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Microbial Colonization in a New Intensive Care Burn Unit

A Prospective Cohort Study

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• Renovation of an existing intensive care burn facility required closure for ten months. An interim eight-bed open intensive care ward (B) was established in a burn convalescence ward. The renovated unit (A) contained nine single-bed intensive care rooms and seven intermediate-level care beds in four rooms. Patients admitted to unit A were treated as a cohort. The first 25 admissions to unit A and the last 25 admissions to ward B meeting the inclusion criteria were compared. Microbial colonization was monitored by a fixed protocol of admission and multiple weekly sputum, wound, stool, and urine cultures. During intensive care, both cohorts exhibited the same incidence of gram-negative wound, sputum, and urine colonization. Occurrence of antibiotic-resistant organisms was the same. No evidence of bacterial cross-contamination was observed between A and B. A continuation of *Providencia stuartii* and *Pseudomonas aeruginosa* (type 15) endemics occurred in B. The collected data demonstrate that the A cohort was colonized with new, similar but distinct gram-negative organisms and indicate that cohort separation may be a practical way of eliminating endemic resistant gram-negative organisms from burn units.
(Arch Surg 1985;120:217-224)

Altered microbial ecology following burn injury is the result of the interaction of endogenous and exogenous microbial flora with injury-induced physical and immunobiologic host defects. The microbial component of this equation is, at present, the variable most easily influenced by therapy. Inappropriate growth of organisms is addressed, when possible, with antimicrobial agents. The continual effort to develop more active antimicrobials demonstrates the ability of organisms to adapt to treatment and perpetuate the adaptation.

The accumulation of resistant opportunists in the clinical burn care environment has been previously reported from this and other burn care facilities.[1-10] Successful methods for blocking such accumulation and spread by patient isolation and conditioned physical environments have been reported.[11-16] Despite the reported success, no single method of microbial containment of endemic organisms is commonly used.

The recent reconstruction of the intensive care unit of our burn center was accomplished while the center continued to receive patients. A temporary eight-bed open intensive care area was established in a physically separate ward located on the same hospital floor. The reconstruction required a ten-month use of the temporary facility. On completion, the new unit contained nine single-bed intensive care rooms and seven intermediate-level care beds in four rooms.

We attempted to observe the nature of microbial colonization in a cohort of patients separately admitted to this new facility within a burn center where strains of *Providencia stuartii* and an unusual serotype of *Pseudomonas aeruginosa* were endemic to the ward being used for burn care. The cohort admitted to the new facility was compared with a control cohort of patients treated under the same regimen who had been admitted to the eight-bed open intensive care area.

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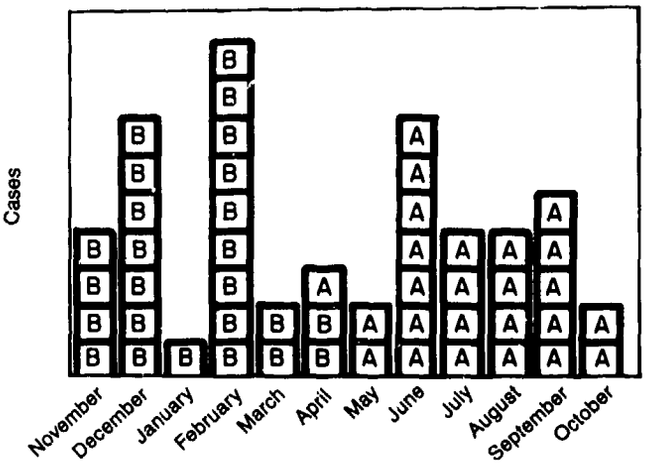

Fig 1.—Distribution of admission month to study. Each block represents one admission.

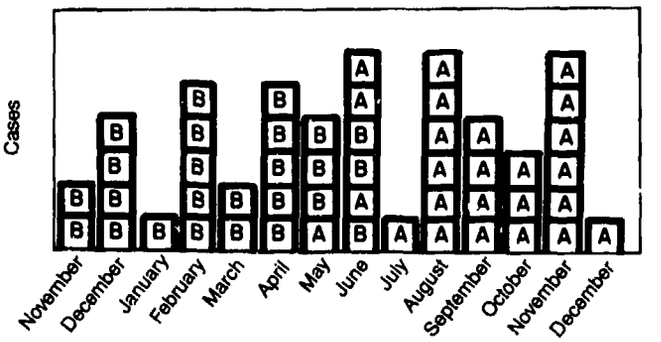

Fig 2.—Distribution of month of patient completion of study.

Table 1.—Demographic Comparison of Patients Studied

	Cohort A (New Unit)	Cohort B (Old Ward)
Age, yr (range)	37.0 (19-68)	40.0 (19-69)
Burn size, % (range)	45.6 (20-84)	45.8 (21.5-85)
Sex, M/F	23/2	21/4
Days in intensive care (range)	26.0 (4-70)	22.5 (3-105)
Total days (range)	49.0 (4-124)	47.0 (8-122)
Survival*	16 L, 9 D	14 L, 11 D

*L indicates lived; D, died.

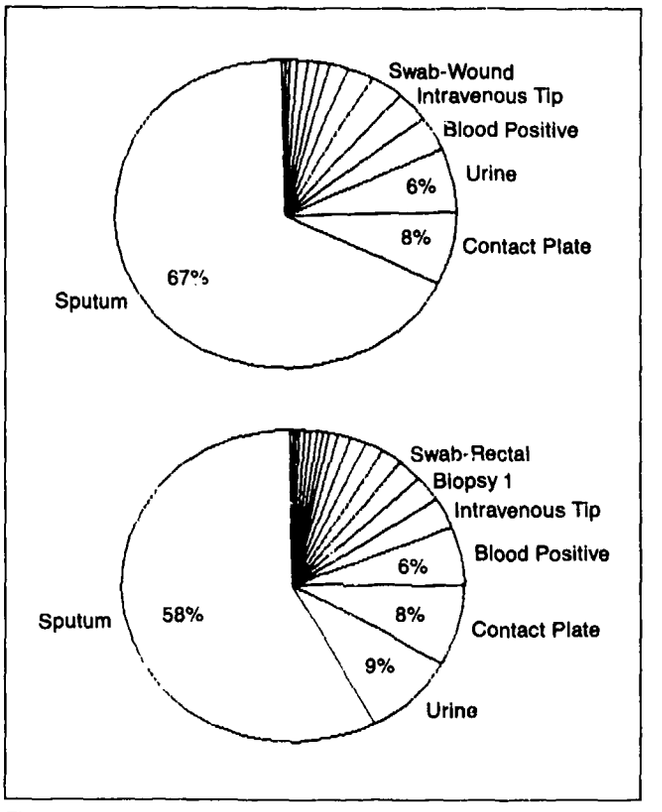

Fig 3.—Relative frequency of positive specimen sources in A (top) and B (bottom) cohorts.

This report demonstrates that endemic burn ward flora can be eliminated by cohort admission. Additionally, within the limits of the ten-month observation period in the new unit, there has been no accumulation of persistent bacterial flora.

METHODS AND MATERIALS

Cohort Design

Entry criteria for patients in this study were admission to our center within seven days of injury; burn size greater than or equal to 20% of the body surface; and age between 18 and 80 years. The last 25 patients meeting the criteria admitted to the open-bed temporary unit were designated cohort B. The first 25 patients meeting the criteria admitted to the new unit were designated cohort A. On discharge from intensive care, members of both cohorts were admitted to the same convalescent area. Colonization data were compared during the intensive phase of treatment.

Microbial Surveillance

Cultures were taken according to a fixed protocol from admission to discharge. Wound, urine, stool, and sputum cultures were taken on admission. For the first 30 days after admission, or longer if the patient remained in the intensive care unit, patients had sputum cultures taken three times per week and surface wound cultures, urine cultures, and stool cultures taken two times per week. Following 30 days of admission and transfer to the convalescence ward, sputum, stool, and urine were cultured once per week. Specimens were cultured by standard techniques; sputum and stool specimens were also plated on an additional plate of MacConkey agar containing 20 mg/L of gentamicin sulfate. Stool cultures with gentamicin-resistant organisms or *P aeruginosa* isolated were examined for other antibiotic resistances. Blood cultures, wound biopsy, or other specimens requiring invasive procedures were ordered on the basis of clinical indications by the attending physician. Organisms isolated within three days of admission were considered as not being acquired within this center and were excluded from analysis. This culture protocol was in effect prior to the start of this study.

Culture results were placed into an ongoing microbial surveillance computerized data base system designed for immediate physician access of past and pending cultures as well as a referral base for epidemiologic study. As is the practice at this institute, all culture results, including those positive for normal flora for nonburned patients, were reported. Antibiotic sensitivity tests

Table 2.—Patient Colonization by Gram-negative Aerobic Rods

Organism	Cohort A	Cohort B
Pseudomonas aeruginosa	6	12
Escherichia coli	10	8
Enterobacter cloacae	9	7
Klebsiella pneumoniae	7	7
Proteus mirabilis	3	4
Citrobacter diversus	3	3
Providencia stuartii	0	4
Enterobacter agglomerans	3	0
Pseudomonas putida	0	3
Acinetobacter anitratus	0	2
Citrobacter freundii	0	2
Enterobacter aerogenes	1	1
Klebsiella oxytoca	1	1
Alcaligenes faecalis	1	0

Table 4.—Patient Colonization by Selected Gram-positive Cocci and Yeasts*

Organism	Cohort A	Cohort B
Staphylococcus aureus		
ICU	14	19
Total	18	20
Enterococcus species		
ICU	7	11
Total	9	13
Candida species		
ICU	12	7
Total	14	12

*ICU indicates intensive care unit.

Table 3.—Patient Colonization by Gram-negative Aerobic Rods*

	Cohort A	Cohort B
Sputum		
ICU	16	18
Total	19	23
Wound		
ICU	11	14
Total	15	18
Urine		
ICU	4	4
Total	7	9

*ICU indicates intensive care unit.

Table 5.—Patients With Positive Blood Cultures

Organism	Cohort A	Cohort B
Candida (non-*albicans*)	5	4
Pseudomonas aeruginosa	2	5
Staphylococcus epidermidis	3	4
Candida albicans	3	2
Staphylococcus aureus	3	1
Escherichia coli	1	2
Klebsiella pneumoniae	1	2
Enterobacter cloacae	0	2
Streptococcus pneumoniae	0	2
Enterococcus species	0	2
Klebsiella ozaenae	0	1
Bacillus species	1	0
Citrobacter diversus	1	0
Flavobacterium 2L	1	0

were performed on *Staphylococcus aureus*, *P aeruginosa*, the predominant gram-negative isolate from sputum, wound, or urine culture, organisms isolated from the MacConkey gentamicin plate, all bacteria isolated from blood cultures, and any other isolate as requested. *Pseudomonas aeruginosa* isolates with a distinct antibiotic resistance pattern for each patient were serotyped using autoclaved suspensions and the international typing system (DIFCO).

Enteric specimens selected for particular antibiotic resistance patterns were examined for plasmid content by agarose gel electrophoresis. Conjugative transfer was attempted by agar matings and selection with appropriate combinations of antibiotics.[10] Matings were attempted into *Escherichia coli* C600 or a nalidixic acid–resistant mutant of an *Enterobacter cloacae* isolate from a patient in cohort B.

Data for epidemiologic review were retrieved using a computer-based system (Digital VAX-11 Datatrieve System). The retrieval system uses any one of the field names in the data base (subject headings) or any combination of fields to find desired records. Field names in the data base included such headings as patient name, patient location, date of specimen, source of specimen, organism isolated, quantity of organism isolated in the specimen, antibiotic sensitivity to tested drugs, the pattern of sensitivity to tested antibiotics, and the quantitative measurement of sensitivity to tested drugs (zone size).

Statistical comparisons of the frequencies of patient occurrences were made using Fisher's exact test (two-tailed).

Clinical Staffing

Prior to the anticipated completion date of the reconstruction, a plan was made to phase staffing into the new unit on a cumulative admission basis. That is, following admission of the first patient a nurse team was assigned to the new unit to care for that patient. Each series of teams moved from the open-bed unit to the new unit after two or more days' absence from all patient care. Such incremental staffing continued until the close of the B unit. Once assigned to the new unit, direct-care nursing staff did not return to the old unit. Other support personnel such as physical therapists, occupational therapists, and respiratory therapists remained on one of the other service as much as possible, but when such personnel were required on both units during the same day, the progression was from the A to the B cohort. Roentgenographic and other bedside teams were also directed to work on the new unit prior to working in the open unit. Physicians were aware of the staffing efforts and attempted to follow the same directional activity. Gown, glove, and mask procedures were the same on both services. Personnel in direct patient contact changed such attire when leaving the bedside. A separate gown change was made when travel was necessary between units.

RESULTS

Patients were admitted to this study over a 12-month period. The distribution of dates of admission to each cohort is presented in Fig 1. The 50 patients fitting the entry

Table 6.—Patient Colonization by Resistant Gram-negative Aerobic Rods*

Antibiotic	Cohort A	Cohort B
Gentamicin		
ICU	3	9
Total	4	10
Ticarcillin		
ICU	7	13
Total	8	15
Sulfonamide		
ICU	14	13
Total	19	16

*ICU indicates intensive care unit.

Table 8.—Patient Colonization With *Providencia* Plasmid Pattern*

Organism	Cohort A	Cohort B
Providencia stuartii		
ICU	0	2
Total	0	3
Enterobacter cloacae		
ICU	0	3
Total	1	3
Klebsiella pneumoniae		
ICU	0	1
Total	0	1
Enterobacter aerogenes		
ICU	0	1
Total	0	1
Citrobacter diversus		
ICU	0	1
Total	0	1
Total patients		
ICU	0	6†
Total	1	7

*ICU indicates intensive care unit.
†$P<.03$.

criteria were selected from a total of 167 admissions. The time of exit from the hospital for the cohorts is presented in Fig 2. A demographic comparison of the two cohorts is given in Table 1. As can be seen, there was no significant difference between the two groups in mean age, mean burn size, sex distribution, intensive care days, total days of hospitalization, or survival.

The frequency of culturing was similar in both cohorts. During the intensive phase of care, 962 and 792 isolates were recovered from cohort A and cohort B, respectively. Gram-negative aerobic rod (GNR) isolates numbered 262 and 371 in the A and B cohorts, respectively. The relative frequency of specimen sources yielding GNRs is presented in Fig 3. As expected from the sampling protocol, the sputum was the most common source for both cohorts. A comparison of the overall colonization frequencies for GNRs is given in Table 2. The table represents the ten most common colonizing GNRs in both cohorts. Statistical analysis for differences in the frequency of patient colonizations showed no significant preponderance. The frequency of

Table 7.—Pattern of Zones of Inhibition*

Antibiotic	*Enterobacter cloacae* Blood	*Enterobacter cloacae* Sputum	*Providencia stuartii* (Mean, 44 Isolates)
Amikacin	23.3 (S)	15.4 (I)	23.2 (S)
Gentamicin	22.2 (S)	6.0 (R)	6.0 (R)
Tobramycin	22.2 (S)	6.0 (R)	8.2 (R)
Ticarcillin	26.4 (S)	6.0 (R)	7.4 (R)
Mezlocillin	24.0 (S)	6.0 (R)	18.6 (S)
Piperacillin	25.0 (S)	6.0 (R)	20.0 (S)
Cefoxitin	20.3 (S)	6.0 (R)	23.3 (S)
Cefamandole	21.6 (S)	11.2 (R)	25.0 (S)
Moxalactam	28.6 (S)	28.6 (S)	31.5 (S)
Kanamycin	23.0 (S)	6.0 (R)	8.4 (R)
Neomycin	19.8 (S)	18.2 (S)	18.7 (S)
Streptomycin	17.2 (S)	18.8 (S)	17.9 (S)
Chloramphenicol	18.8 (S)	6.0 (R)	6.0 (R)
Sulfadiazine	6.0 (R)	6.0 (R)	6.0 (R)

*S indicates sensitive; I, intermediate; R, resistant.

colonization of the two cohorts by site of isolation is given in Table 3. In addition to patients colonized during the intensive phase of care (ICU), patients colonized for the first time after transfer to the convalescent ward were added to the ICU patients and presented as a total. Analysis of frequency of colonization was performed only during the intensive phase. Again there was no difference in patient colonization between cohorts. Colonization with *S aureus*, *Enterococcus* species and *Candida* species was also compared. Data are given in Table 4. The occurrence of patient colonization by those organisms was also not different between cohorts. In summary, on a taxonomic basis the occurrence of colonization of patients admitted to the older open ward and of those patients admitted to the new separate room unit was not different.

Blood cultures were positive in 12 patients in cohort A and 11 patients in cohort B. There were six patients in each cohort whose bacteremia was confined to a single organism. The number of patients with episodes of bacteremia due to differing organisms was six in the A cohort and five in the B cohort. Organisms recovered and the number of patients with positive blood cultures are given in Table 5. There was no significant difference in the number of patients with positive cultures or a predominant organism in either cohort. Of a total of 360 blood cultures, 75 cultures were positive in cohort A. Cohort B had 424 blood cultures taken, with 67 organisms recovered in 62 positive cultures.

The number of patients colonized with antibiotic-resistant GNR was examined next. The antibiotics gentamicin, ticarcillin, and sulfadiazine were examined as representative of the major classes of antibiotics used in the care of both cohorts. In addition, resistances to these antibiotics were the hallmark of *Providencia* and a type 15 *P aeruginosa* strain endemic in the burn center. The relative frequency of occurrence of antibiotic-resistant GNR is given in Table 6. There was a suggestion of decreased incidence of gentamicin and ticarcillin resistance in the A

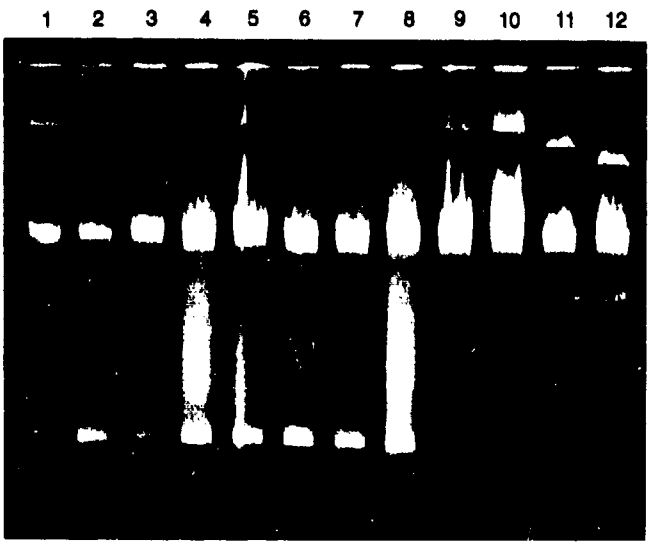

Fig 4.--Agarose gel electrophoresis patterns of lysates prepared from endemic *Providencia* (lane 1); *Enterobacter cloacae* blood isolate (lane 2); *E cloacae* mated to *P stuartii* (lanes 3 through 8); control plasmids (lanes 9 through 12).

cohort. The difference was not significant. Sulfonamide resistance as represented by sulfadiazine resistance was high in both cohorts.

Providencia stuartii was recovered in three patients in the B cohort and was not observed after February 1982. In early March, a gentamicin-resistant *E cloacae* was recovered from a sputum culture of a patient in the B cohort. Review of the patient's microbiology and treatment data showed the patient had a positive blood culture for *E cloacae* on his third postburn day. The patient was treated for ten days with aminoglycoside and β-lactam antibiotics, to which the organism was sensitive. The resistant organism was recovered 13 days following discontinuation of the antibiotic therapy. The antibiotic sensitivity of the blood isolate, the sputum isolate, and the last 44 *P stuartii* isolates of 1983 are given in Table 7. As can be seen, the blood isolate was sensitive to most tested drugs, whereas the sputum isolate had similar patterns to the *Providencia* strains and additional β-lactam resistances.

The similarity of the resistant *E cloacae* isolate to the *Providencia* isolates suggested the possibility that a plasmid transfer had occurred. An attempt was made to transfer the previously characterized 80-megadalton *Providencia* plasmid to the gentamicin-sensitive *E cloacae* blood isolate. A nalidixic acid–resistant mutant of the *E cloacae* was isolated by direct plating onto tryptic soy agar containing 100 mg/L of nalidixic acid. This mutant was mated with *P stuartii*.[10] The mating was successful with a frequency of transfer of 10^{-7} per donor cell. A gel electropherogram of the mated strains and progeny is presented as Fig 4. The *Providencia* plasmid is in the first lane; the blood isolated *E cloacae* is in the second. Lanes 3 through 8 are clones isolated after the mating. Lanes 9 through 12 contain control molecular weight marker plasmids at 96, 80, 60, and 34 megadaltons, respectively. One *Enterobacter* clone (lane 3) maintained the large plasmid and small plasmid of the

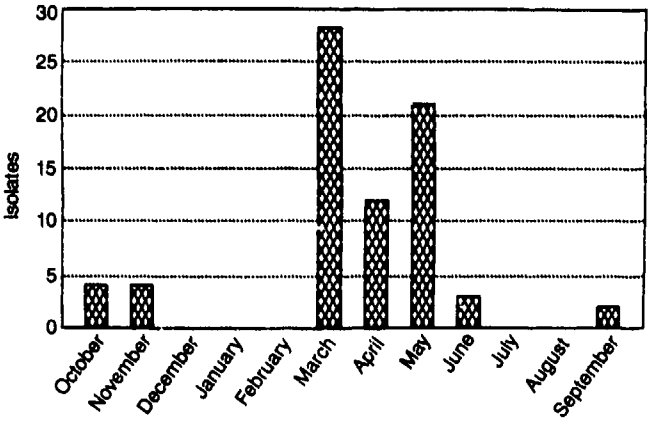

Fig 5.—Occurrence of non-*Providencia* enteric species with *Providencia* plasmid phenotype.

blood recipient strain, whereas the other clones eliminated the larger plasmid. Interestingly, the *Providencia* plasmid transferred resistance to β-lactam antibiotics like that seen in the sputum isolate. These resistances were not expressed in the *Providencia* donor. This phenomenon of host-specific expression of β-lactam resistance was also true in *E coli* transconjugants. All in vitro transconjugants expressed antibiotic sensitivity patterns identical to the sputum isolate.

Gram-negative rods from both cohorts were examined for antibiotic resistance patterns that mimicked the *Providencia* and *Enterobacter* patterns. Table 8 shows that six patients had organisms from five different species isolated that matched the *Providencia* pattern. Colonization occurred in one patient in the A cohort after transfer to the convalescent ward with an *E cloacae* strain that matches the pattern and also carries the 80-megadalton plasmid and expresses the same aminoglycoside resistance enzyme pattern as the *Providencia* strains. The occurrence of these strains is shown in Fig 5. The isolate in September was from the convalescent patient from the A cohort. Bacteria containing that plasmid have not been found since the discharge of that patient.

Each patient with a *P aeruginosa* isolate with a distinct antibiotic pattern was serologically examined. The endemic type 15 strain occurred in nine patients in the B cohort and was not seen in the A cohort ($P<.01$).

The overall consequences that the opening of the A cohort had on the two endemic organisms being followed are presented in Figs 6 and 7. Figure 6 shows the number of patients colonized with *P stuartii* during the last two years; colonization occurred in one patient in November colonized on admission to the new unit with a gentamicin-sensitive strain. This strain remained with that patient. Figure 7 shows that after more than five years type 15 *P aeruginosa* has been eliminated from this burn center following the move to the new ICU.

COMMENT

The purpose of this study was to examine the effects of admitting a cohort of burn patients into a new intensive care unit located in a burn center occupied with patients col-

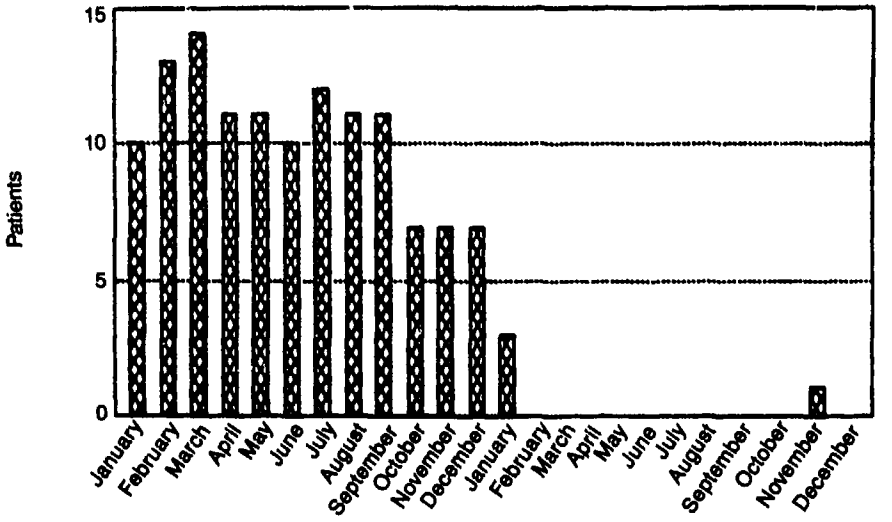

Fig 6.—Patients colonized with *Providencia stuartii* (endemic strain).

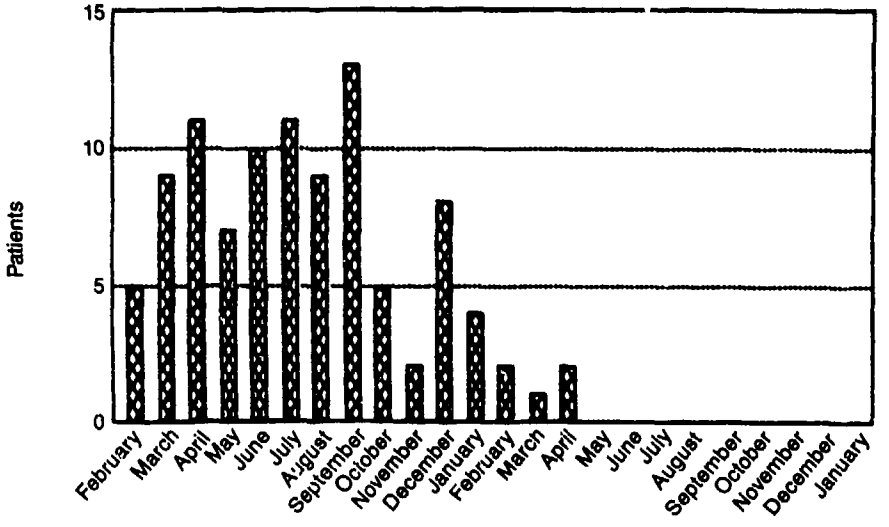

Fig 7.—Frequency of patients colonized with *Pseudomonas aeruginosa* (type 15).

onized with identifiable endemic antibiotic-resistant flora. The data collected clearly show that cohort admission and incremental staffing of the new unit prevented cross-contamination with the endemic flora. The data also demonstrate that colonization with residual endemic flora occurred in patients in the convalescent care area several months after the closing of the open-ward intensive care area. This fact is perhaps our most important observation. The patients with the least probability of becoming infected were a reservoir. Burn care facilities with areas of nonintensive care should include these areas in their microbial surveillance and infection control practices.[17]

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Discussion

BRUCE G. MACMILLAN, MD, Cincinnati: First, were all environmental cultures in cohort B negative? I presume that is something that the authors will want to comment on since contamination from these areas has been a problem in the past.

Second, was *P stuartii* and *P aeruginosa* type 15 colonization of cohort B related to cross-infection? I think that cross-infection should be a major concern in the design of new units and for the care of patients being treated in them. If we can avoid cross-infection by one means or another, resistance patterns can be eliminated.

Third, were the availability and use of hand-washing facilities the same in both cohort A and cohort B areas? The experience of the Cincinnati unit, Shriners Burns Institute, with two four-bed ICUs for the treatment of patients with acute injuries, has confirmed that rigid adherence to hand and forearm washing and reverse isolation techniques has been able to control the spread of endemic infections. Hand and forearm washing, in our experience, has been the most important feature in accomplishing this end.

STANLEY M. LEVENSON, MD, Bronx, NY: I would ask Dr McManus to clarify one point: the gown, glove, and masking technique. I started off with the idea, and perhaps it was so, that the routine of hand washing, and fresh gown, mask, and gloves, was done in between treating each patient particularly in the new unit. What confused me was the statement that the gown change was made when going from one unit to the other; therefore, it was not clear to me that in fact in the new unit a new gown, mask, and gloves were used when going from one patient to another.

MARIA D. ALLO, MD, Baltimore: Does Dr McManus believe that the separate rooms in his new unit had any influence on the development or the lack of development of resistant strains in that unit?

DR A. T. MCMANUS: First, as I stated in "Methods and Materials" section, when patients were colonized within the first three days of admission, the organisms were excluded; but there were no *Providencia* organisms on admission, and there were no type 15 *Pseudomonas* organisms on admission.

Ninety-five percent of our microbial surveillance is patient monitoring. We do about 5% environmental culturing: air, sinks, floors. These two gram-negative organisms could never, over the five years that I have tracked them, be found anywhere specifically in the environment, which leads to the next question.

As I have stated, I believe that the reservoir for these organisms was the patient population itself, and it is simply a matter of transferring resistant organisms from one patient to the next, which is what we have apparently stopped, at least with these two organisms.

In the new unit, there is a sink in every room, and there are two sinks at the nursing station, as compared with two sinks for eight beds in the old unit. I think that does make a difference.

When it was necessary for primary patient care personnel, ie, a nurse, to move from one ward to the next, there was a gown available just outside the cubicle area. That is where they would change gowns, and they would change when they went to the other ward. It was not the gown that they were wearing during patient care.

DR LEVENSON: Between patients, was there a new gown put on for each?

DR MCMANUS: Yes.

With reference to the value of single units, I do not think that answer is totally in yet, but I can say that in the 12 months that we have been in the new unit there have been no predictable common flora. So I think, at least in the one year's experience, we have not developed a resistant burn wound flora.

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