

Nonfermentative Bacilli: Evaluation of Three Systems for Identification

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Three systems for the identification of nonfermentative bacilli were evaluated for their rapidity and accuracy of identification of 217 strains. Two of the systems, API 20E (API) and Oxi/Ferm tube (OxiF), are available as kits; the oxidative attack (OA) system is not commercially available. The overall accuracies of the OA, API, and OxiF systems were 91, 69, and 50%, respectively. Identification within 48 h was achieved for 98% of the strains by OA, for 50% by API, and for 18% by OxiF. Most of the organisms that were either misidentified or not identified by API and OxiF were those nonfermentative bacilli which are relatively more fastidious or rarely encountered or both. All three systems accurately identified nonfermentative bacilli commonly isolated at Olive View Medical Center, namely, *Pseudomonas aeruginosa*, *Acinetobacter anitratus*, *Pseudomonas maltophilia*, *Acinetobacter lwoffii*, saccharolytic flavobacteria (CDC IIB), moraxellae, *Pseudomonas fluorescens*, and *Pseudomonas putida*. The OA system identified 100% of the above organisms correctly, API identified 99.4%, and OxiF identified 99.3%. Since these organisms comprise 92% of the total number of nonfermentative bacilli isolated at Olive View Medical Center, we conclude that both API and OxiF may be useful alternatives to conventional methods, based on accuracy of identification alone. These two systems were considered substantially inferior to the OA system when both accuracy and rapidity of identification were taken into account.

Nonfastidious nonfermentative gram-negative bacilli (NFB) are isolated in many laboratories more frequently than *Haemophilus* species and make up 12 to 16% of all aerobic gram-negative bacilli encountered (1). Since NFB are often etiologically significant, it is imperative that they be identified completely and accurately. In an attempt to accomplish these objectives, a number of commercial houses have developed systems (kits) for the identification of these organisms. Two such kits, the API 20E (API; Analytab Products Inc., Plainview, N.Y.) and the Oxi/Ferm (OxiF) tube (Roche Diagnostics, Div. Hoffman-LaRoche, Inc., Nutley, N.J.), have recently received considerable attention. This is in part because their numerically coded identifications are based on studies of large numbers of strains. For this reason these systems were expected to yield results which were accurate.

Results of studies that previously evaluated these two systems vary considerably, primarily because investigators used different NFB species (3, 4, 6, 10-14, 22, 23). We studied 217 NFB strains with the API and OxiF systems and the oxidative attack (OA) system of Otto and Pickett (16) because we were interested in testing the

efficacy of these kits and the comparative performance of the OA system. We compared the rapidity with which identifications were achieved and determined the accuracy of results compared with those obtained by using conventional methods (24).

MATERIALS AND METHODS

Organisms. A total of 217 cultures were used in this study; these cultures consisted of organisms freshly isolated at Olive View Medical Center (40%) and stock cultures from the culture collection of the U.S. Food and Drug Administration District Laboratory (60%). Many of the stock cultures were obtained from M. J. Pickett, University of California at Los Angeles, and had been held at -80°C in glycerol broth (5). A total of 13 reference strains were included in this study; 3 strains were from the American Type Culture Collection (ATCC) and 10 strains were obtained from R. E. Weaver, Center for Disease Control (CDC), Atlanta, Ga. The ATCC strains were *Pseudomonas vesicularis* 11426, *P. aeruginosa* 27853, and *P. diminuta* 19146. The CDC strains were *P. maltophilia* A2897, *P. aeruginosa* 1615, *P. putida* KC941, *P. fluorescens* 7311(2), *Alcaligenes odorans* C1789, *A. faecalis* B4256, *Bordetella bronchiseptica* C1828, *Bordetella* group IVc-2 B2443, *Moraxella osloensis* B4087, and *P. vesicularis* AB-102.

Conventional system. All media were prepared according to the formulas of King et al. (8, 9). The inoculated media were incubated at 35°C for up to 7 days, with the exception of the motility and gelatin media, which required room temperature incubation. The following tests were used: triple sugar iron agar for dextrose fermentation and H₂S production; enriched nitrate broth (with inverted tube) for detection of nitrite and nitrogen gas production; motility test medium; nutrient gelatin; 2% tryptone broth (read with xylene extraction) for indole production; esculin agar (modified); Christensen urea agar; Simmons citrate; MacConkey agar; King medium B (fluorescence was observed under a 366-nm ultraviolet light); nitrite broth (with inverted tube); oxidative-fermentative basal medium with 1% carbohydrate; Moeller basal medium with 1% L-lysine, L-arginine or L-ornithine; and brucella or Trypticase soy agar for pigment production. Flagella were stained by the method of Clark (2). Carbohydrates tested included glucose, xylose, D-mannitol, lactose, sucrose, maltose, salicin, sorbitol, fructose, and galactose. Oxidase was detected by smearing (with a platinum loop) 24-h cell paste from growth on an agar surface onto a dried filter paper impregnated with tetramethyl-*p*-phenylenediamine hydrochloride (17). Production of a dark purple color within 10 s was considered positive. Isolates were identified according to CDC data (9, 24) and other sources (7, 20, 21).

OxiF system. The procedures for inoculation and incubation followed the recommendations of the manufacturer. The inoculum was taken from plates of

brucella (BBL Microbiology Systems, Cockeysville, Md.) or blood agar incubated for 18 to 24 h at 35°C. After inoculation of the tube and before reinsertion of the inoculating wire, a Trypticase soy agar or brucella agar plate was streaked and allowed to incubate overnight at 35°C. These plates were allowed to stand at room temperature to enhance pigment production and to serve as a source of inoculum for supplemental tests. Although 32 supplemental tests are suggested by the manufacturer, only 24 were needed in the study. Nine tests involved observations (test codes WRI, AKX, AKD, PYO, BRP, YEP, RG, RU, and OD), six were antimicrobial susceptibility tests (PB, AMP, KAN, PEN, NOV, and NA), and nine were other tests (42, GEL, MAC, MOT, FLA, CET, ANG, SR, and NUT) (Table 1).

A four-digit "ID value" was generated from the initial tube and oxidase tests, and the organism was identified by using the OxiF code manual. Additional tests were performed as specified by the manual.

API system. Performance of the API tests followed the procedure described in the directions of the manufacturer. In the beginning considerable difficulty was experienced in performing the confirmation tests for the negative nitrite test, because the printed instructions were brief. However, consultation with and training by the representative of the manufacturer solved this problem. Seventeen additional tests were needed in this study. They are listed in order of frequency in Table 1. These tests were performed when the 9-digit profile generated by the strip and complementary tests did not identify the strain. When the analytical profile

TABLE 1. Additional tests required by the three systems for identification

API			OxiF ^a			OA	
Test code	Test	No. of strains	Test code	Test	No. of strains	Test	No. of strains
42C	42°C, growth	54		Antimicrobial susceptibility		Gelatinase	4
Flag	Flagellum stain	44	PB	Polymyxin B	15	Flagellum stain	1
Acet	Acetamide, growth	42	PEN	Penicillin	16		
SS	SS agar, growth	35	AMP	Ampicillin	9		
Escu	Esculin hydrolysis	18	KAN	Kanamycin	7		
Cet	Cetrimide, growth	16	NOV	Novobiocin	4		
Gel	Gelatinase	15	NA	Nalidixic acid	3		
NaCl	6.5% NaCl, growth	12					
NO ₂	Repeat nitrite test	9	MOT	Motility	78		
Lec	Lecithinase	8	MAC	MacConkey agar, growth	29		
10%L	10% Lactose slant, acid	8	FLA	Flagellum stain	28		
DNA	Deoxyribonuclease	7	42	42°C, growth	20		
PB	Polymyxin B sensitivity	5	GEL	Gelatinase	17		
Mac	MacConkey agar, growth	3	ANG	Anaerobic growth	12		
TSI	Butt of TSI agar, growth ^b	3	CET	Cetrimide, growth	6		
Pen	Penicillin sensitivity	1	SR	Salt tolerance (6.5% NaCl)	1		
Yel	Yellow pigment	1	NUT	Nutrient broth, growth	1		
Mot	Repeat motility	1					
Urea	Repeat urease	1					
No. of strains requiring additional tests/total no.		114/217 (53) ^c			154/217 (71)		5/217 (2)

^a The following observations which did not constitute additional testing were required: yellow pigment (YEP), 97 strains; alkaline xylose (AKX), 84; brown pigment (BRP), 59; wrinkled colonies (WRI), 27; fluorescent pigments (PYO), 20; runny growth (RG), 16; rapid urea (RU), 9; odor (OD), 9; alkaline dextrose (AKD), 7.

^b TSI, Triple sugar iron.

^c Numbers in parentheses are percentages.

index did not list the profile number, the API computer service was consulted.

OA system. This system consists of two compartmentalized plastic boxes, each containing 11 substrates and a control, and several conventional tests (15). The boxes were inoculated and incubated as originally described (16). The carbohydrates tested were glucose, xylose, D-mannitol, lactose, sucrose, maltose, salicin, sorbitol, *m*-inositol, melibiose, and fructose. The organic acid salts and amino acids tested were acetamide, allantoin, saccharate, tartrate, β -alanine, *p*-hydroxybenzoate, benzoate, betaine, butyrate, norleucine, and glutamine. The conventional tests consisted of the following: triple sugar iron agar for H₂S production; oxidase (paper strip method); motility-nitrate medium (test code, M-N) for detection of motility and reduction of nitrate to nitrite, N₂, or amines; fluorescence-lactose-denitrification medium (test code, FLN) for detection of fluorescence, acidification of lactose, and reduction of nitrate or nitrite or both to N₂; 42°C growth (brucella broth); lysine decarboxylase (test code, LDC), gluconate, urease, and indole production by the buffered single-substrate method of Pickett and Pedersen (18, 19); esculin hydrolysis (15); and growth on MacConkey agar. Pigment production was determined by using brucella agar incubated at 35°C, followed by room temperature incubation.

RESULTS

Table 2 shows the time required for identification (correct or incorrect) by each of the three systems. The API 48-h column does not include the time required for a response from the computer service. This varied from a few hours to overnight, depending upon the time of day that the data were telephoned in. When further tests were required, identification could be delayed by 24 h or more. No 24-h column is given for OxiF because the manufacturer makes no claim for identification before 48 h of incubation. The OxiF >48-h figures represent those strains which required additional tests rather than simple observations, such as runny growth or alkalization of xylose, which were available at 48 h.

Of the fluorescent NFB studied, at 48 h the API, OxiF, and OA systems identified 36, 3, and 87% of the strains, respectively. Additional tests were required by API for 64% of the strains, by OxiF for 97% of the strains, and by OA for 13% of the strains. Gelatinase was the only additional test required by the OA system.

With the saccharolytic group, 43 and 62% of the strains required more than 48 h for definitive identification when tested by API and OxiF, respectively. By comparison, all strains were identified in 48 h by the OA system.

With the pigmented organisms, 42 and 65% of the strains tested by API and OxiF, respectively, needed more than 48 h for completion of all tests. No strains required more than 48 h for identification by the OA system.

With the weakly saccharolytic group, 55, 22, and 100% of the strains were identified within 48 h by API, OxiF, and OA, respectively. Similar, but not identical, results were obtained with the nonsaccharolytic NFB. API identified 44%, OxiF identified 14%, and OA identified 99% of the strains in 48 h. The overall rapidity of identification, determined in terms of the percentage of strains identified within 48 h, was 50% for API, 18% for OxiF, and 98% for OA.

The principal additional tests that were required, in order of frequency, for the API system were: 42 C, Flag, Acet, SS, Escu, Cet, Gel, and NaCl (Table 1). For OxiF the tests were: BRP (which required 3 days of incubation at room temperature), MOT, MAC, FLA, 42, GEL, PEN, PB, and ANG. The API profiles for 67 strains (31%) were not in the code book and required the use of the computer service. The OA system required two additional tests: four gelatinase, and one flagellum stain. Additional tests were required for 114 strains (53%) by API, for 154 strains (71%) by OxiF, and for 5 strains (2%) by OA.

The identification accuracy of the systems is shown in Table 3. It should be noted that the OxiF coding manual gave only one species, *A. faecalis*, for all three *Alcaligenes* species. Also, the designation *Flavobacterium* species was applied to both *F. meningosepticum* and CDC group IIb. All three systems identified *Moraxella* species to the genus level only.

All *P. aeruginosa* strains tested were non-mucoid and were accurately identified by all systems. One strain of *P. fluorescens* was identified as *P. putida* by API (the additional test for lecithinase was negative). One strain of *P. putida* was identified by OxiF as *P. maltophilia*. A different strain of *P. putida* was identified by API as *P. aeruginosa*. All of the other strains of *P. fluorescens* and *P. putida* were identified correctly by the systems.

Pseudomonas cepacia and *P. maltophilia* in the saccharolytic group presented few problems in identification. API and OA gave correct identification for all strains of these two species tested. OxiF incorrectly identified one strain of *P. cepacia* as *P. maltophilia* and one strain of *P. maltophilia* as *Achromobacter* species. *Pseudomonas pickettii* was identified to the genus level only by API (100%) and OxiF (75%). The OA system identified all strains of *P. pickettii* to the species level. In this group, *Pseudomonas stutzeri* presented the most problems in identification for both API and OxiF. Four strains were variously identified by the API system as *Moraxella*, *Achromobacter*, *P. putida*, and *Pseudomonas pseudoalcaligenes*. The OxiF system failed to identify five strains of *P. stutz-*

TABLE 2. Time required for completion of identification tests

Organism	No. of strains	% of strains completed ^a							
		API			OxiF ^b		OA		
		24 h	48 h	>48 h	48 h	>48 h	24 h	48 h	>48 h
Fluorescent									
<i>P. aeruginosa</i>	9	11	55	33	11	89	100	0	0
<i>P. fluorescens</i>	8	0	25	75	0	100	25	50	25
<i>P. putida</i>	13	0	23	77	0	100	46	39	15
Total	30	3	33	64	3	97	57	30	13
Saccharolytic									
<i>P. cepacia</i>	7	29	71	0	57	43	100	0	0
<i>P. stutzeri</i>	15	0	47	53	7	93	60	40	0
<i>P. maltophilia</i>	11	0	64	36	64	36	91	9	0
<i>P. pickettii</i>	4	0	0	100	25	75	25	75	0
Total	37	5	51	43	38	62	73	27	0
Pigmented									
CDC IIk-1	10	0	40	60	60	20	80	20	0
CDC IIk-2	10	0	50	50	40	60	100	0	0
CDC Ve-2	9	0	56	33	44	56	56	44	0
<i>P. vesicularis</i>	8	0	0	100	13	87	100	0	0
<i>F. meningosepticum</i>	7	0	100	0	71	29	86	14	0
<i>Flavobacterium</i> IIf	11	9	82	9	64	36	9	91	0
Total	55	4	55	42	35	65	68	32	0
Weakly saccharolytic									
<i>P. acidovorans</i>	7	0	14	86	0	100	42	58	0
<i>P. pseudoalcaligenes</i>	2	0	0	100	0	100	50	50	0
<i>A. anitratus</i>	13	15	85	0	23	77	100	0	0
<i>A. xylooxidans</i>	4	0	0	100	25	75	0	100	0
Vd (<i>Achromobacter</i> sp.)	2	0	50	50	100	0	0	100	0
Total	28	7	48	44	22	78	63	37	0
Nonsaccharolytic									
<i>P. diminuta</i>	15	0	7	93	0	100	0	93	7
<i>P. alcaligenes</i>	5	0	0	100	0	100	25	75	0
<i>P. putrefaciens</i>	5	40	0	60	80	20	100	0	0
<i>A. lwoffii</i>	4	0	100	0	50	50	100	0	0
<i>B. bronchicanis</i>	4	0	50	50	25	75	25	75	0
CDC IVc-2	2	0	50	50	0	100	0	100	0
<i>A. faecalis</i>	3	0	0	100	0	100	66	34	0
<i>A. odorans</i>	6	0	0	100	0	100	100	0	0
<i>A. denitrificans</i>	3	0	34	66	66	34	0	100	0
<i>Flavobacterium</i> IIf	9	0	100	0	0	100	100	0	0
<i>Moraxella</i> species	11	0	82	18	0	100	91	9	0
Total	67	3	41	56	14	86	58	41	1
Total	217	4	46	50	18	82	63	35	2

^a Numbers do not necessarily imply correct identification.

^b OxiF makes no claim for 24 h identification.

eri. Four strains were misidentified as *P. cepacia*, *P. maltophilia*, *Achromobacter xylooxidans*, and *Achromobacter* sp. Failure to produce wrinkled colonies was the principal cause of either no identification or an incorrect identification by OxiF. One strain of *P. stutzeri* was not identified by OA; the others were correctly identified.

The pigmented group was accurately identified by all systems, if one considers the designation *Flavobacterium* species adequate for both *F. meningosepticum* and CDC group IIf.

API and OxiF identified 100% of these strains to the genus level. API correctly identified 57% of the *F. meningosepticum* strains, with one strain being identified as IIf and two strains named *Flavobacterium* species. Correct identification of CDC group IIf strains (designated *Flavobacterium* species) was 82% for API. One strain was misidentified as *F. meningosepticum*, and one was identified as CDC group IIf. OxiF placed all strains of *F. meningosepticum* and CDC group IIf in *Flavobacterium* species. OA correctly identified all *F. meningosepticum* strains, but

TABLE 3. Accuracy of identification by the three systems

Organism	No. of strains	% correctly identified to species level (genus level) by:		
		API	OxiF	OA
Fluorescent				
<i>P. aeruginosa</i>	9	100	100	100
<i>P. fluorescens</i>	8	87 (100)	100	100
<i>P. putida</i>	13	92 (100)	92 (100)	100
Total	30	93 (100)	97 (100)	100
Saccharolytic				
<i>P. cepacia</i>	7	100	86 (100)	100
<i>P. stutzeri</i>	15	73 (87)	40 (53)	93
<i>P. maltophilia</i>	11	100	80	100
<i>P. pickettii</i>	4	0 (100)	0 (75)	100
Total	37	78 (94)	57 (79)	97
Pigmented				
CDC Iik-1	10	60	60	100
CDC Iik-2	10	60 (90)	40	100
CDC Ve-2	9	67	78	89
<i>P. vesicularis</i>	8	88	100	100
<i>F. meningosepticum</i>	7	57 (100)	0 (100)	100
<i>Flavobacterium</i> IIb	11	82 (100)	0 (100)	72 (100)
Total	55	69 (84)	45 (78)	93 (98)
Weakly saccharolytic				
<i>P. acidovorans</i>	7	57 (86)	0 (86)	71
<i>P. pseudoalcaligenes</i>	2	0 (100)	0 (100)	100
<i>A. anitratus</i>	13	100	100	100
<i>A. xylooxidans</i>	4	50	25	100
Vd (<i>Achromobacter</i> sp.)	2	100	100	100
Total	28	75 (89)	57 (86)	93
Nonsaccharolytic				
<i>P. diminuta</i>	15	27 (67)	0 (93)	100
<i>P. alcaligenes</i>	5	20 (60)	0 (100)	80
<i>P. putrefaciens</i>	5	80 (100)	80 (100)	100
<i>A. lwoffii</i>	4	100	100	100
<i>B. bronchicanis</i>	4	75 (100)	75	100
CDC IVc-2	2	100	0	100
<i>A. faecalis</i>	3	67 (100)	100	100
<i>A. odorans</i>	6	33 (100)	0 (83)	100
<i>A. denitrificans</i>	3	33 (66)	0 (100)	100
<i>Flavobacterium</i> IIc	9	89 (100)	89	100
<i>Moraxella</i> species ^a	11	0 (90)	0 (100)	0 (100)
Total	67	65 (88)	31 (85)	82 (99)
Total	217	69	50	91

^a No system claims to identify moraxellae to the species level.

misidentified three CDC group IIb strains as CDC group IIf. Of the CDC group Iik-1 strains, 60% were correctly identified by API and OxiF; 100% of these strains were accurately identified by OA. Three Iik-1 strains were misidentified by API as *Moraxella* sp., *Acinetobacter lwoffii* (although oxidase positive), and *Pseudomonas* species. The fourth strain was not identified. OxiF misidentified two strains as *Flavobacterium* species, one as *P. vesicularis* and one as CDC Ve-2. All strains were correctly identified by OA. The CDC Iik-2 strains gave similar results. API and OxiF gave 60 and 40% correct identifications, respectively; 100% were correctly identified by OA. API placed two strains in the Iik group, but

did not distinguish between Iik-1 and Iik-2. One strain was identified as *P. cepacia*, and one strain was identified as Iik-1 by API. The six misidentifications by OxiF were *P. cepacia* (three strains), M-4 (two strains), and *Achromobacter* (one strain). Three CDC Ve-2 strains (33%) were incorrectly identified by API as *P. cepacia* (A375), *P. putida* (A373), and "not identified" (A342). Two of these three strains were misidentified by OxiF as *P. maltophilia* (A342) and *P. fluorescens* (A373). One strain (A373) was not identified by OA. All strains of *P. vesicularis* were correctly identified by OxiF and OA. API gave no identification for one strain; the other seven were correctly identified. The

OxiF identification for six of the strains depended upon the production of diffusible brown pigment, which required 3 additional days of incubation at room temperature to appear.

In the weakly saccharolytic group, *Acinetobacter anitratum* was correctly identified by all systems. Two of the seven *Pseudomonas acidoovorans* strains were not identified by OA and were named *P. putida* and Ilk-1 by API and OxiF. One strain was identified as *Pseudomonas testosteroni* by API; the remaining four were correctly identified. OxiF identified five strains to the genus level only. The two strains of *P. pseudoalcaligenes* were labeled *Pseudomonas* species by OxiF. API placed one strain in the *Pseudomonas alcaligenes*-*P. pickettii* group; the other strain was identified as *P. diminuta*. Two of the four *A. xylooxidans* strains were identified by API as *Alcaligenes denitrificans*. The others were correctly identified. OxiF identified two strains as *A. faecalis*, one as *P. vesicularis* and one correctly. The OA system correctly identified all strains of these two species. The two strains of CDC biogroup Vd were correctly identified as *Achromobacter* species by all systems.

P. diminuta, in the nonsaccharolytic group, posed a problem in identification with both the OxiF and API systems. With API only 4 of 15 strains were correctly identified. No identification was possible with three strains, two strains were named *Pseudomonas* "other", two strains were called CDC group IVc-2, one strain was identified as *P. stutzeri*, and another was identified as *P. denitrificans*. One strain was placed in the *P. alcaligenes*-*P. testosteroni* category and could not be further differentiated. The last unidentified strain was placed in the *P. diminuta*-*P. vesicularis* group. OxiF identified 13 strains to the genus level only. One strain was misidentified as *Alcaligenes* species, and a second strain (ATCC 19146) was identified as CDC group Ilk-1. OA correctly identified all strains to the species level. Four of the five *P. alcaligenes* strains were not identified beyond the genus level by OxiF. The last strain was designated *P. maltophilia*. API correctly identified one strain. The other four strains were identified as *P. testosteroni*, *P. pseudoalcaligenes*, *A. odorans*, and *P. pseudoalcaligenes*-*P. alcaligenes* group. OA correctly identified four strains but did not identify the fifth strain. Four of the five *Pseudomonas putrefaciens* strains were correctly identified by API. The fifth strain was identified as *Pseudomonas* species. OxiF gave identical results; OA identified all strains correctly. All systems correctly identified all strains of *A. lwoffii*. Three of the four strains of *Bordetella bronchicanis* (formerly *B. bronchiseptica*) were

correctly identified by both API and OxiF. The fourth strain was identified by API as CDC group IVc-2 and by OxiF as *A. faecalis*. All strains were correctly identified by OA. Both IVc-2 strains studied were correctly identified by API and OA. OxiF misidentified both strains, calling them *A. faecalis* and *B. bronchiseptica*. OxiF places all *Alcaligenes* species in the *A. faecalis* group. Within this limitation, OxiF correctly identified 100% of *A. faecalis* (species level) and *A. denitrificans* (genus level). One of the six *A. odorans* strains was misidentified as *Moraxella* species; the others were correctly identified by OxiF. API misidentified one *A. faecalis* strain (as *A. denitrificans*), one *A. odorans* (as *A. denitrificans*), and two *A. denitrificans* (as *B. bronchiseptica* and *A. denitrificans*-*A. faecalis*). Three strains of *A. odorans* were called *Alcaligenes* species. The OA system correctly identified all strains of the three *Alcaligenes* species studied. Of the CDC group IIf strains, 89% were correctly identified by API. One strain was termed *Flavobacterium* species. OxiF identified eight of the nine strains as *Flavobacterium* species. The last strain was misidentified as *Pasteurella multocida*. OA correctly identified all strains. *Moraxellae* were identified to the genus level only by all systems. API placed one strain in the *Brucella*-*Moraxella* category and did not identify it further. All other strains were correctly identified. OxiF and OA correctly identified all strains to the genus level.

DISCUSSION

The selection of any one identification system is, at best, difficult. Clinical microbiologists must consider several factors: (i) the accuracy of the system, (ii) rapidity of identification, (iii) economic factors (cost of materials, work and time involved, storage), and (iv) the appropriateness of the system in a given laboratory, i.e., its integration into the daily routine. Since there is no system presently available that is superior in all respects, any decision must involve compromise.

Based on the above considerations, we decided to compare the accuracy and rapidity of three systems. Our results indicate that the OA system is superior to API and OxiF with respect to both parameters (accuracy and rapidity). The reasons are threefold: (i) the OA system utilizes a large number of substrates; (ii) it is highly sensitive; and (iii) it was designed solely for identification of NFB. The sensitivity of the OA system has been attributed to the low buffering capacity of the basal media coupled with the use of large inocula which provide the system with preformed enzymes (16). The overall accuracy

of the OA system was 94% (ranging from 100% with the fluorescent pseudomonads to 91% with nonsaccharolytic NFB). In comparison, the accuracy levels of the API and OxiF systems varied from a high of 93 and 97%, respectively, for the fluorescent group to a low of 65 and 31%, respectively, for the nonsaccharolytic organisms. Mean accuracy levels were 69% for API and 50% for OxiF. These figures do not reflect the actual effectiveness of the systems since the distribution of organisms studied differed significantly from their relative frequencies of occurrence in ordinary clinical laboratories. Indeed, if the data regarding accuracy are adjusted to indicate how the systems would have performed in a clinical laboratory, it appears that the API, OxiF, and OA systems would have been able to identify 96.3, 95.3, and 99.8% of the NFB encountered, respectively (Table 4). The estimates of the probable performance of the API and OxiF systems are in good agreement with the actual performance recently reported by Oberhofer (13). Frequently, however, reaching a final species level identification entailed, for both API and OxiF, performance of a sizable number of additional tests, which was both costly and time consuming.

The rapidity with which the systems yielded definitive species or biogroup identification varied greatly. Within 48 h OA identified 100% of

the saccharolytic, pigmented, and weakly saccharolytic groups, 99% of the nonsaccharolytic NFB, and 87% of the fluorescent group. The API and OxiF systems identified, in the same time period, 56 and 38% of the saccharolytic, 59 and 35% of the pigmented, 55 and 22% of the weakly saccharolytic, 44 and 14% of the nonsaccharolytic, and 36 and 3% of the fluorescent NFB, respectively.

In summary, it appears from our data, as well as from those of other investigators (3, 4, 6, 10-14, 22, 23), that both API and OxiF accurately identify commonly isolated NFB, specifically *P. aeruginosa*, *A. anitratus*, *P. maltophilia*, *A. lwoffii*, CDC group IIb, *Moraxella* species, *P. fluorescens*, and *P. putida*. At Olive View Medical Center these species comprised 92% of the total NFB isolated during 1978. Therefore, on the basis of accuracy alone, we conclude that API and OxiF offer clinical laboratories convenient and acceptable alternatives to conventional methods for the identification of the above organisms. However, neither system would be completely satisfactory for high-volume or reference laboratories where more fastidious and rare NFB are regularly encountered. In contrast, the OA system offers a highly accurate and rapid tool for the identification of most of the NFB isolated from clinical specimens.

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TABLE 4. Accuracy of identification of the three systems^a

Organism	Frequency of isolation (%) ^b	Accuracy of identification (%) ^c		
		API	OxiF	OA
<i>P. aeruginosa</i>	74	100 ^d	100	100
<i>A. anitratus</i>	6	100	100	100
<i>P. maltophilia</i>	3	100	80	100
<i>A. lwoffii</i>	2	100	100	100
<i>Flavobacterium</i> IIb	2	100	100	100
<i>Moraxella</i> sp.	2	90	100	100
<i>P. fluorescens</i>	2	87	100	100
<i>P. putida</i>	1	92	92	100
<i>P. stutzeri</i>	1	73	40	93
CDC Ve-2	1	67	78	89
<i>Flavobacterium</i> IIc	1	89	89	100
<i>P. cepacia</i>	1	100	86	100
<i>P. pickettii</i>	1	0	0	100
All others	3	53	34	98
Total	100	96.3	95.3	99.8

^a These data were generated by adjusting the distribution of NFB species studied to the distribution of species found at the Olive View Medical Center Laboratory.

^b Frequency of isolation as a percentage of the total nonfermenters isolated at Olive View Medical Center in 1978.

^c Expressed as percentage of strains correctly identified, irrespective of temporal considerations.

^d Data are those shown in Table 3.

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