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Critical Evaluation of the AutoMicrobic System Gram-Negative Identification Card for Identification of Glucose-Nonfermenting Gram-Negative Rods

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During a 6-month study we critically evaluated the accuracy of the AutoMicrobic system Gram-Negative Identification Card (Vitek Systems, Inc., Hazelwood, Mo.) in identifying glucose-nonfermenting gram-negative bacilli by testing 419 selected isolates in parallel with a conventional reference method. Of 356 isolates included in the AutoMicrobic system profile, a total of 307 (86.2%) were correctly identified, 36 (10.1%) were not identified, and 13 (3.7%) were misidentified. Fifty-eight of 63 (92%) isolates not included in the profile were correctly reported as "unidentified organisms." Overall, if the first-choice identification was always accepted, only 18 (4.3%) isolates would have been incorrectly reported. When first-choice identifications appended with the special message "questionable biopattern" were rejected, and organisms were screened for characteristic odor and antimicrobial susceptibility before final acceptance of the AutoMicrobic system report, the number of misidentifications was reduced to 5 (1.2%). The average time to identification with the AutoMicrobic system Gram-Negative Identification Card was 15 h. This compares favorably with the 65 h required by the reference method.

In early 1984, Vitek Systems, Inc. (Hazelwood, Mo.) replaced the EBC-Plus card used with their AutoMicrobic system (AMS) for the automated identification of gram-negative bacilli with a new Gram-Negative Identification (GNI) card. Together with an upgraded software package, the GNI card expanded the spectrum of glucose-fermenting and -nonfermenting gram-negative bacilli (NFGNB) that could be identified with the AMS. We were particularly interested in evaluating the ability of the new system to identify NFGNB. Five previous groups of investigators (5, 6, 17, 33, 38) have reported on the usefulness of the older EBC-Plus card in the identification of this group of organisms. One (38) utilized the GNI card to test isolates whose identification with the EBC-Plus card differed from that obtained by a reference method. To date, however, there have been no comprehensive published evaluations of the AMS GNI for the identification of NFGNB.

The accuracy of any bacterial identification system is a function of the particular mixture of isolates tested. Data generated by the testing of unselected consecutive clinical isolates are skewed by the disproportionate presence of the commonly encountered and more readily identified bacterial species. Although this may better reflect the usefulness of the identification system in routine laboratory work, it fails to reveal the specific strengths and weaknesses of the system for the identification of individual species or groups of organisms. We attempted to test equivalent numbers of strains from each group or, when availability of isolates permitted, species of NFGNB listed in the AMS GNI data base (16, 17, 35); in addition, we sought to challenge the system with a significant number of unlisted species. This initial phase of the study was followed by an evaluation of the AMS GNI in routine laboratory use.

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MATERIALS AND METHODS

Microorganisms. Of the 419 NFGNB tested during the first phase of the study at the Seattle Veterans Administration Medical Center, 39 were fresh clinical isolates obtained from the clinical microbiology laboratories of Seattle Veterans Administration Medical Center, the University of Washington Hospital, and the Seattle Children's Orthopedic Hospital. The remaining 380 isolates were from the frozen reference collections of the Seattle Veterans Administration Medical Center (323 isolates), Children's Orthopedic Hospital (2 isolates), University of Washington Hospital (17 isolates), Harborview Medical Center, Seattle (24 isolates), and Genetic Systems Corp., Seattle (14 isolates). A tabulation, by group and genus, of the various species tested is found in Table 1. Each isolate was initially subcultured twice onto sheep blood agar plates (30°C) to ensure purity and good log-phase growth.

During the second phase of the study all NFGNB recovered from specimens submitted to the Seattle Veterans Administration Medical Center microbiology laboratory for culture, which could not be readily identified by rapid conventional procedures, were tested on the AMS GNI system.

Conventional identification method. All organisms tested in the AMS GNI test system were simultaneously identified using two subsets of biochemical tests from among those recommended by Gilardi (15, 16) and Rubin et al. (29). The primary 18-test battery consisted of motility, oxidase, MacConkey agar, nitrate, nitrite, DNase, *o*-nitrophenyl-β-D-galactopyranoside, gelatin, lysine, arginine, indole, urea, and oxidation-fermentation (O-F) basal medium with 1% glucose, maltose, mannitol, and xylose.

The collation of the results obtained by the primary tests and the subsequent bacterial identification was accomplished by using an adaptation of a binary coding system

TABLE 1. Isolates studied in the evaluation

Group (no. of isolates)	Organism		No. tested	Data base species[a]
	Genus or strain	Species		
Fluorescent (94)	*Pseudomonas*	*aeruginosa*	39	*
		fluorescens	21	*
		putida	34	*
Pseudomallei (40)	*Pseudomonas*	*pseudomallei*	1	*
		cepacia	32	*
		pickettii 1	3	*
		pickettii 2	3	*
		pickettii 3	1	*
Stutzeri (32)	*Pseudomonas*	*stutzeri*	31	*
		mendocina	1	*
Acidovorans (16)	*Pseudomonas*	*acidovorans*	12	*
		testosteroni	4	
Diminuta (44)	*Pseudomonas*	*diminuta*	3	
		vesicularis	5	*
		maltophilia	36	*
Alcaligenes (13)	*Pseudomonas*	*alcaligenes*	7	
		pseudoalcaligenes	6	
Other pseudomonads (10)	*Pseudomonas*	*putrefaciens*	6	*
	Ve-1		2	*
	Ve-2		2	*
				*
Bordetella-alcaligenes (61)	*Bordetella*	*bronchicanis*	7	*
	Alcaligenes	*denitrificans*	10	
		odorans	30	*
		faecalis	6	
	CDC IVc-2		6	
	CDC IVe		2	
Achromobacter-agrobacterium (25)	*Achromobacter*	Vd-1	4	*
		xylosoxidans	19	*
	Agrobacterium	*tumefaciens*	2	*
Flavobacterium (17)	*Flavobacterium*	*breve*	1	*
		meningosepticum	3	*
		multivorum	1	*
		odoratum	10	*
	CDC IIf		2	
Acinetobacter (50)	*Acinetobacter*	*alcaligenes*	5	*
		lwoffii	13	*
		calcoaceticus	27	*
		hemolyticus	5	*
Moraxella (13)	*Moraxella*	*nonliquifaciens*	5	
		phenylpyruvica	2	
		osloensis	5	
		urethralis	1	
	CDC M-5		5	

[a] Asterisks indicate that the species is included in the AMS GNI system data base.

described by Dito et al. (10). Results of the 18 primary tests were divided into six sets of 3 tests each. Each digit of the final six-digit biotype represented the summation of the numerical scores given to each positive biochemical test within a set. This resulted in the production of a unique numerical code for any given constellation of biochemical results. This was checked against a computer-generated listing of codes, the associated bacterial species capable of producing these biochemical reactions, their biotype frequencies, and an unweighted identification probability factor. The biotype frequencies, indicating the likelihood that the organism listed would have that particular biochemical

pattern, were calculated by multiplying by one another the positivity rates for each biochemical test in the battery (15, 16). The identification probability factor was calculated by dividing the biotype frequency for each tested organism by the sum of the biotype frequencies for that particular code. This method of calculation assumes that each test result is independent of the results of the other tests in the battery and that all organisms have the same isolation frequency. An identification was considered confirmed if the identification probability exceeded 0.950. Organisms yielding identification probabilities of less than 0.950 were identified by utilizing a battery of secondary tests. These included phenylalanine, acetate, salmonella-shigella agar, growth at 42°C, citrate, *Pseudomonas* agar F and P (Difco Laboratories, Detroit, Mich.), esculin, starch, cetrimide, 6.5% salt slant, acetamide, casein, lecithinase, supplemental oxidation-fermentation sugars, Kirby-Bauer disk diffusion susceptibility tests (24), and electron microscopy for flagellar configuration.

AMS identification testing. All isolates were identified with the AMS GNI card in conjunction with the current AMS software program, AMS E01.RIA 19 NOV 84. The inoculum suspensions were prepared in accordance with the manufacturer's instructions (36). A cytochrome oxidase test was done on each organism, and a positive result was recorded by blackening the appropriate spot on the GNI card at the time of inoculation. Results were automatically reported in 4 to 18 h.

The GNI test system is driven by a software package which uses the results of the 30 test wells and oxidase reaction to calculate the biotype frequency for each taxon in the data banks; the two taxa with the largest values are selected as the first- and second-choice organisms. Identification probabilities are calculated for each of these by dividing their biotype frequencies by the sum of the biotype frequencies of all organisms fitting that biopattern. These identification probabilities indicate how well the first and second choices are separated from the other taxa capable of producing that biopattern. The final report is generated as soon as the first-choice organism reaches an identification probability of ≥0.90 and passes a predetermined likelihood screen for that taxon. Final reports are generated at 18 h on all other cards. The final report lists all biochemical reactions and the identification probabilities for the two most likely identifications. A number of special messages may also appear on the final GNI report explaining why an identification could not be made or qualifying the identification reported.

Unidentified organisms are reported in one of two fashions. (i) The first-choice organism identification is reported as "unidentified organism," a special message describes the isolate as saccharolytic or asaccharolytic, and a second-choice identification is offered with a lesser probability figure. (ii) The report consists of a simple "unidentified organism" statement without further message or probability figures.

AMS GNI report interpretation. The AMS GNI reports only presumptive or group identifications for certain bacterial species and suggests further biochemical tests for definitive evaluation. For the purposes of this study, *Pseudomonas fluorescens*, *P. putida*, and *P. mendocina* strains identified as the *P. fluorescens-putida-mendocina* group and *Flavobacterium breve* identified as *Flavobacterium* species by the AMS GNI were considered to have been correctly identified. *Acinetobacter calcoaceticus* biotype *anitratus* and *A. calcoaceticus* biotype *lwoffii* were accepted as correct identification of the hemolytic strains *A. hemolyticus* and *A. alcaligenes*. Similarly, *Pseudomonas pickettii* 1, 2, and 3 (*thomasii*) were all accepted as correct identifications for any of the three *P. pickettii* biotypes. Presumptive identifications of *A. calcoaceticus* biotype *lwoffii*, *Alcaligenes* species (*odorans*), and *Pseudomonas pseudomallei* were considered final. In all cases, the time required for the AMS GNI to generate a report was considered equivalent to identification time. An AMS report of "nonviable organism" was considered equivalent to an "unidentified organism" report with no second-choice organism listed.

Data analysis. The identification of the NFGNB by the AMS GNI was evaluated by comparing the AMS results with those determined by conventional identification. The results for those isolates contained in the AMS GNI data banks are presented in a "crosstabs" (12) comparison table (Table 2).

RESULTS

Identification categories. Sixty-three of the 419 tested organisms belonged to species not included in the AMS data base. Five of these isolates were incorrectly identified to the species level (two as *Pseudomonas acidovorans*, three as *Flavobacterium odoratum*) with an identification probability of <0.90. The remaining 58 (92%) were appropriately reported as unidentified. Of the 356 test organisms included in the data base, 270 (75.8%) were identified with a first-choice identification probability of ≥0.90, 50 (14%) were identified with a first-choice probability of <0.90, and 36 isolates (10.1%) were not identified.

High-probability species identification. Of the 356 test organisms belonging to species included in the AMS data base, 320 (89.9%) were identified to the species level (Table 2). In 270 of these the identification probability was reported to be ≥0.90. Only four (1.5%) of these identifications were incorrect (Table 2). Two of these would have been recognized as such by a competent technologist working under normal laboratory conditions. One was a kanamycin-sensitive *P. putida*, incorrectly identified by the AMS as *Pseudomonas aeruginosa*; the second was an *A. odorans* with typical colonial morphology and fruity odor identified as *Pseudomonas stutzeri* by the AMS. The other two misidentifications would have gone undetected. One was a *Pseudomonas cepacia* reported by AMS as a CDC Ve-2 and the other a *Flavobacterium multivoram* incorrectly identified as a *Pseudomonas paucimobilis*.

Low-probability species identification. Of the 50 isolates included in the AMS data base with first-choice identification probabilities of <0.90, 41 (82%) were correctly identified and 9 (18%) incorrectly identified (Table 2). Two of the nine incorrectly identified organisms were fluorescent pseudomonads correctly reported at the group level (Table 2); both errors were detected by kanamycin susceptibility testing. Of the remaining seven, three were misidentified as *A. odorans*, a microbe readily detected on solid media by its strong fruity odor. All three misidentified *P. stutzeri* isolates would have been correctly reported at 0.60, 0.83, and 0.93 probability levels had the maltose well not been read as negative. No other specific biochemical test or species of organism was disproportionately involved in the misidentifications.

Special messages or message codes accompanied 29 of the 50 low-probability reports. Nearly one-half (19 of 41) of the correct reports included a "good confidence, marginal separation" message; only 2 of the 9 incorrect reports were so labeled. Conversely, a "questionable biopattern" message

TABLE 2. Comparison of conventional identification methods and AMS GNI for 356 isolates[a]

Conventional identification	Total no. of strains	AMS GNI identification																																
		P. aeruginosa	*P. fluorescens*	*P. putida*	*P. pseudomallei*	*P. cepacia*	*P. pickettii* 1	*P. pickettii* 2	*P. pickettii* 3	*P. stutzeri*	*P. mendocina*	*P. acidovorans*	*P. vesicularis*	*P. maltophilia*	*P. putrefaciens*	Ve-1	Ve-2	*P. paucimobilis*	*B. bronchicanis*	*A. odorans*	*Achromobacter* Vd-1	*A. xylosoxidans*	*A. tumefaciens*	*F. breve*	*F. meningosepticum*	*F. multivorum*	*F. odoratum*	*A. alcaligenes*	*A. lwoffii*	*A. calcoaceticus*	*A. hemolyticus*	Unidentified as first-choice organism identification	Unidentified as only organism identification
P. aeruginosa	39	35	1																															3
P. fluorescens	21	1	19																															1
P. putida	34	①		33																														
P. pseudomallei	1																																	1
P. cepacia	32					27											①																	4
P. pickettii 1	3							2												1														
P. pickettii 2	3							2	1																									
P. pickettii 3	1																																	1
P. stutzeri	31									24		1								2												3		1
P. mendocina	1										1																							
P. acidovorans	12											9																				3		
P. vesicularis	5												4													1								
P. maltophilia	36													35																				1
P. putrefaciens	6														3																			3
Ve-1	2															2																		
Ve-2	2																2																	
P. paucimobilis	0																																	
B. bronchicanis	7																	5														2		
A. odorans	30									①										29														
Achromobacter Vd-1	4																				3													1
A. xylosoxidans	19																					18												1
A. tumefaciens	2																						2											
F. breve	1																							1										
F. meningosepticum	3																								3									
F. multivorum	1																	1																
F. odoratum	10																									5						4		1
A. alcaligenes	5																										3							2
A. lwoffii	13																①											10						2
A. calcoaceticus	27									1																			24					2
A. hemolyticus	5																													5				

[a] Species included in the AMS GNI data base. Numbers in boldface type represent identifications reported with probabilities of ≥0.90.

TABLE 3. Accuracy of identification of 419 isolates using appended messages and screening procedures to determine acceptance or rejection of AMS GNI reports

AMS GNI identification	≥0.9 Identification probability or "good confidence, marginal separation," no. (%)	All first choice except "questionable biopattern," no. (%)	All first choice No. (%)
Correct	285 (68.0)	304 (72.6)	307 (73.3)
Incorrect			
Without screen[a]	6 (1.4)	13 (3.1)	18 (4.3)
With screen	2 (0.5)	5 (1.2)	8 (1.9)
Unidentified			
Without screen	128 (30.5)	102 (24.3)	94 (22.4)
With screen	312 (31.5)	110 (26.3)	104 (24.8)

[a] Presence or absence of fruity odor, kanamycin susceptibility.

occurred on 5 of the 9 incorrect reports and 3 of the 41 correct reports.

Unidentified organisms. Of the 36 "unidentified" isolates belonging to species listed in the AMS GNI system data base, 12 (2 *Bordetella bronchicanis*, 4 *F. odoratum*, 3 *P. acidovorans*, and 3 *P. stutzeri*) had a second-choice identification with a low identification probability listed. One of these identifications was correct to the species level; two were correct to the group level. The four unidentified strains of *F. odoratum* represented 40% of the total number of isolates of this species examined during the study. All 12 reports from this group of organisms were appended with a message indicating whether or not the organism was saccharolytic; five of these messages proved incorrect.

In 24 of the 36 "unidentified" organisms, no second choice was listed. Since 19 of the 24 were shown by conventional biochemical testing to be relatively active organisms, it was felt that reports of this nature might signal a failure of the inoculating technique. These were accordingly retested with the AMS. In one-half (12 of 24), the identification remained unchanged. Eight organisms were correctly identified to the species level of ≥0.90 probability, and two more were identified at <0.90 probability. Two *Pseudomonas cepacia* isolates were incorrectly identified as *Agrobacterium tumefaciens* at a confidence of 0.99.

Overall accuracy. Ignoring identification probabilities, the first listed identification choice was correct in 307 of 419 (73.3%) instances and incorrect in 18 (4.3%); no identification was made on the remaining 94 (22%) isolates (Table 3). If all AMS-generated first-choice identifications except those appended with a "questionable biopattern" were accepted, the number of misidentifications would have been reduced to 13. Screening of all organisms for the characteristic fruity odor of *A. odorans* and *F. odoratum* and for the kanamycin resistance of *P. aeruginosa* reduced the number of misidentifications to 5 in 419 (1.2%). Alternatively, if reports with first-choice identification probabilities of <0.90 were accepted only if appended with the "good confidence, marginal separation" message, the number of misidentifications would have been reduced to 6. Screening of isolates for fruity odor and kanamycin susceptibility would further reduce the number of misidentifications to 2 in 419 (0.5%).

Time to identification. The AMS GNI system required an average of 14 h to identify the 270 isolates it reported with high-probability (≥0.90) biopatterns; the average identification time for this same group of organisms with the reference method was 44 h. The AMS GNI system took the full 18-h testing period to report on the remaining 149 microbes. For the 41 organisms in this subset which the AMS correctly identified and the 14 which it misidentified, reference testing took an average of 79 and 101 h, respectively. For the 94 isolates reported as unidentified by the AMS method, reference identification took an average of 112 h. For 60 of these 94 organisms, the primary battery of tests in the reference method had to be supplemented with procedures chosen from the secondary battery to achieve a definitive identification. The average time to report for these 60 was 145 h. The addition of oxidation-fermentation (O-F) basal medium with 1% fructose and electron microscopy or flagellar stain to our standard test battery would have resulted in the identification of 22 of the 60 isolates within 48 h. The further addition of acetate, citrate, phenylalanine, and 42°C growth would have led to the identification of 30 of the 60 within the same period. Overall, the average time required by the reference method for the identification of all 419 organisms was 65 h.

Use of the AMS GNI system in routine laboratory work. After the completion of the first phase of the study, the AMS GNI system was used in the clinical laboratory for the routine identification of all NFGNB other than pigmented strains of *P. aeruginosa*. A total of 130 isolates, including 79 nonpigmented *P. aeruginosa*, 23 *Pseudomonas maltophilia*, 3 *P. cepacia*, 2 *P. putida*, 1 *P. fluorescens*, 3 *A. odorans*, 2 *Moxarella nonliquifaciens*, 3 *A. lwoffii*, 10 *A. calcoaceticus* var. *anitratus*, 1 *Agrobacterium xylosoxidans*, 1 *Flavobacterium meningosepticum*, 1 *F. odoratum*, and 1 *Achromobacter* Vd-1 were examined. Rejection of first-choice identifications with a "questionable biopattern" special message resulted in correct identification of 122 (93.8%) of the isolates in an average time of 11 h. Six organisms (4.6%) (1 *P. aeruginosa*, 1 *P. fluorescens*, 2 *P. maltophilia*, 1 *P. putida*, and 1 *A. calcoaceticus*) were misidentified; the remaining two failed identification.

DISCUSSION

The NFGNB have become an important cause of nosocomial infections in immunocompromised patients (33). The frequency with which these organisms produce epidemic outbreaks and display resistance to antimicrobial agents (13, 14, 21, 40) underscores the importance of their rapid identification and antimicrobial susceptibility testing. Unfortunately, identification of many NFGNB is complicated by their fastidious nature and limited biochemical activity.

There have been numerous commercial systems introduced that are capable of identifying aerobic and facultative gram-negative bacilli (1, 2, 8, 11, 19, 20, 23, 25–28, 30, 31, 37, 39). Most incorporate conventional media and biochemical tests into a single strip or a microtiter test plate. They offer ease of inoculation, computer-assisted interpretation of results, simple quality control procedures, and standardization of identification methods from laboratory to laboratory. However, since most of these depend on the activity of organisms in conventional media, they reliably identify only the most biochemically active NFGNB (1, 2, 27, 28, 30, 37). Most of these kits require subjective interpretation of subtle color changes in the medium wells; precision, therefore, varies from observer to observer (2, 19, 25, 28).

Recently, several automated or semiautomated bacterial identification systems have been introduced which claim the capacity to identify NFGNB as well as *Enterobacteriaceae*

(3–7, 9, 17, 18, 22, 32–34, 38). All offer the advantage of automated reading and recording of biochemical results, thus eliminating subjective interpretation of endpoints. The AMS has the additional advantage of being fully automated once the 30-well card has been inoculated and introduced into the reader-incubator module. All systems have been limited in the number of NFGNB they can identify to the species level (7, 32–34). The reported accuracy of these systems for the identification of the NFGNB included in their data bases has generally been quite high (3–7, 9, 17, 18, 22, 32–34, 38). Their effectiveness in routine clinical use, however, has been compromised not only by the limited number of NFGNB that they are capable of recognizing, but also by their tendency to falsely identify non-data-base organisms (7, 33). In one study of the AMS EBC-Plus (33), over one-third of non-data-base organisms were inappropriately identified.

With the introduction of the AMS GNI, the number of NFGNB species or groups of species this system was capable of identifying increased from 7 to 30 (35). Although one investigator (38) has utilized the AMS GNI card to test all isolates whose identification by EBC-Plus differed from the reference technique, our study is the first to compare the AMS GNI card directly with a standard reference method in the identification of a balanced spectrum of NFGNB. We found the accuracy of the card to be high. Of the 356 tested organisms contained in the data base, only 13 (3.7%) were misidentified; 36 (10.1%) remained unidentified. Further, the AMS GNI correctly reported 58 of 63 (92%) of those species not found in the AMS database as "unidentified." Overall, although nearly one-quarter of the test organisms could not be identified by the AMS, only 4.3% were incorrectly identified to the species level. These results are impressive when one considers that the isolates were tested under "worst case" conditions. We purposely attempted to select equal numbers of isolates from within every organism group found in the AMS data base as well as large numbers of organisms not contained within the data base. Moreover, we excluded from the testing all pigmented strains of *P. aeruginosa* identified by colonial morphology, cytochrome oxidase reaction, and antimicrobial susceptibility profile. Finally, we accepted all first-choice identifications regardless of identification probability.

The number of erroneous results could have been substantially reduced (4.3 to 1.5%) by accepting only high-probability ($\geq$0.90) identifications. However, this course would have resulted in a decrease in the number of correct identifications from 307 to 266 and a corresponding increase in the number of "unidentified" organisms. If only low-probability identifications accompanied by the "good confidence, marginal separation" message had been accepted, the number of errors would have decreased from 18 to 6 (1.4% error rate), whereas the number of correct identifications would have shown a smaller decline than that described above (307 to 285). In our judgment, the most satisfactory approach was to accept all first-choice identifications unless accompanied by the message, "questionable biopattern." This decreased the number of errors from 18 to 13 and had a negligible impact on correct identifications (307 versus 304). Furthermore, 8 of the resulting 13 errors were detected by screening isolates for odor and antimicrobial susceptibility patterns, reducing the total number of misidentifications to 5 (1.2%).

Undoubtedly, the results obtained during the second phase of our study more closely reflect the performance that can be expected in routine clinical laboratory work than those obtained during the first phase. One hundred and thirty consecutive NFGNB isolates (pigmented *P. aeruginosa* excluded) were examined as they were recovered in the clinical laboratory. All first identifications were accepted as final unless they were accompanied by a "questionable biopattern" message. In 122 instances (93.8%) these identifications proved to be correct; two organisms (1.5%) failed identification. This represents a substantial improvement vis-à-vis the first phase of the study. This is not surprising, since most of the commonly isolated species of NFGNB tend to be biochemically active and thus more readily identified. Interestingly, the misidentification rate (4.6%) was higher than that observed during the first phase. The erroneously characterized organisms belonged to species with very high correct identification rates in the earlier study, suggesting that there had been some minor degradation in technical performance during the clinical phase of testing. The GNI card package insert (36) contains a warning that variations in bacterial inoculum density may affect accuracy of identification. During the first phase of our study, all inoculum suspensions were prepared by a single technologist; a number of technologists were involved in the clinical study. It is conceivable that this resulted in some variation in the inoculum density during the clinical study and contributed to the slightly higher error rate observed.

Regardless of the identification criteria utilized, a small but significant number of NFGNB fail to be identified in the AMS GNI. In a clinical laboratory setting these isolates would, presumably, require evaluation by alternate procedures. In our study such organisms proved difficult to characterize, requiring an average of 112 h with our reference method. When this time is added to the 18 to 24 h required for their initial testing on the AMS, the average turnaround time would be over 130 h. This time period could have been substantially shortened if it had been recognized that organisms failing identification on the AMS GNI would require a more comprehensive battery of conventional tests than were present in our initial panel. Specifically, the addition of 6 tests to our 18-test initial battery would have decreased the subset of the 94 unidentified isolates requiring supplemental reference tests from 60 to 30 and decreased the average time to identification from 112 to 81 h. Thus, the loss of one working day engendered by the initial testing of these isolates on the AMS system could be recovered by utilizing more exhaustive conventional procedures.

In summary, we found the AMS GNI, used in conjunction with simple screening tests and system-generated special messages, to be a rapid and reliable method of identifying NFGNB. Definite characterization of organisms failing identification with the AMS GNI often requires exhaustive testing by conventional methods.

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