



#### **Conference** Paper

# Lichen-like Symbiotic Associations of Wood-decaying Fungi and Algae. I. Biodiversity and Ecology of Photobionts

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#### Abstract

The article presents new data on the taxonomical, morphological and ecological composition and species diversity of symbiont algae associated with xylotrophic fungi. The largest part of symbionts (86%) are eukaryotic algae belonging to the divisions *Chlorophyta* (68% of total number of species), *Ochrophyta* (9%) and *Charophyta* (8%). The prokaryotic algae, or *Cyanoprokaryota*, make up the remaining 14% of species. The eukaryotic algae are an obligatory component of mycetobiont communities, whereas *Cyanoprokaryota* are the optional, facultative part. Out of 46 mycetobiont algae genera, 29 (or 64%) are single-species taxa, while 15 (32%) genera include two or three species. Two genera – *Chlamydomonas* and *Klebsormidium* – are represented by 6 and 4 species, respectively. The majority of mycetobiont algae have coccoid (41%) and trichal (33%) thalli, colonial-coccoid (18%) and monadic (8%) algae are rarely observed. All algae species belong to widespread epiphytic, soil and lichenophilic groups that do not require symbiosis with fungi. Obligatory mycetobionts were not observed during the study. Communities of mycetobiont algae have host-specificity and high geographical and individual variability.

**Keywords:** wood-decaying fungi, algae and *Cyanoprokaryota*, biodiversity, ecology, symbiosis

#### 1. Introduction

The widely observed presence of algae in basidiocarps of xylotrophic basidiomycetes remains poorly studied. Only a few articles address the issue. In one of the first related studies, Burdsall and Volk [1] reported two species of one-celled algae observed in basidiocarps of *Oxyporus nobilissimus* W. B. Cooke. In articles by Zavada, Simoes [2]

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and Zavada et al. [3], 4 species of green algae (Hormidium sp., Stichococcus bacillaris Nägeli, Chlorococcum sp. and Trebouxia sp.) were found in basidiocarps of Trametes versicolor (L.) Lloyd. In another work by Videv et al. [4] reported 10 species: 9 belonged to green algae and one, Klebsormidium dissectum (F. Gray) Y. Ettl. et Gärtner, to Charophyta. Stoyneva et al. [5] described the growth of 4 green algae species, Desmococcus vulgaris, Trebouxia arboricola, Stichococcus bacillaris and S. minutus, in basidiocarps of Fomes fomentarius (L.) Fr. In total, 16 taxons of algae are found in basidiocarps of xylotrophic fungi, according to mentioned articles. Their interaction type with the fungi remains under a question. Some researchers [4, 5] say that algae developing on basidiocarps cannot be viewed as symbionts but should be rather seen as epiphytic, epimicotic organisms. However, there is convincing evidence of symbiosis between algae and *T. versicolor*, referred to as 'possible demi-lichenization of the basidiocarps' [2, 3], an interaction which is similar to those that mycobionts and photobionts have in lichens. We think that the relationship between mycetobiont algae and fungi should be characterized as 'associative symbiosis', in which the algae receives protection from excessive insolation and CO<sub>2</sub> and H<sub>2</sub>O from respiration and, in return, the fungus gets an additional source of carbon and nitrogen nutrition to wooden substrate [6]. In Zavada et al.'s [3] article and our own works, it has been experimentally proven that products of algal photosynthesis are found in fungi [7, 8] and that the fixation of molecular nitrogen happens when prokaryotic blue-green algae (Cyanoprokaryota) are present in basidiocarps [9].

Therefore, associations between wood-destroying fungi and algae require the most careful study, as this symbiosis has unique and significant importance for a biosphere. The biodiversity and ecology of symbiotic algae in such associations should be given special attention.

### 2. Methods

The objects of research mycetobiont algae in basidiocarps of wood-destroying fungi, were collected from four locations: (a) pine-birch pre-forest-steppe ( $56^{\circ}36'5''$  N,  $61^{\circ}3'24''$  E), (b) dark coniferous forests of southern taiga, the Central Urals ( $57^{\circ}21'01''$  N,  $58^{\circ}41'54''$  E), (c) the Valdai Upland ( $57^{\circ}57.76'$  N,  $33^{\circ}20.34'$  E) and (d) taiga forests of the Komi Republic ( $61^{\circ}34'$  N,  $50^{\circ}36'$  E). Before microscopic examination of the algae, the basidiocarps were moistened with distillate for two days. After we divided them into two parts, each got a different treatment for 7 days: one was moistened with 3N BBM nutrient medium for green algae, while a Bg11 nutrient medium for *Cyanoprokaryota* 

was applied to the other. The algae (surface abrading probes and sections of a basidiocarp) were examined under a Nikon Eclipse 80i microscope (magnification x400, x1000). For better identification of algal species diversity, we transferred fragments of basidiocarps and washings from their surface into agar and liquid nutrient media cultures: 3N BBM and Bg11 were used for green algae and *Cyanoprokaryota*, respectively. Algae were identified using Russian and foreign published literature and taxonomic keys [10–12]. The part of the species which requires observations during different stages of reproduction and development were determined only to the genus level.

### 3. Results

Our findings (Table 1) show that mycetobiont algae are found in the basidiocarps of many xylotrophic basidiomycetes, though they mostly inhabit annual or overwintering basidiocarps: very rarely, they inhabit perennial ones. Fungi with algae in basidiocarps have a diverse ecology: they decompose deciduous and coniferous wood, cause white and brown rot. The Cerrena unicolor and Trichaptum pargamenum species have the highest diversity of mycetobiont algae (31–34 species). The basidiocarps of Stereum subtomentosum, Trichaptum abietinum and T. fuscoviolaceum have 20-22 species of algae. 11 to 15 symbiont algae species are found in Stereum hirsutum, Trametes gibbosa, T. ochracea and T. pubescens. Most fungi have no more than 10 species of algae in their basidiocarps. The data analysis shows that eukariotic algae are obligatory components in basidiocarps of all fungi species, whereas Cyanoprokaryota serve as a small and additional (facultative) part. Eukaryotic algae mainly or exclusively belong to the chlorophyta division in most fungi species: only in one, Gloeophyllum sepiarium, are members from the group are absent (Table 1). As shown earlier by our research [6], the proportion of basidiocarps with algae varies from 30% to 90% between fungi species, indicating that such a relationship is not obligatory.

The taxonomical and morphological characteristics of mycetobiont algae are presented in Table 2. The largest number of species (64% or 86%) belong to eukaryotic algae from the *Chlorophyta* (51 species, 68%), *Ochrophyta* (7 species, 9%) and *Charophyta* (6 species, 8%) divisions: 15% of species belong to *Cyanoprokaryota*. From 46 genera of mycetobiont algae, 29 (or 64%) consist of one species, while 15 have two or three species. *Chlamydomonas* and *Klebsormidium* are the most diverse genus, with 6 and 4 species, respectively. The average species diversity within genus is 1.7.



TABLE 1: The list of xylotrophic fungi with mycetobiont algae in basidiocarps (total number of species/number of species belonging to *Chlorophyta/Charophyta/Ochrophyta/Cyanoprokaryota* divisions, respectively).

Xylotrophic Fungi	Mycetobiont Algae
Bjerkandera adusta (Willd.) P. Karst. (awd)*	9/5/0/1/3
Cerrena unicolor (Bull.) Murrill (awd)	31 / 23 /1 / 4 / 3
Chondrostereum purpureum (Pers.) Pouzar (awd)	7/7/0/0/0
Datronia mollis (Sommerf.) Donk (awd)	3/3/0/0/0
Echlerialla deglubens (Berk. & Br.) D.A. Reid (awd)	3/3/0/0/0
Fomes fomentarius (L.) Fr. (pwd)	2/2/0/0/0
Gloeophyllum sepiarium (Wulfen) P. Karst. (abc)	4/0/1/1/2
Lenzites betulina (L.) Fr. (awd)	8/7/0/0/1
<i>Onnia leporina</i> (Fr.) H. Jahn (awc)	4/3/0/0/1
Phellinus chrysoloma (Fr.) Donk (pwc)	4/3/0/0/1
Ph. igniarius (L.) Quél. (pwd)	2/2/0/0/0
Ph. tremulae (Bondartsev) Bondartsev & P. N. Borisov (pwd)	3/2/0/0/1
Steccherinum ochraceum (Pers.) Gray (awd)	7/7/0/0/0
Stereum hirsutum (Willd.) Pers. (awd)	11 / 10 / 1 / 0 / 0
S. subtomentosum Pouzar (awd)	21 / 17 / 2 / 1 / 1
Trametes gibbosa (Pers.) Fr. (awd)	13 / 12 / 0 / 0 / 1
T. hirsuta (Wulfen) Lloyd (awd)	7/7/0/0/0
<i>T. ochracea</i> (Pers.) Gilb. & Ryvarden (awd)	15 / 10 / 0 / 1 / 4
T. pubescens (Schumach.) Pilát (awd)	12/8/1/1/1
<i>T. trogii</i> Berk. (awd)	9/6/1/2/0
T. versicolor (L.) Lloyd (awd)	9/7/1/1/0
<i>Trichaptum abietinum</i> (Dicks.) Ryvarden (awc)	22 / 18 / 2 / 2 / 0
<i>T. fuscoviolaceum</i> (Ehrenb.) Ryvarden (awc)	20/ 18/ 1 / 0 / 1
<i>T. pargamenum</i> (Fr.) G. Cunn. (awd)	34 / 28 / 4 / 2 / 0
Source: Authors' own work.	
Note: * – annual basidiocarp (a), perennial (p); brown rot (b), wh deciduous (d) or coniferous (c) wood.	ite rot (w); decomposers of

Most algae have a coccoid (41%) and trichal (33%) thallus: colonial-coccoid (18%) and monadic (8%) forms are rare. All mycetobiont species are eurybiotic with a wide distribution: pedobionts and hydrobionts (*Chlamydomonas sp.*, etc.), epiphytes (*Desmococcus olivaceus*, etc.), aerophytes and photobionts of ascomycete lichens (*Pseudococcomyxa simplex, Stichococcus bacillaris*, etc.). There are no obligatory myce-tobiont algae, which indicates the facultative nature of the relationship between the algae and fungi.

Only a few mycetobiont algae have a low degree of selectivity and can be found in many fungi species. For example, the *Interfilum terricola*, *Pseudococcomyxa simplex* and *Stichococcus bacillaris* species occur in basidiocarps of all fungi from the *Trametes* 

Genus	Number Species, thallus type *	Genus	Number Species, thallus type
Chlorophyta		Chlorophyta	
Bracteacoccus	1, C	Tetracystis	2, CC
Chlamydomonas	6, m	Trebouxia	2, C
Chlorella	з, с	Ulothrix	2, t
Chlorococcum	1, C	Ochrophyta	
Chloroidium	2, C	Bumilleriopsis	1, t
Chlorosarcinopsis	1, CC	Characiopsis	2, C
Coenochloris	з, сс	Eustigmatos	1, C
Coenocystis	1, CC	Heterococcus	1, t
Desmococcus	1, CC	Tribonema	1, t
Dictyochloropsis	1, C	Vischeria	1, C
Diplosphaera	1, CC	Charophyta	
Elliptochloris	з, с	Cylindrocystis	1, C
Interfilum	2, t	Klebsormidium	4, t
Leptosira	2, t	Mesotaenium	1, C
Mychonastes	1, C	Cyanoprokaryota	
Мугтесіа	з, с	Anabaena	1, t
Neochlorosarcina	1, CC	Aphanocapsa	1, CC
Neocystis	1, CC	Calothrix	1, t
Neospongiococcum	з, с	Chroococcus	1, C
Parietochloris	1, C	Desmonostoc	1, t
Pseudococcomyxa	2, C	Hassallia	1, t
Scotiellopsis	1, C	Nostoc	3, t
Spongiococcum	1, CC	Phormidium	1, t
Sporotetras	1, CC	Scytonema	1, t
Stichococcus	2, t		

TABLE 2: Taxonomical and morphological characteristics of mycetobiont algae dwelling in basidiocarps of xylotrophic fungi.

Note: \* – coccoid (c), colonial-coccoid (cc), monadic (m), trichal (t).

genus (Table 3). However, the opposite group of algae, found only in 1 or 2 fungi species from a genus, is also observed. The members of this group are the green algae species *Diplosphaera chodatii, S. minor, Cyanoprokaryota – Hassallia byssoidea, Nostoc commune* and *Scytonema ocellatum*. Most likely, this group consists of both random and, possibly, highly selective species. In any case, they determine the specificity of mycetobiont algae communities [13]. This can be seen in the variation of mycetobiont algae richness between fungi species, particularly in the genus *Trametes* where the number of associated algae species varies from 7 to 15 species (Table 3).

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The species composition of mycetobiont algae varies between basidiocarps belonging to the same fungi species: in other words, high individual variability is present. For example, basidiocarps of *Trichaptum pargamenum* (collected at the same time and in the same place, the Komi Republic, the middle-taiga forests) could have from 5 to 10 species of algae, but only 2 – *Pseudococcomyxa simplex* and *Stichococcus bacillaris* – are present in all basidiocarps. Such species as *Dictyochloropsis* sp., *Interfilum terricola*, *Klebsormidium pseudostichococcus*, *Leptosira sp.*, *Trebouxia sp.* and *Vischeria helvetica*, are noted only in one of the basidiocarps. The majority of species (*Chlamydomonas sp.*, *Coenocystis oleifera* (Broady) Hindák, *Desmococcus olivaceus*, *Diplosphaera chodatii*, *Elliptochloris sp.*, *E. subsphaerica*, *Klebsormidium nitens* (Kützing) Lokhorst, *Myrmecia bisecta* Reisigl and *Sporotetras polydermatica*) occur in two or three basidiocarps of *Trichaptum pargamenum*. Therefore, each basidiocarp has its own particular algal composition – the Sörensen-Chekanovsky species similarity coefficient lies at the range from o.3 to o.6.

The geographic variability of mycetobiont algae composition is also observed [14]. For example, out of 16 species of algae found in the *Trichaptum pargamenum* basidiocarps from the middle taiga forests of the Komi Republic, only 6 occur in the basidiocarps of the same species in the Central Urals: *Chlamydomonas sp., Interfilum terricola, Desmococcus olivaceus, Pseudococcomyxa simplex, Stichococcus bacillaris* and *Trebouxia sp.* High individual and geographical variability, again, proves the non-obligatory connection of algae and fungus.

Mycetobiont algae	Species					
	1	2	3	4	5	6
Chlorophyta						
Chlamydomonas sp.			+	+	+	
<i>Chlorella vulgaris</i> f. <i>globosa</i> V. M. Andreyeva	+					
<i>Chloroidium saccharophilum</i> (W. Krüger) Darienko, Gustavs, Mudimu, Menendez, Schumann, Karsten, Friedl & Proschold	+	+	+			
<i>Coenochloris oleifera</i> (Broady) I. Kostikov, T. Darienko, A. Lukesová, & L. Hoffmann	+					
<i>Desmococcus olivaceus</i> (Persoon ex Acharius) J. R. Laundon	+	+		+		
Diplosphaera chodatii Bialosukniá	+				+	+
Elliptochloris reniformis (S. Watanabe) H. Ettl & G. Gärtner				+		+
E. subsphaerica (Reisigl) Ettl & Gärtner	+					

TABLE 3: Mycetobiont algae of the basidiocarps of the *Trametes* genus species: 1 – *T. gibbosa*, 2 – *T. hirsuta*, 3 – *T. ochracea*, 4 – *T. pubescens*, 5 – *T. trogii*, 6 – *T. versicolor*.

Mycetobiont algae	Species					
Interfilum terricola (J. B. Petersen) Mikhailyuk, Sluiman, Massalski, Mudimu, Demchenko, Friedl & Kondratyuk	+	+	+	+	+	+
Leptosira sp.			+		+	
<i>Mychonastes homosphaera</i> (Skuja) Kalina et Puncochárová				+		
<i>Myrmecia incisa</i> Reisigl				+		
<i>Myrmecia</i> sp.						+
Neochlorosarcina sp.	+					
Pseudococcomyxa simplex (Mainx) Fott	+	+	+	+	+	+
<i>Sporotetras polydermatica</i> (Kützing) I. Kostikov, T. Darienko, A. Lukesová, & L. Hoffmann		+	+	+		
Stichococcus bacillaris Nägeli	+	+	+	+	+	+
S. minor Nägeli	+		+			
Tetracystis macrostigmata Nakano			+			
Trebouxia sp.	+					+
Ulothrix variabilis Kützing			+			
Ochrophyta						
Bumilleriopsis sp.				+		
Characiopsis sp.			+			+
Eustigmatos magnus (J. B. Petersen) D. J. Hibberd					+	
Vischeria helvetica (Vischer & Pascher) D. J. Hibberd					+	
Charophyta						
Klebsormidium pseudostichococcus (Heering) H. Ettl & Gärtner				+	+	
<i>Mesotaenium</i> sp.						+
Cyanoprokaryota						
Desmonostoc muscorum (C. Agardh ex Bornet & Flahault) Hrouzek & Ventura	+					
Hassallia byssoidea Hassal ex Bornet et Flahault			+			
Nostoc cf. punctiforme (Kützing ex Hariot) Hariot			+			
N. commune Vaucher ex Bornet et Flahault			+			
Scytonema ocellatum Lyngbye ex Bornet & Flahault			+	+		
The number of species of algae in basidiocarps	13	7	15	12	9	9
Source: Authors' own work.						

## 4. Conclusion

Xylotrophic *Basidiomycetes* and mycetobiont algae inhabiting of their basidiocarps form multicomponent symbiotic associations consisting of a fungus (mycobiont) and several species of algae associated with it (photobionts). Mycobionts, as a rule, are fungi



with an annual, annual-wintering basidiocarps, and less often – perennial. The main, obligatory component of photobionts is green algae: other algal groups, such as *Ochrophyta*, *Charophyta* and *Cyanoprokaryota* are optional, not obligatory. The composition of mycetobiont algae in basidiocarps is not stable. It varies between different fungi species, the regions where where they grow and within individuals. Although the symbiotic relationship between fungi and mycetobiont algae is not obligatory, it is beneficial for both: the algae have some protection from the environment and receive additional H<sub>2</sub>O and CO<sub>2</sub> during their hosts' respiration, while the fungi get an additional source of carbon and nitrogen nutrition. In our opinion, the symbiotic associations of xylotrophic fungi and mycetobiont algae correspond to our notions of basidial lichens and should be considered as such.

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