Attack Rates of Acute Nephritis after Type 49 Streptococcal Infection of the Skin and of the Respiratory Tract

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ABSTRACT Prospective studies in a population of American Indian children during an outbreak of acute nephritis associated with the Type 49 Group A streptococcus permitted a comparison of attack rates of renal complications after infection at different sites and at different ages. Acute nephritis or unexplained hematuria developed in 10 of 42 children (23.8%) with Type 49 streptococcal skin infection, in 2 of 44 (4.5%) with Type 49 throat infection, and in 3 of 16 (18.8%) with simultaneous Type 49 infection at both sites. The higher attack rate of nephritis and hematuria in children with pyoderma indicates that skin lesions played a direct and quantitatively greater role than respiratory infection in the pathogenesis of acute nephritis during this outbreak. Skin infections with the Type 49 strain were followed by evidence of renal complications more often in children younger than 6.5 yr (9 of 21 or 43%) than in older children (1 of 21 or 5%). Attack rates of renal complications after Type 49 skin infection were approximately equal in males and females.

INTRODUCTION

In 1966 a second outbreak of acute nephritis occurred among children at the Red Lake Indian Reservation in Minnesota, concomitant with the reappearance and spread of the Type 49 Group A streptococcus which had been responsible for a larger nephritis epidemic in the same population 13 yr earlier (1).

Longitudinal studies of healthy preschool and school age children, directed toward documenting the occurrence of streptococcal infections and detecting the frequency of renal complications, were initiated shortly before and continued throughout the more recent outbreak. These data allow an evaluation of the relative roles of skin and of respiratory infection by the Type 49 strain in the pathogenesis of acute nephritis. This report presents an analysis of the attack rates of acute nephritis and hematuria after infection at these two sites and an examination of other factors such as age and sex in relationship to the frequency of renal complications after infection of the skin with this nephritogenic strain.

METHODS

Population. Children participating in the Operation Headstart school program were enrolled in the study in early July 1966. When a nephritis outbreak became evident 1 month later, the study group was enlarged by the addition of approximately 100 siblings and contacts of children with acute nephritis. In mid September, the group was further enlarged by adding one of the two classes in each of grades one through six of the Red Lake Elementary School. From July through September, schools and homes were visited each week; thereafter, school visits were made every other week. The Headstart children were followed for a full calendar year but the contact and elementary school groups were followed only through December 1966.

Laboratory studies. At every examination, cultures were obtained of the throat and anterior nares and of any skin lesion suspected to be infected. Cultures were processed as previously described (2) and the plates were quantitated for β-hemolytic streptococci as follows: 1+ for 1-10 colonies; 2+ for 11-50 colonies; 3+ for over 50 colonies; and 4+ for over 50 colonies with a preponderance of streptococcal growth. Three or more colonies from each positive culture

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TABLE I
Distribution by Site of Infection, Sex, and Age of Children in Attack Rate Analysis

<table>
<thead>
<tr>
<th>Site of Type 49 infection</th>
<th>No. of children</th>
<th>Sex</th>
<th>Age in years*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Male</td>
<td>Female</td>
</tr>
<tr>
<td>Skin</td>
<td>42</td>
<td>18</td>
<td>24</td>
</tr>
<tr>
<td>Throat</td>
<td>44</td>
<td>25</td>
<td>19</td>
</tr>
<tr>
<td>Skin-throat</td>
<td>16</td>
<td>5</td>
<td>11</td>
</tr>
<tr>
<td>Total study group</td>
<td>102</td>
<td>48</td>
<td>54</td>
</tr>
</tbody>
</table>

* Calculated to nearest month at beginning of ARP ("at risk period").

were isolated for grouping and all Group A streptococci were identified for M- and T-protein by methods reported earlier (1, 2) and with sera obtained from the National Communicable Disease Center, Atlanta, Ga., and from the Central Public Health Laboratory at Colindale in London. Blood for streptococcal antibodies was obtained from a number of children in July 1966, at the beginning of the study, and again in September 1966. Antistreptolysin O (3), antideoxynucleoside B (4, 5) and antinicotinamide adenine di-

nucleotidase (6-8) were determined as previously described. At each examination a urine test was sent by the Hem-a-

combistix method (Ames Co., Inc., Elkhart, Ind.). Previous studies (2) have shown this to be a highly sensitive screening test for hematuria, detecting less than 10 red blood cells (RBC)/mm³ of urine. When a specimen tested positively for blood with the Hema-

combistix, the sediment of a freshly collected urine was examined microscopically, most often by one of the authors or by a physician at the Red Lake Public Health Service Hospital. Children suspected of having acute nephritis on either clinical or laboratory grounds were usually hospitalized at the University of Minnesota for further studies. All hospitalized patients had renal biopsies and the diagnosis of acute glomerulonephritis was based upon pathological findings by light microscopy as described earlier (1). A detailed description of the clinical, laboratory, and pathologic findings on these patients will be published elsewhere (9, 10).

Criteria for attack rate analysis. A child was considered at risk of developing acute nephritis secondary to Type 49 infection when he was observed for at least 3 wk after acquiring infection as defined below. Minimal observations during such an "at risk period" (ARP) were: (a) initial cultures; (b) initial urinalysis; and (c) followup urinalysis 3 wk later, but most children had additional cultures and urine tests during the minimal ARP of 3 wk. The period was extended for as long as the child had a urinalysis at least every 3rd wk. When no urinalysis was performed for 3 consecutive wk, the ARP was considered ended at the time of the last urinalysis. Each child was analyzed for only the 1st ARP, although seven of the 102 included in this analysis experienced a second infection with Type 49 streptococci at the same or at a separate site. A complete analysis of these and other data suggesting the occurrence of nephritis of the skin with the same serological type will be presented elsewhere.

Medical records at the Red Lake Public Health Service Hospital of all children considered for this analysis were reviewed for evidence of previous acute nephritis or other renal disease. No evidence of chronic renal or urinary tract disease was encountered and girls with previous transient bouts of pyuria associated with acute symptoms of urinary tract infection were not included since there was no reason to suspect that this would be associated with recrudescent hematurna at the time of streptococcal infection. Medical treatment was administered by the Public Health Service physicians. Exudative pharyngitis and extensive skin lesions were usually treated with systemic or oral antibiotics whereas less extensive skin lesions were often treated topically. Children receiving antimicrobial therapy within the first 3 wk of the ARP were excluded from the analysis unless such treatment was clearly without effect on the presence of Group A streptococci in cultures of throats and (or) skin lesions.

Children otherwise qualifying for analysis were excluded for the following reasons: previous acute nephritis (one child); treatment during ARP (two children); unexplained baseline hematuria (three children).

For 84 of the 102 children accepted for analysis, one or more negative complete urinalyses were on record at the Red Lake Hospital and (or) one or more negative Hema-

combistix tests had been performed by us before the ARP. 11

TABLE II
Response of Three Streptococcal Antibodies* in Children with Group A Streptococcal Throat "Infection" and "Questionable Infection"†

<table>
<thead>
<tr>
<th>Interpretation of throat culture</th>
<th>No. of</th>
<th>1+</th>
<th>2+</th>
<th>3+</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>children</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Infection</td>
<td>15§</td>
<td>13</td>
<td>11</td>
<td>4</td>
</tr>
<tr>
<td>Questionable infection</td>
<td>7</td>
<td>2</td>
<td>1</td>
<td>0</td>
</tr>
</tbody>
</table>

* Antistreptolysin O, antideoxynucleoside B, antinicotinamide adenine dinucleotidase.
† "Infection" defined as minimum of one 3+ culture or series of 2+ cultures. "Questionable infection" defined as recovery of no more than one 2+ culture or a series of 1+ cultures.
§ "Infection" group includes six children with Type 49 throat infection included in the attack rate analysis. Other nine children were similarly infected with Type 49 but for other reasons did not qualify for analysis or were infected with other Group A streptococci.


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of the remaining 18 children without a more complete examination had a negative partial urinalysis (sugar and albumin) on record. Seven children with no baseline urinalyses whatever were accepted for the study because six began the ARP with a negative Hema-combi-stix and the remaining child, a 17 yr old female, had an initially positive test which could be attributed to menses and her subsequent tests were negative.

Type 49 skin infection was defined as the presence of the epidemic strain (Type 49) in cultures of material from purulent skin lesions. The nature of these superficial lesions is described elsewhere (2, 9) and every child in this group yielded one or more cultures containing 3 + streptococci from his lesion(s).

Type 49 throat infection was defined as the isolation of the epidemic strain from at least one throat culture with 3 + streptococci or from a series of throat cultures with 2 + or greater growth. Throat cultures with only 1 + streptococci were disregarded. Similarly, the isolation of Type 49 organisms from nose cultures was disregarded, as the origin and significance of nasal Group A streptococci is often uncertain. Previous studies indicate that they may be transitory residents (11) or secondary to pharyngitis (12) or to pyoderma (2).

Type 49 combined skin-throat infection was considered present when a child met the criteria for both skin and throat infection as defined above, either simultaneously or in succession without an intervening absence of the epidemic strain.

RESULTS

Of approximately 600 children examined from July 1966 to June 1967, 102 qualified for this analysis of attack rates. The distribution of the skin, throat, and skin-throat groups and other characteristics of the population under analysis are shown in Table I.

Validity of criteria for streptococcal infection. For purposes of this analysis, the definition of skin infection related to the Type 49 streptococcus posed no particular problem. Recovery of the epidemic strain was associated with a manifest clinical lesion and streptococci were consistently isolated in large numbers from skin lesions (3 + or more at least once in each patient). Moreover, previous data indicate that a significant response of antibodies to one or more streptococcal extracellular products usually occurred in association with infection so defined (13).

A valid definition of streptococcal pharyngitis proved to be more difficult. Only 8 of 44 children in the throat infection group and 2 of 16 children in the skin-throat group (10/60 or 17% of the combined groups) were known to manifest signs and symptoms attributable to streptococcal respiratory infection. Since overt clinical manifestations of respiratory infection were rare, it was necessary to rely primarily upon bacteriological and serological data for the definition of throat infection. In Table II the results of serial antibody titers from bleedings taken before and after the ARP are shown. Children who fulfilled the bacteriological criteria for throat infection are compared with children who had positive throat cultures which contained too few streptococcal colonies to meet these criteria ("questionable infection"). 13 of 15 children (80%) classified as having throat infection developed a significant (0.2 log) increase in one or more streptococcal antibodies. Since only two of seven children in the questionable group developed a similar response, it seems likely that many children with these bacteriologic findings represent chronic carriers rather than acute infection. These data suggest that most children defined as having Type 49 throat infection on the strength of bacteriological findings had bona fide infection as expressed by an antibody response.

Comparability of groups at risk with regard to length and number of observations. It was important to establish that the duration and frequency of observations were not different from group to group. If one group were followed significantly longer or more closely than another, this might be expected to result in a higher frequency of observed complications. As shown in Table III, the periods of observation (ARP) were longer in the throat infection group (mean 8.8 wk) than in the skin infection or skin-throat groups (means of 6.0 and 5.1 wk respectively), and during the ARP the throat infection group

<table>
<thead>
<tr>
<th>Group</th>
<th>Duration of ARP</th>
<th>No. of times cultures in ARP</th>
<th>No. of urinalyses in ARP</th>
<th>Average interval between cultures</th>
<th>Average interval between urinalyses</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Range</td>
<td>Mean</td>
<td>Median</td>
<td>Range</td>
<td>Mean</td>
</tr>
<tr>
<td>Skin</td>
<td>1–23*</td>
<td>6.0</td>
<td>4.8</td>
<td>1–17</td>
<td>5.0</td>
</tr>
<tr>
<td>Throat</td>
<td>3–36</td>
<td>8.8</td>
<td>7.0</td>
<td>2–10</td>
<td>3.2</td>
</tr>
<tr>
<td>Skin-throat</td>
<td>3–10</td>
<td>5.1</td>
<td>5.1</td>
<td>2–11</td>
<td>4.6</td>
</tr>
</tbody>
</table>

Table III

Comparison of Children with Type 49 Infection of Different Sites with Respect to Length and Number of Observations while at Risk

* "At risk periods" (ARP) shorter than 3 wk were confined to children who developed proven or probable acute nephritis, since such children were considered no longer at risk when hematuria occurred under these conditions.
was cultured less frequently. Other differences were not statistically significant. The longer period of observation in children with throat infection did not appear prejudicial however, since the complication rate was lowest in this group. More importantly, the average frequency of urinalysis in the throat group (one every 1.4 wk) was not significantly different than in the other two groups (one every 1.1 wk in both). Therefore, existing differences between the groups in duration or frequency of observations did not appear a likely source of bias.

**Attack rates of acute nephritis and hematuria (Table IV).** Among the 102 children at risk, five cases of biopsy-proven acute nephritis developed. Three additional children not available for hospitalization excreted significant numbers of RBC and also casts (RBC and[or] granular) and are considered instances of probable acute nephritis. Seven of these eight cases occurred in the groups with skin and skin-throat infection and it is of interest that the rates of acute nephritis in these two groups were similar (12%) and noticeably higher than in children with throat infection (2.3%). However, all children who developed a positive Hema-combistix test were not available for hospitalization and several did not have a microscopic urine examination at an appropriate time. For these reasons and because of the small numbers involved, differences in the rates of occurrence of proven and probable nephritis are probably not suitable for statistical analysis.

The Hema-combistix test was performed repeatedly in all children at risk and is a more objective measurement of the true incidence of renal complications. As shown in Table IV, the total rate of hematuria (including proven and probable nephritis and unexplained hematuria) was higher in the skin infection group (23.8%) and skin-throat group (18.8%) than in the group with throat infection alone (4.5%). When the skin infection and throat infection groups were compared (Fig. 1), the incidence of hematuria was significantly higher in the former group ($P = 0.01$). The attack rate of hematuria in the group with skin infection was significantly greater than in the throat infection group when the small number of children with combined skin-throat infection were added to the skin infection group ($P < 0.02$) or added to the throat infection group ($P = 0.03$).

**Clinical features of acute nephritis.** Of the eight children who developed either biopsy-proven or probable acute nephritis, only two manifested overt clinical evidence, both in the group with skin infection. One developed facial edema and had a significant weight loss during hospitalization. The other had mild, transient hypertension which subsided spontaneously. The full spectrum of signs and symptoms of acute nephritis during this outbreak is described in detail elsewhere [9].

**Pattern of unexplained hematuria in children at risk.** Data on the seven children who developed a positive Hema-combistix with either a negative examination for casts or no microscopic examination are summarized in Table V. The shortest duration of a positive test for hematuria was 2 consecutive wk (patient D. S.) and the longest was at least 5 consecutive wk (F. N.). All children in this category eventually reverted to a persistently negative Hema-combistix and none at any time developed signs or symptoms of acute nephritis.

**Influence of other factors on attack rate of hematuria after skin infection.** Besides the site of Type 49 streptococcal infection, the only other variable which appeared to influence the incidence of nephritis and hematuria was
age. As shown in Fig. 2, in children with skin infection, the attack rate of total hematuria was significantly greater in those under the median age of 6.5 yr (9 of 21 or 43%) than in older children (1 of 21 or 5%). Although more older children were at risk during the latter part of the study, this did not appear to bias the results. Attack rates were uniformly low in the older age group and consistently high in the younger group regardless of the period of the study.

The total number of females was slightly higher than males in this study as a result of the inclusion of a larger number of girls with either skin or skin plus throat infection. However, attack rates of renal complications by sex appeared to be approximately equal regardless of the site of the preceding infection (Table VI).

Although Type 49 streptococci persisted longer in the throat than in skin lesions, there was no evidence that persistence of the organism influenced the development of acute nephritis. From the data available it was possible to calculate the minimum mean duration of recovery of the epidemic strain, approximately 5.5 wk in throat cultures and 2.5 wk in skin lesion cultures. The presence of a positive culture at the other site did not appear to affect the duration of persistence at either site. In the two children with throat infection only who developed acute nephritis or hematuria, Type 49 streptococci were recovered for less than 1 wk and less than 2 wk. In the children with skin infection or skin-throat infection who developed acute nephritis or hematuria, the epidemic organism persisted in the skin for an average of 3 wk and in the respiratory tract for 1–2 wk.

Occurrence of other Group A streptococci in children who developed hematuria or acute nephritis. Strains of Group A streptococci in addition to Type 49 were isolated during the ARP from a number of children who developed evidence of hematuria or acute nephritis. Strains with the T-agglutination pattern 5/27/44 (with or without an M 11 reaction) were isolated from two such children. Strains with a T-agglutination pattern of 3/B3264 and a type 41 M reaction were obtained from two children. An M 31 strain was isolated from one patient and M 26 strains (with the T-agglutination pattern

### TABLE V

**Pattern of Unexplained Hematuria**

<table>
<thead>
<tr>
<th>Patient Initials</th>
<th>Site of Type 49 Infection</th>
<th>Time in weeks after beginning of ARP</th>
<th>Urinary Sediment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>0  1  2  3  4  5  6  7  8  9  10</td>
<td></td>
</tr>
<tr>
<td>C. B. Skin-throat</td>
<td>+  +  -  +  -  -  -  0  0  0  0</td>
<td>N.E.*</td>
<td></td>
</tr>
<tr>
<td>K. D. Skin</td>
<td>-  +† -  +  -  0  0  0  -  -  -</td>
<td>3–7 RBC, Occ. WBC</td>
<td></td>
</tr>
<tr>
<td>G. G. Skin</td>
<td>0  +  +  -  0  -  -  0  0  -</td>
<td>N. E.</td>
<td></td>
</tr>
<tr>
<td>W. M. Throat</td>
<td>0  -  +  +  +  -  -  0  -  0</td>
<td>N. E.</td>
<td></td>
</tr>
<tr>
<td>F. N. Skin</td>
<td>0  -  +† +  +  +  +  +§ -  -  -</td>
<td>Occ. RBC and WBC</td>
<td></td>
</tr>
<tr>
<td>E. R. Skin</td>
<td>0  +  +  +  -  +  0  -  -  -  - 0</td>
<td>N. E.</td>
<td></td>
</tr>
<tr>
<td>D. S. Skin</td>
<td>+  +  0  -  0  -  -  -  0  -</td>
<td>N. E.</td>
<td></td>
</tr>
</tbody>
</table>

+ = a positive test for blood; 0 = negative test; — = no test done.

* Not examined.
† Time of microscopic examination.
§ F. N. was negative at 14 wk and on numerous occasions thereafter.

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**Figure 2** Influence of age on attack rates of acute nephritis and hematuria in children with Type 49 streptococcal skin infection. Children younger than the median age of 6.5 years (left bar) are compared with older children (right bar). Prob. AGN = probable acute glomerulonephritis; prov. AGN = proven acute glomerulonephritis.

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but in lent (15, 19) occurs in children with respiratory infection, was probably a rare occurrence in this outbreak, in view of the age of the population involved and the normal baseline urine examinations in the children at risk. Hematuria in the absence of underlying renal disease also occurs during streptococcal pharyngitis caused by both nephritogenic and nonnephritogenic Group A strains, but this is transient and usually lasts less than a week (20). The persistence of positive Hema-combitest tests for blood for 2–5 or more wk in children with “unexplained hematuria” strongly suggests that several had subclinical acute nephritis. Therefore, the incidence of “proven” and “probable” acute nephritis in children with Type 49 skin infection (12%) probably represents a minimal attack rate.

The preponderance of pyoderma over pharyngitis as the antecedent infection during this outbreak of acute nephritis prompts consideration of whether Type 49 streptococci are capable of establishing respiratory in-

**Table VI**

**Occurrence of Renal Complications by Sex and Site of Preceding Infection**

<table>
<thead>
<tr>
<th>Site of Type 49 infection</th>
<th>Male</th>
<th>Female</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total No.</td>
<td>Acute nephritis or hematuria</td>
</tr>
<tr>
<td>Skin</td>
<td>18</td>
<td>5</td>
</tr>
<tr>
<td>Throat</td>
<td>25</td>
<td>1</td>
</tr>
<tr>
<td>Skin-throat</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td>Skin plus Skin-throat</td>
<td>23</td>
<td>6 (26%)</td>
</tr>
</tbody>
</table>
fection which results in nephritis. In a review of experiences with Type 49 strains from several continents, Maxted, Fraser, and Parker emphasized the frequent recovery of strains of this type from populations with pyoderma-associated nephritis, but also recorded instances of nephritis apparently secondary to Type 49 respiratory infection (21). Other reports confirm the potential virulence and nephritogenicity of Type 49 in the respiratory tract (scarlet fever and nephritis) (22), and it should be noted that scarlet fever was prevalent during the original Red Lake epidemic of Type 49-associated nephritis in 1953 (23). An earlier experience reported 10 yr. before Type 49 was first recognized may also represent the association of nephritis and scarlet fever due to this type, as suggested by a similar T-protein antigen (24).

In view of the experience cited above of acute nephritis after Type 49 streptococcal respiratory infection, it is surprising that nephritis followed throat infection so infrequently at Red Lake in 1966. A possible but unlikely explanation is that children harboring the epidemic strain in the pharynx represented the carrier state rather than true infection. Although the complete combination of clinical and serological as well as bacteriological data with which to define infection was lacking in many patients, available serial antibody titers suggest that most of these children experienced true respiratory infection. Additional, more extensive serological information also indicates that Type 49 respiratory infection was common during this outbreak (13).

That Type 49 is not unique among Group A streptococci in the propensity to produce skin infection and acute nephritis is suggested by the recent report of Dillon, Reeves, and Maxted (25) who isolated a Group A streptococcus with an unusual combination of antigens (M 2 and T 25/Imp 19) from patients with impetigo and nephritis. The organism was rare in uncomplicated impetigo or in respiratory infection among individuals drawn from the same population in Alabama (25). Potter, Moran, Poon-King, and Earle, in studies conducted in Trinidad, have compiled similar evidence for a new streptococcal serotype with a capacity for pyoderma and nephritis (26) and Johnson, Baskin, Beachey, and Stollerman have tentatively identified still another similar streptococcus (27). It should be emphasized, however, that for most “impetigo strains” of Group A streptococci, first characterized in England (28) and subsequently shown to be associated with endemic pyoderma in the United States (2, 29), there is little or no evidence for nephritogenic capacity.

The sex distribution of the patients is of some interest in view of the predominance of males in most reported series of cases of acute nephritis (14, 18). The present study shows approximately equal attack rates for renal complications among males and females regardless of the preceding site of infection. Whether this is a peculiarity of infection of the skin as compared with infection of the throat with a nephritogenic strain is not known since the number of children who developed hematuria after throat infection with Type 49 is so low in the present study, and previous studies have not analyzed sex as a factor in the attack rate of nephritis after infection of the upper respiratory tract.

In one earlier study of epidemic infection with a nephritogenic Group A streptococcus, reported by Stetson, Rammelkamp, Krause, Kohen, and Perry (20), the attack rate was carefully defined in terms of the numerator (nephritis) and denominator (infection). Although this outbreak involved a different Group A streptococcus (type 12), a different site of infection (pharynx), a different age group (young adults), and a uniform population by sex ((all males), the attack rate of nephritis (11.4%) was remarkably close to that reported here for skin infections with Type 49 streptococci in children. Nevertheless, there is indirect evidence that the attack rate of acute nephritis may vary widely after infection with a single nephritogenic type, whether due to bacterial, host, or environmental factors (16). Besides the site of infection, the other factor evident at Red Lake in 1966 was the influence of age; nephritis after Type 49 skin infection developed significantly more often in children younger than 6.5 yr. In view of the many variables, known and unknown, which influence the development of acute nephritis, an attack rate approximating that reported here may or may not occur at another time or in a different population infected with a nephritogenic Group A streptococcus.

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