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PROBLEMS IN THE IDENTIFICATION OF LICHEN PHOTOBIONTS

Probleme bei der Bestimmung von Flechtenalgen

by

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Key words: Lichen photobionts, identification, *Stichococcus, Trentepohlia, Trebouxia,* cultures.

Schlagwörter: Flechtenalgen, Bestimmungsprobleme, *Stichococcus, Trentepohlia, Trebouxia,* Kulturen.

Summary: An accurate identification of a lichen photobiont especially to the level of species should be undertaken by the study of unialgal cultures. Problems in the identification of *Stichococcus bacillaris* as the photobiont of *Chaenotheca furfuracea, Trentepohlia aurea* from *Gyalecta jenensis* and *Trebouxia* sp. as the photobiont of the lichen *Dactylina ramulosa* are discussed.

Zusammenfassung: Zur exakten Bestimmung von Flechtenalgen auf Artniveau sind Kulturen unumgänglich. Probleme bei der Bestimmung von Photobionten wie *Stichococcus bacillaris* aus *Chaenotheca furfuracea, Trentepohlia aurea* aus *Gyalecta jenensis* und *Trebouxia* sp. aus *Dactylina ramulosa* werden diskutiert.

Introduction

More than 60% of lichen photobionts belong to unicellular green algae, including so controversial genera like *Trebouxia* or *Coccomyxa.* Also several filamentous or sarcinoid taxa occur as single cells within the lichen thallus, e.g. *Trentepohlia, „Protococcus"* or *Leptosira.* In some cases an identification of photobionts in situ to its genus should be possible with simple squash preparation. But proper identification needs some type of culture, the methods well documented in literature. In this study the photobionts of *Chaenotheca furfuracea (Stichococcus), Gyalecta jenensis (Trentepohlia)* and *Dactylina ramulosa*

(*„Protococcus"*) were investigated.

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Earlier reports on the photobionts of the investigated lichens can be found in LYNGE (1933), RATHS (1938), TSCHERMAK (1941), TSCHERMAK-WOESS (1988), TIBELL (1978, 1980), POELT & VEZDA (1977, 1981).

Materials and Methods

Lichen specimens were collected from the following localities:

- o) *Chaenotheca furfuracea*: Sellrain valley near Innsbruck, among roots of Picea abies
- o) *Gyalecta jenensis*: Leutasch, Gaistal, surface of limestone rocks
- o) *Dactylina ramulosa*: Ötztal, Obergurgl, 2800m, soil.

Algal cells were observed with light microscope in situ from fresh material (squash preparation) and in cultures on BBM-agar plates under standard conditions (13° Celsius, cool-white fluorescent lamps (1300-3000 lux) on a 12/12 h L/D cycle) after 21 days of incubation. Also liquid medium was used, following ETTL & GÄRTNER (1995).

For EM microscopy cells were fixed:

a) in 3% glutaraldehyd in 0,1 m cacodylate buffer

b) in 1% aqueous O_sO_4 in 0,1 m cacodylate buffer,

dehydrated in alcohol/acetone and embedded in Spurr's resine; ultrathin sections were stained with uranyl acetate and lead citrate (REYNOLDS 1963).

Results and discussion

***Stichococcus bacillaris* NÄGELI as photobiont in *Chaenotheca furfuracea* (Fig. 1):**

As reported by TIBELL (1978), TSCHERMAK (1941) and other authors, the filamentous genus *Stichococcus (Charophyceae, Klebsormidiales)* is a common photobiont of coniocarpic lichens. This ubiquitous alga is well known as a member of the terrestrial algal flora. The taxonomy of the species is based mainly on the cell morphology and the development of filaments. *Stichococcus bacillaris* scarcely changes the cell morphology from lichen into culture, therefore the identification of characteristic criteria - cell morphology, lipid vacuoles at both poles of the cells and the pyrenoid - is simple. 5 varieties have been reported by RATHS (1938) and demonstrate the variability of *Stichococcus bacillaris* in nature. Especially the presence or absence of a pyrenoid has been discussed controversially. With JKJ solution the pattern of starch granules surrounding the pyrenoid can be determinded, but this is hardly visible in the light microscope. With TEM the pyrenoid (arrow) with some lenticular starch grains can be seen. For further information on the pyrenoid see SILVERBERG

(1975). Keys for the determination of *Stichococcus* species can be found in ETTL & GÄRTNER (1995) or HINDÁK (1996).

***Trentepohlia aurea* (L.)MARTIUS as photobiont in *Gyalecta jenensis* (Fig. 2):**

Well known since REINKE (1896) and detailed studied by TSCHERMAK (1941) this green alga changes the morphology +/- very little.

Also in lichenized condition short branched filaments can be seen. But seldom reproduction by swarmers/gametes occurs under natural conditions within the lichen. Only time-consuming cultivation can elucidate the taxonomic situation of *Trentepohlia* algae from lichens (but also from free-living state!). Some characteristic taxonomic criteria of *T. aurea* can be seen from Fig. 2, following BACHMANN (1992).

***Trebouxia sp.* as photobiont in *Dactylina ramulosa* (Fig. 3):**

Up to now the photobiont of this arctic-alpine lichen was characterised as „protococcoid" (POELT & VEZDA 1977). As the generic name „*Protococcus*" should not be used any longer (LAUNDON 1985, GÄRTNER & INGOLIĆ 1989, ETTL & GÄRTNER 1995, GÄRTNER 1992), we have to check these photobionts in details. „*Protococcus*"-like algae should form sarcinoid (= packets of cells) and short filamentous stages of development, which are only observed under culture conditions. The green algal genera *Apatococcus, Desmococcus, Coccobotrys* and *Diplosphaera* are synonym with *Protococcus* (or *Pleurococcus),* their diacritic features can be found in ETTL & GÄRTNER (1995). The photobiont of *Dactylina ramulosa* from high alpine areas of the Ötztaler Alps belongs certainly to the genus *Trebouxia* (for details see Fig. 3). The cultivated photobiont was quite recently isolated and needs further investigations. According to our current state of knowledge it seems to be an unknown species until now.

The results of our studies confirm again the necessity of cultures for an identification of photobionts at species level, as stated recently by FRIEDL & BÜDEL (1996).

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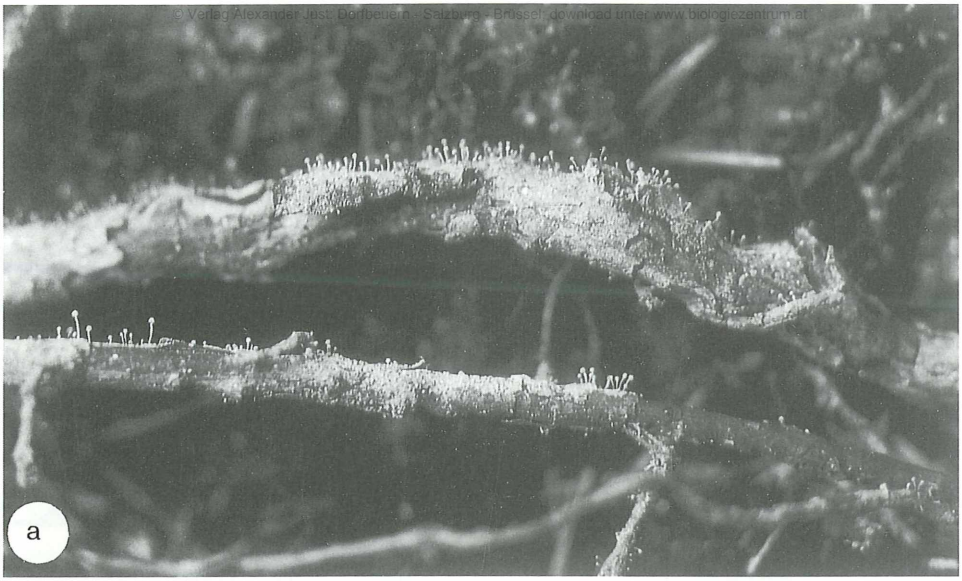
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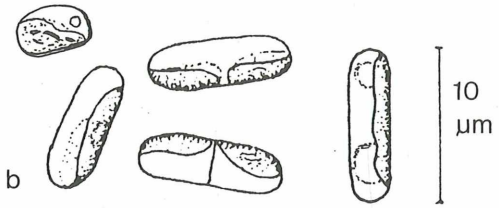
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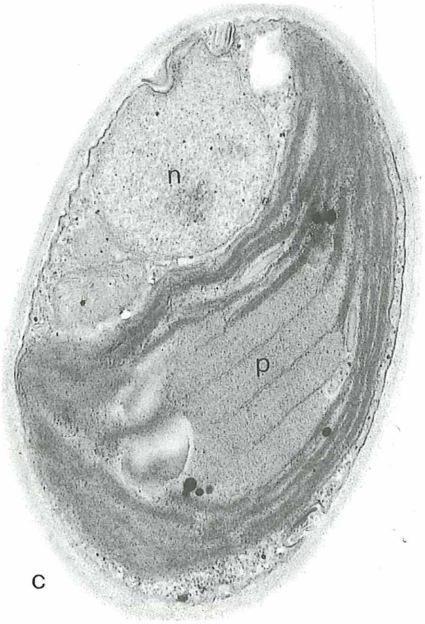
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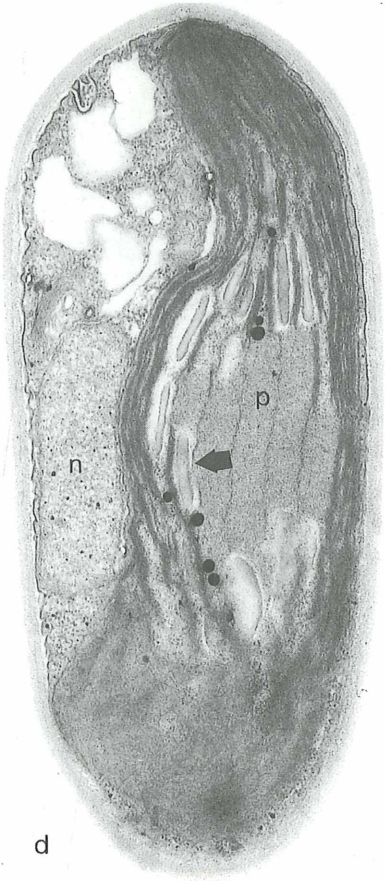
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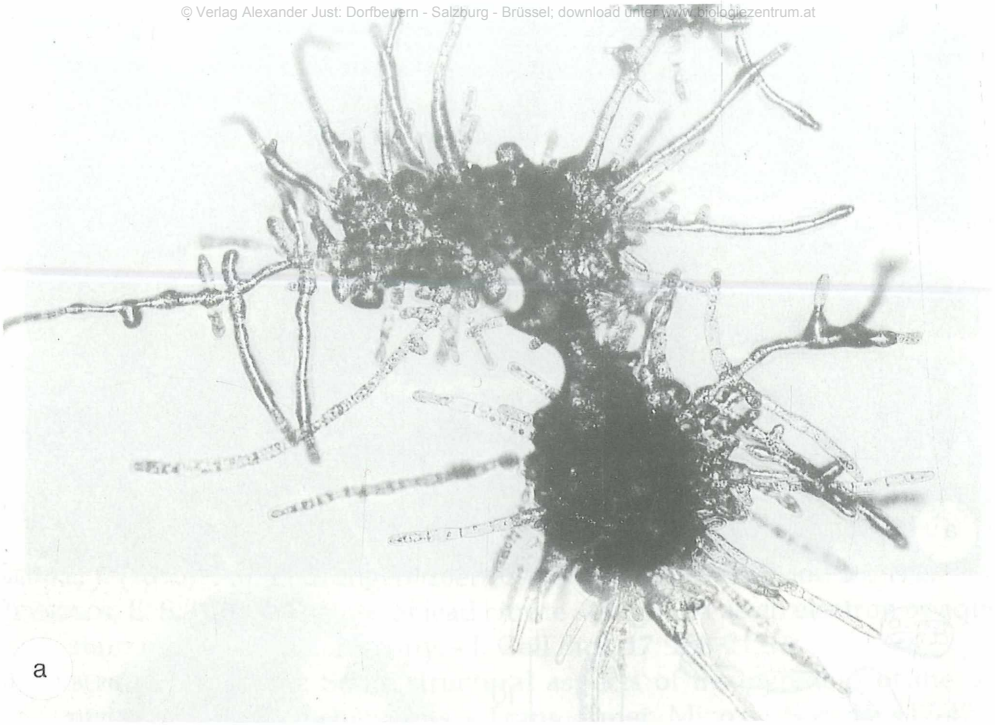


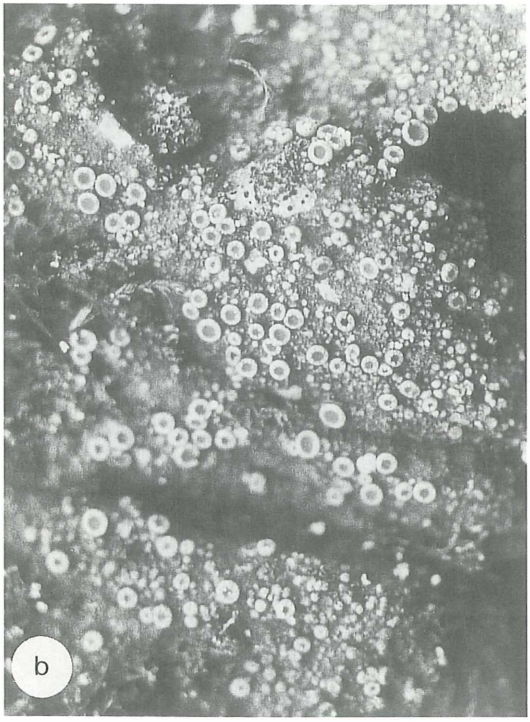


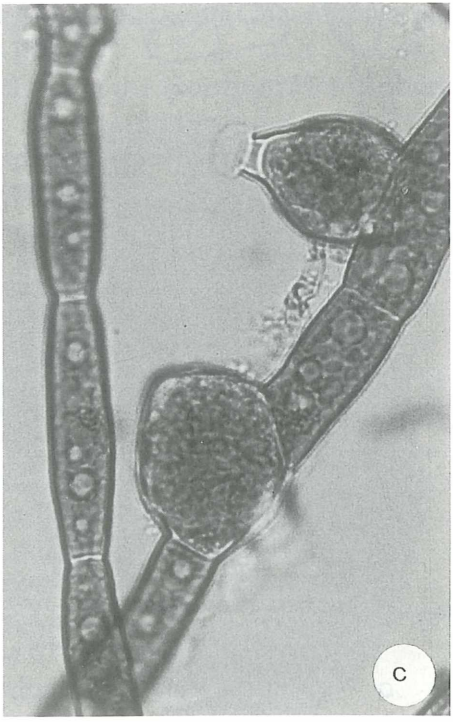


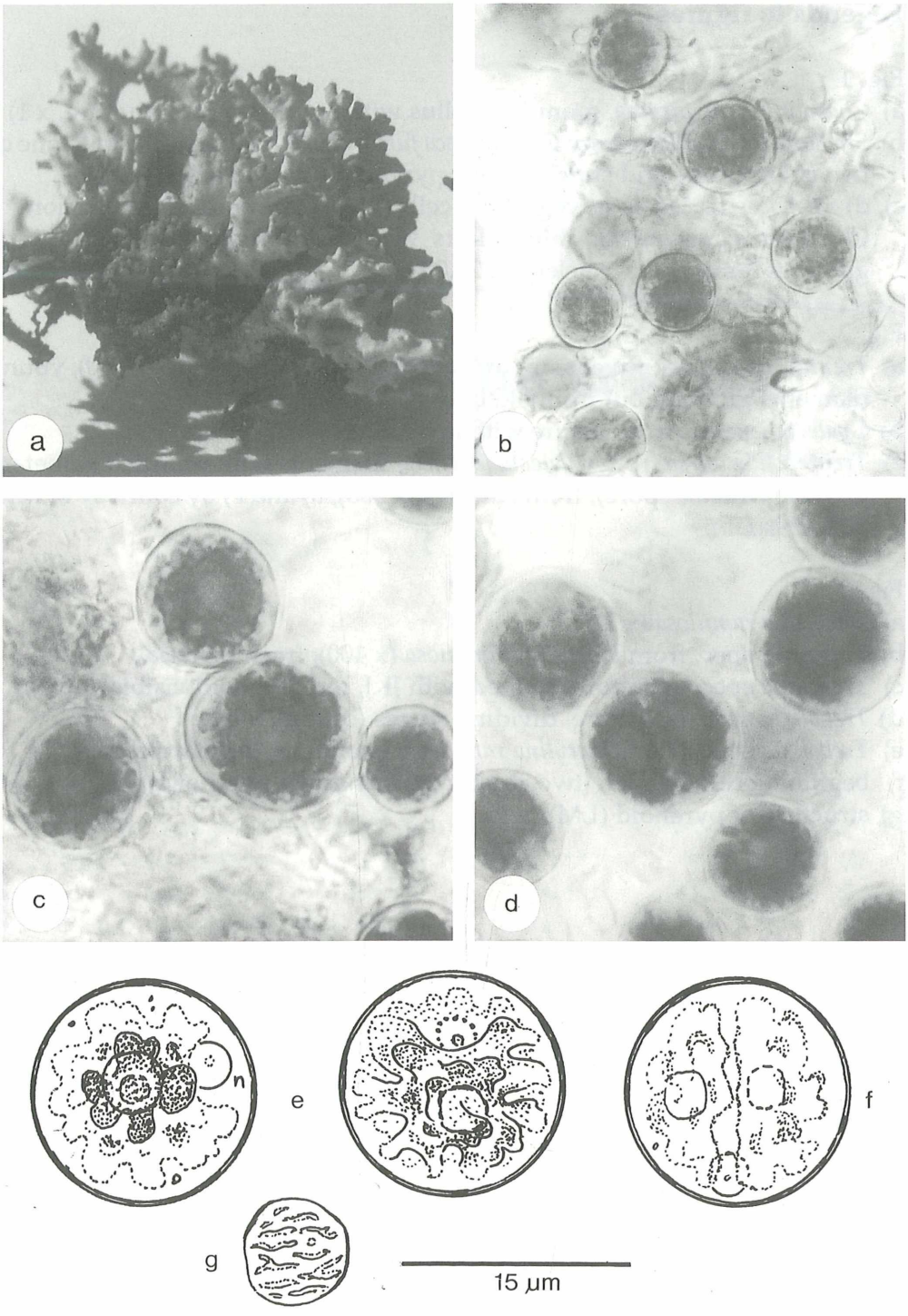


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Fig. 1
a) *Chaenotheca furfuracea,* granular thallus with apothecia (from nature, x 1)
b) *Stichococcus bacillaris* from *Chaenotheca furfuracea* in culture; cells with one or two chloroplasts and starch grains, pyrenoid hardly visible;
c), d) *Stichococcus bacillaris,* vegetative cells (EM), from culture (phycobiont of *Chaenotheca furfuracea);* chloroplasts with one pyrenoid (p) and starch granules (arrow); n = nucleus; x 28.000

Fig. 2
a) *Trentepholia aurea* (phycobiont of *Gyalecta jenensis)* in culture (x 160); young filaments growing out from old ones;
b) *Gyalecta jenensis* from nature with apothecia (x 4);
c) *Trentepohlia aurea* from *Gyalecta jenensis,* filaments with rounded gametangia (one with exit pore); from cultures (x 360); a) and c) by courtesy of Mrs. G. BACHMANN;

Fig. 3
a) *Dactylina ramulosa* (x 2);
b) *Trebouxia* spec. from *Dactylina ramulosa* (x 400); fresh material;
c) *Trebouxia* spec. in culture, stained with JKJ, the pyrenoid visible (x 630);
d) *Trebouxia* spec. in culture, dividing stages (centre), x 630;
e) *Trebouxia* spec. from *Dactylina ramulosa,* in culture, vegetative cells;
f) beginning cell division (two chloroplasts and two pyrenoids);
g) structure of pyrenoid (LM);

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