Prokaryotic Biodiversity in Marine versus Terrestrial Ecosystems: Methylobacteria and Research Ethics

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Abstract

Prokaryotic microbes (bacteria) are the most numerous organisms on Earth. In marine environments, cyanobacteria of the genus Prochlorococcus and heterotrophic prokaryotes of the family Pelagibacteraceae are of importance, and in terrestrial ecosystems, plant-associated methylobacteria are very abundant. Here, we summarize the discovery and description of a microbe isolated from the common cord moss, Methylobacterium funariae, as described by Schauer and Kutschera (2011). Based on samples of M. funariae which were provided to colleagues as a gift, our isolate was described without our knowledge and permission for a second time, under another name. We discuss this wasteful and duplicative publication of data generated twice from the same bacterial isolate with respect to the ethics for the conduct of research in the biological sciences.

Despite their minute size, bacteria are the most numerous and diverse forms of life in the world. In marine ecosystems, phototrophic cyanobacteria of the genus Prochlorococcus and heterotrophic prokaryotes of the family Pelagibacteraceae, also known as the ‘SAR11 bacterial clade’, are among the most abundant prokaryotes [1]. However, despite their proliferation throughout the world’s oceans, the taxonomy of these important bacteria is still unclear. In terrestrial ecosystems, members of the alpha-proteobacterial genus Methylobacterium are abundant epiphytic endophytes that have been isolated from the leaf surfaces of bryophytes, mosses, ferns, and seed plants. Methylobacteria are capable of growing on a variety of one-carbon compounds, such as methanol, which is emitted from the stomatal pores of the leaves. About 40 bacterial species (i.e., defined strains) of these pink-pigmented, Gram-negative prokaryotes have been described so far [2]. In this personal account, we describe a case study of bacterial systematics that sheds light on current problems in publication ethics [3].

In 2003, we isolated methylobacteria from the upper surface of the thallus of the liverwort Marchantia polymorpha. The “primitive” land plants were collected from Bergpark Wilhelmshöhe (Kassel, Germany). Later, we described one of our lab strains, labelled ‘JT1’, as novel species (Methylobacterium marchantiae) [4].

One year before (May 2002), we had isolated another strain (F3.2) from the phylloids of the common cord moss Funaria hygrometrica, collected in the Bot. Garden of the University of Kassel, where sunflower plants were raised (Figure 1), and cultivated these microbes in the Plant Physiology laboratories of the Institute of Biology. This isolate was described informally as Methylobacterium funariae in the PhD thesis of the second author [5], and later, supplemented by additional data, as a new species in a peer-reviewed journal [6].

In his “Validation list no. 145”, Euzéby [7] proposed that our strain F3.2 should be assigned to the taxon Methylobacterium bullatum sp. nov [8], but added the following footnote: "Based on the strain history of the type strain of Methylobacterium bullatum, it appears that strain F3.2 has also been used as the type strain of 'Methylobacterium funariae' (Schauer and Kutschera, 2011)" [6]. In Feb. 2012, we received a pertinent e-mail from a colleague (D. P. Kelly, University of Warwick, UK; 15th February, 2012): "I have a query about Methylobacterium funariae. While making a survey of the 16S rDNA gene accessions of all the published species, I found that strain F3.2 has been described twice with different names (M. funariae and M. bullatum) by separate groups at Kassel. Can you tell me how this happened, and which of the names is likely to be validated in the International Journal of Systematic and Evolutionary Microbiology (IJSEM)? I am wondering if the earlier report (April 2011) will have taxonomic precedence, or whether the report by Hoppe et al. [8] will be the validated name, as they deposited the type strain in the German Collection of Microorganisms and Cell Cultures (DSMZ), as required by the international nomenclatural..."
rules”. Moreover, a recent report [9] misinterpreted the two-fold description of our isolate F3.2, and erroneously discussed “two” separate bacterial species. However, our unpublished DNA sequence-based phylogenetic trees document that *M. funariae* and *M. bullatum* are identical bacterial species. In this editorial, we elucidate the mysterious two-fold description of our isolate *Methylobacterium* F3.2 (Figure 1).

In our first research paper that was published in the year when we isolated lab strain *Methylobacterium* F3.2, we characterized the unique phenotype of our “*Methylobacterium* sp. that was isolated from the phylloid” [10]. Moreover, we quantified the ability of *Methylobacterium* sp.F3.2 to secrete cytokinines, using a *Funaria hygrometrica*-protonemata-bioassay. In two subsequent reports [11,12], *Methylobacterium* sp. F3.2 was further characterized with respect to its capability to synthesize and secrete the phytosphere auxin. Based on these extensive studies, “*Methylobacterium* sp. isolated from phylloids of *F. hygrometrica*” [10] was described as *M. funariae* [5,6].

In our article which was published ten years ago [10], we depicted scanning electron micrographs that document the rugged outer surface of *Methylobacterium* sp. F3.2 (see Figure 1, Inset). To further characterize the fimbriae-like structures on the outer wall layer of this isolate, we provided samples of *Methylobacterium* sp. F3.2 to the microbiology lab of the Institute of Biology, University of Kassel (Principal Investigator: F. Schmidt). However, without our knowledge and permission, our colleagues used our un-described microbial taxon to work on a second species description. They were informed that our isolate had already been named *M. funariae* by Schauer (2009) [5], and that a corresponding species description was in preparation. It should be noted that the first author of this second paper [8] proposed to share the data, publish them, and give a valid species description in the IJSEM. We agreed to the proposal of T. Hoppe, but the PI of the microbiology lab (F. Schmidt) declined our offer (the second “author” on the Hoppe et al.-article [8] is a technician without academic qualification).

As a result, two species descriptions were published [6,8], and although our paper appeared in print first, and Schauer (2009) [5] had already described this taxon two years earlier, the name “*M. bullatum*” has been accepted by IJSEM [7]. We are aware of the fact that, according to the Bacterial Code 1990 Revision, the “date of effective publication does not determine priority” (Rules 23b and 27).

In a recent editorial published in the IJSEM, Kämpfer [13] described a case of falsification of ‘Certificates of Deposit’ and reminded the readers that the IJSEM is a member of the Committee on Publication Ethics. We feel that the “*M. bullatum* case” described here is clearly a violation of the ethical standards in biology, since, according to the “Codes and policies for research ethics”, scientists should avoid duplicate publication and not deceive colleagues [3]. We hope that such a wasteful and two-fold description of the same bacterial isolate, provided by the discoverers of this taxon as a gift to colleagues, will not happen again.

Finally, we want to point out that methylobacteria are among the most abundant heterotrophic microbes of the above-ground phytosphere (stems, leaves) in terrestrial ecosystems. Moreover, they also inhabit the rhizosphere of all land plants investigated so far [14]. Hence, these microbes may be viewed as ecological counterparts to the marine *Pelagibacteraceae*, which are likewise members of the alphaproteobacteria [1,14]. However, more experimental work is required to further corroborate our conclusion that the marine SAR11 bacterial clade [1] and land plant-associated methylobacteria [14] are in fact the dominant heterotrophic prokaryotic microbes in their respective ecosystems.

**References**