Paradoxical Erythroid Hypoplasia and Anemia in Standardbred Racehorses Following Recombinant Human Erythropoietin Administration

R. J. Piercy, MA VetMB; C. J. Swardson, DVM, PhD; and K. W. Hinchcliff, BVSc, PhD

The administration of recombinant human erythropoietin to horses may be associated with severe debilitating disease. In some horses, antibodies may be produced that cross react with endogenous equine erythropoietin, resulting in erythroid hypoplasia and anemia. Authors' addresses: The Ohio State Veterinary Teaching Hospital, Columbus, OH 43210 (Piercy and Hinchcliff) and The College of Veterinary Medicine and Biochemical Sciences, Colorado State University, Fort Collins, CO 80523 (Swardson). © 1997 AAEP.

1. Introduction
Anecdotal reports have implicated the use of recombinant human erythropoietin (rhEPO) to enhance the athletic ability of racehorses by stimulating erythrocyte production in the bone marrow.1,2 Deaths in human athletes have been attributed to the abuse of this recombinant hormone, and rumors exist concerning horse morbidity and mortality following its use. This paper describes two separate clinical cases seen with disease associated with documented rhEPO administration and gives an explanation for some of these previously unsubstantiated rumors. It is therefore of major significance to trainers and racetrack veterinarians.

2. Materials and Methods
Two 3-year-old Standardbred racehorses were presented to the Ohio State Veterinary Teaching Hospital with a history of poor performance and anemia following the administration of rhEPO (at least two 4000 IU doses each). A full physical examination and hematological and biochemistry profile were performed. Additional tests included urine and fecal analyses, complement fixation tests for piroplasmosis, a Coggin's test for equine infectious anemia virus, iron binding studies, a direct antiglobulin (Coombs) test, and sternebral bone marrow biopsy. Mouse bone marrow cells were cultured in the presence of the serum of the affected horses to determine whether the serum would inhibit the erythropoietin-dependent growth and differentiation of erythroid colony-forming units (CFU-E) and burst-forming units (BFU-E). Serum from a healthy horse was used as a control.

3. Results
The gelding and colt had hematocrits of 16% and 24%, serum iron concentrations of 210 µg/dl and 304 µg/dl (normal 73–140 µg/dl), and percentage iron

NOTES
saturations of 87.9% and 94.7%, respectively. Routine laboratory tests failed to reveal the cause of the anemia in each horse; plasma fibrinogen and differential white cell counts were within normal limits and tests for infectious causes of the anemia were negative. Sternebral bone marrow core biopsy and cytology indicated myeloid-to-erythroid ratios of 6.7:1 in the gelding and 3.2:1 in the colt. The cytology in each case was consistent with erythroid specific hypoplasia.

Serum from each horse, unlike serum from a healthy control horse, inhibited the rhEPO-induced activity of mouse bone marrow CFU-E and BFU-E in vitro. The degree of inhibition in each case was inversely proportional to the amount of exogenous rhEPO added to the serum, suggesting the presence of an inhibitory substance, probably anti-rhEPO antibodies in the serum of each horse.

4. Treatment and Follow-Up
Each horse was discharged from the hospital with instructions to receive oral dexamethasone (0.05 mg/kg) once daily and to have rhEPO administration discontinued. The steroid dose was to be adjusted according to clinical response. Five months following initial presentation, both horses were back in training and the gelding's hematocrit had returned to 35%.

5. Discussion
Failure to determine a cause of the anemia in each horse with routine tests led us to pursue a diagnosis using bone marrow analysis and in vitro studies of the suppressive effects of serum on the rhEPO-dependent proliferation of erythroid progenitors in culture. The histories of rhEPO administration and the presence of an inhibitory factor in the serum of each anemic horse led us to suspect that anti-rhEPO antibodies had been formed. It is likely that these antibodies cross reacted with the horses' endogenous erythropoietin, thereby causing erythroid hypoplasia. The formation of anti-rhEPO antibodies has been suggested to occur following its administration to dogs and cats\(^1\) and in a horse,\(^2\) however, to our knowledge, this is the first time that the likely presence of antibodies in horses has been substantiated by the analysis of hematological effects in vitro.

References