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I. Taxonomy, ecology, isolation and identification

Aeromonas taxonomy

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Key words. Aeromonas; taxonomy.

The taxonomy of the aeromonads has been the subject of scrutiny and reclassification by several investigators in an attempt to clarify the position of this group of organisms and their relationship to similarly described but obviously separate genera. Recommendations have varied from recognition of one genus comprising three species to the proposal of a complex subspeciation[3, 10]. Other than the obvious need for accurate nomenclature and classification, there are pragmatic reasons to seek improvement in the classification of *Aeromonas,* e.g., in clinical medicine, where physicians tend to refer to all aeromonads as *Aeromonas hydrophila* and, in fact, most of the early medical literature concerning *Aeromonas* used this nomenclature only. We now know such simplification to be unrealistic in light of Popoff and Véron's contributions[9], which showed that *A. hydrophila* and *A. punctata* constitute a single species, *A. hydrophila.* A second, previously undescribed group of strains was also described and named *A. sobria.* The human medical significance of *A. sobria* was demonstrated when it was isolated from a scuba diver's wound infection[5], and in a study of the comparative occurrence and virulence of *A. hydrophila* and *A. sobria* from human and environmental sources[2].

The species on which there is agreement is *A. salmonicida.* It differs from other aeromonads as follows: non-motile, no growth at 37 °C, produces a brown, water-soluble pigment on nutrient agar. In Bergey's Manual of Systematic Microbiology[7] this species is subspeciated into *salmonicida, achromogenes,* and *masoucida.*

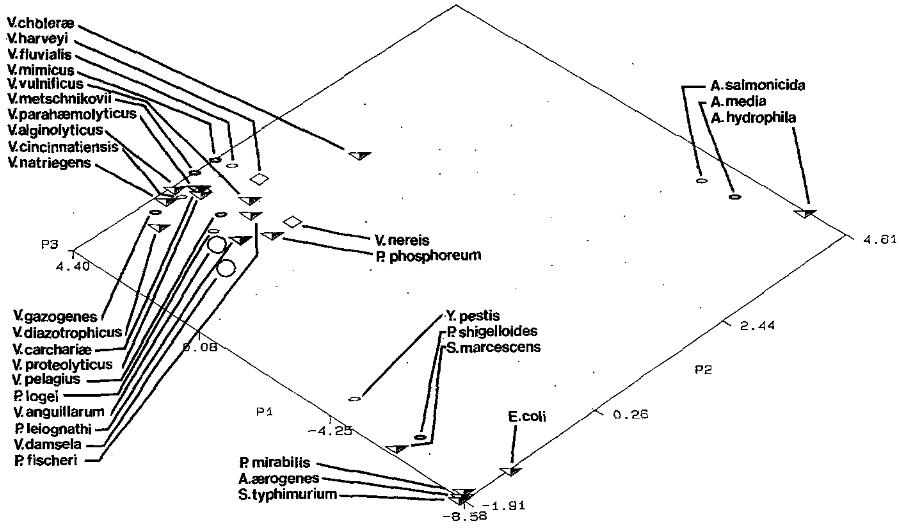
Based upon differential, phenotypic, and genetic characteristics, Popoff and Véron recommended establishment of a third species, *A. caviae,* which is included amongst the motile aeromonads[8]. In summary, five species and three sub-species of *Aeromonas* are recognized at the time of this writing.

Definition

Aeromonas consists of straight cells, rod-shaped and with rounded ends to coccoid. Resting stages are not known. Generally motile by a single flagellum. Some species are non-motile. Metabolism of glucose is both respiratory and fermentative. Oxidase positive and resistant to the vibriostatic agent 2,4,diamino-6,7-diisopropylpteridine (0/129). The mol% G + C of the DNA is 57–63 (Bd,T_m).

Minimal identifying and differential characters

The common characteristics which define the motile aeromonads are possession of motility, morphology, growth in nutrient broth at 37 °C, indole production, fermentation of sucrose and



Overhead view of a four-dimensional plot of evolutionary relationships amongst species of the gamma subdivision of the Rhodobacteria[6, 11]. In this projection, principal components 1 and 2 are given by the X and Y axes, respectively, while principal component 4 is indicated through the use of symbols. Principal component 3, i.e., the Z axis, is lost in this projection.

mannitol, breakdown of inositol, and oxidase activity (table 1). Differential characteristics for the motile aeromonads include esculin hydrolysis, growth in KCN broth, L-histidine and L-arginine utilization, L-arabinose utilization, fermentation of salicin, gas from glucose, and H_2S from cysteine (table 2).

DNA analyses

DNA hybridization studies have verified the distinction of the three species, *A. hydrophila, A. sobria* and *A. caviae*. Within these species there are at least seven DNA hybridization groups, three in *A. hydrophila*, two in *A. caviae*, and at least two in *A. sobria*. Thus far these DNA-related new groups have not been distinguished phenotypically one from the other[8].

Similarly, Farmer et al.[4], using DNA hybridization (hydroxyapatite, ^{32}P, 60C), found only 5 of 60 *Aeromonas* strains to be highly related to the type strain of *A. hydrophila* (70% or greater). Similarly, the type strain of *A. sobria* was highly related to only two other strains, suggesting that many strains of the motile, mesophilic group of *Aeromonas* belong to species other than *A. hydrophila, A. sobria*, and *A. caviae*. There appear to be nine to twelve different DNA hybridization groups.

Table 1. Identifying characteristics for motile aeromonads

Catalase	+
Oxidase	+
Motility	+
Morphology	Rods in singles and pairs
Growth in nutrient broth at 37 °C	+
Arginine dihydrolase	+
Ornithine decarboxylase	–
Indole production	+
Fermentation:	
sucrose and mannitol	+
dulcitol, rhamnose, xylose,	–
raffinose, inositol, and adonitol	–
Breakdown of inositol	–
NO_3 reduction to NO_2	+
Growth in peptone H_2O without NaCl	+
0/129 resistance	+
Starch, gelatin, ONPG, RNA and DNA hydrolysis	+
Tween 80 esterase	+

Table 2. Minimal identifying characteristics for the motile aeromonads

Characteristics	*A. hydrophila*	*A. sobria*	*A. caviae*
Esculin hydrolysis	+	–	+
Growth in KCN broth	+	–	+
L-histidine utilization	+	–	+
L-arginine utilization	+	–	+
L-arabinose utilization	+	–	+
Fermentation of salicin	+	–	+
Gas from glucose	+	+	–
H_2S from cysteine	+	+	–

5S RNA sequence analysis

Results of 5S RNA sequence analysis showed that *Aeromonas* spp. should be included in a family, Aeromonadaceae, separate from the Enterobacteriaceae and the Vibrionaceae[1]. Molecular genetic information compiled to date, including results of 16S ribosomal ribonucleic acid cataloging and 5S ribosomal ribonucleic acid sequence analyses, suggest that *Aeromonas* demonstrate an evolutionary divergence which is significantly at variance with that of other members of the Vibrionaceae[6]. An indication of the phylogenetic diversity among vibrios, aeromonads, and enterics can be gleaned from the figure.

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0014-4754/87/040349-02$1.50 + 0.20/0

Plesiomonas: Taxonomy, classification and enterotoxin production

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Key words. Plesiomonas taxonomy; *Plesiomonas* classification; *Plesiomonas* enterotoxin; *Aeromonas/Plesiomonas.*

Encountered in clinical specimens and the natural environment, *Plesiomonas shigelloides* has been implicated as a potential diarrheal agent in man, but has been overshadowed in importance by related bacteria in the genera *Aeromonas* and *Vibrio*, resulting in its neglect. Consequently, a good deal of uncertainty remains with regard to the taxonomy, ecology and pathogenicity of this member of the family Vibrionaceae. In fact, recent phylogenetic data, based on 5S rRNA sequencing, suggest a closer relationship of *P. shigelloides* with the Enterobacteriaceae rather than the Vibrionaceae. The closest relationship amongst the genera of the Enterobacteriaceae for *P. shigelloides* is with *Proteus mirabilis*[2,3]. Furthermore, a serological relationship between *P. shigelloides* and *Shigella* has been reported[6]. To clarify the taxonomy of *P. shigelloides*, phenotypic characterization was carried out, emphasizing extracellular enzyme profiles. Several investigators have reported production of enterotoxin(s) by *P. shigelloides*[1,4]. Enterotoxin of *P. shigelloides* is not believed to be related immunologically to cholera toxin (CT). We have deter-