

Bronchopulmonary dysplasia: understanding of the underlying pathological mechanisms

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The role of the clinical pathological dialogue in problem solving

Guest Editors: Gavino Faa, Vassilios Fanos, Peter Van Eyken

Abstract

Bronchopulmonary dysplasia (BPD) is a chronic lung disease occurring in preterm infants, typically before 28 weeks of gestational age, characterized by a prolonged need for supplemental oxygen or positive pressure ventilation. The normal stages of lung development and their relation to the timing of preterm birth is strategic in order to understand the pathogenesis of BPD. In embryonic and pseudoglandular stages the lungs arise from the anterior foregut as a bud where the branching morphogenesis generate a tree-like network of airways. The canalicular stage is characterized by increasing proliferation of distal lung epithelial cells and rapid expansion of the intra-acinar capillaries. The complexity of the airways increases, secondary crests begin to form and full maturation of the alveolus occurs during the saccular and the alveolar stages. Mesechyme components, especially elastin and myofibroblast, display a major role in normal lung development. BPD is thought to result after an acute insult to the neonatal lung following therapy with oxygen supplementation and mechanical ventilation. Chorioamnionitis, infections and genetic susceptibility are hypothesized to contribute to the injury that affect the normal human lung development. Abnormalities in the mesenchyme were consistently seen in association with inhibition of alveolarization. The pathological features that characterize BPD are complex and differ according with the disease progression. Alveolar simplification, interstitial fibrosis, septal thickness, large airways, smooth muscle hypertrophy, fetal artery persistence and decrease in the arterial number can be histologically observed. In conclusion, in order to reach a complete clinical-pathological diagnosis, the correlation of the pathological features with the fundamental steps of lung morphogenesis and a strict dialogue between the neonatologist and the perinatal

pathologist are required. Given these conditions, in our experience, a better understanding of the underlying pathological mechanisms of BPD might provide insight into development of new therapeutic and preventive strategies.

Keywords

Bronchopulmonary dysplasia, lung development, pathological features, histochemistry, immuno-histochemistry.

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Introduction

Bronchopulmonary dysplasia (BPD) was first described as a lung injury in preterm infants resulting from oxygen (O₂) supplementation and mechanical ventilation [1]. The pathophysiology of BPD has been extensively reviewed in recent years and subsequent research has shown that the very preterm lung, especially in smaller and more immature infants, can be injured either by O₂ or mechanical ventilation [2]. BPD results from an interference with normal lung development, triggered by the abnormal environment [3], and consequential inhibition of lung alveolar and vascular development [2]. The most important risk factor for BPD is represented by decreasing gestational age at birth. Indeed practically all infants at 22-23 weeks of gestational age develop BPD, being at a much earlier stage of lung development [4]. Moreover, chorioamnionitis, and postnatal infection have been hypothesized to affect normal human lung development [3] as demonstrated by the increase of the risk of BPD in these patients [5]. Additional risk factors for BPD, including chronic hypoxia, aberrations in vascular endothelial growth factor (VEGF) signaling and pulmonary vascular dysfunction, have been suggested [5, 6]. The aim of this work is to review the molecular pathway and the pathological features that characterized the development of BPD, in order to provide additional

understanding of the role played by different genetic and epigenetic factors in the insurgence and progression of BPD.

Normal human lung development

In order to identify the processes implicated in BPD insurgence, considering the normal stages of lung development and their relation to the timing of preterm birth is strategic (**Fig. 1**).

Embryonic stage

Human lungs, together with the trachea, arise from the anterior foregut endoderm, a tissue that generates multiple organs, including the respiratory system, esophagus, thyroid and liver [7]. The lung first appears as a bud from the primitive foregut at 3 weeks gestation [5]. Extroversion of endodermal epithelial cells results in the formation of the trachea and two lung buds, from which the embryonic lung develops. During this stage, the trachea completes its separation from the esophagus. The two lung buds undergo a highly regulated branching process defined as branching morphogenesis [7].

Pseudoglandular stage

Branching morphogenesis generate a tree-like network of airways with thousands of terminal branches [7]. Thus, through a series of new dichotomous branching the conducting airways of the lung are fully formed by 16-18 weeks gestation [8]. Moreover, the terminal bronchioles have branched into two or more respiratory bronchioles, each giving rise to clusters of acinar tubules and buds which will form the future alveolar ducts and alveoli, respectively [5].

Canalicular stage

The terminal branches narrow and form clusters of epithelial sacs that will later develop into alveoli in preparation for respiration at birth [7]. The acinar tubules and buds elongate, split, and expand, while the surrounding mesenchyme progressively thins between 18-26 weeks gestation. This stage is characterized by increasing proliferation of distal lung epithelial cells and rapid expansion of the intra-acinar capillaries. The primitive, cuboidal distal lung epithelium differentiates into immature type-II alveolar cells, able to produce surfactant, and type-I flat cells, closed to the capillaries, in charge

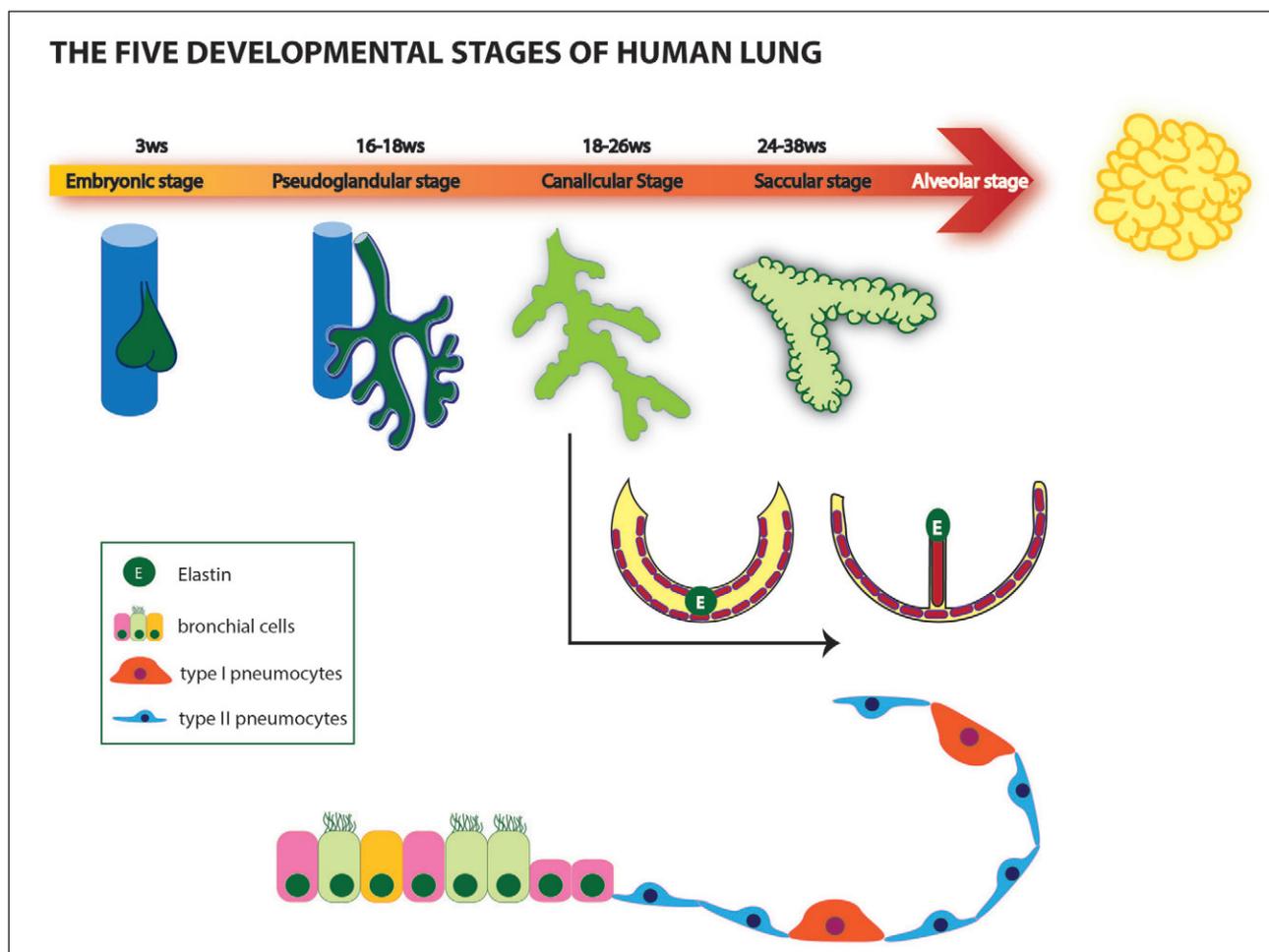


Figure 1. Schematic representation of human lung development.

to establish the gas exchange. This is most likely the earliest period of lung development impacted by preterm birth, being the period of extensive distal airspace structural and microvascular development. The presence of functional gas-exchange unit allows viability of the preterm infant [9].

Saccular stage

The pulmonary acinus mainly expands between 24-38 weeks gestation. Additional branching, elongation, and expansion of the acinar tubules and buds sets up thin-walled alveolar saccules and ducts [5]. From about 28 weeks, the complexity of the saccules increases, secondary crests begin to form. Extensive vasculogenesis within the developing terminal saccules occurs, along with a decline in airspace wall thickness [10]. Thinning of the surrounding mesenchyme together with the further differentiation of cuboidal acinar epithelial cells into type-I flat cells decreases the depth required for O_2 diffusion and increases the functional gas-exchange

volume, so that, by the end of the saccular stage, the total lung volume has increased dramatically, paralleling a progressive thinning of mesenchyme [8]. By 36 weeks alveoli are uniformly present and the total number of alveoli increases exponentially in proportion to gestational age [10]. Infants born prior to 28 weeks gestation are at highest risk of developing BPD, suggesting that disruptions occurring in saccular morphogenesis are primarily responsible for BPD pathogenesis [4].

Alveolar stage

Full maturation of alveoli occurs during the alveolarization stage [7]. The surface area of the respiratory epithelium exponentially increases from birth through early childhood. Alveoli first appear in the human saccular lung at approximately 28 weeks, then primary septa trigger the insurgence of secondary alveolar septa, and they slowly expand to approximately a fifth of the normal adult numbers by the end of gestation [11, 12].

Alveolarization mainly occurs between birth and 6 months of age, continuing throughout the first 2-3 years of life and in adulthood. Secondary septa originate from the thick saccular wall and contain a double capillary network and a central core of delicate connective tissue [12, 13]. Finally, the pulmonary microvasculature characterized by double capillary network matures towards a single capillary network, decreasing the depth of the gas diffusion barrier. Interference with any of the above components of mesenchymal maturation results in failure of secondary septation [5]. In a premature infant, this process may be disrupted by prenatal and postnatal inflammatory processes, aggravated by supplemental O₂ and positive pressure ventilation [10]. Preterm birth occurs weeks before the secondary alveolar septation is expected, thus alterations in the developing saccules persists, inhibiting future alveolarization [5].

The role of mesenchyme

During all stages of endodermal development, the lung mesoderm develops and interacts with the lung endoderm to promote branching and differentiation and to generate the various cells lineages within the lung, including airway and vascular smooth muscle cells and pericytes [7]. The new formed mesenchyme plays a key role in lung development leading the early lung development and influencing the late lung remodelling. Lung mesenchyme induces airway branching and differentiation into type-II alveolar epithelial cells [5]. The lung mesenchyme is primarily composed of fibrous structural proteins, such as type-I and IV collagen, elastin, fibronectin, and laminins, produced by fibroblasts, providing a supportive scaffold for the airspaces and airways. Elastin and collagen provide the supportive structure for the airways and alveolar spaces of the lung parenchyma, whereas fibronectins and laminins link cells to the matrix [14]. A supportive network made of glycosaminoglycans, including hyaluronic acid and heparan sulfate, and proteoglycans regulate water and growth factors, giving the right consistency to the developing lung tissue [15]. Additional mesenchymal proteins, such as tenascin and thrombospondin, exert influence on lung cells, through integrins, and modulate adhesion, migration, and cell survival [16]. Alterations in mesenchyme-associated proteins and growth factors have been associated with the development of impaired alveolarization [10]. Thus, mesenchyme is an important component that could potentially

disrupt normal lung morphogenesis. Indeed, factors that predispose to BPD, as mechanical ventilation, infection and hyperoxia, can interfere with mesenchymal structure and function and have been associated with inhibition of normal lung development [5].

Elastin

The mesenchymal structural protein elastin is the trigger site of the secondary crests into the airspaces emerging from the primary septa [12, 13]. Elastic fibers display a vital role in alveolar septation and are necessary for alveolarization in the normal lung development. Tropoelastin gene expression first appears in the pseudoglandular stage and peaks at the time of alveolarization, being localized in the site of growing secondary septal tips into the saccular airspaces. Significant increases in elastin content in elongating secondary septal tips coincide with formation and maturation of the secondary septa [17].

Myofibroblasts

Alpha smooth muscle actin (α SMA) expressing myofibroblasts mainly located within the secondary crests produce the extracellular matrix proteins that elongate the secondary septa in order to split the saccular airspace into the mature alveolus [5]. Myofibroblasts were identified as the major contributor of elastin production inside the growing secondary septal tips [18]. Loss of platelet-derived growth factor alpha (PDGFA) in the developing lung resulted in failure of α SMA-myofibroblast migration into the saccular airways, associated with a complete lack of elastin in the growing septa, confirming that these specialized fibroblasts are responsible for elastin deposition in the developing lung [19].

Pathogenesis of bronchopulmonary dysplasia

BPD was defined as O₂ dependence at 36 weeks post-menstrual age [20]. BPD is thought to result from an acute insult to the neonatal lung following therapy with high concentrations of O₂ and mechanical ventilation with high positive pressures. High concentrations of O₂ and positive pressure ventilation are responsible for cellular injury to the immature lung and subsequent impairment and inhibition of lung alveolar and vascular development. Direct cellular injury occurs

from reactive oxygen species that are not detoxified by the antioxidant defence system of the immature host [21]. The pathogenesis of BPD is multifactorial with inflammation playing a key role.

Chorioamnionitis

Preterm infants with BPD born after pregnancies complicated by chorioamnionitis may have been exposed to infection at early stages of development. However, in clinical practice, it is difficult to determine exactly when intrauterine infection first appears and is clinically relevant [5]. Infection in the intrauterine compartment is defined by histological presence of polymorphonuclear cells in choriodecidual space (**Fig. 2**), fetal membranes, and cord. Microbiologic positive culture, molecular detection methods, and biochemical elevated amniotic fluid cytokines are also helpful for the diagnosis of chorioamnionitis. The fetal inflammatory response has been defined histologically by fetal vasculitis and funisitis both characterized

by polymorphonuclear infiltration of the chorionic vessels or umbilical cord [22]. Biochemically, elevated umbilical cord concentrations of IL-1 β , IL-6 and TNF- α are detected [23]. The markers of inflammation are elevated in preterm infants with BPD. Neutrophils, macrophages, mast cells, and eosinophils are increased in the airspaces. IL-8 and macrophage protein 1 are increased while anti-inflammatory cytokines, such as IL-10 are decreased [24]. The fetal inflammatory response also involves upregulation of adhesion molecules (ICAM-1, ICAM-3, and E-selectin), matrix metalloproteinases (MMP-1 and MMP-9), angiogenic factors such as VEGF, and acute phase protein (C-reactive protein, CRP) in venous blood in the first few days of life [25]. These changes indicate that the fetus is capable of initiating a complex cascade of immune responses to microbial invasion. Anyway, infection, along with ventilator-induced trauma and hyperoxia, initiates an inflammatory cascade, which acts on the immature lung where cytokines displaying a key role in the initiation, propagation and resolution

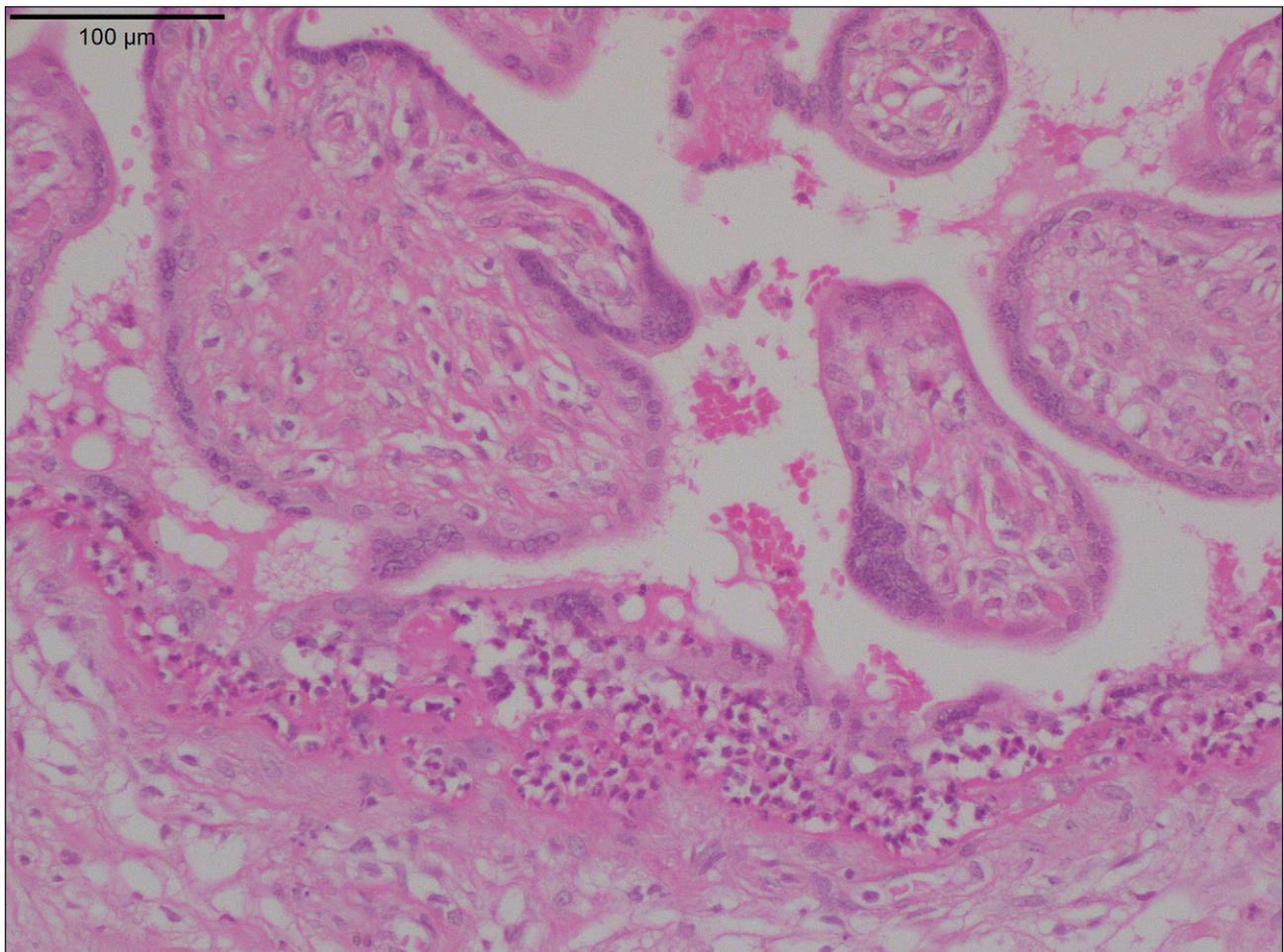


Figure 2. 23 weeks and 2 days: chorioamnionitis maternal response; hematoxylin and eosin (H&E), 20 HPF.

of this process [26]. Inflammation interferes with normal anatomic development of airway and alveoli causing abnormal healing in the premature infant ending with lung tissue damage further exacerbated by immaturity [27]. Mycoplasma species, including ureaplasma, are the most common microbes isolated from infected amniotic fluid, placentas, and the respiratory tracts of preterm infants [28] and more likely associated with postnatal sepsis [24]. Ureaplasma ability to induce inflammation in the previous sites is undeniable. Knowledge of the biology of ureaplasmas and their behavior in the respiratory tract of preterm neonates suggests that lung disease associated with these organisms is not necessarily due to direct damage from the bacteria themselves, but rather to their potent stimulation of proinflammatory cytokines (TNF- α , IL-1 β , and IL-8) or alternatively, to the blockage of counterregulatory cytokines (IL-6 and IL-10). Infections contribute to the pathogenesis of early and persistent lung dysfunction and risk for BPD [24]. Several recent investigations have examined the relationship between ureaplasma colonization of the neonatal respiratory tract and release of inflammatory mediators that may be involved in the pathogenesis of BPD [28].

Genetic susceptibility

There appears to be a genetic component too, as infants with a similar degree of lung immaturity and exposure to the very same “environmental factors” such as O₂, ventilation may or may not develop BPD. Different outcomes reported in infants matched for gestation, duration of mechanical ventilation and nutritional status, also suggest a genetic risk for the development of respiratory distress [29]. Genetic factors including race, gender, and family history of airway hyper-responsiveness have also been found to play a role in development of BPD. Genetic polymorphisms in the population may result in increased risk of developing BPD as shown for RDS. Recently, research has focused on identifying the genetic contribution to risk of intrauterine infection, preterm birth, and BPD. Elevated mid-trimester vaginal IL-1 β has been shown to be associated with increased risk for spontaneous preterm birth [30]. In women who had a preterm birth, the combination of clinical chorioamnionitis and IL-10 (-1082)*G allele was associated with an increased risk for delivery before 29 weeks gestation, suggesting a gene-environment interaction [31]. Twin concordance studies have suggested that the contribution of genetic

risk to BPD is high, accounting for 35-65% risk for the outcome. To date, studies have focused on candidate genes that encode components of the innate immune system, antioxidants, determinants of lung and vascular development and surfactant proteins. Although a number of single nucleotide polymorphisms have been described as associated with an increased or decreased risk for BPD, the associations have not been replicated in subsequent studies. This may be due to the challenges of enrolling an adequate sample size in the preterm population, to racial/ethnic heterogeneity in populations, to variations in clinical practice and to the multifactorial pathogenesis of BPD [32].

Pathological features

Depending on the degree of lung injury, the damage can resolve with growth, resulting in normal lung architecture, otherwise repair of the lung injury occurs by fibrosis, thus resulting in typical pathological features of BPD. The pathological picture of BPD is a tissue characterized by areas of normal lung architecture interspersed with areas of abnormal lung [21]. Pathologic findings of lungs of infants died with BPD, include inflammation, airway fibrosis and smooth muscle hypertrophy, associated with alveolar collapse, hyperinflation and interstitial fibrosis [33]. All these features are associated with the arrest of normal alveolar septation and pulmonary microvasculature development, according with the stage of lung development at birth, supporting the theory that BPD might result from an inhibition of normal lung maturation [34]. Autopsy samples from infants died with BPD have been described, and abnormalities in the mesenchyme were consistently seen in association with inhibition of alveolarization [35]. Pathology of the BPD lung from the pre-surfactant era was remarkable for presence of airway injury, airway inflammation and parenchymal fibrosis.

Three pathological phases of BPD have been described [36] (**Fig. 3**).

Phase 1 (acute)

The acute phase occurs within 4 days and is characterized by severe epithelial injury in airways and terminal air spaces, interstitial and intra-alveolar edema, followed by necrosis of the bronchiolar lung lining cells and plugging, which may lead to occlusion of bronchiolar lumens by necrotic debris, lymphatic dilatation and persistence of fetal artery (**Fig. 3A**).

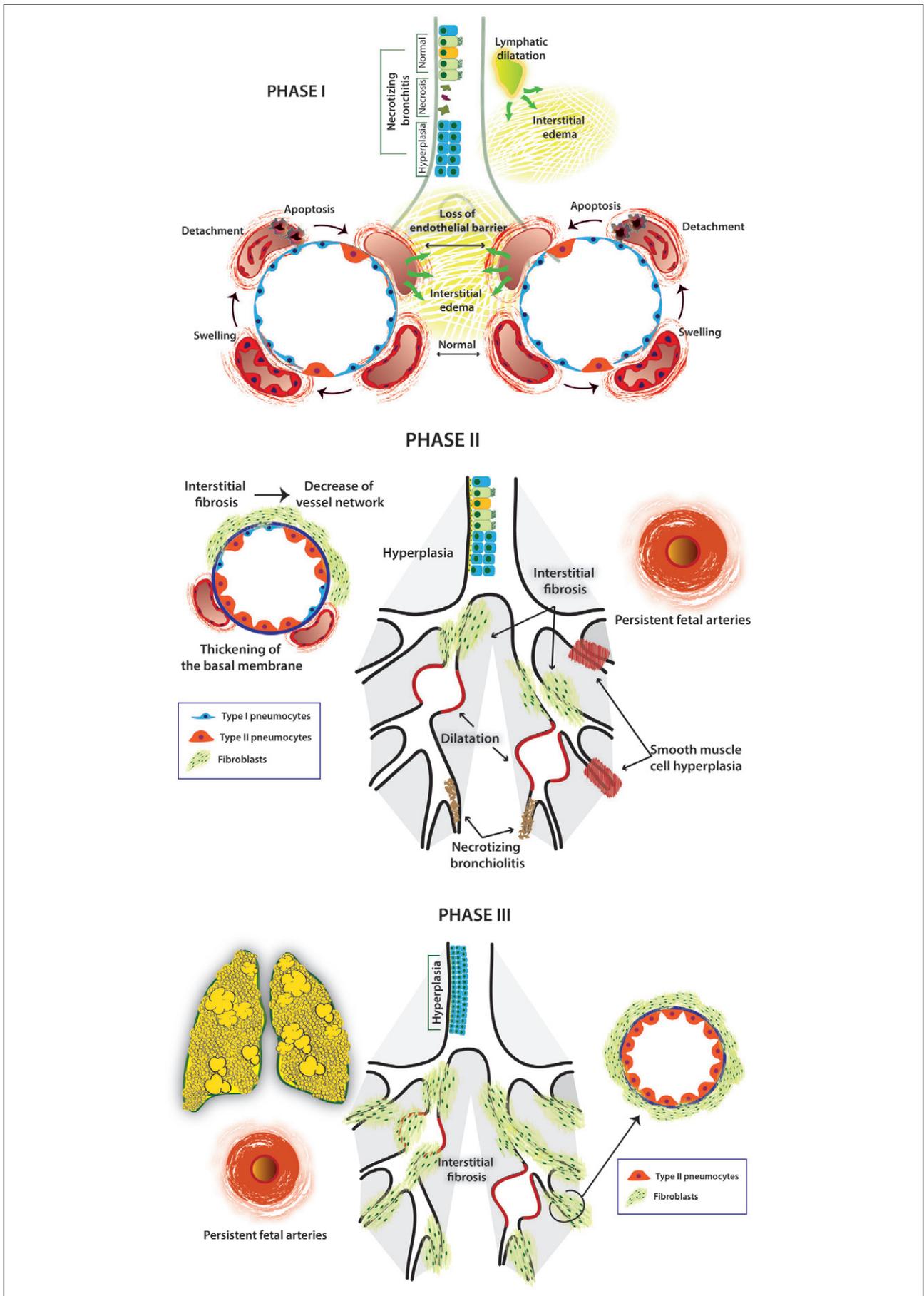


Figure 3. Schematic representation of the three pathological phases of BPD.

Phase 2 (subacute)

Before 1 month squamous metaplasia, necrotizing bronchiolitis, periairway fibrosis, reparative changes in subacute phase, leading to bronchiolar obliteration, bronchiolectasis and tracheal stenosis may be observed. Other lesions include bronchiolar smooth muscle hyperplasia with extension to lobule and prominent bronchiolar musculature, type II cell hyperplasia, thickening of the basal membrane, increasing interstitial fibrosis, decrease of vessel network and persistent of fetal artery (**Fig. 3B**).

Phase 3 (chronic)

After 1 month, lobular remodelling with inappropriate regeneration and repair and hyper-expansion of the lung, non-uniform inflation of the lung, combination of enlarged alveoli and interstitial fibrosis, dysplastic mucosal cells with prominent bronchiolar musculature, medial hypertrophy of lung artery characterize the chronic phase. At all phases, pulmonary interstitial air may be present (**Fig. 3C**).

Infants in the era of gentle ventilation and conservative O₂ supplementation demonstrate diffuse alveolar simplification due to a lack of secondary septation. They continue to display abnormalities in the mesenchyme, manifested by interstitial fibrosis and interalveolar septa thickening. However, mesenchymal abnormalities are more diffuse and homogenous than in the past and are now clearly associated with the arrested alveolarization. Preterm infants born during the surfactant era have continued to display abnormalities in the mesenchyme in association with inhibited alveolarization. Both non-surfactant-treated and surfactant-treated infants show alveolar simplification in association with interstitial fibrosis [37]. The increasing respiratory support in infants at risk for BPD have increased the total lung elastic tissue and the abnormal septal thickness in association with decreased internal surface area and alveolar duct diameter compared to gestational age-matched controls. Infants at greatest risk for BPD and need for respiratory support have demonstrated increased total collagen content in the lung as well [38]. In contrast to the delicate interstitial collagen fiber network normally seen in the developing lung, infants with BPD had thickened, tortuous and disorganized collagen fibers [5]. The most consistent residual finding in these infants is alveolar septal

fibrosis; the extent of this alveolar septal fibrosis is strikingly variable, with moderate to severe fibrosis in one area and normally inflated and or hyper-inflated lung in the adjacent sub-lobule or lobe [39]. There is more uniform inflation and less marked fibrosis and both small and large airways are free of epithelial squamous metaplasia, smooth muscle hypertrophy and fibrosis, as compared to lungs of infants that did not receive surfactant. Arrest in acinar development is observed in both the lungs of the surfactant treated as well as untreated patients affected by BPD [37]. Along with decreased alveolar number, a decrease in the arterial count has also been reported in BPD patients, so that the alveolar/arterial ratio remained normal [21].

Discussion

BPD represents the typical example of complications occurring in neonates mainly related to immaturity, reflecting a poor transition from intrauterine to extrauterine life, disarrangement in lung development being associated with toxic damage induced by O₂ therapy. This pulmonary disorder represents a leading cause of morbidity and mortality in neonates, and particularly in preterm neonates, whose lungs are characterized by morphological marker of architectural and cellular immaturity [5]. As a consequence, the histological approach to these lungs needs a differentiation between histological changes mainly due to an incomplete development and pathological lesions related to therapeutic interventions. Based on our experience, we may state that the histological interpretation of the preterm lung in the clinical setting of BPD is complex, and requires the knowledge of the main physiological changes occurring in the human lung during development at the different gestational ages. The histopathological approach to the neonatal lung in the setting of BPD includes two steps: i) the preliminary definition of the degree (and possibly the retard) of physiological pulmonary development; ii) subsequently, to focus on the pathological lesions possibly related to associated pathological events or to therapeutic interventions, including mechanical ventilation and O₂ therapy [36].

Another finding emerging from our data is the absence of any pathognomonic histological change of BPD. In the vast majority of cases, the pulmonary histological pattern may be defined as “compatible” with BPD, but not as “specific” of the disease. Therefore, the correlation of the histological changes

with the clinical and laboratory data is mandatory for a correct and complete clinical-morphological diagnosis of BPD. In fact, the definition of BPD is essentially based on clinical data (a chronic lung disease affecting neonates, characterized by associated lung injury and repair, mainly related to O₂ toxicity) and with few pathological data (altered alveolarization and vascular maldevelopment).

Taken all these data together, which is the role of the pathologist when a suspicion of BPD is prospected, based on the clinical history?

Confirm (or not) the clinical diagnosis

First, the histological examination should confirm (or not) the clinical diagnosis performed by the neonatologist, mainly based on persistent O₂ requirement and mechanical ventilation greater than 28 days after birth, and on the radiological pulmonary picture. To this end, the histological evaluation of pulmonary maturation (**Fig. 4**), including incomplete alveolarization and the persistence of fetal arteries, associated with alveolar

and or interstitial emphysema related to chronic mechanical ventilation, should be considered the most useful microscopic features [36, 39].

Determine the stage of development of the disease

After confirming the clinical diagnosis, the neonatal pathologist should determine the stage of development of the disease. As stated before, three pathological phases have been described, which may be associated with the different clinical stages of the disease, including the acute, subacute, and chronic or late stage. In the first phase, the histological picture is characterized by severe epithelial injury, affecting both the alveolar epithelium as well as the bronchial and bronchiolar epithelial cells. Histology of the second phase is characterized by reparative changes, including type II hyperplasia in alveolar spaces, squamous metaplasia of bronchial and bronchiolar epithelial cells, and variable bronchiolar obliteration and bronchiolectasis. In the third, or end-phase, the histological picture is characterized by lobular remodeling with a combination of

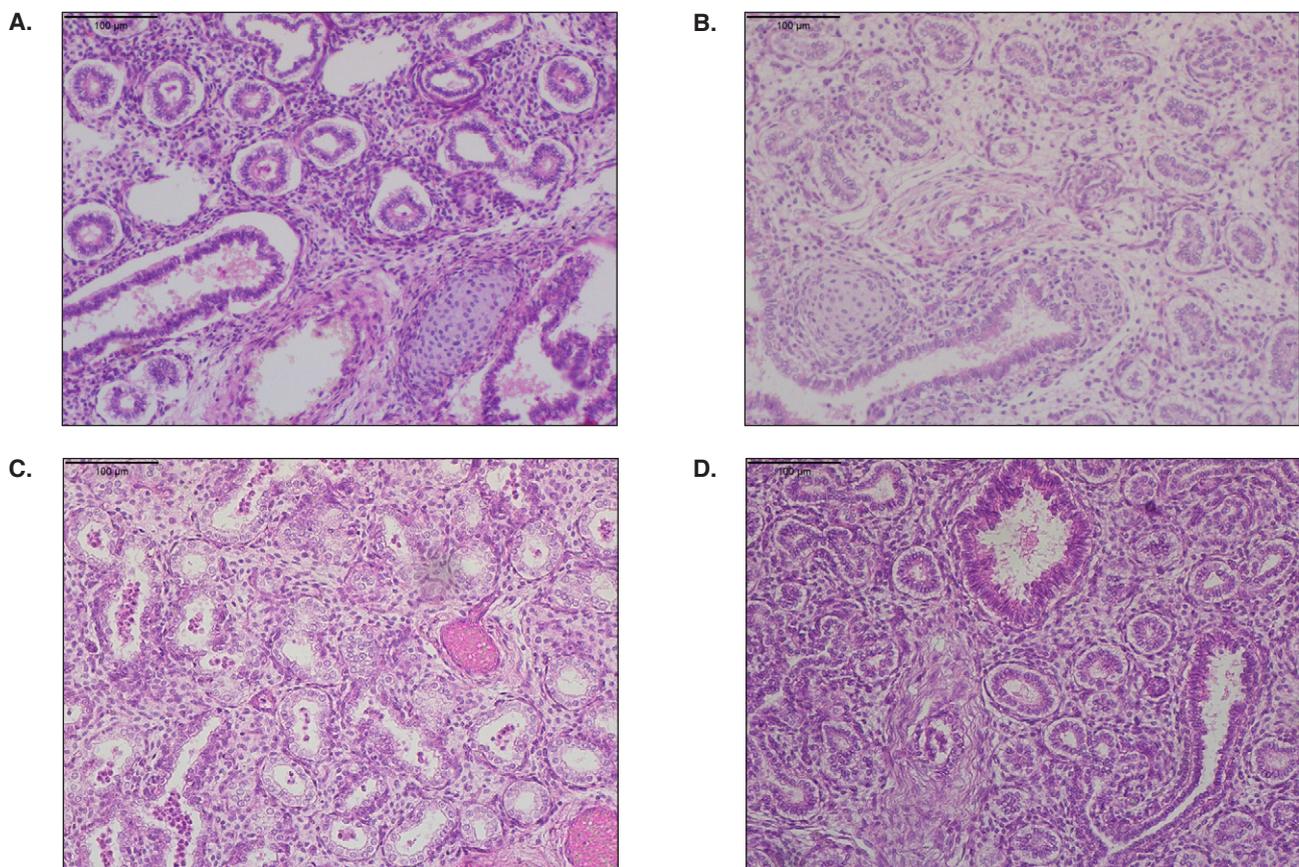


Figure 4. Lung histological section of fetuses died because of chorioamnionitis at different gestational age: **A)** 14-15 weeks; **B)** 16-17 weeks; **C)** 19-20 weeks with neutrophils inside air spaces; **D)** 20 weeks that look like 16-17 weeks, in fact all the pictures are at the same lung development stage; hematoxylin and eosin (H&E), 20 HPF.

simplified and enlarged alveoli resulting from a block in the postnatal saccular septation, with areas of atelectasis, and interstitial fibrosis [36, 39].

Apply histochemical stains

For a better definition of the pathological stage of BPD in every affected newborn, some histochemical stains may be useful (**Fig. 5**).

- i. Silver stain may show the extent of reticular fibers present in the pulmonary parenchyma, the highest degree of reticulin fibers being detected in immature lungs.
- ii. Weigert stain for elastic fibers may also help in the evaluation of the degree of maturation of the neonatal lung, elastic fibers being absent in cases of severe immaturity and progressively increasing till full lung maturation at birth.

The amount of elastic fibers may be considered as a histochemical marker of lung maturity and, associated with the degree of surfactant expression, as a sign of structural disposition to survival in the extrauterine environment. For a complete morpho-functional evaluation of the lung development, the evaluation of the amount of elastic fibers is mandatory. In fact, in the absence of elastin, alveolar septation does not occur, ending with a block in the alveolarization phase [12, 13, 17]. Moreover, the retard in the deposition of elastic fibers is responsible for a decreased elasticity of the neonatal lung, which renders the organ more susceptible to tissue damage related to mechanical ventilation. A collagen trichrome stain may be useful when the third phase of BPD is diagnosed, in order to better underline the degree of fibrosis affecting the neonatal lung [39].

Apply immunohistochemistry

Immunohistochemistry, introduced in the recent years in the analysis of lungs in newborns affected by BPD, may be very useful even in the evaluation of the degree of lung development. Immunoreactivity for type A and/or type B surfactant may help in the evaluation of lung maturation. Both proteins should be first detected in the alveolar epithelium and in the alveolar spaces, their presence being associated with adequate surface tension in the alveolus and with functional maturity of the lung [40]. Moreover, surfactant A and B immunoreactivity should be detected in the bronchiolar and bronchial epithelium, the presence of reactive cells being a sign of Clara cell development and differentiation. In the advanced stages of BPD, immunostaining for surfactant A and B should help in the evaluation of the alveolar damage, allowing a precise evaluation of the degree of type II pneumocyte hyperplasia, following apoptosis or necrosis of type I pneumocytes. Another antibody to be utilized in the evaluation of lung affected by BPD is the anti-smooth muscle actin (SMA) antibody [5]. Immunostaining with SMA antibodies may be useful for the evaluation of the medial wall of pulmonary arteries, medial hypertrophy of lung arteries being associated with immaturity, pulmonary hypertension, and with the third evolutive stage of the disease. Anti-SMA antibodies may also allow a better interpretation of the extent and degree of the peri-bronchial and peribronchiolar musculature hypertrophy, another feature typical of the advanced stages of BPD. Recently, the use of neuroendocrine markers have been proposed in the immunohistochemical evaluation of the neonatal lung affected by BPD. The number of neuroendocrine cells results increased in

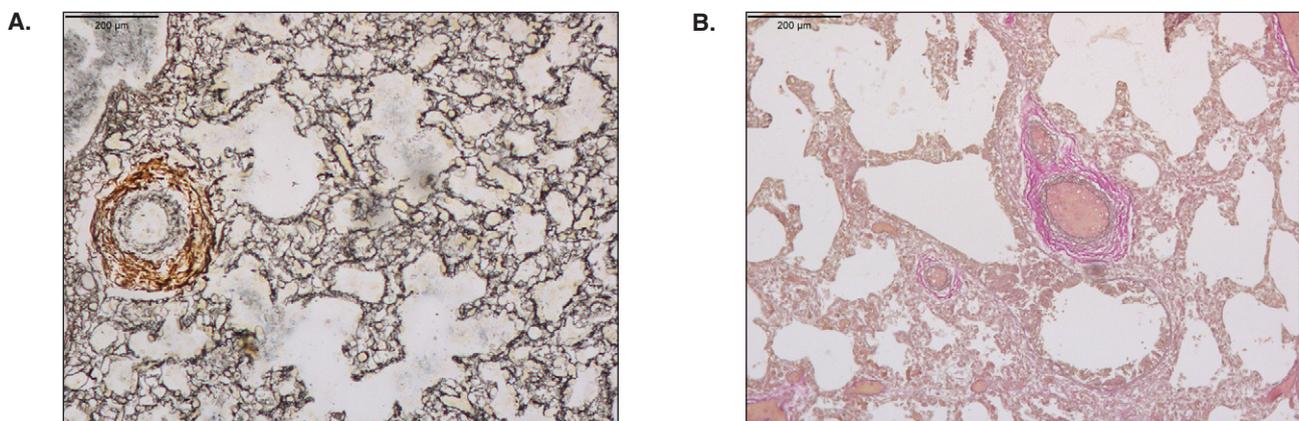


Figure 5. 36 weeks of gestational age lung of a newborn died after 1 day, showing the characteristic features of BPD: **A)** Reticulin stain; **B)** Waighert stain. 10 HPF.

BPD [21]. Relative numbers of gastrin-releasing peptide (GRP)-positive neuroendocrine cells normally decrease over the first postnatal months, and are markedly decreased in premature infants dying of RDS, thought to reflect neuroendocrine cell degranulation. In contrast, neuroendocrine cells are increased in bronchioles of infants dying with BPD at 2 weeks to 6 months of age [41]. Consistent with the increased number of neuroendocrine cells, urine GRP was also elevated in urine sample in \leq 28-week gestation infants who later developed BPD [42]. GRP is excreted as a stable peptide in the urine. Urinary GRP levels are positively correlated with bronchoalveolar lavage (BAL) GRP levels and are associated with an increased risk of BPD. Urine GRP for screening might permit early therapeutic interventions to reduce disease progression and could provide a target for new preventive therapies [20]. The GRP elevation is closely correlated with impaired respiratory function with increased oxygenation index, and also arrested alveolar number with alveolar wall thickening, decreased secondary alveolar septa, and blunted capillary tubulogenesis. Remarkably, postnatal inhibition of GRP with a blocking anti-GRP antibody prevented the functional and histological changes of BPD in animal models. These observations suggest that GRP could be an important therapeutic target to decrease BPD prevalence and later pulmonary morbidity [20, 43].

Conclusion

In conclusion, given the complexity of the pathological features that characterize BPD, two conditions may be underlined, in order to may reach a complete clinical-pathological diagnosis. First, the knowledge of the fundamental steps of lung morphogenesis, in order to discriminate between the developmental and the acquired pathological changes, all overlapping in the histological picture. Second, a strict dialogue between the neonatologist and the perinatal pathologist, in order to correlate the clinical data with the microscopic features, allowing a better explanation of the events occurred in the clinical setting. Given these conditions, in our experience, every case of BPD may represent the basis for a research project in this fascinating field of neonatology, with the aim that a better understanding of the underlying pathological mechanisms of BPD might provide insight into development of new therapeutic and preventive strategies.

Declaration of interest

The Authors declare that there is no conflict of interest.

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