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Identification of *Aeromonas* strains of clinical origin by the Omnilog system

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ABSTRACT

The genus Aeromonas belongs to the family Aeromonadaceae and the class of γ-roteobacteria. The identification of some species is problematic because the classification into phenotypic groups on the basis of conventional biochemical tests differs from that obtained in genetic groups. Given this situation, the purpose of our study was to identify 108 strains of Aeromonas original clinical, biochemical basis, through the system Omnilog.

Key words: *Aeromonas*- identification- environnement- Omnilog-clinical.

INTRODUCTION

The genus *Aeromonas* belongs to the family Aeromonadaceae and the class of γ-roteobacteria (Harf_Monteil and Monteil, 2007). It includes 17 species distributed. *Aeromonas* species are widely distributed in aquatic environments (Abbey and Etang, 1988a; Abbey and Etang, 1988b; Nam and Joh, 2007) in freshwater (Wu and *al.*, 2007) and wastewater (Sechi and *al.*, 2002). They are also detected in various foods (Clity and *al.*, 2003). Some species are pathogens strict or opportunistic pathogens for humans in whom they are responsible for many infections (Laganowska and Kaznowski, 2004; Nam and Joh, 2007; Sechi and *al.*, 2002; Watts and *al.*, 2000). The three *Aeromonas* species of clinical interest and causing more than 85% of human infections are *Aeromonas hydrophila*, *Aeromonas caviae* and *Aeromonas veronii bv sobria* (Harf_Monteil and Monteil, 2007; Janda, 1991; Janda and *al.*, 1996). The identification of some species is problematic because the classification into phenotypic groups on the basis of conventional biochemical tests differs from that obtained in genetic groups (Junda and *al.*, 1996). Given this situation, the purpose of our study was to identify 108 strains of *Aeromonas* original clinical, biochemical basis, through the system Omnilog.

MATERIALS AND METHODS

2.1.Collection strains

One hundred and eight strains of clinical origin collected through the collaboration of the College of Bacteriology Virology and Hospital Hygiene, which includes many hospitals in

France, were studied. These strains were grown on agar sheep blood BioMérieux, Marcy l'Etoile, France) for 24 hours at 37° C. Their belonging to the family and aeromonadaceae genus *Aeromonas* has been verified by various biochemical tests:

Gram staining, oxidase test, type and respiratory resistance vibriostatique compound O/129. The strains were then stored at -80° C in brain-heart broth containing 25% glycerol.

2.2 Identification of strains by Omnilog

The BIOLOG system (BIOLOG, Hayward CA, USA) for biochemical identification of microorganisms has been used on all strains studied. The BIOLOG system relies on the ability of a strain to metabolize each substrate. This ability is measured by the presence or absence of a violet color over time. This coloration is due to oxidation of tetrazolium violet by cellular respiration of bacteria. The protocol was performed as specified by the supplier "organic" by the laboratory AES Chemunex (Bruz, France).

Two successive subcultures of the strains were made on agar sheep blood BioMérieux, Marcy l'Etoile, France). A bacterial suspension was performed in 12 mL broth inoculation (GN2/GP2, BIOLOG, and Hayward CA, USA) to obtain an inoculum turbidity equivalent to 52% (50-55) transmission measured turbidity. Microplates of 96 wells ready for use and composed of 95 carbon substrates were inoculated with 150 µL of inoculum per well. The incubation was done at 30 ° C for 22 h. The plates were read from the fourth hour of incubation and steadily during the 22 h. The result of the identification was obtained by comparing the profile of each strain with the profile of strains of each species of *Aeromonas* in this database. The results were recorded on the Software ID BIOLOG computer and printed at the end of the confirmation of the species.

RESULTS AND DISCUSSION

The results of the identification Omnilog showed that the 108 strains of clinical origin, 31 (29%) were identified as belonging to the species *A.hydrophila*, 26 (24%) in the case A.caviae 17 (16%) in the case *A.veronii*, 12 (11%) in the case *A.media*, 10 (9%) in the case *A.allosaccharophila*, 5 (4.5%) in the. *A veronii bv sobria* species, 3 (2.5%) in the case *A.sobria*, 2 (2%) in the case *A.enteropelogenes*, 1 (1%) in the case *A.encheleia* and 1 (1%) in the case *A.jandaei*. The biochemical identification of *Aeromonas* strains was based on 2 systems identification: the system of the API 20E gallery made previously in the laboratory and in the context of our study, the system Omnilog.

According to the results and referring to the identification by sequencing the gene rpoB (β subunit RNA polymerase), the identification Omnilog able to identify 71.5% of clinical strains of *A. veronii*. Strains of this species are identified *by the* Api 20E gallery as belonging to the group of species *A.hydrophila / A.caviae / A.sobria1 and A.hydrophila* / *A*.caviae / *A.sobria*2 with a clinical strain identified as belonging to *Vibrio*.

To the strains of *A. hydrophila,* identification Omnilog gives over 75% of clinical strains. These strains are identified by the gallery Api20E as belonging to the group of species *A.hydrophila*/*A.caviae*/*A.sobria1* and *A.hydrophila* / *A.caviae*/*A.sobria2*, and 2 unidentified strains. Strains in a clinic. *Caviae* are identified by the system Omnilog over 86%, while identifying the gallery Api20E identify them as belonging to the group of species *A.hydrophila* /

A.caviae / *A.sobria1* and *A.hydrophila*/*A.caviae*/*A.sobria2*, and 6 strains as belonging to the genus *Vibrio*.

All these results show that the identification of *Aeromonas* appears problematic in using the system of the gallery Api20E. Indeed, the system software Api 20E includes only very few species of *Aeromonas* (*A.hydrophila, A.caviae, A.sobria, A.salmonicida subsp salmonicida*) unlike Omnilog system. The system seems Omnilog outperform gallery Api20E is what corresponds to the results of the study by Watts and *al*., (2000). Furthermore, reading is rapid (4-22h), but the system remains costly for a laboratory of medical bacteriology. Compared to the molecular identification, the system Omnilog is less efficient because its database does not systematically change which corresponds to the results of studies by Watts and *al*., (2000) and De Paolis and Lippi, (2008) that worked on *Corynebacterium* and *Bacillus*

CONCLUSION

The biochemical identification by the system allows the identification Omnilog the status of the case and gives the result with a detailed data sheet on the strain identified (probability, percentage of similarity and distance with existing strains in the database software that is updated after each use of the system).

Despite this, the system Omnilog we have not identified a number of strains studied mostly they are of clinical interest. In addition, the system Omnilog remains a major investment and costly for a laboratory routine.

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