Safety and Pharmacokinetics of Recombinant Human Superoxide Dismutase Administered Intratracheally to Premature Neonates With Respiratory Distress Syndrome

Warren N. Rosenfeld, MD*; Jonathan M. Davis, MD*; Lance Parton, MD*; Susan E. Richter, MPA*; Anita Price, MD†; Edith Flaster, MS‡; and Nadim Kassem, MD§

ABSTRACT. Objective. As a first step in the evaluation of recombinant human CuZn superoxide dismutase (rhSOD) in the prevention of neonatal lung injury, safety and pharmacokinetics of intratracheally (IT) administered rhSOD were studied.

Methods. Twenty-six preterm infants weighing 750 to 1250 g with respiratory distress syndrome were studied in three sequential groups (placebo, 0.5, and 5 mg/kg). Placebo or rhSOD was administered IT 30 minutes after the first surfactant dose. Serial blood and urine studies, rhSOD levels, tracheal aspirate fluid (TAF) markers of acute inflammation, radiographs, and ultrasounds were performed over the 28-day study period.

Results. Serum SOD concentrations were similar at baseline for all three groups (geometric mean 0.2, upper-lower limit 0.1 to 0.2 µg/mL). In the 0.5-mg/kg group, levels were highest at 12 hours (geometric mean 0.7, upper-lower limit 0.5 to 0.8 µg/mL) and returned to baseline by day 3. In the 5-mg/kg group, levels were highest at 6 hours (geometric mean 3.0, upper-lower limit 2.3 to 4.0 µg/mL) and returned to baseline by day 4. Concentrations of SOD in TAF were also similar at baseline for all three groups (geometric mean 0.2, upper-lower limit 0.2 to 0.3 µg/mL). There were no significant increases in the placebo group, but levels in the 0.5-mg/kg group were highest when first sampled at 24 hours (geometric mean 1.1, upper-lower limit 0.8 to 1.4 µg/mL) and returned to baseline by day 3. In the 5-mg/kg group, levels were also highest when sampled at 24 hours (geometric mean 1.4, upper-lower limit 0.9 to 2.1 µg/mL) and returned to baseline by day 4. Urine levels were highest at 12 hours in both the 0.5-mg/kg (geometric mean 1.3, upper-lower limit 1.0 to 1.7 µg/mL) and 5-mg/kg infants (geometric mean 6.4, upper-lower limit 3.9 to 10.4 µg/mL) and decreased significantly by days 2 to 3. rhSOD activity assays (serum, TAF, and urine) demonstrated that the enzyme still possessed significant activity. No adverse effects of rhSOD were found. TAF neutrophil chemotactic activity and albumin concentrations, important acute lung injury markers, were significantly lower in the high-dose rhSOD group compared with the other groups.

Conclusions. Data suggest that a single IT dose of rhSOD results in significant increases in both concentrations and activity of the antioxidant in serum, TAF, and urine for 2 to 3 days. The enzyme appears to be well tolerated, and TAF inflammatory markers are reduced after administration. This has important implications in rhSOD trials to prevent acute and chronic lung injury in preterm neonates. Pediatrics 1996;97:811–817; pharmacokinetics, antioxidants, superoxide dismutase, respiratory distress syndrome.

ABBREVIATIONS. BPD, bronchopulmonary dysplasia; rhSOD, recombinant human CuZn superoxide dismutase; IT, intratracheally; TAF, tracheal aspirate fluid; RDS, respiratory distress syndrome; SOD, superoxide dismutase.

Bronchopulmonary dysplasia (BPD) is a chronic lung disease that develops in infants treated with oxygen and mechanical ventilation for a primary lung disorder. BPD has been traditionally defined as oxygen requirement with an abnormal chest radiograph at 28 days of life.12 BPD can affect 20 to 60% of premature neonates who are born each year with respiratory distress syndrome (RDS).3 The development of BPD is associated with significant mortality and morbidity, such as repeated hospitalizations and developmental handicaps.4 With the increasing survival of many premature infants, BPD is emerging as the most common sequel of neonatal intensive care.5 The etiology of BPD is poorly understood. It was initially postulated that exogenous surfactant replacement therapy (by lowering oxygen and ventilator requirements) would lessen the incidence and severity of BPD in infants with RDS. However, recent surfactant trials have demonstrated that despite an improvement in clinical course and overall survival, the incidence of BPD has not been substantially affected.3 This suggests that the pathogenesis of BPD is multifactorial and associated with various causative factors. Because the lungs of premature infants with RDS are exposed to supraphysiological oxygen concentrations during therapy, it seems logical that an oxidative insult could be an important component of the injury process. BPD has been hypothesized to begin as acute inflammatory changes secondary to toxic free oxygen radicals which then evolve into chronic lung disease.6 7

The premature neonate may be particularly vulnerable to oxidant damage because endogenous antioxidant enzyme activity may be deficient at birth.8 9 Preliminary animal and human studies have suggested that acute and chronic lung injury may be

From the Departments of *Pediatrics (Neonatology) and †Radiology and the CardioPulmonary Research Institute, Winthrop-University Hospital, SUNY Stony Brook School of Medicine, Mineola and Stony Brook, New York; and ‡Bio-Technology General Corporation, Iselin, New Jersey. Received for publication Feb 21, 1995; accepted Jul 10, 1995. Reprint requests to (W.N.R.) Department of Pediatrics, Winthrop-University Hospital, 259 First Street, Mineola, NY 11501. PEDIATRICS (ISSN 0031 4005). Copyright © 1996 by the American Academy of Pediatrics.
ameliorated by the administration of one of these antioxidants, specifically superoxide dismutase (SOD).\textsuperscript{10-13} SOD is a low molecular weight CuZn-containing protein ($M_\text{r} = 32,600$) that is present in all mammalian cells. The only known function of the enzyme is to catalyze the conversion of toxic superoxide anions (oxygen-free radicals) to potentially less toxic hydrogen peroxide and water. SOD activity has been detected in natural lung surfactants but is absent in commercial surfactant preparations.\textsuperscript{14} It is possible that augmenting antioxidant enzyme activity may minimize cell damage and inflammatory changes in the lung and prevent the development of acute and chronic injury.

As the first step in the evaluation of the use of SOD to prevent BPD, this study was designed to assess the safety and pharmacokinetics of recombinant human CuZn SOD (rhSOD) administered intratracheally (IT) to premature neonates with RDS who are at high risk for developing BPD. Pharmacokinetics of rhSOD were determined in serum, urine, and tracheal aspirate fluid (TAF) to determine whether dosages shown to be efficacious in our animal studies would result in comparable SOD concentrations in human neonates.\textsuperscript{15} Patients were also monitored for potential side effects and adverse outcomes. Markers of acute lung injury in TAF were also evaluated to determine whether IT administration of rhSOD could mitigate the inflammatory response and the development of acute lung injury.

**METHODS**

**Subjects**

Patients were enrolled in this placebo-controlled, nonblinded, dose-ranging study at two participating hospitals (Winthrop-University Hospital and University Hospital at Stony Brook) from April to December 1993. A total of 28 infants were studied in three sequential groups. The first group (n = 12) received placebo (saline) in an unblinded fashion (requested as a reference/control group by the Food and Drug Administration). Saline was used for the placebo because rhSOD is routinely diluted in saline. The second (n = 8) and third (n = 8) groups received 0.5 mg/kg and 5 mg/kg of rhSOD, respectively. As per Food and Drug Administration requirements, the groups were studied sequentially and not randomized. Patients were considered eligible if they were less than or equal to 24 hours of age, weighed 750 to 1520 g at birth, required intubation and mechanical ventilation for treatment of RDS (clinical and radiographic criteria), and had received “rescue” surfactant therapy (Surfactan) within the first 24 hours of life. Infants were eligible to receive additional dosages of surfactant if clinically indicated after rhSOD administration. Parental informed consent was obtained before study entry. The study was approved by the institutional review boards at both hospitals.

Patients were excluded from the study if evidence of congenital infection, major congenital anomalies (chromosomal, cardiac, pulmonary, and renal), or perinatal asphyxia was present.

**Drug Administration**

The rhSOD used in this study was supplied by Bio-Technology General Corporation (Belin, NJ) under Food and Drug Administration approval (IND 28,225). The rhSOD was produced in Escherichia coli by using recombinant DNA technology. The amino acid composition and sequence of the resultant product is identical to human SOD, although it lacks an NH$_2$-terminal acetyl group and is defined as an analog. rhSOD preparation has 4000 U of activity per mg of enzyme.

The two separate dosages of rhSOD were provided as 1 mg (for the 0.5-mg/kg dose group) or 10 mg (for the 5-mg/kg dose group) in 2 mL of saline. The first group of infants received IT saline (1 mL/kg, total volume), the next group of patients received 0.5 mg/kg rhSOD IT (suspended in 1 mL/kg), and the final group received 5 mg/kg (1 mL/kg) of rhSOD. Drug or placebo was administered in two IT 0.5-mL/kg aliquots over a 1- to 2-minute period 30 to 60 minutes after surfactant administration (because of potential drug-drug interactions when combining the two agents, the surfactant and rhSOD were given separately). Laboratory evaluations obtained for safety and pharmacokinetics are listed in Table 1.

**Collection of Blood and TAF**

Blood (0.5 mL) was obtained for the measurement of serum concentrations of SOD before drug administration (t = 0, from umbilical cord blood) from all 28 infants and at 6, 12, 24, 48, 72, 120, and 168 hours post-administration only in the infants receiving rhSOD. TAF was obtained by instilling 1 mL of saline into the endotracheal tube and suctioning the fluid into a leukems trap. The catheter was then rinsed with an additional 1.5 mL of saline. The first TAF was collected before surfactant administration (baseline). Initial postdrug TAF collections were conducted 24 hours after placebo or rhSOD administration to prevent premature removal of drug. Subsequent TAF collections were obtained from all infants on days 2, 5, 7, 9, 12, 14, 16, 18, 21, 24, and 28. TAF was only collected if infants continued to require intubation and mechanical ventilation. TAF was obtained immediately before any elective extubation. The TAF was then centrifuged at 8000 rpm for 2 minutes to pellet the cells. The supernatant was removed and divided into three aliquots and frozen at −70°C for SOD concentration and activity and for assays of pulmonary inflammation. The cell pellet was resuspended in 100 μL of saline then trypan blue was added and the cell count obtained by using a hemocytometer. Cell differentials were determined by using cytospin centrifugation and staining with Diff-Quik.

**Urine Sample Collection**

Each urine sample collected consisted of the total urine voided over a specified collection interval. Urine samples were collected and pooled only in infants receiving rhSOD from 1 to 12, 12 to 24, and every 12 hours after rhSOD administration for the first week of life.

**rhSOD Concentrations and Activity**

Serum, TAF, and urine samples were analyzed for total SOD (endogenous and exogenous) concentrations by radioimmunoassay by using a monoclonal antibody specific for rhSOD. The radioimmunoassay was performed by first labeling 100 μg of rhSOD with$^{125}$I by using 1 μCi of Bolton Hunter Reagent (Amersham, Arlington Heights, IL). The solution was then diluted to 15 mL with phosphate-buffered saline with 0.1% bovine serum albumin. A total of 100 μL of sample or standard (10 to 2500 μg rhSOD/L) was added to 100 μL of the$^{125}$I-labeled rhSOD solution and 100 μL of mouse anti-rhSOD antibody (Bio-Technology General Corp.). After the solution was incubated at 4°C for 12 hours, it was then incubated at 37°C for 1 hour to allow for the formation of a antibody-antigen complex. Next, 1 mL of charcoal suspension was added to the tubes. After 5 minutes of incubation, the tubes were centrifuged at 3000 rpm for 5 minutes to precipitate the antibody-antigen complex. A 100 μL aliquot of each supernatant was counted for radioactivity in a gamma counter. The supernatant concentrations were calculated by using an internal standard of tritiated bovine serum albumin.

**TABLE 1.** Data Obtained at Baseline and Throughout the Study

| Physical examination including blood pressure, respiration, and temperature (daily) |
| Chest radiograph (days 1 and 28) |
| Arterial blood gases (as clinically indicated) |
| CBC with differential, platelet count, and reticulocytes (days 1, 3, 7, 14, 21, and 28) |
| Serum Na$^+$, K$^+$, Cl$^-$, HCO$_3^-$, Ca$^{2+}$, glucose, BUN, and creatinine (days 1, 3, 7, 14, 21, and 28) |
| SCOT, alkaline phosphatase, bilirubin, total protein, and albumin (days 1, 7, 14, 21, and 28) |
| Serum rhSOD level (6, 12, and 24 hours, days 2, 3, 5, and 7) |
| Urinalysis (days 1, 3, 7, 14, 21, and 28) |

**Abbreviations:** BUN, blood urea nitrogen; SCOT, serum glutamic-oxaloacetic transaminase.
50 μL of rabbit anti-mouse whole serum (Sigma Chemical Co., St. Louis, MO; 1/40 dilution) was added. This solution was incubated at room temperature for 30 minutes and then 300 μL of 20% polyethylene glycol 8000 (Sigma Chemical Co.) was added. After the solution was incubated at room temperature for 20 minutes, centrifugation was performed at 1500 × g for 20 minutes. The supernatant was counted in an LKB 1282 CompuGamma radiodino-

Biochemical Assessments of Lung Inflammation

Neutrophils for chemotaxis were collected from an adult vol-
unteer and the assay performed by a modification of the pro-
dure of Boyum.76 After anticoagulation, agglutination, and sedi-
mementation of red blood cells, the white cell layer was washed and
centrifuged. One milliliter of neutrophils (4 × 10^7 cells/mL) was
cultured with 30 μCi of ^3H]thionin for 48 hours. The supernatant was counted in an LKB 1282 CompuGamma radiodino-

Concentrations of albumin (representing endothelial/epithelial
integrity) in TAF were measured by ELISA and elastase activity
was determined by the elastin-agar plate method and by cleavage
of a peptide substrate (described previously).77

Statistical Analyses

Concentrations of SOD and analyses of inflammatory markers
over time were examined by using analysis of variance. Log
transformation was used to better normalize the data (values
expressed as geometric means with upper and lower confidence
limits). Correlation of concentration and activity of SOD was
analyzed by using regression analyses. The incidence of various
complications was analyzed by using χ^2 and Fisher’s exact test.
Duration of ventilatory support and chest radiograph scores were
analyzed with analysis of variance and Student’s t-tests.

RESULTS

Patient Population

Twenty-eight patients were originally enrolled in the
trial. One placebo infant was excluded after de-
tection of a diaphragmatic hernia on day 2 of life (an
initial chest radiograph did not show the anomaly).
This left the placebo group with 11 patients. One
infant was excluded from the high-dose rhSOD
group when initial blood studies (unknown at the
time of admission) revealed congenital syphilis. This
left the 5-mg/kg rhSOD group with seven patients.

There were no statistical differences between the
placebo and treatment groups with respect to sex,
gestational age, birthweight, or race (Table 2). How-
ever, initial score for neonatal acute physiology18
were significantly lower in the placebo group com-
pared with the 0.5-mg/kg group (P = .02) and ap-


davored significance compared with the 5-mg/kg
group (P = .09), suggesting that the severity of ill-
ness was higher at birth in both rhSOD treatment
groups. Note that mothers of five infants in the pla-
cele group, three infants in the low-dose group, and

Pharmacokinetics

Serum, TAF, and Urine rhSOD Concentrations

Initial pretreatment concentrations of human
SOD in TAF were similar in all three groups (geo-
metric mean 0.2, upper-lower limit 0.2 to 0.3 μg/
ml; 1 SEM). Although there were no significant
increases in the placebo group (insufficient sam-
ple were available after day 1), total human SOD
concentrations (representing total endogenous hu-
man SOD and rhSOD) increased significantly in
both treatment groups (Fig 1). Levels were highest
when first sampled at 24 hours in the 0.5-mg/kg
group (geometric mean 1.1, upper-lower limit 0.8
to 1.4 μg/ml) and the 5-mg/kg group (geometric
mean 1.4, upper-lower limit 0.9 to 2.1 μg/ml).
SOD concentrations remained elevated until day 3
for the 0.5-mg/kg group and day 4 for 5-mg/kg
group. By using an exponential decay model, the
TAF elimination constant for the β-phase was 1.5
days.

Serum concentrations of SOD were also similar in
all three groups before treatment (geometric mean
0.2, upper-lower limit 0.1 to 0.2 μg/ml) (Fig 2).
Serial levels were performed only in infants who
received rhSOD. In the 0.5-mg/kg group levels were
highest at 12 hours (geometric mean 0.7, upper-lower
limit 0.5 to 0.8 μg/ml) and returned to baseline by
24 hours. In infants receiving 5-mg/kg of rhSOD
levels were highest at 6 hours (geometric mean 3.0,
upper-lower limit 2.3 to 4.0 μg/ml) and did not
return to baseline until day 3. By using an exponen-
tial decay model, the serum elimination constant
for the β-phase for the 5-mg/kg group was 3 days.

Urine was not obtained before drug or placebo
administration. Only the urine of infants receiving
rhSOD was analyzed (Fig 3). In the 0.5-mg/kg group
urine levels were highest at 12 hours (geometric
mean 1.3, upper-lower limit 1.0 to 1.7 μg/ml) and
decreased significantly by day 2. The 5-mg/kg group
had the highest urine concentrations at 12 hours
(geometric mean 6.4, upper-lower limit 3.9 to 10.4
μg/ml). Levels decreased significantly by day 3. In
the 0.5-mg/kg group a mean of 29.0 ± 7.3% of the
administered dose was eventually recovered in the

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<th>TABLE 2. Study Population</th>
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<td>Group (n)</td>
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<tr>
<td>Placebo (11)</td>
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<td>0.5 mg/kg (8)</td>
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* Values are mean ± SD.
† Values are mean ± SEM; SNAP, score for neonatal acute physiology.
‡ P < .05 compared with placebo.
§ P = .09 compared with placebo.
Fig 1. TAF SOD concentrations (geometric mean ± SEM). TAF SOD concentrations were similar in all groups before drug administration. There were no increases in the placebo group. Levels were highest in both rhSOD groups when first sampled at 24 hours. Levels remained elevated in 0.5-mg/kg group on day 3 and for the 5-mg/kg group by day 4.

urine collected on days 1 to 3, 5, and 7. Twelve percent was collected in the first 12 hours post-dose with decreasing recovery over the first week. A similar pattern was seen for the 5-mg/kg group in which 22 ± 3.5% of the administered dose was recovered over the same time period with 9.3% collected in the first 12 h.

The concentration of SOD present in TAF, serum, and urine was determined by radioimmunoassay. Several samples from each experimental group were randomly selected and analyzed for SOD activity. A regression analysis comparing concentration to activity revealed an R^2 value of 0.86.

TAF Markers of Acute Inflammation

The migration of normal adult human neutrophils to any chemotactic stimulus present in TAF was analyzed by using the neutrophil chemotaxis assay. Activity was similar for all three groups at baseline. Percentage of migration was significantly less in the high-dose group compared with the low-dose group over the first week of life (P = .02) (Fig 4). No comparison to the placebo group could be made because most infants from this group were extubated by day 2. Albumin concentrations were similar in all three groups at baseline. The levels from both rh-SOD-treated groups decreased significantly over the first week of life (P < .05 both groups) (Fig 5). Total cell counts and elastase activity were similar in all three groups at baseline and did not change significantly over the first week of life. Data were analyzed over the first week of life because most infants from all three groups (especially the placebo group) were weaned from mechanical ventilation by 2 to 4 days of

Fig 2. Serum SOD concentrations (geometric mean ± SEM). Serum concentrations of SOD were similar in all groups before drug administration. In the 0.5-mg/kg group, levels were highest at 12 hours and returned to baseline by 24 hours. In the 0.5-mg/kg group, levels were highest at 6 hours and returned to baseline by day 3.
Fig. 3. Urine SOD concentrations (geometric mean ± SEM). Urine concentrations of SOD were highest at 12 hours. Levels decreased significantly by day 2 in the 0.5-mg/kg group and by day 3 in the 5-mg/kg group.

age and relatively little data were available at later time points for statistical analyses.

Clinical Outcome Variables

No specific side effects of rhSOD treatment were noted. One infant died in the placebo group at 2 hours of life secondary to a liver hemorrhage. An infant in the 0.5-mg/kg group developed necrotizing enterocolitis on day 12 of life and died on day 29, 1 day after the study period ended.

BPD was defined as oxygen dependency (greater than or equal to 25% inspired oxygen or 1/4 L/minute nasal cannulae for at least 21 of the first 28 days of life) at 28 days of life and an abnormal chest radiograph (ie, Edward’s score greater than or equal to 4).10 None of the placebo patients, one patient in the low dose, and two patients in the high-dose rhSOD group developed BPD. None of the infants in the trial had an Edward’s radiographic score of more than 4.

Two infants in each group had retinopathy of prematurity diagnosed at 4 to 6 weeks of life (none greater than stage 2). One infant in the placebo group, one infant in the low-dose group, and one infant in the high-dose rhSOD group developed a clinically significant (greater than grade II) intraventricular hemorrhage. Four infants in the placebo group, five in the low-dose group, and three in the high-dose rhSOD group developed clinical and echocardiographic evidence of patent ductus arteriosus. All cases responded to indomethacin, except for one placebo infant who required surgical ligation.

There were no significant differences between the three groups with regard to total days in oxygen and duration of mechanical ventilatory support (ventilation and continuous positive airway pressure). When all laboratory parameters were analyzed, only one difference was found. The number of infants with elevated potassium (greater than 6.5 mEq/L) was significantly increased in the placebo group (P < .01) compared with both rhSOD treatment groups.

DISCUSSION

Clinical and laboratory evidence continues to accumulate, implicating oxygen free radicals in the pathogenesis of acute and chronic lung injury.22,23 The introduction of surfactant replacement therapy in premature infants with RDS has resulted in a reduction in the overall severity of BPD, but the incidence has not been substantially affected.3 In fact, the improvement in survival of many very low birth weight infants has actually increased the prevalence of BPD.5 Strategies to prevent BPD have been limited and have generally been unsuccessful. Vitamin E supplementation was not effective in the past22,23 and the current emphasis has centered on the use of corticosteroids.24-26 The results of a number of clinical trials have been inconclusive, especially in view of the significant variability in study populations, dosing strategies, and duration of treatment. None of the studies were able to demonstrate a clear and unequivocal improvement in outcome (ie, survival) with the use of corticosteroids. In addition, corticosteroids are known to possess many significant side effects in preterm infants.27 Clearly, safe and effective alternatives need to be developed that will specifically target the lung injury process in infants with RDS. An interest in antioxidants has increased recently with the demonstration of significant SOD activity associated with natural surfactants, which is absent in commercial preparations.34

SOD has been used to prevent lung injury in several animal models and in human neonates. Rosenfeld et al35 had previously shown that subcutaneous administration of bovine SOD to premature infants with RDS (before the introduction of surfactant replacement therapy) increased serum levels of SOD and decreased the clinical and radiologic severity of BPD. However, other studies using systemic bovine SOD were discontinued after the development of recombinant human SOD, which offered distinct advantages such as purity and hypoallergenicity. In addition, studies of newborn piglets treated with 48 hours of hyperoxia and mechanical ventilation demonstrated that IT administration strategies were more efficacious and had fewer side effects compared with systemic administration.10,28 Davis et al10 demonstrated that IT administration of 5 mg/kg of rhSOD resulted in prolonged serum, TAF, and lung tissue concentrations and significantly reduced markers of acute inflammation in TAF. This provided the impetus for this study because the early inflammatory changes in the TAF of preterm neonates appears to be a major determinant in the pathogenesis of BPD.67

This study demonstrated that rhSOD can be given by the IT route with significant increases in serum, TAF, and urine SOD activity achieved with dosages of 0.5 mg/kg and 5 mg/kg for 2 to 4 days. Serum and TAF concentrations observed in newborns were comparable to those previously seen in the piglet studies with these dosages.10 In addition, the piglet studies revealed that TAF concentrations of rhSOD appeared to correlate well with bronchoalveolar lavage and lung tissue concentrations. Initial concerns
Fig 4. Chemotaxis (TAF) concentrations (mean ± SEM). Chemotactic activity TAF to normal adult neutrophils was measured by % migration and was similar in all three groups before rhSOD administration. Percentage of migration was significantly less in the 5-mg/kg group compared with the 0.5-mg/kg group ($P = .02$).

over the short half-life of systemically administered SOD, reported in adults to be 4 to 8 minutes, was not apparent in the premature neonate. It appears that the pulmonary clearance or metabolism of IT rhSOD is significantly delayed in the premature neonate. This may be due in part to recent findings in neonatal piglets that IT-administered rhSOD is rapidly taken up by various cell types in the lung (airway and alveolar) and localizes intracellularly within 30 minutes of administration.29

No direct toxicity of rhSOD was found. However, potential toxicities that may have had a low incidence may not have been seen due to the small numbers of patients studied. Animal studies had demonstrated renal dysfunction when large dosages of intravenous rhSOD (50 mg/kg) were administered as a bolus.28 No abnormalities of renal function were detected in serum or urine of any rhSOD-treated infant. In fact, creatinine clearance in the 5-mg/kg rhSOD group exceeded normal values established for very low birthweight infants.30

This study, although controlled, did not permit determination of efficacy. The number of patients in each group was small and control and treatment groups were not randomly selected. Single dosages of rhSOD may not be sufficient to protect the lung

Fig 5. Albumin (TAF) concentrations (mean ± SEM). TAF albumin was similar in all groups before rhSOD administration. Levels for both rhSOD groups decreased significantly over the first week of life ($P < .05$ for both groups).
from prolonged exposure to oxygen and mechanical ventilation, even though acute inflammatory markers associated with BPD were significantly diminished in the 5-mg/kg group. Future studies with multiple dosing regimens with a much larger number of patients will be required before efficacy can be determined. In addition, rhSOD may need to be encapsulated in liposomes and administered IT to enhance cellular uptake and maximize efficacy.34 This approach is similar to the evaluation of surfactant replacement therapy in neonatal RDS. Single dosages of exogenous surfactant resulted in significant physiologic improvements but no differences in outcome.35 Significant improvements in outcome were only found after multiple dosages of exogenous surfactant were used. It is interesting that significant hyoperkalemia did not occur in any of the rhSOD-treated infants. It could be hypothesized that the rhSOD may have prevented cellular damage, resulting in the release of potassium. It may also be that renal excretion of rhSOD had a positive diuretic or other effect on the premature kidney, enabling greater excretion of potassium to occur.

BPD has become the major sequela of neonatal intensive care. Its cost in terms of mortality, morbidity, and the expenditure of health care dollars is great. Previous interventions for the prevention of BPD have not proven to be efficacious. A new direction in treatment, augmentation of antioxidant defenses, needs to be studied further. This preliminary phase I study provides the basis for these additional investigations. The ability to increase antioxidant activity may also be important in the treatment of other disease processes in premature infants where free radical injury has also been implicated, such as intraventricular hemorrhage.32 Prophylactic treatment with antioxidants may also help reduce the incidence of this significant neonatal problem.

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