Role of coxsackievirus B_4 in the pathogenesis of acute glomerulonephritis

M. Bayatpour, M.D., F.A.A.P., A. Zbitnew, B.A., M.SC., G. Dempster, M.D., M.R.C. PATH. and K. R. Miller, M.D., F.R.C.P.[C], *Saskatoon, Sask.*

Summary: Coxsackievirus B_4 was isolated from the throat, nose, blood, stools and urine of a 9-year-old boy with acute glomerulonephritis and a pneumonitis. Neutralization test showed a greater than fourfold rise in the antibody titre to coxsackievirus B_4. The antistreptolysin O titre was elevated, but the complement component was within the normal range. The importance of the coxsackievirus B_4 in the pathogenesis of acute glomerulonephritis is clearly indicated; however, further investigations are needed to understand the details of the virus-kidney interaction.

Résumé: *Le rôle du coxsackievirus B_4 dans la pathogénie de la glomérulonéphrite aiguë*

Le coxsackievirus B_4 a été isolé dans le pharynx, le nez, le sang, les fèces et l'urine d'un garçon de 9 ans souffrant d'une glomérulonéphrite aiguë et de pneumonite. Les épreuves de neutralisation ont révélé une augmentation plus que quadruple du titre d'anticorps au coxsackievirus B_4. Le titre d'antistreptolysine O était élevé, mais le complément demeurait dans les limites normales. Cette étude a permis d'indiquer clairement l'importance du rôle du coxsackievirus B_4 dans la pathogénie de la glomérulonéphrite aiguë, mais il faudra procéder à des études plus poussées pour comprendre le mécanisme détaillé de l'interaction virus-rein.

From the Departments of Bacteriology, Medical Microbiology and Pediatrics, University of Saskatchewan and University Hospital, Saskatoon, Saskatchewan

M. Bayatpour, Lecturer and Clinical Virologist, Department of Bacteriology, University of Saskatchewan, and Department of Medical Microbiology, University Hospital
A. Zbitnew, Virologist, Department of Medical Microbiology
G. Dempster, Professor and Head, Department of Bacteriology, University of Saskatchewan, and Department of Medical Microbiology, University Hospital
K. R. Miller, Assistant Professor, Department of Pediatrics

Reprint requests to: Dr. M. Bayatpour, Assistant Professor, Dept. of Pediatrics, Child Development Clinic, Meharry Medical College, Nashville, Tennessee 37208

Acute glomerulonephritis has been known to be an immunological reaction of the host to a preceding infection due to beta-hemolytic streptococcus.[1-3] The question of the possible role of viruses as etiological agents in this condition has been studied recently.[4-8] Burch *et al*[8-10] in two different sets of experimental studies, reported the role of enteroviruses (echovirus type 9 and coxsackievirus B_4) in the pathogenesis of acute glomerulonephritis.

We report a case of acute glomerulonephritis in which we isolated coxsackievirus B_4 from blood, urine and stool, and from cultures of throat and nose. We found also serological evidence of recent beta-hemolytic streptococcus infection.

Case history

A 9-year-old boy was admitted to the local hospital on October 9, 1971 with headache, vomiting and a temperature of 103° F. He complained also of pain in the left lower chest for the previous 24 hours. There was no history of preceding upper respiratory tract or other infection. Chest x-ray showed evidence of right upper lobe consolidation for which he was treated with oral ampicillin. He did not improve but continued to have intermittent fever and headache. On October 14 the colour of his urine was described as being dark red. On October 16 intravenous (IV) fluids were started and the ampicillin was given intravenously. His temperature decreased to 101°F. from 103° by the next day. However, he developed a convulsion which was predominantly on the left side. He was treated with phenobarbital, diphenylhydantoin, paraldehyde and diazepam. After the convulsions had been brought under control, the patient was transferred to University Hospital.

Physical examination

The patient was well developed, appeared well nourished, and was in no acute distress. He appeared drowsy but was responsive to verbal stimuli. His blood pressure ranged from 140/100 to 150/106 mm. Hg; his weight was 63 lbs. Ocular fundi were normal. He was found to have a dental cavity with abscess formation. The rest of the physical examination yielded findings within normal limits.

Laboratory data

The hemoglobin was 12.3 mg./100 ml., erythrocyte count 4,200,000 and leukocyte count 3700. The urine was cloudy red; protein ranged from 250 to 649 mg./100 ml. and there were 100 erythrocytes and 50 to 100 leukocytes per high-power field. Blood urea was 50 to 60 mg./100 ml. and continued at that level until November 9 when it dropped to 38 mg.; on November 15 it was 26 mg. Electrolyte levels were within normal limits. Serum creatinine was normal; creatinine clearance test was not done. Total protein was 6.5 g./100 ml. and albumin 2.5 g./100 ml. Serum cholesterol was 192 mg./100 ml. Immunodiffusion done on October 17, 1971 yielded the following: IgG 1800, IgM 48, IgA 270 and C_3 130 mg./100 ml. The C_3 level on October 20 and 28 and November 15 was 120, 190 and 180 mg./100 ml. Antistreptolysin O titre (ASOT) on admission was 300 Todd units; it rose to 400 on two occasions and on November 15 decreased to 200 Todd units. Erythrocyte sedimentation rate ranged from 60 to 110 mm./hr. for the first three weeks; on November 15 it was 40 mm./hr. Electrocardiogram appearance was within normal limits. Chest x-ray showed right upper lobe infiltration.

Bacteriology

Blood and urine cultures yielded no growth. No pathogenic bacteria were isolated from the throat or the dental abscess.

Viral study

Specimens submitted to the laboratory on October 17, 18 and 22 for viral studies included blood, cerebrospinal fluid, nose swab, throat swab, clean-catch midstream urine, stool, and acute (Oct. 18) and convalescent sera (collected 23 days apart.)

Coxsackievirus B_4 was isolated from blood, urine, stool, nose and throat swabs, but not from CSF. The typing of the virus was done in cell cultures with specific coxsackievirus B_4 rabbit serum obtained from Microbiological Associates.

The acute phase serum had a neutralization titre of less than 1:4 using 100 TCD_{50} prototype coxsackievirus B_4 whereas the titre of the convalescent serum was 1:64.

Hospital course

The boy was treated with bed rest, low salt and protein diet, antihypertensive agents and antibiotics. Blood pressure returned to normal level in a few days. His weight remained constant throughout the hospital course and no edema was evident. Severe proteinuria as well as microscopic hematuria persisted through-

out the period he was in hospital. He was discharged on November 18, 1971.

Discussion

Although the number of reported human cases of acute glomerulonephritis due to viruses is small, there is a great deal of experimental work which supports the possible role of viruses in the pathogenesis of the condition.[8-10]

Yuceoglu, Berkovich and Minkowitz[4] reported acute glomerulonephritis in a set of twins to be associated with echovirus type 9. The virus was isolated from the stool of one twin and the neutralizing antibody level rose fourfold or more. The second twin had a high neutralizing antibody titre during the convalescent period.

Our patient presents a typical case of acute glomerulonephritis with high blood pressure, proteinuria, hematuria and mild hypertensive encephalopathy, with a high level of blood urea nitrogen. X-ray showed evidence of right upper lobe infiltration unresponsive to antibiotics. We were unable to isolate any bacterial pathogens from cultures of blood, throat, dental abscess or urine, but these samples had been collected during the period of antibiotic treatment. The ASOT was high, indicating recent infection with beta-hemolytic streptococcus. However, the presence of a high ASOT is not by itself an indication of kidney involvement, as has been reported by others.[11-12] The complement component was within the normal range, which is in contrast to what is reported in the case of post-streptococcal infection.[13]

The findings of (a) clinical and laboratory evidence of acute glomerulonephritis; (b) viral isolation from the blood, urine, stool and cultures of nose and throat; (c) pneumonitis; and (d) normal serum complement, made us conclude that our patient must have had a recent streptococcal infection which was followed by coxsackievirus B_4 infection, the virus attacking the kidney as well as the lung. We suggest that:

1. The virus is playing the main role in the pathogenesis of the acute glomerulonephritis.
2. The virus is important in activating the so-called "lodged streptococcal antigens" within the kidney.
3. The preceding streptococcal infection predisposes to localization and multiplication of the virus within the renal tissue.

The above speculations would be settled best by employing immunofluorescence tests for coxsackievirus B_4 as well as streptococcus on a kidney biopsy.

It is our hope that by reporting this patient, interest will be stimulated in the investigation of the role of viruses in the pathogenesis of acute glomerulonephritis. Viruses are common infectious agents in childhood[14,15] and the kidney is one of the most susceptible organs for viral propagation. Therefore their role in the pathogenesis of renal disease requires further investigation.

The authors wish to thank Drs. R. S. Ubhi and G. H. Jonat of Biggar, Saskatchewan for referring the patient, and Miss Judith McQuoid for her technical assistance.

References

1. Jennings R: *Natural history of acute glomerulonephritis.* Presented at Seventeenth Annual Conference on the Kidney, Airlie House, Virginia, October 1965
2. Kaplan EL, Anthony BF, Chapman SS, et al: Epidemic of glomerulonephritis associated with Type 49 streptococcus pyoderma. *Am J Med* 48: 28, 1970
3. Treser G, Semar M, McVicar M, et al: Antigenic streptococcal components in acute glomerulonephritis. *Science* 163: 676, 1969
4. Yuceoglu AM, Berkovich S, Minkowitz S: Acute glomerulonephritis associated with ECHO virus Type 9 infection. *J Pediatr* 69: 603, 1966
5. Sun SC, Burch GE, Sohal RS, et al: Coxsackie B_4 viral nephritis in mice and its autoimmune-like phenomena. *Proc Soc Exp Biol Med* 126: 882, 1967
6. Jensen MM: Virus and kidney disease (review). *Am J Med* 43: 897, 1967
7. Virus and kidney (annotation). *Lancet* I: 801, 1968
8. Viruses and renal disease (editorial). *JAMA* 204: 539, 1968
9. Burch GE, Chu KC, Sohal RS: Glomerulonephritis induced in mice by ECHO 9 virus. *N Engl J Med* 279: 1420, 1968
10. Burch GE, Sun SC: Viral nephritis (editorial). *Am Heart J* 75: 1, 1968
11. Lange K, Treser G, Sagel I, et al: Routine immunohistology in renal disease. *Ann Intern Med* 64: 25, 1966
12. McCluskey R, Vassali P, Gallo G, et al: An immunofluorescent study of pathogenic mechanisms of glomerular diseases. *N Engl J Med* 274: 695, 1966
13. Freedman SO: *Clinical Immunology.* New York, Harper and Row, 1971, pp 157-170; 368
14. Burch GE, Sun SC, Chu KC, et al: Interstitial and Coxsackie virus B myocarditis in infants and children. *JAMA* 203: 1, 1968
15. Burch GE, Sun SC, Colcolough HL, et al: Coxsackie B viral myocarditis and valvulitis identified in routine autopsy specimens by F. A. techniques. *Am Heart J* 74: 13, 1967