

# EVALUATING MANAGEMENT PRACTICES FOR LOG-GROWN SHIITAKE PRODUCTION IN MIDWESTERN AGROFORESTRY

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**ABSTRACT.**—Two experiments evaluating outdoor shiitake cultivation practices in the Central United States were established at the same site in December 1999 and May 2000. Natural fruiting began in August 2000. We compare two response variables: traditional Biological Efficiency and W/UV (the weight of harvested mushrooms in g per 100-cm<sup>3</sup> of initial undisclored wood volume). Use of W/UV provided clearer statistical distinction among experimental factors. Significant differences were detected in year 2000 data among shiitake strains, substrate log species, and spawn forms. We expect shifts in pattern of treatment differences as the experiments proceed.

Values derived from woodland ownership can be enhanced by capitalizing on natural associations of specialty fungi with trees. Mushroom production can be integrated with most management objectives and provides profitable avenues for recycling low-value forestry by-products. A research and demonstration project has been initiated at the University of Missouri Center for Agroforestry (UMCA) to develop and/or identify best management practices for woodland cultivation of specialty mushroom fungi, including shiitake (*Lentinula edodes* (Berk.) Pegler). Although about two-thirds of Central States woodland mushroom growers have other primary sources of income, nearly all consider mushroom cultivation a part of their career, and nearly all grow shiitake (Bruhn and others 2000).

Biological challenges to shiitake cultivation involve selection and development of "best management practices" for improving efficiency of outdoor mushroom production under various sets of local conditions. While a great deal of descriptive literature exists (e.g., Ito 1978, Stamets and Chilton 1983, Chang and Miles 1989, Przybylowicz and Donoghue 1990, Kozak and Krawczyk 1993, Stamets 1993, Ikegaya 1997), published experimental evaluations of outdoor management factors relevant to the Central States region are scarce (e.g., Abe 1989,

Bratkovich 1991, Tokimoto and others 1998). We present here an experimental approach capable of distinguishing the effects of field treatments, and first results of two experiments. We also discuss the shortcomings of so-called Biological Efficiency as a response variable for scientific evaluation of log-grown shiitake production, and we suggest use of an alternative response variable, W/UV (the weight of harvested mushrooms in g per 100-cm<sup>3</sup> of initial undisclored wood volume).

## RESEARCH GOAL AND OBJECTIVES

Our long-term research goals are to identify the best management practices for woodland cultivation of shiitake in temperate agroforestry, and to determine the associated productivity.

Specific research objectives of the two experiments presented here are to:

- 1) compare the productivity of several strains of shiitake;
- 2) compare the productivity of several readily available host tree species;
- 3) compare the productivity of the most commonly available forms of spawn (inoculum);
- 4) evaluate the productivity associated with early winter and spring inoculation;
- 5) evaluate the relative productivity of immediate vs. delayed inoculation;

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*Citation for proceedings:* Van Sambeek, J.W.; Dawson, J.O.; Ponder, F., Jr.; Loewenstein, E.F.; Fralish, J.S., eds. 2003. Proceedings, 13<sup>th</sup> Central Hardwood Forest conference; 2002 April 1-3; Urbana, IL. Gen. Tech. Rep. NC-234. St. Paul, MN: U.S. Department of Agriculture, Forest Service, North Central Research Station. 565 p. [Peer-reviewed paper from oral presentation].

- 6) compare the results obtained from comparable General Linear Model analyses using two different response variables: traditional "biological efficiency" (BE, the fresh weight of harvested mushrooms expressed as a percentage of initial log dry weight) and W/UV (the fresh weight of harvested mushrooms in g per 100-cm<sup>3</sup> of undiscolored wood volume).

## MATERIALS AND METHODS

### Study Site and Experimental Design

Two separate experiments were established in early December 1999 and May 2000 at the University of Missouri-Columbia's Horticulture and Agroforestry Research Center (HARC), New Franklin, MO. The experiments are located on a gentle north-facing slope (16 to 24 percent, averaging 21 percent) under hardwood shade.

The experimental units are logs 1-m long and 8- to 23-cm diameter, cut from the stems of healthy, living, pole-size trees. Both experiments included logs cut from sugar maple (*Acer saccharum* Marsh.), white oak (*Quercus alba* L.), and northern red oak (*Q. rubra* L.). All logs were cut from relatively straight stem sections free of heart rot, and with intact bark. Aluminum tags identifying the source tree and stem position were attached to each log at the time of harvest. For example, substrate logs 1-1 and 1-2 are the first and second logs (numbered from the stump upward) cut from tree number 1. These tags are used to relate the mushrooms harvested from each log to that log's descriptive data and the treatments applied to it. For the early winter-initiated experiment (Experiment 1), all logs were cut from trees harvested dormant (10-12 November, after passage of autumn color). Inoculation was conducted 8-10 December. In the spring-initiated experiment (Experiment 2), logs cut from trees harvested dormant in mid-February (approximately 13-weeks prior to inoculation) are compared with logs cut from trees harvested 2-5 May. All logs were inoculated in May.

The surface area (SA) of each substrate log was calculated from its length and diameters outside bark at both ends. Undiscolored wood volume was calculated for each log from its length and the diameters inside bark and inside undiscolored wood at both ends. The numbers and diameters of live branch stubs on each log were also recorded. The initial dry weight of each log was estimated from its initial fresh weight and the fresh-to-dry-weight ratio of a 1-cm thick slice collected from the end of each log at the

time of harvest; for example, a single slice taken between logs 1-1 and 1-2 at the time of felling and bucking was used to represent the initial fresh-to-dry-weight ratio of both logs.

The same three fungal strains are evaluated in both experiments. All inoculum (spawn) was purchased from Field and Forest Products, Peshtigo, WI. Strains included in both experiments represent one each selected for its fruiting potential during cold weather (CW-50), warm weather (WW-44), or over a wide temperature range (WR-46).

Two spawn forms of each shiitake strain are being evaluated in each experiment. Standard sawdust and wooden dowel spawn (Przybyłowicz and Donoghue 1990, Kozak and Krawczyk 1993), both sealed with hot cheese wax, are being compared in Experiment 1. Standard sawdust (sealed with hot cheese wax) and waxless "thimble" spawn (molded sawdust-based plugs with a thin styrofoam cap) are being compared in Experiment 2.

Logs were inoculated as follows. For all spawn forms, holes were drilled approximately 15-cm apart in rows 5-cm apart around the log circumference with a Makita high speed drill (8,000 rpm). A diamond pattern was created by offsetting the holes in alternate rows by approximately 7.5-cm. Holes for sawdust and thimble spawns vs. dowel spawn placement were drilled 2.5-cm deep and 12-mm in diameter vs. 1.9-cm deep and 8.5-mm in diameter, respectively, using screw-tip bits. The screw tip guides the drill bit safely, and the high speed of the drill strips the wood around the screw tip when the bit reaches the pre-set stop depth, permitting very fast hole creation and easy drill extraction (Przybyłowicz and Donoghue 1990, Kozak and Krawczyk 1993). Very hot (smoking) molten cheese wax was used to seal traditional sawdust and dowel inocula. If the wax is not hot enough to sizzle on contact with the inoculum, it may not provide a lasting seal over the spawn (Przybyłowicz and Donoghue 1990, Kozak and Krawczyk 1993). The styrofoam cap on thimble spawn is designed to provide an effective seal without application of cheese wax.

Each experiment is arranged as a randomized complete block design. Experiments 1 and 2 comprise 9 and 8 blocks, respectively, with one log of each species assigned randomly to a treatment combination and a location within each block. Logs were laid approximately 10-cm above the ground parallel to the forest floor,

oriented perpendicular to the slope contour with centers 30-cm apart, on paired wooden rails placed parallel to the slope contour. Rails were laid over black perforated plastic Weed Block® landscape fabric (Easy Gardener, Inc., Waco, TX). This landscape fabric has functioned well to reduce weed growth among the substrate logs. Leaf and woody litter were removed from the fabric when it contacted the substrate logs. The forest floor around the experimental blocks was mowed as necessary to a height of approximately 5-cm with a heavy-duty brush mower, to further prevent forest floor vegetation from overgrowing the substrate logs.

Air temperature at the study site is being measured 66-cm above ground level, using a Campbell Scientific, Inc. (Logan, UT) micrologger with sensors. Degree days are calculated on a 4° C basis from the average of daily minimum and maximum temperatures. Daily total precipitation is monitored in the open at the HARC weather station, approximately 400-m from the experimental site.

### Harvesting and Productivity

The weight and grade of each mushroom harvested is being credited to its source log. Harvest frequency (as often as daily) is intended to maximize grade yield. Individual mushrooms were harvested by twisting and or bending the base of the stem to separate the mushroom stem from the log at or just below the log surface. It is hypothesized that mushrooms harvested in this manner remain fresher longer, and that this method leaves less debris on the log to attract pests (Joe Krawczyk, Field & Forest Products, Inc., personal communication).

Individual mushrooms were assigned to grades 1, 2, or cull (Kozak and Krawczyk 1990). To summarize, Grade 1 mushrooms have approximately round, complete caps 2.5- to 10-cm in diameter, 50–80 percent expanded, less than 10 percent pitting, but free of rodent damage. Gills are white, unbruised, and clean. Grade 1 mushrooms are neither excessively dry nor water-soaked. A Grade 2 mushroom is any usable mushroom which does not meet specifications for Grade 1, and a Cull mushroom is any unusable mushroom. The distinction between Grade 2 and Cull mushrooms varies, depending on the end uses available to a given grower. The analyses presented here are based on total fresh weight of harvested mushrooms, regardless of grade.

### Models

All statistical analyses were conducted using SAS/STAT System Release 8.0 (SAS Inst., Inc., Cary, NC, USA). SAS PROC CORR was used to evaluate the extent of correlation among harvest-related and log characteristic variables. SAS PROC GLM was used to analyze the effects of treatments and log characteristics on productivity in each experiment during the year 2000. Two response variables were evaluated: the traditional “biological efficiency” (BE, the fresh weight of mushrooms produced from a substrate log expressed as a percentage of initial log dry weight) and W/UV (the fresh weight of mushrooms produced per 100-cm<sup>3</sup> of undiscolored wood volume). Both of these response variables were calculated from total harvested mushroom weight. Crop grade distribution will be analyzed after collection of another year’s data. A third model used W/UV as the response variable, but included substrate log SA as a covariate. Tukey’s HSD test ( $\alpha = 0.05$ ) was used to evaluate treatments where ANOVA detected significant differences (Dowdy and Wearden 1983).

### RESULTS

Fruiting commenced naturally in Experiments 1 and 2 in early and late August 2000, respectively, and continued into November 2000 until approximately day of year (DOY) 300 (fig. 1). In Experiment 1, strain WR46 began fruiting in response to August rains (after DOY 220), whereas strains CW50 and WW44 fruited little until the rains of October 2000 (after DOY 280, figs. 1 and 2). In Experiment 2, WR46 fruited only weakly in late September 2000, and all three strains responded to October rains with increased fruiting (figs. 1 and 2). Thus, with early winter inoculation, WR46 appeared to complete spawn run earlier than Strains CW50 and WW44, whereas this difference was much less pronounced following late spring inoculation.

During 2001, fruiting commenced naturally in both experiments in April (after DOY 90) and continued into late December (data are shown for only March through July in fig. 3). However, in both experiments, CW50 fruited substantially in April 2001, whereas WR46 and WW44 began fruiting substantially in May 2001 (fig. 3). Fruiting in early 2001 appeared to be more responsive to accumulated degree days than to precipitation events (figs. 3 and 4).

At the end of the first growing season, W, BE, and W/UV for each log were all strongly and positively correlated with one another (table 1).

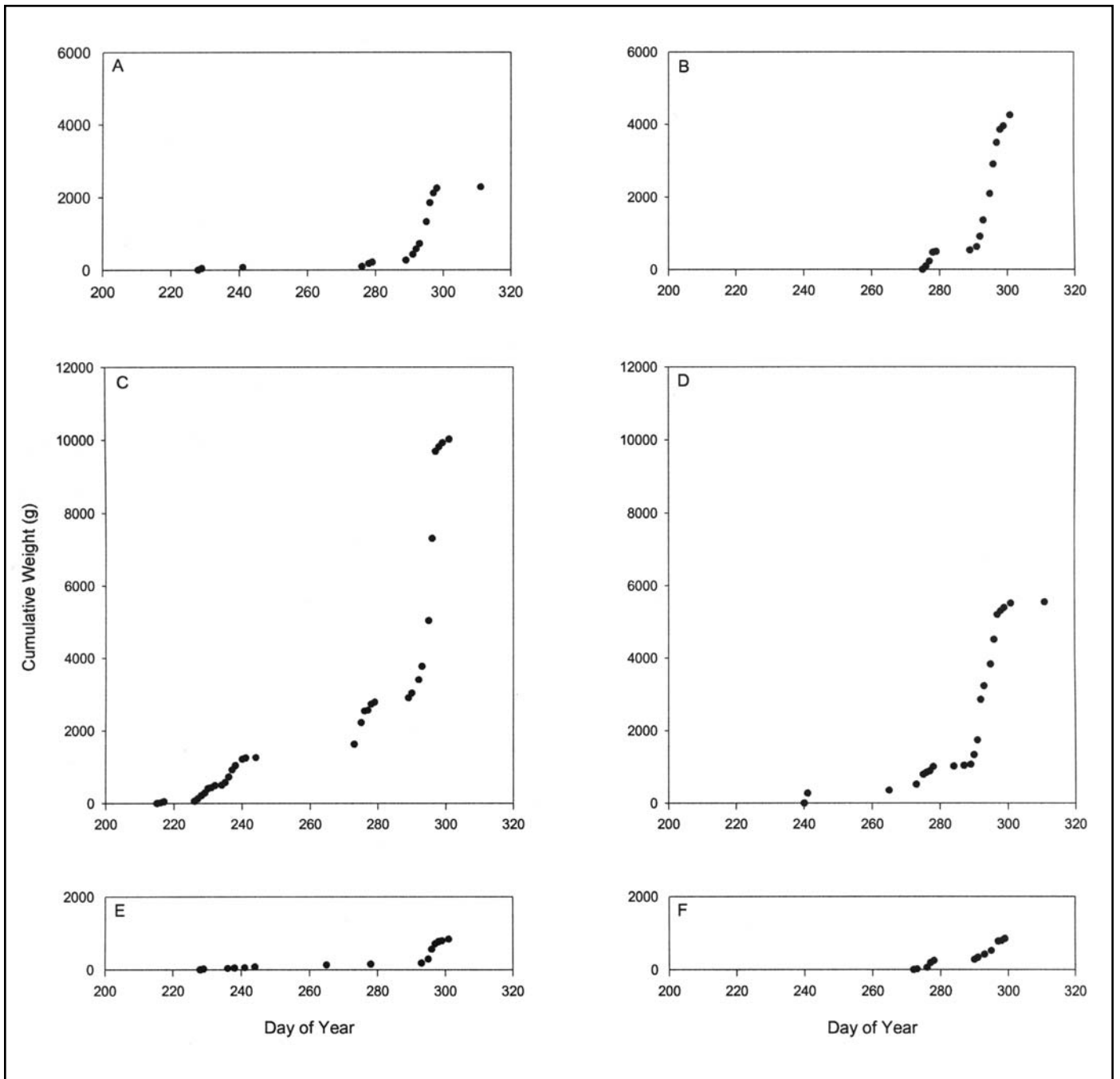


Figure 1.—*Shiitake* production during year 2000 by Cold Weather, Wide Range, and Warm Weather *shiitake* strains inoculated during early December 1999 (panels A, C, and E, respectively) and by the same strains inoculated during May 2000 (panels B, D, and F, respectively).

In both experiments, W was not correlated with substrate log SA, whereas BE and W/UV were both negatively correlated with SA. W was weakly correlated (Experiment 1) or not correlated (Experiment 2) with UV, whereas BE and W/UV were not correlated and negatively correlated, respectively, with UV in both experiments. Neither W nor W/UV was correlated with the number or total diameter of live branches

severed from substrate logs, and BE was only weakly correlated with total severed branch diameter. Substrate log SA and UV are strongly and positively correlated with each other in both experiments, and both are negatively correlated with substrate log branchiness.

ANOVA model results for Experiments 1 and 2 using BE as the response variable are presented

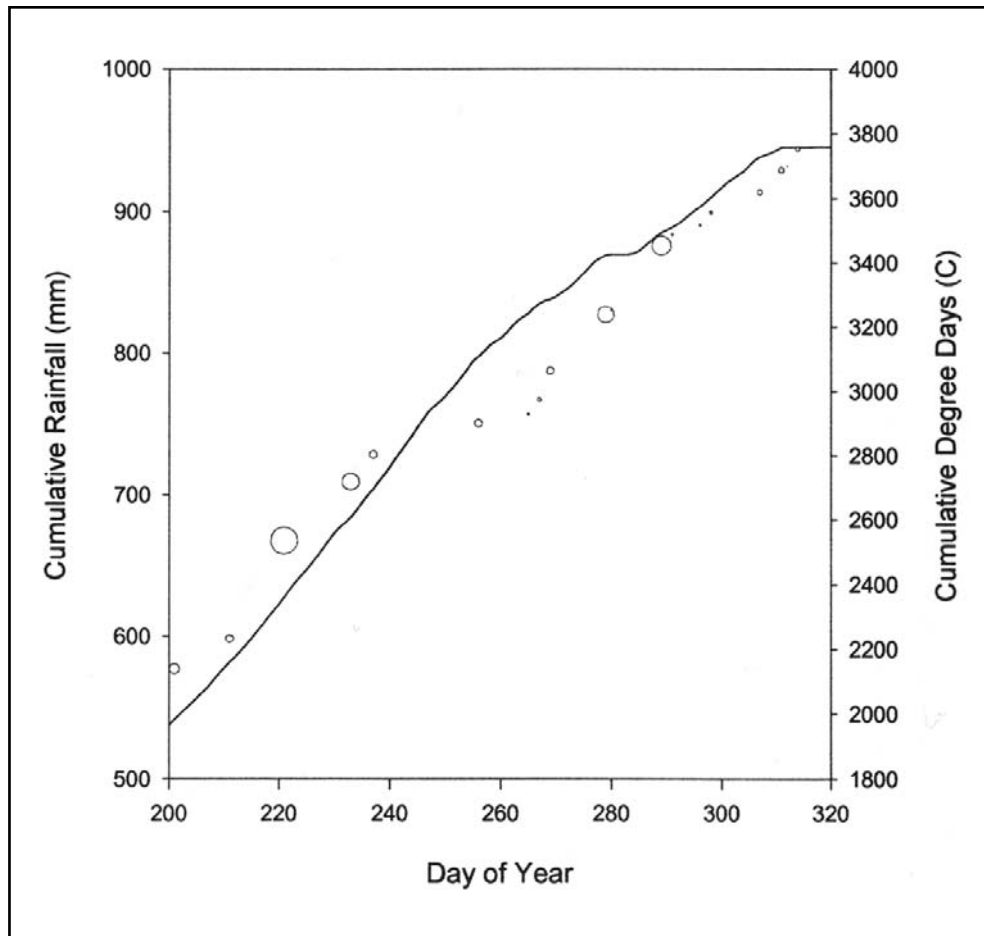


Figure 2.—Cumulative precipitation (each circle represents a precipitation event, for which circle diameter is proportionate to precipitation amount) and cumulative degree days (solid line, 4° C basis) during year 2000.

in Appendix table 1. Corresponding model results using W/UV as the response variable are presented in Appendix table 2. The corresponding models presented in Appendix table 3 with W/UV as the response variable include substrate log SA as a covariate. In each experiment, all three models detected the same highly significant differences in productivity among shiitake strains (table 2). In both experiments, WR46 outproduced both CW50 and WW44 (fig. 1). In Experiment 2, CW50 also outproduced WW44. The additional resolution among the three strains achieved in Experiment 2 is probably due to the greater variation in Experiment 1 productivity resulting from the significantly different performance of the two spawn forms employed in Experiment 1 (see below and table 2). A similar pattern appears to have developed through July 2001 (fig. 3).

Differences in productivity among substrate log species were not as pronounced in Experiment

1 as in Experiment 2 (table 2), again probably due to the greater variation in Experiment 1 productivity resulting from the significantly different performance of the two spawn forms employed. Because BE vs. W/UV express productivity as functions of total substrate log weight vs. undisclored wood volume, it isn't surprising that sugar maple BE exceeded that of white oak (though not significantly greater than red oak) for year 2000 in Experiment 1. Nor is it surprising that red oak W/UV exceeded that of sugar maple (though not significantly greater than white oak) for year 2000 in Experiment 1. However, in Experiment 2, both BE and W/UV for white oak exceeded values for sugar maple during year 2000.

During year 2000, sawdust spawn outperformed dowel spawn in Experiment 1 (table 2). Waxless "thimble" spawn performed as well as traditional sawdust spawn during year 2000 in Experiment 2. The distinction between the

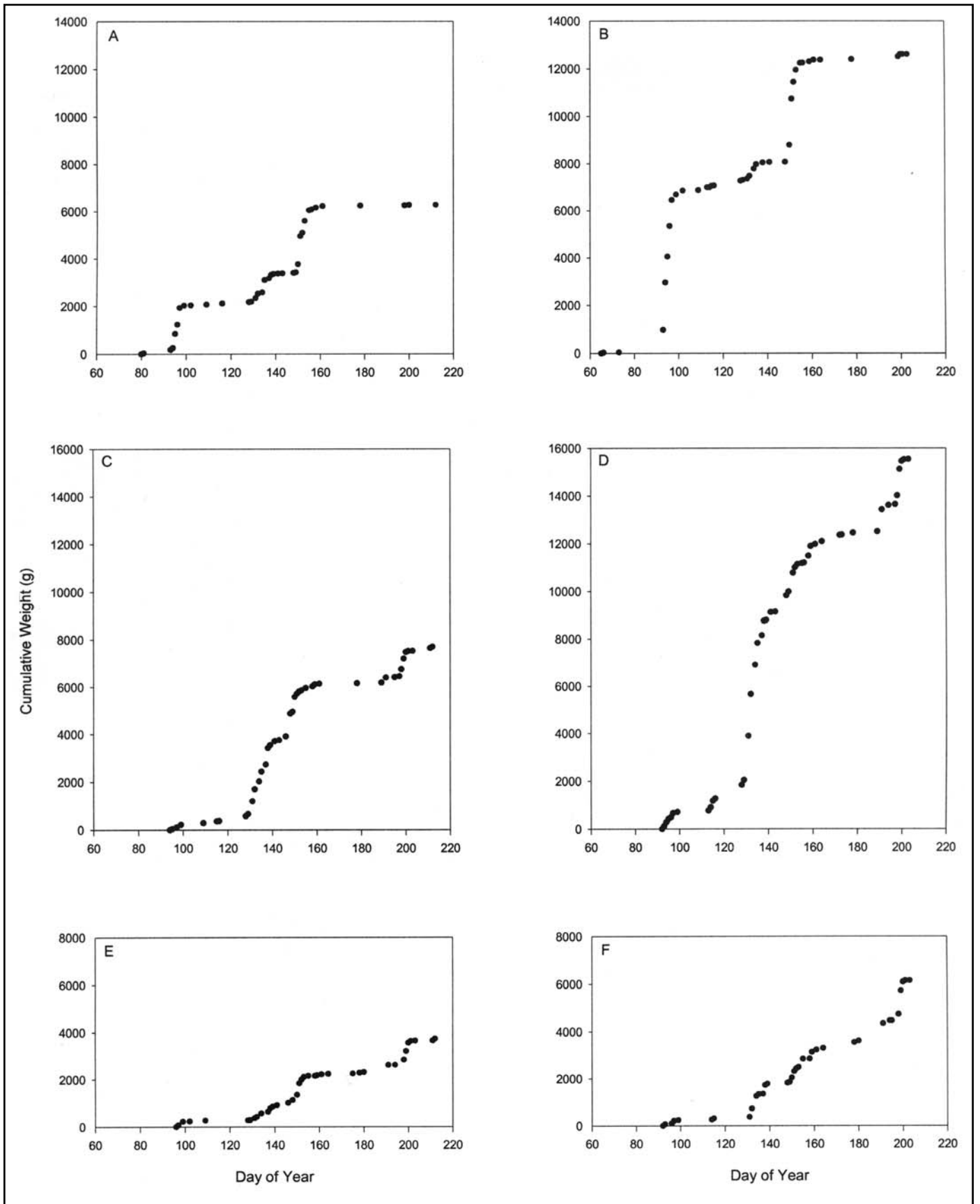


Figure 3.—*Shiitake* production during year 2001 (through July) by Cold Weather, Wide Range, and Warm Weather *shiitake* strains inoculated during early December 1999 (panels A, C, and E, respectively) and by the same strains inoculated during May 2000 (panels B, D, and F, respectively).

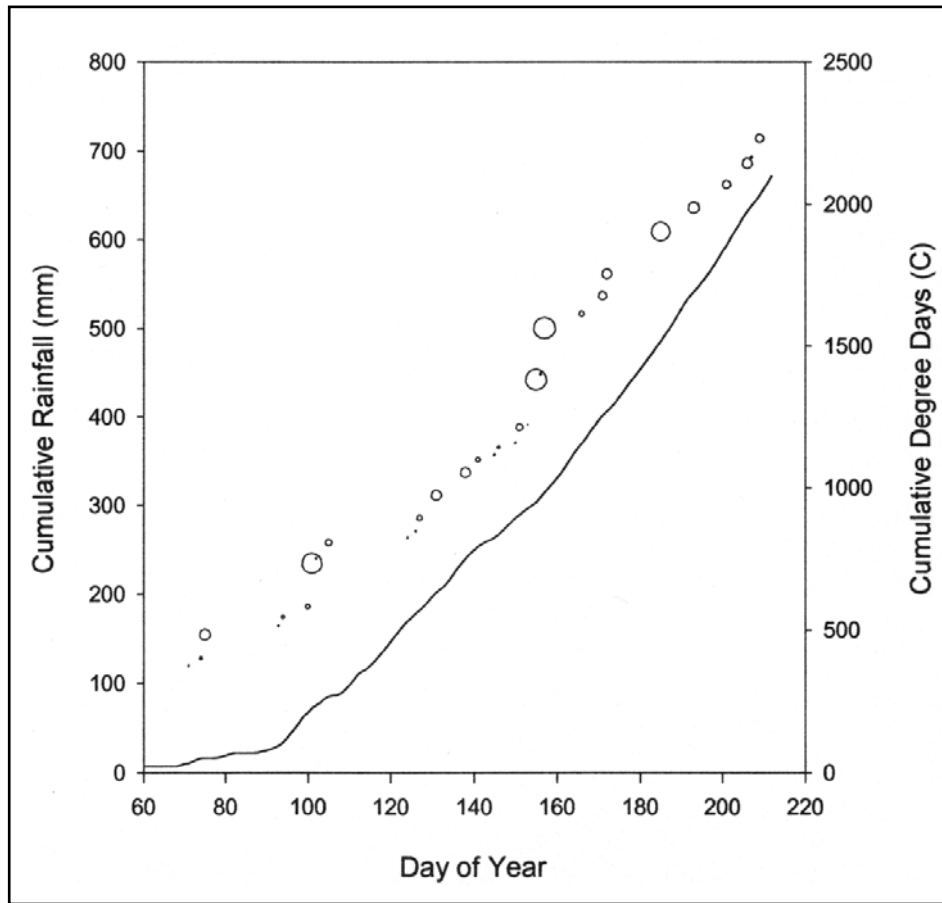


Figure 4.—Cumulative precipitation (each circle represents a precipitation event, for which circle diameter is proportionate to precipitation amount) and cumulative degree days (solid line, 4° C basis) during year 2001 through July.

performance of sawdust and dowel spawn in Experiment 1 was stronger when productivity was measured as W/UV than as BE.

Delayed inoculation (13 weeks following harvest) had no effect on productivity during year 2000 measured either as BE or as W/UV (table 2). A significant block effect was detected for year 2000 fruiting in Experiment 2 only, and only for W/UV. Of the eight blocks, block 3 was more productive than blocks 4 and 7.

A stronger interaction between shiitake strain and substrate log species was detected in both experiments when productivity was measured as W/UV rather than as BE (table 2). A significant interaction between shiitake strain and spawn form was detected only in Experiment 1 and only for W/UV. A substrate log species interaction with inoculation delay was detected in Experiment 2 only for BE.

## DISCUSSION

Our studies of outdoor log-grown shiitake production are designed to contribute to development of best management practices for growers in the Central States. The experiments presented here have an expected duration of 4 years, through year 2003. In the course of evaluating BE and W/UV, and comparing their relationships with experimental factors throughout the life of these two experiments, we expect to see some shifts in the relative productivity associated with the experimental factors (i.e., shiitake strain, substrate species, spawn form, and inoculation delay), including development of significant statistical interactions among factors. While it is premature to draw conclusions based solely on the first season's data, the novel analytical approach described here will provide improved insight into the biology and management of log-grown shiitake mushroom production.

Table 1.—Correlation coefficients  $R^2$  and significance levels for two measures of mushroom productivity with selected substrate log characteristics for 2000 (first field season) in each of two experiments<sup>1</sup>

	W <sup>3</sup>	SA <sup>4</sup>	UV <sup>5</sup>	W/UV <sup>6</sup>	N of stubs <sup>7</sup>	Stub diam. <sup>8</sup>
<b>Experiment 1 (inoculated early December 1999)</b>						
BE <sup>2</sup>	0.848 ***	-0.296 ***	-0.020 ns	0.825 ***	0.068 ns	0.087 ns
W		-0.059 ns	0.193 *	0.780 ***	-0.033 ns	-0.025 ns
SA			0.503 ***	-0.239 **	-0.428 ***	-0.391 ***
UV				-0.172 *	-0.348 ***	-0.303 ***
W/UV					0.222 ns	0.110 ns
<b>Experiment 2 (inoculated May 2000)</b>						
BE	0.864 ***	-0.204 ***	-0.096 ns	0.868 ***	0.109 ns	0.121 *
W		0.073 ns	0.081 ns	0.795 ***	0.068 ns	0.096 ns
SA			0.598 ***	-0.148 *	-0.288 ***	-0.240 ***
UV				-0.231 ***	-0.183 **	-0.129 *
W/UV					0.109 ns	0.102 ns

<sup>1</sup>Tabular symbols indicate significance at  $P \leq 0.05$  (\*),  $P \leq 0.01$  (\*\*),  $P \leq 0.001$  (\*\*\*), not significant (ns).  
<sup>2</sup>BE, Biological Efficiency, the fresh weight of all mushrooms harvested from a substrate log expressed as a percentage of initial log dry weight.  
<sup>3</sup>W, Weight, the total fresh weight (g) of shiitake mushrooms harvested from each substrate log.  
<sup>4</sup>SA, Surface Area, the surface area (cm<sup>2</sup>, outside bark) of each substrate log.  
<sup>5</sup>UV, Undiscolored wood volume, the volume of undiscolored wood (cm<sup>3</sup>) in each substrate log, estimated from diameter inside bark and undiscolored wood thickness at both ends of each log.  
<sup>6</sup>W/UV, the weight of harvested mushrooms (g) per 100-cm<sup>3</sup> of initial undiscolored wood volume.  
<sup>7</sup>Number of Stubs, the number of branch stubs alive at the time of harvest on each substrate log.  
<sup>8</sup>Stub Diameter, the total diameter (cm) of all branch stubs alive at the time of harvest on each substrate log.

Table 2.—Patterns of significance detected by three ANOVA models of shiitake production during 2000 applied to two experiments

Source of variation <sup>1</sup>	EXPERIMENT 1 <sup>2</sup>			EXPERIMENT 2 <sup>2</sup>		
	BE None	W/UV None	W/UV SA	BE None	W/UV None	W/UV SA
Shiitake Strain (SS)	*** <sup>3</sup>	***	***	***	***	***
Log Species (LS)	*	*	**	***	***	***
SS x LS	*	***	***	**	***	***
Inoculum Form (IF)	*	***	***	ns	ns	ns
SS x IF	ns	**	***	ns	ns	ns
LS x IF	ns	ns	ns	ns	ns	ns
Inoculation Delay (ID)	—	—	—	ns	ns	ns
SS x ID	—	—	—	ns	ns	ns
LS x ID	—	—	—	*	ns	ns
IF x ID	—	—	—	ns	ns	ns
Block	ns	ns	ns	ns	*	*
Log Surface Area	—	—	***	—	—	*

<sup>1</sup>Variables included: Shiitake Strain (3 - Cold Weather, Wide Range, Warm Weather); Log Species (3 - *Acer sacharum*, *Quercus alba*, *Q. rubra*); Inoculum Form (2 - sawdust and dowel inocula used December 1999, sawdust and thimble inocula used May 2000); Inoculation Delay (2 - only for May 2000 inoculation, no delay or 4-mo delay post-harvest); Block (9 for December 1999 experiment, 8 for May 2000 experiment); Surface Area (the surface area outside bark of each substrate log, cm<sup>2</sup>).  
<sup>2</sup>Experiments 1 and 2, respectively, represent logs inoculated either in early December 1999 or May 2000. Model response variables were either Biological Efficiency (BE, the fresh weight of all mushrooms harvested from a substrate log expressed as a percentage of initial log dry weight) or Weight/Undiscolored wood volume (W/UV, total weight of harvested mushrooms (g) per 100-cm<sup>3</sup> of undiscolored wood volume for individual shiitake logs. Log Surface Area (SA) was either included in the model as a covariate or not (None).  
<sup>3</sup>Tabular symbols indicate significance of F: \*,  $P \leq 0.05$ ; \*\*,  $P \leq 0.01$ ; \*\*\*,  $P \leq 0.001$ ; ns, not significant; —, not tested.



We were motivated to compare BE and W/UV as alternative response variables for modeling log-based shiitake mushroom productivity for two reasons. First, differences between red and white oaks and maples in stemwood anatomy and heartwood composition may result in different rates and patterns of substrate log decomposition by shiitake mycelium, resulting in different efficiencies of wood energy conversion into mushroom biomass. Certain tree genera (including oaks) routinely produce highly discolored heartwood through the deposition of toxic extractives in dying sapwood cells. These toxic extractives are responsible for the natural decay resistance of heartwood (Zabel and Morrell 1992). Other tree genera (including maples) normally produce much less decay-resistant heartwood, which is undiscolored due to its lower extractive content. Maple heartwood darkens only through the living tree's compartmentalization response to stem injury (Shigo 1984) and microbial colonization (Shigo 1972). The heartwoods of white oaks, red oaks, and maples have been rated resistant, moderately susceptible, and susceptible to decay, respectively (Scheffer and Cowling 1966).

For the above reasons, we hypothesize that a larger percentage of the volume of maple logs compared to oak logs may be more efficiently utilized by shiitake. If this is the case, then BE (which is based on total log weight) may provide incomplete insight into differences in productivity between oaks and maples. Through consideration of both BE and W/UV as response variables, we expect to both document and explain differences in shiitake productivity between sugar maple and red and white oak. To the extent that undiscolored heartwood and sapwood are better energy sources for shiitake growth than discolored heartwood, W/UV should provide additional useful insight for substrate species comparisons.

Second, the proportion of decay-resistant discolored heartwood in oak logs generally increases as log diameter increases. This phenomenon biases attempts to compare the effects of treatments if they are applied to logs of different sizes. Here again, W/UV may provide useful insight into the effects of log size as well as management practices.

Another interesting substrate log characteristic under study is surface area (SA), which directly measures the space available for mushroom production. Substrate log SA also indirectly reflects the tendency of a log to dry out between

wetting events. Though SA was not correlated with W during year 2000 (table 1), SA was significantly correlated with substrate log productivity in year 2000 analyses of W/UV (table 2). We will continue to evaluate the relationship between SA and productivity. The information gained should help us to determine the effect of substrate log size on productivity.

The poor correlations for year 2000 shiitake harvest (W) with either SA or UV probably reflect the early stages of these experiments. We anticipate that these correlations will become significant as the experiments progress and the strains more completely utilize the substrate logs. We are evaluating the correlations between shiitake productivity and substrate log branchiness. Severed branches represent both points of potential invasion by competitor fungi and locations from which excessive substrate drying may take place. As might be expected this early in an experiment, there was no apparent effect of branchiness in the year 2000 analysis.

Productivity by the warm weather strain WW44 in year 2000 of both our experiments clearly lagged behind that of the other two strains, and this trend appeared to continue less markedly through July 2001. Jo Krawczyk (Field & Forest Products, Inc., personal communication) indicated that productivity by WW44 increases more gradually compared with other WW46 and CW50, and that WW44 tends to continue to fruit for a year or two longer than the other strains. We will be able to determine if this is the case under conditions common for central Missouri. Also, the years 2000 and 2001 growing seasons were not particularly warm, and it would be useful to document the performance of WW44 during a warmer growing season. Strain CW50 is performing well in these experiments. This strain fruited substantially in early April 2001 (approximately DOY 93, as much as a month earlier than WR46 or WW44). In fact, some form of protection from spring frost and desiccation prior to overstory foliation would have enhanced yields of all three strains during March and April 2001, when many mushroom primordia were lost to frost and dry weather.

We expect that the substrate species effect on productivity will become clearer as the experiments mature. Because pole-size sugar maple stems characteristically contain less discolored heartwood than do oak stems, one might expect shiitake to decompose sugar maple logs more completely, leading to higher BE values for maple logs than for oak logs. That seems to be

the case already in Experiment 1. Nevertheless, because it may take shiitake mycelium longer to penetrate deeply enough to access all the non-discolored wood in maple logs, it isn't surprising that W/UV appears to be greater for oak than for maple initially. If maple logs are consumed faster and/or more completely due to their less decay-resistant heartwood, use of sugar maple may provide growers a faster and/or greater return on their investment.

The only difference in productivity during year 2000 attributable to spawn form developed in Experiment 1, where traditional sawdust-based spawn outproduced solid wood (dowel) spawn. Solid wood spawn has been recommended for autumn inoculations in colder climates where winter temperatures might result in frost-heaving of more traditional sawdust-based spawn (Kozak and Krawczyk 1993). While we have noticed some winter disturbance of sawdust spawn, the disturbance does not resemble frost heave. Rather, we attribute this disturbance to birds because disturbances are most often conical gouges in the spawn. Six species of woodpeckers have been observed in the study area. No difference developed during year 2000 between the performances of traditional sawdust and thimble spawn.

No difference in productivity has yet developed between logs harvested dormant and inoculated 13-weeks later compared with logs harvested just after bud-break and inoculated immediately. We are concerned that the process of aging cut logs might favor development of *Hypoxylon atropunctatum* and/or other competing wood decay fungi. Although competing wood decay fungi have not been a serious problem yet, we will evaluate fruiting by competing fungi on each log to serve as a covariate to model the impact of competition on shiitake production.

#### ACKNOWLEDGMENT

The U.S. Environmental Protection Agency and the USDA Agricultural Research Service, Dale Bumpers Small Farms Research Center, Booneville, AR funded this work. This research was also, in part, supported by the Missouri Agricultural Experiment Station project number PSSL 0112. The results presented are the sole responsibility of the Principal Investigators and may not represent the policies or positions of the funding agencies.

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## APPENDICES

Appendix Table 1.—Results of ANOVA using Biological Efficiency of individual shiitake logs in harvest year 2000 as the response variable to detect significant differences among treatments for Experiment 1 (initiated early December 1999) and for Experiment 2 (initiated May 2000)<sup>1</sup>

Source of variation	df	SS	F-value	Significance of F
<b>EXPERIMENT 1:</b>				
Model	21	71.932	4.75	<0.0001
Shiitake Strain (SS)	[2]	41.877	29.06	<0.0001
Log Species (LS)	[2]	4.669	3.24	0.0422
SS x LS	[4]	9.084	3.15	0.0163
Inoculum Form (IF)	[1]	3.882	5.39	0.0218
SS x IF	[2]	3.963	2.75	0.0675
LS x IF	[2]	0.730	0.51	0.6038
Block	[8]	7.838	1.36	0.2197
Error	135	97.258		
Total	156	169.190		
<b>EXPERIMENT 2 :</b>				
Model	26	63.898	3.96	<0.0001
Shiitake Strain (SS)	[2]	29.334	23.62	<0.0001
Log Species (LS)	[2]	9.966	8.02	0.0004
SS x LS	[4]	9.755	3.93	0.0042
Inoculum Form (IF)	[1]	0.068	0.11	0.7412
SS x IF	[2]	2.895	2.33	0.0995
LS x IF	[2]	0.701	0.56	0.5696
Inoculation Delay (ID)	[1]	2.249	3.62	0.0583
SS x ID	[2]	1.236	1.00	0.3712
LS x ID	[2]	4.207	3.39	0.0355
IF x ID	[1]	0.238	0.38	0.5368
Block	[7]	3.890	0.89	0.5113
Error	236	146.579		
Total	262	210.477		
<sup>1</sup> Biological Efficiency (BE), the fresh weight of all mushrooms harvested from a substrate log expressed as a percentage of initial log dry weight.				

Appendix Table 2.—Results of ANOVA using Weight/Undiscolored wood volume for individual shiitake logs for year 2000 harvest as the response variable to detect significant differences among treatments for Experiment 1 (initiated early December 1999) and Experiment 2 (initiated May 2000)<sup>1</sup>

Source of variation	df	SS	F-value	Significance of F
<b>EXPERIMENT 1:</b>				
Model	21	582.952	9.08	<0.0001
Shiitake Strain (SS)	[2]	332.392	54.33	<0.0001
Log Species (LS)	[2]	19.149	3.13	0.0469
SS x LS	[4]	89.492	7.31	<0.0001
Inoculum Form (IF)	[1]	41.230	13.48	0.0003
SS x IF	[2]	33.872	5.54	0.0049
LS x IF	[2]	7.844	1.28	0.2808
Block	[8]	45.886	1.87	0.0690
Error	135	412.944		
Total	156	995.896		
<b>EXPERIMENT 2:</b>				
Model	26	109.405	5.33	<0.0001
Shiitake Strain (SS)	[2]	41.848	26.51	<0.0001
Log Species (LS)	[2]	22.908	14.51	<0.0001
SS x LS	[4]	16.570	5.25	0.0005
Inoculum Form (IF)	[1]	0.093	0.12	0.7322
SS x IF	[2]	3.559	2.25	0.1072
LS x IF	[2]	1.869	1.18	0.3078
Inoculation Delay (ID)	[1]	1.338	1.69	0.1942
SS x ID	[2]	1.341	0.85	0.4289
LS x ID	[2]	2.118	1.34	0.2634
IF x ID	[1]	1.649	2.09	0.1496
Block	[7]	14.005	2.53	0.0156
Error	235	185.496		
Total	261	294.902		
<sup>1</sup> Weight/Undiscolored wood volume, total weight of harvested mushrooms (g) per 100-cm <sup>3</sup> of undiscolored wood volume for individual shiitake logs.				

Appendix Table 3.—Results of ANOVA using Weight/Undiscolored wood volume for individual shiitake logs for year 2000 harvest as the response variable (and log surface area as a covariate) to detect significant differences among treatments for Experiment 1 (initiated early December 1999) and Experiment 2 (initiated May 2000)<sup>1</sup>

Source of variation	df	SS	F-value	Significance of F
<b>EXPERIMENT 1:</b>				
Model	22	615.326	9.85	<0.0001
Shiitake Strain (SS)	[2]	308.822	54.37	<0.0001
Log Species (LS)	[2]	30.063	5.29	0.0061
SS x LS	[4]	75.145	6.61	<0.0001
Inoculum Form (IF)	[1]	38.765	13.65	0.0003
SS x IF	[2]	41.592	7.32	0.0010
LS x IF	[2]	6.857	1.21	0.3023
Block	[8]	38.943	1.71	0.1006
Surface Area	[1]	32.374	11.40	0.0010
Error	134	380.571		
Total	156	995.896		
<b>EXPERIMENT 2:</b>				
Model	27	113.913	5.45	<0.0001
Shiitake Strain (SS)	[2]	41.160	26.61	<0.0001
Log Species (LS)	[2]	20.548	13.28	<0.0001
SS x LS	[4]	15.582	5.04	0.0007
Inoculum Form (IF)	[1]	0.161	0.21	0.6484
SS x IF	[2]	3.322	2.15	0.1191
LS x IF	[2]	2.849	1.84	0.1609
Inoculation Delay (ID)	[1]	2.451	3.17	0.0763
SS x ID	[2]	1.146	0.74	0.4779
LS x ID	[2]	1.572	1.02	0.3636
IF x ID	[1]	1.388	1.79	0.1817
Block	[7]	13.830	2.55	0.0149
Surface Area	[1]	4.507	5.83	0.0165
Error	234	180.989		
Total	261	294.902		
<sup>1</sup> Weight/Undiscolored wood volume, total weight of harvested mushrooms (g) per 100-cm <sup>3</sup> of undiscolored wood volume for individual shiitake logs; surface area, the surface area (cm <sup>2</sup> , outside bark) of each substrate log.				