Hyperuricemia and Reticulocytopenia in Association With Autoimmune Hemolytic Anemia in Two Children

Wasil A. Jastaniah, MBBS, FRCPC, Sheila L. Pritchard, BM, BS, FRCPC, John K. Wu, MBBS, MSc, FRCPC, and Louis D. Wadsworth, MBChB, FRCPC, FRCPath

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Abstract
Hyperuricemia developed in 2 children with autoimmune hemolytic anemia with reticulocytopenia at a time of hemolytic crisis. One likely cause of hyperuricemia is the destruction of nucleated RBC precursors by autoantibodies. It is advised that patients with autoimmune hemolytic anemia with reticulocytopenia be examined for hyperuricemia. This might explain the reason for reticulocytopenia and might prevent unnecessary bone marrow procedures. When hyperuricemia is present, supportive therapy might be needed to prevent renal damage.

Autoimmune hemolytic anemia (AIHA) is a relatively uncommon disorder. The exact incidence is unknown but is estimated at less than 0.2 per 100,000 children younger than 20 years. The peak incidence in children occurs in the preschool-age group. The cause is primary (idiopathic) or secondary to underlying diseases such as lymphoproliferative disorders, autoimmune disorders, and infections. Idiopathic AIHA is more common in females and has a peak incidence in the fourth and fifth decades of life.

Depending on the antibody characteristics, AIHA is subclassified into warm antibody AIHA, cold agglutinin syndrome, biphasic antibody AIHA, and drug-induced AIHA. In children, warm antibody AIHA and biphasic antibody AIHA constitute 79% of cases. This is preceded by an acute infection or immunization in more than 50% of patients. The diagnosis is made by demonstrating a positive direct antiglobulin test (DAT) result and/or indirect antiglobulin test (IAT) result, anemia combined with reticulocytosis, hyperbilirubinemia, decreased serum haptoglobin level, elevated lactate dehydrogenase (LDH) level, and/or hemoglobinuria or hemosiderinuria. Hemolysis is primarily an extravascular process. Bone marrow examination rarely is performed but usually shows erythroid hyperplasia. Hyperuricemia has not been reported in association with childhood AIHA. We describe 2 children with severe AIHA, reticulocytopenia, and hyperuricemia.

Case Reports

Case 1
A 15-month-old previously healthy boy was examined because of a 2-day history of vomiting, lethargy, pallor, decreased
oral intake, decreased urine output, and a recent history of intermittent cough. A fine, papular rash developed on his face and trunk 1 week before he was examined and resolved spontaneously. There was no history of fever, diarrhea, weight loss, night sweats, or bleeding. No history of travel, infectious contact, or drug or toxin exposure could be obtained. Dietary history and medical history were unremarkable. Developmental milestones were appropriate for age. Immunizations were up-to-date with the last immunization, measles, mumps, and rubella vaccine, given at 12 months of age. His parents owned a taxidermy shop, but their child had limited exposure to the reagents and chemicals used. The family history was otherwise unremarkable.

At first examination, the boy was pale, lethargic, and showed evidence of heart failure. His temperature was 38°C, pulse was 190 beats per minute, respiratory rate was 50 breaths per minute, and oxygen saturation was 100% in room air. His liver was palpable 3 cm below the costal margin (liver span, 6 cm), and the spleen was enlarged 3 cm below the costal margin. Lymphadenopathy, dysmorphic features, and skin rash were not detected, and the remainder of the examination findings were normal.

He had evidence of AIHA with the following laboratory findings: hemoglobin level, 2.8 g/dL (28 g/L); unconjugated bilirubin level, 1.5 mg/dL (25 µmol/L); LDH level, 971 U/L (reference interval, 500-920 U/L); plasma free hemoglobin level, 0.48 g/L (reference interval, <0.107 g/L); serum haptoglobin level, less than 10 mg/dL (<0.1 g/L; reference interval, 30-200 mg/dL [0.3-2 g/L]); and an inappropriately low absolute reticulocyte count, 92 × 10³/µL (92 × 10⁹/L; reference interval, 40-120 × 10³/µL [40-120 × 10⁹/L]); the corrected reticulocyte count was 23 × 10³/µL (23 × 10⁹/L).²

The DAT result was positive with polyspecific antihuman globulin, anti-IgG was positive (2+), and anti-complement factor 3d was negative using gel card technology (Micro Typing Systems, Pompano Beach, FL). The IAT gave a 2+ reaction with 1 of 2 screening cell panels; a full RBC panel showed variable positive results (weak to 2+) with 6 of 16 cells, but no definite antibody specificity could be assigned. The eluate was panreactive. These results were compatible with the presence of a warm autoantibody. The uric acid level was elevated at 9.4 mg/dL (552 µmol/L; reference interval, 1.8-5.1 mg/dL [105-300 µmol/L]), the urea level was 3 mmol/L (reference range, 1.8-6.1 mmol/L), and the creatinine was 0.3 mg/dL (28 µmol/L; reference range, 0.1-0.6 mg/dL [10-50 µmol/L]); the urinalysis showed large amounts of amorphous urate crystals, but hemosiderin, hemoglobin, urobilinogen, and bilirubin were not detected.

Further workup included a glucose-6-phosphate dehydrogenase screen; pyruvate kinase screen; and vitamin B₁₂, folic acid, and homocysteine levels; all were normal. Viral serologic tests for parvovirus B19 IgG and IgM and Epstein-Barr virus (EBV) capsid antigen IgG and IgM and nasopharyngeal aspirates for viral cultures were all negative. Cytomegalovirus IgM was negative, but IgG was positive. The antinuclear antibody test result was negative, and anti-double-stranded DNA was less than 3.6 IU/mL (reference interval, 0-3.6 IU/mL).

A bone marrow aspirate and biopsy were performed to determine the cause of the reticulocytopenia and hyperuricemia. The marrow aspirate showed increased cellularity with marked erythroid hyperplasia; the majority of the RBC precursors were at the intermediate and late normoblastic stages. There was mild dyserythropoiesis with fewer than 10% of the erythroblasts containing 2 or more nuclei. Iron staining showed absent stainable iron (not an uncommon finding at this age) and no ringed sideroblasts. The bone marrow biopsy showed erythroid hyperplasia with dyserythropoiesis but no evidence of multilineage dysplasia, metastatic infiltration, or lymphoproliferative disease. The results of bone marrow cytogenetic studies were normal.

The chest radiograph showed cardiomegaly but no mediastinal mass or perihilar lymphadenopathy. Abdominal ultrasound showed an enlarged liver but normal echo texture with no focal lesions and an enlarged spleen but no evidence of lymphadenopathy or an abdominal mass.

He was given 2 transfusions with the least incompatible packed RBCs. A total volume of approximately 20 mL/kg of body weight from 2 units was divided into 6 aliquots; these each were transfused over 3 hours. The shelf-age of the transfused aliquots was 21 to 27 days at the time of transfusion. The immediate pretransfusion hemoglobin was 2.8 g/dL (28 g/L), and after 4 aliquots were transfused, his hemoglobin level was 6.9 g/dL (69 g/L). He received 2 more aliquots, and 24 hours after the last aliquot was transfused, his hemoglobin level was 5.9 g/dL (59 g/L). He tolerated the transfusions well with no transfusion reactions. The uric acid level continued to increase, reaching a level of 14.9 mg/dL (878 µmol/L). Hydration was started, and he was given rasburicase (0.2 mg/kg per day, intravenously), a recombinant urate oxidase, for 2 days, and his blood chemistry values were observed closely. Figure II. No corticosteroid therapy was initiated. He was discharged from the hospital on day 7 of his illness in a stable condition and was followed up weekly as an outpatient with no transfusions required. The absolute reticulocyte count was 696 × 10³/µL (696 × 10⁹/L) on day 8, the uric acid level remained normal (3.6 mg/dL [215 µmol/L]), and his hemoglobin level was normal (11.7 g/dL [117 g/L]) by day 34 of his illness. By day 62, he had recovered completely with a negative DAT result and a hemoglobin level of 13.2 g/dL (132 g/L).

Case 2

A 9-year-old previously healthy boy was examined several months after the previous case with a 6-day history of low energy, followed by fever, then jaundice, pallor, and
dark-colored urine. This was associated with intermittent body aches. There was no history of weight loss or night sweats. No history of travel, infectious contact, drug, or toxin exposure was elicited. Dietary history and medical history were unremarkable. He was in grade 4 with no concerns about school performance. Immunizations were up-to-date, with the last immunization, varicella-zoster vaccine, given at 5 years of age. The family history was unremarkable.

On admission to the hospital he was pale, lethargic, and jaundiced. His temperature was 37°C, pulse was 110 beats per minute, respiratory rate was 22 breaths per minute, and oxygen saturation was 100% in room air. His liver was tender and palpable about 3 cm below the costal margin, and the spleen was enlarged 4 cm below the costal margin. He had shotty cervical lymphadenopathy and enlarged tonsils but no other lymph node enlargement. The remainder of the examination findings were unremarkable.

He had evidence of AIHA with the following laboratory findings: hemoglobin level, 5.6 g/dL (56 g/L); unconjugated bilirubin level, 5.3 mg/dL (91 µmol/L); LDH level, 678 U/L; and an inappropriately low absolute reticulocyte count, 135 × 10^3/µL (135 × 10^9/L; reference range, 30-180 × 10^3/µL [30-180 × 10^9/L]); the corrected reticulocyte count was 61 × 10^3/µL (61 × 10^9/L).^5^

The DAT result was positive with polyspecific antihuman globulin, anti-IgG, anti-C3, and anti-IgM but negative with anti-IgA using gel card technology (Micro Typing Systems). The eluate was nonreactive vs screening cells. The results of Donath-Landsteiner test (using a modified 2-stage technique) and cold agglutinin titer were negative. The results were compatible with the presence of a warm autoantibody.

The uric acid level was checked because of the previous case and was found to be elevated, 5.3 mg/dL (312 µmol/L; reference range for age, 2.0-5.0 mg/dL [120-295 µmol/L]; blood chemical testing results were normal, including sodium level, 138 mEq/L (138 mmol/L); urea level, 6 mmol/L (reference range, 2.5-6.4 mmol/L); creatinine level, 0.6 mg/dL (51 µmol/L; reference range, 0.3-0.7 mg/dL [30-60 µmol/L]); and bicarbonate, 27 mEq/L (27 mmol/L), indicating that the hyperuricemia was not related to dehydration.

The urine analysis was positive for urobilinogen and trace blood with a urine RBC count of 1 and the presence of
amorphous urate crystals. Further workup showed a positive Monospot test and a positive EBV semiquantitative polymerase chain reaction with fewer than 100 copies per milliliter of serum. The antinuclear antibody screen was negative. Abdominal ultrasound confirmed the presence of hepatosplenomegaly but no other abnormality. Chest radiograph findings were normal.

Prednisone and folic acid therapy had been started at an outside hospital, and the patient was referred to us because of relative reticulocytopenia and a continued fall in the hemoglobin level. The next day, his hemoglobin level decreased to 4.4 g/dL (44 g/L) and the reticulocyte count to 85 × 10^3/µL (85 × 10^9/L); the corrected reticulocyte count was 28 × 10^3/µL (28 × 10^9/L). The unconjugated bilirubin level was 5.7 mg/dL (97 µmol/L), and the LDH was 2,772 U/L. This coincided with a continued increase in his uric acid level to 6.2 mg/dL (367 µmol/L).

The prednisone was continued, allopurinol was started, and the patient was given 1 transfusion with the least incompatible packed RBCs. A total volume of approximately 10 mL/kg of body weight from 1 unit was transfused over 4 hours. The shelf-age of the transfused unit was 16 days at the time of transfusion. The immediate pretransfusion hemoglobin level was 4.8 g/dL (48 g/L), and approximately 12 hours after transfusion, his hemoglobin level was 8.4 g/dL (84 g/L). He tolerated the transfusion well with the only reaction being anxiety. His reticulocyte count started increasing a few days later, and this coincided with a fall in the uric acid level.

Figure 2 (Case 2) The inverse relation between reticulocyte count (triangles, × 10^9/L) and uric acid level (squares, µmol/L) might suggest that hyperuricemia is secondary to nucleated RBC destruction rather than increased bone marrow compensation. The upper dotted line reflects the upper limit of normal for uric acid level and the bottom line reflects the upper limit of normal for the reticulocyte count. Values are given as Système International units; for conversions to conventional units, see the legend for Figure 1.

The cause of reticulocytopenia in AIHA is not well established, but there is evidence that it is mediated immunologically, whether associated with hyperplasia or hypoplasia of marrow cells. Reticulocytopenia in AIHA might reflect a concomitant accelerated immune-mediated destruction or autoantibody-induced apoptosis of RBC precursors within the bone marrow, a temporary suppression of bone marrow activity secondary to infection, and/or a delayed bone marrow response to the hemolytic event.

In an attempt to study the mechanism of reticulocytopenia in a patient with AIHA, reticulocytopenia, and marrow erythroid hyperplasia, Conley et al. performed differential IAT tests, phagocytic-index assays, and ferrokinetic studies. The differential IAT involved testing the eluate from the patient’s RBCs against reticulocyte-rich and reticulocyte-poor fractions of normal RBCs. Monoctytic-macrophage phagocytic-index assays involved incubation of reticulocyte-rich preparations of normal RBCs sensitized with the patient’s serum antibody. There was no evidence of preferential in vitro destruction of reticulocytes. The ferrokinetic studies (plasma iron clearance and RBC incorporation of radioactive iron measured simultaneously with RBC survival testing) showed retention of radioactive iron in the marrow.
and failure of incorporation of iron into circulating RBCs. Remission of anemia was associated with reticulocytosis, the appearance of radioactive iron in circulating erythrocytes, and a decrease in titer of the serum autoantibody, suggesting that the antibody reacted with marrow cells, preventing their release into the blood. The mechanism by which release of RBCs from the bone marrow was inhibited is unknown. The possibility that the antibody had some effect on the maturing RBC precursors, retarding their proliferation or preventing their extrusion into the circulation, was postulated.

In case 1, a bone marrow DAT was performed by centrifugation of washed, EDTA-suspended bone marrow cells in a microhematocrit tube. DAT was performed on the buffy coat and the bottom RBC layer. The buffy coat, which contains the erythroblasts and reticulocytes, showed positivity, but the RBC layer below the buffy coat had a negative DAT result. The differential positivity between the buffy coat and the lower RBC layer suggests the presence of an antibody directed more toward immature erythrocytes and erythroblasts; however, weak results also were seen with a control marrow sample, which raises uncertainty about the validity of this conclusion.

The presence of reticulocytopenia in association with AIHA aggravates the severity of anemia and increases the need for RBC transfusions.

Hyperuricemia has been reported in association with some congenital hemolytic anemias such as sickle cell anemia. The hyperuricemia in these patients usually is the consequence, not the cause, of renal impairment. Decreased renal function in hemolytic anemias might be due to the presence of free hemoglobin from intravascular hemolysis, proximal tubular dysfunction secondary to oxidative stress, and/or hemodynamic alteration and inflammatory response secondary to the release of immune complexes. Hyperuricosuria has been reported in sickle cell anemia, presumably secondary to the increased synthesis of nucleic acids occurring as part of the erythropoietic response to hemolysis, but hyperuricemia occurs only in patients in whom altered renal tubular function has developed, and it probably is due to hypoxic renal tubular damage caused by sickling.

We can find only 1 report of hyperuricemia in a patient with AIHA; it occurred in a 70-year-old woman in whom lymphoma subsequently developed. Herein we describe 2 children with severe AIHA of the warm autoantibody type associated with reticulocytopenia, increasing uric acid levels, and urinary urate crystals but no renal failure. The severity of anemia and reticulocytopenia paralleled an increase in the uric acid level; remission of anemia was associated with reticulocytosis and normalization of the uric acid level (Figures 1 and 2). Transfusion, hydration, and rasburicase or allopurinol therapy resulted in stabilization of their conditions, clearance of urate crystals, and, possibly, prevention of renal failure.

Reticulocytopenia in association with AIHA portends a serious prognosis. Hyperuricemia, although this may suggest an underlying leukemic or lymphoproliferative process, can be seen in AIHA and might initiate or potentiate renal injury. The high uric acid level in the patients described herein probably was related to the destruction of nucleated RBCs by autoantibodies. A prospective analysis of a large series of patients with severe AIHA would be required to determine the relationship among hyperuricemia, reticulocytopenia, and AIHA. In the light of the present cases, it is advised that the serum uric acid level and urinalysis be checked in patients with hemolytic anemia (especially when associated with reticulocytopenia). The presence of hyperuricemia in a child with AIHA and reticulocytopenia might explain the reason for the reticulocytopenia and possibly prevent unnecessary bone marrow procedures. Supportive therapy with hydration and uric acid–lowering agents might be required in patients with hyperuricemia, especially during active hemolysis, to prevent renal injury.

References


