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Diversified Agriculture Part 1: Simplified and Lower Cost Methods for Mushroom Cultivation in Africa

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Abstract

Mushroom cultivation in more developed countries has evolved from an art into huge agri- business by way of the most innovative production technology and biotechnology available. However, there are still less-developed areas of the world where access to these technological advances are either unavailable or too expensive to utilize.

West Africa exemplifies this problem, and a low cost mushroom production program would have compounded benefits for the region. This process would allow the use of secondary crops to be produced in a short time frame from agricultural by-products derived from the primary agriculture of the region, such as millet and sorghum straw, cassava peelings, or virtually any other cellulose source. These and other potential raw materials are currently being disposed of as waste. After the production of edible or medicinal mushroom crops are harvested from these agricultural wastes, the residual mushroom substrate represents a bioconversion from the non- nutritional cellulosic material to an edible fungal matrix of high protein content which can be utilized as nutritious cattle and goat fodder as well as feedstock for tilapia farming. This diversified approach results in three successive cash crops from any agricultural operation where previously there had been only the one primary crop.

The oyster mushroom complex (*Pleurotus ostreatus* and other *Pleurotus* and Hypsizygus species) appear to be the best candidates for production in the West African climate. These mushrooms are primary cellulose decomposers, and can grow on almost any plant material substrate including banana waste, coffee residue, sugar cane baggasse, paper or cardboard waste, river grass, sawdust and nearly all other agricultural waste.

The purpose of this paper is to present low-cost, low-technology methods applicable to small- scale village mushroom production. The process obtained from this research represents the least training and smallest capital and infrastructure investment to implement a mushroom farming operation, while suggesting the easiest system to grow locally acclimatized mushroom strains for a large and valued mushroom crop in the shortest possible time with minimal financial risk for the farmers.

Introduction

A universal need for all mankind is food. Reliable supplies of low cost, nutritious and readily available food for the population are the key items to a stable and healthy society. Until this basic need is met, poverty alleviation and economic development cannot take place. To achieve the goal of stable, healthy societies, mankind over most of the world has evolved

from hunter- gatherer societies to agriculture, aquaculture and animal husbandry for production of their necessary foodstuffs. These farming endeavors have developed over centuries of social evolution, originally utilizing local plants, fish and animals as the raw inputs to make these production systems work. However, in the last century or two this has begun to change, with plants and animals from far and wide being imported into new locals as superior candidates for cultivation over the plants and animals found originally in that region.

This paper presents the first step in this expanded cultivation scheme, which is Mushroom Production through utilization of low cost, technologically simple methods that can be easily implemented at the village level in any country, be it highly developed or the poorest country on earth. Our main goals in this research were to develop real world solutions to the problems faced in implementing these cultivation systems. It is easy to imagine a system where if you have capital resources of X, Y and Z, you can achieve great results, but what about those peoples that do not have even the minimum resources of X, Y and Z?. It is to these people that we have directed this research. Using only agricultural waste which we have diverted from the disposal stream, and commonly available tools, equipment and supplies, we will show in this paper how anyone of even the most modest means can produce an economically viable crop of wholesome and nutritious food with which to feed their families and villages; the surplus of which can be exported to others as a cash crop. At the end of the first step of this diversified agriculture program the remaining mushroom waste and spent substrate can be utilized as nutritious fodder for goats, cattle, horses, camels, sheep, and fish farming. This allows a third crop, where initially there was only one. To put this in perspective, consider the typical case of a cassava farmer in Africa: the cassava is grown and harvested, and the plant waste and cassava peeling are dumped back on the ground. The next crop to be harvested by the farmer is the next crop of cassava. By implementing the system proposed here, the cassava peelings and plant waste becomes the feedstock for the mushroom cultivation which yields a second crop (mushrooms) in approximately three weeks, long before the next crop of cassava would be ready for harvest. The waste materials from the mushroom production then becomes a highly nutritious feed for animals and fish such as goats, cows and Tilapia, producing a third crop, where there originally had been but the one primary crop. This increase in efficiency with little additional input of labor or capital has perhaps the greatest potential for elevating the economic status of the rural farmer across much of the world.

Mushroom cultivation represents a very basic natural process, that of fungal decay. In nature the primary function of plants is to act as the molecular assemblers. Plants take simple compounds like water and carbon dioxide and using sunlight as energy they assemble these into complex organic compounds such as proteins, carbohydrates and fats. Upon the death of these plants, the fungi move in and become the molecular disassemblers. That is to say the fungi take these complex compounds like the proteins and carbohydrates and disassemble them back into the simple compounds which we started with, those compounds like water and carbon dioxide. As these simple compounds are released back into the environment by the fungi, they become available as raw materials for the next generation of plant growth. In the process of running through their life cycle, the fungi reproduce as do all other living things. In the case of fungi, the reproductive organ can take several forms. One particular class of fungus, the basidiomycetes, produce a large fleshy reproductive organ (or fruit body as it is named) which we know more commonly as the mushroom. Many of these mushrooms are choice edibles, and in fact represent very high quality nutrition, often equaling or exceeding meat in quantities of protein and essential amino acids. [1]

The basis of mushroom cultivation is the breakdown of cellulose. The cell wall structure of virtually all plants is a fibrous structure composed of cellulose and hemicellulose, surrounded by a structural compound called lignin. The lignin wraps around the cellulose fibers like plastic wrap. This makes for a very strong structure, allowing a tree to stand upright for hundreds of years. The cellulose and hemicellulose are made of sugars which are great sources of food, but these are protected by the lignin wrapping, which is a very stable compound and difficult to breakdown. Only a few organisms can breakdown the lignin and utilize it as a food source, thus exposing the underlying cellulose and hemicellulose for food use by other organisms. The best known and most effective of these lignin-breakdown organisms are known as the white rot fungi, of which the Oyster mushrooms are the prime examples. Oyster mushrooms are a closely related species complex, comprised of many species within the genera *Hypsyzygus* and *Pleurotus*. Many different species of Oyster mushrooms have evolved in different locations around the world, and have become more or less specialized in degrading different raw material cellulose sources (the substrate) at different temperatures, oxygen and light levels (acclimatization).

A word on species and strains: There are many "Oystereque" mushrooms, with similar growth characteristics; morphology (looks) edibility, flavor, shelf life, etc. Some of these are very closely related to each other, even being given the same genus and species name such as the common tree oyster, *Pleurotus ostreatus*. However, not all Tree oysters are completely alike even though they may have the same genus and species name. For example, one

may be growing on an oak tree in the temperate region, while another is growing on an oil palm in the tropics.

These two mushrooms may be the same species but they are different strains. They will each have evolved different enzymes to degrade their respective substrates, and they will have different growth parameters having to do with the temperature, humidity and sunlight intensity in the regions where they evolved. To try to grow the northern climate oak mushroom on a palm tree in the tropics would yield very disappointing results. Then there are other Oysteresque mushrooms that are not quite as closely related, but which still share most of the characteristics with the other Oyster mushrooms. These have names like *Hypsyzygus ulmarius*, the Elm Oyster, so named because it usually grows on Elm trees. This does not mean that it will ONLY grow on Elm trees, just that this is the usual native habitat.

Choosing the best species and strains of Oyster mushrooms is the single most important step in creating a successful mushroom cultivation program. There is no one strain that is any better than another strain, it all just depends on the climate and the substrate and the cultivation methods used. The only way to determine the best species and strain for growing in any particular situation is to experiment with several types of Oyster mushrooms and record the production time and yield for each strain in that particular circumstance. Generally, a local strain of mushroom isolated from nearby the growing location is the best choice, as this local strain will have become acclimatized to the local environment and the local competitors for the food source.

For the purpose of this research, we choose several strains of oyster mushrooms to indicate the possibility of different strains being utilizable in this cultivation method, and to show that the results obtained in this research were not strain specific. It should not be assumed that the results shown here will necessarily transfer 100% to another location, strain or substrate, although the general guidelines shown here will certainly apply. Anyone desiring to follow this methodology of mushroom cultivation is encouraged to experiment with different species and strains, and to include locally acclimatized strains whenever possible. In some cases such as northern climates, it is not unusual to see several different species or strains cycled through a farm over the course of the year as the seasons change. This way, one can assure the maximum yield both in the hot season and in the cold season by selecting appropriate strains according to prevailing temperature. Likewise, it is often preferable to cycle different strains through the farm seasonally depending on substrate availability. If you have a strain that grows well on straw, you should use that one at grain harvest time when straw is readily available, but if your only substrate is river grass or cassava peeling, by all means be flexible in switching to strains particular to these substrates so as to maximize your harvest.

The most basic concept of mushroom cultivation is that we need to produce an environment in the substrate that is selectively preferential to the growth of our target species of mushroom, and less amenable to other types of microorganisms and pests. This involves sterilization (completely killing any other organisms that are present in the substrate which would compete with the mushrooms for utilization of the substrate as food) or pasteurization (killing off the majority of competitive organisms). Mushroom cultivation in more developed countries has evolved from an art into huge agri-business by way of the most innovative production technology and biotechnology available. This usually means the use of large pressure chambers and high temperature steam for sterilization of the substrate. However, steam and the associated equipment are both expensive and technically demanding in terms of training and education.

There are many less-developed areas of the world where access to these technological advances are either unavailable or too expensive to utilize. In those areas it would be much more practical to look at ways to pasteurize rather than sterilize the substrate, and by methods other than the use of steam. This can be done in many ways, such as using commonly available substances such as soap or hydrated lime to pasteurize the substrate. The mechanism by which these chemicals work is through the rapid change in osmotic pressure, causing the majority of microorganisms present to burst from the change in osmotic pressure. Once the substrate is treated in this way, and the majority of the microorganisms are killed, the substrate is suitable for the introduction of our target species, the Oyster mushroom.

Another potential treatment method is simply to soak the substrate in water for a week or so. This results in a rapid bloom of the bacteria that rapidly consume the readily available simple sugars. Once the sugars are exhausted after about 5 days of fermentation, the bacterial bloom dies off due to lack of any more simple nutrient availability. This leaves a substrate which is more selective to the higher cellulose degrading organisms such as Oyster Mushrooms. This is the simplest method of substrate preparation; however it is the least effective of any methods we have tried. It does work, but the chemical treatment methods are much more effective and not considerably more difficult or expensive.

Materials and Methods

Substrates Used

Unshredded wheat straw (whole stalk), shredded wheat straw (reduced in a hammer mill to small particle size).

Wheat straw is the post-harvest stalk of *Triticum aestivum* and Ground maize cobs (*Zea mays*) were used as the substrates. All straw was obtained from S&W feeds, Carson City, NV. Ground maize cob obtained from Benson's Feed and Tack, Carson City, NV.

Species and Strains Used

Although there are several fungi capable of white rot degradation, the following genus and species in the *Tricholomataceae* family were used in this experiment: *Pleurotus pulmonarius* AX strain, *Hypsizygous ulmarium* ELM 1 strain, *Pleurotus D'jamor* PDJ strain, *Pleurotus Sajar-caju* PSAJ strain, and *Pleurotus ostreatus* TL strain. All cultures used in this research are commercially used in mushroom farming, and were all provided by Aloha Medicinals Inc. of Carson City, Nevada.

Spawn Used

Spawn is the seed stock which is used for inoculating the substrate for mushroom production. The spawn for this research was generated under the usual conditions which will be known to anyone familiar with spawn production. Spawn is generally grown on grain or sawdust. The spawn substrate used for this project was White Sorghum grain, which was cooked with a measured quantity of water to achieve a moisture content of approx. 55%. A small amount of ground oyster shell was added to control the pH. Approx. 400g oyster shell per 100kgs of dry grain was added, along with 102kgs of water, which resulted in a pH of 7.0. After cooking with water and oyster shell, gypsum was added to keep the individual grains of sorghum separated and reduce the possibility of anaerobic conditions forming in the spawn. The cooked, gypsum treated grain was filled into glass jars of 1liter size until jars were approx. ¾ full, which measured an average of 454g of weight per bottle. The top of the jars were covered with a filter made of thick paper and a metal lid screwed over the filter paper. The metal lid has a single central hole drilled, measuring approx. 25mm in diameter. This allows gas exchange of the spawn substrate while eliminating the introduction of any foreign organisms during the spawn grow-out period. These filled, capped jars were then sterilized in an autoclave at 17 psi steam pressure (approx. 1.2 bar) for a period of 2.5hours. After the sterilized jars were cooled overnight, a small piece of inoculum of the appropriate strain was introduced into each jar under sterile conditions, as will be known by anyone familiar with the art of spawn making. The inoculated grain jars were then grown for 10 to 12 days before use, which resulted in completely colonized grain of only the target species of mushroom. The short growth period of 10 to 12 days means there was some percentage of unconverted grain starch remaining in the spawn, which becomes a nutritional amendment to the final mushroom substrate, raising the nutrient level of the straw without raising the risk of contamination. The concept of using young grain spawns as both inoculum and nutrition amendment is important to the simplified cultivation process presented here. Simply adding a rich nutrient amendment to an otherwise poor nutrient base like straw or maize cobs will increase the risk of contamination by unwanted competitive organisms, which would lead to reduced yield of the target mushroom fruit bodies. By using the grain spawn as mentioned here, the grain is already fully colonized by the target mushroom strain, which in turns eliminates all competitors within that jar, while still providing a rich nitrogen and starch base for quick initial growth in the final fruiting substrate for the mushroom organism.

Spawning

Spawning is the term used in inoculating the final substrate in order to grow mushrooms. In this research 2.73 kg (6 pounds) of treated straw was added to a growing bag for each strain and treatment type. All treatment methods and strains were done in triplicate time three, or in other words a minimum of nine bags were used for each treatment method and strain, insuring statistical reliability of the data generated.

The growing bags in this research were spawned with the addition of 454 g (one pound) of spawn per bag, added alternately with a layer of straw as the bags were filled. The layers of straw were approx. 50 mm thick, then a sprinkling of spawn, before adding the next layer of straw, followed by another layer of spawn until the bags were full. This represents a spawn to substrate ratio of approximately 1:6. This is considered a heavy rate of spawning, but any spawn ratio from 1: 5 up to 1: 40 can be used. It is generally considered that the higher the rate of spawning (the more spawn used per bag) the better the results in consistency and yield, while a lower rate of spawning (less spawn per bag) is cheaper, but with a correspondingly lower yield of mushrooms per bag. The spawn rate chosen in farm practice is usually a reflection on the price and availability of spawn in the local region. Whenever possible, it is always better to use more spawn rather than less.

Growing Bags used

Clear polyethylene bags of approx. 300 mm x 700 mm x 2 mil thickness were used as growing containers for this experiment. The bags were filled with substrate and spawn, with the excess air squeezed out and the bags sealed by twisting a short metal wire or twine around the top. After 48 hours post-filling, the bags were ventilated by puncturing repeatedly with a knife blade. The knife blade was 20 mm wide and ventilation holes were made on a random spacing

approx. 100 mm apart all over the surface of the bag. These holes both allowed gas exchange and a place for the mushrooms to erupt from when fruiting.

Substrate Treatments

A control batch of substrate for comparisons was made by first soaking the whole straw for 24 hours in barrels of water, and then draining the straw by setting it on racks and covering by newspaper for two hours resulting in approx. 60% moisture content in the straw. The straw was then treated with steam at atmospheric pressure for 2 hours (maintaining ~100 degrees C). The bins were then tipped so that the excess water condensed from the steam treatment could drain off for two hours while cooling. This straw was filled into growing bags and spawned in an identical fashion to the experimental bags and grown in the same room and under identical conditions throughout the trial period. This method of steam treatment is well known in the mushroom industry and is well understood by anyone familiar with mushroom farming, and so forms the comparison for all other substrate treatment methods. Straw treated with steam had no detergent, lime or other substances added.

A second method of substrate conditioning used was the natural fermentation of one bale of wheat straw (approx. 30 kgs) in water. The straw was soaked in barrels containing 200L of clean tap water for seven days with no other substances added. The straw was removed from the barrels after seven days and placed on racks for two hours, covered with newspaper in order to drain excess water and allow out-gassing and aeration of the straw. As can be imagined, after soaking this straw for seven days in the sun it was a stinky, sticky mess, but actually had a fairly low microorganism load due to exhaustion of the simple sugars and other readily utilizable food sources upon which the first generation of decay organisms depend. This fermented straw was layered with spawn (454 g bottle) in four plastic bags for each test strain until a total weight for each bag was 2.73 kg (6 pounds). When the bags reached the proper weight, they were compressed by hand and tied off using segments of light metal wire. These bags were kept outside in the shade for two weeks of observation and then moved inside a climate controlled grow room. While this treatment method did produce edible mushrooms, it did not result in commercially viable quantities of mushrooms being produced. This method of substrate preparation is not very effective when compared to the other methods utilized in this trial, and the data on this treatment method is not shown in the results.

Two more bales (approx. 30 kgs ea) of unshredded wheat straw were soaked in several barrels containing 200L water each and treated with one of the following substances for 24 hours:

- 355 g per 200 L water of hydrated lime (Calcium hydroxide)(Rockwell Lime Co. Manitowoc, WI, 54220).
- 710 mL per 200 L water 5% Chlorine Bleach (Clorox Co. Oakland, CA 94612.
- 120 g clothes washing powder per 200 L water (Tide brand clothes washing detergent, Procter & Gamble, Cincinnati, OH).
- 240 g per 200 L water Tri Sodium Phosphate (Savogran Co. Norwood MA 02062).

After soaking in the above solutions for 24hrs, the straw was laid out on top of large metal racks and drained in the same manner as the fermented straw.

Shredding Straw and Maize Cobs

When shredding the straw for the second portion of this experiment, dry straw was run through a 50 HP Jacobsen Hammer Mill for shredding. The shredded straw was soaked and processed in the same fashion as the regular unshredded straw. The bags used for the shredded straw were hand compressed more tightly than the regular straw after inoculating each, due to the smaller particle size of the substrate. This additional compression was possible due to the increased density of the shredded straw.

In addition, a drum containing 118 mL bleach was mixed with 200L water and filled with shredded maize cob. Once the maize cob was sufficiently drained of excess water by straining through small mesh chicken wire, the material was filled into identical growing bags and spawned. For spawning, bottles containing 454 g of spawn were mixed intermittently with the maize cob in a manner similar to that described for the straw. The maize cob substrate treated with bleach resulted in yields very similar to the straw substrate. The outcome for the maize cob substrate is not shown in the results simply in the interest of conserving space. It is notable that the information regarding straw can be used representatively with maize waste and with most other cellulosic agricultural waste.

All of the shredded straw bags and the maize cob bags were compressed by hand and tied with string, wire or rubber bands. They were then placed on racks in the same growing room as the unshredded straw bags. The growing room was maintained at a temperature of 21 to 25 degrees C. Humidity was maintained with the use of humidifiers from 80% to 90% RH. Light cycle was natural shaded light, approx. 12hrs per day of light and dark.

Picking Procedure

Mushrooms were checked daily and picked at specific times in their growth cycle: eg. When the edges of the

caps were flattening out or showing signs of slight curling. The mushrooms were picked, weighed, and put into a refrigerator. Pictures and weights were taken and recorded at each harvest.

Observation Sheets

Observations were taken bi-weekly in order to note the difference and similarities between growth for each treatment method and individual strains. Characteristics noted included the amount of straw sprouts, bacterial contamination, competitive mold contamination, mycelial growth, number of primordial formation (not yet matured fruit bodies called pins) and fruit body clusters, and the weight of the harvest.

Bag Disposal

Bags that became overly contaminated by bacteria or competitive molds were weighed using an accurate lab scale (Yamato Corp. Colorado Springs, CO 80906) and disposed of immediately to avoid contamination of the remaining bags. After the remaining bags passed two months of active harvest, they were also weighed and disposed of.

Biological Efficiency Equations

Biological efficiencies (BE) for each strain were attained by dividing the average fresh weight totals for that treatment method and strain type by the average dry weight of substrate and multiplied by 100. This resulting figure is given as a percentage, with the figure of 100% BE equal to 2.5 kgs of dry weight substrate yielding 2.5 kgs fresh weight mushrooms. In this study, it was not uncommon to see BE's in the neighborhood of 150% to 180%.

Results and Discussion

In the third world, protein-energy malnutrition (PEM) affects 500 million people and kills 10 million annually [2]. To avoid ill health from certain diseases, and death caused from nutritional deficiency, daily dietary protein consumption is essential [3,4].

In countries that suffer from widespread protein deficiency, available potential food sources are generally full of plant fibers from the terrestrial biomass. Wood and straw make up about 80% of this biomass, which equates to approximately 800 billion tons worldwide, an extremely large reservoir [4-6]. Although four stomached mammals (ruminants) such as cows, goats and sheep produce the enzymes necessary to digest the cellulose present in plants such as grasses and hay, they are unable to directly utilize the biggest cellulose reservoir; woody biomass, which remains untouchable because of the lignin that is structurally wrapped around the cellulose. Lignin is a complex chemical compound found in plant cell walls that is cross-linked with other cell wall components, and acts as an integral part of the secondary cell walls of plants [7] and some algae [3]. As a biopolymer lignin is unusual because of its assorted composition and lack of defined primary structure. Lignin is most commonly noted for support through strengthening of wood (xylem cells) in trees [5,6,8] and is generally only degradable by highly specialized enzymes associated with very few organisms [8]. Therefore, this molecule is also generally associated with reduced digestibility which helps defend against pathogens and pests [9]. In order to make use of this cellulose, the lignin first needs to be stripped, leaving the cellulose exposed. As mentioned above, all forms of White-rot fungi such as Pleurotus ostreatus are able to oxidize the non- digestible lignin, setting free the cellulose and giving both the fungi and other potential consumers such as ruminants ready access to this significant nutrient reservoir.

As will be seen from the graphs and illustrations, this experiment clearly shows the practicality of raising mushrooms on low cost (or free) agricultural waste such as straw, that has been treated by simple and low cost methods such as soaking with a bit of lime or washing powder. In fact, the yields seen with these low cost treatment methods were not substantially different than the results seen in the steam treated control group. The biggest factor in yields was the density of the substrate, with shredded straw showing considerably higher yields than was seen with unshredded straw.

The results of this experiment show no significant difference between the mushroom marketability and characteristics such as fruit body, yield, taste, color, smell, etc. of the low cost, low technological methods described here as compared to the high technological steam-based methods used with most mushroom farming in developed nations today.

Implementing a low tech mushroom farming approach and low cost mushroom production program in West Africa would have compounded benefits for the region. This process would allow the use of secondary crops to be produced in a short time frame from agricultural by- products derived from the primary agriculture of the region. These and other potential raw materials are currently being disposed of as waste. After the production of edible or medicinal mushroom crops from these agricultural wastes, the residual mushroom substrate represents a bioconversion from the non-nutritional cellulosic material to an edible fungal matrix of high protein content which can be utilized as nutritious cattle and goat fodder as well as feedstock for tilapia farming. This diversified approach results in three successive cash

crops from any agricultural operation where previously there had been only the one primary crop.

The oyster mushroom complexes used for this experiment appear to be one of the best candidates for production in the West African climate. These mushrooms are primary lignin/cellulose decomposers, and can grow on almost any plant material available for substrate including banana waste, coffee residue, sugar cane baggasse, cassava peelings, paper or cardboard waste, river grass, sawdust and nearly any other agricultural waste.

Other studies have been successful in revealing semi-low-tech methods for recycling agricultural waste and utilizing white-rot fungi for the production of fodder for ruminants which showed fast and reliable conditioning of the substrate [10]. This type of study was also able to help in the revitalization of the local grazing areas from being arid and dry from overgrazing to lush and green. To take this idea one step further, we explored different detergent solutions as a method for pasteurization to find which will provide the best protection against outside contamination, while still producing the highest yield. In this way farmers will be able to produce less expensive fodder for their herds while having a commodity that will feed and support their families as well as raise their economic status and maintain the land. This type of technique has been shown to decrease the land needed to maintain healthy livestock and increase employment availability. An addition to these obviously stated facts, the first harvest of oyster mushrooms occurs only 18 to 21 days after inoculation. There are few other crops that can give the farmer an income in less than 3 weeks. Many of the graphs employed are calculated using the values found for biological efficiency (BE). BE is the amount of growth determined by a comparison between the dry weight of substrate compared to the fresh weight of the mushrooms harvested. BE indicates the total percentage of growth in terms of mushroom fruit bodies. The figures below indicate a significant increase in BE when using shredded straw as the substrate for all the treatment types. In fact, substrate density appears to be the greatest determining factor in mushroom yield, at least for straw substrate and the strains of oyster mushrooms tested in this experiment.

Figure 1a shows the steam treatment comparison between unshredded and shredded straw. Steam treatment was used as a control in this experiment for both shredded and unshredded straw as it is a known and regularly used method for substrate sterilization. The AX strain was found to have an efficiency of 129.8% on shredded straw and 34% on unshredded straw, which is a 95.8% difference. The ELM 1 strain had an efficiency of 175.6% on shredded straw and 43.3% on unshredded straw, a 132.3% difference. The PDJ strain had an efficiency of 102.3% on shredded straw and 24.6% on unshredded straw, a 77.7% difference. The PSAJ strain had an efficiency of 119.1% on shredded straw and 30.5% on unshredded straw, an 88.6% difference. As seen in this graph, the biological efficiency for any of the Pleurotus strains using the shredded straw as a substrate and steam as the treatment gives a substantially higher yield than using unshredded straw as the substrate.

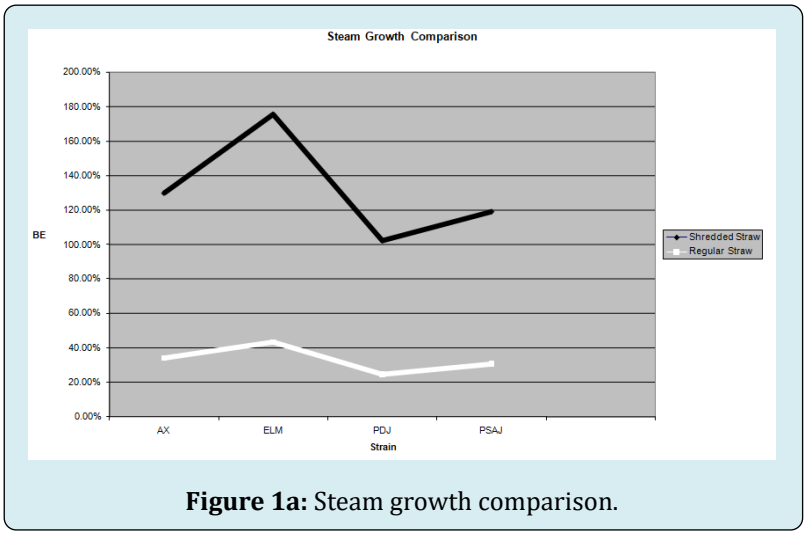

Figure 1a: Steam growth comparison.

Figure 1a shows a direct comparison between the Bio efficiencies of shredded straw to regular unshredded straw for each of the representative Pleurotus strains, AX, ELM, PDJ, and PSAJ using an autoclave for the steam sterilization method (122°C 1 bar pressure).

Figure 1b shows the Bleach treatment comparison between unshredded and shredded straw using 710 ml bleach diluted with 200L water. The AX strain was found to have a biological efficiency of 129.3% on shredded straw and 46.2% on unshredded straw, an 83.1% difference; while the

ELM 1 strain had an efficiency of 129.1% on shredded straw and 67.6% on unshredded straw, an 83.1% difference. The PDJ strain had an efficiency of 84.2% on shredded straw and 25.1% on unshredded straw, a 59.1% difference; while the PSAJ strain had an efficiency of 96.6% on shredded straw and 25.6% on unshredded straw, a 71% difference.

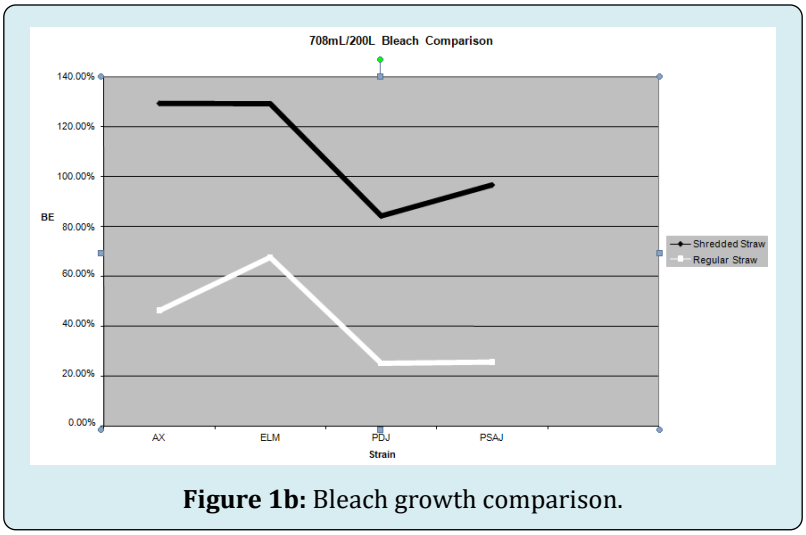

Figure 1b: Bleach growth comparison.

Figure 1b shows a direct comparison between the Bio efficiencies of shredded straw to regular unshredded straw for each of the representative Pleurotus strains, AX, ELM, PDJ, and PSAJ using the 708 ml bleach to 200L of water sterilization method.

Figure 1c shows the Lime treatment comparison between unshredded and shredded straw using 355 g lime diluted with 200 L water. The AX strain was found to have an efficiency of 126.7% on shredded straw and 33.2% on unshredded straw, a difference of 93.5%. The ELM 1 strain had an efficiency of 168.9% on shredded straw and 53.2% on unshredded straw, a difference of 115.7%. The PDJ strain had an efficiency of 165.9% on shredded straw and 23.14% on unshredded straw; a difference of 142.76%.The PSAJ strain had an efficiency of 117.1% on shredded straw and 29.23% on unshredded straw, a difference of 87.87%.

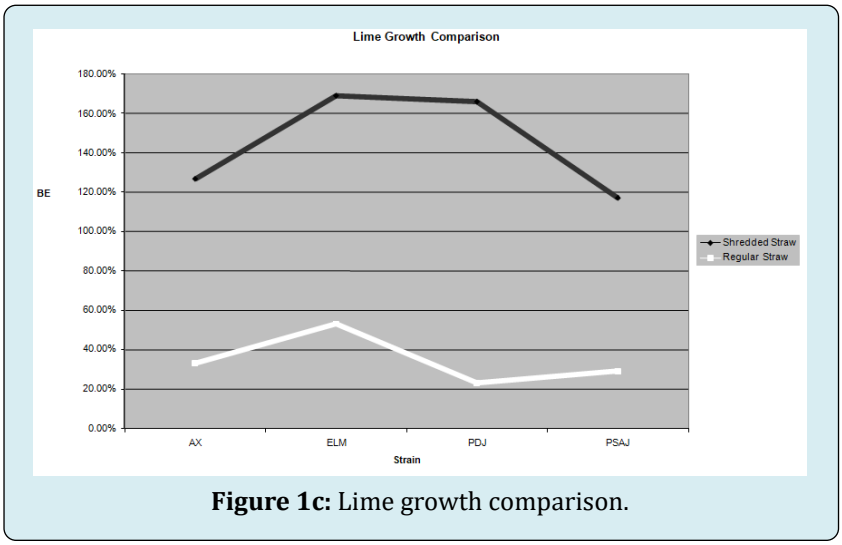

Figure 1c: Lime growth comparison.

Figure 1c shows a direct comparison between the Bio efficiencies of shredded straw to regular unshredded straw for each of the representative Pleurotus strains, AX, ELM, PDJ, and PSAJ using the 355 ml lime to 200L of water sterilization method.

Figure 1d shows the Tide brand washing powder treatment comparison between the unshredded and shredded straw using the 118.3 ml tide washing powder diluted with 200 L water. The AX strain was found to have an efficiency of 134.7% on shredded straw and 79.5% on

unshredded straw; a difference of 55.2%. The ELM 1 strain had an efficiency of 128% on shredded straw and 37.9% on unshredded straw; a difference of 90.1%. The PDJ strain had an efficiency of 83.2% on shredded straw and 23.13% on unshredded straw; a difference of 60.07%. The PSAJ strain had an efficiency of 87% on shredded straw and 39.7% on unshredded straw; a difference of 47.3%. As seen in graphs 1a through 1d, the biological efficiency for any of the test strains using the shredded straw as a substrate gives a noticeably higher yield than using unshredded straw as the substrate for all of the treatment methods.

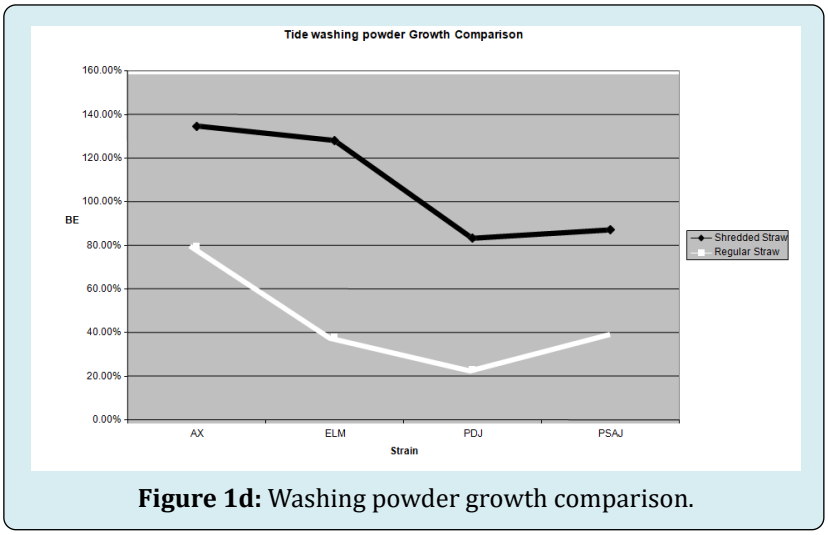

Figure 1d: Washing powder growth comparison.

Figure 1d shows a direct comparison between the Bio efficiencies of shredded straw to regular unshredded straw for each of the representative Pleurotus strains, AX, ELM, PDJ, and PSAJ using the 118.3 ml tide to 200L of water sterilization method.

Figure 2 shows the direct biological efficiency comparisons for shredded straw versus unshredded straw for each of the four test strains used. As seen in this graph, none of the regular unshredded straw strains for any of the treatment methods reached a biological efficiency higher than 80% while none of the shredded straw strains reached a biological efficiency lower than 83.20%. It is interesting to note that the steam treatment method did not have the highest bio- efficiency for all representative strains; which shows the potential for varying growth requirements among this family of mushroom.

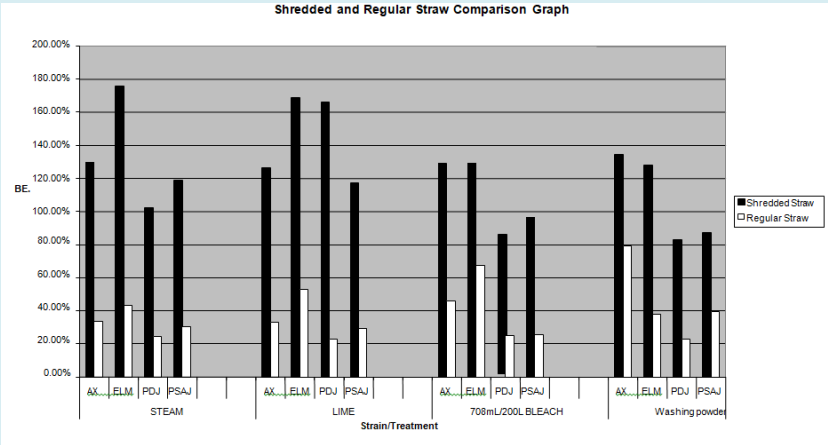

Figure 2: Unshredded straw vs. Shredded straw biological efficiency comparison graph Shredded and Regular Straw Comparison Graph.

Figure 2 shows the direct comparison of Bio-efficiencies for each of the four Pleurotus strains grown using the four treatment methods for shredded straw versus regular straw.

Figure 3 shows a direct comparison between the quantity of fruit bodies (fresh weight) picked in grams for both the unshredded and shredded straw for each of the treatment methods Steam, Lime, Bleach, and Washing Powder. This graph includes only representative Pleurotus strains grown on both the shredded and unshredded substrate. It is interesting to note that the regular straw gave higher yield average for ELM 1 treated with bleach and AX treated with washing powder only, all the other strains treated with any of the treatment methods remained that the shredded straw gave better yield in terms of fresh weight.

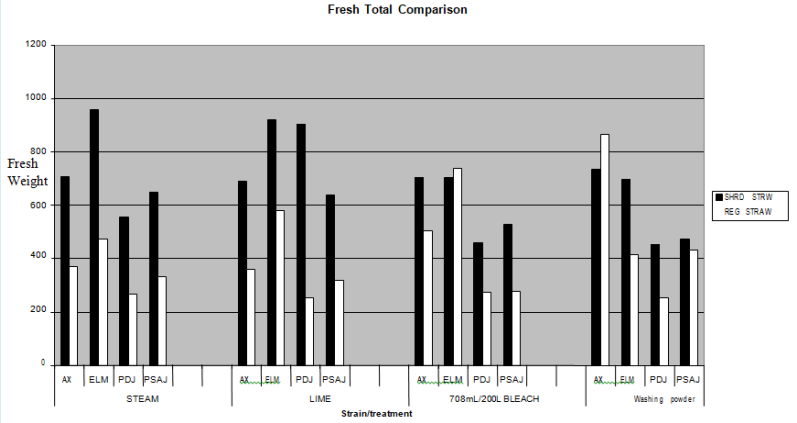

Figure 3: Regular straw vs. Shredded straw fresh weight totals Fresh Total Comparison.

Figure 3 shows a direct comparison between the amount of fruit bodies (fresh weight) picked, in grams, for both the regular unshredded and shredded straw for each of the treatment methods Steam, Lime, Bleach, and Tide.

Figure 4 shows the comparative all-strain average biological efficiencies for each treatment method for both the regular unshredded and shredded straw substrates. The Steam treatment averaged a biological efficiency of 132% on shredded straw and 63% on unshredded straw; the bleach treatment averaged a biological efficiency of 110% on shredded straw and 73% on unshredded straw; the lime treatment averaged a biological efficiency of 145% on shredded straw and 69% on unshredded straw; and the washing powder treatment averaged a biological efficiency of 108% on shredded straw and 90% on unshredded straw.

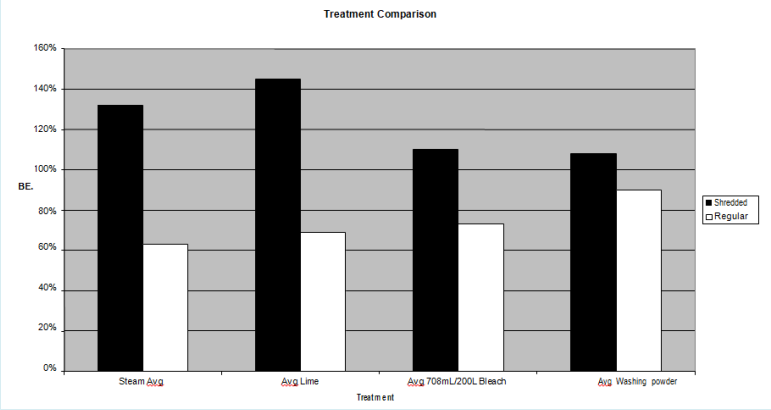

Figure 4: Regular straw vs. Shredded straw treatment averages Treatment Comparison.

Figure 4 shows the comparative all-strain average bio-efficiencies for each treatment method for both the regular unshredded and shredded straw sub straights.

Figure 5 shows the average biological efficiencies of both unshredded straw and shredded straw substrates for each representative strain. The AX strain averaged a biological efficiency of 130% on shredded straw and 62% on unshredded straw; ELM 1 averaged a biological efficiency of 150% on shredded straw and 67% on unshredded straw; PDJ

strain averaged a biological efficiency of 109% on shredded straw and 42% on unshredded straw; and the PSAJ strain averaged a biological efficiency of 105% on shredded straw and 51% on unshredded straw.

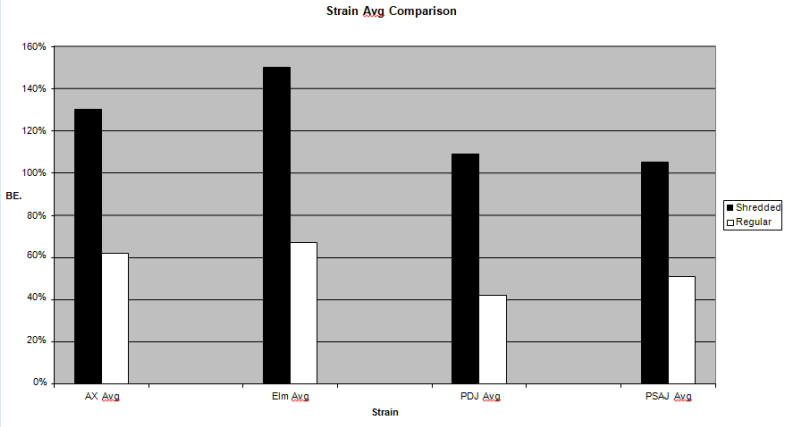

Figure 5: Unshredded straw vs. shredded straw strain averages.

Figure 5 shows the average bio-efficiencies of both regular unshredded straw and shredded straw substrates for each representative Pleurotus strain.

Figure 6 shows representative photographs of the Pleurotus fruit bodies on the densely packed shredded straw sub straight using a variety of treatment methods; notice that the quantity of fruit body clusters on each of the bags are more numerous than the quantity of fruit bodies shown in figure 7; this is due to the more densely packed shredded straw substrate. Picture 1 is a bag of the AX strain treated with washing powder. Picture 2 is a bag of the PSAJ strain also treated with the washing powder treatment method. Picture 3 shows bags of the PDJ and AX strains both treated with washing powder. Picture 4 shows fruit body clusters of an ELM 1 bag treated with the steam treatment method.

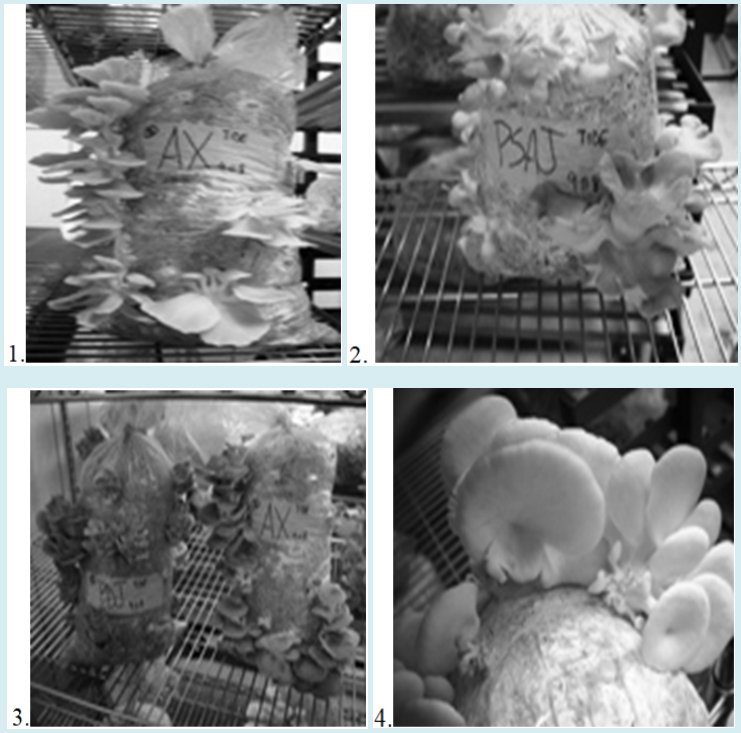

Figure 6: Photos of shredded straw mushrooms.

Figure 6 shows representative photographs of the Pleurotus fruit bodies on the densely packed shredded straw substrate using a variety of treatment methods

Figure 7 shows representative pictures of fruit bodies growing on unshredded straw treated with the four representative treatment methods. The pictures in this figure can be compared to the pictures in figure 6 in order to see the difference in growth between shredded and unshredded straw when used as the substrate. Picture 1 shows the AX strain fruit bodies treated with washing powder. Picture 2 shows the ELM 1 strain fruit bodies treated with steam. Picture 3 shows PDJ strain fruit bodies treated with washing powder. Picture 4 shows the PSAJ strain fruit bodies treated with lime.

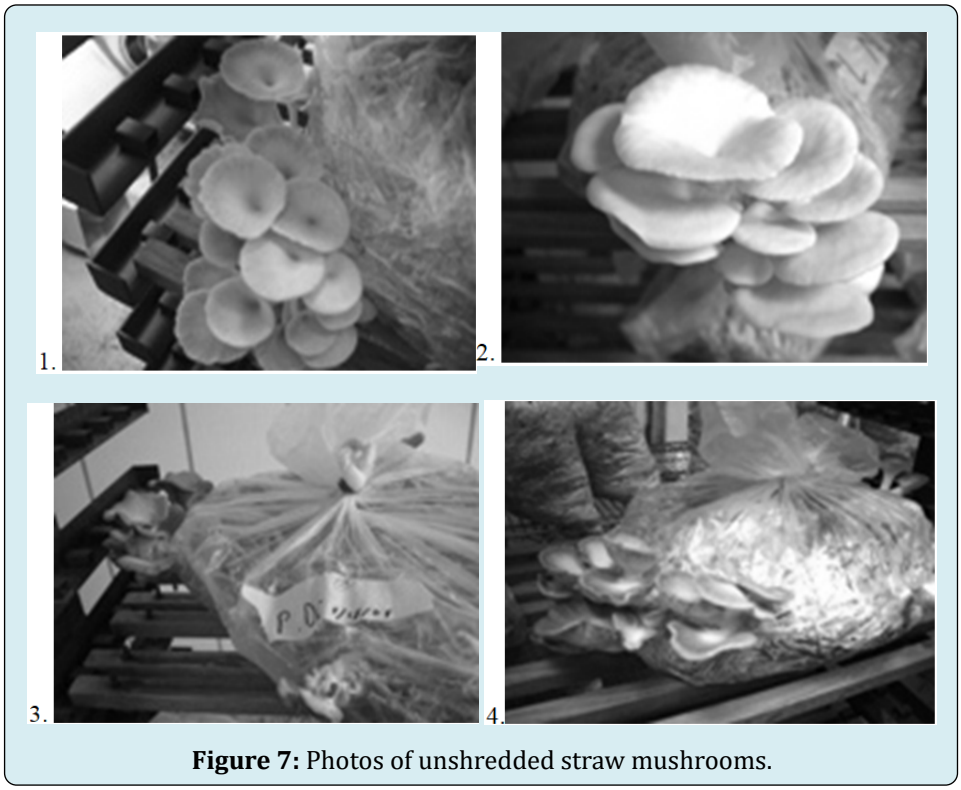

Figure 7: Photos of unshredded straw mushrooms.

Figure 7 shows representative pictures of fruit bodies growing on regular unshredded straw treated with the four representative treatment methods.

Conclusion

Until now each farm cultivation operation has almost always been looked upon as a "stand alone" operation, which is an operation complete in and of it-self, whether it is for plants, animals or fish. Raw materials come in, a crop is produced and the waste byproducts are disposed of. It does not need to, and should not be like this. In fact, all of nature is a continuous flow of building up followed by decay, with the residual waste from each step of the process forming the raw material basis for the life forms to follow. Following this natural process, a system of diversified agriculture has been developed and is presented where several successive crops can be raised one after another, each crop depending upon the waste materials from the previous step to act as the raw materials for the next step. In this fashion, several food crops can be successfully grown, harvested, and used so that hunger and malnutrition need not be a factor while the people of a country, city, town or village grow and develop.

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