

ORIGINAL ARTICLE

# Inhaled Nitric Oxide in Premature Infants with the Respiratory Distress Syndrome

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## ABSTRACT

### BACKGROUND

Inhaled nitric oxide improves gas exchange, decreases pulmonary vascular lability, and reduces pulmonary inflammation. We hypothesized that the use of inhaled nitric oxide would decrease the incidence of chronic lung disease and death in premature infants with the respiratory distress syndrome.

### METHODS

We conducted a randomized, double-blind, placebo-controlled study of the effect of inhaled nitric oxide during the first week of life on the incidence of chronic lung disease and death in premature infants (less than 34 weeks' gestation) who were undergoing mechanical ventilation for the respiratory distress syndrome. Infants were randomly assigned to receive inhaled nitric oxide (10 ppm on day 1, followed by 5 ppm for six days) or inhaled oxygen placebo for seven days. We further randomly assigned the infants in each group to receive intermittent mandatory or high-frequency oscillatory ventilation.

### RESULTS

A total of 207 premature infants were enrolled. In the group given inhaled nitric oxide, 51 infants (48.6 percent) died or had chronic lung disease, as compared with 65 infants (63.7 percent) in the placebo group (relative risk, 0.76; 95 percent confidence interval, 0.60 to 0.97;  $P=0.03$ ). There was no significant difference between the nitric oxide and placebo groups in the overall incidence of intraventricular hemorrhage and periventricular leukomalacia (33.3 percent and 38.2 percent, respectively), but the group given inhaled nitric oxide had a lower incidence of severe intraventricular hemorrhage and periventricular leukomalacia (12.4 percent vs. 23.5 percent; relative risk, 0.53; 95 percent confidence interval, 0.28 to 0.98;  $P=0.04$ ). The type of ventilation had no significant effect on the outcome.

### CONCLUSIONS

The use of inhaled nitric oxide in premature infants with the respiratory distress syndrome decreases the incidence of chronic lung disease and death.

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**C**HRONIC LUNG DISEASE REMAINS THE primary long-term pulmonary complication among premature infants and is associated with pulmonary hypertension as well as abnormalities of postnatal alveolarization and neovascularization.<sup>1</sup> In addition to having impaired growth,<sup>2</sup> infants with chronic lung disease may have poor long-term cardiopulmonary function, an increased susceptibility to infection,<sup>3</sup> and a sharply increased risk of abnormal neurologic development.<sup>4</sup>

Nitric oxide attenuates pulmonary vascular disease, inflammation, and pulmonary hypertension in newborns with lung injury.<sup>5-7</sup> Accordingly, we hypothesized that the use of inhaled nitric oxide would decrease the incidence of chronic lung disease and death in premature infants with the respiratory distress syndrome who were receiving mechanical ventilation. We conducted a randomized, double-blind, placebo-controlled study of inhaled nitric oxide in such infants during their first week of life.

## METHODS

This study was approved by the institutional review board of the University of Chicago. Written informed consent was obtained from the parents of all infants at the time of entry into the study.

### CRITERIA FOR ELIGIBILITY

Although chronic lung disease predominantly affects infants born at less than 28 weeks' gestation with birth weights under 1000 g,<sup>8</sup> larger premature infants with the respiratory distress syndrome severe enough to require mechanical ventilation and surfactant are at greater risk for chronic lung disease and death than their counterparts who are not undergoing mechanical ventilation. Accordingly, all premature infants who were less than 72 hours old, who had been born at less than 34 weeks' gestation and had a birth weight of less than 2000 g, and who were being cared for at the University of Chicago Children's Hospital were eligible for the study. Eligible infants had received a clinical diagnosis of the respiratory distress syndrome, requiring tracheal intubation, mechanical ventilation, and exogenous surfactant (Survanta, Abbott Laboratories). Infants were excluded if they had major congenital malformations or hydrops fetalis.

### STUDY DESIGN AND RANDOMIZATION

The study was a single-site, randomized, double-blind, placebo-controlled trial with a 2-by-2 factorial design. To ensure that the birth-weight distribu-

tions were similar among the groups, we used five 250-g birth-weight categories for randomization. Infants were randomly assigned within each stratum, according to a permuted-block design, to receive inhaled nitric oxide (INOmax, INO Therapeutics) or oxygen placebo and either intermittent mandatory ventilation or high-frequency oscillatory ventilation (model 3100A, SensorMedics).

Treatment with nitric oxide was initiated at a dose of 10 ppm by continuous inhalation for the first day (12 to 24 hours) followed by 5 ppm for six days. To ensure that changes in the study gas concentration occurred in the presence of the study investigators, treatment with the study gas was weaned during the daytime. When study gas was discontinued, it was resumed if the partial pressure of arterial oxygen (PaO<sub>2</sub>) decreased by more than 15 percent, and the dose was decreased by 1 ppm every six hours. This complication rarely occurred. Study gas was delivered by means of a proprietary delivery and monitoring unit (INOvent, Datex-Ohmeda), shielded so the identity of the gas was known only to the respiratory therapist and the study safety monitor. For infants receiving placebo, the respiratory therapist performed mock maneuvers to simulate changing nitric oxide concentrations.

### VENTILATOR STRATEGIES

Decisions about the management of ventilator and oxygen therapy were made by clinicians according to the usual protocol. The oxygenation index —  $(100 \times \text{the fractional inspiratory oxygen concentration} \times \text{the mean airway pressure [in centimeters of water]}) \div \text{PaO}_2 \text{ [in mm Hg]}$  — was calculated. Intermittent mandatory ventilation was begun at a rate of 40 breaths per minute, a peak inspiratory pressure sufficient to inflate the chest, and a positive end-expiratory pressure of 4 to 6 cm of water. High-frequency oscillatory ventilation was begun at a mean airway pressure 2 cm of water above that required during initial stabilization, an amplitude sufficient to jiggle the chest wall to the level of the umbilicus, and a frequency of 10 to 15 Hz. The mean airway pressure was adjusted to keep lung inflation at approximately nine posterior ribs on chest radiography. With both ventilatory strategies, PaO<sub>2</sub> was maintained between 50 and 90 mm Hg and the partial pressure of arterial carbon dioxide was maintained between 35 and 55 mm Hg. Infants whose birth weight was less than 1250 g were treated with prophylactic indomethacin to prevent or attenuate patent ductus arteriosus.

During the treatment period, the ventilatory

mode could be changed if the clinical staff thought that the patient's clinical condition was so critical that an alternative mode should be tried. In that case, treatment with the study gas was stopped. No crossover between groups was permitted, nor was inhaled nitric oxide administered to any premature infant who met the entry criteria but was not enrolled. Data from infants in whom the ventilatory mode was changed were analyzed on an intention-to-treat basis. Parents could withdraw their infant from the study at any time. In infants who were extubated within seven days, treatment was stopped one hour before extubation.

#### **HYPOTHESES AND OUTCOMES**

The primary hypothesis was that inhaled nitric oxide would decrease the incidence of chronic lung disease and death among premature infants who were undergoing mechanical ventilation. We defined the primary outcome measure as death or chronic lung disease (among surviving infants). Chronic lung disease was prospectively diagnosed by investigators who were unaware of the treatment assignments in infants who required supplemental oxygen as their usual daily therapy at 36 weeks' postmenstrual age and who had a chest radiograph showing persistent parenchymal lung disease.<sup>9</sup> Infants who died before discharge or by six months of age, whichever came later, were included in the analysis.

We performed additional post hoc analyses to improve our understanding of the influence of the ventilator strategy, the initial severity of lung disease, and birth weight on the effects of inhaled nitric oxide on chronic lung disease and death. The incidences of complications associated with the development of bleeding (a potential complication of nitric oxide therapy) or chronic lung disease were determined by investigators who were unaware of the infants' treatment assignments. To assess bleeding, we measured the incidence of pulmonary hemorrhage and the combined incidence of severe intraventricular hemorrhage (defined by a Papile grade of III or IV<sup>10</sup>) and periventricular leukomalacia.<sup>11</sup> Pulmonary hemorrhage was defined as clinically significant, bloody tracheal secretions temporally associated with a new pulmonary infiltrate and worsening pulmonary function.

To assess complications associated with chronic lung disease, we measured the incidences of pulmonary interstitial emphysema and pneumothorax. Because of the effect of nitric oxide on transitional pulmonary vascular resistance, we tracked the inci-

dence of symptomatic patent ductus arteriosus, defined by the need for indomethacin therapy or surgery in an infant with echocardiographically confirmed patent ductus arteriosus.

To determine whether the change in the incidence of chronic lung disease was associated with a decreased duration of mechanical ventilation or hospitalization, we measured these variables as well. Cranial ultrasonograms, obtained routinely during the first two weeks of life and before discharge, as well as all chest radiographs, were interpreted by an attending pediatric radiologist who was unaware of the infants' treatment assignments. The incidences of necrotizing enterocolitis, late-onset sepsis, retinopathy of prematurity, and hydrocephalus — all important complications of prematurity — were also systematically tracked, with outcomes identified by investigators who were unaware of the infants' treatment assignments.

#### **SAFETY MONITORING**

The methemoglobin concentration was measured daily, and the safety and data monitoring committee was informed of any instance in which the methemoglobin concentration exceeded 5 percent. Infants who had elevated methemoglobin concentrations on reexamination were to have study gas stopped. An interim, blinded analysis was performed by the safety and data monitoring committee at the midpoint of the study. The committee approved continuation of the study.

#### **STATISTICAL ANALYSIS**

Assuming an incidence of chronic lung disease and death of 60 percent, we determined that approximately 200 infants would need to be enrolled to provide the study with 80 percent power to detect a reduction in the incidence of more than 20 percent in the group given inhaled nitric oxide, with a two-sided type I error of 0.05. We conducted the analysis according to the intention-to-treat principle. Clinical and demographic characteristics of the two study groups were compared with the use of Pearson chi-square tests for categorical variables and two-sample t-tests or Wilcoxon rank-sum tests for continuous data.<sup>12</sup> If the expected number of observations was less than 5, Fisher's exact test was used instead. We also analyzed clinical outcomes using a generalized linear model with logarithmic link to obtain relative risks and corresponding 95 percent confidence intervals. We calculated adjusted relative risks, controlling for ventilation strategy using the generalized linear model. The interactions between birth

weight, oxygenation index, type of ventilation, and treatment group were also examined.<sup>13</sup> All reported P values are two-sided.

INO Therapeutics was not involved in the study design, safety monitoring, data analysis and interpretation, or manuscript preparation.

## RESULTS

### BASE-LINE CHARACTERISTICS AND CLINICAL COURSE

From October 1998 to October 2001, 207 premature infants receiving care at the University of Chicago Children's Hospital underwent randomization.

Demographic and base-line clinical characteristics did not differ significantly between the control group and the group given inhaled nitric oxide (Table 1). The birth-weight distribution was as follows: 72 infants (34.8 percent) weighed less than 750 g, 57 (27.5 percent) weighed 751 to 1000 g, 33 (15.9 percent) weighed 1001 to 1250 g, 18 (8.7 percent) weighed 1251 to 1500 g, and 27 (13.0 percent) weighed more than 1500 g.

Of the 207 randomized infants, 2 died before receiving any study medication and 1 never received study gas; all 3 were in the nitric oxide group. In five other infants, the assigned mode of ventilation was changed because of worsening clinical condition. Three of these infants (all of whom were assigned to inhaled nitric oxide) were switched from intermittent mandatory ventilation to high-frequency oscillatory ventilation. Of the other two infants (both of whom were assigned to placebo), one was switched from high-frequency oscillatory ventilation to intermittent mandatory ventilation, and the other was switched from intermittent mandatory ventilation to high-frequency oscillatory ventilation.

Three infants had elevated methemoglobin concentrations. None exceeded 7 percent, and none were elevated on reevaluation. Nitrogen dioxide, which was continuously monitored throughout the study, was never reported to be elevated (greater than 2 ppm).

Thirty-six infants were successfully extubated during the treatment period and therefore received study gas for fewer than seven days. Twenty of these infants were assigned to placebo gas, one of whom subsequently had chronic lung disease. Two other infants in the placebo group were extubated before the placebo was started. Among the 16 infants in the group given inhaled nitric oxide who were successfully extubated during the treatment period, chronic lung disease developed in 1.

### PRIMARY OUTCOME

In the group given inhaled nitric oxide, 51 of 105 infants (48.6 percent) died or had chronic lung disease, as compared with 65 of 102 infants (63.7 percent) in the placebo group (relative risk, 0.76; 95 percent confidence interval, 0.60 to 0.97; P=0.03) (Table 2). Similar results were obtained when we excluded the three infants who underwent randomization but never received study gas. The mode of ventilation had no significant effect on the primary outcome. Among the 102 infants who were treated with high-frequency oscillatory ventilation, 61 (59.8

**Table 1. Base-Line Characteristics.\***

Characteristic	Inhaled Nitric Oxide (N=105)	Placebo (N=102)
Birth weight — g	1017±369	949±387
Gestational age — wk	27.4±2.5	27.0±2.8
Age at study entry — hr		
Median	12.9	14.0
Interquartile range	7.0–25.2	7.6–28.5
Male sex — no. (%)	63 (60.0)	56 (54.9)
Mother's racial or ethnic group — no. (%) †		
Black	71 (67.6)	74 (72.6)
White	18 (17.1)	12 (11.8)
Other	16 (15.2)	16 (15.7)
Born at study hospital — no. (%)	80 (76.2)	80 (78.4)
Antenatal corticosteroids — no. (%) ‡	58 (56.3)	52 (52.0)
Cesarean section — no. (%)	54 (51.4)	58 (56.9)
Apgar score at 1 minute		
Median	5	5
Interquartile range	3–6	3–6
Apgar scores at 5 min		
Median	7	7
Interquartile range	6–8	6–8
Small for gestational age — no. (%)	6 (5.7)	9 (8.8)
Initial oxygenation index §		
Median	7.3	6.8
Interquartile range	4.1–12.3	4.4–12.7
Early-onset sepsis — no. (%) ¶	5 (4.8)	12 (11.8)
Surfactant — no. of doses	2.3±0.9	2.2±1.0

\* Plus-minus values are means ±SD.

† The racial or ethnic group was self-reported.

‡ Data were missing for two infants in the nitric oxide group and two in the placebo group.

§ The oxygenation index was calculated as  $(100 \times \text{the fractional inspiratory oxygen concentration} \times \text{the mean airway pressure}) \div \text{the partial pressure of arterial oxygen}$ .

¶ Early-onset sepsis was defined as sepsis that occurred within 24 hours after birth.

percent) died or had chronic lung disease, as compared with 55 of 105 infants (52.4 percent) treated with intermittent mandatory ventilation (relative risk, 1.14; 95 percent confidence interval, 0.90 to 1.45; P=0.28). After adjustment for the type of study gas, the relative risk was 1.12 (95 percent confidence interval, 0.88 to 1.42). There was no significant interaction between the type of study gas and the type of ventilation (P=0.11) (Table 3).

For a better understanding of the birth-weight subgroups that benefited most from inhaled nitric

oxide, we performed post hoc analyses stratified according to birth weight. No significant interaction was observed between the type of study gas and the birth-weight subgroup (Table 4).

To evaluate whether inhaled nitric oxide differentially benefited infants with mild, as compared with severe, initial respiratory disease, we performed a post hoc analysis, stratified according to the severity of disease, dividing infants into two groups on the basis of whether their initial oxygenation index was less than the median of the initial oxygenation

**Table 2. Primary Outcome.\***

Outcome	Inhaled Nitric Oxide (N=105) no. (%)	Placebo (N=102) no. (%)	P Value	Relative Risk (95% CI)	Adjusted Relative Risk (95% CI)†
Death or chronic lung disease	51 (48.6)	65 (63.7)	0.03	0.76 (0.60–0.97)	0.77 (0.60–0.98)
Death	16 (15.2)	23 (22.5)	0.18	0.68 (0.38–1.20)	0.68 (0.38–1.20)
Survival	89 (84.8)	79 (77.5)			
Survival without chronic lung disease	54 (60.7)	37 (46.8)			
Survival with chronic lung disease	35 (39.3)	42 (53.2)	0.07	0.74 (0.53–1.03)	0.75 (0.54–1.05)

\* CI denotes confidence interval.

† Values were adjusted for the type of ventilation.

**Table 3. Primary Outcome According to the Type of Ventilation and Severity of Disease.**

Variable	Inhaled Nitric Oxide	Placebo	Relative Risk (95% CI)*	P Value for Interaction
<i>no. (%)</i>				
<b>Type of ventilation</b>				0.11
Conventional ventilation	51	54		
Survival without chronic lung disease	31 (60.8)	19 (35.2)		
Death or survival with chronic lung disease	20 (39.2)	35 (64.8)	0.61 (0.41–0.90)	
High-frequency oscillatory ventilation	54	48		
Survival without chronic lung disease	23 (42.6)	18 (37.5)		
Death or survival with chronic lung disease	31 (57.4)	30 (62.5)	0.92 (0.67–1.26)	
<b>Initial oxygenation index†</b>				0.02
<6.94 (median)	50	49		
Survival without chronic lung disease	32 (64.0)	16 (32.7)		
Death or survival with chronic lung disease	18 (36.0)	33 (67.3)	0.53 (0.35–0.81)	
≥6.94	51	48		
Survival without chronic lung disease	21 (41.2)	20 (41.7)		
Death or survival with chronic lung disease	30 (58.8)	28 (58.3)	1.01 (0.72–1.40)	

\* CI denotes confidence interval.

† Data on the initial oxygenation index were missing for four infants in the nitric oxide group and five in the placebo group.

**Table 4. Primary Outcome According to Birth Weight.\***

Variable	Total No. of Infants	Inhaled Nitric Oxide (N=105)		Placebo (N=102)
		no. (%)		
Birth weight, ≤750 g	72	32	40	
Survival without chronic lung disease		7 (21.9)	4 (10.0)	
Death or survival with chronic lung disease		25 (78.1)	36 (90.0)	
Birth weight, 751–1000 g	57	28	29	
Survival without chronic lung disease		14 (50.0)	11 (37.9)	
Death or survival with chronic lung disease		14 (50.0)	18 (62.1)	
Birth weight, 1001–1500 g	51	30	21	
Survival without chronic lung disease		18 (60.0)	12 (57.1)	
Death or survival with chronic lung disease		12 (40.0)	9 (42.9)	
Birth weight, >1500 g	27	15	12	
Survival without chronic lung disease		15 (100.0)	10 (83.3)	
Death or survival with chronic lung disease		0	2 (16.7)	

\* There was not a significant interaction between birth weight and the primary outcome ( $P=0.36$ ).

index (6.94). As compared with placebo gas, inhaled nitric oxide significantly decreased the risk of the primary outcome by 47 percent among infants whose oxygenation index was below the median but not among infants whose initial oxygenation index was at or above the median (Table 3). This disease-severity-specific interaction was significant ( $P=0.02$ ).

#### SECONDARY OUTCOMES

The incidence of pulmonary hemorrhage did not differ significantly between the two groups (Table 5). The overall incidence of intraventricular hemorrhage and periventricular leukomalacia did not differ significantly between the groups (38.2 percent in the placebo group and 33.3 percent in the group given inhaled nitric oxide,  $P=0.46$ ). However, as compared with placebo gas, inhaled nitric oxide significantly decreased the incidence of severe intraventricular hemorrhage and periventricular leukomalacia (risk reduction, 47 percent;  $P=0.04$ ) (Table 5).

The incidences of pneumothorax, pulmonary interstitial emphysema, and symptomatic patent ductus arteriosus did not differ significantly between the groups. There were also no significant differences between groups in the incidences of other common complications of prematurity, including necro-

tizing enterocolitis, late-onset sepsis, retinopathy of prematurity, and hydrocephalus (Table 5).

Among the infants who survived, the median duration of mechanical ventilation was 16 days (interquartile range, 8 to 37) in the group given inhaled nitric oxide, as compared with 28.5 days (interquartile range, 8 to 48) in the placebo group ( $P=0.19$ ). The median length of hospitalization was 76 days (interquartile range, 44 to 97) in the control group, as compared with 65 days (interquartile range, 42 to 88) in the group given inhaled nitric oxide ( $P=0.22$ ).

#### DISCUSSION

In this randomized, controlled study, early treatment with inhaled nitric oxide improved long-term pulmonary outcomes in premature infants with the respiratory distress syndrome, decreasing the incidence of the combined end point of chronic lung disease and death. In addition, inhaled nitric oxide decreased the incidence of severe intraventricular hemorrhage and periventricular leukomalacia, the primary cause of serious, long-term neurologic disability in this population.

Analysis of the data according to the mode of ventilation showed a significant decrease in the risk of chronic lung disease and death in the group receiving nitric oxide and intermittent mandatory ventilation but not in the group receiving nitric oxide and high-frequency oscillatory ventilation. However, because the study did not have sufficient power to detect a significant interaction, conclusions cannot be drawn regarding the question of whether the benefit of inhaled nitric oxide is restricted to infants receiving intermittent mandatory ventilation.

Nitric oxide is an important mediator of both normal lung development and pulmonary vascular tone<sup>14–17</sup> and may be important in the optimization of ventilation-perfusion matching. In animal models of chronic lung disease, the expression of nitric oxide synthase is decreased in both small-airway epithelium and distal-pulmonary-artery endothelium.<sup>18,19</sup> In addition, other nitric oxide-dependent processes may be important in preventing chronic lung disease, including enhancement of pulmonary surfactant activity,<sup>20</sup> inhibition of neutrophil infiltration and retention,<sup>21,22</sup> inhibition of cytokines,<sup>23</sup> and prevention of neomuscularization and airway remodeling.<sup>6,24</sup>

In contrast with previous randomized trials, we studied all premature infants with respiratory distress syndrome who required mechanical ventila-

**Table 5. Secondary Outcomes.**

Outcome	Inhaled Nitric Oxide (N=105)	Placebo (N=102)	Relative Risk (95% CI)*	P Value†
	no. (%)			
Severe intraventricular hemorrhage and periventricular leukomalacia	13 (12.4)	24 (23.5)	0.53 (0.28–0.98)	0.04
Pneumothorax	11 (10.5)	16 (15.7)	0.67 (0.33–1.37)	0.27
Pulmonary interstitial emphysema	28 (26.7)	35 (34.3)	0.78 (0.51–1.18)	0.23
Pulmonary hemorrhage	4 (3.8)	7 (6.9)	0.56 (0.17–1.84)	0.37‡
Symptomatic patent ductus arteriosus	20 (19.0)	26 (25.5)	0.75 (0.45–1.25)	0.27
Necrotizing enterocolitis§	13 (12.4)	6 (5.9)	2.10 (0.83–5.32)	0.11
Late-onset sepsis¶	54 (51.4)	50 (49.0)	1.05 (0.80–1.38)	0.73
Retinopathy of prematurity	6 (5.7)	10 (9.8)	0.58 (0.22–1.54)	0.27
Hydrocephalus	12 (11.4)	10 (9.8)	1.17 (0.53–2.58)	0.71

\* CI denotes confidence interval.

† P values were calculated with the use of Pearson's chi-square test, except when otherwise specified.

‡ The P value was calculated with the use of Fisher's exact test.

§ Necrotizing enterocolitis was defined by a Bell's stage of 2 or greater.

¶ Late-onset sepsis was defined as sepsis that occurred after 24 hours of age.

|| Retinopathy of prematurity was defined by an international classification stage of 3 or greater.

tion. In our previous study of premature infants weighing less than 1000 g who had relatively mild initial disease (as defined by an oxygenation index of less than 4), the risk of chronic lung disease was nearly 25 percent.<sup>25</sup> Of the infants who were given a diagnosis of chronic lung disease, 28 percent had mild or moderate lung disease initially and might therefore have benefited from therapies such as inhaled nitric oxide. We found that infants with milder disease (as defined by an oxygenation index of less than 6.94) were the ones likely to benefit from inhaled nitric oxide, although this finding was derived from a post hoc analysis and must be interpreted cautiously. Additional studies are needed to identify the subpopulations of infants who would benefit most from inhaled nitric oxide and to determine the optimal dose and duration of therapy.

In addition to decreasing the risk of chronic lung disease and death, inhaled nitric oxide decreased the risk of severe intraventricular hemorrhage and periventricular leukomalacia by 47 percent, as compared with placebo gas. The overall incidence of intraventricular hemorrhage and periventricular leukomalacia did not differ significantly between the groups, however, suggesting that inhaled nitric oxide did not prevent this complication but instead reduced its severity. Nitric oxide, by decreasing right

ventricular afterload, may attenuate venous stasis and subsequent infarction of the fragile germinal-matrix arteriovenous rete.<sup>26</sup> In addition, nitric oxide-induced reduction of platelet aggregation<sup>27,28</sup> may limit venous thrombosis.<sup>26</sup> Finally, inhaled nitric oxide inhibits cytokines,<sup>23</sup> which may also have a role in the pathogenesis of intraventricular hemorrhage and periventricular leukomalacia.<sup>29</sup>

Concern about the safety of inhaled nitric oxide has centered on the possibility of bleeding, methemoglobinemia, and oxidative stress. Inhaled nitric oxide increases bleeding times in adults<sup>30</sup> and term infants,<sup>31,32</sup> and an early, uncontrolled study of inhaled nitric oxide in premature infants showed a disturbing incidence of intraventricular hemorrhage.<sup>33</sup> However, subsequent studies have reported incidences of intraventricular hemorrhage no different from those among untreated infants.<sup>34–36</sup> Our results provide further reassurance in this regard. Methemoglobinemia has been reported in term infants treated with high concentrations of inhaled nitric oxide (80 ppm) but not in infants receiving less than 20 ppm.<sup>7,37,38</sup> Inhaled nitric oxide could subject the lung to increased oxidative stress, which contributes to a variety of lung injuries.<sup>39</sup> Although elevated concentrations of 3-nitrotyrosine may be present in lung-lavage fluid from infants after prolonged ex-

posure to nitric oxide (10 days or more),<sup>40</sup> increases have not been observed in term infants who received inhaled nitric oxide for fewer than 10 days.<sup>39</sup>

In conclusion, when initiated soon after birth, treatment with low-dose inhaled nitric oxide reduces the incidence of chronic lung disease and death among premature infants with the respiratory distress syndrome. The use of nitric oxide may also decrease the risk of severe intraventricular hemorrhage and periventricular leukomalacia, which are important neonatal complications associated with prematurity.

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