

Detection of bacteriuria in a well-woman screening unit

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General practitioners commonly encounter painful and frequent micturition amongst their female patients. A diagnosis of urinary tract infection (UTI) is often made, subject to confirmation by urinary bacteriological studies. Unfortunately people who have no other symptom or sign of urinary infection may void urine containing large numbers of bacteria and the precise significance of such an 'asymptomatic bacteriuria' is unknown.

It has been suggested that it may precede the clinical signs of UTI (Turner, 1961) and it has been observed in latent periods of the disease or after incomplete healing of an active infection (Williams *et al.*, 1969).

Since a urine specimen may contain bacteria because of either infection or contamination, Kass (1956) introduced the concept of significant bacteriuria. Human urine is an excellent medium for bacterial growth (Asscher *et al.*, 1966) and so bacteria can multiply rapidly in the bladder urine. Quantitative estimation of the bacteria in the urine, therefore, enables the distinction between contamination and infection to be made. Carefully taken specimens from an uninfected urinary tract will seldom give counts above 10,000 per ml urine (*Drug and Therapeutics Bulletin*, 1971), whereas organisms which have multiplied in the bladder urine before voiding will give counts of greater than 100,000 per ml (a 'significant bacteriuria').

The introduction of simple methods for the collection of urine specimens with minimal contamination and their subsequent examination by quantitative bacteriological procedures, has made it possible, without undue inconvenience or cost, to investigate large groups of people for UTI. The difficulty is that there are no established objective criteria by which the harm caused by UTI can be assessed, and there is considerable controversy over the desirability of such 'screening'.

Since in those situations where it is now known that UTI can be sinister, the hazards have only recently been recognised (Smellie, 1967; Whalley, 1967; Gruneberg *et al.*, 1969), we have preferred to assume that UTI is capable of causing damage, even if we cannot prove it. With this assumption we set out to determine the prevalence of UTI in a group of non-pregnant women, and to examine the effect of incubation temperature on a simple method of screening for bacteriuria.

Method

1. Population

The women tested were those who attended the BUPA women's screening unit in London between 1 March and 1 July, 1972. As this is a private sector clinic, there is selection by social class, all the patients being from classes I, II or III.

During her visit the woman's history was obtained, with particular reference to the genitourinary system. Examination of the breasts and pelvis was performed after a sample of urine had been collected. The history and examination were performed by one of three specially trained nurses, under the supervision of a doctor. During this survey there was no modification of the normal procedures except in the collection of the urine sample.

2. Collection of urine samples

The use of cleansing agents in the collection of midstream specimens of urine has been widely debated, but since even small amounts of antiseptic contaminating the urine may reduce the bacterial count sufficiently to mask infection (Roberts *et al.*, 1967), we adopted the method described by Brumfitt and Reeves (1968), using only sterile water.

Using a 'dip and read' strip test ('Labstix') the nurse tested each sample for pH, protein,

blood, glucose and ketones; all positives being checked in the laboratory. She then effected urine culture using a dip-slide method ('Uricult'). 'Uricult' is a semi-quantitative culture method containing a dip-slide coated on one side with cystine lactose electrolyte deficient medium (CLEM) and on the other with MacConkey agar.

The dip-slide was removed from its tube, dipped into the urine specimen to cover completely the agar surfaces, drained and stood end-on on clean blotting paper to absorb excess urine. The slide was then replaced in its tube, which was tightly closed and labelled with the patient's name and index number. Two such tubes were obtained for each patient and these, with a sterile specimen of urine, were taken to the laboratory on the hour.

3. Laboratory procedures

At the laboratory the 'Uricult' tubes were incubated for 24 hours in a vertical position, one at 37°C and one at room temperature (roughly constant at 22°C). After incubation any growth was compared with the maker's 'colony density model' to determine whether it was significant (>100,000 per ml urine), doubtfully significant (10,000–100,000) or not significant (<10,000). For the sake of convenience these groups will be indicated by the numbers 3, 2, 1 respectively. 'No growth' (represented by the number 0) was generally included with group 1.

All the laboratory work was performed by the same experienced technician (V.H.), and positive results were made known to the patient's family doctor.

Results

1. General characteristics of the sample

There were 828 non-pregnant women admitted to the study; 689 (83 per cent) were married. The age range was wide (18 to 77) and positively skewed, the mean age being 47.2 years with a standard deviation of 12.1 years. Analysis confirmed that they formed a representative sample of the usual population attending the medical centre.

Four groups were identified: group A, 638 patients (77.1 per cent) without symptoms; group B, 50 patients (6.0 per cent) with recent but not current urinary symptoms (i.e. within the preceding year); group C, 27 patients (3.3 per cent) with current symptoms suggestive of UTI (i.e. strangury, dysuria, haematuria or loin pain with frequency); and group D, 113 patients (13.6 per cent) with any other current urinary symptoms.

The mean age of the 140 women who admitted to having urinary symptoms at the time of examination (groups C and D combined) was 50.6 years, which was significantly different ($p < 0.001$) from that of the patients with no current history (groups A and B combined). The mean age of group C was found to be 47.1 years. This was not significantly different from that of the patients with no current history.

2. Prevalence of bacteriuria

Bacterial growth was classified as described earlier and for each patient the most significant value observed was used in the analysis. Table 1 compares the results of incubation for people

TABLE 1
RESULTS OF URINE CULTURE IN PATIENTS WITH CURRENT URINARY HISTORY

<i>Growth</i>	<i>Significant</i>	<i>Doubtfully significant</i>	<i>Insignificant</i>	<i>Total</i>
<i>Patient group</i>	(3)	(2)	(1)	
Group C	5 (18.5%)	4 (14.8%)	18 (66.7%)	27 (100%)
Group D	5 (4.4%)	13 (11.5%)	95 (84.1%)	113 (100%)
Total	10 (7.1%)	17 (12.1%)	113 (80.7%)	140 (99.9%)

with current symptoms suggestive of urinary infection (group C) and for those with other urinary symptoms (group D). The results are significantly different ($\chi^2 = 7.07$; 2df; $p < 0.05$), suggesting that group D is probably non infective.

TABLE 2
RESULTS OF URINE CULTURE

<i>Growth</i>	<i>Significant</i>	<i>Doubtfully significant</i>	<i>Insignificant</i>	<i>Total</i>
<i>Patient group</i>	(3)	(2)	(1)	
Group C	5 (18.5%)	4 (14.8%)	18 (66.7%)	27 (100%)
Groups A, B, D	38 (4.7%)	84 (10.5%)	679 (84.8%)	801 (100%)
Total	43 (5.2%)	88 (10.6%)	697 (84.2%)	828 (100%)

Table 2 compares the results of incubation of group C's urines with the results of all other cultures in the survey (groups A, B and D combined) when 18.5 per cent of these probably-infected urines showed a significant bacteriuria against only 4.7 per cent of other urines, a highly significant difference ($\chi^2=11.02$; 2df; $p<0.005$).

A further analysis (table 3) revealed that there was no difference in the distribution of bacteriuria in the groups A, B and D ($\chi^2=5.56$; 4df; $0.2 < p < 0.3$).

TABLE 3
RESULTS OF URINE CULTURE IN PATIENTS WITHOUT URINARY SYMPTOMS SUGGESTIVE OF UTI

<i>Growth</i>	<i>Significant</i>	<i>Doubtfully significant</i>	<i>Insignificant</i>	<i>Total</i>
<i>Patient group</i>	(3)	(2)	(1)	
Group A	31 (4.8%)	61 (9.6%)	546 (85.6%)	638 (100%)
Group B	2 (4%)	10 (20%)	38 (76%)	50 (100%)
Group D	5 (4.4%)	13 (11.5%)	95 (84.1%)	113 (100%)
Total	38 (4.7%)	84 (10.5%)	679 (84.8%)	801 (100%)

The overall prevalence of significant bacteriuria in our study was 5.2 per cent. Although 81.5 per cent of group C's urines did not show a significant bacteriuria, their histories revealed that all the women in this group were receiving treatment for UTI from their general practitioner at the time of examination and thus we might expect results of this type. Of possibly greater importance is the finding of a 4.7 per cent prevalence of asymptomatic bacteriuria.

In all groups we noted a prevalence rate for doubtfully significant bacteriuria of about 10–20 per cent.

3. Relationship between bacteriuria and urinary albumin and blood

Tables 4 and 5 show that the presence of albuminuria or haematuria is independent of the degree of bacteriuria (table 4: $\chi^2=1.65$; 2df; $0.4 < p < 0.5$; table 5: $\chi^2=1.59$; 2df; $0.4 < p < 0.5$).

TABLE 4
ALBUMINURIA BY RESULT OF URINE CULTURE

<i>Growth</i>	<i>Significant</i>	<i>Doubtfully significant</i>	<i>Insignificant</i>	<i>Total</i>
<i>Albumin</i>	(3)	(2)	(1)	
Present	2	5	22	29
Absent	41	83	675	799
Total	43	88	697	828

TABLE 5
HAEMATURIA BY RESULT OF URINE CULTURE

<i>Growth</i>	<i>Significant</i>	<i>Doubtfully significant</i>	<i>Insignificant</i>	<i>Total</i>
<i>Blood</i>	(3)	(2)	(1)	
Present	2	1	16	19
Absent	41	87	681	809
Total	43	88	697	828

4. Effect of incubation temperature on growth of bacteria

For each of the 43 patients for whom a significant result was obtained, the quantity $X=(U_{37}-U_{22})$ was calculated, where U_{37} and U_{22} are the coded values representing the growth densities observed for that patient on incubation at 37°C and at room temperature respectively. X can therefore assume any of the values -3, -2, -1, 0, 1, 2, 3. If we hypothesise that there is no real difference in growth at the two temperatures, then observed differences will be merely random errors and X should be distributed symmetrically with a large number of zeros and numbers falling rapidly as we diverge from zero.

The actual distribution of X found in our study is shown in table 6, the two culture media being treated separately. In both cases the distribution is highly positively skewed, only one

TABLE 6
DISTRIBUTION OF $X=(U_{37}-U_{22})$

<i>X</i>	-3	-2	-1	0	1	2	3	<i>Total</i>
<i>Medium</i>								
CLED	—	—	1	14	15	2	11	43
MacConkey agar	—	—	—	20	9	11	3	43

negative value being observed in the case of CLED and none at all in the case of MacConkey agar. Under the assumption of symmetry, these observed distributions are so unlikely that we can reject our initial hypothesis and conclude that there is a genuine difference in growth at the two temperatures, incubation at 37°C tending to produce denser colonies.

Identical results at the two temperatures were found in only 14 cases (32·6 per cent) for cultures grown on CLED medium and in 20 cases (46·5 per cent) for those grown on MacConkey agar. With CLED, as many as 11 cultures (25·6 per cent) which were significant at 37°C showed no growth at room temperature. The corresponding figure for MacConkey agar was three (seven per cent).

Discussion

The consultation rate for UTI has been estimated at 11 per 1,000 consultations (Logan and Cushion, 1958), corresponding to a prevalence rate of four per cent in females (Asscher, 1970). Our figure of 5·2 per cent is generally comparable with this, even though the condition is said to be less common in the higher social classes (Henderson *et al.*, 1962; Turck *et al.*, 1962).

Group D had current urinary symptoms not regarded by us as suggestive of urinary infection. The patients in this group had a higher mean age than every other group in this study, and a low prevalence of significant bacteriuria which was related more to that of groups A and B than to group C.

The patients in group D were probably sufferers of long-standing disabilities such as urge incontinence whose prevalence increases with age. The overall prevalence rate of asymptomatic

significant bacteriuria was assessed at 4·7 per cent which again is comparable with many other studies (Asscher, 1970).

Since the false positive rate of the 'Uricult' technique is about ten per cent (Jackaman *et al.*, 1973) and that of our method of collecting a specimen of urine is approximately 20 per cent (Kass, 1962), we can postulate a true prevalence of 3·4 per cent.

The low prevalence rate of 18·5 per cent for symptomatic significant bacteriuria can have a variety of explanations although the likeliest is that, since the women were under their general practitioner's care at the time of examination, treatment had reduced the number of pathogens present in the bladder urine.

When a woman complains of dysuria or some such symptom one of the first actions taken is usually chemical analysis of her urine. The results of such testing could not be related to the outcome of culture in our study.

When dip-inoculum urine cultures were first introduced it was hoped that general practitioners would be able to determine the presence of bacteriuria themselves, relying on the bacteriology laboratory only for identification of significant cultures. Experience has shown that 'Uricult' is an efficient method of achieving this (Mackey and Sandys, 1965; Guttman and Naylor, 1967; Jackaman *et al.*, 1973), but that doctors using this technique would require a special chamber for correct incubation. It has been suggested that incubation of such a culture at room temperature is as effective as incubation at the conventional 37°C (Arneil *et al.*, 1970; Arneil, 1970) despite the fact that gram-positive cocci fail to grow at room temperature and could result in a failure rate of up to 15 per cent (Maskell and Polak, 1970).

A series of 308 cultures examined after incubation at room temperature for 24 hours produced an eight per cent error rate when compared with the results of 18 hours further incubation at 36°C (Mann and Sandys, 1970). In our study we found that more than 50 per cent of results of cultures at room temperature differed from the corresponding growth at 37°C, and we would therefore recommend that, unless incubation at 37°C can be performed by the doctor in his surgery, the 'Uricult' tubes should be sent by post to the nearest bacteriology laboratory for correct processing.

Summary

Urine samples from 828 women were examined for bacteriuria, using the bacterial growth on 'Uricult' dip-slides as a measure. Of the 801 women without current symptoms suggestive of urinary tract infection, 4·7 per cent were found to have significant bacteriuria compared with 5·2 per cent of the whole group. Comparison of the growth on 'Uricult' at 37°C and room temperature for each patient confirmed that the correct incubation temperature is essential for acceptable results.

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