

## URINARY WHITE CELLS AND THE SYMPTOMS OF THE OVERACTIVE BLADDER

### Hypothesis / aims of study

A surprising observation, nevertheless true, is that the literature does not contain reports of rigorous tests of whether urinary infection, or inflammation of the bladder, plays any role in the overactive bladder (OAB).

The conventional method for diagnosing urine infection is to despatch a midstream urine sample for culture. The result is usually judged positive if  $\geq 10^5$  ml<sup>-1</sup> colony forming units (cfu) of a known urinary pathogen are isolated. This is the criterion of Kass who in 1957<sup>1</sup> was reporting on normal, asymptomatic women. Recognising the absence of symptoms in the Kass sample, Stamm and Hooton<sup>2</sup>, having studied women with symptomatic, cystitis with dysuria showed that the diagnostic threshold in this situation should be  $\geq 10^2$  cfu ml<sup>-1</sup>. Regrettably, the automated laboratory methods used today at best achieve a threshold of  $\geq 10^4$  cfu ml<sup>-1</sup>. Thus, several studies have shown that midstream urine (MSU) culture misses 50% of all genuine, symptomatic urine infections. It could be argued that overactive bladder symptoms have much in common with those from cystitis and that we should not be reassured by the conventional MSU. The dipstick tests, which uses chemical indicators to identify leucocyte esterase and nitrite have been calibrated to the threshold of if  $\geq 10^5$  cfu ml<sup>-1</sup>, so their sensitivity is in doubt. In 1965 the threshold criterion of  $\geq 10$  white cells (wbc) per  $\mu$ l, of fresh, unspun, urine for diagnosing significant urine infection was described after an impressive experimental series. This threshold has been validated in many subsequent studies. The microscopy of a fresh, unspun specimen of urine, using a haemocytometer chamber in the clinic setting is the correct method for detecting significant pyuria ( $\geq 10$  wbc  $\mu$ l<sup>-1</sup>) is a little laborious and requires skill, so it is not commonly effected. Nevertheless, given the background of evidence, the OAB narrative will remain incomplete until this experiment be achieved. This matter has been given greater urgency in the wake of the discovery of the P2X<sub>2</sub> and P2X<sub>3</sub> receptors in bladder urethelium. These ionic channels are opened by ATP binding. ATP release is not a dominant aspect of the normal human urethelium. However, an inflammatory reaction involving the urethelium would result is very significant ATP release by the cells. This study deployed microscopy of fresh, unspun urine specimens, using a haemocytometer chamber, to assess the prevalence of pyuria  $\geq 10$  wbc  $\mu$ l<sup>-1</sup> in patients presenting with overactive bladder. Using a blinded design the results were compared with MSU culture results.

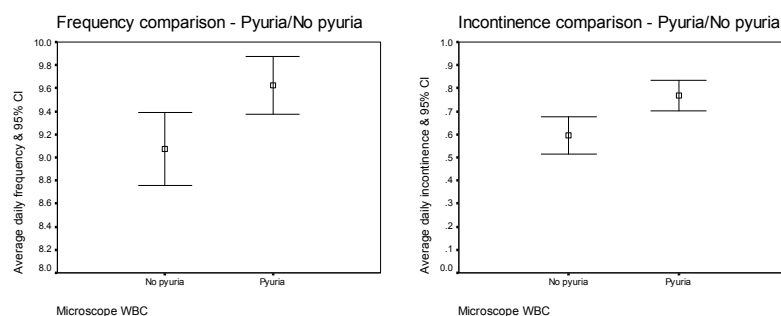
### Study design, materials and methods

The study was approved by the ethical committee.

Patients presenting with overactive bladder provided a fresh urine sample in the clinic by the MSU method. The sample was examined immediately by dipstick and by microscopy for white cells using a haemocytometer. An aliquot was sent for laboratory culture. The microscopist and the microbiologist were blind to each others' results. The patients' symptoms were recorded and they were treated for an overactive bladder using an antimuscarinic agent and bladder retraining. Patients, who were found to have significant pyuria, were treated with antibiotics, without reference to the culture, in-line with probabilistic advice available in the literature. The patients were followed up and at each subsequent visit they were reassessed using the identical methods. Thus data were collected at presentation and following treatment. For the purposes of this experiment the microscopy result for pyuria was used as the reference gold standard. The data were examined by means of frequency tables. Between groups differences were assessed parametrically using paired and independent sample t-tests.

### Results

The study was conducted in an incontinence clinic over a four year period April 2003 to March 2007. 785 patients were studied, mean age=54 (sd=19) 719 females and 68 males. At presentation 452 patients (58%) showed significant pyuria. However during the observation period, 579 (74%) demonstrated significant pyuria at some time ( $\geq 10$  wbc  $\mu$ l<sup>-1</sup>). Only 93 (21%) of the 452 patients presenting with pyuria produced a significant urine culture at  $\geq 10^5$  cfu ml<sup>-1</sup>. Provided that leucocyte esterase test results of  $\geq$ Trace were taken as positive, 371 of pyuria patients (82%) showed a positive dipstick test. The nitrite test was positive in only 36 (8%) of the patients with significant pyuria. The comparison of average 24-hour urinary frequency (95% CI diff; 0.96, 0.15, p=0.011) and incontinence (95% CI diff; 0.28, 0.67, p<0.001) between those with and without pyuria at presentation showed significant differences; see figures. Paired analysis in patients with pyuria and following resolution showed similarly significant differences



### Interpretation of results

These data demonstrate significant pyuria ( $\geq 10$  wbc  $\mu$ l<sup>-1</sup>) detected in between 58% and 74% of patients with overactive bladder symptoms. Whilst a probable cause for this could be chronic urine infection this is unproven and the

MSU cultures, albeit imperfect were negative in nearly 80%. The dipstick leucocytes erase results add credence to the results of microscopy method, nowadays uncommonly used in clinical practice. The results of the Nitrite tests indicate a low level of urea-splitting organism activity in these samples. The differences in symptoms between those with or without pyuria support the proposition that this hitherto unreported pyuria in patients with overactive bladder symptoms may be clinically significant

#### Concluding message

These data have identified evidence of an inflammatory reaction in the urine of between 58% and 74% of patients presenting with overactive bladder symptoms. The origins of this response require elucidation, chronic low-grade cystitis is a plausible option. Inflammation in the urethelium should be associated with increase epithelial ATP release, which in turn could be germane to the pathophysiology of the overactive bladder<sup>3</sup>.

#### References

1. *Arch.Intern.Med.* 1957;100:709-1
2. *N.Engl.J.Med.* 1982;307:463-8.
3. *Br.J.Pharmacol.* 2006;147 Suppl 2:S132-S143.

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**HUMAN SUBJECTS:** This study was approved by the Whittington and Moorfields Research Ethics Committee and followed the Declaration of Helsinki Informed consent was obtained from the patients.