

PSEUDOMONAS PSEUDOMALLEI IN THE SURFACE WATER OF SINGAPORE

By R. N. T. Thin*

and

M. Groves**, G. Rapmund** and M. Mariappan**

INTRODUCTION

The Gram negative rod *Pseudomonas pseudomallei* causes melioidosis which is endemic in South East Asia. Although the first report of this disease was published in 1912 by Whitmore and Krishnaswamy, the epidemiology is not yet clearly defined. Recent work indicates that *Ps pseudomallei* is a normal inhabitant of the environment (Strauss, Groves, Mariappan and Ellison, 1969). The micro-organism has been isolated from soil and surface water in South Vietnam (Chambon, 1955), Northern Australia (Laws and Hall, 1964), East Malaysia (Strauss, Jason and Mariappan, 1967) and West Malaysia (Strauss, Groves, Mariappan and Ellison, 1969). This paper reports the first isolation of *Ps pseudomallei* from surface water in Singapore. The work was carried out in 1967 following the diagnosis of melioidosis in a patient in Singapore (Thin, Brown, Stewart and Garrett, 1970).

METHOD

One hundred and thirty six specimens of surface water were collected in the city of Singapore and in the southern part of the island. After two small pilot collections most (80%) of the samples were collected over a two week period. Twenty-one specimens were collected from drains serving an area of forest, 44 from puddles and drains around buildings, 43 from puddles and drains in roads and streets, and 28 specimens were taken from puddles on sports fields.

The samples were collected in sterile containers and sent to the United States Army Medical Research Unit at the Institute for Medical Research in Kuala Lumpur. There they were examined by the hamster inoculation method described by Ellison, Baker and Mariappan (1969). Five weanling hamsters were inoculated intraperitoneally with 2.0 ml. from each water sample. Heart blood from all hamsters dying between the first and seventh day after inoculation was plated on to nutrient agar containing 3% glycerol and

1:200,000 crystal violet, and incubated at 37 Centigrade for 48 hours. The identity of colonies showing typical morphology of *Ps pseudomallei* was further assessed by growth on MacConkey's agar and failure to grow on SS agar. Subsequently identification was confirmed at the Division of Veterinary Medicine, Walter Reed Institute for Research, Washington D.C.

RESULTS

Ps pseudomallei was isolated from 8 of the 136 specimens. One positive specimen was from a stream draining forest near Dunearn Road and two were from storm drains, one in Orchard Road and one in Nassim Road. Five positive samples came from low lying sports fields near Ayer Raja Road. With the exception of the first specimen they were all collected on the same day shortly after heavy rain had started to fall.

TABLE I
DETAILS OF POSITIVE SPECIMENS

Category of Terrain	No. Collected	No. Positive	Percentage
Forest	21	1	4.8
Around buildings	44	0	0.0
Roadside drains	43	2	4.6
Sports fields	28	5	18.0
TOTAL	136	8	5.9

DISCUSSION

As the collection of water samples was not systematic only tentative conclusions may be drawn from these results. However they do show that *Ps pseudomallei* is present in the surface water on Singapore Island, and that the organism was more readily isolated from low lying sports fields than from other categories of terrain.

* British Military Hospital, Singapore.

** Institute for Medical Research, Kuala Lumpur, Malaysia.

West Malaysia has been more extensively studied than other countries (Strauss, Groves, Mariappan and Ellison, 1969). There *Ps pseudomallei* was isolated most often from wet rice fields and cleared areas; forests gave low isolation rates. The low lying sports fields in our study correspond with cleared areas in Malaysia.

Seven of our positive specimens were collected shortly after rain had started to fall. In another study of Malaysia the same group of workers (Strauss *et al.*, 1970) showed that *Ps pseudomallei* was more frequently recovered from surface water in months with heavy rainfall than during dry months. The pattern of positive isolates in Singapore resembles the findings in Malaysia.

Ps pseudomallei is widely distributed in West Malaysia and in the heavily contaminated areas many people are exposed to the organism. Although clinical melioidosis is rare, a serological study of Malaysians (Strauss, Alexander, Rapmund, Gan and Dorsey, 1969) revealed antibodies in a significant number of persons. Antibodies in healthy people are thought to be due to previous melioidosis, and in the population studied the infection was presumed to have taken a mild, unrecognised, self limiting course. Thus though clinical melioidosis is rare, mild forms may be relatively common.

This preliminary study was prompted by the diagnosis of melioidosis in a patient in Singapore. Although only a small area was studied the findings suggest that *Ps pseudomallei* may be widely distributed over the island. Few cases of clinical melioidosis have been reported from Singapore, but the findings in Malaysia suggest that the condition may be more common than is generally realised. It is therefore recommended that melioidosis is considered in the differential diagnosis of any obscure infective condition presenting in Singapore.

SUMMARY

Eight of 136 samples of surface water collected on Singapore Island contained *Pseudomonas pseudomallei*, the organism causing melioidosis. *Ps*

pseudomallei has not been reported from surface water of Singapore before. The micro-organism was found more frequently in samples collected from low lying sports fields than from other types of terrain. Most of the positive specimens were collected shortly after rain had started to fall. These observations are similar to findings in West Malaysia. As melioidosis may be more common than is generally realised, this condition should be considered in the differential diagnosis of any obscure infective illness presenting in Singapore.

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REFERENCES

1. Chambon, L. (1955): "Isolement du bacille de Whitmore a partir du milieu exterieur." Ann. Inst. Pasteur., 89, 229.
2. Ellison, D. W., Baker, H. J. and Mariappan, M. (1969): "Melioidosis in Malaysia I: A method for isolation of *Pseudomonas pseudomallei* from soil and surface water." Amer. J. Trop. Med. and Hyg., 18, 694.
3. Laws, L. and Hall, W. T. K. (1964): "Melioidosis in animals in North Queensland IV: Epidemiology." Aust. Vet. J., 40, 309.
4. Strauss, J. M., Alexander, A. D., Rapmund, G., Gan, E. and Dorsey, A. E. (1969): "Melioidosis in Malaysia III: Antibodies to *Pseudomonas pseudomallei* in the human population." Amer. J. Trop. Med. and Hyg., 18, 703.
5. Strauss, J. M., Ellison, D. W., Gan, E., Jason, S., Marcarelli, J. L. and Rapmund, G. (1970): "Melioidosis in Malaysia IV: Intensive ecological study of Carey Island, Selangor, for *Pseudomonas pseudomallei*." Med. J. Malaya, 24, 94.
6. Strauss, J. M., Groves, M. G., Mariappan, M. and Ellison, D. W. (1969): "Melioidosis in Malaysia II: Distribution of *Pseudomonas pseudomallei* in soil and surface water." Amer. J. Trop. Med. and Hyg., 18, 698.
7. Strauss, J. M., Jason, S. and Mariappan, M. (1967): "*Pseudomonas pseudomallei* in soil and surface water of Sabah, Malaysia." Med. J. Malaya, 22, 31.
8. Thin, R. N. T., Brown, M., Stewart, J. B. and Garrett, C. J. (1970): "Melioidosis: A report of 10 cases." Quart. J. Med., 39, 115.
9. Whitmore, A. and Krishnaswamy, C. S. (1912): "An account of the discovery of a hitherto undescribed infective disease occurring among the population of Rangoon." Indian Med. Gaz., 47, 262.