Human gonadotropin-releasing hormone receptor-activated cellular functions and signaling pathways in extra-pituitary tissues and cancer cells (Review)

ARTURO AGUILAR-ROJAS[1] and MAIRA HUERTA-REYES[2]

[1]Unidad de Investigación Médica en Medicina Reproductiva, Unidad Médica de Alta Especialidad (UMAE) en Ginecología y Obstetricia No. 4 'Dr. Luis Castelazo Ayala', Instituto Mexicano del Seguro Social (IMSS), Av. Río Magdalena No. 289, Col. Tizapán San Ángel, 01090 México, D.F.; [2]Centro de Investigación Biomédica del Sur (CIBIS), IMSS, Argentina No. 1, Col. Centro, 62790 Xochitepec, Morelos, México

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Abstract. Human gonadotropin-releasing hormone receptor (GnRHR) and its natural ligand human gonadotropin-releasing hormone (GnRH) were initially described as signaling complexes that play a key role in reproductive functions. By binding to specific receptors present on pituitary gonadotropes, GnRH regulates the sperm and ovum maturation, as well as steroidogenesis within the context of the hypothalamus-hypophysis axis. The expression of GnRH and its receptor has clearly been established in many extra-pituitary organs. Some of them are tumors from non-reproductive tissues such as liver, larynx, pancreas, colon, lymphoma, kidney, skin, blood and brain as well as tissues from reproductive track, for example ovary, endometrium, prostate and breast or tumors derived from these organs. Expression of GnRH and its receptor in these organs has gained much attention and several research groups have established their role during cell proliferation and cell motility. Although the signaling pathways and their effector proteins in these samples remain unclear, the molecular mechanism employed for GnRH and its receptor in extra-pituitary tissues could be related with non-classical GnRHR-signaling pathways. In the present review, we explore the vast literature reported on GnRH and GnRHR principally in tumors, describing how cross-talk between GnRHR and growth factor receptor, the coupling between GnRHR and many G proteins depending on cell context, and the regulation of several proteins associated with cell proliferation and cell motility are employed by GnRHR/GnRH to regulate their extra-pituitary activities.

Correspondence to: Dr Arturo Aguilar-Rojas, Research Unit in Reproductive Medicine, IMSS, Unidad Médica de Alta Especialidad (UMAE) en Ginecología y Obstetricia No. 4 'Dr. Luis Castelazo Ayala', Sexto piso, Av. Río Magdalena No. 289, Col. Tizapán San Ángel, 01090 México, D.F., México
E-mail: a_aguilar@correo.unam.mx

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Contents

1. Introduction

Function of GnRHR and GnRH in pituitary. Human gonadotropin-releasing hormone type I [denominated here as GnRH and also known as Luteinizing hormone-releasing hormone (LHRH)], is a hypothalamic decapeptide (1,2). It specifically binds to the gonadotropin-releasing hormone receptor type I (referred here as GnRHR and also known as LHRH receptor), which is a member of G-protein coupled receptors (GPCR) (2,3). In conjunction, both molecules comprise one of the most important signaling complexes that control sperm and ovum maturation, as well as steroidogenesis in gonads, by means of production and release of pituitary gonadotropins, luteinizing hormone (LH) and Follicle-stimulating hormone (FSH) (1,2). LH and FSH are structured by two non-covalently linked α and ß subunits (4). Within a species, α-subunits are identical, while ß-subunits differ and confer physiological heterodimeric hormone specificity.

After GnRH receptor activation, synthesis and release of the α and the two separate ß chains of FSH and LH are promoted (5). At present, many molecular details employed during this signaling process are well known. GnRH is produced in anterior hypothalamus while GnRHR is expressed mainly in gonadotrope membrane in adenohypophysis (6). After GnRH binds, GnRHR via $G_{q/11}$ protein produces Phospholipase C-ß (PLC) activation with the consequent

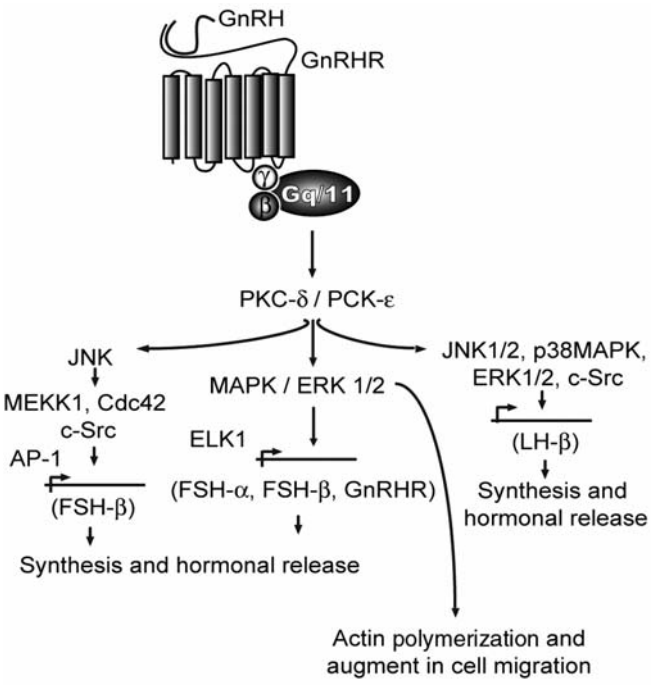

Figure 1. The signaling pathway activated after gonadotropin-releasing hormone receptor (GnRHR) activation in pituitary cells. The binding between gonadotropin-releasing hormone (GnRH) and GnRHR, produce PKC activation and the concomitant production and release of α and ß subunit of LH and FSH. GnRHR activation caused changes in cellular morphology derivative by actin polymerization after ERK1/2 activity with a high increase in cell motility as well as detach cell behavior. Activation is represented by solid arrows (↓).

increase in second messengers inositol 1,4,5-triphosphate (IP3) and diacylglycerol (DAG), and calcium (Ca^{2+}) release. This increase in DAG and Ca^{2+} is able to activate at PKC-δ or PKC-ε (7), triggering the synthesis and release in pulse fashion of LH and FSH via some members of the mitogen-activated protein kinase family (MAPK) (7,8) (Fig. 1).

Gene expression of LH and FSH by GnRHR via MAPK pathway. The MAPK family is composed of several isoforms in mammals: Extracellular signal-regulated kinase (ERK) (ERK1/2, -3/4, -5, -7/8); Jun N-terminal kinase [(JNK)1/2/3]; p38 MAPK [a/b/g(ERK6)/d], and big MAPK (BMK, ERK5) (8-12). Each of these signaling pathways is constituted of a few classes of serine/threonine kinase proteins, which are activated in a sequential manner. Their principal function is to regulate gene expression either by activating nuclear transcription factor or by phosphorylating down-stream cytoplasmatic kinases (13). MAPK pathways are activated by many stimuli, including receptor tyrosine kinases (RTK) and GPCR (14). MAPK activation for RTK is associated with many effector molecules, such as Grb2, an adaptor molecule, and the guanine exchange factor (GEF) mSos. These proteins produce Ras activation followed by Raf-1, MAPK kinase (MAPKK; MEK), and MAPK. In the case of GPCR, these receptors are linking with MAPK by several systems, including PKC-dependent and -independent pathways, through the sequential activation of the proteins (15,16).

The gene expression of LH and FSH subunits via GnRHR-MAPK has been demonstrated by several assays carried out in pituitary cells or in the αT3-1 gonadotrope-derived cell line. These reports showed that either ERK1/2 (8,17) or MAPK are markedly activated during GnRH-pulse activation (18,19) via PKC (20-23) with concomitant mRNA expression of FHS-α, FSH-ß subunit, as well as mRNA of GnRH receptor (19,24) (Fig. 1). In the case of α-subunit, this transcription gene is produced at least by two unrelated response elements. One is a permissive GnRH tissue-specific enhancer designated by pituitary glycoprotein hormone basal elements (PGBE), which is able to bind the transcription factor ELK1 (24), and the other comprises the GnRH-responsive element (GnRH-RE) which is sufficient to allow GnRH responses (25,26).

Another MAPK family member that is activated by GnRH is JNK (27). This signaling pathway reported by FSH-ß subunit employs the AP-1 transcription factor, which is activated by the tyrosine kinase c-Src, the small G protein Cdc42 and MEK kinase 1 (MEKK1) (27,28) (Fig. 1). It has also been shown that multiple MAPK-family members, such as JNK 1/2, p38MAPK, and ERK1/2, as well as c-Src, are involved in GnRH-promoted LH ß-subunit gene expression (29) (Fig. 1).

MAPK activation through GnRHR has been demonstrated by gonadotropes, but the signaling cascade could depend on cell type (17). For GT1-7, a hypothalamus-derived cell, ERK activation occurs via cross-talking between GnRHR and epidermal growth factor receptor (EGFR) (30). Once ERK protein is activated, the canonical pathway begins. For other cell lineages, cellular response via GnRHR-MAPK is very different. In the case of HEK293 cells, ERK activation depends on its binding to focal adhesion kinase (FAK) and c-Src at the adhesion complex that produces cytoskeleton remodeling (31). At the same time, in several cell lines, GnRHR is also able to be coupled with several G proteins (32-35). For COS-7 cells, MAPK regulation couples with G_i protein and EGFR, c-Src, ERK and JNK as the next step in signal transduction (36,37).

2. Function of GnRHR in extra-pituitary tissues

Synthesis and release of FSH and LH in pituitary cells is the principal GnRH receptor function. However, in non-pituitary tissues, tumor derived from non-reproductive tissues and tumors from reproductive tissues (ovary, endometrium, prostate and breast) (38), GnRH and GnRHR are expressed and associated with many novel cellular responses.

GnRHR in non-pituitary tissues. Identification of GnRH and GnRHR mRNA by RT-PCR from ovary sample was observed in granulosa-lutea consorting with its expression in follicular growth (39). The coding sequence for GnRH receptor in ovary was identical to that reported by gonadotropes and was able to control corpus luteum function either *in vitro* or *in vivo* (40,41). In human granulosa-luteal cells (hGLCs), treatment with a GnRH agonist (GnRHa) promoted down-regulation of both estrogen receptor (ER) subunit α and ß via the PKC system. These results showed that GnRH may contribute to control of granulosa-luteal cell function, regulating ER subunit α and ß expression in human ovary (42).

The GnRHR gene is also expressed in human placenta, (43). A dose-response effect of human chorionic gonadotropin

Table I. Characteristics of GnRHR and GnRH in cancer cells.

- GnRHR and GnRH are related with proliferation and migration in cancer cells
- Expression of GnRHR and GnRH in cancer cells is very low
- There are high-affinity/low-affinity binding sites by GnRH in cancer cells
- There is no dichotomy of GnRH agonist and antagonist in cancer cells
- According with the amount of GnRHR in cell surfaces and GnRH in the media, these exert a bi-phasic impact over its extra-pituitary functions

(hCG) secretion was observed in the culture system from placental explants treated with GnRH ranging from 10^{-9} to 10^{-7} mol/l (44). In primary culture of cytotrophoblast cells, choriocarcinoma cell line (JEG-3) and immortalized extravillous trophoblasts cells (IEVT), expression of an mRNA identical to its pituitary counterpart was exhibited, and an increase in GnRHR mRNA levels was observed after GnRHa treatment. This effect was reversed by adenylate cyclase inhibitor or PKA inhibitor, concluding that GnRH plays a regulatory function through PKC and PKA pathways in placenta (45). Another important extrahypothalamic GnRH function has been demonstrated in cultured stromal cells (46). GnRH agonist was able to abolish the tissue inhibitor function of metalloproteinase-1 and -3 (TIMP-1, TIMP-3), which are responsible for inhibiting matrix metalloproteinase (MMP) activity (47), providing evidence that GnRH in trophoblast cells may modulate cell invasion via MMP regulation (46,47).

In rat ventral-prostate organ culture and prostate-cancer derived cells, the effect of GnRH was demonstrated on cell proliferation. GnRH receptor was able to antagonize either the testosterone actions on cell proliferation, tissue growth or it induced nuclear translocation of non-active androgen receptors (48).

The GnRH receptor is expressed in T cells (49). In this case, treatment with GnRH was able to encourage gene transcription of a 67-kDa non-integrin laminin receptor, namely 67-kDa LR, and also furthers cell adhesion to laminin and cell migration *in vitro* and *in vivo* (49).

GnRHR in tumors. Despite that GnRHR and GnRH transcripts from tumor cells are equivalent to pituitary coding region and protein (3,33,50), there are important functional differences between them (38). In nearly all tumor cell lines, receptor expression is low in comparison with that of gonadotropes (38,51-54). Another important difference is related to GnRH and its receptor-binding ability. In pituitary cells, there are binding sites for GnRH with nanomolar dissociation constants (K_d) (high affinity) (55,56). In certain tumor cells, only high-affinity GnRH-binding sites could be detected (57,58), or in other cases, solely low affinity GnRH-binding sites with micromolar K_d values were reported (51,56,58-61). Finally, the dichotomy of GnRH agonist and antagonist does not exist in tumor cells (62) (Table I). These discrepancies could possibly explain the functional differences present between tumor and pituitary cells after receptor activation.

Function of GnRHR in tumors from non-reproductive tissues. Specific binding sites by GnRH have been reported in several tumor specimens and cancer cell lines originating from organs, which are not part of the reproductive system. These include liver (63), larynx (64), pancreas (65), colon (66), lymphoma (67), kidney (68,69), skin (70,71), leukemia (49) and brain (72). The function of GnRH and GnRHR in these tumors has been associated with inhibition of proliferation in a time- and dose-related manner in respond to various molecular form of GnRH (63-72). The response to GnRH in pancreatic tumors has been shown to be specific, it was demonstrated by the absences of binding sites for GnRH in membranes from non-tumoral human pancreas tissues (65,73). Experiments carried out with cytotoxic radicals linking to GnRH agonists, showed a potent antitumor effect over xenografts of GnRHR-expressing cells from colon carcinoma, xenografts of GnRHR-expressing cells from non-Hodking's lymphoma and xenografts of GnRHR-expressing cells from human renal cell carcinoma (RCC) (66,69,74-76). The inhibition of growth of RCC xenografts in nude mice by administration of GnRH antagonist (Cetrorelix) was accompanied by a marked decrease in the number of EGF binding sites (Fig. 2A) (77). In melanoma-derived cells BLM and Me15392, GnRHR activation by Zoladex is able to support a high diminution in cell proliferation and matrigel invasion (70,71); in addition, in the case of leukemia, either overexpression on cell surfaces of 67-kDa LR cell surfaces or an increase in migration into bone marrow and spleen after GnRH stimulation was observed (49) (Fig. 2A).

Function of GnRHR in tumors from reproductive tissues. The presence of GnRHR and GnRH has been demonstrated in tumor of reproductive tract as ovarian cancer (57,58,78,79), prostate cancer (53,80-82), and breast cancer (59,83,84). The function of these molecules in these cells remains unclear, but has been associated with cell proliferation, and migratory and invasive capacity (71,85).

GnRHR and ovarian cancer. Numerous studies have demonstrated GnRH/GnRHR expression in ovarian cancer in *in vitro* and *in vivo* assays (41,79). In 70% of primary ovarian cancers and 83% of primary endometrial cancers, there is expression of high-affinity/low-capacity binding sites for GnRH receptors in their surfaces, and according to recent clinical data, receptor expression is a favorable prognostic factor (51,86,87). In the majority of cases, receptor activation was related with cell-proliferation inhibition (79,86). For

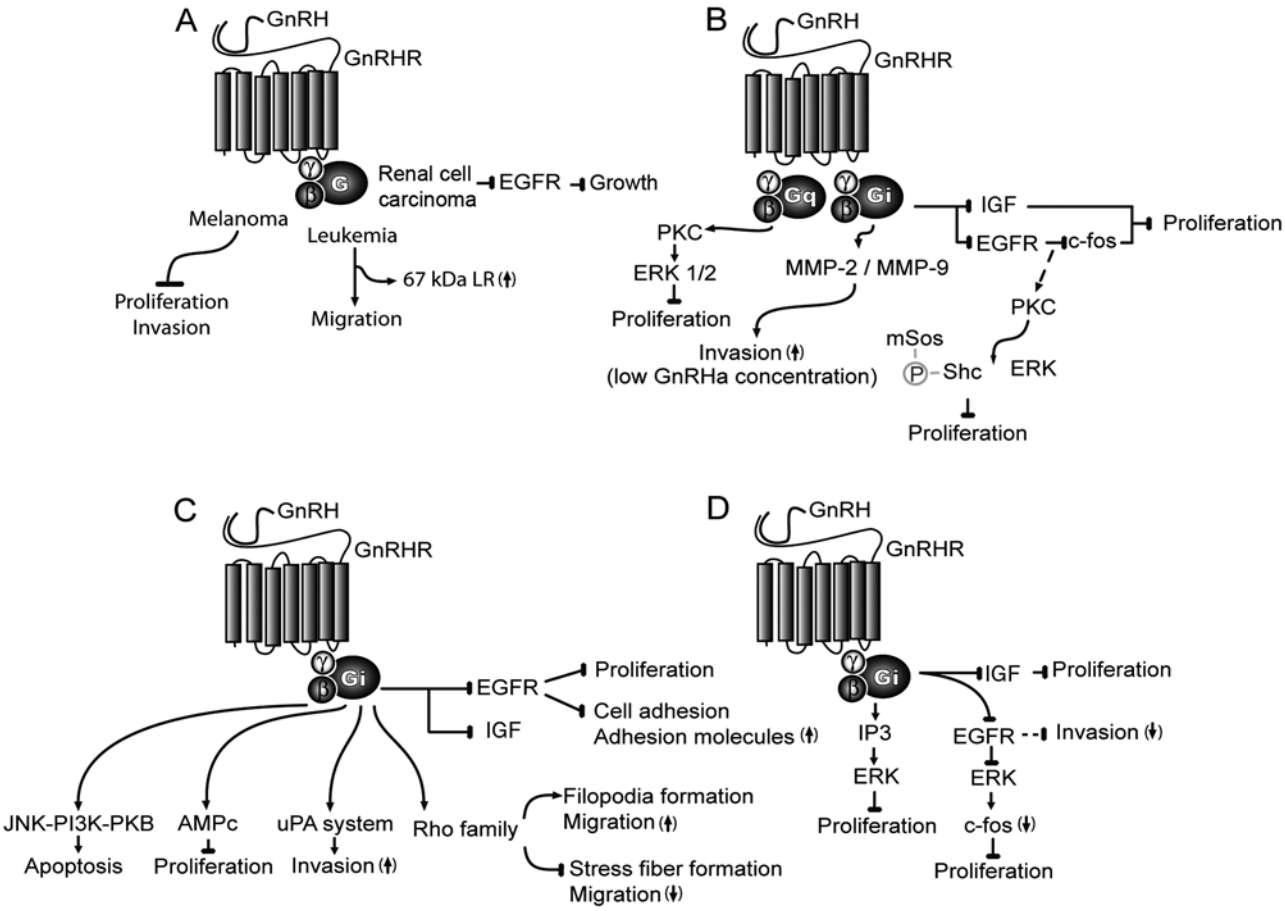

Figure 2. Signaling pathways activated by gonadotropin-releasing hormone receptor (GnRHR) in tumors from non-reproductive tissues and tumors of the reproductive tract. (A) In non-reproductive tissues GnRHR produced effects over cell proliferation and cell invasion. The signaling pathways activated by GnRHR in ovarian cancer (B), prostate cancer (C) and breast cancer (D). Coupling between GnRHR with Gi protein, cross-talk between GnRHR and growth factors receptors, and its association with different effector proteins could explain the versatility in extra-pituitary receptor function. Activation is represented by solid arrows, inhibition by a cut line, and non-well-described, by a dotted line.

human endometrial cancer cell lines (HEC-1A, Ishikawa, HHUA), GnRHR stimulation was able to down-regulate the cellular proliferation of these tumors via the PKC system (58,88). This effect was misplaced when neutralizing antibodies against GnRH were employed (89). The signaling pathway by which GnRHR acts in ovarian cancer is not well-detailed, but many reports have focused on understanding the manner in which this system works. Coupling between GnRHR and G_i protein in the membrane of surgically removed ovarian carcinoma and human ovarian cancer cells (Caov-3) raised the possibility that the antimitogenic action of this receptor may take place via the coupling of both molecules (33,90-92) (Fig. 2B). This interaction was linked with ERK activation and high phosphorylation levels of mSos, with the concomitant association between mSos and Shc (92) (Fig. 2B). On the other hand, mitogenic signal transduction and cell proliferation of the GnRHR effect could be associated with cross-talk between this receptor and growth factor receptors. In some ovarian cancer cells (EFO-21 and -27), GnRHR is also coupled with G_i protein, and their activation promote phosphotyrosine phosphatase action on EGFR (33). In DU-145 cells, this receptor abrogates EGFR-induced c-fos expression and reduces the concentration of EGF-binding sites, resulting in down-regulation of cellular proliferation (93) (Fig. 2B). In nude mouse model, reduction of insulin-like growth factor I (IGF) and EGFR after GnRH analog treatment was reported (94) (Fig. 2B). In addition, GnRHR is mainly coupled with G_i in ovarian-tumor cells, nevertheless in EFO-21 the anti-proliferative effect of this receptor was transduced by G_q protein (33). Activation of this G_q protein could also be related with the antiproliferative effect observed in SKOV-3 and Ovacr-3 cells, in which pronounced activation of ERK1/2 via PKC after GnRH was demonstrated (95) (Fig. 2B).

The effect of GnRHR and GnRH on the invasion ability of ovarian cancer has been investigated. For ovarian-cancer cell lines, Caov-3 and OVCAR-3, JNK activation was critical for up-regulation of MMP-2 and MMP-9 after GnRHa administration. During *in vitro* assays a low concentration of GnRHa increased cell motility and invasiveness, but no prominent effect was shown at high concentration (96) (Fig. 2B). These results support the idea that GnRHR possesses a bi-phasic impact on cellular function in tumors from non-pituitary cells (38,96) (Table I).

GnRHR and prostate cancer. Specific GnRH receptors have been found in nearly 80% malignant prostate tumors (53,97,98). In these tumor cells, GnRH production was also demonstrated, suggesting a local paracrine/autocrine system

(80). In rat prostate-cancer model, different values to GnRH receptor binding and capacities were found (99). On the other hand, specific GnRH binding sites have been reported for many tumor cell lines, such as LNCaP, an androgen-sensitive prostatic cancer cell line (100), DU145, a human androgen-independent cancer cell line (101), and PC-82, an androgen-dependent cell line (102). Similarly, the existence of this receptor was demonstrated in mouse and rat nude models (103,104), as well as in human prostate cancer biopsy (99). Based on the above evidence, several research groups have been employing GnRH analogs as antineoplastic drugs (102,105-109). GnRHR and GnRH in prostate cancer are able to promote a high diminution in cellular proliferation (100,101). However, the molecular mechanism responsible for this inhibition is unknown; notwithstanding this, a possible direct interaction between GnRH analogs and growth factor-receptor activity must be involved. Utilizing LNCaP and DU145 cells, down-regulation for EGF receptor after GnRHa treatment has been exhibited (93). Similar results were observed in DU145 cells after GnRHR activation, in which important inhibition of IGF mitogenic action was observed (110) (Fig. 2C). The signaling pathway involved in this latter process is uncertain, but at least a GnRHR-G_i-AMPc system has been associated (82,111) (Fig. 2C). Another possibility for explaining the antiproliferative effect exhibited by the GnRH/GnRHR system in prostate cancer comprises direct induction of apoptotic signaling (37, 112,113). The signaling pathway associated with this effect is unknown; however, a previous report supports the association with JNK and the phosphatidylinositol 3'-kinase (PI3K)-protein kinase B (PKB) pathway (37) (Fig. 2C).

GnRHR and its ligand are associated with invasion and metastatic potential by inducing actin cytoskeleton remodeling. In the DU145 cell, GnRHa was able to inhibit fibroblast growth factor (FGF)-stimulated cell proliferation, invasion and the ability of these cells to recover from a cytotoxic insult by exposure to etoposide, a topoisomerase II inhibitor (108). In TSU-Pr1 cells, GnRH induced filopodia formation and increases cell migration, while in DU145 cells this hormone promotes stress-fiber formation and abolishes cell migration (114). In this latter case, the mechanism activated by GnRH to regulate cell migration and actin cytoskeleton was related with small G protein from the Rho family (Fig. 2C). The effect of GnRH on cell invasion appears to be mediated by several specific steps. In DU145 and PC3 cells, it occurs by means of the regulation of the urokinase-type plasminogen activator (uPA) system (115) and increases in cell-cell adhesion molecules (116,117) (Fig. 2C). Collectively the information suggests that GnRHR activation could be associated with cell proliferation and motility in prostate cancer cells.

GnRHR and breast cancer. Specific GnRH binding sites were reported in biopsies of primary human breast carcinoma tissues (59). Nearly 50% of breast cancer specimens possess GnRH binding sites (118). For different human breast cancer cell lines (ZR-75-1, MDA-MB-231, Sk Br 3, MDA-MB-157 and MCF-7) GnRH-binding sites were reported (119). GnRHR and GnRH expression in tumoral and non-tumoral human mammary gland was tested, and no statistical differences were found at mRNA level for either the hormone or this receptor (84). The physiological function of GnRH and its agonist in breast cancer has been reported to inhibit the growth of cells in culture (56). In nude mouse models, complete regression of human breast carcinomas have been demonstrated experimentally (120-122), and its expression could be associated with a protective effect on the chemotherapeutic drug-produced apoptotic effect (123). Employing the adenovirus expression system, it was possible to produce a high-affinity binding site against Buserelin (1.4 nM) and high receptor levels at the surface in the MCF-7 cell line. This cellular model was able to support increasing levels of second messenger IP3 and ERK1/2 activation and a decrease in cellular proliferation after GnRH activation (60) (Fig. 2D). In addition, cellular proliferation decreased after addition of Buserelin or other GnRH agonists, this depended either on the amount of GnRH at cell surfaces or on receptor functionality (61,124). Concerning the molecular mechanism employed by GnRHR in breast cancer cells to reduce cell proliferation in MCF-7 cells, the direct inhibitory effects of Buserelin on breast cancer cells was mediated, at least in part, by an antagonists effect on the EGF receptor (125) and IGF receptor (126) (Fig. 2D). Complementary experiments were carried out in *in vivo* and *in vitro* models. In a mouse model treated with GnRH agonist, expression of both growth factor receptors was decreased (127). In 4OH-tamoxifen-resistant MCF-7 and T47D-TR cells, some GnRH analogs were able to abolish cell proliferation, blocking EGF-receptor autophosphorylation, ERK1/2 activation (128), and reducing EGF-induced c-fos protein expression (129) (Fig. 2D).

Finally, reduction in the metastatic potential of several breast cancer cell lines by GnRH agonists has been demonstrated. In different breast cancer cells lines (HCC70, MCF-7, MDA-MB-435, MDA-MB-453 and T47-D), their invasive capability was increased when these were co-cultured with hOB or MG63 osteosarcoma cells. This effect was reverted after GnRH analog treatments, demonstrating receptor ability to down-regulate cell motility in this cellular model (130) (Fig. 2D).

3. Action of GnRHR during cell motility and invasion

The complexity of GnRHR/GnRH regulation has made it attractive for considering the involvement of multiple transducer or regulatory systems. As mentioned previously, receptor coupling with different G proteins have been demonstrated. For example, association among GnRHR-G_q, G_{11}, G_i and G_s, in multiple mammalian cell lines such as COS7, HEK293, GH3, HeLa and CHO-K1 (35) had been shown. The G_i protein is coupled with GnRHR in GT1-7 cells and primary hypothalamic cells (34). The G_q, G_{11}, G_{14} and G_{15} proteins are associated in the GGH31-cell rat gonadotrope (32). In tumor cells, GnRHR could be mainly coupling with the G_i protein (33,82). The versatility in receptor coupling with many G proteins might be explained within a specific cell-type context; according to cell lineage, a different receptor conformation and signaling complex might be produced (17). This information clearly suggests a possible mechanism of the manner in which GnRHR could be associated with different actions in extra-pituitary cells (35).

Collectively the information suggests that GnRHR and GnRH form part of a sophisticated signaling pathway that is involved with many aspects of cell behavior, relating to cellular proliferation, but also with metastasis in tumor cells.

In αT3-1, a gonadotrope-derived cell line, GnRHR activity caused acute and dramatic changes in a cellular morphology induced by actin polymerization after ERK1/2 activation. At the same time, GnRHR was able to produce a high increase in cell motility, as well as detached cell behavior in this cellular model (131) (Fig. 1). HEK293 stably expressing GnRHR cells from rat, was able to produce either high extracellular matrix adhesion or rich actin structure or FAK, c-Src and ERK1/2 activation. It has also been demonstrated that Rac, but not RhoA, was active during GnRHR signaling (31).

In invasive human trophoblasts after GnRHR activation an increase was observed in the expression ratio of the urokinase-type plasminogen activator (uPA) and a concomitant decrease in plasminogen activator inhibitor (PAI-1) (132). Both proteins are members of a complex system related with cell migration, angiogenesis, wound healing, embryogenesis, tumor cell dissemination and metastasis in a variety of solid tumors (133). In addition, in trophoblasts GnRHR was able to increase MMP-2 and -9 at mRNA and protein levels and decreased their endogenous inhibitors TIMP-1 at mRNA and protein levels (134). This MMP activation is not only related with cell motility behavior, but also with cell proliferation via cross-talking between GnRHR and EGFR (135).

Clearly, the effect on cell motility and cell attachment was shown by cross-talk between EGFR/GnRHR. In breast cancer cells, administration of several GnRH analogs was able to produce a clear loss in EGFR function (128). In the prostate cancer cell line DU-145, GnRHR was able to produce high down-regulation in matrigel matrix invasion; this could be associated with down-regulation in EGFR expression and increases in cell-cell adhesion molecules such as E-cadherin, α- and ß-catenin and p120 (116) (Fig. 2D).

The effect of GnRHR on cell motility and cytoskeleton must be linked with several actin-associated molecules, such as GTPases from the Rho family (136,137). The principal function of these small G proteins is to modulate the cytoskeleton during cell locomotion in normal and tumor cells. This group is made up of six members, in which the majority of studies have been conducted on Rho, Rac and Cdc42 (138). The cellular function of Rho is to develop stress fiber and focal adhesion assembly (139). Rac is the main regulator of actin polymerization at the cellular membrane level, supporting lamellipodia and membrane-ruffling formation (140). Finally, Cdc42 supports filopodia develop-ment (141). The GTPase activity of these proteins is modulated by guanine diphosphate (GDP) or guanine triphosphate (GTP) nucleotide binding. In the active form, these proteins bind GTP, and in the inactive form, GTP is hydrolyzed to GDP. Conversion between both states is regulated by union with accessory proteins. Guanine nucleotide exchange factors (GEF) are proteins involved in the exchange of GDP by GTP. GTPase activating proteins (GAP) comprise the responsible molecules of GTP hydrolysis (142).

Many authors have established the link between activation of Rho family members and GnRHR; nonetheless, details in signaling pathways are unknown, suggesting new and interesting research opportunities.

4. Conclusions

Over the past decade, the presence of GnRH and GnRHR has been reported either in non-pituitary tissues or human tumors from reproductive and non-reproductive tissues or derived cell lines. These reports have associated GnRHR and GnRHR with sophisticated signaling pathways and novel functions. Coupling between this receptor and many G proteins, as well as cross-talk with a certain growth factor such as EGFR, could explain how these proteins are able to control cellular proliferation and metastatic potential in non-pituitary and tumor cells. A vast body of literature suggests that GnRH and GnRHR may control their mitogenic effect via MAPK or by the induction of apoptotic signaling. On the other hand, there must be multiple targets of GnRHR for modulating the cytoskeleton during cell motility; proteins from the Rho family or their regulator proteins could be associated. The mechanisms by which diverse members of the Rho family are associated with diverse cytoskeleton proteins will aid in clarifying how these fine-tuned interactions regulate cell mobility during GnRHR activation. In this regard, understanding these novel cellular processes at the cellular and molecular levels will lead to development of new therapeutic resources in the control of tumor growth and cancer dissemination.

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