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Letter to the Editor

Trichlorobacter thiogenes Should Be Renamed as a *Geobacter* Species

In a recent paper by De Wever et al. (2), it is proposed that the recently isolated microorganism strain K1 be assigned to a new genus within the delta *Proteobacteria*. Those authors state that the 16S ribosomal DNA (rDNA) sequence of strain K1 places it within a "cluster of mixed taxonomic affiliation" within the delta *Proteobacteria*. From this statement it is apparent that De Wever et al. (2) are unaware of several previous phylogenetic analyses of this group within the delta *Proteobacteria*, most notably, a study by Lonergan et al. (5). As De Wever et al. (2) note, the 16S rDNA sequence of strain K1 is nearly identical (99% sequence identity) to a 16S rDNA sequence recovered from a bioreactor that they misidentify as "environmental sp. 2" but that is actually an environmental sequence first described as a population type 1 sequence (*Desulfuromonas*-like sp.) (1) and listed by both GenBank and the Ribosomal Database Project (RDP) II (9) release 7.1 as *Desulfuromonas* sp. (GenBank accession number M80618). If De Wever et al. (2) had known about the study by Lonergan et al. (5), they would have realized that detailed analysis of this sequence has demonstrated that it rests squarely within the *Geobacter* cluster of the family *Geobacteraceae* (5). Our own analysis of the strain K1 sequence confirms not only that the overall sequence of strain K1 is closely related to organisms in the *Geobacter* cluster (Fig. 1) but also that the sequence contains the signature secondary structures characteristic of the *Geobacter* cluster (5).

At this time, the *Geobacter* cluster contains only one organism, *Pelobacter propionicus*, that does not have the genus designation *Geobacter* (Fig. 1). There are four more species of *Pelobacter* within the *Geobacteraceae* family, but these other *Pelobacter* species are interspersed throughout two other genera outside the *Geobacter* cluster. Thus, it is clear that the genus *Pelobacter* is not phylogenetically coherent and that organisms in the *Geobacter* cluster cannot be renamed *Pelobacter* as this would result in organisms of different phylogenetic clusters being placed within the same genus. It has been suggested that the simplest way to avoid confusion with the phylogeny is to place *P. propionicus* in the genus *Geobacter* (5).

Therefore, designation of strain K1 as a new genus within the *Geobacter* cluster, as suggested by De Wever et al. (2), creates havoc within an otherwise logical grouping of organisms, comprised of predominantly a single genus within a phylogenetically coherent cluster. Designation of a new genus might be warranted if strain K1 had some unique physiological characteristics that distinguished it from previously described *Geobacter* species, but this does not appear to be the case. Strain K1 uses acetate as an electron donor for the reduction of S^0 and fumarate, and it has been suggested that it might also use Fe(III) as an electron acceptor (2). The oxidation of acetate coupled to the reduction of S^0 and Fe(III) is one of the defining physiological characteristics of *Geobacter* species, and many *Geobacter* species can also use fumarate as an electron acceptor for acetate oxidation (6, 7, 8). Strain K1 does reductively dechlorinate trichloroacetic acid, a physiological capacity not previously reported for *Geobacter* species. However, to our knowledge, no other organisms in the *Geobacter* cluster have been evaluated for the ability to carry out this reaction. Therefore, it is not clear that strain K1 is unique among organisms in the *Geobacter* cluster in this ability. Furthermore, De Wever et al. (2) suggest that at least part of the reductive dechlorination observed with strain K1 is the result of strain K1 reducing S^0 to sulfide, with the subsequent reduction of the trichloroacetic acid by sulfide. Since all organisms in the *Geobacter* cluster have the ability to reduce S^0 to sulfide, it is likely that all *Geobacter* species have the ability to dechlorinate trichloroacetic acid via this electron shuttling mechanism.

Furthermore, even if all the organisms in the *Geobacter* cluster other than strain K1 were found to not be able to reductively dechlorinate trichloroacetic acid, precedence suggests that the designation of a new genus would not be warranted. When the first organism in the *Desulfuromonas* cluster of the *Geobacteraceae* found to have the ability to carry out reductive dechlorination was described (4), it was not assigned to a new genus; rather, it was designated a new species in the genus *Desulfuromonas* (3). This is consistent with the concept of not cluttering phylogenetically coherent groups with multiple genus designations.

In summary, designating strain K1 as a new genus in the *Geobacter* cluster at this time advances neither clarity in phylogeny or understanding of physiology. It is suggested that, once the characterization of the physiology of strain K1 is completed, it be designated a new species in the genus *Geobacter*.

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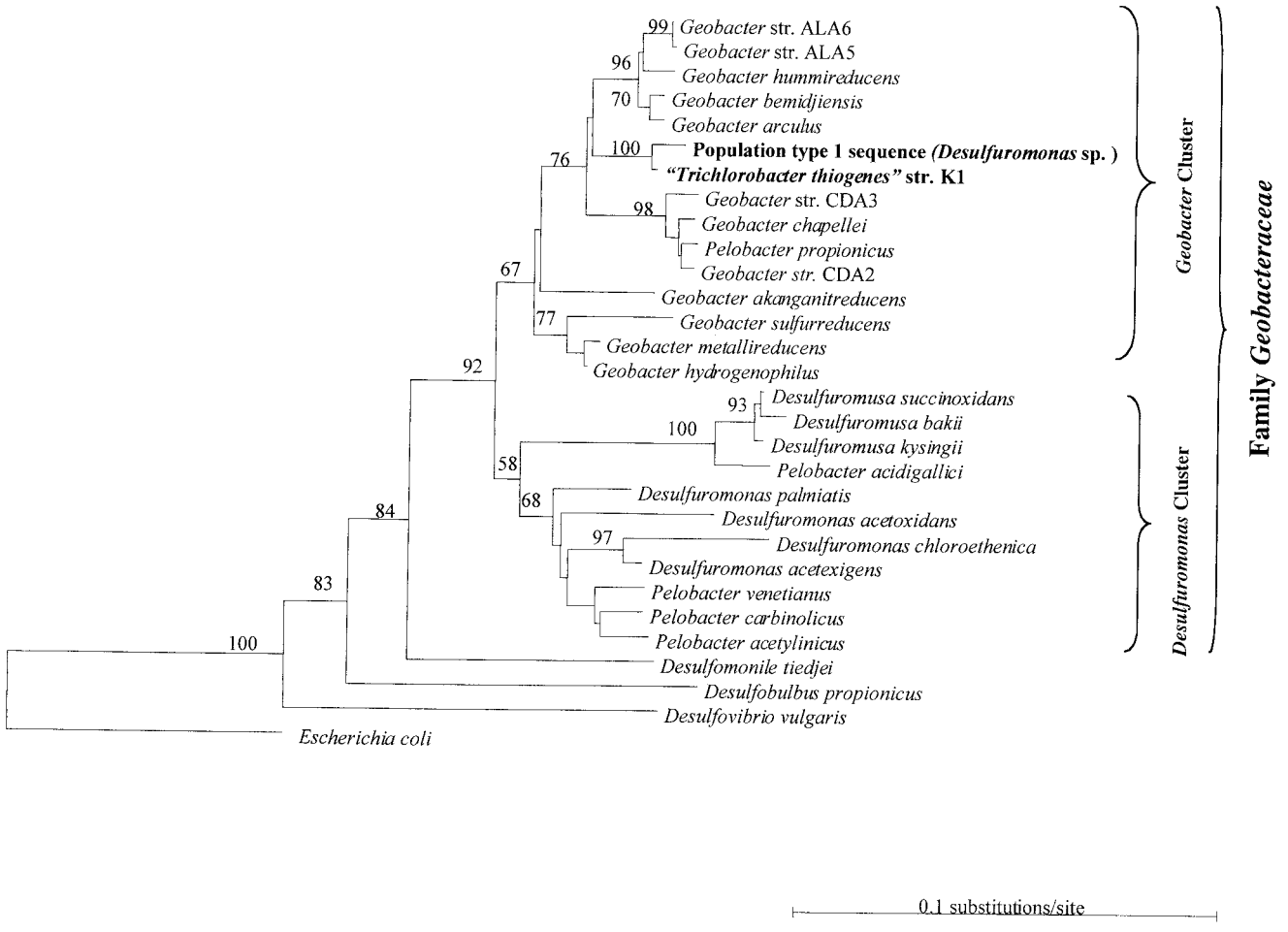

FIG. 1. Phylogenetic tree inferred from 16S rRNA sequences showing the phylogenetic placement of "*Trichlorobacter thiogenes*" strain K1. Phylogenetic relationships shown here were inferred by using neighbor joining and Kimura two-parameter genetic distances in TREECON (10). Bootstrap values above 60 are shown adjacent to branch nodes and were calculated from 100 resampled data sets using neighbor joining. The scale bar shows the number of expected nucleotide substitutions per site per unit of branch length. Operational taxonomic units shown on the tree were obtained from GenBank and the RDP databases. A similar tree topology was generated for trees constructed using maximum-likelihood and maximum-parsimony methods (data not shown).

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Authors' Reply

The genus *Geobacter* has only two species with standing in bacterial nomenclature: *G. metallireducens* and *G. sulfurreducens*. According to the rules of the International Code of Nomenclature of Bacteria, neither the two additional *Geobacter* species from Lonergan et al. (2), nor the other four *Geobacter* species in Fig. 1, nor the proposed family name "*Geobacteraceae*" from Lonergan et al. has been validly published. Therefore, according to the Code, they have no standing. In fact, we are unable to find even a GenBank sequence record for "*G. hummireducens*" or "*G. bemidjiensis*", and the GenBank rRNA records for "*G. arculus*" and "*G. akanganitreducens*" indicate that they are unpublished! A quick search of Medline drew a blank for all four species. In any event, the Code is clear; validation of a name or new combination is the responsibility of the name's author.

The "*Geobacter* cluster" contains only three valid species: *Pelobacter propionicus* and the two *Geobacter* species. From phylogenetic analysis, strain K1 is most closely related to *P. propionicus* (94% rRNA sequence similarity), while the two species of *Geobacter* form a separate cluster (92 and 93% similarity to K1). Although 16S rRNA similarity should not be used as the sole criterion for division of taxa, these values are well within the range of differences seen for related genera.

We are not sure about the relevance of the name we used for

the *Desulfuromonas*-like bioreactor sequence (GenBank accession number M80618), but the original publication states that "the GenBank accession numbers for population type 1 and population type 2 partial 16S rRNA sequences are, respectively, M80617 and M80618" (1). In addition, the RDP annotates their corresponding record as "population type 2."

We were aware of the work of Lonergan et al. (2). In fact, we included the 16S sequences for their two proposed *Geobacter* species in our analysis. If we offended by not citing that work, we apologize but note in partial defense that the practice of referencing sequences by GenBank accession number as opposed to original work is quite common.

Snoeyenbos-West et al. apparently take offense at our calling this group of organisms a "cluster of mixed taxonomic affiliation;" however, this seems to be in accordance with the conclusions of Lonergan et al., who suggest a reassessment of the affiliation of five species in the proposed "*Geobacteraceae*" family. We note that none of these reassessments have taken place.

Since the number of strains belonging to this "family" is currently growing, we suggest a full taxonomic reassessment of the entire group, including full phenotype comparisons and species-level molecular discrimination, e.g., by *gyrB* phylogeny analysis and DNA-DNA hybridization, published in valid manner according to the international rules of nomenclature so that the taxa are properly recognized. It is rather common for such reassessments to occur once the number of strains and their diversity have grown for a more informed analysis. It may well be that it is more appropriate for strain K1 to be reassigned as a dechlorinating species to a single generic group, but this conclusion is premature given the current status of reporting.

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