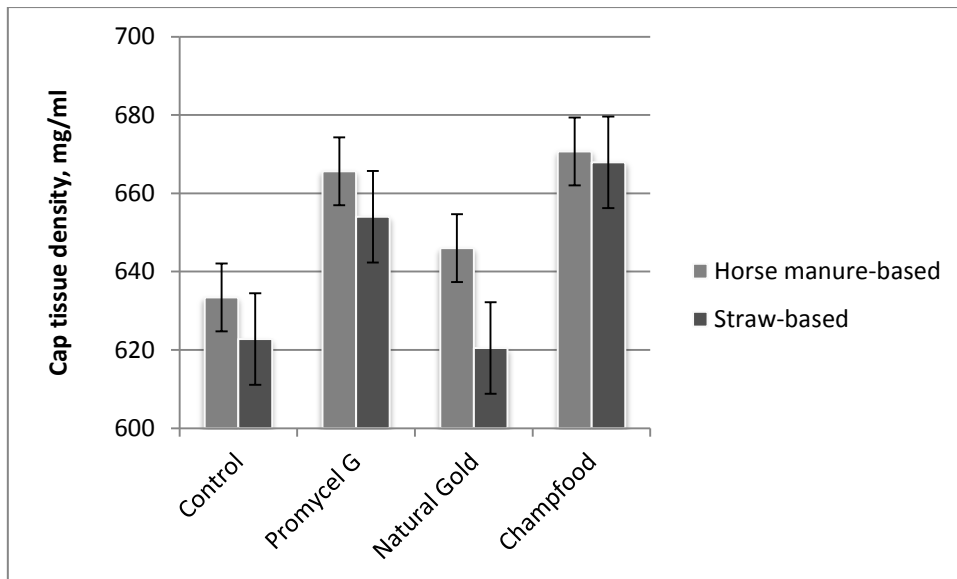


Figure 5: The effects of supplements on mushroom density (and texture) (strain A15) on the two compost types: Horse-manure-based and straw-based, from Trial Two. Error bars show SED

Effect of Supplements on Mushroom Dry Weight/Fresh Weight Ratio)

Supplementation did not change the dry matter content of the mushrooms (dry weight/fresh weight ratios) Figure 6. However, the horse manure-based compost produced mushrooms with higher dry matter than the straw-based compost. Further experimentation will determine whether this is a general trend or may represent batch to batch variability.

Also flush 1 mushrooms were found to have a higher dry matter content than flush two mushrooms, 8.0% (flush 1) compared with 7.4% (flush two).

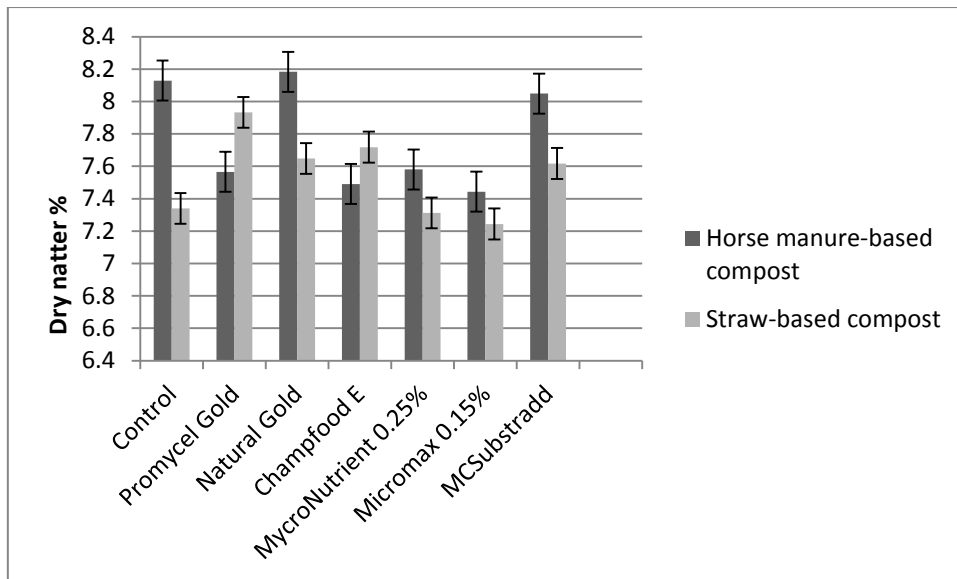


Figure 6: The effects of compost type on mushroom dry matter content (strain A15) on the two compost types: Horse-manure-based and straw-based, from Trial Two. Error bars show SED

Explanation of Mushroom Colour Measurements

Three colour parameters were measured: L, 'a' and 'b' using a Minolta meter.

L represents the overall whiteness to darkness on a black and white scale (L of 100 is pure white and an L of 0 is matt black; the more discoloured the mushrooms the lower the L value).

'a' represents the redness (positive numbers) to greenness (negative numbers). For mushrooms the figure is close to zero and so it is not very informative.

'b' represents the yellowness (positive numbers) to blueness (negative numbers).

Effects of Supplements on L Colour Values

There were no clear effects of supplements on the L value of mushroom colour.

Statistical analyses show that the straw-based compost produced whiter mushrooms (higher L value) in the second flush than horse manure-based compost (Figure 7). However no difference in whiteness was detected in first flush mushrooms between the two compost types.

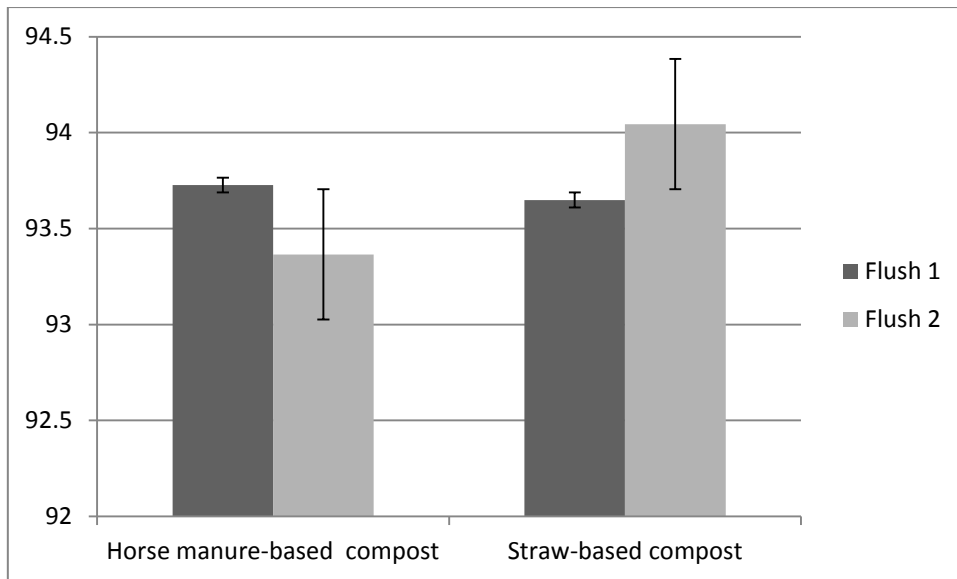


Figure 7: The effects of compost type on mushroom colour (L value) (strain A15) on the two compost types: Horse-manure-based and straw-based, from Trial Two. Error bars show SED

Effects of Supplements on ‘a’ colour values

The values the ‘a’ colour parameter were too close to zero to draw a meaningful interpretation or conclusion.

Effects of Supplements on ‘b’ colour values

The straw-based compost produced less yellow mushrooms (lower ‘b’ value) than the horse manure-based compost on first flush mushrooms, on both tops and sides of the mushroom caps (Figure 8).

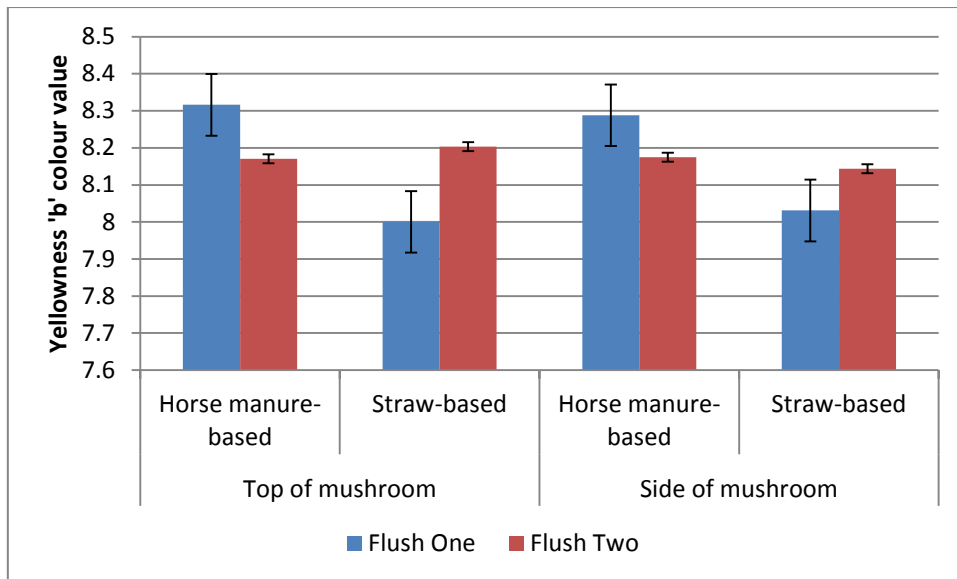


Figure 8: The effects of compost type on “yellowness” mushroom colour (b value) on the tops and sides of the mushroom cap (strain A15) on the two compost types: Horse-manure-based and straw-based, from Trial Two. Error bars show SED

Discussion

The addition of protein-based supplements to phase 3 composts increased mushroom yield. This is consistent with the view that mushroom composts (straw- or horse-manure-based) have a deficiency in nitrogen which can be overcome by protein addition. We have not examined different rates of supplementation, so we are unable to comment on optimum rates. The use of two protein-lipid mixtures (Natural Gold and ProMycel Gold) increased yield by approximately the same amount as the protein-only supplements. There is currently insufficient information to comment on any advantages of different formulations. Information from the microarray analysis in trial three may shed new light on different formulations and how the supplements work.

In terms of mushroom quality, protein-based supplements increased mushroom cap density but had no effect on the dry matter content or mushroom colour. Cap density relates directly to texture (an important quality attribute), the higher the density the better the ‘bite’ (McGarry & Burton, 1994). Cap density is determined by the number of cells in a given volume. The dry matter content is determined by amount of material (DNA, protein,

carbohydrates, lipids etc) within a cell. There is no inconsistency that the cap density increases while there is no change to the dry matter content.

Earlier work (Royse and Beelman, 2008; Desrummaux, Calus and Sedeyn, 2000) suggested that mushroom compost had deficiencies in mineral nutrients and that supplementing with Micromax can improve yields. We have not been able to repeat this finding. A possible reason for the inconsistency is that the raw materials of European mushroom compost (straw, horse and chicken manures) are subjected to greater optimisation through fertilizer and animal supplement applications than the raw materials in North America (where Royse and Beelman are based).

The work of this project is to examine supplementation to composts, it is therefore not surprising that we did not see a beneficial effect of MycroNutrient which is marketed as a casing supplement.

Thorough statistical analyses have revealed that for trial two, the type of phase 3 compost (horse manure-based or straw-based) had significant effects on mushroom yields, colour and dry matter content. We wish to emphasise that in this experiment we have examined only a single batch of each compost type and are not therefore in a position to make an overall conclusion or comment on this issue.

Conclusions

Compost supplement addition to phase 3 composts is widespread in UK and Europe but this is the first study to analyse their effects on mushroom quality parameters and attempt to identify their mode of action using microarray technology.

Protein-based supplementation to phase 3 composts has a significant effect on increasing mushroom yields and mushroom cap density. Both of these characteristics should have major impacts for the mushroom industry. Both increased yields and density can lead to greater financial rewards to growers. Also increased density which also means increased texture (as mushroom firmness or 'bite') represents an improvement in a major quality characteristic which will be appreciated by consumers.

When trial three is completed and analysed, then conclusions can be made on the effects of supplements on different batches of composts and their mechanisms of action.

Knowledge and Technology Transfer

Progress on this project (M 56) has been presented to the HDC Mushroom Panel at meetings on 2 July 2013, 14 January 2014 and 2 July 2014.

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