BIOCELL
2004, 28(1): 1-11

ISSN 0327 - 9545
PRINTED IN ARGENTINE

Review

Uterine mast cells: A new hypothesis to understand how we are born

M. Isolde Rudolph[1], I. Gina Rojas[2], and Alicia B. Penissi[3]

1. Departamento de Farmacología, Facultad de Ciencias Biológicas, Universidad de Concepción, Casilla 160-C, Concepción, Chile.
2. Departamento de Estomatología Quirúrgica, Facultad de Odontología, Universidad de Concepción, Casilla 160-C, Concepción, Chile.
3. Instituto de Histología y Embriología " Dr. Mario H Burgos" (IHEM-CONICET), Facultad de Ciencias Médicas, Universidad Nacional de Cuyo, Casilla de Correo 56, (5500) Mendoza, Argentina.

Key words: uterine mast cells, labor, myometrium contractions, cervical ripening

ABSTRACT: Birth is the result of complex, well-defined, and coordinated events, that are tightly regulated by endocrine, nervous, and immune responses, and take place primarily in the female reproductive tract. Various mechanisms and mediators involved in pregnancy, labor, and delivery, are highly conserved among different mammalian species and mast cells emerge as potential and crucial participants in these processes, as it is discussed in this review.

I. An overview of human pregnancy, labor and delivery

Optimal well-being of the neonate depends on the exact timing of delivery. Infant immaturity, which results from premature birth is one of the greatest worldwide health problems to be solved today. In fact, premature birth contributes significantly to perinatal death and neonatal morbidity. As a risk factor, immaturity can strongly contribute to neurologic, psychiatric and pulmonary disorders, which may become evident at preschool and adolescent years of premature children. Treatment of these alterations requires high demand of economic and human resources at health centers around the world. On the other hand, postdate infants have an increased incidence of intrauterine demise, and it is not uncommon to end the risk of survival of a postdate fetus through a cesarean delivery, which also implies higher costs for the healthcare system. However, prevention and management of premature labor is more difficult to attain. This has caused obstetricians to develop increased interest and demand for new approaches and methodologies to diagnose and prevent premature labor, leading to more basic and clinical research in this area. In addition, more educational programs aiming at better care and integration of the mother and child, as well as enhanced nutritional support and control of the fetus during prenatal life are being implemented.

Address correspondence to: Dr. M. Isolde Rudolph. Departamento de Farmacología, Facultad de Ciencias Biológicas, Universidad de Concepción, Casilla 160-C, 4080831 Concepción, CHILE.
E-mail: mrudolph@udec.cl
Received on December 19, 2003. Accepted on February 10, 2004

As a clinical procedure, normal labor is well characterized. Nevertheless, knowledge about the specific physiological signals initiating myometrium contractions and evoking cervix maturation are not fully understood. In fact, the development of effective therapeutic procedures to prevent or reduce the incidence of premature birth depends on understanding the mechanisms that control the interaction between endocrine, biochemical, neural and immune responses regulating pregnancy, labor and delivery. During 99% pregnancy, the myometrium is quiescent, without contractions, maintaining a basal tone for optimal blood irrigation to the fetus. Contractions start at the end of pregnancy when the fetus is fully developed, and they seem to be stimulated by a complex mechanism of positive feedback. Therefore once started, contractions become very difficult to stop by the therapeutic methods available up to date. Simultaneously with increased uterine contractions, the cervix, which normally is rigid and closed, starts to soften and dilate allowing the fetus, which is being expelled from the uterus by the vigorous contractions, to descend.

It has been difficult to indicate a common pathway for the currently accepted hypothesis of how birth occurs on the base of studies performed in different mammalian species. However, in spite of the differences, some cellular and molecular mechanisms have been found to be conserved among different species, and they may contribute to a better insight of the physiological processes involved in human labor and delivery.

The aim of this review is to summarize biochemical, pathological and pharmacological evidences that support the theory that uterine mast cells (MCs) play a central role in the initiation and progression of uterine contractions and cervical ripening. Information is also provided to understand how uterine mast cells can be activated by the well known endogenous labor inducers: estrogens, oxytocin and corticotropin-releasing hormone (CRH).

II. Mast Cells

Identified by Paul Ehrlich in the late 1870s, MCs are important effector cells of the immune system, whose primary function is to stimulate host defense against potentially harmful agents, such as pathogens and other environmental insults (Galli *et al.*, 1999). MCs are ubiquitous throughout the body, with the exception of solid bone and cartilage, and are found at higher density in areas of environmental exposure such as skin and mucosa. Besides its role in host defense, the MC has been recognized as a versatile effector cell that serves as a key link for the interactions among nervous, immune and endocrine systems (Cocchiara *et al.*, 1997b).

Although MCs are most often associated with pathological disorders related to IgE-dependent immediate hypersensitivity, like allergy and asthma, they have important functions in many other biological responses. These include normal and altered tissue remodeling, blood vessel formation, clearance of parasite and bacterial infections, cell proliferation, heart disease, wound healing, and acute and chronic inflammatory disorders (Galli *et al.*, 1987; Metcalfe *et al.*, 1997; Penissi *et al.*, 2003b; Puxeddu *et al.*, 2003). In addition, the potential of MCs to release their granular contents, which involve a diverse array of uterotonic and inflammatory mediators, makes them powerful mediator cells with the potential to regulate uterine contractility and cervical ripening, respectively (Rudolph *et al.*, 1992; Rudolph *et al.*, 1993; Rudolph *et al.*, 1997a; Shum *et al.*, 2002).

Human MCs are derived from pluripotent stem cells of hematopoietic origin, that leave the bone marrow and circulate in peripheral blood as immature committed progenitors (Galli *et al.*, 1994). MCs continue to differentiate and mature in diverse peripheral tissues where they acquire special characteristics influenced by the tissue type and the microenvironmental conditions of the host organ, a phenomenon that leads to MC heterogeneity. MCs express the receptor for stem cell factor (SCF) receptor or c-kit, that binds to SCF, the single most important growth factor for MCs, which promotes their growth, maturation, differentiation, survival, migration, and degranulation (Galli *et al.,* 1994; Lemura *et al.*, 1994).

Human MCs can be classified into two subpopulations according to their content of specific serine proteases known as tryptase and chymase. MCs expressing predominantly tryptase are designated as $MC_{T,}$ those expressing both, tryptase and chymase, are designated as MC_{TC}. MC_T are key for inflammation and the immune response, with a primary role in host defense. They are usually found in anatomical sites of high exposure to pathogens, such as mucosal surfaces, blood vessels, nerves, glands, serosal cavities, and epithelial surfaces. On the other hand, MC_{TC} predominate in the gastrointestinal tract, skin, synovium, and subcutaneous tissues, contributing more to tissue remodeling and angiogenesis, rather than to host defense. Increased number of MC_T are found in allergic and parasitic diseases, and surrounding some tumors, whereas MC_{TC} predominate in fibrotic alterations (Church and Levy-Schaffer, 1997;

Cabanillas-Saez *et al.*, 2002). The murine counterparts of these subtypes have been classified according to tissue distribution, namely mucosal MCs (MMC), present in the epithelium and in the sub-epithelial lamina propria, and connective tissue MCs (CTMC), found predominantly in submucosa. MMC and CTMC are histochemically, ultrastructurally, and functionally different. They also vary in their pharmacological properties, for example in their sensitivity to MC secretagogues, such as Compound 48/80, and anti-allergic compounds, such as Sodium Cromoglicate (Wershil and Galli, 1991; McNeil and Austen, 1995; Stenton *et al.*, 1998).

The activity of mature MCs can be regulated by a variety of compounds. Stimulating compounds induce MCs to differentially release and synthesize a heterogeneous group of mediators that differ in their potency and biological activities. These mediators are pleiotropic and redundant, i.e., each mediator has more than one function, and they may overlap in their biologic activity. These MC mediators can increase vasodilation and vasopermeability, as well as evoke smooth muscle contractions, tissue remodeling, wound healing, and gastrointestinal protection (Rudolph *et al.*, 1992; Rudolph *et al.*, 1993; Armetti *et al.*, 1999; Penissi *et al.*, 2003 a, b; Puxeddu *et al.*, 2003). Therefore, MCs may elicit a myriad of pro-inflammatory and anti-inflammatory effects, depending on the tissue microenvironment and type of stimuli.

MC mediators are classified in three groups, depending on their origin and biological activities (Nilsson and Schwartz, 1995). The first group are the preformed secretory granule-associated mediators, such as histamine, serotonin (5HT), proteases, proteoglycans, cytokines, and other individual mediators. They are found in a crystalline complex ionically bound to a proteoglycan matrix, which are exocytosed to the extracellular media within seconds or minutes after MC activation. The second group are the newly synthesized arachidonic acid-derived mediators, such as prostaglandins D_2 and E_2, leukotriene C, thromboxane B_2, and platelet activating factor (PAF). These mediators are also key participants in acute inflammation, although they can also evoke anti-inflamatory responses (Harris and Phipps, 2002). Synthesis and release of these compounds start within minutes, as a consequence of phospholipase A_2 and cyclooxygenase (COX)-1 and -2 activation within MC lipid and granule membrane bodies (Schmauder-Chock and Chock, 1989; Massey *et al.*, 1991). The third group is composed of newly synthesized cytokines, chemokines and growth factors, which are secreted at later stages. These mediators are crucial for the late phase reaction, that develops in a couple of hours after the initiation of the inflammatory response. Among this group of MC-derived mediators is the pro-inflammatory cytokine, TNF-α, which is critical for inflammatory and allergic reactions. TNF-α promotes leukocyte recruitment and activation by stimulating endothelial expression of adhesion molecules and chemokines, via up-regulation of the transcription factor NF-kB (Tkaczyk *et al.*, 1996). Studies by Gordon & Galli (1990) showed that, unlike T cells and macrophages, MCs constitutively express and store TNF-α. Furthermore, *de novo* TNF-α production is significantly greater in MCs as compared to macrophages (Gordon and Galli, 1990). MCs also produce IL-5, the chemokines IL-8 and RANTES, and granulocyte-macrophage colony-stimulating factor (GM-CSF), which are involved in leukocyte priming, activation and recruitment. GM-CSF is also involved in priming of eosinophils and in increasing their life-span (Okayama *et al.*, 1997). Among the anti-inflammatory cytokines produced by MCs are IL-4, IL-5, and IL-13 (Brightling *et al.*, 2003). It is worth mentioning that MCs also produce nerve growth factor (NGF) (Leon *et al.*, 1994), a key neurotrophic factor, that stimulates the inflammatory response. For further information on MC mediators and function reviews by Metcalfe (1997), Galli *et al.*(1999), and Krishnaswamy *et al.* (2001) are recommended.

II.1. Mast cells in the uterine microenvironment

In myometrium, endometrium, and cervix, MCs are found in high numbers, specially localized around blood and lymph vessels, and nerves (Jeziorska *et al.*, 1995; Mori *et al.*, 1997; Cabanillas-Saez *et al.*, 2002). MCs are also found in close association with myometrial smooth muscle cells, both in mice and humans (Padilla *et al.*, 1990; Rudolph *et al.*, 1993). Regarding MC phenotypes, both MC_T or "immune system-related" MCs, and MC_{TC} or "non-immune system related" MCs are found in similar proportion. Furthermore, uterine MCs share morphological features common to skin and lung MCs (Massey *et al.*, 1991).

In mouse myometrium, MC number and histamine content, a marker of MC concentration in uterine tissue, change according to the stage of the estrous cycle, being higher during diestrus than in estrus, suggesting that MC activity may be regulated by endocrine mechanisms (Padilla *et al.*, 1990; Rudolph *et al.*, 1997b). In addition, the myometrium of pregnant mice show a

steady increase in MC density and histamine content, specially during the second half of pregnancy. Peak MC density and histamine levels are reached by the end of the gestational period (Padilla *et al*., 1990; Tabb, 1994). Histamine content returns to basal, pre-gestational levels after delivery, suggesting a massive activation of MCs during parturition (Padilla *et al*., 1990).

Activation of uterine MCs evokes uterine contractions, through the release of histamine, 5HT and lipid-mediators, such as PGD_2, leukotrienes and PAF (Massey et al., 1991; Rudolph *et al*., 1997a,b). Histamine and 5HT contract the human uterus which becomes hyperresponsive to these compounds by the end of pregnancy (Cruz *et al.*, 1989; Bytautiene *et al.*, 2003). Histamine and 5HT, in combination with $PGF_{2\alpha}$, potentiate each other in evoking smooth muscle contractions of the myometrium. Thus, MCs by making these endogenous compounds available in the uterine tissue in a concurrent fashion, potentiate their effects as compared to their individual actions (Rudolph *et al*., 1992; Rudolph *et al*., 1993).

Uterine MCs release several cytokines, growth factors, and chemokines that are crucial for stimulating inflammation (Costa *et al.*, 1994; Cocchiara *et al.*, 1997a). An important pro-inflammatory cytokine is TNF-α, which stimulates expression of E-selectin, an adhesion molecule that is up-regulated in the vascular endothelium of the myometrium of women in labor (Walsh *et al.*, 1991; Schmith *et al.*, 2002). Up-regulation of E-selectin expression has been linked to leukocyte accumulation in the myometrium during labor (Thompson *et al.*, 1999). These findings, added to the condition of estrogen predominancy during parturition, have led to the hypothesis that the TNF-α that initiates the inflammatory cascade in the uterus may be produced by infiltrating MCs (Roby and Hunt, 1995; Hunt *et al.*, 1997). Therefore, it is highly possible that MCs may be key contributors to uterine "inflammatory-state", both at the myometrium and cervix during human parturition (Bokstrom *et al.*, 1997; Thompson *et al*., 1999).

Inflammation leads to a microenvironment that favors uterine contractility. Pro-inflammatory enzymes, such as COXs and nitric oxide synthases (NOS), are up-regulated and activated, increasing prostaglandin and nitric oxide production (Sautebin *et al.*, 1995). MC activation and leukocyte accumulation are important sources of reactive oxygen and nitrogen species (Evans *et al.*, 1995; Huang *et al.*, 1995). High nitric oxide (NO) concentration, which may occur under estrogen predominance, stimulates both uterine MCs to release uterotonic compounds, and smooth muscle myometrial cells to synthesize PGE_2 (Martinez *et al.*, 1999; Motta *et al.*, 1999). High concentrations of NO in an inflammatory environment, as it occurs in uterine cervix during labor, also favor cervical ripening (Chwalisz and Garfield, 1998). It is possible that under such conditions, the high concentration of oxygen and nitrogen reactive species may favor the synthesis of peroxynitrite, a strong MC activator, as it has been shown for other tissues (Konopka *et al.*, 2001). Finally, MC activation releases trypase, a serine-protease, that could favor cervical ripening through its collagenolytic activity (Fajardo and Pejler, 2003).

Uterine MCs, like peritoneal and genito-urinary MCs, are sensitive to estrogen (Pang *et al.*, 1995). Results by our group showed that uterine contractions evoked by estrogen predominance are mediated by MC activation (Martinez *et al*., 1999). The mechanism of estrogen action on uterine MCs is believed to involve MC sensitization for further activation by other endogenous stimuli (Cocchiara *et al.*, 1992).

Uterine MCs are also sensitive to the action of oxytocin, which in turn inhibits the uptake of 5HT into MCs (Rudolph *et al.* 1998). Finally, studies by Madhappan et al. (Madhappan *et al.*, 2003) suggest an effect of CRH on uterine MC activation as a mechanism to evoke uterine contractions, whereas the relaxant hormones, relaxin and progesterone, are believed to inhibit uterine MC activation (Masini *et al.*, 1994; Huang *et al*., 1995).

III. Effects of the estrogen/progesterone ratio on the uterus and mast cell activation

The balance between the plasmatic concentrations of estrogen and progesterone are critical for pregnancy prolongation or termination. Alterations in this endocrine balance determine labor onset or inhibition. In domestic mammalian species, such as cow, mouse, rat, sheep, guinea pig, rabbit and sow, progesterone withdrawal stimulates the onset of labor. In humans and in sub-primate species, such as macaque or baboon, plasmatic progesterone concentration increases during pregnancy. However, by the end of pregnancy, the rate of estrogen synthesis is greater than that of progesterone production. Therefore, as a result, estrogens predominate over progesterone (Pasqualini and Kincl, 1985).

Progesterone is believed to exert an overall control of uterine quiescence and cervical integrity, while estrogens seem to be vital for myometrium and cervix preparation for delivery (Lepert, 1995; Weiss, 2000). Regarding their fetal origin and ubiquity of actions, it

is possible that estrogen production represents a primary event, positioned at the beginning of the labor cascade, that affects myometrium contractility and cervix ripening (Umezaki *et al.*, 1993; Challis and Lye, 1994). In the myometrium, estrogens stimulate a series of cellular and molecular changes that increase uterine contractility, sensitize the uterus to uterotonic compounds, and coordinate the contractile events during labor. They stimulate hyperplasia and hypertrophy of smooth muscle cells, the synthesis of key contraction associated proteins (CAPs), metabolic enzymes, and ATP (Lye, 1996; Hatthachote and Gillespie, 1999). In addition, the formation of gap junctions between adjacent smooth muscle cells, which are also promoted by estrogens, facilitate the synchronization of uterine contractions (Garfield, 1994). Estrogens are also involved in cervical ripening, a mechanism in which 5HT may be involved (Wilcox *et al.*, 1992).

The actions of estrogenic hormones in the uterus are mediated by the estrogen receptor alpha, a member of the family of nuclear receptors that function as ligand-activated transcription factors. Upon activation, the estrogen receptor binds to estrogen-response-elements in the DNA, often located in the 5' flanking region of estrogen responsive genes. These DNA sequences function as enhancers of transcription, conferring estrogen the ability to up-regulate gene expression (Katzenellenbogen, 1996). Estrogens, acting via the estrogen receptor alpha, regulate growth, differentiation, and function of reproductive tissues including the uterus. Regarding its action on MCs, estrogens potentiate MC degranulation and histamine release, with a latency period of 30 min (Cocchiara *et al*., 1992). Therefore, it is highly probable that estrogens also exert direct non-genomic effects on uterine MCs, via membrane estrogen receptors.

IV. Effects of the Oxytocin-Serotonin pathway on myometrium contractility and cervical ripening

Oxytocin, recognized for its many physiological effects and pharmacological applications, has been long considered to be essential for parturition, due to its stimulatory effects on myometrium contractility and cervical ripening (Zingg *et al.*, 1995). However, some doubts have emerged regarding the real role of oxytocin in inducing labor, since it is known that oxytocin plasmatic concentration does not increase until the very end of labor (Leake *et al.*, 1981). Furthermore, labor and delivery is not affected in mice that lack oxytocin (Imamura *et al.*, 2000). Nevertheless, it is possible that plasmatic levels of oxytocin do not reflect the real concentration of this hormone in uterine tissue. It has been found that uterine tissue from pregnant rats synthesizes oxytocin, which could result in increased local levels of this hormone (Lefebvre *et al.*, 1992b; Lefebvre *et al.*, 1992a). Another important aspect is the increased number of oxytocin receptors in uterine tissue that is induced by estrogens and the mechanical stretch of the uterus (Ou *et al.*, 1998). This contributes to increased sensitivity and contractile response of the myometrium to the action of oxytocin. Furthermore, it has been shown that pharmacological inhibition of oxytocin receptors delays delivery, suggesting that the regulatory function of these receptors is more important than the actual oxytocin concentration in the uterine microenvironment (Mitchell and Smid, 2001).

Oxytocin is known to induce uterine contractility by three mechanisms. First, by a direct mechanism, that involves activation of oxytocin receptors on plasma membranes of smooth muscle myometrium (Soloff and Sweet, 1982; Magocsi and Penniston, 1991). Second, by an indirect mechanism, involving the action of oxytocin on the endometrial epithelium, which leads to arachidonic acid metabolism and production of platelet activating factor (PAF) and PGs, specifically PGE_2 and $PGF_{2\alpha}$, which contribute to myometrium contractions (Edgerton *et al.*, 1996; Asselin *et al.*, 1997). However, this mechanism has been questioned, since chorion metabolic activity is able to block the spread of PGs from fetal membranes to the myometrium and cervix (McCoshen *et al.*, 1990; Roseblade *et al.*, 1990). The third mechanism of oxytocin action involves 5HT, a strong uterotonic compound (Rudolph *et al*., 1992; Rudolph *et al*., 1993) whose synthetic analogs (e.g, ergometrine) are used in the clinical setting to reduce hemorrhage by inducing post-partum uterine contractions (Hollingsworth *et al.*, 1988). In addition, 5HT participates in cervical ripening and uterine tissue remodeling by a mechanism involving direct and indirect up-regulation of matrix metalloproteinase (MMP)-13 (Passaretti *et al.*, 1996; Spangaard *et al.*, 1997; Shum *et al*., 2002; Hattori *et al.*, 2003).

It has been shown that oxytocin increases extracellular availability of 5HT in smooth muscle myometrial cells, through the inhibition of 5HT uptake by MCs, the single reservoir of this autacoid in mouse uterus (Rudolph *et al.*, 1998). Interestingly, this effect depends on estrogen predominance in the uterine microenvironment (Rudolph *et al*., 1998). In humans, difficulties to find 5HT-positive MCs in uterine tissue have led to question a possible potentiation between

5HT and oxytocin in human myometrium. However, unpublished results by our group have shown the presence of 5HT-positive MCs in human uterus and cervix. Therefore, it seems important to consider the presence of oxytocin receptors in human uterine MCs, since oxytocin could exert and inhibitory effect on 5HT uptake by MCs leading to increased extracellular levels of this mediator in the human uterus. Clinical results from the use of inhibitors of 5HT uptake, such as Fluoxetine, during pregnancy are contradictory. While some studies have found no association between Fluoxetine intake and premature labor (Goldstain, 1995; Goldstain *et al.*, 1997), Chambers *et al*. (1996) showed that Fluoxetine administered in the third trimester of pregnancy increases premature delivery. Further research in this area will help clarify the clinical significance of 5HT uptake by MCs on premature labor.

V. Effects of Corticotropin-Releasing Hormone on labor and mast cell activation

During the last 20 years, special interest has been placed on Corticotropin Releasing Hormone (CRH), an hypothalamus-derived hormone responsible for adrenocorticotropin hormone (ACTH) production. CRH is a key regulatory component of the stress response, which is also suspected to coordinate the transition from pregnancy to normal parturition (Majzoub *et al.*, 1999). Recently it has been found that CRH belongs to a larger family of stress-related peptides, the urocortins (Lewis *et al.*, 2001). Near the end of pregnancy the placental concentration of CRH increases exponentially. Similar increases in CRH have been observed in response to stressors arising either at the placenta, the mother, or the fetus. This CRH increase may in fact cause premature delivery. Interestingly, this effect is only observed in humans and primates, since they have the ability to express CRH in placenta, fetal membranes, and maternal deciduas. Furthermore, these tissues become the major source of CRH after the second trimester of pregnancy (Robinson *et al.*, 1989). Recent findings have also demonstrated that CRH and its receptors are expressed in many other tissues and organs, such as skin, endocrine glands, immune system, pancreas, liver, and gastrointestinal tract (reviewed by Slominski *et al.*, 2001). An hypothesis in which CRH stimulates both fetal maturation and parturition by stimulating the production of ACTH, glucocorticoids, and dehydroepiandrosterone sulfate (an estrogen precursor) by the fetal hypothalamic-pituitary-adrenal axis has been developed (Schwartz, 1997; Majzoub *et al*., 1999). On the other hand, as a negative feedback mechanism, estrogens may inhibit placental CRH production (Ni *et al.*, 2002).

Myometrial cells express the CRH receptors R_1 and R_2, which belong to the Gs-coupled seven transmembrane receptor family (Slominski *et al.*, 2001). It has been reported that receptor R_1 activation stimulates the constitutive synthesis of NOS (Aggelidou *et al.*, 2002), which may be responsible for vasodilation in uterus and placenta, and for increased $PGF_{2\alpha}$ and PGE_2 production by fetal membranes and decidua (Jones and Challis, 1989; Clifton *et al.*, 1995; Aggelidou *et al*., 2002). An endogenous agonist with greater specificity for R_2 receptors is urocortin, a structurally CRH-related peptide, which is produced by the female reproductive organs only during pregnancy (Singh *et al.*, 1999). This compound stimulates PGE_2 and ACTH production, and has CRH-like effects on myometrial contractility, i.e., increased uterine contractions during delivery (Petraglia *et al.*, 1996). CRH also stimulates inducible NOS and stimulates inflammation, as shown in ovaries and endometrium (Chrousos *et al.*, 1998; Cantarella *et al.*, 2001). These effects seem to be mediated by MC activation in uterine tissue (Madhappan *et al*., 2003), as it has been previously shown for MCs in skin and blood vessels (Cocchiara *et al*., 1997b; Theoharides *et al.*, 1998). Since skin MCs are structurally similar to uterine MCs, it is highly probable that uterine MCs could also express CRH receptors. However, this remains to be demonstrated. Recent evidences also point to uterine MCs not only as targets of CRH actions, but also as a potential source for this hormone in the periphery (Kempuraj *et al.*, 2003).

VI. The New Hypothesis and Model

A series of allergic and inflammatory reactions are characterized by MC activation, leading to a cascade of events involving leukocyte recruitment and activation, with the subsequent release of pro-inflammatory mediators. The actions of these mediators are redundant and have overlapping effects that finally lead to the development of an allergic or inflammatory reaction, respectively. Which is important to point out is that these allergic and inflammatory reactions have a point of no return, where it becomes impossible to therapeutically stop them. Most of these mediators and signal pathways have been identified in women in labor and pre-term labor. Therefore, it has been accepted that the cellular and molecular mechanisms that allow the transition from

a pregnancy stage to labor and delivery stages have common pathways with allergic and inflammatory reactions, in which MCs play a central role.

In fact, experimental, clinical, and pharmacological evidences point to the MC as an effective link among endocrine, paracrine, and autocrine responses in uterine tissue, with a central role for MCs in tilting the balance from a pregnancy stage to labor and delivery stages. In this aspect, MCs may constitute the last resource to pharmacologically prevent the cascade of reactions that lead to the onset of labor.

Evaluation of the most common pathophysiological conditions for premature labor also confirm this hypothesis. Allergic reactions (Klein *et al.*, 1984), as well as extrauterine and intra-uterine infections, are common clinical situations in which MC degranulation evokes myometrial contractions that could terminate pregnancy (Romero *et al.*, 1989; Fidel *et al.*, 1997). Pregnant women affected by systemic mastocytosis, a disease characterized by a pathological increase in MC numbers in different organs, also present obstetric manifestations of preterm labor and delivery (Metcalfe and Akin, 2001). Most of these cases of preterm labor have little or no influence of endocrine factors.

On the other hand, b-adrenergic agonists, commonly used as tocolytic agents for the treatment of preterm labor, as well as classic anti-inflammatory agents, such as corticosteroids and non-steroidal anti-inflammatory drugs, are also used with certain degree of success in the prevention of premature labor in the pathophysiological conditions described above. These drugs stabilize MCs, preventing their activation and the release of MC mediators in response to several compounds (Krishnaswamy *et al.*, 1997; Martinez *et al.*, 1999). Therefore, similar to β-adrenergic agonists and anti-inflammatory drugs, MC stabilizers, may be effective in slowing down the activation of the mechanisms leading to labor and delivery.

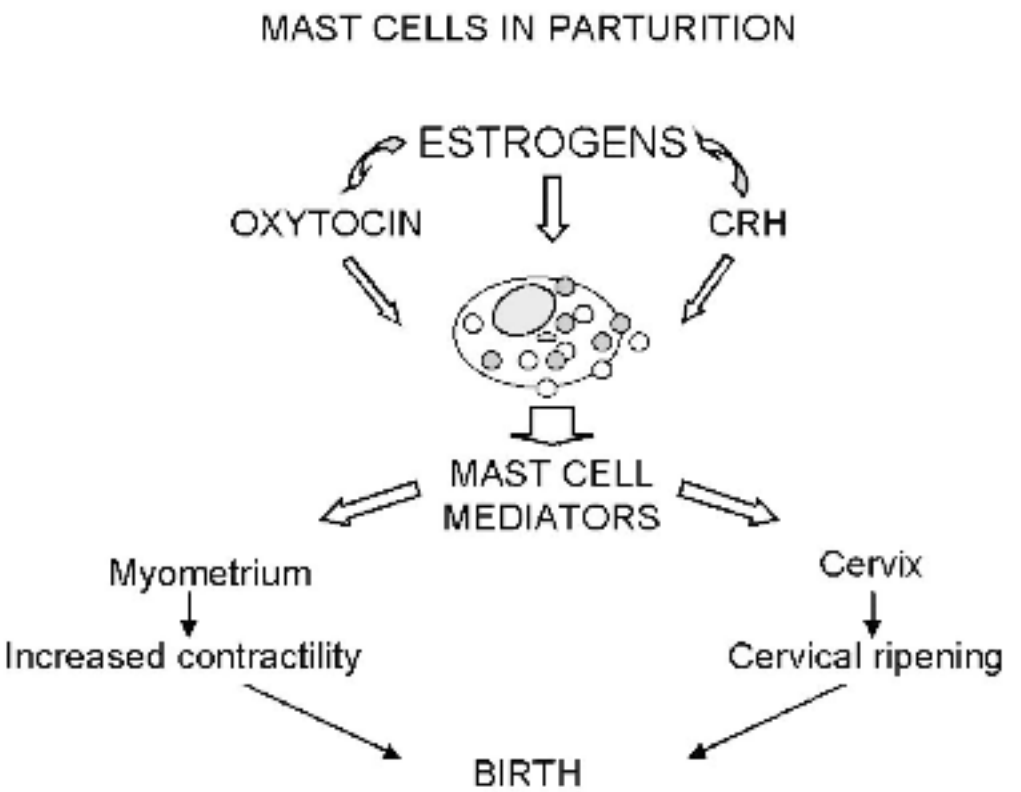

FIGURE 1. A simplified scheme of the proposed role of MCs in parturition. Estrogens, oxytocin, and CRH are positive regulators of MC activation, and therefore specifically potentiate labor-inducing actions via MCs. On the other hand, progesterone and relaxin (not included in the scheme) can potentiate their own relaxing effects on the myometrium by stabilizing uterine MCs. Alternative or parallel pathways, such as the direct regulatory action of estrogens on myometrial CAPs, or the stimulatory effect of oxytocin on uterine contractions and cervix ripening are not represented to maintain the simplicity of the scheme.

Nitric oxide has also been found to prolong gestation in women with preterm labor (Lees *et al.*, 1994). This compound is well known as a potent smooth-muscle cell and blood vessel relaxant. However, nitric oxide can exert a contractile action in myometrium depending upon other endocrine factors present in the uterine microenvironment. Uterus relaxation, which occurs under progesterone predominance, is mediated not only by a direct action of NO on smooth muscle cells, but also by stabilizing MCs (Yallampalli *et al.*, 1993; Coleman, 2002). On the other hand, uterine contractions, which are evoked under estrogen predominance, have been shown to be mediated by activation of prostanoid synthesis and uterine MC activation (Franchi *et al.*, 1994; Martinez *et al*., 1999; Motta *et al*., 1999). Therefore, successful use of NO-donors as therapeutic agents will depend on the time of pregnancy and the endocrine status of the uterus.

The analysis of the mechanisms of action of some "old-fashion" therapeutic approaches to resolve stillbirth with uterine inertia also help to confirm this hypothesis. Stillbirth with uterine inertia is a pathological condition in which endocrine signals fail to evoke labor and delivery, due to the lack of fetal estrogens precursors. Under these circumstances, PGs and their analogues are extensively used (Chung *et al.*, 1999). However, during the 70´s, when PGs were not available, drugs such as ethodin were used to evoke labor. Ethodin, a polybasic amine that could also be described as an allergic compound, was known as "an effective drug" in inducing PG synthesis and uterine contractions that mimic a physiological labor in stillbirth with uterine inertia (Olund *et al.*, 1980; Schubert and Cullberg, 1987). A study aimed to understand the mechanism of action of this drug, demonstrated that contractions were evoked through a direct stimulation of uterine MCs (Rudolph *et al*., 1997a). This action over MCs points to the fact that endocrine signals for labor and delivery, such as estrogens, oxytocin and CRH, may be bypassed by directly activating uterine MCs.

A number of experimental approaches using anti-cytokine therapy have failed to stop the symptoms of premature labor (Fidel *et al*., 1997). Therefore, these results help confirm the hypothesis of irreversibility of the labor process once full MC activation is attained. Once activated, a rapid release of MC mediators activate new pathways, and exert redundant and overlapping actions on the target tissues, i.e. the myometrium and cervix, to promote labor and delivery.

Based on the evidences previously described, a new model is proposed for the role of MCs on parturition (Fig. 1).

In spite of the current knowledge in this area, further basic and clinical research remains to be performed to fully assess the role of MCs in normal and abnormal labor and delivery. However, considering the MC as a link between endocrine and inflammatory responses in the uterine tissue may help to understand, at least in part, the complex process leading to pregnancy ending and parturition. This may provide new approaches for therapeutic intervention and assurance of a better well-being and safety for the mother and newborn.

VIII. Summary and perspectives

In summary, it is proposed that MCs represent the link between endocrine signals and the physiological cascade of local reactions leading to the onset of labor and delivery. Local uterine reactions such as myometrium contraction and cervical ripening may be achieved by the direct activation of uterine MCs. Therefore, MCs may represent a new target for novel therapeutic agents capable of stimulating (MCs stimulators) or inhibiting (MCs stabilizers) both pregnancy and parturition. Confirmation of these results awaits clinical trials with suitable compounds that can be safely administered to the pregnant mother and fetus.

Aknowledgements

This work has been supported by FONDECYT, Chile (1020458).

References

Aggelidou E, Hillhouse EW, Grammatopoulus DK (2002). *Up-regulation of nitric oxide synthase and modulation of guanylate cyclase activity by corticotropin-relesing hormone but not urocortin II or urocortin II in cultured human pregnant myometrial cells*. Proc Natl Acad Sci USA. 99: 3300-3305.

Armetti L, Admon D, Solomon A, Levi-Schaffer F (1999). *mast cells and eosinophils. Their role in tissue remodeling*. ACI International. 11: 22-28.

Asselin E, Drolet P, Fortier MA (1997). *Cellular mechanism involved during oxytocin-induced prostaglandin F2a production in endometrial epithelial cells in vitro: Role of cyclooxygenase-2.* Endocrinology. 138: 4798-4805.

Bokstrom H, Brannstrom M, Alexandersson M, Norstrom A (1997). *Leukocyte subpopulations in human uterine cervical stroma at early and term pregnancy.* Hum Reprod. 12: 586-590.
Brightling C, Symon F, Holgate S, Wardlaw A, Pavord I, Bradding P (2003). *Interleukin-4 and -13 expression is co-localized to mast cells within the airway smooth muscle in asthma.* Clin Exp Allergy. 33: 1711-1716.
Bytautiene E, Vedernikov YP, Saade GR, Romero R, Garfield RE (2003). *Effect of histamine on phasic and tonic contractions of isolated uterine tissue from pregnant women*. Am J Obstet Gynecol. 188: 774-778.
Cabanillas-Saez A, Schalper JA, Nicovani SM, Rudolph MI (2002). *Characterization of mast cells according to their content of tryptase and chymase in normal and neoplastic human uterine cervix.* Int J Gynecol Cancer. 12: 92-98.
Cantarella G, lempercur L, Lombardo G, Chiarenza A, Pafumi C, Zappala G, Bernardini R (2001). *Divergent effects of corticotropin releasing hormone on endothelial cell nitric oxide synthase are associated with different expression of CRH type 1 and 2 receptors.* Br J Pharmacol. 134: 837-844.
Challis JRG, Lye SJ (1994). *Parturition.* In: The Physiology of Reproduction. E Knobil, JD Neil, Eds. Raven Press, New York; Vol 2.
Chambers CD, Johnson KA, Dick LM, Felix RJ, Jones KL (1996). *Birth outcomes in pregnant women taking fluoxetine*. N Engl J Med 335: 1010-1015.
Chrousos GP, Torpy D, Gold PW (1998). *Interactions between the hypothalamic-pituitary-adrenal axis and the female reproductive systems.* Ann Intern Med. 129: 229-240.
Chung YP, Hwa HL, Ko TM (1999). *Laminaria and sulprostone in second trimester pregnancy termination for fetal abnormalities*. Int J Gynecol Obstetr. 65: 47-52.
Church MK, Levy-Schaffer F (1997). *The human mast cell.* J Allergy Clin Immunol. 99: 155-160.
Chwalisz K, Garfield RE (1998). *Role of nitric oxide in the uterus and cervix: implications for the management of labor.* J Perinat Med. 26: 448-457.
Clifton VL, Read MA, Leitch IM, Giles WB, Boura AL, Robinson PJ, Smith R (1995). *Corticotropin-releasing hormone-induced vasodilatation in the human fetal-placental circulation: involvement of nitric oxide-cyclic guanosine 3′,5′-monophosphate pathway.* J Clin Endocrinol Metab. 80: 2888-2893.
Cocchiara R, Bongiovanni A, Albeggiani G, Azzolina A, Geraci D (1997a). *Sustance P selectively activates TNFa mRNA in rat uterine ummune cells: a neuroimmune link.* Neuroreport. 8: 2961-2964.
Cocchiara R, Bongiovanni A, Albeggiani G, Azzolina A, Lampiasi N, Di Blasi F, Geraci D (1997b). *Inhibitory effect of neuraminidase on SP-induced histamine release and TNF-alpha mRNA in rat mast cells: evidence of a receptor-independent mechanism.* J Neuroimmunol. 75: 9-18.
Cocchiara R, Albeggianni G, Di Trapani G, Azzolina A, Lampiasi N, Rizzo F, Diotallevi L, Gianaroli L, Geraci D (1992). *Oestradiol enhances in vitro the histamine release induced by embrionic histamine-releasing factor (EHGF) from uterine mast cells*. Human Reprod. 7: 1036-1041.
Coleman J (2002). *Nitric oxide: a regulator of mast cell activation and mast cell-mediated inflammation.* Clin Exp Immunol. 129: 4-10.
Costa JJ, Burd PR, Metcalfe DD (1994). *Mast cell cytokines.* In: The mast cell in health and disease, MA Kaliner and DD Metcalfe, Eds., Marcel Dekker, New York, pp. 443-466.
Cruz MA, González C, Acevedo CG, Sepúlveda WH, Rudolph MI (1989). *Effects of histamine and serotonin on the contractility of isolated pregnant and nonpregnant human myometrium.* Gynecol Obstet Invest. 28: 1-4.
Edgerton LA, Kaminski MA, Silvia MJ (1996). *Changes in uterine secretion of prostaglandin F2alpha in response to oxytocin during estrous cycle, early pregnancy and estrogen-induced pseudopregnancy in swine.* Biol Reprod. 55: 657-662.
Evans CH, Stefanovic-Racic M, Lancaster J (1995). *Nitric oxide and its role in orthopaedic disease.* J Clin Orthop Rel Res. 312: 275-294.
Fajardo I, Pejler G (2003). *Human mast cell beta-tryptase is a gelatinase.* Journal of immunology (Baltimore, Md : 1950). 171: 1493-1499.
Fidel PL, Romero R, Cutright J, Wolf N, Gomez R, Araneda H, Ramirez M, Yoon BH (1997). *Treatment with the interleukin-1 receptor antagonist and soluble tumor necrosis factor receptor Fc fusion protein does not prevent endotoxin-induced preterm parturition in mice.* J Soc Gynecol Invest. 4: 22-26.
Franchi A, Chaud M, Rettori V, Suburo A, McCann S, Gimeno M (1994). *Role of nitric oxide in eicosanoid synthesis and uterine motility in estrogen-treated rat uteri.* Proc Natl Acad Sci USA. 91: 539-543.
Galli SJ, Wershil BK, Mekori YA (1987). *Analysis of mast cell function in biological responses not involving IgE.* Int Arch Allergy Appl Immunol. 82: 269-271.
Galli SJ, Zsebo KM, Geissler EN (1994). *The kit ligand, stem cell factor.* Adv Immunol. 55: 1-96.
Galli SJ, Maurer M, Lantz CS (1999). *Mast cells as sentinels of innate immunity*. Curr Opin Immunol. 11: 53-59.
Garfield RE (1994). *Role of cell-to-cell coupling in control of myometrial contractility and labor.* In: Control of uterine contractility RE Garfield and TN Tabb, Eds, CRC Press Inc., Boca Raton, FLA. pp. 39-81.
Goldstain DJ (1995). *Effects of third trimester fluoxetine exposure on the newborn*. J Clin Psychopharmacol. 15: 417-420.
Goldstain DJ, Sundell KL, Corbin LA (1997). *Birth outcomes in pregnant women taking fluoxetine (letter).* N Engl J Med. 336: 872-873.
Gordon JR, Galli SJ (1990). *Mast cells as a source of both preformed and immunologically inducible TNK/cachectin.* Nature. 19: 274-276.
Harris S, Phipps R (2002). *Prostaglandin D(2), its metabolite 15-d-PGJ(2), and peroxisome proliferator activated receptor-gamma agonists induce apoptosis in transformed, but not normal, human T lineage cells.* Immunology. 105: 23-.
Hatthachote P, Gillespie JI (1999). *Complex interactions between sex steroids and cytokines in the human pregnant myometrium: Evidence for an autocrine signaling system at term.* Endocrinology. 140: 2533-2540.
Hattori Y, Nerusu KC, Bhagavathula N, Brennan M, Hattori N, Murphy HS, Su LD, Wang TS, Johnson TM, Varani J (2003). *Vascular expression of matrix metalloproteinase-13 (collagenase-3) in basal cell carcinoma.* Exp Mol Pathol. 74: 230-237.
Hollingsworth M, Edwards D, Miller M (1988). *Ergometrine a partial agonist at 5HT receptors in the uterus isolated from estrogen-primed rat.* . Eur J Pharmacol. 158: 79-84.
Huang J, Roby K, Pace J, Russell S, Hunt J (1995). *Cellular localization and hormonal regulation of inducible nitric oxide synthase in cycling mouse uterus.* J Leukoc Biol.. 57: 27-35.
Hunt JS, Miller L, Roby KF, Huang J, Platt JS, DeBrot BL (1997). *Female steroid hormones regulate production of pro-inflammatory molecules in uterine leukocytes.* J Reprod Immunol. 35: 87-99.
Imamura T, Luedke CE, Vogt SK, Muglia LJ (2000). *Oxytocin modulates the onset of murine parturition by competing ovarian and uterine effects*. Am J Physiol. 279: R1051-R1057.

Jeziorska M, Salamonsen LA, Woolley DE (1995). *Mast cell and eosinophil distribution and activation in human endometrium throughout the menstrual cycle.* Biol Reprod. 53: 312-320.

Jones SA, Challis JRG (1989). *Local stimulation of prostaglandin production by corticotrophin-releasing hormone in human female membranes and placenta.* Biochem Biophys Res Commun. 159: 192-199.

Katzenellenbogen BS (1996). *Estrogen receptors: Bioactivities and interactions with cell signalling pathways*. Biol Reprod. 54: 287-293.

Kempuraj D, Papadopoulou N, Lytinas M, Huang M, Kandere-Grzybowska K, Madhappan B, Boucher W, Christodoulou S, Athanassiou A, Theoharides T (2003). *Corticotropin-Releasing Hormone (CRH) and its Structurally Related Urocortin (Ucn) are Synthesized and Secreted by Human Mast Cells.* Endocrinology.(e- pub)

Klein VR, Harris AP, Abraham RA, Niebyl JR (1984) *Fetal distress during maternal systemic allergic reaction*. Obstet Gynecol. 64: 155-175.

Konopka T, Barker J, Bamford T, Guida E, Anderson R, Stewart A (2001). *Nitric oxide synthase II gene disruption: implications for tumor growth and vascular endothelial growth factor production.* Cancer Research. 61: 3182-.

Krishnaswamy G, Kelley J, Johnson D, Younberg G, Stone W, Huang S-K, Bieber J, Chi DS (2001). *The human mast cell: functions in physiology and disease*. Front Biosci. 6: 1109-1127.

Krishnaswamy G, Lakshman T, Miller AR, Srikanth S, Hall K, Huang SK, Suttles J, Smith JK, Stout R (1997). *Multifunctional cytokine expression by human mast cells: regulation by T cell membrane contact and corticoids.* J Interferon Cytokine Res. 17: 167-176.

Leake RD, Weitzman RE, Glatz TH, Fisher DH (1981). *Plasma oxytocin concentration in men, non-pregnant women, and pregnant women before and during spontaneous labor.* J Clin Endocrinol Metab. 53: 730-733.

Lees C, Campbell S, Jauniaux E, Brown R, Ramsay B, Gibb D, Moncada S, Martin JF (1994). *Arrest of preterm labour and prolongation of gestation with glyceryl trinitrate, a nitric oxide donor*. Lancet. 343: 1325-1326.

Lefebvre DL, Giaid A, Zingg HH (1992a). *Expression of the oxytocin gene in rat placenta.* Endocrinology. 130: 1185-.

Lefebvre DL, Giaid A, Bennett H, Lariviere R, Zingg HH (1992b). *Oxytocin gene expression in rat uterus.* Science. 256: 1553-.

Lemura A, Tsai M, Ando A, Wershil BK, Galli SJ (1994). *The c-kit ligand, stem cell factor, prometes mast cell survival by suppressig apoptosis*. Am J Pathol. 144: 321-328.

Leon A, Buriani A, Dal Toso R, Fabris M, Romanello S, Aloe L, Levi-Montalcini R (1994). *Mast cells synthezise, store, and release nerve growth factor.* Proc Natl Acad Sci USA. 91: 3739-3743.

Lepert PC (1995). *Anatomy and physiology of cervical ripening.* Clin Obstet Gynecol. 38: 267-279.

Lewis K, Li C, Perrin MH, Blount A, Kunitake K, Donaldson C, Vaughan J, Reyes TM, Gulyas J, Fischer W, Bilezikjian L, Rivier J, Sawchenko PE, Vale WW (2001). *Identification of urocortin III, an additional member of the corticotropin-releasing factor (CRF) family with high affinity for the CRF2 receptor.* Proc Natl Acad Sci U S A. 98: 7570-7575.

Lye SJ (1996). *Initiation of parturition.* Anim Reprod Sci. 42: 495-503.

Madhappan B, Kempuraj D, Christodoulou S, Tsapidikidis S, Boucher W, Theorarides TC (2003). *High levels of intrauterine corticotropin-releasing hormone, urocotrin, tryptase, and interleukin-8 in spontaneous abortions.* Endocrinology. 144: 2285-2290.

Magocsi M, Penniston JC (1991). *Oxytocin pretreatment of pregnant rat uterus inhibits Ca uptake in plasma membrane and sarcoplasmic reticulum.* Biochem Biophys Acta. 1063: 7-12.

Majzoub JA, Mcgregor JA, Lockwood CJ, Smith R, Snyder-Tagart M, Schulkin J (1999). *A central theory of preterm and term labor: Putetive role for corticotropin-releasing hormone*. Am J Obstet Gynecol. 180: S232-S241.

Martinez L, Gimeno MF, Villán L, Villar M, Rudolph MI (1999). *Nitroprusside stimulates contractility and the synthesis of 14C-acetylated PAF-like substances in estrogen primed-mouse uterine horns.* Prostaglandins Other Lipid Mediat. 57: 49-62.

Masini E, Bani D, Bigazzi M, Mannaioni PF, Bani-Sacchi T (1994). *Effects of relaxin on mast cells. In vitro and in vivo studies in rats and guinea pigs*. J Clin Invest. 94: 1974-1980.

Massey WA, Guo C-B, Dvorak AM, Hubbard WC, Bhagavan BS, Cohan VL, Warner JA, Kagey-Sobotka A, Lichtenstein LM (1991). *Human uterine mast cells. Isolation, purification, characterization, ultraestructure, and pharmacology.* J Immunol. 147: 1621-1627.

McCoshen JA, Hoffman DR, Kredentser JV, Araneda C, Johnston JM (1990). *The role of fetal membranes in regulating prosuction, transport, and metabolism of prostaglandin E2 during labor.* Am J Obstet Gynecol. 163: 1632-1640.

McNeil HP, Austen KF (1995). *Biology of the mast cell.* In: Samter´s Immunologic Diseases. Eds. MM Frank, KF Austes, HN Claman, ER Unanue Eds. Little. Brown and Co., NY. pp. 185-204.

Metcalfe D, Baram D, Mekori Y (1997). *Mast cells.* Physiol Reviews. 77: 1033-1079.

Metcalfe DD, Akin C (2001). *Mastocytosis: molecular mechanisms and clinical disease heterogeneity.* Leuk Res. 25: 577-582.

Mitchell BF, Smid B (2001). *Oxytocin and its receptor in the process of parturition*. J Soc Gynecol Invest. 8: 122-133.

Mori A, Nakayama K, Suzuki J, Nikaido T, Isobe M, Fujii S (1997). *Analysis of stem cell factor for mast cell proliferation in the human myometrium.* Mol Hum Reprod. 3: 411-418.

Motta AB, Gonzalez ET, Rudolph MI, Gimeno MF (1999). *Regulation of prostaglandin production by nitric oxide in rat smooth muscle myometrial cells.* Prostaglandins Leukot Essent Fatty Acids. 60: 73-76.

Ni X, Nicholson RC, King BR, Chan E-C, Read MA, Smith R (2002). *Estrogen represses whereas the estrogen-antagonist ICI 182780 stimulates placental CRH gene expression*. J Clin Endocrinol Metab. 87: 3774-3778.

Nilsson G, Schwartz LB (1995). *Mast cell heterogeneity: Structure and mediators.* In: Asthma and Rinitis.. WW Busse, ST Holgate, Eds., Blackwell Sci, Boston. pp. 195-208.

Okayama Y, Kobayashi H, Ashman L, Holgate S, Church M, Mori M (1997). *Activation of eosinophils with cytokines produced by lung mast cells.* Int Arch Allergy & Immunol. 114 Suppl 1: 75-77.

Olund A, Kindahl H, Oliw E, Lindgren A, Larsson B (1980). *Prostaglandins and thromboxanes in amniotic fluid during rivanol-induced abortion and labor.* Prostaglandins. 19: 701-800.

Ou C-W, Chen Z-Q, Qi S, Lye SJ (1998). *Increased expression of the rat myometrial oxytocin receptor messenger ribonucleic acid during labor requires both mechanical and hormonal signals.* Biol Reprod. 59: 1055-1061.

Padilla L, Reinicke K, Montesino H, Villena F, Asencio H, Cruz MA, Rudolph MI (1990). *Histamine content and mast cell distribution in mouse uterus: The effect of sexual hormones, gestation and labor*. Cell Mol Biol. 36: 93-100.

Pang X, Cotreau-Bibbo MM, Sant GR, Theoharides TC (1995). *Bladder mast cell expression of high affinity oestrogen receptors in patients with interstitial cystitis.* Br J Urol. 75: 151-161.

Pasqualini JR, Kincl F (1985). *Hormones and the fetus.* Pergamon Press, Oxford.

Passaretti TV, Wilcox BD, Jeffrey JJ (1996). *Serotonin regulation of gene expression in uterine extracellular matrix: reciprocal effects on collagens and collagenase.* Mol Cell Endocrinol. 120: 125-132.

Penissi A, Rudolph M, Piezzi R (2003a). *Role of mast cells in gastrointestinal mucosal defense.* Biocell. 27: 163-172.
Penissi A, Rudolph I, Fogal T, Piezzi R (2003b). *Changes in duodenal mast cells in response to dehydroleucodine.* Cells Tissues Organs. 173: 234-241.
Petraglia F, Fluorio P, Gallo R (1996). *Human placenta and fetal membranes express human urocortin mRNA and peptide.* J Clin Endocrino Metab. 81: 3807-3810.
Puxeddu I, Piliponsky A, Bachelet I, Levi-Schaffer F (2003). *Mast cells in allergy and beyond.* International Journal of Biochemistry & Cell Biology. 35: 1601-1605
Robinson BG, Arbiser JL, Emanuel RL, Majzoub JA (1989). *Species-specific placental corticotropin-releasing hormone messenger RNA and peptide expression.* Mol Cell Endocrinol. 61: 337-341.
Roby KF, Hunt JS (1995). *Myometrial tumor necrosis factor alpha: cellular localization and regulation by estradiol and progesterone in the mouse*. Biol Reprod. 52: 509-515.
Romero R, Sirtori M, Oyarzun E, Avila C, Mazor M, Callahan R, Sabo V, Athanassiadis AP, Hobbins JC (1989). *Infection and labor. V. Prevalence, microbiology, and clinical significance of intraamniotic infection in women with preterm labor and intact membranes*. Am J Obstet Gynecol. 161: 817-824.
Roseblade CK, Sullivan MHF, Khan H, Lumb MR, Elder MG (1990). *Limited transfer of prostaglandin E2 across the fetal membranes before and after labor.* Acta Obstet Gynecol Scand. 69: 399-403.
Rudolph MI, Bardisa L, Cruz MA, Reinicke K (1992). *Mast cell mediators evoke contractility and potentiate each other in mouse uterine horns*. Gen Pharmacol. 23: 833-836.
Rudolph MI, Reinicke K, Cruz MA, Gonzalez C, Bardisa L (1993). *Distribution of mast cells and the effect of their mediators on contractility in human myometrium*. Br J Obstet Gynecol. 100: 1125-1130.
Rudolph MI, Cabanillas A, Gómez P, Garcia MA, Villán L (1997a). *On the mechanism of action of ethodin in inducing myometrium contractions.* Gen Pharmacol. 28: 381-385.
Rudolph MI, Garcia MA, Sepúlveda M, Brandan E, Reinicke K, Nicovani S, Villán L (1997b). *Ethodin: Pharmacological evidence of the interaction between smooth muscle and mast cells in the myometrium.* J Pharmacol Exper Ther. 282: 256-261.
Rudolph MI, Oviedo C, Martínez L, Reinicke K, Villar M, Villán L (1998). *Oxytocin inhibits the uptake of serotonin into uterine mast cells.* J Pharmacol Exper Ther. 287: 389-394.
Sautebin L, Lalenti A, Ignarro A, Rosa MD (1995). *Modulation by nitric oxide of prostaglandin biosynthesis in the rat uteri.* Br J Pharmacol. 114: 323-328.
Schmauder-Chock E, Chock SP (1989). *Localization of cyclooxygenase and prostaglandin E2 in the secretory granule of the mast cell.* J Histochem Cytochem. 37: 1319-1328.
Schmith A, Goepfert C, Feitsma K, Buddecke E (2002). *Lovastatin-stimulated superinduction of E-selectin, ICAM-1 and VCAM in TNFa activated human vascular endothelial cells.* Atherosclerosis. 164: 57-64.
Schubert W, Cullberg G (1987). *Second trimester abortion by rivanol with or without cervical dilatators.* Acta Obstet Gynecol Scand. 66: 549-550.
Schwartz LB (1997). *Understanding human parturition.* Lancet. 350: 1792-1793.
Shum JKS, Melendez JA, Jeffrey JJ (2002). *Serotonin-induced MMP-13 production is mediated via Phspholipase C, protein kinase C, and ERK1/2 in rat uterine smooth muscle cells.* J Biol Chem. 277: 42830-42840.
Singh LK, Boucher W, Pang X, Letourneau R, Seretakis D, Green M, Theoharides TC (1999). *Potent mast cell degranulation and vascular permeability triggered by urocortin through activation of CRH receptors.* J Pharmaol Exp Ther. 288: 1349-1356.
Slominski A, Wortsman J, Pisarchik A, Zbytek B, Linton EA, Mazurkiewicz JE, Wei ET (2001). *Cutaneous expression of corticotropin-releasing hormone (CRH), urocortin, and CRH receptors.* FASEB J. 15: 1678-1693.
Soloff MS, Sweet P (1982). *Oxytocin inhibition of (Ca-Mg)ATPase activity in rat myometrial plasma membranes.* J Biol Chem. 257: 10687-10693.
Spangaard H, Knudsen UB, Uldberg N, Jerzioska M, Woolley DE, Danielsen CC (1997). *Mast cells in cervical ripening- an immunohistochemical and biochemical study in rats.* Eur J Obstet Gynecol Reprod Biol. 73: 91-97.
Stenton GR, Vliagoftis H, Befus D (1998). *Role of intestinal mast cells in modulating gastrointestinal pathophysiology.* Ann Allergy Asthma Immunol. 85: 1-15.
Tabb NT (1994). *Immune control of myometrial contractility: Role of mast cells*. In: Control of uterine contractility. RE Garfield, NT Tabb, Eds. CRC Press, Boca de Raton, FL. pp. 355-373.
Theoharides TC, Singh LK, Boucher W, Pang X, Letourneau R, Webster E, Chrousos GP (1998). *Corticotropin-releasing hormone induces skin mast cell degranulation and increased vascular permeability, a possible explanation for its proinflammatory affects.* Endocrinology. 139: 403-413.
Thompson AJ, Telfer JF, Young A, Campbell S, Stewart CJR, Cameron IT, Greer IA, Norman JE (1999). *Leukocytes infiltrate the myometrium during human parturition: further evidence that labor is an inflammatory process.* Hum Reprod. 14: 229-236.
Tkaczyk C, Frandji P, Botros HG, Poncet P, Lapeyre J, Peronet R, David B, Mecheri S (1996). *Mouse bone marrow-derived mast cells and mast cell lines constitutively produce B cell growth and differentiation activities.* J Immunol. 157: 1720-1728.
Umezaki H, Valenzuela GJ, Hess DL, Ducsay CA (1993). *Fetectomy alters maternal endocrine and uterine activity rhythms in rhesus macaques during late gestation.* Am J Obstet Gynecol. 169: 1435-1441.
Walsh LJ, Trinchieri G, Waldorf HA, Whitaker D, Murphy GF (1991). *Human dermal mast cells contain and relese tumor necrosis factor alpha, which induces endothelial leukocyte adhesion molecule 1*. Proc Natl Acad Sci USA. 88: 4220-4224.
Weiss G (2000). *Endocrinology of parturition*. J Clin Endocrinol Metab. 85: 4221-4225.
Wershil BK, Galli SJ (1991). *Gastrointestinal mast cells. New approaches for analyzing their function in vivo.* Gastroenterol Clin NA. 20: 613-627.
Wilcox BD, Ridelek-Fitzgerard L, Jeffrey JJ (1992). *Regulation of collagenase gene expression by serotonin and progesterone in rat uterine smooth muscle cells*. J Biol Chem. 267: 20752-20757.
Yallampalli C, Izumi H, Byam-Smith M, Garfield R (1993). *An L-arginine-nitric oxide-cyclic guanosine monophosphate system exists in the uterus and inhibits contractility during pregnancy*. Am J Obstet Gynecol. 170: 175-185.
Zingg HH, Rozen F, Chu K, Larcher A, Arian A, Richard S, Lefebvre D (1995). *Oxytocin and oxytocin receptor gene expression in the uterus*. Recent Progr Hormone Res. 50: 255-273.