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3 **Problems With Procaryote**

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5 Norman R. Pace

6 MCD Biology, CU-Boulder

7 nrpace@colorado.edu

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10 Scientific foundations must be based on facts, not untested conjectures. Good
11 conjectures are ones that suggest experimental tests and concomitant refinements of
12 knowledge. Bad conjectures accepted without test can stultify progress. Beliefs built only
13 on conjecture can lead to flawed understanding and hollow teaching.

14

15 Biologists of the 20th Century embraced conjecture in the form of what I will call the
16 procaryote-eucaryote model for biological organization and evolution. That model, explicit
17 in the language, posits that fundamentally there are two kinds of organisms, procaryotes
18 and eucaryotes. Furthermore, the prefix “pro-” injects the connotation that procaryotes
19 preceded eucaryotes because “pro-“ means before. This procaryote-eucaryote model
20 dominates our textbooks and discourse in matters of microboal taxonomy and deep
21 evolution.

22

23 The thesis of this article is that the notion of procaryote is now obsolete and
24 counterproductive. I will: (1) trace the source of the procaryote-eucaryote model; (2) show
25 why it is wrong scientifically; and (3) document the damage it has done to our perception of
26 important biological issues. I conclude that “procaryote” needs to be removed from the
27 lexicon of biology.

28

29 **The essence of the argument**

30

31 The procaryote-eucaryote concept is a hypothesis that has been proven wrong by
32 three decades of results in molecular phylogeny and biochemistry. These results,
33 summarized below, show that life’s diversity falls into three fundamentally distinct

34 phylogenetic domains: bacteria, eucarya (eucaryotes) and archaea. Moreover, the results
35 show that archaea and eucaryotes evolved independently of the bacterial line of descent.
36 Thus, there is no such thing as a “procaryotic” relatedness group of organisms in any
37 phylogenetically based taxonomy. It makes no sense to lump bacteria with archaea
38 phylogenetically. Furthermore, the molecular tree reveals no group of organisms that
39 preceded eucaryotes.

40

41 The accompanying diagrams contrast the procaryote-eucaryote and three-domain
42 models for the large scale of biological organization and the course of evolution.

43

44 **Where procaryote started**

45

46 The fallacy of procaryote is not a new issue. The history of the procaryote concept
47 and its incorporation as dogma into the conventional wisdom of biology has been reviewed
48 (4,8, 9).

49

50 In 1962, R.Y. Stanier and C.B. van Niel, seeking a “concept of a bacterium,”
51 proposed “The distinctive property of bacteria and blue-green algae is the prokaryotic
52 nature of their cells...” (5). By “prokaryotic nature” they meant their postulated “common
53 cellular organization”. Thus, by negative definition, lack of a nuclear membrane meant
54 procaryote. The choice of language, procaryote and eucaryote, explicitly invoked the
55 evolutionary model.

56

57 Beyond selection of terminology, Stanier and van Niel did not specify an evolutionary
58 model. Stanier, with colleagues M. Douderoff and E.A. Adelberg, approached the issue
59 subsequently in the 2nd edition of their popular textbook “The Microbial World” (1963) (6).

60

61 “It is not too unreasonable to consider that the bacteria and blue-green
62 algae represent *vestiges of a stage in the evolution of the cell* which, once it
63 achieved a eucaryotic structure in the ancestors of the present-day higher
64 protists, did not undergo any further fundamental changes through the
65 entire subsequent course of biological evolution.” (p. 85).

66

67 And they concluded:

68
69 “All these organisms [“lower protists”] share the distinctive structural
70 properties associated with the procaryotic cell (Chapter 4 [above]), and we
71 can therefore safely infer a common origin for the whole group in the remote
72 evolutionary past ...” (p. 409).

73
74 The procaryote-eucaryote model was welcomed by biologists and hailed - finally
75 there was a scientific definition of bacteria. But procaryote was never described
76 scientifically because the properties that define the two primary taxonomic groups were
77 exclusionary, not specific. The origins of the two groups were guesswork. Yet, the
78 procaryote model was incorporated into the conventional wisdom of biology with little
79 question and without test. I speculate that easy acceptance by biologists was because it
80 neatly fit 19th Century concepts of evolution prevalent in textbooks even today, with
81 monera at the base of a tree of complex eucaryotes.

82

83 **Disproof of the concept of procaryote**

84

85 The first experimental test of the procaryote-eucaryote hypothesis came with the
86 introduction of molecular phylogeny and Carl Woese’s landmark comparisons of small-
87 subunit (16S) ribosomal RNA (rRNA) sequences, in 1977 (7). In contrast to the
88 physiological and morphological properties traditionally used in microbial taxonomy,
89 comparison of gene sequences provided an objective metric for evolutionary diversity.
90 Simply put, the extent of sequence difference between orthologous genes in different
91 organisms is a measure of evolutionary distance and relationship. Consequently,
92 comparisons of sequences can be used to extract maps of evolutionary relatedness of
93 organisms, phylogenetic trees. Ribosomal RNA sequences, because of their ubiquity and
94 high degree of conservation, became the gold standard for relating all of life. Moreover,
95 the rRNA genes have not undergone appreciable lateral transfer, as have some genes.
96 The phylogenetics of the other elements of the nucleic acids-based information processing
97 system are congruent with that of the rRNA. Consequently, the rRNA tree tracks the
98 evolution of the cell’s genetic machinery, at the very least.

99

100 The results of rRNA-based molecular phylogeny studies have been reviewed
101 extensively (e.g. 1, 2, 9). Analyses of many rRNA and now genome sequences have
102 solidified Woese's early findings of three primary relatedness groups, the phylogenetic
103 domains of bacteria, eucarya and archaea. Woese originally dubbed this latter group
104 "archaebacteria," although the sequences and biochemical correlates would show that the
105 traditional bacteria and archaea are fundamentally distinct kinds of organisms.

106

107 The pattern of the three-domain tree (figure) shows that there is no phylogenetically
108 coherent group that can be described as procaryotic. The pattern of the molecular tree
109 also shows that none of the primary domains is derived from another. The molecular
110 results prove that the major eucaryotic organelles, mitochondria and chloroplasts, had their
111 origins among bacteria (not shown in the figure). But the eucaryotic nuclear line of
112 descent was of primordial origin, as old as the archaeal line and derived from neither
113 bacteria nor archaea.

114

115 **The relationships of the phylogenetic domains**

116

117 The location of the "root" of the universal tree, the position of the last universal
118 common ancestral state, is critical to the legitimacy of the term procaryote. This root
119 cannot be inferred from rRNA data. However, other phylogenetic results put the root of the
120 molecular tree on the line between the bacterial radiation and the separation of the
121 eucaryal and archaeal lineages (3). This is indicated as "origin" in the diagram of the
122 three-domain tree in the figure. The position of the root of the tree fundamentally
123 separates bacteria and archaea. There is no phylogenetically based grouping that
124 contains bacteria and archaea. There can be no such thing as a procaryote in any
125 phylogenetic sense.

126

127 The phylogenetic differentiation of the bacterial, archaeal and eucaryal lines of
128 descent is supported by many biochemical correlates. For instance, whereas bacteria use
129 sigma factors to control transcription initiation, both archaea and eucarya use a different
130 mechanism, TATA-binding proteins. As another example, bacteria wrap their DNA in a
131 variety of basic proteins, while eucaryotes and many archaea both use histones. The early
132 common history and such similarities do not mean that archaea are rudimentary

133 eucaryotes. Vast evolutionary history separates archaeal and eucaryotic cells, and they
134 have fundamental differences. Eucaryotes (and bacteria), for instance, make their
135 membranes from ester-linked lipids, whereas archaea use ether-linked lipids.

136

137 **The damage of procaryote: it's more than terminology**

138

139 Conflict between the procaryote-eucaryote and three-domain models is not simply
140 one of terminology, the names we give to organisms. So what does it matter if we lump
141 together bacteria and archaea? Maybe, some would argue, we can use the term
142 "procaryote" informally, to refer to small organisms that are not members of the domain
143 eucarya, non-eucaryotes. However, this is not satisfactory terminology because it
144 confuses an invalid scientific term with a colloquial one.

145

146 The terminology also matters because it perpetuates incorrect information about
147 important biological concepts, of the structure of biological diversity and the course of
148 evolution. Progress in biology requires a proper perception of life's natural organization
149 and evolution. The procaryote notion distorts and misleads. Taken literally, it would have
150 stipulated that experiments to probe distinctions between archaea and bacteria would not
151 be necessary. Procaryote smothered interest in microbial evolution and obstructed
152 acceptance of archaea, the first test of the procaryote hypothesis. It permeates our
153 textbooks and journals with subtle and not so subtle misinformation. Thus, the dogma of
154 the procaryote has damaged and can continue to damage microbiology by retarding or
155 even denying progress. It elicits false concepts and misdirects inquiry. There is no place
156 for procaryote in modern scientific discourse.

157

158 **What to do about it?**

159

160 The facts are on the table, but it will take time for the terminology of procaryote to
161 disappear from our textbooks and language. Nonetheless, the process needs to start and
162 it needs to be catalyzed. We microbiologists need to take the lead in rectifying the
163 misconceptions because we are closest to the problem of how to understand and describe
164 highly disparate organisms.

165

166 An early challenge to microbiologists is to stop using the term “procaryote.” This is
167 hard to do because of long conditioning. Those tempted still to use it must realize,
168 however, that they saddle their students with misconception and muddy their thinking
169 about important biological problems. An alternative catch-all term that I use and
170 recommend is “microbe”, which captures the microbial quality of size and includes as well
171 the poorly acknowledged microbial eucaryotes. But beyond that it is necessary to be more
172 specific. The adjective “procaryotic” is almost always misleading; for example, it is
173 scientifically more correct and far more illuminating to distinguish bacterial and archaeal
174 transcription than it is to lump them together into “procaryotic transcription.”

175

176 How should we teach this issue in the context of the currently pervasive use of
177 “procaryote” in textbooks and journals? In fact, the discordance between currently
178 emerging data and the conventional thought on deep evolutionary relationships is a
179 wonderful example of how science, biology in this case, is an ongoing, living process.
180 Bringing the subject to students shows them how new information based on experimental
181 evidence can change in fundamental ways how we understand important natural
182 processes. Dealing with the procaryote-eucaryote issue provides good example of
183 weighing specific models, hypotheses for testing in the face of experimental data.
184 Phylogenetic trees, maps of evolutionary relationships, are not hard to understand in
185 essence. They are abstract, to be sure, but provide graphic information that is readily
186 interpreted by students. The three domains concept of course poses many questions, but
187 it also provides a solid foundation for progress toward answering those questions.

188

189 **Acknowledgements**

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192 Goldenfeld and Corrie Detweiler, for their contributions to this article.

193

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233 **Figure legend:**

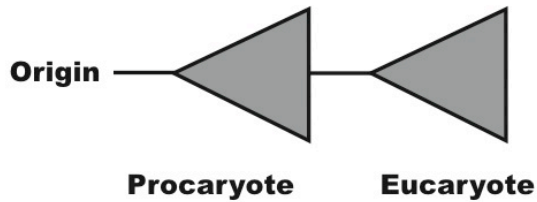
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235 **Models for the pattern of life's evolution.** The wedges represent the genetic radiations
236 from the base of the respective primary relatedness groups. **A** represents the procaryote-
237 eucaryote model and **B** the three-domain model based on molecular properties.

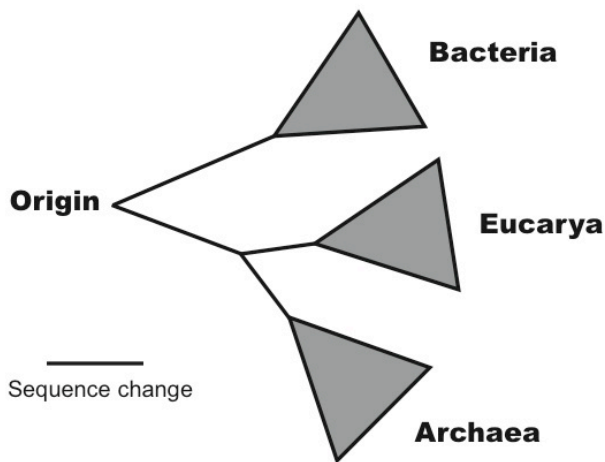
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A. Procaryote-Eucaryote Model



B. Three-Domain Model



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