ERJ open research
THE BEST IN OPEN ACCESS BASIC, TRANSLATIONAL & CLINICAL RESPIRATORY RESEARCH

Early View

Review

Early Prediction of Bronchopulmonary Dysplasia: Can Noninvasive Monitoring Methods be Essential?

Xuewei Cui, Jianhua Fu

Please cite this article as: Cui X, Fu J. Early Prediction of Bronchopulmonary Dysplasia: Can Noninvasive Monitoring Methods be Essential?. *ERJ Open Res* 2023; in press (https://doi.org/10.1183/23120541.00621-2022).

This manuscript has recently been accepted for publication in the *ERJ Open Research*. It is published here in its accepted form prior to copyediting and typesetting by our production team. After these production processes are complete and the authors have approved the resulting proofs, the article will move to the latest issue of the ERJOR online.

Copyright ©The authors 2023. This version is distributed under the terms of the Creative Commons Attribution Non-Commercial Licence 4.0. For commercial reproduction rights and permissions contact permissions@ersnet.org

Early Prediction of Bronchopulmonary Dysplasia: Can Noninvasive Monitoring Methods be Essential?

Xuewei Cui, Jianhua Fu*

Department of Pediatrics, Shengjing Hospital of China Medical University, Shenyang, China

***Corresponding authors:** Jianhua Fu. Department of Pediatrics, Shengjing Hospital of China Medical University, Shenyang, China. E-mail: liying58522@163.com

Summary

This review provided a better understanding of early noninvasive prediction methods, which is essential for constructing predictive indicators of subsequent outcomes, and developing cutting-edge prediction models

Abstract

Despite remarkable breakthroughs in diagnosis and treatment, the prevalence of bronchopulmonary dysplasia (BPD) in preterm infants and the consequent mortality have remained high over the last half-century. The pathophysiology of BPD is complicated, with several causes. In addition, infants with severe BPD are predisposed to a variety of complications that need multidisciplinary collaboration during hospitalization and post-discharge home treatment. Consequently, early prediction, precise prevention, and individualized management have become the cornerstones of therapeutic care of preterm infants with BPD, thereby improving patient survival and prognosis. BPD has an operational clinical description; however, it has various clinical phenotypes and endotypes, making accurate prediction challenging. Currently, most approaches for predicting BPD in preterm infants include invasive collection of biofluids, which is inappropriate in fragile neonates. Consequently, researchers and clinicians are becoming more interested in noninvasive monitoring for BPD prediction. Comprehensive assessments of pertinent research, however, remain scarce. In this review, we compared many noninvasive monitoring techniques that contribute to early prediction of BPD development in premature infants.

Keywords: bronchopulmonary dysplasia, early prediction, noninvasive, biomarkers, monitoring methods, preterm infants

Background

With continuous advancements in perinatal medicine and neonatal intensive care unit (NICU) medical procedures over the last three decades, there has been a steady rise in the survival rate of preterm infants, particularly very-low-birth-weight infants (VLBWIs) and extremely-low-birth-weight infants (ELBWIs) [1]. Bronchopulmonary dysplasia (BPD) was initially identified by Northway et al. in 1967 [2]. Despite impressive advancements in the diagnosis and management of BPD, there has not been a substantial drop in its incidence during the subsequent five decades [3], with up to 45% of premature infants at less than 29 weeks of gestational age (GA) suffering from BPD [4]. Presently, the National Institute of Child Health and Human Development (NICHD) (2001 and 2018) criteria for BPD are widely used [5, 6]. Since BPD has an operational clinical description, it is impossible to apply an objective definition of the disease to reliably predict its prognosis in terms of mortality and morbidity [7]. The “old” BPD has become less common due to the use of pulmonary surfactant (PS) and relatively sophisticated mechanical ventilation protocols, whereas the “new” BPD is characterized by alveolar hypoplasia, which is identified by a decrease in the number and an increase in the size of alveoli, simplification of structure, and pulmonary microvascular dysplasia. Furthermore, the etiology of BPD is multifactorial. Different potential prenatal and postnatal risk factors may contribute to BPD [6] (Fig. 1).

BPD remains one of the primary causes of mortality in very preterm infants (VPIs). Premature infants with BPD have a significantly higher morbidity of complications, high incidence of long-term neurodevelopmental prognosis, and severe compromise on quality of life. Therefore, early prediction, accurate prevention, and individual management of BPD have become major challenges in the field of perinatal and neonatal medicine [8]. Ideal BPD biomarkers would target causative pathways involved in BPD pathogenesis, such as biomarkers of immunomodulatory and inflammatory responses, angiogenic growth, epithelial damage and fibrosis, oxidative stress, vascular injury, and pulmonary hypertension. Among them, early noninvasive detection of BPD using measures such as

biofluids biomarkers, exhaled breath condensates, radiographical studies, lung function, and future individualized biomarkers "OMICS," have been the focus of extensive investigation in the field. Although noninvasive methods may not precisely represent the lung and blood concentrations of the respective biomarkers, the application of noninvasive methods will not damage the integrity of the skin and mucous membranes of the infants and can reduce their pain and facilitate continuous measurements. However, comprehensive reviews of pertinent research in the field remain scarce. Therefore, this review aimed to investigate recent advancements in noninvasive monitoring approaches for early prediction of BPD progression and severity within the first week of life, even at birth, with the goal of improving personalized therapy and short- and long-term prognoses (Fig. 2).

1 Biofluid Biomarkers

In recent years, there has been a rapid progress in research on the pathophysiology of BPD, with the focus largely being on inflammation, oxidative stress, pulmonary interstitial fibrosis, epithelial differentiation. Furthermore, the biomarker potential of different cytokines and growth factors that influence lung development or may be associated with lung damage in preterm infants have been investigated [9]. For the most part, the noninvasive prediction approach has been studied using umbilical cord blood (UCB), urine, or tracheal aspirate (TA) samples [10, 11].

UCB Biomarkers

In the examination of preterm infants prone to BPD, the UCB is possibly one of the simplest and earliest samples to obtain. The UCB screening provides information about fetal disturbances that cause BPD in preterm births. However, they may not precisely represent lung concentrations of the respective biomarker, and it is not feasible to determine whether pulmonary cells produced or released them (Table 1).

1.1.1 Immunomodulatory and Inflammatory Factors

Interleukin (IL)-6 is a cytokine associated with inflammation, immunological control that binds to receptors and acts on the signaling membrane glycoprotein (gp) 130, activating a cascade of downstream signals and regulating gene expression [12]. A study evaluating the levels of IL-6 and soluble gp130 (Sgp130) in the UCB of 134 preterm infants found that these biomarkers may be used as independent predictors of BPD. The area under curve (AUC) was 0.849 when IL-6 was > 46.125 pg/ml, demonstrating a positive predictive efficacy on BPD [13].

CD4+ T-cells play an essential role as immune system mediators. It is well accepted that CD4+ regulatory T-cells (Tregs) have an immunological suppressive function, and are essential in maintaining immune homeostasis [14]. In addition, Tregs are engaged in a variety of chronic, inflammation-mediated respiratory disorders [15]. Pagel compared Tregs frequencies in newborns at < 29 weeks of GA with and without BPD (n = 57 and 56, respectively). Increased Treg frequencies were seen in preterm infants with BPD on day of life (DOL) 4-10 (P = 0.0343) [16]. Mishra et al. [17] detected CD4+ T-cells in UCB mononuclear cells in preterm infants with GA of <32 weeks. The total number of Tregs and non-Tregs in preterm infants with moderate BPD was considerably lower than the number of the aforementioned cells in those with mild or no BPD, indicating that BPD is caused by inflammatory alterations ($P < 0.05$).

1.1.2 Angiogenic Growth Factors

Given that pulmonary microvascular dysplasia is a potential etiology of “New BPD”, aberrant angiogenesis may have a role in the onset of BPD [18]. During angiogenesis, the angiopoietin (ANG)/Tie-2 ligand/receptor system communicates with the vascular endothelial growth factor (VEGF) pathway to determine the development of blood vessels [19].

A prospective study of 102 preterm infants (GA < 32 weeks) found that ANG-1 concentrations of UCB were considerably lower while levels of endostatin were significantly higher in infants with BPD than in those without BPD ($p < 0.001$). The levels of ANG-1 in the UCB were inversely

associated with endostatin levels in newborns diagnosed with BPD (r = -0.48; p = 0.008) [20]. Endostatin, a 20-kDa angiogenesis inhibitor that is a proteolytic fragment of collagen XVIII's C-terminal non-triple helical domain, plays a role in angiogenesis. In VLBWIs with GA ＜ 32 weeks, those who developed BPD had higher circulating levels of endostatin in the UCB than those who did not develop BPD (100.7±29.7 ng/mL vs. 85.6±28.7 ng/mL; p = 0.029) [21]. These findings suggest that prenatally reduced ANG-1 and high endostatin levels in the UCB are risk factors for the development of BPD.

Placental growth factor (PlGF), a VEGF family member, regulates angiogenesis via the modification of VEGF activity by competing to bind with fms-like tyrosine kinase 1 (Flt-1). Apart from its angiogenic action, the overexpression of PlGF in transgenic mice increased alveolar type II cell death, resulting in expanded airspace and pulmonary emphysema, which is pathologically comparable to BPD [22]. Tsao et al. [23] revealed that a higher level of PlGF in UCB was significantly and independently associated with a higher risk of BPD. The PlGF cut-off value for predicting BPD was 17 mg/dl, with a specificity of 95%, a sensitivity of 53%, and a positive predictive value of 83%. However, extremely preterm infants with BPD-PH at a corrected age of 36 weeks had significantly lower UCB levels of PlGF and granulocyte-colony stimulating factor (G-CSF) than the subgroups of infants with BPD only and those without BPD or PH. Analysis of the receiver operator characteristic (ROC) curve demonstrated that reduced levels of PIGF and G-CSF were good indicators of BPD-PH (AUC= 0.83 and 0.76, respectively) [24].

Endoglin (ENG), a co-receptor of transforming growth factor (TGF) family members such as TGF-β1 and TGF-β3, is highly expressed in cell membranes of vascular endothelium [25]. Placental ENG is upregulated in preeclampsia, releasing soluble ENG (sENG) into the maternal circulation. sENG, which is a truncated form of the extracellular domain of ENG, has an antiangiogenic effect that may inhibit TGF-ß signaling in the vasculature [26]. Circulating sENG, which has antiangiogenic

properties, may promote endothelial dysfunction [27]. Elevated concentrations of sENG (>3420 pg/mL) in UCB were associated with an increased risk of severe or moderate BPD in the preeclampsia group, as determined by multivariate logistic regression analysis in a retrospective cohort study of 199 singleton newborns (GA < 32 weeks, adjusted odds ratio [OR] 11.9, P = 0.006) [28].

Endothelial colony-forming cells (ECFCs) are a type of circulating and resident endothelial cells that may self-renew and generate new vessels. Abnormal ECFCs function may lead to alveolar growth retardation [29]. Baker et al. [30] discovered that the ECFCs level in the UCB was significantly lower in preterm infants who later had moderate or severe BPD than in those who did not (P<0.001). All patients with BPD had lower ECFCs than those without BPD (P<0.05).

During the initial phases of lung development, fibroblast growth factor-10 (FGF-10) modulates branching morphogenesis. FGF-10 promotes the elongation and branching of developing airways [31]. A low concentration of FGF-10 in UCB is independently related to the development of BPD and its severity [32].

1.1.3 Epithelial and Fibrotic Markers

The generation of alveoli during lung development is dependent on the synthesis, maturity, and remodeling of pulmonary epithelial cell markers as well as extracellular matrix proteins [33-35]. These two may be involved in alveolar hypoplasia, which is a crucial pathologic characteristic of the "New BPD".

Epithelial cells known as Clara cells border the respiratory and terminal bronchioles. Clara cell secretory protein (CCSP) of 10–16 kDa (CC16 or CC10) belongs to the family of secretoglobins [36]. CC16 has been demonstrated to play a role in lung repair by significantly reducing pulmonary inflammation and improving airway regeneration in animal studies, in addition to stimulating epithelial proliferation and protecting against oxidative stress [37, 38]. Schrama et al. [39] inferred that preterm infants with low CC16 concentrations in UCB were at an increased risk of developing

BPD. Low CC16 levels may indicate the onset of lung damage, an important factor in BPD progression.

Krebs yon den Lundgen-6 (KL-6) is a high-molecular-weight gp that is predominantly produced by type II alveolar cells and induces lung fibrosis by acting as a fibroblast chemotactic factor [40]. A case-control study including 74 preterm infants of GA <32 weeks revealed that the cord plasma KL-6 concentrations in the BPD group were substantially higher than those in the non-BPD group ($P<0.05$). It were shown to be strongly linked with the duration of oxygen exposure ($r = 0.502$, $P = 0.024$). This demonstrates that KL-6 is elevated in BPD and objectively represents disease severity [41].

Matrix metalloproteinases (MMPs) have crucial roles in a variety of pathological processes, including inflammation, cardiovascular problems, and respiratory system disorders [42]. Tissue inhibitors of metalloproteinases (TIMPs) regulate the action of MMPs. MMP-9, a core component of the MMP family, is a prominent element of the basement membrane of the airways [43]. Preterm infants with moderate/severe BPD have greater MMP-9/TIMP-1 ratios in the UCB than those with mild/no BPD [44].

1.1.4 Other Factors

Vitamin D, a steroid hormone, has antifibrotic, antioxidative, and anti-inflammatory properties. In addition, it stimulates the production of alveolar surfactants, and is involved in cell proliferation, cell differentiation, and fetal lung development and maturation [45, 46]. Moreover, vitamin D insufficiency at birth is associated with poor respiratory outcomes, particularly BPD in preterm births [47, 48]. The phenolic hydroxyl group linked to the benzene ring of vitamin E is primarily responsible for its antioxidant effects [49]. UCB levels of vitamins D and E were significantly lower in the BPD group than in the non-BPD group, and this difference was also associated with BPD severity. The vitamin D and E levels were shown to be inversely linked with the period of oxygen

support required for preterm newborns with BPD [50]. A retrospective research revealed that a 25(OH)D level of 15.7 ng/ml in the UCB was predictive of BPD development (AUC = 0.585, 95% CI: 0.523-0.645, P = 0.016) [51]. The positive predictive value was 57.1% , whereas the negative predictive value was 63.3%.

Urinary Biomarkers

Convenience, noninvasiveness, and the ability to get a large volume of specimens for repeated measurements are all benefits of urine screening tests [52]; however, the lack of information on urine protein profiles in preterm infants and high variability of protein concentrations present challenges when trying to standardize urinary biomarkers [53] (Table 2).

1.1.5 Inflammatory Markers

Leukotriene E4 (LTE4) is an inflammatory marker that is associated with the severity of bronchial asthma and may be identified in the blood and urine. It is recognized as a stable and dependable byproduct of the destruction of the cell membrane's phospholipid layer [54]. According to a study, urinary LTE4 concentrations on DOL 7 were higher in "atypical" BPD than in "classic" BPD (P = 0.006). These findings imply that continuous inflammatory process may be a key factor in "atypical" BPD [55].

B-2-microglobulin (B2M) is a low-molecular-weight protein that is expressed on the surface of lymphocytes and macrophages, as a part of the class I major histocompatibility complex (MHC), and is amplified by systemic immunological activation [56]. Shima et al. [57] investigated the relationship between B2M urine excretion levels at birth and the presence of BPD in preterm (24–28 weeks) singleton newborns and discovered that those with BPD had substantially higher median B2M levels than those without BPD (P<0.0001). After adjusting for GA and other confounding variables, the determined cut-off value of urine B2M at birth (19.6×10^4 mg/ g Cr; AUC, 0.800; P<0.0001, a positive predictive value of 69% and a negative predictive value of 85%) was associated with the development of BPD.

1.1.6 Oxidative Stress-Related Markers

Beginning with acute inflammatory damage (as in respiratory distress syndrome [RDS]), oxidative stress contributes to lung injury that ultimately results in pulmonary microvascular remodeling, poor alveolarization, and ultimately, BPD [58]. Multiple studies have demonstrated the role of oxidative stress in the development of BPD [59].

8-hydroxy-2'-deoxyguanosine (8-OHdG) is the most widely used biomarker of endogenous oxidative stress-related DNA damage [60]. We found that urine 8-OHdG levels were substantially higher in the BPD group from DOL 7–28 than in the control group ($P<0.05$) in a prospective cohort study of 165 preterm babies. Furthermore, 8-OHdG (DOL 14–28) levels were shown to be directly correlated with the development of BPD ($P<0.05$) [61]. Additionally, one urine lipid oxidation product, 11-dehydro-thromboxane B2 (TXB2), was higher in BPD participants ($P=0.05$) [62].

1.1.7 Pulmonary Neuroendocrine Markers

BPD has been associated with an increase in pulmonary neuroendocrine overexpressed bombesin-like peptide (BLP). Furthermore, in BPD, BLP regulates alveolarization and angiogenesis [63]. The correlation between hyperoxia, airway remodeling, and pulmonary neuroendocrine hyperplasia has also been shown in neonatal rat models of hyperoxia exposure [64]. During DOL 1–4, BPD preterm infants had a urine BLP level of more than 20,000 pg/mg Cr ($P \leq 0.001$). After adjusting for all confounding variables, multivariable logistic regression analysis demonstrated that increased urine BLP levels were correlated with BPD ($P \leq 0.001$) [65].

Infants with BPD have persistent numbers of neuroendocrine cells that produce Gastrin-releasing peptide (GRP) in their bronchioles [66]. GRP may modulate BPD in a mechanical way, probably as an oxidant sensor. Urinary GRP concentration in the first postnatal week was positively linked with the diagnosis of BPD at 36 weeks postmenstrual age (PMA) (90 pmol/mg Cr VS. 67 pmol/mg Cr, P

= 0.005). Biomarkers of oxidative stress including allantoin and 8-OHdG were related to GRP levels over 80 pmol/mg Cr, as were BPD, and ventilation and oxygen support durations [67].

1.1.8 Circulatory Stress Marker

N-terminal pro-brain natriuretic peptide (NT-proBNP) is secreted by cardiac myocytes in response to volume overload. The use of serum NT-proBNP as a screening biomarker for multiple neonatal diseases, such as diaphragmatic hernia, RDS, hemodynamically significant patent ductus arteriosus (HsPDA), PH, and BPD, has generated increased research interest in recent years [53]. Moreover, the levels of NT-proBNP in the serum and urine are identical [68]. According to our pilot investigation, urine NT-proBNP levels between DOL 7 and 28 were significantly higher in the BPD group than in the control group ($P<0.05$), positively correlated with mechanical ventilation and oxygen exposure duration (r: 0.175-0.505, $P<0.05$), and were significantly associated with BPD development ($P<0.05$) [61].

TA Biomarkers

Studies on BPD biomarkers have largely been conducted using TA and bronchoalveolar lavage fluid (BALF) samples because they provide direct access to the pulmonary compartment [69]. Although endotracheal intubation for mechanical ventilation is intrusive, tracheal suctioning is administered as required to preserve airway patency in intubated newborns and it is relatively noninvasive. The convenience of sample acquisition via routine endotracheal suctioning is one of the key benefits of TA examination, even though that leads to a lack of sampling of initially non-intubated infants who may subsequently develop BPD and may not necessarily represent the distal lung parenchymal milieu [70]. (Supplemental Table 1).

1.1.9 Inflammatory Markers

Proinflammatory cytokines IL-1, -1β, -6, -8, -16, and tumor necrosis factor α (TNFα) in earlier studies [71], and interferon γ (IFNγ), IL1/IL6 ratios, and IL1/IL1ra ratios in more recent studies [72, 73], have been shown to be elevated in the TA and to predict adverse pulmonary outcomes,

respiratory failure, and, in particular, the development of BPD in preterm infants. Furthermore, investigators have hypothesized that newborns who develop BPD have a diminished capacity to produce enough quantities of IL-10 [74].

Monocyte chemoattractant protein (MCP) controls the recruitment of mononuclear cells to the lung and their subsequent activation [74]. The levels of MCP-1, -2, and -3 in the TA were higher in infants with BPD than in those without BPD [75, 76]. Nuclear factor-kappaB (NF-κB) plays a critical role in regulating major proinflammatory mediators [77]. Its activation is elevated in the TA of preterm infants with BPD [69, 78].

1.1.10 Fibrotic and epithelial markers

In the first 2–3 weeks of life, low concentrations of TIMP-1 and TIMP-2 in the TA, high levels of MMP-8 and MMP-9, and ratio of MMP-9/TIMP-1 were attributed to the acute inflammatory process in RDS and later development of BPD in preterm infants [11, 79, 80]. The TA of premature births with BPD demonstrated an increase in TGF-β1 expression [81].

Cathepsin K (CatK) is a cysteine proteinase that has been shown to degrade matrix collagen and elastin. It has been found to be highly expressed in osteoclasts and, more recently, in lung tissue [82]. CatK expression in infants with BPD was dramatically downregulated from DOL 9–13 compared to that in controls in the TA ($P<0.05$) [83].

1.1.11 Oxidative stress-related markers

8-OHdG levels in the TA were comparatively higher in the BPD group than in the non-BPD group on DOL 1 and 28, and they remained elevated on DOL 28 in the BPD group ($P<0.05$) as reported in a birth cohort study [84]. Furthermore, predicting the development of BPD with TA 8-OHdG levels on DOL 1 (cut-off, 4.4 ng/mg) yielded an AUC of 0.77, as determined by an ROC curve analysis [84].

1.1.12 Angiogenic Growth Factors

Lung capillary maturation is a key process regulated by VEGF, which also plays a role in normal and pathologic vasculogenesis and angiogenesis. VEGF acts on cells through two different tyrosine kinase receptors, VEGF receptor (VEGFR)-1 and VEGFR-2. In VLBWIs needing oxygen and assisted mechanical ventilation, Hasan et al. [85] discovered that low VEGF levels in the TA, in combination with high soluble VEGFR (sVEGFR)-1 levels on DOL1, were biological indicators of BPD development. In addition, preterm newborns with later BPD development had significantly increased sphingosine 1-phosphate (S1P) concentrations in the TA on DOL 1 [86].

ANG-2 is an angiogenic growth factor that destabilizes blood vessels [87]. Higher ANG-2 levels in TA have been correlated with the incidence of BPD or mortality in ventilated preterm newborns [88].

Endothelin-1 (ET-1) is an effective endothelial-derived vasoconstrictor and bronchoconstrictor with the highest concentration in the lungs [89]. The ET-1 concentrations in newborns with BPD were considerably higher than those who did not eventually develop BPD in the first week of life [90].

Furthermore, higher levels of FGF-2 in the TA on DOL1 were correlated to a poorer prognosis (BPD/death) [91].

1.1.13 Others

Mesenchymal stromal cells (MSCs) isolated from TA of newborns express lung-specific genes, differ from lung fibroblasts, and produce pro-inflammatory cytokines [92]. In a newborn mouse lung injury model, intravenous or intra-alveolar MSCs transplantation was related to decreased inflammation and fibrosis in the lung [93]. The colony/cluster ratio of MSCs in the TA was considerably lower in BPD infants, according to a cross-sectional investigation ($P=0.002$). In addition, the colony/cluster ratio was a significant variable linked with BPD (95% CI: 1.6-2.98, $P = 0.029$).

Heat shock protein-70 (Hsp-70) is essential for protein folding, protein stability, and cellular homeostasis. The BPD group had considerably higher TA Hsp-70 concentrations on DOL 1 and 28 than the non-BPD group. In addition, an elevated TA Hsp-70 threshold value (above 149.1 ng/mg)

had a 71.9% sensitivity and 93.5% specificity for the diagnosis of BPD, demonstrating a significant predictive value [94].

Exhaled Breath Condensates

Inflammation of the respiratory system is a hallmark of BPD; in addition, measuring inflammatory markers in exhaled breath condensates may be a valuable noninvasive method. Infants who are either ventilated or not may have their exhaled breath condensate analyzed using gas chromatography and mass spectrometry for diagnostic purposes [95].

By monitoring end-tidal carbon monoxide (ETCO), the levels of an inflammatory biological marker (carbon monoxide) were determined [96]. May et al. [97] concluded in prospective research that the evaluation of ETCO on DOL 14 enhanced the prediction of BPD when birth weight was taken into consideration. ETCO levels on DOL 14 were consistently higher in newborns with BPD. This result re-emphasized the significance of continuing inflammation in the development of BPD.

In preterm infants with BPD, levels of fractional exhaled nitric oxide (FeNO) were distinctly higher than in those without BPD ($P = 0.006$). In addition, there was a significant association of elevated exhaled nitric oxide (eNO) concentrations with BPD (95%CI: 1.397-4.824, P=0.000) [98]. In preterm newborns with existing BPD, especially those with moderate or severe BPD, there was an increase in eNO levels, which may be indicative of an inflammatory process [99].

Radiographical Studies

Since 1967, radiological imaging has been an essential technique for identifying BPD. Image diagnosis and prediction are complicated in BPD. Plain chest radiography (CXR) and computed tomography (CT) scanning remain the most widely used imaging modalities in BPD [100]. Radiological innovations, especially in magnetic resonance imaging (MRI) and lung ultrasound (LUS), have the potential to provide new quantitative measures of pulmonary function and structure in BPD [101] (Supplemental Table 2).

Chest Radiography

CXR is the primary line of radiological investigation in neonates due to a negligible dose of ionizing radiation, and is thus, widely regarded as safe for serial monitoring of infants [102]. However, CXR only offers a flat image of the chest (two-dimensional, usually a coronal view) and is not sufficiently capable of detecting subtle cases of pulmonary diseases [103].

In a prospective study of 400 VLBWIs, in the first 6 hours after birth, the presence of a small thymus, as measured by the ratio between the transverse diameter of the cardiothymic image at the level of the carina and that of the thorax (CT/T) < 0.28, distinguished newborns with BPD [104]. Furthermore, 89.5% of newborns requiring oxygen dependency at 36 weeks PMA exhibited leaky lung syndrome with hazy-to-opaque lungs or cystic BPD with bubbly alterations on CXR at that time [105]. Kim et al. [106] reported that the pattern of interstitial pneumonia on the CXR on DOL 7 was considerably greater in the BPD group ($P<0.001$). It was independently related to BPD or death prediction (OR: 4.0, 95% CI: 1.1–14.4) and may be used to identify high-risk VLBWIs who may benefit from prevention strategies against BPD.

The chest radiograph thoracic area (CRTA), which can be used to predict how much the pulmonary mechanics has changed, is a sign of hyperinflation [107]. After controlling for potential confounders, the CRTA in the first 72 h of life was higher in the moderate/severe BPD group ($P<0.001$) and mild BPD group ($P<0.05$) than in the no BPD group [108]. Enhanced CRTA in BPD was shown to be substantially associated with oxygenation impairment, suggesting that CRTA may be utilized as a straightforward radiographic diagnostic index to evaluate BPD severity [109].

The CXR scoring criteria was designed by Toce et al. [110] and Greenough et al. [111]. It was discovered that the scoring system used for radiographs on DOL 7 correlated better with BPD diagnosis [112]. The relationship between the scoring system and the clinical severity of BPD was substantial ($r = 0.78$, $P<0.001$) [113].

Chest CT scanning

Because of its high three-dimensional anatomical resolution of the lung parenchyma and significant tissue contrast, CT is a common imaging modality for the evaluation of respiratory diseases in adults [114]. Although CT observations were more specific and objective for preterm infants with BPD than CXR, the continuous use of chest CT in infants has been limited, largely due to the consequent ionizing radiation exposure in neonates who may be radiosensitive throughout development [115]. With the recent advancement of low-dose CT protocols, it is believed that newborn pulmonary imaging with CT would become more commonplace [116, 117].

Based on the quantitative investigation of pulmonary abnormalities in BPD, Shin et al. [114] developed a novel high-resolution computed tomography (HRCT) scoring system for BPD that consists of the hyperaeration and parenchymal scores. The HRCT score was more strongly correlated ($r = 0.646$, $P<0.001$) with the clinical severity of BPD than the CXR score. In contrast, another research found no significant difference in the average HRCT scores of moderate and severe BPD [118]. Moreover, CT scoring systems were directly associated with a variety of adverse physiological and clinical outcomes in neonates with BPD, such as the duration of oxygen exposure, incidence of desaturation events during sleep, severity of wheezing, and rate of decline in forced expiratory volume in one second [119, 120]. In conclusion, CT scans have not yet been clearly characterized in their efficacy as a predictor of BPD in preterm newborns, however, they may be useful for predicting the severity and long-term respiratory consequences of BPD.

.

Chest MRI

As a non-ionizing modality, MRI is appropriate for pediatric imaging, especially routine and serial examination. Historically, MRI of the lung has been restricted owing to technological limitations resulting primarily from the lung parenchyma's essentially low proton density and rapid signal decay [121]. Recent improvements in MRI software acquisition methodologies have substantially

eliminated these limitations, and neonatal pulmonary MRI is currently being developed as a method that yields new imaging-based biomarkers [122, 123].

Ultrashort echo-time (UTE) MRI is the most common technique for MRI imaging of pulmonary structures [124]. The use of neonatal pulmonary scoring of UTE MRI improves the independent assessment of structural abnormalities of BPD [125]. Forster et al. [126] discovered that higher T2 and lower T1 relaxation times were correlated with an elevated risk of BPD and its severity at near-term age (mean age at imaging: 36 weeks PMA) with an AUC of 0.80 [127]. MRI might play a vital role in the characterization of certain phenotypes and could ultimately replace CT as the method for BPD imaging with high resolution.

LUS

LUS is accessible at the bedside, can be done immediately and serially, has a high interobserver agreement, and does not involve ionizing radiation. It can evaluate dynamic shifts over time and identify them even before CXR [128]. In addition, LUS has been demonstrated to be effective for pulmonary maintenance in VPIs and appears to be a potential technique for BPD prediction and diagnosis [129].

Compared to CT and MRI, perinatal LUS lacks a consistent acquisition method and cannot discriminate lung parenchyma, pulmonary vasculature, and lung function in newborns with BPD [101, 130]. According to Aldecoa-Bilbao et al. [131], VPIs with BPD had substantially higher LUS score at admission, on DOL 7, and DOL 28. Patients with BPD demonstrated greater consolidations and pleural line anomalies than those without BPD ($P<0.001$). LUS on DOL 7 accurately predicted the NICHD 2001-BPD (AUC = 0.87, $P<0.001$) and Jensen 2019-BPD (AUC = 0.80, $P<0.001$). After the first week of life, the LUS score, pleural line anomalies, and consolidations may aid in the diagnosis of BPD in VPI and prediction of its severity.

Lung Function

There is substantial evidence linking preterm delivery to reduced lung capacity, lower lung compliance, increased airway obstruction and lung resistance, impaired gas exchange, and early reduction of respiratory function [120, 132]. Due to BPD, structural abnormalities such as alveolar simplicity, decreased number of alveoli, and enlarged alveoli are major pathophysiological basis for impaired lung function that may continue throughout childhood and contribute to chronic obstructive pulmonary disease in adulthood [133-135]. Studies have shown that early diagnosis of pulmonary function abnormalities in preterm newborns is advantageous for early prediction of BPD development [136].

May et al. [97] prospectively assessed lung function using functional residual capacity (FRC), compliance (Crs), and resistance (Rrs) of the respiratory system on DOL 3 and 14. Results for FRC and Crs on DOL 3 and 14, and Rrs on DOL 3 were substantially different between preterm infants with and without BPD. Another research demonstrates that decreased FRC in the first 72 h after delivery is a substantial predictor of moderate to severe BPD [108]. On DOL 7, 14, and 28, those with mild and moderate BPD had higher effective airway resistance per kilogram (Reff/kg), lower tidal volume per kilogram (TV/kg), FRC, ratio of time to peak tidal expiratory flow to total expiratory time (% T-PF), and ratio of volume to peak tidal expiratory flow to total expiratory volume (% V-PF) than those without BPD ($P<0.05$). This demonstrated the value of plethysmography for dynamic monitoring of pulmonary function in evaluating lung development in neonates with BPD [137].

However, there is no consensus on the early lung function assessment that best predicts the development of BPD, presumably because various methodologies have been utilized in different studies [138] (Supplemental Table 2).

“OMIC” Biomarkers

Due to recent developments in molecular genetics and next-generation sequencing, such as whole exome sequencing and whole genome sequencing, the investigation of BPD at a molecular “omic” level without bias is now possible [139]. Genetic inheritance and epigenetic mechanisms play a role in determining BPD vulnerability. Individualized and precise BPD prediction and management is made feasible by the “omics” sciences. This review focuses on genomic, proteomic, metabolomic, and microbiomic indicators in noninvasive biofluids for BPD (Table 3).

Genomics and Epigenetics

Several studies have demonstrated that genetic variables have a substantial role in the development of BPD, ranging from 53 % to 82 % [140, 141]. Scientists have tried to determine how the complex interactions between genetic and environmental factors lead to BPD. Genomic analysis may help in the future by identifying newborns with a greater risk of this condition so that they may get personalized therapy [142]. Furthermore, epigenetic processes may regulate genetic expression by either activating or silencing the genetic material encoded by the genome [143].

Fujioka et al. [144] obtained genomic DNA from 97 VLBWIs' umbilical cords, UCB, or buccal mucosa. The genotype -634C > G was significantly related to BPD. The frequency of the G allele was substantially greater in the BPD group than in the non-BPD group ($P = 0.02$). Multivariate logistic regression revealed that VEGF-634G > C G alleles were independent BPD risk factors. Exosomal miRNA (miR) signals were discovered using a prospective cohort analysis of the TA collected from extremely premature newborns shortly after delivery. Significantly lower levels of miR-876-3p were associated with severe BPD [145].

Proteomics

Multiple specific proteins have been involved in the pathophysiology of BPD. Proteomic analysis may aid in the research of protein networks that provide real-time clinical status and protein function modification. However, studies using unbiased proteome analysis for BPD are not yet available [146, 147].

Ahmed et al. [148] gathered urine samples from extremely-low-gestational-age neonates (ELGANS) within 72 h of delivery, and they used this to construct a high throughput urine proteomics approach. A total of 94 proteins had substantial abundance differences between BPD and healthy controls, and 20 proteins were directly linked to BPD and/or other infantile lung diseases (for example, chitinase-3-like protein-1, MMP-9, and so on), as determined by analyzing the urine proteome.

Metabolomics

Dyslipidemia (decreased major choline-containing phospholipids) and metabolic immaturity (increased oxylipins PGE1, PGE2, PGF2a, 9- and 13-HOTE, 9- and 13-HODE, and 9- and 13-KODE) were found to be positively associated with BPD severity and PH development in the analysis of UCB of 42 preterm infants using a multiplatform metabolomics strategy [149].

In recent research, Rogosch et al. [150] demonstrated that smell prints of volatile organic compounds assessed with an electronic nose varied between the TAs from preterm newborns who later had BPD and those who did not. Metabolites in fatty acid activation, and those involved in androgen and estrogen formation and metabolism, were found to be enriched in the TA; in addition, there were tendencies toward reduction in the fatty acid beta-oxidation pathway in newborns at high risk for BPD, according to a mummichog metabolomic pathway study [151]. Furthermore, neonates with BPD had increased concentrations of histidine, glutamic acid, citrulline, glycine, and isoleucine in their TA during the first week of life, and higher levels of acyl carnitines C16-OH and C18:1-OH [152].

Lactate, taurine, trimethylamineN-oxide (TMAO), myoinositol (which increased), and gluconate (which decreased) were identified as significant discriminant metabolites in the urine of BPD newborns during the first 24 to 36 h of life [153]. Pintus et al. [154] collected urine samples on DOL7 and examined them with ^{1}H-nuclear magnetic resonance (^{1}H-NMR). Alanine and betaine (both of which were shown to be elevated in the BPD group), TMAO, lactate, and glycine (decreased in the

BPD group), were identified as metabolites that differentiated the BPD group from the non-BPD group.

Microbiomics

The microbiota plays an essential role in maintaining the host's normal levels of immunity, hormones, and metabolism. Although most of these bacteria cannot be cultivated, high-throughput sequencing of the bacterial 16S rRNA gene has enabled the identification of the whole microbiome by facilitating its identification, alignment, sorting, and classification using publicly accessible taxonomic databases [155]. Recent research has shown that the early airway microbiome may stimulate the maturing pulmonary immune system and dysbiosis of a normal airway microbiome may lay the foundation for BPD and future pulmonary disease.

Furthermore, numerous studies have shown that an increased microbial community turnover with age and decreased microbiome diversity may be related with the development of BPD [156, 157]. Lal et al. [158] examined the TA microbiome in 23 ELBWIs and discovered an increase in Proteobacteria in the first week of birth. Conversely, Firmicutes, such as genus Lactobacillus, which is known to have strong anti-inflammatory properties and regulate alveolar development, was less common in BPD-prone newborns than in BPD-resistant newborns ($P<0.05$). A longitudinal investigation demonstrated that mechanically ventilated preterm infants who were later diagnosed with severe BPD acquired fewer Staphylococcus on DOL 1 and had a relatively higher abundance of Ureaplasma [159]. Potential biomarkers for BPD include an elevated abundance of Stenotrophomonas and increased level of sn-glycerol 3-phosphoethanolamine on DOL 1. Stenotrophomonas, which may affect the content of the lower airway microbiome through its metabolite sn-glycerol 3-phosphoethanolamine, is linked to BPD development and severity [160]. This result raised the possibility that the metabolome is affected by the metabolic activity of the airway microbiome.

Conclusions

There is a demand for objective, precise, and reliable early predictive methods to understand the development of BPD, construct predictive indicators of subsequent outcomes, and guide innovative therapeutics to avoid or reduce the severity of BPD in preterm newborns. Existing noninvasive methods for predicting the incidence and severity of BPD at a very early stage, even right after birth, were reviewed in this study. However, no established universally accepted indicator has been found. This may be because the sample sizes of the studies in the review were not large enough. Confirmative investigations with larger sample sizes and high-quality data and the evaluation of the function of noninvasive biomarkers in the identification of BPD would increase the potential for therapeutic treatments. In the future, better understanding of the pathogenesis of the disease and the potential for the development of novel biomarkers for early detection, prevention, and treatment of BPD can be gained through the application of cutting-edge prediction models that integrate clinical information with risk factors, noninvasive predictors, and "omic" approaches.

Methodology

PubMed searches using MeSH keywords from 1967 to 2022 yielded the data for this review. We searched through: "Bronchopulmonary Dysplasia" OR "BPD" AND "Early Prediction" OR "Early Diagnosis" AND "Biomarkers" OR "biomarkers" AND "Noninvasive" OR "Non-invasive" AND "Monitoring" OR "Detection" AND "Preterm infants" OR "Preterm newborns" OR "Premature infants" and other similar terms to prevent missing important literature.

Funding

This work was supported by grants from the National Natural Science Foundation of China (No. 82071688), and the Key R&D Guidance Plan Projects in Liaoning Province (2020JH1/10300001).

Conflict of Interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Author Contributions

XC and JF designed the overall study; XC drafted the manuscript and performed the systematic literature search. JF critically reviewed the manuscript for intellectual content. Both authors assisted with manuscript preparation and contributed to the study's conception and design. Both authors read and approved the final manuscript.

Table 1. Umbilical cord blood biomarkers associated with BPD

Biomarker	Physiologic Role	Levels associated with Increased BPD Risk (refs)
Immunomodulatory and inflammatory factors		
IL-6	Proinflammatory cytokine	Higher (AUC:0.849, cutoff: 46.125 pg/ml) [13]
Sgp130	IL-6 signaling membrane protein	Higher [13]
T-reg	Anti-inflammatory cytokine	Lower [16]
G-CSF	Proinflammatory cytokine	Lower in BPD-PH [24]
Angiogenic Growth Factors		
ANG-1	Pro-angiogenesis	Lower [20]
Endostatin	Anti-angiogenesis	Higher in BPD and BPD-PH [20, 21]
PlGF	Pro-angiogenesis, increase alveolar type II cell death	Higher in BPD (cutoff: 17 mg/dl) [23] Lower in BPD-PH [24]
sENG	Anti-angiogenesis	Higher [28]
ECFCs	Pro-angiogenesis	Lower [30]
FGF-10	Airway branching and lengthening promotion	Lower [32]
Epithelial and Fibrotic Markers		

Biomarker	Physiologic Role	Levels associated with Increased BPD Risk (refs)
CCSP (CC16 or CC10)	Non-ciliated epithelial cells lining the respiratory and terminal bronchioles	Lower [39]
KL-6	Fibroblast chemotactic factor	Higher [41]
MMP-9/TIMP-1	Protease/antiprotease of extracellular matrix	Higher [44]
Other factors		
Vitamin D	Antifibrotic, antioxidative, and anti-inflammatory effects	Lower (AUC:0.585, cutoff: 15.7 ng/ml) [50, 51]
Vitamin E	Antioxidant	Lower [50]

IL: Interleukin; AUC: area under curve; Sgp130: soluble glycoprotein 130; Tregs: CD4+ regulatory T-cells; G-CSF: granulocyte-colony stimulating factor; BPD-PH: BPD-associated pulmonary hypertension; ANG-1: angiopoietin-1; PlGF: Placental growth factor; sENG: soluble endoglin; ECFC: Endothelial colony-forming cell; FGF-10: fibroblast growth factor-10; CCSP: Clara cell secretory protein; KL-6: Krebs yon den Lundgen-6; MMP-9/TIMP-1: Matrix metalloproteinase-9/ Tissue inhibitors of metalloproteinase.

Table 2. Urinary biomarkers associated with BPD

Biomarker	Physiologic Role	Levels associated with Increased BPD Risk (refs)
Inflammatory markers		
LTE4	Inflammatory marker	Higher in "atypical" BPD [55]
B2M	An alternative to proinflammatory cytokines	Higher (AUC:0.8, cutoff: $19.6*10^4$ mg/g Cr) [57]
Oxidative stress-related markers		
8-OHdG	Endogenous oxidative stress-related DNA damage marker	Higher [61]
11-dehydro-TXB2	Lipid oxidation product	Higher [62]
Pulmonary neuroendocrine markers		
BLP	Pulmonary neuroendocrine marker	Higher [65]
GRP	Oxidant sensor	Higher (AUC:0.575, cutoff: 80 pmol/mg Cr) [67]
Circulatory stress marker		
NT-proBNP	Circulatory stress marker	Higher [61]

LTE4: Leukotriene E4; B2M: B-2-microglobulin; AUC: area under curve; 8-OHdG: 8-hydroxy-2'-deoxyguanosine; DNA: deoxyribonucleic acid; TXB2: thromboxane B2; BLP: bombesin-like peptide; GRP: Gastrin-releasing peptide; NT-proBNP: N-terminal pro-brain natriuretic peptide.

Table 3. Noninvasive "OMIC" biomarkers associated with BPD

Biomarker	Specimen	Levels associated with Increased BPD Risk (refs)
Genomics and epigenetics		
VEGF-634G > C G alleles	umbilical cords, UCB or buccal mucosa	Higher [144]
miR-876-3p	TA	Lower [145]
Proteomics		
Chitinase-3-like protein-1, MMP9	Urine	Higher [148]
Metabolomics		
Choline-containing phospholipids	UCB	Lower [149]
Oxylipins (PGE1, PGE2, PGF2a, 9- and 13-HOTE, 9-and 13-HODE, 9- and 13-KODE)	UCB	Higher [149]
Volatile organic compounds detected by "electronic nose"	TA	Differ [150]
Metabolites of fatty acid oxidation and androgen and estrogen formation	TA	Higher [151]
Histidine, glutamic acid, citrulline, glycine, isoleucine,	TA	Higher [152]

Biomarker	Specimen	Levels associated with Increased BPD Risk (refs)
acyl carnitines C16-OH and C18:1-OH		
Lactate, taurine, TMAO, myoinositol	Urine	Higher within 24-36 h of life [153]
Gluconate	Urine	Lower [153]
TMAO, lactate, glycine	Urine	Lower at DOL 7 [154]
Alanine and betaine	Urine	Higher [154]
Microbiomics		
Firmicutes (Genus Lactobacillus)	TA	Lower [158]
Staphylococcus	TA	Lower [159]
Proteobacteria	TA	Higher [158]
Ureaplasma	TA	Higher [159]
Stenotrophomonas	TA	Higher [160]

VEGF-634G: vascular endothelial growth factor-634G; UCB: umbilical cord blood; miR-876-3p: miRNA-876-3p; TA: Tracheal aspirate; MMP9: Matrix metalloproteinase-9; TMAO: trimethylamineN-oxide; DOL: Day of life.

References

[1] Thébaud B, Goss KN, Laughon M, et al. Bronchopulmonary dysplasia. Nat Rev Dis Primers 2019; 5: 78.

[2] Northway WH, Jr., Rosan RC and Porter DY. Pulmonary disease following respirator therapy of hyaline-membrane disease. Bronchopulmonary dysplasia. N. Engl. J. Med. 1967; 276: 357–368.

[3] Jobe AH and Abman SH. Bronchopulmonary dysplasia: a continuum of lung disease from the fetus to the adult. Am J Respir Crit Care Med 2019; 200: 659–660.

[4] Stoll BJ, Hansen NI, Bell EF, et al. Trends in care practices, morbidity, and mortality of extremely preterm neonates, 1993-2012. Jama 2015; 314: 1039–1051.

[5] Jobe AH and Bancalari E. Bronchopulmonary dysplasia. Am J Respir Crit Care Med 2001; 163: 1723–1729.

[6] Higgins RD, Jobe AH, Koso-Thomas M, et al. Bronchopulmonary dysplasia: executive summary of a workshop. J Pediatr 2018; 197: 300–308.

[7] Steinhorn R, Davis JM, Göpel W, et al. Chronic pulmonary insufficiency of prematurity: developing optimal endpoints for drug development. J Pediatr 2017; 191: 15–21.e11.

[8] Bancalari E, Jain D and Jobe AH. Prevention of bronchopulmonary dysplasia: are intratracheal steroids with surfactant a magic bullet? Am J Respir Crit Care Med 2016; 193: 12–13.

[9] Lal CV and Schwarz MA. Vascular mediators in chronic lung disease of infancy: role of endothelial monocyte activating polypeptide II (EMAP II). Birth Defects Res A Clin Mol Teratol 2014; 100: 180–188.

[10] Bhandari A and Bhandari V. Biomarkers in bronchopulmonary dysplasia. Paediatr Respir Rev 2013; 14: 173–179.

[11] Bhandari A and Bhandari V. Pitfalls, problems, and progress in bronchopulmonary dysplasia. Pediatrics 2009; 123: 1562–1573.

[12] Hirani D, Alvira CM, Danopoulos S, et al. Macrophage-derived IL-6 trans-signalling as a novel target in the pathogenesis of bronchopulmonary dysplasia. Eur Respir J 2022; 59: 2002248.

[13] Tian XY, Zhang XD, Li QL, et al. Biological markers in cord blood for prediction of bronchopulmonary dysplasia in premature infants. Clin Exp Obstet Gynecol 2014; 41: 313–318.

[14] Cao C, Ma T, Chai YF, et al. The role of regulatory T cells in immune dysfunction during sepsis. World J Emerg Med 2015; 6: 5–9.

[15] Mohr A, Atif M, Balderas R, et al. The role of FOXP3(+) regulatory T cells in human autoimmune and inflammatory diseases. Clin Exp Immunol 2019; 197: 24–35.

[16] Pagel J, Twisselmann N, Rausch TK, et al. Increased regulatory T cells precede the development of bronchopulmonary dysplasia in preterm infants. Front Immunol 2020; 11: 565257.

[17] Misra R, Shah S, Fowell D, et al. Preterm cord blood $CD4^+$ T cells exhibit increased IL-6 production in chorioamnionitis and decreased $CD4^+$ T cells in bronchopulmonary dysplasia. Hum Immunol 2015; 76: 329–338.

[18] Gilfillan M, Das P, Shah D, et al. Inhibition of microRNA-451 is associated with increased expression of Macrophage Migration Inhibitory Factor and mitgation of the cardio-pulmonary phenotype in a murine model of Bronchopulmonary Dysplasia. Respir Res 2020; 21: 92.

[19] Sahni M and Bhandari V. Patho-mechanisms of the origins of bronchopulmonary dysplasia. Mol Cell Pediatr 2021; 8: 21.

[20] Mohamed WA, Niyazy WH and Mahfouz AA. Angiopoietin-1 and endostatin levels in cord

plasma predict the development of bronchopulmonary dysplasia in preterm infants. J Trop Pediatr 2011; 57: 385–388.

[21] Janér J, Andersson S, Kajantie E, et al. Endostatin concentration in cord plasma predicts the development of bronchopulmonary dysplasia in very low birth weight infants. Pediatrics 2009; 123: 1142–1146.

[22] Tsao PN, Su YN, Li H, et al. Overexpression of placenta growth factor contributes to the pathogenesis of pulmonary emphysema. Am J Respir Crit Care Med 2004; 169: 505–511.

[23] Tsao PN, Wei SC, Su YN, et al. Placenta growth factor elevation in the cord blood of premature neonates predicts poor pulmonary outcome. Pediatrics 2004; 113: 1348–1351.

[24] Mestan KK, Gotteiner N, Porta N, et al. Cord blood biomarkers of placental maternal vascular underperfusion predict bronchopulmonary dysplasia-associated pulmonary hypertension. J Pediatr 2017; 185: 33–41.

[25] Adu-Gyamfi EA, Ding YB and Wang YX. Regulation of placentation by the transforming growth factor beta superfamily. Biol Reprod 2020; 102: 18–26.

[26] Rana S, Burke SD and Karumanchi SA. Imbalances in circulating angiogenic factors in the pathophysiology of preeclampsia and related disorders. Am J Obstet Gynecol 2022; 226: S1019–s1034.

[27] Lawera A, Tong Z, Thorikay M, et al. Role of soluble endoglin in BMP9 signaling. Proc Natl Acad Sci U S A 2019; 116: 17800–17808.

[28] Kim DH, Shin SH, Kim EK, et al. Association of increased cord blood soluble endoglin with the development of bronchopulmonary dysplasia in preterm infants with maternal preeclampsia. Pregnancy Hypertens 2018; 13: 148–153.

[29] Alphonse RS, Vadivel A, Fung M, et al. Existence, functional impairment, and lung repair potential of endothelial colony-forming cells in oxygen-induced arrested alveolar growth.

Circulation 2014; 129: 2144–2157.

[30] Baker CD, Balasubramaniam V, Mourani PM, et al. Cord blood angiogenic progenitor cells are decreased in bronchopulmonary dysplasia. Eur Respir J 2012; 40: 1516–1522.

[31] Mathew R. Signaling pathways involved in the development of bronchopulmonary dysplasia and pulmonary hypertension. Children 2020; 7: 100.

[32] Mohamed WA and Aseeri MA. Cord blood fibroblast growth factor-10 as a possible predictor of bronchopulmonary dysplasia in preterm infants. J Neonatal Perinatal Med 2014; 7: 101–105.

[33] Kolesnichenko OA, Whitsett JA, Kalin TV, et al. Therapeutic potential of endothelial progenitor cells in pulmonary diseases. Am J Respir Cell Mol Biol 2021; 65: 473–488.

[34] Mižíková I, Pfeffer T, Nardiello C, et al. Targeting transglutaminase 2 partially restores extracellular matrix structure but not alveolar architecture in experimental bronchopulmonary dysplasia. Febs j 2018; 285: 3056–3076.

[35] Witsch TJ, Niess G, Sakkas E, et al. Transglutaminase 2: a new player in bronchopulmonary dysplasia? Eur Respir J 2014; 44: 109–121.

[36] Wang K, Huang X, Lu H, et al. A comparison of KL-6 and Clara cell protein as markers for predicting bronchopulmonary dysplasia in preterm infants. Dis Markers 2014; 2014: 736536.

[37] Bustos ML, Mura M, Marcus P, et al. Bone marrow cells expressing clara cell secretory protein increase epithelial repair after ablation of pulmonary clara cells. Mol Ther 2013; 21: 1251–1258.

[38] Bustos ML, Mura M, Hwang D, et al. Depletion of bone marrow CCSP-expressing cells delays airway regeneration. Mol Ther 2015; 23: 561–569.

[39] Schrama AJ, Bernard A, Poorthuis BJ, et al. Cord blood Clara cell protein CC16 predicts the development of bronchopulmonary dysplasia. Eur J Pediatr 2008; 167: 1305–1312.

[40] Dilli D, Özyazici A, Dursun A, et al. Predictive values of plasma KL-6 in bronchopulmonary dysplasia in preterm infants. Turk J Med Sci 2017; 47: 621–626.

[41] Kim DH, Kim HS, Shim SY, et al. Cord blood KL-6, a specific lung injury marker, correlates with the subsequent development and severity of atypical bronchopulmonary dysplasia. Neonatology 2008; 93: 223–229.

[42] Fernandes JC, Schemitt EG, Da Silva J, et al. Combination of trans-resveratrol and ε-viniferin induces a hepatoprotective effect in rats with severe acute liver failure via reduction of oxidative stress and MMP-9 expression. Nutrients 2021; 13: 3677.

[43] Yang M, Chen BL, Huang JB, et al. Angiogenesis-related genes may be a more important factor than matrix metalloproteinases in bronchopulmonary dysplasia development. Oncotarget 2017; 8: 18670–18679.

[44] Fukunaga S, Ichiyama T, Maeba S, et al. MMP-9 and TIMP-1 in the cord blood of premature infants developing BPD. Pediatr Pulmonol 2009; 44: 267–272.

[45] Gil Á, Plaza-Diaz J and Mesa MD. Vitamin D: classic and novel actions. Ann Nutr Metab 2018; 72: 87–95.

[46] Chen L, Wilson R, Bennett E, et al. Identification of vitamin D sensitive pathways during lung development. Respir Res 2016; 17: 47.

[47] Lu T, Liang B, Jia Y, et al. Relationship between bronchopulmonary dysplasia, long-term lung function, and vitamin D level at birth in preterm infants. Transl Pediatr 2021; 10: 3075–3081.

[48] Kim I, Kim SS, Song JI, et al. Association between vitamin D level at birth and respiratory morbidities in very-low-birth-weight infants. Korean J Pediatr 2019; 62: 166–172.

[49] Szymańska R, Nowicka B and Kruk J. Vitamin E - occurrence, biosynthesis by plants and functions in human nutrition. Mini Rev Med Chem 2017; 17: 1039–1052.

[50] Ge H, Liu W, Li H, et al. The association of vitamin D and vitamin E levels at birth with

bronchopulmonary dysplasia in preterm infants. Pediatr Pulmonol 2021; 56: 2108–2113.

[51] Yu H, Fu J and Feng Y. Utility of umbilical cord blood 25-hydroxyvitamin D levels for predicting bronchopulmonary dysplasia in preterm infants with very low and extremely low birth weight. Front Pediatr 2022; 10: 956952.

[52] Yamasaki K, Hata J, Ide T, et al. Urinary N-terminal pro-B-type natriuretic peptide as a biomarker for cardiovascular events in a general Japanese population: the Hisayama Study. Environ Health Prev Med 2021; 26: 47.

[53] Iliodromiti Z, Christou E, Vrachnis N, et al. Serum and urinary N-terminal pro-brain natriuretic peptides as biomarkers for bronchopulmonary dysplasia of preterm neonates. Front Pediatr 2020; 8: 588738.

[54] Ban GY, Kim SH and Park HS. Persistent eosinophilic inflammation in adult asthmatics with high serum and urine levels of leukotriene E(4). J Asthma Allergy 2021; 14: 1219–1230.

[55] Joung KE, Kim HS, Lee J, et al. Correlation of urinary inflammatory and oxidative stress markers in very low birth weight infants with subsequent development of bronchopulmonary dysplasia. Free Radic Res 2011; 45: 1024–1032.

[56] Rosen D, Lee JH, Cuttitta F, et al. Accelerated thymic maturation and autoreactive T cells in bronchopulmonary dysplasia. Am J Respir Crit Care Med 2006; 174: 75–83.

[57] Shima Y, Nishimaki S, Nakajima M, et al. Urinary β-2-microglobulin as an alternative marker for fetal inflammatory response and development of bronchopulmonary dysplasia in premature infants. J Perinatol 2011; 31: 330–334.

[58] Elkabany ZA, El-Farrash RA, Shinkar DM, et al. Oxidative stress markers in neonatal respiratory distress syndrome: advanced oxidation protein products and 8-hydroxy-2-deoxyguanosine in relation to disease severity. Pediatr Res 2020; 87: 74–80.

[59] Choi Y, Rekers L, Dong Y, et al. Oxygen toxicity to the immature lung-part I:

pathomechanistic understanding and preclinical perspectives. Int J Mol Sci 2021; 22: 11006.

[60] Yzydorczyk C, Mitanchez D, Buffat C, et al. [Oxidative stress after preterm birth: origins, biomarkers, and possible therapeutic approaches]. Arch Pediatr 2015; 22: 1047–1055.

[61] Cui X and Fu J. Urinary biomarkers for the early prediction of bronchopulmonary dysplasia in preterm infants: A pilot study. Front Pediatr 2022; 10: 959513.

[62] Parad RB, Breeze JL, Terrin N, et al. Differences in clinical and laboratory biomarkers for short and long-term respiratory outcomes in preterm neonates. Pediatr Pulmonol 2021; 56: 3847–3856.

[63] Subramaniam M, Bausch C, Twomey A, et al. Bombesin-like peptides modulate alveolarization and angiogenesis in bronchopulmonary dysplasia. Am J Respir Crit Care Med 2007; 176: 902–912.

[64] Shenberger JS, Shew RL and Johnson DE. Hyperoxia-induced airway remodeling and pulmonary neuroendocrine cell hyperplasia in the weanling rat. Pediatr Res 1997; 42: 539–544.

[65] Cullen A, Van Marter LJ, Allred EN, et al. Urine bombesin-like peptide elevation precedes clinical evidence of bronchopulmonary dysplasia. Am J Respir Crit Care Med 2002; 165: 1093–1097.

[66] Degan S, Lopez GY, Kevill K, et al. Gastrin-releasing peptide, immune responses, and lung disease. Ann N Y Acad Sci 2008; 1144: 136–147.

[67] Voynow JA, Fisher K, Sunday ME, et al. Urine gastrin-releasing peptide in the first week correlates with bronchopulmonary dysplasia and post-prematurity respiratory disease. Pediatr Pulmonol 2020; 55: 899–908.

[68] Kurihara N, Miwa M, Matsuzaki Y, et al. Usefulness of measurement of urinary N-terminal pro-brain natriuretic peptide in neonatal period. Pediatr Int 2011; 53: 608.

[69] Thompson A and Bhandari V. Pulmonary biomarkers of bronchopulmonary dysplasia. Biomark Insights 2008; 3: 361–373.

[70] Yazdi SM, Patel EU, Richardson CD, et al. Dexamethasone alters tracheal aspirate T-cell cytokine production in ventilated preterm infants. Children 2021; 8: 879.

[71] Wickramasinghe LC, van Wijngaarden P, Tsantikos E, et al. The immunological link between neonatal lung and eye disease. Clin Transl Immunology 2021; 10: e1322.

[72] Aghai ZH, Saslow JG, Mody K, et al. IFN-γ and IP-10 in tracheal aspirates from premature infants: relationship with bronchopulmonary dysplasia. Pediatr Pulmonol 2013; 48: 8–13.

[73] Liao J, Kapadia VS, Brown LS, et al. The NLRP3 inflammasome is critically involved in the development of bronchopulmonary dysplasia. Nat Commun 2015; 6: 8977.

[74] Bose CL, Dammann CE and Laughon MM. Bronchopulmonary dysplasia and inflammatory biomarkers in the premature neonate. Arch Dis Child Fetal Neonatal Ed 2008; 93: F455–461.

[75] Baier RJ, Loggins J and Kruger TE. Monocyte chemoattractant protein-1 and interleukin-8 are increased in bronchopulmonary dysplasia: relation to isolation of Ureaplasma urealyticum. J Investig Med 2001; 49: 362–369.

[76] Liang Z, Zhang X, Liu Y, et al. SEMA3A protects against hyperoxia-induced lung injury in a bronchopulmonary dysplasia model of newborn rat by inhibiting ERK pathway. Allergol Immunopathol (Madr) 2021; 49: 8–15.

[77] Chen S, Wu Q, Zhong D, et al. Caffeine prevents hyperoxia-induced lung injury in neonatal mice through NLRP3 inflammasome and NF-κB pathway. Respir Res 2020; 21: 140.

[78] Aghai ZH, Kode A, Saslow JG, et al. Azithromycin suppresses activation of nuclear factor-kappa B and synthesis of pro-inflammatory cytokines in tracheal aspirate cells from premature infants. Pediatr Res 2007; 62: 483–488.

[79] Cederqvist K, Sorsa T, Tervahartiala T, et al. Matrix metalloproteinases-2, -8, and -9 and

TIMP-2 in tracheal aspirates from preterm infants with respiratory distress. Pediatrics 2001; 108: 686–692.

[80] Ekekezie I, Thibeault DW, Simon SD, et al. Low levels of tissue inhibitors of metalloproteinases with a high matrix metalloproteinase-9/tissue inhibitor of metalloproteinase-1 ratio are present in tracheal aspirate fluids of infants who develop chronic lung disease. Pediatrics 2004; 113: 1709–1714.

[81] Aly H, Mansi Y, Ez El Din Z, et al. Mesenchymal stromal cells and TGF-β1 in tracheal aspirate of premature infants: early predictors for bronchopulmonary dysplasia? J Perinat Med 2019; 47: 470–477.

[82] Altiok O, Yasumatsu R, Bingol-Karakoc G, et al. Imbalance between cysteine proteases and inhibitors in a baboon model of bronchopulmonary dysplasia. Am J Respir Crit Care Med 2006; 173: 318–326.

[83] Knaapi J, Lukkarinen H, Kiviranta R, et al. Cathepsin K expression is diminished in infants with bronchopulmonary dysplasia. Acta Paediatr 2006; 95: 1298–1300.

[84] Hsiao CC, Chang JC, Tsao LY, et al. Correlates of elevated interleukin-6 and 8-hydroxy-2'-deoxyguanosine levels in tracheal aspirates from very low birth weight infants who develop bronchopulmonary dysplasia. Pediatr Neonatol 2017; 58: 63–69.

[85] Hasan J, Beharry KD, Valencia AM, et al. Soluble vascular endothelial growth factor receptor 1 in tracheal aspirate fluid of preterm neonates at birth may be predictive of bronchopulmonary dysplasia/chronic lung disease. Pediatrics 2009; 123: 1541–1547.

[86] Hendricks-Muñoz KD, Xu J and Voynow JA. Tracheal aspirate VEGF and sphingolipid metabolites in the preterm infant with later development of bronchopulmonary dysplasia. Pediatr Pulmonol 2018; 53: 1046–1052.

[87] Amelio GS, Provitera L, Raffaeli G, et al. Proinflammatory endothelial phenotype in very

preterm infants: a pilot study. Biomedicines 2022; 10: 1185.

[88] Aghai ZH, Faqiri S, Saslow JG, et al. Angiopoietin 2 concentrations in infants developing bronchopulmonary dysplasia: attenuation by dexamethasone. J Perinatol 2008; 28: 149–155.

[89] El Shemi MS, Tawfik S, Khafagy SM, et al. Endothelin 1 as a predictor marker for bronchopulmonary dysplasia in preterm neonates with respiratory distress syndrome. J Neonatal Perinatal Med 2017; 10: 79–83.

[90] Niu JO, Munshi UK, Siddiq MM, et al. Early increase in endothelin-1 in tracheal aspirates of preterm infants: correlation with bronchopulmonary dysplasia. J Pediatr 1998; 132: 965–970.

[91] Ambalavanan N and Novak ZE. Peptide growth factors in tracheal aspirates of mechanically ventilated preterm neonates. Pediatr Res 2003; 53: 240–244.

[92] Bozyk PD, Popova AP, Bentley JK, et al. Mesenchymal stromal cells from neonatal tracheal aspirates demonstrate a pattern of lung-specific gene expression. Stem Cells Dev 2011; 20: 1995–2007.

[93] Chen J, Chen Y, Du X, et al. Integrative studies of human cord blood derived mononuclear cells and umbilical cord derived mesenchyme stem cells in ameliorating bronchopulmonary dysplasia. Front Cell Dev Biol 2021; 9: 679866.

[94] Hsiao CC, Lee CH, Yang RC, et al. Heat shock protein-70 levels are associated with a state of oxidative damage in the development of bronchopulmonary dysplasia. Front Pediatr 2021; 9: 616452.

[95] Anyanwu AC, Bentley JK, Popova AP, et al. Suppression of inflammatory cell trafficking and alveolar simplification by the heme oxygenase-1 product carbon monoxide. Am J Physiol Lung Cell Mol Physiol 2014; 306: L749–763.

[96] Lin H and Wang X. The effects of gasotransmitters on bronchopulmonary dysplasia. Eur J Pharmacol 2020; 873: 172983.

[97] May C, Patel S, Kennedy C, et al. Prediction of bronchopulmonary dysplasia. Arch Dis Child Fetal Neonatal Ed 2011; 96: F410–416.

[98] Zhang Z, Wu W, Hou L, et al. Cytokines and exhaled nitric oxide are risk factors in preterm infants for bronchopulmonary dysplasia. Biomed Res Int 2021; 2021: 6648208.

[99] May C, Williams O, Milner AD, et al. Relation of exhaled nitric oxide levels to development of bronchopulmonary dysplasia. Arch Dis Child Fetal Neonatal Ed 2009; 94: F205–209.

[100] Vanhaverbeke K, Van Eyck A, Van Hoorenbeeck K, et al. Lung imaging in bronchopulmonary dysplasia: a systematic review. Respir Med 2020; 171: 106101.

[101] Higano NS, Ruoss JL and Woods JC. Modern pulmonary imaging of bronchopulmonary dysplasia. J Perinatol 2021; 41: 707–717.

[102] Arai H, Ito T, Ito M, et al. Impact of chest radiography-based definition of bronchopulmonary dysplasia. Pediatr Int 2019; 61: 258–263.

[103] Arai H, Ito M, Ito T, et al. Bubbly and cystic appearance on chest radiograph of extremely preterm infants with bronchopulmonary dysplasia is associated with wheezing disorder. Acta Paediatr 2020; 109: 711–719.

[104] De Felice C, Latini G, Del Vecchio A, et al. Small thymus at birth: a predictive radiographic sign of bronchopulmonary dysplasia. Pediatrics 2002; 110: 386–388.

[105] Hyödynmaa E, Korhonen P, Ahonen S, et al. Frequency and clinical correlates of radiographic patterns of bronchopulmonary dysplasia in very low birth weight infants by term age. Eur J Pediatr 2012; 171: 95–102.

[106] Kim HR, Kim JY, Yun B, et al. Interstitial pneumonia pattern on day 7 chest radiograph predicts bronchopulmonary dysplasia in preterm infants. BMC Pediatr 2017; 17: 125.

[107] Gilfillan M, Bhandari A and Bhandari V. Diagnosis and management of bronchopulmonary dysplasia. Bmj 2021; 375: n1974.

[108] May C, Prendergast M, Salman S, et al. Chest radiograph thoracic areas and lung volumes in infants developing bronchopulmonary dysplasia. Pediatr Pulmonol 2009; 44: 80–85.

[109] Dassios T, Curley A, Krokidis M, et al. Correlation of radiographic thoracic area and oxygenation impairment in bronchopulmonary dysplasia. Respir Physiol Neurobiol 2016; 220: 40–45.

[110] Toce SS, Farrell PM, Leavitt LA, et al. Clinical and roentgenographic scoring systems for assessing bronchopulmonary dysplasia. Am J Dis Child 1984; 138: 581–585.

[111] Greenough A, Kavvadia V, Johnson AH, et al. A simple chest radiograph score to predict chronic lung disease in prematurely born infants. Br J Radiol 1999; 72: 530–533.

[112] Greenough A, Thomas M, Dimitriou G, et al. Prediction of outcome from the chest radiograph appearance on day 7 of very prematurely born infants. Eur J Pediatr 2004; 163: 14–18.

[113] Li R and Zhang J. Diagnostic value of chest CT combined with x-ray for premature infants with bronchopulmonary dysplasia. Medicine (Baltimore) 2018; 97: e9723.

[114] Shin SM, Kim WS, Cheon JE, et al. Bronchopulmonary dysplasia: new high resolution computed tomography scoring system and correlation between the high resolution computed tomography score and clinical severity. Korean J Radiol 2013; 14: 350–360.

[115] Miglioretti DL, Johnson E, Williams A, et al. The use of computed tomography in pediatrics and the associated radiation exposure and estimated cancer risk. JAMA Pediatr. 2013; 167: 700–707.

[116] Semple T, Akhtar MR and Owens CM. Imaging bronchopulmonary dysplasia-a multimodality update. Front Med (Lausanne) 2017; 4: 88.

[117] Sun J, Zhang Q, Hu D, et al. Feasibility study of using one-tenth mSv radiation dose in young children chest CT with 80 kVp and model-based iterative reconstruction. Sci Rep 2019; 9:

12481.

[118] Akcan AB, Oygucu SE, Arslan AG, et al. High-resolution computed tomography scores in cases of bronchopulmonary dysplasia. Biomed Res Int 2022; 2022: 5208993.

[119] van Mastrigt E, Kakar E, Ciet P, et al. Structural and functional ventilatory impairment in infants with severe bronchopulmonary dysplasia. Pediatr Pulmonol 2017; 52: 1029–1037.

[120] Simpson SJ, Turkovic L, Wilson AC, et al. Lung function trajectories throughout childhood in survivors of very preterm birth: a longitudinal cohort study. Lancet Child Adolesc Health 2018; 2: 350–359.

[121] Wielpütz M and Kauczor HU. MRI of the lung: state of the art. Diagn Interv Radiol 2012; 18: 344–353.

[122] Voskrebenzev A and Vogel-Claussen J. Proton MRI of the lung: how to tame scarce protons and fast signal decay. J Magn Reson Imaging 2021; 53: 1344–1357.

[123] Willmering MM, Robison RK, Wang H, et al. Implementation of the FLORET UTE sequence for lung imaging. Magn Reson Med 2019; 82: 1091–1100.

[124] Hahn AD, Higano NS, Walkup LL, et al. Pulmonary MRI of neonates in the intensive care unit using 3D ultrashort echo time and a small footprint MRI system. J Magn Reson Imaging 2017; 45: 463–471.

[125] Higano NS, Spielberg DR, Fleck RJ, et al. Neonatal pulmonary magnetic resonance imaging of bronchopulmonary dysplasia predicts short-term clinical outcomes. Am J Respir Crit Care Med 2018; 198: 1302–1311.

[126] Renz DM, Herrmann KH, Kraemer M, et al. Ultrashort echo time MRI of the lung in children and adolescents: comparison with non-enhanced computed tomography and standard post-contrast T1w MRI sequences. Eur Radiol 2022; 32: 1833–1842.

[127] Förster K, Ertl-Wagner B, Ehrhardt H, et al. Altered relaxation times in MRI indicate

bronchopulmonary dysplasia. Thorax 2020; 75: 184–187.

[128] Razak A and Faden M. Neonatal lung ultrasonography to evaluate need for surfactant or mechanical ventilation: a systematic review and meta-analysis. Arch Dis Child Fetal Neonatal Ed 2020; 105: 164–171.

[129] Alonso-Ojembarrena A and Lubián-López SP. Lung ultrasound score as early predictor of bronchopulmonary dysplasia in very low birth weight infants. Pediatr Pulmonol 2019; 54: 1404–1409.

[130] Loi B, Vigo G, Baraldi E, et al. Lung ultrasound to monitor extremely preterm infants and predict bronchopulmonary dysplasia. A multicenter longitudinal cohort study. Am J Respir Crit Care Med 2021; 203: 1398–1409.

[131] Aldecoa-Bilbao V, Velilla M, Teresa-Palacio M, et al. Lung ultrasound in bronchopulmonary dysplasia: patterns and predictors in very preterm infants. Neonatology 2021; 118: 537–545.

[132] Thunqvist P, Tufvesson E, Bjermer L, et al. Lung function after extremely preterm birth-A population-based cohort study (EXPRESS). Pediatr Pulmonol 2018; 53: 64–72.

[133] Verheggen M, Wilson AC, Pillow JJ, et al. Respiratory function and symptoms in young preterm children in the contemporary era. Pediatr Pulmonol 2016; 51: 1347–1355.

[134] Davidson LM and Berkelhamer SK. Bronchopulmonary dysplasia: chronic lung disease of infancy and long-term pulmonary outcomes. J Clin Med 2017; 6: 4.

[135] Moschino L, Bonadies L and Baraldi E. Lung growth and pulmonary function after prematurity and bronchopulmonary dysplasia. Pediatr Pulmonol 2021; 56: 3499–3508.

[136] Martinez FD. Early-life origins of chronic obstructive pulmonary disease. N. Engl. J. Med. 2016; 375: 871–878.

[137] Zhang J, Zhang LP, Kang L, et al. [Dynamic observation of pulmonary function by plethysmography in preterm infants with bronchopulmonary dysplasia]. Zhongguo Dang Dai

Er Ke Za Zhi 2019; 21: 1153–1158.

[138] Greenough A and Pahuja A. Updates on functional characterization of bronchopulmonary dysplasia - the contribution of lung function testing. Front Med 2015; 2: 35.

[139] Dai D, Chen H, Dong X, et al. Bronchopulmonary dysplasia predicted by developing a machine learning model of genetic and clinical information. Front Genet 2021; 12: 689071.

[140] Bhandari V, Bizzarro MJ, Shetty A, et al. Familial and genetic susceptibility to major neonatal morbidities in preterm twins. Pediatrics 2006; 117: 1901–1906.

[141] Lavoie PM, Pham C and Jang KL. Heritability of bronchopulmonary dysplasia, defined according to the consensus statement of the national institutes of health. Pediatrics 2008; 122: 479–485.

[142] Lal CV and Ambalavanan N. Genetic predisposition to bronchopulmonary dysplasia. Semin Perinatol 2015; 39: 584–591.

[143] Avci E, Sarvari P, Savai R, et al. Epigenetic mechanisms in parenchymal lung diseases: bystanders or therapeutic targets? Int J Mol Sci 2022; 23: 10546.

[144] Fujioka K, Shibata A, Yokota T, et al. Association of a vascular endothelial growth factor polymorphism with the development of bronchopulmonary dysplasia in Japanese premature newborns. Sci Rep 2014; 4: 4459.

[145] Lal CV, Olave N, Travers C, et al. Exosomal microRNA predicts and protects against severe bronchopulmonary dysplasia in extremely premature infants. JCI Insight 2018; 3: e93994.

[146] Hamvas A, Deterding R, Balch WE, et al. Diffuse lung disease in children: summary of a scientific conference. Pediatr Pulmonol 2014; 49: 400–409.

[147] Lal CV, Bhandari V and Ambalavanan N. Genomics, microbiomics, proteomics, and metabolomics in bronchopulmonary dysplasia. Semin Perinatol 2018; 42: 425–431.

[148] Ahmed S, Odumade OA, van Zalm P, et al. Urine proteomics for noninvasive monitoring of

biomarkers in bronchopulmonary dysplasia. Neonatology 2022; 119: 193–203.

[149] La Frano MR, Fahrmann JF, Grapov D, et al. Umbilical cord blood metabolomics reveal distinct signatures of dyslipidemia prior to bronchopulmonary dysplasia and pulmonary hypertension. Am J Physiol Lung Cell Mol Physiol 2018; 315: L870–l881.

[150] Rogosch T, Herrmann N, Maier RF, et al. Detection of bloodstream infections and prediction of bronchopulmonary dysplasia in preterm neonates with an electronic nose. J Pediatr 2014; 165: 622–624.

[151] Lal CV, Kandasamy J, Dolma K, et al. Early airway microbial metagenomic and metabolomic signatures are associated with development of severe bronchopulmonary dysplasia. Am J Physiol Lung Cell Mol Physiol 2018; 315: L810–l815.

[152] Piersigilli F, Lam TT, Vernocchi P, et al. Identification of new biomarkers of bronchopulmonary dysplasia using metabolomics. Metabolomics 2019; 15: 20.

[153] Fanos V, Pintus MC, Lussu M, et al. Urinary metabolomics of bronchopulmonary dysplasia (BPD): preliminary data at birth suggest it is a congenital disease. J Matern Fetal Neonatal Med 2014; 27 Suppl 2: 39–45.

[154] Pintus MC, Lussu M, Dessì A, et al. Urinary (1)H-NMR metabolomics in the first week of life can anticipate BPD diagnosis. Oxid Med Cell Longev 2018; 2018: 7620671.

[155] Schloss PD, Westcott SL, Ryabin T, et al. Introducing mothur: open-source, platform-independent, community-supported software for describing and comparing microbial communities. Appl Environ Microbiol 2009; 75: 7537–7541.

[156] Lohmann P, Luna RA, Hollister EB, et al. The airway microbiome of intubated premature infants: characteristics and changes that predict the development of bronchopulmonary dysplasia. Pediatr Res 2014; 76: 294–301.

[157] Pammi M, Lal CV, Wagner BD, et al. Airway microbiome and development of

bronchopulmonary dysplasia in preterm infants: a systematic review. J Pediatr 2019; 204: 126–133.e122.

[158] Lal CV, Travers C, Aghai ZH, et al. The airway microbiome at birth. Sci Rep 2016; 6: 31023.

[159] Wagner BD, Sontag MK, Harris JK, et al. Airway microbial community turnover differs by BPD severity in ventilated preterm infants. PLoS One 2017; 12: e0170120.

[160] Xu Q, Yu J, Liu D, et al. The airway microbiome and metabolome in preterm infants: potential biomarkers of bronchopulmonary dysplasia. Front Pediatr 2022; 10: 862157.

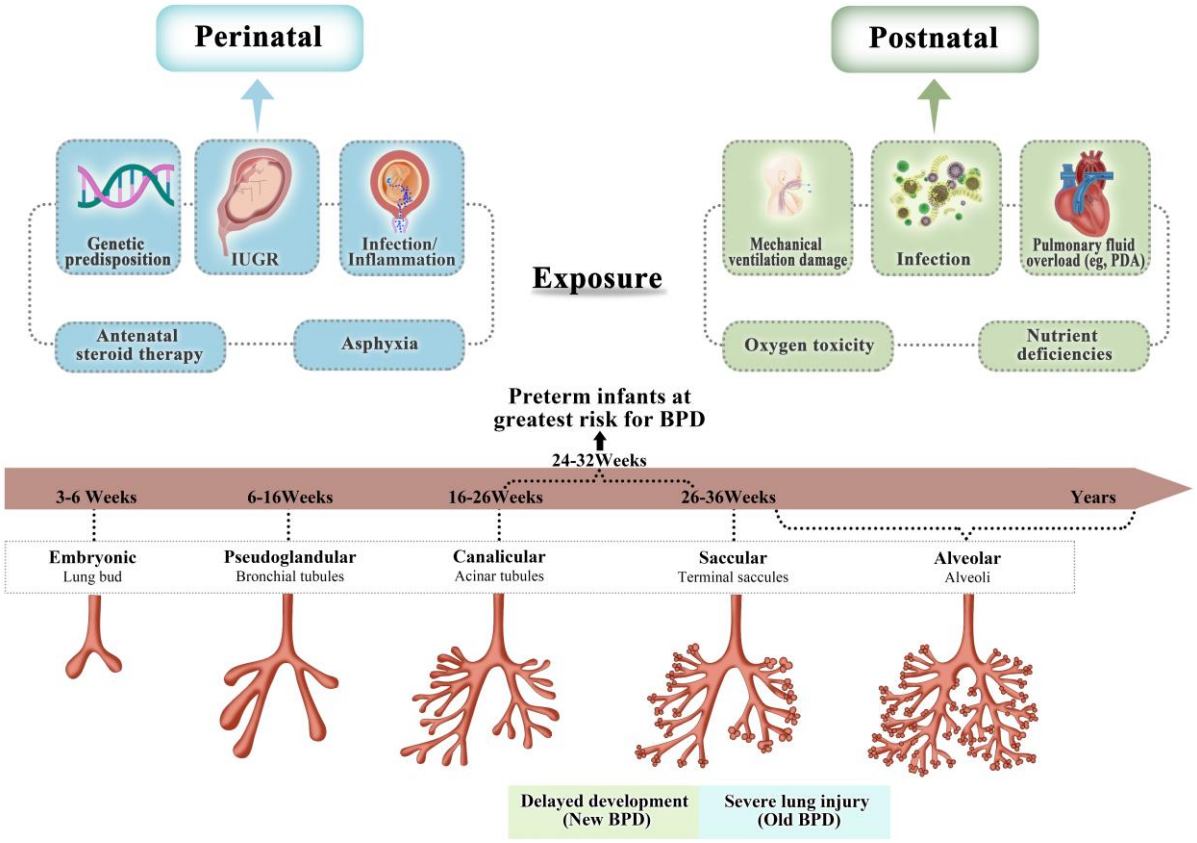
Figure legends

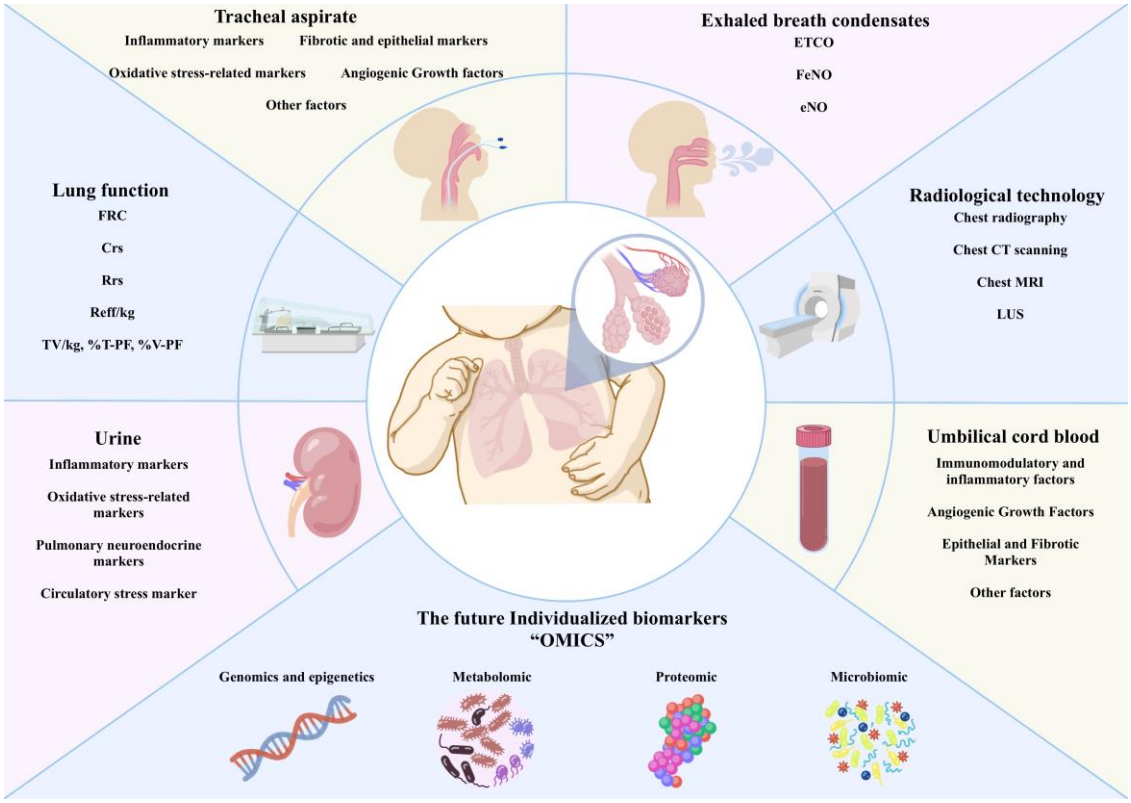
Figure 1. Lung morphogenesis and different prenatal and postnatal risk factors contributing to BPD. A schematic diagram depicting the development of the lungs from the embryonic stage until alveolarization. Changes in lung anatomy as gestation advances are shown. The proportion of "old BPD" with "severe lung damage" decreases, whereas the proportion of "new BPD" with "delayed lung development" increases. Preterm infants born at a gestational age of 24–32 weeks have a significant risk of developing BPD. Different potential prenatal (genetic predisposition, infection/inflammation, IUGR, asphyxia, and antenatal steroid therapy) and postnatal (mechanical ventilation damage, oxygen toxicity, nutrient deficiencies, infection, and pulmonary fluid overload) risk factors may contribute to BPD.

Abbreviations: BPD: bronchopulmonary dysplasia; IUGR: intrauterine growth restriction.

Figure 2. Noninvasive monitoring approaches for predicting BPD development and severity. The use of several biomarkers (such as inflammatory markers, fibrotic and epithelial markers, oxidative stress-related markers, and angiogenic growth factors) that are detectable in the umbilical cord blood, urine, or tracheal aspirate have been suggested for identifying preterm infants at a high risk for BPD. Exhaled breath condensates, lung function, and radiological innovations have the potential to provide new quantitative measures of pulmonary function and structure in BPD. The "omics" sciences have emerged as a promising new approach for BPD forecasting

Abbreviations: FRC: functional residual capacity; Crs: compliance; Reff/kg: resistance per kilogram; Rrs: resistance; TV/kg: lower tidal volume per kilogram; %T-PF: ratio of time to peak tidal expiratory flow to total expiratory time; %V-PF: and ratio of volume to peak tidal expiratory flow to total expiratory volume; eNO: elevated exhaled nitric oxide; ETCO: end-tidal carbon monoxide; FeNO: fractional exhaled nitric oxide; CT: computed tomography; MRI: magnetic resonance imaging; LUS: lung ultrasound


Perinatal
Postnatal
Genetic predisposition
IUGR
Infection/ Inflammation
Antenatal steroid therapy
Asphyxia
Exposure
Mechanical ventilation damage
Infection
Pulmonary fluid overload (eg, PDA)
Oxygen toxicity
Nutrient deficiencies
Preterm infants at greatest risk for BPD
24-32Weeks
3-6 Weeks
6-16Weeks
16-26Weeks
26-36Weeks
Years
Embryonic
Lung bud
Pseudoglandular
Bronchial tubules
Canalicular
Acinar tubules
Saccular
Terminal saccules
Alveolar
Alveoli
Delayed development (New BPD)
Severe lung injury (Old BPD)


Tracheal aspirate
Inflammatory markers
Fibrotic and epithelial markers
Oxidative stress-related markers
Angiogenic Growth factors
Other factors
Exhaled breath condensates
ETCO
FeNO
eNO
Lung function
FRC
Crs
Rrs
Reff/kg
TV/kg, %T-PF, %V-PF
Radiological technology
Chest radiography
Chest CT scanning
Chest MRI
LUS
Urine
Inflammatory markers
Oxidative stress-related markers
Pulmonary neuroendocrine markers
Circulatory stress marker
Umbilical cord blood
Immunomodulatory and inflammatory factors
Angiogenic Growth Factors
Epithelial and Fibrotic Markers
Other factors
The future Individualized biomarkers
"OMICS"
Genomics and epigenetics
Metabolomic
Proteomic
Microbiomic

Supplemental Table 1 Tracheal aspirate biomarkers associated with BPD

Biomarker	Physiologic Role	Levels associated with Increased BPD Risk (refs)
Inflammatory markers		
IL-1, -1β, -6, -8, -16, TNFα	Proinflammatory cytokine	Higher (71)
IFNγ, IL1/IL6 ratios, IL1/IL1ra ratios	Proinflammatory cytokine	Higher (72, 73)
IL-10	Anti-inflammatory cytokine	Lower (74)
MCP	Recruitment and activation of mononuclear cells	Higher (75, 76)
Nuclear factor-kappa B	Regulating major proinflammatory mediators	Higher (69, 78)
Fibrotic and epithelial markers		
MMP-8, MMP-9, MMP-9/TIMP-1 ratio	Protease/ antiprotease of extracellular matrix	Higher (11, 79, 80)
TIMP-1, TIMP-2	Antiprotease of extracellular matrix	Lower (11, 79, 80)
TGF-β1	Profibrotic	Higher (81)
CatK	Protease of extracellular matrix	Lower (83)
Oxidative stress-related markers		
8-OHdG	Endogenous oxidative stress-related DNA damage marker	Higher (AUC:0.77, cutoff: 4.4 ng/mg) (84)
Angiogenic Growth factors		
VEGF	Pro-angiogenesis	Lower (85)
sVEGFR-1	Angiogenesis	Higher (85)
S1P	Cell proliferation and differentiation marker	Higher (86)
ANG-2	Angiogenesis	Higher (88)
ET-1	Endothelium-derived vasoconstricting factor	Higher (90)
FGF-2	Extracellular matrix degradation	Higher (91)
Others		

Biomarker	Physiologic Role	Levels associated with Increased BPD Risk (refs)
MSCs	Amelioration of inflammation and lung fibrosis	Lower colony/cluster ratio (81)
Hsp-70	Cytoprotective capacity	Higher (cutoff: 149.1 ng/mg) (94)

IL: Interleukin; TNF α : Tumor Necrosis Factor α ; IFN γ : Interferon γ; MCP: Monocyte chemoattractant protein; MMP-9: Matrix metalloproteinase-9; TIMP: Tissue inhibitors of metalloproteinase; TGF-β1: Transforming growth factor β1; CatK: Cathepsin K; 8-OHdG: 8-hydroxy-2'-deoxyguanosine; AUC: Area under curve; VEGF: Vascular endothelial growth factor; sVEGFR-1: Soluble vascular endothelial growth factor receptor-1; S1P: Sphingosine 1-phosphate; ANG-2: Angiopoietin-2; ET-1: Endothelin-1; FGF2: Fibroblast growth factor-2; MSCs: Mesenchymal stromal cells; Hsp-70: Heat shock protein-70

Supplemental Table 2 Radiographical studies and lung function associated with BPD

Biomarker	Levels associated with Increased BPD Risk (refs)
Chest radiography	
Thymus	Smaller (CT/T <0.28) after birth (104)
Leaky lung syndrome, cystic BPD	Higher in newborns requiring oxygen dependency at 36 weeks of PMA (105)
Interstitial pneumonia pattern	Higher (106)
CRTA	Higher (108)
CXR Scoring system	Higher (113)
Chest CT scanning	
HRCT scoring system	strongly correlated with the severity of BPD (114)
CT scoring system	Higher in a variety of adverse outcomes in neonates with BPD (119, 120)
Chest MRI	
UTE MRI scoring system	Higher T2 and lower T1 relaxation times (127)
LUS	
LUS scoring system	Higher (131)
Consolidations and pleural line anomalies	Higher (131)
Lung function	
FRC	Lower (97, 108, 137)
Crs	Lower (97)
Rrs	Higher (97)
Reff/kg	Higher (137)
TV/kg, %T-PF, %V-PF	Lower (137)

CT/T: cardiothymic to thorax ratio; PMA: postmenstrual age; CRTA: chest radiograph thoracic area; CXR: chest radiography; HRCT: high-resolution computed tomography; UTE: Ultrashort echo-time; MRI: magnetic resonance imaging; LUS: lung ultrasound; FRC: functional

residual capacity; Crs: compliance; Rrs: resistance; Reff/kg: resistance per kilogram; TV/kg: tidal volume per kilogram; %T-PF: ratio of time to peak tidal expiratory flow to total expiratory time; %V-PF: ratio of volume to peak tidal expiratory flow to total expiratory volume.