

ORIGINAL ARTICLE

Early Inhaled Nitric Oxide Therapy in Premature Newborns with Respiratory Failure

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ABSTRACT

BACKGROUND

The safety and efficacy of early, low-dose, prolonged therapy with inhaled nitric oxide in premature newborns with respiratory failure are uncertain.

METHODS

We performed a multicenter, randomized trial involving 793 newborns who were 34 weeks of gestational age or less and had respiratory failure requiring mechanical ventilation. Newborns were randomly assigned to receive either inhaled nitric oxide (5 ppm) or placebo gas for 21 days or until extubation, with stratification according to birth weight (500 to 749 g, 750 to 999 g, or 1000 to 1250 g). The primary efficacy outcome was a composite of death or bronchopulmonary dysplasia at 36 weeks of postmenstrual age. Secondary safety outcomes included severe intracranial hemorrhage, periventricular leukomalacia, and ventriculomegaly.

RESULTS

Overall, there was no significant difference in the incidence of death or bronchopulmonary dysplasia between patients receiving inhaled nitric oxide and those receiving placebo (71.6 percent vs. 75.3 percent, $P=0.24$). However, for infants with a birth weight between 1000 and 1250 g, as compared with placebo, inhaled nitric oxide therapy reduced the incidence of bronchopulmonary dysplasia (29.8 percent vs. 59.6 percent); for the cohort overall, such treatment reduced the combined end point of intracranial hemorrhage, periventricular leukomalacia, or ventriculomegaly (17.5 percent vs. 23.9 percent, $P=0.03$) and of periventricular leukomalacia alone (5.2 percent vs. 9.0 percent, $P=0.048$). Inhaled nitric oxide therapy did not increase the incidence of pulmonary hemorrhage or other adverse events.

CONCLUSIONS

Among premature newborns with respiratory failure, low-dose inhaled nitric oxide did not reduce the overall incidence of bronchopulmonary dysplasia, except among infants with a birth weight of at least 1000 g, but it did reduce the overall risk of brain injury. (ClinicalTrials.gov number, NCT00006401.)

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EARLY REPORTS OF INHALED NITRIC OXide therapy showed acute and sustained improvement in oxygenation in term newborns with persistent pulmonary hypertension.^{1,2} Subsequently, randomized, controlled trials of inhaled nitric oxide in term newborns with persistent pulmonary hypertension confirmed that this selective pulmonary vasodilator improves oxygenation and reduces the need for extracorporeal membrane oxygenation.^{3,4} Previous studies suggested that inhaled nitric oxide can improve gas exchange in premature newborns with hypoxemia caused by the respiratory distress syndrome or persistent pulmonary hypertension.⁵⁻⁸ Although the Food and Drug Administration (FDA) approved inhaled nitric oxide for use in term newborns, the safety and efficacy of such therapy in premature newborns with respiratory failure remain unproven.

In addition to its effects on gas exchange and pulmonary hypertension, in several laboratory studies, inhaled nitric oxide decreased early lung inflammation and oxidant stress,⁹⁻¹¹ maintained surfactant activity,¹² and improved lung structure in diverse models of chronic lung disease, known as bronchopulmonary dysplasia.¹³⁻¹⁵ These findings suggest that inhaled nitric oxide may potentially reduce early lung injury leading to bronchopulmonary dysplasia.¹⁶ Whether early treatment with inhaled nitric oxide can attenuate the risk or severity of bronchopulmonary dysplasia in premature infants remains unclear.

Laboratory and clinical studies have suggested that high doses of inhaled nitric oxide may increase the risk of bleeding,^{17,18} a paramount concern because of the predisposition of premature newborns to intracranial hemorrhage. Although an early multicenter, randomized, controlled trial of low-dose inhaled nitric oxide in premature newborns with severe hypoxemic respiratory failure showed no increased risk of intracranial hemorrhage,⁸ two recent studies reported contradictory findings regarding the safety and efficacy of such treatment.^{19,20} One trial showed a decrease in the combined outcome of death or bronchopulmonary dysplasia and severe intracranial hemorrhage or periventricular leukomalacia after treatment with inhaled nitric oxide.¹⁹ However, another trial found that inhaled nitric oxide reduced the risk of bronchopulmonary dysplasia only in infants with a birth weight of 1000 g or more and increased the risk of severe intracranial

hemorrhage or periventricular leukomalacia in infants with a birth weight below 1000 g.²⁰

We hypothesized that prolonged treatment with low-dose (5 ppm) inhaled nitric oxide would reduce the combined end point of death and bronchopulmonary dysplasia in premature newborns, without increasing the incidence or progression of severe intracranial hemorrhage or periventricular leukomalacia. We also hypothesized that the effect of inhaled nitric oxide therapy may be dependent on the degree of prematurity. We performed a multicenter trial that randomly assigned premature newborns with respiratory distress after stratification by birth weight within the clinical center either to treatment with inhaled nitric oxide or to placebo.

METHODS

ORGANIZATION OF THE STUDY AND ELIGIBILITY CRITERIA

We conducted the study at 16 centers with clinical experience in inhaled nitric oxide therapy and at a coordinating center. The study was approved by institutional review boards at each institution and by the FDA under an investigational new drug exemption. Criteria for enrollment included a gestational age of 34 weeks or less, birth within the previous 48 hours, respiratory failure requiring endotracheal intubation and mechanical ventilation, and a birth weight of 500 to 1250 g. Exclusion criteria were lethal congenital anomalies or congenital heart disease (including an atrial septal defect larger than 1 cm and a ventricular septal defect larger than 2 mm), active pulmonary hemorrhage, unevacuated pneumothorax, or an expected duration of ventilation of less than 48 hours. Infants were enrolled after written informed consent was obtained from a parent or guardian.

STUDY DESIGN

Randomization was stratified according to center and birth weight (500 to 749 g, 750 to 999 g, and 1000 to 1250 g), balanced in blocks of two or four within strata on the basis of a planned enrollment of 792 patients. Randomization numbers were linked to masked cylinders of inhaled nitric oxide or placebo study gas identified only by sequence numbers.

After randomization, the ventilator circuit was configured to allow delivery of inhaled nitric oxide at 5 ppm or nitrogen placebo through a shield-

ed INOvent device (INO Therapeutics). This shielding allowed visualization of the set dose of nitric oxide but not the readout of the nitric oxide or nitrogen dioxide analyzers. Study gas was delivered for 21 days or until extubation.

Infants underwent mechanical ventilation with either standard neonatal time-cycled, pressure-limited ventilators (Sensormedics 3100A High Frequency Oscillator, Sensormedics) or high-frequency devices (Infant Star HFV, Infrasonics). The only ventilator prohibited was the Life Pulse High Frequency Ventilator (Bunnell), because of limited information on the accuracy of delivered inhaled nitric oxide concentrations. Ventilation strategies and management were left to the discretion of the neonatologist.

The primary outcome measure was the combined end point of death or bronchopulmonary dysplasia. Bronchopulmonary dysplasia was defined as the need for supplemental oxygen or mechanical ventilation at 36 weeks of postmenstrual age, plus abnormal findings on chest radiography, assessed by a centrally read standardized scoring system.²¹ Secondary outcomes included grade 3 or 4 intracranial hemorrhage, periventricular leukomalacia, and ventriculomegaly.

CRANIAL ULTRASONOGRAPHY STUDIES

Cranial ultrasonography was performed before enrollment (to determine the baseline incidence and severity of intracranial hemorrhage),²² at 7 to 14 days of age, and at more than 30 days of age (to determine the progression of intracranial hemorrhage with treatment and to identify new hemorrhages and periventricular leukomalacia). A diagnosis of incident intracranial hemorrhage in an infant with hemorrhage at baseline required a worsening of the condition over the baseline level. Intracranial hemorrhage was classified according to the most severe grade occurring after baseline, and ultrasonography readings were masked to assignment.

STATISTICAL ANALYSIS

The planned enrollment of 792 infants was based on an estimated reduction of 10 percentage points in the combined end point of death or bronchopulmonary dysplasia in the group that received inhaled nitric oxide, for an incidence of 50 percent, as compared with 60 percent in the placebo group, given a statistical power of 80 percent, with a two-sided alpha of 0.05. Critical values were prespeci-

fied for two interim analyses ($P=0.001$), maintaining the overall type I error for the final data analysis at 0.048. Safety analyses were conducted by the data and safety monitoring board at semi-annual meetings and through routine monitoring of all serious adverse events. Significance levels for these analyses were not prespecified, and unadjusted P values are reported for these analyses.

Detailed safety analyses were conducted after 284 infants had been enrolled. At the time of this interim safety analysis, a favorable effect on grade 3 or 4 intracranial hemorrhage was detected in the group that received inhaled nitric oxide. Subsequently, this protective effect was monitored closely. After the Neonatal Network trial of inhaled nitric oxide was prematurely terminated in September 2003, the data and safety monitoring board and National Heart, Lung, and Blood Institute reviewed the safety data and did not recommend stopping the present trial.

Binomial data were analyzed with the use of the chi-square test or Fisher's exact test, where appropriate. Continuous data were compared with the use of Student's t -test or the Wilcoxon test for data that were not normally distributed. Analyses controlling for birth-weight strata, site, and other covariates were conducted with the use of generalized estimating equations based on the ProcGenmod procedure (SAS). The analysis plan adjusted for study site and randomization strata with the use of the Cochran-Mantel-Haenszel test (augmented with more powerful statistical approaches as confirmatory analyses). Generalized estimating equations were used to provide parametric-model adjustment for these design effects. A P value of less than 0.05 was considered to indicate statistical significance for all analyses except for the primary outcome, for which a P value of 0.048 was used to accommodate interim monitoring.

INO Therapeutics provided the study gas, site monitoring after forms were faxed to the coordinating center, and INOvent devices for the trial. The company remained unaware of the results and was not involved in the design of the study, data analysis or interpretation, or the preparation of the manuscript.

RESULTS

Between March 31, 2001, and June 17, 2005, 793 newborns underwent randomization: 398 to receive

inhaled nitric oxide and 395 to receive placebo; 384 weighed 500 to 749 g, 280 weighed 750 to 999 g, and 129 weighed 1000 to 1250 g (Fig. 1). Of the 4562 newborns who were prospectively screened for this study, 1921 met all eligibility criteria, and 793 were enrolled. The most common reasons for ineligibility included categories of “not intubated” or “expected early extubation” (i.e., within 48 hours). The most common reason for failure to enroll eligible infants was parental refusal (43 percent), followed by failure to obtain consent (20 percent), usually because of logistical issues.

There were no significant differences between groups in birth weight, gestational age, sex, race or ethnic group, the use of antenatal corticosteroids, or Apgar scores (Table 1). More infants in the placebo group were delivered by cesarean section ($P=0.03$), but there were no significant differences in the rates of chorioamnionitis, pre-eclampsia, or placental hemorrhage. There were also no significant differences at baseline in ventilator requirements, levels of arterial blood gases, the use of surfactants, or the frequency or severity of intracranial hemorrhage. The median duration of drug treatment was 14 days (range,

0 to 24) in the group that received inhaled nitric oxide and 12 days (range, 0 to 22) in the placebo group.

For the overall study population, the combined end point of death or bronchopulmonary dysplasia did not differ significantly between the study groups (71.6 percent in the group receiving inhaled nitric oxide and 75.3 percent in the placebo group, $P=0.24$) (Table 2). However, there was a significant interaction ($P<0.001$) between birth-weight strata and treatment. Prespecified subgroup analyses according to birth-weight strata showed significant reductions in the combined primary outcome ($P=0.004$) and in bronchopulmonary dysplasia alone ($P=0.001$) with inhaled nitric oxide therapy in the group with a birth weight of 1000 to 1250 g; death rates did not significantly differ between the group receiving inhaled nitric oxide and the placebo group in this birth-weight stratum (Table 2). There were no significant differences in the incidence of the combined end point of death or bronchopulmonary dysplasia between groups in the other birth-weight strata.

Infants who received low-dose inhaled nitric oxide had a lower incidence of periventricular

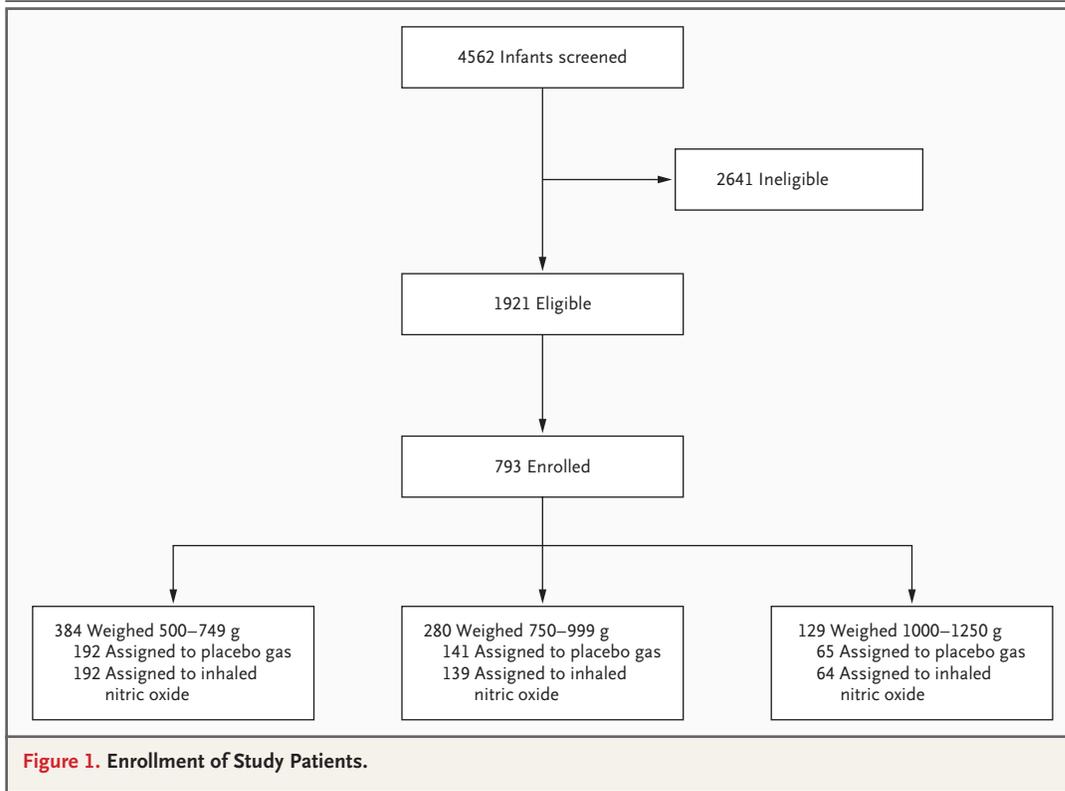


Table 1. Baseline Characteristics of Patients.*

Characteristic	Inhaled Nitric Oxide (N=398)	Placebo (N=395)	P Value
Birth weight — g	796±190	788±185	0.54
Birth-weight strata			
500–749 g	642±76	639±71	0.66
750–999 g	851±71	843±71	0.36
1000–1250 g	1129±68	1113±77	0.21
Gestational age — wk	25.6±1.7	25.6±1.8	0.86
Male sex — no. (%)	211 (53.0)	216 (54.7)	0.64
Mother's race or ethnic group — no./total no. (%)†			0.77
White	249/397 (62.7)	234/394 (59.4)	
Black	94/397 (23.7)	98/394 (24.9)	
Hispanic	41/397 (10.3)	48/394 (12.2)	
Other	13/397 (3.3)	14/394 (3.6)	
Inborn — no./total no. (%)	296/397 (74.6)	299/395 (75.7)	0.71
Antenatal corticosteroids — no./total no. (%)	310/395 (78.5)	290/394 (73.6)	0.11
Apgar score — median (interquartile range)			
At 1 min	4 (0–9)	4 (0–9)	0.71
At 5 min	7 (0–9)	7 (1–10)	0.24
Maternal complications — no./total no. (%)			
Cesarean section	248/398 (62.3)	276/395 (69.9)	0.02
Chorioamnionitis	76/397 (19.1)	58/394 (14.7)	0.07
Preeclampsia	65/397 (16.4)	64/394 (16.2)	0.95
Multiple gestation	96/397 (24.2)	107/394 (27.2)	0.34
Diabetes‡	24/397 (6.0)	15/394 (3.8)	0.16
Antepartum hemorrhage	64/397 (16.1)	58/394 (14.7)	0.61

leukomalacia than did those receiving placebo (5.2 percent vs. 9.0 percent, $P=0.048$) (Table 3). No significant interaction with birth-weight strata was found for periventricular leukomalacia in the overall population. However, there was a significant interaction between birth-weight strata and the combined outcome of severe intracranial hemorrhage or periventricular leukomalacia ($P=0.04$), with the largest reduction in this outcome with inhaled nitric oxide therapy in the stratum of infants weighing 750 to 999 g ($P=0.006$). Overall, ventriculomegaly was observed in 5.2 percent of infants in the group that received inhaled nitric oxide and in 8.9 percent of those in the placebo group ($P=0.05$). However, the interaction with birth-weight strata was not significant in the overall population.

We also performed post hoc analyses to examine the effect of race or ethnic group on the

combined outcome of death, intracranial hemorrhage, or periventricular leukomalacia and found no significant interaction between race and outcomes after treatment with inhaled nitric oxide ($P=0.99$). Although nonwhite infants had slightly higher rates of the combined outcome, the effects of inhaled nitric oxide on this combined outcome appeared to be similar in whites and nonwhites (data not shown).

During the study, there were no significant differences between the groups in the rates of serious adverse events, including air leak, pulmonary hemorrhage, the need for medical or surgical treatment of patent ductus arteriosus, necrotizing enterocolitis, threshold retinopathy of prematurity, or sepsis. There were also no significant differences between groups in the rates of postnatal use of corticosteroids, treatment with respiratory medications at 36 weeks of postmen-

Table 1. (Continued.)

Characteristic	Inhaled Nitric Oxide (N=398)	Placebo (N=395)	P Value
Age at randomization — hr	30.5±13.4	30.1±13.2	0.65
Oxygenation index [§]	5.4±5.2	5.8±6.7	0.30
FiO ₂	0.4±0.2	0.4±0.2	0.82
Arterial blood gas			
PaO ₂ — mm Hg	63.9±25.6	64.3±29.7	0.81
PaCO ₂ — mm Hg	47.6±13.2	47.4±10.6	0.77
pH	7.3±0.1	7.3±0.1	0.79
Surfactant before randomization — no./total no. (%)	319/398 (80.1)	304/395 (77.0)	0.27
Type of ventilator — no./total no. (%)			0.93
Conventional	280/393 (71.2)	276/389 (71.0)	
High-frequency	113/393 (28.8)	113/389 (29.0)	
Pulmonary hemorrhage before randomization — no.	1	0	
Intracranial hemorrhage — no./total no. (%)			
None	296/392 (75.5)	280/392 (71.4)	0.41
Grade 1 or 2	72/392 (18.4)	86/392 (21.9)	
Grade 3 or 4	24/392 (6.1)	26/392 (6.6)	

* Plus-minus values are means ±SD. Percentages may not total 100 because of rounding. FiO₂ denotes the fraction of inspired oxygen, PaO₂ the partial pressure of arterial oxygen, and PaCO₂ the partial pressure of arterial carbon dioxide.

† Race or ethnic group was self-reported by a parent or guardian.

‡ Diabetes includes both preexisting and gestational diabetes mellitus.

§ The oxygenation index equals the mean airway pressure multiplied by the FiO₂ multiplied by 100, with the result divided by the PaO₂.

strual age (Table 4), or in the duration of ventilator therapy or hospitalization. A transient elevation of methemoglobin occurred in two infants in the group receiving inhaled nitric oxide and resolved without specific therapy.

DISCUSSION

In this multicenter, randomized, controlled trial of inhaled nitric oxide therapy in premature newborns, we found that low-dose (5 ppm) inhaled nitric oxide therapy, administered within 48 hours after birth for a median of 14 days, did not decrease the overall risk of a composite end point of death or bronchopulmonary dysplasia in infants with birth weights between 500 and 1250 g who required mechanical ventilation. However, inhaled nitric oxide therapy significantly decreased the risk of bronchopulmonary dysplasia and the combined end points by 50 percent and 40 percent, respectively, in premature newborns with birth weights of 1000 to 1250 g. Nitric oxide therapy

was associated with a decreased risk of brain injury in the overall population. The risk of serious adverse events was not increased with inhaled nitric oxide therapy in the population overall or within any of the birth-weight strata. These findings suggest that early treatment with low-dose inhaled nitric oxide therapy can safely and effectively improve neurologic and respiratory outcomes in some premature infants.

During the past decade, debate over the safety of inhaled nitric oxide therapy in premature newborns has focused on concern that such treatment might increase the risk of intracranial hemorrhage, predominantly because of the potential adverse effects on platelet adhesion. Our rationale for the use of low-dose inhaled nitric oxide was based on the results of previous laboratory and clinical studies suggesting that this dose could optimize the beneficial vasoactive and antiinflammatory effects while it reduced potential adverse effects on platelet adhesion. Severe intracranial hemorrhage, periventricular leukomalacia, and

Table 2. Incidence of Death or Bronchopulmonary Dysplasia at 36 Weeks of Postmenstrual Age.

Variable	Inhaled Nitric Oxide (N=398)	Placebo (N=395)	P Value	Relative Risk (95% CI)*
	no./total no. (%)			
All patients				
Death	78/394 (19.8)	98/392 (25.0)	0.08	0.79 (0.61–1.03)
Bronchopulmonary dysplasia	212/326 (65.0)	210/309 (68.0)	0.43	0.96 (0.86–1.09)
Death or bronchopulmonary dysplasia	282/394 (71.6)	295/392 (75.3)	0.24	0.95 (0.87–1.03)
Birth weight of 500–749 g				
Death	55/191 (28.8)	66/189 (34.9)	0.20	0.82 (0.61–1.11)
Bronchopulmonary dysplasia	113/144 (78.5)	100/132 (75.8)	0.59	1.04 (0.91–1.18)
Death or bronchopulmonary dysplasia	162/191 (84.8)	159/189 (84.1)	0.85	1.01 (0.92–1.10)
Birth weight of 750–999 g				
Death	15/138 (10.9)	24/139 (17.3)	0.13	0.63 (0.35–1.15)
Bronchopulmonary dysplasia	82/125 (65.6)	76/120 (63.3)	0.71	1.04 (0.86–1.25)
Death or bronchopulmonary dysplasia	95/138 (68.8)	95/139 (68.3)	0.93	1.01 (0.86–1.18)
Birth weight of 1000–1250 g				
Death	8/65 (12.3)	8/64 (12.5)	0.97	0.98 (0.39–2.46)
Bronchopulmonary dysplasia	17/57 (29.8)	34/57 (59.6)	0.001	0.50 (0.32–0.79)
Death or bronchopulmonary dysplasia	25/65 (38.5)	41/64 (64.1)	0.004	0.60 (0.42–0.86)

* CI denotes confidence interval.

ventriculomegaly are among the most serious consequences of prematurity and the most important predictors of late neurodevelopmental sequelae in premature newborns.²³ We found that early, low-dose inhaled nitric oxide decreased the incidence of these abnormalities in the central nervous system, consistent with observations from our pilot trial⁸ and results of a previous single-center trial by Schreiber et al.¹⁹ In contrast to these findings, Van Meurs et al.²⁰ reported that inhaled nitric oxide increased the risk of severe intracranial hemorrhage or periventricular leukomalacia in premature newborns with severe respiratory failure and a birth weight below 1000 g. This conclusion, however, was based on the interpretation of a single cranial ultrasonographic examination at 28 days of age, among a sample smaller than originally planned because of early termination of the study.

Intracranial hemorrhage generally occurs within the first seven days of life, making it difficult to quantify the incidence of hemorrhage that is potentially attributable to inhaled nitric oxide with the use of late cranial ultrasonography studies alone. Indeed, the study by Ballard et al.,²⁴

which appears elsewhere in this issue of the *Journal*, enrolled patients after the first week of life, and was unable to draw any conclusions about the neuroprotective effect of nitric oxide in the study population. We evaluated the rates of intracranial hemorrhage at baseline, at 7 to 14 days, and at 30 days or later. If we had relied solely on ultrasonography performed at 30 days or later, these results would have suggested an even larger benefit of inhaled nitric oxide (a 38 percent reduction in intracranial hemorrhage). In contrast to our use of 5 ppm of inhaled nitric oxide, Van Meurs et al. treated many infants with 10 ppm of inhaled nitric oxide. It is possible that inhaled nitric oxide in premature newborns acts within a narrow therapeutic range, particularly in the infants with severe respiratory failure whose condition is the least stable.

The precise mechanisms by which inhaled nitric oxide might improve lung function are uncertain. The ability of low-dose inhaled nitric oxide to reduce early neutrophil accumulation in the lung in patients with the acute respiratory distress syndrome may be important, since neutrophils play an important role in the inflamma-

Table 3. Incidence of Primary Outcomes According to Cranial Ultrasonography.*

Variable	Inhaled Nitric Oxide	Placebo	P Value	Relative Risk (95% CI)
	no./total no. (%)			
All patients				
Grade 3 or 4 ICH, PVL, or ventriculomegaly	64/366 (17.5)	87/364 (23.9)	0.03	0.73 (0.55–0.98)
Grade 3 or 4 ICH or PVL	61/372 (16.4)	80/366 (21.9)	0.06	0.75 (0.56–1.02)
Grade 3 or 4 ICH	49/398 (12.3)	63/394 (16.0)	0.14	0.77 (0.54–1.09)
PVL	19/365 (5.2)	32/356 (9.0)	0.048	0.58 (0.33–1.00)
Ventriculomegaly	19/364 (5.2)	32/359 (8.9)	0.05	0.58 (0.37–1.01)
Death or grade 3 or 4 ICH	112/394 (28.4)	140/392 (35.7)	0.03	0.80 (0.65–0.98)
Death, grade 3 or 4 ICH, or PVL	120/392 (30.6)	151/391 (38.6)	0.02	0.79 (0.65–0.96)
Birth weight of 500–749 g				
Grade 3 or 4 ICH, PVL, or ventriculomegaly	37/177 (20.9)	38/168 (22.6)	0.70	0.92 (0.62–1.38)
Grade 3 or 4 ICH or PVL	34/179 (19.0)	35/170 (20.6)	0.71	0.92 (0.60–1.41)
Grade 3 or 4 ICH	29/192 (15.1)	27/191 (14.1)	0.79	1.07 (0.66–1.73)
PVL	10/175 (5.7)	14/165 (8.5)	0.32	0.67 (0.31–1.47)
Ventriculomegaly	11/173 (6.4)	13/166 (7.8)	0.60	0.81 (0.37–1.76)
Death or grade 3 or 4 ICH	75/191 (39.3)	83/189 (43.9)	0.36	0.89 (0.70–1.14)
Death, grade 3 or 4 ICH, or PVL	77/191 (40.3)	89/189 (47.1)	0.18	0.86 (0.68–1.08)
Birth weight of 750–999 g				
Grade 3 or 4 ICH, PVL, or ventriculomegaly	17/131 (13.0)	36/137 (26.3)	0.006	0.49 (0.29–0.83)
Grade 3 or 4 ICH or PVL	17/129 (13.0)	36/135 (26.7)	0.006	0.49 (0.29–0.83)
Grade 3 or 4 ICH	13/141 (9.2)	27/139 (19.4)	0.02	0.47 (0.26–0.88)
PVL	5/130 (3.8)	14/133 (10.5)	0.04	0.37 (0.14–0.99)
Ventriculomegaly	3/131 (2.3)	12/133 (9.0)	0.02	0.25 (0.07–0.88)
Death or grade 3 or 4 ICH	25/138 (18.1)	42/139 (30.2)	0.02	0.60 (0.39–0.93)
Death, grade 3 or 4 ICH, or PVL	29/137 (21.2)	47/139 (33.8)	0.02	0.63 (0.42–0.93)
Birth weight of 1000–1250 g				
Grade 3 or 4 ICH, PVL, or ventriculomegaly	10/60 (16.7)	13/61 (21.3)	0.52	0.78 (0.37–1.64)
Grade 3 or 4 ICH or PVL	10/62 (16.1)	9/59 (15.3)	0.86	1.07 (0.47–2.46)
Grade 3 or 4 ICH	7/65 (10.8)	9/64 (14.1)	0.57	0.77 (0.30–1.93)
PVL	4/60 (6.7)	4/58 (6.9)	0.96	0.97 (0.25–3.68)
Ventriculomegaly	5/60 (8.3)	7/60 (11.7)	0.54	0.71 (0.24–2.13)
Death or grade 3 or 4 ICH	12/65 (18.5)	15/64 (23.4)	0.49	0.79 (0.40–1.55)
Death, grade 3 or 4 ICH, or PVL	14/64 (21.9)	15/63 (23.8)	0.80	0.92 (0.48–1.74)

* CI denotes confidence interval, ICH intracranial hemorrhage, and PVL periventricular leukomalacia. ICH was defined by the maximum grade at baseline, at follow-up on days 7 to 14, and at 30 days by ultrasonography; ventriculomegaly and PVL were defined by ultrasonography at 30 days.

tory cascade, contributing to lung injury and the evolution of bronchopulmonary dysplasia.^{25–30} In addition, laboratory studies suggest that inhaled nitric oxide therapy modulates lung injury through other means, including the down-regulation of oxidant stress, the production of inflammatory cytokines, and the induction of apoptosis.

We found that low-dose inhaled nitric oxide reduced the incidence of bronchopulmonary dysplasia in infants with a birth weight of 1000 g or more. However, there was no significant reduction in the incidence of bronchopulmonary dysplasia among infants with lower birth weights. The study by Van Meurs et al. also suggested that

Table 4. Incidence of Secondary Outcomes.

Variable	Inhaled Nitric Oxide (N = 398)	Placebo (N = 395)	P Value
	no./total no. (%)		
Air leak	25/398 (6.3)	24/395 (6.1)	0.94
Pulmonary hemorrhage	24/398 (6.0)	26/395 (6.6)	0.75
Symptomatic patent ductus arteriosus			
Medical treatment	215/398 (54.0)	212/395 (53.7)	0.92
Surgical ligation	86/398 (21.6)	86/395 (21.8)	0.96
Necrotizing enterocolitis	53/379 (14.0)	46/369 (12.5)	0.54
Threshold retinopathy*	66/398 (16.6)	60/395 (15.2)	0.59
Postnatal corticosteroids	222/369 (60.2)	204/365 (55.9)	0.24
Sepsis	139/381 (36.5)	118/369 (32.0)	0.19
Medications at 36 wk			
Bronchodilators	62/309 (20.1)	60/298 (20.1)	0.98
Corticosteroids	47/308 (15.3)	37/298 (12.4)	0.31
Diuretics	113/309 (36.6)	113/298 (37.9)	0.73
Medications among survivors			
Bronchodilators	56/298 (18.8)	55/283 (19.4)	0.84
Corticosteroids	43/298 (14.4)	32/283 (11.3)	0.26
Diuretics	105/298 (35.2)	104/283 (36.7)	0.70

* Threshold retinopathy of prematurity was defined as a condition requiring interventional therapy.

even brief therapy with inhaled nitric oxide decreased the incidence of bronchopulmonary dysplasia in premature newborns weighing more than 1000 g but not in smaller infants.²⁰ In addition, the study by Schreiber et al., which demonstrated a significant reduction in the combined end point of death and bronchopulmonary dysplasia with inhaled nitric oxide therapy, studied preterm infants with an average birth weight of 992 g.¹⁹ In contrast, the average birth weights in our trial and the study by Van Meurs et al. were 792 g and 839 g, respectively. Another important difference between our trial and the study by Van Meurs et al. was the duration of inhaled nitric oxide treatment (a median of 14 days vs. a mean of 76 hours).

Various factors other than inhaled nitric oxide, including the stage of lung development at birth, influence the need for supplemental oxygen at 36 weeks of postmenstrual age. The multifactorial nature of lung injury and repair at the extremes of prematurity may limit the efficacy of any single intervention, and the targeting of 36 weeks of postmenstrual age for an evaluation of the pul-

monary reparative capacity of newborns with extremely low birth weights may not be an adequate end point. Longer follow-up is needed to assess whether low-dose inhaled nitric oxide will reduce late pulmonary complications in premature newborns.

Mechanisms through which inhaled nitric oxide therapy might provide neuroprotection in the premature newborn are uncertain and warrant further study. One possibility is that inhaled nitric oxide modulates circulating cells (including neutrophils, monocytes, and platelets) as they transit the pulmonary circulation; the down-regulation of lung-derived cytokines that is induced by inhaled nitric oxide may also reduce the injury of distant organs.^{31,32} Another possible mechanism may relate to the distal delivery to the central nervous system of nitric oxide or nitric oxide-related metabolites through the systemic circulation through pathways mediated by red cells or proteins.^{33,34}

In conclusion, early, low-dose inhaled nitric oxide did not reduce the risk of bronchopulmonary dysplasia in premature newborns with respi-

ratory failure and a birth weight of 500 to 1250 g, but it did reduce the risk among infants with a birth weight of 1000 g or more and the risk of brain injury in the overall population. Long-term follow-up studies of these infants are ongoing to determine later pulmonary and neurocognitive outcomes of early inhaled nitric oxide therapy.

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APPENDIX

The following investigators also participated in this study: **National Heart, Lung, and Blood Institute** — M. Berberich, G. Zheng, N. Geller, C. Hunt; **Data and Safety Monitoring Board:** University of Rochester Medical Center, Rochester, N.Y. — D. Phelps; Johns Hopkins University, Baltimore — M. Allen; Emmes, Rockville, Md. — S. Carter; New York Academy of Medicine, New York — A. Fleischman; State University of New York, Buffalo — F. Morin; **Site Coordinators:** University of Colorado Health Sciences Center and Children's Hospital, Denver — S. Moreland, N. Waas, L. Fashaw, D. Rodden, K. Hale, G. Addison, S. Collins, K. Novak, A. Reed, B. Pruckler; Vanderbilt University Medical Center, Nashville — A. Law, S. Steele; Medical University of South Carolina, Charleston — K. Mathis; University of North Carolina, Chapel Hill — B. Gogri, L. Anderson, G. Bose; University of Southern California, Los Angeles — R. Mayoral, B. Jones, H. Chinchilla; Utah Valley Regional Medical Center, Provo, Utah — K. Wood, B. Smith; Duke University Medical Center, Durham, N.C. — K. Auten; Magee Women's Hospital, University of Pittsburgh, Pittsburgh — J. Jones, J. Lisak; Pennsylvania Hospital, Philadelphia — T. Mancini, A. Schwoebel, M. Grous; Children's Hospital of Oklahoma, Oklahoma City — M. McCoy, D. McCann, K. Corff; University of Iowa Hospitals and Clinics, Iowa City — K. Johnson, G. Cress; St. Joseph's Hospital, Phoenix — E. Ramthun; University of Connecticut Health Center, Farmington — K. Jennings; Loma Linda University Medical Center, Loma Linda, Calif. — L. Dalton, L. Machain; Children's Hospitals and Clinics of St. Paul, St. Paul, Minn. — P. Meyers, R. Gertz; Children's Hospitals and Clinics of Minneapolis, Minneapolis — M. Maxwell; **Data Coordinating Center:** D. Mitchell, W. Gehring, T. Van Duzer, T. Borreck, G. Hansen, C. Bender, C. Daniel.

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