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Investigating Wood Decaying Fungi Diversity in Central Siberia, Russia Using ITS Sequence Analysis and Interaction with Host Trees

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Abstract: Wood-decay fungi (WDF) play a significant role in recycling nutrients, using enzymatic and mechanical processes to degrade wood. Designated as a biodiversity hot spot, Central Siberia is a geographically important region for understanding the spatial distribution and the evolutionary processes shaping biodiversity. There have been several studies of WDF diversity in Central Siberia, but identification of species was based on morphological characteristics, lacking detailed descriptions and molecular data. Thus, the aim of this study was to identify WDF in Central Siberia, regarding the degradation of host trees based on both morphological and molecular analyses. We collected 106 WDF samples from Krasnoyarsk and the Republic of Khakassia in 2014 and 2017, and identified a total of 52 fungal species from six main host tree genera. In order to assess the host preference of the WDF, we examined previous literature, and data from this study. We confirmed a division in host preference of WDF between gymnosperms and angiosperms. DNA-based identification and host preference assessment of the WDF provide preliminary data on WDF diversity and their role in nutrient cycles in the ecosystem of Central Siberia. To fully understand WDF diversity in Central Siberia, continuous long-term surveys, including DNA sequence data, are needed.

Keywords: Central Siberia; ITS sequence; host preference; wood-decay fungi

1. Introduction

Wood-decay fungi (WDF) decompose dead wood or attack living trees as a pathogen. Wood decomposition is a crucial biological process that breaks down complex molecules and recycles nutrients back to the soil [1,2]. Through the wood decay cycle, young trees grow and replace dead or decaying trees. As such, WDF are excellent ecosystem engineers that play a pivotal role in recycling nutrients in a forest ecosystem, and in providing a viable habitat for many other organisms [3,4].

Although WDF are not part of a natural taxonomic group, they form a number of well-defined groups within Ascomycota and Basidiomycota [5,6]. The majority of WDF typically form a fruitbody

and belong to the class Agaricomycetes in Basidiomycota. Polypore decay fungi also form a polyphyletic group, with representatives in the orders Hymenochaetales, Polyporales, and Gloeophyllales. Gilled pleurotoid and corticoid decay fungi also form polyphyletic groups with representatives in orders Hymenochaetales, Corticiales, and Russulales [7,8]. WDF degrade wood using both enzymatic and mechanical processes, and based on the type of decay of the wood lingo-cellulose material, WDF fall into two groups: brown-rot fungi and white-rot fungi [3].

Russia, the largest country in the world, has a variety of environments and landforms such as arctic tundra, forest tundra, taiga, mixed and deciduous forests, broad-leaved forests, steppes, semi-deserts, and subtropics [9]. There are three biodiversity hotspots in Russia; the North Caucasus, South Siberia, and the Far East [10]. Recently, biologists from various backgrounds have shown a great interest in Central Siberia as a biodiversity hot spot [11,12]. As a geographic link between Europe and Asia, Central Siberia is an important region for understanding the spatial distribution and the evolutionary processes of a diversity of organisms, including WDF. WDF have been well-surveyed in the Western and Northern Siberian Plains [13–16], the Ural Mountains [17,18], the Altai-Sayan Mountains [19], and in several islands and peninsulas [20,21], but there is limited information from Central Siberia. Recent studies in Central Siberia consist of reports of WDF and their corresponding hosts without detailed descriptions or molecular data [15,22,23]

As many WDF produce a relatively rigid fruiting body, they are often identified based on morphological characteristics such as pileus color, pore size, basidiospore shape, cystidia shape, hyphal system, and type of rot [24,25]. However, species identification based on morphology alone has been shown to be inaccurate, due to morphological similarities and the tendency of morphological characters to undergo convergent evolution [26,27]. With an increase in available sequence data in public databases, sequence-based identification has become prevalent [28–30]. Sequence-based identification enables taxonomists to distinguish morphologically similar organisms, and to identify fruiting bodies that have no unique characteristics, are premature, or are fragmented in pieces. The internal transcribed spacer (ITS) region has been widely used as the primary fungal barcode [31] for sequence-based identification of fungi.

The majority of WDF are considered to have a broad range of hosts, but some species show close affinity and preference for specific host trees [32,33]. Investigating host preference is helpful in understanding the ecology of a particular area, because WDF contribute to the nutrient cycle by breaking down the host tree. The main objective of this study was to survey WDF in Central Siberia involved in the degradation of host tree species and identify them to species, based on their morphology and ITS sequence analysis. To investigate the major wood decayers in this area, we evaluated the WDF species lists from previous literature, along with the data from this study.

2. Materials and Methods

2.1. Sampling Sites and Determining the Major Wood-Decaying Fungi

Wood-decay fungi were collected in Central Siberia: three sites near Krasnoyarsk in September 2014, and four sites at the Republic of Khakassia in August 2017 (Figure 1). A total of 106 fruiting bodies of WDF were sampled from six plant species: *Abies sibirica* (29 fruiting bodies), *Betula pendula* (59), *Larix siberia* (6), *Pinus sylvestris* (5), *Populus tremula* (3), and *Salix alba* var. *sericea* (4). All samples were dried in air-vented ovens at 55 °C for 3–4 days, then deposited at the Seoul National University Fungus Collection (SFC).

To investigate the major wood-decaying fungi in Central Siberia, we combined our data and the WDF lists from previous literature [15,22,23]. We investigated whether WDF species are host tree specific by categorizing species at the genus level of the host trees, focusing on the six main genera (*Abies, Betula, Larix, Pinus, Populus,* and *Salix*).



Figure 1. Location of the sampling sites in Central Siberia: open circles and squares - this study, closed circles and squares - previous literature. •Central Siberia (Kotiranta and Shiryaev, 2016); ▲Tuva Republic, Southern Siberia (Kotiranta et al., 2016); ■Yenisein River basin, East Siberia (Shiryaev and Kotiranta, 2016); ○Krasnoyarsk, Central Siberia; □Republic of Khakassia.

2.2. Morphological and Molecular Approaches to WDF Identification

The specimens were initially identified based on their macro- and micromorphological characters. For the microscopic structure observation, dried tissues were rehydrated in 3% (w/v) KOH, stained in 1% (w/v) phloxine and Melzer's reagent (IKI), and then observed using a Nikon Eclipse 80i optical microscope (Nikon, Tokyo, Japan). Fungal nomenclature was based on the current information on Index Fungorum [34] to reflect the legitimate name.

Genomic DNA was extracted from the inner tissues of the fruiting body using a modified CTAB extraction protocol [35]. The ITS region was amplified using the primer set ITS1F/ITS4 [36]. PCR was performed in a C1000 thermal cycler (Bio-Rad, Richmond, CA, USA) using the Maxime PCR PreMix-StarTaq (Intron Biotechnology Inc., Seoul, Korea). The PCR conditions were slightly modified from the previously described method [37]: 95 °C for 5 min, followed by 35 cycles of 95 °C for 40 s, 55 °C for 40 s, and 72 °C for 1 min, and a final extension step at 72 °C for 10 min. The PCR products were electrophoresed on 1% agarose gel and purified for sequencing using the ExpinTM PCR Purification Kit (GeneAll Biotechnology, Seoul, Korea), according to the manufacturer's instructions. DNA sequencing was performed in both forward and reverse directions using the PCR primers at Macrogen (Seoul, Korea), using an ABI Prism 3700 genetic analyzer (Life Technologies, Gaithersburg, MD). Sequences were assembled and proofread using MEGA 6 [38]. All consensus sequences were matched with a reference sequence using BLAST in the NCBI GenBank database.

3. Results

3.1. WDF Identification

WDF with obvious and distinct morphological characters were identified on the spot during the collection. In deciduous forests, identification of the following species was possible: *Cerrena unicolor*, *Daedaleopsis tricolor*, *Fomes fomentarius*, *Fomitopsis betulina*, *Fomitopsis pinicola*, *Ganoderma applanatum*, *Innotus obliquus*, *Irpex lacteus*, *Phlebia tremellosa*, *Plicaturopsis crispa*, *Stereum subtomentosum*, *Trametes hirsuta*, *Trametes versicolor*, and *Trichaptum biforme*. Species often found and identified in coniferous forests were as follows: *Fomitopsis pinicola*, *Hymenochaete cruenta*, *Rhodofomes cajanderi*, and *Trichaptum abietinum*. The remaining specimens could not be identified to the species-level based only on their macro- and micromorphologies, so they were identified based on the sequence analysis of the ITS region.

Based on the sequence analysis, the 106 WDF samples were identified to 52 species (Table 1). All BLAST searches, with two exceptions, showed over 98% similarity to the respective reference species. The two exceptions are as follows: SFC20170809-03 matched to *Ph. igniarius* with 96% similarity, and SFC20140922-07 matched to *Postia* sp. 3 with 96.7% similarity. Although 96% similarity is relatively low to distinguish a fungus at the species level, microscopic features of SFC20170809-03 corresponded to *Ph. igniarius*, the species identified in BLAST. Specimen SFC20140922-07 remained identified as *Postia* sp. due to an inconclusive morphological discrimination. Of the 52 species, 27 species were previously unrecorded from Central Siberia.

3.2. Host Preference

In our research area, *Abies sibirica* and *Betula pendula* were the dominant host species, and most WDF were collected from these two species. Eighteen WDF species were collected from *A. sibirica* and 26 species were collected from *B. pendula*, while only two to four species were identified from each of the other four host plants (Table 1). A total of 23 WDF species were found on gymnosperms, with *Fomitopsis pinicola*, *Phaeolus schweinitzii*, and *Trichaptum abietinum* being the most commonly found species. *F. pinicola* and *Pha. schweinitzii* commonly occurred on both *A. sibirica* and *Pin. sylvestris*. In contrast, 30 WDF species were found on angiosperms. The dominant species collected were *Fomes fomentarius*, *Fomitopsis betulinus*, *Ganoderma applanatum*, *Inonotus obliquus*, *Trametes versicolor*, and *Trichaptum biforme*. Major species found on each host genus are represented in Figure 2.

To evaluate the host specificity, the records of WDF and their corresponding hosts from the previous literature (271 species), and the data from this study (52 species), were analyzed together (Figure 3a). A total of 130 WDF specifically grew on angiosperms, 143 on gymnosperms, and 20 grew on both (Figure 3b). There were no WDF species detected across all host plant species. Host preference of WDF was stronger on gymnosperms than on angiosperm trees; not many WDF found on a coniferous tree were found on other host genera. *Fomitopsis pinicola* and *Trichaptum abietinum* were the only species commonly found across all three genera of the gymnosperm (Figure 3c). On the other hand, a higher number of WDF species were found on two or more hosts among the deciduous tree genera. The following 11 WDF were confirmed to be involved in the decomposition of all angiosperms: *Bjerkandera adusta, Cerrena unicolor, Fomes fomentarius, Ganoderma applanatum, Gloeoporus dichrous, Irex lacteus, Polyporus leptocephalus, Schizophyllum commune, Scopuloides rimosa, Trametes hirsuta, and Tra. versicolor (Figure 3d). The list of fungal species for each host plant is provided (See Supplementary Table S1).*

Fungal Species	Kr1	Kr2	Kr3	Kh1	Kh2	Kh3	Kh4	Specimen No. (Acc No.)	Closest Match (Accession Number)	Similarity (%)
Antrodia gossypium (=Fibroporia gossypium)						Ab	Ab	SFC20170811-08 (MT044421)	Fibroporia gossypium (HM590880)	100
Antrodia hingganensis							Ab	SFC20170811-04 (MT044428)	Antrodia hingganensis (KC595893)	99.8
Antrodia xantha							Ab	SFC20170811-17 (MT044427)	<i>Antrodia xantha</i> strain (DQ491424)	100
Botryobasidium subcoronatum			Be		Be	Be		SFC20170809-07 (MT044390)	Botryobasidium subcoronatum (AJ389790)	100
Cerrena unicolor			Be			Be		SFC20140920-13 (MT044429)	Cerrena unicolor (KP135305)	99.8
Climacodon pulcherrimus						Be	Be	SFC20170810-20 (MT044416)	Climacodon pulcherrimus (KY948784)	99.8
Coniophora puteana		Ab			Ab			SFC20170809-26 (MT044400)	Coniophora puteana (AM293074)	99.6
Daedaleopsis tricolor			Be		Be		Be	SFC20140920-08 (MT044430)	Daedaleopsis tricolor (MG696213)	100
Fomes fomentarius			Be		Be	Be	Be	SFC20170809-15 (MT044396)	Fomes fomentarius (EF155499)	100
Fomitiporia punctata			Sa		Sa			SFC20140922-20 (MT044431)	Fomitiporia punctata (KX639630)	99.6
Fomitopsis betulina (=Piptoporus betulinus)			Be		Be	Be	Be	SFC20170809-09 (MT044392)	Piptoporus betulinus (JX109856)	99
Fomitopsis pinicola	Ab		Pi		Be		Ab	SFC20140919-02 (MT044432)	Fomitopsis pinicola (KX524505)	100
Ganoderma applanatum	Ро			Ро	Be		Be	SFC20170809-27 (MT044401)	Ganoderma applanatum (KY364256)	99.6
Gloeocystidiellum convolvens (=Gloeopeniophorella convolvens)			Ве			Ве		SFC20140920-19 (MT044433)	Gloeopeniophorella convolvens (KY848506)	99.8
Gloiothele sp.					Ab		Ab	SFC20170811-07 (MT044420)	Gloiothele sp. (KJ713991)	99.6
Hapalopilus rutilans	Ab				Ab		Ab	SFC20170811-05 (MT044419)	Hapalopilus rutilans (KX752625)	98.9
Hericium coralloides						Be		SFC20170810-04 (MT044405)	Hericium coralloides (MG735348)	99.8

Table 1. List of wood-decay fungi collected from Krasnoyarsk and the Republic of Khakassia, Russia.

Fungal Species	Kr1	Kr2	Kr3	Kh1	Kh2	Kh3	Kh4	Specimen No. (Acc No.)	Closest Match (Accession Number)	Similarity (%)
Heterobasidion annosum			Ab					SFC20140922-17 (MT044434)	Heterobasidion annosum (MH859050)	100
Hymenochaete cruenta					Ab		Ab	SFC20170811-12 (MT044423)	Hymenochaete cruenta (JQ279595)	100
Hypsizygus marmoreus				Ро				SFC20170809-28 (MT044402)	Hypsizygus tessulatus (KP192620)	100
Inonotus leporinus	Ab							SFC20140919-04 (MT044435)	Inonotus leporinus voucher (FJ775542)	99.5
Inonotus obliquus			Be		Be	Be		SFC20140922-21 (MT044436)	Inonotus obliquus (KP004970)	100
Irpex lacteus		Sa				Be		SFC20170810-15 (MT044413)	Irpex lacteus strain (JX311924)	100
Kuehneromyces mutabilis					Ab			SFC20170809-25 (MT044399)	Kuehneromyces mutabilis (AY354218)	100
Laetiporus montanus					La			SFC20170809-01 (MT044387)	Laetiporus montanus (KX354466)	99.8
Neolentinus lepideus						Be		SFC20170810-14 (MT044412)	Neolentinus lepideus (HM536098.1)	99.5
Onnia tomentosa						La		SFC20170810-01 (MT044403)	Onnia tomentosa (KF996518)	99.9
Phaeolus schweinitzii	Ab					Pi		SFC20170810-18 (MT044415)	Phaeolus schweinitzii (FR686570)	99.6
Phellinus igniarius					Sa			SFC20170809-03 (MT044388)	Phellinus igniarius (GQ383711)	96
Phellinus laricis =Porodaedalea laricis)						La	Ab	SFC20170810-07 (MT044408)	Porodaedalea laricis (FJ775569)	98.9
Phellinus cinereus							Be	SFC20170811-16 (MT044426)	Phellinus cinereus (AY340047)	100
Phellinus laevigatus			Be		Be	Be		SFC20170809-14 (MT044395)	Phellinus laevigatus (AY340053)	100
Phellinus lundellii			Be		Be		Be	SFC20170811-03 (MT044418)	Phellinus lundellii (AY340058)	100
Phlebia tremellosa			Be		Be			SFC20170809-08 (MT044391)	Phlebia tremellosa (LN611126)	99.3
Pleurotus ostreatus (=Pleurotus pulmonarius)			Be		Be	Be		SFC20170810-10 (MT044410)	Pleurotus pulmonarius (KP867918)	100

Table 1. Cont.

Fungal Species	Kr1	Kr2	Kr3	Kh1	Kh2	Kh3	Kh4	Specimen No. (Acc No.)	Closest Match (Accession Number)	Similarity (%)
Plicaturopsis crispa					Ве		Be	SFC20170811-10 (MT044422)	Plicaturopsis crispa (DQ494686)	99
Postia balsamea						La		SFC20170810-11 (MT044411)	Postia balsamea (JF950570)	99.1
Postia caesiosimulans							Ab	SFC20170811-14 (MT044424)	Postia caesiosimulans (MG137061)	99.2
Postia fragilis					Ab			SFC20170809-23 (MT044398)	Postia fragilis (JF950573)	99.6
Postia lateritia							Ab	SFC20170811-15 (MT044425)	Postia lateritia (JF950566)	97.6
Postia sp.			Pi					SFC20140922-07 (MT044437)	<i>Postia</i> sp. (KJ668468)	96.7
Rhodofomes cajanderi (=Fomitopsis cajanderi)					La	La		SFC20170810-06 (MT044407)	Fomitopsis cajanderi (DQ491413)	100
Stereum hirsutum			Be		Be			SFC20170809-10 (MT044393)	Stereum hirsutum (AY854063)	99.8
Stereum subtomentosum			Be			Be	Be	SFC20140920-21 (MT044438)	Stereum subtomentosum (KR673461)	98.5
Trametes hirsuta					Ве	Be		SFC20170810-16 (MT044414)	Trametes hirsuta (KF573009)	99.6
Trametes pubescens						Be		SFC20170810-09 (MT044409)	Trametes pubescens (JN164960)	99.3
Trametes versicolor			Be		Be			SFC20170809-05 (MT044389)	Trametes versicolor (JN164965)	99.7
Trichaptum abietinum	Ab	Pi	Pi		Ab	Ab	Ab	SFC20170809-22 (MT044397)	Trichaptum abietinum (AY781273)	99.8
Trichaptum biforme			Be		Be	Be		SFC20170810-02 (MT044404)	Trichaptum biforme (MH245095)	99.8
Vitreoporus dichrous (=Gloeoporus dichrous)					Ве	Be		SFC20170810-21 (MT044417)	Gloeoporus dichrous (MG572751)	100
Xylodon flaviporus						Be		SFC20170810-05 (MT044406)	Xylodon flaviporus (AF145585)	100
Xylodon radula					Ab		Ab	SFC20170809-11 (MT044394)	Xylodon radula (KP814413)	99.5
The number of samples	6	3	20	2	29	25	21	(

Table 1. Cont.



Figure 2. The major species found on each host genus in Central Siberia. a–c (Abies sibirica):
(a) Antrodia gossypium, (b) Fomitopsis pinicola, (c) Hymenochaete cruenta; d,e (Larix siberia): (d) Laetiporus montanus, (e) Rhodofomes cajanderi; f–h (Pinus sylvestris): (f) Fomitopsis pinicola, (g) Phaeolus schweinitzii, (h) Trichaptum abietinum; i–l (Betula pendula): (i) Fomitopsis betulina, (j) Inonotus obliquus, (k) Phellinus laevigatus, (l) Phellinus lundellii; m,n (Populus tremula): (m) Ganderma applanatum, (n) Hypsizygus marmoreus; o,p (Salix alba var. sericea): (o) Fomitiporia punctate, (p) Phellinus igniarius.

4. Discussion

We identified 52 WDF species from six main host genera (*Abies, Betula, Larix, Pinus, Populus,* and *Salix*) located in Central Siberia [22,39]. Despite the large number of WDF reported from Central Siberia, very few have available sequence data. As the majority of WDF are macrofungi, many can be identified based on their morphological characters. However, some macrofungi can be difficult to identify when they are collected in an immature state or do not have distinguishing morphological characteristics. In addition, depending on the environment, the shape of many macrofungi within the same species can be different [37,40]. Thus, sequence-based identification can be an alternative approach for distinguishing morphologically similar species and reducing the rate of misidentification.

The community of WDF species collected from Central Siberia was largely consistent with those from all around the world. The reference ITS sequences used for identification were of specimens from Europe, North America, and East Asia (South Korea, China, and Japan), and a limited number from Russia. Considering the number of WDFs reported from Central Siberia, sequence information available from the public database is low (Table S1). A total of 25 WDF species matched with species that were described in previous records with only morphological characters [15,22,23], while 27 WDF species were identified as new records to Central Siberia. However, when looking at the identified species in several genera, our results differed from previous studies. Such discrepancy may be true differences based on different sampling sites or artifacts of misidentification. For example, *Phellinus*

species are difficult to identify morphologically to the species level, and sequence data can be useful to confirm their identity to determine why there is a discrepancy. In addition, the provision of sequence data from this area is necessary to study the population genetics and phylogeography of these species.



Figure 3. The number of wood-decay fungi (WDF) associated with different host tree species in Central Siberia. (**a**) Number of WDF species associated with six major host tree genera, combining data from this study with previous studies. (**b**) Venn diagram illustrating the number of WDF occurring on angiosperm and gymnosperm. (**c**) Venn diagram showing the number of WDF found on main host trees of gymnosperm (*Abies* spp., *Larix* spp., *Pinus* spp.). (**d**) Venn diagram showing the number of WDF found on main host trees of angiosperm (*Betula* spp., *Populus* spp.).

There have been many studies focusing on host preference or host specialization of WDF to evaluate the distribution and the ecological impacts of plant-related fungi [41,42]. Since fungal host preference can affect spreading and population dynamics [33,43,44], understanding this aspect is important for studying the biological diversity of an ecosystem. Generally, WDF prefer either gymnosperm or angiosperm hosts [45], and this phenomenon has also been observed in Central Siberia. Brown-rot fungi were mainly found on gymnosperms, while most white-rot fungi were found on angiosperm, in accordance with previous studies [42,46]. Brown-rot fungi have an apparent preference for gymnosperm substrates and favor colder climates for survival and propagation [2]. Correspondingly, members of Antrodia, Gloeophyllum, and Postia likely play pivotal roles in degrading the Central Siberian coniferous forests. Heterobasidion annosum and Phaeolus schweinitzii are two well-known conifer pathogens [47,48] that might threaten the coniferous forests of Central Siberia. However, many other species found in this area are also known worldwide to cause a decay in gymnosperms [49–52], including Dichomitus squalens, Stereum sanguinolentum, Trichaptum abietinum, Trichaptum fuscoviolaceum, Tubulicrinis calothrix, and Tubulicrinis subulatus (Table 1; Table S1). For white-rot fungi, several species were exclusively found on angiosperms: Bjerkandera adusta, Cerrena unicolar, Fomes fomentarius, Irpex lacteus, Trametes hirsuta, and Tra. versicolor. These species are known to inhabit angiosperm branches and trunks [49,50].

Although *Trichaptum abietinum* is known as a powerful white-rotter, it was detected mostly on gymnosperms in this study (Table 1). This species overwhelmingly preferred gymnosperms and acted as a pine indicator species, growing on *Picea*, *Pinus*, and *Abies* [53]. In the case of *Rhodofomes cajanderi*,

as reported to occur commonly on dead coniferous wood [54], it was frequently observed growing on a conifer, *Larix siberia*. However, for *Hapalopilus rutilans*, known to grow on most deciduous woods [55], it instead preferred to the host genus *Abies* in Siberia.

Three species frequently found in this survey (*Fomitopsis betulina*, *Inonotus obliquus*, and *Phellinus laevigatus*; Figure 2i–l) exhibited strong specialization on the host genus *Betula*. In support of this observation, the same results were found in previous studies in Central Siberia (Table S1). The generic name of *Fomitopsis betulina* was recently transferred from *Piptoporus betulinus*, based on its morphological characters and multi-gene analyses [56]. Tsuneda and Kennedy [57] confirmed that *Piptoporus betulinus* (current name *Fomitopsis betulina*) occurred only on *Betula* species as a typical host-specific fungus. For *Fomes fomentarius* and *Stereum subtomentosum*, which have been reported to occur on various hardwoods [51,52], *Betula* seems to be preferred in Central Siberia.

Fomitiporia punctate and *Phellinus igniarius* (Figure 20,p) were frequently found on *Salix* in this region. *Fomitiporia punctate*, known as a harmful plant pathogen in regions growing Mediterranean vine, has been recorded as parasites of *Salix*, *Sorbus*, and *Vitis* in Europe and the USA [58]. *Phellinus igniarius* is also known to grow on *Salix* in Europe [59]. Apart from sharing the same host, *Fomitiporia punctate* and *Phellinus laevigatus* also have superficially similar morphology. The former, however, does not have setae and strongly dextrinoid and cyanophilous basidiospores [60], which distinguishes it from the latter.

In conclusion, WDF have been widely studied to monitor and assess biodiversity in forests [61]. It is known that a fruiting body of WDF is a better indicator compared to a non-fruiting mycelia, in that it reflects population persistence as a life-history stage [32]. Thus, our DNA-based identification of host preference of WDF in Central Siberia provides a good basis to further study the spatial distribution and the evolutionary process of WDF in Europe and Asia, and to understand the nutrient cycle in ecosystems. With that, a persistent long-term ecological investigation and survey of WDF in Central Siberia based on molecular data is necessary.

Supplementary Materials: The following are available online at http://www.mdpi.com/2071-1050/12/6/2535/s1, Table S1: The list of WDF from six main host tree genera in previous literature and current study.

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