

ORIGINAL ARTICLE

# Neurodevelopmental Outcomes of Premature Infants Treated with Inhaled Nitric Oxide

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## ABSTRACT

### BACKGROUND

Chronic lung disease and severe intraventricular hemorrhage or periventricular leukomalacia in premature infants are associated with abnormal neurodevelopmental outcomes. In a previous randomized, controlled, single-center trial of premature infants with the respiratory distress syndrome, inhaled nitric oxide decreased the risk of death or chronic lung disease as well as severe intraventricular hemorrhage and periventricular leukomalacia. We hypothesized that infants treated with inhaled nitric oxide would also have improved neurodevelopmental outcomes.

### METHODS

We conducted a prospective, longitudinal follow-up study of premature infants who had received inhaled nitric oxide or placebo to investigate neurodevelopmental outcomes at two years of corrected age. Neurologic examination, neurodevelopmental assessment, and anthropometric measurements were made by examiners who were unaware of the children's original treatment assignment.

### RESULTS

A total of 138 children (82 percent of survivors) were evaluated. In the group given inhaled nitric oxide, 17 of 70 children (24 percent) had abnormal neurodevelopmental outcomes, defined as either disability (cerebral palsy, bilateral blindness, or bilateral hearing loss) or delay (no disability, but one score of less than 70 on the Bayley Scales of Infant Development II), as compared with 31 of 68 children (46 percent) in the placebo group (relative risk, 0.53; 95 percent confidence interval, 0.33 to 0.87;  $P=0.01$ ). This effect persisted after adjustment for birth weight and sex, as well as for the presence or absence of chronic lung disease and severe intraventricular hemorrhage or periventricular leukomalacia. The improvement in neurodevelopmental outcome in the group given inhaled nitric oxide was primarily due to a 47 percent decrease in the risk of cognitive impairment (defined by a score of less than 70 on the Bayley Mental Developmental Index) ( $P=0.03$ ).

### CONCLUSIONS

Premature infants treated with inhaled nitric oxide have improved neurodevelopmental outcomes at two years of age.

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OVER THE PAST DECADE, ADVANCES IN neonatal–perinatal medicine have resulted in increased survival rates among premature infants.<sup>1</sup> Despite therapies such as antenatal corticosteroids and surfactant replacement, however, the rates of birth-weight–specific abnormal neurodevelopmental outcomes have remained constant.<sup>2,3</sup> Other therapies, such as postnatal corticosteroids,<sup>4</sup> have been associated with abnormal neurodevelopment. The disabling neurologic conditions — cerebral palsy, hearing loss, and blindness — adversely affect the long-term neurodevelopmental outcomes among premature infants. However, even premature infants who have relatively uncomplicated neonatal courses are at substantial risk for developmental delays in cognition and motor skills.<sup>5</sup>

In a double-blind, randomized, placebo-controlled, single-center trial, we demonstrated that inhaled nitric oxide significantly decreased the incidence of chronic lung disease and death by 24 percent in premature infants with the respiratory distress syndrome.<sup>6</sup> Inhaled nitric oxide also decreased the incidence of severe intraventricular hemorrhage and periventricular leukomalacia by 47 percent. Because these pulmonary and neurologic conditions are associated with abnormal neurodevelopmental outcomes, we hypothesized that premature infants who had received inhaled nitric oxide would have improved neurodevelopmental outcomes. Accordingly, we prospectively studied the neurodevelopmental outcomes among this cohort of infants at two years of age.

## METHODS

### PRIMARY OUTCOME

This study was a prospective, longitudinal follow-up of the neurodevelopmental outcomes among premature infants who were randomly assigned to receive inhaled nitric oxide or placebo. The primary outcome, as specified in the protocol for this follow-up study, was an abnormal neurodevelopmental outcome at two years of age. We used three neurodevelopmental outcomes<sup>7</sup>: disability, defined as cerebral palsy, bilateral blindness, or bilateral hearing loss; delay, defined by a score on the Mental or Psychomotor Developmental Index of the Bayley Scales of Infant Development II that was at least 2 SD below the mean (i.e., less than 70) without disability; and normal, defined by the absence of both disability and delay. An abnormal neurodevelop-

mental outcome was defined as either disability or delay. The study was approved by the institutional review board of the University of Chicago. Written informed consent was obtained from the children's parents.

### INITIAL STUDY

The initial study, conducted at the University of Chicago from October 1998 to October 2001, was a single-center, randomized, placebo-controlled trial of 207 premature infants requiring mechanical ventilation and surfactant-replacement therapy (mean [ $\pm$ SD] gestational age, 27.2 $\pm$ 2.7 weeks). Treatment with inhaled nitric oxide (INO Therapeutics) was initiated at a dose of 10 ppm for the first day, followed by a dose of 5 ppm for six days or until extubation. A total of 105 patients received inhaled nitric oxide, and 102 patients received placebo. Infants were also randomly assigned to receive intermittent mandatory ventilation or high-frequency oscillatory ventilation.

### FOLLOW-UP

Close contact with the original cohort was maintained, and children were scheduled for evaluation at one and two years of age, corrected for prematurity. Data for the present study were collected at two years of corrected age. Patients were evaluated at the University of Chicago Neonatal High-Risk Follow-up Clinic. Medical records, including visual and hearing examinations, were reviewed. Maternal race or ethnic group was self-reported, as recorded in the maternal medical record. Socioeconomic status was determined by interviewing the mothers at follow-up. The children's height, weight, and head circumference were measured. A physical examination was performed by a pediatrician, a neurologic examination was performed by a pediatric neurologist, and infant development was assessed by a certified neonatal occupational therapist, dedicated to the High-Risk Clinic. Infant development was assessed with the use of the Bayley Scales of Infant Development II Mental and Psychomotor Developmental Indexes,<sup>8</sup> adjusted for prematurity. Examiners and parents were unaware of the patients' treatment assignments in the initial study.

Patients were screened for cerebral palsy with the use of the following criteria<sup>9</sup>: abnormalities in neuromotor tone, deep tendon reflexes, and either coordination or movement; delay in motor milestones; and abnormalities in primitive reflexes and postural reactions. Patients with spastic hemiple-

gia, diplegia, hemidiplegia, or quadriplegia received a diagnosis of cerebral palsy.<sup>10</sup>

Patients were screened for visual impairment by means of a parental questionnaire, and the diagnosis was confirmed by a pediatric ophthalmologist. Blindness was defined as a corrected visual acuity worse than 20/200. Hearing loss was defined as impairment requiring a hearing aid.

The proportions of children in the two groups who had cognitive impairment (defined by a score of less than 70 on the Bayley Mental Developmental Index), performance impairment (defined by a score of less than 70 on the Bayley Psychomotor Developmental Index), or both were compared. However, since the Bayley Scales of Infant Development II system does not assign numerical values for scores of less than 50, the proportion of children in each group with scores of less than 50 was also calculated.

#### STATISTICAL ANALYSIS

Clinical and demographic characteristics of the study groups were compared by means of Fisher's exact tests for categorical variables and two-sample t-tests or Wilcoxon rank-sum tests for continuous variables. To control for sex and corrected age in the analysis of anthropometric measures, z scores were generated with the use of the Centers for Disease Control and Prevention (CDC) growth charts.<sup>11</sup>

The primary outcome was analyzed with the use of a generalized linear model with a logarithmic link (log-binomial model) to obtain relative risks and corresponding 95 percent confidence intervals. Using this model, we calculated adjusted relative risks, controlling individually for potential confounders: birth weight, sex, and (as measures of socioeconomic status) whether the mother had graduated from high school and the presence or absence of any employed person in the household. Adjustments were also made for the presence or absence of prolonged (more than seven days) postnatal exposure to corticosteroids, the type of ventilation used (high-frequency oscillatory or intermittent mechanical), and the initial oxygenation index, calculated by means of the following equation:  $100 \times \frac{\text{the fraction of inspired oxygen} \times \text{mean airway pressure (in centimeters of water)}}{\text{partial pressure of arterial oxygen (in millimeters of mercury)}}$ . Because of the relatively few events in some subgroups, it was impossible to adjust for all potential confounders simultaneously in the multivariate analysis.

Therefore, we controlled only for the confounders found to be significant predictors of neurodevelopmental outcome: birth weight and sex.

To better characterize factors mediating the primary outcome, we adjusted the primary outcome separately and simultaneously for the potential intermediate variables of the presence or absence of chronic lung disease and intraventricular hemorrhage or periventricular leukomalacia. In cases in which the log-binomial model did not converge, a modified Poisson regression was fitted.<sup>12</sup> All analyses were prespecified, unless noted as post hoc. All P values are two-sided.

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## RESULTS

#### PATIENT DISPOSITION

Of the 207 infants enrolled in the initial study, 168 were alive at two years of age (89 in the group given inhaled nitric oxide and 79 in the placebo group). A total of 29 infants were lost to follow-up: 13 families could not be reached, and 16 families were contacted but did not come to the clinic. The 29 infants lost to follow-up had greater birth weights ( $1255 \pm 453$  g vs.  $994 \pm 360$  g,  $P < 0.001$ ) and gestational ages ( $28.6 \pm 3.0$  weeks vs.  $27.4 \pm 2.5$  weeks,  $P = 0.03$ ) than those who were included in the follow-up study.

From January 2000 to February 2004, 139 patients from the initial study were evaluated. Three patients underwent neurologic examinations but not Bayley testing. Two of the three patients were identified as having cerebral palsy, categorized as disabled, and included in the analysis. The third patient (who had received inhaled nitric oxide) was identified as not having cerebral palsy but was excluded from the analysis, since it could not be determined whether the child had normal or delayed neurodevelopment. Accordingly, 138 patients (70 who had received inhaled nitric oxide and 68 who had received placebo; 82 percent of survivors) were analyzed.

#### CHARACTERISTICS OF THE FOLLOW-UP COHORT

In the follow-up cohort, 89 of the 168 patients who were alive at two years of age (53 percent) had received inhaled nitric oxide, similar to the proportion of patients enrolled in the original study (51

percent). The baseline characteristics of the infants at the time of enrollment in the initial study did not differ significantly between study groups (Table 1). In contrast, the group given inhaled nitric oxide had a decreased incidence of severe intraventricular hemorrhage or periventricular leukomalacia (P=0.02) (Table 1). There was no significant difference in the mean corrected age at follow-up (24.9±7.9 months in the group given inhaled nitric oxide and 25.2±8.4 months in the placebo group, P=0.84). Maternal characteristics did not differ significantly in terms of important factors

**Table 1. Characteristics of the Follow-up Cohort and Their Mothers.\***

| Characteristic                                                      | Inhaled Nitric Oxide (N=70) | Placebo (N=68) | P Value |
|---------------------------------------------------------------------|-----------------------------|----------------|---------|
| <b>Infants</b>                                                      |                             |                |         |
| Birth weight — g                                                    | 1026±366                    | 958±356        | 0.27    |
| Gestational age — wk                                                | 27.5±2.4                    | 27.2±2.6       | 0.50    |
| Corrected age at follow-up — mo                                     | 24.9±7.9                    | 25.2±8.4       | 0.84    |
| Male sex — no. (%)                                                  | 39 (56)                     | 33 (49)        | 0.50    |
| Initial oxygenation index†                                          |                             |                |         |
| Median                                                              | 6.6                         | 7.2            | 0.37    |
| Interquartile range                                                 | 4.0–11.5                    | 4.5–14.3       |         |
| 1-Minute Apgar score                                                |                             |                |         |
| Median                                                              | 5                           | 5              | 0.65    |
| Interquartile range                                                 | 3–6                         | 3–6            |         |
| 5-Minute Apgar score                                                |                             |                |         |
| Median                                                              | 7                           | 7              | 0.54    |
| Interquartile range                                                 | 7–8                         | 6–8            |         |
| Antenatal corticosteroids — no./total no. (%)                       | 40/69 (58)                  | 38/67 (57)     | 1.0     |
| Surfactant — no. of doses                                           | 2.2±0.9                     | 2.3±1.0        | 0.44    |
| Prolonged postnatal exposure to corticosteroids — no. (%)‡          | 6 (9)                       | 6 (9)          | 1.0     |
| At end of original study — no. (%)‡                                 |                             |                |         |
| Chronic lung disease§                                               | 27 (39)                     | 37 (54)        | 0.09    |
| Severe intraventricular hemorrhage or periventricular leukomalacia¶ | 6 (9)                       | 16 (24)        | 0.02    |
| <b>Household</b>                                                    |                             |                |         |
| Mother has less than high-school education — no./total no. (%)      | 11/66 (17)                  | 9/62 (15)      | 0.81    |
| Single parent — no./total no. (%)                                   | 35/66 (53)                  | 26/65 (40)     | 0.16    |
| Foster care — no./total no. (%)                                     | 6/70 (9)                    | 6/68 (9)       | 1.0     |
| Household without an employed person — no./total no. (%)            | 18/67 (27)                  | 12/68 (18)     | 0.30    |
| Maternal race or ethnic group — no./total no. (%)                   |                             |                |         |
| Black                                                               | 44/70 (63)                  | 52/68 (76)     | 0.23    |
| White                                                               | 14/70 (20)                  | 8/68 (12)      |         |
| Other                                                               | 12/70 (17)                  | 8/68 (12)      |         |

\* Plus-minus values are means ±SD.

† The initial oxygenation index was calculated by means of the following equation: 100 × the fractional inspiratory oxygen concentration × mean airway pressure (in centimeters of water) ÷ partial pressure of arterial oxygen in millimeters of mercury).

‡ Prolonged exposure was defined as more than seven days.

§ Chronic lung disease was defined as the need for supplemental oxygen at 36 weeks' postmenstrual age plus abnormal findings on chest radiography.

¶ Severe intraventricular hemorrhage or periventricular leukomalacia was defined by a Papile grade of III or IV.<sup>13</sup>

|| Race or ethnic group was self-reported.

known to influence neurodevelopmental outcomes, including the level of education, marital status, employment status, and race or ethnic group.<sup>5</sup>

**NEURODEVELOPMENTAL OUTCOMES**

Patients treated with inhaled nitric oxide had approximately half the risk of having an abnormal neurodevelopmental outcome as those in the placebo group (relative risk, 0.53; 95 percent confidence interval, 0.33 to 0.87; P=0.01). Eight patients (12 percent) in the placebo group were identified as disabled, as compared with six patients (9 percent) in the group given inhaled nitric oxide. Of the 14 disabled patients, 1 (in the placebo group) had isolated hearing loss. The remainder had cerebral palsy, and two of these (in the placebo group) were also blind. All six of the patients with cerebral palsy

in the group given inhaled nitric oxide were diplegic. In the placebo group, two were diplegic, four hemiplegic, and one quadriplegic. The percentage of patients categorized as having developmental delays was higher in the placebo group than in the group given inhaled nitric oxide (34 percent vs. 16 percent).

We also assessed associations between the neurodevelopmental outcome and other factors, including birth weight, sex, whether the mother had graduated from high school, and the presence or absence of any employed person in the household. Lower birth weight and male sex were associated with an increased risk of an abnormal neurodevelopmental outcome, but low socioeconomic status was not (Table 2). We also considered relationships between the primary outcome and the type of ven-

**Table 2. Risk Factors for the Primary Outcome of Abnormal Neurodevelopment at Two Years of Age.**

| Risk Factor                                                                            | Neurodevelopmental Outcome |                 | Relative Risk (95% CI)* | P Value |
|----------------------------------------------------------------------------------------|----------------------------|-----------------|-------------------------|---------|
|                                                                                        | Normal (N=90)              | Abnormal (N=48) |                         |         |
| Birth weight                                                                           |                            |                 |                         |         |
| Mean — g                                                                               | 1047±374                   | 891±316         |                         |         |
| Per 100-g increment                                                                    |                            |                 | 0.91 (0.85–0.99)        | 0.02    |
| Sex — no./total no. (%)                                                                |                            |                 |                         |         |
| Male                                                                                   | 39/72 (54)                 | 33/72 (46)      | 2.02 (1.21–3.36)        | 0.007   |
| Female                                                                                 | 51/66 (77)                 | 15/66 (23)      | 1.00                    |         |
| Maternal education — no./total no. (%)                                                 |                            |                 |                         |         |
| Less than high school                                                                  | 15/20 (75)                 | 5/20 (25)       | 0.73 (0.33–1.63)        | 0.61    |
| High school or higher                                                                  | 71/108 (66)                | 37/108 (34)     | 1.00                    |         |
| Household without an employed person — no./total no. (%)                               |                            |                 |                         |         |
| Yes                                                                                    | 19/30 (63)                 | 11/30 (37)      | 1.17 (0.50–2.74)        | 0.83    |
| No                                                                                     | 69/103 (67)                | 34/103 (33)     | 1.00                    |         |
| Type of ventilation — no./total no. (%)                                                |                            |                 |                         |         |
| High-frequency oscillatory                                                             | 44/66 (67)                 | 22/66 (33)      | 0.92 (0.58–1.46)        | 0.86    |
| Intermittent mechanical                                                                | 46/74 (62)                 | 26/74 (35)      | 1.00                    |         |
| Prolonged postnatal exposure to corticosteroids — no./total no. (%)                    |                            |                 |                         |         |
| Yes                                                                                    | 8/12 (67)                  | 4/12 (33)       | 0.94 (0.41–2.16)        | 1.0     |
| No                                                                                     | 80/124 (65)                | 44/124 (35)     | 1.00                    |         |
| Chronic lung disease — no./total no. (%)                                               |                            |                 |                         |         |
| Yes                                                                                    | 33/64 (52)                 | 31/64 (48)      | 2.11 (1.29–3.43)        | 0.002   |
| No                                                                                     | 57/74 (77)                 | 17/74 (23)      | 1.00                    |         |
| Severe intraventricular hemorrhage or periventricular leukomalacia — no./total no. (%) |                            |                 |                         |         |
| Yes                                                                                    | 10/22 (45)                 | 12/22 (55)      | 1.76 (1.10–2.81)        | 0.05    |
| No                                                                                     | 80/116 (69)                | 36/116 (31)     | 1.00                    |         |

\* CI denotes confidence interval.

tilation used and the presence of prolonged postnatal exposure to corticosteroids (more than seven days), chronic lung disease, and severe intraventricular hemorrhage or periventricular leukomalacia. Of these factors, only chronic lung disease and severe intraventricular hemorrhage and periventricular leukomalacia were associated with an increased risk of an abnormal neurodevelopmental outcome (Table 2).

Because other factors contributed to an abnormal neurodevelopmental outcome, we assessed the extent to which these factors accounted for the significant effect of inhaled nitric oxide. We first adjusted the relative risk of the primary outcome for the confounding factors of birth weight, sex, whether the mother had graduated from high school, the presence or absence of any employed person in the household, type of ventilation, and whether there was prolonged postnatal exposure to corticosteroids. After adjustment for each of these factors separately, the risk of an abnormal neurodevelopmental outcome remained essentially unchanged among patients treated with inhaled nitric oxide as compared with those in the placebo group (Table 3).

Furthermore, in a post hoc analysis simultaneously adjusting for both birth weight and sex, the risk of an abnormal neurodevelopmental outcome remained essentially unchanged (Table 3).

Inhaled nitric oxide may have decreased the incidence of an abnormal neurodevelopmental outcome through its effects on the incidences of chronic lung disease and severe intraventricular hemorrhage or periventricular leukomalacia. However, the decreased risk of an abnormal neurodevelopmental outcome persisted after separate adjustment for these intermediate variables (Table 3). In addition, simultaneous, post hoc adjustment for the presence of chronic lung disease and severe intraventricular hemorrhage or periventricular leukomalacia minimally attenuated the relative risk in the group given inhaled nitric oxide, as compared with the placebo group (0.60 vs. 0.53), indicating that a reduced risk of chronic lung disease, severe intraventricular hemorrhage or periventricular leukomalacia, or both outcomes does not fully explain the effect of inhaled nitric oxide on neurodevelopmental outcome. Because of the relatively small size of our cohort and small number of events, simultaneous adjustment for all four significant confounding and intermediate variables was not possible.

In the original study, inhaled nitric oxide preferentially decreased the incidence of chronic lung disease and death among premature infants having oxygenation indexes below the median of 6.94. The median oxygenation index in the follow-up cohort was 6.8. For the comparison of the group given inhaled nitric oxide with the placebo group, the relative risk of an abnormal neurodevelopmental outcome was 0.52 (95 percent confidence interval, 0.26 to 1.01) in the group with an initial oxygenation index of less than 6.94 and 0.38 (95 percent confidence interval, 0.16 to 0.93) for those with an index of 6.94 or greater. However, the interaction between the initial oxygenation index and the primary outcome in this post hoc analysis was not significant ( $P=0.59$ ).

#### BAYLEY SCORES

Because we observed few cases of disability and the distribution of these cases did not differ significantly between groups, inhaled nitric oxide may have improved neurodevelopmental outcome solely through a decrease in neurodevelopmental delay. However, in this study, a diagnosis of neurodevelopmental delay excluded children with cerebral palsy. Thus, we performed a post hoc analysis to

**Table 3. Primary Outcome Adjusted for Potential Confounders and Intermediate Variables.**

| Potential Confounders and Intermediate Variables                                                                        | Relative Risk of Abnormal Neurodevelopmental Outcome (95% CI)* | P Value |
|-------------------------------------------------------------------------------------------------------------------------|----------------------------------------------------------------|---------|
| None                                                                                                                    | 0.53 (0.33–0.87)                                               | 0.01    |
| <b>Potential confounders</b>                                                                                            |                                                                |         |
| Birth weight                                                                                                            | 0.57 (0.35–0.93)                                               | 0.02    |
| Sex                                                                                                                     | 0.52 (0.32–0.82)                                               | 0.006   |
| Mother's graduation from high school                                                                                    | 0.48 (0.28–0.82)                                               | 0.007   |
| Household without an employed person                                                                                    | 0.49 (0.29–0.82)                                               | 0.006   |
| Type of ventilation                                                                                                     | 0.53 (0.33–0.87)                                               | 0.01    |
| Prolonged postnatal exposure to corticosteroids                                                                         | 0.53 (0.33–0.87)                                               | 0.01    |
| Simultaneous adjustment for birth weight and sex                                                                        | 0.55 (0.35–0.88)                                               | 0.01    |
| <b>Potential intermediate variables</b>                                                                                 |                                                                |         |
| Severe intraventricular hemorrhage or periventricular leukomalacia                                                      | 0.55 (0.34–0.89)                                               | 0.01    |
| Chronic lung disease                                                                                                    | 0.59 (0.36–0.95)                                               | 0.03    |
| Simultaneous adjustment for chronic lung disease and severe intraventricular hemorrhage or periventricular leukomalacia | 0.60 (0.38–0.96)                                               | 0.03    |

\* CI denotes confidence interval.

compare Bayley scores of all children regardless of disability diagnosis (Table 4). As compared with the placebo group, the group given inhaled nitric oxide had approximately half the risk of either cognitive impairment (defined by a score of less than 70 on the Mental Developmental Index) or performance impairment (defined by a score of less than 70 on the Psychomotor Developmental Index), or both (relative risk, 0.55; 95 percent confidence interval, 0.33 to 0.93; P=0.03). This difference was the result of a decreased incidence of cognitive impairment in the group given inhaled nitric oxide, as compared with the placebo group (relative risk, 0.53; 95 percent confidence interval, 0.29 to 0.94; P=0.03). The

risk of performance impairment did not differ significantly between groups (relative risk, 0.73; 95 percent confidence interval, 0.33 to 1.61; P=0.48). The distributions of scores for the Mental and Psychomotor Developmental Indexes in the group given inhaled nitric oxide and the placebo group are shown in Figure 1.

**ANTHROPOMETRIC MEASUREMENTS**

Median height and head circumference were similar in the two groups, as were their respective CDC-referenced, median z scores (Table 4). In contrast, the group given inhaled nitric oxide had a significantly higher median weight than the placebo group

**Table 4. Neurodevelopmental and Anthropometric Outcomes.**

| Outcome                                             | Inhaled Nitric Oxide (N=70) | Placebo (N=68) | P Value |
|-----------------------------------------------------|-----------------------------|----------------|---------|
| <b>Abnormal neurodevelopmental outcome</b>          | 17 (24)                     | 31 (46)        | 0.01    |
| Disability — no. (%) <sup>*</sup>                   | 6 (9)                       | 8 (12)         |         |
| Cerebral palsy                                      | 6 (9)                       | 7 (10)         | 0.78    |
| Blindness                                           | 0                           | 2 (3)          | 0.24    |
| Hearing loss                                        | 0                           | 1 (1)          | 0.49    |
| Delay without disability — no. (%)                  | 11 (16)                     | 23 (34)        |         |
| <b>Bayley scores<sup>†</sup></b>                    |                             |                |         |
| Mental Developmental Index score <70 — no. (%)      | 13 (19)                     | 24 (36)        | 0.03    |
| Psychomotor Developmental Index score <70 — no. (%) | 9 (13)                      | 12 (18)        | 0.48    |
| Either score <70 — no. (%)                          | 16 (23)                     | 28 (42)        | 0.03    |
| Both scores <70 — no. (%)                           | 6 (9)                       | 8 (12)         | 0.58    |
| <b>Anthropometric measures</b>                      |                             |                |         |
| <b>Height</b>                                       |                             |                |         |
| Median — cm                                         | 84.5                        | 83.9           | 0.55    |
| Interquartile range — cm                            | 81.2 to 88.5                | 81 to 88.3     |         |
| z Score for height for age <sup>‡</sup>             | -0.23                       | -0.59          | 0.32    |
| Interquartile range for z score                     | -0.83 to 0.36               | -1.25 to 0.41  |         |
| <b>Weight</b>                                       |                             |                |         |
| Median — kg                                         | 11.7                        | 10.8           | 0.04    |
| Interquartile range — kg                            | 10.5 to 13.5                | 9.5 to 12.2    |         |
| z Score for weight for age <sup>‡</sup>             | -0.49                       | -1.07          | 0.02    |
| Interquartile range for z score                     | -1.51 to 0.61               | -2.25 to -0.38 |         |
| <b>Head circumference</b>                           |                             |                |         |
| Median — cm                                         | 47.4                        | 47.5           | 1.0     |
| Interquartile range — cm                            | 46 to 49                    | 46 to 49       |         |
| z Score for head circumference for age <sup>‡</sup> | -0.33                       | -0.48          | 0.73    |
| Interquartile range for z score                     | -1.20 to 0.64               | -1.45 to 0.65  |         |

\* Some patients had more than one disability.

† Two patients (one in each group) did not undergo Bayley testing.

‡ A z score is the deviation from the mean value of the sex-specific and age-specific reference population, divided by the standard deviation for the reference population. It was generated with the use of the Centers for Disease Control and Prevention growth charts.

( $P=0.04$ ). Furthermore, the group given inhaled nitric oxide had significantly higher age- and sex-adjusted z scores for weight ( $P=0.02$ ) (Table 4).

## DISCUSSION

In this follow-up study of premature infants who were randomly assigned at birth to receive inhaled nitric oxide or placebo for seven days, patients treated with inhaled nitric oxide had a significantly lower risk of an abnormal neurodevelopmental outcome at two years of age. Increased birth weight and female sex also reduced the risk of an abnormal neurodevelopmental outcome. However, the beneficial effect of inhaled nitric oxide persisted after simultaneous adjustment for these confounding variables.

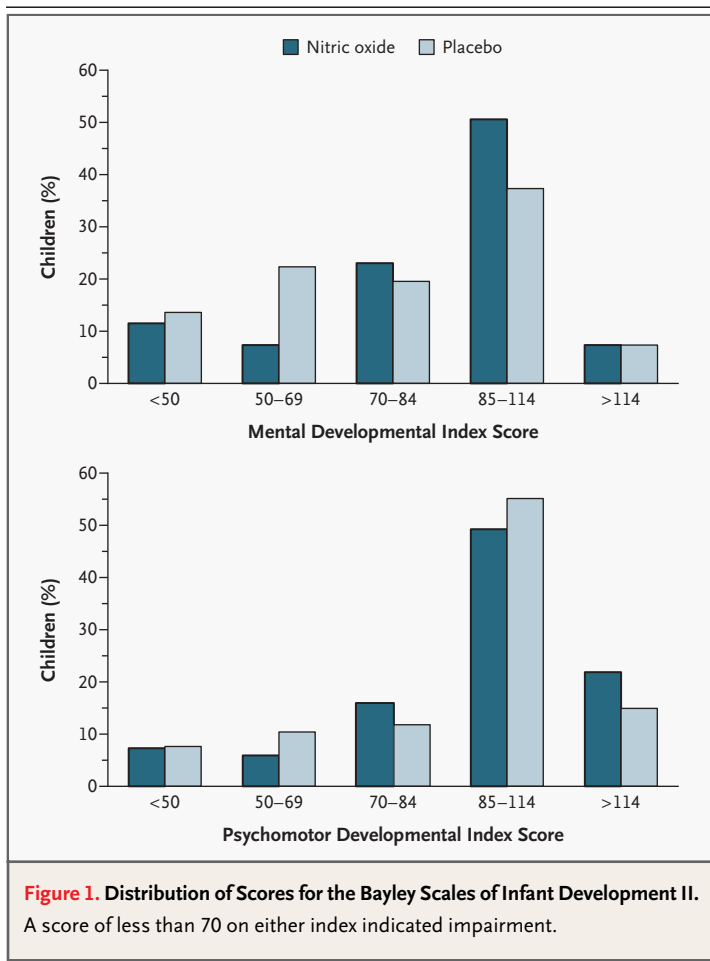
We have previously shown that inhaled nitric oxide decreases the risk of chronic lung disease and death, as well as of severe intraventricular hem-

orrhage or periventricular leukomalacia.<sup>6</sup> These conditions were also risk factors for an abnormal neurodevelopmental outcome. However, the effect of inhaled nitric oxide persisted after simultaneous adjustment for these potential intermediate variables. This observation indicates that the beneficial effect of inhaled nitric oxide on neurodevelopment is not simply explained by effects on chronic lung disease and severe intraventricular hemorrhage or periventricular leukomalacia.

The population for which we report the incidence of abnormal neurodevelopmental outcomes and cerebral palsy includes only infants who required mechanical ventilation and surfactant-replacement therapy during the newborn period. Consequently, comparison of these incidences with those in other reports is difficult, since these incidences are generally reported for all premature newborn infants, and only about 50 percent of infants weighing less than 1500 g at birth require mechanical ventilation and surfactant replacement<sup>14</sup>; the rates of these neurologic sequelae are considerably lower in premature infants who do not require such treatments.<sup>15,16</sup> Nonetheless, the rate of an abnormal neurodevelopmental outcome in our control group is similar to disability rates previously reported among children with very low birth weights and extremely low birth weights.<sup>17</sup> The incidence of cerebral palsy in our control group is also similar to previously reported rates.<sup>5,10,18</sup>

In our original study, the effect of inhaled nitric oxide on the combined incidence of chronic lung disease and death was confined to infants with initial oxygenation indexes that were less than the median. In contrast, our present data did not show a significant effect of the initial oxygenation index on the relationship between inhaled nitric oxide and neurodevelopmental outcome.

We observed that patients treated with inhaled nitric oxide had a significantly lower risk of having Bayley scores of less than 70. This difference was due to the significantly lower percentage of children in the inhaled nitric oxide group who had cognitive impairment. These findings suggest that inhaled nitric oxide primarily improves cognitive outcome. Developmental testing of premature infants at two years of age with the use of the Bayley Scales of Infant Development II provides an early opportunity to estimate future developmental capabilities.<sup>19,20</sup> However, the correlation of developmental assessments with ultimate developmental achievement is greater for assessments at five



and eight years of age.<sup>21</sup> Consequently, continued follow-up of this cohort will provide data on the robustness of the effect of inhaled nitric oxide on neurodevelopment.

How might inhaled nitric oxide given to premature infants at birth result in improved neurodevelopmental status at two years of age? Although the head circumferences of both groups of infants were similar and in the normal range, infants treated with nitric oxide had improved somatic growth and may have been healthier overall than infants given placebo, permitting better neurodevelopment. Alternatively, studies have suggested that inhaled nitric oxide may be delivered to sites outside the lung.<sup>22-24</sup> Thus, treatment with nitric oxide may have directly affected the brain through mechanisms involving the cerebral vasculature<sup>25</sup> or neuronal maturation.<sup>26-28</sup> Nonetheless, the mechanisms by which inhaled nitric oxide improves neurodevelopmental outcome remain unclear. Comparison of data from this and our previous study<sup>6</sup> with the results reported by Van Meurs et al. in this issue of the *Journal*,<sup>29</sup> suggests that further research is need-

ed to define the optimal dose of inhaled nitric oxide and the duration of treatment in premature infants. It will be of interest to see whether a difference emerges between the groups in neurodevelopmental outcomes.

In conclusion, premature infants treated with inhaled nitric oxide throughout the first week of life, as described in our study, had better neurodevelopmental status at two years of age than their placebo-treated counterparts. By increasing the likelihood of a premature infant's survival without neurodevelopmental delay, inhaled nitric oxide may significantly improve the quality of life of both the child and his or her family and may decrease the societal burden of caring for these high-risk children.

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