Antimicrobial resistance of clinical isolates from 1987 to 1993 in Hong Kong

JM Ling, AF Cheng

A total of 122 968 specimens were culture-positive from 1987 to 1993 in a general hospital in Hong Kong, with skin and soft tissue specimens being the most common. Gram negative organisms were most frequently isolated and of these, *Escherichia coli* was the most common. Approximately 60% of *E coli* isolates were resistant to ampicillin. Imipenem was the most active of the β-lactam antibiotics against coliforms and *Acinetobacter* spp., and ceftazidime was most active against *Pseudomonas aeruginosa*. Coliform resistance to gentamicin was high, at approximately *20%*. The newer quinolones were quite active against the coliforms and *P aeruginosa*. Twenty-six per cent of *Staphylococcus aureus* were found to be resistant to methicillin. Resistance to ampicillin and chloramphenicol was high in *Haemophilus influenzae* isolates (27% and 13%, respectively), and more penicillin-resistant *Streptococcus pneumoniae* were found, increasing from nil in 1987 to 10% of *S pneumoniae* isolates in 1993.

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Introduction

Bacterial resistance has been emerging worldwide.[1] This has both clinical and financial implications for the therapy of infected patients.[2,3] Continuous surveillance is necessary to monitor changes in antimicrobial susceptibilities. Such information is important locally, to guide clinicians in their choice of therapy, and internationally, to contribute to the global picture of antimicrobial resistance.

This also provides a basis for hospitals to amend their antibiotic policy, with such changes being an important method for combatting the development of resistance.[4,5] Susceptibility data on specific organisms have been reported previously in Hong Kong.[6-10] However, there have been only two reports[11,12] on the prevalence of bacteria in clinical specimens and their resistance to antimicrobial agents. Consequently, we decided to analyse the distribution and antimicrobial resistance of individual bacterial organisms isolated from specimens collected in a teaching hospital in Hong Kong from 1987 to 1993.

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Materials and methods

The Prince of Wales Hospital (PWH) is a 1400-bed general teaching hospital situated in the New Territories, serving a population of approximately one million.[13] It also serves as a referral centre for other hospitals.

Bacterial isolates

Single patient isolates from specimens submitted to the Department of Microbiology at the PWH from 1987 to 1993 were studied. These were cultured and identified according to standard methods.[14,15]

Antimicrobial susceptibility testing

This was performed by the disc diffusion method and interpreted by the comparative method.[16] Antibiotic discs as listed (Tables 3 and 4) were from various suppliers (Becton Dickinson & Co., Maryland, US; MAST Laboratories Ltd., Merseyside, UK; Oxoid and Unipath Ltd., Basingstoke, UK). Detection of methicillin resistance in *S aureus* was performed on Mueller-Hinton agar (Oxoid, Basingstoke, UK) according to the method of French et al.[8] Penicillin-resistant *S pneumoniae* was detected using oxacillin (1 μg) in addition to a penicillin disc. Minimal inhibitory concentration of penicillin using the agar dilution method[16] was performed on any isolate which gave an annular

radius of less than 6 mm. Iso-sensitest agar (Oxoid, Basingstoke, UK) was used for non-fastidious bac teria and as a base for fastidious organisms. Chocolate agar was used for fastidious organisms and lysed horse blood agar for *Streptococcus* spp. (other than enterococci). Control strains included *E coli* NCTC10418 and NCTC11560; *P aeruginosa* NCTC10662; *S aureus* NCTC6571; and *H influenzae* NCTC11931.

Analysis of results

The data centre of the hospital generated monthly reports of the distribution of non-duplicate organisms isolated and their susceptibility to different antimicrobial agents. All information was then entered into a computer spreadsheet program and analysed.

Results

Table 1 shows the type or site of patient specimens from which microorganisms were cultured. A total of 122 968 specimens were positive for bacteria, fungi or parasites. The specimens which most frequently grew a potential pathogen were those from skin or soft tissue sites, constituting 29%, followed by urinary tract (24%) and lower respiratory tract (13%) isolates. Specimens from the genital tract, upper respiratory tract, eye, gastrointestinal tract, cerebrospinal fluid, body fluids, and bile which gave a positive culture result constituted less than 10% of the total culture-positive specimens.

Table 1. Type or site of patient specimens from which microorganisms were isolated

Type/site of specimen	No.	(%)
Skin and soft tissue	35 561	(29)
Urine	29 827	(24)
Lower respiratory tract	16 434	(13)
Genital tract	10 927	(9)
Ear, nose and throat	7873	(6)
Blood	5913	(5)
Eye	3611	(3)
Stool	3300	(3)
Other specimens*	9522	(8)
Total	122 968	(100)

* Includes cerebrospinal fluid and other body fluids, bile, and miscellaneous specimens

Table 2 shows the organisms isolated from different specimens. Stool parasites were not included as these were detected by microscopy and not cultured. Aerobic gram negative organisms were the most frequent isolate found (56%), followed by aerobic gram positive organisms (29%). Most of the fungi isolated belonged to *Candida* spp. (12%) Anaerobes constituted approximately 2%. *Escherichia coli* was the organism most often isolated, constituting 16% of the total, followed by *S aureus* (13%), enterococci (8%), *Pseudomonas* spp. and *Klebsiella* spp. (both approximately 7%). The other organisms constituted 6% or less of the total isolated.

Skin and soft tissue yielded similar proportions of gram negative organisms (47%) and gram positive organisms (44%) because *S aureus* (27%) was often isolated. Organisms from urine specimens yielded predominantly gram negative isolates (73%) with *E coli* being the most common (35%), followed by enterococci (14%), *Klebsiella* spp. (11%) and *Proteus* spp. [including *Morganella* spp. and *Providencia* spp. (10%)]. This profile is similar to that for growth from blood cultures, with gram negative isolates predominating (62%); *E coli* being the organism most frequently isolated (24%). The second most common organism from blood cultures was *S aureus* (12%). Most *Candida* spp. were isolated from the female genital and urinary tracts.

Table 3 shows the percentage resistance of coliforms to various antimicrobial agents. Approximately 60% of *E coli* and *Proteus* spp. were found to be resistant to ampicillin, but this was reduced to almost half in the presence of a β-lactamase inhibitor (sulbactam or clavulanic acid). The presence of either inhibitor significantly reduced the level of ampicillin resistance in *Klebsiella* spp. (97% vs 20% to 30%). However, a β-lactamase inhibitor only minimally reduced ampicillin resistance in the Enterobacters (i.e. *Enterobacter* spp., *Citrobacter* spp. and *Serratia* spp.). Resistance to the second-generation cephalosporins, cefaclor and cefuroxime, was low in *E coli* (5% to 7%), slightly higher in *Klebsiella* spp. (11% to 13%), and significant in *Proteus* spp. (34% to 42%). Resistance to ceftazidime and aztreonam was low in these three groups. Only imipenem was highly active against the Enterobacters. It was surprising to find that 11% of *Proteus* spp. were resistant to imipenem as this is more than would be expected.

The level of resistance to gentamicin and tobramycin was high among these four coliforms (10% to 31%) but resistance to netilmicin was low (4% to

Table 2. Distribution of microorganisms isolated from patient specimens

Organism	Skin and soft tissue	Urine	Blood	Others	Total
	No. (%)	No. (%)	No. (%)	No. (%)	No. (%)
Aerobes					
Gram negative organisms					
Escherichia coli	3432 (9.7)	10 322 (34.6)	1442 (24.4)	4231 (8.2)	19 427 (15.8)
Pseudomonas spp.	3329 (9.4)	1823 (6.1)	366 (6.2)	3759 (7.3)	9277 (7.5)
Klebsiella spp.	2143 (6.0)	3159 (10.6)	560 (9.5)	3043 (5.9)	8905 (7.2)
Enterobacters	2062 (5.8)	2300 (7.7)	285 (4.8)	2569 (5.0)	7216 (5.9)
Proteus spp.	2101 (5.9)	2989 (10.0)	266 (4.5)	887 (1.7)	6243 (5.1)
Acinetobacter spp.	2526 (7.1)	907 (3.0)	267 (4.5)	2310 (4.5)	6010 (4.9)
Haemophilus influenzae	158 (0.4)	2 (0.0)	27 (0.5)	3474 (6.7)	3661 (3.0)
Salmonella spp.	31 (0.0)	27 (0.0)	214 (3.6)	2404 (4.7)	2676 (2.2)
Others	986 (2.8)	139 (0.5)	245 (4.1)	3918 (7.6)	5288 (4.3)
Total	16 768 (47.2)	21 668 (72.6)	3672 (62.1)	26 595 (51.5)	68 703 (55.9)
Gram positive organisms					
Staphylococcus aureus	9418 (26.5)	929 (3.1)	680 (11.5)	5458 (10.6)	16 485 (13.4)
Enterococci	3020 (8.5)	4209 (14.1)	287 (4.9)	2218 (4.3)	9734 (7.9)
Group A streptococci	383 (1.1)	2 (0.0)	34 (0.6)	263 (0.5)	682 (0.6)
β-haemolytic streptococci (other)	985 (2.8)	358 (1.2)	230 (3.9)	2232 (4.3)	3805 (3.1)
α-haemolytic streptococci	1151 (3.2)	118 (0.4)	258 (4.4)	872 (1.7)	2399 (2.0)
non-haemolytic streptococci	602 (1.7)	170 (0.6)	38 (0.6)	394 (0.8)	1204 (1.0)
Streptococcus pneumoniae	61 (0.2)	0 (0.0)	139 (2.4)	889 (1.7)	1089 (0.9)
Others	105 (0.3)	125 (0.4)	82 (1.4)	130 (0.3)	442 (0.4)
Total	15 725 (44.2)	5911 (19.8)	1748 (29.6)	12 456 (24.1)	35 840 (29.1)
Fungi					
Candida spp.	1127 (3.2)	2151 (7.2)	101 (1.7)	10 791 (20.9)	14 170 (11.5)
Others	54 (0.2)	25 (0.0)	4 (0.0)	180 (0.3)	263 (0.2)
Total	1181 (3.4)	2176 (7.3)	105 (1.8)	10 971 (21.2)	14 433 (11.7)
Anaerobes					
Anaerobic bacilli	1192 (3.4)	0 (0.0)	341 (5.8)	341 (0.7)	1874 (1.5)
Anaerobic cocci	689 (1.9)	0 (0.0)	46 (0.8)	52 (0.0)	787 (0.6)
Total	1881 (5.3)	0 (0.0)	387 (6.5)	393 (0.8)	2661 (2.2)
Others	6 (0.0)	72 (0.2)	1 (0.0)	1252 (2.4)	1331 (1.1)
Total	35 561 (100.0)	29 827 (100.0)	5913 (100.0)	51 667 (100.0)	122 968 (100.0)

5%) with the exception of the Enterobacters in which 14% were resistant. Amikacin was the most active of the aminoglycosides against these four organisms.

Susceptibility to the quinolones and co-trimoxazole was mainly tested on strains isolated from the urinary tract. There was a moderately low level of resistance to the newer quinolones (1% to 9%), but a high level of resistance to co-trimoxazole (25% to 41%).

Table 4 shows the percentage resistance of *P aeruginosa* and *Acinetobacter* spp. to various anti-

Table 3. Percentage resistance of coliforms to various antimicrobial agents

Antimicrobial agents	% Resistance (No. tested)							
	E coli		*Klebsiella* spp.		Enterobacters		*Proteus* spp.	
β-lactams								
Ampicillin	58	(19 123)	97	(8712)	89	(7214)	65	(6186)
Unasyn	30	(5827)	28	(1603)	50	(1095)	26	(1404)
Augmentin	27	(4193)	20	(1253)	66	(816)	36	(1126)
Cefaclor	5	(7310)	13	(2328)	56	(1485)	42	(2268)
Cefuroxime	7	(18 455)	11	(8722)	52	(6834)	34	(6062)
Ceftazidime	3	(5717)	7	(3856)	36	(3674)	5	(2321)
Imipenem	0	(7340)	0	(4323)	3	(3911)	11	(2861)
Aztreonam	2	(1176)	7	(855)	30	(807)	3	(509)
Aminoglycosides								
Gentamicin	20	(15 617)	11	(7731)	24	(7058)	14	(5268)
Netilmicin	5	(13 767)	4	(6358)	14	(5301)	4	(4391)
Tobramycin	19	(3953)	11	(2758)	31	(2603)	10	(1564)
Amikacin	0	(10 112)	0	(6058)	3	(5079)	1	(3560)
Quinolones								
Nalidixic acid	6	(10 202)	13	(3074)	21	(2096)	12	(2785)
Ofloxacin	2	(8785)	4	(2548)	9	(1878)	2	(2386)
Ciprofloxacin	1	(6511)	2	(1833)	6	(1305)	1	(1593)
Others								
Co-trimoxazole	41	(18 351)	25	(8716)	36	(6833)	40	(6054)

microbial agents. Except for carbenicillin and ofloxacin, the antibiotics tested were active against 90% or more of *P aeruginosa* isolates, with ceftazidime being the most active. For those that were resistant, 19% or more of isolates showed resistance to the antibiotics tested, with the exception of ceftazidime and piperacillin, to which 11% and 14% were resistant, respectively. The acinetobacters also had high resistance levels, with 20% and up to 91% being resistant to the antibiotics tested, except imipenem and amikacin, to which 3% and 8% were resistant, respectively.

There was a slight change in resistance to gentamicin in the coliforms, *P aeruginosa*, and *Acinetobacter* spp., although resistance to ofloxacin increased from 1987 to 1993. Methicillin and gentamicin resistance in *S aureus* appears to have declined, approximately from 20% to 30%. Ofloxacin resistance has increased from approximately 10% to 30%. The level of resistance to fusidic acid remained low at 2% to 4%. All strains were sensitive to vancomycin. Ampicillin-resistant *H influenzae* increased to 31% and chloramphenicol resistance also rose (7% to 20%). However, the level of resistance to co-trimoxazole and cefaclor, although high (up to 42% and 20%), declined in 1993 to 11% and 2%, respectively.

There was an increase in resistance to penicillin in *S pneumoniae* isolates from nil in 1987/1988 to 10% in 1993. All penicillin-resistant strains were isolated from sputum. Resistance to tetracycline, chloramphenicol, and erythromycin remained high at 71%, 24%, and 17%, respectively. Resistance to cefaclor was low, at an average of 3%.

A comparison of antimicrobial resistance in gram negative organisms as reported in various studies[11,12,17] is shown in Table 5. Generally, the level of antimicrobial resistance found in our study is similar to previous findings[11] except for the increased resistance to cefuroxime, ofloxacin, and co-trimoxazole seen in *E coli* and *Klebsiella* spp., and to carbenicillin in *P aeruginosa*. Hospital and community isolates of

Table 4. Percentage resistance of *Pseudomonas aeruginosa* and *Acinetobacter* spp. to various antimicrobial agents

Antimicrobial agents	% Resistance (No. tested)		
	P aeruginosa	*Pseudomonas* spp.	*Acinetobacter* spp.
β-lactams			
Carbenicillin	19 (6856)	31 (2215)	61 (3283)
Piperacillin	7 (6862)	14 (2216)	65 (3280)
Ceftazidime	4 (6872)	11 (2294)	76 (3674)
Imipenem	6 (5044)	19 (1070)	3 (3652)
Aztreonam	7 (1185)	32 (385)	91 (922)
Aminoglycosides			
Gentamicin	10 (6903)	29 (2381)	44 (5761)
Netilmicin	7 (5422)	25 (1713)	22 (4309)
Tobramycin	7 (5681)	30 (1843)	29 (5761)
Amikacin	6 (6881)	24 (2264)	8 (5007)
Quinolones			
Ofloxacin	35 (304)	46 (208)	20 (594)
Ciprofloxacin	8 (85)	32 (78)	20 (317)

E coli and *Klebsiella* spp. were similarly resistant. Community isolates of the Enterobacters, *Proteus* spp., and *P aeruginosa* showed less resistance, with the exception of the quinolones, to which they were more resistant. Organisms in this locality were much more resistant than those isolated in the United Kingdom, particularly to the aminoglycosides. The proportion of ampicillin-resistant *E coli* isolates was similar.

Discussion

This study shows that the most common types of specimens from hospital patients which yielded a positive culture result were those from skin and soft tissue sites (29%) and the urinary tract (24%). This contrasts with the situation in the community, where the respiratory and urinary tracts were the most common sources.[12] It was expected that gram negative organisms would constitute more than 50% of the total organisms isolated. *Escherichia coli* was the most frequent isolate. Gram positive organisms constituted approximately 30% of the total, with *S aureus* being frequently found, particularly from skin and soft tissue sites. Of the organisms isolated, 12% were fungi, and most were *Candida* spp.

It was not surprising to find that approximately 60% of *E coli* and *Proteus* spp. were resistant to ampicillin, as ampicillin was the most frequently prescribed antibiotic at the PWH (approximately 30 000 500-mg doses of ampicillin and 40 000 250-mg doses of amoxicillin monthly). Although the inclusion of a β-lactamase inhibitor can reduce resistance to ampicillin, approximately 30% of *E coli* and *Proteus* spp. were still resistant. The second-generation cephalosporins, of which cefuroxime was the most commonly prescribed (more than 6 000 750-mg doses monthly), were reliably active against *E coli* but not against other coliforms. Of the β-lactam drugs, imipenem was the most active, although 11% of *Proteus* spp. were resistant to it.

Resistance to gentamicin and tobramycin was high, with approximately 20% of isolates showing resistance. Surprisingly, more coliforms than *P aeruginosa* were found to be resistant to gentamicin. Although many coliforms showed resistance to nalidixic acid, most were sensitive to the newer quinolones. As nearly 40% were resistant to co-trimoxazole, this drug may be unsuitable for the treatment of urinary tract infections caused by gram negative organisms. Apart from carbenicillin, all β-lactams and aminoglycosides tested were active against *P aeruginosa* but none were reliably active against other *Pseudomonas* spp. Most of the drugs tested were not active against *Acinetobacter* spp., with the exception of imipenem and amikacin.

Concomitant with increased use of the newer quinolones, an increased level of resistance to these drugs was seen in the gram negative organisms and *S aureus*. More restricted use would help keep the resistance level low. The problem of methicillin-resistant *S aureus* (MRSA) has stabilised compared with the situation in the 1980s when 25% to 30% of *S aureus* isolates were MRSA.[8] This is probably attributable to the use of netilmicin instead of gentamicin since 1989, as most MRSA are susceptible to netilmicin.[8] Although all strains were sensitive to vancomycin and most to fusidic acid, the former should be used as a reserve drug and the latter in combination with another anti-staphylococcal agent to prevent development of resistance.

Although it is worrying to find resistance to penicillin in the pneumococci, all penicillin-resistant strains isolated were from sputum and may not represent true pathogens. Resistance to tetracycline has remained high.[6] Fewer resistant organisms were isolated in the community[12] than in the hospital, with the exception of the quinolone-resistant organisms. This is probably due to the frequent use of quinolones in the community (unpublished observation). There were also many more resistant isolates found in this locality than in other parts of the world.[17,18]

Resistance to ampicillin, co-trimoxazole, and gentamicin have remained high in this locality. The newer quinolones are still active against most gram negative bacteria. Ampicillin resistance was also high among *H influenzae* and penicillin resistance in *S pneumoniae* is beginning to pose a threat. By contrast, the problem of MRSA has stabilised.

Table 5. Comparison of the percentage resistance to antimicrobial agents found in gram negative organisms

Antimicrobial agents	% Resistance																
	E coli				*Klebsiella* spp.			Entero-bacters		*Proteus* spp.			*P aeruginosa*			*Pseudo-monas* spp.	
	A*	B†	C‡	D§	A	B	C	A	C	A	C	D	A	B	C	A	D
β-lactams																	
Ampicillin	58	55	63	44	97	89	95	89	61	65	18	15	-	-	-	-	-
Cefuroxime	7	1	10	-	11	8	10	52	33	34	18	9	-	-	-	-	-
Ceftazidime	3	-	0	-	7	-	10	36	0	5	0	-	4	-	11	11	1
Carbenicillin	-	-	-	-	-	-	-	-	-	-	-	-	19	23	-	31	8
Aminoglycosides																	
Gentamicin	20	19	17	3	11	18	10	24	6	14	14	2	10	12	4	29	5
Tobramycin	19	-	-	-	11	-	-	31	-	10	10	-	7	11	-	30	4
Quinolones																	
Nalidixic acid	6	6	19	-	13	11	19	21	11	12	27	-	-	-	-	-	-
Ofloxacin	2	0	6	-	10	-	10	9	6	2	0	-	35	-	15	46	-
Ciprofloxacin	1	-	-	4	2	-	-	6	-	1	-	1	8	-	-	32	6
Others																	
Co-trimoxazole	41	34	35	20	25	26	24	36	0	40	27	45	-	-	-	-	-

* A = this study
† B = Lim W, et al. J HK Med Assoc 1991.[11]
‡ C = Ling JM, et al. HK Pract 1993.[12]
§ D = MacGowan AP, et al. J Antimicrob Chemother 1993.[17]

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