Failure of Urinary Beta-Glucuronidase Activity to Localize the Site of Urinary Tract Infection

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The differentiation of renal from bladder bacteriuria is difficult on clinical grounds alone. To evaluate the correlation between site of infection and urinary beta-glucuronidase activity, 46 patients with well documented recurrent bacteriuria were studied by bilateral ureteral catheterization. Urinary beta-glucuronidase activity was also determined in 46 control subjects. In general, asymptomatic patients with renal bacteriuria, either unilateral or bilateral, had levels of enzyme activity in their urine comparable to patients with infection confined to the bladder and to normals. Only 4 of 25 patients with renal bacteriuria had significant elevations of urinary beta-glucuronidase. After localization of infection, 9 of 10 patients treated with kanamycin, a potentially nephrotoxic drug, developed significant elevations of urinary beta-glucuronidase. The results of these studies indicate that determination of beta-glucuronidase activity in urine is not useful in predicting the site of infection in patients with bacteriuria but may find a role in screening for early nephrotoxicity.

The differentiation of renal from bladder bacteriuria is difficult on clinical grounds alone, especially in the asymptomatic patient. Since recognition of renal involvement may be useful not only in defining groups of patients for epidemiological and therapeutic studies, but also for guiding the duration and intensity of treatment in an individual patient, an accurate method for localizing the site of urinary tract infection is needed.

In the past, several laboratory tests, which have included selective staining of the urinary sediment (12), and observation of the sediment after intravenous administration of bacterial pyrogen (9) or adrenal corticosteroid (8) have been employed to distinguish between upper and lower urinary tract infection. In addition, measurements of O-specific hemagglutinating and other antibody titters (2, 3) and of maximal concentrating ability (13) have also been proposed as methods for differentiating renal involvement in chronic bacteriuria.

Although these procedures may be helpful in characterizing groups of patients, none is specific in individual patients, and to date only bilateral ureteral catheterization has been shown to localize infection with relative certainty (14, 15). However, ureteral catheterization cannot be justified for the routine evaluation of patients with recurrent bacteriuria, and there has been a continued search for less cumbersome methods of localizing infection.

Because beta-glucuronidase is found in high concentrations in the mammalian kidney (10) and can be readily assayed in the urine, this enzyme has been studied as a possible indicator of parenchymal renal disease. For example, Bank and Bailine (1) found concentrations of beta-glucuronidase elevated in the urine of patients with active pyelonephritis but not in patients with bladder infection. A limitation of these earlier studies has been the definition of the site of urinary tract infection based primarily upon clinical parameters rather than upon precise localization within the urinary tract. The purpose of the present study was to define the urinary activity of beta-glucuronidase in a group of patients with recurrent or persistent bacteriuria in whom the site of infection was localized by ureteral catheterization.

MATERIALS AND METHODS

Forty-six patients, 44 women and 2 men, with well documented recurrent infections of the urinary tract were studied by cystoscopy and bilateral ureteral catheterization employing urological and bacteriological techniques described previously from this laboratory (15). Although many of the patients had
symptoms of urinary tract infection in the past, none had acute flank pain, chills, or fever at the time of localization of infection. Urine for enzyme determinations was obtained at the time of localization of infection, and serial observations were made in the majority of patients from urine collected during and after antimicrobial therapy.

Beta-glucuronidase activity was determined by a modification of the method of Fishman and Bernfield (6). Urine (0.1 ml) was incubated with 0.1 ml of 0.01 M phenolphthalein glucuronide in 0.8 ml of 0.2 M acetate buffer (pH 5.0) for 22 hr at 37 C. The reaction was stopped with 5.0 ml of 0.95 M carbonate buffer (pH 10.4), and the optical density was compared to that of a phenolphthalein standard at 550 nm. Micrograms of phenolphthalein liberated per milliliter of urine per hour (Fishman units) were divided by the urine creatinine (milligrams per milliliter) to correct for variations in urine volume and to allow comparison with urine samples collected randomly from 46 normal control subjects without bacteriuria. This enzyme-creatinine ratio is expressed as units of beta-glucuronidase activity and correlates well with the total enzyme excretion for the 24-hr period (7).

RESULTS

The results of beta-glucuronidase activity determined in 21 patients with bladder bacteriuria, 25 patients with unilateral or bilateral renal bacteriuria, and 46 normal individuals are summarized in Fig. 1. The mean beta-glucuronidase activity in the urine of 46 control subjects was 5.1 ± 2.9 units with the accepted upper limit of normal (99% confidence limit) at 13.5 units. This did not differ significantly from the activities in urine of patients with bladder bacteriuria (6.6 ± 3.1 units) or renal bacteriuria (8.7 ± 5.3 units). Although not depicted, 14 patients with unilateral renal bacteriuria had levels of activity in their urine comparable to 11 patients with bilateral involvement. Furthermore, the activity in urine from the infected side in patients with unilateral infection did not differ significantly from the activity in the noninfected side. Although four patients with renal bacteriuria (three with unilateral and one with bilateral infection) did demonstrate activity in their urine above 13.5 units, two of these patients had diabetic glomerulosclerosis, one had a renal calculus, and one was found to have a bladder carcinoma in addition to upper tract bacteriuria.

In contradistinction to the lack of correlation between the site of bacteriuria and elevated urinary enzyme activity, initiation of therapy with kanamycin did result in a prompt rise in urinary beta-glucuronidase activity in 9 of 10 patients treated with this potentially nephrotoxic drug. The mean peak activity was 28.8 units and was attained during the first 3 days of treatment. In 8 of 9 patients, the activity of enzyme in the urine returned to pretreatment levels within 4 days of cessation of therapy with kanamycin. Patients treated with other antimicrobial agents which included ampicillin, nalidixic acid, nitrofurantoin, cephalaxin, and cephaloglycin showed no elevations in beta-glucuronidase activity.

DISCUSSION

The results of the present study demonstrate that, unlike the findings in patients with acute pyelonephritis, asymptomatic patients with renal bacteriuria do not have elevations of urinary beta-glucuronidase activity. An associated abnormality of the urinary tract or concomitant renal parenchymal disease was present in the only four patients in our study who did have elevated enzyme activity. These observations are similar to those made recently by Pfau and co-workers (11), who reported their findings in 33 patients with infection carefully localized by ureteral catheterization. On the other hand, our results appear to differ with the observations of Bank and Bailine (1). This apparent discrepancy may be due to patient selection since we excluded from study patients with acute symptomatic illness. However, it is particularly the group of patients without symptoms or pyelographic abnormalities in which differentiation of the site of infection is difficult on clinical grounds alone.
The elevation of beta-glucuronidase in the urine of patients with "active" kidney disease has been attributed to liberation of the enzyme from lysosomes of injured renal tubular cells. It is of interest that after localization of infection 9 of 10 patients in our study did develop significant elevations of this enzyme in their urine during treatment with kanamycin, a potentially nephrotoxic drug, whereas elevations were not detected in patients receiving other antimicrobials. The elevation in enzyme activity was most marked at the onset of kanamycin therapy and returned to initial levels promptly upon cessation of treatment. A similar effect on beta-glucuronidase activity in the urine has been noted with other nephrotoxins (4), and an elevated level of activity in urine appears to be a sensitive indicator of early refection in patients undergoing renal homotransplantation (7).

In experimental models of pyelonephritis, Coonrod and Paterson (5) observed that urinary beta-glucuronidase was elevated with the onset of infection and decreased with resolution of tubular injury. In their studies, both staphylococcal pyelonephritis and injections of mercuric chloride produced tubular lesions in the rat that were associated with transitory elevations in the excretion of beta-glucuronidase. Conversely, elevations of enzyme failed to occur in the milder enterococcal model, suggesting to these authors that considerable tissue destruction must be present to produce elevations of the enzymes. If these observations in experimental animals are applicable to the situation in man, it appears that most patients with asymptomatic bacteriuria emanating from the upper tract have a minimal inflammatory lesion in the renal parenchyma. However, it is also possible that asymptomatic patients defined as having upper tract infection by ureteral urine culture may have involvement only of the ureter or renal pelvis. If the infections were luminal or superficial, such patients might more closely resemble patients with infection confined to the bladder. In either case, it appears unlikely on the basis of these findings that determination of beta-glucuronidase activity will find much place in differentiating renal from bladder bacteriuria in most patients with urinary tract infection. Perhaps, it may have a role in screening for early renal tubular injury due to nephrotoxins and other causes.

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LITERATURE CITED

1. Bank, N., and S. H. Bailine. 1965. Urinary beta-glucuronidase activity in patients with urinary tract infection. N. Engl. J. Med. 272:70–75.
2. Brenner, D. A., K. F. Fairley, C. O’Keeffe, and P. Kincaid-Smith. 1969. The serum antibody response in renal and bladder infections. Med. J. Aust. 1:1069–1071.
3. Clark, H., A. R. Ronald, and M. Turck. 1971. Serum antibody response in renal versus bladder bacteriuria. J. Infect. Dis., in press.
4. Coonrod, D., and P. Y. Paterson. 1969. Urinary beta-glucuronidase activity. I. Enzyme assay conditions and response to mercuric chloride in rats. J. Lab. Clin. Med. 73:6–16.
5. Coonrod, D., and P. Y. Paterson. 1969. Urinary beta-glucuronidase in renal injury. II. Excretion patterns in experimental staphylococcal pyelonephritis in rats. J. Lab. Clin. Med. 73:17–24.
6. Fishman, W. H., and P. Bernfield. 1955. Glucuronidases, p. 262–269. In S. P. Colowick and N. O. Kaplan (ed.), Methods in enzymology, vol. 1. Academic Press Inc., New York.
7. Leonard, C. D., and R. E. Cutler. 1970. Urinary beta-glucuronidase in renal homotransplantation. Transplantation 19:196–1971.
8. Little, P. J., and H. E. deWardener. 1962. The use of prednisolone phosphate in the diagnosis of pyelonephritis in man. Lancet 1:1145–1149.
9. Pears, M. A., and B. J. Houghton. 1959. Response of infected urinary tract to bacterial pyrogen. Lancet 2:1167–1172.
10. Paul, W., A. Schaprio, and H. Gonick. 1967. Studies of human kidney and urinary beta-glucuronidase. Enzymol. Biol. Clin. 8:47–66.
11. Pfau, A. A. Aabkenazi, and T. G. Sacks. 1968. The value of urinary tract infections. Israel J. Med. Sci. 4:1249–1253.
12. Poirier, K. P., and G. G. Jackson. 1957. Characteristics of leukocytes in urine sediment in pyelonephritis: correlation with renal biopsies. Amer. J. Med. 23:579–586.
13. Ronald, A. R., R. E. Cutler, and M. Turck. 1969. Effect of bacteriuria on the renal concentrating mechanisms. Ann. Int. Med. 70:723–733.
14. Stamey, T. H., D. E. Govan, and J. M. Palmer. 1965. The localization and treatment of urinary tract infections. The role of bactericidal urine levels as opposed to serum levels. Medicine 44:1–36.
15. Turck, M., A. R. Ronald, and R. G. Petersdorf. 1968. Relapse and reinfection in chronic bacteriuria. II. The correlation between site of infection and pattern of recurrence in chronic bacteriuria. N. Engl. J. Med. 278:422–427.