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Ureaplasma and BPD

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Abstract

Ureaplasma is an organism with low virulence and is a commensal of the lower genito-urinary tract in females. From here, it can gain entry in the amniotic fluid to cause inflammation in the amniotic compartment during pregnancy. *Ureaplasma* spp. are the most common organisms isolated from women with chorioamnionitis. *Ureaplasma* spp. are associated with increased risk for preterm labor and morbidity in the preterm neonate. However, there is some controversy regarding the importance of *Ureaplasma* in the pathogenesis of bronchopulmonary dysplasia (BPD). This article will review the microbiology of *Ureaplasma*, host innate immune responses, and the pathology of lung injury in animal models of *Ureaplasma* chorioamnionitis. We will review epidemiological studies of *Ureaplasma* and BPD in preterm infants and efficacy of antibiotics in preventing preterm labor and BPD.

The *Ureaplasmas* are generally regarded as commensals of the lower genital tract in both males and females^{1,2}. However, *Ureaplasma* infection of the upper genital tract during pregnancy is associated with adverse pregnancy outcomes including preterm births and neonatal morbidity³.

Microbiology of *Ureaplasma* Species

Ureaplasma spp. are among the smallest free-living, self-replicating microorganisms. They have an extremely low G+C content of 25.5% (or the highest A+T composition) within open reading frames of any prokaryotes sequenced to date. *Ureaplasmas* have evolved from Gram-positive bacteria by degenerative evolution to lose the peptidoglycan cell wall⁴. As their name suggests, *Ureaplasmas* utilize urea as their sole source of carbon, producing ammonia as a metabolic product⁵. Currently, there are 2 identified species of *Ureaplasmas* that infect humans that are divided into 14 antigenically distinct Serovars⁶. Prior to 2000, the 14 Serovars were considered to be the single species *U. urealyticum*. Their separation into *U. parvum* (Serovars 1, 3, 6 and 14) and *U. urealyticum* (Serovars 2, 4, 5, 7–13) is based on phylogenetic evidence⁷. However, the new taxonomic classification has not been fully adopted in the literature and the 14 Serovars are often referred to as *U. urealyticum*.

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Virulence of *Ureaplasma* Species

Several proteins have been proposed as virulence factors. The *mba* gene encodes for MBA, the major surface-exposed lipoprotein. MBA is thought to be the major virulence factor of *Ureaplasma* spp. and is the predominant antigen recognized by the host immune system during infection⁴. *Ureaplasmas* can alter the expression of their MBA in order to evade host immune responses and maintain chronicity of infection^{8–10}. *Ureaplasma* Phospholipase A and C activities were identified by *in vitro* hydrolysis of an artificial phosphate ester¹¹. The phospholipases could potentially generate prostaglandins – a known trigger of labor¹¹. Similarly, an IgA protease activity, which could destroy mucosal IgA, was demonstrated in *Ureaplasma*¹². However, genome analyses of all the *Ureaplasma* Serovars did not find sequences for either the IgA protease or phospholipase A or C¹³. However there was a gene encoding a phospholipase D domain containing protein¹³. The urease activity of *Ureaplasma* generates ammonia from the cleavage of urea⁵, which can cause toxicity to host tissues due to change in pH. To date there is no definitive identification of specific virulence factors in *Ureaplasma*.

Recent studies demonstrated that horizontal gene transfer occurs in *Ureaplasma* isolated from clinical samples, resulting in genetic hybrid forms of *Ureaplasma* Serovars, implying unstable genotypes during the course of infection¹⁴. Nevertheless, there is no evidence of Serovar specific pathogenic effects or severity of infection^{10,13}. An experiment in the sheep illustrates the instability of *Ureaplasma* phenotype. Sheep were given an intra-amniotic injection with either a virulent or non-virulent-derived *Ureaplasma* clones (both *Ureaplasma parvum* Serovar 6) at 55 days of gestation. Virulence was defined as *Ureaplasma* recovered from amniotic fluid of sheep with severe chorioamnionitis or minimal chorioamnionitis. Although both *Ureaplasma* isolates caused chronic colonization and fetal inflammation, the severity of chorioamnionitis or fetal inflammation was not different after 70d of colonization of the fetal compartment¹⁵. Therefore the original phenotype of *Ureaplasma* virulence was not sustained during the chronic infection. Virulence and persistence will also be influenced by the ability of microorganisms to form biofilms. The majority of clinical isolates of *Ureaplasma* form biofilms¹⁶.

Ureaplasma as Perinatal Pathogens Causing Preterm Birth

The microbial invasion of amniotic cavity associated with preterm birth is relatively unique among infectious diseases in humans. The microbes are mostly opportunistic commensal vaginal organisms of low pathogenicity in otherwise healthy women. Polymicrobial growth is common from amniotic fluid from chorioamnionitis, and *Ureaplasma* spp. are the organisms most frequently isolated, although they rarely cause infections elsewhere^{13,17}. The strongest evidence that *Ureaplasma* can cause preterm labor is from experiments in Rhesus macaques. Intra-amniotic injection of *Ureaplasma parvum* or the related organism *Mycoplasma hominis* induced chorioamnionitis, fetal inflammation and preterm labor¹⁸. In human pregnancies, several studies demonstrate the association of *Ureaplasma* species and preterm labor. Using PCR amplification of 16S ribosomal DNA, *Ureaplasma* species were the most common microbial genus identified in amniotic fluid of women with preterm premature rupture of membranes¹⁷. These organisms can be isolated in the amniotic fluid in the second trimester and can remain clinically silent for several weeks to months¹⁹. Gerber et al sampled amniotic fluid by amniocentesis from 254 asymptomatic women at 15–17 weeks' gestation, and *Ureaplasma* species were identified in 11% subjects. Preterm labor occurred in 59% of *Ureaplasma*-positive women compared with 4% *Ureaplasma*-negative women. Preterm births were also significantly higher in *Ureaplasma*-positive women compared to *Ureaplasma*-negative women²⁰. Isolation of *Ureaplasma* from the amniotic fluid was associated with chorioamnionitis and inflammatory mediators in the amniotic fluid

– known triggers of preterm labor^{21,22}. The current hypothesis for infection related preterm labor is that the organisms from lower genital tract (e.g. *Ureaplasma*) ascend through the cervix into the choriodecidual space. The localized inflammation in this region traverses through the chorion-amnion and disseminates in the amniotic fluid. This leads to a generalized inflammation of the amnion and the chorio-decidua resulting in the production of pro-inflammatory mediators such as interleukin-1beta (IL-1 β), IL-6 and prostaglandins that ultimately initiate preterm labor²³.

Host Response to *Ureaplasma* Infection

Microbial recognition by innate immune systems can be mediated by a variety of germline-encoded receptors, including Toll-like receptors (TLRs), RIG-like receptors (RLRs), Nod-like receptors (NLRs), and cytosolic DNA sensors such as the HIN200 family member AIM2²⁴. *Ureaplasma sp.* lack a gram-negative or gram-positive bacterial cell wall, thus are devoid of lipopolysaccharides or peptidoglycans – the microbial products that are potent activators of TR4 (lipopolysaccharide or LPS) and the TLR2 or NOD1/2 (peptidoglycan) pathways. Nevertheless, placental leukocytes or neonatal monocytes exposed *in vitro* to *Ureaplasma* spp. induce the release of inflammatory cytokines^{25,26}. Shimizu et al²⁷ showed that *U. parvum* lipoproteins, including the multi-banded antigen activate nuclear factor-kappa B (NF- κ B) in reporter cell lines via TLR1, TLR2 and TLR6 signaling. Peltier et al.²⁸ found that the macrophage-stimulating activity from *U. urealyticum* is mainly due to lipoproteins, and signaling involving TLR2 or TLR4 receptors. Mechanisms of immune alterations induced by *Ureaplasma in vivo* have not been identified. However, the *Ureaplasmas* do induce antibody production in both humans and animals^{15,29}.

Ureaplasmas are generally considered to be microorganisms with low virulence. However, it is conceivable that in immunodeficient hosts, the organism may have enhanced virulence. Surfactant protein-A (SP-A) is an innate host defense molecule that is secreted by the lung and is deficient in preterm infants³⁰. Compared to wild type mice, SP-A deficient mice had delayed clearance of *Ureaplasma* from the lungs, increased of inflammatory cells and pro-inflammatory cytokine expression³¹. This observation may be relevant to preterm fetuses and neonates who will have low levels of SP-A and other innate host defense factors in the lungs.

Ureaplasma and Immune Modulation

Chronic infections can induce a state of immune paralysis called endotoxin tolerance. Thus leukocytes from patients with sepsis or recovering from typhoid fever can have endotoxin tolerance^{32, 33}. We reported that in sheep repeated intra-amniotic injections of LPS (endotoxin), a constituent of cell wall of gram negative bacteria, induces tolerance to a variety of toll-like receptor agonists in the preterm fetus^{34–36}. Interestingly, chronic intra-amniotic colonization/infection by *Ureaplasma* also profoundly diminished responses to intra-amniotic LPS in the preterm fetal sheep³⁷ (Figure 1). A benefit of endotoxin tolerance is decreased inflammation in the host and therefore decreased organ injury. However, endotoxin tolerance also can increase susceptibility to infections due to suppression of innate immune responses. These observations in preterm fetuses may have direct relevance to immune responses of newborns at risk of developing bronchopulmonary dysplasia (BPD).

Ureaplasma Species and Lung Inflammation in the Developing Lung – Animal Studies

Ureaplasma infection of the developing lung has been studied primarily in sheep and non-human primates. Intra-amniotic injection of *Ureaplasma* in early gestation sheep resulted in

efficient colonization and a 5-log increase in the *Ureaplasma* in amniotic fluid counts that persisted for 3 months to term with very little overt adverse effects in the ewe, consistent with a commensal-like host response¹⁵. Interestingly, after a chronic exposure to intra-amniotic *Ureaplasma*, all of the fetal lungs were colonized¹⁰. Although *Ureaplasma* was not cultured from either the fetal or maternal blood after an intra-amniotic injection, about 50% of the ewes had an IgG antibody response^{10,15}. Although chorioamnionitis of varying severity was demonstrated in a majority of the sheep, about 10% of the fetal membranes showed no chorioamnionitis despite recovery of high titers of *Ureaplasma* from the amniotic fluid over 3 months¹⁰. These experiments illustrate the complexities in understanding the host response to a *Ureaplasma* exposure.

Recruitment to and activation of inflammatory cells in the fetal lung could be detected as early as 3d after intra-amniotic injection of live *Ureaplasma* in sheep³⁸ (Figure 2). Both monocytes and neutrophils increased, and MHCII expression in the monocytes increased 14d after intra-amniotic *Ureaplasma* injection, consistent with maturation of the monocytic cells³⁸. The inflammatory cell infiltration was focal in nature and no areas of consolidation or “pneumonia” were detected. This modest microscopic inflammation would not be detectable by standard X-rays. The inflammatory infiltrate was accompanied by modest increases in the pulmonary expression of the pro-inflammatory cytokines/chemokines IL-1 β , IL-6 and IL-8 within 1 week that persisted for at least 6 weeks^{39,40}.

The modest lung inflammation was followed by the counter-intuitive observation of significant increases in lung gas volumes and surfactant lipids in the preterm fetal sheep. This early lung maturation was first detected in the preterm fetal lungs 3 weeks after intra-amniotic *Ureaplasma* injection and persisted for 10 weeks despite continuous exposures⁴¹ (Figure 3). Although these striking effects on lung physiology are consistent with clinical “lung maturation”, they probably represent “dysmaturation” since the improved lung physiology was accompanied by evidence of impaired lung development³⁸. Fourteen days after intra-amniotic *Ureaplasma* injection, preterm fetal sheep delivered at 80% gestation had decreased elastic foci and increased smooth muscle around bronchioles and pulmonary artery/arterioles³⁸ (Figure 4). These changes in elastin and smooth muscle are similar to those reported for infants with BPD⁴². Therefore the benefits from acute improvement in lung physiology were somewhat offset by changes in lung architecture. However, alveolar and pulmonary vascular development was no different between the controls and fetus exposed to *Ureaplasma* for 70 d⁴³. Therefore the pathologic changes detected at 14d were not sustained during more extended exposures, suggesting that repair mechanisms must be operative.

In Rhesus macaques, intra-amniotic injection of *Ureaplasma* caused chorioamnionitis, preterm labor and delivery within 15d¹⁸. Both *Ureaplasma parvum* and the related organism *Mycoplasma hominis* given by the intra-amniotic route caused fetal pneumonia characterized by increased neutrophils and macrophages and alveolar type II cell proliferation indicating injury¹⁸. In another experiment, pregnant baboons were given intra-amniotic *Ureaplasma* at 65% gestation. The preterm baboons were delivered operatively 2–3d later and were given mechanical ventilation for 14d. Half of the neonatal baboons cleared *Ureaplasma* from their airways, while the remaining half had persistent *Ureaplasma* colonization of the lungs⁴⁴. The neonatal baboons with persistent *Ureaplasma* colonization had lung inflammation and worse lung function compared to those animals that cleared the *Ureaplasma* from the lungs. Lung pathology at autopsy of the persistently *Ureaplasma* positive at 14d revealed a mixed monocytic, lymphocytic and to a lesser extent neutrophilic infiltrate in the lungs⁴⁴. In general, the lung pathology after intrauterine *Ureaplasma* exposure was more severe in non-human primates compared to sheep, suggesting species differences in susceptibility to *Ureaplasma*.

Ureaplasma Species and Lung Inflammation in the Developing Lung – Human Studies

The lung pathology of archived autopsy specimens from *Ureaplasma* infected preterm infants demonstrated increased lung fibrosis, elastic fiber accumulation, smooth muscle actin, and increased tumor necrosis factor alpha (TNF α) and transforming growth factor beta 1 (TGF β 1) immunoreactivity^{45,46}. Elevated maternal and fetal or newborn antibody titers to *Ureaplasma* correlated with an increased incidence of stillbirth and infants with fatal neonatal respiratory disease²⁹. The findings from these limited human studies are consistent with lung pathology described in animal studies.

Pathogenesis of BPD

The most predictive risk factors for development of BPD are gestational age, birth-weight, duration of respiratory support, fraction of inspired oxygen (FiO₂), race and gender⁴⁷. Although these risk factors hold true on a population basis, individual infants who develop BPD have different trajectories toward developing BPD. For instance, if the need for supplemental oxygen is used as a proxy for lung injury, then three patterns (or subsets) of BPD are discernible^{48,49}. In a study of preterm infants <28 week gestation at birth, 20% of the infants received a consistently low FiO₂, 38% had pulmonary deterioration (low initial FiO₂ followed by increased oxygen need), and the remaining 43% had consistently high FiO₂ needs⁴⁸. The incidence of BPD was 17% in the consistently low FiO₂ group, 51% in the pulmonary deterioration group, and 67% in the early and persistent pulmonary dysfunction group. These patterns of oxygen use are apparent soon after birth and must result from complex interactions between antenatal exposures and postnatal management⁵⁰. For example, the chronic, indolent chorioamnionitis associated with very preterm birth can induce early lung maturation and cause endotoxin tolerance with a suppressed inflammatory response to a secondarily inflammatory exposure (Figure 1). However, chorioamnionitis associated with more severe lung inflammation may cause a pneumonia that is seldom distinguished clinically from respiratory distress syndrome (RDS) and/or may interfere with a surfactant treatment response⁵¹. Fetal growth restriction greatly increases the risk of BPD⁵². Subsequent early life experiences such as lung injury with the initiation of ventilation are further variables that contribute to the initial severity of the lung disease⁵⁰. However, some antenatal exposures such as chorioamnionitis could ameliorate inflammation due to a tolerance phenomenon and have beneficial effects on surfactant production and lung maturation. Conceptually therefore, these different insults during fetal or neonatal epochs can interact in a synergistic fashion to increase the inflammation or conversely may dampen the net inflammatory exposure. The interactions between the different insults can therefore modulate the risk for BPD.

Clinical and Epidemiological Studies of Ureaplasma and BPD

Ureaplasma spp. are the organisms most frequently associated with chorioamnionitis and very preterm delivery⁵³. However, the contribution of Ureaplasma or chorioamnionitis to BPD is less clear. Several single center studies reported the association of Ureaplasma in the respiratory secretions of newborns with the development of BPD^{54–58}. Some studies used the BPD definition of oxygen supplementation at 28d, while others used the more current definition of BPD as the need for oxygen supplementation at 36 week post-menstrual age. However, other studies of mechanically ventilated preterm infants did not associate Ureaplasma in the respiratory tract with BPD^{59–62}. Similarly, some studies reported an association of BPD with chorioamnionitis^{63,64}, but more recent reports do not find a clear association^{65,66}. Indeed some studies found that chorioamnionitis correlated with a decrease in the development of BPD⁶⁷.

To better define the role of *Ureaplasma* in the development of BPD, Schelonka et al performed a meta-analysis of published studies using stringent entry criteria⁶⁸. Twenty-three studies with an aggregate of 2216 infants reported BPD using the 28 week definition, and 8 studies with 751 infants reported BPD using the 36 week definition. Overall, the relative risk for BPD in *Ureaplasma* positive infants was 1.6 (C.I. 1.1–2.3) for BPD at 36 weeks, or 2.8 (C.I. 2.3–3.5) for BPD at 28d compared to *Ureaplasma* negative group. However, there was substantial heterogeneity in the studies with the greatest contribution to the BPD association from the studies reporting fewer than 100 infants, suggesting a reporting bias⁶⁸.

Why might there be such heterogeneity in the studies? Different patient populations, divergent ventilator management practices, non-uniform *Ureaplasma* culture techniques and publication bias may contribute to the discrepant results. However, another possible explanation is that *Ureaplasma* may cause both some benefit and some harm, and the differential contribution of harm vs. benefit in different populations may explain the variation. *Ureaplasma* in preterm neonates is acquired antenatally. Experiments in sheep clearly demonstrate that chorioamnionitis caused by *Ureaplasma* results in increased lung compliance^{39–41}. At least one study in preterm infants also reported a decreased incidence of RDS in infants with *Ureaplasma* isolated from tracheal aspirates⁵⁸. Further, mechanical ventilation is a major contributor to BPD and more gentle ventilation (i.e. continuous positive airway pressure or CPAP) tends to decrease the incidence of BPD^{69–73}. Therefore, it is possible that *Ureaplasma* induced increases in surfactant and thus the improved lung function may decrease the need for mechanical ventilation and therefore afford protection against BPD. Alternatively, *Ureaplasma* may be a priming insult and mechanical ventilation following *Ureaplasma* may increase lung injury. Indeed, a recent study of 122 preterm infants < 1000g birth weight from Japan demonstrated no association between *Ureaplasma* cultured in the tracheal aspirate with BPD. However, the infants that received prolonged mechanical ventilation for ≥ 2 weeks and were *Ureaplasma* positive had an increased risk for BPD compared to *Ureaplasma* negative infants on prolonged mechanical ventilation⁷⁴. This synergy between *Ureaplasma* colonization and subsequent injurious exposures in preterm infants may partly explain the variable increased risk for BPD in different patient populations.

Treatment with Antimicrobial Agents

Since *Ureaplasma* is the most common organism isolated in women with chorioamnionitis it is instructive to review antibiotic trials for chorioamnionitis. Antenatal trials of antimicrobial use in the setting of preterm labor have yielded mixed results. Mercer et al.⁷⁵, randomized 614 women with preterm premature rupture of membranes between 24–32 weeks to ampicillin plus erythromycin or placebo for a 7d treatment course⁷⁵. The women randomized to antibiotics had a higher latency and the infants had a lower rate of RDS, necrotizing enterocolitis (NEC) and BPD. In the ORACLE I randomized trial, 4286 women with preterm premature rupture of membranes <37 weeks gestation were randomized to erythromycin only, amoxicillin plus clavulanic acid only, both, or placebo⁷⁶. Any antibiotic use was associated with increased latency but only erythromycin decreased the composite outcome of death or BPD or intraventricular hemorrhage (IVH) or periventricular leukomalacia⁷⁶. The amoxicillin and clavulanic acid combination increased the incidence of NEC. In the ORACLE II trial, the same antibiotic regimens (or placebo) as ORACLE I was given to 6295 women <37 week gestation with intact membranes but spontaneous preterm labor⁷⁷. In this trial, antibiotic use for 10d decreased maternal infections but had no benefit for neonatal mortality or morbidity. Additionally, results of a 7-yr follow-up of the ORACLE II trial found that children whose mothers randomized to either erythromycin or amoxicillin-clavulanic acid had increased rates of cerebral palsy, suggesting the use of antibiotics during pregnancy may have some inherent risks⁷⁸. The current recommendations

by the American College of Obstetrics and Gynecology (ACOG) is to use antibiotics as prophylaxis for group B streptococcal infection and use broad spectrum antibiotics for women with preterm premature rupture of membranes or for women with diagnosed acute perinatal infections⁷⁹. However, ACOG recommends not using antibiotics for women with preterm labor with intact membranes⁷⁹.

The pharmacokinetics of maternal antibiotic treatments are not well understood in the amniotic fluid or the fetus⁸⁰. In particular, the penetration of macrolides to the amniotic compartment after a systemic administration appears to be poor in sheep⁸¹, which might explain the lack of efficacy of antibiotics in women with intact membranes. However, in a Rhesus macaque model of chorioamnionitis induced by intra-amniotic injection of *Ureaplasma*, maternal intravenous administration of azithromycin effectively delayed preterm delivery and reduced fetal lung inflammation⁸². These experiments with disparate results in different species reinforce the need for further work to elucidate pharmacokinetics and efficacy of macrolides to treat the fetal compartment.

A limited number of studies report the use of macrolides for eradication of *Ureaplasma* in neonates at risk for developing BPD. Infants in two trials with limited numbers of infants <30 weeks gestation on mechanical ventilation were treated with erythromycin. *Ureaplasma* status was unknown at the time of initiation of antibiotics in the Lyon⁸³ et al. study. In contrast, Jonsson et al.⁸⁴ recruited only infants with *Ureaplasma* positive tracheal aspirates. Both studies failed to show a benefit of erythromycin for prevention of death or BPD. Another macrolide, azithromycin is actively concentrated in the macrophages and has anti-inflammatory effects. Ballard et al⁸⁵ randomized 220 mechanically ventilated preterm infants 1250g in the first 3d of life. They treated the infants with azithromycin or placebo for up to 6 weeks. Overall, azithromycin did not protect against death or BPD, but in the subgroup of infants who had *Ureaplasma* isolated in tracheal aspirates, azithromycin decreased BPD from 94% in the placebo arm to 73% in the antibiotic arm. Recently, a randomized trial from Turkey evaluated clarithromycin in 74 preterm infants 750–1250g at birth who tested positive for *Ureaplasma* in the nasopharynx in the first 3d of life⁸⁶. Clarithromycin treatment for 10d decreased the incidence of BPD. Although no significant side-effects were reported in these studies, antibiotics must be used with caution in preterm neonates since prolonged antibiotic exposure has been associated with increased rates of NEC or late onset sepsis^{87,88}.

Summary and Conclusions

Ureaplasma species are the organisms most associated with chorioamnionitis and preterm delivery. Although there is controversy regarding the role of *Ureaplasma* in BPD, experimental evidence and clinical/epidemiological data demonstrate that *Ureaplasma species* can increase the risk for BPD in preterm infants. Antenatal exposure to *Ureaplasma* increases surfactant production, increases lung volumes and induces pulmonary inflammation with disordered alveolar development in preterm sheep. Experimental evidence demonstrates that *Ureaplasma* can suppress signaling by LPS and thus modulate the innate immune system. In preterm infants, *Ureaplasma* colonization can prime the respiratory tract and synergistically increase ventilator induced lung injury. Treatment with macrolide antibiotics for *Ureaplasma* in preterm infants has not met with much success.

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Figure 1. Immune modulation: Chronic exposure to Ureaplasma (UP) downregulated LPS responses in preterm fetal Sheep

IL-1 β mRNA in fetal lung was measured by rt-PCR after different intraamniotic injections at different intervals prior to preterm delivery at 125d (80% gestation). The mean mRNA level in the control group was set to 1. mRNA levels of each group were expressed as fold increase relative to controls. Exposure to UP alone did not induce IL-1 β expression but prevented LPS induced IL-1 β expression (* $p < 0.05$ vs control). Figure based on data from³⁷.

Figure 2. Modest time-dependent fetal lung inflammation induced by UP in preterm fetal Sheep
A: Intraamniotic injection of *Ureaplasma parvum* serovar 3 into the amniotic cavity induced an increase in IL-8 mRNA in the fetal lung. **B:** Neutrophils in broncho-alveolar lavage fluid. Results are given per kilogram body weight. The *Ureaplasma parvum* serovar 3 injection induced a pulmonary inflammation at 3 days, 7 days, 14 days or 70 days before preterm delivery. *, $p < 0.05$ versus control. Figure based on data from³⁸⁴¹.

Figure 3. Increases in lung gas volume induced by UP in preterm fetal Sheep

The lung gas volume at 40 cm H₂O increased after intraamniotic injection of *Ureaplasma parvum* serovar 3 at 3 days, 7 days, 14 days or 70 days before preterm delivery. *, $p < 0.05$ versus control. . Figure based on data from³⁸.

Figure 4. Lung remodeling induced by UP in preterm fetal Sheep

Elastin foci in the lung after intraamniotic injection of *Ureaplasma parvum* serovar 3 after 3, 7 and 14 days. The number of elastin foci in the control group was 12 and set to 1 for ease of comparison. Changes are shown as fold increase or decrease over control. *, $p < 0.05$ versus control. Figure based on data from³⁸.