# Epidemiology of human parvovirus B19 in children with sickle cell disease

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Human parvovirus (HPV) B19 causes significant morbidity and mortality in children with sickle cell disease (SCD), but little data are published about the epidemiology of HPV B19 infection and its associated complications in this patient population. In this study, prevalence and incidence rates of HPV B19 were determined in 633 patients with SCD followed at The Children's Hospital of Philadelphia between November 1996 and December 2001. Thirty percent (30%) were HPV B19 immunoglobulin G (IgG) positive at first testing, and the 70% without evidence of past HPV B19 infection were tested annually. One hundred ten patients developed evidence of HPV B19 infection for an incidence rate of 11.3 per 100 patient-years. Sixty-eight episodes of HPV B19–induced transient red cell aplasia occurred with the following clinical events: fever (89.7%), pain (61.8%), acute splenic sequestration (19.1%), and acute chest syndrome (11.8%). Pain, fever, and acute splenic sequestration were more frequent events with acute HPV B19 infections compared with acute events in uninfected patients. The results of this epidemiologic study, the largest and most comprehensive to date, justify the development of HPV B19 prevention strategies to diminish the frequent and often severe complications associated with HPV B19 infections in patients with SCD. (Blood. 2004;103:422-427)

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## Introduction

Human parvovirus (HPV) B19, a common childhood infection, frequently causes transient red cell aplasia (TRCA) in children with sickle cell disease (SCD).[1-3] Although the outcome of some TRCA episodes in children with SCD is benign, many are treated with red cell transfusions to reduce the risk of circulatory collapse from severe anemia.[1,2] Furthermore, other HPV B19–related complications, including acute splenic sequestration (ASS),[4] hepatic sequestration,[5,6] acute chest syndrome (ACS),[7,8] nephrotic syndrome,[9,10] meningoencephalitis, and stroke,[11] also may occur. These complications or untreated severe anemia could result in chronic medical conditions or death.[1,7,12,13] Since HPV B19 was found to be the cause of TRCA in 1981,[14,15] clinicians have been eagerly awaiting the development of a vaccine or other prevention strategies that would diminish the morbidity and mortality of these infections.[16] Accurate epidemiologic data on the frequency of HPV B19 infection and its associated complications in children with SCD are essential for assessing the potential effect of viral prevention programs in this patient population.

The primary objectives of this study were to estimate the incidence and prevalence of HPV B19 infection in a large population of US children with SCD and to determine the frequency of TRCA and other complications in children acutely infected with HPV B19. Special populations such as children receiving chronic transfusions and hydroxyurea therapy were included. Secondary objectives were to determine risk factors for TRCA, red cell transfusions during TRCA, and prolonged hospitalization during TRCA. This information will have medical and economic implications for designing prevention strategies for HPV B19 infection in children with SCD.

## Patients, materials, and methods

### Patients

Starting in November 1996, a yearly screening program for HPV B19 was instituted for all children with SCD followed at the Sickle Cell Center (SCC) of the Children's Hospital of Philadelphia (CHOP). Specimens for simultaneous HPV B19 immunoglobulin M (IgM) and IgG were collected during routine clinic visits starting at age 12 months and during acute illness visits and hospitalizations for suspected episodes of TRCA regardless of age. Children younger than 12 months of age were not tested during routine clinic visits because of the likelihood that a positive serum HPV B19 IgG would represent passive maternal antibody as opposed to evidence of past infection in the child. Yearly routine HPV B19 serologic tests were discontinued when IgG titers became positive.

TRCA episodes were defined as a 30% or greater reduction from the baseline hemoglobin level (acute anemic event according to the Cooperative Study for Sickle Cell Disease [CSSCD]) and severe reticulocytopenia.[17] In addition, ACS, ASS, and stroke were defined according to the CSSCD.[17] These SCD complications were considered related to HPV B19 infection if they occurred 4 weeks before or after the first documented positive HPV B19 IgM for any individual patient. Although serum HPV B19 IgM titers can be positive for 12 weeks,[18] this 8-week period was selected to minimize overestimation of possible HPV B19–related complications. Criteria for red cell transfusion therapy during TRCA episodes were clinician dependent and, therefore, were not standardized. However, standard clinical practice at the SCC at CHOP was to reserve red cell transfusion therapy for those patients who exhibited signs of cardiovascular stress.

Medical charts were reviewed retrospectively, according to Institutional Review Board standards, to obtain information on patient demographics, sibling disease status, chronic transfusion therapy, hydroxyurea therapy,

TRCA episodes, and steady-state hematologic values. Hemoglobin levels and reticulocyte counts were reviewed on all children to ensure identification of TRCA episodes, particularly those episodes that were not associated with hospitalization or HPV B19 infection, and mild hematologic changes associated with HPV B19 infection.

## HPV B19 IgG and IgM serology

HPV B19 IgG and IgM antibody testing was performed on serum collected from 2 to 4 mL coagulated blood submitted on each patient. All sera were stored at 2°C to 8°C for up to 7 days, pending the completion of tests, and were frozen at −20°C for long-term storage. Repeat freezing and thawing of specimens was avoided. Prior to December 2000, HPV B19–specific IgM and IgG antibodies were detected by using indirect immunofluorescence assays (IFAs) manufactured by Diasorin (Stillwater, MN). After December 2000, IgG and IgM sandwich enzyme immunoassays (EIAs) manufactured by Biotrin International (Dublin, Ireland) were substituted when the Diasorin product was discontinued. All assays were performed according to the manufacturers' written instructions, and appropriate HPV B19–specific IgG and IgM positive and negative controls were tested with each batch of patient specimens. Seroconversion was defined as positive HPV B19 IgM, with or without positive IgG, or positive HPV B19 IgG alone in a patient with previously negative serology.

## Statistical analysis

Chi square tests were used to compare SCD genotype, sex, age groups, chronic transfusion therapy, hydroxyurea therapy, and sibling with SCD between children for whom first HPV B19 results were positive and children for whom first HPV results were negative. The Wilcoxon rank sum test was used to compare age at first HPV B19 serologic test between children with positive results and children with negative results. Mean steady-state hemoglobin levels and reticulocyte counts were calculated by using the 2 most recent values obtained the year prior to positive HPV B19 serology. The most recent routine hemoglobin (Hb) F values prior to positive HPV B19 serology were used as baseline Hb F values. The time of acute HPV B19 infection in children who developed positive HPV B19 serologic tests and TRCA episodes was estimated to occur at the time of presentation with severe anemia. The time of acute HPV B19 infection in children who developed positive IgM HPV B19 serologic tests without TRCA episodes was estimated to occur 3 months prior to a positive titer. The time of HPV B19 infection in children who developed positive HPV B19 IgG without positive IgM was estimated to occur at the midpoint between the last negative titer and the first positive titer. Statistical analysis was performed using Stata 7.0 (StataCorp, College Station, TX). Statistical significance was declared if $P < .05$.

Survival analysis incorporating left truncation with right censoring was used to determine incidence rates for HPV B19 infection. When cumulative incidence rates were calculated, left censoring was added.[19] The date of the first HPV B19 serologic test was the time when a subject was first observed. Kaplan-Meier event-free survival curves were calculated, and event-free survival distributions were compared by using the log-rank test. Cox regression models were used to explore associations between prognostic variables such as SCD genotype and event-free survival. Univariate odds ratios and 95% confidence intervals were calculated for SCD genotype, age, and other baseline evaluations to estimate transfusion risk and prolonged hospitalization in the group of children experiencing TRCA. Logistic regression models were used to assess the possible associations between variables such as SCD genotype and TRCA episode requiring transfusion therapy. Univariate odds ratios, 95% confidence intervals, and logistic regression models were used to compare clinical events in children acutely infected with HPVB19 with events in uninfected children.

## Results

### Patient characteristics

Between November 1996 and December 2001, 657 children with SCD aged 12 months and older were followed routinely at the SCC at CHOP. A total of 633 patients had serum collected for 1406 HPV B19 serologic tests. Of the 24 subjects without serologic tests, 1 was not 12 months old at the time of his last clinic visit but was 1.6 years old at the end of the study period, 2 transferred to other institutions before HPV B19 specimens were obtained, 3 were lost to follow-up, and 18 were missed. Demographics on all study subjects are presented in Table 1. The SCD genotype distribution in the study population was similar to the general population with SCD. The 3 patients with rare or "other" SCD genotypes were as follows: SO-Arab (1), SD (1), and SS, $\alpha^{\text{G-Philadelphia}}$ (1).

### HPV B19 serology

Of the 633 children, 187 (29.5%) had positive IgG and negative IgM HPV B19 titers at first testing (cohort A) (Table 2). One set of triplets developed acute TRCA episodes because of HPV B19 prior to 12 months of age. When these children were tested at 12 months according to the clinical protocol, they had evidence of past HPV B19 infection and were included in cohort A. Of the 446 children who were IgM and IgG negative or IgM positive at first testing

## Table 1. Demographics of study population grouped by first HPV B19 results

| | Total population | Cohort A* | Cohort B† | P‡ |
|---|---|---|---|---|
| Total, n | 633 | 187 | 446 | |
| **SCD type, n (%)** | | | | NS |
| SCD-SS | 409 (64.6) | 117 (62.5) | 292 (65.4) | |
| SCD-SC | 145 (22.9) | 43 (23.0) | 102 (22.9) | |
| SCD-Sβ⁺ | 58 (9.2) | 19 (10.3) | 39 (8.7) | |
| SCD-Sβ° | 18 (2.8) | 7 (3.7) | 11 (2.5) | |
| SCD-other | 3 (0.5) | 1 (0.5) | 2 (0.5) | |
| Male sex, n (%) | 335 (52.9) | 88 (47.1) | 247 (55.4) | NS |
| Median age at first HPV B19 test, y (range) | 7.3 (0.9-21.7) | 11.2 (1.1-21.7) | 5.4 (0.9-20.9) | < .001 |
| **Chronic therapy, n (%)** | 91 (14.4) | 13 (6.9) | 78 (17.5) | < .001 |
| Transfusion | 57 | 8 | 49 | |
| Hydroxyurea | 34 | 5 | 29 | |
| Sibling with SCD, n (%) | 147 (23.2) | 47 (25.1) | 100 (22.4) | NS |

HPV B19 indicates human parvovirus B19; SCD, sickle cell disease; NS, not significant.

*Children with positive IgG and negative IgM for HPV B19 at first testing.

†Children with positive IgM or negative IgM and IgG at first testing.

‡*P* represents comparisons between cohorts A and B.

**Table 2. HPV B19 point prevalence, incidence, and cumulative prevalence rate by age groups**

| Age group, y | No. of children at study entry | Children with past HPV B19 infection at study entry, no. (%)* | Incidence rate of HPV B19 during the study period per 100 patient-years (95% CI)† | Cumulative HPV B19 infection rate (%) |
|---|---|---|---|---|
| 1-5 | 227 | 17 (7.5) | 10.0 (5.7-14.3) | By age 5 y (26.0) |
| 5-10 | 178 | 56 (31.5) | 12.1 (7.4-16.9) | By age 10 y (47.0) |
| 10-15 | 128 | 55 (43.0) | 11.8 (6.0-17.7) | By age 15 y (64.1) |
| 15-21 | 100 | 59 (59.0) | 10.6 (3.1-18.0) | By age 20 y (73.2) |
| All | 633 | 187 | 11.3 (8.2-14.4) | |

*Cohort A.
†Cohort B.

(cohort B), 372 (83.4%) had serial HPV B19 serology during the observation period. Of these children, 110 (29.6%) seroconverted during the study period. Cohort B was younger than cohort A ($P < .001$) and had a larger proportion of children on chronic transfusion or hydroxyurea therapy ($P = .002$). For cohort B, the mean length of follow-up was 2.3 years (standard deviation [SD] ±1.8). The incidence rate of HPV B19 in cohort B was 11.3 per 100 patient-years (95% confidence interval [CI], 8.2-14.4). Incidence was highest in the 5- to 10-year age group (Table 2). Fifty percent of those who seroconverted during the study period developed positive HPV B19 serology by 6.6 years of age. SCD genotype, sex, and age at first HPV B19 serologic test did not affect seroconversion. Nine of 29 (31.0%) children receiving hydroxyurea seroconverted, and 7 of 49 (14.3%) on chronic transfusion therapy seroconverted.

### Acute infection with TRCA

Sixty-eight TRCA episodes because of HPV B19 infection occurred during the study period; 65 in cohort B and 3 in cohort A. Five children with SCD-SS in cohort B did not show evidence of HPV B19 infection with TRCA episodes and were not included in the "TRCA on study" analysis. Two of these children did not have follow-up titers after the TRCA episodes, although one had a brother who seroconverted 1 week after a TRCA episode. The other 2 children (aged 10 months and 4.6 years at the time of TRCA episodes) required red cell transfusions. However, titers during the episodes and over several years of follow-up have been repeatedly negative.

The 68 TRCA episodes associated with HPV B19 occurring during the study period, were examined further to elucidate the clinical course and management of TRCA episodes and to estimate the frequency of other complications (Table 3). Several children in this group require further description. One child who had hematologic evidence of the recovery phase of an acute HPV B19 infection and a positive IgM for HPV B19 was included in the TRCA group. Two children, one with SCD-SC and one with SCD-Sβ⁺ thalassemia, were first diagnosed with SCD at the time of acute HPV B19 infections. Steady-state hemoglobin and reticulocyte values for these 2 children were obtained at least 3 months after the acute infections. Seven children were on hydroxyurea therapy, and one child was on chronic transfusion therapy. The median age of children with HPV B19–associated TRCA was 7.6 years (range,

**Table 3. Clinical course of children with TRCA episodes associated with HPV B19**

| | Total population, N = 68 | SCD-SS,* N = 47 | SCD-SC,* N = 14 | Other,† N = 7 | P* |
|---|---|---|---|---|---|
| **Chief complaint, %** | | | | | |
| Fever | 42.7 | 51.1 | 35.7 | 0 | NS |
| Pain | 39.7 | 25.5 | 57.1 | 100 | .049 |
| Pallor | 5.9 | 4.4 | 7.1 | 0 | NS |
| **Laboratory findings** | | | | | |
| Median initial Hb level, g/dL | 5.8 | 5.0 | 8.1 | 8.0 | < .0001 |
| Median initial reticulocyte, % | 1.0 | 1.2 | 0.4 | 0.7 | NS |
| Median lowest Hb level, g/dL | 4.8 | 4.5 | 5.6 | 7.6 | .0002 |
| Median lowest reticulocyte, % | 0.6 | 0.7 | 0.3 | 0.7 | NS |
| Mean Δ hemoglobin level, g/dL (SD) | 4.1 (1.4) | 3.7 (1.0) | 5.4 (1.8) | 3.5 (0.8) | < .001 |
| Neutropenia, % | 17.9 | 21.3 | 15.4 | 0 | NS |
| Thrombocytopenia, % | 26.5 | 14.9 | 64.3 | 28.6 | .001 |
| **SCD complications, %** | | | | | |
| Painful episode | 61.8 | 46.8 | 92.9 | 100 | .002 |
| Acute chest syndrome | 11.8 | 6.4 | 28.6 | 14.3 | .042 |
| Acute splenic sequestration | 19.1 | 8.5 | 50.0 | 28.6 | .002 |
| Fever while hospitalized | 89.7 | 87.2 | 100.0 | 85.7 | NS |
| **Management, %** | | | | | |
| Transfusion for TRCA | 72.1 | 85.1 | 57.1 | 14.3 | .025 |
| Parenteral antibiotics | 88.2 | 85.1 | 100 | 85.7 | NS |
| Opioids | 51.5 | 36.1 | 78.6 | 100 | .007 |
| Mean days hospitalized (SD) | 4.5 (2.2) | 4.2 (1.8) | 5.7 (2.7) | 4.4 (±3.1) | .009 |
| PICU admission, % | 13.2 | 14.9 | 14.3 | 0 | NS |
| Negative HPV B19 serology at presentation, % | 16.2 | 23.4 | 0 | 0 | .054 |

SCD indicates sickle cell disease; TRCA, transient red cell aplasia; HPV B19, human parvovirus B19; PICU, pediatric intensive care unit; NS, not significant.
*Percentages and P values represent comparisons between SCD-SS and SCD-SC groups.
†Category includes 2 patients with SCD-Sβ° and 5 patients with SCD-Sβ⁺.

0.7-19.2). Children with SCD-SC were older than those with SCD-SS, 12.6 years and 6.6 years, respectively ($P = .008$), although these 2 groups did not differ by age at first HPV B19–positive serology.

Most of the children with TRCA episodes complained of fever or pain at presentation, whereas only a small number sought medical attention because of complaints related to acute anemia such as pallor, fatigue, or headache. In most cases, the presence of TRCA was not suspected until the discovery of a lower than expected reticulocyte count obtained as part of evaluation of acute illness. Table 3 summarizes the clinical course and management of these TRCA episodes grouped by patient genotype. Eleven of the 68 children (16.2%), all SCD-SS, had negative HPV B19 IgM and IgG titers during TRCA episodes but had positive IgM or IgG titers at follow-up (median, 0.9 months later). All children with SCD-SC or SCD-S$\beta^+$ thalassemia with TRCA had positive HPV B19 titers at the time of TRCA episodes.

All 68 children with TRCA had at least one complete blood count with a differential during the course of the illness. Hematologic findings are summarized in Table 3. The median absolute neutrophil count (ANC) was 950 in the neutropenic group (range, 352-1456). Children with SCD-SC were thrombocytopenic more frequently than those with SCD-SS ($P = .001$). The median platelet count was 92 000/$\mu$L ($92 \times 10^9$/L) in the thrombocytopenic group (range, 27 000-146 000/$\mu$L [27-$146 \times 10^9$/L]). No patient developed abnormal bleeding. Thrombocytopenia was associated with SCD-SC ($P = .001$) and ASS ($P < .001$), but neutropenia was not ($P = .366$). ACS and ASS were frequent events during TRCA episodes, occurring in 11.8% and 19.1% of patients respectively (Table 3). Children with SCD-SC had more ACS and ASS events compared with the SCD-SS group even when patient age was considered ($P = .042$ and .002, respectively). Stroke and meningitis occurred separately in 2 children with SCD-SS. One child with SCD-SS developed nephrotic syndrome, and one pregnant adolescent with SCD-SS had a spontaneous first-trimester abortion.

During the study period, 76 acute events occurred in children acutely infected with HPV B19 compared with 4262 acute events in children who were not acutely infected. Acute HPV B19 infections were associated with higher frequencies of fever and ASS in univariate analyses and fever, ASS, and painful episodes in multivariable analysis adjusted for age and SCD genotype (Table 4). When a subgroup of children who presented with fever or pain

was analyzed, ACS was more common in children with acute HPV B19 than in those children who were not infected. This observation may reflect the high rate of ACS in children who present with wheezing or cough in our patient population. When children with SCD-SC were analyzed separately, acute HPV B19 infection was associated with a higher frequency of fever, pain, ACS, and ASS. Older age in this subpopulation was associated with more complications in univariate and multivariable analyses.

The only risk factor for seroconversion in the group of children who developed positive HPV B19 serology was having a sibling with a recent HPV B19 infection (odds ratio [OR], 2.97; 95% CI, 1.29-6.81; $P = .010$). The only risk factor for TRCA in children with SCD who seroconverted was a high mean steady-state reticulocyte count ($P = .039$). The majority (72.1%) of TRCA episodes required red cell transfusions. Five of 7 children receiving hydroxyurea therapy who developed TRCA episodes required red cell transfusions as well as one child receiving chronic transfusion therapy. Risk factors associated with red cell transfusion during TRCA episodes were SCD-SS genotype (OR, 7.62; 95% CI, 2.34-24.79; $P = .001$), low mean steady-state Hb level (OR, 0.47; 95% CI, 0.31-0.72; $P < .001$), and high mean steady-state reticulocyte count (OR, 1.2; 95% CI, 1.08-1.43; $P = .002$). Children with SCD-SS with higher baseline Hb F values had a lower frequency of TRCA episodes requiring transfusion (OR, 0.74; 95% CI, 0.57-0.97; $P = .027$). All children with negative serologic tests during TRCA episodes who later developed HPV B19 IgM required red cell transfusions.

The median length of hospitalization for TRCA episodes because of HPV B19 was 4 days (range, 0-5). Children with SCD-SC were hospitalized longer, 5.7 days, compared with 4.3 days in the group with SCD-SS ($P = .009$). Acute chest syndrome (OR, 5.57; 95% CI, 1.03-30.07; $P = .046$), neutropenia (OR, 4.63; 95% CI, 1.25-17.06; $P = .021$), and thrombocytopenia (OR, 3.34; 95%CI, 1.09-10.22; $P = .035$) were associated with prolonged hospitalization (defined as hospitalization $> 4$ days) in univariate analyses. ACS was the only variable to approach statistical significance in multivariable analysis.

### Acute infection with mild to moderate exacerbation of anemia

Few patients were evaluated for acute HPV B19 infections that were associated with mild to moderate exacerbation of anemia. Six children, 4 with SCD-SS, 1 with SCD-S$\beta^0$ thalassemia, and 1 with

**Table 4. Comparison of acute events in children with and without acute HPV B19 infections**

|  | Univariate analysis | | | Multivariable analysis | | |
|---|---|---|---|---|---|---|
|  | OR | 95% CI | *P* | OR | 95% CI | *P* |
| **Total study population\*** | | | | | | |
| ACS | 1.2 | 0.6-2.4 | .57 | 1.8 | 0.9-3.6 | .09 |
| ASS | 9.4 | 5.1-17.4 | < .01 | 11.4 | 5.8-22.1 | < .01 |
| Painful episode | 1.2 | 0.7-1.9 | .36 | 1.9 | 1.1-3.2 | .02 |
| Fever | 2.1 | 1.3-3.4 | < .01 | 3.2 | 1.9-5.6 | < .01 |
| **SCD-SC population†** | | | | | | |
| ACS | 4.5 | 1.4-14.5 | .01 | 9.3 | 2.0-42.4 | < .01 |
| ASS | 49.5 | 16.4-149.4 | < .01 | 50.7 | 14.1-182.4 | < .01 |
| Painful episode | 2.4 | 0.8-7.0 | .11 | 4.6 | 1.1-18.9 | .03 |
| Fever | 1.7 | 0.6-4.8 | .36 | 5.4 | 1.5-20.4 | .01 |
| **Presentation with fever and/or pain‡** | | | | | | |
| ACS | 2.3 | 1.2-4.6 | .02 | 2.3 | 1.1-4.7 | .02 |
| ASS | 12.9 | 6.5-25.2 | < .01 | 13.3 | 6.6-26.8 | < .01 |

*This group represents 76 events with acute HPV B19 and 4262 events without acute HPV B19; multivariable analysis was adjusted for age and SCD genotype.
†This group represents 16 events with acute HPV B19 and 757 events without acute HPV B19; multivariable analysis was adjusted for age.
‡This group represents 65 events with acute HPV B19 and 3696 events without acute HPV B19; multivariable analysis was adjusted for SCD genotype and age.

SCD-S$\beta^+$ thalassemia, developed a 10% to 20% decrease from steady-state hemoglobin values with severe reticulocytopenia during acute HPV B19 infection. All children were hospitalized for fever or pain, but none developed ACS or ASS.

### Acute infections without exacerbation of anemia

A sizable proportion of children with SCD developed positive HPV B19 serology without seeking medical attention for acute illness. Thirty-nine (34.5%) children seroconverted during the course of this study but did not have documented exacerbation of anemia with severe reticulocytopenia: SCD-SS (22), SCD-S$\beta^0$ thalassemia (1), SCD-SC (12), and SCD-S$\beta^+$ thalassemia (4). Two of these children with SCD-SS were on hydroxyurea therapy, and 6 were on chronic transfusion therapy. Children with SCD-SC who seroconverted without developing TRCA were younger, 6.1 years compared with 11.5 years in the SCD-SC group that did develop TRCA ($P = .003$). This pattern was not detected in any other SCD genotype.

### Special patient subgroups

*Siblings.* The secondary attack rate in families with multiple children with SCD was 56.3%. Eight sibling pairs seroconverted simultaneously within 0.7 to 3.7 weeks of one another. Another sibling pair, not included in the estimate of secondary attack rate, seroconverted 75.6 weeks apart. The risk of acute infection with HPV B19 after exposure to an HPV B19–infected sibling with SCD was increased (OR, 2.97; 95% CI, 1.29-6.81; $P = .010$).

*Chronic transfusion therapy.* Forty-nine children in cohort B were on chronic red cell transfusion therapy during the course of this study. Their median age, 5.2 years (range, 1.0-18.2), was similar to that of other children in cohort B ($P = .591$). Twelve developed positive HPV B19 serologic tests, and those children with positive HPV B19 IgM were considered acutely infected. Children with positive HPV B19 IgG without positive IgM may have been infected during the study period or may have passively acquired HPV B19 IgG from red cell transfusion. To distinguish between these 2 groups, repeat HPV B19 titers were obtained on all children with positive IgG, negative IgM, and recent red blood cell transfusions. Seven children were considered infected during the study period on the basis of initial and repeated serologic testing. Therefore, the incidence rate of HPV B19 infection in children with SCD-SS receiving chronic transfusion therapy was 5.9 per 100 patient-years (95% CI, 1.0-15.0) compared with 11.9 per 100 patient-years (95% CI, 7.6-16.2) in those not receiving transfusions or hydroxyurea therapy ($P = .06$).

## Discussion

This study, the largest to date, documents the incidence and prevalence of HPV B19 infection in children with SCD. The point prevalence rate at entry in this study, 29.8%, is similar to a Jamaican report involving a cohort of children with SCD-SS (37%) and a small cohort of children with SCD followed in Brooklyn, New York (34.5%).[2,20] Yet both the point prevalence rate and the period prevalence rate in this study (47%) are lower than the infection rate reported in a second Jamaican cohort, 63%.[3] The differences between the prevalence rates in this and the Jamaican reports may be explained by differences in the median age of the subjects at the time of initial and final serologic testing, differences in the percentage of patients who were HPV B19 positive at study entry, and differences in HPV B19 epidemic frequencies in Jamaica

and the northeastern United States.[2,3] In the Jamaican study, there were interval breaks, sometimes years, between HPV B19 cases. In our study, the longest period without a HPV B19 case is 5 months. Therefore, the pattern of HPV B19 infections in our geographic region, although epidemic at times, is endemic as well with a higher number of sporadic cases in the United States than in Jamaica.

Human parvovirus B19 infection is thought to be an infection transmitted most frequently by school-aged children. Our data support this observation because 47.0% of children younger than 10 years of age were infected by the end of the observation period. This rate is higher than rates observed in healthy hosts from the United Kingdom and Australia where 27% to 28% are infected by 11 years of age.[21,22] Although the incidence and prevalence of HPV B19 infection in children with SCD in this study are high, they may be underestimated because of the lack of follow-up serologic tests in a small proportion of patients in this study.

As expected, children with siblings with acute HPV B19 infections were more likely to become infected with HPV B19 compared with those children without acutely infected siblings. The 56.3% secondary attack rate in the group of children with siblings with SCD is similar to the 60% secondary attack rate in the Jamaican cohort.[2] In families in which more than one child has SCD, siblings should be tested promptly and monitored for TRCA when another sibling is diagnosed with an acute HPV B19 infection.

Although previous reports describe a broad range of non-TRCA complications in children with SCD, they did not determine whether these complications were more frequent in children with acute HPV B19 and SCD. However, this study demonstrates that children acutely infected with HPV B19 are more likely to develop fever, painful episodes, and ASS than uninfected children. These associations were even higher in children with SCD-SC, and included ACS.

Children with SCD genotypes associated with mild disease as well as those receiving transfusion or hydroxyurea therapy have been overlooked frequently in studies of HPV B19 infection, yet their clinical course can be very severe. Fifty percent of the children with SCD-SC or SCD-S$\beta^+$ thalassemia who developed HPV B19 infections developed TRCA. The clinical course of children with SCD-SC and HPV B19 infection deserves special attention. Although many HPV B19 infections were asymptomatic in this group, once worsening anemia developed other SCD complications occurred frequently. Older children with SCD-SC and acute HPV B19 infection seemed to develop a more severe clinical course when compared with younger children. Further studies to better define the clinical course and the immunologic response to HPV B19 infection in children with SCD-SC are needed to determine why morbidity appears higher in this group of children.

Children receiving chronic transfusion or hydroxyurea therapy should have lower red blood cell turnover compared with those with SCD-SS not receiving therapy; therefore TRCA with acute HPV B19 infections might be less common. However, 7 of 9 children receiving hydroxyurea and 1 of 7 children receiving chronic transfusions developed TRCA, and 6 of these children required transfusions. Therefore, children with SCD receiving these therapies who develop acute HPV B19 infection require close follow-up similar to their untreated counterparts.

Children with SCD-SC and SCD-S$\beta^+$ thalassemia seem to seek medical attention later in the course of illness than do those with SCD-SS or SCD-S$\beta^0$ thalassemia. The entire former group had

positive HPV B19 serology by the time they sought medical attention, which may be because of lower red cell turnover in this group, thus lengthening the time to symptomatic anemia. In addition, this subgroup may be less likely to seek medical attention for febrile illness unless fever persists or is accompanied by pain. Neutropenia and thrombocytopenia were common in children with acute HPV B19 infections. Although thrombocytopenia[1,23-25] and neutropenia[1,24] have been observed in patients with SCD and HPV B19 infection, the high frequency in this study may have been due to the inclusion of patients with SCD-SC who may be more likely to develop these hematologic changes because of lower steady-state white cell and platelet counts or ASS. However, the fact that a larger number of children with SCD genotypes associated with mild disease did not develop mild to moderate hemoglobin decreases was surprising because many patients sought medical attention for reasons other than symptomatic anemia.

The false-positive rate in the subgroup of children receiving chronic transfusions was higher than expected, given the high sensitivity and specificity of serologic tests used in this study.[26] This previously unrecognized finding may reflect the passive transfer of HPV B19 IgG through transfused units of blood and the high sensitivity of current HPV B19 serologic tests. Repeated HPV B19 serologic testing is recommended in children who test IgG positive following red cell transfusions because of the high rate of transfusion-acquired HPV B19 IgG in this group of patients. Although HPV B19 can be transmitted through blood products,[27,28]

children with SCD-SS on chronic transfusion therapy had a trend toward a lower HPV B19 incidence rate than children who were not receiving chronic transfusions. This observation requires further investigation.

The percentage of children with HPV B19 infection not associated with worsening anemia was 34.5% in this study compared with 20% to 24% in other reports.[2,3] This higher rate most likely reflects the inclusion of children with SCD genotypes associated with mild disease and those with SCD-SS receiving chronic transfusions or hydroxyurea therapy. In fact, when the untreated group of children with SCD-SS is considered alone, 22.6% were "asymptomatic" or had subclinical HPV B19 infection.

In conclusion, this study has established the prevalence and incidence of HPV B19 in children with SCD in the northeastern United States. In addition, it emphasizes the high morbidity associated with HPV B19 infections in children with SCD. Data from this report support the need for development of a vaccine that primarily protects young children where the incidence of acute HPV B19 infections is high as well as older children with SCD-SC who may be at greater risk of major HPV B19–related morbidity.

## Acknowledgment

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