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Phylogenetic analysis of some Hymenochaetaceae members

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Abstract: *The Hymenochaetaceae family of Phylum Basidiomycotina is characterized by 28 genera and 400 species to date. Phylogenetic analysis of various members of this family was attempted using morphological and molecular traits. Thirty samples belonging to Hymenochaetaceae were collected from Pune region. Morphological studies were carried out using 14 quantitative and 70 qualitative characters. Out of these, 8 quantitative characters of the 30 samples along with the same characters from previously reported genera of this family were used in Principle Component Analysis (PCA) in an attempt to group the samples in different clusters. In addition DNA sequence data corresponding to 80S ribosomal large subunit rRNA (28S) and 70S (mitochondrial) ribosomal small subunit rRNA (16S) was collected for the same 30 samples. Sequence data for the same rRNA molecules was also downloaded from NCBI site for 12 reported genera of Hymenochataceae. The sequences were aligned using MEGA (version 4) software and a phylogenetic tree was constructed using maximum parsimony (PAUP 4.0). Results indicated that clustering achieved using DNA sequence data differed from that using morphological data. Probable causes for this discrepancy could be that the evolution rate at phenotypic and molecular levels may differ and / or that the phenotype is determined by environmental factors in addition to genetic factors.*

Keywords: *Hymenochaetaceae, Phylogenetic analysis*

I. Introduction

Family Hymenochaetaceae forms a cosmopolitan group of wood inhabiting fungi that metabolize lignin for their growth and reproduction. It consists of 28 genera and 400 species exhibiting saprophytic and parasitic features. It is characterised by xanthochroic reaction, yellow to deep brown trama, generative hyphae without clamp connection and frequent occurrence of setae. The description of hymenochaetacean genera is based on characters pertaining to morphology, anatomy, sexuality, nuclear behaviour, pigmentation, and ecology and is rather imprecise. In this work we have attempted to identify 30 samples collected from Pune and Switzerland on the basis of information available on 12 genera of Hymenochaetaceae using DNA sequence data (rDNA) as well as morphological and anatomical data (1,2,3,4,5).

II. Material and methods

25 samples were collected from Pune region such as Pune University, Empress Garden, Papel Seminary (Nagar road), Katraj Ghat, NDA reserved forest, Taljai, Khadakwasla, Sinhgad, Lohagad, Mulshi, Dongarwadi, Lonavala, Bhimashankar, and road side old avenue trees in Pune city. 5 samples of Hymenochaetaceae members were obtained from Dr. Valérie Hofstetter, Agroscope Changins-Wädenswil Research station Institute, Switzerland. Morphological studies were carried out using 11 quantitative characters (Table 1) of the 30 collected samples and other genera of this family reported in literature were subjected to Principle Component Analysis (PCA) in an attempt to group the samples in different clusters. Cladistic analysis was also done using the same morphological characters using Pearson correlation and single-linkage distance method (MINITAB software). In addition, DNA sequence data corresponding to 80S ribosomal large subunit rRNA (28S) and 70S (mitochondrial) ribosomal small subunit rRNA (16S) was collected for the same 30 samples (6.7.8). Sequence data for the same rRNA molecules was also downloaded from NCBI site for 12 reported genera of Hymenochataceae. The sequences were aligned using ClustalW method (MEGA 4 software) and a phylogenetic tree was constructed using maximum parsimony (PAUP 4.0).

III. Results and Discussion

The PCA analysis did not lead to distinct clusters for the samples. However the Cladistic analysis of morphological data showed distinct grouping between temperate and tropical genera (Fig 1). All temperate samples showed similarity with the type specimens chosen for the analysis. Of the tropical samples, 15 formed a group with the *Phellinus* type species: *P. laevigatus*, *P. tremulae* and *P. lundelli* while 8 samples formed an individual cluster, which did not include any type genera.

The dendrogram generated using 80S ribosomal large subunit (LSU) rRNA (28S) region showed 10 distinct clusters out of which 9 clusters included at least one type genus (Fig 2a). The dendrogram generated using 70S (mitochondrial) ribosomal small subunit (mitSSU) rRNA (16S) showed 8 clusters which included the type genera (Fig 2b). *Fomitopsis* and *Coltricia* formed outgroups. None of the samples clustered with *Hymenochaetae*, *Coltricia*, *Onnia* and *Porodaedalea*. The tropical samples showed similarity to either *Phellinus*, *Inonotus*, and *Fulvifomes* and the clustering based on morphological and molecular data differed (Table 2).

IV. Figures and Tables

Table 1

Number	**Quantitative Characters**
1	Area of basidiocarp in square cm
2	no of pores / mm
3	Width of **Contexual** skeletal hyphae in µm
4	Width of **Hymenial** skeletal hyphae in µm
5	mean length of basidiospore in µm
6	mean breadth of basidiospore in µm
7	context layer height in mm
8	margin width in mm
9	no. of strata
10	length of pore tube in mm
11	length of setae in µm

Table 2

Number	**Genus in Hymenochaetaceae**	**Distribution of samples no.**		**Geographical location**
		Molecular	Morphological	
1	Fomitopsis	173	173	Switzerland
2	Fuscoporia	58	58	Switzerland
		21	-	Pune
3	Fomitiporia	62	62	Switzerland
4	Coltricia	-	-	-
5	Inonotus	49		Pune
		50		Pune
		51	51	Pune
		53	53	Pune
			59	Switzerland
6	Phylloporia	59	-	Switzerland
7	Phellinus igniarius	55	55	Switzerland
8	Phellinus	-	2, 7, 8, 10, 11, 12, 17, 21, 26, 29, 36, 43, 45, 145, 153	Pune
9	Fulvifomes	1, 2, 11, 26, 28, 29, 36, 45, 145, 153		

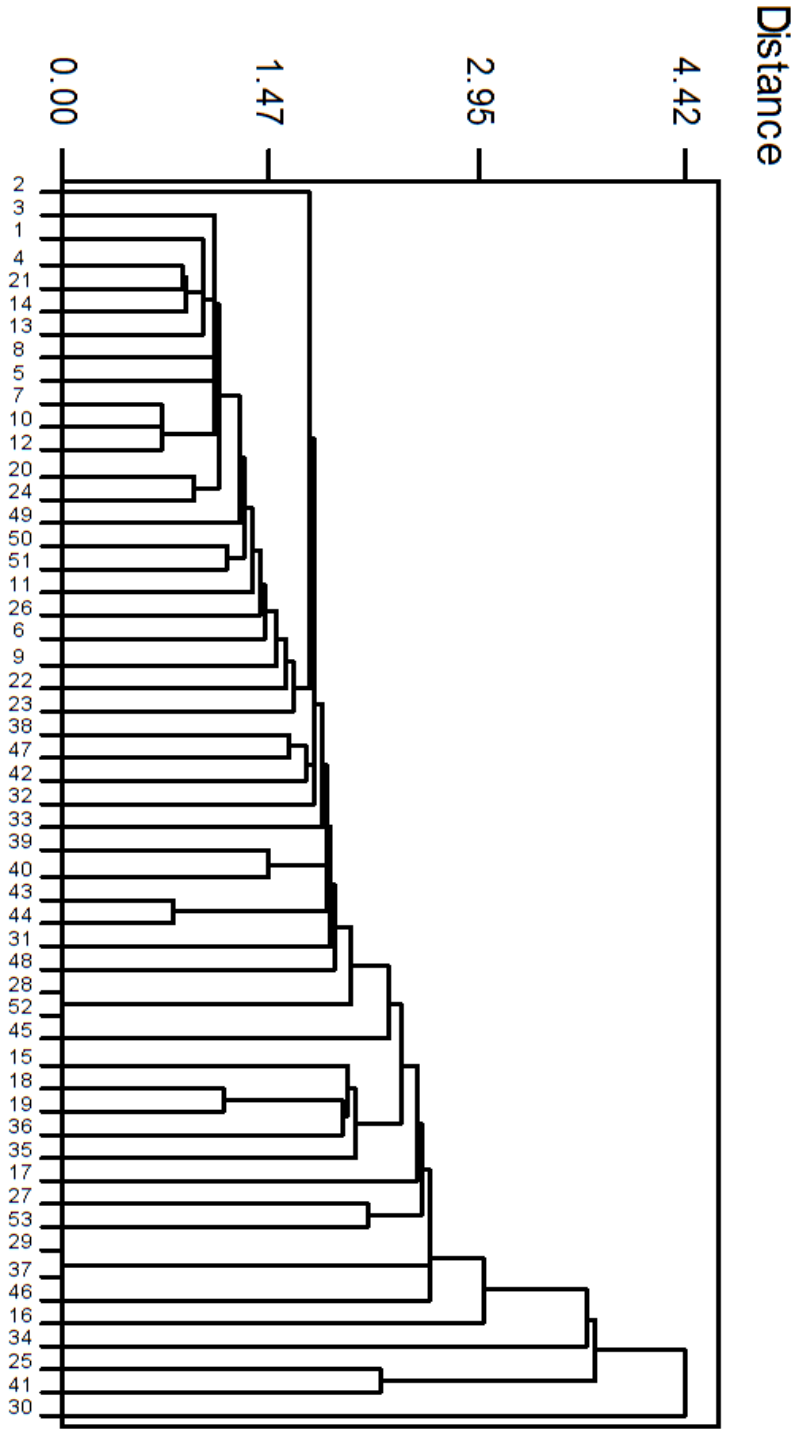

Fig 1. PCA analysis using Morphological character data

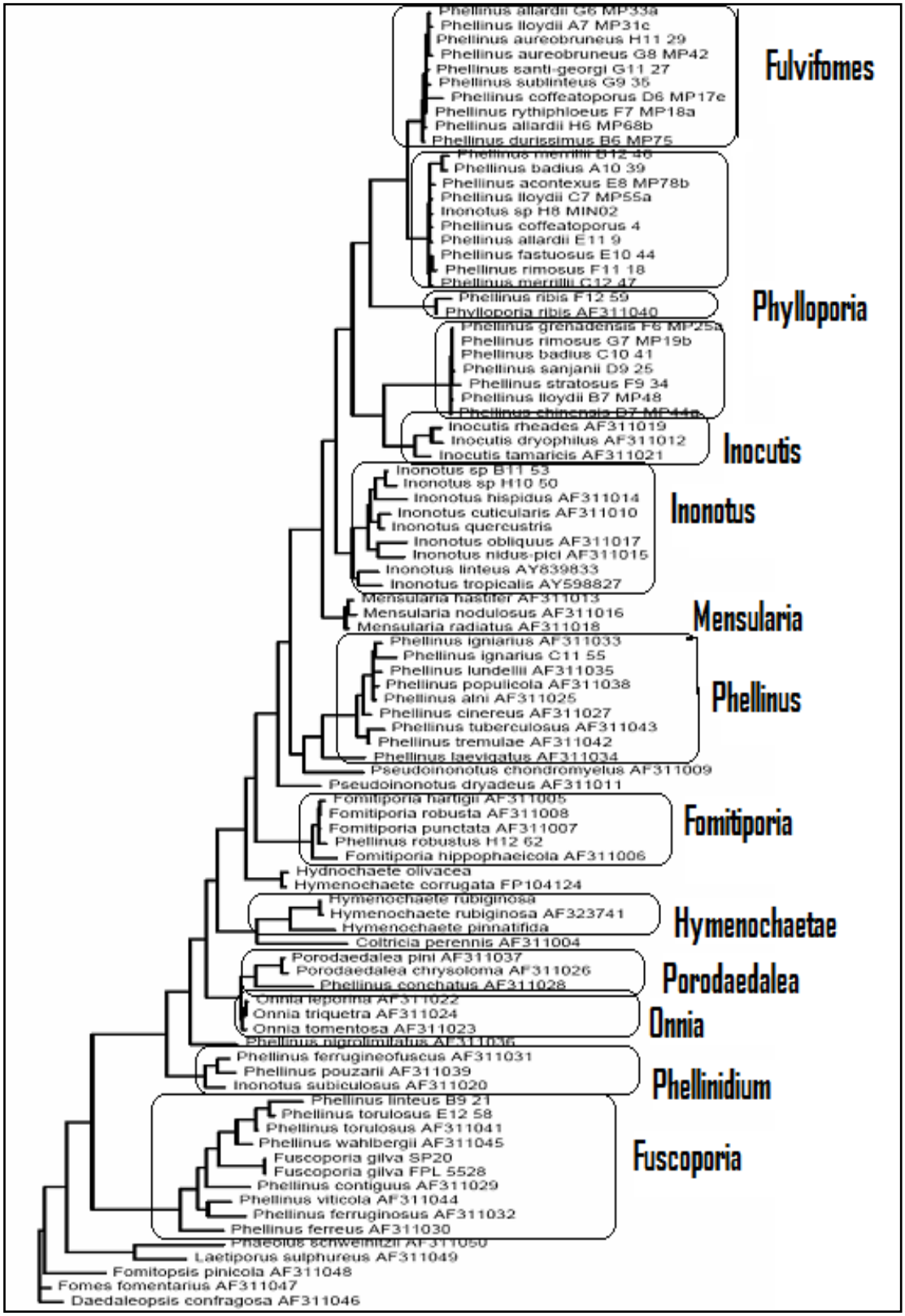

Fig 2a: Dendrogram generated using 80S ribosomal large subunit (LSU) rRNA (28S)

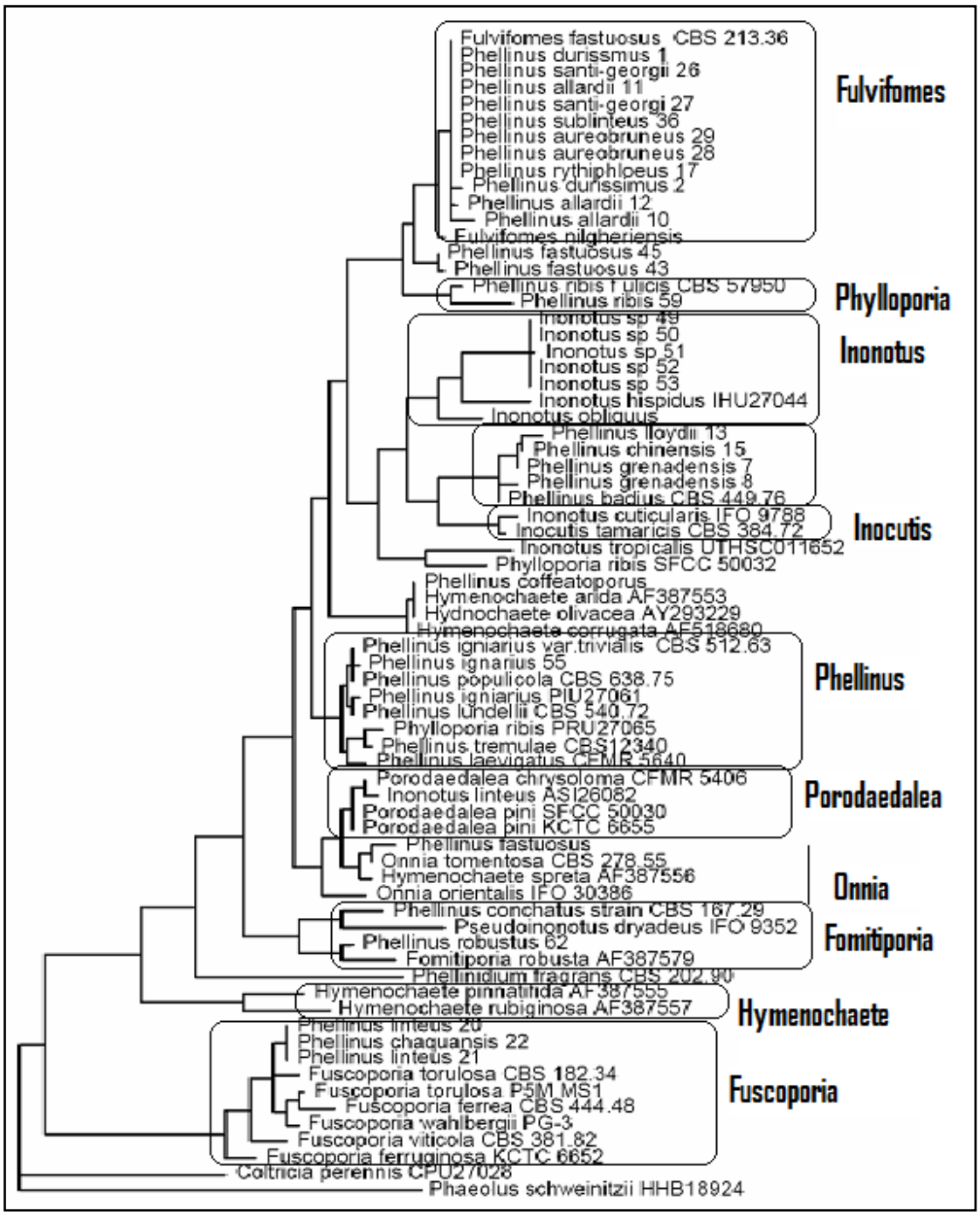

Fig 2b: Dendrogram generated using 70S (mitochondrial) ribosomal small subunit (mitSSU) rRNA (16S)

V. Conclusion (11 Bold)

Results indicated that clustering achieved using DNA sequence data differed from that using morphological data. Fulvifomes was classified as a sub-genus of Phellinus based on morphology, anatomy, pigmentation, and karyology (9-14). Later they were identified as two distinct genera on the basis of molecular data as well as the presence / absence of setae (13). Our data showed a distribution of 15 samples under Phellinus on the basis of morphological data, while molecular data showed the distribution of 10 of these same samples under Fulvifomes. Discrepancy in the conclusions drawn from morphological and molecular data could have arisen due to different rates of evolution at phenotypic and molecular levels and / or the role of environmental factors in determining the phenotype besides genetic factors.

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