

Preliminary Survey Of Wood-Associated Fungi In Southeast O'ahu Of Hawai'i Using DNA-Based Identification

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INTRODUCTION

Hawai'i is a biological hotspot with a variety of climates and habitats. While fungal species diversity has been more extensively studied in Hawai'i than other Pacific Islands (e.g. see Gilbertson et al. 2002), there remain many species unreported in the literature. This project attempted to capture a small portion of Hawai'i's fungal diversity in southeast O'ahu by identifying fruit bodies using molecular techniques. The efficacy of using the internal transcribed spacer (ITS) and large subunit (LSU) regions of the nuclear ribosomal DNA (rDNA) to identify Hawai'ian fungi is discussed.

METHODS

Fungi were collected from the Honolulu Mauka trail system, Maunawili, and Waikiki in southeast O'ahu. Samples from most collections were cultured on potato-dextrose agar (PDA) and benomyl-dichloran-streptomycin (BDS) medium, which were sent with the accompanying fruiting bodies to the Rocky Mountain Research Station (RMRS) for DNA-based identification. The RMRS extracted DNA from each collection and conducted PCR and sequencing using fungal ITS and LSU primers. Sequence data were compared to similar sequences in the NCBI GenBank® database and specimens were tentatively

identified using the closest named match in the database. Sequences that matched non-wood associated fungi and were not similar to morphological identifications of fruiting body collections were not further analyzed.

RESULTS

We tentatively identified 15 specimens using the ITS region, LSU region, or both. Of those specimens where both ITS and LSU sequences were successfully obtained, 75% matched species-level identifications. The other 25% were identified as different but related genera, depending on the region of the rDNA sequenced (Table 1).

DISCUSSION

Fungal LSU and ITS sequences are well represented in GenBank®, making them useful in preliminary identifications of fungi. This study targeted fungi with distinctive fruit bodies (Figure 1) and all samples had at least a strong genus-level match to similar sequences in GenBank®. Due to low sequence variation for some taxonomic groups in the ITS and LSU regions, such as species in the genus *Ganoderma* (Moncalvo et al. 1995 A & B), species-level identification using these regions is not always conclusive. However, these regions did provide relatively quick and easy taxonomic delineations and could lead to more precise molecular and morphological studies on relevant specimens.

Comprehensive, updated species lists are useful to land managers, allowing them to track species composition and movement over time. Currently, molecular tools are the most practical means of identifying fungi for non-

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mycologists. The increasing availability of more advanced tools in the field of metagenomics could allow managers to quickly understand species composition of larger communities. To use molecular tools to full advantage, however, reference materials identified by taxonomic experts and relevant, easily searchable DNA sequence databases must be maintained. It is likely only a fraction of Hawai'ian fungi have

been recorded and for many species there are no preserved specimens or DNA records. This study tentatively identified four species previously unrecorded in Hawai'i: *Microporus xanthopus*, *Inonotus rickii*, *Panus* spp. and *Coriopsis rigida*. Further morphological and molecular analyses are needed to confirm these identifications.

Table 1. ITS and LSU identifications of O'ahu fungi. Yellow highlighted rows indicate a species match in both regions, white indicates no match, and gray indicates incomplete data.

ITS1 +5.8S + ITS2 regions				LSU D-Domain region		
Isolate ID	Closest GenBank Match	Maximum Identity Score	Closest GenBank Match Accession Number	Closest GenBank Match	Maximum Identity Score	Closest GenBank Match Accession Number
FB1	<i>Fuscoporia gilva</i>	95%	FJ481039	<i>Fuscoporia gilva</i>	99%	HQ328525
FB2	<i>Earliella scabrosa</i>	99%	JN164992	<i>Earliella scabrosa</i>	99%	JN164793
FB3	<i>Microporus xanthopus</i>	98%	JX290074	Not Available		
FB7	Not Available			<i>Fuscoporia gilva</i>	99%	HQ328525
FB8	<i>Microporus xanthopus</i>	98%	JX290074	<i>Microporus xanthopus</i>	99%	JX290071
FB9	<i>Physisporinus vitreus</i>	95%	JN182920	<i>Rigidoporus microporus</i>	100%	AY333795
FB13	<i>Earliella scabrosa</i>	99%	JN165006	<i>Earliella scabrosa</i>	100%	JN164793
FB14	<i>Ganoderma gibbosum</i>	99%	EU273513	<i>Fomes fomentarius</i>	98%	DQ208419
FB17	<i>Physisporinus vitreus</i>	95%	JN182920	<i>Rigidoporus microporus</i>	99%	AY333795
FB19	<i>Inonotus rickii</i>	96%	GU111921	Not Available		
FB21	<i>Inonotus rickii</i>	96%	KC479129	Not Available		
FB22	<i>Panus</i> sp.	99%	KJ195662	Not Available		
FB23	<i>Polyporus arcularius</i>	99%	AF516524	<i>Polyporus arcularius</i>	99%	AF393067
FB24	<i>Coriopsis rigida</i>	99%	JF894112	<i>Coriopsis rigida</i>	99%	KC867454
FB25	<i>Trametes sanguinea</i>	99%	JN164981	Not Available		

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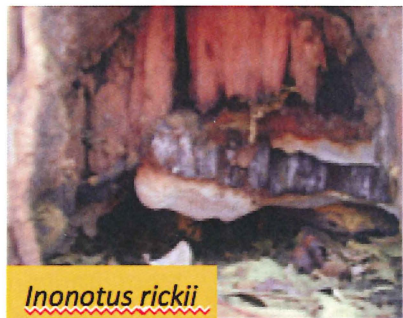
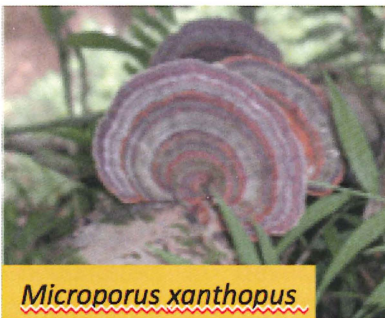


Figure 1. Wood rotting fungi from O'ahu and corresponding ITS- and LSU-sequence based identifications. Fungi that resulted in different GenBank species matches between ITS and LSU gene regions are denoted by sample number only (Photos by F. Brooks).