Evidence

Études

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Risk factors for resistance to "first-line" antimicrobials among urinary tract isolates of *Escherichia coli* in children

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Abstract

Background: There are increasing concerns regarding antimicrobial resistance in Canada. Data are limited on the prevalence, patterns of resistance and risk factors associated with resistant organisms, including coliforms, in children. This study was done to address these issues as they relate to urinary tract isolates of *Escherichia coli* in a tertiary care pediatric centre in Ottawa.

Methods: A surveillance study was conducted from December 1992 to December 1994. Susceptibility testing of urinary tract isolates of *E. coli* was performed using a panel of antimicrobial agents. A case–control study was also conducted for subjects with isolates resistant to trimethoprim–sulfamethoxazole (T-S), this drug being used a representative "first-line" agent.

Results: A total of 1636 consecutive isolates were obtained from 967 subjects. Of the 1636 isolates, 736 (45.0%) were resistant to ampicillin, 514 (31.4%) were resistant to T-S, 363 (22.2%) were resistant to both ampicillin and T-S, and 27 (1.7%) were resistant to both ampicillin and gentamicin. In the case–control study 274 children with isolates resistant to T-S were matched with 274 children who had T-S-sensitive isolates obtained during the study period or the preceding or subsequent 6 months. Multivariate analyses indicated that subjects who had received antimicrobials for more than 4 weeks in the previous 6 months were about 23 times more likely to have isolates resistant to T-S than were subjects without this risk factor (odds ratio [OR] 23.4, 95% confidence interval [CI] 12.0–47.6). Children with genitourinary tract abnormalities were 2.4 times more likely to have resistant isolates than those without such abnormalities (95% CI 1.2–4.5). Compared with children who had no hospital admissions in the previous year, those with 1 admission in that period were more likely to have resistant isolates (OR 2.3, 95% CI 1.4–7.5), as were those with 2 or more admissions in that period (OR 3.2, 95% CI 1.1–4.8). Compared with children aged 2–6 years, children under 2 years of age were less likely to have resistant isolates (OR 0.3, 95% CI 0.2–0.8).

Interpretation: Selective antimicrobial pressure and multiple admissions to hospital were among the risk factors associated with antimicrobial resistance. The finding of a low but definite level of resistance to both ampicillin and gentamicin is important for the selection of empiric therapy for sepsis in neonates. The role of inexpensive first-line agents in the outpatient treatment and prevention of urinary tract infections requires re-examination, particularly in children who have recently received antimicrobial therapy.

There is growing concern regarding the changing patterns of antimicrobial resistance among bacterial pathogens. Resistant organisms have emerged worldwide owing to several factors related to the genetic nature of the organisms and selective antimicrobial pressure in humans and animals.[1] Humans play the role of vectors in the spread of resistant organisms across international borders. Currently, human and animal reservoirs of resistant bacteria exist, with the potential

for widespread dissemination of resistant organisms.[2,3] Selective antimicrobial pressure has played a role in the emergence of specific patterns of resistance.[4,5] In some regions of the world, organisms have become resistant to many agents frequently used in the outpatient setting,[6–9] which may limit the usefulness of relatively inexpensive agents. The possibility of the emergence of resistant organisms in children is of concern because there are fewer treatment options available to them compared with adults, given the unavailability of quinolones for routine use in children.[10]

There are concerns about the frequency of antimicrobial use and the growing prevalence of colonization and infection by resistant organisms among groups in crowded environments, such as child care facilities and hospitals.[9] Attendance at child care facilities and the previous use of antimicrobials have been found to be risk factors for otitis media caused by penicillin-resistant pneumococci,[11] and antibiotic use within the previous month has been reported to be associated with bacteremia due to this resistant organism.[12]

Data are limited on the prevalence, specific patterns of resistance and risk factors associated with resistant coliforms isolated from children. Besides the potential implications for the therapy and outcome of coliform-related infections, such information would likely be useful in reinforcing efforts to educate physicians on the appropriate use of commonly prescribed antimicrobials. Consequently, we performed a study to determine the prevalence of resistance to "first-line" antimicrobial agents among urinary isolates of *Escherichia coli* in a pediatric population in both inpatient and outpatient settings, and to determine the factors associated with antimicrobial resistance. First-line agents refer to orally administered antimicrobials normally prescribed as initial therapy or prophylaxis for ambulatory infections.

Methods

The study was conducted at the Children's Hospital of Eastern Ontario, Ottawa. This is a 158-bed regional tertiary care pediatric centre serving the population of eastern Ontario and western Quebec. The study period was from December 1992 to December 1994.

The study design consisted of a descriptive component involving all urinary tract *E. coli* isolates obtained during the study period and a case–control study. Case subjects consisted of all children who had isolates that were resistant to trimethoprim–sulfamethoxazole (T-S). Control subjects were selected from children who had T-S-sensitive isolates obtained during the study period or the preceding or subsequent 6 months. Laboratory procedures were the same for case and control subjects. There was 1 control subject per case subject. Each subject represented 1 isolate (i.e., there were no duplicates in the case–control study). In selecting control subjects we chose the first child from among a sequential list of those with sensitive isolates who matched the case subject based on the period when the isolates were obtained. Matching was done before the subjects' medical records were reviewed by a research nurse, who was not previously involved in the care of patients at the hospital.

Urine samples were collected and transported to the hospital's Division of Bacteriology in sterile containers. All consecutive urinary tract isolates of *E. coli* were identified and subjected to susceptibility testing with the Vitek automated system (BioMerieux Vitek, Inc., Hazelwood, Mo.). The breakpoints (in micrograms per millilitre) were based on the National Committee for Clinical Laboratory Standards guidelines[13] and were as follows: ampicillin 2–8; gentamicin ≤ 0.5–1; T-S ≤ 10; ticarcillin ≤ 16; cefazolin ≤ 8; cefotaxime ≤ 8; nitrofurantoin ≤ 32; and norfloxacin ≤ 4. The standard operation of the Vitek system involves programming to detect the presence or absence or bacterial growth, thus enabling susceptibility testing to be performed on all isolates regardless of colony count. The Kirby–Bauer disc diffusion method[14] was used for comparison on a subset of samples.

In univariate analyses, we analysed proportions using the α^2 test or Fisher's exact test where appropriate. Continuous variables were compared by means of Student's *t*-test if the data were distributed normally and a nonparametric procedure (Kruskal–Wallis test) if the distribution was skewed. Multiple logistic regression was performed, with variables that were significant at a *p* value of 0.25 being examined in the multivariate model. The variables that remained in the final model were those that achieved a *p* value of 0.05 or less.

Results

During the study period 1636 consecutive urinary tract isolates of *E. coli* from 967 different subjects were examined for susceptibility patterns. Testing of a randomly selected subset of 55 isolates with both the Vitek system and the Kirby–Bauer method indicated excellent agreement (98%) between the 2 methods (kappa value 0.94).[15]

Table 1 shows the prevalence of resistance to selected antimicrobials. Similar values were obtained when all resistant isolates were expressed as a proportion of the total number of isolates versus when the prevalence of at least 1

Table 1: Prevalence of urinary tract *Escherichia coli* isolates resistant to antimicrobials among children seen between December 1992 and December 1994

	No. (and % of cases)	
Antimicrobial*	All resistant isolates as a proportion of total isolates *n = 1636*	At least 1 resistant isolate as a proportion of total subjects† *n = 967*
Ampicillin	736 (45.0)	433 (44.8)
T-S	514 (31.4)	274 (28.3)
Ampicillin and T-S	363 (22.2)	195 (20.2)
Gentamicin	47 (2.9)	28 (2.9)
Ampicillin and gentamicin	27 (1.7)	12 (1.2)
Cefazolin	41 (2.5)	34 (3.5)
Cefotaxime	2 (0.1)	2 (0.2)
Nitrofurantoin	30 (1.8)	17 (1.8)
Norfloxacin	11 (0.7)	2 (0.2)
Ticarcillan	650 (39.7)	385 (39.8)

Note: T-S = trimethoprim–sulfamethoxazole.
*Categories are not mutually exclusive.
†Duplicate isolates resistant to T-S were excluded.

resistant isolate was examined after duplicates were removed. When all resistant isolates were examined as a proportion of the total number of isolates, 736 isolates (45.0%) were resistant to ampicillin, 514 (31.4%) to T-S and 363 (22.2%) to both ampicillin and T-S. Low rates of resistance were found for both ampicillin and gentamicin, and for nitrofurantoin, norfloxacin and cefotaxime.

We identified 274 children with *E. coli* isolates resistant to T-S and matched them with 274 control subjects. The 2 groups were comparable with respect to the female:male sex ratios (2.4:1 and 2.3:1 for the case subjects and control subjects respectively).

The mean age (and standard deviation [SD]) of the case subjects was 7.2 (4.7) years and of the control subjects, 5.8 (5.4) years ($p < 0.001$). Children under 2 years of age were less likely than those aged 2–6 years to have resistant isolates (odds ratio [OR] 0.3, 95% confidence interval [CI] 0.2-0.4, $p < 0.001$) (Table 2). The difference between children aged 2–6 years and those over 6 years of age was not statistically significant.

Children who had received antimicrobial treatment for more than 4 weeks in the preceding 6 months were more likely to have resistant isolates than were children who had not received prolonged antibiotic treatment (OR 13.9, 95% CI 8.2–23.5, $p < 0.001$) (Table 2). Patients with genitourinary tract abnormalities were almost 4 times more likely to have resistant isolates than children without such abnormalities (OR 3.9, 95% CI 2.7–5.7, $p < 0.001$). Univariate analyses also indicated that several variables potentially associated with prolonged antimicrobial use, including spina bifida, genitourinary reflux, the presence of a malignant disorder (nominally significant) and antibiotic prophylaxis for immunodeficiency, were associated with T-S resistance (Table 2).

The mean number (and SD) of hospital admissions in the previous year among the case subjects was 0.96 (2.4), compared with 0.30 (0.8) among the control subjects ($p < 0.001$). Children with 1 hospital admission in the previous year were twice as likely to have a resistant isolate as children who had no hospital admissions in the previous year (OR 2.0, 95% CI 1.2–3.1). Children with 2 or more admissions were 4 times more likely to have a resistant isolate than those who had no admissions in the previous year (OR 4.1, 95% CI 2.2–7.4). There was no significant difference between the case and control subjects in the proportion of children with pyelonephritis.

The relation between T-S resistance and resistance to other antimicrobials was also examined by univariate analyses (Table 3). These analyses indicated that isolates resistant to ampicillin were more likely to be resistant to T-S (OR 8.2, 95% CI 5.5–12.2, $p < 0.001$). Isolates that were resistant to gentamicin were also more likely to be resistant to T-S (OR 14.7, 95% CI 2.0–301.9, $p < 0.001$). Similar findings were also found for isolates resistant to ticarcillin and cefazolin.

The independent effects of several variables from Table 2 were determined with multiple logistic regression (Table 4). More than 4 weeks of antimicrobial therapy or prophylaxis in the preceding 6 months remained significantly associated with resistance (OR 23.4, 95% CI 12.0–47.6, $p < 0.001$), as did genitourinary tract abnormalities (OR 2.4, 95% CI 1.2–4.5, $p = 0.01$). Children with 2 or more hospi-

Table 2: Characteristics of children with *E coli* isolates resistant to T-S compared with control subjects in univariate analyses

Variable	No. of children: Case subjects *n = 274*	No. of children: Control subjects *n = 274*	OR (and 95% CI)
Female sex	191	192	1.0 (0.7–1.4)
Age, yr*			
< 2	42	104	0.3 (0.2–0.4)
2–6 (reference)	81	50	1.0
> 6	150	120	0.8 (0.5–1.2)
Inpatient	53	85	0.5 (0.4–0.8)
Genitourinary tract abnormality	216	137	3.9 (2.7–5.7)
Spina bifida	90	37	3.1 (2.0–4.9)
Genitourinary reflux	61	42	1.7 (1.1–2.6)
Malignant disorder	9	2	4.7 (1.0–21.8)
Pyelonephiritis	18	25	0.7 (0.4–1.4)
Antibiotic therapy for > 4 wk in past 6 mo	136	19	13.9 (8.2–23.5)
Any antibiotic prophylaxis	138	21	12.9 (7.8–21.4)
Prophylaxis for urinary tract infection	101	17	24.1 (13.4–41.2)
Prophylaxis for immunodeficiency	9	1	15.5 (1.9–123.3)
No. of hospital admissions in past yr†			
0 (reference)	165	218	1.0
1	53	36	2.0 (1.2–3.1)
≥ 2	49	16	4.1 (2.2–7.4)

Note: OR = odds ratio, CI = confidence interval.
*One missing value.
†Eleven missing values.

Table 3: Univariate anaylses of the likelihood that *E. coli* isolates resistant to T-S also showed resistance to selected antimicrobials

Antimicrobial	No. of children: Case subjects	No. of children: Control subjects*	OR (and 95% CI)
Ampicillin	203	71	8.2 (5.5–12.2)
Cefazolin	14	1	14.7 (2.0–301.9)
Cefotaxime	1	0	3.0 (0.1–999.9)
Gentamicin	14	1	14.7 (2.0–301.9)
Nitrofurantoin	8	3	2.7 (0.7–13.1)
Norfloxacin	4	0	9.1 (0.7–999.9)
Ticarcillin	194	68	7.4 (4.6–10.9)

*For cells containing 0, an adjustment was made, with 0.5 being added to each cell to compute the odds ratios.

tal admissions in the previous year were 3.2 times more likely to have resistant isolates than children with no admissions in the previous year (95% CI 1.1–4.8, $p = 0.03$), and children with 1 admission in the previous year were also more likely to have a resistant isolate than those with no admissions (OR 2.3, 95% CI 1.4–7.5, $p = 0.008$). Children under 2 years of age were less likely to have a resistant isolate than those aged 2–6 years (OR 0.3, 95% CI 0.2–0.8, $p = 0.001$). Those over 6 years of age were also less likely to have a resistant isolate than those aged 2–6 years (nominally significant).

We added ampicillin susceptibility to the full logistic regression model. This model showed that children with ampicillin-susceptible isolates were less likely than children with ampicillin-resistant isolates to have isolates resistant to T-S (OR 0.1, 95% CI 0.05–0.2, $p < 0.001$). The results were similar to that of the model as summarized in Table 4, with the exception that age was not significant when ampicillin susceptibility was included in the model.

Interpretation

Our results indicate that a high proportion of genitourinary *E. coli* isolates from children were resistant to the first-line agents ampicillin and T-S. Categories of patients found to be at increased risk for having isolates resistant to T-S included those receiving antimicrobial prophylaxis because of genitourinary abnormalities, malignant disorders and immunodeficiency. Given the paucity of Canadian data on antibiotic resistance in children, our findings contribute to the background information needed to devise strategies to reduce antibiotic resistance. In this context, the results have defined a target group for control, namely, children receiving antibiotic prophylaxis.

The Vitek system enabled us to obtain susceptibility data for a cross-section of children with and without urinary tract infection, as our goal was to evaluate all *E. coli* isolates, not merely those present in significant numbers consistent with a diagnosis of urinary tract infection. An alternative strategy would have been to study fecal samples from a cross-section of children.

T-S was used as a representative first-line agent. It should be noted that resistance to T-S was associated with resistance to other, pharmacologically unrelated agents commonly used to treat ambulatory infections, including ampicillin, orally administered first-generation cephalosporins (e.g., cephalexin) that are equivalent in activity to cefazolin, and norfloxacin. Thus, the main risk factors associated with T-S resistance would likely be associated with resistance to other agents, such as ampicillin. In the future, one approach that may assist physicians in determining the probability of resistance for each patient is the creation of a matrix in which various risk factors are combined to create particular patient profiles.

Table 4: Multivariate modelling of factors associated with T-S resistance

Variable	OR (and 95% CI)
Antibiotic therapy for > 4 wk in past 6 mo	23.4 (12.0–47.6)
Genitourinary tract abnormality	2.4 (1.2–4.5)
No. of hospital admissions in past yr	
0 (reference)	1.0
1	2.3 (1.4–7.5)
≥ 2	3.2 (1.1–4.8)
Age, yr	
< 2	0.3 (0.2–0.8)
2–6 (reference)	1.0
> 6	0.5 (0.2–1.0)

Owing to concerns about the use of quinolones in children,[10] there are fewer agents for oral use available to treat outpatient urinary tract infections in children than in adults. Breakthrough bacteriuria due to resistant organisms may limit the usefulness of first-line agents such as T-S,[16] one of the agents most frequently used to prevent recurrent urinary tract infections.[17] Combination antimicrobial prophylaxis in a subpopulation of girls with breakthrough urinary tract infections has been suggested.[18] Our results support the inclusion of nitrofurantoin in such combinations.[18]

We found that children admitted to hospital 1 or more times in the previous year were more likely to have resistant isolates than those with no admissions in the previous year. This finding reinforces the importance of strategies to limit the spread of resistant organisms within closed settings, such as hospitals. The finding of isolates showing resistance to both ampicillin and gentamicin is of concern with respect to the potential for spread to pregnant women and hence to neonates. The combination of ampicillin and gentamicin is commonly used to treat nonmeningeal bacterial infections in neonates.

Compared with children aged 2–6 years, children under 2 years of age in our study were less likely to have isolates resistant to T-S. Those over 6 years of age were also less likely to have resistant isolates (nominally significant). Although many children at child care facilities are 2–6 years of age, we did not evaluate attendance at child care facilities in enough detail to enable meaningful analyses. However, studies have shown that many children attending such facilities carry resistant strains of *E. coli*[19] and have high rates of respiratory infections, including otitis media,[20] the latter providing the rationale used by some physicians to prescribe multiple and prolonged courses of antibiotics to this group of children.

We found that, in the population studied, prolonged selective antimicrobial pressure is associated with resistance. The results suggest that the role of inexpensive first-line antimicrobials in the outpatient treatment of urinary tract infections will have to be re-examined in relation to treatment outcomes, particularly in children who have recently received antimicrobial therapy. The clinical outcome of children with urinary tract infections due to resistant *E. coli* requires further study.

We limited the number of variables used to match case subjects with control subjects so that we could compare as

many important potential risk factors as possible. In this context, we felt that it was most important to match the period when the samples were collected, insofar as there may be differences in circulating strains of *E. coli* or differences in laboratory procedures over time. In any event, the laboratory procedures used were uniform throughout the study period. Although molecular typing was not performed, the antibiotic resistance patterns observed during the study period did not suggest the presence of a specific strain of *E. coli* confined to one particular period but not another.

The implications of resistance to first-line agents are more far reaching than any immediate difficulty in treating urinary tract infections due to resistant *E. coli*. Given the similarity in the patient populations and clinical practices across Canadian tertiary care pediatric centres, it is likely that results similar to ours would be found in other centres. Carefully planned active surveillance systems are needed to monitor antimicrobial resistance in pediatric centres in Canada. Such systems should ideally be designed to enable the linking of clinical and epidemiologic risk factors with emerging laboratory data.

We recommend that children in the risk groups identified by our study have their antimicrobial regimens periodically reassessed in relation to the proposed surveillance data. In cases in which prophylaxis for urinary tract infection is felt to be necessary, it would be prudent to consider implementing a strategy of alternating drugs, with particular agents being excluded for prolonged periods. This strategy of avoiding the use of specific antimicrobials has been shown to be beneficial in restoring susceptibility rates among certain organisms in other groups of patients.[21,22]

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