[Frontiers in Bioscience 13, 5794-5809, May 1, 2008]

# Prostaglandins in labor – a translational approach

**Abdul H. Khan[1], Ray J. Carson[2], Scott M. Nelson[1]**

*[1]Reproductive and Maternal Medicine, Division of Developmental Medicine, University of Glasgow, Glasgow Royal Infirmary, 10 Alexandra Parade, Glasgow, G31, 2 ER, U.K., [2]Biomolecular Sciences, Faculty of Health and Life Sciences, Coventry University, Priory Street, Coventry, CV1 5FB, U.K.*

## TABLE OF CONTENTS



## 1. ABSTRACT

The mechanisms involved in the initiation of human labor are largely unknown. Understanding the molecular pathways is fundamental in both the development of effective therapeutic strategies and intervention to prevent preterm labor. Prostaglandins are bioactive lipids and members of the eicosanoids family, derived from arachidonic acid, which act in a paracrine or autocrine manner and function via binding to specific G-protein-coupled receptors, activating intracellular signaling and gene transcription. Prostaglandins have a central role in the maintenance of pregnancy and initiation of labor, with the change from uterine quiescence to a contractile state facilitated by differential expression of prostaglandin receptors within the myometrium and fetal membranes. Clinical evidence for the key role of prostaglandins in human parturition is evident from their successful exploitation as exogenous agents for the induction of labor and the role of prostaglandin synthase inhibitors as a preventative therapy for preterm labor. This review aims to focus on prostaglandin synthesis and metabolism and how differential regulation of prostaglandins and their receptors in gestational tissues interact in the initiation of labor.

## 2. INTRODUCTION

Prostaglandins were first extracted from semen, the prostate gland and seminal vesicles by Goldblatt and Von Euler in the 1930s, with purification of the first prostaglandin isomers in the 1950s and 60s by Bergström and colleagues (1). Further characterization has demonstrated that prostaglandins are 20 carbon chain unsaturated fatty acids (eicosanoids) synthesized from arachidonic acid (AA; 5, 8,11,14-eicosatetraenoic acid) via the enzymatic action of phospholipases and cyclooxygenase (COX). Prostaglandin synthesis and activation in gestational tissues is now recognized as one of the fundamental signals responsible for labor (2). In this review, we will discuss recent developments in our understanding of the synthesis of prostaglandins, their metabolism and transport, the interaction with receptors and the downstream signaling pathways with respect to the physiology of labor.

## 3. PROSTAGLANDIN BIOSYNTHESIS AND TRANSPORT

### 3.1. Biosynthesis of prostaglandins

Prostaglandin synthesis can be broadly divided into three main steps. In the first step, AA is liberated from

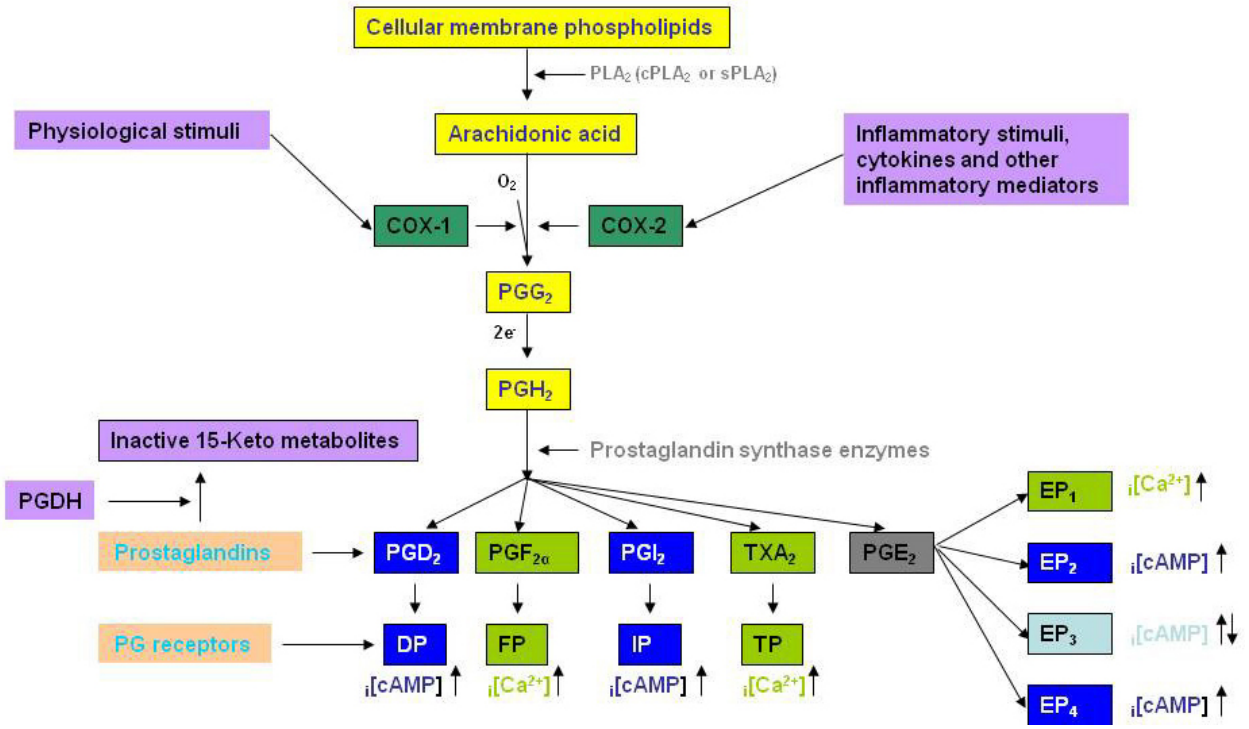


**Figure 1.** Prostaglandin biosynthesis pathways. Arachidonic acid liberated from cellular membrane phospholipids by the action of phospholipases $A_2$ and cyclooxygenase are the rate-limiting steps in the biosynthesis of PGs. COX-1 and COX-2 convert arachidonic acid to $PGH_2$, with further conversion to specific PGs depending on the specific PG synthases. Prostaglandin dehydrogenase (PGDH) metabolizes bioactive PGs into inactive keto-metabolites. The PG receptors for $PGD_2$, $PGF_{2\alpha}$, $PGI_2$, $TXA_2$ and $PGE_2$ are DP, FP, IP, TP and EP respectively. There are four receptor-subtypes for $PGE_2$ (EP1-4). Their actions are mediated via increases in intracellular calcium level (FP, TP, $EP_1$), increases in cAMP level (DP, IP, $EP_2$, $EP_4$) and increases or decreases in intracellular cAMP levels.

the sn-2 position of cellular membrane phospholipids (Figure 1). As levels of AA are generally very low in cells, thus its rate of liberation can limit prostaglandin synthesis. AA may itself also act as an important signaling molecule regulating its release and prostaglandin production (3-5), in addition to its role as the principal precursor in the biosynthesis of prostaglandins. The liberation of AA from membranes is by the action of specific acyl phospholipases, with phospholipase $A_2$ ($PLA_2$) enzymes catalyzing the hydrolysis of the sn-2 ester bonds of gylcerophospholipids and thereby achieving the release of AA. With respect to parturition there is evidence that the activity of $PLA_2$ is increased with labor at term in human placenta, fetal membranes and decidua, consistent with $PLA_2$ mediating release of uterine arachidonic acid (6-8). Of the several phospholipases $A_2$ enzymes, only type IIa secretory $PLA_2$ (14 kDa) and type IV cytosolic $PLA_2$ (85 kDa) enzymes have been characterized in human gestational tissues (9), however at present their relative contributions with respect to the observed increased PG synthesis at the onset of labor is still unknown. $cPLA_2$ is found to be ubiquitously expressed and has specificity for phospholipid substrates that contain AA at sn-2 position (10). The roles of the different $PLA_2$ enzymes and their relative functions and regulation of expression have been the subject of considerable recent study and are beyond the scope of this review, with the reader directed to recent literature (11-19).

In addition to the $PLA_2$ pathway, phospholipase C (PLC) enzyme forms diacylglycerol that can be cleaved to generate AA via a mono- or diglycerol lipase (20). The action of PLC coupled to diacylglycerol lipase, could provide a further indirect mechanism for the release of arachidonic acid for prostaglandin biosynthesis during parturition (21, 22).

The next step is the oxidation of AA and subsequent reduction to an intermediate endoperoxide prostaglandin $H_2$ ($PGH_2$). Production of $PGH_2$ is the rate limiting step in the synthesis of prostaglandins, with this reaction catalyzed by the prostaglandin H synthase (PGHS) enzyme, a 72-kD heme protein containing both cyclooxygenase (COX) and peroxidase activity. PGHS is also known as COX and PTGS (Prostaglandin-endoperoxide synthase), with the latter representing the most recent consensus nomenclature. To date, three isoforms of PGHS enzyme have been characterized; PGHS-1, PGHS-2 and PGHS-3, also known as cyclooxygenase COX -1 (PTGS1), COX-2 (PTGS2) and COX-3 (PTGS3) respectively, are responsible for PG synthesis, however, to date only COX-1 and COX-2 have been implicated in human parturition (23, 24).

COX-1 and -2 are both capable of catalyzing a cyclooxygenase and a peroxidase reaction, in which AA

and two molecules of oxygen are converted to PGG2, with subsequent reduction to PGH2 by the addition of two electrons (Figure 1). The cyclooxygenase reaction occurs in a hydrophobic channel in the core of the COX enzyme and the peroxidase reaction occurs at a heme-containing active site located near the protein surface (25). The two isoforms of human COX-1 and -2, although having approximately the same number of amino acids; 599 and 604 respectively (26), only demonstrate 60% homology at the nucleic and amino acid levels. Moreover, COX-1 contains a sequence of 17 amino acids in its amino terminus that is absent in COX-2, whereas COX-2 has an additional insert of 18 amino acids in its carboxy terminus (27). It is these alterations in COX structure that facilitate differential regulation. Indeed COX-1 is constitutively expressed in all cell types and functions as a housekeeping enzyme, and may therefore considered to be 'constitutive', whereas COX-2 is an 'inducible' isoform, found at variable levels in tissues, with induction of activity by cytokines, growth factors and by a variety of other agonists (28, 29). COX-2 is also activated at an approximately ten fold lower concentration of hydroperoxide than that of COX-1 (30, 31), further facilitating COX-2 to function independently of COX-1 in cells expressing both isoforms. Despite COX-1 and -2 having significant similarities in terms of cyclooxygenase active site structures, catalytic mechanisms and kinetics, there are two major structural differences. First, the cyclooxygenase active site of COX-2 is larger and more accommodating than COX-1, with this difference in size exploited to facilitate the development of selective COX-2 inhibitors. Second, although COX-1 and -2 have similar $K_m$ and $V_{max}$ values with arachidonic acid, COX-1 at low AA concentrations exhibits negative allosterism; this difference may facilitate COX-2 to compete effectively for newly released AA when both the isozymes are expressed in the same cell (25). For example, AA at 1μM concentration stimulates prostaglandin synthesis by human embryonic kidney 293 cells expressing COX-2, whereas concentrations of more than 10μM are required in order to stimulate prostaglandin synthesis in cells expressing only COX-1 (32).

The transport and post-transcriptional regulation of COX-2 mRNA is of critical importance in controlling the expression of the COX-2 gene and consequently may have significant functional impact. The nuclear export receptor chromosome maintenance region 1 (CRM1) is required to transport the COX-2 mRNA from the nucleus to the rough endoplasmic reticulum (33, 34), and a specific inhibitor of this pathway, leptomycin B, inhibits the transport and decreases the expression of COX-2 gene in mammary cancer cells (35). Although these data suggest that exploitation of inhibition of the CRM1 receptor may be useful in controlling the pathogenic expression of COX-2, to date the role of this receptor in human parturition has not been explored.

The third and final step in the prostaglandin synthesis pathway is the conversion of the PG intermediate, $PGH_2$ to one of the biologically active prostaglandins, namely; D ($PGD_2$), E ($PGE_2$), F ($PGF_{2\alpha}$), thromboxane ($TXA_2$), and prostacyclin ($I_2$) series with these reactions catalyzed by prostaglandin D synthase (PGDS), prostaglandin E synthase (PGES), prostaglandin F synthase (PGFS), thromboxane synthase (TXS) and prostacyclin synthase (PGIS) (36) respectively.

With respect to human parturition the role of these enzymes, the final step in prostaglandin synthesis continue to be elucidated, with multiple isoforms for most now being recognized. Indeed two distinct isoforms of PGDS have been characterized and cloned (37, 38); lipocalin-type PGDS (L-PGDS) and a cytosolic protein hemopoietic PGDS (H-PGDS) enzyme. In pregnancy H-PGDS was localized in human placenta; in contrast, L-PGDS was detected in amniotic fluid with further up-regulation with the onset of human labor (39). Changes in the L-PGDS level in human cervico-vaginal fluid has been suggested as a possible indicator of membrane rupture during pregnancy (40).

To date, four isoforms of PGES have been characterized; two are membrane bound PGES (mPGES-1 and mPGES2), one cytosolic (cPGES) and one is glutathione S-transferase (GST-μ) (41). cPGES primarily converts COX-1 derived $PGH_2$ to $PGE_2$ and is responsible for immediate PG biosynthesis. In contrast, mPGES-1 may be induced in response to pro-inflammatory stimuli and is primarily associated with COX-2 when there is limited availability of AA, however, it can also couple to COX-1 where AA is available, thereby facilitating the delayed biosynthesis of $PGE_2$. (42). Relative to mPGES-1, mPGES-2 is structurally different and can couple with both COX-1 and COX-2. Lastly although the GST-μ isoform has been implicated in PG production; whether it is coupled with either COX-1 or COX-2 (41), and has a role in human labor has yet to be established.

Two isoforms of PGFS have been characterized and reported; one known as lung-type PGFS (or PGFS I) (43, 44) and the other liver-type PGFS (or PGFS II) (45, 46). Data on PGFS regulation have been limited to *in vitro* studies mostly in bovine species (47, 48), with a decrease in PGFS mRNA expression in amnion (49) was noted after dexamethasone induced labor, but in contrast no changes were observed in either the myometrium or endometrium of sheep after induced and spontaneous labor (49). In contrast PGIS (50) and TXS (51-53), membrane-bound hemoproteins and members of the cytochrome P450 family have been implicated in human labor with PGIS shown to be expressed by human myometrial smooth muscle (54) with additional localization to vascular smooth muscle and endothelial cells within the lower uterine segment, although expression decreased with advancing gestation (55). Expression of TXS has been shown in the human endometrial and myometrial cells as well as in uterine blood vessels (56) and also in placental villi, fetal membranes, umbilical cord and myometrium (57).

### 3.2. Prostaglandin transport

Synthesized prostaglandins are organic anions with a pKa value of about 5 and exist primarily as charged species at a physiological pH (58, 59). After biosynthesis PGs can cross the plasma membrane and this rapid

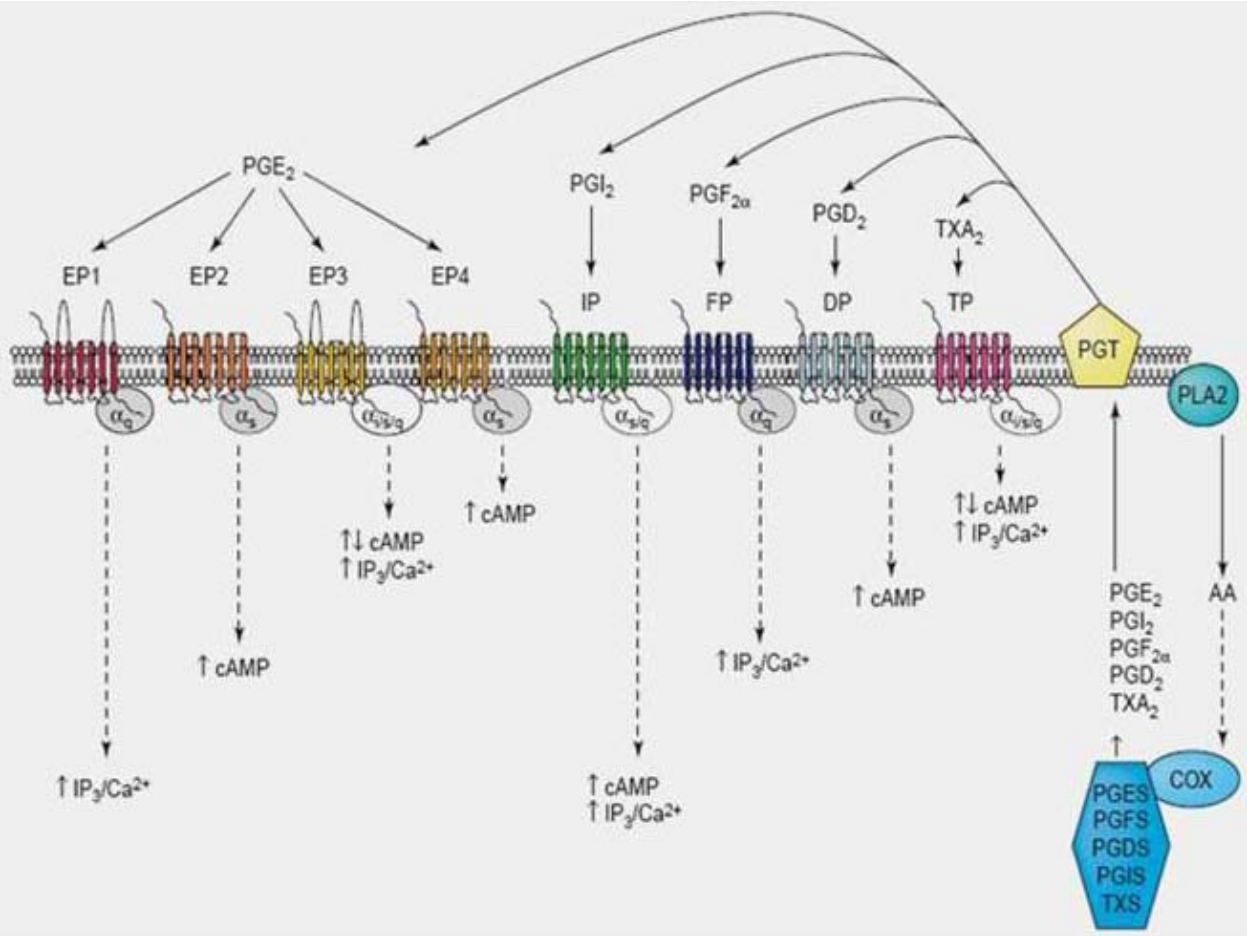


**Figure 2.** Prostaglandins with target receptor subtypes and signaling mechanisms. Arachidonic acid (AA) is released from plasma membrane phospholipids by phospholipase $A_2$ ($PLA_2$) and used by COX enzymes and specific synthase enzymes, such as prostaglandin D synthase (PGDS), PGES, PGFS, PGIS thromboxane synthase (TXS), to form prostaglandin $D_2$ ($PGD_2$), $PGE_2$, $PGF_{2\alpha}$, $PGI_2$ and thromboxane $A_2$ ($TXA_2$), respectively. These molecules are actively transported out of the cell by means of a prostaglandin transporter (PGT), where they exert an autocrine or paracrine effect by coupling to their respective heptahelical transmembrane receptors, DP, $EP_1$–$EP_4$, FP, IP and TP, to activate second messengers, such as cyclic AMP (cAMP) and inositol (1,4,5)-trisphosphate ($IP_3$), and intracellular signaling cascades. Reproduced with permission from (193).

transportation into cells is facilitated by prostaglandin transporter (PGT) (60), a member of the 12-trans-membrane organic anion-transporting polypeptides superfamily (61, 62) (Figure 2). The affinity of PGT for PGs is high; with $K_m$ values for $PGE_2$ and $PGF_{2\alpha}$ of 50-95 nM (63, 64), notably the PG concentrations are probably in this range when presented to the transporter in tissues (61), as $PGE_2$ concentration in human lung have been measured at 70 nM and in human semen this value increases up to 70 μM (65). Although, PGT has been identified in human placenta, endometrium and decidualized endometrial stromal cells (60, 66), it has not yet been reported in the human uterus in relation to pregnancy and labor. However, it is believed that PGT may facilitate PG transfer across uterine compartments and their re-uptake after release (67). Indeed the studies identifying the re-uptake of PGs via PGT in prostaglandin synthesizing cells suggests that PG signaling may be similar to synaptic signaling (68), with potential reuse of PGs as signaling moieties.

### 3.3. Concentrations of prostaglandins in tissues

Due to the labile nature of prostaglandins they are difficult to measure, however, concentration of 15d-$PGJ_2$ and $\Delta^{12}$-$PGJ_2$ have been reported in the picomolar-to-nanomolar range in the medium of 3T3-L1 preadipocytes and in human urine respectively (69-71), with plasma PGE and PGF concentrations of 0.7nM (72). Maternal human plasma levels of PGFM, a $PGF_{2\alpha}$ metabolite, increase considerably with advancing gestation with further increases associated with labor at term. In contrast, no significant difference were found in plasma levels in preterm labor when compared with term not in labor (73, 74), suggesting a gestational rise in addition to the observed increases with labor. Human amniotic fluid $PGE_2$ concentrations have also been shown to increase 10 fold, with a concomitant 3-fold rise in $PGF_{2\alpha}$ (75). Similarly the concentration of 9 α,11β-$PGF_2$, a $PGD_2$ derivative with equal myometrial contractile potency as $PGF_{2\alpha}$, (76, 77)

demonstrated a two fold increase in amniotic fluid concentrations with human labor (78). Lastly some studies have reported increased decidual cells PG concentrations with the onset of labor at term (79, 80), with others reporting no change (81, 82). These widespread changes in concentrations of PG with human labor, would appear to be consistent with high concentrations having a contractile effect on myometrial smooth muscle. Certainly PGs appear to exhibit dose-dependent effects with potentially relaxatory effects at lower dose ranges and contractile effects at higher concentrations (83). Consistent with this in non-pregnant human myometrium, $PGE_2$ appears to have a variable effect on contractility (84) with a biphasic response in pregnant human myometrium (85). These concentration dependent responses in conjunction with alterations in receptor levels may facilitate the tight regulation of contractile responses and enable the dynamic transition of myometrium from a quiescent to an active phenotype prerequisite for labor.

## 4. PROSTAGLANDIN RECEPTORS IN PREGNANCY AND LABOR

The action of prostaglandins are predominantly mediated via specific receptors, including the four prostaglandin E receptor subtypes $EP_{1\text{-}4}$, the DP receptor for prostaglandin $D_2$, and the FP receptor for prostaglandin $F_{2\alpha}$. It is based on their historical effects on smooth muscle cells and on their second messenger system (Figure 1), that these prostaglandin receptors have been divided into relaxatory ($EP_2$, $EP_4$, DP, IP) and contractile ($EP_1$, $EP_3$, FP, TP) groups. This differential response is mediated via differential coupling of PG receptors to either $G_s$ or Gi proteins (Figure 2). Activation of $G_s$ stimulates adenylate cyclase resulting in an increase in cAMP levels, with subsequent relaxation of smooth muscle. Conversely, activation of $G_i$ coupled PG receptors inhibits adenylate cyclase, reduces cAMP production resulting in an increase in intracellular calcium with initiation of an action potential and contraction of smooth muscle (86). Notably although the effects of PGs are predominantly mediated through G-protein coupled plasma membrane receptors, data on neonatal porcine brain and adult rat liver has shown that $PGE_2$ may also modulate gene transcription via perinuclear envelope expression of $EP_1$, $EP_3$ and $EP_4$ receptors (87).

To date the role of the various forms of PG receptors continues to be elucidated with increasing recognition that for any given type additional splice variants may also exist. For example several isoform of $EP_3$ receptor subtypes have been reported (88, 89), with generation of subtypes by alternative splicing at the carboxy-terminal tail of nine encoded transcripts; $EP_{3\text{-}I(1a)}$, $EP_{3\text{-}I(1b)}$, $EP_{3\text{-}II}$, $EP_{3\text{-}III}$, $EP_{3\text{-}IV}$, $EP_{3\text{-}V}$, $EP_{3\text{-}VI}$, $EP_{3\text{-}e}$ and $EP_{3\text{-}f}$. Notably in PG receptors, it is the carboxy-terminal tail that is important in mediating G protein coupling, and thus, changes in this region seems to alter the coupling affinity of particular receptor subtypes. Specifically, $EP_3$ is coupled to $G_i$ and hence can mediate a contractile effect; however, splice variants ($EP_{3\text{-}II}$ and $EP_{3\text{-}IV}$) may lead to different cellular responses depending on coupling to $G_s$ and/or $G_q$ respectively (90, 91).

Prostaglandin E receptor isoforms also show differential spatial expression in myometrium with significantly increased $EP_1$ expression in laboring human lower segment myometrium at term (92), with an up-regulation of $EP_3$ in fundal tissue. In contrast $EP_2$ mRNA expression was significantly increased in the lower segment of non laboring preterm human myometrium as compared to term samples, potentially reflecting the role of $EP_2$ in the maintenance of uterine quiescence (93). Furthermore mRNA expression of the four non-contractile $EP_3$ splice variants ($EP_{3I\text{-}1b}$, $EP_{3\text{-}II}$, $EP_{3\text{-}III}$ and $EP_{3\text{-}IV}$) was down-regulated with increasing gestation in both upper segment and the lower segment of human myometrium (92). Given the well established paradigm that labor involves the switch from myometrial quiescence to an active contractile state, this orchestrated up-regulation of contractile receptors, with concurrent loss of the relaxatory receptors and a decrease in intracellular cAMP levels would be consistent. Anatomical differences in location of the prostaglandin receptors may also be important, with studies in baboons demonstrating an abundance of the contractile $EP_1$ and $EP_3$ receptors in fundal tissue, in contrast to the predilection for the relaxatory $EP_2$ and $EP_4$ receptors for the lower uterine segment (94), where they may be partly responsible for relaxation of the uterus to allow smooth passage of the fetus and help in delivery of the baby.

Although there are dramatic changes in PG receptor expression with labor, substantive changes also occur with the initial transition to a pregnant state with maintenance of pro-quiescent changes across gestation (92). Indeed during human pregnancy, a decrease in the number of contractile PG receptors have been reported (95-97), with $EP_3$ expression in pregnant human myometrium 40% less than in non-pregnant samples. FP expression is also decreased during human pregnancy by approximately 45%, although these changes are not sustained as expression increases with labor (98, 93). In rats similar increases with labor were noted, although a decrease with pregnancy per se was not noted (99). Certainly these gestational and labor associated changes suggest a role for FP receptors in maintaining and then switching the myometrial phenotype. The noted concomitant increase in relaxatory $EP_2$ receptor responses during pregnancy would also facilitate uterine quiescence (96). In addition increased coupling of $G_{\alpha s}$ to adenylate cyclase with subsequent increases in relaxatory cAMP formation throughout gestation as compared to the reduction in coupling observed with labor, may also potentially act as a triggering mechanism for the initiation of labor (100).

At present there is a limited amount of published data available on the regulation of prostaglandin receptors as related to uterine physiology. Certainly steroids have been implicated in PG receptor regulation in rats. With increased FP and decreased $EP_2$ mRNA expression noted after progesterone receptor antagonism by RU486 (99). Similarly, estrogen antagonism decreased FP mRNA, in contrast to the enhanced FP expression with administration of exogenous estradiol (99). Conversely supplementation with progesterone alone increased $EP_2$ expression, and when this was combined with estradiol, the increase was

even greater (99). These studies suggest that steroids have a role in the regulation of prostaglandin receptors, at least in rats, and these effects may underlie some of the observed effects of progesterone on preterm birth (101).

With respect to additional mechanisms of regulation of prostaglandin receptors, *in vitro* myometrial cell cultures are known to express $EP_2$ receptors, with stimulation by IL-1β inducing $EP_4$ expression (102), suggesting that pro-inflammatory cytokines which have been implicated in the pathophysiology of labor may partly regulate myometrial activity via prostaglandin receptor up-regulation. PG receptors may also exhibit auto-regulation via their own ligands, with $PGF_{2\alpha}$ shown to decrease the number of binding sites in the rat myometrium, consistent with ligand-induced receptor down regulation (103). Our own recent gene array studies of human parturition also showed a 10 fold down-regulation in $EP_3$ receptors expression in lower segment laboring human myometrium compared to non laboring samples (104), consistent with the published mRNA and immunohistochemistry data (92).

## 5. ROLE OF PROSTAGLANDINS AND ITS REGULATION IN LABOR

### 5.1. Prostaglandins and labor at term

It is clear that prostaglandins have an important role to play in parturition, with exogenous prostaglandins used for abortion (105), cervical ripening (101) and induction of labor (106, 107). Conversely inhibition of prostaglandin synthesis can delay the onset of labor, reduces uterine contractions, and prolongs the process of labor (108, 109). Certainly endogenous prostaglandin production has been implicated in human parturition with levels in maternal plasma, amniotic fluid and urine all known to rise prior to the onset of labor, with continued elevations and subsequent plateauing of levels with parturition (101, 110-112). The primary sites of this PG synthesis are the fetal membranes and decidua (80, 113), however, myometrial and cervical contributions may also be important with prostaglandin E and prostaglandin $F_{2\alpha}$ increasing in a time-dependent manner during late gestation. Altered expression of the synthetic enzymes has been noted with labor, with lower segment human myometrium PGF synthase expression decreased by 3-fold ($P<0.02$), in contrast to a 6-fold increase in cervical PGES expression (104). The c-PGES and m-PGES-1 isoforms have also been identified in human fetal membranes and placenta to show differential expression with labor (114-116), with similar up-regulation of expression of m-PGES1 and m-PGES-2 in human myometrium (55, 117), (118). Notably m-PGES activity may be further enhanced via the positive feedback of $PGE_2$ enhancing IL-1β-induced mPGES-1 expression (119), this loop will also enable continuous production of $PGE_2$ by invading monocytes with the downstream effect of myometrial stimulation. In addition, human amnion, chorion and decidua primarily produce $PGE_2$, further propagating this cycle, in contrast to the decidua which mainly produces $PGF_{2\alpha}$. This production of $PGE_2$, $PGF_{2\alpha}$, and other prostanoids by the fetoplacental unit would appear to relate to the observed increased contractile activity during labor. Further, any stimulus (hypoxia, infection, exposure to oxytocin, hypertonic solutions, prostaglandins, or arachidonic acid) facilitating $PGE_2$ synthesis in the fetal membrane is likely to induce the formation of $PGF_{2\alpha}$ in decidua and myometrium and clinically translate to a stimulated myometrial phenotype. Notably however, since naturally occurring prostaglandins in the human body are rapidly metabolized, induction of labor will require a sustained presence of PGs (120).

### 5.2. Cyclooxygenase (COX) enzymes and gene expression in labor

Despite the apparent central role for PGs in the process of parturition, analysis of gene expression profiles of fundal myometrium from pregnant women before and after the onset of labor have not demonstrated an alteration in fundal COX-2 expression in association with spontaneous labor (121, 122), with human microarray data similarly not demonstrating any change in COX-1 or COX-2 expression in lower segment myometrial samples (123). Furthermore quantitative RT-PCR and protein studies have also not shown any change in COX-1 or COX-2 expression in human laboring myometrium both at term and preterm (124). However, studies are not consistent with COX-2 mRNA expression reported as increased in human fundal myometrium prior to the onset of term labor (121). These inconsistencies in results may be partially attributable to infection, as myometrial COX-2 expression was noted to be increased in women with chorioamnionitis (121), and IL-1β stimulates COX-2 transcription via activation of NF-κB in human myometrial cells (125). Moreover in a mouse model of inflammation-mediated preterm labor, there was a significant increase in uterine COX-2, but not COX-1 gene expression, with inhibition of COX-2 abolishing LPS-induced preterm labor (126).

In addition to these changes within the myometrium, COX-2 has also been implicated in cervical ripening. We observed an 11-fold ($P<0.01$) increase in COX-2 expression in laboring human cervix (104), with others noting differential regulation of COX-2 in endocervix mucosa as compared with cervical stroma (121). Immunohistochemical studies suggest that it is this differential localization to the endocervical glandular epithelial cells that are responsible for the observed up-regulation of COX-2 in the endocervix during human labor at term (121). In addition to these local cervical changes COX-2 protein expression also appears to be spatially regulated within the uterus with a distinct positive concentration gradient from fundus to cervix (127). Given that COX-2 expression is predominantly localized to macrophages, the major cell subtype responsible for the cervical inflammatory influx, this may underlie the observed gradient. Further increases within the cervix are possible due to additional localization primarily within cervical epithelial cells, activated endothelial cells and stromal fibroblasts, with numbers of positive COX-2-stromal fibroblasts per tissue cross-section increased in cervical tissues in labor (121).

Several studies have examined the relative contributions of COX-1 and -2 in the process of labor, hypothesizing that COX-2 is predominantly responsible for

the observed labor associated increases in PG production. Admittedly although expression of COX-1 enzyme has previously been reported to be constant throughout gestation in the lower segment human myometrium (122), recent analysis of mRNA expression suggests that COX-1 levels decline precipitously towards term in fetal membranes (128). Conversely, the mRNA level of COX-1 is reported to increase about three to four fold in late pregnancy, starting at approximately 35 weeks of gestation. The level of COX-1 reaches a peak at term, with no observed increase with the onset of labor. Conversely, mRNA levels of COX-2 are very low throughout pregnancy, but increase dramatically about six fold at the same time as COX-1. However, COX-2 mRNA level in amnion continues to rise during labor (129, 130). Similarly, COX-2 activity in the fetal membranes, chorio-decidua and myometrium significantly increases with advancing gestation (124, 131).

It would appear that these increases in COX-2 mRNA are tissue specific and mediated by transcriptional control in amnion and post-transcriptional control in chorion (128). Certainly COX-2 activation is NF-κB dependent, with two NF-κB response elements located within the COX-2 promoter, whereas, COX-1 has none (132). It is this NF-κB dependent pathway that interleukin-1beta (IL-1β) could potentially exploit as IL-1 β increases NF-κB activity in human amnion (133) , the main site of COX-2 activity, and myometrium, while also repressing progesterone receptor activity. Indeed IL-1β induced COX-2 expression in lower segment human fibroblast cells is suppressed by progesterone (134), with this effect mediated by reduced recruitment of NF-κB p65 to both proximal and distal NF-κB elements within the COX-2 promoter and concurrent increased expression of the NF-κB inhibitor Iκ-Bα (135). Clear evidence for the role of NF-κB in COX-2 regulation in human myometrial cells, has been achieved by inhibition of Iκ-Bα degradation with the result of impaired NF-κB nuclear translocation, suppression of COX-2 mRNA and protein expression, and inhibition of $PGE_2$ and $PGI_2$ synthesis (125). Collectively these data supports a concept whereby the pro-inflammatory state associated with both term and preterm labor potentially facilitates the induction of labor in a COX-2 and prostaglandin-dependent manner. With these changes being principally brought about by increased NF-κB activation and a decline in progesterone receptor coactivators (136), with the overall effect of overcoming the quiescent state that is maintained by progesterone throughout gestation (137).

### 5.3. Factors affecting prostaglandin production during labor

#### 5.3.1. Prostaglandin metabolism

Prostaglandin dehydrogenase (PGDH) has a key role to play in the physiology of labor, as PGDH determines the concentration of biologically active PGs within tissues, due to its role as a key metabolizer of prostaglandins. This metabolic activity of PGDH is dependent on an oxidized form of nicotinamide adenine dinucleotide (NAD+), to transform PGs into biologically inactive 15-keto compounds. These molecules are subsequently converted to 13,14-dihydro-15-keto derivatives (Figure 1) by Δ13,14-reductase, which are the stable inactive PG metabolites (138). Consequently the distribution of PGDH within gestational tissues may directly affect the concentration of PGs that reach the myometrium.

Chorion is the principal site of PGDH activity, with subsequent degradation of $PGE_2$ and $PGF_{2\alpha}$ into their inactive metabolites (139). Thus, the chorion may act as an in-built protective barrier minimizing the free transfer of bioactive PGs produced by the amnion or indeed even the chorion from passing to the underlying decidua and myometrium (49, 140, 141). Consistent with this, increases in $PGE_2$ concentration in amnion after labor do not result in an increase in $PGE_2$ concentration in myometrium as assessed *in vitro* (142). By contrast, a decrease in the metabolic activity of PGDH, and consequently, an increase in the concentration of bioactive PGs at the onset of labor both at term and preterm could allow $PGE_2$ to reach myometrium (143, 144), enabling contractions and thereby inducing preterm labor (145). Despite this clear evidence for a potentially significant role for PGDH in regulating PG trafficking, assessment of the permeability of intact-amnion-chorion-decidua revealed that 72% of $PGE_2$ remained in the active form and was not metabolized after passage across this barrier, with no alteration in the barrier characteristics after the onset of spontaneous labor (146).

PGDH expression is regulated by additional signals with pro-inflammatory cytokines and infectious agents including IL-1β and LPS inducing a reduction in PGDH expression. This regulation of PGDH may be responsible for the increased availability of bioactive prostaglandins which have been implicated in infection-induced preterm labor (147). In addition, PGDH may be regulated at a transcriptional level by the competing activities of progesterone and cortisol (148). A potential model whereby during pregnancy, fetal membrane PGDH activity is maintained by progesterone acting either through low level of progesterone receptors or via glucocorticoid receptors (GR) is subsequently reversed at term by elevated levels of cortisol competing with progesterone for GR with subsequent inhibition of PGDH transcription and activity is attractive. This would also enable local withdrawal of progesterone action within human intrauterine tissues, without requiring changes in systemic and circulating levels of progesterone. Notably glucocorticoids also induce the expression of prostaglandin synthesizing enzymes within the amnion and chorion directly, by up-regulating COX-2 expression and also indirectly via intermediary actions of paracrine effectors including CRH (148).

In addition to these effects within myometrium we have also shown that PGDH expression was down-regulated 11-fold ($P<0.01$) in the laboring human cervix at term (104). This cervical decrease in PGDH enzyme expression in association with active labor may act to increase the concentration of bioactive PGs within the human cervix, thereby facilitating cervical remodeling and dilation. Potentially localized reductions in PGDH activity may be present in those patients with premature cervical

softening, or conversely PGDH activity may be particularly high in those patients with an unfavorable cervix, presenting with a low Bishop score and poor progression at the onset of labor (149). If indeed PGDH crucially determines the passage and availability of biologically active prostaglandins from amnion and chorion to underlying cervix, then pharmacological manipulation of PDGH activity may represent a novel therapeutic intervention for not only preterm labor, but also cervical incompetence and induction of labor (149).

### 5.3.2. Stretch

It is well recognized that the uterus is subject to stretch throughout pregnancy, with mechanical stretch inducing myometrial smooth muscle hypertrophy and increases in tensile strength. Mechanical stretch, in addition to protein synthesis, can also activate the transcription of specific genes in particular; Connexin 43, oxytocin receptor, ion channels and FP receptors thereby rendering the myometrium responsive to uterotonins - a key step in the initiation of labor (150). Indeed stretching of human myometrium *in-vitro* has been reported to release prostaglandins (151). Furthermore, mechanical stretching of human lower uterine segment and cervix, with a rubber balloon, has shown induction of cervical softening and parturition at term, due to increases in the levels of $PGF_{2\alpha}$ in amniotic fluid (152) and plasma (153). Indeed COX-2 seems to be one of the target genes activated by mechanical stress, with myometrial COX-2 and $PGE_2$ expression increased with twins or polyhydramnios (154) and COX-2 and downstream $PGE_2$ expression increased after tonic stretch of primary human uterine myocytes (155). Pathway analysis to determine the principal players has suggested that this stretch related induction of COX-2 activity is via activation of the activator protein-1 (AP-1) system (155). Similarly stretch of primary human amnion epithelial cells is also associated with an increase in COX-2 expression and $PGE_2$ synthesis, again via activation of AP-1 and NF-κB transcription factors (156). The regulation of these stretch induced responses is unclear, however, data from animal studies suggest that progesterone may partially contribute by blocking stretch-induced responses, with little induction of gene expression until after functional progesterone withdrawal has occurred (150).

### 5.3.3. Oxytocin

Prostaglandins also have a close functional interaction with oxytocin, an important modulator of myometrial function (157). Myometrial oxytocin receptor activation is coupled with the activation of the phosphoinositide cycle, producing diacylgylcerol (DAG) and inositol trisphosphate ($IP_3$). DAG is a source of arachidonic acid, furthermore $IP_3$ via an influx of extracellular calcium by receptor operated channels increases intracellular $[Ca^{2+}]$, activating $PLA_2$ thereby further increasing AA release. In addition to these immediate changes infusion of oxytocin for longer periods to augment or induce labor may also initiate the myometrial transcription of COX-2 gene via the MAPK signaling pathway, ensuring continuous prostaglandin production (158). Furthermore although oxytocin and PGs can both independently stimulate myometrium (159), the response of myometrial strips to oxytocin is significantly increased after pre-treatment with prostaglandins (160).

### 5.3.4. Cytokines

Pro-inflammatory cytokines including IL-1β have been widely implicated in the initiation and progression of human labor both at term and preterm (161). This may be potentially through interactions with PGs as IL-1β is a potent agonist of prostaglandin synthesis in human myometrial cells, primarily via the induction of $PLA_2$ and COX-2 enzymes, with IL-1β stimulated cultured human myometrial cells exhibiting a dose- and time-dependent increase in $PGI_2$ and $PGE_2$ production, to the order of 15- to 23-fold (162). Furthermore mRNA level of $PGE_2$ synthesizing enzymes are associated with IL-1β in human labor supporting this pathway as a key factor (117).

## 6. TOCOLYTIC STRATEGIES

### 6.1. Clinical use of enzyme inhibitors in preterm labor

Given these critical roles for PGs in labor, the obvious clinical intervention has been the prevention of preterm labor by inhibition of COX enzymes. Certainly non-specific COX inhibitors, such as indomethacin, have reduced preterm birth, and increased gestational age at birth and birthweight (163). However, the major problem with such agents is due to their low molecular weight and high lipid solubility with the inevitable consequence that they readily cross the placenta. Due to their non-specific nature once in the fetal circulation they may cause premature closure of the fetal ductus arteriosus or due to the role of PGs regulating renal blood vessels may induce fetal renal failure. The use of specific COX-2 inhibitors to block the pro-inflammatory effects of PGs, but allow the constitutive production of PGs for homeostatic mechanisms would appear to have been a logical step forward. However trials with specific COX-2 inhibitors have reported serious fetal side-effects even though they have delayed labor to a certain extent. A meta-analysis of two trials found that COX-2 inhibitors did not show any improved maternal or neonatal outcome compared to non-specific COX inhibitors (163-165). Consistent with meta-analysis a randomized controlled trial of rofecoxib (a specific COX-2 inhibitor) found a significant, but reversible, effect on fetal urine production rate and on the ductus arteriosus, with no reduction in the incidence of early preterm birth (166). In contrast a prospective observational study by the same group of the use of nimesulide in the prevention of preterm labor suggested that it had a significant benefit for high risk cases with no long-term side-effects on the fetus (167). Although in patients with preterm prelabor rupture of membranes the actual incidence of preterm birth was increased by the use of COX-2 inhibitors (168). Therefore at present COX-2 inhibitors are not fulfilling their promise as therapeutic agents for preterm birth. However, it is not clear whether the side-effects of COX-2 inhibitors are due to COX-2-dependent mechanisms or to COX-1 inhibition and their limited ability to differentiate between prostaglandins for maternal and fetal physiological and developmental processes and those that are responsible for labor (169). Secondly selective COX-2 inhibitors also only

block the cyclooxygenase activities while leaving peroxidise activities intact (170). Notably the formation of a COX-1 and COX-2 heterodimer has recently been implicated in the closure of the ductus arteriosus in mice (171), which may explain the observed differential tissue sensitivity to non-steroidal anti-inflammatory drugs (NSAIDs) and potentially offers a new approach to therapeutically exploit the formation of prostaglandins. Future exploration of the role of COX inhibition has been facilitated by the development of a novel selective COX-2 inhibition mouse model (171). These COX-2 Y385F mice in combination with COX-2 knock-out mice, may allow precise assignment of the respective roles of both cyclooxygenase and peroxidise activities in physiological as well as pathophysiological conditions, and help in separating out non-COX NSAID-related activities (170).

### 6.2. Prostaglandin F receptor antagonists: THG113 and THG113.31

$PGF_{2\alpha}$ plays a key role in parturition, and it is an effective smooth muscle contractile agent (172), having a significant stimulatory effect on myometrium contractility. THG113 is a selective, non-competitive, reversible and potent FP receptor antagonist (173, 174), which works by blocking the interaction of FP receptor with $G_{\alpha q}$, consequently inhibiting the further increase in intracellular calcium level (175). A variant of the FP receptor antagonist, THG113.31 has been reported to delay significantly endotoxin-induced preterm labor in mouse (174) and in RU486-induced preterm labor in sheep (49). The mechanism by which THG113.31 inhibits contractions in the uterus remains to be fully elucidated, however, recently, it has been reported that THG113.31 induced a rapid concentration- dependent relaxation on spontaneous human uterine contractions, suggesting that the activation of $BK_{Ca}$ channels, the predominant $K^+$ channel found in the human myometrium (176-180), and not FP receptors were responsible for mediating the relaxatory effect of THG113.31 in the human myometrium (181). At present clinical studies are urgently required to determine whether these agents will play a role in the pharmacological armamentarium for the prevention of preterm birth.

## 7. COX AND PG RECEPTOR KNOCK-OUT MOUSE MODELS

Despite the structural similarities in COX-1 and COX-2, they are encoded differently with COX-1 encoded by a >28 kb gene, in contrast to ~8 kb for COX-2(182, 183), although the organization of exons and introns is similar for the two with the COX-1 locus containing an extra intron, intron-1, that gives rise to COX-3 by alternative splicing (23). To date manipulation of these loci for COX-1 and COX-2 has resulted in knock-out mice being developed with COX-2 knock-out mice exhibiting a major disruption of reproductive capacity and are left infertile (184). In contrast COX-1 knock-out mice although fertile, have delayed spontaneous parturition at term (185), but respond normally to preterm labor induced with LPS, suggesting that the mechanisms of inflammation-induced preterm and term labor differ in mice (126). In a microsomal PGES-1 knock-out mouse LPS induced preterm labor was also not delayed, possibly due to compensation by microsomal PGES-2 (186). $cPLA_2$ knock-out mice have been reported to have small litters and usually dead pups (187, 188), furthermore, $cPLA_2$ knock-out mice do not undergo labor at term, although labor can be achieved/ rescued by administering the progesterone receptor antagonist, RU486 (189). This delayed onset of labor in $cPLA_2$ knock-out female mice is supportive of $cPLA_2$ being an important component in the initial signaling cascade of murine luteolysis and thereby labor, consistent with $cPLA_2$ acting as a direct catalyzing enzyme for AA release with coupling of COX to $PGF_{2\alpha}$ synthesis, luteolysis and labor induction (188), however the direct relevance to humans may be limited due to luteolysis in humans not being attributable to PGs.

Mice lacking each type and subtype of the PG receptors have now been produced with research ongoing to elucidate their respective roles (190). $EP_4$ knock-out mice demonstrate failure of closure of the ductus arteriosus as neonates, suggesting a crucial involvement of this receptor sub-type (190), however, they do not appear to demonstrate any defect in parturition. In contrast mice lacking the FP receptor exhibit a complete failure of parturition at term, although the fetuses are phenotypically normal (191). Much like $cPLA_2$ knock-out mice, both COX-1 and FP receptor knock-out mice fail to initiate spontaneous labor (191, 192), with studies ongoing on the other PG receptor null mice.

## 8. SUMMARY AND PERSPECTIVE

Despite significant research in elucidating the role of PGs in labor, we have yet to fully understand their contribution to the physiology of term and preterm labor. Continued focus on receptor regulation and activation is essential as prostaglandins may be a final common pathway in human parturition. Notably however human labor is a multistep process, with significant redundancy in molecular pathways, therefore the role of other important mediators like cytokines should not be overlooked, and indeed combined therapeutic interventions may be the key to preventing preterm delivery.

## 9. ACKNOWLEDGMENTS

AHK was supported by an Overseas Research Scholarship and a postgraduate research scholarship, Faculty of Medicine, University of Glasgow.

## 10. REFERENCES

1. D. L. Simmons, R. M. Botting and T. Hla: Cyclooxygenase isozymes: the biology of prostaglandin synthesis and inhibition. *Pharmacol Rev*, 56, 387-437 (2004)
2. W. Gibb: The role of prostaglandins in human parturition. *Ann Med*, 30, 235-41 (1998)
3. S. B. Abramson, J. Leszczynska-Piziak and G. Weissmann: Arachidonic acid as a second messenger. Interactions with a GTP-binding protein of human neutrophils. *J Immunol*, 147, 231-6 (1991)

4. Y. Bromberg and E. Pick: Unsaturated fatty acids stimulate NADPH-dependent superoxide production by cell-free system derived from macrophages. *Cell Immunol*, 88, 213-21 (1984)
5. R. Graber, C. Sumida and E. A. Nunez: Fatty acids and cell signal transduction. *J Lipid Mediat Cell Signal*, 9, 91-116 (1994)
6. W. Farrugia, M. A. Aitken, F. van Dunne, M. H. Wong, S. P. Brennecke, K. F. Scott and G. E. Rice: Type II phospholipase A2 in human gestational tissues: subcellular distribution of placental immuno- and catalytic activity. *Biochim Biophys Acta*, 1166, 77-83 (1993)
7. A. Lopez Bernal, G. E. Newman, P. J. Phizackerley, G. Bryant-Greenwood and J. W. Keeling: Human placental phospholipase A2 activity in term and preterm labour. *Eur J Obstet Gynecol Reprod Biol*, 43, 185-92 (1992)
8. T. Okazaki, N. Sagawa, J. E. Bleasdale, J. R. Okita, P. C. MacDonald and J. M. Johnston: Initiation of human parturition: XIII. Phospholipase C, phospholipase A2, and diacylglycerol lipase activities in fetal membranes and decidua vera tissues from early and late gestation. *Biol Reprod*, 25, 103-9 (1981)
9. D. M. Slater, S. Zervou and S. Thornton: Prostaglandins and prostanoid receptors in human pregnancy and parturition. *J Soc Gynecol Investig*, 9, 118-24 (2002)
10. J. D. Clark, L. L. Lin, R. W. Kriz, C. S. Ramesha, L. A. Sultzman, A. Y. Lin, N. Milona and J. L. Knopf: A novel arachidonic acid-selective cytosolic PLA2 contains a Ca(2+)-dependent translocation domain with homology to PKC and GAP. *Cell*, 65, 1043-51 (1991)
11. M. G. Farina, S. Billi, G. Leguizamon, C. Weissmann, T. Guadagnoli, M. L. Ribeiro and A. M. Franchi: Secretory and cytosolic phospholipase A2 activities and expression are regulated by oxytocin and estradiol during labor. *Reproduction*, 134, 355-64 (2007)
12. K. A. Freed, E. K. Moses, S. P. Brennecke and G. E. Rice: Differential expression of type II, IV and cytosolic PLA2 messenger RNA in human intrauterine tissues at term. *Mol Hum Reprod*, 3, 493-9 (1997)
13. M. Koyama, S. Ito, A. Nakajima, K. Shimoya, C. Azuma, N. Suehara, Y. Murata and H. Tojo: Elevations of group II phospholipase A2 concentrations in serum and amniotic fluid in association with preterm labor. *Am J Obstet Gynecol*, 183, 1537-43 (2000)
14. M. Lappas, M. Permezel, H. M. Georgiou and G. E. Rice: Regulation of phospholipase isozymes by nuclear factor-kappaB in human gestational tissues *in vitro*. *J Clin Endocrinol Metab*, 89, 2365-72 (2004)
15. M. Lappas and G. E. Rice: Phospholipase A2 isozymes in pregnancy and parturition. *Prostaglandins Leukot Essent Fatty Acids*, 70, 87-100 (2004)
16. E. K. Moses, K. A. Freed, S. P. Brennecke and G. E. Rice: Distribution of the phospholipase A2 receptor messenger RNA in human gestational tissues. *Placenta*, 19, 35-40 (1998)
17. G. E. Rice: Secretory type II phospholipase A2 and the generation of intrauterine signals. *Reprod Fertil Dev*, 7, 1471-9 (1995)
18. I. Kudo and M. Murakami: Phospholipase A2 enzymes. *Prostaglandins Other Lipid Mediat*, 68-69, 3-58 (2002)
19. M. Lappas, M. Permezel, H. M. Georgiou and G. E. Rice: Type II phospholipase A2 in preterm human gestational tissues. *Placenta*, 22, 64-9 (2001)
20. J. Balsinde, M. A. Balboa, P. A. Insel and E. A. Dennis: Regulation and inhibition of phospholipase A2. *Annu Rev Pharmacol Toxicol*, 39, 175-89 (1999)
21. G. C. Di Renzo, J. M. Johnston, T. Okazaki, J. R. Okita, P. C. MacDonald and J. E. Bleasdale: Phosphatidylinositol-specific phospholipase C in fetal membranes and uterine decidua. *J Clin Invest*, 67, 847-56 (1981)
22. G. A. Bala, N. R. Thakur and J. E. Bleasdale: Characterization of the major phosphoinositide-specific phospholipase C of human amnion. *Biol Reprod*, 43, 704-11 (1990)
23. N. V. Chandrasekharan, H. Dai, K. L. Roos, N. K. Evanson, J. Tomsik, T. S. Elton and D. L. Simmons: COX-3, a cyclooxygenase-1 variant inhibited by acetaminophen and other analgesic/antipyretic drugs: cloning, structure, and expression. *Proc Natl Acad Sci U S A*, 99, 13926-31 (2002)
24. I. Morita: Distinct functions of COX-1 and COX-2. *Prostaglandins Other Lipid Mediat*, 68-69, 165-75 (2002)
25. W. L. Smith, D. L. DeWitt and R. M. Garavito: Cyclooxygenases: structural, cellular, and molecular biology. *Annu Rev Biochem*, 69, 145-82 (2000)
26. W. L. Xie, J. G. Chipman, D. L. Robertson, R. L. Erikson and D. L. Simmons: Expression of a mitogen-responsive gene encoding prostaglandin synthase is regulated by mRNA splicing. *Proc Natl Acad Sci U S A*, 88, 2692-6 (1991)
27. D. L. DeWitt, E. A. Meade and W. L. Smith: PGH synthase isoenzyme selectivity: the potential for safer nonsteroidal antiinflammatory drugs. *Am J Med*, 95, 40S-44S (1993)
28. T. Hla and K. Neilson: Human cyclooxygenase-2 cDNA. *Proc Natl Acad Sci U S A*, 89, 7384-8 (1992)
29. T. Kosaka, A. Miyata, H. Ihara, S. Hara, T. Sugimoto, O. Takeda, E. Takahashi and T. Tanabe: Characterization of the human gene (PTGS2) encoding prostaglandin-endoperoxide synthase 2. *Eur J Biochem*, 221, 889-97 (1994)
30. J. H. Capdevila, J. D. Morrow, Y. Y. Belosludtsev, D. R. Beauchamp, R. N. DuBois and J. R. Falck: The catalytic outcomes of the constitutive and the mitogen inducible isoforms of prostaglandin H2 synthase are markedly affected by glutathione and glutathione peroxidase(s). *Biochemistry*, 34, 3325-37 (1995)
31. R. J. Kulmacz and L. H. Wang: Comparison of hydroperoxide initiator requirements for the cyclooxygenase activities of prostaglandin H synthase-1 and -2. *J Biol Chem*, 270, 24019-23 (1995)
32. M. Murakami, T. Kambe, S. Shimbara and I. Kudo: Functional coupling between various phospholipase A2s and cyclooxygenases in immediate and delayed prostanoid biosynthetic pathways. *J Biol Chem*, 274, 3103-15 (1999)
33. S. Hutten and R. H. Kehlenbach: CRM1-mediated nuclear export: to the pore and beyond. *Trends Cell Biol*, 17, 193-201 (2007)

34. F. Higashino, M. Aoyagi, A. Takahashi, M. Ishino, M. Taoka, T. Isobe, M. Kobayashi, Y. Totsuka, T. Kohgo and M. Shindoh: Adenovirus E4orf6 targets pp32/LANP to control the fate of ARE-containing mRNAs by perturbing the CRM1-dependent mechanism. *J Cell Biol*, 170, 15-20 (2005)
35. B. C. Jang, U. Munoz-Najar, J. H. Paik, K. Claffey, M. Yoshida and T. Hla: Leptomycin B, an inhibitor of the nuclear export receptor CRM1, inhibits COX-2 expression. *J Biol Chem*, 278, 2773-6 (2003)
36. S. Narumiya, Y. Sugimoto and F. Ushikubi: Prostanoid receptors: structures, properties, and functions. *Physiol Rev*, 79, 1193-226 (1999)
37. Y. Kanaoka, K. Fujimori, R. Kikuno, Y. Sakaguchi, Y. Urade and O. Hayaishi: Structure and chromosomal localization of human and mouse genes for hematopoietic prostaglandin D synthase. Conservation of the ancestral genomic structure of sigma-class glutathione S-transferase. *Eur J Biochem*, 267, 3315-22 (2000)
38. Y. Urade and O. Hayaishi: Biochemical, structural, genetic, physiological, and pathophysiological features of lipocalin-type prostaglandin D synthase. *Biochim Biophys Acta*, 1482, 259-71 (2000)
39. R. J. Helliwell, J. A. Keelan, K. W. Marvin, L. Adams, M. C. Chang, A. Anand, T. A. Sato, S. O'Carroll, T. Chaiworapongsa, R. J. Romero and M. D. Mitchell: Gestational age-dependent up-regulation of prostaglandin D synthase (PGDS) and production of PGDS-derived antiinflammatory prostaglandins in human placenta. *J Clin Endocrinol Metab*, 91, 597-606 (2006)
40. Y. Shiki, K. Shimoya, Y. Tokugawa, T. Kimura, M. Koyama, C. Azuma, Y. Murata, N. Eguchi, H. Oda and Y. Urade: Changes of lipocalin-type prostaglandin D synthase level during pregnancy. *J Obstet Gynaecol Res*, 30, 65-70 (2004)
41. M. Murakami and I. Kudo: Recent advances in molecular biology and physiology of the prostaglandin E2-biosynthetic pathway. *Prog Lipid Res*, 43, 3-35 (2004)
42. M. Murakami, H. Naraba, T. Tanioka, N. Semmyo, Y. Nakatani, F. Kojima, T. Ikeda, M. Fueki, A. Ueno, S. Oh and I. Kudo: Regulation of prostaglandin E2 biosynthesis by inducible membrane-associated prostaglandin E2 synthase that acts in concert with cyclooxygenase-2. *J Biol Chem*, 275, 32783-92 (2000)
43. T. Suzuki-Yamamoto, M. Nishizawa, M. Fukui, E. Okuda-Ashitaka, T. Nakajima, S. Ito and K. Watanabe: cDNA cloning, expression and characterization of human prostaglandin F synthase. *FEBS Lett*, 462, 335-40 (1999)
44. K. Watanabe, R. Yoshida, T. Shimizu and O. Hayaishi: Enzymatic formation of prostaglandin F2 alpha from prostaglandin H2 and D2. Purification and properties of prostaglandin F synthetase from bovine lung. *J Biol Chem*, 260, 7035-41 (1985)
45. L. Y. Chen, K. Watanabe and O. Hayaishi: Purification and characterization of prostaglandin F synthase from bovine liver. *Arch Biochem Biophys*, 296, 17-26 (1992)
46. T. Suzuki, Y. Fujii, M. Miyano, L. Y. Chen, T. Takahashi and K. Watanabe: cDNA cloning, expression, and mutagenesis study of liver-type prostaglandin F synthase. *J Biol Chem*, 274, 241-8 (1999)
47. C. W. Xiao, J. M. Liu, J. Sirois and A. K. Goff: Regulation of cyclooxygenase-2 and prostaglandin F synthase gene expression by steroid hormones and interferon-tau in bovine endometrial cells. *Endocrinology*, 139, 2293-9 (1998)
48. C. W. Xiao, B. D. Murphy, J. Sirois and A. K. Goff: Down-regulation of oxytocin-induced cyclooxygenase-2 and prostaglandin F synthase expression by interferon-tau in bovine endometrial cells. *Biol Reprod*, 60, 656-63 (1999)
49. J. J. Hirst, H. C. Parkington, I. R. Young, H. K. Palliser, K. G. Peri and D. M. Olson: Delay of preterm birth in sheep by THG113.31, a prostaglandin F2alpha receptor antagonist. *Am J Obstet Gynecol*, 193, 256-66 (2005)
50. S. Hara, A. Miyata, C. Yokoyama, R. Brugger, F. Lottspeich, V. Ullrich and T. Tanabe: Molecular cloning and expression of prostacyclin synthase from endothelial cells. *Adv Prostaglandin Thromboxane Leukot Res*, 23, 121-3 (1995)
51. K. Ohashi, K. H. Ruan, R. J. Kulmacz, K. K. Wu and L. H. Wang: Primary structure of human thromboxane synthase determined from the cDNA sequence. *J Biol Chem*, 267, 789-93 (1992)
52. R. F. Shen and H. H. Tai: Thromboxanes: synthase and receptors. *J Biomed Sci*, 5, 153-72 (1998)
53. C. Yokoyama, A. Miyata, H. Ihara, V. Ullrich and T. Tanabe: Molecular cloning of human platelet thromboxane A synthase. *Biochem Biophys Res Commun*, 178, 1479-84 (1991)
54. P. Moonen, G. Klok and M. J. Keirse: Immunohistochemical localisation of prostaglandin endoperoxide synthase and prostacyclin synthase in pregnant human myometrium. *Eur J Obstet Gynecol Reprod Biol*, 19, 151-8 (1985)
55. D. Giannoulias, N. Alfaidy, A. C. Holloway, W. Gibb, M. Sun, S. J. Lye and J. R. Challis: Expression of prostaglandin I(2) synthase, but not prostaglandin E synthase, changes in myometrium of women at term pregnancy. *J Clin Endocrinol Metab*, 87, 5274-82 (2002)
56. M. L. Swanson, Z. M. Lei, P. H. Swanson, C. V. Rao, S. Narumiya and M. Hirata: The expression of thromboxane A2 synthase and thromboxane A2 receptor gene in human uterus. *Biol Reprod*, 47, 105-17 (1992)
57. B. Wetzka, W. Schafer, F. Kommoss, H. Bettendorf, R. Nusing, M. Breckwoldt and H. P. Zahradnik: Immunohistochemical localization of thromboxane synthase in human intrauterine tissues. *Placenta*, 15, 389-98 (1994)
58. A. Avdeef, K. J. Box and K. Takacs-Novak: pH-metric log P. 6. Effects of sodium, potassium, and N-CH3-D-glucamine on the octanol-water partitioning of prostaglandins E1 and E2. *J Pharm Sci*, 84, 523-9 (1995)
59. T. J. Roseman and S. H. Yalkowsky: Physicochemical properties of prostaglandin F2 alpha (tromethamine salt): solubility behavior, surface properties, and ionization constants. *J Pharm Sci*, 62, 1680-5 (1973)
60. J. Kang, P. Chapdelaine, P. Y. Laberge and M. A. Fortier: Functional characterization of prostaglandin transporter and terminal prostaglandin synthases during decidualization of human endometrial stromal cells. *Hum Reprod*, 21, 592-9 (2006)
61. V. L. Schuster: Molecular mechanisms of prostaglandin transport. *Annu Rev Physiol*, 60, 221-42 (1998)
62. V. L. Schuster: Prostaglandin transport. *Prostaglandins Other Lipid Mediat*, 68-69, 633-47 (2002)

63. S. Itoh, R. Lu, Y. Bao, J. D. Morrow, L. J. Roberts and V. L. Schuster: Structural determinants of substrates for the prostaglandin transporter PGT. *Mol Pharmacol*, 50, 738-42 (1996)
64. N. Kanai, R. Lu, J. A. Satriano, Y. Bao, A. W. Wolkoff and V. L. Schuster: Identification and characterization of a prostaglandin transporter. Science, 268, 866-9 (1995)
65. Von U.S. Euler and R. Eliasson: Prostaglandins. Academic Press, New York and London (1967)
66. J. Kang, P. Chapdelaine, J. Parent, E. Madore, P. Y. Laberge and M. A. Fortier: Expression of human prostaglandin transporter in the human endometrium across the menstrual cycle. J Clin Endocrinol Metab, 90, 2308-13 (2005)
67. S. K. Banu, J. A. Arosh, P. Chapdelaine and M. A. Fortier: Expression of prostaglandin transporter in the bovine uterus and fetal membranes during pregnancy. Biol Reprod, 73, 230-6 (2005)
68. Y. Bao, M. L. Pucci, B. S. Chan, R. Lu, S. Ito and V. L. Schuster: Prostaglandin transporter PGT is expressed in cell types that synthesize and release prostanoids. Am J Physiol Renal Physiol, 282, F1103-10 (2002)
69. L. C. Bell-Parikh, T. Ide, J. A. Lawson, P. McNamara, M. Reilly and G. A. FitzGerald: Biosynthesis of 15-deoxy-delta12,14-PGJ2 and the ligation of PPARgamma. J Clin Invest, 112, 945-55 (2003)
70. M. Fukushima: Prostaglandin J2--anti-tumour and anti-viral activities and the mechanisms involved. Eicosanoids, 3, 189-99 (1990)
71. Y. Hirata, H. Hayashi, S. Ito, Y. Kikawa, M. Ishibashi, M. Sudo, H. Miyazaki, M. Fukushima, S. Narumiya and O. Hayaishi: Occurrence of 9-deoxy-delta 9,delta 12-13,14-dihydroprostaglandin D2 in human urine. *J Biol Chem*, 263, 16619-25 (1988)
72. R. M. Zusman, D. Spector, B. V. Caldwell, L. Speroff, G. Schneider and P. J. Mulbow: The effect of chronic sodium loading and sodium restriction on plasma prostaglandin A, E, and F concentrations in normal humans. *J Clin Invest*, 52, 1093-8 (1973)
73. M. D. Mitchell, A. P. Flint, J. Bibby, J. Brunt, J. M. Arnold, A. B. Anderson and A. C. Turnbull: Plasma concentrations of prostaglandins during late human pregnancy: influence of normal and preterm labor. *J Clin Endocrinol Metab*, 46, 947-51 (1978)
74. S. M. Sellers, M. D. Mitchell, J. G. Bibby, A. B. Anderson and A. C. Turnbull: A comparison of plasma prostaglandin levels in term and preterm labour. *Br J Obstet Gynaecol*, 88, 362-6 (1981)
75. P. C. MacDonald and M. L. Casey: The accumulation of prostaglandins (PG) in amniotic fluid is an aftereffect of labor and not indicative of a role for PGE2 or PGF2 alpha in the initiation of human parturition. *J Clin Endocrinol Metab*, 76, 1332-9 (1993)
76. H. Giles, M. L. Bolofo, S. J. Lydford and G. R. Martin: A comparative study of the prostanoid receptor profile of 9 alpha 11 beta-prostaglandin F2 and prostaglandin D2. *Br J Pharmacol*, 104, 541-9 (1991)
77. T. Suzuki-Yamamoto, H. Yokoi, Y. Tsuruo, K. Watanabe and K. Ishimura: Identification of prostaglandin F-producing cells in the liver. *Histochem Cell Biol*, 112, 451-6 (1999)
78. M. D. Mitchell, M. C. Chang, T. Chaiworapongsa, H. Y. Lan, R. J. Helliwell, R. Romero and T. A. Sato: Identification of 9alpha,11beta-prostaglandin F2 in human amniotic fluid and characterization of its production by human gestational tissues. *J Clin Endocrinol Metab*, 90, 4244-8 (2005)
79. E. R. Norwitz, P. M. Starkey, A. Lopez Bernal and A. C. Turnbull: Identification by flow cytometry of the prostaglandin-producing cell populations of term human decidua. *J Endocrinol*, 131, 327-34 (1991)
80. E. A. Willman and W. P. Collins: Distribution of prostaglandins E2 and F2 alpha within the foetoplacental unit throughout human pregnancy. *J Endocrinol*, 69, 413-9 (1976)
81. D. M. Olson, K. Skinner and J. R. Challis: Prostaglandin output in relation to parturition by cells dispersed from human intrauterine tissues. *J Clin Endocrinol Metab*, 57, 694-9 (1983)
82. Y. Sadovsky, D. M. Nelson, L. J. Muglia, G. A. Gross, K. C. Harris, A. Koki, J. L. Masferrer and L. M. Olson: Effective diminution of amniotic prostaglandin production by selective inhibitors of cyclooxygenase type 2. *Am J Obstet Gynecol*, 182, 370-6 (2000)
83. A. H. Khan, A. Ashwani, T. Javed, S. M. Nelson and R. J. Carson: Effect of femto to nano molar concentrations of prostaglandin analogues on pregnant rat uterine contractility. *Eur J Pharmacol*, 581, 185-190 (2008)
84. D. J. Crankshaw and R. Dyal: Effects of some naturally occurring prostanoids and some cyclooxygenase inhibitors on the contractility of the human lower uterine segment *in vitro*. *Can J Physiol Pharmacol*, 72, 870-4 (1994)
85. M. P. Embrey and D. L. Morrison: The effect of prostaglandins on human pregnant myometrium *in vitro*. *J Obstet Gynaecol Br Commonw*, 75, 829-32 (1968)
86. R. A. Coleman, W. L. Smith and S. Narumiya: International Union of Pharmacology classification of prostanoid receptors: properties, distribution, and structure of the receptors and their subtypes. *Pharmacol Rev*, 46, 205-29 (1994)
87. M. Bhattacharya, K. Peri, A. Ribeiro-da-Silva, G. Almazan, H. Shichi, X. Hou, D. R. Varma and S. Chemtob: Localization of functional prostaglandin E2 receptors EP3 and EP4 in the nuclear envelope. *J Biol Chem*, 274, 15719-24 (1999)
88. M. Kotani, I. Tanaka, Y. Ogawa, T. Usui, N. Tamura, K. Mori, S. Narumiya, T. Yoshimi and K. Nakao: Structural organization of the human prostaglandin EP3 receptor subtype gene (PTGER3). *Genomics*, 40, 425-34 (1997)
89. A. Schmid, K. H. Thierauch, W. D. Schleuning and H. Dinter: Splice variants of the human EP3 receptor for prostaglandin E2. *Eur J Biochem*, 228, 23-30 (1995)
90. M. Kotani, I. Tanaka, Y. Ogawa, T. Usui, K. Mori, A. Ichikawa, S. Narumiya, T. Yoshimi and K. Nakao: Molecular cloning and expression of multiple isoforms of human prostaglandin E receptor EP3 subtype generated by alternative messenger RNA splicing: multiple second messenger systems and tissue-specific distributions. *Mol Pharmacol*, 48, 869-79 (1995)

91. T. Namba, Y. Sugimoto, M. Negishi, A. Irie, F. Ushikubi, A. Kakizuka, S. Ito, A. Ichikawa and S. Narumiya: Alternative splicing of C-terminal tail of prostaglandin E receptor subtype EP3 determines G-protein specificity. *Nature*, 365, 166-70 (1993)
92. S. Astle, S. Thornton and D. M. Slater: Identification and localization of prostaglandin E2 receptors in upper and lower segment human myometrium during pregnancy. *Mol Hum Reprod*, 11, 279-87 (2005)
93. J. Brodt-Eppley and L. Myatt: Prostaglandin receptors in lower segment myometrium during gestation and labor. *Obstet Gynecol*, 93, 89-93 (1999)
94. G. C. Smith, W. X. Wu and P. W. Nathanielsz: Effects of gestational age and labor on expression of prostanoid receptor genes in baboon uterus. *Biol Reprod*, 64, 1131-7 (2001)
95. J. Senior, K. Marshall, R. Sangha, G. S. Baxter and J. K. Clayton: *In vitro* characterization of prostanoid EP-receptors in the non-pregnant human myometrium. *Br J Pharmacol*, 102, 747-53 (1991)
96. J. Senior, K. Marshall, R. Sangha and J. K. Clayton: *In vitro* characterization of prostanoid receptors on human myometrium at term pregnancy. *Br J Pharmacol*, 108, 501-6 (1993)
97. J. Senior, R. Sangha, G. S. Baxter, K. Marshall and J. K. Clayton: *In vitro* characterization of prostanoid FP-, DP-, IP- and TP-receptors on the non-pregnant human myometrium. *Br J Pharmacol*, 107, 215-21 (1992)
98. T. Matsumoto, N. Sagawa, M. Yoshida, T. Mori, I. Tanaka, M. Mukoyama, M. Kotani and K. Nakao: The prostaglandin E2 and F2 alpha receptor genes are expressed in human myometrium and are down-regulated during pregnancy. *Biochem Biophys Res Commun*, 238, 838-41 (1997)
99. Y. L. Dong and C. Yallampalli: Pregnancy and exogenous steroid treatments modulate the expression of relaxant EP(2) and contractile FP receptors in the rat uterus. *Biol Reprod*, 62, 533-9 (2000)
100. G. N. Europe-Finner, S. Phaneuf, A. M. Tolkovsky, S. P. Watson and A. Lopez Bernal: Down-regulation of G alpha s in human myometrium in term and preterm labor: a mechanism for parturition. *J Clin Endocrinol Metab*, 79, 1835-9 (1994)
101. I. Z. MacKenzie and M. P. Embrey: Intravaginal PGE2 gel prior to labor induction. *Adv Prostaglandin Thromboxane Res*, 8, 1477-81 (1980)
102. T. L. Erkinheimo, K. Saukkonen, K. Narko, J. Jalkanen, O. Ylikorkala and A. Ristimaki: Expression of cyclooxygenase-2 and prostanoid receptors by human myometrium. *J Clin Endocrinol Metab*, 85, 3468-75 (2000)
103. M. Molnar and F. Hertelendy: PGF2 alpha and PGE2 binding to rat myometrium during gestation, parturition, and postpartum. *Am J Physiol*, 258, E740-7 (1990)
104. S. S. Bollapragada, R. Youssef, A. H. Khan, F. Jordan, A. Young, I. A. Greer, J. E. Norman and S. M. Nelson: Prostaglandin system gene expression during term labour. *J Obstet and Gynaecol*, 27, S61 (2007)
105. S. M. Karim, S. D. Sharma, G. M. Filshie, J. A. Salmon and P. A. Ganesan: Termination of pregnancy with prostaglandin analogs. *Adv Biosci*, 9, 811-30 (1973)
106. M. Y. Dawood: Pharmacologic stimulation of uterine contraction. *Semin Perinatol*, 19, 73-83 (1995)
107. S. M. Karim: Physiological role of prostaglandins in the control of parturition and menstruation. *J Reprod Fertil Suppl*, 16, Suppl 16:105-1 (1972)
108. J. W. Aiken: Aspirin and indomethacin prolong parturition in rats: evidence that prostaglandins contribute to expulsion of fetus. *Nature*, 240, 21-5 (1972)
109. J. R. Vane: Inhibition of prostaglandin synthesis as a mechanism of action for aspirin-like drugs. *Nat New Biol*, 231, 232-5 (1971)
110. V. Curbelo, R. Bejar, K. Benirschke and L. Gluck: Premature labor. I. Prostaglandin precursors in human placental membranes. *Obstet Gynecol*, 57, 473-8 (1981)
111. S. M. Karim and J. Devlin: Prostaglandin content of amniotic fluid during pregnancy and labour. *J Obstet Gynaecol Br Commonw*, 74, 230-4 (1967)
112. S. M. Karim and S. D. Sharma: Oral administration of prostaglandins for the induction of labour. *Br Med J*, 1, 260-2 (1971)
113. D. M. Olson, J. E. Mijovic and D. W. Sadowsky: Control of human parturition. *Semin Perinatol*, 19, 52-63 (1995)
114. N. Alfaidy, M. Sun, J. R. Challis and W. Gibb: Expression of membrane prostaglandin E synthase in human placenta and fetal membranes and effect of labor. *Endocrine*, 20, 219-25 (2003)
115. J. W. Meadows, A. L. Eis, D. E. Brockman and L. Myatt: Expression and localization of prostaglandin E synthase isoforms in human fetal membranes in term and preterm labor. *J Clin Endocrinol Metab*, 88, 433-9 (2003)
116. J. W. Meadows, B. Pitzer, D. E. Brockman and L. Myatt: Differential localization of prostaglandin E synthase isoforms in human placental cell types. *Placenta*, 25, 259-65 (2004)
117. S. R. Sooranna, P. L. Grigsby, N. Engineer, Z. Liang, K. Sun, L. Myatt and M. R. Johnson: Myometrial prostaglandin E2 synthetic enzyme mRNA expression: spatial and temporal variations with pregnancy and labour. *Mol Hum Reprod*, 12, 625-31 (2006)
118. S. Astle, R. Newton, S. Thornton, M. Vatish and D. M. Slater: Expression and regulation of prostaglandin E synthase isoforms in human myometrium with labour. *Mol Hum Reprod*, 13, 69-75 (2007)
119. F. Kojima, S. Kato and S. Kawai: Prostaglandin E synthase in the pathophysiology of arthritis. *Fundam Clin Pharmacol*, 19, 255-61 (2005)
120. C. H. Egarter and P. Husslein: Biochemistry of myometrial contractility. *Baillieres Clin Obstet Gynaecol*, 6, 755-69 (1992)
121. J. C. Havelock, P. Keller, N. Muleba, B. A. Mayhew, B. M. Casey, W. E. Rainey and R. A. Word: Human myometrial gene expression before and during parturition. *Biol Reprod*, 72, 707-19 (2005)
122. D. Giannoulias, F. A. Patel, A. C. Holloway, S. J. Lye, H. H. Tai and J. R. Challis: Differential changes in 15-hydroxyprostaglandin dehydrogenase and prostaglandin H synthase (types I and II) in human

pregnant myometrium. *J Clin Endocrinol Metab*, 87, 1345-52 (2002)
123. K. E. Bethin, Y. Nagai, R. Sladek, M. Asada, Y. Sadovsky, T. J. Hudson and L. J. Muglia: Microarray analysis of uterine gene expression in mouse and human pregnancy. *Mol Endocrinol*, 17, 1454-69 (2003)
124. D. M. Slater, W. J. Dennes, J. S. Campa, L. Poston and P. R. Bennett: Expression of cyclo-oxygenase types-1 and -2 in human myometrium throughout pregnancy. *Mol Hum Reprod*, 5, 880-4 (1999)
125. A. R. Belt, J. J. Baldassare, M. Molnar, R. Romero and F. Hertelendy: The nuclear transcription factor NF-kappaB mediates interleukin-1beta-induced expression of cyclooxygenase-2 in human myometrial cells. *Am J Obstet Gynecol*, 181, 359-66 (1999)
126. G. Gross, T. Imamura, S. K. Vogt, D. F. Wozniak, D. M. Nelson, Y. Sadovsky and L. J. Muglia: Inhibition of cyclooxygenase-2 prevents inflammation-mediated preterm labor in the mouse. *Am J Physiol Regul Integr Comp Physiol*, 278, R1415-23 (2000)
127. C. Sparey, S. C. Robson, J. Bailey, F. Lyall and G. N. Europe-Finner: The differential expression of myometrial connexin-43, cyclooxygenase-1 and -2, and Gs alpha proteins in the upper and lower segments of the human uterus during pregnancy and labor. *J Clin Endocrinol Metab*, 84, 1705-10 (1999)
128. R. F. Johnson, C. M. Mitchell, W. B. Giles, A. Bisits and T. Zakar: Mechanisms regulating prostaglandin H2 synthase-2 mRNA level in the amnion and chorion during pregnancy. *J Endocrinol*, 188, 603-10 (2006)
129. A. Fuentes, E. P. Spaziani and W. F. O'Brien: The expression of cyclooxygenase-2 (COX-2) in amnion and decidua following spontaneous labor. *Prostaglandins*, 52, 261-7 (1996)
130. D. Slater, W. Dennes, R. Sawdy, V. Allport and P. Bennett: Expression of cyclo-oxygenase types-1 and -2 in human fetal membranes throughout pregnancy. *J Mol Endocrinol*, 22, 125-30 (1999)
131. J. J. Hirst, F. J. Teixeira, T. Zakar and D. M. Olson: Prostaglandin endoperoxide-H synthase-1 and -2 messenger ribonucleic acid levels in human amnion with spontaneous labor onset. *J Clin Endocrinol Metab*, 80, 517-23 (1995)
132. T. Lindstrom and P. Bennett: Transcriptional regulation of genes for enzymes of the prostaglandin biosynthetic pathway. *Prostaglandins Leukot Essent Fatty Acids*, 70, 115-35 (2004)
133. Y. Lee, V. Allport, A. Sykes, T. Lindstrom, D. Slater and P. Bennett: The effects of labour and of interleukin 1 beta upon the expression of nuclear factor kappa B related proteins in human amnion. *Mol Hum Reprod*, 9, 213-8 (2003)
134. J. A. Loudon, C. L. Elliott, F. Hills and P. R. Bennett: Progesterone represses interleukin-8 and cyclo-oxygenase-2 in human lower segment fibroblast cells and amnion epithelial cells. *Biol Reprod*, 69, 331-7 (2003)
135. J. C. Condon, D. B. Hardy, K. Kovaric and C. R. Mendelson: Up-regulation of the progesterone receptor (PR)-C isoform in laboring myometrium by activation of nuclear factor-kappaB may contribute to the onset of labor through inhibition of PR function. *Mol Endocrinol*, 20, 764-75 (2006)
136. J. C. Condon, P. Jeyasuria, J. M. Faust, J. W. Wilson and C. R. Mendelson: A decline in the levels of progesterone receptor coactivators in the pregnant uterus at term may antagonize progesterone receptor function and contribute to the initiation of parturition. *Proc Natl Acad Sci U S A*, 100, 9518-23 (2003)
137. V. C. Allport, D. Pieber, D. M. Slater, R. Newton, J. O. White and P. R. Bennett: Human labour is associated with nuclear factor-kappaB activity which mediates cyclo-oxygenase-2 expression and is involved with the 'functional progesterone withdrawal'. *Mol Hum Reprod*, 7, 581-6 (2001)
138. R. T. Okita and J. R. Okita: Prostaglandin-metabolizing enzymes during pregnancy: characterization of NAD(+)-dependent prostaglandin dehydrogenase, carbonyl reductase, and cytochrome P450-dependent prostaglandin omega-hydroxylase. *Crit Rev Biochem Mol Biol*, 31, 101-26 (1996)
139. P. Y. Cheung and J. R. Challis: Prostaglandin E2 metabolism in the human fetal membranes. *Am J Obstet Gynecol*, 161, 1580-5 (1989)
140. S. Nakla, K. Skinner, B. F. Mitchell and J. R. Challis: Changes in prostaglandin transfer across human fetal membranes obtained after spontaneous labor. *Am J Obstet Gynecol*, 155, 1337-41 (1986)
141. M. H. Sullivan, C. K. Roseblade and M. G. Elder: Metabolism of prostaglandin E2 on the fetal and maternal sides of intact fetal membranes. *Acta Obstet Gynecol Scand*, 70, 425-7 (1991)
142. J. A. McCoshen, D. R. Hoffman, J. V. Kredentser, C. Araneda and J. M. Johnston: The role of fetal membranes in regulating production, transport, and metabolism of prostaglandin E2 during labor. *Am J Obstet Gynecol*, 163, 1632-40 (1990)
143. R. K. Sangha, J. C. Walton, C. M. Ensor, H. H. Tai and J. R. Challis: Immunohistochemical localization, messenger ribonucleic acid abundance, and activity of 15-hydroxyprostaglandin dehydrogenase in placenta and fetal membranes during term and preterm labor. *J Clin Endocrinol Metab*, 78, 982-9 (1994)
144. C. A. van Meir, S. G. Matthews, M. J. Keirse, M. M. Ramirez, A. Bocking and J. R. Challis: 15-hydroxyprostaglandin dehydrogenase: implications in preterm labor with and without ascending infection. *J Clin Endocrinol Metab*, 82, 969-76 (1997)
145. J. R. G. Challis, S. G. Matthews, W. Gibb and S. J. Lye: Endocrine and paracrine regulation of birth at term and preterm. *Endocr Rev*, 21, 514-50 (2000)
146. P. R. Bennett, G. V. Chamberlain, L. Patel, M. G. Elder and L. Myatt: Mechanisms of parturition: the transfer of prostaglandin E2 and 5-hydroxyeicosatetraenoic acid across fetal membranes. *Am J Obstet Gynecol*, 162, 683-7 (1990)
147. N. L. Brown, S. A. Alvi, M. G. Elder, P. R. Bennett and M. H. Sullivan: Interleukin-1beta and bacterial endotoxin change the metabolism of prostaglandins E2 and F2alpha in intact term fetal membranes. *Placenta*, 19, 625-30 (1998)
148. F. A. Patel and J. R. Challis: Cortisol/progesterone antagonism in regulation of 15-hydroxysteroid dehydrogenase activity and mRNA levels in human chorion

and placental trophoblast cells at term. *J Clin Endocrinol Metab*, 87, 700-8 (2002)
149. J. R. Challis, F. A. Patel and F. Pomini: Prostaglandin dehydrogenase and the initiation of labor. *J Perinat Med*, 27, 26-34 (1999)
150. S. J. Lye, J. Mitchell, N. Nashman, A. Oldenhof, R. Ou, O. Shynlova and L. Langille: Role of mechanical signals in the onset of term and preterm labor. *Front Horm Res*, 27, 165-78 (2001)
151. F. K. Kloeck and H. Jung: *In vitro* release of prostaglandins from the human myometrium under the influence of stretching. *Am J Obstet Gynecol*, 115, 1066-9 (1973)
152. Y. Manabe, T. Okazaki and A. Takahashi: Prostaglandins E and F in amniotic fluid during stretch-induced cervical softening and labor at term. *Gynecol Obstet Invest*, 15, 343-50 (1983)
153. Y. Manabe, S. Yoshimura, T. Mori and T. Aso: Plasma levels of 13,14-dihydro-15-keto prostaglandin F2 alpha, estrogens, and progesterone during stretch-induced labor at term. *Prostaglandins*, 30, 141-52 (1985)
154. G. Leguizamon, J. Smith, H. Younis, D. M. Nelson and Y. Sadovsky: Enhancement of amniotic cyclooxygenase type 2 activity in women with preterm delivery associated with twins or polyhydramnios. *Am J Obstet Gynecol*, 184, 117-22 (2001)
155. S. R. Sooranna, Y. Lee, L. U. Kim, A. R. Mohan, P. R. Bennett and M. R. Johnson: Mechanical stretch activates type 2 cyclooxygenase via activator protein-1 transcription factor in human myometrial cells. *Mol Hum Reprod*, 10, 109-13 (2004)
156. A. R. Mohan, S. R. Sooranna, T. M. Lindstrom, M. R. Johnson and P. R. Bennett: The effect of mechanical stretch on cyclooxygenase type 2 expression and activator protein-1 and nuclear factor-kappaB activity in human amnion cells. *Endocrinology*, 148, 1850-7 (2007)
157. G. Jenkin: Oxytocin and prostaglandin interactions in pregnancy and at parturition. *J Reprod Fertil Suppl*, 45, 97-111 (1992)
158. M. Molnar, J. Rigo, Jr., R. Romero and F. Hertelendy: Oxytocin activates mitogen-activated protein kinase and up-regulates cyclooxygenase-2 and prostaglandin production in human myometrial cells. *Am J Obstet Gynecol*, 181, 42-9 (1999)
159. M. Ivanisevic, J. Djelmis and D. Bukovic: Review on prostaglandin and oxytocin activity in preterm labor. *Coll Antropol*, 25, 687-94 (2001)
160. H. C. Brummer: Further studies on the interaction between prostaglandins and syntocinon on the isolated pregnant human myometrium. *J Obstet Gynaecol Br Commonw*, 79, 526-30 (1972)
161. G. E. Rice: Cytokines and the initiation of parturition. *Front Horm Res*, 27, 113-46 (2001)
162. H. M. Todd, V. L. Dundoo, W. R. Gerber, C. A. Cwiak, J. J. Baldassare and F. Hertelendy: Effect of cytokines on prostaglandin E2 and prostacyclin production in primary cultures of human myometrial cells. *J Matern Fetal Med*, 5, 161-7 (1996)
163. J. King, V. Flenady, S. Cole and S. Thornton: Cyclo-oxygenase (COX) inhibitors for treating preterm labour. *Cochrane Database Syst Rev*CD001992 (2005)
164. R. J. Sawdy, S. Lye, N. M. Fisk and P. R. Bennett: A double-blind randomized study of fetal side effects during and after the short-term maternal administration of indomethacin, sulindac, and nimesulide for the treatment of preterm labor. *Am J Obstet Gynecol*, 188, 1046-51 (2003)
165. C. S. Stika, G. A. Gross, G. Leguizamon, S. Gerber, R. Levy, A. Mathur, L. M. Bernhard, D. M. Nelson and Y. Sadovsky: A prospective randomized safety trial of celecoxib for treatment of preterm labor. *Am J Obstet Gynecol*, 187, 653-60 (2002)
166. K. M. Groom, A. H. Shennan, B. A. Jones, P. Seed and P. R. Bennett: TOCOX--a randomised, double-blind, placebo-controlled trial of rofecoxib (a COX-2-specific prostaglandin inhibitor) for the prevention of preterm delivery in women at high risk. *Bjog*, 112, 725-30 (2005)
167. R. J. Sawdy, K. M. Groom and P. R. Bennett: Experience of the use of nimesulide, a cyclo-oxygenase-2 selective prostaglandin synthesis inhibitor, in the prevention of preterm labour in 44 high-risk cases. *J Obstet Gynaecol*, 24, 226-9 (2004)
168. M. M. Slattery and J. J. Morrison: Preterm delivery. *Lancet*, 360, 1489-97 (2002)
169. J. A. Loudon, K. M. Groom and P. R. Bennett: Prostaglandin inhibitors in preterm labour. *Best Pract Res Clin Obstet Gynaecol*, 17, 731-44 (2003)
170. Y. Yu and C. D. Funk: A novel genetic model of selective COX-2 inhibition: comparison with COX-2 null mice. *Prostaglandins Other Lipid Mediat*, 82, 77-84 (2007)
171. Y. Yu, J. Fan, X. S. Chen, D. Wang, A. J. Klein-Szanto, R. L. Campbell, G. A. FitzGerald and C. D. Funk: Genetic model of selective COX2 inhibition reveals novel heterodimer signaling. *Nat Med*, 12, 699-704 (2006)
172. D. H. Wright, D. Abran, M. Bhattacharya, X. Hou, S. G. Bernier, A. Bouayad, J. C. Fouron, A. Vazquez-Tello, M. H. Beauchamp, R. I. Clyman, K. Peri, D. R. Varma and S. Chemtob: Prostanoid receptors: ontogeny and implications in vascular physiology. *Am J Physiol Regul Integr Comp Physiol*, 281, R1343-60 (2001)
173. D. M. Olson, D. B. Zaragoza, M. C. Shallow, J. L. Cook, B. F. Mitchell, P. Grigsby and J. Hirst: Myometrial activation and preterm labour: evidence supporting a role for the prostaglandin F receptor--a review. *Placenta*, 24 Suppl A, S47-54 (2003)
174. K. G. Peri, C. Quiniou, X. Hou, D. Abran, D. R. Varma, W. D. Lubell and S. Chemtob: THG113: a novel selective FP antagonist that delays preterm labor. *Semin Perinatol*, 26, 389-97 (2002)
175. D. M. Olson and C. Ammann: Role of the prostaglandins in labour and prostaglandin receptor inhibitors in the prevention of preterm labour. *Front Biosci*, 12, 1329-43 (2007)
176. K. Anwer, C. Oberti, G. J. Perez, N. Perez-Reyes, J. K. McDougall, M. Monga, B. M. Sanborn, E. Stefani and L. Toro: Calcium-activated K+ channels as modulators of human myometrial contractile activity. *Am J Physiol*, 265, C976-85 (1993)
177. R. N. Khan, B. Matharoo-Ball, S. Arulkumaran and M. L. Ashford: Potassium channels in the human myometrium. *Exp Physiol*, 86, 255-64 (2001)
178. R. N. Khan, S. K. Smith and M. L. Ashford: Contribution of calcium-sensitive potassium channels to

NS1619-induced relaxation in human pregnant myometrium. *Hum Reprod*, 13, 208-13 (1998)
179. R. N. Khan, S. K. Smith, J. J. Morrison and M. L. Ashford: Properties of large-conductance K+ channels in human myometrium during pregnancy and labour. *Proc Biol Sci*, 251, 9-15 (1993)
180. R. Schubert and M. T. Nelson: Protein kinases: tuners of the BKCa channel in smooth muscle. *Trends Pharmacol Sci*, 22, 505-12 (2001)
181. H. C. Doheny, M. J. O'Reilly, D. J. Sexton and J. J. Morrison: THG113.31, a specific PGF2alpha receptor antagonist, induces human myometrial relaxation and BKCa channel activation. *Reprod Biol Endocrinol*, 5, 10 (2007)
182. S. B. Appleby, A. Ristimaki, K. Neilson, K. Narko and T. Hla: Structure of the human cyclo-oxygenase-2 gene. *Biochem J*, 302 ( Pt 3), 723-7 (1994)
183. B. S. Fletcher, D. A. Kujubu, D. M. Perrin and H. R. Herschman: Structure of the mitogen-inducible TIS10 gene and demonstration that the TIS10-encoded protein is a functional prostaglandin G/H synthase. *J Biol Chem*, 267, 4338-44 (1992)
184. H. Lim, B. C. Paria, S. K. Das, J. E. Dinchuk, R. Langenbach, J. M. Trzaskos and S. K. Dey: Multiple female reproductive failures in cyclooxygenase 2-deficient mice. *Cell*, 91, 197-208 (1997)
185. C. D. Loftin, D. B. Trivedi and R. Langenbach: Cyclooxygenase-1-selective inhibition prolongs gestation in mice without adverse effects on the ductus arteriosus. *J Clin Invest*, 110, 549-57 (2002)
186. K. Kubota, T. Kubota, D. Kamei, M. Murakami, I. Kudo, T. Aso and I. Morita: Change in prostaglandin E synthases (PGESs) in microsomal PGES-1 knockout mice in a preterm delivery model. *J Endocrinol*, 187, 339-45 (2005)
187. J. V. Bonventre, Z. Huang, M. R. Taheri, E. O'Leary, E. Li, M. A. Moskowitz and A. Sapirstein: Reduced fertility and postischaemic brain injury in mice deficient in cytosolic phospholipase A2. *Nature*, 390, 622-5 (1997)
188. N. Uozumi, K. Kume, T. Nagase, N. Nakatani, S. Ishii, F. Tashiro, Y. Komagata, K. Maki, K. Ikuta, Y. Ouchi, J. Miyazaki and T. Shimizu: Role of cytosolic phospholipase A2 in allergic response and parturition. *Nature*, 390, 618-22 (1997)
189. D. J. Dudley, D. W. Branch, S. S. Edwin and M. D. Mitchell: Induction of preterm birth in mice by RU486. *Biol Reprod*, 55, 992-5 (1996)
190. F. Ushikubi, Y. Sugimoto, A. Ichikawa and S. Narumiya: Roles of prostanoids revealed from studies using mice lacking specific prostanoid receptors. *Jpn J Pharmacol*, 83, 279-85 (2000)
191. Y. Sugimoto, A. Yamasaki, E. Segi, K. Tsuboi, Y. Aze, T. Nishimura, H. Oida, N. Yoshida, T. Tanaka, M. Katsuyama, K. Hasumoto, T. Murata, M. Hirata, F. Ushikubi, M. Negishi, A. Ichikawa and S. Narumiya: Failure of parturition in mice lacking the prostaglandin F receptor. *Science*, 277, 681-3 (1997)
192. R. Langenbach, S. G. Morham, H. F. Tiano, C. D. Loftin, B. I. Ghanayem, P. C. Chulada, J. F. Mahler, C. A. Lee, E. H. Goulding, K. D. Kluckman, H. S. Kim and O. Smithies: Prostaglandin synthase 1 gene disruption in mice reduces arachidonic acid-induced inflammation and indomethacin-induced gastric ulceration. *Cell*, 83, 483-92 (1995)
193. H. N. Jabbour and K. J. Sales: Prostaglandin receptor signalling and function in human endometrial pathology. *Trends Endocrinol Metab*, 15, 398-404 (2004)

**Key Words:** Prostaglandins, labor, COX-1, COX-2, PTGS, Prostaglandin Receptors, Prostaglandin Dehydrogenase, Review

**Send correspondence to:** Abdul H. Khan, Reproductive and Maternal Medicine, Division of Developmental Medicine, University of Glasgow, Glasgow Royal Infirmary, 10 Alexandra Parade, Glasgow, G31 2ER, U.K., Tel: 441412114376, Fax: 44141211552 073, E-mail: khan.abdulh@gmail.com

http://www.bioscience.org/current/vol13.htm