



Maximizing Log Based Shiitake Mushroom Production by Determining Optimal Fruiting Conditions

2014 Final Report

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Summary

Nicholas Laskovski, owner/founder of Dana Forest Farm and Bridgett Jamison, Graduate Student at UVM collaborated to perform on-farm research at Dana Forest Farm in Waitsfield, VT between the years of 2011 and 2014. The research goal was to (1) determine optimal fruiting times and soaking conditions for the Northeastern production of log-based shiitake mushroom cultivation (2) test alternative methods for determine log moisture content and (3) determine the viability of using a non-destructive pH indicator to estimate mycelium colonization. Ultimately, our goal was to determine conditions which would increase shiitake yields whilst determining a organized approach to better manage shiitake production. By increasing yield, our research would allow current log based shiitake farmers to save time and money while increasing farm revenue. By understanding specific management techniques, decisions in when or how to force fruit bolts at different times of the day may allow farmers to better manage their laying yard amongst other farm ventures. New or upcoming shiitake farmers could justify adding or increasing log-based shiitake cultivation to their current farm or forests, ultimately diversifying operations, creating a more sustainable economic future for farmers of the Northeast.

In 2011, our research tested several methods for determining mycelial colonization and moisture levels within inoculated shiitake logs (aka bolts). In 2012, we continued our colonization research as well as created time trials for full water immersion (aka shocking) of bolts to 'force fruit' shiitake from inoculated logs. In 2013, additional research trials were attempted; however fruiting results were compromised by deer invasion. We requested a non-monetary extension to attempt to gather another trial or two of log shocking in the summer of 2014. Unfortunately, deer damage occurred for a second year in a row and fruiting data was completely removed by deer. Fencing and noise device attempts were made and the deer still managed to penetrate the barriers to gain access to fruiting bolts.

Despite setbacks, our research did yield interesting results. We determined that logs soaked for 24 hours produce more mushrooms than logs shocked for shorter and longer durations however there was no significant difference between the 6, 12, 24, and 48 hour treatments. Also we found that log moisture levels are directly related to rates of shiitake mycelial colonization and that smaller logs colonize faster than larger logs by volume. We tested the potential of using a log moisture meter and increment borer to estimate long moisture content and determined that neither method is precise or accurate. We found the pH indicators can be used to estimate mycelium colonization however more research is needed in order to determine if a pH indicator, applied to the surface of the log, could accurately predict when logs are ready for harvest and how large the harvest will be. Based on our initial trials and tests, we confirmed that *Trichoderma* does negatively affect shiitake colonization. Lastly, we developed and tested an innovative method of shocking logs using a tractor/ metal basket. This technique has the potential to greatly reduce the physical burden and time consuming nature shocking usually imposed on a farmer.

Although we were not able to complete all of the research goals we set out to due to an unforeseen pest problem, our research was an overall success. Our research regarding the use of the moisture meters and log shocking duration was used in the recently published, and widely distributed, Best Management Practices for Log-Based Shiitake Cultivation. Our preliminary research into the use of the pH indicator to estimate mycelium colonization was extremely innovative and may serve as a foundation for graduate level research into shiitake mushroom production. Lastly our research was frequently referenced in posts on the popular Mushrooms Listserv based out of the University Vermont.

Introduction

Nicholas Laskovski and Bridgett Jamison teamed up to conduct research on a farm level SARE grant after working together on a University level SARE research grant between 2010 and 2012. Allen Matthews was a lead on that project which led to his involvement as an advisor for their farm research. Nicholas is the founder/owner of Dana Forest Farm, located in central Vermont.

Dana Forest Farm provided the optimum research location to conduct shiitake research as it is a small commercial shiitake operation with needed infrastructure in place for shiitake production. Dana Forest Farm is an off grid farm/ homestead aimed at producing shiitake mushrooms using the most sustainable practices available, minimizing fossil fuel energy inputs, and maximizing food production for local markets.

Bridgett Jamison was a graduate student at UVM during the course of this research.

There has been a resurgence of interest in cultivating shiitakes on whole logs, because of certain small scale advantages. Log based mushroom production is well-suited to a broad range of land management objectives and can be a profitable method of diversifying income using low-value forestry by-products. Compared to sawdust substrate procedures, the log growth method is simpler and less expensive. It requires less energy input and is more environmentally friendly. Despite the advantages of log-based

production, growers are frustrated by the lack of standardization. We are specifically interested in investigating the final stage in shiitake production during which growers “shock” logs by submerging them in water in order to encourage fruiting. The overall research goal was to provide shiitake farmers with useful information and techniques that could be used to increase shiitake yields.

Objectives/Performance Targets

The objective of this study was to improve shiitake mushroom productivity and production efficiency and thereby increase potential profits of small scale log-based growers. More specially our goals were threefold:

(Objective 1)

We aimed to determine if the pH indicator Bromophenol Blue (BPB) can be successfully used to estimate mycelial colonization within the logs and predict future fruiting potential thereby providing farmers and researchers a simple, effective, inexpensive field-based method for evaluating mycelium maturity that could be used to determine if and when their logs are ready to fruit.

(Objective 2)

We aimed to determine the viability of using an increment borer and/or log moisture meter to determine log moisture content. Moisture content of a log is closely linked to net shiitake production; farmers and researchers need to routinely measure log moisture content when making management decisions. The traditional method of determining log moisture content involves removing a round slice from the log (cookie) and measuring the moisture of the slice by weighing, drying, and reweighing. However, this process is fairly destructive and time consuming. Using a moisture meter and/or increment borer would be less time consuming and destructive.

(Objective 3)

We aimed at investigating the final stage in shiitake production during which growers “shock” logs by submerging them in water in order to encourage fruiting. Variables that have been shown to dramatically impact production rates are: the degree of colonization by mycelium, and the ratio of water to air in the log following shocking, the length of time the log was submerged for, the history of outside air temperature vs. water temperature .

Methods

In the spring of 2011, we felled thirty sugar maple trees. Trees were cut into three-foot lengths generating 500 three-foot long logs with a diameter between 4 and 6 inches. The bolts were inoculated on either 4/17/11 or 5/1/11 with Wide Range stain (W46) following standard procedures as written in the Best Management Practices for Log-based Shiitake Cultivation publication authored in 2013. First a series of small holes were drilled into the surface of the log approximately four inches apart. Each hole was tightly packed with sawdust spawn and covered in cheese wax to ensure optimal mycelium growing conditions and reduce contamination by other fungi species. Logs were then placed in a laying yard using the crib stack. One hundred logs were randomly selected for the experiment. Each log was labeled using a durable metal tag. Initial log weight, diameter, and length were recorded. We accomplished our performance target in regard to bolt inoculation and selection with one exception. After reviewing other log based shiitake production experiments and the experimental objectives, we decided to include 100 logs, rather than 160 logs in the experiments. Treatments remained identical however, the number of replications per treatment

were reduced from 15 to 10. We decided this was appropriate given that the standard deviation between shiitake mushroom production per log is fairly low.

(Objective One)

After nearly one year of undisturbed colonization, we removed a “cookie” slice from each log. Cookies circumference was measured and cookies were weighed, dried for 72 hours at 105 degrees Celsius, and reweighed using a portable digital scale. This information was used to calculate log density and moisture content. A dilute 1% solution of BPH sprayed on each log section; each slice was then carefully photographed in a specially designed light box. The dye clearly discolored the areas of the log that had not been affected by shiitake mycelium blue (Figure 1). Using the imaging software, ImageJ, we measured the exact area of blue wood” and “yellow wood”. This information was used to determine the exact degree of mycelium colonization. We then plotted the degree of colonization (measure using digital image analysis) against and first year log yields and log density.

(Objective Two)

Our experiment was designed to track the rate of mycelium growth within log by monitoring the weight loss of the log over time. This requires having an estimate of the log’s moisture content during each sampling event. There are many ways of measuring moisture content in a log. The traditional method involves removing a round slice from the log (cookie) and measuring the moisture of the slice by weighing, drying, and reweighing. However, this process is fairly destructive and time consuming. There are many other ways of measuring moisture that are less time consuming and destructive. One way we had proposed in our experimental design involving using a unique type of hand drill called an increment corer. We planned on removing a 6-10 cm core from each log; we would then measure the percent moisture within the core by weighing, drying and reweighing the core. We would then use this value for percent moisture to estimate the percent moisture within the entire log. Another idea recommended to us was using a hammer corer to more rapidly remove a smaller core from the uppermost 3 cm. Lastly, we had the option of using a unique electronic moisture meter designed for measuring moisture of timber to a depth of 5 cm. In the spring of 2011, we tested the three techniques mentioned above against each other.

(Objective Three)

In 2012, 2013, and 2014 logs were shocked with the aim of estimating the relationship between shocking duration and yield. Logs were randomly divided into four groups to be subjected to four durations of soaking: 6 hours, 12 hours, and 24 hours and 48 hours. Mushrooms were collected from the logs seven days after the soaking event in order to calculate total mass of mushrooms obtained per log. A few representative mushrooms were selected to determine the mass moisture content of the mushrooms. Yield results were compared against other variables including initial moisture content, log age, and degree of mycelium colonization to determine how these factors influence production.

In 2013, we repeated our time trial shocking experiment. The results from fruiting from the shocking trial were compromised due to deer harvesting almost 100% of fruiting mushrooms before they were ready for harvest (Images and 1 and 2).

In 2014, we repeated our time trial shocking experiment again. The results from fruiting from the shocking trial were compromised due to deer harvesting almost 100% of fruiting mushrooms before they were ready for harvest. Measures were taken this year in anticipation of deer, however the fence and noise makers were ineffective. We decided against an earlier trial in the beginning of the summer (June) due to our commercial logs being used as a relative test deciding whether we should shock the research logs. All of our shiitake were late to fruit this year and we didn't want to shock mushroom logs too early in the season and not be able to have any harvestable results.

Outcomes and Impacts

(Objective One)

The pH indicator BPB clearly delineated area of the log that had and had not been colonized with shiitake mushroom mycelium. Digital imaging technology was able to, with great accuracy; estimate the exact degree of mycelium colonization. However, there was no relationship between the amount of shiitake mycelium colonization estimated using digital image analysis of the BPB treated sections and the shiitake production. Therefore, we can conclude that a BPB treatment cannot be used to predict when a log is ready to fruit (Figure 1). We hypothesize the pH indicator BPB indiscriminately dyed areas of the log that had been colonized by mycelium regardless of whether the mycelium were beneficial shiitake mycelium or harmful competitor mushroom mycelium. Therefore, it overestimated the degree of shiitake mycelium colonization.

Interestingly, the amount of mycelium colonization estimated using digital image analysis of the BPB treated sections could be used to successfully predict the log density. Therefore, we know the BPB treatment can accurately measure the rate of mycelium colonization. (Figure 2)

Although BPB cannot be used to estimate potential shiitake yields, it may have valuable research applications. It proved to be a fast, environmentally friendly, non-destructive, and very precise measure of estimating mycelium colonization.

(Objective Two)

In the spring of 2011, we tested the three techniques mentioned above against each other. The results of our trials are provided in Figure 3. Our trials showed that log moisture was too variable to be assessed used either an electronic probe or through small subsamples (cores). As a confirmation, we conducted another trial comparing the measure of percent moisture using a hammer corer against the standard cookie method. See Figure 4. Although, there is a definitive relationship, we realized that the variability was too large to make the hammer corer a viable alternative to the cookie method. After seeing results of this trial, we agreed to use the traditional cookie method to measure log moisture. Our next question was (1) how large a cookie would be needed and (2) from how deep within the log will we need to cut to get accurate consistent results. To answer these questions, we conducted another trial. This time we cut two cookies from one side of log (each approximately 4 cm thick) and measured the moisture in each of the cookie. The results indicated that there was no difference in the moisture content between a cookie taken on the inside and outside of a log. Therefore, we decided to collect cookies from the outside each bolt during each sampling event. This reduces the overall destructiveness of our sampling.

Although our attempts to develop a more efficient and less destructive method of measuring log moisture content were unsuccessfully, our experiment provided useful results. We were able to definitively report that expensive moisture meter probes cannot be used to make management decisions in a shiitake production operation. We were also shown that log moisture levels were far too variable to measured using a small sample. These questions had arisen numerous times over the mushrooms listserv and at mushroom growing conferences from shiitake farmers.

(Objective Three)

We determined that the duration which a log is soaked/shocked impacts the shiitake production. Logs dunked repeatedly but never soaked produced significantly less shiitake mushrooms than logs soaked for 24 hours. The greatest yields of mushrooms and number of mushrooms were on obtained on logs soaked for 24 hours. However, there was no significant difference between logs soaked for 6, 12, 24, and 48 hours in shiitake mushroom production. (See Figure 5)

By tracking the change in log density over time, we were able to estimate the rate of mycelium colonization over time. Our results showed that the rate the bolt density change was not correlated to production. We also plotted the total percent change in density over two years against the shiitake production. There was no relationship (See Figure 6) These results indicated that shiitake production is not related to the rate of mycelium colonization. During the beginning of the study, we measured the diameter of each log. We used this information to calculate the surface area of the end of the log. We plotted this information against the shiitake production in order to determine if there is a correlation between log size and production. Our results showed there was no relationship between log diameter and total shiitake mushroom production (See Figure 7)

We had hoped to repeat these trials for three more years in order determine if optimal shocking duration changes as the log ages and if the relationship between log density, log diameter, and shiitake production change over time. However, we were unable to harvest mushrooms in 2013 and 2014 due to a deer infestation.

We begrudgingly admit that the pervasiveness of deer attacking our shiitake was a surprise. In all of our collective years growing shiitake outdoors on logs, and through conversations with other growers throughout the Northeast during our research, this was a difficult surprise to swallow. We did our best to protect our logs during this past year but unfortunately our efforts fell short.

Other Interesting Findings:

Based on our initial trails and tests, we confirmed that Trichoderma does negatively affect shiitake colonization (Figure 8).

Accomplishments

Although we were not able to accomplish all we had set out to accomplish, our experiment was, nonetheless, a success. We address the question of how to measure log moisture by thoroughly testing a variety of creative methods. We verified that a pH indicator will differentially stain areas of the log that were and were not colonized by shiitake mycelium and that imaging software can easily quantify these areas. In addition, we provided some basic information regarding the impact of log size, shocking duration, and density of ultimate shiitake yields.

A design factor in our experiments allowed us to successfully trial shocking treatments using a tractor/ metal basket which enabled us to group shiitake logs in a way which allowed us to keep our logs well organized. See figure 10.

Potential Contributions

The practice of log-based shitake cultivation in the Northeast is still in its early stages relative to many other aspects of farming. First recorded shiitake cultivators in North America date only back to 1983. There are many more questions regarding management decisions. Our study was able to provide valuable insight into a few of these problems. First our study alluded to the likelihood that 24 hours of soaking is the optimal soaking length for common northeastern species of logs. However, because the data was not significant, additional trials need to be conducted again before the result can be verified. We also verified that moisture meters, increment borers, and hammer corer are not capable of accurately measuring log moisture content. We briefly investigated the interesting possibility that the pH indicator could be useful in monitoring mycelium production. Lastly, we confirmed that trichoderma, a competitive mold, will appreciable negatively affect yields. Up until our experiment, the general consensus was that trichoderma would not have a significant impact on yields.

Publications/Outreach

We were not able to create a formal publication outlining our research; the consumption of mushrooms by deer significantly limited our data set. However, our research was shared in a number of other venues with the public.

Our research was prominently featured in the recently published “Best Management Practices for Shiitake Mushroom production in the Northeast” (See Attachment). We wrote segments regarding deer and shiitake pests, the use of moisture meters, and appropriate soaking times for logs.

We also used our research to answer numerous questions that arose on the Mushrooms Listserv based out the University of Vermont. This popular listserv has over 100 members. Most of whom are shiitake growers in the Northeast.

Future Recommendations

We believe that additional research into the use of BPB should be conducted. This simple, cheap, and non-toxic pH stain clearly delineated the areas of the log that were colonized and the areas uncolonized.

This could be used in the future to assess the damage by competitive fungi, or may prove effective at predicting appropriate fruiting times if a cookie from deeper into the interior of the log used.

We also believe that additional research needs to be conducted to determine if the optimal soaking duration changes give the age and size of the logs. More replicates would be needed to decrease the experimental variation and increase the experimental power. Many growers and scientists have predicted that smaller and younger logs should be soaked for shorter period compared with older, larger logs. This hypothesis could not be effectively tested in our experiment due to deer infestation and has not, to our knowledge, been tested by other researchers.

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Attachments:

Figure 10. Tractor / Basket Shocking: http://mysare.sare.org/assocfiles/991983IMG_5601.JPG

Figure 1 - Example of a log dyed blue and yellow from the pH indicator BMB:
<http://mysare.sare.org/assocfiles/9919682013 bmp colonization.JPG>

Image 1 - Photo of recently shocked logs from which deer had eaten all of the shiitake mushrooms. :

[http://mysare.sare.org/assocfiles/991968Shiitake damage by deer.JPG](http://mysare.sare.org/assocfiles/991968Shiitake%20damage%20by%20deer.JPG)

Image 2 - Photograph of logs from which deer had eaten all the mushrooms. :

[http://mysare.sare.org/assocfiles/991968fruiting research post deer.jpg](http://mysare.sare.org/assocfiles/991968fruiting%20research%20post%20deer.jpg)

Figure 1 - Regression analysis between percent mycelium colonization estimated using BMB pH indicator and shiitake yeild. The relationship is not significant and therefore BMB stain cannot be used to estimate potencial yields. : [http://mysare.sare.org/assocfiles/991976BPB based estimate of mycium colonization versus shiitake production.jpg](http://mysare.sare.org/assocfiles/991976BPB%20based%20estimate%20of%20mycium%20colonization%20versus%20shiitake%20production.jpg)

Figure 3 - Measure of the moisture content of individual logs taken using three different methods: hammer corer, increment corer, and moisture meter. : [http://mysare.sare.org/assocfiles/991976Moisture readings from three methods.jpg](http://mysare.sare.org/assocfiles/991976Moisture%20readings%20from%20three%20methods.jpg)

Figure 4 - Relationship between measures of moisture taken using the cookie method and the hammer borer. Although the relationship is significant, measures using the hammer borer are not accurate enough to used to measure log moisture.: [http://mysare.sare.org/assocfiles/991976Regression of moisture readings.jpg](http://mysare.sare.org/assocfiles/991976Regression%20of%20moisture%20readings.jpg)

Figure 5 - Bar graph indicating the mean mass of the mushrooms from logs soaked for different durations, : [http://mysare.sare.org/assocfiles/991976Shocking duration and production.jpg](http://mysare.sare.org/assocfiles/991976Shocking%20duration%20and%20production.jpg)

Figure 6 - Correlation between percent change in log density and shiitake mushroom production.: [http://mysare.sare.org/assocfiles/991976Change in density versus shiitake production.jpg](http://mysare.sare.org/assocfiles/991976Change%20in%20density%20versus%20shiitake%20production.jpg)

Figure 7 - Relationship between log diameter and shiitake mushroom production. Contry to popular assumption larger logs did not produce more mushrooms.:

[http://mysare.sare.org/assocfiles/991976991976 Diameter and Production.jpg](http://mysare.sare.org/assocfiles/991976991976%20Diameter%20and%20Production.jpg)

Figure 8 - Bar graph displaying the impact of trichoderma infestation on shiitake yields. :

[http://mysare.sare.org/assocfiles/991976Impact of Tricoderma.gif](http://mysare.sare.org/assocfiles/991976Impact%20of%20Tricoderma.gif)

Best Management Practices for Shiitake Cultivation:

http://mysare.sare.org/assocfiles/992128Shiitake_BMP_Guide.pdf

Figure 2 - Regression between change in log density and estimate of mycelium colonization based from BMB stain. There is a slightly signifcant relationship and therefore the stain is capable of correctly estimating mycelium colonization/ : [http://mysare.sare.org/assocfiles/991976Change in density versus percent of funal colonization.jpg](http://mysare.sare.org/assocfiles/991976Change%20in%20density%20versus%20percent%20of%20funal%20colonization.jpg)

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