Studies on the Vibrios and the Pseudomonas living in the Kinko Bay I : On the Similarity Value

Mem. Fac. Fish., Kagoshima Univ.
Vol. 21, No. 1, pp. 109~112 (1972)

Studies on the *Vibrios* and the *Pseudomonas* living in the Kinko Bay. I
On the Similarity Value

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Summary

In the previous paper, the authors[1] reported the isolation method of the bacteriophages living in the Kinko Bay, describing some characters of the host bacteria. The greater part of the organisms was classified as either *Pseudomonas* or *Vibrios*. In the study, the similarity between the individuals among those organisms was mainly discussed. The results obtained were as follows:

1) Similarity values calculated between A-4 and A-6; A-6 and B-4; A-4 and B-4, classified as *Pseudomonas* respectively, were in the range from 81% to 90%.

2) Of the three strains, A-4 was markedly similar to *1055-1* strain recognized as marine *Pseudomonas*, isolated at 694 m depth in North Pacific Ocean, their similarity values being above 93%.

3) The similarity value between the both strains B-5 and B-8, was above 90%. Similarly, that between the both strains B-1 and *Vibrio parahaemolyticus* was also above 90%, while the value between the former two strains and the latter two strains was only 70% or so.

4) The B-1 is assumed to be a *Vibrio*, which was not ascertained in the previous study. In this examination, it was found to be markedly similar to the *V. parahaemolyticus*. Its similarity value was caluculated to be 93%, therefore B-1 is to be determined as a biotype of *V. parahaemolyticus*.

In the previons paper, of six isolates only B-1 was not identified by Bain and Shewan's classification because it had peritrichous flagella. However, in this examination, B-1 was assumed to be a *Vibrio* judging from its high similarity, reaching 93%.

According to the Figure made by the similarity, the six isolates A-4, A-6, B-4, B-5, B-8, and standard organisms such as *1055-1* and *V. parahaemolyticus*, among them *1055-1*, A-4, A-6, and B-4 were *Pseudomonas;* B-5 and B-8 were *Vibrios*, and B-1 was assumed to be a biotype of *V. parahaemolyticus*, respectively.

Experimentals

Organisms: The six isolates from the Kinko Bay were studied. They were all attacked by the bacteriophages. The details of their isolation and some of the characters were reported in another paper.[1]

Determination of properties: One hundred properties of each strain were examined. The hundred articles examined were shown in Table 1. All examinations were carried out by using the medium containing 3% NaCl; while the examinations listed

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Table 1. Properties used in the characterization of the isolates from the Kinko Bay

General

Gram stain
Growth on SS agar
Neutral red test
NO_3 reduction
Indole production
Cytochrome oxidase
H_2S production
Pigment production
Luminescence
Lysis by SLS *1
Chitine decomposition*3
Starch hydrolysis
Phenylalanine deaminase
MR test
Enterotoxin sensitivity *2
Growth on McConkeys
Growth on Koser's citrate
Growth on B. G. agar
Litmus milk
V. P. test
NH_3 production
Swarming
Haemolysis
Gelatin liquifaction
Catalase
Urease
Lysozyme *4

Morphology

Form and size
Motility
Colony form
Colony color
Flagellation

Physiology

Growth in NaCl; 0, 0.5, 1.0, 3.0, 6.0, 7.5, 10.0, and 12.0%
LiCl-replacement with NaCl
KCl-replacement with NaCl
Growth inhibition by various pH; 4.5, 5.4, 8.0, and 9.0.
Growth at various temperatures; 0, 20, 30, 37, 45, and 55 C.

Metabolism

Organic acids: Lactic acid, oxalic acid, succinic acid, Malic acid, Citric acid, Tartaric acid, and Acetic acid.
Amino acids: Tryptophan, Ornithine, Glysine, Arginine, Lysine, Cystein, Glutamic acid, Histidine, and Proline.
Carbohydrates: Arabinose, Serobiose, Fructose, Lactose, Glucose, Multose, Mannose, Rhamnose, Sucrose, Xylose, Inuline, Inosit, Cellulose, Sorbit, and Galactose.

Antibiotics: Penicillin, Oleandomycin, Chloramphenicol, Dihydrostreptomycin, Colistin, Erythromycin, Leucomycin, Tetracycline, Kanamycine, Sulfaisoxazole, Sulfaguanidine, Sulfamide, Romedin, Homosulfamide, Sulfaisomidine, 2-4-di-amino-6-7-diisopropyl pteridine. *5

with asterisks were done as follows; *1 SLS-lysis: SLS-lysis of the test organisms was judged by the measurement of turbidity decrease occassioned by the addition of 0.05 ml of 0.5 Mol SLS aquatic solution into 7 ml of the culture, after 24 hour incubation. *2 Enterotoxin: Enterotoxin producing *Staphylococcus aureus* was used. The activity of the toxin was examined by the same method as applied usually in the disc method on the antibiotics. *3 Chitine decomposition: This

was examined according to the method published by Zo'Bell and Rittenberg[2] *4 Lysozyme lysis: This was done mainly due to the biochemical method[3]. *5 Growth inhibition by the antibiotics: This was all determined by the discs purchased from Eiken Kagaku Co. LTD.

The similarity value was calculated by the following equation; %s=NsP (NsP +Nd) × 100 where %s=Similarity coeffecient; NsP=number of similar positive matches; and Nd=number of disimilar matches.

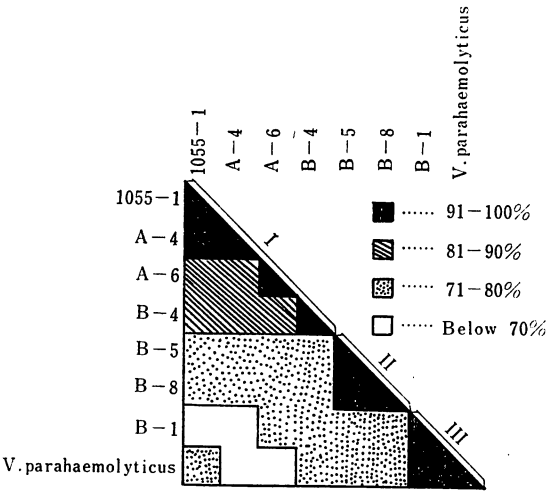

Fig. 1. The similarity values obtained by the use of one hundred features, of 8 strains; 6 isolates from the Kinko Bay and two standard strains *1055-1* and *V. parahaemolyticus* respectively
I *Psendomonas*
II *Vibrios*
III *V. parahaemolyticus* and its biotype

Results and Discussion

The similarity values %s of each strain was calculated and the results obtained were shown in Figure 1. As shown in the Figure, the similarity value between A-4, A-6, B-4, and *1055-1* respectively was above 85 %. On the other hand, among the reciprocal strains of *Vibrios*, both B-5 and B-8 demonstrated high similarity of above 90 %; whereas, the similarity value between these strains and B-1 was only 70 % or so. B-1 was much similar with *V. parahaemolyticus* used for standard strain, similarity value being 93 %. In the examination, the authors have demonstrated that some marine isolates are capable of living in the sea covering a wide area, that is, the A-4 harvested in the sea shore near the Kinko Bay is so much similar to *1055-1* harvested in the North Pasific Ocean.

Considering the tide current mixed with both the Kuroshio and the Oyashio in anywhere of the ocean, such a wide ranged bacterial distribution seems to be of great interest.

Referrences

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