Physiology and predictors of impaired gas exchange in infants with bronchopulmonary dysplasia

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At a Glance Commentary:

Scientific Knowledge on the Subject: Assessment of impaired gas exchange may provide a continuous outcome measure for sensitive and equitable determination of severity of bronchopulmonary dysplasia (BPD). Previous gas exchange studies in BPD infants used

small cohorts and targeted moderate-severe BPD. These studies show right shift of the peripheral oxyhemoglobin saturation (SpO_2) versus inspired oxygen partial pressure (P_IO_2) curve and reduced ventilation-perfusion ratio reliably predict hypoxaemia in preterm infants breathing air, and further, that many infants also have a right-left shunt.

What This Study Adds to the Field: We provide measures of right shift, ventilation/perfusion and shunt, across the full spectrum of lung disease in a large (n=219) group of preterm infants. Shift increases and ventilation/perfusion decreases with increased severity of BPD as defined by the NIH classification of BPD. Shunt is primarily a feature of infants with moderate-severe BPD who require supplemental oxygen. Non-invasive bedside assessment of shift, ventilation/perfusion and shunt provide physiological continuous outcome measures of severity of respiratory disease in very preterm infants with/without BPD independent of altitude and unit practices. Routine analysis of the SpO_2/P_IO_2 curve may improve accuracy of BPD severity classification and provide a sensitive continuous outcome measure for clinical trials evaluating pulmonary outcomes.

*This article has an online data supplement, which is accessible from this issue's table of content online at www.atsjournals.org

Contributors' Statements:

Dr J Svedenkrans collected and analysed the data, interpreted the data, drafted the initial manuscript, performed literature search, drafted the figures, and approved the final manuscript as submitted. Dr B Stoecklin assisted with the REDCap database design and development, collected and analysed the data, interpreted the data, reviewed and revised the manuscript, and approved the final manuscript as submitted. Prof JG Jones developed the algorithms used for the calculation of shunt, shift and V_A/Q, reviewed and revised the manuscript, and approved the final manuscript as submitted. Prof D Doherty contributed to the overall study design and successful funding, advised on the statistical approach, reviewed the statistical analyses, reviewed and approved the final manuscript as submitted. Assoc/Prof J J Pillow was the principal investigator obtaining funding, leading study design including development of the REDCap database, obtained the study funding, verified all statistical calculations, interpreted the data, critically reviewed and revised the manuscript, and approved the final manuscript as submitted.

Abstract

Rationale

A sensitive outcome measure for infants with bronchopulmonary dysplasia would facilitate clinical benchmarking, enhance epidemiological understanding, evaluation of clinical interventions, and outcome prediction.

Objectives

Non-invasive assessment of pulmonary gas exchange in preterm infants with and without bronchopulmonary dysplasia to grade disease severity and to identify determinants of impaired gas exchange.

Methods

Prospective observational study in very preterm infants. Inspired oxygen pressure was decreased stepwise to achieve oxygen saturation from 95-86%. Right shift, ventilation/perfusion ratio and right-left shunt were derived from the resulting oxygen dissociation curve and compared to current disease severity classification. Potential determinants of shift, ventilation/perfusion and shunt were identified using principal components analysis and multiple linear regression.

Measurements and Main Results

219 infants with median (IQR) gestation of $28^{0}(26^{0}\text{-}29^{0})$ weeks had a valid study at $35^{4}(34^{7}\text{-}39^{3})$ weeks' postmenstrual age. Shift increased and ventilation/perfusion decreased as severity of bronchopulmonary dysplasia increased. Infants with moderate-severe disease also had increased shunt. Extent of impaired gas exchange overlapped between severity groups. Infants requiring mechanical support but no supplemental oxygen at 36 weeks' postmenstrual age had similar values of shift, ventilation/perfusion and shunt to preterm infants without bronchopulmonary dysplasia. Lower gestation and increased duration of invasive ventilation independently predicted increased shift, decreased ventilation/perfusion and increased shunt. Shift was the most sensitive and specific index of the severity of bronchopulmonary dysplasia.

Conclusions

Most infants with bronchopulmonary dysplasia have impaired oxygenation quantified by a simple, sensitive bedside testShift of the SpO_2/P_IO_2 curve may be useful for prediction and measurement of preterm infant respiratory outcomes.

Abstract Word Count: 250

Key words for indexing: infant, premature; ventilation, mechanical; oximetry; ventilation-perfusion ratio; oxygen inhalation therapy.

Introduction

Bronchopulmonary dysplasia (BPD) is a respiratory disorder in preterm infants characterised by impaired development of alveoli and pulmonary capillaries. Impaired development of the lung parenchyma may have life-long implications for respiratory health (1). In this regard, and oddly without mentioning arterial oxyhemoglobin saturation (SaO_2), the National Institute of Health (NIH) defined BPD in 2001 as a need for supplemental oxygen for at least 28 days. Again without mention of SaO_2, the NIH defined severity of BPD in very preterm infants by the level of supplemental oxygen required at 36 weeks' postmenstrual age (PMA). The definition of severe BPD includes a need for pressure support regardless of P_IO_2. The odd conclusion is that following these criteria, infants breathing air have mild BPD; moderate BPD is a P_IO_2 of 22-29 kPa, and severe BPD is a $P_IO_2 \geq 30$ kPa or pressure respiratory support, even without supplemental oxygen (2).

Walsh and colleagues added another confusing dimension by proposing an alternative "physiological" BPD classification (3), that ultimately defined BPD as "a $SpO_2 \geq 90$ % at 36 w PMA (4)"; confusing because SpO_2 in healthy infants is ≥ 96 %. Hence the Walsh definition misclassifies infants with a mild impairment of oxygenation as normal.

More recently, a NICHD workshop on BPD in 2016 proposed a revised definition of BPD (5). However, the suggested definition of BPD continues to define BPD by its treatment rather than by functional or pathophysiological basis, and does not address the limitations of the old 2001 definition outlined above.

An alternative, non-invasive approach for assessing pulmonary outcomes after preterm birth measures SpO_2 when P_IO_2 is reduced stepwise, and from the shape and position of the SpO_2 vs P_IO_2 curve derives the right shift from the position of the oxyhemoglobin dissociation curve, reduced ventilation/perfusion ratio (V_A/Q), and shunt (Figure 1) (6, 7). This approach was initially described in 1993,(6) then first applied to neonates in 2001(8). Subsequently, shift of the SpO_2 vs P_IO_2 curve was used to determine the P_IO_2 needed to achieve a SpO_2 between 86 % and 94 %, as an objective measure of BPD severity in

infants with moderate to severe BPD (9). Previous studies in preterm infants are limited by the small cohort size (≤32 infants) (9-12), a focus on infants with moderate to severe BPD,(9-11) and compensation for adult rather than fetal hemoglobin.(9-12). Full utility and application of the SpO_2 vs P_IO_2 approach requires application of the methodology across the full range of severity of BPD.

We derived the right shift of the SpO_2 vs P_IO_2 curve, reduced V_A/Q, and right to left shunt in a large cohort of preterm infants representing the NICHD spectrum of BPD severity: we used paired measurements of SpO_2 and P_IO_2 at 36 weeks' postmenstrual age in 219 infants less than 32 weeks' gestation. We ascertained the factors that might influence impaired gas exchange at 36 weeks' postmenstrual age. We hypothesised that a) this approach would highlight the inconsistencies between the NIH BPD classification levels and functional impairment of oxygenation; and that b) gestation and duration of invasive ventilation would be the primary independent factors influencing shift of the SpO_2 vs. P_IO_2 curve at 36 w postmenstrual age, whilst the presence of significant right to left shunt would be restricted to infants with moderate to severe BPD as defined by the NIH (2). Some of the results of these studies were reported previously in the form of abstracts (13, 14).

Methods

Study design and ethics approval

This study was a prospective evaluation of BPD severity in unsedated preterm infants at 36 w postmenstrual age. The study was approved by the Women and Newborn Health Service Human Research Ethics Committee (HREC: 1883EW and 20130193EW) and the University of Western Australia (RA/4/1/5942 and RA/4/1/426).

Study participants

Preterm infants were recruited from the Neonatal Critical Care Unit at King Edward Memorial Hospital for Women in Perth, Western Australia (KEMH) (Figure 2). Eligible infants were those born at KEMH before 32 w gestation between 21st of July, 2013 and 8th of January, 2017 with informed consent from a parent or guardian. All included infants were a part of the Preterm Infant Functional and Clinical Outcomes (PIFCO) study (ACTRN126130010627181). Infants were excluded if they had a major congenital malformation.

Outcome assessment

Infants were tested at 36 w PMA or immediately prior to hospital discharge, whichever occurred sooner. Infants who were intubated, or clinically unstable at 36 w PMA were tested within three weeks, as soon as clinical instability had resolved. Infants were studied supine during sleep, or during quiet consciousness.

Paired measurements of SpO_2 and P_IO_2

All studies were obtained at sea level at which the percentage of inspired oxygen closely approximates P_IO_2 (kPa). Inspired oxygen concentration was measured using a calibrated oxygen analyser (Model: AX-300, Teledyne Analytical Instr. California). SpO_2 was measured from the infant's right hand (MasimoSET® Radical-7™, Masimo Corporation, Frenchs Forest/NSW); a valid SpO_2 measurement was defined as a good pulse wave form on the monitor without movement artefact. Measurements of SpO_2 and P_IO_2 were recorded digitally (Powerlab, ADInstruments, Bella Vista, NSW) and monitored and analysed within LabChart (v7, ADInstruments).

The test was usually performed using a head box with a continuous fresh gas flow of 6 L/min commencing at the prescribed P_IO_2. Infants receiving CPAP or humidified high flow with supplemental oxygen above 25 kPa were tested bedside using adjustments to the air/oxygen blender whilst continuing on the prescribed respiratory support with a closed mouth.

P_IO_2 was reduced in ~5 steps of 1-3 kPa, to achieve SpO_2 ranging from ~95-86 %. Average SpO_2 and P_IO_2 were determined from a one minute recording obtained four minutes after each change in P_IO_2. P_IO_2 was decreased below 21 kPa for infants with a SpO_2 of ≥ 90 % in air, by mixing air (20·8 kPa O_2) with a mixture of 14 kPa oxygen in nitrogen (BOC, Perth, Western Australia, Australia). The lowest permissible P_IO_2 and SpO_2 were 14 kPa and 86 % respectively. Hemoglobin was determined from a capillary or venous blood gas obtained within three days of the study.

The SpO_2 vs P_IO_2 curve was plotted from paired values of SpO_2 and P_IO_2 and compared to the expected SpO_2 vs P_IO_2 curve, using the neonatal oxyhemoglobin dissociation curve as reference (see Figure 1). Shift, V_A/Q and right to left shunt were derived using the Lockwood algorithm, which derives results for each dataset from both 1) a two compartment model (shunt, shift and V_A/Q of a single homogeneous ventilated compartment) and 2) a three compartment model (shunt, V_A/Q for each of two homogeneous ventilated compartments and the relative perfusion of each ventilated lung region) (15). The current hemoglobin level was incorporated in the computations.

Data management and statistical methods

Recruitment and study data were collected prospectively and managed using Research Electronic Data Capture (REDCap) software hosted at The University of Western Australia (16). Analysis included all infants with a valid study.

Descriptive statistics are presented as mean (SD) for variables with normal distribution and with median (IQR) for variables without normal distribution. Infant characteristics (excluded and tested) were performed using t-test, Mann-Whitney U-test and chi^2-test, as appropriate. The relation between the outcome variables (shift, V_A/Q and shunt) and the infant's NIH BPD classification were determined using one way ANOVA. The threshold level that optimised sensitivity and specificity for defining mild, moderate or severe BPD was calculated from a receiver operating curve as $\sqrt{(1-\text{Sensitivity})^2+(1-\text{Specificity})^2}$.

Potential explanatory variables for the outcome variables of shift, V_A/Q and shunt were examined for normal distribution (Shapiro-Wilk) and collinearity (Variance Inflation Factor). Skewed data were transformed to meet assumptions required for linear regression. Postnatal explanatory variables collinear with maturity at birth were regressed against gestation, and the unstandardized residuals saved as an independent linear measure of the variable of interest.

Potential independent perinatal factors influencing shift, V_A/Q and shunt were identified from univariate regression. Principal component analysis was used to examine the key variance factors (Eigenvalue >1·0) within the cohort and to address residual multiple collinearity between potential explanatory variables. The factor with the highest score of the rotated component matrix and least overlap with other factors was selected for stepwise multiple linear regression. Model fit is reported as adjusted R^2. Effect size is reported as B (95 % CI). Statistical significance was defined as $p<0·05$. Data were analysed using SPSS (v25; IBM Corp, USA).

Results

Cohort description

The flow pathway describing eligibility, recruitment and enrolment into the study is shown in Figure 2. Characteristics of eligible, recruited and studied infants are shown in Table 1. Studied infants are further characterised according to 2001 NIH BPD severity classification in Table 2.

Relation of oxygen dissociation curve outcome variables to NIH BPD Severity Classification

Tests were completed within 25-30 minutes (5 steps of 5 min each). A valid test was obtained from each infant tested. Shift, V_A/Q and shunt derived from the neonatal oxyhemoglobin curve are shown according to NIH BPD classification in Figure 3; the median (IQR) shift, V_A/Q and shunt are presented in Table E1 (online data supplement). Data from all included infants were best fitted to a two compartment model,

indicative of homogeneous gas exchange (15). Outcomes using the adult oxyhemoglobin curve as a reference are provided in Table E2 (online data supplement) for comparative purposes.

Shift was increased and V_A/Q was decreased in infants with mild, moderate or severe BPD compared to no BPD, and for infants with severe BPD compared to mild BPD. Shift was also increased in moderate compared to mild BPD. The difference in shift and V_A/Q between infants with moderate and severe BPD was not statistically significant.

To understand the basis for the overlap in 95 % confidence intervals between moderate and severe BPD, we characterised the severe BPD group (n=33) according to oxygen requirement alone: 3 infants had received less than 28 d supplemental oxygen and were not receiving supplemental oxygen at 36 w PMA; 3 had received at least 28 d supplemental oxygen but were not receiving supplemental oxygen at 36 w PMA; 11 received at least 28 d supplemental oxygen but were requiring less than 30 % oxygen at 36 w PMA; with the remaining 16 infants all requiring at least 30 % oxygen at 36 w PMA. Severe BPD infants not requiring supplemental oxygen at 36 w PMA had significantly lower shift and higher V_A/Q values than the severe BPD infants requiring oxygen (Table E3, online data supplement). Infants classified as having severe BPD by the NIH criteria who were not receiving oxygen at 36 w PMA had shift and V_A/Q values that were not statistically different to infants with no BPD ($p = 0.175$ and $p = 0.389$ respectively), suggesting absence of significant parenchymal pathology despite their BPD classification.

Shunt was increased in infants with severe BPD compared to infants with no BPD or mild BPD, and in infants with moderate BPD compared to infants with no BPD. There was no difference in shunt between infants with no BPD or mild BPD, or between moderate and severe BPD. Shunt in infants with severe BPD not requiring oxygen at 36 w PMA (Table E3, online data supplement) was not significantly different from shunt in infants with no BPD or mild BPD ($p = 1.0$ and $p = 1.0$). There was no difference in shunt between infants classified as severe BPD requiring or not requiring supplemental oxygen ($p = 0.153$).

Shift was the outcome variable with the greatest area under the curve as an indicator of the presence of any BPD or moderate-severe BPD on ROC analysis (Table 3). Restriction of the ROC analysis to define a threshold shift value for moderate to severe BPD according to requirement for supplemental oxygen at 36 w PMA alone resulted in a small increase in the threshold shift level, with increased sensitivity and increased specificity. Specificity of the ROC was lower for the threshold shift value discriminating between infants with no BPD and any BPD.

Univariate Analyses for Potential Explanatory Variables and Principle Components Analyses

Univariate analyses of antenatal factors, perinatal characteristics, markers of disease severity, nutrition, growth and maturity at test relative to shift, V_A/Q and shunt are shown in Table E4 (online data supplement). A graph of shift versus both gestation and duration of ventilation relative to oxygen requirements at 36 w PMA is shown in Figure E1 (online data supplement). Principal components analysis identified multi-collinearity between these variables and grouped potential influential perinatal factors into four key factors defining postnatal nutrition (average daily protein, fluid and caloric intake over the first month of life), postnatal illness severity (duration of mechanical ventilation, postnatal steroids and airleak), maturation at birth (gestation), and health status at test (non-invasive ventilation and postmenstrual age at test) as shown in Table E5 (online data supplement).

Multiple linear Regression

Shift and V_A/Q were principally defined by gestational age and duration of mechanical ventilation (Table 4): together these explanatory variable accounted for 34.4 % of the variability in shift and 23.7 % of the variability in V_A/Q. Shunt was principally defined by duration of mechanical ventilation, and to a lesser extent by gestation, which together accounted for 19.2 % of the total variability in measurement of shunt (Table 4).

Discussion

We derived three indices of gas exchange (right shift of the SpO_2 vs P_IO_2 curve, reduced V_A/Q and shunt) using paired measurements of SpO_2 and P_IO_2 obtained at 36 weeks' postmenstrual age in 219 preterm infants less than 32 weeks' gestation. Key observations include the utility of shift to provide a continuous measure of impaired gas exchange, and the identification of shunt as a marker of impaired gas exchange in infants with moderate to severe BPD. Importantly, we show that infants with mild BPD using the NIH classification of BPD (oxygen for at least 28 days but not at 36 w postmenstrual age)(2) have a significant impairment in gas exchange despite being classified as "no BPD" by the Walsh test (4).

Whereas summary measures of shift, ventilation-perfusion ratio and shunt differed between BPD severity levels, there was variance within each group and considerable overlap between the NIH classification levels. This overlaps suggests that the NIH severity classification incompletely identifies or differentiates functional pulmonary pathophysiology after preterm birth. The most important explanatory variables accounting for variance in shift, V_A/Q and shunt were maturity at birth (gestation) and disease severity, exemplified by duration of mechanical ventilation after adjusting for gestational age.

Strengths of our study include the large cohort size and the use of sub-atmospheric oxygen concentrations to obtain outcome measures across the no BPD to severe BPD spectrum (2). These features distinguish this study from previous smaller ($\leq$ 32 infants), and underpowered investigations (9, 11, 12), and facilitated identification of physiological thresholds for shift, V_A/Q and shunt that align with the NIH classification of BPD severity (2).

We identified distinct differences between NIH BPD severity levels for shift except in the change from moderate to severe BPD. The significant changes in shift and V_A/Q in infants with mild BPD compared to infants with no BPD is an important observation highlighting the failings of the Walsh physiological classification of BPD (4). Non-separation of the summary measure of shift for moderate and severe BPD was due in part to the inclusion of infants classified as severe BPD with requirement for mechanical

respiratory support but no, minimal, or moderate impairment of gas exchange, as illustrated in Figure E1. We did not include a measure of airway function or control of breathing in our assessment. However, the need for ongoing positive pressure in this subset of infants may have an alternative physiological basis than parenchymal lung disease. The pathophysiological basis of respiratory support requirements is an important consideration for future development of a BPD classification to inform ongoing clinical management and outcome prediction.

Recruitment was biased to target a higher proportion of more immature infants than observed in a geographic cohort of preterm births before 32 weeks. However, the even spread of gestational age and disease severity across the cohort improved study power to identify the primary explanatory factors defining the magnitude of our outcome variables.

The studies were well tolerated by all infants. Hypoxic gas mixtures are used routinely pre-flight for assessment of need for supplemental oxygen during air travel, including our preterm infant population (17, 18). Stepwise reduction of P_IO_2 commencing the test with a SpO_2 of at least 94 %, ensured sufficient measurements were obtained to describe the upper inflection of the SpO_2 vs. P_IO_2 curve. Infants on pressure support and a P_IO_2 > 25 % remained on pressure support during the test, to avoid precipitating clinical instability. Maintenance of pressure support in the face of oxygen requirement ensured we captured the gas exchange capacity of the infant without confounding by small airway collapse. Whereas the theoretical impact of commonly used continuous positive airway pressures (5-8 cmH_2O) on pressure of inspired oxygen is negligible, the effect of such positive pressure on shift, V_A/Q and shunt needs formal evaluation.

The algorithm used to calculate shift, V_A/Q and shunt adjusts for current hemoglobin level. These outcome measures were derived using a reference curve based on fetal rather than adult hemoglobin. The fetal curve lies to the left of the adult curve and accounts for the differences between reported values of shift and V_A/Q in our population and those reported previously (9). Our approach is justified as the neonatal curve produced better fit to our data and the level of HbF is related mostly to postmenstrual age

rather than gestation and the number of previous blood transfusions received (19, 20). Compared to the cohort described by Jones et al, all of our data was best fitted to a two-compartment model, indicating homogeneous lung disease across the whole cohort, independent of disease severity. Non requirement of three-compartment modelling in our study may suggest a different population group (21), or may indicate stenting of the airways and reduction of airway obstruction resulting from the use of CPAP or humidified high flow in infants with severe BPD.

Our finding that gestation is the principal independent determinant of shift and V_A/Q and also influences shunt confirms current understanding of the negative impact of premature transition to *ex utero* life on growth and development of the lung. These determinants of shift and V_A/Q differ from those reported by Dassios et al (12). However, their smaller study (n = 24) was undertaken across a wider range of postmenstrual age (29-36 w), did not consider collinearity between potential influential factors (e.g. gestation and birth weight; postmenstrual age and study weight), and did not include regression analysis to identify independent associations. Illness severity defined by the duration of invasive ventilation not accounted for by gestation was also an important independent contributor to impaired gas exchange. Taken together, we conclude that treatments that reduce illness severity and decrease postnatal co-morbidity may achieve significant improvements in pulmonary outcomes.

Although V_A/Q is derived from shift, the latter had higher sensitivity and specificity for threshold values than either V_A/Q or shunt. Accordingly, shift may provide the best diagnostic index for assessing disease severity as a clinical benchmarking tool or as a sensitive outcome determinant for clinical trials. Importantly, shift may be determined from single paired measurements of SpO_2 and P_IO_2 without the need for an extended oxygen reduction test as undertaken in this study. Hence, shift of the oxyhemoglobin dissociation curve offers the ease of a simple, rapid and quantitative bedside assessment of impaired gas exchange that is easily adjusted for altitude (9).

Our study has several limitations. Our endpoint selection of 36 w PMA precludes comparison of preterm outcomes against those obtained in a healthy term cohort. Previously published outcomes for shift, V_A/Q

and shunt for term born infants at a median 3 days postnatal age indicate lower shift, higher V_A/Q and similarly low values of shunt relative to the preterm infants without BPD in our study.(22) Although measurements in these term infants were analysed using a reference curve accounting for adult rather than fetal hemoglobin, they may also indicate potential for improvement in gas exchange with more advanced lung development and/or maturation.

Secondly, tests were performed at a slightly lower postmenstrual age in clinically stable infants compared to more immature and sicker babies due to early discharge or inter-hospital transfers. This spread of postmenstrual age may have reduced differences between different disease severity levels. However, post-menstrual age had a non-significant contribution to the regression model and hence the effect of this bias is likely minimal within the current study.

Conclusions

Shift of the SpO_2 vs P_IO_2 curve, V_A/Q and shunt provide a simple and safe continuous physiological outcome measure for quantifying the full range of severity of lung disease in very and extremely preterm infants at 36 weeks' postmenstrual age, independent of altitude and local oxygen prescribing protocols. The presence or absence of these functional impairments are described poorly by the current BPD classifications, which perplexingly define BPD severity from non-standardised, clinician prescribed treatment using arbitrary thresholds. Even preterm infants with no BPD have mild impairment of gas exchange at 36 weeks' postmenstrual age. Infants with mild BPD have worse gas exchange than preterm infants with no BPD, despite classification as no BPD according to the Walsh physiological classification of BPD (4). Further, evidence of increased shunt in infants with moderate to severe BPD may signal a need for incorporating cardiological follow-up into planning for healthcare after initial hospital discharge.

Future studies should evaluate the repeatability and reproducibility of these measures, evaluate the use of this technique to quantify improvement or deterioration in gas exchange over time, identify its utility for clinical benchmarking, prediction of poor respiratory outcomes and as a sensitive outcome to assess the effect of clinical interventions on lung development. Validation of right shift of the SpO_2/P_IO_2 curve from

a single paired measurement of SpO_2 and P_IO_2 will facilitate global utilisation of this objective assessment for these purposes.

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Figure Legends

Figure 1. Oxygen Saturation vs. Oxygen Pressure. The figure shows a plot of oxygen saturation vs. oxygen pressure in a preterm infant (open squares, solid line) compared with a predicted normal curve (long dashed line) in a healthy infant alongside the fetal oxygen dissociation curve (ODC, short dashed line). In this example the preterm infant's curve gave a right shift of 13.5 kPa from the position of the ODC at the steepest part of the curve below 90 % SpO_2, a V_A/Q of 0.55, and a shunt of 19 %. Dotted reference lines for 90 % SpO_2 and 21 kPa oxygen are shown.

Figure 2. Flowchart describing the inclusion pathway of the studied infants. Of 1088 infants born during the study period, 43 had major congenital malformations, leaving 1045 eligible infants. Parents of 508 infants were not approached, and parental consent was not given for an additional 248 infants, resulting in 289 infants enrolled. 219 of the 291 enrolled infants had a completed test. Reasons for non-completion of the test included early transfer to another hospital (n = 59), deceased or too sick to be tested (n = 6), withdrawal of consent prior to study (n = 1), and other (n = 4).

Figure 3. Shift, VA/Q and shunt for included infants relative to NIH BPD classification. Mean values and 95 % confidence intervals for A) shift, B) V_A/Q, and C) shunt for infants with no BPD, mild BPD, moderate BPD and severe BPD. Accompanying dot plot shows the spread of individual measurements.

Tables

Table 1. Comparison of patient demographics for eligible and recruited infant subpopulations

				Included in the PIFCO cohort		p value (95 % CI)	
	all eligible infants n=1045	not enrolled n=756	enrolled n=289	tested n=219	not tested n=70	not enrolled vs enrolled	tested vs not tested
male (n, % male)	600 (57·4)	415 (54.9)	185 (64·0)	138 (63·0)	47 (67·1)	0·008	0·531
GA (median, IQR)	29·3 (27·1,31·0)	30·0 (27·0,31·0)	28·0 (26·0,30·0)	28.0 (26·0, 9·0)	29·0 (27·0,31·0)	<0·001 (1·0,2·0)*	0·001 (0·0,2·0)*
BW (mean, SD)	1217 (401)	1260 (415)	1104 (336)	1070 (318)	1209 (369)	<0·001 (103·2,210·4)†	0·002 (49·3,237·0)†
BW z-score (mean, SD)	0.01 (0·89)	0·00 (0·90)	0·01 (0·86)	0·02 (0·87)	-0.01 (0·83)	0·877 (-0·130,0·111)†	0·799 (-0·260,0·198)†
Any choriamnionitis (n, %)	153 (14·6)	109 (14·4)	44 (15·2)	37 (16·9)	7 (10·0)	0·741	0·162
Any antenatal steroid (n,%)	1002 (95·9)	721 (95·4)	281 (97·2)	212 (96·8)	69 (98·6)	0·175	0·433
Any postnatal steroids (n, %)	49 (4·7)	33 (4·4)	16 (5·5)	14 (6·4)	2 (2·9)	0·423	0·260
MV (d; median, IQR)	0·3 (0·0·18)	0·3 (0·0, 1·6)	0·5 (0, 3·5)	0·6 (0, 4·9)	0·15 (0·0, 1·0)	<0·001 (-0·30, 0·00)*	0·005 (-0·5, 0·0)*
Moderate to severe BPD (n, %)	136 (13·0)	70 (9·3)	66 (22·8)	57 (26·0)	9 (12·9)	<0·001	0·022

GA – Gestational age, BW – Birth weight, BPD – Bronchopulmonary dysplasia; MV – mechanical ventilation; * Mann-Whitney U Test, †t-test.

Table 2. Detailed Characteristics of Studied Infants

	all tested infants	no BPD	mild BPD	moderate BPD	severe BPD
N	219	133	29	24	33
MATERNAL FACTORS					
Any maternal betamethasone (n, %)	213 (96·8)	128 (96·2)	28 (96·6)	24 (100)	33 (100)
Rupture of membranes > 72 h (n, %)	37 (16·9)	28 (21·1)	4 (13·8)	1 (4·2)	4 (12·1)
Fever > 38.0 ^{0}C in labour (n, %)	13 (5·9)	5 (3·8)	3 (10·3)	1 (4·2)	4 (12·1)
Histological choriamnionitis (n, %)	121 (55·3)	62 (46·6)	20 (69·0)	17 (70·8)	22 (66·7)
BIRTH & EARLY POSTNATAL TREATMENT					
Male (n, %)	138 (63·0)	83 (62·4)	20 (69·0)	19 (79·2)*	16 (48·5)
Gestational age (mean, SD)	27·9 (26·0, 29·6)	29·1 (1·6)	25.7 (1·5)*	26·1 (1·9)*	25·6 (1·6)*
Birth weight (mean, SD)	1071 (318)	1221 (277)	908 (275)*	825 (161)*	788 (209)*
Birth weight z-score (mean, SD)	0·07 (0·85)	-0·03 (0·79)	0·45 (0·71)*	0·17 (0·98)*	0·10 (1·00)*
APGAR at 5 min (median, IQR)	8 (7, 9)	8 (7, 9)	7 (7, 9)	8 (7, 9)	7 (7, 9)
Multiple birth (n, %)	52 (23·7)	38 (28·6)	8 (27·6)	3 (12·5)	3 (9·1)
Intubated in 1st 48 h (n, %)	161 (73·5)	77 (56·9)	29 (91·9)*	24 (100)*	31 (93·5)*
Surfactant (n, %)	159 (72·6)	75 (56·4)	29 (100)*	24 (100)*	31 (93·9)*
MATURITY AT TEST					
Postnatal age (d; mean, SD)	53·1 (18·8)	41·6 (12·4)	68·9 (11·8)*	69·8 (12·2)*	73·6 (11·7)*
PMA at test (w; median, IQR)	35·4 (34·7, 39·3)	35·0 (34·6, 35·6)	35·6 (35·1, 36·3)*	35·8 (35·4, 36·1)*	36·1 (35·5, 36·7)*
POSTNATAL GROWTH AND NUTRITION					
36 w PMA weight (g; mean, SD)	2346 (433)	2195 (373)	2620 (383)*	2570 (372)*	2547 (479)*
36 w PMA weight z-score (mean, SD)	-0·68 (0·90)	-0·79 (0·88)	-0·23 (0·87)*	-0·47 (0·76)*	-0·75 (0·97)*
36 w PMA length (cm; mean, SD)	44·2 (2·16)	43·9 (2·08)	44·8 (2·08)	44·6 (1·67)	44·5 (2·80)
36 w PMA length z-score (mean, SD)	-0·99 (0·87)	-0·87 (0·86)	-0·97 (0·76)*	-1·15 (0·71)*	-1·38 (0·86)*
Weight z-score change birth to 36 w PMA (mean, SD)	-0·75 (0·71)	-0·76 (0·59)	-0·68 (0·84)	-0·63 (0·88)	-0·87 (0·86)
Fluid intake 1st 28 d (mL/kg/d; mean, SD)	150 (7·3)	152 (7·0)	149 (5·0)*	147 (8·4)*	147 (7·5)*
Caloric intake 1st 28 d (kcal/kg/d; mean, SD)	110 (13·3)	116 (10·0)	104 (12·6)*	98·3 (11·0)*	97·7 (11·0)*
Protein intake 1st 28 d (g/kg BW/d; mean, SD)	3·4 (0 ·3)	3·5 (0·3)	3·3 (0·4)*	3·2 (0·3)*	3·2 (0·3)*
Hb (median, IQR)	103 (92, 115)	100 (90, 114)	101 (92, 115)	108 (98, 119)	111 (101, 126)
DISEASE SEVERITY					
Mechanical ventilation (d; median, IQR)	0·6 (0·0, 4·9)	0·3 (0·0, 0·7)	4·3 (1·0, 16·9)*	6·6 (1·0, 25·1)*	32.0 (4·3, 44·9)*
Non-invasive ventilation (d; median, IQR)	43·3 (8·0, 61·5)	15·2 (3·0, 42·2)	64·4 (51·5, 73·9)*	63·0 (55·8, 74·2)*	69·1 (52·4, 78·8)*
Supplemental oxygen (d; median, IQR)	5·0 (0·2, 56·3)	0·5 (0·0, 3·4)	50·0 (35·0, 64·7)*	80·8 (58·4, 95·9)*	106 (78·4, 137)*
Any postnatal steroids (n, %)	14 (6·4)	0 (0·0)	0 (0·0)	1 (4·2)	13 (39·4)*
Any hyaline membrane disease (n, %)	199 (90·9)	114 (85·7)	29 (100·0)	24 (100·0)	32 (97·0)
Any air leak (n, %)	14 (6·4)	4 (3·0)	1 (3·4)*	1 (4·2)*	8 (24·2)*
Any pulmonary hemorrhage (n, %)	9 (4·1)	1 (0·8)	1 (3·4)*	5 (20·8)*	2 (6·1)*
Any sepsis (n, %)	33 (15·1)	6 (4·5)	6 (20·7)*	10 (41·7)*	11 (33·3)*
Any intraventricular hemorrhage (n, %)	43 (19·6)	20 (15·0)	7 (24·1)	7 (29·2)	9 (27·3)
Any periventricular leukomalacia (n, %)	2 (0·9)	0 (0·0)	0 (0·0)	0 (0·0)	2 (6·1)
Any necrotising enterocolitis (n, %)	5 (2·3)	1 (0·8)	2 (6·9)	0 (0·0)	2 (6·1)

BPD – bronchopulmonary dysplasia; Hb – hemoglobin; PMA – postmenstrual age; * significantly different to 'No BPD'.

Table 3: Thresholds for Shift, V_A/Q and Shunt for Any BPD and Moderate-Severe BPD

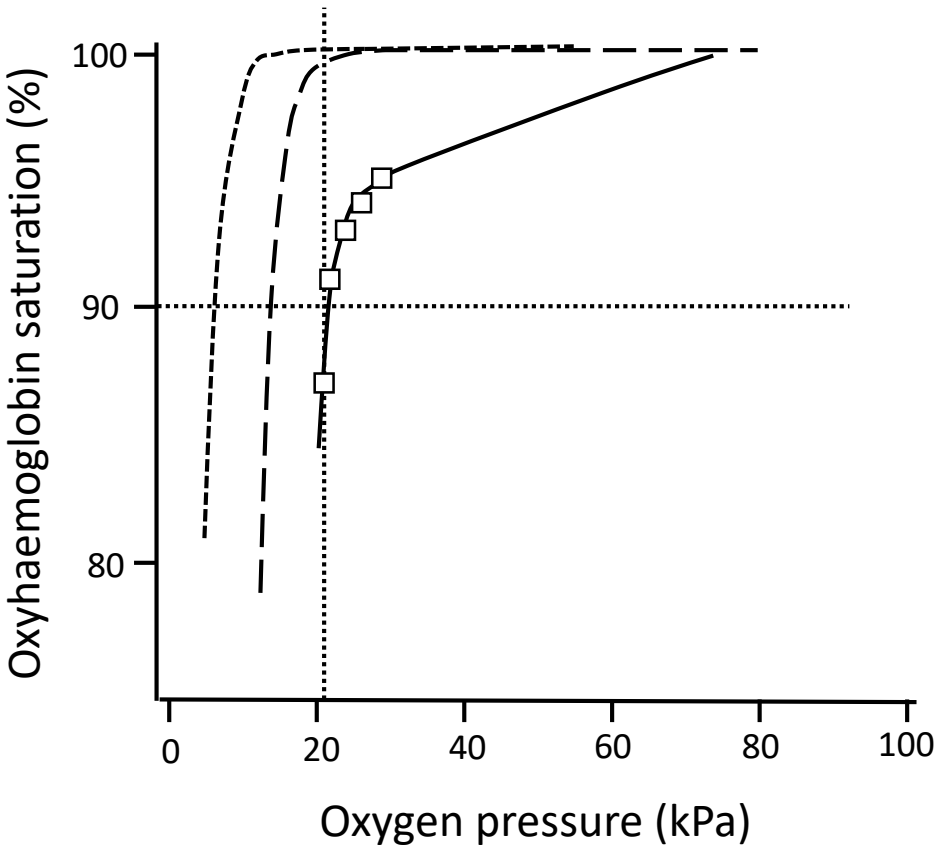
Measure	Comparison Group	AUC	95% CI	Threshold	Sensitivity %	Specificity %
Shift (kPa)						
	Any BPD	0·899	(0·858, 0·940)	11·35	78·0	79·8
	Mod-Severe BPD					
	All	0·922	(0·879, 0·966)	12·15	82·5	86·4
	O_2 @ 36 w PMA	0·951	(0·913, 0·989)	12·31	92·7	88·2
V_A/Q						
	Any BPD	0·864	(0·814, 0·915)	0·55	78·8	76·9
	Mod-Severe BPD					
	All	0.881	(0·824, 0·937)	0·54	80·7	78·4
	O_2 @ 36 w PMA	0·921	(0·875, 0·966)	0·51	82.9	87·6
Shunt (%)						
	Any BPD	0·683	(0·606, 0·761)	4·86	58·8	72·4
	Mod-Severe BPD					
	All	0·753	(0·668, 0·837)	5·25	70·8	78·8
	O_2 @ 36 w PMA	0·789	(0·696, 0·882)	8·14	68·3	87·1

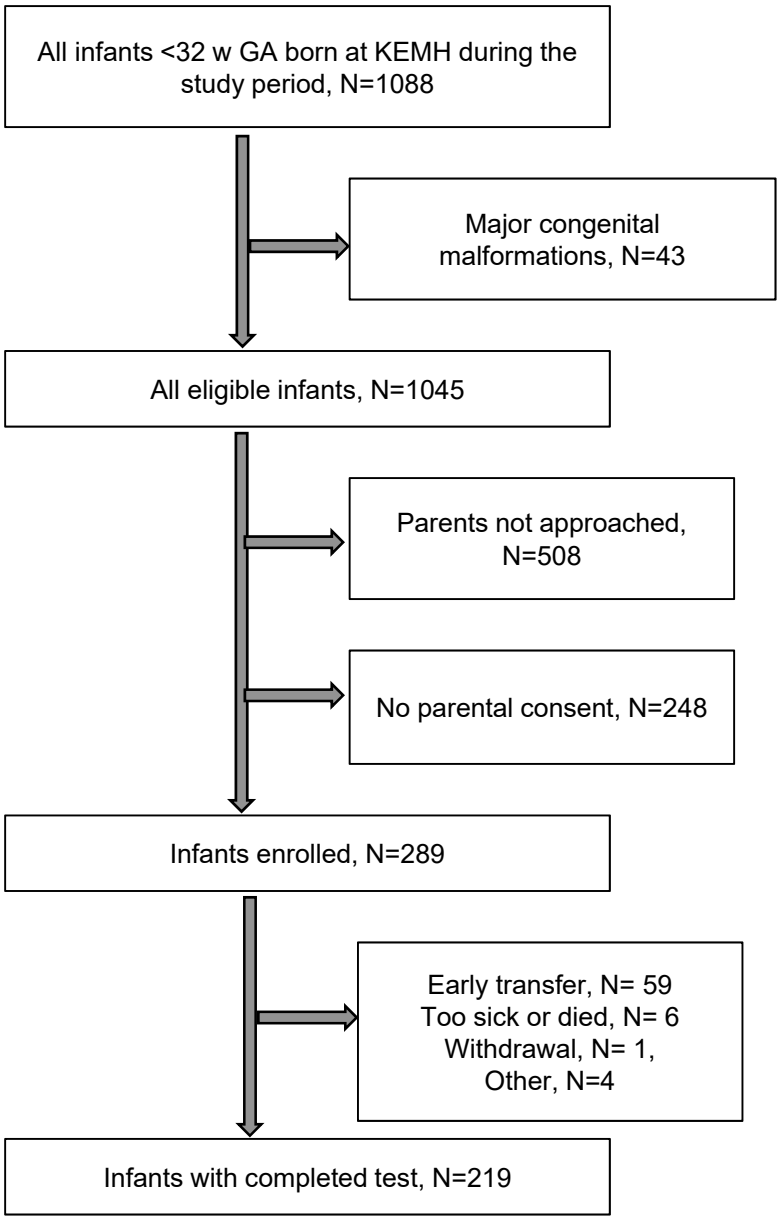
AUC, area under the receiver operating curve; BPD, bronchopulmonary dysplasia; CI, confidence interval; PMA, postmenstrual age.

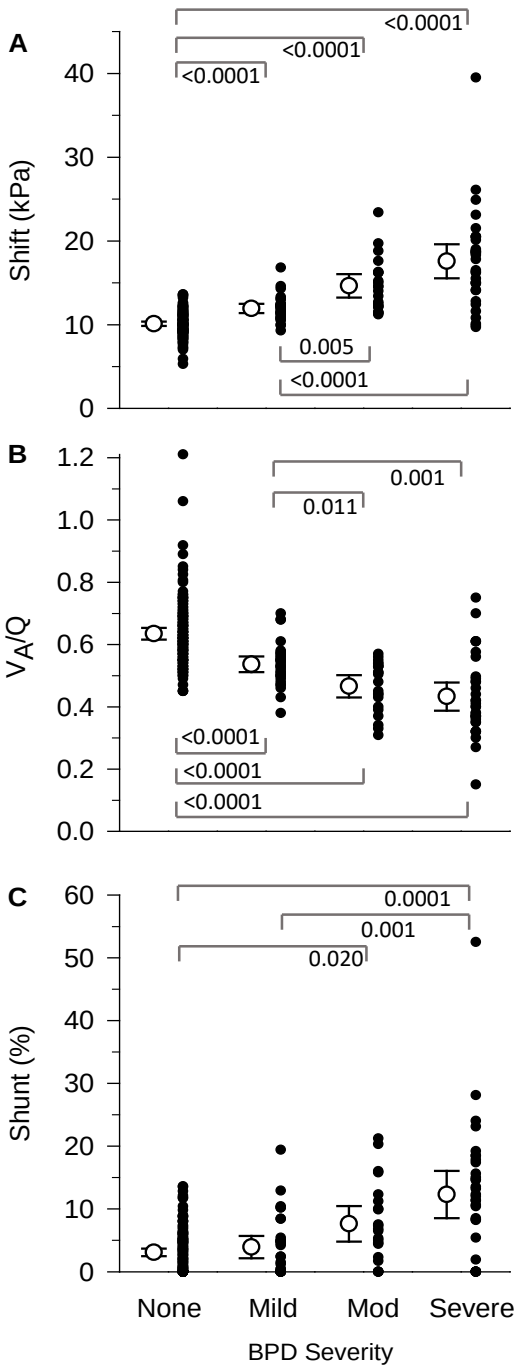
Table 4: Multivariable Analyses

Outcome Variable	Explanatory Variables	Adjusted R^2	Coefficient (B)	SE	95 % CI	p value
Right shift (kPa)	Constant		32·3	2·535	27·3, 37·3	<0·0001
	Gestational age (w)	0·192	-0·732	0·091	-0·91, -0·55	<0·0001
	Duration of mechanical ventilation (d)*	0·152	0·130	0·018	0·09, 0·17	<0·0001
V_A/Q	Constant		-0·187	0·094	-0·407, 0·032	0·094
	Gestational age (w)	0·162	0·021	0·003	0·013, 0·028	<0·0001
	Duration of mechanical ventilation (d)*	0·063	-0·003	0·001	-0·004, -0·001	<0·0001
	Average daily protein 1st 28 d (g/kg)	0·012	0·057	0·.027	0·003, 0·110	0.038
Shunt (%)	Constant		24.5	4·99	14.7, 34.4	<0·0001
	Duration of mechanical ventilation (d)*	0·136	0·209	0·035	0·137, 0·274	<0·0001
	Gestational age (w)	0·056	-0·703	0·181	-1·057, -0·348	0·0001

SE, standard error; CI, confidence interval. *unstandardised residual of regression against gestational age.


Oxyhaemoglobin saturation (%)
100
90
80
0
20
40
60
80
100
Oxygen pressure (kPa)


All infants <32 w GA born at KEMH during the study period, N=1088
Major congenital malformations, N=43
All eligible infants, N=1045
Parents not approached, N=508
No parental consent, N=248
Infants enrolled, N=289
Early transfer, N= 59
Too sick or died, N= 6
Withdrawal, N= 1,
Other, N=4
Infants with completed test, N=219


A
Shift (kPa)
0
10
20
30
40
<0.0001
<0.0001
<0.0001
0.005
<0.0001
B
V_A/Q
0.0
0.2
0.4
0.6
0.8
1.0
1.2
0.001
0.011
<0.0001
<0.0001
<0.0001
C
Shunt (%)
0
10
20
30
40
50
60
0.0001
0.001
0.020
None
Mild
Mod
Severe
BPD Severity

Physiology and predictors of impaired gas exchange in infants with bronchopulmonary dysplasia

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Online Data Supplemental Figure

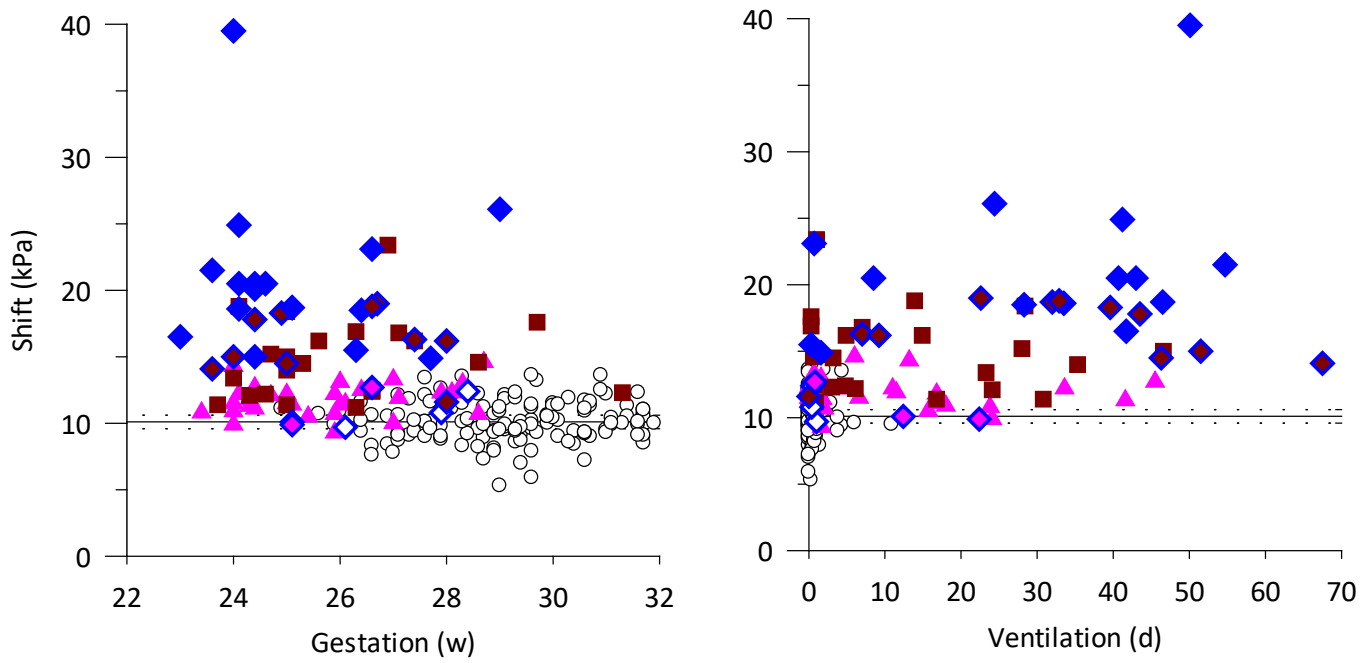

Figure E1. Major determinants of right shift of the SpO_2/P_IO_2 curve. Figure shows relation between measured right shift of the SpO_2/P_IO_2 and gestation (left panel) and duration of invasive ventilation (right panel). Symbols show NIH BPD classification: no BPD (open circle); mild BPD (pink triangle); moderate BPD (brown square) and severe BPD (diamond). The severe BPD symbols (diamond symbols, blue border) are color filled according to whether infants required oxygen for less than 28 d (white) or according to oxygen requirement at 36 w PMA in those infants requiring supplemental oxygen for at least 28 d: air (pink), < 30 % (brown) and ≥ 30 % (blue). The mean (95 % CI) for the no BPD group is shown as a solid (dashed) line for reference. This figure highlights the heterogeneity of oxygen requirements in the severe BPD group, in part accounting for the wide range of shift values in this group.

Physiology and predictors of impaired gas exchange in infants with bronchopulmonary dysplasia

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Online Data Supplemental Text

Table E1: Summary values of Shift, V_A/Q and Shunt relative to NICHD BPD Classification using the fetal oxyhemoglobin dissociation curve as a reference.

	no BPD	mild BPD	moderate BPD	severe BPD
Shift	10·0 (9·2, 11·1)	11·9 (10·9, 12·4)*[b]	14·3 (12·2, 16·2)*†	17·8 (14·1, 20·3)*†
V_A/Q	0·62 (0·56, 0·69)	0·53 (0·50, 0·57)*	0·49 (0·40, 0·53)*†	0·40 (0·36, 0·49)*†
Shunt	2·0 (0·0, 5·2)	2·4 (0·0, 5·4)	6·7 (2·2, 11·5)*	12·0 (3·7, 17·6)*†

V_A/Q – ventilation perfusion ratio; Kruskal-Wallis; Values are median (IQR). * $p< 0.05$ compared to no BPD; † $p< 0.05$ compared to mild BPD.

Table E2: Summary values of Shift, V_A/Q and Shunt relative to NICHD BPD Classification using the adult oxyhemoglobin dissociation curve as a reference.

	no BPD	mild BPD	moderate BPD	severe BPD
Shift (kPa)	8·4 (7·7, 9·6)	10·5 (9·2, 11·1)*	13·0 (11·2, 15·0)*†[c]	17·0 (13·5, 18·5)*†
V_A/Q	0·72 (0·64, 0·79)	0·58 (0·56, 0·67)*	0·51 (0·42, 0·55)[c]	0·42 (0·35 ,0·50)[c]
Shunt (%)	0·0 (0·0, 0·0)	0·0 (0·0, 0·0)	0·0 (0·0, 8·1)*†	8·1 (1·6, 11·8)*†‡

V_A/Q – ventilation perfusion ratio; Kruskal-Wallis; Values are median (IQR). Values within a row that do not share the same superscripted letter are significantly different from each other. * $p< 0.05$ compared to no BPD; † $p< 0.05$ compared to mild BPD; ‡ $p<0.05$ compared to moderate BPD.

Table E3: Summary values of shift, V_A/Q and shunt for infants classified as severe BPD according to need for oxygen at 36w PMA.

	no supplemental O_2 (n=6)	supplemental O_2 (n=25)	p value
Shift (kPa)	11·2 (9·7,18·7)	18·8 (16·6, 20·9)*	<0·001
V_A/Q	0·59 (0·54, 0·65)	0·38 (0·35, 0·44)*	<0·001
Shunt (%)	1.0 (0·0,17·5)	12·3 (8·3, 18·4)	0·153

V_A/Q – ventilation perfusion ratio; Mann-Whitney U; Values are median (IQR). Note that infants not receiving supplemental oxygen but classified as having severe BPD due to need for ongoing invasive or non-invasive respiratory support had lower shift, and higher V_A/Q than those infants with a continuing need for oxygen (supplementary Table 1). * compared to infants classified as severe BPD but not requiring supplemental O_2 at 36 w PMA.

Table E4: Pearson correlations ® of shift, V_A/Q and shunt with potential explanatory factors

	Shift		V_A/Q		Shunt	
	r	**p value**	**r**	**p value**	**r**	**p value**
MATERNAL FACTORS						
Any maternal betamethasone	0·074	0·278	-0·042	0·538	0·094	0·168
No. of courses maternal betamethasone	-0·038	0·586	0·029	0·671	0·012	0·865
Rupture of membranes > 72 h	-0·075	0·268	0·03	0·663	-0·046	0·5
Fever > 38.0 C in labour	0·065	0·34	0·014	0·84	0·053	0·433
Histological chorioamnionitis	0·055	0·416	-0·091	0·181	-0·098	0·149
BIRTH & EARLY POSTNATAL TREATMENT						
Intubated in 1st 48 h	0·042	0·54	-0·024	0·723	0·075	0·267
Multiple birth?	-0·095	0·162	0·088	0·193	-0·103	0·13
Male	-0·082	0·228	0·066	0·328	0·062	0·362
APGAR at 5	-0·103	0·128	0·104	0·123	-0·105	0·122
Surfactant given	**0·264**	<0·0001	**-0·237**	<0·0001	**0·186**	0·006
Number of surfactant doses	**0·257**	<0·0001	**-0·228**	0·001	**0·199**	0·003
MATURITY						
GA	**-0·43**	<0·0001	**0·421**	<0·0001	**-0·223**	<0·001
Postmenstrual age at test	**0·206**	0·002	**-0·189**	0·005	**0·205**	0·002
GROWTH AND NUTRITION						
Birth weight Z score	0·066	0·329	-0·05	0·461	0·061	0·370
Weight Z score at test	0·065	0·338	-0·057	0·406	0·036	0·594
Length Z score at test	-0·097	0·156	0·092	0·180	-0·051	0·461
Weight z-score change birth to 36 w PMA	0·002	0·979	-0·010	0·879	-0·023	0·733
Average fluid intake 1st 28 d	**-0·282**	<0·0001	**0·229**	0·001	**-0·175**	0·010
Average caloric intake 1st 28 d	**-0·451**	<0·0001	**0·409**	<0·0001	**-0·268**	0·042
Average protein intake 1st 28 d	**-0·310**	<0·0001	**0·286**	<0·0001	**-0·138**	<0·0001
DISEASE SEVERITY						
Mechanical ventilation	**0·584**	<0·0001	**-0·460**	<0·001	**0·432**	<0·0001
Non-invasive ventilation	**0·477**	0·002	**-0·450**	0·005	**0·298**	0·003
Any postnatal steroids	**0·470**	<0·0001	**-0·305**	<0·0001	**0·418**	<0·0001
Total days of postnatal steroids	**0·419**	<0·0001	**-0·250**	<0·001	**0·295**	<0·0001
Any hyaline membrane disease	0·118	0·083	-0·112	0·098	**0·155**	0·022
Any airleak	**0·344**	<0·0001	**-0·237**	<0·001	**0·139**	0·040
Any pulmonary haemorrhage	**0·147**	0·030	**-0·159**	0·018	0·103	0·130
Any sepsis	**0·258**	<0·001	**-0·276**	<0·0001	**0·151**	0·026
Any intraventricular haemorrhage	0·017	0·804	0·010	0·883	0·039	0·563
Any periventricular leukomalacia	0·076	0·265	-0·082	0·226	0·104	0·126
Any necrotising enterocolitis	**0·137***	0·043	-0·103	0·128	0·044	0·517

V_A/Q – ventilation perfusion; PMA – postmenstrual age. Significant correlations are in bold font.

Table E5: Rotated Principal Components Analysis Matrix

	Component			
Right shift	**1**	**2**	**3**	**4**
Duration of ventilation*	**0·864**	-0·200	0·105	-0·082
Any postnatal steroids	0·817	-0·096	-0·207	-0·005
Any airleak	0·701	-0·034	-0·196	0·063
Average fluid intake 1st 28 d	0·062	**0·767**	0·062	-0·146
Average protein intake 1st 28 d	-0·067	0·748	0·337	0·110
Average caloric intake 1st 28 d	-0·251	0·639	0·593	0·047
Any NEC	0·109	-0·528	0·185	0·059
Any Sepsis	0·237	-0·467	-0·367	-0·067
Gestation	-0·156	0·124	**0·858**	0·060
Any surfactant	0·012	-0·012	-0·774	0·157
Postmenstrual age at test	0·313	-0·123	-0·158	**0·738**
Duration of non-invasive ventilation*	-0·495	-0·204	-0·099	0·673
Any pulmonary haemorrhage	0·127	-0·341	-0·214	-0·464
V_A/Q				
Duration of ventilation*	**0·870**	-0·198	-0·146	-0·077
Any postnatal steroids	0·818	-0·115	0·193	-0·011
Any airleak	0·70	-0·038	0·202	0·052
Average fluid intake 1st 28 d	0·037	0·817	-0·198	0057
Average protein intake 1st 28 d	-0·083	**0·827**	0·095	-0·210
Average caloric intake 1st 28 d	-0·259	0·713	-0·503	0·014
Any Sepsis	0·238	-0·484	0·338	-0·054
Gestation	-0·148	0·217	**-0·852**	0·065
Any surfactant	0·006	-0·085	0·782	0·149
Postmenstrual age at test	0·324	-0·091	0·160	**0·736**
Duration of non-invasive ventilation*	-0·482	-0·184	0·078	0·691
Any pulmonary haemorrhage	0·123	-0·377	0·181	-0·451
Shunt				
Duration of ventilation*	**0·880**	-0·167	-0·096	0·120
Any postnatal steroids	0·810	-0·154	0·138	0·110
Any airleak	0·677	-0·092	0·147	0·038
Duration of non-invasive ventilation*	-0·648	-0·138	-0·092	0·533
Average protein intake 1st 28 d	-0·071	**0·851**	-0·132	0·089
Average caloric intake 1st 28 d	-0·287	0·782	-0·329	-0·091
Average fluid intake 1st 28 d	0·058	0·774	0·225	0019
Any Sepsis	0·213	-0·553	0·310	-0·001
Number of surfactant doses	0·013	-0·001	0·784	0·033
Gestation	-0·208	0·378	**-0·677**	-0·148
Postmenstrual age at test	0·193	-0·059	-0·002	**0·852**
Any hyaline membrane disease	0·004	-0·065	-0·081	-0·379

*unstandardized residual versus gestation; rotation method varimax with Kaiser normalization. Bold font indicates significant factor used for multivariable linear regression.