Prescribed burning reduces species richness of wood-decay fungi in the forests of northwest Arkansas

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Abstract

The present study, which was carried out at three localities in the Ozark Mountains of northwest Arkansas, investigated the effects of prescribed burning on wood-decomposing fungi using samples of decaying woody debris (DWD) placed in plastic incubation chambers. One of the localities had not been subjected to recent prescribed burning, whereas the other localities contained both an unburned area as well as an area recently subjected to burning. In all three localities, small pieces of decaying woody debris (DWD) were collected, placed in the incubation chambers and the latter kept moist for a period of time. Pieces of DWD collected in the areas subjected to burning typically displayed evidence of considerable charring. Fruiting bodies appearing in the incubation chambers were removed and identified by sequencing of ribosomal DNA region. A total of 101 specimens representing 80 different taxa were recorded in the entire investigation, but the numbers of both specimens and taxa were appreciably higher for the unburned collecting sites. As such, the data obtained indicate that prescribed burning lowers the species richness of the wood-decomposing fungi associated with DWD at a particular locality. The unique aspect of the present study was the use of incubation chambers to characterize the taxa of fungi associated with CWD.

1. Introduction

The decomposition of wood (coarse woody debris) by the enzymatic activity of what are referred to as wood-decay fungi in forest ecosystems is an essential part of the carbon cycle in nature. In addition to recycling the elements in wood, the decaying wood creates an important habitat for wildlife, insects, mycorrhizal tree roots, other fungi, and an incredible diversity of microorganisms. As such, any factor that affects the decomposition of wood potentially has profound consequences for the overall diversity and functioning of forest ecosystems. However, there have been relatively few studies that have examined the impact of prescribed burning on wood-decay fungi, although prescribed burning is extensively used as a management technique in forests such as those in northwest Arkansas (Larsen et al., 1982). Studying the wood-decay fungi associated with forest ecosystems has one major constraint. Identification of the fungi involved is based almost exclusively upon features of the fruiting bodies, and these are not always available in the field. The research approach described herein makes use of incubation chambers to encourage the production of fruiting bodies from fungal mycelia already present pieces of coarse woody debris collected in the field but brought back to the laboratory. Among the advantages are that samples can be collected at any time of the year and some of the fungi appearing on the samples are small forms that are likely to be overlooked in nature.

As such, the aim of this study was to investigate the effect of prescribed burning wood-decomposing fungi. The unique aspect of the present study was the use of plastic incubation chambers to describe the taxa of fungi associated with DWD. Specimens of the fruiting bodies of wood-decomposing fungi can be collected in the field, but the appearance of fruiting bodies is dependent upon the prevailing weather conditions. Moreover, those species which produce fleshy fruiting bodies are likely to be underrepresented because the fruiting bodies do not persist for very long in nature. Presumably, placing pieces of DWD in incubation chambers.
and keeping the latter moist would promote the growth of any mycelia already present in the DWD, with the ultimate production of fruiting bodies. To our knowledge, no previous study of wood-decomposing fungi has used this technique.

The present study was carried out at three localities in the Ozark Mountains of northwest Arkansas. These were Devil’s Den State Park (35°46′29″N, 94°14′45″W, elevation 476 m), the Buffalo National River (36°10′41″N and 92°25′34″W, elevation 153 m), and Pea Ridge National Military Park (36°27′15″N, 94°02′05″W, elevation 439 m), and the climate of northwest Arkansas is mild in the winter and hot and humid in the summer. The forests are dominated in a mixture of several species of hickory (Carya ovata [Mill.] K. Koch, C. texana Buckley and C. tomentosa Sarg.), oak (Quercus alba L., Q. velutina Lam., Q. stellata Wangenh. and Q. rubra L.) and with an admixture of other species.

Decaying woody debris (DWD) consists of woody substrates resulting from tree branches, trunks and roots in numerous stages of decomposition. The accumulation of DWD in a particular forest depends upon a number of factors, including weather conditions, animal and disease efficiency, forest composition and prescribed burning intervals. The decomposition of DWD releases important nutrients such as sulfur, nitrogen and phosphorus into the soil, particularly near the end of the decomposition process. DWD has a high concentration of these nutrients (Larsen et al., 1982).

Prescribed burning (also called controlled burning) is a management technique used to decrease fuel load and to return a disturbance regime to landscapes which historically were subjected to fire. Theoretically, the organisms that occur in the area burned are subjected to the direct effects of fire. For example, plants can be killed when the aboveground portions are burned. Large species of animals can avoid the fire but many small ones (e.g., invertebrates) cannot. The extent to which prescribed burning affects the fungi associated with DWD is still understudied.

Presumably, prescribed burning has an indirect impact on wood-decay fungi as a result of what happens to DWD when the latter is subjected to fire. Theoretically, the organisms that occur in the area burned are subjected to the direct effects of fire. For example, plants can be killed when the aboveground portions are burned. Large species of animals can avoid the fire but many small ones (e.g., invertebrates) cannot. The extent to which prescribed burning affects the fungi associated with DWD is still understudied.

Table 1

| Time/hrs | 250 °C | 300 °C | 350 °C | 400 °C | 500 °C |
|----------|--------|--------|--------|--------|--------|
| 1        | 93.6   | 33.3   | 27.8   | 22.3   | –      |
| 8        | 44.1   | 30.7   | 23.5   | –      | –      |
| 24       | 35.5   | 27.2   | 18.9   | 7.8    | –      |

3. Molecular characterization of fungi

DNA was extracted from dried fungal tissue using a Wizard genomic purification kit (Promega Corporation, Madison, Wisconsin). The internal transcribed spacer (ITS) region of ribosomal DNA was targeted for PCR amplification with the primers ITS1 (5′-TCCGTAAGGTTCACCAATCGG-3′) and ITS4 (5′-TCTCCGCTATTGATATGC-3′) (White et al., 1992). PCR amplifications were performed in a Applied Biosystems thermocycler with 35 cycles programmed for initial denaturation at 95 °C for 5 min, denaturation at 95 °C for 45 sec, annealing at 50 °C for 45 sec, followed by final extension at 72 °C for infinite time. Amplified products were analysed and confirmed using 1% agarose gel electrophoresis. Furthermore, Sanger sequencing was performed with identified amplicons and later BLAST search was done against the various taxon available in NCBI database (www.ncbi.nlm.nih.gov). Moreover, the species were recorded at 95% sequence identity and above, whereas genus level was identified at less than 95% identity. The author of each fungal taxa was determined using Index Fungorum (http://www.indexfungorum.org/Names/Names.aspx).

4. Results and discussion

A total of 101 specimens were collected from the 60 incubation chambers. These represented 80 taxa (Table 2), the majority of which could be identified to the level of species. Seventy-four taxa were reported from samples of DWD from unburned areas, whereas only six taxa were collected from samples of DWD from burned areas (Fig. 3). The taxa collected belong to 34 different families, with representatives of the Mycenaceae, Polyporaceae, Omphalotaceae and Xylariaceae the most common. The Marasmiaceae was the most diverse family with eight different species, and five taxa were recorded from samples of DWD from unburned areas. The majority of taxa was determined using Index Fungorum (http://www.indexfungorum.org/Names/Names.aspx).
information relating to its effects on wood-decomposing fungi, especially in the forests in the northwest Arkansas. The data generated in the present study indicated that the number of wood-decomposing fungi was appreciably higher on unburned DWD as compared to burned (and usually highly charred) DWD. There was a clear difference in the number of wood-decomposing fungi between unburned and charred decaying woody debris (Figs. 3 and 4).

Taxa of wood-decomposing fungi are clearly affected by prescribed burning, and a few species appear to tolerate burning better than others. For example, five species each of Gymnopus and Marasmius were recorded only on DWD from unburned areas of the Pea Ridge Military National Park and the Buffalo National River (Fig. 5). They were not recorded on charred DWD collected from burned areas of these same two localities. However, Perenniporia ohiensis, Mycena aurantia-marginata and Blastobotrys sp. were recorded on samples of DWD collected from burned areas but were absent on samples from unburned areas. In contrast, a few taxa were recorded on samples of DWD from both the burned and

| Study area                        | Number of specimens | Number of taxa |
|----------------------------------|---------------------|----------------|
| Devil’s Den State Park (control) | 34                  | 30             |
| Pea Ridge National Military Park | 33                  | 27             |
| (unburned area)                  | 7                   | 5              |
| Pea Ridge National Military Park | 24                  | 17             |
| (burned area)                    |                      |                |
| Buffalo National River (unburned | 3                   | 1              |
| area)                            |                      |                |
| Buffalo National River (burned   |                      |                |
| wood)                            |                      |                |
| Total                            | 101                 | 80             |

Fig. 1. Stages in the collection of coarse woody debris. A. Wood divided into four pieces, B. Two pieces were prepared for each chamber for prescribed burning, and C. Coarse woody debris conducted for prescribed burning. (Photo by author).

Fig. 2. Two pieces were placed in each incubation chamber. A: Coarse woody debris incubated before burning and B: Coarse woody debris incubated after burning. (Photo by author).

Fig. 3. Variation in the number of wood-decay fungi between unburned and charred coarse woody debris. Note: PRP, Pea Ridge Military National Park; DDP, Devil’s Den State Park; PFR, the Buffalo National River.
| Taxon                                      | CDD | CUBF | CBBF | CUPR | CBPR |
|-------------------------------------------|-----|------|------|------|------|
| Agaricus pinsitus Fr.                     |     | X    |      |      |      |
| Blastobotrys. sp. 1                       |     |      | X    |      |      |
| Clitocybe subditopoda Peck                |     | X    |      |      |      |
| Coprinus albivorus Bogar                  |     | X    |      |      |      |
| Cordyceps confragosa (Mains) G.H. Sung, J.M. Sung, Hywel-Jones & Spatafora |     |      | X    |      |      |
| Cryptococcus yokahamensis Alshahi, Satoh & Makimura |     |      |      | X    |      |
| Diatrype stigma (Hoffm.) Fr.              |     |      |      |      | X    |
| Echleriella sp. 1                         |     |      |      |      | X    |
| Galella rufa (Schwein.) Nannf. & Korf     | X   |      |      |      |      |
| Gymnopilus fimbrius (Peck) Halling        |     |      |      |      |      |
| Gymnopilus dryophilus (Bull.) Murrill     | X   |      |      |      |      |
| Gymnopilus earlæe Murrill                 | X   |      |      |      |      |
| Gymnopilus junquileus R.H. Petersen & J.L. Mata | X   |      |      |      |      |
| Gymnopilus luxurians (Peck) Murrill       |     | X    |      |      |      |
| Hyphodontia tropica Sheng H. Wu           |     |      |      |      | X    |
| Lachnum virgineum (Batsch) P. Karst.     | X   |      |      |      |      |
| Marasmieus juniperinus Murrill            | X   |      |      |      |      |
| Marasmius cohaerens (Pers.) Cooke & Quél.| X   |      |      |      |      |
| Marasmius graminicolor Spec.              | X   |      |      |      |      |
| Marasmius pulcheripes Peck                | X   |      |      |      |      |
| Marriannea sp. 1                          |     |      |      | X    |      |
| Merulius incarnatus Schwein.              | X   |      |      |      |      |
| Mycena acicula (Schaeff.) P. Kumm.       | X   |      |      |      |      |
| Mycena aurantiomarginata (Fr.) Quél.     |     |      |      | X    |      |
| Mycena haematopus (Pers.) P. Kumm.       | X   |      |      |      |      |
| Mycena polygramma (Bull.) Gray            | X   |      |      |      |      |
| Mycena sp. 1                              | X   | X    |      |      |      |
| Mycena thymicola Velen.                   | X   |      |      |      |      |
| Nectria mariannaeae Samuels & Seifert     | X   |      |      |      |      |
| Panus conchatus (Bull.) Fr.               | X   | X    |      |      |      |
| Panus lecomtei (Fr.) Corner               | X   |      |      |      |      |
| Panus nestriginosus Drechsl-Santos & Wartchow | X   |      |      |      |      |
| Perenniporia ohiensis (Berk.) Ryvarden   | X   |      |      |      |      |
| Pezizomycetes sp. 1                       | X   |      |      |      |      |
| Physalacria bambusae Hono.                | X   |      |      |      |      |
| Physisporinus vitreus (Pers.) Karst.      | X   |      |      |      |      |
| Pleurotus floridanus Singer               | X   |      |      |      |      |
| Pleurotus ostreatus (Jacq. ex Fr.) P. Kumm. | X   |      |      |      |      |
| Pluteus romellii (Britzelm.) Sacc.        | X   |      |      |      |      |
| Pluteus sp. 1                             | X   |      |      |      |      |
| Pluteus thomsonii sensu Singer            | X   |      |      |      |      |
| Polyporus tuberaster (Jacq. ex Pers.) Fr. | X   |      |      |      |      |
| Resupinatus alboniger (Pat.) Singer       | X   | X    |      |      |      |
| Resupinatus appicatus (Batsch) Gray       | X   |      |      |      |      |
| Rhizopus oryzae Went & Prins. Geerl.     | X   |      |      |      |      |
| Rigidosporus pouzarii Vampola & Vlasák    | X   |      |      |      |      |
| Rigidosporus sp. 1                        | X   |      |      |      |      |
| Schizophyllum commune Fr.                 | X   |      |      |      |      |
| Schizopyllum radiatum Fr.                 | X   |      |      |      |      |
| Schizospora sp. 1                         | X   |      |      |      |      |
| Scutellinia crinita (Bull.) Lambotte.     | X   |      |      |      |      |
| Scutellinia sp. 1                         | X   |      |      |      |      |
| Simocybe sp. 1                            | X   |      |      |      |      |
| Simplexillum lanosoniveum (J.F.H. Beyma) Zare & W. Gams | X   |      |      |      |      |
| Stereum sanguinolentum (Alb. & Schwein.) Fr. | X   |      |      |      |      |
| Stereum sp. 1                             | X   |      |      |      |      |
| Trametopsis cervina (Schwein.) Tomlovs ký | X   |      |      |      |      |
| Trametopsis sp. 1                         | X   |      |      |      |      |
| Trichaptum fuscoviolaceum (Ehrenb.) Ryvarden | X   |      |      |      |      |
| Trichoderma gamssii Samuels & Druzhin.    | X   |      |      |      |      |
| Trichoderma sp. 1                         | X   |      |      |      |      |
| Trichoderma viride Pers.                  | X   |      |      |      |      |
| Trogia sp. 1                              | X   |      |      |      |      |
| Truncospora ohiensis (Berk.) Pilát        | X   |      |      |      |      |
| Truncospora sp. 1                         | X   |      |      |      |      |
| Xylaria cornu-damae (Schwein.) Berk.      | X   |      |      |      |      |
| Xylaria sp. 1                             | X   |      |      |      |      |
| Xylaria sp. 2                             | X   |      |      |      |      |
unburned areas. The most prominent examples were *Polyporus tuberaster* and *Panus conchatus*. In addition, to the fruiting bodies that were collected and identified, mycelia that never produced fruiting bodies appeared on several of the samples of charred DWD.

Presumably, based on the results of the present study and other studies reported in the literature, the heat from prescribed burning results in the direct destruction of fruiting bodies and kills mycelia present in DWD while also creating rather unfavorable conditions for the post-burning inoculation by the spores or mycelia of other fungi (Parmeter, 1977, Zak, 1991). In the two localities where prescribed burning takes place, it could be observed that larger pieces of DWD (e.g., stumps and logs) were characterized by the conspicuous absence of macroscopic (i.e., easily visible with the naked eye) fruiting bodies. Under the confined space of an incubation chamber, the inoculation potential of other fungi would be expected to be very low, certainly lower than the conditions that exist in the field. However, it would appear that the results obtained from incubation with respect to inoculation are comparable to what typically occurs under natural conditions in the field.

In summary, the data reported herein support the concept that prescribed burning has a noticeable effect upon wood-decay fungi, resulting in an appreciable reduction in the numbers of both fruiting bodies and taxa. Documenting the extent of this reduction is difficult when relying solely on observations and collections of fungi under field conditions, but the use of incubation chambers would seem to represent a way of obtaining a relatively solid body of data, especially in comparative studies.

**Declaration of Competing Interest**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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**Fig. 4.** Variation in the number of wood-decay fungi between unburned and charred coarse woody debris.

**Fig. 5.** Selected species of wood-decay fungi documented from the incubation chambers. A. *Xylaria hypoxylon*, B. *Polyporus tuberaster*, C. *Trichaptum fuscoviolaceum*, and D. *Mycena leaiana*. (Photo by the senior author).
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