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Evaluation of Current Species Definition for Prokaryotes Using Multigene Phylogeny

Ji-Hye Han, Sung Heun Cho, and Seung Bum Kim*

Department of Microbiology, School of Bioscience and Biotechnology, Chungnam National University

The concept of species may vary in different organisms, and that for prokaryotic organisms has been a main subject of debate in prokaryotic taxonomy. Currently the most widely accepted concept of prokaryotic species would be the phylogenetic species based on 16S rRNA gene sequences. However, the rRNA based phylogeny alone cannot provide solution on how to define species, although the simple sequence similarity below a certain level ensures distinction of species. The relatedness between genomic DNAs is currently used as an ultimate operational standard for species distinction [1]. However, despite that the 70% rule looks to have its own rationale, some practical problems of this rule, both experimental and conceptual, are also apparent [3, 4].

Phylogeny based on multigene analysis seems to gain popularity, and through this approach, ways to avoid the tedious DNA-DNA relatedness experiments are sought for. Multigene phylogeny has merits in that more parts of the genome can be utilized in the inference of phylogenetic relationships, and that the sequence data can be made available through public databases [1, 2, 6]. In this study, popular genetic markers were used in the phylogentic analysis of the three genera belonging to the family *Streptomycetaceae* and the resultant data were compared. The markers employed were the genes encoding small and large subunit ribosomal RNAs (5S, 16S and 23S), β-subunit of DNA gyrase (*gyrB*), and β-subunit of RNA polymerase (*rpoB*).

Different levels of correlation were found between the markers, thus implying that the phylogenetic relationship among the organisms would vary depending on the selection of markers. However, the overall topology of the phylogenetic trees based on *gyrB* and *rpoB* sequences was similar to that based 16S rDNA. In contrast, the 23S rDNA based tree showed that one genus, *Streptacidiphilus*, was split into two clades, one clustered with its neighboring genus, *Kitasatospora*. The degree of variation, or nucleotide substitution rate, was highest in the *gyrB* sequences, and lowest in the 23S rRNA gene sequences. The 16S rDNA and *rpoB* exhibited similar range of nucleotide substitution rates.

The analysis of concatenated sequences using five genes gave different views from that using 16S rDNA alone, and in particular the phylogenetic relationships between *Kitasatospora* and *Streptacidiphilus*. The precise tree topology for *Streptomycetaceae* family would be expected by addition of more sequences, in particular those of *Streptomyces*.

Multigene phylogeny has two important advantages over the single gene approach - higher resolution and higher correlation with genome similarity. Nonetheless, the problem of how close is a species in terms of sequence similarity or evolutionary relatedness remains. Still, it is now obvious that reliance on single gene phylogeny is a dangerous choice, and thus that multigene phylogeny would give better solution, though the problem of which genes to choose remains. At the same time, species based on quantitative measurement essentially creates borderline problem, and controversies over the prokaryotic species concept will not likely disappear in near future.

References

1. Wayne L, DJ Brenner, RR Colwell, and 9 others. *Int J Syst Bacteriol* **37**,463 (1987).
2. Hatano K, T Nishii, and H Kasai. *Int J Syst Evol Microbiol* **53**,1519 (2003).
3. Stackebrandt E and J Ebers. *Microbiology Today* **nov,** 152 (2006).
4. Ciccarelli FD, T Doerks, C von Mering, CJ Creevey, B Snel, and & P Bork. 2006. *Science* **311**,1283 (2006).
5. Guo Y, W Zheng, X Rong, and Y Huang. *Int J Syst Evol Microbiol* 58,149 (2008).
6. Yamamoto S and S Harayama. *Int J Syst Evol Microbiol* **48**,813 (1998).