

## BACTERIAL COLONIZATION IN UROTHELIUM OF WOMEN WITH OVERACTIVE BLADDER SYNDROME

### Hypothesis / aims of study

Pyuria has been reported in 35 % of women with overactive bladder (OAB) when catheter specimen of urine (CSU) is tested compared to 7 % of asymptomatic controls. Of these women with pyuria 35% were culture positive at  $10^5$ (1). It has also been shown that urothelial biopsies from patients with symptoms of overactive bladder have evidence of chronic inflammation even when the urine was free of pyuria and infection. Therefore there is growing evidence that OAB is associated with cystitis and infection potentially plays an important aetiological or exacerbatory role.

Electron microscopic studies in mouse bladders have shown that *Escherichia coli* have the ability to invade superficial bladder epithelium and mature into biofilms, creating pod like bulges (biopods) on the bladder surface. (2) These biopods contain bacteria in a polysaccharide rich extracellular matrix and are capable of evading host defences creating a chronic quiescent reservoir in the bladder and serve as a source for recurrent cystitis.

Biofilms have been shown to stain intensely with periodic acid –Schiff (PAS) stain because of the polysaccharide rich extracellular matrix surrounding the bacteria in corneal biopsies.(3)

Therefore our aim was to study the bladder biopsies of women with intractable overactive bladder symptoms with PAS and examine for the presence of biofilms and rule out infection as an etiological factor.

### Study design, materials and methods

Women with overactive bladder symptoms who did not respond to conservative management of bladder retraining, anti muscarinics and antibiotics were recruited. The patients also completed a three day bladder diary and a quality of life questionnaire (KHQ). The women had previously undergone investigations with urine microscopy, culture and sensitivity which were negative. The women also underwent saline or video urodynamics studies and cystoscopy. The cystoscopy was performed under general anaesthetic with rigid cystoscopes. Biopsies were taken with cold cup biopsy forceps. Superficial bladder biopsy was taken for PAS staining and another biopsy of bladder wall was taken for routine histopathological examination. The biopsy taken for PAS staining was fixed with 3%glutaraldehyde in cacodylate buffer and then stained with PAS stain and processed. Informed consent was obtained from these patients

### Results

Fifteen women were recruited and the urodynamic diagnoses are shown below in Table 1.

| Number of symptomatic patients | Urodynamic diagnosis                          |
|--------------------------------|---|
| 6                              | Detrusor overactivity                         |
| 2                              | Urodynamic stress incontinence                |
| 5                              | Bladder pain                                  |
| 1                              | Normal  |
| 1*                             | *urodynamics not done but had symptoms of OAB |

Table 1: Urodynamic diagnoses of the women in the study

None of the women were found to have positive urine cultures at the time of the cystoscopy.

The routine histopathology revealed mild chronic cystitis in 10 patients and moderate chronic cystitis in 5 patients of the symptomatic group and also evidence of mild chronic cystitis in one asymptomatic patient using the standardised technique previously described (4).

PAS staining revealed presence of biofilms (biopods) in 8 patients (figure 1). PAS staining was negative (no biofilms identified) (figure 2) in remaining 7 symptomatic patients.

In patients who had positive PAS staining there was also evidence of severe trigonitis, erythema and trabeculation on cystoscopy.

Figure 1: PAS Staining of superficial bladder biopsy showing presence of biofilms in umbrella cells of urothelium (magenta spots in the superficial layer)

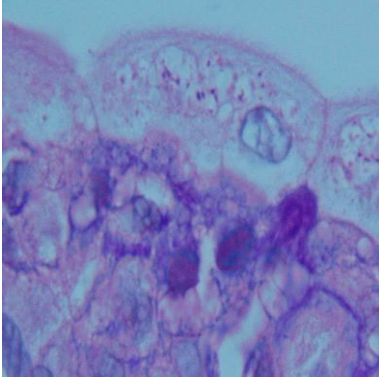
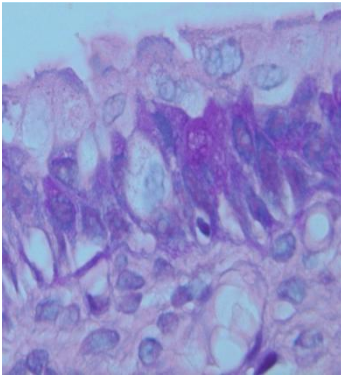


Figure 2: PAS Staining of superficial bladder biopsy showing presence of PAS negative umbrella cells



Interpretation of results

The presence of biopods in urothelium of half of the symptomatic group of patients supports the study on animal models of the presence of a chronic bacterial reservoir in the bladder which possibly contributes to overactive bladder symptoms.. This infection may be an important aetiological or exacerbation factor for overactive bladder

Concluding message

This is the first time biofilms have been shown with PAS staining in the urothelium of human with lower urinary tract symptoms. We believe that these bacteria may contribute to ongoing low grade inflammation in the bladder mucosa which produces or exacerbates overactive bladder symptoms and there is a need to establish the viability of these pathogens by culture and electron microscopy to further investigate this finding

References

1. Neurourol Urodyn 2007; 26(5):656-7
2. Science. 2003 Jul 4; 301(5629):105-7.
3. Ophthalmology. 2001 Jun; 108(6):1088-92.

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|--|---------------------------------|
| <b><i>Specify source of funding or grant</i></b>               | <b>None</b>                     |
| <b><i>Is this a clinical trial?</i></b>                        | <b>No</b>                       |
| <b><i>What were the subjects in the study?</i></b>             | <b>HUMAN</b>                    |
| <b><i>Was this study approved by an ethics committee?</i></b>  | <b>Yes</b>                      |
| <b><i>Specify Name of Ethics Committee</i></b>                 | <b>St. Mary's REC committee</b> |
| <b><i>Was the Declaration of Helsinki followed?</i></b>        | <b>Yes</b>                      |
| <b><i>Was informed consent obtained from the patients?</i></b> | <b>Yes</b>                      |