REVIEW

Contribution of inflammation to lung injury and development

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Inflammation interferes with lung development in model systems and is present chronically in the lungs of preterm infants who develop bronchopulmonary dysplasia (BPD). Antenatal inflammation is very commonly associated with preterm deliveries, but there is generally minimal information about the duration, intensity, or organisms associated with chorioamnionitis. In preterm lamb models, chorioamnionitis causes a lung injury similar to BPD and also causes clinical lung maturation. Continuous exposure of the developing lung before and after delivery to inflammation may be central to the development of BPD.

The idea that inflammation may contribute to lung disease in preterm infants began with observations that oxygen exposure and ventilator mediated lung injury caused inflammation. Ogden *et al*[1] reported in 1983 that large numbers of inflammatory cells were in tracheal aspirates of infants that develop bronchopulmonary dysplasia (BPD). Proinflammatory cytokines, prostanoids, and reactive oxygen species were subsequently identified as mediators of lung injury caused by oxygen and mechanical ventilators in mature lungs.[2]

The human lung develops as an outgrowth of the foregut endoderm and thereafter undergoes progressive branching categorised into the following stages: embryonic (3–7 weeks), pseudoglandular (5–17 weeks), canalicular (16–26 weeks), saccular (24–38 weeks), and alveolar (32 weeks to 18 months post natal).[3] Thus the process of secondary septation (alveolarisation) and terminal microvascular development begins before term birth in humans. In comparison, rodent lungs alveolarise after term birth, and overexpression of multiple different proinflammatory cytokines or growth factors in mouse lungs after birth will cause inflammation and disrupt alveolar septation and microvascular development.[4] These results in transgenic animals clearly show that inflammation can interfere with normal lung development. In this article, we will review the clinical and animal experimental data on the connection between prenatal and postnatal inflammation and BPD.

EXPOSURE OF THE PRETERM LUNG TO INFLAMMATION AND ANTI-INFLAMMATION

We define lung inflammation as increased inflammatory cells in the airspaces and lung tissue that are producing proinflammatory mediators such as hydrogen peroxide, interleukin 1 (IL1), and interleukin 8 (IL8). In clinical practice, lung inflammation is detected by inflammatory cells and mediators in tracheal aspirates from intubated preterm infants. Lung inflammation may be a constant problem that begins before delivery with chorioamnionitis and continues as a result of routine care practices (ventilation and oxygen exposure) and adverse clinical events, such as nosocomial infection (fig 1). The proinflammation is counterbalanced by anti-inflammatory effects of corticosteroids, because about 80% of preterm infants are exposed to antenatal corticosteroids. In fetal sheep, maternal betamethasone initially suppresses the inflammatory responses of fetal plasma monocytes and suppresses endotoxin induced chorioamnionitis and lung inflammation.[5] However, seven days after maternal corticosteroid treatment, the inflammatory responses of plasma monocytes increase and the lung inflammation associated with chorioamnionitis increases.[6] Postnatal corticosteroids also acutely decrease indicators of inflammation in tracheal aspirates of infants with BPD, but subsequent effects on inflammatory responses have not been evaluated.[2] Furthermore, both antenatal and postnatal corticosteroids inhibit alveolar septation in multiple animal models. The therapeutic conundrum is that both proinflammatory and anti-inflammatory mediators disrupt alveolarisation and to date no treatments are available to promote alveolarisation. Inflammation is common in the lungs of the preterm fetus and newborn, and complex interactions between inflammation and corticosteroids remain unexplored mechanistically. Indomethacin, another anti-inflammatory agent, does not seem to decrease the risk of BPD in preterm infants.[7]

ANTENATAL INFLAMMATION CHORIOAMNIONITIS

Most deliveries before 30 weeks gestation are associated with histological chorioamnionitis, which is often clinically silent.[8] The more preterm the delivery, the more often histological chorioamnionitis is detected[9] (fig 2). Most infants delivered before 30 weeks gestation also have amniotic fluid that is culture positive for low pathogenic organisms such as *Ureaplasma* and *Mycoplasma*.[8] Recent reports further show that about 10% of amniotic fluids collected for

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Abbreviations: BPD, bronchopulmonary dysplasia; CPAP, continuous positive airway pressure; IL, interleukin; RDS, respiratory distress syndrome

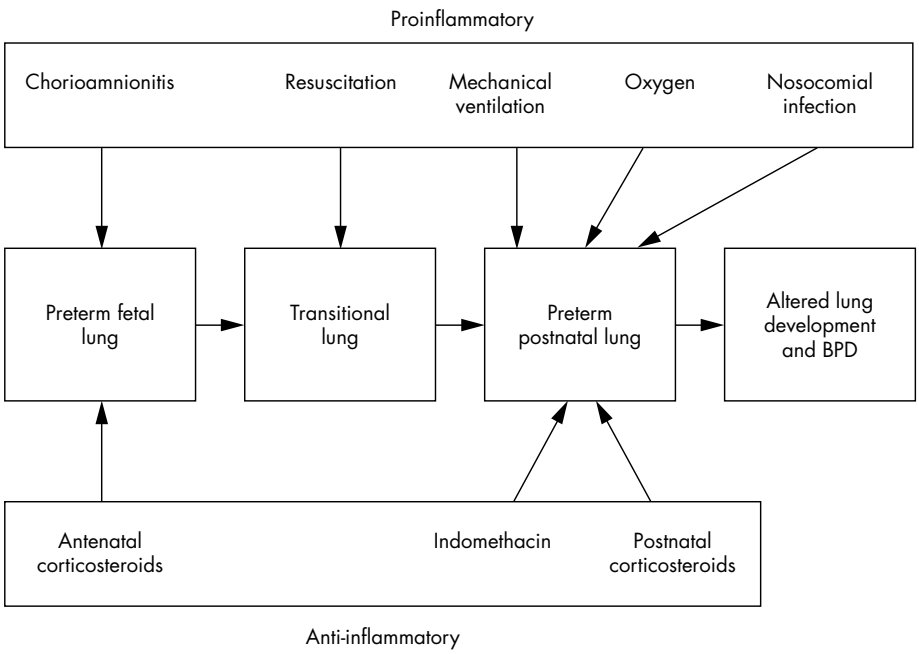

Figure 1 Proinflammatory and anti-inflammatory influences on the fetal and preterm lung.

genetic assessments at about 18 weeks gestation were polymerase chain reaction (PCR) positive for *Ureaplasma*. Recently, Steel *et al*[10] used a bacterial 16S ribosomal RNA probe to show that only 10% of fetal membranes from 29 preterm deliveries had no bacteria deep in the membranes. More problematic for the diagnosis of chorioamnionitis, some membranes had bacteria and no inflammation. In contrast, another recent report used the same molecular probe and found good congruity between PCR identification of organisms and histological chorioamnionitis.[11] Chorioamnionitis is a major diagnostic problem: clinical chorioamnionitis does not correlate well with histological chorioamnionitis or culture positive amniotic fluid, and some preterm deliveries may have organisms with neither histological nor culture positive chorioamnionitis. Chorioamnionitis (however diagnosed) may often be a chronic and indolent process that has been present for weeks or months before the onset of preterm labour. Chorioamnionitis represents a maternal and/or fetal inflammatory response that, in most very low birthweight deliveries, is of indeterminate duration, undefined severity, and generally caused by unidentified organisms.

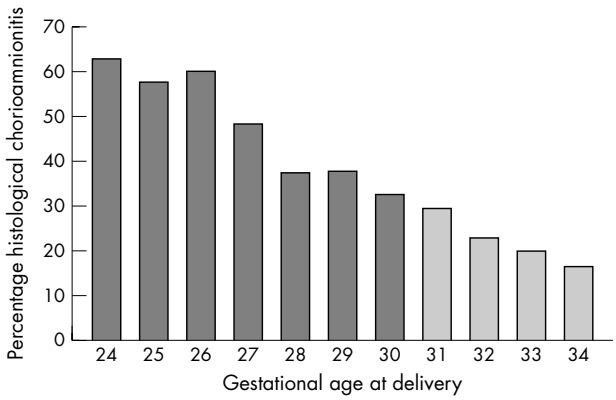

Figure 2 Percentage of infants delivered at the University of Sydney with placental membranes with histological chorioamnionitis. The graph includes 3928 deliveries. Redrawn from Lahra and Jeffery.[9]

RESPONSES OF THE FETAL LUNG TO CHORIOAMNIONITIS

Fetal responses to chorioamnionitis vary depending on the severity of the inflammation and the populations of infants studied. Raised concentrations of proinflammatory cytokines such as IL1β, IL6, and IL8 in amniotic fluid measured within five days of preterm delivery predicted the subsequent development of BPD. However, the predictive values for BPD were better for raised concentrations of cord plasma IL6 than for IL6 in the amniotic fluid.[12] Gomez *et al*[13] proposed that chorioamnionitis associated with a fetal inflammatory response (increased IL6 concentrations in cord blood) was equivalent to the acute respiratory distress syndrome (ARDS) in adults with systemic inflammatory mediators targeting the lungs. Severe histological chorioamnionitis is associated with a higher risk of subsequent BPD than is mild histological chorioamnionitis or no chorioamnionitis[14] (fig 3). In one small series, ventilated preterm infants exposed to histological chorioamnionitis had proinflammatory cytokines in their tracheal aspirates soon after birth and an increased risk of BPD.[15] In contrast, chorioamnionitis predicted a decreased risk of BPD for a population of preterm infants, unless the infants were ventilated or developed nosocomial sepsis after birth.[16] In several reports, histological chorioamnionitis and culture positivity for *Ureaplasma* were associated with a decreased incidence of respiratory distress syndrome (RDS) but an increased risk of BPD.[15] The associations of chorioamnionitis with a decrease in RDS and an increase in BPD are not consistent across the literature, probably because of the imprecision of the diagnosis of the severity, duration, and organisms associated with chorioamnionitis and the different populations of infants included in the reports.

CHORIOAMNIONITIS IN ANIMAL MODELS

The diagnosis of chorioamnionitis in humans is imprecise and poorly characterised. Therefore we have created models of chorioamnionitis in fetal sheep to explore how fetal exposure to inflammation can influence the lung. Intra-amniotic injections of *Escherichia coli* endotoxin resulted in large increases in inflammatory cells in the chorioamnion and amniotic fluid and increased expression of IL1β, IL6, and IL8.[17] The fetal lung responded with an acute but modest

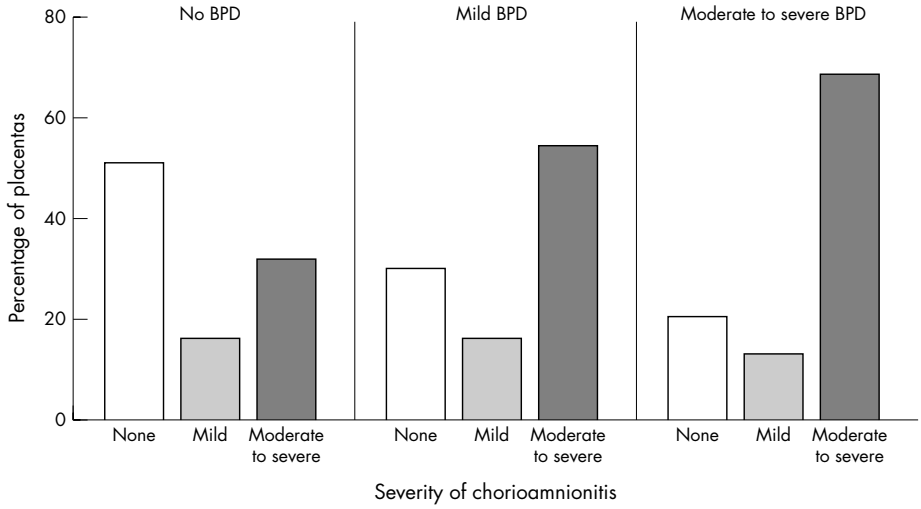

Figure 3 Relation between severity of histological chorioamnionitis and the occurrence of bronchopulmonary dysplasia (BPD) in very low birthweight infants. Definitions of severity of chorioamnionitis and original data from Viscardi *et al*.[14]

generalised inflammatory response (inflammatory cell influx and expression of proinflammatory cytokines one to two days after intra-amniotic endotoxin). Intra-amniotic endotoxin did not induce a large systemic inflammatory response or an increase in plasma cortisol.[18] The fetal lung inflammation was associated with decreased microvascular development and decreased alveolar septation by seven days, resulting in mild BPD-type anatomical changes.[19] However, the inflammation resulting from single intra-amniotic doses of endotoxin or the proinflammatory cytokine IL1 resolved with striking lung maturation as indicated by increased surfactant and improved lung function and no indications of progressive injury.[20] These results show that intra-amniotic inflammation can induce lung maturation (less RDS) by a cortisol independent pathway and cause anatomical changes consistent with BPD. The experimental studies validate the concept that fetal exposure to inflammation can have profound effects on the fetal lung. Surprisingly, repeated or 28 day continuous endotoxin exposures did not progressively injure the fetal sheep lung, showing that the fetus can also modulate inflammation.[21]

INJURY RESULTING FROM RESUSCITATION

The preterm lung can also be easily injured by mechanical ventilation after delivery because lung structure is immature with decreased amounts of collagen and elastin, low lung gas volumes, and surfactant deficiency.[4] The research on ventilation and lung injury is clear: ventilation of lungs from low functional residual capacities or to volumes that overstretch the lungs results in inflammation from stretch/volume mediated injury. Although less information is available for preterm lungs, Bjorklund *et al*[22] and Wada *et al*[23] showed that initiation of ventilation in preterm lambs with large volumes severely injures the lungs. Ventilation of preterm lungs without positive end expiratory pressure also increases proinflammatory cytokine expression.[24] The variables that may contribute to lung injury with the initiation of ventilation at birth (volume, positive end expiratory pressure, oxygen concentration, gas humidity, and temperature) have not been individually evaluated for their contributions to lung injury in the preterm requiring resuscitation at birth. Each variable can induce inflammation, and most resuscitations are performed with 100% oxygen or a gas mixture that may not be heated or humidified and without tidal volume monitoring or control of positive end expiratory pressures.

POSTNATAL VENTILATION AND OXYGEN EXPOSURE

The experimental literature clearly shows that prolonged ventilation of the developing lung will cause BPD-type anatomical changes associated with persistent inflammation.[25] Exposure of lungs to high concentrations of oxygen also causes severe inflammation. Although different styles of mechanical ventilation can be more or less injurious, an endotracheal tube that bypasses normal airway protective mechanisms can introduce infection, which will contribute to the risk of BPD. The clinical literature supports the correlation of an increased risk of BPD with high and persistent levels of multiple proinflammatory mediators in tracheal aspirates.[2] Corticosteroid treatment can acutely decrease mediator concentrations and improve lung function, a response that is consistent with decreased inflammation. For infants who require mechanical ventilation, the current practice is to use ''adequate'' positive end expiratory pressure (no practical way to know what is adequate), low tidal volumes, and the acceptance of higher P_{CO_2} levels to decrease the risk and severity of BPD.[26] No studies have demonstrated that ventilation styles that may cause less severe BPD also decrease lung inflammation.

Non-invasive continuous positive airway pressure (CPAP) has been proposed as a way to prevent BPD in infants that do not require mechanical ventilation.[27] The initiation of CPAP from birth did decrease some indicators of lung inflammation in preterm lambs relative to mechanical ventilation.[28] Chronic CPAP also resulted in minimal anatomical changes in preterm baboon lungs.[29] The general consensus is that CPAP decreases the risk and severity of BPD, although many infants receiving CPAP continue to develop BPD. There is no information about how prolonged use of CPAP may modulate inflammatory responses in the preterm lung.

POSTNATAL SEPSIS

Nosocomial infections causing sepsis and/or pneumonia are very common in preterm infants. Sepsis events must target the lungs because postnatal sepsis is associated with progression of BPD.[30] These infectious events may be a two way street: systemic infection results in lung inflammation analogous to the adult RDS, and inflammation of the preterm lung can become systemic with mechanical ventilation.[31]

THE CONCEPT OF CONTINUOUS INFLAMMATION CONTRIBUTING TO BPD

If the incidence of chorioamnionitis in humans and the results from animal models are correct, then most very low birthweight infants born at less than 30 weeks gestation have had inflamed lungs before delivery. Although resuscitation of the preterm lung with positive pressure ventilation can cause inflammation, the effects of resuscitation on the already inflamed lung have not been evaluated. Short term ventilation of the previously inflamed fetal lamb lung increased monocyte and lymphocyte recruitment to the lungs, indicating an increased innate immune response.[32] Corticosteroids and prior inflammation each modulate subsequent inflammatory responses by the preterm lung. However, the effects must depend on timing and chronicity of the exposures. For example, a single exposure of the fetal lung to a proinflammatory mediator will result in augmented monocyte responses while repeated exposures paralyse monocyte responses.[33] Understanding the net effects of clinical interventions on inflammatory responses and how those responses modulate lung development is the challenge for the future. In the mean time, it is just logical to decrease the proinflammatory exposures to the preterm as much as possible.

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