

# **Diversity of Group B Streptococcus Serotypes Causing Urinary Tract Infection in Adults**

Kimberly B. Ulett,<sup>1†</sup> William H. Benjamin Jr.,<sup>2,3†</sup> Fenglin Zhuo,<sup>4</sup> Meng Xiao,<sup>4</sup> Fanrong

Kong,<sup>4</sup> Gwendolyn L. Gilbert,<sup>4</sup> Mark A. Schembri,<sup>5</sup> and Glen C. Ulett<sup>2,6\*</sup>

<sup>1</sup> Departments of Medicine, <sup>2</sup> Microbiology, and <sup>3</sup> Pathology, University of Alabama at Birmingham, Birmingham AL 35294

<sup>4</sup> Centre for Infectious Diseases and Microbiology, Institute of Clinical Pathology and Medical Research, Westmead, NSW, Australia 2145

<sup>5</sup> School of Chemistry and Molecular Biosciences, University of Queensland, Brisbane QLD, Australia 4072

<sup>6</sup> School of Medical Sciences, Centre for Medicine and Oral Health, Griffith University Gold Coast Campus, QLD, Australia 4222

Short Title: GBS Urinary Tract Infection

Abstract: 250 words

Text: 3158 (excluding abstract)

<sup>†</sup>Contributed equally

\*Correspondence: Glen C Ulett, Department of Microbiology, University of Alabama at Birmingham, 845 19<sup>th</sup> Street South, AL 35294-2170; phone +1 205 996 7695; fax +1 205 934 0605; email [gulett@uab.edu](mailto:gulett@uab.edu)

## ABSTRACT

25 Serotypes of group B streptococcus (GBS) that cause urinary tract infection (UTI) are poorly characterized. We conducted a prospective study of GBS UTI in adults to define the clinical and microbiological characteristics of these infections including which serotypes cause disease. Patients who had GBS cultured from urine over a one year period were grouped according to symptoms, bacteriuria, and urinalysis. Demographic  
30 data were obtained by reviewing medical records. Isolates were serotyped by latex agglutination and multiplex PCR-reverse line blot (mPCR/RLB). Antibiotic susceptibilities were determined by disc-diffusion. GBS was cultured from 387/34,367 consecutive urine samples (1.1%); 62 patients had bacteriuria  $>10^7$  cfu/L and  $\geq 1$  UTI symptom; of these 31 had urinary leukocyte esterase and pyuria (others not tested), 50 (81%) had symptoms  
35 consistent with cystitis and 12 (19%) of pyelonephritis. Compared with controls (who had GBS isolated without symptoms) a prior history of UTI was an independent risk factor for disease. Increased age was also significantly associated with acute infection. Serotyping results were consistent between latex agglutination and mPCR/RLB for 331/387 (85.5%) isolates; 22 (5.7%) and 7 (1.8%) isolates were nontypeable with antisera and by  
40 mPCR/RLB, respectively; 45/56 (80.4%) isolates with discrepant results were typed by mPCR/RLB as serotype V. Serotypes V, Ia, and III caused most UTI; II, Ib, and IV were less common. Nontypeable GBS was not associated with UTI. Erythromycin (39.5%) and clindamycin (26.4%) resistance was common. We conclude that a more diverse spectrum of GBS serotypes than previously recognized causes UTI with the exception of  
45 nontypeable GBS.

Key Words: Group B Streptococcus; *Streptococcus agalactiae*

## INTRODUCTION

Group B streptococcus (GBS) is a leading cause of infection in newborns, pregnant women, and older persons with chronic medical illness (3, 8). In addition to maternal cervicovaginal colonization and neonatal infection that results from vertical transmission of bacteria from mothers to their infants, GBS can also cause urinary tract infection (UTI). The spectrum of GBS UTI includes asymptomatic bacteriuria (ABU), cystitis, pyelonephritis, urethritis, and urosepsis (6, 8, 10, 20, 23, 26). GBS ABU is particularly common among pregnant women, although those most at risk for cystitis due to GBS appear to be the elderly and immunocompromised individuals (8, 9, 25). Despite the uropathogenic nature of GBS, the clinical and microbiological features of GBS UTI, including risk factors for disease, and whether there is a tendency for particular GBS serotypes to cause UTI are poorly understood.

Clinically, UTI due to GBS may be indistinguishable from UTI caused by other uropathogens (25). However, a recent study of multiple uropathogens and host characteristics highlighted unique frequencies of host characteristics in UTI groups as defined by the causal organism (37). This suggests that the clinical and microbiological features of UTI may differ depending on the infecting uropathogen. GBS colonization of the urinary tract in women most likely occurs by an ascending route from the vagina where GBS can persist asymptotically. While the overall prevalence of GBS UTI in the adult population remains unclear GBS bacteriuria during pregnancy occurs at rates between 1-3.5% (4, 23, 41). Many of these episodes represent ABU (2, 18), however, GBS ABU is considered a surrogate for heavy maternal colonization (29, 42) and is

70 currently recommended for intrapartum antibiotic chemoprophylaxis (23, 34). In addition,  
up to 7% of pregnancies may be complicated by GBS UTI, and GBS reportedly  
accounts for approximately 10% of all pyelonephritis during pregnancy (25, 28). GBS  
UTI may also contribute to chorioamnionitis (1), premature onset of labor (24), and an  
increased risk of vertical transmission of GBS (29, 42).

75  
Several studies have also reported high rates of GBS UTI in non-pregnant adults  
(8, 9, 25, 39). In one study GBS was cultured from 39% of all cases of symptomatic UTI  
among nursing home residents >70 years of age (40). Other studies have reported that  
GBS UTI may account for up to one third of all invasive infections due to GBS in adults  
80 (9, 12, 19, 26). Several independent surveys have reported recovery of GBS in between  
1-2% of all UTI cases (7, 26, 30). GBS UTI may also account for up to 7% of late-onset  
disease in neonates (43). Thus, while there is increasing amount of data regarding the  
prevalence of GBS UTI in adults little is known regarding the clinical and microbiological  
features associated with these infections or the GBS serotypes that cause UTI. In this  
85 study we carried out a single-center analysis of adult patients at University of Alabama  
at Birmingham Hospital between August 2007 and August 2008 who had GBS cultured  
from urine during routine assessment for UTI to define the clinical and microbiological  
characteristics of GBS UTI including which serotypes cause disease.

## MATERIALS AND METHODS

90 **Patients** – The study subjects were adult patients (>18 yrs) encountered at University of Alabama at Birmingham Hospital between August 2007 and August 2008 who underwent clinical and microbiological assessment for UTI because of symptoms indicating infection or as part of routine patient screening. Urine samples were obtained as clean-catch voided or catheterized samples from all adult patients who underwent  
95 assessment for UTI during the study period. This included inpatients, patients that were evaluated in the emergency department, and patients from various University Hospital outpatient clinics. In cases where GBS was cultured from urine (any count) the medical records for each patient were reviewed for presenting symptoms at the time of sample collection and demographic data including possible risk factors were recorded. A  
100 provisional UTI diagnosis was defined by the presence of single-organism GBS bacteriuria ( $>10^7$  cfu/L) with  $\geq 1$  symptom(s) that included dysuria, increased urinary frequency and/or urgency, fever  $>38^\circ\text{C}$ , flank pain and/or lumbar tenderness. In cases where urinalysis (UA) was performed UTI was confirmed on the basis of positive urinary leukocyte esterase and pyuria ( $\geq 10^7$  wbc/HPF; non-spun). These are generally-accepted  
105 criteria for the diagnosis of UTI (11, 14, 21). Patients were grouped into probable GBS UTI where UA was not performed and confirmed cases where (positive) UA data was available. This study was performed in accordance with the ethical standards of the University of Alabama Birmingham committee on human experimentation and the Helsinki Declaration. The need for specific informed consent was waived by the  
110 institutional review board of the University of Alabama at Birmingham.

**Controls** – To conduct an analysis of risk factors for GBS UTI we identified a group of control subjects who were defined as having GBS isolated from urine incidentally. These individuals were selected on the basis of low grade GBS bacteriuria ( $<10^7$  cfu/L) in the absence of symptoms. This allowed an assessment of the factors associated with acute GBS UTI versus asymptomatic genitourinary colonization, which is prevalent among healthy adult women. Among the total patient cohort who had GBS isolated from urine there were 51 individuals who satisfied these criteria and all were included as controls. We also performed alternative comparisons using a control group of all 325 individuals who did not satisfy the criteria defined for GBS UTI ('all others'; subjects who were GBS culture-positive but did not have acute symptomatic UTI as defined by the exclusion criteria).

**Bacterial Isolates** – All isolates were cultured from clean-catch voided urine or a catheterized urine sample. Isolates were identified by typical colony morphology on TSA 5% sheep blood agar plates (BD), were tested for catalase, and grouped using the Remel PathoDx latex agglutination kit. Capsular serotyping of each GBS isolate was performed by latex agglutination using commercial antisera from SSI (Denmark) as described elsewhere (36). Antimicrobial susceptibility testing was performed by disc diffusion according to methods of the Clinical and Laboratory Standards Institute. One isolate per patient was analyzed and stored at  $-80^{\circ}\text{C}$  in 15% glycerol in Todd Hewitt Broth. Strains were grown at  $37^{\circ}\text{C}$  on Todd Hewitt agar or in Todd Hewitt Broth.

**Molecular serotyping** – Molecular serotypes were determined at the Centre for

135 Infectious Diseases and Microbiology, Institute of Clinical Pathology and Medical Research, Sydney NSW, for each GBS isolate (17). The molecular serotype (MS) identification of ten serotypes (Ia, Ib, II-VIII) was determined, using a multiplex PCR and reverse line blot (mPCR/RLB) hybridization assay targeting a GBS species-specific gene (*cfb*) and serotype-specific sequences in various genes: *cpsH* for MS Ia, Ib, III, IV, 140 V, VI and IX; *cpsK* for MS II; *cpsM* for MS VII; and *cpsJ* for MS VIII. Methods for extraction of GBS genomic DNA are described elsewhere (15) along with sequences of oligonucleotides, primers, probes and optimal PCR and hybridization conditions (17).

**Discrepancies between latex agglutination and mPCR/RLB** – Isolates were tested

145 independently by the two methods, without knowledge of the results of the other method. When discrepancies were identified, both were repeated and, if discrepancies persisted, a portion of *cpsE-cpsF-cpsG* gene cluster was sequenced (~800 bp) to confirm the result (44). The mPCR/RLB result was accepted as the definitive result.

150 **Statistical Analysis** – We used SPSS v9.0 (SPSS Inc., Chicago) for statistical

analyses. Differences in categorical variables (i.e. risk factors) between case patients and control subjects were examined by Pearson Chi-squared ( $\chi^2$ ) analysis or Fisher's exact test where the expected value of any cell was below 5. For continuous variables without normal distribution (i.e., age), Mann–Whitney U test was used. Gender 155 comparisons were performed using population data (equal group sizes assumed) from the US Census Bureau for Birmingham, AL (male-female ratio 0.857). The quantitative

effects of covariates on variables identified as significant by univariate analysis were evaluated by logistic regression to identify independent risk factors for GBS UTI. Results are given as adjusted odds ratios (OR). P values  $<.05$  were regarded as significant.

## RESULTS

**Study Population** – A total of 34,367 urine samples were collected from patients who underwent assessment for UTI during the study period. These samples represented a total patient cohort of inpatients (39%), outpatients (19.6%), women in obstetrics care (27.4%), and patients who presented to emergency (14%). GBS was cultured from 387 patients (1.1%) representing both clean-catch voided (346/387; 89.4%) and catheterized samples (41/387; 10.6%). The demographic data for the cohort of 387 GBS-positive patients are summarized in Table 1. The mean age for the 387 patients was 46 yrs (range: 18-95 yrs) and 322/387 (83.2%) were female. The most common co-morbid conditions were: urological (21.4%), cardiac (17.6%), neurological (15.5%), endocrine (8.5%), and pulmonary (7.8%). The cohort of 387 patients with urine cultures positive for GBS included inpatients (18.8%), outpatients (52.4%), women in obstetrics care (21%), and patients who presented to emergency (7.8%). Among the 387 patients with urine cultures positive for GBS, 207 (53.5%) had single-organism GBS cultured, and 210 (54.3%) underwent UA. The most common co-cultured organisms were *E. coli* (6.7%), coagulase negative staphylococci (5.7%), diptheroids (4.4%), *Lactobacillus* spp. (3.4%), *Klebsiella pneumoniae* (2.8%), *Staphylococcus aureus* (2.6%), and viridans-type streptococci (2.6%).

**Cases of GBS UTI** – Among 387 patients with urine cultures positive for GBS, 62 patients had single-organism GBS bacteriuria  $>10^7$  cfu/L concurrent with  $\geq 1$  UTI symptom(s) and were defined as probable GBS UTI. Among these, 50/62 patients (81%) had clinical and microbiological features consistent with cystitis versus 12/62

(19%) for pyelonephritis (Table 1). All 31 of the 62 patients who had UA performed, had positive results based on positive urinary leukocyte esterase and pyuria (UA +ve; Table 1). None of the 62 cases were associated with an indwelling urinary catheter and only one patient had GBS concurrently isolated from blood representing probable urosepsis.

**Risk Factors Analysis** – Comparing 62 case patients with 51 controls ('controls', Table 1) the mean age of case patients was significantly greater ( $53 \pm 19$  vs.  $30 \pm 12$ ;  $P < .001$ ).

However, there were no significant differences in the prevalence rates of previously identified (and several other potential) risk factors including limited mobility, diabetes mellitus, chronic kidney disease, and the presence of an indwelling urinary catheter between case patients and control subjects (Table 1). GBS UTI case patients were significantly more likely to have had a prior history of UTI compared to controls (OR, 3.1, 95% CI, 1.1-8.7) and be female (OR, 3.8, 95% CI, 1.6-8.7). A prior history of UTI was highly significant among pyelonephritis case patients (50.0%, 6/12, vs. 11.8%, 6/51,  $P = .007$ ) compared to controls, and was an independent risk factor for GBS pyelonephritis (OR, 10.1, 95% CI, 1.9-54.4). Significantly fewer women of child-bearing age (defined as  $<40$  yrs) with GBS UTI were pregnant compared with women of child-bearing age who were controls (37.5%, 6/16, vs. 81.4%, 35/43,  $P = .003$ ). No significant effects of race were observed (data not shown). Overall, these results were consistent with an alternative analysis where we compared 62 GBS UTI case patients to 'all-other patients' who were GBS culture-positive but did not have acute symptomatic GBS UTI as defined by the exclusion criteria ( $n=325$ ; data not shown).

**GBS serotypes and capsular sequence types** – Twenty two of 387 (5.7%) isolates were nontypeable (NT) by latex agglutination and 7 (1.8%) were NT by mPCR/RLB. Results were consistent, between the two methods for 331/387 (85.5%) isolates. After retesting by both methods, 45/56 (80.4%) isolates for which results were discrepant, were serotyped by mPCR/RLB as serotype V, after being NT (16) or identified as other serotypes (VIII, 15 isolates; II, 6 isolates; and Ia, Ib, IV and III, 2 isolates each) by latex agglutination. Four isolates that were NT by mPCR/RLB were identified, using antisera, as serotypes II (2 isolates), IV (1 isolate) and V (1 isolate). Mismatches for other isolates were (latex agglutination-mPCR/RLB shown): II-Ia (2 isolates), II-III (1 isolate), IV-II (1 isolate), NT-Ia (2 isolates), and NT-Ib (1 isolate). A total of 3 isolates were identified as NT by both methods. Discrepant results for the most common mismatch (VIII-V) were confirmed by sequencing of the mPCR amplicon. The mPCR/RLB serotyping results were accepted as definitive and are shown in Table 2.

The most prevalent serotypes among the 62 cases of GBS UTI were V, Ia, and III (Table 2), which together account for 76% of cases. Serotypes II, Ib, and IV accounted for the remaining 24% of cases. Serotypes III and V GBS were the only serotypes that were more frequently isolated from UTI case patients compared to controls (21% vs 10% and 32% vs 20%, respectively, Table 2) but the differences were not significant. The trend of an increased prevalence of serotype III GBS was statistically significant when we compared 62 GBS UTI case patients with ‘all-other patients’ who were GBS culture-positive but did not have acute symptomatic GBS UTI as defined by the exclusion criteria (21%, 13/62, vs. 10.8%, 35/325,  $P=.026$ ) but there was no difference in

the prevalence of serotype V GBS between case patients and 'all-other patients' (32%, 20/62, vs. 32.3%, 105/325, P=.994).

230

**Antibiotic Susceptibilities** – Disc diffusion analysis of all 387 GBS isolates demonstrated that they were uniformly susceptible to amoxicillin with clavulanic acid, penicillin, linezolid, and chloramphenicol. A considerable proportion of isolates was resistant to clindamycin (102/387, 26.4%) and erythromycin (153/387, 39.5%) and the  
235 majority was resistant to tetracycline (311/387, 80.4%), cefoxitin (343/387, 88.7%), and trimethoprim with sulfamethoxazole (380/387, 98.3%).

## DISCUSSION

The uropathogenic potential of GBS prompted the current study to better define the clinical and microbiological features of GBS UTI and the serotypes that are predominately associated with disease. Among 387 patients who had GBS cultured from urine we identified 62 UTI cases on the basis of single-organism bacteriuria and symptoms; diagnosis was confirmed in half of these cases where UA was performed. Diagnostic strategies for UTI vary substantially between clinicians (11, 14, 21), however, patients with a combination of symptoms have a high probability of UTI (5, 27). Pyuria concurrently with bacteriuria may constitute diagnostic criteria (35), as does  $>10^6$  cfu/L of a uropathogen for cystitis (27, 33). We regarded single-organism GBS bacteriuria  $>10^7$  cfu/L and  $\geq 1$  UTI symptom as probable UTI, and used urinary leukocyte esterase with significant pyuria as confirmatory for diagnosis. Our criteria excluded contaminated samples and low-grade ABU, which are especially common among urine cultures positive for GBS (2, 28, 29) and elderly individuals (13). The majority of 62 GBS UTI cases in this study were uncomplicated cystitis although diagnostic features for pyelonephritis (27) identified this in almost 20% of cases overall. It is noteworthy that our exclusion criteria limited the GBS UTI case definition to only those patients with single-organism GBS bacteriuria. Based on the prevalence of single-organism cultures and the ratio of GBS UTI to ABU (1.2), it is likely that up to one third and one quarter of the 387 urine cultures positive for GBS in this study represent GBS UTI and ABU, respectively. The total isolation rate of GBS from 34,367 urine cultures of 1.1% in this study is consistent with prior reports on the prevalence of GBS UTI among adults (7, 26, 30).

260 Several patients in this study with multiple symptoms of UTI and positive UA findings had GBS bacteriuria between  $10^7$ - $10^8$  cfu/L, which is consistent with reports that up to 30% of women with cystitis present with bacteriuria  $<10^8$  cfu/L. In addition, there were several symptomatic patients who presented with clinical features consistent with acute cystitis including positive UA findings but who were excluded from our case  
265 definition of GBS UTI because of single-organism bacteriuria of only between  $10^6$ - $10^7$  cfu/L at the time of analysis. In one such patient GBS bacteriuria increased four-fold over a six hour period, highlighting the dynamic nature of bacteriuria as an indicator of GBS UTI. These findings support the notion that low-grade bacteriuria ( $>10^6$  cfu/L) of a uropathogen including GBS may be indicative of cystitis in symptomatic individuals (5,  
270 27, 33).

Urinary tract abnormalities, chronic renal failure (26), diabetes mellitus (32), and corticosteroid use (9) are among risk factors previously associated with GBS UTI. In this study the rate of diabetes and chronic kidney disease among GBS UTI case patients did  
275 not differ significantly from control subjects who had GBS isolated from urine in the absence of symptoms. Multivariate analysis however revealed significant effects of a prior history of UTI, which compares to studies that have identified a history of UTI as a risk factor for disease caused by other uropathogens including *Enterobacter cloacae* (37). Increased age was also significantly associated with GBS UTI.

280 Non-typable and serotype III GBS have been identified as causes of UTI (23, 29). In this study, serotype III was the only type more frequently associated with acute

disease compared to other serotypes. Future molecular studies will be important to determine the clonal types of serotype III uropathogenic GBS since only a few clones  
285 cause early- and late-onset neonatal disease (38). Recovery of multiple additional serotypes from case patients, particularly of serotypes II, V, and Ia, shows for the first time that a broader spectrum of GBS serotypes than previously recognized causes UTI. Notably, NT GBS accounted for no cases of GBS UTI in this study in contrast to prior reports (23, 29). Molecular serotyping is more specific than antisera-based assays (17),  
290 which are confounded by variable expression and partial sharing of GBS capsular antigens. It is possible that high prevalence rates of NT GBS among patients with UTI in prior studies may represent novel molecular serotypes that could not be classified by agglutination approaches. Very few GBS isolates are NT by mPCR/RLB; those that are can usually be shown to have mutations in the serotype-specific target region. The high  
295 proportion of molecular serotype V isolates that were NT or gave conflicting results with latex agglutination, is consistent with results of a recent study (16). In a large collection GBS NT isolates, we showed that serotype V was the most common molecular serotype identified. Reactions of these isolates with discordant antisera (especially serotype VIII) may reflect low-titer of antisera and/or cross-reactivity. These findings underscore the  
300 limitations of agglutination serotyping approaches for some GBS in epidemiological surveys (15).

A high rate of resistance to macrolides was observed among GBS isolates in this study. These findings are consistent with a recent survey of GBS in the United States,  
305 where Manning et al. demonstrated higher than expected frequencies of macrolide

resistance among GBS in non-pregnant women (22). Trends of increasing antibiotic resistance may reflect clonal dissemination and horizontal transfer of resistance genes among GBS, which occurs among certain GBS serotypes (31).

310           In summary, GBS UTI in this study occurred mostly as uncomplicated cystitis in women over the age of fifty in the absence of chronic underlying disease but was associated with a prior history of UTI. GBS UTI was caused by a more diverse spectrum of serotypes than previously recognized with the exception of NT GBS. Relatively high rates of treatment failure and poor clinical outcomes have been associated with GBS  
315   UTI (26). Further longer term surveillance studies will help to better define the clinical features and serotypes associated with these important infections.

## **ACKNOWLEDGEMENTS**

This study was supported with grants from the National Health and Medical Research Council (455901, 569674), and an Australian Academy of Sciences Fellowship (G.C.U.).

320 We thank Elisabeth Adderson and David Briles for helpful discussions.

## REFERENCES

1. **Anderson, B. L., H. N. Simhan, K. M. Simons, and H. C. Wiesenfeld.** 2007. Untreated asymptomatic group B streptococcal bacteriuria early in pregnancy and chorioamnionitis at delivery. *Am J Obstet Gynecol* **196**:524 e1-5.
- 325 2. **Aungst, M., J. King, A. Steele, and M. Gordon.** 2004. Low colony counts of asymptomatic group B streptococcus bacteriuria: a survey of practice patterns. *Am J Perinatol* **21**:403-7.
3. **Baker, C. J.** 2000. Group B Streptococcal infections, p. 222-37. *In* D. L. Stevens and E. L. Kaplan (ed.), *Streptococcal Infections. Clinical aspects, microbiology, and molecular pathogenesis.* Oxford University Press, New York.
- 330 4. **Baker, C. J.** 1997. Group B streptococcal infections. *Clin Perinatol* **24**:59-70.
5. **Bent, S., B. K. Nallamothu, D. L. Simel, S. D. Fihn, and S. Saint.** 2002. Does this woman have an acute uncomplicated urinary tract infection? *Jama* **287**:2701-10.
- 335 6. **Bronsema, D. A., J. R. Adams, R. Pallares, and R. P. Wenzel.** 1993. Secular trends in rates and etiology of nosocomial urinary tract infections at a university hospital. *J Urol* **150**:414-6.
7. **de Mouy, D., R. Fabre, and J. D. Cavallo.** 2007. [Community-acquired urinary tract infections in 15 to 65 years old female patients in France. Susceptibility of *E. coli* according to history: AFORCOPI-BIO network 2003.]. *Med Mal Infect* **37**:594-598.
- 340 8. **Edwards, M. S., and C. J. Baker.** 2005. Group B streptococcal infections in elderly adults. *Clin Infect Dis* **41**:839-47.

9. **Falagas, M. E., E. S. Rosmarakis, I. Avramopoulos, and N. Vakalis.** 2006.  
345 Streptococcus agalactiae infections in non-pregnant adults: single center  
experience of a growing clinical problem. Med Sci Monit **12**:CR447-51.
10. **Farley, M. M., R. C. Harvey, T. Stull, J. D. Smith, A. Schuchat, J. D. Wenger,  
and D. S. Stephens.** 1993. A population-based assessment of invasive disease  
due to group B Streptococcus in nonpregnant adults. N Engl J Med **328**:1807-11.
- 350 11. **Hay, A. D., and T. Fahey.** 2002. Clinical diagnosis of urinary tract infection. Jama  
**288**:1229; author reply 1230-1.
12. **Hernaiz, C., N. Anton, J. I. Alos, B. Orden, M. A. Orellana, J. Colomina, J.  
Redondo, and J. L. Gomez-Garces.** 2004. [Clinical significance of  
Streptococcus agalactiae isolation from urine samples of outpatients from health  
355 care centers]. Enferm Infecc Microbiol Clin **22**:89-91.
13. **Juthani-Mehta, M.** 2007. Asymptomatic bacteriuria and urinary tract infection in  
older adults. Clin Geriatr Med **23**:585-94, vii.
14. **Kaufmann, J., and G. A. Modest.** 2002. Clinical diagnosis of urinary tract  
infection. Jama **288**:1229-30; author reply 1230-1.
- 360 15. **Kong, F., S. Gowan, D. Martin, G. James, and G. L. Gilbert.** 2002. Serotype  
identification of group B streptococci by PCR and sequencing. J Clin Microbiol  
**40**:216-26.
16. **Kong, F., L. M. Lambertsen, H. C. Slotved, D. Ko, H. Wang, and G. L. Gilbert.**  
2008. Use of phenotypic and molecular serotype identification methods to  
365 characterize previously nonserotypeable group B streptococci. J Clin Microbiol  
**46**:2745-50.

17. **Kong, F., L. Ma, and G. L. Gilbert.** 2005. Simultaneous detection and serotype identification of *Streptococcus agalactiae* using multiplex PCR and reverse line blot hybridization. *J Med Microbiol* **54**:1133-8.
- 370 18. **Le, J., G. G. Briggs, A. McKeown, and G. Bustillo.** 2004. Urinary tract infections during pregnancy. *Ann Pharmacother* **38**:1692-701.
19. **Lefebvre, N., E. Forestier, M. Mohseni-Zadeh, V. Remy, O. Lesens, C. Kuhnert, V. Poindron, P. Riegel, Y. Piemont, D. Christmann, and Y. Hansmann.** 2007. [Invasive *Streptococcus agalactiae* infections in non-pregnant adults.]. *Med Mal Infect.*
- 375 20. **Lefevre, J. C., J. P. Lepargneur, R. Bauriaud, M. A. Bertrand, and C. Blanc.** 1991. Clinical and microbiologic features of urethritis in men in Toulouse, France. *Sex Transm Dis* **18**:76-9.
21. **Libbus, M. K.** 2002. Review: specific combinations of symptoms effectively rule in the diagnosis of urinary tract infection based on history alone. *Evid Based Nurs* **5**:119.
- 380 22. **Manning, S. D., K. E. Schaeffer, A. C. Springman, E. Lehotzky, M. A. Lewis, L. M. Ouellette, G. Wu, G. M. Moorer, T. S. Whittam, and H. D. Davies.** 2008. Genetic diversity and antimicrobial resistance in group B streptococcus colonizing young, nonpregnant women. *Clin Infect Dis* **47**:388-90.
- 385 23. **McKenna, D. S., S. Matson, and I. Northern.** 2003. Maternal group B streptococcal (GBS) genital tract colonization at term in women who have asymptomatic GBS bacteriuria. *Infect Dis Obstet Gynecol* **11**:203-7.

24. **Moller, M., A. C. Thomsen, K. Borch, K. Dinesen, and M. Zdravkovic.** 1984.  
390 Rupture of fetal membranes and premature delivery associated with group B  
streptococci in urine of pregnant women. *Lancet* **2**:69-70.
25. **Muller, A. E., P. M. Oostvogel, E. A. Steegers, and P. J. Dorr.** 2006. Morbidity  
related to maternal group B streptococcal infections. *Acta Obstet Gynecol Scand*  
**85**:1027-37.
- 395 26. **Munoz, P., T. Coque, M. Rodriguez Creixems, J. C. Bernaldo de Quiros, S.  
Moreno, and E. Bouza.** 1992. Group B Streptococcus: a cause of urinary tract  
infection in nonpregnant adults. *Clin Infect Dis* **14**:492-6.
27. **Nicolle, L. E.** 2008. Uncomplicated urinary tract infection in adults including  
uncomplicated pyelonephritis. *Urol Clin North Am* **35**:1-12, v.
- 400 28. **Persson, K., B. Bjerre, L. Elfstrom, S. Polberger, and A. Forsgren.** 1986.  
Group B streptococci at delivery: high count in urine increases risk for neonatal  
colonization. *Scand J Infect Dis* **18**:525-31.
29. **Persson, K., K. K. Christensen, P. Christensen, A. Forsgren, C. Jorgensen,  
and P. H. Persson.** 1985. Asymptomatic bacteriuria during pregnancy with  
405 special reference to group B streptococci. *Scand J Infect Dis* **17**:195-9.
30. **Persson, K. M., M. Grabe, P. Kristiansen, and A. Forsgren.** 1988. Significance  
of group B streptococci in urine cultures from males and non-pregnant females.  
*Scand J Infect Dis* **20**:47-53.
31. **Puopolo, K. M., D. C. Klinzing, M. P. Lin, D. L. Yesucevitz, and M. J.  
410 Cieslewicz.** 2007. A composite transposon associated with erythromycin and  
clindamycin resistance in group B Streptococcus. *J Med Microbiol* **56**:947-55.

32. **Ronald, A.** 2003. The etiology of urinary tract infection: traditional and emerging pathogens. *Dis Mon* **49**:71-82.
33. **Rubin, R. H., E. D. Shapiro, V. T. Andriole, R. J. Davis, and W. E. Stamm.** 1992. Evaluation of new anti-infective drugs for the treatment of urinary tract infection. Infectious Diseases Society of America and the Food and Drug Administration. *Clin Infect Dis* **15 Suppl 1**:S216-27.
34. **Schrag, S., R. Gorwitz, K. Fultz-Butts, and A. Schuchat.** 2002. Prevention of perinatal group B streptococcal disease. Revised guidelines from CDC. *MMWR Recomm Rep* **51**:1-22.
35. **Shaikh, N., N. E. Morone, J. Lopez, J. Chianese, S. Sangvai, F. D'Amico, A. Hoberman, and E. R. Wald.** 2007. Does this child have a urinary tract infection? *Jama* **298**:2895-904.
36. **Slotved, H. C., J. Elliott, T. Thompson, and H. B. Konradsen.** 2003. Latex assay for serotyping of group B Streptococcus isolates. *J Clin Microbiol* **41**:4445-7.
37. **Tabibian, J. H., J. Gornbein, A. Heidari, S. L. Dien, V. H. Lau, P. Chahal, B. M. Churchill, and D. A. Haake.** 2008. Uropathogens and host characteristics. *J Clin Microbiol* **46**:3980-6.
38. **Tong, Z., F. Kong, B. Wang, X. Zeng, and G. L. Gilbert.** 2007. A practical method for subtyping of Streptococcus agalactiae serotype III, of human origin, using rolling circle amplification. *J Microbiol Methods* **70**:39-44.
39. **Toumi, A., A. Ferjani, H. Ben Abdallah, and J. Boukadida.** 2006. [Streptococcus agalactiae in nonpregnant adults]. *Tunis Med* **84**:161-4.

- 435 40. **Trivalle, C., E. Martin, P. Martel, B. Jacque, J. F. Menard, and J. F. Lemeland.**  
1998. Group B streptococcal bacteraemia in the elderly. *J Med Microbiol* **47**:649-  
52.
41. **Whitney, C. G., S. Daly, S. Limpongsanurak, M. R. Festin, K. K. Thinn, T.**  
**Chipato, P. Lumbiganon, J. Sauvarin, W. Andrews, and J. E. Tolosa.** 2004.  
440 The international infections in pregnancy study: group B streptococcal  
colonization in pregnant women. *J Matern Fetal Neonatal Med* **15**:267-74.
42. **Wood, E. G., and H. C. Dillon, Jr.** 1981. A prospective study of group B  
streptococcal bacteriuria in pregnancy. *Am J Obstet Gynecol* **140**:515-20.
43. **Yagupsky, P., M. A. Menegus, and K. R. Powell.** 1991. The changing spectrum  
445 of group B streptococcal disease in infants: an eleven-year experience in a  
tertiary care hospital. *Pediatr Infect Dis J* **10**:801-8.
44. **Zhao, Z., F. Kong, G. Martinez, X. Zeng, M. Gottschalk, and G. L. Gilbert.**  
2006. Molecular serotype identification of *Streptococcus agalactiae* of bovine  
origin by multiplex PCR-based reverse line blot (mPCR/RLB) hybridization assay.  
450 *FEMS Microbiol Lett* **263**:236-9.

**Table 1 – Adult patients who had GBS isolated from urine during routine assessement for UTI at University of Alabama Hospital between August 2007-2008**

|                                  | Total<br>Specimens<br>(n=387) <sup>a</sup> | GBS UTI Cases <sup>b</sup>    |                              |               | Controls <sup>d</sup><br>(n=51) | P<br>(All cases<br>vs controls) |
|----------------------------------|--|-------------------------------|------------------------------|---------------|---------------------------------|---------------------------------|
|                                  |  | UA +ve <sup>c</sup><br>(n=31) | UA ND <sup>c</sup><br>(n=31) | All<br>(n=62) |                                 |                                 |
| Age (mean years; range)          | 46; 18-95                                  | 54; 19-82                     | 52; 19-93                    | 53; 19-93     | 30; 18-64                       | < 0.001 <sup>g</sup>            |
| Female sex                       | 322 (83)                                   | 25 (81)                       | 27 (87)                      | 52 (84)       | 46 (90)                         | 0.002 <sup>h</sup>              |
| Symptoms                         |  |                               |                              |               |                                 |                                 |
| - Dysuria                        | 68 (17.6)                                  | 18 (58.1)                     | 17 (54.8)                    | 35 (56.5)     | 0 (0)                           | ND                              |
| - Frequency                      | 57 (14.7)                                  | 11 (35.5)                     | 12 (38.7)                    | 23 (37.1)     | 0 (0)                           | ND                              |
| - Flank pain                     | 35 (9.0)                                   | 7 (22.6)                      | 7 (22.6)                     | 14 (22.6)     | 0 (0)                           | ND                              |
| - Fever                          | 15 (4.0)                                   | 4 (13.0)                      | 2 (6.0)                      | 6 (10.0)      | 0 (0)                           | ND                              |
| - C/W cystitis <sup>e</sup>      | 130 (33.6)                                 | 25 (80.6)                     | 24 (77.4)                    | 50 (81.0)     | 0 (0)                           | ND                              |
| - C/W pyleonphritis <sup>f</sup> | 65 (16.8)                                  | 6 (19.4)                      | 7 (22.6)                     | 12 (19.0)     | 0 (0)                           | ND                              |
| Pregnant                         | 99 (30.1)                                  | 1 (4)                         | 5 (18.5)                     | 6 (11.5)      | 35 (76)                         |                                 |
| Possible Risk factors            |  |                               |                              |               |                                 |                                 |
| - Limited mobility               | 13 (3.4)                                   | 1 (3.2)                       | 0 (0)                        | 1 (1.6)       | 0 (0)                           | 1.000                           |
| - Diabetes mellitus              | 91 (23.5)                                  | 5 (16.1)                      | 4 (12.9)                     | 9 (14.5)      | 9 (17.6)                        | 0.651                           |
| - Chronic kidney disease         | 72 (18.6)                                  | 4 (12.9)                      | 0 (0)                        | 4 (6.5)       | 7 (13.7)                        | 0.219                           |
| - Indwelling urinary catheter    | 4 (1.0)                                    | 0 (0)                         | 0 (0)                        | 0 (0)         | 0 (0)                           | ND                              |
| - Altered mental status          | 14 (3.6)                                   | 3 (9.7)                       | 2 (6.5)                      | 5 (8.1)       | 0 (0)                           | 0.063 <sup>i</sup>              |
| - Prior History of UTI           | 76 (19.6)                                  | 9 (29.0)                      | 9 (29.0)                     | 18 (29.0)     | 6 (11.8)                        | 0.032 <sup>j</sup>              |
| Pure GBS isolated                | 207 (53.5)                                 | 31 (100)                      | 31 (100)                     | 62 (100)      | 15 (29.4)                       | ND                              |

|                                       |            |           |           |           |           |    |
|---------------------------------------|------------|-----------|-----------|-----------|-----------|----|
| GBS count >10 <sup>7</sup> cfu/L      | 319 (82.4) | 31 (100)  | 31 (100)  | 62 (100)  | 0 (0)     | ND |
| Mean GBS count (x 10 <sup>7</sup> /L) | 4.7 ± 3.6  | 7.4 ± 3.5 | 6.1 ± 3.1 | 6.7 ± 3.4 | 0.5 ± 0.3 | ND |
| UA done                               | 210 (54.3) | 31 (100)  | 0 (0)     | 31 (50)   | 9 (17.3)  | ND |
| - Pyuria                              | 114 (54.3) | 31 (100)  | ND        | 31 (100)  | 0 (0)     | ND |
| - Leukocyte esterase                  | 122 (58.1) | 31 (100)  | ND        | 31 (100)  | 0 (0)     | ND |
| - Hematuria                           | 74 (35.2)  | 21 (67.7) | ND        | 21 (67.7) | 2 (22)    | ND |
| - +ve and C/W UTI <sup>d-f</sup>      | 91 (43.3)  | 31 (100)  | ND        | 31 (100)  | 0 (0)     | ND |

---

NOTE. Data are no. (%) of patients, unless otherwise indicated.

<sup>a</sup>. Consecutive urine specimens sent for culture, from which GBS was isolated.

455 <sup>b</sup>. Patients with ≥1 symptom(s) of UTI and pure growth of GBS >10<sup>7</sup> cfu/L.

<sup>c</sup>. UA: +ve (consistent with UTI) positive leukocyte esterase and pyuria; ND not done.

<sup>d</sup>. Subjects without symptoms from whose urine GBS was isolated in counts <10<sup>7</sup>/L.

<sup>e</sup>. Symptoms consistent with (C/W) cystitis: dysuria and/or frequency.

460 <sup>f</sup>. Symptoms C/W pyelonephritis: dysuria and/or frequency plus flank pain and/or fever >38°C.

<sup>g</sup>. By Mann–Whitney U test.

<sup>h</sup>. By Pearson  $\chi^2$  analysis. Gender comparisons performed using population data (equal group sizes) from the US Census Bureau for Birmingham (male-female ratio 85.7).

<sup>i</sup>. By Fisher's exact test.

465 <sup>j</sup>. By forward stepwise logistic regression subsequent to Pearson  $\chi^2$  analysis.

**Table 2 – Molecular serotyping data for GBS UTI isolates in this study**

| GBS Serotype    | All  |   | GBS UTI Cases <sup>b</sup>                   |                               |          |      | Controls <sup>d</sup><br>( <i>n</i> =51) |                   |
|-----------------|--|---|--|-------------------------------|----------|------|--|-------------------|
|                 | Specimens<br>( <i>n</i> =387) <sup>a</sup> |   |  |                               |          |      |  |                   |
|                 |  | <i>UA</i> +ve <sup>c</sup><br>( <i>n</i> =31) | <i>UA</i> ND <sup>c</sup><br>( <i>n</i> =31) | <i>All</i><br>( <i>n</i> =62) |          |      |  |                   |
|                 | <i>n</i>                                   | (%)   | <i>n</i>                                     | (%)                           | <i>n</i> | (%)  | <i>n</i>                                 | (%)               |
| Ia              | 81   | (21)  | 6  | (19)                          | 8        | (26) | 14                                       | (23)              |
| Ib              | 31   | (8)   | 2  | (7)                           | 3        | (10) | 5  | (10)              |
| II              | 69   | (18)  | 5  | (16)                          | 3        | (10) | 7  | (11)              |
| III             | 48   | (12) <sup>e</sup>                             | 8  | (26)                          | 5        | (16) | 13                                       | (21) <sup>e</sup> |
| IV              | 24   | (6)   | 2  | (7)                           | 1        | (3)  | 3  | (5)               |
| V               | 125  | (32)  | 8  | (26)                          | 12       | (37) | 20                                       | (32)              |
| VI              | 0  | (0)   | 0  | (0)                           | 0        | (0)  | 0  | (0)               |
| VII             | 0  | (0)   | 0  | (0)                           | 0        | (0)  | 0  | (0)               |
| VIII            | 2  | (1)   | 0  | (0)                           | 0        | (0)  | 0  | (0)               |
| NT <sup>f</sup> | 7  | (2)   | 0  | (0)                           | 0        | (0)  | 0  | (0)               |
| IX              | 0  | (0)   | 0  | (0)                           | 0        | (0)  | 0  | (0)               |

NOTE. Data are no. (%) of GBS isolates, unless otherwise indicated.

<sup>a</sup>. Consecutive urine specimens sent for culture, from which GBS was isolated.

<sup>b</sup>. Patients with ≥1 symptom(s) of UTI and pure growth of GBS >10<sup>7</sup> cfu/L.

470 <sup>c</sup>. UA: +ve (consistent with UTI) positive leukocyte esterase and pyuria; ND not done.

<sup>d</sup>. Subjects without symptoms from whose urine GBS was isolated in counts <10<sup>7</sup>/L.

<sup>e</sup>. Difference between the prevalence of serotype III among all GBS UTI case patients and all other non-GBS UTI cases (n=325), significant by Pearson  $\chi^2$  analysis (P=0.026).

<sup>f</sup>. These isolates were identified using antisera as: NT (3), II (2), IV (1) and V (1).