Anaerobic infections of the urinary tract: are they being missed?

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Abstract

Routine anaerobic culture of urine identified the urinary tract as the primary focus of sepsis in a postoperative patient with *Bacteroides fragilis* septicaemia. Specimens of urine from six other symptomatic patients grew $> 10^8$ cfu/litre of a *Bacteroides* species in pure growth. The significance of these isolates is discussed. Multipoint technology and the availability of anaerobic work stations have facilitated anaerobic culture and reduced its cost. The incorporation of anaerobic culture of urine into routine laboratory practice may be clinically valuable and should be considered.

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Standard procedural guidelines for the diagnosis of urinary tract infections recommend primary culture on a single medium such as cystine-lactose-electrolyte deficient (CLED) agar.^{1,2} Although there are reports of infection with fastidious organisms,³⁻⁵ procedures which would identify such infections are rarely incorporated into routine practice, either because the significance of these isolates remains in doubt or because of the additional cost. The use of multipoint technology enabled us to increase the range of primary inoculation media at little extra cost. When culture on blood agar incubated in an anaerobic atmosphere was included to facilitate the identification of haemolytic streptococci, anaerobic infection was identified more often than anticipated. We present a case history and report on the anaerobic isolates obtained from midstream urine specimens over a six month period.

Laboratory methods

Specimens of urine from adults were collected into 25 ml capacity sterile universal containers containing 1g boric acid preservative (Ross Laboratories, Macclesfield, Cheshire, UK). Uncentrifuged urine was examined using an inverted microscope (Rant and Shepherd, PHLS, Norwich, UK) and a 1 µl loopful cultured on CLED agar, incubated for 18 hours in air at 37°C. Specimens showing > 100 leucocytes/µl on microscopy were selected for further testing using a 19 pin multipoint inoculator (MAST Diagnostics, Bootle, Merseyside, UK). The media inoculated included a set of biochemical substrates for isolate identification, antibiotic containing media for breakpoint susceptibility tests, an *Escherichia coli*

seeded plate for the detection of antimicrobial substances, heated blood ("chocolate") agar incubated for 18 hours in 5% carbon dioxide at 37°C, and 5% horse blood in Columbia agar incubated in an anaerobic work station (WISE, Don Whitley, Shipley, W Yorkshire, UK). All media were supplied by MAST Laboratories and prepared in accordance with their instructions. Anaerobic isolates from urine were reported when $> 10^8$ colony forming units (cfu)/litre were obtained in pure growth.

Blood cultures were processed using a BacT-Alert microbial detection system (Organon-Teknika, Cambridge, UK). The recommended sample was 20 ml blood divided equally between two bottles containing enriched tryptone soya broth, one of which was vented. When microbial growth was detected a sample from the bottle was subcultured onto heated blood agar and 5% horse blood in Columbia agar base incubated as described above. Anaerobic isolates were identified using API 21A (bioMerieux UK, Basingstoke, UK). When indicated, confirmation of identity and further typing of isolates were carried out at the PHLS Anaerobic Reference Laboratory, Cardiff.

Case report

A 29 year old woman had a vaginal hysterectomy for grade 3 cervical intraepithelial neoplasia extending to the resection line of a previous cone biopsy. She gave no history of symptoms related to the urinary tract. She was a heavy smoker (20 cigarettes a day). The operative procedure was uneventful. Prophylactic antibiotics were not given. A Foley urinary catheter was introduced postoperatively and retained for 48 hours.

On the first postoperative day, the patient developed a productive cough and was pyrexial at 38.5°C. On clinical examination, bronchospasm was noted on the left side. There was no evidence of infection at any other site. She was treated for a presumed respiratory tract infection with oral amoxycillin but remained unwell, with pyrexia increasing to 39.6°C. On the third postoperative day blood cultures were taken and specimens of sputum and midstream urine sent for laboratory examination. Antibiotic treatment was changed to parenteral cefuroxime and metronidazole per rectum. Parenteral gentamicin was begun when blood cultures were reported as showing Gram negative rods. These were identified as *Bacteroides fragilis*. The specimen of midstream urine was described as smelling offensive, showed pyuria (over 100 leucocytes/µl) and

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Table 1 Midstream urine specimens with $>10^8$ cfu/litre *Bacteroides* spp: clinical details and results of microscopy

Patient	Age (years)	Sex	Leucocytes/ μ l	Symptoms	Associated factors
1	67	F	>2000	Dysuria, backache	Bladder tumour
2	46	F	>2000	Frequency	Bladder tumour
3	30	F	>2000	Dysuria	Pregnancy
4	62	F	>100	Frequency	None
5	21	F	>2000	Dysuria	None
6	86	F	>2000	Not known	Not known

yielded a heavy growth ($> 10^{10}$ cfu/litre) of *Bacteroides fragilis*. Further phenotypic tests indicated that the isolates from blood and urine were indistinguishable. The patient continued on antibiotics for a further three days and was discharged well on the ninth postoperative day.

AUDIT OF ANAEROBIC CULTURE OF URINE

Retrospective review of midstream urine culture results over a six month period showed six of 19 429 yielded $> 10^8$ cfu/litre (usually $> 10^{10}$ cfu/litre) of a *Bacteroides* species in pure growth. All patients were female and had significant pyuria (table 1). Clinical details were unavailable in one patient (patient 6), who died in renal failure six months later. All the other patients were symptomatic. In two patients malignant bladder tumours were subsequently identified. Repeat specimens were not received from any of these patients.

Not infrequently, the anaerobically incubated horse blood agar and "chocolate" agar plates showed a significant growth of an isolate which did not appear on the CLED plates but which might have been expected to do so. These isolates, which included coliforms and *Streptococcus pyogenes*, are not reviewed here.

Discussion

Anaerobes colonise the distal urethra and vagina so could easily contaminate a midstream specimen of urine.⁵ Since all seven

patients described in this report were female, and since bladder specimens were not obtained, it might be argued that the anaerobic isolates obtained were urethral or vaginal contaminants. Criteria for significance included symptoms related to the urinary tract, pyuria, isolation of the organism in pure growth and in one instance concurrent bacteraemia. These are criteria applied when assessing the significance of the common urinary pathogens and so may reasonably be applied to anaerobic isolates. The isolation of anaerobes in numbers exceeding 10^{10} cfu/litre is also suggestive of multiplication in the bladder rather than of contamination.

Anaerobes are sensitive to oxygen tension in the bladder, but infection may become established in association with tumours or tissue damage and following instrumentation. Anaerobic infections are, however, rarely reported. This is presumably because anaerobic culture of urine is not undertaken routinely—primary culture for anaerobes is not considered cost-effective. The use of multipoint technology and the increased capacity of anaerobic cabinets compared with anaerobic jars have facilitated anaerobic culture and reduced its cost. Incorporation of anaerobic culture of urine into routine laboratory practice should therefore be considered. Anaerobic infection may account for otherwise unexplained symptoms related to the urinary tract. Repeat cultures and exclusion of vaginal infection are indicated in the first instance and the possibility of an associated tumour should be considered if infection is confirmed.

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