A New Bacterial Disease on Orchids *Dendrobium sp.* Caused by *Pseudomonas gladioli*[1]

Samerchai Chuenchitt,[2] Wallapa Dhirabhava,[2] Surang Karnjanarat[2]
Dara Buangsuwon[2] and Tsutomu Uematsu[3]

ABSTRACT

A new bacterial disease of *Dendrobium* sp. was observed in the farms of the Nongkham district during the rainy season of 1981. The disease occurred in every orchid farm in this area. The young seedlings and new leaves of mature plants in several varieties of *Dendrobium* sp. were particularly damaged by the disease. The diseased leaves showed generalized water-soaked rot lesions and wet slimy decay symptoms. But in the dry season, dry symptoms prevailed. At first the wet lesions appeared on the leaves and expanded very rapidly. Finally, the infected leaves became yellow or dark brown and dropped off. Nine cultures of the causal bacterium were isolated from the diseased leaves of the seedlings and mature plants. All isolates were pathogenic on *Dendrobium* seedlings causing leaf rot, and the symptoms were identical with those occurring under natural conditions. Examination of the bacteriological characteristics revealed that the causal bacteria were identical with *Pseudomonas gladioli* which causes soft rot disease of *Gladiolus* sp., *Allium* sp., *Iris* sp., and *Freesia* sp. For the determination of species, it was deemed necessary to compare the pathogenicity of the organism with that of a strain of *P. gladioli* by cross-inoculation on *Dendrobium* sp. and *Gladiolus* sp.

INTRODUCTION

In the early rainy season at the end of April of 1981, samples of orchid plants showing evidence of bacterial disease were received from the Bangkok Flower Center in Bangkok. Field observations were made in the Nongkham district which is the largest orchid-producing area in Thailand. Results indicated that the disease was very severe and had thus far been restricted to *Dendrobium* spp. Losses were estimated at 50% or more.

According to the Approved List of Bacterial Names (Skerman et. al., 1980) and the List of Pathovar Names (Dye et. al., 1980), bacterial pathogens reported on orchids are the following four species : *Erwinia carotovora* subsp. *carotovora* (Jones 1901) causing soft rot, *Pseudomonas cattleyae* (Pavaring 1911) causing corm and basal leaf rot, *E. cy-*

1 The paper was conducted under one of the collaborative research projects between the Department of Agriculture, Thailand and Tropical Agriculture Research Center, Japan.

2 Plant Pathology and Microbiology Division, Department of Agriculture, Bangkhen, Bangkok 9, Thailand.

3 Tropical Agriculture Research Center, Tsukuba, Ibaraki, Japan.

pripedii (Hori 1911) causing brown rot, and *P. andropogonis* (Smith 1911) causing firm rot disease. The disease observed in the Nongkham district showed symptoms similar to those of corm and basal leaf rot disease which was described by Ark and Thomas or brown rot disease which was described by Sutton et al.. However, the causal bacterium isolated from the plants in the Nongkham district differed from both pathogens in their morphology, physiology and pathogenicity.

Results of the investigation into the nature of the disease and the bacteriological properties of the causal organism are presented in this paper.

MATERIALS AND METHODS

Bacterial cultures : The nine cultures used in this experiment were isolated from naturally infected seedlings and mature plants of orchids, *Dendrobium* spp. in five orchid farms in Nongkham district during the rainy season of 1981.

Isolation and culture medium : The causal organism was isolated from water-soaked areas of freshly infected leaves and stem of young seedling by means of the dilution agar plate method. The medium used for isolation and subculture was the semisynthetic potato agar medium (PSA) (Wakimoto and Yoshii, 1955). The composition of PSA medium was as follows : One liter of 300 g potato decoction ; $Ca(NO_3)_2 . 4H_2O$, 0.5g; $Na_2HPO_4 . 12H_2O$, 2.0g; peptone, 5.0g; sucrose, 15.0g; agar 12-15g; pH 6.8-7.0.

Pathogenieity tests : The bacteria that had been isolated and stored on PSA slant were allowed to grow for 24 hrs. before use. The healthy *Dendrobium* sp. plants which were grown under the roof and under full sunlight were used for the inoculation tests. They were inoculated by wounding with a five needle-bundle smeared with bacteria. The sterile water from the sterile medium of PSA was used as control instead of the bacterial suspension. Each isolate was inoculated on separate orchid plants, belonging to *Dendrobium* sp. The inoculated plants were covered with plastic bags for 19-20 hrs. They were kept under weak sunlight after the bags were taken out. Disease development was evaluated 2 to 3 days later by measuring the length of the water-soaked lesions that formed at the inoculation site. The rotting ability of the bacteria to potato tubers, onion bulbs, carrot slice, lettuce leaves, lime fruits and *Amallilis* sp. plants was also determined. Slices from mature onion, potato and carrot with sterilized surface were placed on moistened filter paper in petri dishes and inoculated with a loopful of bacteria on the outer surface of the slices. Lettuce leaves removed from the inner parts of head lettuce and lime fruits with sterilized surface were placed in a moist chamber and inoculated by the needle prick inoculation method. All inoculated samples were kept in the dark at 28°C. *Amallilis* sp. plants were inoculated by wounding with a five needle-bundle smeared with bacteria in the same manner as for the orchid plants. The development of a necrotic zone or soft rot lesion of at least 10.0 mm beyond the inoculation

site was considered positive. Pathogenicity test to the other kinds of orchid plants, Vanda princess blue, *Ascocenda* sp., Oncidium golden shower, *Phalaenopsis* sp. and *Rhyncostylis gigantea* (alba) was also covered out in the same manner as for *Dendrobium* sp.

Bacterial characteristics : The Gram reaction was performed according to the method of Ryu (1940). Flagellation and cell form were studied by electron microscopic observation of bacteria from 19-20 hrs.-old culture on nutrient agar (Difco) slant in accordance with the method of Schaad (1980). Colony color and form on YDC and NA, production of fluorescent pigment, growth ability on CVP and D-1 agar were observed as described by Sands et al. (1980). The accumulation of poly-β-hydroxybutyric acid was analysed by the method of Stainer et al.(1966). Oxygen requirement, oxidase activity, arginine dihydrolase, levan production, pectolytic enzyme production, and organic compound utilization were determined as described by Sands et al. (1980). Gelatin liquefaction was performed in gelatin gels in applying the plate method described by Kelman and Dickey (1980). Spore formation, starch hydrolysis, indole production and nitrate reduction were tested by the methods described in the Manual for the Indentification of Medical Bacteria (Cowan and Steel, 1965). The tests for growth ability at 41°C and 4°C were the same as those described by Sands et al. (1980) and temperature relationships such as maximum, optimum and minimum temperatures were analysed with a temperature gradient biophotorecorder (Tokyo Kagaku Sangyo Co. Ltd., Tokyo).

RESULTS

Symptoms : Characteristic symptoms of the disease in the fields were as follows : wet rotting of leaves, poor growth of young seedlings and lack of leaves on mature plants. The lesions were primarily observed on the upper part of the leaves, and infected leaves displayed water-soaked, dark green to yellow lesions, wet rot (Fig. 2), with the wet rot area rapidly expanding to cover the whole leaf. Finally, the infected leaves turned yellowish brown or dark brown (Fig. 3) and fell off. Lesions were usually confined to the leaves only. When severe infection occurred in young shoots, the stem became dark-green, collapsed and the leaf parts above also died. The disease was also seen on young seedlings in nursery beds which showed dark green rot (Fig. 1). Seedling rot spread rapidly and patches of infected seedlings were seen in seedling beds. These symptoms were observed on varieties of *Dendrobium* spp. but were absent on the other kinds of orchid plants. The disease was generally observed in the rainy season and the incidence was low in the dry season in the Nongkham district.

Pathogenicity tests : Water-soaked lesions with dark-green color appeared one day after inoculation around the inoculation site (Fig. 4). This lesion enlarged gradually, turned yellow or yellowish brown, and finally showed wet decay all over the leaf similar to that found in natural infection (Fig. 5). Inoculation on stem of young seedlings also produced dark-green to brownish, water-soaked lesions (Fig. 6) which had

increased in size and caused die-back of the infected shoots at temperatures of 30-32° and under high moisture conditions.

Other kinds of orchid plants such as *Rhyncostylis gigantea* (alba), *Phalaenopsis* sp., Vanda princess blue, *Ascocenda* sp., and Oncidium golden shower seemed to be resistant to this bacterium in comparison with *Dendrobium* sp. However, *Phalaenopsis* sp. and *Rhyncostylis gigantea* (alba) showed water-soaked symptoms in the initial stage but later on the lesions became dark brown and did not expand. Vanda princess blue, *Ascocenda* sp. and Oncidium golden shower also showed brown to dark brown necrotic lesions without water-soaked symptoms at the inoculation sites. These necrotic (or dry rot) lesions did not expand to cover the whole leaf. They were limited to the adjacent area around the inoculation point with a size of below 1cm in diameter in the dry season.

All the isolates tested caused rot of onion bulbs, potato tubers, lettuce leaves, carrot slices and leaves of *Amallilis* sp. The rotted area expanded slowly on all specimens. None of the isolates caused rot on lime fruits (Table 1).

Bacteriological characteristics : The bacterial isolates used were Gram-negative, aerobic, rod shaped, motile with 1-2 bipolar flagella, spores and capsules were not formed. On NA medium they produced opaque colonies which were surrounded by a full margin, had a smooth surface and were pale yellow in color reaching 2-3 mm diam. after 3 to 4 days. On YDC agar medium, the isolates produced brownish yellow colonies. They could not grow on CVP and D-1 agar media. They grow in peptone water and yeast extract broth, and the minimum, optimum and maximum temperatures for growth were 8-10°C. 32-34°C and 41-42°C in these liquid media respectively. All produced diffusible green pigment in King's B agar but no fluorescent pigment. Poly -β-hydroxybutyrate inclusions were observeb. Oxidase was positive. Indole, arginine dihydrolase production and nitrate reduction were negative. Levan, pectolytic enzyme and arbutin hydrolase production were positive. Gelatin was liquefied but starch was not hydrolyzed. All cultures utilized glucose, sucrose, D-arabinose, trehalose, L-rhamnose, maltose, cellobiose, lactose, inulin, sorbitol, inositol, erythritol, mannitol, betaine, n-propanol, L-lactate and D-tartrate. Levulinate was not utilized by any of the isolates, and malate, geraniol and β-alanine were not utilized by some isolates (Table 1).

DISCUSSION

High incidence leaf and seedling rot of orchid plants, *Dendrobium* spp. has been very severe in many orchid farms in Thailand. Field observations showed that the disease occurrence and spread seemed to depend largely on the temperature and moisture conditions because the disease occurred heavily only under warm and high moisture conditions such as those prevailing in the rainy season. Very few definite cases of the disease were observed in the dry season. At

present, however, the conditions affecting the disease development remain to be elucidated.

Microscopic observations showed that the diseased specimens appeared to have abundant bacteria in the advancing margin of the water-soaked lesions. Thus for the following bacteria have been found to cause orchid diseases : *P. cattleyae* causing leaf spot and bud rot on *Cattleya* spp. and *Phalaenopsis* spp. (Ark and Thomas, 1946), *E. cypripedii* causing brown rot on *Cypripedii* spp. (Sutton et. al., 1960); *E. carotovora* subsp. *carotovora* causing soft rot of *Cattleya* spp. and *Phalaenopsis* spp. (Matsumoto and Okabe, 1931); *P. andropogonis* causing firm rot on *Phalaenopsis*, *Dendrobium* and *Vanda* spp. (Oshiro et. al., 1964) In Thailand, the occurrence of bacterial soft rot caused by *E. corotovora* subsp. *carotovora* has been recognized on *Phalaenopsis* spp. (Chandrasrikul, 1977; Saifa, 1975). The symptoms observed in the Nongkham district were remarkably similar to those of bacterial diseases such as leaf spot and bud rot which were reported by Ark and Thomas(1946) and brown rot which was reported by Sutton et al.(1960). However, the present disease occurred on a different genus, *Dendrobium* spp. All isolates which had shown a pathogenicity on *Dendrobium* sp. were definitely placed in the genus *Pseudomonas* based on the analysis of bacterial characteristics (Table 1 - 1). According to the key to the genus *Pseudomonas* defined in Bergey Manual and to Hildebrand et al. (1973) and Sands et al. the results obtained in the current study suggest that the isolates belong to *P. gladioli* with in the non-fluorescent group of pseudomonads, and that they are different from *P. cattleyae* and *P. andropogonis*, as compared with the description of Ark and Thomas(1946), Goto and Starr(1971), respectively(Table 1). The isolates from *Dendrobium* spp. differed in the utilization of some organic compounds from the description by Hildebrand et al. (Table1-2). Differences in the utilization of organic compounds might be due to the range of variations of bacterial strains such as those reported by the authors (Mishagi and Grogan, 1969 ; Sands et. al, 1980) who described similar variations among the isolates of other phytopathogenic pseudomonads.

P. gladioli which was first reported on *Gladiolus* spp. caused rot of gladiolus corms (Severini, 1913). *P. marginata* and *P. alliicola* which are considered as the synonyms of *P. gladioli*had been isolated from decayed onion, *Gladiolus* spp. and *Iris* spp. to which they were thought to be pathogenic. *P. gladioli* had been also isolated from *Freesis* spp. causing neck rot symptoms (Tsuchiya and Muko1963). Cross inoculation tests to host plants using the present isolates and taxon strains should be performed for the identification of the pathovar. At present, cross inoculation tests were not performed due to the difficulty in growing the *Gladiolus* spp., *Iris* spp. and *Freesia* spp. in the tropics. However, all isolates from *Dendrobium* spp. produced severe rot on onion bulbs, potato tubers etc. (Table 2).

From the above mentioned bacteriological characteristics and pathogenicity tests, it is proposed that the isolates of *Dendrobium* spp. should be considered as *P. gladioli* and that *Dendrobium* sp. is an additional host for *P. gladioli*.

ACKNOWLEDGEMENTS

The authors wish to express their gratitude to Mr. P. Titatarn, former Director-General, Dr. R. Syamananda, Deputy Director-General, Miss A. Chandrasrikul, Director of Plant Pathology and Microbiology Division, Dr. P. Giatgong, Chief of Mycology Branch, Department of Agriculture, Bangkhen, Bangkok, for providing facilities for this study. They also wish to thank Mrs. K. Kamjaipai, Bangkok Flower Center in Bangkok for drawing their attention to the problem and providing samples. Mr. N. Nabheerong., and Dr. S. Tantayaporn, research staffs in Plant Pathology and Microbiology Division for their in valuable cooperation.

LITERATURE CITED

Ark, P.A. and H.E. Thomas. 1946. Bacterial leaf spot and bud rot of Orchids caused by *Phytomonas cattleyae*. *Phytopathology*. 36 : 695-698.

Bergey's manual of determinative bacteriology, 8th ed. 1974. The Williams and Wilkins Comp., Baltimore.

Chandrasrikul, A. 1977. Diseases and pests of Ornamental plants. Bangkok, The Thai Watthana Panich Comp. 163pp. (In Thai).

Cowan, S.T. and K.J. Steel. 1965. Manual for the identification of medical bacteria. 2nd ed. Cambridge Univ. Press 217 pp.

Dye, D.W., J.E. Broadbury, M. Goto, A.C. Hayward, R.A. Elliott and M.N. Schroth. 1980. Pathovar names and pathotype or neopathotype strains approved by the ISPP committee on taxonomy of phytopathogenic bacteria in International standards for naming pathovars of phytopathogenic bacteria and a list of pathovar names and pathotype strains. *Rev. Pl. Pathol.* 59 : 159-168.

Goto, M. and M.P. Starr. 1971. A Comparative study of *Pseudomonas andropogouis*, *P. stizolobii* and *P. alboprecipitans*. *Ann. Phytopathol. Soc. Japan* 37 : 233-241.

Hildebrand, D.C., N.J. Palleroni and M. Doudroff. 1973. Synonymy of *Pseudomodas gladioli* Severini 1913 and *Pseudomonas marginata* (McCulloch 1921) Stapp 1928. *J. Syst. Bacteriol.* 23 : 433-437.

Kelman, A. and R.S. Dickey. 1980. Soft rot or 'carotovora' group in Laboratory Guide for Identification of Plant Pathogenic Bacteria, ed. by Schaad, N.W. *Bacteriology Committee of American Phytopathological Society*. St. Paul. Minesota p. 31-35.

Matsumoto, T. and N. Okabe. 1931. On the causal organisms of Bacterial soft rot of Kotyo-Ran, *Phaenopsis aphrodite* Reich B. f. *J. Soc. Trop. Agr.* 3 : 117-134.

Misaghi, I. and R.G. Grogan. 1969. Nutritional and biochemical comparisons of plant-pathogenic and saprophytic fluorescent pseudomonads. *Phytopathology*. 59 : 1436-1450.

Nishiyama, K., T. Kusaba, K. Ohta, K. Nahata and A. Ezuka. 1979. Bacterial black rot of Tulip caused by *Pseudomonas andropogonis*. *Ann. Phytopathol. Soc. Japan.* 45 : 668-674. (In Japanese with English summary)

Oshiro. L.S., R.B. Hine and S. Goto. 1964. The identification of *Pseudomonas andropogonis* as the cause of firm rot disease of the terete Vanda Orchids in Hawaii. *Pl. Dis. Reptr.* 48 : 736-740.

Ryu, E. 1940. A simple method of differentiation between Gram-positive and Gram-negative organisms without staining. *Kitasato Arch. Expt. Med.* 17 : 58-63.

Saifa, Y. 1975. Studies on bacterial top rot of corn and soft rot of Orchids genus Phalaenopsis. Bangkok : Graduate Studies, Kasetsart Univ. 61 pp. Thesis (M.S. in Agriculture). (In Thai with English summary)

Sands, D.C., M.N. Schroth and D.C. Hildebrand. 1970. Taxonomy of phytopathogenic pseudomonads. *J. Bact.* 101 : 9-23.

Sands, D.C., M.N. Schroth and D.C. Hildebrand. 1980. Pseudomonas in Laboratory Guide for Identification of Plant Pathogenic bacteria, edited by Schaad, N.W. *Bacteriology Committee American Phytopathological Society*. St. Paul, Minnesota. p. 36-44.

Schaad, N.W. 1980. Initial identification of common genera in Laboratory Guide for Identification of Plant Pathogenic Bacteria, ed. by Schaad, N.W. *Bacteriology Committee of American Phytopathological Society*. St. Paul, Minnesota. p. 1-11.

Severini, G. 1931. Inttornoalle attivita enzimatiche didue bacteria pathogeni per le piante. *Ann. Bot.* 11 : 441-452. (In Rome with English summary)

Skerman, V.B.D., V. Mcgowan and P. H.A. Sneath. 1980. Approved list of Bacterial names. American Society for Microbiology, Washington D.C. 420 pp.

Stainer, R.Y., N.J. Palleroni and M. Doudroff, 1966. The aerobic pseudomonads : a toxonomic study. *J. Gen. Microbiol.* 43 : 159-271.

Sutton, D.D., P.A. Ark and M.P. Starr. 1960. The causal agent of Bacterial brown rot of Cypripedium Orchids. *Phytopathology*. 50 : 182-186.

Tabei, H. and A.J. Quimio. 1978. Bacterial brown spot of Phalaenopsis orchids in the Philippines. *JARO*. 12 (4) : 241-242.

Tsuchiya, Y. and H. Muko. 1963. Occurrence of neck rot disease of *Freesia* sp. in Japan. Chukan hokoku of Plant Pathology Section. *NIAS*. No. 16 : 82-84 (In Japanese)

Wakimoto, S. and H. Yoshii. 1955. Quantitative determination of the population of a bacteria by phage technique. *Sci. Bull. Fac. Agr. Kyshu Univ.*, 15 : 161 - 169. (In Japanese with English summary)

Table 1-1 Characteristics of the isolates from *Dendrobium* spp. as compared with *P. gladioli*, *P. cattleyae* and *P. andropogonis*.

Character	Present isolates[a]	Pseudomonas gladioli (H)[b]	(S)[c]	(T)[d]	cattleyae (A)[e]	andropogonis (G)[f]
Gram reaction	−	−	−	−	−	−
Spore formation	−	−	−	−	−	−
Oxygen requirment	+	+	+	+	+	+
Fluorescent pigment production	−	−	−	−	−	−
Poly-β-hydroxybutyrate	+	+	+			
Arginine hydrolase production	−	−	−			
Nitrate reduction	−	−	−	−	+	−
Oxidase activity	+	+	V			
Indole production	−		−	−	−	−
Gelatine liquefaction	+				−	−
Starch hydrolysis	−	−			+	+
Pectolytic enzyme production	+	V				
Levan production	+					−
Arbutin hydrolysis	+					
Growth at 41°C (Max. temp.)	+(42°C)	+	+			−
Optimum temperature	32–34°C	30–35°C			25°–35°C	25–30°C
Growth at 4°C (Min. tempt.)	−(8–10°C)	−				
Capsules	−	−				
Size	ND	0.6 by 2.3 to 2.8 um			0.4 to 0.6 by 2.4 um	0.8 by 1.9 um
Flagella	One or two bipolar	One or more bipolar			One or two bipolar	One
Pigment on NA media	pale yellow	plae yellow occasional orange, and certain substrates reddish.			Grayish white	White
Colony color on YDC agar	Yellowish brown	Yellowish brown				
Growth on CVP and D-1 agar	−	−				

Table 1-2 Utilization of organic compounds by the isolates from *Dendrobium* spp. as compared with *Pseudomonas gladioli*, *P. cattleyae* and *P. andropogonis*

Compounds tested	Present isolates[a]	Pseudomonas gladioli (H)[b]	(S)[c]	(T)[d]	cattleyae (A)[e]	andropogonis (G)[f]
Carbohydrates						
Glucose	+	+	+	+	+	+
Sucrose	+	+	+	+	+	−
D-Arabinose	+	+	+	+	+	
Trehalose	+	+	+	+		
L-Rhamnose	+(W)[h]	−	−			V
Maltose	+(W)	−		+		−
Cellobiose	+	+	+	+		
Lactose	+	−		+	+	+
Inulin	+	−		−		
Alcohol, polyalcohol, and glycols						
Geraniol	V(+4,W)	−				
Sorbitol	+	+	+	+		+
Inositol	+	+				V
Erythritol	+	+				
Mannitol	+	+	+	+	+	+
n-Propanol	+	+	−			
Organic acids						
Malate	(V+2,W)	−				
L-Lactate	+	+				
Levulinate	−	−	−			
D-Tartrate	+	+	+			−
Alphalic amino acids						
β-Alanine	V(+3,W)	+				
Betaine	+					

a) Nine isolates from *Dendrobium* spp.

b) Data from Hildebrand et al.[7]

c) Data from Sands et al[16].

d) Data from Tsuchiya and Muko[23].

e) Data from Ark and Thomas[1].

f) Data from Goto and Starr[6].

g) Variable (Number of positive isolates.)

h) Poor growth.

Table 2 Pathogenicity of the present isolates to various plants as compared with *P. gladioli*, *P. cattleyae* and *P. andropogonis*.

Inoculated plants and organ	Present isolates[a]	Pseudomonas gladioli (B)[i]	gladioli (T)[c]	cattleyae (A)[e]	andropogonis (O)[d]
Orchid plants					
Dendrobium sp.	+				+
Rhyncostylis sp.	+			+	+
Phalaenopsis sp.	+			+	+
Vanda sp.	+				+
Ascocenda sp.	+				
Oncidium sp.	+				
Cattleya sp.	ND[l]			+	
Onion bulbs					
Allium cepa L.	+	+	+		
Freesia plants					
Freesia refracta Klatt.	ND		+		
Amallilis plants					
Amallilis sp.	+				
Tulip plants					
Tulipa gesneriana L.	ND				(+)[k]
Potato tubers					
Solanum tuberosum L.	+		+		
Lettuce leaf					
Lactuca sativa L.	+		+		
Carrot tubers					
Daucus carota L. *sativa* DC.	+		+		
Gladiolus plants					
Gladiolus sp.	ND	+	+		
Iris plants					
Iris sp.	ND	+	+		
Sorghum plants					
Holcus sorghum	ND				
Lemon fruits					
Citrus sp.	_[m]	_			

i) Data from bergey's manual.[2]

j) Data from Oshiro et al.[12]

k) Dato from Nishiyama et al.[11]

l) Not deter mined. m) Lime fruits were used.

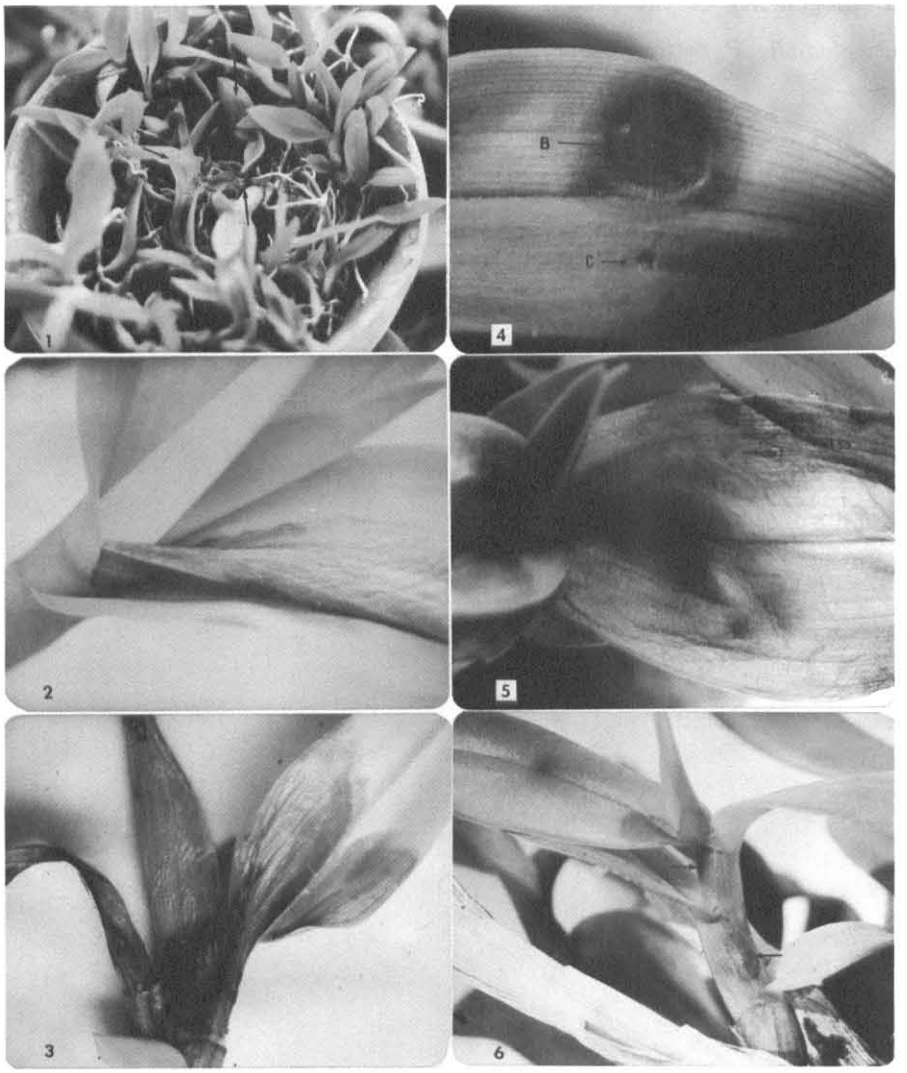

Fig. 1. Wet rot and decayed symptoms on young seedling in the seedling bed. (arrow : disease seedlings)

Fig. 2. Natural symptoms of bacterial leaf rot of *Dendrobium* sp., note the slight yellowish brown soft rot lesion on the leaf.

Fig. 3. Severe decayed symptoms in the field.

Fig. 4. (B) Water-soaked symptom produced by wound inoculation with present isolate on the leaf of *Dendrobium* sp. (20 hrs after inoculation).

Fig. 5. (C) Wet decayed symptom at the advanced stage (4 days after inoculation)

Fig. 6. Water-soaked symptom produced by wound inoculation to the sheath of *Dendrobium* sp. (arrow : inoculation points).