Selective deficiency of antibody responses to polysaccharide antigens in a child mosaic for partial trisomy 1 (46,XX,dup (1) (q12 → q23)/46,XX)

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The selective inability to mount antibody responses to polysaccharide antigens occurs infrequently and usually is seen in association with other defects of the immune system, such as IgG subclass deficiency, functional asplenia, and Wiskott-Aldrich syndrome. This report describes a selective deficiency of antipolysaccharide immunity in a child mosaic for partial trisomy 1. The association of these two extremely rare conditions in a single patient may suggest a role for a gene on chromosome 1 in the humoral immune response to polysaccharide antigens.

CASE REPORT

The patient, the 3100 gm product of a full-term gestation, was born to a 16-year-old black adolescent. The infant had multiple congenital anomalies, including Pierre Robin syndrome with cleft palate, scoliosis, bilateral mental hypoplasia of the ear canals, hypertrophic obstructive cardiomyopathy, Wolff-Parkinson-White syndrome, severe mental retardation, and cerebral palsy. Ophthalmologic examination revealed myopia and astigmatism with normal visual acuity. The patient had no cataracts, glaucoma, or retinal abnormalities. Auditory examination by evoked potentials revealed no response from the left ear and a mild conductive hearing deficit on the right.

Cytogenetic analysis of lymphocyte cultures from the proband revealed two cell lines [46,XX, dir dup (1) (pter → q23;q12 → q23;q23 → qter)/46,XX]. In the first cell line (39/50 = 78%) a direct duplication in the long arm of chromosome 1 was present, the duplicated segment being 1q12 → 1q23 (Figure). The second cell line (11/50 = 22%) had a normal 46,XX karyotype. The proband was thus mosaic for trisomy of the q12 → q23 portion of chromosome 1. Cytogenetic studies of the proband's mother revealed a normal karyotype. The father was not available for analysis.

When the patient was 4 days of age, midgut volvulus secondary to malrotation developed, and surgery was required. Subsequent surgical procedures included tracheostomy for airway management, gastrostomy tube placement for management of gastroesophageal reflux and recurrent aspiration pneumonia, insertion of a ventriculoperitoneal shunt to control hydrocephalus, cleft palate repair, and tympanostomy tube placement for chronic otitis media. When the patient was 1 month of age, scleral icterus developed in her mother, who was also found to have hepatitis B. The infant, who was symptom free, subsequently had positive test results for both hepatitis B surface antigen and e antigen; her antigen test results have remained persistently positive, but she has had only minimal elevations of liver enzymes.

The child had episodes of pneumococcal bacteremia at ages 20 and 26 months, recurrent otitis media, and several episodes of Pseudomonas tracheitis. Immunizations were up-to-date, including the Haemophilus influenzae b vaccine (HibVax, from Connaught Laboratories, Inc., Swiftwater, Pa.), which was given at age 24 months. The patient was treated with phenobarbital, phenytoin, and theophylline, as well as with metaproteroxenol and cromolyn sodium administered by inhalation. She has continued to grow well, and her height and weight have remained within the normal range.

The mother's family history was negative for miscarriages, early infant deaths, congenital anomalies, or unusual susceptibility to infection. The mother denied alcohol or drug exposure during pregnancy. The father's family history was unobtainable. The parents are unrelated, and the patient has no siblings.

At age 36 months, the child became febrile and was brought to the hospital. There were coarse rhonchi throughout the lungs, but the physical examination findings were otherwise unchanged from baseline findings. The leukocyte count was 8900/mm³ with 41% neutrophils, 45% lymphocytes, and 4% monocytes. The hemoglobin value was 14.0 gm/dl, the hematocrit 41.4%, and the platelet count 244,000/mm³. Lumbar puncture revealed 33 leukocytes/mm³ with 73% polymorphonuclear cells, 99 erythrocytes/mm³, glucose concentration 95 mg/dl (5.3 mmol/L), and protein concentration 55 mg/dl. Cultures of blood were positive for Streptococcus pneumoniae, and counterimmunoelectrophoresis of cerebrospinal fluid was positive for S. pneumoniae antigen.
A diagnostic evaluation for underlying immunodeficiency was begun. Leukocyte counts and differential cell counts were normal on multiple occasions. Serum concentrations of immunoglobulins were normal: IgG 565 mg/dl, IgA 255 mg/dl, IgM 47 mg/dl, IgG1 321 mg/dl, IgG2 135 mg/dl, IgG3 90 mg/dl, and IgG4 31 mg/dl. The antidiaphtheria toxoid antibody level was 5 IU/ml by indirect hemagglutination (>0.01 IU/ml is protective), and the antitetanus antibody level was 3.0 IU/ml by latex agglutination (>0.01 IU/ml is protective). Lymphocyte enumeration was within normal limits: CD2 (total T cells) 67%, CD4 (T helper cells) 34%, CD8 (T cytotoxic/suppressor cells) 32%, and CD20 (B cells) 10% with an absolute lymphocyte count of 3465/mm³. Responses to the mitogens phytohemagglutinin, concanavalin A, Staphylococcus aureus protein A, and pokeweed mitogen were within normal limits. The CH₅₀ titer was 198% of the reference control, no Howell-Jolly bodies were visible on examination of the peripheral blood smear, and a liver-spleen scan was normal. The hemoglobin electrophoresis had a normal (AA⁺) pattern. A serologic test did not detect human immunodeficiency virus; cytomegalovirus was undetectable by culture of the urine.

METHODS

The IgG and IgM antibodies to the *H. influenzae* type b capsular polysaccharide were measured by enzyme-linked immunosorbent assay using Hib coupled to tyramine. Antibody was quantitated by comparison with a reference serum (kindly supplied by Dr. George R. Siber, Dana-Farber Cancer Institute, Boston, Mass.).

Sera were analyzed for antibodies to 12 pneumococcal antigens by radioimmunoassay, as previously described. A standard curve was produced with the use of 10,000 cpm of capsular polysaccharide, internally labeled with carbon 14, in 0.5 ml of diluent. The diluent consisted of phosphate-buffered saline solution (pH 7.4), to which 2.5% fetal calf serum and 0.25% phenol were added. Assays were performed with 0.025 ml serum. After being mixed with antigen, antigen-antibody complexes were precipitated by the addition of equal volumes of saturated ammonium sulfate at 37°C. The precipitate was dissolved in 0.05 ml of 10% detergent (Triton X-100), and radioactivity was measured by liquid scintillation counting.

RESULTS

Antibody titers to pneumococcal antigens were quantitated before and 4 weeks after the patient’s immunization with Pneumovax-23 vaccine (Merck Sharp & Dohme, West Point, Pa.) at age 43 months (Table). After immunization, the patient had no detectable antibody to seven pneumococcal serotypes and only negligible responses to four others (types 4, 7F, 8, and 19F). She had protective antibody levels (≥200 to 300 mg/dl) only to the type 3 serotype, and the level of this antibody did not significantly increase with immunization.

The IgG and IgM antibody levels were measured in response to multiple immunizations with *H. influenzae* type b vaccines (Table). The child had been immunized with HibVax vaccine (polysaccharide antigen) at age 24 months but had less than protective levels (<200 ng/ml) of antibody when tested at age 38 months. She was reimmunized with HibVax vaccine at age 39 months and with ProHIBit vaccine (polysaccharide conjugated to diphtheria toxin, from Connaught Laboratories) at age 43 months. In both cases, she had marginal responses to immunization with barely twofold increases in IgM titer. Protective antibody levels (≥200 ng/ml) were not achieved after the second immunization with polysaccharide alone but required a subsequent immunization with an *H. influenzae* type b polysaccharide–diphtheria toxoid conjugate vaccine. In comparison, 12
**Table.** Antibody responses to polysaccharide vaccines

<table>
<thead>
<tr>
<th>Pneumococcal antigen</th>
<th>Antibody (ng/ml)†</th>
<th>Response to H. influenzae b vaccine</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Before</td>
<td>After</td>
</tr>
<tr>
<td>Type 1</td>
<td>0.0</td>
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<tr>
<td>Type 3</td>
<td>208.6</td>
<td>322.3</td>
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<tr>
<td>Type 4</td>
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<td>Type 6A</td>
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<td>Type 7F</td>
<td>62.5</td>
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<td>Type 8</td>
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<tr>
<td>Type 19F</td>
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<td>49.8</td>
</tr>
<tr>
<td>Type 23F</td>
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<td>0.0</td>
</tr>
</tbody>
</table>

*Pneumovax 23 = 23-valent pneumococcal vaccine.

†Minimum protective antibody levels are generally considered to be ≥200 to 300 ng/ml.

‡After one immunization, 12 age-matched (2- to 4-year-old) normal control subjects had 6400 ± 3760 ng/ml IgM antibody (mean ± SD). Minimum protective antibody levels are generally considered to be ≥150 to 200 ng/ml.

Age-matched (2- to 4-year-old) normal control subjects had a mean IgM antibody level of 6400 ng/ml (range 2370 to 13,100 ng/ml) after their first immunization with *H. influenzae* vaccine.

**DISCUSSION**

This child has an impaired antibody response to polysaccharide antigens despite having normal levels of serum immunoglobulins, normal levels of IgG subclasses, and normal responses to protein antigens such as diphtheria and tetanus toxoids. She did not respond to 11 of 12 pneumococcal serotypes in Pneumovax vaccine and had only a marginal response to one serotype. Two immunizations with an *H. influenzae* type b polysaccharide vaccine failed to induce protective levels of serum antibody. A third immunization with an *H. influenzae*—diphtheria toxoid conjugate vaccine induced protective levels of IgM, but this response was approximately 10% to 20% of the normal response to the first dose of polysaccharide vaccine in children 2 years of age or older. Further functional evidence for humoral immune dysfunction in this child is the history of two episodes of pneumococcal bacteremia and one episode of pneumococcal meningitis with bacteremia.

Impairment of antibody responses restricted to polysaccharide antigens has most frequently been reported in patients with selective deficiencies of IgG2 or IgG4 or both. Impaired antipolsaccharide antibody responses may also occur in other primary immunodeficiency diseases, such as common variable immunodeficiency, ataxia-telangiectasia, the hyperimmunoglobulinemia E syndrome, and the Wiskott-Aldrich syndrome, disorders that this patient did not have. To our knowledge, there have been reports of only three patients with selective inability to respond to polysaccharide antigens. Ambrosino et al. described a 30-year-old man with recurrent pneumococcal pneumonia who had normal serum levels of immunoglobulins and normal antibody responses to protein antigens but impaired antibody responses to polysaccharide vaccines. A second patient was a 7-year-old boy who was in remission after treatment for acute lymphocytic leukemia. He, too, had defective antipolsaccharide immunity with normal serum immunoglobulin levels and normal antibody response to protein antigens. The third patient was a 2-year-old boy who had very low or absent antibody responses to *H. influenzae* type b and pneumococcal vaccines, and also had an associated finding of functional asplenia. Only the first of these three cases occurred in the absence of other immunologic or hematologic abnormalities.

The karyotype of the proband’s father is unknown, but the presence of a normal cell line in the patient suggests that each parent contributed a normal number 1 chromosome at fertilization and that the duplication of 1q12→1q23 was a postzygotic event. It is theoretically possible that the abnormal chromosome 1 was lost from the original cell line and that nondisjunction of the remaining chromosome 1 produced an apparently normal karyotype. However, in the normal cell line the heterochromatic region of one chromosome 1 was slightly larger than that of its homolog (not shown), suggesting that the two chromosomes were, in fact, different and not two copies of a single chromosome.
Approximately 30 patients with partial trisomy of the long arm of chromosome 1 have been reported, but none has had the exact chromosomal duplication of q12 → q23. However, Mertens et al.\textsuperscript{10} described a dysmorphic male infant with a duplication in the region of q11 → q22. The patient, who was 11 months of age at the time of the report, was not noted to have had an unusual history of infections.

Three genes of immunologic interest have been mapped to the portion of chromosome 1 that was duplicated in our patient.\textsuperscript{11} C-reactive protein and amyloid P component map within the region q12 → q23, and a B cell activation marker (blast-1) maps to the region 1 cen → q32. Duplications of these genes would not appear to explain the propensity to infection observed in our patient. However, it is possible that the structure or function of any of these genes may have been disrupted during the DNA breakage and reunion that produced the aberrant chromosome 1 in this patient. In this regard, levels of two gene products in the duplicated region, C-reactive protein and coagulation factor V, were quantitatively normal in our patient.

Although it is not possible to prove that the immunodeficiency and chromosomal duplication are related, we recommend that any patient with one of these findings be investigated for the presence of the other.

REFERENCES