Review

Immune cells in term and preterm labor

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Labor resembles an inflammatory response that includes secretion of cytokines/chemokines by resident and infiltrating immune cells into reproductive tissues and the maternal/fetal interface. Untimely activation of these inflammatory pathways leads to preterm labor, which can result in preterm birth. Preterm birth is a major determinant of neonatal mortality and morbidity; therefore, the elucidation of the process of labor at a cellular and molecular level is essential for understanding the pathophysiology of preterm labor. Here, we summarize the role of innate and adaptive immune cells in the physiological or pathological activation of labor. We review published literature regarding the role of innate and adaptive immune cells in the cervix, myometrium, fetal membranes, decidua and the fetus in late pregnancy and labor at term and preterm. Accumulating evidence suggests that innate immune cells (neutrophils, macrophages and mast cells) mediate the process of labor by releasing pro-inflammatory factors such as cytokines, chemokines and matrix metalloproteinases. Adaptive immune cells (T-cell subsets and B cells) participate in the maintenance of fetomaternal tolerance during pregnancy, and an alteration in their function or abundance may lead to labor at term or preterm. Also, immune cells that bridge the innate and adaptive immune systems (natural killer T (NKT) cells and dendritic cells (DCs)) seem to participate in the pathophysiology of preterm labor. In conclusion, a balance between innate and adaptive immune cells is required in order to sustain pregnancy; an alteration of this balance will lead to labor at term or preterm.

Keywords: B cells; cytotoxic T cells; dendritic cells; macrophages; mast cells; neutrophils; NKT cells; parturition; preterm delivery; regulatory T cells; T cells

Introduction

Pregnancy demonstrates the capabilities of the human immune system. The fetus, a semi-allogeneic graft, grows and develops within the mother without succumbing to immunological rejection, a process which depends on the proper establishment of fetomaternal tolerance. This tolerance is initiated by the presentation of the paternal–fetal antigen from semen and is facilitated by seminal plasma factors. Antigen is processed by dendritic cells (DCs) and then presented to T cells in the uterine draining lymph nodes. As a result, antigen-specific regulatory T cells (Tregs) proliferate in order to create peripheral tolerance towards fetal antigens and allow conceptus implantation.

Treg numbers are maintained through pregnancy, creating a tolerogenic anti-inflammatory state or hyporesponsiveness towards paternal antigens until late gestation. During late pregnancy, we have proposed that circulating maternal leukocytes (innate and adaptive) are recruited into reproductive tissues (cervix and myometrium) and to the maternal/fetal interface (decidual tissues) by chemotactic processes, where a pro-inflammatory state develops and leads to labor and delivery of the baby. It is thought that the premature activation of this pro-inflammatory pathway can lead to a breakdown of fetomaternal tolerance and play a role in the induction of labor, which subsequently can result in preterm birth.

Preterm birth is a major determinant of neonatal mortality and morbidity. In 2011, 11.7% of all births in the United States were diagnosed as preterm. Among problems occurring after preterm birth are chronic respiratory illnesses, neurodevelopmental disorders and long-term cognitive impairment. However, the mechanisms that lead to preterm birth/labor are poorly understood. The main goal of this review is to summarize the innate and adaptive immune cell components that participate in term and in preterm labor, clarifying the contributions of resident, adaptive leukocytes to the physiological or pathological...
activation of parturition. Achieving a deeper understanding of the innate and adaptive immune cell components involved in preterm labor might allow us to develop strategies to prolong pregnancy and thereby improve pregnancy outcomes.

**INNATE IMMUNE CELLS IN TERM AND PRETERM LABOR**

Labor is an inflammatory process. A number of studies in humans and mice have reported the presence of inflammatory neutrophils and macrophages in the uterus, decidua, cervix and fetal membranes during labor. The spreading and homing of these granulocytes is facilitated by chemokines and cellular adhesion molecules. Additionally, mast cells are present in the uterus and cervix during late gestation and may contribute to the process of labor. Uterine contractions, cervical ripening and dilation, and rupture of the fetal membranes (ROM) are processes which must occur in a timely fashion for a successful delivery. Innate immune cells have been linked to these processes by various studies, and this section aims to discuss the possible roles for these cells in the processes of term and preterm labor.

**Neutrophils**

Neutrophil numbers are higher in the circulation of women who undergo labor than in those who do not undergo labor. Labor-related granulocytes are activated since they exhibit an increased ability to migrate. The presence of neutrophils in reproductive tissues at term and their ability to migrate to this region during labor have been well documented in both humans and rodents. Neutrophils participate in the process of labor by releasing pro-inflammatory cytokines and secreting matrix metalloproteinases (MMPs); however, their role in each anatomical compartment seems to be unique and will be discussed further below.

In the myometrium, mRNA levels of CXCL8, a neutrophil chemoattractant, are higher in women at term during labor than in women without labor, suggesting that neutrophils are more abundant in the myometrium during labor. However, it was recently demonstrated in a murine model of infection-induced preterm birth (intratrue administration of lipopolysaccharide (LPS)) that there is no increase in the neutrophil numbers (Gr-1+ cells) in the myometrium 6 h after LPS administration. Preliminary studies in our laboratory put forward evidence on the role of myometrial neutrophils during LPS-induced preterm birth. We found that LPS administration in the peritoneum causes high rates of preterm birth and this is associated with increases in proportion and in absolute numbers of neutrophils in the myometrium (NGL, unpublished data). This discrepancy between studies may be due to the fact that we collected tissues prior to delivery (24 h after LPS injection) instead of 6 h after the intratrue administration of LPS. Nonetheless, the potential role of myometrial neutrophils in the process of labor needs continuing exploration.

Neutrophils are proposed to play a central role in the cervical ripening process, although recent studies have indicated that this is likely not the case. In mice, neutrophil numbers in the cervix do not vary from 15 days postcoitum (dpc) to the time of cervical ripening (late 18 dpc). It has been consistently reported that there are no differences in the number of cervical neutrophils between women without labor with cervical ripening and women who had not undergone the ripening process. However, the number of neutrophils is higher in the cervixes of women who have just completed a vaginal delivery following spontaneous labor at term than in women who were in the first trimester of pregnancy. This finding supports the new hypothesis that neutrophil function is required shortly after parturition, in the phase of postpartum tissue repair.

There have been several studies in mice implicating decidual neutrophils in the process of infection-induced preterm birth. A large influx of neutrophils into the decidua and myometrium is observed during LPS-induced preterm labor and during term labor; however, this increment was not seen in a non-infectious model of preterm birth (caused by mifepristone). Another study reported a sevenfold increase in neutrophils in the decidua after 6 h of intrauterine LPS administration. Despite these findings, the role of neutrophils as a causative agent of preterm labor is questioned since the depletion of these cells does not alter the timing or success of labor and does not prevent LPS-induced preterm birth. Neutrophil depletion prior to LPS administration did, however, reduce the amount of a key pro-inflammatory cytokine, IL-1β, in the uteroplacental tissues. This finding is relevant since systemic administration of IL-1β leads to preterm birth in mice. These results suggest that neutrophils are not a necessary component in infection-induced preterm birth, yet they may be required in inflammation-induced preterm birth.

In human decidual tissues, the number of neutrophils was higher in women with preterm labor associated with chorioamnionitis than in women with term gestations (with and without labor) and in women with spontaneous preterm labor/birth without chorioamnionitis. The maternal origin of these decidual leukocytes (e.g., neutrophils) in preterm labor/birth associated with acute chorioamnionitis was proven by FISH. Maternal cells could be recruited into this maternal/fetal interface by decidual-derived chemokines, such as CXCL8. Human decidual neutrophils release several inflammatory mediators and MMPs, which degrade the extracellular matrix of the fetal membranes during both term and preterm labor. Taken together, these data suggest that decidual neutrophils contribute to the physiological ROM and pathological preterm premature rupture of membranes (PPROM) during term and preterm labor.

**Macrophages**

Macrophages are among the primary innate immune cells that contribute to the processes of term and preterm labor, and their roles have been studied in humans, mice and rats. Macrophages are significant during late gestation primarily due to their secretory products, which include MMPs, IL-1β, IL-6, TNF-α and nitric oxide (NO). These versatile leukocytes are being extensively studied to deepen our understanding of the
parturition process. We discuss below the possible effector actions of macrophages in term and preterm labor.

Macrophages play a relevant role in the uterus during parturition. In mice, the number of uterine macrophages at 15 dpc (4 days prior to birth) was significantly higher than in non-pregnant controls though these numbers dropped to non-pregnant levels one day prior to birth. This trend for macrophages to decrease immediately prior to labor correlates with another study, performed in rats, which found that NO synthesis in the uterus was elevated during pregnancy but reduced during term labor, NO, which can be produced by macrophages, has been demonstrated to inhibit myometrial contractions. Altogether, these results suggest that a decrease in macrophages, and the resultant reduction in NO, is required for the onset of labor.

Although the aforementioned studies indicate that macrophage numbers decrease in the uterus prior to labor, a study in rats found concentrations of CCL2, a monocyte/macrophage chemoattractant, increased in the myometrium near the time of delivery in comparison to earlier points of gestation and during RU486-induced preterm labor. Additionally, macrophages may exert effects on the uterus during parturition through the release of pro-inflammatory cytokines, such as TNF-α, which are able to upregulate uterine activation proteins, allowing the uterus to prepare for labor. These findings suggest that macrophages are instead recruited into the uterus during labor.

Ripening and dilation of the cervix are the next steps in parturition after the initiation of uterine contractions, an inflammatory response has been associated with these processes. During pregnancy at term, but before the onset of labor, women with a ripened cervix were found to have greater numbers of cervical macrophages in comparison to women who were not undergoing cervical ripening. A murine model similarly found an increased proportion of macrophages in the cervix the day before birth (18 dpc) in comparison to mid/late gestation (15 dpc). A large number of cervical macrophages was also found in antepartum and in LPS-induced preterm labor. These data suggest the possible involvement of macrophages in cervical remodeling.

Although their presence suggests that macrophages play a role in the cervix during the process of labor, the characterization of these cells also supports this theory. Murine cervical macrophages expressing markers associated with adhesion (CD11b<sup>high</sup>) and migration (CD54) were lower prior to birth (18 dpc) than in mid/late gestation (15 dpc). However, macrophages that express markers associated with MMP activation (CD147) and cell matrix remodeling (CD169) are significantly higher at 18 dpc than at 15 dpc. These results suggest that cervical macrophages are probably not migrating or binding to vessels prior to birth, but are instead remodeling and degrading the extracellular matrix, which are important processes in ripening of the human cervix. The fact that cervical leukocytes (e.g., macrophages) secrete MMP-9 at term pregnancy and that macrophage depletion prevents LPS-induced preterm birth in mice, suggests that macrophages are a main source of MMP-9 and contribute to the process of labor at both term and preterm stages. Moreover, macrophage-derived cytokines IL-1β and TNF-α increase the levels of MMP-1, MMP-3 and MMP-9, which may be another pathway whereby macrophages participate in the cervical ripening process. Despite the evidence above, it is important to point out that several studies have contrarily suggested that macrophages are not necessary for cervical ripening in mice, but play a role in postpartum repair. Further research on the human cervix during labor and preterm labor must be performed in order to come to a definitive conclusion.

Macrophages could also play a role in the rupture of the fetal membranes since macrophages are recruited by these tissues and produce MMP-9. MMP-9 concentrations are significantly increased in the fetal membranes during labor, preterm labor and PPROM, which directly links this enzyme to physiological ROM and pathological PPROM. Additionally, pro-inflammatory cytokines released by macrophages during labor can regulate the further release of MMPs by the fetal membranes, suggesting another mechanism whereby macrophages may contribute to ROM and PPROM.

Macrophages also reside in the decidua near or during the time of labor. In human decidual tissues, macrophage proportions are higher at term than in preterm gestations without labor. Macrophage tissue density is even greater in decidua from women who delivered term and preterm with labor in comparison to women who delivered at term without labor. In mice, the proportion of decidual macrophages increases prior to birth (18 dpc) in comparison to mid/late gestation (15 dpc). Altogether, these results suggest that decidual macrophages have a role prior to the onset of labor.

Macrophages are also implicated in the etiology of preterm labor since CCL2 concentrations are increased in the amniotic fluid of women delivering preterm, both in the presence and absence of intra-amniotic infection, in comparison to women delivering at term. One of the most significant indicators of the role of macrophages in preterm labor was the demonstration that the depletion of macrophages in pregnant mice protected these animals from LPS-induced preterm birth. Ultimately, macrophages are potentially involved in several processes during parturition. The precise role of this cell type in labor remains disputed, yet much evidence gives credibility to their putative roles. Further studies are required to fully elucidate the roles of macrophages in the physiological process of labor and the pathological induction of preterm labor.

Currently, we are investigating the role of macrophages during preterm birth using animal models. Our preliminary data suggest that the plasticity of these cells at the maternal/fetal interface is unique, and that besides participating in the process of labor, macrophages play a central role in the maintenance of fetomaternal tolerance during late pregnancy.

**Mast cells**

Mast cells (MCs) are also important innate immune effector cells during late gestation and labor due to their secretion of...
mediators. Fast-acting MC mediators are histamine, serotonin, heparin, proteoglycans, proteases, prostaglandins and leukotrienes. MCs also secrete the long-term modulators IL-1β, IL-3, IL-5, IL-6 and TNF-α. Moreover, human MCs induce the expression of endothelial adhesion molecules, and express several chemokine receptors. This combination of MC recruitment and up-regulation of cellular adhesion molecules allows MCs to localize within the uterus and cervix, where they may play a role in the development of a pro-inflammatory environment. Due to their presence and actions in cervical tissue during late gestation, MCs and histamine have been associated with the stimulation of cervical contractility; however, MCs have been detected in higher proportions in postpartum than during late gestational cervix which indicates a greater role for this cell type in postpartum uterine cervical repair than during labor. We therefore focus the section below on discussing the role of MCs and their mediators in the uterus during term and preterm labor.

MC degranulation releases mediators which likely play major roles in the process of labor by remodeling uterine smooth muscle cells and stimulating uterine contractions. The release of histamine and serotonin has been linked to uterine contractility since MCs reside adjacent to smooth muscle in the myometrium. Indeed, during murine pregnancy MCs are more abundant in the myometrium than in the endometrium. The degranulation of MCs in vitro, utilizing compound 48/80, induces greater uterine contractility in tissue from late gestation than in non-pregnant uterine tissue. In addition, the tissue density of human uterine MCs is greater during pregnancy than in the non-pregnant state, which also suggests that uterine MCs modulate myometrial contractility during late pregnancy. A link between MCs and allergy has been suggested as a mechanism of preterm labor/birth, since MCs are one of the cells effecting immediate hypersensitivity reactions and allergic disease and allergies play a central role in uterine contractions. Furthermore, pre-treatment of guinea pigs with a histamine H1 receptor antagonist decreased the rate of preterm birth induced by an allergic reaction. This finding suggests a vital role for histamine, and therefore MCs, in the processes of term and preterm labor.

A recent study contradicts the notion of MC involvement in labor. This study found that, in human myometrial tissues, the abundance of MCs was similar at mid-pregnancy and during labor. In MC deficient KitW-sh mice, labor still occurs and leukocyte recruitment into the myometrium is not different from wild-type controls. A possible explanation is that MCs are not the sole leukocyte recruiters, and the pro-inflammatory cascade can be upregulated by other leukocyte subpopulations even in the absence of MCs. Further research is needed in order to clarify the role of mast cells during term and preterm labor.

**ADAPTIVE IMMUNE CELLS IN TERM AND PRETERM LABOR**

The adaptive immune system creates memory and responds to specific antigens. During pregnancy, the adaptive immune limbs of both the mother and the fetus must tolerate each other in order to maintain pregnancy until term. A breakdown of this fetomaternal tolerance may lead to labor. In term pregnancy, lack of the tolerogenic state results in physiologic labor. However, a premature retreat of this tolerogenic state might lead to preterm labor.

**T cells**

During pregnancy, maternal T cells recognize fetal antigens through interactions with antigen-presenting cells. Fetal antigen-specific T cells maintain fetomaternal immune tolerance across pregnancy. We previously proposed that maternal circulating T cells infiltrate into the maternal/fetal interface prior to delivery and during labor at term. Decidual T cells are activated and have both a regulatory and an effector phenotype. The next section further addresses the putative roles of specific T-cell subsets during late pregnancy and in term and preterm labor.

**Effect T cells.** We recently provided evidence that decidual CD4+ T cells are involved in term parturition. Specifically, we demonstrated that decidual CD4+ T cells are more abundant in term than in preterm gestations without labor. These T cells express CD45RO, but not CD45RA, which suggests that they are memory cells that were generated early in pregnancy when fetal–antigen presentation occurs. We also demonstrated that decidual CD4+ T cells express IL-1β, TNF-α and MMP-9 during spontaneous labor at term. The fact that decidual T cells express activation markers such as CD25 and labor mediators implicated in both term and preterm labor suggests that the adaptive limb of the immune system participates during labor.

Additionally, we demonstrated that during term labor T cells are preferentially recruited into the rupture zone of the fetal membranes by chemotactic processes facilitated by CXCL10 and CCL5. However, T-cell attraction to the rupture zone was significantly diminished in premature ROM cases. These data suggest that T-cell recruitment into the maternal/fetal interface is required for term pregnancy, and the dysregulation of this recruitment may lead to pathological rupture of membranes.

Th17 cells (CD3+CD4+IL-17A+) also congregate in human decidua, and their tissue density is higher in cases of chorioamnionitis than in cases without chorioamnionitis. This finding further supports the idea that pro-inflammatory adaptive immune cells at the maternal/fetal interface are associated with chorioamnionitis, which can lead to preterm labor/birth. Studies in our laboratory are currently exploring the potential role for this T-cell subset in preterm labor using LPS-induced and RU486-induced preterm birth models.

Fetal T cells might also play a role during preterm labor. Memory fetal T cells (CD45RO RA) are present in higher proportions in cord blood from cases of preterm labor compared to term labor. Fetal T cells are also activated (CD25+CD69+) during preterm labor. Indeed, acute chorioamnionitis, a leading cause of preterm deliveries, is associated with an increase in cord blood T-cell chemokines

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**Cellular & Molecular Immunology**
(CXCL9, -10 and -11). These results suggest that fetal T cells can contribute to the pathophysiology of preterm labor.

Cytotoxic T cells (CTLs) are present at the maternal/fetal interface in term gestations in the absence of labor, where they express perforin and granzyme B. In placenta, CTLs are abundant in cases with villitis of unknown etiology and express T-cell chemokine receptors (CXCR3 and CCR5). In peripheral circulation, CD300a+ CTLs have an effector memory phenotype, and their proportion is higher in women with chronic chorioamnionitis than in women without this lesion. Taken together, these data suggest that CTLs may participate in pathological inflammation associated with preterm birth, but their role during spontaneous labor at term and preterm requires further exploration.

Tregs

There are two main Treg subsets: thymic Tregs (tTregs) and extrathymic or peripheral Tregs (pTregs). During pregnancy, CD4+ pTregs have been categorized into four subsets: DR<sup>high</sup>+CD45RA<sup>-</sup>, DR<sup>low</sup>+CD45RA<sup>-</sup>, DR<sup>-</sup>CD45RA<sup>+</sup> and naive DR<sup>-</sup>CD45RA<sup>-</sup> Tregs within their total pTreg pool. Indeed, the suppressive activity of pTregs is strongly reduced in term and preterm labor, which is correlated with a reduction in the expression of HLA-DR in preterm cases. This suggests that the lack of suppressive function during late pregnancy could trigger the onset of parturition at term and preterm gestations.

At term pregnancy, Tregs are found at the maternal/fetal interface, however, the targeted depletion of CD25<sup>+</sup> Tregs within their total pTreg pool. Preliminary data from our laboratory demonstrates that systemic ablation of Tregs by targeting FoxP3<sup>+</sup> cells leads to pregnancy failure during early gestation. A separate study suggested that regulatory T cells are not required in late pregnancy; however, the targeted depletion of CD25<sup>+</sup> cells is not specific for Tregs. Preliminary data from our laboratory demonstrates that systemic depletion of FoXP3<sup>+</sup> cells during late gestation does lead to pregnancy complications (NGL, unpublished data).

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Additional unpublished data from our laboratory demonstrate that LPS-induced preterm labor causes an expansion of CD4<sup>+</sup> Tregs in the spleen and thymus but a reduction of uterine CD4<sup>+</sup> Tregs (NGL, unpublished data). We also found that the administration of vaginal progesterone, a clinical strategy to prevent preterm birth in women with a short cervix, increases the proportion of decidual CD4<sup>+</sup> Tregs (NGL, unpublished data). Altogether, these data suggest that preterm birth is characterized by altered proportions of CD4<sup>+</sup> Tregs at the maternal/fetal interface and that natural progesterone can restore the number of these cells during late pregnancy, preventing preterm birth.

B cells

A few years ago, we suggested a role for B cells during term labor since the fetal membranes from laboring women who delivered at term exhibit B-cell attraction in vitro. Current preliminary data demonstrate that B cells are indeed present in the decidua and cord blood at term and preterm stages (NGL, unpublished data). However, the role of B cells in the processes of term and preterm labor is still under investigation.

Several studies have linked various B cell subsets to pregnancy. B1 cells are present in lower proportions in maternal blood during pregnancy and return to non-pregnant proportions post-partum. However, B2 cell frequencies are unchanged by pregnancy in the peripheral blood of women. Regulatory B cells exist during early and late gestation and release IL-10. Regulatory B cells are potential immune players in the development of immunological tolerance, and their presence during mid-gestation may be important in sustaining pregnancy until labor. B10 cells suppress TNF-α secretion by CD4<sup>+</sup> T cells during pregnancy, and this may regulate the inflammatory state prior to labor. Furthermore, B cells isolated from term placenta produce increased amounts of asymmetric IgG upon stimulation by IL-6, IL-10 and IL-4. Therefore, an abnormal disruption of B cell-derived cytokine and asymmetric antibody production could play a part in disturbing fetal tolerance and possibly in eliciting preterm labor.

BRIDGES BETWEEN THE INNATE AND ADAPTIVE IMMUNE SYSTEMS IN TERM AND PRETERM LABOR

Immune tolerance involves both the innate and adaptive immune systems. Therefore, fetomaternal tolerance must involve the participation of immune cells that bridge the innate and adaptive immune systems, such as DCs and natural killer T (NKT) cells. The roles of these cells during late gestation, labor and preterm labor are discussed below.

**NKT cells in term and preterm labor**

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**Currently, we are investigating the function and phenotypic characteristics of these cells during term and preterm labor.**

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secretes large quantities of IL-4 (Th2) and IFN-γ (Th1) upon TCR activation, and their activation has been shown to have roles in activating NK cells, B cells and T cells. As a result of their immunological effects and their presence during gestation, iNKT cells may participate in pathological or physiological responses during late gestation.

iNKT cells have been involved in the induction of an increased cytotoxic state during human pregnancy complications such as preeclampsia. The proportion of iNKT cells expressing activation markers (CD69), perforin and IFN-γ is increased in the blood of pre-eclamptic women in comparison to pregnant women without this pathology. Although the aforementioned study did not address preterm labor, these results indicate that pro-inflammatory iNKT cells are increased during late gestational pregnancy complications; this Th1-like environment could potentially disrupt fetomaternal tolerance and lead to preterm labor.

The role of iNKT cells in the induction of LPS-induced pre-term birth has been studied in iNKT cell deficient (Jα18−/−) mice. The injection of LPS at 15 dpc caused preterm birth in wild-type mice but not in iNKT cell-deficient mice, suggesting that iNKT cells modulate the process of labor induced by microbial products. Conversely, the stimulation of iNKT cells in vivo by injection of α-galactosylceramide during late gestation (16 dpc) induced early preterm birth (17 dpc), which may be due to an expansion of NK1.1+CD3εδ+ NKT cells in the uterus.

In contrast, studies conducted in our laboratory found that the activation of iNKT cells during late gestation (16 dpc) through α-galactosylceramide administration induced late preterm birth (birth at 18 dpc, 24 h post-injection), which is relevant since 70% of all preterm births in women fall under this category (NGL, unpublished data). Current experiments in our laboratory are addressing the immune mechanisms whereby iNKT cell activation leads to late preterm birth.

DCs in term and preterm labor

DCs are specialized in antigen recognition and presentation. DCs exhibit properties that include induction of antigen-specific T-cell activation, T-cell suppression, Treg generation and peripheral tolerance. Lymphoid CD8α+ DCs (DCs1) induce a Th1 response whereas myeloid CD8α- DCs (DCs2) elicit a Th2 response. A third type of DCs is the inflammatory DCs which initiate a Th1 response as well. Due to their immunomodulatory properties, these three subsets of DCs are relevant in the study of fetomaternal tolerance and inflammation during labor and preterm labor.

DCs contribute to fetomaternal tolerance during early pregnancy. In mice, uterine DCs have a DC2 phenotype at 15 dpc, which suggests that these cells contribute to the tolerogenic state by inducing a local anti-inflammatory (Th2) response during late gestation. Later in pregnancy (17.5 dpc), the predominant DC subset in the uterus is CD11c+CD8α- MHCIγ (immature phenotype). The fact that immature DCs express the anti-inflammatory cytokine IL-10, a potential early biomarker of preterm birth, suggests that these cells may participate in the etiology of preterm labor. Moreover, in T and B cell-deficient mice (Rag1−/−) injected with LPS to induce preterm birth, uterine DC activation was observed, suggesting the participation of DCs in the induction of labor. Further research is needed in order to establish a role for DCs during late gestation, labor and preterm labor.

CONCLUSIONS

During late pregnancy, paternal–fetal antigen-specific memory T cells (including Tregs) participate in the maintenance of fetomaternal peripheral tolerance. Collectively, these cells create an anti-inflammatory environment which will sustain pregnancy. We suggest the following pathway could lead to labor: (1) activation of innate and adaptive immune cells increases their migratory ability; (2) reproductive tissues and the maternal/fetal interface actively recruit the activated cells through the release of chemokines such as CXCL10, CXCL8, CCL2 and CCL5; and (3) infiltrating leukocytes amplify the pro-inflammatory microenvironment at the maternal/fetal interface leading to labor.

A triggered stimulus (e.g., infection/inflammation, sterile inflammation, stress, etc.) can cause the premature activation of this pathway, eliciting a shift from an anti-inflammatory to a pro-inflammatory microenvironment and consequently preterm labor (Figure 1).

An overview of the innate and adaptive immune cells in reproductive tissues and at the maternal/fetal interface during term and preterm labor is shown in Figure 2. Neutrophils are present in the cervix, myometrium, fetal membranes and decidua at term pregnancy; however, their density increases in the myometrium and decidua in term labor and infection-associated preterm labor. Neutrophils are present in the cervix and participate in the repair process during the postpartum period. Macrophages are present in the cervix, myometrium, fetal membranes and decidua at term pregnancy and their density increases in all these tissues, except the cervix, during term and preterm labor. Cervical macrophages also seem to participate in postpartum repair processes. Mast cells are found in cervical and myometrial tissues during late gestation; however, their roles during term and preterm labor are unclear. Effector CD4+ T cells are present in decidual tissues during term labor, and decidual Th17 cells also seem to be involved in the pathology of preterm labor. CTLs are found in term pregnancy and in placental tissues in cases with villitis of unknown etiology; however, their role during labor is unknown. The fetal membranes exhibit B-cell recruitment during term labor, and B cells are found in decidual tissues and cord blood; however, their role in preterm labor is still under investigation. Finally, in myometrial tissues, NKT cell and DC activation seem to be involved in the pathophysiology of preterm labor.

Overall, collaboration between the innate and adaptive limbs of the immune system is required to sustain pregnancy until term. A disruption of either limb at term may lead to physiological labor, and an untimely disruption could result in pathological preterm labor. Research targeting the immune cells involved in the process of labor might reveal new strategies to prevent preterm labor and consequently preterm birth.
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1 Finn R, St Hill CA, Davis JC, Hipkin LJ, Harvey M. Feto-maternal bidirectional mixed lymphocyte reaction and survival of fetal allograft. Lancet 1977; 2: 1200–1202.
2 Robertson SA, Skinner RJ, Care AS. Essential role for IL-10 in resistance to lipopolysaccharide-induced preterm labor in mice. J Immunol 2006; 177: 4888–4896.
3 Robertson SA, Sharkey DJ. The role of semen in induction of maternal immune tolerance to pregnancy. Semin Immunol 2001; 13: 243–254.
4 Robertson SA, Mau VJ, Tremellen KP, Seamark RF. Role of high molecular weight seminal vesicle proteins in eliciting the uterine inflammatory response to semen in mice. J Reprod Fertil 1996; 107: 265–277.
5 Beer AE, Scott JR, Billingham RE. Histoincompatibility and maternal immunological status as determinants of feto-placental weight and litter size in rodents. J Exp Med 1975; 142: 180–196.
6 Aluvihare VR, Kallikourdis M, Betz AG. Regulatory T cells mediate maternal tolerance to the fetus. Nat Immunol 2004; 5: 266–271.
7 Rowe JH, Erteigt JM, Xin L, Way SS. Pregnancy imprints regulatory memory that sustains energy to fetal antigen. Nature 2012; 490: 102–106.
8 Smith RN, Powell AE. The adoptive transfer of pregnancy-induced unresponsiveness to male skin grafts with thymus-dependent cells. J Exp Med 1977; 146: 899–904.
9 Chauvat G, Voisin GA, Escalier D, Robert P. Facilitation reaction (enhancing antibodies and suppressor cells) and rejection reaction (sensitized cells) from the mother to the paternal antigens of the conceptus. Clin Exp Immunol 1979; 35: 13–24.
10 Chauvat G, Petitbarat M, Dubanchet S, Rahmati M, Ledee N. Tolerance to the foetal allograft? Am J Reprod Immunol 2010; 63: 624–636.
11 Gomez-Lopez N, Estrada-Gutierrez G, Jimenez-Zamudio L, Vega-Sanchez R, Vadillo-Ortega F. Fetal membranes exhibit selective leukocyte chemotactic activity during human labor. J Reprod Immunol 2009; 80: 122–131.
12 Gomez-Lopez N, Vadillo-Perez L, Nessim S, Olson DM, Vadillo-Ortega F. Choriodecidua and amnion exhibit selective leukocyte chemotaxis during term human labor. Am J Obstet Gynecol 2011; 204: 5.
13 Gomez-Lopez N, Hernandez-Santiago S, Lobb AP, Olson DM, Vadillo-Ortega F. Normal and premature rupture of fetal membranes at term delivery differ in regional chemotactic activity and related chemokine/cytokine production. Reprod Sci 2013; 20: 276–284.
14 Gomez-Lopez N, Vega-Sanchez R, Castillo-Castrejon M, Romero R, Cubiero-Arreola K, Vadillo-Ortega F. Evidence for a role for the adaptive immune response in human term parturition. Am J Reprod Immunol 2013; 69: 212–230.
15 Gomez-Lopez N, Tanaka S, Zaem Z, Metz GA, Olson DM. Maternal circulating leukocytes display early chemotactic responsiveness during late gestation. BMC Pregnancy Childbirth 2013; 13(Suppl 1): S8.
16 Gomez-Lopez N, Guilbert LJ, Olson DM. Invasion of the leukocytes into the fetal–maternal interface during pregnancy. J Leukoc Biol 2010; 88: 625–633.
17 Romero R, Brody DT, Oyarzun E, Mazor M, Wu YK, Hobbins JC et al. Infection and labor. III. Interleukin-1: a signal for the onset of parturition. Am J Obstet Gynecol 1989; 160: 1117–1123.
18 Romero R, Espinoza J, Goncalves LF, Kusanovic JP, Friel LA, Nien JK. Inflammation in preterm and term labor and delivery. *Semin Fetal Neonatal Med* 2006; 11: 317–326.

19 Romero R, Espinoza J, Kusanovic JP, Gotsch F, Hassan S, Erez O *et al.*. The preterm parturition syndrome. *BJOG* 2006; 113(Suppl 3): 17–42.

20 Trombol SM, Klimaviciute A, Byström B, Chromek M, Brauner A, Ekman-Ordeberg G. Non-infected preterm parturition is related to increased concentrations of IL-6, IL-8 and MCP-1 in human cervix. *Reprod Biol Endocrinol* 2005; 3: 39.

21 Gonzalez JM, Franzke CW, Yang F, Romero R, Girardi G. Complement activation triggers metalloproteinases release inducing cervical remodeling and preterm birth in mice. *Am J Pathol* 2011; 179: 838–849.

22 Lee J, Romero R, Xu Y, Kim JS, Topping V, Yoo W *et al.*. Leukocytes infiltrate the myometrium during human labor at term. *Hum Reprod* 2003; 18: 41–45.

23 Martin JA, Hamilton BE, Ventura SJ, Osterman MJ, Kirmeyer S, Goldenberg RL, Hauth JC, Andrews WW. Intrauterine infection and preterm delivery. *PLoS ONE* 2011; e16806.

24 McCann MC. The contribution of low birth weight to infant mortality and childhood morbidity. *N Engl J Med* 1985; 312: 82–90.

25 Mackler AM, Iezza G, Akin MR, McMillan P, Yellon SM. Macrophage infiltration of the human and rat decidua during pregnancy. *Hum Reprod* 1999; 14: 229–236.

26 McCann MC. The contribution of low birth weight to infant mortality and childhood morbidity. *N Engl J Med* 1985; 312: 82–90.

27 Thomson AJ, Telfer J, Young A, Campbell S, Stewart CJ, Cameron IT *et al.*. Leukocytes infiltrate the myometrium during human parturition: further evidence that labour is an inflammatory process. *Hum Reprod* 1999; 14: 229–236.

28 Biollapragada S, Youssef R, Jordan F, Greer I, Norman J, Nelson S. Term labor is associated with a core inflammatory response in human fetal membranes, myometrium, and cervix. *Am J Obstet Gynecol* 2009; 200: 104.e101–104.111.

29 Osman I, Young A, Ledingham MA, Thomson AJ, Jordan F, Greer I *et al.*. Leukocyte density and pro-inflammatory cytokine expression in human fetal membranes, decidua, cervix and myometrium before and during labor at term. *Hum Reprod* 2003; 9: 41–45.

30 Bokstom H, Brannstrom M, Alexandersson M, Norstrom A. Leukocyte subpopulations in the human uterine cervical stroma at early and term pregnancy. *Hum Reprod* 1997; 12: 586–590.

31 Payne KJ, Clyde LA, Weldon AJ, Milford TA, Yellon SM. Residency and activation of myeloid cells during remodeling of the prepartum murine cervix. *Biol Reprod* 2012; 87: 106.

32 Young A, Thomson AJ, Ledingham M, Jordan F, Greer I, Norman JE. Immunolocalization of proinflammatory cytokines in myometrium, cervix, and fetal membranes during human parturition at term. *Biol Reprod* 2002; 66: 445–449.

33 Mackler AM, Iezza G, Akin MR, McMillan P, Yellon SM. Macrophage trafficking in the uterus and cervix proceeds parturition in the mouse. *Biol Reprod* 1999; 61: 879–883.

34 Hamilton S, Oomomian Y, Stephen G, Shynlova O, Tower CL, Garrod A *et al.*. Macrophages infiltrate the human and rat decidua during term and preterm labor: evidence that decidual inflammation precedes labor. *Biol Reprod* 2012; 86: 39.

35 Rudolph MI, Bardis L, Cruz MA, Reinicke K. Mast cell mediators evoke contraction and potentiate each other in mouse uterine horns. *Gen Pharmacol* 1992; 23: 833–836.

36 Garfield RE, Bytautiene E, Vedenikov YP, Marshall JS, Romero R. Modulation of rat uterine contractility by mast cells and their mediators. *Am J Obstet Gynecol* 2000; 183: 118–125.

37 Bytautiene E, Vedenikov YP, Saade GR, Romero R, Garfield RE. IgE-independent mast cell activation augments contractility of nonpregnant and pregnant guinea pig myometrium. *Int Arch Allergy Immunol* 2008; 147: 140–146.

38 Bytautiene E, Vedenikov YP, Saade GR, Romero R, Garfield RE. Endogenous mast cell degranulation modulates cervical contractility in the guinea pig. *Am J Obstet Gynecol* 2002; 186: 438–445.

39 Romero R, Mazor M, Munoz H, Gomez R, Galasso M, Sherer DM. The preterm labor syndrome. *Ann NY Acad Sci* 1994; 734: 414–429.

40 Yuan M, Jordan F, McInnes IB, Harnett MM, Norman JE. Leukocytes are primed in peripheral blood for activation during term and preterm labour. *Mol Hum Reprod* 2009; 15: 713–724.

41 Shynlova O, Ned-Roderique T, Li Y, Dorogin A, Nguyen T, Lye SJ. Infiltration of myeloid cells into decidua is a critical early event in the labour cascade and post-partum uterine remodelling. *J Cell Mol Med* 2013; 17: 311–324.

42 Junqueira LC, Zugai M, Montes GS, Toledo OM, Krizstjan RM, Shigihara KM. Morphologic and histochemical evidence for the occurrence of collagenolysis and for the role of neutrophilic polymorphonuclear leukocytes during cervical dilation. *Am J Obstet Gynecol* 1980; 138: 273–281.

43 Osmers R, Rath W, Adelmann-Grill BC, Fittkau C, Kulozik M, Szeverenyi M *et al.*. Origin of cervical collagenase during parturition. *Am J Obstet Gynecol* 1992; 166: 1455–1460.

44 Romero R, Ceska M, Avila C, Mazor M, Behnke E, Lindley I. Neutrophil attractant/activating peptide-1/interleukin-8 in term and preterm parturition. *Am J Obstet Gynecol* 1991; 165: 813–820.

45 Osmers RG, Blaser J, Kuhn W, Tschesche H. Interleukin-8 synthesis and the onset of labor. *Obstet Gynecol* 1995; 86: 223–229.

46 Winkler M, Fischer DC, Ruck F, Marx T, Kaiserling E, Oberpichler A *et al.*. Parturition at term: parallel increases in interleukin-8 and proteinase concentrations and neutrophil count in the lower uterine segment. *Hum Reprod* 1999; 14: 1096–1100.

47 Helmg BR, Romero R, Espinoza J, Chiaiwaropangs T, Bujold E, Gomez R *et al.*. Neutrophil elastase and secretory leukocyte protease inhibitor in prelabour rupture of membranes, parturition and intra-amniotic infection. *J Matern Fetal Neonatal Med* 2002; 12: 237–246.

48 Rinaldi SF, Catalano RD, Wade J, Rossi AG, Norman JE. Decidual neutrophil infiltration is not required for preterm birth in a mouse model of infection-induced preterm labor. *J Immunol* 2014; 192: 2315–2325.

49 Gomez-Lopez N, Mial T, Robertson SA. Infection-induced preterm delivery does not involve a shift in macrophage polarization from the M2→M1 phenotype but implicates a maternal pro-inflammatory state. *Am J Reprod Immunol* 2014; in press.

50 Yellon SM, Ebner CA, Eloitw MA. Medroxyprogesterone acetate modulates remodeling, immune cell census, and nerve fibers in the cervix of a mouse model for inflammation-induced preterm birth. *Reprod Sci.* 2009; 16: 257–264.

51 Sakamoto Y, Moran P, Bulmer JN, Learle RF, Robson SC. Macrophages and non granulocytes are involved in cervical ripening. *J Reprod Immunol* 2005; 66: 161–173.

52 Timmons BC, Mahendroo MS. Timing of neutrophil activation and expression of proinflammatory markers do not support a role for neutrophils in cervical ripening in the mouse. *Biol Reprod* 2006; 74: 236–245.

53 Timmons B, Akins M, Mahendroo M. Cervical remodeling during pregnancy and parturition. *Trends Endocrinol Metab* 2010; 21: 353–361.

54 Shynlova O, Ned-Roderique T, Li Y, Dorogin A, Lye SJ. Myometrial immune cells contribute to term parturition, preterm labour and post-partum involution in mice. *J Cell Mol Med* 2013; 17: 90–102.

55 Romero R, Mazor M, Tartakovsky B. Systemic administration of interleukin-1 induces preterm parturition in mice. *Am J Obstet Gynecol* 1991; 165: 969–971.

56 Steel HH, O’Donoghue K, Kernea NL, Sullivan MH, Edwards AD. Maternal origin of inflammatory leukocytes in preterm fetal membranes, shown by fluorescence in situ hybridisation. *Placenta* 2005; 26: 672–677.

57 Dudley DJ, Trautman MS, Mitchell MD. Inflammatory mediators regulate interleukin-8 production by cultured gestational tissues: evidence for a cytokine network at the chorio-decidual interface. *J Clin Endocrinol Metab* 1993; 76: 404–410.
58 Vadillo OF, Gonzalez AG, Furth EE, Lei H, Muschel RJ, Stetler-Stevenson WG et al. 92-kd type IV collagenase (matrix metalloproteinase-9) activity in human amnionchorion increases with labor. Am J Pathol 1995; 146: 148–156.

59 Athayde N, Romero R, Gomez R, Maymon E, Pacora P, Mazor M et al. Matrix metalloproteinases-9 in preterm and term human parturition. J Matern Fetal Med 1999; 8: 213–219.

60 Maymon E, Romero R, Pacora P, Gomez R, Athayde N, Edwin S et al. Human neutrophil collagenase (matrix metalloproteinase 8) in parturition, premature rupture of the membranes, and intrauterine infection. Am J Obstet Gynecol 2000; 183: 94–99.

61 Hibbs JB Jr, Taintor RR, Vavrin Z, Rachlin EM. Nitric oxide: a cytotoxic activated macrophage effector molecule. Biochem Biophys Res Commun 1988; 157: 87–94.

62 Pavlov O, Pavlova O, Ailamazyan E, Selkov S. Characterization of cytokine production by human term placenta macrophages in vitro. Am J Reprod Immunol 2008; 60: 556–567.

63 Huang WC, Sala-Newby GB, Susana A, Johnson JL, Newby AC. Classical macrophage activation up-regulates several matrix metalloproteinases through mitogen activated protein kinases and nuclear factor-kappaB. PLoS ONE 2012; 7: e42507.

64 Buhimschi I, Ali M, Jain V, Chwalisz K, Garfield RE. Differential regulation of nitric oxide in the rat uterus and cervix during pregnancy and labour. Hum Reprod 1996; 11: 1755–1766.

65 Izumi H, Yallampalli C, Garfield RE. Gestational changes in L-arginine-induced relaxation of pregnant rat and human myometrial smooth muscle. Am J Obstet Gynecol 1993; 169: 1327–1337.

66 Shynlova O, Tsui P, Dorogin A, Lye SJ. Monocyte chemoattractant protein-1 (CCL-2) integrates mechanical and endocrine signals that mediate term and preterm labor. J Immunol 2008; 181: 1470–1479.

67 Fang X, Wong S, Mitchell BF. Effects of LPS and IL-6 on oxytocin receptor in non-pregnant and pregnant rat uterus. Am J Reprod Immunol 2000; 44: 65–72.

68 Gonzalez JM, Dong Z, Romero R, Girardi G. Cervical remodeling/ ripening at term and preterm delivery: the same mechanism initiated by different mediators and different effector cells. PLoS ONE 2011; 6: e26877.

69 Hassan SS, Romero R, Tarca AL, Draghici S, Pineles B, Bugrim A et al. Signature pathways identified from gene expression profiles in the human uterine cervix before and after spontaneous term parturition. Am J Obstet Gynecol 2007; 197: 250.e251–250.257.

70 Hassan SS, Romero R, Tarca AL, Nhan-Chang CL, Vaisbuch E, Erez O et al. The transcriptome of cervical ripening in human pregnancy before the onset of labor at term: identification of novel molecular functions involved in this process. J Matern Fetal Neonatal Med 2009; 22: 1183–1193.

71 Stygar D, Wang H, Vadic YS, Ekman G, Eriksson H, Sahlin L. Increased level of matrix metalloproteinases 2 and 9 in the ripening process of the human cervix. Biol Reprod 2002; 67: 889–894.

72 Watari M, Watari H, DiSanto ME, Chacko S, Shi GP, Strauss JF, 3rd. Pro-inflammatory cytokines induce expression of matrix-metabolizing enzymes in human cervical smooth muscle cells. Am J Pathol 1999; 154: 1755–1762.

73 Word RA, Landrum CP, Timmons BC, Young SG, Mahendroo MS. Transgene insertion on mouse chromosome 6 impairs function of the uterine cervix and causes failure of parturition. Biol Reprod 2005; 73: 1046–1056.

74 Gonzalez JM, Xu H, Chai J, Ofori E, Elowitz MA. Preterm and term cervical ripening in G101 Mice (Mus musculus): similar or divergent molecular mechanisms? Biol Reprod 2009; 81: 1226–1232.

75 Vadillo-Ortega F, Hernandez A, Gonzalez-Avila G, Bermejo L, Iwata K, Strauss JF 3rd. Increased matrix metalloproteinase activity and reduced tissue inhibitor of metalloproteinases-1 levels in amniotic fluids from pregnancies complicated by premature rupture of membranes. Am J Obstet Gynecol 1996; 174: 1371–1376.

76 Athayde N, Edwin SS, Romero R, Gomez R, Maymon E, Pacora P et al. A role for matrix metalloproteinase-9 in spontaneous rupture of the fetal membranes. Am J Obstet Gynecol 1998; 179: 1248–1253.

77 Xu P, Alfady N, Challis JR. Expression of matrix metalloproteinase (MMP)-2 and MMP-9 in human placenta and fetal membranes in relation to preterm and term labor. J Clin Endocrinol Metab 2002; 87: 1353–1361.

78 Braunsteiner AG, Nowak RA. Cytokines regulate matrix metalloproteinases in human uterine endometrial fibroblast cells through a mechanism that does not involve increases in extracellular matrix metalloproteinase inducer. Am J Reprod Immunol 2006; 56: 201–214.

79 Esplin MS, Romero R, Chaiworaopongs T, Kim YM, Edwin S, Gomez R et al. Monocyte chemotactic protein-1 is increased in the amniotic fluid of women who deliver preterm in the presence or absence of intra-amniotic infection. J Matern Fetal Neonatal Med 2005; 17: 365–373.

80 Menzies FM, Shepherd MC, Vibbs RJ, Nelson SM. The role of mast cells and their mediators in reproduction, pregnancy and labour. Hum Reprod Update 2011; 17: 383–396.

81 Abbas AK, Lichtman AH, Pillai S. Cellular and Molecular Immunology. 7th ed. Philadelphia, PA: Elsevier/Saunders, 2012.

82 Walsh LJ, Trinchieri G, Waldorf HA, Whitaker D, Murphy GF. Human dermal mast cells contain and release tumor necrosis factor alpha, which induces endothelial leukocyte adhesion molecule 1. Proc Natl Acad Sci USA 1991; 88: 4220–4224.

83 Juremalm M, Nilsson G. Chemokine receptor expression by mast cells. Chem Immunol Allergy 2005; 87: 130–144.

84 Bosquiaux VL, Durando M, Varayoud J, Ramos JG, Rodriguez HA, Munoz-de-Toro M et al. Macrophage density in the pregnant uterine cervix is modulated by mast cell degranulation. J Reprod Immunol 2005; 65: 147–158.

85 Bytautiene E, Vedeninkov YP, Saade GR, Romero R, Garfield RE. Degranulation of uterine mast cell modifies contractility of isolated myometrium from pregnant women. Am J Obstet Gynecol 2004; 191: 1705–1710.

86 Menzies FM, Higgins CA, Shepherd MC, Vibbs RJ, Nelson SM. Mast cells reside in myometrium and cervix, but are dispensable in mice for successful pregnancy and labor. Immunol Cell Biol 2012; 90: 321–329.

87 Rudinh MI, Reinicke K, Cruz MA, Gallardo V, Gonzalez C, Bardisa L. Distribution of mast cells and the effect of their mediators on contractility in human myometrium. Br J Obstet Gynaecol 1993; 100: 1125–1130.

88 Padilla L, Reinicke K, Monteso H, Villena F, Asencio H, Cruz M et al. Histamine content and mast cells distribution in mouse uterus: the effect of sexual hormones, gestation and labor. Cell Mol Biol 1990; 36: 93–100.

89 Garfield RE, Irani AM, Schwartz LB, Bytautiene E, Romero R. Structural and functional comparison of mast cells in the pregnant versus nonpregnant human uterus. Am J Obstet Gynecol 2006; 194: 261–267.

90 Bytautiene E, Vedeninkov YP, Maner WL, Saade GR, Romero R, Garfield RE. Challenge with ovalbumin antigen increases uterine and cervical contractile activity in sensitized guinea pigs. Am J Obstet Gynecol 2008; 199: 658.e651–658.e656.

91 Bytautiene E, Romero R, Vedeninkov YP, El-Zeky F, Saade GR, Garfield RE. Induction of premature labor and delivery by allergic reaction and prevention by histamine H1 receptor antagonist. Am J Obstet Gynecol 2004; 191: 1356–1361.

92 Erlebacher A, Vencato D, Price KA, Zhang D, Glimcher LH. Constraints in antigen presentation severely restrict T cell recognition of the allogeneic fetus. J Clin Invest 2007; 117: 1399–1411.

93 Gomez-Lopez N, Vadillo-Perez L, Hernandez-Carbajal A, Godines-Enriquez M, Olson DM, Vadillo-Ortega F. Specific inflammatory microenvironments in the zones of the fetal membranes at term delivery. Am J Obstet Gynecol 2011; 205: 16.

94 Sindram-Trujillo A, Scherjon S, Kannhai H, Roelen D, Claas F. Increased T-cell activation in decidua parietalis compared to
decidua basalis in uncomplicated human term pregnancy. Am J Reprod Immunol 2003; 49: 261–268.

95 Tilburgs T, Scherjon SA, Roelen DL, Claas FH. Decidual CD8^{+}/CD28^{−} T cells express CD103 but not perforin. Hum Immunol 2009; 70: 96–100.

96 Tilburgs T, Scherjon SA, van der Mast BJ, Haasnoot GW, Versteeg VDV-MM, Roelen DL et al. Fetal-maternal HLA-C mismatch is associated with decidual T cell activation and induction of functional T regulatory cells. J Reprod Immunol 2009; 82: 148–157.

97 Tilburgs T, Schonkeren D, Eikmans M, Nagtzaam NM, Datema G, Swings GM et al. Human decidual tissue contains differentiated CD8^{+} effector-memory T cells with unique properties. J Immunol 2010; 185: 4470–4477.

98 Sindreau-Trujillo AP, Scherjon SA, van Hulst-van Miert PP, Kanhai HH, Roelen DL, Claas FH. Comparison of decidual leukocytes following spontaneous vaginal delivery and elective cesarean section in uncomplicated human term pregnancy. J Reprod Immunol 2004; 62: 125–137.

99 Romero R, Parvisi ST, Oyarzun E, Mazor M, Wu YK, Avila C et al. Amniotic fluid interleukin-1 in spontaneous labor at term. J Reprod Med 1990; 35: 235–238.

100 Romero R, Mazor M, Brandt F, Sepulveda W, Avila C, Cotton DB et al. Interleukin-1 alpha and interleukin-1 beta in preterm and term human parturition. Am J Reprod Immunol 1992; 27: 117–123.

101 Romero R, Mazor M, Sepulveda W, Avila C, Copeland D, Williams J. Tumor necrosis factor in preterm and term labor. Am J Obstet Gynecol 1992; 166: 1576–1587.

102 Elliott CL, Loudon JA, Brown N, Slater DM, Bennett PR, Sullivan MH. IL-1beta and IL-8 in human fetal membranes: changes with gestational age, labor, and culture conditions. Am J Reprod Immunol 2001; 46: 260–267.

103 Nakashima A, Ito M, Yoneda S, Shiozaki A, Hidaka T, Saito S. Circulating and decidual Th17 cell levels in healthy pregnancy. Am J Reprod Immunol 2010; 63: 104–109.

104 Ito M, Nakashima A, Hidaka T, Okabe M, Bac ND, Ina S et al. A role for IL-17 in induction of an inflammation at the fetomaternal interface in preterm labour. J Reprod Immunol 2010; 84: 75–85.

105 Byrne JA, Stankovic AK, Cooper MD. A novel subpopulation of primed T cells in the human fetus. J Immunol 1994; 152: 3098–3106.

106 Luciano AA, Yu H, Jackson LW, Wolfe LA, Bernstein HB. Preterm labor and chorioamnionitis are associated with neonatal T cell activation. PLoS ONE 2011; 6: e0016698.

107 Kim MJ, Romero R, Kim CJ, Tarca AL, Chhaya S, Lalaneusse C et al. Villitis of unknown etiology is associated with a distinct pattern of chemokine up-regulation in the fetomaternal and placental compartments: implications for conjoint maternal allograft rejection and maternal anti-fetal graft-versus-host disease. J Immunol 2009; 182: 3919–3927.

108 Tilburgs T, Roelen DL, van der Mast BJ, van Schip JJ, Kleijburg C, de Groot-Swings GM et al. Differential distribution of CD4^{+} CD25^{bright} and CD8^{+} CD28^{−} T-cells in decidua and maternal blood during human pregnancy. Placenta 2006; 27: 25.

109 Xu Y, Tarquini F, Romero R, Kim CJ, Tarca AL, Bhattacharyya S et al. Peripheral DC300a^{+} CD8^{+} T lymphocytes with a distinct cytotoxic molecular signature increase in pregnant women with chronic chorioamnionitis. Am J Reprod Immunol 2012; 67: 184–197.

110 Steinborn A, Schmitt E, Kiselewicz A, Rechenberg S, Seissler N, Mahnke K et al. Pregnancy-associated diseases are characterized by the composition of the systemic regulatory T cell (Treg) pool with distinct subsets of Tregs. Clin Exp Immunol 2012; 167: 84–98.

111 Schober L, Radnai D, Schmitt E, Mahnke K, Sohn C, Steinborn A. Term and preterm labor: decreased suppressive activity and changes in composition of the regulatory T-cell pool. Immunol Cell Biol 2012; 90: 935–944.

112 Kiselewicz A, Schair A, Schmitt E, Hug F, Haensch GM, Meuer S et al. A distinct subset of HLA-DR^{−} regulatory T cells is involved in the induction of preterm labor during pregnancy and in the induction of organ rejection after transplantation. Clin Immunol 2010; 137: 209–220.

113 Gomez-Lopez N, Laresgoiti-Servitje E. T regulatory cells: regulating both term and preterm labor? Immunol Cell Biol 2012; 90: 919–920.

114 Tilburgs T, Roelen DL, van der Mast BJ, de Groot-Swings GM, Kleijburg C, Scherjon SA et al. Evidence for a selective migration of fetus-specific CD4^{+} CD25^{bright} regulatory T cells from the peripheral blood to the decidua in human pregnancy. J Immunol 2008; 180: 5737–5745.

115 Samstein RM, Josefowicz SZ, Arvey A, Treuting PM, Rudensky AY. Extrathymic generation of regulatory T cells in placental mammary glands. Nat Immunol 2011; 12: 29–38.

116 Shima T, Sasaki Y, Itoh M, Nakashima A, Ishii N, Sugamura K et al. Regulatory T cells are necessary for implantation and maintenance of early pregnancy but not late pregnancy in allogeneic mice. J Reprod Immunol 2010; 85: 121–129.

117 Arenas-Hernandez M, St-Louis D, Romero R, Hassan S, Gomez-Lopez N. Endotoxin expands the pool of regulatory T cells in pregnancy but not in the non-pregnant state. Reprod Sci 2014; 21: 130A.

118 Hassan SS, Romero R, Vidyadhari D, Fusey S, Baxter JK, Khandelwal M et al. Vaginal progesterone reduces the rate of preterm birth in women with a sonographic short cervix: a multicenter, randomized, double-blind, placebo-controlled trial. Ultrasound Obstet Gynecol 2011; 38: 18–31.

119 Nak A, Rutledge N, Romero R, Hassan S, Gomez-Lopez N. Vaginal Progesterone, but not 170HP-C, induces changes in decidua macrophages and regulatory T cells. Reprod Sci 2014; 21: 332A.

120 Bhat NM, Mithal A, Bieber MM, Herzenberg LA, Teng NN. Human CD5^{+} CD8^{+} lymphocytes (B1 cells) decrease in peripheral blood during pregnancy. J Reprod Immunol 1995; 28: 53–60.

121 Jensen F, Muzzio D, Soldati R, Fest S, Zencilussen AC. Regulatory B10 cells restore pregnancy tolerance in a mouse model. Biol Reprod 2013; 89: 90.

122 Rolle L, Memarzadeh Tehran M, Morell-Garcia A, Raeya Y, Schumacher A, Hartig R et al. Cutting edge: IL-10-producing regulatory B cells in early human pregnancy. Am J Reprod Immunol 2013; 70: 448–453.

123 Canellada A, Farber A, Zencilussen AC, Gentile T, Dokmetjian J, Keil A et al. Interleukin regulation of asymmetric antibody synthesized by isolated placental B cells. Am J Reprod Immunol 2002; 48: 275–282.

124 Kawanoto T, Cui J, Koezuka Y, Toura I, Kaneko Y, Motoki K et al. CD1d-restricted and TCR-mediated activation of valpha14 NK T cells by glycosylceramides. Science 1997; 278: 1626–1629.

125 Jenkinson HJ, Wainwright SD, Simpson KL, Perry AC, Fociadou P, Holmes CH. Expression of CD1D mRNA transcripts in human choriocarcinoma cell lines and placentaly derived trophoblastic cells. Immunology 1999; 96: 649–655.

126 Boyson JE, Rybalov B, Koopman LA, Exley M, Bal SP, Racke FK et al. CD1d and invariant NKT cells at the human maternal–fetal interface. Proc Natl Acad Sci USA 2002; 99: 13741–13746.

127 Kumar V, Delovitch TL. Different subsets of natural killer T cells may vary in their roles in health and disease. Immunology 2014; 15: 12247.

128 Boyson JE, Nagarkatti N, Nizam L, Exley MA, Strominger JL. Gestation-stage-dependent mechanisms of invariant natural killer T cell-mediated pregnancy loss. Proc Natl Acad Sci USA 2006; 103: 4580–4585.

129 Wang S, Li C, Kawamura H, Watanabe H, Abo T. Unique sensitivity to alpha-galactosylceramide of NKT cells in the uterus. Cell Immunol 2002; 215: 98–105.

130 Cantarella R, St-Louis D, Milovic T, Romero R, Gomez-Lopez N. NKT cell activation leads to late preterm birth. Reprod Sci 2014; 21: 230A.

131 Dang Y, Beckers J, Wang CR, Heyborne KD. Natural killer 1.1^{+} alpha beta T cells in the periimplantation uterus. Immunol 2000; 101: 484–491.
132 Mrakovcic-Sutic I, Simin M, Radic D, Rukavina D, Radojevic-Stasic B. Syngeneic pregnancy induces overexpression of natural killer T cells in maternal liver. Scand J Immunol 2003; 58: 358–366.

133 Singh N, Hong S, Scherer DC, Serizawa I, Burdin N, Kronenberg M et al. Cutting edge: activation of NK T cells by CD1d and alpha-galactosylceramide directs conventional T cells to the acquisition of a Th2 phenotype. J Immunol 1999; 163: 2373–2377.

134 Carnaud C, Lee D, Donnars O, Park SH, Beavis A, Koezuka Y et al. Cutting edge: cross-talk between cells of the innate immune system: NKT cells rapidly activate NK cells. J Immunol 1999; 163: 4647–4650.

135 Miko E, Szereday L, Barakonyi A, Jarkovich A, Varga P, Szekeres-Bartho J. The role of invariant NKT cells in pre-eclampsia. Am J Reprod Immunol 2008; 60: 118–126.

136 Li LP, Fang YC, Dong GF, Lin Y, Saito S. Depletion of invariant NKT cells reduces inflammation-induced preterm delivery in mice. J Immunol 2012; 188: 4681–4689.

137 Banchereau J, Steinman RM. Dendritic cells and the control of immunity. Nature 1998; 392: 245–252.

138 Steinman RM, Turley S, Mellman I, Inaba K. The induction of tolerance by dendritic cells that have captured apoptotic cells. J Exp Med 2000; 191: 411–416.

139 Maldonado-Lopez R, de Smedt T, Michel P, Godfroid J, Pajak B, Heirman C et al. CD8alpha1 and CD8alpha2 subclasses of dendritic cells direct the development of distinct T helper cells in vivo. J Exp Med 1999; 189: 587–592.

140 Pulendran B, Smith JL, Caspary G, Brasel K, Pettit D, Maraskovsky E et al. Distinct dendritic cell subsets differentially regulate the class of immune response in vivo. Proc Natl Acad Sci USA 1999; 96: 1036–1041.

141 Schlitzer A, Ginhoux F. Organization of the mouse and human DC network. Curr Opin Immunol 2014; 26: 90–99.

142 Piaks V, Bimberg T, Berkutzki T, Sela S, BenYashar A, Kalchenko V et al. Uterine DCs are crucial for decidua formation during embryo implantation in mice. J Clin Invest 2008; 118: 3954–3965.

143 Bizargity P, Bonney EA. Dendritic cells: a family portrait at mid-gestation. Immunology 2009; 126: 565–578.

144 Blois SM, Alba Soto CD, Tometten M, Klapp BF, Margni RA, Arck PC. Lineage, maturity, and phenotype of uterine murine dendritic cells throughout gestation indicate a protective role in maintaining pregnancy. Biol Reprod 2004; 70: 1018–1023.

145 Ruiz RJ, Jailo N, Murphey C, Marti CN, Godbold E, Pickler RH. Second trimester maternal plasma levels of cytokines IL-1Ra, IL-6 and IL-10 and preterm birth. J Perinatol 2012; 32: 483–490.

146 Bizargity P, Del Rio R, Phillippe M, Teuscher C, Bonney EA. Resistance to lipopolysaccharide-induced preterm delivery mediated by regulatory T cell function in mice. Biol Reprod 2009; 80: 874–881.

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