

Correspondence

An objective measure of antibiotic use for febrile illness in a rural paediatric population using high performance liquid chromatography

Sir,

High variability in antibiotic use has been observed worldwide, some of which highlights inappropriate prescribing behavior^{1,2} and could account for the emerging antimicrobial resistance³. Widespread antibiotic use for minor ailments⁴ as well as harmful side-effects and additional financial burdens due to irrational antibiotic prescribing have been reported⁵. In spite of respective guidelines for antibiotic use available in different hospitals in India, there exists an appreciable gap in the attitudes and prescribing patterns of both prescribers and dispensers at the various levels of health care⁶. Children with acute febrile illness are frequently seen in secondary care hospitals after receiving prior treatment at home, in a primary health centre, a local clinic or a private hospital. An important consequence of prior antimicrobial medication is the masking of meningitis cultures which results in underestimating meningitis even in the presence of raised cell counts or an inability to identify the causative organism.

This study investigated a paediatric population (<2 yr of age) who presented to the secondary care centre within two to four days of onset of fever. Earlier use of antibiotics was qualitatively assessed by High Performance Liquid Chromatography (HPLC) urine analysis at the first visit. HPLC is an objective, specific and sensitive method for the identification of antibiotics in urine⁷. These results were also compared to answers given by the parent(s) in response to a questionnaire regarding medication given prior to presenting at the centre.

This was a bi-centre study conducted at the Community Health and Development Hospital, Vellore, Tamil Nadu and Post Graduate Institute of Medical Education and Research, Chandigarh.

Institutional Review Board clearance was obtained in both centres. From January to March 2007 a total of 64 rural children were recruited from those attending the out patient facility in each secondary health care study centre. Recruitment was based on age (less than 2 yr), a history of acute febrile illness (greater than 2 days duration but less than 4 days), seen for the first time in the study hospital(s) and with signed informed consent from the parents. The most responsible member accompanying the child, verbally answered a questionnaire conducted by the study physician. The questionnaire included demographics, symptoms at the time of presentation and information about earlier medication for the same episode. Details on how the medicines were obtained were also noted. Urine was collected from the child, transported on ice and stored at -20°C until analysis. The specimens were assayed for trimethoprim /sulphamethoxazole, amoxycillin, ciprofloxacin, cephalixin and cefuroxime in the Clinical Pharmacology Unit, Christian Medical College, Vellore. The antibiotics of interest have half-lives ranging from 1-13 h⁸. The antibiotic with the longest half life will be cleared from the body within four days after it is discontinued. Therefore even if the child had completed a full course of any one of these antibiotics two days prior to the onset of this febrile illness, there would be no interference from that antibiotic treatment in the urine specimen. All specimens were assayed by isocratic HPLC with ultraviolet detection and column temperature maintained at 30°C. All assays used a Discovery (Supelco[®], USA) C18, 5 micron, (250 x 4.6 mm) column. Drug extraction from urine was performed using solid phase extraction (SPE) cartridges (Strata[®], USA). All assays were developed such that there was no interference from commonly prescribed drugs. Stability of the antibiotics in urine showed that amoxycillin, trimethoprim/sulphamethoxazole, cephalixin and cefuroxime were stable at -20°C up to three weeks and

ciprofloxacin for 1 wk. The mobile phases were: for amoxicillin - acetonitrile (ACN) and phosphate buffer (PB) (100 mM, pH 3.0) (1:4), ciprofloxacin - ACN and PB (14.5 mM, pH 6.9) (1:3.55), trimethoprim/sulphamethoxazole - methanol and PB (14.5 mM, pH 3.0) (1:3.55), cephalexin and cefuroxime - methanol and PB (100 mM, pH 7.0) (1:4). Detection was at 227 nm for amoxicillin and cephalosporins and 215 nm for ciprofloxacin and trimethoprim /sulphamethoxazole.

Sixty four children (30 from Vellore, 34 from Chandigarh) with presenting complaints of fever alone (51), cough (25) and a combination of symptoms which included fever, cough, cold and other minor symptoms (21) were recruited into the study. Twenty (31%) of the 64 children, 10 each from Vellore and Chandigarh were shown by HPLC to have antibiotic in the urine (95% CI: 19.7, 42.3). Other medications (from questionnaire) included paracetamol (19), cough syrup (2), anti-asthmatics (3) and promethazine (1). Fifty-eight parents (90%) answered that prior treatment was given for the same episode of fever (95% CI: 82.7, 97.4). Five parents said they had not given any medicine for this episode and one did not answer. The Table shows how the medicines were obtained by the 58 parents who had said their child had received prior treatment and by the 20 children who were positive in the urine analysis. The antibiotic most commonly detected by HPLC was amoxicillin (11 patients) followed by trimethoprim/sulphamethoxazole (9 patients) and then cephalexin and cefuroxime (4 patients each). Ciprofloxacin was not detected in either of the centers. From which seven urine specimens were positive for a combination of antibiotics, many of which included amoxicillin.

This study raises many concerns; firstly, these results are a conservative estimate since not all antibiotics were tested for by HPLC. Therefore we

can expect that more than one third of children with a febrile illness will have had prior treatment with antibiotics. This has serious implications for diagnoses or treatments relying upon microbiological cultures. Secondly, the detection of multiple antibiotics in the urine may be due to parents visiting multiple centres for the treatment of their child or buying antibiotics over the counter. This raises questions regarding the thoroughness of enquiry and education of the parent by those prescribing or dispensing the drugs. Thirdly, only half of the children whose urine proved positive for antibiotic, were given medication obtained with a prescription and a high proportion of parents gave medicines that were left over and in most cases were unaware of its content. It must be noted that many, if not most of the families, have an extremely low daily income and low literacy level. Some limitations of this study are that false negatives could exist for the following reasons: a) A very small peak occurring in the HPLC chromatogram at the retention time of the drug may have been considered insufficient to be classified as positive. b) Three antimicrobials quoted by the parents were not assayed for. These were ofloxacin, furazolidone/metronidazole and cefpodoxime.

This study highlights the importance of continuing medical education on rational antibiotic prescribing for medical personnel employed in local clinics. Pharmacies should also be discouraged in selling antibiotics over the counter without a prescription from a qualified medical person. However, both of these are long term issues that require the co-operation of those involved. As an immediate response continual education by community health teams, especially to new parents, regarding the use and abuse of antibiotics can be imparted.

It would be interesting to expand the urine screen to incorporate more antibiotics especially cephalosporins

Table. Parents response as to how the medicines were obtained

How the medication was obtained	Prescription only (A)	Pharmacy with no prescription (B)	Left over from a previous illness (C)	A + B	A + C	B + C	Not answered
Number (58)	20	5	21	2	6	1	3
Percentage (95% CI) in %	34.5 (22.3, 46.7)	8.6 (1.4, 15.8)	36.2 (23.8, 48.6)	3.4 (0, 8.1)	10.3 (2.5, 18.1)	1.7 (0, 5.0)	5.2 (0, 10.9)
Percentage with a positive urine analysis (95% CI) in %	35 (14.1, 55.9)	10 (0, 23.2)	30 (9.9, 50.1)	5 (0, 14.6)	10 (0, 23.2)	nil	5 (0, 14.6)

* 5% of positive urine analysis were from children whose parents responded that medication was not given

and study resistance patterns in the respective communities at more rural centres in India.

Conflict of interest: Nil.

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