Characterization of Strains of *Pseudomonas maltophilia* Which Do Not Require Methionine

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Nine strains of *Pseudomonas maltophilia* which do not require methionine are described. Methionine-requiring and -nonrequiring strains of *P. maltophilia* belong to different biovars, which are designated I and II, respectively; they differ from each other with respect to phenotypic characteristics, cellular fatty acid composition, deoxyribonucleic acid base composition, and deoxyribonucleic acid-deoxyribonucleic acid hybridization. The relationship of *P. maltophilia* to *Xanthomonas* species is also discussed.

Pseudomonas maltophilia strains have been isolated from a wide variety of sources, including soil, water, foods, and clinical specimens (7). Recently, Debette et al. (2, 3) reported that *P. maltophilia* represented about 25% of the non-fermentative, gram-negative rods isolated from soil in France, and Katoh and Suzuki (17) showed that a considerable number of *P. maltophilia* strains were isolated from manured soils in Japan. Furthermore, in the course of isolation of aromatic compound- and hydrocarbon-assimilating bacteria, Komagata et al. (1, 12) encountered *P. maltophilia* strains, although these strains did not assimilate the compounds. These observations suggest an important ecological role for this species in the element cycle in nature. On the other hand, it is known that *P. maltophilia* is an opportunistic pathogen of humans (7). Thus, taxonomic studies of *P. maltophilia* are required for a better understanding of the function of this species in nature.

The requirement for methionine or other sulfur-containing amino acids by *P. maltophilia* was first reported by Iizuka and Komagata (12) and was confirmed by other investigators (6, 33, 35, 42). Although this characteristic has been used as an aid in the identification of strains of this species, Hugh and Gilardi (7) reported that strains of *P. maltophilia* which do not require methionine accounted for approximately 4% of the strains that he studied. We found strains of *P. maltophilia* that do not require methionine among strains held by culture collections as flavobacteria. Katoh and Suzuki (17) also reported the isolation of such strains of *P. maltophilia* from soils. Therefore, the requirement for methionine should be interpreted cautiously in the identification of pseudomonads.

This report deals with the identification of nine strains of *P. maltophilia* that do not require methionine on the basis of phenotypic charac-

teristics, cellular fatty acid composition, ubiquinone system, deoxyribonucleic acid (DNA) base composition, and DNA-DNA hybridization.

MATERIALS AND METHODS

Bacterial strains. We studied the type strain of *P. maltophilia* KS 0001 (=ATCC 13637) (8), *P. maltophilia* KS 0002 (=ATCC 17806 = IAM 1554), which is the type strain of *Pseudomonas melanogenum* (sic) (a later subjective synonym of *P. maltophilia* [9, 21]), *Flavobacterium rigense* KS 0129 (32), *Flavobacterium rhenanus* KS 0130 and KS 0131 (32), and *Flavobacterium ferrugineum* KS 0132 (19). Strains KS 0186 and KS 0189 were isolated from a phenol enrichment culture by Ikemoto and Komagata, and strains KS 0193, KS 0194, KS 0195, KS 0196, and KS 0197 were isolated by Katoh and Suzuki (17), National Institute of Agricultural Sciences, Tokyo, Japan, from manured soils. *Xanthomonas* sp. KS 0053, first reported as an aromatic compound-utilizing bacterium (1), was identified as a strain of *P. maltophilia* in this study. One strain each of *Xanthomonas campestris*, *Xanthomonas citri*, and *Xanthomonas oryzae*, which were obtained from the National Institute of Agricultural Sciences, Tokyo, were employed as controls. Brief histories and several corresponding culture collection designations of these strains are listed in Table 1.

Identification methods. The characteristics of the strains were determined by previously described methods (21). Poly- β -hydroxybutyrate accumulation was observed by the method of Williamson and Wilkinson (37). Nucleoside phosphotransferase was determined by the thin-layer chromatography procedure (4, 18) instead of the electrophoresis method (20, 24). The assimilation of carbon compounds was determined in the basal medium of Stanier et al. (35) with and without the addition of DL-methionine (50 mg/liter). Growth patterns in defined media with and without methionine were determined with a Bio-Log apparatus (Duan, Paris, France).

Cellular fatty acid composition. Cells cultured in nutrient broth at 30°C for 24 h with shaking were used for analysis of cellular fatty acids. Cellular fatty acid composition and similarity on the basis of com-

position were determined as described previously (13, 14). On the basis of these similarity values, a dendrogram was drawn by a computer according to the arithmetic average linkage-clustering method (34).

Ubiquinone system. Ubiquinones were determined by the method of Yamada et al. (38).

DNA base composition. Purified DNA was extracted from cells grown in nutrient broth at 30°C for 24 h with shaking by the method of Saito and Miura (31) with minor modifications. DNA base composition was calculated from melting temperature (23), as determined in 0.1× SSC (1× SSC is 0.15 M NaCl plus 15 mM trisodium citrate, pH 7.0) with a Gilford model 250 spectrophotometer. DNAs from *Micrococcus luteus* IAM 1056 (=ATCC 4698) and *Escherichia coli* IAM 1268 (=strain B = ATCC 11303), which contain 72.0 and 50.9 mol% guanine plus cytosine, respectively, were used as references.

DNA-DNA hybridization. DNA-DNA hybridization experiments were carried out at 62°C by a previously reported method (16). Labeled DNA was extracted from cells grown in the presence of [6-³H]-thymidine (10 µCi/ml). The heat stabilities of the hybrids were determined by heating the filters after counting at 75°C in 2× SSC containing 0.1% sodium dodecyl sulfate for 1 h, followed by washing and counting.

RESULTS

Phenotypic characteristics. The strains studied were gram-negative rods which were

motile by means of a tuft of polar flagella. They grew aerobically on nutrient agar, and the growth was yellow. The accumulation of poly-β-hydroxybutyrate was not observed in the cells grown on nutrient agar. Gelatin was liquefied. Catalase was produced. All strains except *F. rigense* KS 0129 produced deoxyribonuclease and lysine decarboxylase. The only strain to produce oxidase was *F. rigense* KS 0129, which was weakly positive. The isomer produced by nucleoside phosphotransferases was 5'-nucleotide. Acid was produced aerobically from various carbohydrates, but the strains were not identical in this regard (Table 2). Table 3 shows the growth patterns of 14 strains on 31 carbon compounds. Growth was observed with some carbon sources only after a rather long incubation time (up to 3 weeks). Methionine-requiring strains grew with malonate and sucrose but not with DL-β-hydroxybutyrate; conversely, strains that did not require methionine grew with DL-β-hydroxybutyrate but not with malonate or sucrose. On the basis of the above-mentioned characteristics and the growth patterns with carbon compounds with and without added methionine, the following were regarded as *P. maltophilia* strains that did not require methionine: *F. rigense* KS 0129, *F. rhenanus* KS 0130 and KS 0131, *F. ferrugineum* KS 0132, and strains KS 0194, KS 0196, and KS 0197.

TABLE 1. Bacterial strains studied

Species	Strain designations ^a					Taxonomic status	Original strain designation	Reference
	KS no.	AJ	ATCC	IAM	NIAS			
<i>P. maltophilia</i>	KS 0001	2082	13637			Type		8
<i>P. maltophilia</i>	KS 0002	2220	17806	1554			<i>P. melanogena</i> YF-3	21
<i>Xanthomonas</i> sp.	KS 0053	2790					R-5-b	1
<i>F. rigense</i>	KS 0129 ^b	2472				Type	F-45 (Shiga)	32
<i>F. rhenanus</i>	KS 0130 ^b	2474					F-11 (Shiga)	32
<i>F. rhenanus</i>	KS 0131 ^b	2475					S-11 (Shiga)	32
<i>F. ferrugineum</i>	KS 0132 ^b	2500					P-187 (Komagata and Ogawa)	19
Isolate	KS 0186						NP9-16-2	
Isolate	KS 0189						KP9-6-2-1	
Isolate	KS 0193						2F5 (Katoh)	17
Isolate	KS 0194 ^b						2K1 (Katoh)	17
Isolate	KS 0195						6I11 (Katoh)	17
Isolate	KS 0196 ^b						6K2 (Katoh)	17
Isolate	KS 0197 ^b						6K8 (Katoh)	17
<i>P. aeruginosa</i>	KS 0025	2116	10145	1514		Type		
<i>X. campestris</i>	KS 0198				⊗ 1-1-1			
<i>X. citri</i>	KS 0199				⊗ 1-2-1			
<i>X. oryzae</i>	KS 0200				⊗ 1-5-1			

^a AJ, Central Research Laboratories, Ajinomoto Co., Inc, Kawasaki, Japan; ATCC, American Type Culture Collection, Rockville, Md.; IAM, Institute of Applied Microbiology, University of Tokyo, Tokyo, Japan; NIAS, National Institute of Agricultural Sciences, Tokyo, Japan.

^b Strains that do not require methionine.

TABLE 2. Acid production from sugars and sugar alcohol by 14 strains of *P. maltophilia*

Sugar or sugar alcohol ^a	Acid production by strain ^b													
	KS 0001	KS 0002	KS 0053	KS 0129	KS 0130	KS 0131	KS 0132	KS 0186	KS 0189	KS 0193	KS 0194	KS 0195	KS 0196	KS 0197
Glucose	+	+ ^w	+	+	+	+	-	+	+	+ ^w	+	+ ^w	+	+
Mannose	+	+	+	+	+	+	+	+	+	+	+	+	+	+
Fructose	+	+	+	+	+	+	+ ^w	+	+	+	+	+	+	+
Sucrose	+	-	-	-	-	-	-	-	-	-	-	-	-	-
Maltose (open)	+	+	+	+	+	+	+ ^w	+	+	+	+	+	+	+
Maltose (closed)	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Cellobiose	+	-	-	-	-	-	-	+ ^w	+ ^w	+ ^w	+	-	+ ^w	+
Lactose	+	-	-	-	-	-	-	-	-	-	-	-	-	-
Xylose	-	-	+ ^w	+ ^w	-	-	-	-	-	-	+ ^w	-	+ ^w	+ ^w
Mannitol	-	-	-	-	-	-	-	-	-	-	-	-	-	-

^a Incorporated into the medium of Hugh and Leifson (8).^b Symbols: +, acid produced; +^w, weak production of acid; -, acid not produced.

It is noteworthy that the requirement for methionine depended upon the carbon compounds employed. Lowe and Ingledew (22) also observed this phenomenon in a strain of *P. maltophilia*. As Table 4 shows, *F. rhenanus* KS 0131 grew without methionine at the expense of fumarate, L-histidine, and citrate but failed to grow without methionine at the expense of L-proline. In addition to these compounds, strains KS 0194, KS 0196, and KS 0197 grew on cellobiose without methionine. The lack of a methionine requirement was confirmed with a Bio-Log apparatus (Fig. 1). The quantity of methionine in the culture medium affected the growth of *P. maltophilia* strains. As Fig. 2 shows, the growth curves of *P. maltophilia* KS 0001 varied with the concentration of methionine. The concentration of methionine for optimal growth also varied from strain to strain. In general, the concentration of DL-methionine required for the growth of methionine-requiring *P. maltophilia* strains seems to be 50 mg/liter or more.

Cellular fatty acid composition. The fatty acids of *P. maltophilia* KS 0001 and KS 0002 were composed mainly of a branched-chain i-C_{15:0} acid, a saturated straight-chain C_{16:0} acid, an unsaturated straight-chain C_{16:1} acid, and an unknown acid (anteiso-C₁₅ ?) (Fig. 3). The cellular fatty acid composition of *P. maltophilia* was different from that of *Pseudomonas aeruginosa*, as previously reported (14, 25-27). *F. rhenanus* KS 0131, which did not require methionine, has a composition similar to that of *P. maltophilia* KS 0001. Methionine-requiring strains and strains that did not require methionine shared almost the same kinds of fatty acids, but some quantitative differences were found between the two types (Table 5). Furthermore, it is of interest that the cellular fatty acid compositions of *X. campestris* KS 0198 and *X. citri* KS

0199 were similar to that of *P. maltophilia*. However, the composition of *X. oryzae* KS 0200 was different from those of *P. maltophilia* strains (Fig. 4). Similarity values based on cellular fatty acid compositions showed that methionine-requiring strains and strains that did not require methionine matched at the 82% level (Fig. 5).

Ubiquinone system. A total of 18 strains, including *X. campestris* KS 0198, *X. citri* KS 0199, and *X. oryzae* KS 0200, showed the presence of ubiquinone Q₈, but *P. aeruginosa* KS 0025 contained Q₉.

DNA base composition. The DNA base composition of 10 *P. maltophilia* strains was in the range of 63 to 67 mol% guanine plus cytosine (Table 6). Some differences were found between the three methionine-requiring strains and the seven strains that did not require methionine, but the values did not distinguish between the two groups at the species level.

DNA-DNA hybridization. The 10 *P. maltophilia* strains studied seemed to cluster into five groups on the basis of DNA-DNA homology indexes (Table 6). According to the index at 75°C, KS 0001, the type strain of *P. maltophilia*, showed a relatedness of approximately 49 to 56% to *P. maltophilia* KS 0002 and *Xanthomonas* sp. KS 0053 and approximately 30 to 36% to the seven strains that did not require methionine. *P. maltophilia* KS 0002 showed a high homology index to *Xanthomonas* sp. KS 0053, a low index to *P. maltophilia* KS 0001, and even lower indexes to the seven strains that did not require methionine. On the other hand, *F. rigense* KS 0131, a strain that did not require methionine, showed high homology indexes to *F. rhenanus* KS 0130 and *F. ferrugineum* KS 0132, slightly lower indexes to *F. rigense* KS 0129 and strains KS 0194 and KS 0196, and lower indexes to

TABLE 3. Assimilation of carbon compounds by 14 strains of *P. maltophilia*

Strain	Assimilation of:													
	D-Xy-lose	Glu-cose	Fruc-tose	Man-nose	Mal-tose	Su-crose	Lac-tose	Cello-biose	Acetate	Succinate	Fumarate	DL-Malate	DL-Lactate	Pyruvate
KS 0001	-	+	+	+	+	+	+	+	+	+	+	+	+	+
KS 0002	+	+	+	+	+	+	+	+	+	+	+	+	+	+
KS 0053	+	+	+	+	+	+	+	+	-	+	+	+	+	+
KS 0186	-	+	+	+	+	+	+	+	+	+	+	+	+	+
KS 0189	-	+	+	+	+	+	+	+	+	+	+	+	+	+
KS 0193	+	+	+	+	+	+	+	+	+	+	+	+	+	+
KS 0195	-	+	+	+	+	+	+	+	+	+	+	+	+	+
KS 0129 ^b	-	+	+	+	+	+	+	+	+	+	+	+	+	+
KS 0130 ^b	-	+	-	-	+	+	-	-	+	+	+	+	+	+
KS 0131 ^b	-	+	+	-	+	+	-	-	+	+	+	+	+	+
KS 0132 ^b	-	+	-	-	+	+	-	-	+	+	+	+	+	+
KS 0194 ^b	-	+	-	+	+	+	+	+	+	+	+	+	+	+
KS 0196 ^b	-	+	-	+	+	+	+	+	+	+	+	+	+	+
KS 0197 ^b	-	+	-	-	+	+	+	+	+	+	+	+	+	+

^a Symbols: +, growth; +^w, weak growth; -, no growth. The basal medium (35) was supplemented with 50 mg of DL-methionine per liter. Compounds used by none of the strains studied were L-arabinose, gluconate, citraconate, itaconate, mannitol, glycerol, β -alanine, and *p*-hydroxybenzoate.

^b Strains that did not require methionine.

TABLE 4. Growth with carbon compounds with and without methionine by seven strains of *P. maltophilia* that did not require methionine

Strain	Growth with: ^a											
	Fumarate		L-Histidine		L-Proline		Citrate		Cellobiose		Sucrose	
	+M ^b	-M	+M	-M	+M	-M	+M	-M	+M	-M	+M	-M
KS 0129	+	+	+	-	+	+	-	-	+	-	-	-
KS 0130	+	+	+	+	+	+	+	+	-	-	-	-
KS 0131	+	+	+	+	+	-	+	+	-	-	-	-
KS 0132	+	+	+	-	+	+	+	+	-	-	-	-
KS 0194	+	+	+	+	+	+	+	+	+	+	-	-
KS 0196	+	+	+	+	+	+	+	+	+	+	-	-
KS 0197	+	+	+	-	+	+	+	+	+	+	-	-

^a Symbols: +, growth; -, no growth.^b M, Methionine. FIG. 1. Growth patterns of *F. rhenanus* KS 0131 and strains KS 0189 and KS 0196 in media with (+) and without (-) DL-methionine (Met). The basal medium (35) contained 1% maltose, and DL-methionine was added at a concentration of 50 mg/liter. OD, optical density.

methionine-requiring strains. Furthermore, another strain that did not require methionine, *F. rigense* KS 0129, showed a relatedness of approximately 50% to the other six strains that did not require methionine. However, the four reference strains of *P. maltophilia* showed the lowest relatedness to *X. campestris* KS 0198, *X. citri* KS 0199, and *P. aeruginosa* KS 0025.

DISCUSSION

On the basis of the data presented here, the methionine-requiring strains KS 0053, KS 0186, KS 0189, KS 0193, and KS 0195 were identified as members of *P. maltophilia*. Furthermore, strains KS 0129, KS 0130, KS 0131, KS 0132, KS

0194, KS 0196, and KS 0197, which do not require methionine, seem to differ from the methionine-requiring strains with respect to cellular fatty acid composition and DNA-DNA homology.

Numerical analyses of fatty acid compositions have been reported for species in a limited number of genera. Tanaka et al. (36) concluded that *Brucella* species can be differentiated at the level of 74 to 90% similarity of cellular fatty acid composition, and they also pointed out the difficulties in distinguishing species solely on the basis of fatty acid composition. Furthermore, Ikemoto et al. (14) reported high similarity values in the fatty acid compositions of *Pseudomonas* species. However, since methionine-re-

FIG. 2. Effect of various concentrations of DL-methionine (Met) on the growth of *P. maltophilia* KS 0001. Basal medium (35) containing 1% maltose was used. OD, optical density.

FIG. 3. Fatty acid patterns in *P. maltophilia* KS 0001 (A), *F. rhenanus* KS 0131 (B), and *P. aeruginosa* KS 0025 (C).

TABLE 5. Cellular fatty acid compositions of 14 strains of *P. maltophilia* and three *xanthomonad* strains

Species	Strain	Fatty acid composition (%)														C-OH	Δ17	3-OH10	D-OH	18:1
		i-11 ^a	i-14	14:0	i-15	a-15 ^b	15:0	A-OH	i-16	16:0	B-OH	16:1	17:0	17:1	18:1					
<i>P. maltophilia</i>	KS 0001	5 ^b	T	4	34	16	2	1	1	13	2	13	2			1	1	1	1	1
	KS 0002	5	1	3	38	11	1	1	1	13	2	14	2			T	1	2	1	2
	KS 0053	6	2	7	33	23	2	2	1	6	1	11	1			1	1	1	1	T
	KS 0186	3	1	6	33	13	2	1	1	17	2	10	3			2	2	1	1	2
	KS 0189	5	1	4	40	22	1	2	2	9	3	7	2			2	2	T	T	T
	KS 0193	3	2	5	29	17	3	1	2	10	1	15	2			1	1	1	1	1
	KS 0195	3	1	2	30	24	3	1	3	7	2	12	7			1	1	1	1	2
	KS 0129 ^c	5	9	1	28	6	3		18	4	2	11	13			2	2	T	T	T
	KS 0130 ^c	6	18	7	19	4	4		6	12	2	13	4			1	1	1	T	T
	KS 0131 ^c	5	5	3	33	6	6		7	10	2	11	10			1	1	T	T	T
	KS 0132 ^c	4	14	3	27	6	7		8	8	2	9	7			1	2	T	T	T
	KS 0194 ^c	6	9	2	32	9	8		6	6	2	10	8			1	2	T	T	T
	KS 0196 ^c	4	19	2	18	8	4		15	7	1	8	7			2	2	T	T	T
	KS 0197 ^c	4	12	2	24	6	5		11	8	1	11	10			2	2	T	T	T
<i>X. campestris</i>	KS 0198	3	2	1	15	26	4	1	12	4	5	16	5			1	2	1	1	1
<i>X. citri</i>	KS 0199	5	1	T	25	10	2	2	10	2	2	24	11			T	1	1	2	T
<i>X. oryzae</i>	KS 0200	2	T	T	5	1	1	1	T	17	2	37	28			T	T	1	1	4

^a i, iso acid; a, anteiso acid; Δ, cyclopropane acid; OH, hydroxy acid. A-OH, B-OH, C-OH, and D-OH are unidentified hydroxy acids.
^b Numbers are percentages of total acids. T, Acid present at less than 1%.
^c Strains that did not require methionine.

FIG. 4. Fatty acid patterns in *P. maltophilia* KS 0002 (A), *X. citri* KS 0199 (B), and *X. oryzae* KS 0200 (C).

quiring strains and strains that do not require methionine matched at a fairly high level of similarity of cellular fatty acid composition, the groups could not be separated at the species level solely on the basis of cellular fatty acid composition.

At present, DNA-DNA homology is widely accepted as an expression of genetic relatedness, but it is evaluated in different senses in bacterial taxonomy. Johnson (15) stated the relatedness of anaerobic bacteria from the point of DNA-DNA hybridization as follows: "The genosub-

species would be those strains that have 60 to 70% homology with organisms in the high homology (geno variety) group There is graduation in homology levels from 50 to about 20%. These I have designated as separate, but closely related, geno-species." On the other hand, Gardner and Kado (5) stated their view concerning DNA-DNA homology as follows: "It is not clear at what point on the homology scale the lines for interspecies and intragenus relatedness should be drawn. How far, for instance, can a genus or species diverge in evolution, and still

TABLE 6. DNA base composition and DNA-DNA homologies among *P. maltophilia* strains and related bacteria

Species	Homology group	Strain	Guanine plus cytosine content (mol%)	DNA-DNA homology (%) with:							
				KS 0001		KS 0002		KS 0131		KS 0129	
				62°C	75°C	62°C	75°C	62°C	75°C	62°C	75°C
<i>P. maltophilia</i>	I	KS 0001	66.9	100	100	72	54	34	24	36	28
	II	KS 0002	66.9	63	49	100	100	35	23	36	28
		KS 0053	66.0	67	56	97	96	38	27	43	27
	III	KS 0131 ^a	63.0	45	31	45	37	100	100	56	50
		KS 0130 ^a	62.7	48	30	55	43	100	105	59	47
	IV	KS 0132 ^a	63.7	43	30	52	43	92	96	56	47
		KS 0129 ^a	63.2	48	35	46	37	62	52	100	100
	V	KS 0194 ^a	64.6	37	28	58	48	69	58	66	53
		KS 0196 ^a	63.0	47	36	54	46	67	61	61	56
		KS 0197 ^a	64.8	44	35	57	47	27	18	63	55
<i>X. campestris</i>		KS 0198	65.5	34	25	37	32	23	15	29	21
<i>X. citri</i>		KS 0199	64.3	34	26	36	27	23	15	22	17
<i>P. aeruginosa</i>		KS 0025	64.3	22	9	17	10	20	8	11	6

^a Strains that did not require methionine.

retain its nomenclature? Obviously, such rules must be made with both arbitrary and pragmatic considerations."

Kaneko et al. (16) showed a close relationship among *Bacillus anthracis*, *Bacillus cereus*, and *Bacillus thuringiensis* on the basis of DNA-DNA homology, and, in contrast, they found a sharp separation of *Bacillus subtilis* and related species (unpublished data).

The data presented here suggest that the methionine-requiring group of *P. maltophilia* and the group of *P. maltophilia* that does not require methionine are distinct from each other and that the strains in each group are not necessarily homologous. If the concept of Johnson is applied, the 14 strains of *P. maltophilia* studied might fall into at least two different species. However, we believe that the data are not enough for separation into two species, and further studies may be required. Thus, we agree for the time being with the opinion of Gardner and Kado. Although the 14 strains studied are separable into two clusters on the basis of methionine requirement and some other properties, we conclude that the methionine-requiring strains and the strains that do not require methionine should be treated as members of biovars of *P. maltophilia*, which we designate biovar I and biovar II, respectively.

The close relationship of *P. maltophilia* to xanthomonads has been suggested on the basis of DNA-DNA homology (28), ribosomal ribonucleic acid-DNA homology (29), and fatty acid content of lipopolysaccharides (30) and has been confirmed here by fatty acid composition and DNA-DNA hybridization. Furthermore, these bacteria share the same ubiquinone system, Q₈, whereas *P. aeruginosa* possesses Q₉. Yamada et al. (38-41) pointed out that ubiquinone and menaquinone systems are useful taxonomic characters at the generic level. Therefore, *P. maltophilia* seems to be related to xanthomonads more closely than to pseudomonads on the basis of the characteristics mentioned above. Whereas chromogenic pseudomonads (10, 11, 14) seemingly resemble xanthomonads, they can be clearly differentiated by their cellular fatty acid compositions, as has been revealed in this and in previous work (14).

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REPRINT REQUESTS

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FIG. 5. Dendrogram of 14 *P. maltophilia* strains and three xanthomonad strains based on cellular fatty acid composition. Strains that do not require methionine are marked with asterisks.

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