REPRODUCTION

REVIEW

Focus on Vascular Function in Female Reproduction

Inflammatory pathways in female reproductive health and disease

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Abstract

Inflammation involves alterations to vascular and immune cell function. It is well recognised that many physiological reproductive events such as ovulation, menstruation, implantation and onset of labour display hallmark signs of inflammation. These are orchestrated by specific molecular pathways involving a host of growth factors, cytokines, chemokines and lipid mediators. Resumption of normal reproductive function involves prompt and proper resolution of these inflammatory pathways. Recent literature confirms that resolution of inflammatory pathways involves specific biochemical events that are activated to re-establish homeostasis in the affected tissue. Moreover, initiation and maintenance of inflammatory pathways are the key components of many pathologies of the reproductive tract and elsewhere in the body. The onset of reproductive disorders or disease may be the result of exacerbated activation and maintenance of inflammatory pathways or their dysregulated resolution. This review will address the role of inflammatory events in normal reproductive function and its pathologies.

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Introduction

The Roman encyclopaedist Aulus Cornelius Celsus (ca 25BC–ca 50AD) defined the cardinal signs of inflammation, namely *rubor* (redness), *calor* (increased heat), *tumor* (swelling) and *dolor* (pain). In 1870, Rodulph Virchow highlighted that inflammation is also associated with *functio laesa* or loss of function (Larhammar 1996). In response to a tissue injury or a pathogenic insult, the human body mounts a network of chemical signals that stimulate responses aimed at healing the affected tissue. These signals initiate the activation and chemotaxis of leukocytes from the general circulation to the sites of damage. Inflammatory signals also alter the function of the vasculature and the endothelium to enhance angiogenesis, vascular permeability and the extravasation of leukocytes from the blood to the inflamed tissue (Coussens & Werb 2002, Goswami *et al*. 2008, Serhan *et al*. 2008).

It is becoming increasingly accepted that many normal reproductive processes display hallmark signs of inflammation. Such processes include ovulation, menstruation, implantation and parturition (Goswami *et al*. 2008). All of these events are associated with upregulation in the expression of a host of inflammatory mediators, which include cytokines, growth factors and lipid mediators that influence the growth and function of the immune and vascular compartments (Coussens & Werb 2002, Goswami *et al*. 2008, Serhan *et al*. 2008). Another remarkable feature of the female reproductive tract is its capacity to resolve these inflammatory events rapidly to re-establish normal reproductive function. The resolution of inflammation involves the clearance of leukocytes and tissue debris as well as restoration of mucosal and vascular function in the affected tissue. Until recently, resolution of inflammation was considered a passive process that came about as a result of dissipation in the expression of local inflammatory mediators. However, emerging literature highlights that in response to tissue injury there are specific anti-inflammatory and pro-resolution biochemical pathways that are activated, which facilitate the re-establishment of homeostasis in the affected tissues (Serhan *et al*. 2008). Little is known about the role of pro-resolution pathways in normal reproductive function. However, it is anticipated that in physiological reproductive events (such as

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menstruation, ovulation, implantation and the onset of labour), their expression may be temporally regulated and important in the maintenance of proper reproductive function.

Furthermore, it is well recognised that inflammation-mediated alterations in immune cell and vascular function are important components of many pathologies that include cancer, chronic inflammatory diseases, allergy, asthma, atherosclerosis, autoimmunity, transplant rejection and metabolic and degenerative diseases. Moreover, alterations or disruption in the onset of the pro-resolution pathways may lead to uncontrolled inflammation and the onset of disease, and there is mounting evidence that in animal models of disease the administration of pro-resolution mediators can help control and resolve inflammation (Serhan *et al*. 2008). However, to date the role of pro-resolution pathways in reproductive disorders or their pathologies known to be associated with excessive or premature onset of inflammatory pathways (for example menstrual disorders, infertility, premature onset of labour, cancers of the female reproductive tract) remains to be elucidated.

This review will discuss the role of inflammatory pathways in female reproductive health and will address how alterations in these pathways may contribute to the pathologies of female reproductive function.

Pathways regulating inflammation

In the reproductive tract, the injury and tissue remodelling caused by ovulation, menstruation and parturition trigger the inflammatory cascade. This involves a carefully orchestrated autocrine/paracrine/juxtacrine series of events to facilitate repair, remodelling and the resolution of inflammation which is regulated in a cyclical manner via the sex steroids oestradiol (E_2) and progesterone. Inflammation is an active process which involves the release of inflammatory cytokines, chemokines and peptide growth factors. This establishes a gradient for the recruitment of neutrophils and macrophages to the site of injury. Injury also promotes the activation of the coagulation and fibrinolysis system, which operates in tandem to control clotting and remodelling of the vasculature. This facilitates tissue regeneration and extravasation of neutrophils at the site of injury via dilatation and oedema. Tissue remodelling also involves production of local inflammatory mediators such as kinins, histamine and eicosanoids such as prostanoids (prostaglandins (PGs), prostacyclins and thromboxanes) and leukotrienes.

Immune cells

Approximately 1% of whole human blood comprises leukocytes, more commonly referred to as white blood cells. Leukocytes are divided into two main groups: granulocytes (consisting of neutrophils, basophils and eosinophils) and agranulocytes (consisting of lymphocytes, monocytes and macrophages). Physical injury and inflammation caused by pathogens induce the release of signals such as cytokines, chemokines and growth factors to activate epithelial and endothelial cells, mast cells, macrophages, platelets and neutrophils to facilitate repair (Fig. 1). Inflammatory signals activate haematopoietic stem cells derived from bone marrow to produce monocytes (Ziegler-Heitbrock 2007). Monocytes give rise to macrophages or dendritic cells and are recruited by chemotaxis into the damaged tissue by extravasation. Once in the tissue they can phagocytose cellular debris and pathogens and stimulate lymphocytes (Ziegler-Heitbrock 2007). Neutrophils, which are a key mediator of the inflammatory response, recruit, activate and programme antigen-presenting cells to activate T cells as well as release local mediators to attract monocytes and dendritic cells (Nathan 2006). Neutrophils also generate signals to determine whether macrophages differentiate into a pro- or anti-inflammatory state and are responsible for lymphocyte expansion and lymph node drainage. In addition, neutrophils are key mediators of wound healing and microbial sterilisation as individuals with insufficient neutrophils display poor wound healing, and in severe cases this can be fatal (Nathan 2006). Mast cells are tissue-based inflammatory cells derived from CD34+ pluripotent stem cells and are recruited to the site of injury together with monocytes, macrophages and neutrophils by chemotaxis and are involved in wound healing, defence against pathogens and release of histamines to facilitate vasodilation and oedema associated with inflammation. They also produce the chemoattractant factor, interleukin (IL)16 to recruit CD4+T lymphocytes (Prussin & Metcalfe 2003). In addition to being recruited by inflammatory stimuli, immune cells also amplify and sustain the response by the release of local inflammatory mediators (cytokines, chemokines, growth factors and eicosanoids) at the site of recruitment.

The coagulation and fibrinolysis system

During coagulation, fibrin is deposited to form a clot to limit blood loss (Fig. 1). Inflammatory cytokines such as IL6 are the main mediators of inflammation-induced coagulation (Levi & van der Poll 2005). IL6 in turn induces expression of tissue factor (TF; also known as Factor III (F3) or CD142) via signal transduction pathways such as the MAPK pathway and transcription factors such as early growth response factor 1 (Sampson & Kakkar 2002). TF then promotes thrombin generation via the activation of specific G-protein-coupled protease-activated receptors (PARs1–4; Sampson & Kakkar 2002, Hollenberg *et al*. 2008); leading to conversion of fibrinogen to fibrin (ten Cate *et al*. 1994, van der Poll *et al*. 1994, Belting *et al*. 2005). Thrombin also acts as a potent platelet activator to enhance fibrin deposition and

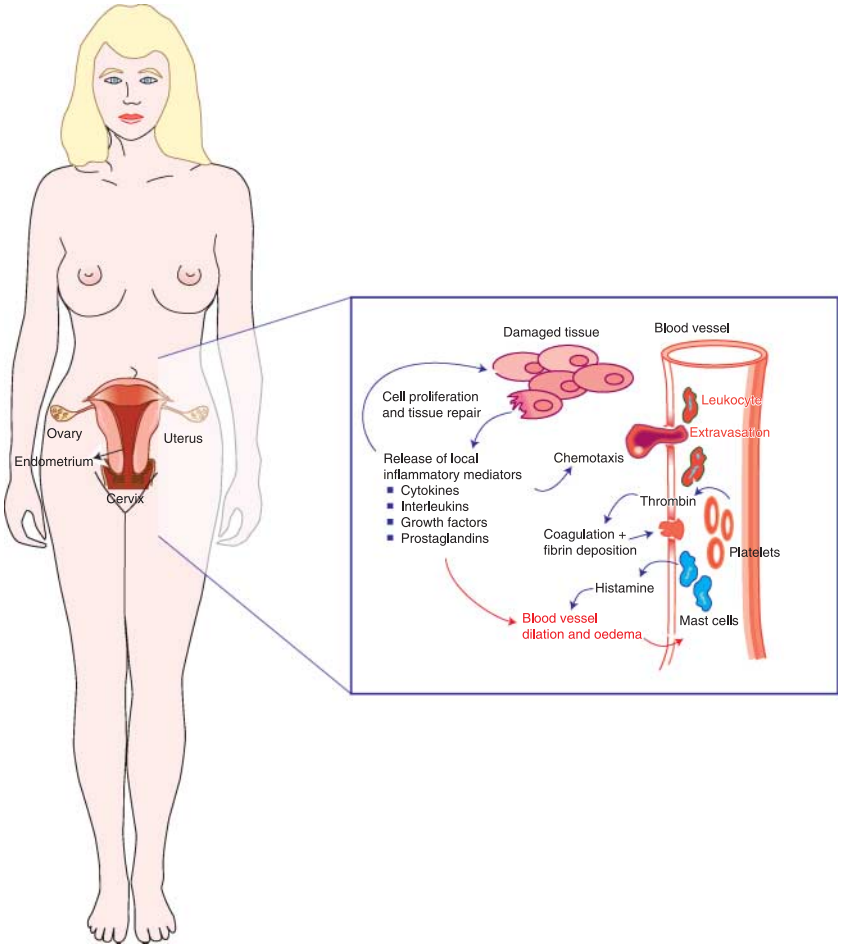

Figure 1 Schematic diagram highlighting the pathways involved in inflammation and tissue repair. Damaged tissue releases a host of local inflammatory mediators including cytokines, interleukins, growth factors and prostaglandins. These mediators activate numerous pathways, which act synergistically to control tissue remodelling and repair. For example, inflammatory mediators are released to facilitate cellular proliferation and repair the damaged tissue and act together with histamine, released from mast cells, to alter vascular tone and facilitate movement of fluid and plasma into the tissue to promote oedema and swelling. This slows blood flow and allows leukocytes to marginate along the endothelium and then extravasate into the damaged tissue by chemotaxis in response to stimuli from local inflammatory mediators. In parallel, thrombin acts as a potent platelet activator to enhance fibrin deposition and remodelling of the vasculature, thereby facilitating angiogenesis.

enhances TF expression by neutrophils, mononuclear cells and macrophages (Levi & van der Poll 2005). The coagulation pathway is tightly regulated by three important anticoagulant pathways, namely the antithrombin, the protein C system and the TF pathway inhibitor (Levi & van der Poll 2005). These pathways are all impaired during inflammation to drive fibrin deposition and coagulation and are implicated in mortality and morbidity under conditions of severe or chronic inflammation (Levi & van der Poll 2005). Following tissue repair, fibrinolysis is induced as the fibrin clot is removed enzymatically by plasmin. Plasmin is produced from plasminogen following the release of plasminogen activators (tissue-type plasminogen activator (PA) and urokinase-type PA) by the cytokines IL1B and tumour necrosis factor α (TNF; Levi & van der Poll 2005). This induction of plasmin from plasminogen is counterbalanced by the induction of PA inhibitor type-(PAI)1 (Levi & van der Poll 2005). Defects in the fibrinolysis cascade due to alteration in the levels of PAI1 can contribute to thrombosis and morbidity due to inadequate fibrin removal.

Vascular changes during inflammation

During inflammation various molecules are produced which promote dilatation of blood vessels and increased vascular permeability. One example is histamine, which is produced from mast cells (Prussin & Metcalfe 2003). Histamine facilitates movement of fluid and plasma into the tissue thereby inducing oedema and swelling. This slows blood flow and allows leukocytes to marginate along the endothelium and then extravasate into the tissue (Fig. 1). In addition, thrombin produced locally within the vasculature is known to promote vasorelaxation in endothelium-containing vessels via the activation of PAR1 and 2 receptors to enhance blood flow to the site (Hollenberg *et al.* 2008). Members of the kallikrein–kinin system of blood proteins, such as bradykinin, are also produced at the site of inflammation by the proteolytic cleavage of a kininogen precursor, high molecular weight kininogen (HMWK or HK). Kinins exert their effects by binding to two classes of G protein-coupled receptors (GPCRs), B1 and B2 (Campos *et al.* 2006).

Whereas B2 receptors are thought to be constitutively expressed, B1 receptors are upregulated during inflammation and are thought to play a role in numerous inflammatory diseases (Campos *et al*. 2006). Activation of both B1 and B2 receptors by kinins such as bradykinin leads to activation of diverse signal transduction pathways including activation of phospholipase Cβ, generation of inositol 1,4,5-trisphosphate and mobilisation of intracellular calcium (Calixto *et al*. 2001, 2004, Campos *et al*. 2006). Furthermore, they activate phospholipase A2 and increase arachidonic acid release giving rise to local production of prostanoids such as PGE_2 (Calixto *et al*. 2001, Campos *et al*. 2006), which in turn can induce expression of B2 receptor. Activation of PARs by TF can also induce vessel sprouting and morphogenesis via the release of vascular endothelial growth factor (VEGF), which under conditions of chronic inflammation may play a role in regulating angiogenesis in disease (Boccaccio & Medico 2006). Inflammatory prostanoids such as PGE_2 and $PGF_{2\alpha}$ in addition to regulation of B2 receptor have also been shown to regulate expression of angiogenic factors such as VEGF via the activation of specific GPCRs like the E prostanoid 2 (EP2) receptor and F prostanoid receptor to promote angiogenesis, proliferation and cytoskeletal reorganisation and cell motility for tissue remodelling (Milne & Jabbour 2003, Sales & Jabbour 2003*a*, Jabbour & Sales 2004, Sales *et al*. 2004, 2008).

Neuroimmunoendocrinology regulation

Immune-endocrine disequilibrium attributed to stress has become a commonly cited factor when discussing unexplained reproductive failures including infertility, impaired oogenesis, miscarriages, preterm labour and impaired fetal development (as reviewed in Nepomnaschy *et al*. (2007)). New multidisciplinary research on brain–body interactions triggered by stress in early pregnancy has shown that maternal biological responses, including localised inflammation in uterine tissue and sustained depression of progesterone production, challenge the endocrine–immune steady state during pregnancy, leading to serious consequences for the fetal environment (Arck *et al*. 2007) This 'pregnancy stress syndrome' is associated with over-activation of the hypothalamic–pituitary–adrenal (HPA) axis which triggers the release of neurohormones, and subsequently the activation of the HPA axis stimulates upregulation of key stress hormones such as corticotrophin-releasing hormone, ACTH and glucocorticoids. The elevated levels of circulating stress hormones consequently lead to altered inflammatory pathways and immune cell function affecting reproductive function (as reviewed in Nakamura *et al*. (2008)).

Inflammatory mediators

Sex steroids

The ovarian sex steroids E_2 and progesterone are responsible for orchestrating the dynamic tissue remodelling observed in the ovary and endometrium during the normal reproductive cycle by activating gene transcription via the specific nuclear E_2 (ER) and progesterone (PR) receptors (Saunders 2005). The expression of ER and PR is under dual control of E_2 and progesterone and is spacio-temporally expressed in the endometrium (as reviewed in Critchley *et al*. (2001), Kelly *et al*. (2001) and Saunders (2005)). The complex role of steroids in inflammation and their regulation of inflammatory mediators has been extensively reviewed elsewhere (Auersperg *et al*. 2001, Critchley *et al*. 2001, Richards *et al*. 2002, Kayisli *et al*. 2004, Saunders 2005, Lea & Sandra 2007, Straub 2007).

Cytokines, chemokines and growth factors

Cytokines are a large family of more than 100 low molecular weight proteins that function as growth and differentiation factors and immune cell modulators. The chemoattractive cytokines or chemokines are a large family grouped on the basis of the arrangement of two N-terminal cysteine residues, CXC and CC, depending on whether the first two cysteine residues have one or more amino acids between them (CX···C) or are adjacent to one another (CC). Roughly 50 chemokines and 20 chemokine receptors have been identified, and they induce diverse responses such as immune cell recruitment, tissue repair and leukocyte extravasation (Thelen & Stein 2008).

In the reproductive tract, coincident with the activation of the coagulation cascade, injury causes inflammatory cytokines to be released locally within tissues (Fig. 1). These cytokines act in an autocrine/paracrine manner to elicit cell-specific events, depending on the temporal nature in which they are released. For example, IL1 is a potent chemotactic cytokine, with pyrogenic and immunomodulatory actions in the reproductive tract (Amjad *et al*. 2006) and is an early response cytokine necessary for wound repair (Salamonsen 2003). IL6, via the IL6 receptor–GP130 complex is one of the most potent cytokines in promoting inflammatory events through expansion and activation of T cells and differentiation of B cells and the acute phase response as well as activation of the coagulation cascade as detailed above (Guazzone *et al*. 2009). IL8 is known for its ability to activate macrophages and recruit neutrophils and T cells; however, it can also act as an autocrine growth factor to promote angiogenesis in endometrial vessels and proliferation of endometrial stromal cells and has been shown to enhance re-epithelialisation of skin grafts as well as to facilitate tissue remodelling (Gimbrone *et al*. 1989, Arici *et al*. 1998*a*,

1998*b*, Kayisli *et al*. 2002, Salamonsen 2003, Ulukus *et al*. 2005). Furthermore, activation of the coagulation cascade directly promotes the production of inflammatory cytokines and chemokines. For example, fibrin can stimulate mononuclear cells and endothelial cells to produce IL6 or IL8 (van der Poll 2001, van der Poll *et al*. 2001), while thrombin can act on endothelial cells to enhance production of IL8, monocyte chemoattractant protein 1 and E-selectin (van der Poll 2001). These ligands can act via their specific receptors on cells within the microenvironment to activate diverse signal transduction cascades, target genes and adhesion molecules and are potential targets for therapeutic intervention for numerous inflammatory diseases (Feldmann 2008).

In addition to cytokines and chemokines, peptide growth factors such as epidermal growth factor (EGF), platelet-derived growth factor (PDGF) and transforming growth factor (TGF) have been identified as paracrine mediators in platelets, macrophages and lymphocytes, where they play a multifactorial role (Auersperg *et al*. 2001). For example, TGFB (TGFβ) and PDGF are potent leukocyte chemoattractants. PDGF released form platelets is a chemoattractant for fibroblasts and stimulates the production of collagenase by fibroblasts to promote remodelling of the extracellular matrix (Sporn & Roberts 1986, Wahl *et al*. 1989). TGFB released from platelets, T lymphocytes and monocytes can induce PDGF, fibroblast growth factor, IL1 and TNF in a host of cell types to regulate tissue repair by controlling the recruitment of new cells and the formation of new matrix and blood vessels necessary for the repair process (Sporn & Roberts 1986, Wahl *et al*. 1989).

Lipid mediators

Glycerophospholipids such as phosphatidylcholine are the main structural eukaryotic membrane lipids (Wymann & Schneiter 2008). Their release by phospholipases gives rise to arachidonic acid and lysophosphatidylcholine (Wymann & Schneiter 2008). Arachidonic acid is then enzymatically converted by cyclooxygenases (COX) to PG, prostacyclins and thromboxanes or by lipoxygenase (LOX) enzymes to leukotrienes (Rajakariar *et al*. 2006, Wymann & Schneiter 2008). Lysophosphatidylcholine is in turn enzymatically converted to lysophosphatidic acid (LPA; Fig. 2; Mills & Moolenaar 2003, Wymann & Schneiter 2008). For many years the role of PGs in inflammation has been ascertained from studies conducted using non-steroidal anti-inflammatory drugs (NSAIDs, which function by blocking COX-catalysed synthesis of prostanoids: PGs, prostacyclins and thromboxanes (Vane & Botting 1998); COX-enzyme-specific knock-out mice (Loftin *et al*. 2002, Rajakariar *et al*. 2006)). At the onset of the inflammatory response, under the regulation of E_2 and progesterone (Critchley *et al*. 2001), a range of growth factors, prostanoids, cytokines, chemokines and ILs promote the production of inflammatory prostanoids by inducing expression of COX enzymes. For example, mast cells and granulocytes recruited to the site of inflammation are activated to release granule contents and promote the production of PGs, thromboxanes and leukotrienes (Wymann & Schneiter 2008). As discussed earlier, bradykinin via its receptor activates phospholipase to promote prostanoid production and thrombin activation of platelets causes production of thromboxane and platelet aggregation. This is balanced by the release of prostacyclin which has a vasodilatory and anti-aggregatory effect on vascular function. Prostanoids mediate these effects in inflammation, following their binding to and activation of specific GPCRs (Fig. 2; Jabbour *et al*. 2006), which are present in a cell-specific manner (Coleman *et al*. 1994). For example, PGE_2 in the CN is induced by inflammatory cytokines IL1 and TNF from activated immune cells and is responsible for regulating fever via the EP3 receptor (Murakami & Kudo 2004). Locally at the site of inflammation, COX enzymes and PGs can promote immune cell infiltration, cellular proliferation and angiogenesis to facilitate tissue remodelling (Fig. 1). LPA via its GPCRs has also been shown to play a role in inflammation in the reproductive tract via the release of cytokines such as IL6 and IL8 and modulation of urokinase-type PA (Ye 2008).

In addition to their pro-inflammatory roles, COX enzymes also play a role in producing anti-inflammatory prostanoids, including PG D2 and 15-deoxy-δ-PG J2, which are thought to play a role in resolution of inflammation (Murakami & Kudo 2004, Rajakariar *et al*. 2006, Scher & Pillinger 2009). The LOX pathway is also thought to contribute towards the resolution of inflammation via the production of anti-inflammatory molecules such as lipoxins and resolvins (Serhan *et al*. 2008).

Inflammatory pathways in reproductive physiology

Ovulation

The hypothesis that mammalian ovulation is comparable to an inflammatory reaction was first proposed by Espey (1980) since many of the molecules responsible for inducing the inflammatory cascade including PGs, leukotrienes, bradykinin, histamine, platelet activating factor and various cytokines have been described in the ovary (Espey 1994). Ovulation is initiated by the LH surge and is controlled by the spacio-temporal expression of specific genes (as reviewed in Richards *et al*. (2002)). The process of ovulation destroys the ovarian surface epithelium and vasculature at the site of oocyte expulsion. Inadequate resolution or remodelling and repair at the site of expulsion are thought to predispose the tissue to neoplastic transformation by the accumulation of genetic mutation following DNA damage (Fleming *et al*. 2006). The ovarian surface is covered by

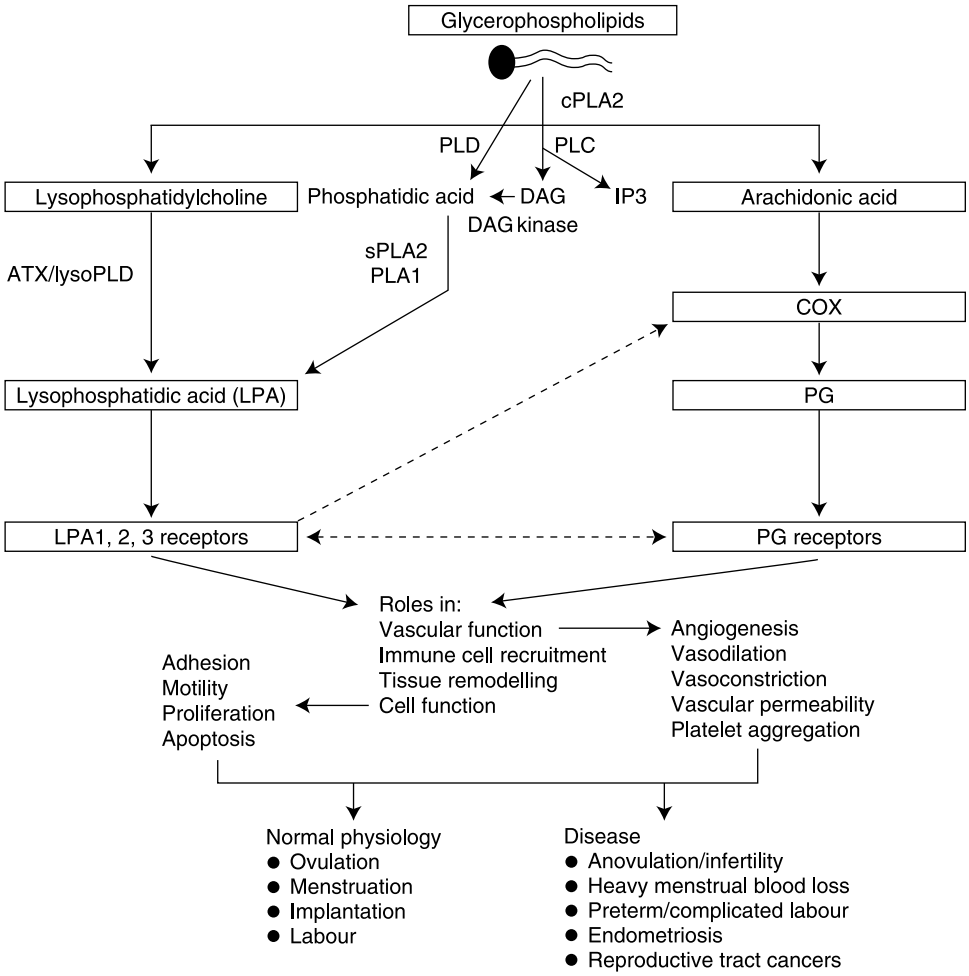

Figure 2 Schematic diagram outlining the pathways regulating lipid signalling. Glycerophospholipids are the main eukaryotic membrane lipids. Their diacylglycerol (DAG) backbone carries a phosphate (phosphatidic acid) esterified to either choline, ethanolamine, serine or inositol. Hydrolysis of membrane lipids by cytosolic phospholipase A2 (cPLA2) gives rise to arachidonic acid and lysophosphatidylcholine. Arachidonic acid is then stereospecifically oxygenated through the cyclooxygenase (COX) pathway, thereby producing prostaglandins (PG). Although the metabolic pathways responsible for LPA biosynthesis are poorly delineated, LPA can be produced by hydrolysis of phosphatidic acid, following its release from glycerophospholipids by phospholipase D (PLD), by soluble phospholipase A2 (sPLA2) or phospholipase A1 (PLA1) or by the cleavage of lysophosphatidyl choline by ATX/lysoPLD. In addition, the phospholipase C (PLC) breakdown products of membrane lipids, DAG and inositol 1,4,5 trisphosphate (IP3) serve as lipid second messengers to promote cell signalling. Once biosynthesised, PG and LPA are released from the cell and exert their biological function via specific G-protein-coupled receptors. Ligand–receptor activation of either LPA or PG pathways can regulate their own biosynthesis and that of each other in a positive feedback manner to regulate inflammatory signalling in normal reproductive physiology as well as disease by altering vascular function, immune cell recruitment, tissue remodelling and cellular function.

a single layer of flat to cuboidal cells referred to as human ovarian surface epithelial cells (OSE or hOSE) or ovarian mesothelium, which is responsible for proteolytic remodelling of the ovarian surface (Auersperg *et al*. 2001). Although receptors for E_2, progesterone and androgen are found on OSE cells, no direct effect of these steroids on OSE cellular proliferation has been observed (Auersperg *et al*. 2001). However, indirect effects of sex steroids on ovarian surface epithelial cell function have been observed *in vitro* via the upregulation of growth factors (Auersperg *et al*. 2001). During the mid-cycle LH surge, leukocytes migrate into the thecal layer and upon ovulation migrate into the granulosa layer coincident with rupture of the basement membrane (Bukulmez & Arici 2000). Following rupture of the ovarian surface epithelium, repair and organisation of the site is necessary to form a corpus luteum. The coagulation cascade is triggered, driven by the release of cytokines, produced locally at the site of tissue damage and by invading leukocytes as well as by growth factors present in the follicular fluid (Fig. 1; Yamada & Gentry 1995, Bukulmez & Arici 2000). At this time, EGF and PDGF are released from platelets and together with the pro-inflammatory cytokines and induction of COX enzymes and prostanoids stimulate OSE growth (Auersperg *et al*. 2001). Invading leukocytes release nitric oxide to facilitate vasodilatation and secretion of IL1 for tissue remodelling (Fleming *et al*. 2006). IL1 has been shown to

upregulate pro-inflammatory genes in OSE cells, including IL6, IL8 and nuclear factor kappa β (NFKB; Rae *et al.* 2004*b*). These in turn can sustain cellular proliferation (Auersperg *et al.* 2001) via the activation of MAPK signalling in a positive feedback manner to facilitate rapid repair of the ovarian surface epithelium. During the repair process, infiltrating macrophages produce TNF, which also facilitates proliferation of OSE cells and produces TNF expression in OSE cells to sustain repair (Auersperg *et al.* 2001). Growth factors, chemokines and cytokines also induce expression of COX enzymes and promote local production of prostanoids (Rae *et al.* 2004*b*). The role of COX enzymes and PGs in the inflammation of ovulation is unclear, since inhibition of COX enzyme with NSAIDs inhibits ovulation completely. However, we can speculate on their role in initiating inflammatory pathways based on the known roles for these molecules as discussed earlier. It is feasible that COX enzymes and PGs promote immune cell recruitment, tissue remodelling and angiogenesis in the ovary post ovulation. Finally, inflammatory stimuli such as IL1A also enhance the steroidogenic environment in granulosa cells and OSE cells to increase 11β-hydroxysteroid dehydrogenase type-1. This enhances conversion of cortisone to cortisol to facilitate repair and counteract the inflammatory response (Rae *et al.* 2004*a*, Fleming *et al.* 2006). During this time, the TGFB family of cytokines present in exudates can exert a growth inhibitory effect on OSE cells by counteracting the proliferative effects of EGF (Auersperg *et al.* 2001).

Menstruation

The human endometrium undergoes extensive remodelling during every menstrual cycle. This process involves the disintegration of the functionalis layer of the endometrium and regeneration and differentiation of a new layer in preparation for an implanting embryo. The features of menstruation are parallel to those of an inflammatory response with the expression of inflammatory cytokines, chemokines and prostanoids. Moreover, there is an abundance of leukocytes in the endometrium prior to the onset of menstruation indicating a role for these factors and cells in the remodelling process.

The actions of PGs on the endometrium result in ischaemia, tissue necrosis and shedding of the endometrium. Endometrial $PGF_{2\alpha}$ and prostacyclin (PGI_2) are highest before the onset of menstruation and induce cyclic blood vessel vasoconstriction and vasodilation respectively (Baird *et al.* 1996). Thromboxane A2 is a potent vasoconstrictor and stimulator of platelet aggregation; whereas, PGI_2 is an inhibitor of aggregation (Salamonsen *et al.* 1999). The level of PGs $PGF_{2\alpha}$ and PGE_2 during the menstrual cycle is regulated by the catabolic enzyme prostaglandin-15-dehydrogenase (PGDH) which is regulated by progesterone, following progesterone withdrawal PGDH expression declines leading to a rise in the levels of PGs peri-menstrually (Norman *et al.* 1991).

In the peri-menstrual period, there is a dramatic influx of inflammatory-type leukocytes: uterine natural killer (uNK) cells, neutrophils, eosinophils, macrophages and activated mast cells (Salamonsen & Lathbury 2000). The origin of uNK cells is unknown, although evidence suggests that peripheral NK cells migrate to the uterus and in the hormone-rich uterine environment they proliferate and differentiate (Moffett-King 2002, Kane *et al.* 2009). Regulation of the migration of leukocytes to the endometrium in response to chemoattractant cytokines and chemokines (Fig. 1) is further enhanced by the action of PGE_2 on the blood vessels to induce capillary leakage (Colditz 1990). Increased expression of chemotactic cytokines and chemokines prior to menstruation plays an important regulatory role in immune cell recruitment, for example IL8 and CCL2. Expression of IL8 in the endometrial epithelial cells and arterioles of the late secretory phase may regulate the recruitment of neutrophils before menstruation (Arici *et al.* 1998*a*). CCL2 has a similar expression pattern with high levels during menstruation and is a potent attractant of macrophages, T cells, NK cells, basophils and mast cells (Jolicoeur *et al.* 1998). The influx of leukocytes into the endometrium and their activation immediately prior to menstruation provides cellular interactions, which are critically important to matrix metalloproteinase (MMP) expression and matrix degradation. For example, eosinophils provide a wide range of secretory products, which activate mast cells. Mast cell activation results in the release of potent regulators such as TNF, histamine and mast cell-specific proteases preceding menstruation (Sivridis *et al.* 2001). The importance of such cellular interactions has been demonstrated by the production and activation of MMP1 and MMP3 by endometrial stromal cells during co-culture with mast cells (Zhang *et al.* 1998).

Finally, after the action of pro-inflammatory mediators and immune cells to induce menstruation, it is essential to regulate menstrual blood flow to allow tissue elimination without excessive bleeding. Regulation is maintained by activation of the haemostatic and fibrinolytic systems to ensure a correct balance of blood coagulation. TF is the primary initiator of haemostasis generating fibrin and leading to clot formation (Fig. 1; Lockwood *et al.* 1993). Fibrinolysis leading to clot degradation is regulated by the availability of plasmin by the action of PA. Progesterone increases TF expression but decreases PA expression, therefore progesterone withdrawal immediately prior to menstruation induces a haemorrhagic environment with decreased clotting and increased fibrinolysis (Casslen *et al.* 1995).

Implantation and placentation

Pro- and anti-inflammatory pathways are involved in the establishment of a receptive endometrium (window of implantation) and also in embryo–endometrium communication. Many cytokines produced by the embryo are pro-inflammatory, suggesting that implantation is a process in which the embryo induces inflammatory pathways in the endometrium. The endometrium responds to these embryonic signals during the window of implantation by enhancing expression of receptivity genes required for embryo adhesion and invasion (Sherwin *et al*. 2007, Evans *et al*. 2009). In preparation for implantation, decidualisation in humans occurs initially during the secretory phase around the spiral arterioles and significantly throughout the endometrium if pregnancy occurs. Decidualisation, in addition to differentiation of endometrial stromal cells into decidual cells, involves initiation of inflammatory events such as infiltration of leukocytes, modification of the extracellular matrix and an increase in vascular permeability (Popovici *et al*. 2006, Hess *et al*. 2007). IL1B and TNF in particular have emerged as candidate genes responsible for the activation of the pro-inflammatory cascade at the fetal–maternal interface (Hess *et al*. 2007). These primary pro-inflammatory cytokines activate production of secondary mediators such as cytokines, chemokines, COX enzymes, PGs and pentraxin 3 (PTX3). PTX3 is novel mediator and plays a key role as an effector and modulator of innate resistance, inflammation and angiogenesis and is localised to perivascular and endothelial cells of first trimester decidua (Garlanda *et al*. 2008). Cytokines and in particular; the IL6 family members (IL11, leukaemia inhibitory factor (LIF) and IL6) play an important role in implantation. *In vitro* studies have shown that their receptors are expressed at the implantation site by several cell types (van Mourik *et al*. 2009). Gene knockout mouse models have demonstrated that both IL11 and LIF play important roles in implantation, and IL6 influences fertility and implantation efficiency (Stewart *et al*. 1992, Robb *et al*. 1998, Jasper *et al*. 2007). LIF is a pro-inflammatory cytokine expressed in the epithelium and decidual stromal cells and is regulated by several inflammatory mediators such as IL1, TNF, leptin, insulin-like growth factor, TGFB (Gonzalez *et al*. 2004, Perrier d'Hauterive *et al*. 2004, Kimber 2005) and more recently prokineticins (Evans *et al*. 2009), suggesting intricate regulation by inflammatory pathways. Several other roles have been described for LIF including immune cell recruitment to the endometrium (Schofield & Kimber 2005) and trophoblast adhesion to extracellular matrix proteins (Tapia *et al*. 2008, Evans *et al*. 2009).

Distinct leukocyte subpopulations are present in the endometrium during implantation. Macrophages, a small number of T cells, and uNK cells predominate in the decidua especially at the sites of trophoblast invasion. Chemokines such as CCL4, CCL7 and CCL13 which recruit these types of leukocytes are upregulated in the endometrial glands during endometrial receptivity and by decidual stromal cells in early pregnancy (Jones *et al*. 2004). At the human implantation site, uNK cells account for 70% of the leukocytes and interact with the allogeneic placenta. By recognising paternal trophoblast ligands, uNK cells may control the extent of placental invasion. The molecular mechanism for the maternal recognition of trophoblast is via the MHC class I molecules HLA-C, HLA-E and HLA-G expressed by the trophoblast cells which are recognised by receptors (such as the killer-cell immunoglobulin-like receptors) expressed on uNK cells (Boyington *et al*. 2001). In humans, it is proposed that this interaction mediates the immune cell response preventing trophoblast over-invasion, but allowing placental access to the maternal blood supply (Moffett-King 2002). There is also a central role for T cell-derived cytokines in the regulation of fetal allograft survival. Changes in the production of hormones such as progesterone and relaxin play a major role in modulating T helper 1/T helper 2 (Th1/Th2)-type cytokine balance (Piccinni *et al*. 2000). Th1-type cytokines promote allograft rejection and compromise pregnancy. The production at the fetal–maternal interface of Th2-type cytokines such as IL4 and IL10 inhibits the Th1 responses and improves fetal survival (Piccinni *et al*. 2001).

Considerable evidence has accumulated indicating that PGs have an important role during implantation (Kennedy *et al*. 2007). PGs are elevated in areas of increased endometrial vascular permeability associated with the initiation of implantation. Further evidence comes from numerous reports that NSAIDs delay or inhibit localised increase in vascular permeability and implantation (Hamilton & Kennedy 1994), and mice with COX2 ablated have multiple reproduction abnormalities including retarded decidualisation (Cheng & Stewart 2003). The type(s) of PG and receptors involved in human embryo implantation still remains unclear and is compounded by animal studies that show species differences (Kennedy *et al*. 2007).

The pro-inflammatory pathways induced during implantation are regulated by anti-inflammatory mediators such as adiponectin and IL10 to prevent excessive inflammation. Adiponectin is a pleiotropic cytokine (Maeda *et al*. 1996) and in addition to playing an important role in regulating energy metabolism and insulin sensitivity (Yamauchi *et al*. 2001), it has been shown to have anti-inflammatory (Brakenhielm *et al*. 2004) and anti-angiogenic activities (Goldstein & Scalia 2004). Adiponectin and the two adiponectin receptors (ADIPOR1 and ADIPOR2) have been shown to be expressed in the epithelial and stromal cells of the endometrium with expression levels of the receptors peaking during the window of implantation (Takemura *et al*. 2006). In the endometrium, adiponectin has been demonstrated to inhibit IL1B-induced expression of IL6

and IL8, suggesting that adiponectin signalling plays a role in regulating pro-inflammatory pathways during implantation (Takemura *et al*. 2006). IL10 is a well characterised anti-inflammatory and immune-modulating cytokine expressed in the endometrium and placenta (Hanna *et al*. 2000). IL10 has the ability to reduce inflammation by inhibiting synthesis of TNF, IL1 and other pro-inflammatory cytokines and chemokines (Moore *et al*. 2001). IL10 null mutant mice demonstrate IL10 as a key regulator of fetal and placental growth (White *et al*. 2004).

Regulation of inflammation during implantation may follow a sequential model in which pro-inflammation is followed by anti-inflammation or there may be a continuous balance between the pro- and anti-inflammatory environments. Despite the characterisation of several mediators of inflammation, the mechanism of inflammatory pathway regulation during implantation is unclear and warrants further investigation.

Labour

There is an emerging evidence that physiological parturition is associated with upregulation of inflammatory pathways. Labour at term is associated with a massive neutrophil and macrophage influx into the myometrium and cervix (Thomson *et al*. 1999, Osman *et al*. 2003). Myometrium, cervix and fetal membranes all release pro-inflammatory cytokines during parturition, with upregulation of pro-inflammatory cytokines being largely but not exclusively confined to invading leukocytes (Ledingham *et al*. 2001, Young *et al*. 2002, Osman *et al*. 2006). Normal labour is associated with upregulation of inflammatory pathways, with NFKB activation appearing to play a key role (Allport *et al*. 2001). More recently, genomic analysis of labouring versus non-labouring uterine and fetal tissue has confirmed that inflammatory genes are among those whose gene expression is most profoundly altered during labour (Haddad *et al*. 2006, Bollopragada *et al*. 2009). The initiating signal(s) that drives these inflammatory events is unknown, although expression of innate immune receptors that receive these signals (the Toll like receptors) is increased towards the end of pregnancy and further increase in labour. Importantly, pro-inflammatory events are not limited to the uterus, with evidence of greater chemotactic ability of leukocytes in peripheral blood in labouring compared with non-labouring women (Yuan *et al*. 2009).

Classically, inflammation is the triad of heat, swelling and pain, and the mechanism by which these events might contribute to the process of parturition is not immediately obvious. However, further consideration suggests that release of pro-inflammatory cytokines may play a major role via stimulation of myometrial contractions. For example, myocytes pre-incubated with IL1B display greater increases in intracellular calcium (which itself is linked to smooth muscle contraction) in response to stimuli compared with myocytes incubated in control media (Tribe *et al*. 2003). IL1B also increases expression of COX2 (itself an initiator of myometrial contractions; Rauk & Chiao 2000) and phosphodiesterase activity in myocytes (Oger *et al*. 2002) again stimulating contractions. Additionally, IL1B administration stimulates preterm labour in a mouse model (Romero & Tartakovsky 1992). In the cervix, IL1B stimulates MMPs (Watari *et al*. 1999), which is likely involved in the process of collagen breakdown during cervical ripening which occurs before and in the early phases of parturition (Yoshida *et al*. 2002). Additionally, leukocytes invading the cervix release nitric oxide, which again induce cervical ripening (Thomson *et al*. 1997, Ledingham *et al*. 2000). A pathway by which inflammation induces parturition might involve initiation of inflammatory stimuli signalling via Toll-like receptors, which results in PG and MMP production in addition to leukocyte invasion into reproductive tissues, and culminating in myometrial contractility, rupture of membranes and cervical ripening (Challis *et al*. 2009).

Although much focus has been on the 'pro-inflammatory' pathways of parturition, there is some evidence that endogenous anti-inflammatory pathways are also active. For example, IL10 levels rise in amniotic fluid during labour in humans (Gotsch *et al*. 2008). We have recently investigated the potential role of anti-inflammatory lipid mediators such as lipoxins in parturition and shown an increase in both synthetic capacity and receptor density of these molecules in the myometrium during parturition (Maldonado-Perez *et al*. 2009; Fig. 3).

Inflammatory pathways in reproductive pathology

Infertility, early pregnancy loss and complications

Aberrant implantation can cause a variety of clinical problems including recurrent miscarriage, intrauterine growth retardation and preeclampsia. The cause of recurrent miscarriage is multifactorial; known causes of maternal defects include coagulation disorders, autoimmune defects and endometrial defects influencing the production of pro-inflammatory cytokines (Laird *et al*. 2003). Pro-inflammatory cytokines IL6, LIF and IL1B are decreased in the endometrium of women with recurrent miscarriage compared with fertile women (von Wolff *et al*. 2000). Impaired endovascular trophoblast invasion is the primary placental defect causing inadequate conversion of the uterine arteries and reduced uteroplacental blood flow, which leads to fetal intrauterine growth restriction and the development of preeclampsia. In addition to endothelial dysfunction, there is evidence of systemic activation of maternal inflammatory cell responses in preeclampsia. There is also increased release of pro-inflammatory cytokines TNF, IL6,

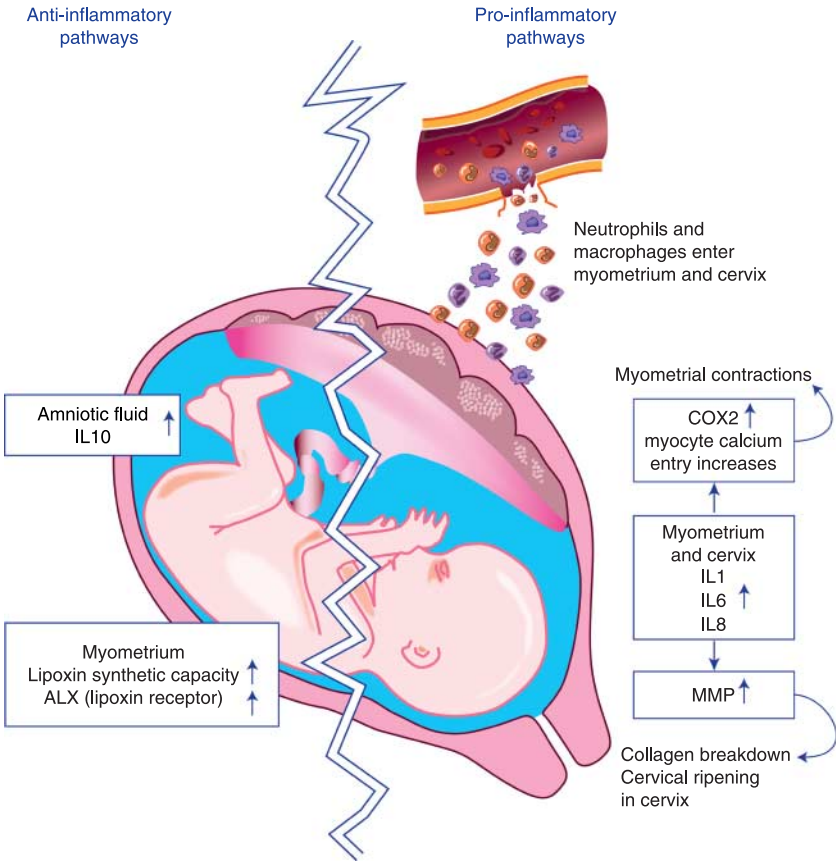

Figure 3 Schematic diagram of the pro- and anti-inflammatory pathways operating in labour. Pro-inflammatory pathways are shown on the right hand side of the diagram. Parturition is associated with the influx of leukocytes (principally neutrophils and macrophages) into the myometrium and cervix during parturition. These cells release cytokines such as interleukin (IL)1, IL6 and IL8. These cytokines upregulate matrix metalloproteinase (MMP) expression and activity in the cervix, causing collagen breakdown and cervical ripening. In the myometrium, IL1 and possibly the other cytokines increase the expression of cyclo-oxygenase (COX)2, leading to increased prostaglandin production. This, together with IL1's activity in increasing calcium entry into myometrial cells will stimulate myometrial contraction. In parallel with these pro-inflammatory events, anti-inflammatory events are also upregulated in labour. Specifically, amniotic fluid IL10 production is increased. In the myometrium, synthetic capacity for the lipoxin pathway and its receptor (ALX) is also upregulated during labour. The upregulation of the anti-inflammatory pathway likely acts as a counterbalance to the pro-inflammatory pathway during the process of parturition. Inappropriate early activation of the pro-inflammatory pathway may initiate preterm labour, and thus strategies to upregulate the anti-inflammatory pathway are of interest in prevention or treatment of preterm birth.

soluble phospholipase A_2 (a mediator of inflammatory reactions) and PTX3 into the circulation-associated preeclampsia (Redman *et al.* 1999, Rovere-Querini *et al.* 2006, Assi *et al.* 2007). Dysregulation of anti-inflammatory mediators has also been reported in these pathologies. IL10 levels are elevated in amniotic fluid at mid-trimester in women with intrauterine growth restriction and elevated in term placenta in women with preeclampsia (Heyborne *et al.* 1994, Rinehart *et al.* 1999). IL10 has also been reported to be aberrantly expressed in decidual T lymphocytes in women with recurrent miscarriage (Piccinni *et al.* 1998). A direct cause-and-effect relationship between a local defect of Th2-type cytokine expression and pregnancy loss has been reported (Piccinni *et al.* 2001). TGFB can inhibit Th1-type responses, which may be detrimental to pregnancy, in addition it is an important regulator of NK cells, down-regulating IFN-γ-induced activation and inflammatory cytokine production. Thus, TGFB actions during implantation are instrumental in the establishment of anti-rejection pathways to embryo survival (Jones *et al.* 2006). Women with high numbers of circulating NK cells have a higher risk of miscarriage as increased infiltration into the decidua of these blood-type NK cells, which express IFN-γ, are known to cause abortion. It has been shown that abortions are probably caused by IFN-γ activating the production of prothrombinase FGL2 in trophoblast and in decidua (Clark *et al.* 2001*a*). This procoagulant leads to fibrin deposition and activation of polymorphonuclear leukocytes that destroy the vascular supply to the placenta (Clark *et al.* 2001*b*).

Recent reports suggest that dysregulation of inflammatory factors play a role in endometriosis-associated reproductive failure (Gupta *et al.* 2008). Endometriosis is a disorder characterised by the proliferation of endometrial tissue outside the uterine cavity following retrograde menstruation of endometrial tissue into the peritoneal cavity. The concentration of inflammatory cytokines (IL1B and TNF) and PGs (PGE_2 and $PGF_{2\alpha}$) produced by peritoneal macrophages (Karck *et al.* 1996) and pro-inflammatory chemokines for monocyte/macrophages (CCL2 and CCL5) and for granulocytes (IL8 and CXCL1) is elevated in women with endometriosis (Ryan *et al.* 1995, Arici *et al.* 1997), although it is unclear whether these pro-inflammatory changes precede or follow endometriosis. Dysregulated production of anti-inflammatory mediators has also has been reported to have an impact on female fertility (Mitchell *et al.* 2005) and to be decreased in the serum of women with endometriosis such as adiponectin (Takemura *et al.* 2005).

Menstrual disorders

Menstrual cycle disorders attributed to a dysfunctional endometrium, include dysmenorrhoea and heavy menstrual blood loss. Dysmenorrhoea is characterised by severe uterine pain during menstruation, and heavy menstrual blood loss is characterised by an abnormally heavy and/or prolonged menstrual period. Primary dysmenorrhoea and heavy menstrual blood loss are diagnosed when symptoms cannot be attributable to other underlying disease, disorder or structural abnormality in the uterus. The role of inflammatory mediators in these pathologies is not well documented except the contribution of aberrant PG production, which has been well reported. Locally produced PGs are elevated prior to menstruation and are considered primary mediators of aberrant menstruation (Sales & Jabbour 2003*a*, Smith *et al*. 2007). Increased PGE_2 relative to $PGF_{2\alpha}$ levels in endometrium and menstrual fluid have been associated with heavy menstrual blood loss, and altered PGI_2 and TXA_2 in the spiral arteries may also contribute to this condition (Lumsden *et al*. 1983). Dysmenorrhoea is associated with uterine hypercontractility resulting in episodes of reduced endometrial blood flow leading to ischaemia and increased pain (Rees 1989). This observation may be a consequence of increased $PGF_{2\alpha}$ production as explants from women with dysmenorrhoea produce more $PGF_{2\alpha}$ in response to arachidonic acid compared with normal endometrial explants (Lundstrom & Green 1978). Further evidence of the role of PGs comes from the administration of COX enzyme inhibitors such as ibuprofen, which have been demonstrated to reduce menstrual blood flow (Makarainen & Ylikorkala 1986) and selective COX2 inhibitors that have been used in the treatment of dysmenorrhoea and heavy menstrual blood loss (Daniels *et al*. 2002).

Complicated labour

The major complication of parturition is preterm labour. Preterm delivery rates in the United Kingdom are in the order of 8%, with over 70% of preterm deliveries following spontaneous preterm labour. The role of infection and inflammation within the amniotic cavity in preterm parturition has been extensively examined with good evidence that the prevalence of infection and/or inflammation is greater the earlier in gestation that preterm labour occurs (i.e. the prevalence of infection and/or inflammation is greater in preterm labour at 28 weeks compared with 34 weeks gestation; Goldenberg *et al*. 2000, Shim *et al*. 2004). Importantly, the likelihood both of preterm delivery and of poor neonatal outcome is greater in the presence of intrauterine inflammation (Shim *et al*. 2004). Fewer studies have examined cervical and myometrial tissues from women in preterm labour, although those that have suggested that inflammatory processes operate here also (Tornblom *et al*. 2004, Osman *et al*. 2006).

The importance of intrauterine inflammation in preterm labour is not confined to the initiation of fetal membrane rupture, cervical ripening, myometrial contractions and preterm delivery. One of the major adverse consequences of infection/inflammation-associated preterm delivery is neonatal brain injury, which manifests as white matter damage. A seminal study in 1997 showed that vaginal inoculation of pregnant rabbits with *Escherichia coli* induced white matter damage in the fetus within 5 days (Yoon *et al*. 1997*b*). In human pregnancy, the same group also showed that periventricular white matter injury was commoner in babies whose mothers had high cytokine levels in amniotic fluid when sampled by amniocentesis prior to delivery (Yoon *et al*. 1997*a*). A clear causal link between non-infective intrauterine inflammation and fetal brain inflammation was shown more recently by Elovitz *et al*. (2006), with the demonstration that intrauterine lipopolysaccharide (LPS) administration in the pregnant mouse stimulates pro-inflammatory cytokine production in the fetal brain.

A key question is whether understanding of these inflammatory pathways can be exploited therapeutically. It has been known for some time that IL1 receptor antagonists can inhibit IL1-induced preterm labour (Romero & Tartakovsky 1992). More recently, exciting data from animal studies have suggested that administration of 'anti-inflammatory' agents could avert not only preterm delivery, but also the risk of neonatal brain damage resulting from exposure to intrauterine inflammation. The first set of these studies focused on IL10 in a preterm labour model induced by intrauterine LPS administration. IL10 administered either on the day of LPS administration or delayed by 24 hours completely abolished preterm delivery (Terrone *et al*. 2001). Further studies by the same group in a rat *E. coli* preterm labour model showed that IL10 also prevented infection-induced white matter injury (Rodts-Palenik *et al*. 2004). More recently, Bennett *et al*. have shown that 15-deoxy-12,14-prostaglandin J2 (15d-PGJ2, an agent which inhibits NFKB and possibly also JNK) averts LPS-induced preterm labour in pregnant mice and LPS-induced NFKB activation in the brain of mouse pups (Pirianov *et al*. 2009).

Although these agents show great promise, they are at present only being trialled in animal models. Clinically, progesterone is the only agent shown to be effective in preventing preterm birth with efficacy proven for selected groups (Dodd *et al*. 2008). The mechanism of action of progesterone is unknown, although there is some evidence *in vitro* of an acute inhibitory effect on myometrial contractions (Ruddock *et al*. 2008). Progestogens may also act to inhibit inflammation, with inhibition of LPS-induced inflammation in human fetoplacental arteries and in myometrium (Gotkin *et al*. 2006) and inhibition of a

physiological rise in mouse myometrial CCL2 *in vivo* (Shynlova *et al*. 2008).

Although there has been little measurement of anti-inflammatory pathways in preterm pathological parturition, it is tempting to speculate that upregulation thereof could be a useful therapeutic strategy to prevent preterm delivery.

Reproductive tract cancers

The relationship between inflammation and cancer dates back to the 19th century, when Virchow first hypothesised that the origin of cancer was at sites of chronic inflammation (Coussens & Werb 2002). Ovarian, uterine and cervical cancers, which arise mainly from the OSE, endometrium and glandular and squamous epithelium of the cervix, are the most common gynaecological malignancies (Forman *et al*. 2003). The incidence of ovarian, uterine and cervical cancer as of 2005 in the United Kingdom are reported to be 17.4, 17.9 and 8.4 women per 100 000 with a mortality rate of 10.1, 3.5 and 2.4 women per 100 000.

It is now well established that infection of the cervix with human papillomavirus (HPV) is the main cause of cervical cancer in women (zur Hausen 2009). However, the aetiology of ovarian and endometrial cancers is multifactorial and less well-defined. In the case of ovarian cancer, it is considered that the inflammatory environment caused by repetitive ovulation over the life time of a women increases the risk of genetic error and mutation during the repair process (Fleming *et al*. 2006), leaving the OSE susceptible to neoplastic transformation in subsequent ovulation and repair cycles. Epidemiological and experimental observations have implicated hormonal fluctuations in sex steroids, and androgen exposure in particular, in the pathogenesis of ovarian cancer; however, the mechanism of actions of steroid hormones in the regulation of ovarian cancer remains unclear (Auersperg *et al*. 2001). Genetic mutations in the *BRCA1* and *BRCA2* genes have also been implicated in the pathogenesis of the disease as they have been associated with a genetically inherited incidence of ovarian cancer (Fleming *et al*. 2006). Endometrial adenocarcinomas on the other hand are associated mainly with post-menopausal women and are thought to arise from excessive E_2 exposure in the setting of endometrial hyperplasia (Persson 2000), although other factors including polycystic ovarian syndrome and obesity are also thought to play a role.

The interplay between inflammation and cancer has been extensively reviewed (Sporn & Roberts 1986, Coussens & Werb 2002, Modugno *et al*. 2005, Goswami *et al*. 2008). In the context of the gynaecological malignancies, inflammation can contribute to the initiation and progression of disease via the release of local mediators, ILs, growth factors and cytokines, to facilitate immune cell recruitment, cell proliferation and angiogenesis and sustain tumour growth. Tissue damage, be it is post-ovulatory damage or damage by chemical carcinogens or viral infection agents can cause activation of the coagulation cascade, as described earlier, and there is now much evidence in support of the coagulation cascade in mediating tumour cell adhesive spreading. For example, TF is increased in cell lines containing inactivating mutations of p53 and PTEN (Boccaccio & Medico 2006) and is thought to enhance tumour metastasis directly by enhancing cell motility as its extracellular domain interacts with several integrins (Belting *et al*. 2005). TF activation of PAR1/integrin $\alpha v\beta 5$ signalling via thrombin can enhance cell motility and metastasis (Belting *et al*. 2005). Furthermore, the TF cytoplasmic domain has been shown to negatively regulate integrin $\alpha 3\beta 1$, which mediates metastatic arrest (Belting *et al*. 2005). Dysregulation of TF phosphorylation and upregulation of PAR1 is thought to contribute to the aggressive behaviour of some cancer cells (Belting *et al*. 2005). The kallikrein–kinin system of proteinase-mediated signalling has also been implicated in cancer. For example, a link has been made between PAR1 receptor expression and mammary tumour cell metastasis and invasion (Hollenberg *et al*. 2008); however the role of these pathways in ovarian, endometrial and cervical cancers needs to be investigated.

We and others have shown that the inflammatory COX–PG axis is elevated in ovarian, endometrial and cervical cancers (Ryu *et al*. 2000, Tong *et al*. 2000, Jabbour *et al*. 2001, Sales *et al*. 2001, 2002, Gupta *et al*. 2003, Sales & Jabbour 2003*a*, 2003*b*, Daikoku *et al*. 2005, Munkarah & Ali-Fehmi 2005, Khunamornpong *et al*. 2009). This pro-inflammatory pathway can be induced by a variety of stimuli, including LPS, cytokines, growth factors and tumour-promoting chemical carcinogens (Modugno *et al*. 2005, Goswami *et al*. 2008). In cervical cancers, a recent study has shown that HPV infection induces expression of COX2 (Subbaramaiah & Dannenberg 2007). Furthermore, PGE_2 has been shown to regulate hormone-dependent diseases of the endometrium by upregulating aromatase expression and local E_2 production, which can in turn upregulate COX enzyme expression (Bulun *et al*. 2000). Elevated PG biosynthesis, as a consequence of elevated COX enzyme expression in ovarian, endometrial and cervical epithelial cells, can promote and sustain tumourigenesis via the activation of specific prostanoid GPCRs and second messenger systems to enhance the expression and delivery of potent growth factors, cytokines and chemokines (Fig. 2; Sales & Jabbour 2003*a*, 2003*b*, Jabbour & Sales 2004, Jabbour *et al*. 2006, Goswami *et al*. 2008). These local mediators enhance the recruitment of immune cells, inhibit apoptosis and enhance cell proliferation, tumour angiogenesis and promote cell migration and metastasis (Fig. 2).

This has led us and other investigators to speculate that inhibition of the inflammatory COX–PG axis could be of

therapeutic relevance for women with gynaecological malignancies. Indeed, the risk of cancer and epithelial ovarian cancer in particular in women on NSAID treatment for at least 6 months is reduced (Fleming *et al*. 2006). This is due to the inhibition of COX enzyme activity and suppression of transcriptional transactivators such as NFKB, which leads to a reduction in the expression of local mediators, such as pro-inflammatory cytokines, growth factors and ILs (Fleming *et al*. 2006, Goswami *et al*. 2008). LPA, which is present in follicular fluid and elevated in ascites from patients with ovarian, endometrial and cervical cancer, is another bioactive lipid that has recently been shown to play a role in reproductive tract pathology. LPA acting via its specific GPCRs has been shown to promote cellular proliferation, growth, migration and survival of ovarian, endometrial and cervical cancer cells by inducing local expression of growth factors and cytokines such as IL6 and IL8 (Fig. 2; Ye 2008). It is anticipated that the activation of the LPA system in parallel to the COX–PG system could enhance the inflammation in reproductive pathologies associated with aberrant expression of LPA or PG receptors (Fig. 2).

Little is known about the role of anti-inflammatory cytokines and lipids in reproductive tract cancer. It would be tempting to speculate that their expression, synthesis and function are suppressed to maintain an exacerbated inflammatory environment conducive for growth and metastases of these cancers. Future work is warranted to address the role for such molecules in female reproductive cancers and their potential exploitation for therapeutic intervention.

Conclusions

It is well accepted now that reproductive processes are regulated by inflammatory events. Tight control of the onset and resolution of these inflammatory events ensures normal reproductive function. Exacerbated or premature activation of inflammation can contribute to disease. Understanding the molecular control of inflammation and its resolution in the reproductive tract may give us insight into how these may be corrected therapeutically in disease.

Declaration of interest

H N Jabbour is a named inventor on several patents for the treatment of endometrial pathologies or preterm labour. J E Norman is a named inventor on a patent for the treatment of preterm labour.

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References

Allport VC, Pieber D, Slater DM, Newton R, White JO & Bennett PR 2001 Human labour is associated with nuclear factor-kappaB activity which mediates cyclo-oxygenase-2 expression and is involved with the 'functional progesterone withdrawal'. *Molecular Human Reproduction* **7** 581–586.

Amjad AI, Soder O & Sultana T 2006 Role of testicular interleukin-1alpha tIL-1alpha in testicular physiology and disease. *Journal of College of Physicians and Surgeons Pakistan* **16** 55–60.

Arck P, Hansen PJ, Mulac Jericevic B, Piccinni MP & Szekeres-Bartho J 2007 Progesterone during pregnancy: endocrine-immune cross talk in mammalian species and the role of stress. *American Journal of Reproductive Immunology* **58** 268–279.

Arici A, Oral E, Attar E, Tazuke SI & Olive DL 1997 Monocyte chemotactic protein-1 concentration in peritoneal fluid of women with endometriosis and its modulation of expression in mesothelial cells. *Fertility and Sterility* **67** 1065–1072.

Arici A, Seli E, Senturk LM, Gutierrez LS, Oral E & Taylor HS 1998*a* Interleukin-8 in the human endometrium. *Journal of Clinical Endocrinology and Metabolism* **83** 1783–1787.

Arici A, Seli E, Zeyneloglu HB, Senturk LM, Oral E & Olive DL 1998*b* Interleukin-8 induces proliferation of endometrial stromal cells: a potential autocrine growth factor. *Journal of Clinical Endocrinology and Metabolism* **83** 1201–1205.

Assi F, Fruscio R, Bonardi C, Ghidini A, Allavena P, Mantovani A & Locatelli A 2007 Pentraxin 3 in plasma and vaginal fluid in women with preterm delivery. *British Journal of Obstetrics and Gynaecology* **114** 143–147.

Auersperg N, Wong AS, Choi KC, Kang SK & Leung PC 2001 Ovarian surface epithelium: biology, endocrinology, and pathology. *Endocrine Reviews* **22** 255–288.

Baird DT, Cameron ST, Critchley HO, Drudy TA, Howe A, Jones RL, Lea RG & Kelly RW 1996 Prostaglandins and menstruation. *European Journal of Obstetrics, Gynecology, and Reproductive Biology* **70** 15–17.

Belting M, Ahamed J & Ruf W 2005 Signaling of the tissue factor coagulation pathway in angiogenesis and cancer. *Arteriosclerosis, Thrombosis, and Vascular Biology* **25** 1545–1550.

Boccaccio C & Medico E 2006 Cancer and blood coagulation. *Cellular and Molecular Life Sciences* **63** 1024–1027.

Bollopragada S, Youssef R, Jordan F, Greer I, Norman J & Nelson S 2009 Term labor is associated with a core inflammatory response in human fetal membranes, myometrium, and cervix. *American Journal of Obstetrics and Gynecology* **200** e101–e111.

Boyington JC, Brooks AG & Sun PD 2001 Structure of killer cell immunoglobulin-like receptors and their recognition of the class I MHC molecules. *Immunological Reviews* **181** 66–78.

Brakenhielm E, Veitonmaki N, Cao R, Kihara S, Matsuzawa Y, Zhivotovsky B, Funahashi T & Cao Y 2004 Adiponectin-induced antiangiogenesis and antitumor activity involve caspase-mediated endothelial cell apoptosis. *PNAS* **101** 2476–2481.

Bukulmez O & Arici A 2000 Leukocytes in ovarian function. *Human Reproduction Update* **6** 1–15.

Bulun SE, Zeitoun KM, Takayama K, Simpson E & Sasano H 2000 Aromatase as a therapeutic target in endometriosis. *Trends in Endocrinology and Metabolism* **11** 22–27.

Calixto JB, Cabrini DA, Ferreira J & Campos MM 2001 Inflammatory pain: kinins and antagonists. *Current Opinion in Anaesthesiology* **14** 519–526.

Calixto JB, Medeiros R, Fernandes ES, Ferreira J, Cabrini DA & Campos MM 2004 Kinin B1 receptors: key G-protein-coupled receptors and their role in inflammatory and painful processes. *British Journal of Pharmacology* **143** 803–818.

Campos MM, Leal PC, Yunes RA & Calixto JB 2006 Non-peptide antagonists for kinin B1 receptors: new insights into their therapeutic potential for the management of inflammation and pain. *Trends in Pharmacological Sciences* **27** 646–651.

Casslen B, Nordengren J, Gustavsson B, Nilbert M & Lund LR 1995 Progesterone stimulates degradation of urokinase plasminogen activator (u-PA) in endometrial stromal cells by increasing its inhibitor and surface expression of the u-PA receptor. *Journal of Clinical Endocrinology and Metabolism* **80** 2776–2784.

ten Cate H, Schenk BE, Biemond BJ, Levi M, van der Poll T, Buller HR & ten Cate JW 1994 A review of studies of the activation of the blood coagulation mechanism in chimpanzees (*Pan troglodytes*). *Journal of Medical Primatology* **23** 280–284.

Challis JR, Lockwood CJ, Myatt L, Norman JE, Strauss JF III & Petraglia F 2009 Inflammation and pregnancy. *Reproductive Sciences* **16** 206–215.

Cheng JG & Stewart CL 2003 Loss of cyclooxygenase-2 retards decidual growth but does not inhibit embryo implantation or development to term. *Biology of Reproduction* **68** 401–404.

Clark DA, Coulam CB, Daya S & Chaouat G 2001*a* Unexplained sporadic and recurrent miscarrage in the new millennium: a critical analysis of immune mechanisms and treatments. *Human Reproduction Update* **7** 501–511.

Clark DA, Yu G, Levy GA & Gorczynski RM 2001*b* Procoagulants in fetus rejection: the role of the OX-2 (CD200) tolerance signal. *Seminars in Immunology* **13** 255–263.

Colditz IG 1990 Effect of exogenous prostaglandin E_2 and actinomycin D on plasma leakage induced by neutrophil-activating peptide-1/interleukin-8. *Immunology and Cell Biology* **68** 397–403.

Coleman RA, Smith WL & Narumiya S 1994 International Union of Pharmacology classification of prostanoid receptors: properties, distribution, and structure of the receptors and their subtypes. *Pharmacological Reviews* **46** 205–229.

Coussens LM & Werb Z 2002 Inflammation and cancer. *Nature* **420** 860–867.

Critchley HO, Kelly RW, Brenner RM & Baird DT 2001 The endocrinology of menstruation – a role for the immune system. *Clinical Endocrinology* **55** 701–710.

Daikoku T, Wang D, Tranguch S, Morrow JD, Orsulic S, DuBois RN & Dey SK 2005 Cyclooxygenase-1 is a potential target for prevention and treatment of ovarian epithelial cancer. *Cancer Research* **65** 3735–3744.

Daniels SE, Talwalker S, Torri S, Snabes MC, Recker DP & Verburg KM 2002 Valdecoxib, a cyclooxygenase-2-specific inhibitor, is effective in treating primary dysmenorrhea. *Obstetrics and Gynecology* **100** 350–358.

Dodd JM, Flenady VJ, Cincotta R & Crowther CA 2008 Progesterone for the prevention of preterm birth: a systematic review. *Obstetrics and Gynecology* **112** 127–134.

Elovitz MA, Mrinalini C & Sammel MD 2006 Elucidating the early signal transduction pathways leading to fetal brain injury in preterm birth. *Pediatric Research* **59** 50–55.

Espey LL 1980 Ovulation as an inflammatory reaction – a hypothesis. *Biology of Reproduction* **22** 73–106.

Espey LL 1994 Current status of the hypothesis that mammalian ovulation is comparable to an inflammatory reaction. *Biology of Reproduction* **50** 233–238.

Evans J, Catalano RD, Brown P, Sherwin R, Critchley HO, Fazleabas AT & Jabbour HN 2009 Prokineticin 1 mediates fetal–maternal dialogue regulating endometrial leukemia inhibitory factor. *FASEB Journal* **23** 2165–2175.

Feldmann M 2008 Many cytokines are very useful therapeutic targets in disease. *Journal of Clinical Investigation* **118** 3533–3536.

Fleming JS, Beaugie CR, Haviv I, Chenevix-Trench G & Tan OL 2006 Incessant ovulation, inflammation and epithelial ovarian carcinogenesis: revisiting old hypotheses. *Molecular and Cellular Endocrinology* **247** 4–21.

Forman D, Stockton D, Moller H, Quinn M, Babb P, De Angelis R & Micheli A 2003 Cancer prevalence in the UK: results from the EUROPREVAL study. *Annals of Oncology* **14** 648–654.

Garlanda C, Maina V, Martinez de la Torre Y, Nebuloni M & Locati M 2008 Inflammatory reaction and implantation: the new entries PTX3 and D6. *Placenta* **29** 129–134.

Gimbrone MA, Obin MS, Brock AF, Luis EA, Hass PE, Hebert CA, Yip YK, Leung DW, Lowe DG, Kohr WJ *et al.* 1989 Endothelial interleukin-8 – a novel inhibitor of leukocyte–endothelial interactions. *Science* **246** 1601–1603.

Goldenberg R, Hauth J & Andrews W 2000 Intrauterine infection and preterm delivery. *New England Journal of Medicine* **342** 1500–1507.

Goldstein BJ & Scalia R 2004 Adiponectin: a novel adipokine linking adipocytes and vascular function. *Journal of Clinical Endocrinology and Metabolism* **89** 2563–2568.

Gonzalez RR, Rueda BR, Ramos MP, Littell RD, Glasser S & Leavis PC 2004 Leptin-induced increase in leukemia inhibitory factor and its receptor by human endometrium is partially mediated by interleukin 1 receptor signaling. *Endocrinology* **145** 3850–3857.

Goswami B, Rajappa M, Sharma M & Sharma A 2008 Inflammation: its role and interplay in the development of cancer, with special focus on gynecological malignancies. *International Journal of Gynecological Cancer* **18** 591–599.

Gotkin JL, Celver J, McNutt P, Shields AD, Howard BC, Paonessa DJ & Napolitano PG 2006 Progesterone reduces lipopolysaccharide induced interleukin-6 secretion in fetoplacental chorionic arteries, fractionated cord blood, and maternal mononuclear cells. *American Journal of Obstetrics and Gynecology* **195** 1015–1019.

Gotsch F, Romero R, Kusanovic JP, Erez O, Espinoza J, Kim CJ, Vaisbuch E, Than NG, Mazaki-Tovi S, Chaiworapongsa T *et al.* 2008 The anti-inflammatory limb of the immune response in preterm labor, intra-amniotic infection/inflammation, and spontaneous parturition at term: a role for interleukin-10. *Journal of Maternal–Fetal and Neonatal Medicine* **21** 529–547.

Guazzone VA, Jacobo P, Theas MS & Lustig L 2009 Cytokines and chemokines in testicular inflammation: a brief review. *Microscopy and Research Technique* **72** 620–628.

Gupta RA, Tejada LV, Tong BJ, Das SK, Morrow J, Dey SK & DuBois RN 2003 Cyclooxygenase-1 is overexpressed and promotes angiogenic growth factor production in ovarian cancer. *Cancer Research* **63** 906–911.

Gupta S, Goldberg JM, Aziz N, Goldberg E, Krajcir N & Agarwal A 2008 Pathogenic mechanisms in endometriosis-associated infertility. *Fertility and Sterility* **90** 247–257.

Haddad R, Tromp G, Kuivaniemi H, Chaiworapongsa T, Kim YM, Mazor M & Romero R 2006 Human spontaneous labor without histologic chorioamnionitis is characterized by an acute inflammation gene expression signature. *American Journal of Obstetrics and Gynecology* **195** e391–e324.

Hamilton GS & Kennedy TG 1994 Uterine vascular changes after unilateral intrauterine infusion of indomethacin and prostaglandin E_2 to rats sensitized for the decidual cell reaction. *Biology of Reproduction* **50** 757–764.

Hanna N, Hanna I, Hleb M, Wagner E, Dougherty J, Balkundi D, Padbury J & Sharma S 2000 Gestational age-dependent expression of IL-10 and its receptor in human placental tissues and isolated cytotrophoblasts. *Journal of Immunology* **164** 5721–5728.

zur Hausen H 2009 Papillomaviruses in the causation of human cancers – a brief historical account. *Virology* **384** 260–265.

Hess AP, Hamilton AE, Talbi S, Dosiou C, Nyegaard M, Nayak N, Genbecev-Krtolica O, Mavrogianis P, Ferrer K, Kruessel J *et al.* 2007 Decidual stromal cell response to paracrine signals from the trophoblast: amplification of immune and angiogenic modulators. *Biology of Reproduction* **76** 102–117.

Heyborne KD, McGregor JA, Henry G, Witkin SS & Abrams JS 1994 Interleukin-10 in amniotic fluid at midtrimester: immune activation and suppression in relation to fetal growth. *American Journal of Obstetrics and Gynecology* **171** 55–59.

Hollenberg MD, Oikonomopoulou K, Hansen KK, Saifeddine M, Ramachandran R & Diamandis EP 2008 Kallikreins and proteinase-mediated signaling: proteinase-activated receptors (PARs) and the pathophysiology of inflammatory diseases and cancer. *Biological Chemistry* **389** 643–651.

Jabbour HN & Sales KJ 2004 Prostaglandin receptor signalling and function in human endometrial pathology. *Trends in Endocrinology and Metabolism* **15** 398–404.

Jabbour HN, Milne SA, Williams ARW, Anderson RA & Boddy SC 2001 Expression of COX-2 and PGE synthase and synthesis of PGE_2 in endometrial adenocarcinoma: a possible autocrine/paracrine regulation of neoplastic cell function via EP2/EP4 receptors. *British Journal of Cancer* **85** 1023–1031.

Jabbour HN, Sales KJ, Smith OP, Battersby S & Boddy SC 2006 Prostaglandin receptors are mediators of vascular function in endometrial pathologies. *Molecular and Cellular Endocrinology* **252** 191–200.

Jasper MJ, Tremellen KP & Robertson SA 2007 Reduced expression of IL-6 and IL-1alpha mRNAs in secretory phase endometrium of women with recurrent miscarriage. *Journal of Reproductive Immunology* **73** 74–84.

Jolicoeur C, Boutouil M, Drouin R, Paradis I, Lemay A & Akoum A 1998 Increased expression of monocyte chemotactic protein-1 in the endometrium of women with endometriosis. *American Journal of Pathology* **152** 125–133.

Jones RL, Hannan NJ, Kaitu'u TJ, Zhang J & Salamonsen LA 2004 Identification of chemokines important for leukocyte recruitment to the human endometrium at the times of embryo implantation and menstruation. *Journal of Clinical Endocrinology and Metabolism* **89** 6155–6167.

Jones RL, Stoikos C, Findlay JK & Salamonsen LA 2006 TGF-β superfamily expression and actions in the endometrium and placenta. *Reproduction* **132** 217–232.

Kane N, Kelly R, Saunders PT & Critchley HO 2009 Proliferation of uterine natural killer cells is induced by human chorionic gonadotropin and mediated via the mannose receptor. *Endocrinology* **150** 2882–2888.

Karck U, Reister F, Schafer W, Zahradnik HP & Breckwoldt M 1996 PGE_2 and $PGF_{2\alpha}$ release by human peritoneal macrophages in endometriosis. *Prostaglandins* **51** 49–60.

Kayisli UA, Mahutte NG & Arici A 2002 Uterine chemokines in reproductive physiology and pathology. *American Journal of Reproductive Immunology* **47** 213–221.

Kayisli UA, Guzeloglu-Kayisli O & Arici A 2004 Endocrine–immune interactions in human endometrium. *Annals of the New York Academy of Sciences* **1034** 50–63.

Kelly RW, King AE & Critchley HO 2001 Cytokine control in human endometrium. *Reproduction* **121** 3–19.

Kennedy TG, Gillio-Meina C & Phang SH 2007 Prostaglandins and the initiation of blastocyst implantation and decidualization. *Reproduction* **134** 635–643.

Khunamornpong S, Settakorn J, Sukpan K, Srisomboon J, Ruangvejvorachai P, Thorner PS & Siriaunkgul S 2009 Cyclo-oxygenase-2 expression in squamous cell carcinoma of the uterine cervix is associated with lymph node metastasis. *Gynecologic Oncology* **112** 241–247.

Kimber SJ 2005 Leukaemia inhibitory factor in implantation and uterine biology. *Reproduction* **130** 131–145.

Laird SM, Tuckerman EM, Cork BA, Linjawi S, Blakemore AI & Li TC 2003 A review of immune cells and molecules in women with recurrent miscarriage. *Human Reproduction Update* **9** 163–174.

Larhammar D 1996 Evolution of neuropeptide Y, peptide YY and pancreatic polypeptide. *Regulatory Peptides* **62** 1–11.

Lea RG & Sandra O 2007 Immunoendocrine aspects of endometrial function and implantation. *Reproduction* **134** 389–404.

Ledingham M, Thomson A, Macara L, Young A, Greer I & Norman J 2000 Changes in the expression of nitric oxide synthase in the human uterine cervix during pregnancy and parturition. *Molecular Human Reproduction* **6** 1041–1048.

Ledingham M, Thomson A, Jordan F, Young A, Crawford M & Norman J 2001 Cell adhesion molecule expression in cervix and myometrium during pregnancy and parturition. *Obstetrics and Gynecology* **97** 235–242.

Levi M & van der Poll T 2005 Two-way interactions between inflammation and coagulation. *Trends in Cardiovascular Medicine* **15** 254–259.

Lockwood CJ, Nemerson Y, Krikun G, Hausknecht V, Markiewicz L, Alvarez M, Guller S & Schatz F 1993 Steroid-modulated stromal cell tissue factor expression: a model for the regulation of endometrial hemostasis and menstruation. *Journal of Clinical Endocrinology and Metabolism* **77** 1014–1019.

Loftin CD, Tiano HF & Langenbach R 2002 Phenotypes of the COX-deficient mice indicate physiological and pathophysiological roles for COX-1 and COX-2. *Prostaglandins & Other Lipid Mediators* **68–69** 177–185.

Lumsden MA, Kelly RW & Baird DT 1983 Primary dysmenorrhoea: the importance of both prostaglandins E_2 and $F_{2\alpha}$. *British Journal of Obstetrics and Gynaecology* **90** 1135–1140.

Lundstrom V & Green K 1978 Endogenous levels of prostaglandin $F_{2\alpha}$ and its main metabolites in plasma and endometrium of normal and dysmenorrheic women. *American Journal of Obstetrics and Gynecology* **130** 640–646.

Maeda K, Okubo K, Shimomura I, Funahashi T, Matsuzawa Y & Matsubara K 1996 cDNA cloning and expression of a novel adipose specific collagen-like factor, apM1 (AdiPose Most abundant Gene transcript 1). *Biochemical and Biophysical Research Communications* **221** 286–289.

Makarainen L & Ylikorkala O 1986 Primary and myoma-associated menorrhagia: role of prostaglandins and effects of ibuprofen. *British Journal of Obstetrics and Gynaecology* **93** 974–978.

Maldonado-Perez D, Jabbour HN, Denison FC & Norman JE 2009 Role of anti-Inflammatory/pro-resolution lipid mediators in myometrium during human labor. *Proceedings of the Society for Gynecological Investigation*, 56th Annual Meeting, Glasgow, United Kingdom, p 475.

Mills GB & Moolenaar WH 2003 The emerging role of lysophosphatidic acid in cancer. *Nature Reviews. Cancer* **3** 582–591.

Milne SA & Jabbour HN 2003 Prostaglandin (PG) F(2alpha) receptor expression and signaling in human endometrium: role of PGF(2alpha) in epithelial cell proliferation. *Journal of Clinical Endocrinology and Metabolism* **88** 1825–1832.

Mitchell M, Armstrong DT, Robker RL & Norman RJ 2005 Adipokines: implications for female fertility and obesity. *Reproduction* **130** 583–597.

Modugno F, Ness RB, Chen C & Weiss NS 2005 Inflammation and endometrial cancer: a hypothesis. *Cancer Epidemiology, Biomarkers & Prevention* **14** 2840–2847.

Moffett-King A 2002 Natural killer cells and pregnancy. *Nature Reviews. Immunology* **2** 656–663.

Moore KW, de Waal Malefyt R, Coffman RL & O'Garra A 2001 Interleukin-10 and the interleukin-10 receptor. *Annual Review of Immunology* **19** 683–765.

van Mourik MS, Macklon NS & Heijnen CJ 2009 Embryonic implantation: cytokines, adhesion molecules, and immune cells in establishing an implantation environment. *Journal of Leukocyte Biology* **85** 4–19.

Munkarah A & Ali-Fehmi R 2005 COX-2: a protein with an active role in gynecological cancers. *Current Opinion in Obstetrics and Gynecology* **17** 49–53.

Murakami M & Kudo I 2004 Recent advances in molecular biology and physiology of the prostaglandin E_2 – biosynthetic pathway. *Progress in Lipid Research* **43** 3–35.

Nakamura K, Sheps S & Arck PC 2008 Stress and reproductive failure: past notions, present insights and future directions. *Journal of Assisted Reproduction and Genetics* **25** 47–62.

Nathan C 2006 Neutrophils and immunity: challenges and opportunities. *Nature Reviews. Immunology* **6** 173–182.

Nepomnaschy PA, Sheiner E, Mastorakos G & Arck PC 2007 Stress, immune function, and women's reproduction. *Annals of the New York Academy of Sciences* **1113** 350–364.

Norman JE, Wu WX, Kelly RW, Glasier AF, McNeilly AS & Baird DT 1991 Effects of mifepristone *in vivo* on decidual prostaglandin synthesis and metabolism. *Contraception* **44** 89–98.

Oger S, Mehats C, Dallot E, Ferre F & Leroy MJ 2002 Interleukin-1β induces phosphodiesterase 4B2 expression in human myometrial cells through a prostaglandin E_2- and cyclic adenosine 3′,5′-monophosphate-dependent pathway. *Journal of Clinical Endocrinology and Metabolism* **87** 5524–5531.

Osman I, Young A, Ledingham M, Thomson A, Jordan F, Greer I & Norman J 2003 Leukocyte density and pro-inflammatory cytokine expression in human fetal membranes, decidua, cervix and myometrium before and during labour at term. *Molecular Human Reproduction* **9** 41–45.

Osman I, Young A, Jordan F, Greer IA & Norman JE 2006 Leukocyte density and proinflammatory mediator expression in regional human fetal membranes and decidua before and during labor at term. *Journal of the Society for Gynecologic Investigation* **13** 97–103.

Perrier d'Hauterive S, Charlet-Renard C, Berndt S, Dubois M, Munaut C, Goffin F, Hagelstein MT, Noel A, Hazout A, Foidart JM *et al.* 2004 Human chorionic gonadotropin and growth factors at the embryonic–endometrial interface control leukemia inhibitory factor (LIF) and interleukin 6 (IL-6) secretion by human endometrial epithelium. *Human Reproduction* **19** 2633–2643.

Persson I 2000 Estrogens in the causation of breast, endometrial and ovarian cancers – evidence and hypotheses from epidemiological findings. *Journal of Steroid Biochemistry and Molecular Biology* **74** 357–364.

Piccinni MP, Beloni L, Livi C, Maggi E, Scarselli G & Romagnani S 1998 Defective production of both leukemia inhibitory factor and type 2 T-helper cytokines by decidual T cells in unexplained recurrent abortions. *Nature Medicine* **4** 1020–1024.

Piccinni MP, Scaletti C, Maggi E & Romagnani S 2000 Role of hormone-controlled Th1- and Th2-type cytokines in successful pregnancy. *Journal of Neuroimmunology* **109** 30–33.

Piccinni MP, Scaletti C, Vultaggio A, Maggi E & Romagnani S 2001 Defective production of LIF, M-CSF and Th2-type cytokines by T cells at fetomaternal interface is associated with pregnancy loss. *Journal of Reproductive Immunology* **52** 35–43.

Pirianov G, Waddington SN, Lindstrom TM, Terzidou V, Mehmet H & Bennett PR 2009 The cyclopentenone 15-deoxy-delta 12,14-prostaglandin J(2) delays lipopolysaccharide-induced preterm delivery and reduces mortality in the newborn mouse. *Endocrinology* **150** 699–706.

van der Poll T 2001 Coagulation and inflammation. *Journal of Endotoxin Research* **7** 301–304.

van der Poll T, Levi M, Hack CE, ten Cate H, van Deventer SJ, Eerenberg AJ, de Groot ER, Jansen J, Gallati H & Buller HR 1994 Elimination of interleukin 6 attenuates coagulation activation in experimental endotoxemia in chimpanzees. *Journal of Experimental Medicine* **179** 1253–1259.

van der Poll T, de Jonge E & Levi M 2001 Regulatory role of cytokines in disseminated intravascular coagulation. *Seminars in Thrombosis and Hemostasis* **27** 639–651.

Popovici RM, Betzler NK, Krause MS, Luo M, Jauckus J, Germeyer A, Bloethner S, Schlotterer A, Kumar R, Strowitzki T *et al.* 2006 *Gene Expression Profiling of Human Endometrial–Trophoblast Interaction in a Coculture Model*, pp 5662–5675.

Prussin C & Metcalfe DD 2003 IgE, mast cells, basophils, and eosinophils. *Journal of Allergy and Clinical Immunology* **111** S486–S494.

Rae MT, Niven D, Critchley HO, Harlow CR & Hillier SG 2004*a* Antiinflammatory steroid action in human ovarian surface epithelial cells. *Journal of Clinical Endocrinology and Metabolism* **89** 4538–4544.

Rae MT, Niven D, Ross A, Forster T, Lathe R, Critchley HO, Ghazal P & Hillier SG 2004*b* Steroid signalling in human ovarian surface epithelial cells: the response to interleukin-1alpha determined by microarray analysis. *Journal of Endocrinology* **183** 19–28.

Rajakariar R, Yaqoob MM & Gilroy DW 2006 COX-2 in inflammation and resolution. *Molecular Interventions* **6** 199–207.

Rauk PN & Chiao JP 2000 Interleukin-1 stimulates human uterine prostaglandin production through induction of cyclooxygenase-2 expression. *American Journal of Reproductive Immunology* **43** 152–159.

Redman CW, Sacks GP & Sargent IL 1999 Preeclampsia: an excessive maternal inflammatory response to pregnancy. *American Journal of Obstetrics and Gynecology* **180** 499–506.

Rees MC 1989 Heavy, painful periods. *Baillière's Clinical Obstetrics and Gynaecology* **3** 341–356.

Richards JS, Russell DL, Ochsner S & Espey LL 2002 Ovulation: new dimensions and new regulators of the inflammatory-like response. *Annual Review of Physiology* **64** 69–92.

Rinehart BK, Terrone DA, Lagoo-Deenadayalan S, Barber WH, Hale EA, Martin JN Jr & Bennett WA 1999 Expression of the placental cytokines tumor necrosis factor alpha, interleukin 1beta, and interleukin 10 is increased in preeclampsia. *American Journal of Obstetrics and Gynecology* **181** 915–920.

Robb L, Li R, Hartley L, Nandurkar HH, Koentgen F & Begley CG 1998 Infertility in female mice lacking the receptor for interleukin 11 is due to a defective uterine response to implantation. *Nature Medicine* **4** 303–308.

Rodts-Palenik S, Wyatt-Ashmead J, Pang Y, Thigpen B, Cai ZW, Rhodes P, Martin JN, Granger J & Bennett WA 2004 Maternal infection-induced white matter injury is reduced by treatment with interleukin-10. *American Journal of Obstetrics and Gynecology* **191** 1387–1392.

Romero R & Tartakovsky B 1992 The natural interleukin-1 receptor antagonist prevents interleukin-1-induced preterm delivery in mice. *American Journal of Obstetrics and Gynecology* **167** 1041–1045.

Rovere-Querini P, Antonacci S, Dell'Antonio G, Angeli A, Almirante G, Cin ED, Valsecchi L, Lanzani C, Sabbadini MG, Doglioni C *et al.* 2006 Plasma and tissue expression of the long pentraxin 3 during normal pregnancy and preeclampsia. *Obstetrics and Gynecology* **108** 148–155.

Ruddock NK, Shi SQ, Jain S, Moore G, Hankins GD, Romero R & Garfield RE 2008 Progesterone, but not 17-alpha-hydroxyprogesterone caproate, inhibits human myometrial contractions. *American Journal of Obstetrics and Gynecology* **199** e391–e397.

Ryan IP, Tseng JF, Schriock ED, Khorram O, Landers DV & Taylor RN 1995 Interleukin-8 concentrations are elevated in peritoneal fluid of women with endometriosis. *Fertility and Sterility* **63** 929–932.

Ryu HS, Chang KH, Yang HW, Kim MS, Kwon HC & Oh KS 2000 High cyclooxygenase-2 expression in stage IB cervical cancer with lymph node metastasis or parametrial invasion. *Gynecologic Oncology* **76** 320–325.

Salamonsen LA 2003 Tissue injury and repair in the female human reproductive tract. *Reproduction* **125** 301–311.

Salamonsen LA & Lathbury LJ 2000 Endometrial leukocytes and menstruation. *Human Reproduction Update* **6** 16–27.

Salamonsen LA, Kovacs GT & Findlay JK 1999 Current concepts of the mechanisms of menstruation. *Baillière's Best Practice & Research. Clinical Obstetrics & Gynaecology* **13** 161–179.

Sales KJ & Jabbour HN 2003*a* Cyclooxygenase enzymes and prostaglandins in pathology of the endometrium. *Reproduction* **126** 559–567.

Sales KJ & Jabbour HN 2003*b* Cyclooxygenase enzymes and prostaglandins in reproductive tract physiology and pathology. *Prostaglandins & Other Lipid Mediators* **71** 97–117.

Sales KJ, Katz AA, Davis M, Hinz S, Soeters RP, Hofmeyr MD, Millar RP & Jabbour HN 2001 Cyclooxygenase-2 expression and prostaglandin E(2) synthesis are up- regulated in carcinomas of the cervix: a possible autocrine/paracrine regulation of neoplastic cell function via EP2/EP4 receptors. *Journal of Clinical Endocrinology and Metabolism* **86** 2243–2249.

Sales KJ, Katz AA, Howard B, Soeters RP, Millar RP & Jabbour HN 2002 Cyclooxygenase-1 is up-regulated in cervical carcinomas: autocrine/-paracrine regulation of cyclooxygenase-2, prostaglandin e receptors, and angiogenic factors by cyclooxygenase-1. *Cancer Research* **62** 424–432.

Sales KJ, Maudsley S & Jabbour HN 2004 Elevated prostaglandin EP2 receptor in endometrial adenocarcinoma cells promotes vascular endothelial growth factor expression via cyclic 3′,5′-adenosine monophosphate-mediated transactivation of the epidermal growth factor receptor and extracellular signal-regulated kinase 1/2 signaling pathways. *Molecular Endocrinology* **18** 1533–1545.

Sales KJ, Boddy SC & Jabbour HN 2008 F-prostanoid receptor alters adhesion, morphology and migration of endometrial adenocarcinoma cells. *Oncogene* **27** 2466–2477.

Sampson MT & Kakkar AK 2002 Coagulation proteases and human cancer. *Biochemical Society Transactions* **30** 201–207.

Saunders PT 2005 Does estrogen receptor beta play a significant role in human reproduction? *Trends in Endocrinology and Metabolism* **16** 222–227.

Scher JU & Pillinger MH 2009 The anti-inflammatory effects of prostaglandins. *Journal of Investigative Medicine* **57** 703–708.

Schofield G & Kimber SJ 2005 Leukocyte subpopulations in the uteri of leukemia inhibitory factor knockout mice during early pregnancy. *Biology of Reproduction* **72** 872–878.

Serhan CN, Chiang N & Van Dyke TE 2008 Resolving inflammation: dual anti-inflammatory and pro-resolution lipid mediators. *Nature Reviews. Immunology* **8** 349–361.

Sherwin JR, Sharkey AM, Cameo P, Mavrogianis PM, Catalano RD, Edassery S & Fazleabas AT 2007 Identification of novel genes regulated by chorionic gonadotropin in baboon endometrium during the window of implantation. *Endocrinology* **148** 618–626.

Shim SS, Romero R, Hong JS, Park CW, Jun JK, Kim BI & Yoon BH 2004 Clinical significance of intra-amniotic inflammation in patients with preterm premature rupture of membranes. *American Journal of Obstetrics and Gynecology* **191** 1339–1345.

Shynlova O, Tsui P, Dorogin A & Lye SJ 2008 Monocyte chemoattractant protein-1 (CCL-2) integrates mechanical and endocrine signals that mediate term and preterm labor. *Journal of Immunology* **181** 1470–1479.

Sivridis E, Giatromanolaki A, Agnantis N & Anastasiadis P 2001 Mast cell distribution and density in the normal uterus-metachromatic staining using lectins. *European Journal of Obstetrics, Gynecology, and Reproductive Biology* **98** 109–113.

Smith OP, Jabbour HN & Critchley HO 2007 Cyclooxygenase enzyme expression and E series prostaglandin receptor signalling are enhanced in heavy menstruation. *Human Reproduction* **22** 1450–1456.

Sporn MB & Roberts AB 1986 Peptide growth factors and inflammation, tissue repair, and cancer. *Journal of Clinical Investigation* **78** 329–332.

Stewart CL, Kaspar P, Brunet LJ, Bhatt H, Gadi I, Kontgen F & Abbondanzo SJ 1992 Blastocyst implantation depends on maternal expression of leukaemia inhibitory factor. *Nature* **359** 76–79.

Straub RH 2007 The complex role of estrogens in inflammation. *Endocrine Reviews* **28** 521–574.

Subbaramaiah K & Dannenberg AJ 2007 Cyclooxygenase-2 transcription is regulated by human papillomavirus 16 E6 and E7 oncoproteins: evidence of a corepressor/coactivator exchange. *Cancer Research* **67** 3976–3985.

Takemura Y, Osuga Y, Harada M, Hirata T, Koga K, Yoshino O, Hirota Y, Morimoto C, Yano T & Taketani Y 2005 Concentration of adiponectin in peritoneal fluid is decreased in women with endometriosis. *American Journal of Reproductive Immunology* **54** 217–221.

Takemura Y, Osuga Y, Yamauchi T, Kobayashi M, Harada M, Hirata T, Morimoto C, Hirota Y, Yoshino O, Koga K *et al.* 2006 Expression of adiponectin receptors and its possible implication in the human endometrium. *Endocrinology* **147** 3203–3210.

Tapia A, Salamonsen LA, Manuelpillai U & Dimitriadis E 2008 Leukemia inhibitory factor promotes human first trimester extravillous trophoblast adhesion to extracellular matrix and secretion of tissue inhibitor of metalloproteinases-1 and -2. *Human Reproduction* **23** 1724–1732.

Terrone DA, Rinehart BK, Granger JP, Barrilleaux PS, Martin JN Jr & Bennett WA 2001 Interleukin-10 administration and bacterial endotoxin-induced preterm birth in a rat model. *Obstetrics and Gynecology* **98** 476–480.

Thelen M & Stein JV 2008 How chemokines invite leukocytes to dance. *Nature Immunology* **9** 953–959.

Thomson A, Lunan C, Cameron A, Cameron I, Greer I & Norman J 1997 Nitric oxide donors induce ripening of the human uterine cervix: a randomised controlled trial. *British Journal of Obstetrics and Gynaecology* **104** 1054–1057.

Thomson A, Telfer J, Young A, Campbell S, Stewart C, Cameron I, Greer I & Norman J 1999 Leukocytes infiltrate the myometrium during human parturition: further evidence that labour is an inflammatory process. *Human Reproduction* **14** 229–236.

Tong BJ, Tan J, Tajeda L, Das SK, Chapman JA, DuBois RN & Dey SK 2000 Heightened expression of cyclooxygenase-2 and peroxisome proliferator-activated receptor-δ in human endometrial adenocarcinoma. *Neoplasia* **2** 483–490.

Tornblom SA, Patel FA, Bystrom B, Giannoulias D, Malmstrom A, Sennstrom M, Lye SJ, Challis JR & Ekman G 2004 15-Hydroxyprostaglandin dehydrogenase and cyclooxygenase 2 messenger ribonucleic acid expression and immunohistochemical localization in human cervical tissue during term and preterm labor. *Journal of Clinical Endocrinology and Metabolism* **89** 2909–2915.

Tribe RM, Moriarty P, Dalrymple A, Hassoni AA & Poston L 2003 Interleukin-1β induces calcium transients and enhances basal and store operated calcium entry in human myometrial smooth muscle. *Biology of Reproduction* **68** 1842–1849.

Ulukus M, Ulukus EC, Seval Y, Zheng W & Arici A 2005 Expression of interleukin-8 receptors in endometriosis. *Human Reproduction* **20** 794–801.

Vane JR & Botting RM 1998 Anti-inflammatory drugs and their mechanism of action. *Inflammation Research* **47** S78–S87.

Wahl SM, Wong H & McCartney-Francis N 1989 Role of growth factors in inflammation and repair. *Journal of Cellular Biochemistry* **40** 193–199.

Watari M, Watari H, DiSanto ME, Chacko S, Shi GP & Strauss JF III 1999 Pro-inflammatory cytokines induce expression of matrix-metabolizing enzymes in human cervical smooth muscle cells. *American Journal of Pathology* **154** 1755–1762.

White CA, Johansson M, Roberts CT, Ramsay AJ & Robertson SA 2004 Effect of interleukin-10 null mutation on maternal immune response and reproductive outcome in mice. *Biology of Reproduction* **70** 123–131.

von Wolff M, Thaler CJ, Strowitzki T, Broome J, Stolz W & Tabibzadeh S 2000 Regulated expression of cytokines in human endometrium throughout the menstrual cycle: dysregulation in habitual abortion. *Molecular Human Reproduction* **6** 627–634.

Wymann MP & Schneiter R 2008 Lipid signalling in disease. *Nature Reviews. Molecular Cell Biology* **9** 162–176.

Yamada M & Gentry PA 1995 Hemostatic profile of bovine ovarian follicular fluid. *Canadian Journal of Physiology and Pharmacology* **73** 624–629.

Yamauchi T, Kamon J, Waki H, Terauchi Y, Kubota N, Hara K, Mori Y, Ide T, Murakami K, Tsuboyama-Kasaoka N *et al.* 2001 The fat-derived hormone adiponectin reverses insulin resistance associated with both lipoatrophy and obesity. *Nature Medicine* **7** 941–946.

Ye X 2008 Lysophospholipid signaling in the function and pathology of the reproductive system. *Human Reproduction Update* **14** 519–536.

Yoon BH, Jun JK, Romero R, Park KH, Gomez R, Choi JH & Kim IO 1997*a* Amniotic fluid inflammatory cytokines (interleukin-6, interleukin-1beta, and tumor necrosis factor-alpha), neonatal brain white matter lesions, and cerebral palsy. *American Journal of Obstetrics and Gynecology* **177** 19–26.

Yoon BH, Kim CJ, Romero R, Jun JK, Park KH, Choi ST & Chi JG 1997*b* Experimentally induced intrauterine infection causes fetal brain white matter lesions in rabbits. *American Journal of Obstetrics and Gynecology* **177** 797–802.

Yoshida M, Sagawa N, Itoh H, Yura S, Takemura M, Wada Y, Sato T, Ito A & Fujii S 2002 Prostaglandin F(2alpha), cytokines and cyclic mechanical stretch augment matrix metalloproteinase-1 secretion from cultured human uterine cervical fibroblast cells. *Molecular Human Reproduction* **8** 681–687.

Young A, Thomson A, Ledingham M, Jordan F, Greer I & Norman J 2002 Immunolocalization of pro-inflammatory cytokines in myometrium, cervix and fetal membranes during human parturition at term. *Biology of Reproduction* **66** 445–449.

Yuan M, Jordan F, McInnes IB, Harnett MM & Norman JE 2009 Leukocytes are primed in peripheral blood for activation during term and preterm labour. *Molecular Human Reproduction* **15** 713–724.

Zhang J, Nie G, Jian W, Woolley DE & Salamonsen LA 1998 Mast cell regulation of human endometrial matrix metalloproteinases: a mechanism underlying menstruation. *Biology of Reproduction* **59** 693–703.

Ziegler-Heitbrock L 2007 The CD14+ CD16+ blood monocytes: their role in infection and inflammation. *Journal of Leukocyte Biology* **81** 584–592.

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