Simultaneous Nosocomial Outbreaks Caused by Multiply Resistant *Klebsiella pneumoniae* Types 2 and 30

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Two simultaneous *Klebsiella pneumoniae* outbreaks resistant to multiple antibiotics are reported. The closely related strains were distinguished by comparing the zone diameters obtained by disk diffusion susceptibility testing but not by interpretations of susceptible, resistant, or intermediate. Similar conclusions were made when minimal inhibitory concentrations were considered. The separation was subsequently confirmed by capsular serotyping into types 2 and 30. The data presented suggest that routine antimicrobial susceptibility patterns obtained from disk diffusion and/or minimal inhibitory concentrations may serve as an inexpensive and reliable typing system for epidemiological investigations.

In August 1977 a new pattern of antibiotic-resistant *Klebsiella pneumoniae* was noted on the genito-urinary (GU) and the medical services (Table 1). The first case on the medical service was in a leukemic patient (patient 1) who had received gentamicin therapy several weeks before the isolation of the resistant organism. *Klebsiella* was not isolated from him previously. Each of the subsequent patients infected on the medical service, with the exception of patients 3 and 6, were in the medical intensive care unit at some time before documentation of infection. All of the patients subsequently infected were in the same room or adjacent beds to a previously infected patient.

K. pneumoniae strains with the same antibiogram were isolated from seven medical patients from the following sites: patients 4, 5, and 6 from sputum; patients 1 and 5 from urine; patient 3 from a cellulitis of the foot; patient 2 from blood; and patients 1, 5, and 7 were shown to be intestinal carriers. Patients 1, 2, 5, and 7 had Foley catheters, and patients 5 and 7 were on type MA-1 respirators. All of the patients had serious underlying disease and five of the seven died during hospitalization. Four of the five patients who died were autopsied and three of them (patients 1, 4, and 7) had the resistant *K. pneumoniae* cultured from lung tissue or abdominal fluid at autopsy.

The pattern of infection or colonization with the second resistant strain (Table 1, patients 8 through 13) in the GU service was somewhat different from that in the medical service although it occurred in the same time period. Before the development of positive cultures, all

of the GU patients had cystoscopy with prophylactic gentamicin before the operation. All of these patients were in the GU ward. With the exception of patient 13, all of the patients had more than one positive urine culture with colony counts greater than 10^5 colony-forming units per ml, but only patient 12 had clinical signs of infection and was treated with amikacin (to which the organisms remained susceptible).

On 20 October a large-scale program of culturing the environment, patients, and personnel was undertaken to determine if there was a common reservoir for the *Klebsiella*. The incorporation of 4 µg of gentamicin per ml into Mueller-Hinton agar plates was used to select for the resistant strains. Of 102 environmental cultures, none was positive for the implicated strains. Of 386 cultures from 10 patients and 45 personnel, only 2 were positive. One was from patient 6 who had previously had positive cultures and the other was from patient 7 who was shown to have gastrointestinal colonization. The latter patient subsequently died, and autopsy cultures grew out the resistant organism from the lungs and pleural fluid.

Different patterns of resistance were noted when the zone diameters from the disk diffusion susceptibilities were compared for the aminoglycoside antibiotics (Table 1). Those of isolates from medical patients were different from those of GU patients. This was not evident when strains were considered only as susceptible, resistant, and intermediate (Table 1). Subsequent minimal inhibitory concentrations (MIC) for 50 of the isolates were determined for gentamicin and amikacin by the method of Washington and

TABLE 1. Patients having positive cultures for strains of *K. pneumoniae*

PATIENT	¹		SERVICE	²										SERO-TYPE
	DATE 1977	SOURCE		AMP	CEP	TET	KAN	GEN	COL	NIT	TOB	AMK	NAL	
1	8/25	Urine	MED	R/6	R/11	S/23	R/10	R/12	S/12	S/18	I/12	S/21	S/23	30
2	8/29	Blood	MED	R/6	R/6	I/15	R/10	R/9	S/11	R/11	R/8	S/18	R/6	30
3	9/7	Cellulitis	MED	R/6	R/12	S/23	R/12	R/11	S/12	S/20	R/11	S/24	S/23	30
4	10/6	Sputum	MED	R/6	R/6	S/18	R/9	R/9	S/11	I/16	R/10	S/17	S/20	30
5	10/11	Sputum	MED	R/6	R/11	S/23	R/12	R/11	S/11	S/17	R/11	S/21	S/23	30
6	10/12	Sputum	MED	R/6	R/8	S/19	R/12	R/11	S/11	R/11	R/11	S/20	R/12	30
7	10/30	Skin	MED	R/6	R/12	S/24	R/12	R/11	S/12	S/19	R/10	S/23	S/23	30
8	8/29	Urine	GU	R/6	I/16	S/21	R/13	R/6	S/11	S/17	R/8	S/20	S/21	2
9	9/7	Urine	GU	R/6	I/15	S/21	R/13	R/6	S/11	S/17	R/6	S/21	S/21	2
10	9/13	Urine	GU	R/6	I/15	S/22	R/10	R/6	S/11	I/15	R/6	S/20	S/21	2
11	9/23	Urine	GU	R/6	I/15	S/20	R/10	R/6	S/11	S/18	R/6	S/21	S/21	2
12	10/12	Urine	GU	R/6	I/15	S/20	R/10	R/6	S/11	S/18	R/6	S/19	S/21	2
13	10/18	Urine	GU	R/6	R/6	S/22	R/6	R/6	S/11	R/11	R/6	S/21	S/23	2

¹ Date of first positive culture.² Antibiogram of disk diffusion susceptibility tests expressed as interpretation of zone of inhibition diameter in millimeters. R, Resistant; S, sensitive; I, intermediate; AMP, ampicillin; CEP, cephalothin; TET, tetracycline; GEN, gentamicin; COL, colistin; NIT, nitrofurantoin; TOB, tobramycin; AMK, amikacin; and NAL, naladixic acid.

Barry (3). The GU strains had MICs greater than 200 µg/ml for gentamicin, whereas the medical strains had MICs of either 10 or 25 µg/ml for that antibiotic. All of the GU and medical isolates had MICs of either 1 or 3 µg/ml for amikacin. Our interpretation that two separate strains caused outbreaks in separate wards was subsequently confirmed by capsular serotyping done by the Enteric Section at the Center for Disease Control, Atlanta, Ga. The capsular serotype of one isolate from each of the 13 patients was determined. The medical strain was type 30 and the GU strain was type 2.

In our study, different mechanisms of resistance to the aminoglycosides appear to have caused the divergent MIC values obtained. MIC values greater than 100 µg/ml usually indicate an R-plasmid-mediated mechanism of resistance, whereas resistance with much lower MIC values usually indicates some other mechanism such as a permeability factor (1). Peter Kresel of Bristol Laboratories determined that a representative isolate of the type 2 strain possessed the aminoglycoside-inactivating enzyme aminoglycoside acetyltransferase, AAC(3)-III, which is believed to be plasmid mediated. He did not determine the mechanism of resistance for the type 30 strain. The different MIC values were also reflected in the zone diameters from the disk diffusion testing for gentamicin (Table 2).

In this case a zone of 6 mm (no zone) corresponded to an MIC of greater than 200 µg/ml, and moderate but resistant zones of 9 to 12 mm corresponded to MICs of 10 or 25 µg/ml. The MIC is more exacting and the method of choice. Since most laboratories use either disk diffusion or MIC for antimicrobial susceptibility testing, we contend that the patterns obtained offer an inexpensive and reliable typing system for epidemiological investigations under certain circumstances. Many laboratories record only the interpretations of the zone sizes and thereby lose a valuable epidemiological marker in the actual zone diameter.

The above interpretation of two simultaneous outbreaks was subsequently confirmed by capsular typing. However, even when the antibiograms are the same, typing may indicate several different strains to be involved in an outbreak. An example of this situation was reported by Perkins, Kundsinn, Bodman, and Gillece after a recent outbreak of antibiotic-resistant *Serratia marcescens* in which all the antibiograms were the same but the serotypes were different (Abstr. Annu. Meet. Am. Soc. Microbiol. 1978, C47, p. 285). A possible explanation for this situation would be an "epidemic" of an R-plasmid.

Capsular types 2 and 30 are closely related serologically (J. J. Farmer, personal communication). We do not know what the possibility of

TABLE 2. Comparison of the susceptibilities of two strains of *K. pneumoniae* to gentamicin

Isolates	Interpretation	Avg zone diam (mm)	Avg MIC (μ g/ml)
Medical	Resistant	10.5	15
GU	Resistant	6	>200

one typing changing into the other is; but we do not believe that this change occurred in our outbreak since two or more mechanisms of resistance to the aminoglycosides appeared to be involved. It can be speculated that if all of the isolates of an outbreak were of the same type, but the resistance mechanisms were shown to be different, one would have to conclude separate but simultaneous outbreaks.

The absence of positive cultures in personnel and in the environment coupled with the clustering of cases implicate person-to-person transmission. Hand carriage was the probable mode of spread, and infected or colonized patients were the probable main reservoir. The supposed presence of bacteria on the hands of personnel

was probably easily eradicated with handwashing (2). Although the original emergence of the multiply drug-resistant organisms was probably related to gentamicin exposure, the subsequent transmission between patients may not have required the same selective pressures.

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