Membrane Progesterone Receptors: Evidence for Neuroprotective, Neurosteroid Signaling and Neuroendocrine Functions in Neuronal Cells

Peter Thomas  Yefei Pang
The University of Texas at Austin Marine Science Institute, Port Aransas, Tex., USA

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Abstract
Membrane progesterone receptors (mPRs) are novel G protein-coupled receptors belonging to the progestin and adipocytokine receptor family (PAQR) that mediate a variety of rapid cell surface-initiated progesterone actions in the reproductive system involving activation of intracellular signaling pathways (i.e. nonclassical actions). The mPRs are highly expressed in the brain, but research on their neural functions has only been conducted in a single neuronal cell line, GT1–7 cells, which have negligible nuclear progesterone receptor (PR) expression. GT1–7 cells express mPR\(\alpha\) and mPR\(\beta\) on their plasma membranes which is associated with the presence of high-affinity, specific \([\text{H}]\)-progesterone receptor binding. The neurosteroid, allopregnanolone, is an effective ligand for recombinant mPR\(\alpha\) with a relative binding affinity of 7.6% that of progesterone. Allopregnanolone acts as a potent mPR agonist on GT1–7 cells, mimicking the progesterone-induced decrease in cAMP accumulation and its antiapoptotic actions at low nanomolar concentrations. The decrease in cAMP levels is associated with rapid progesterone-induced downregulation of GnRH pulsatile secretion from perfused GT1–7 cells. The recent suggestion that mPRs are alkaline ceramidases and mediate sphingolipid signaling is not supported by empirical evidence that TNF\(\alpha\) does not bind to mPRs overexpressed in human cells and that exogenous sphingomyelinase is ineffective in mimicking progesterone actions through mPRs to induce meiotic maturation of fish oocytes. Taken together, these recent studies indicate that mPRs mediate neuroprotective effects of progesterone and allopregnanolone and are also the likely intermediaries in progesterone-induced inhibition of pulsatile GnRH secretion in GT1–7 cells.

Introduction

The central role of two intracellular ligand-activated transcription factors, progesterone receptors PR-A and PR-B, in mediating progesterone’s regulation of diverse female vertebrate reproductive functions through altering gene transcription has been unequivocally demonstrated using PR-selective ligands and PR knockout (PRKO) mice [1]. However, many of progesterone’s actions are too rapid to be readily explained by a genomic mechanism which typically occurs over a time scale of hours, and it is now widely appreciated that progesterone,
like other steroid hormones, can also exert rapid cell surface-initiated actions within minutes through activation of membrane receptors and their associated intracellular signaling pathways [2–5]. For example, alternative (i.e. nonclassical) progesterone actions on sperm motility, oocyte meiotic maturation, granulosa cell apoptosis, immunosuppression of T cells, breast and ovarian cancer cells, GnRH secretion, and reproductive behaviors have been identified and the receptors mediating many of these effects have been biochemically characterized [6–16]. Although some of these alternative progesterone actions are nongenomic, others may ultimately lead to altered gene transcription through activation of second messengers such as MAP kinases and through alteration of PR transactivation through effects on coactivators such as SRC2 [17].

The identities of the receptors mediating many of these nonclassical progesterone actions remain unclear [5, 18]. Activation of PRs located in the cytoplasmic compartment can mediate nonclassical progesterone signaling through an interaction with Src kinase [19], but other receptor mechanisms must be invoked to explain the cell surface-initiated progesterone actions identified in many target tissues. Although progesterone and its metabolite, allopregnanolone, have been shown to induce rapid effects in neural tissues through modulation GABA_A receptor activity [20, 21], rapid progesterone actions in nonneural tissues, which lack GABA_A receptors, must involve alternative receptor mechanisms. There is substantial evidence that a putative membrane receptor, progesterone membrane receptor component 1 (PGMRC1), is a component of nonclassical signaling by progesterone and other hormones [22–24], but its exact role is uncertain and clear evidence that it functions as a specific progesterone receptor is lacking [22, 25, 26]. In contrast, extensive evidence has been obtained by different research groups that wild-type membrane progesterone receptors (mPRs) in a wide range of vertebrate cells as well as recombinant proteins expressed in prokaryotic and eukaryotic systems display high-affinity, specific, displaceable and limited capacity progestin binding characteristic of steroid membrane receptors [5, 15, 25, 27–33]. The mPRs are typically localized on the cell surface and are widely distributed throughout neural, reproductive and nonreproductive vertebrate tissues and are therefore good candidates for the membrane receptors mediating many of these nonclassical cell surface-initiated progesterone actions [27, 34–37]. The mPRs were discovered and first characterized in fish ovaries [15], and three isoforms, mPRα, mPRβ, and mPRγ, were subsequently identified in humans and other vertebrates [27]. The mPRs are 7-transmembrane proteins and activate G proteins; however, they do not belong to the G protein-coupled receptor superfAMILY, but are instead members of the progestin and adipocytokine (PAQR) family [28, 34]. The characteristics and functions of mPRs have been discussed extensively in several recent publications [5, 38–40].

In this short review, potential functions of mPRs in neural tissues will be briefly discussed. New data suggesting that progesterone and the neurosteroid allopregnanolone exert protective effects on mouse neuronal GT1–7 cells through mPRs are discussed as well as recent evidence obtained with this cell line for the involvement of mPRs in progesterone downregulation of GnRH pulsatile secretion. The mPRs have recently been proposed to function as alkaline ceramidases based on the results of bioinformatic analyses and investigations of their functions in heterologous yeast systems [40], but the involvement of mPRs in sphingolipid signaling in vertebrate cells has not been reported. Therefore, new data on potential interactions of TNFα with mPRα, mPRβ, and mPRγ overexpressed in human breast cancer cells and on the ability of exogenous sphingomyelinase to mimic progestin induction of meiotic maturation of fish oocytes through mPRs are also discussed.

**Role of mPRs in the Antiapoptotic Actions of Progestins in Reproductive Tissues**

Progesterone exerts a physiologically important function in the ovary to inhibit apoptosis of granulosa cells. However, the mechanism of progesterone’s antiapoptotic action is unclear because it does not involve activation of the nuclear PR in several cell models [13, 41, 42], but instead is initiated at the cell surface which suggests it is mediated through mPRs [43, 44]. Both PGRMC1 and mPRs have been implicated in these antiapoptotic actions of progestins on granulosa cells [13, 45]. PGRMC1 forms a complex with plasminogen activator inhibitor RNA binding protein (PAIRBP-1) and both of these proteins are localized on the plasma membranes of spontaneously immortalized granulosa cells (SIGCs) [13, 46]. Treatment with either the siRNA or an N-terminal antibody to PGRMC1 blocked the antiapoptotic actions of progesterone on SIGCs, suggesting involvement of PGRMC1 in this progesterone action [47]. Interestingly, progesterone at high concentrations (1 μM) inhibits the expression of the proapoptotic gene BAD and increases the expression of the antiapoptotic BCL2 family member, BCL2A1D,
through PGRMC1 in SIGCs [48]. Decreased cell death and antiapoptotic actions of progesterins have also been shown to be mediated through mPRs in granulosa and breast cancer cells [12, 45]. The mPRα protein is localized on the plasma membranes of teleost granulosa cells and on nuclear PR-negative human breast cancer cells. The demonstration that a specific mPR agonist, Org OD 02-0 [49], mimics the protective effects of progesterin hormones on serum starvation-induced cell death and apoptosis in both granulosa and breast cancer cells indicates mPRs are intermediaries in progesterin-induced cell survival. This protective role of mPRs has been confirmed in both cell types by knockdown studies which show that decreased mPRα protein expression after treatment with mPRα siRNA is associated with decreased [3H]-progesterone binding and a loss of the antiapoptotic actions of progestins [12, 45]. The finding that progesterins activate MAP kinase and Akt through mPRs in both cell types [12, 45] is consistent with mPR's antiapoptotic functions because both MAP kinase and Akt are involved in inhibition of cell death and apoptosis [50]. MAP kinase upregulates antiapoptotic members of the BACL2 family and inhibits BAD [51, 52], which is also inhibited by Akt [53]. Inhibition of apoptosis in breast cancer cells by treatment with the mPR-specific ligand, Org OD 02-0, was also accompanied by an increase in mitochondrial membrane potential which has been shown to correlate with increased cell survival. However, caspase 3 activity was unaltered through mPRs after treatment with Org OD 02-0 [12]. Similarly, caspase gene expression was not altered through PGRMC1 in SIGCs [48]. Thus, progesterins appear to exert their antiapoptotic actions through mPRs and PGRMC1 by similar mechanisms which do not involve activation of caspases.

**Potential Role of mPRs in the Antiapoptotic Effects of Allopregnanolone in the Brain**

There is extensive evidence that progesterone also exerts a variety of protective effects in the brain, including inhibition of apoptosis [54]. Many of these progesterone effects are mediated through its metabolite, allopregnanolone, a neurosteroid synthesized in the central and peripheral nervous systems [54–56]. Allopregnanolone accumulates during the diurnal cycle and during pregnancy in mammals: mouse brain concentrations increasing up to 35 nM during the scotophase and plasma concentrations in women reaching 100 nM during the third trimester of pregnancy [55]. Allopregnanolone does not activate nuclear PRs and is widely believed to be devoid of direct hormonal action through steroid receptors [54, 56, 57]. Instead allopregnanolone is thought to exert its antiapoptotic actions in the brain by binding to GABA\textsubscript{A} receptors, causing their allosteric modulation and an increase in their neuronal cell inhibitory activity, as well as by allosteric modulation of N-methyl-D-aspartic acid (NMDA) receptors, reducing their NMDA-induced excitotoxicity by a noncompetitive mechanism, and also potentially through activation of pregnane X-dependent pathways [54]. However, the results of recent ligand-binding specificity studies with recombinant human mPRα challenge the prevailing view that allopregnanolone does not activate steroid hormone receptors. Allopregnanolone was found to have a relatively high binding affinity, 7.6% that of progesterone, for human mPRs overexpressed in MDA-MB 231 human breast cancer cells [28, 49]. The finding that allopregnanolone, which has been shown to mediate antiapoptotic progesterin actions, can interact with mPRα suggests an alternative mechanism by which this neurosteroid could potentially exert neuroprotective effects in the brain. Initial experiments on potential protective effects of allopregnanolone on cell death and apoptosis through mPRs in neuronal cells have been conducted with a mouse GnRH-secreting neuronal cell line (GT1–7 cells) in which mPR receptor binding and signaling have been partially characterized [58]. The nuclear PR cannot be detected in GT1–7 cells, whereas there is significant expression of mPRα and mPRβ mRNAs and proteins [58]. RT-PCR shows that mPRα mRNA is abundantly expressed in GT1–7 cells, with a lesser amount of mPRβ, and relatively low expression of mPRα and mPRε mRNAs (fig. 1a). Progesterone activates an inhibitory G protein (G\textsubscript{i}) in these cells resulting in downregulation of cAMP, which was abrogated after treatment of the cells with mPRα siRNA, indicating this cAMP response is mediated through mPRα [58]. Therefore, to investigate whether allopregnanolone acts as an mPR agonist, mimicking the actions of progesterone, we investigated its ability to downregulate adenylyl cyclase activity.

Treatment of serum-starved GT1–7 cells with allopregnanolone for 15 min mimicked the inhibitory effects of progesterone on cAMP levels, significantly decreasing cAMP production at low concentrations of 20 nM and 100 nM (fig. 1b). Allopregnanolone also mimicked the inhibitory effects of progesterone on serum starvation-induced cell death and apoptosis (fig. 1c, d). Interestingly, allopregnanolone exerted inhibitory effects on cell death and apoptosis at low physiological concentrations of 100 and 20 nM, respectively, whereas modulatory effects of the
Neurosteroid on GABA<sub>A</sub> receptor functions are usually observed at nanomolar concentrations [20, 59]. These protective actions of allopregnanolone are likely mediated through the most abundant mPR subtype in GT1–7 cells, mPR<sub>α</sub>, and by activation of MAP kinase and Akt, as observed previously in teleost granulosa cells and in PR-negative human breast cancer cells [12, 45]. The mPRs could potentially exert neuroprotective effects throughout the central nervous system because they are widely distributed in the brain and spinal cord [27, 34, 37]. For example, the rat cortex, where progesterone has been shown to exert maximal protective effects through MAP kinase and PI3-kinase pathways [60], shows high expression of mPR<sub>α</sub> and mPR<sub>β</sub> [35]. All five members of the mPR subfamily of PAQRs (class II PAQRs) have been identified in human brains, including a brain-specific homolog, mPR<sub>δ</sub> (PAQR6 [34]). Therefore, it is also important to investigate the interactions of progesterone, allopregnanolone and other neurosteroids with mPR<sub>δ</sub> and the other mPRs (β, γ, ε). In conclusion, these preliminary results suggest a potential involvement of mPRs in the antiapoptotic effects of allopregnanolone in the central nervous system.

**Potential Role of mPRs in Negative Feedback of Progesterone on Gonadotropin-Releasing Hormone Secretion**

Progesterone and neurosteroids influence a wide variety of brain functions including sexual behavior, neuroendocrine function and intracellular second messenger signaling [6, 10, 61]. Studies using the specific PR antagonists RU486 and PRKO mice have clearly implicated the nuclear PRs, PR-A or PR-B, in genomic progesterone actions on LH surges and facilitatory effects on female sexual behavior [62–64], as well as negative feedback progesterone effects on GnRH pulsatile secretion in sheep and rats [65, 66]. However, many neural effects of proges-
terone are too rapid to be mediated by a genomic mechanism [10, 67–70]. The teleost progesterin, 17, 20β, 21-trihydroxy-4-pregnen-3-one (20β-S) decreases GnRH secretion from Atlantic croaker hypothalamic tissue in vitro to 20% that of controls within the initial 10-min incubation period [61]. Some of these rapid progesterone actions in the brain could be mediated by the PR because the receptor has been shown to induce rapid extranuclear progesterone signaling through activation of Src tyrosine kinase [18, 19]. However, other progesterone and neurosteroid actions do not appear to be mediated through PRs. For example, RU486 failed to prevent potentiation of LH secretory responses to multiple GnRH pulses in rats [71], and rapid allopregnanolone effects on lordosis persist in PRKO mice [11]. Allopregnanolone can influence GnRH secretion and other neural functions through modulation of GABA A receptor activity [21, 72, 73], but can also activate mPRs as shown in figure 1. The mPRs and PGRMC1 are expressed in the hypothalamus and other brain regions controlling reproductive functions and therefore are candidates for the intermediaries of some rapid progestin actions on reproductive behavior and neuroendocrine functions [27, 34, 35, 58].

Recently, progesterone injections that resulted in high physiological levels of the hormone, such as those observed during pregnancy and the proestrus progesterone surge, were found to acutely suppress in vivo LH secretion in both wild-type and PRKO ovariectomized mice [58]. This PR-independent progesterone action is exerted at the hypothalamus and specific for this steroid since injections with the same dose of dexamethasone (400 μg/ml) were ineffective. A low dose of allopregnanolone (15 μg/ml) also did not suppress LH secretion [58], but injections with higher amounts of the neurosteroid were not tested, so it is unclear whether allopregnanolone can mimic these progesterone effects.

Superfusion of GT1–7 cells, which express negligible amounts of PR, with progesterone caused a rapid down-regulation of GnRH secretion. This rapid progesterone action was associated with a decrease in cAMP concentration in GT1–7 cells which was attenuated by pretreatment with pertussis toxin, a blocker of G i activation. A subsequent experiment demonstrated progesterone treatment caused G protein activation as assessed by an increase in [35S]-GTPγS cell membrane binding which was immunoprecipitated with a G i α-subunit antibody, confirming that progesterone activates a G i in GT1–7 cells [58]. The finding that the progesterone inhibition of cAMP accumulation in these cells was not mimicked by the selective PR agonist, R5020, indicates this action is independent of the PR. Earlier studies had shown that progesterone and other progestins activate pertussis-sensitive G i through mPRs and mPRβ in a wide variety of cell types including fish oocytes, human myometrial cells, human lymphocytes and human breast cancer cells [15, 17, 28, 74]. Sleiter et al. [58] showed that mPRα and mPRβ proteins are expressed in GT1–7 cells and localized to the plasma membranes where high-affinity, specific [3H]-progesterone binding was detected that was not displaced with 10−6 M R5020, which is characteristic of mPRs. Finally, it was demonstrated that transfection of mPRα siRNA into GT1–7 cells decreased membrane [3H]-progesterone binding and attenuated the inhibitory effect of progesterone on cAMP accumulation. Taken together, these results indicate mPRα is a potential intermediary in these rapid negative feedback effects of progesterone on GnRH pulsatility. However, direct evidence for this role for mPRα such as the demonstration that this action in superfused GT1–7 cells is abolished after transfection with mPRα siRNA has not been obtained due to technical limitations. Therefore, the identity of the progesterone receptor mediating this effect of high progesterone concentrations on GnRH secretion remains unclear. An alternative approach, the use of a specific mPR agonist such as Org OD 02-0, would overcome the problems of using siRNA on GT1–7 cells, and would provide a clear indication of whether mPRα has a role in this neuroendocrine control mechanism.

**Potential Role of mPRs in Sphingolipid Signaling**

Recent bioinformatic studies on the sequence similarities of PAQRs with alkaline ceramidases and other 7-transmembrane proteins suggest they belong to a large diverse family of distantly-related putative transmembrane hydrolases which has been named CREST (alkaline ceramidase, PAQR, Per1, SID-1 and TMEM8) [40, 75]. Alkaline ceramidases catalyze the deacylation of ceramides to yield sphingoid bases, which function as lipid second messengers. On the basis of their finding that the fungal PAQR, Izh2p, has a possible ceramidase function, Lyons and coworkers [76] proposed that PAQRs can act as ligand-activated alkaline ceramidases. Using a heterologous yeast expression system and an endogenous PAQR (Izh2p) driven reporter, these researchers showed that both an alkaline ceramidase inhibitor and supraphysiological concentrations of a homolog of adiponectin, tumor necrosis factor α (TNFα), inhibited signaling through recombinant human adiponectin receptors and mPRβ, mPRγ and mPRδ, but not through mPRα or
They concluded that TNFα is a PAQR antagonist and does not act downstream of the recombinant human receptors because signaling through Izh2p in this reporter system was unaffected by TNFα. However, the relevance of these results obtained in a heterologous yeast system which does not encode mPRs, TNFα members or TNFα receptors to vertebrate physiology remains unclear. It is essential to obtain empirical evidence confirming this action of TNFα in vertebrate cells before proposing mPRs can function as alkaline ceramidases. Therefore, we examined the potential of a high concentration of TNFα (4 μg/ml, 240 nM) to antagonize progesterone actions through mPRs by assessing its ability to compete for specific [3H]-progesterone binding to the receptors overexpressed on the plasma membranes of a nuclear PR-negative human breast cancer cell line, MDA-MB-231 cells. TNFα failed to compete for [3H]-progesterone binding to mPRα, in agreement with the results in the yeast expression system, and its homolog, adiponectin, was also an ineffective competitor of progesterone binding (fig. 2a). However, in contrast to the findings in the yeast system, TNFα also did not display any potential antagonism of progesterone activity through mPRβ and mPRγ because it did not displace specific [3H]-progesterone binding to these receptors (fig. 2b, c). Thus, the results of the competitive binding assays in human cells do not support the suggestion that TNFα is an antagonist or inverse agonist of progesterone actions through mPRs at the receptor level, at least not via a conventional mechanism by interfering with ligand binding.

TNFα and a variety of other factors can induce the production of sphingolipids such as ceramide through several

Fig. 2. The proposed role of mPRs in sphingolipid signaling is not supported by empirical data showing a lack of TNFα binding to mPRs (a–c) and the ineffectiveness of exogenous neutral sphingomyelinase in inducing meiotic maturation of zebrafish oocytes (d). a–c Single point competition with 1 μM progesterone (P4), 4 μg/ml TNFα or 4 μg/ml adiponectin (adip) for [3H]-progesterone binding to plasma membranes of MDA-MB-231 breast cancer cells over expressing mPRα (a), mPRβ (b) or mPRγ (c). Data represent mean percent specific [3H]-P4 binding ± SEM, n = 3. ** p < 0.0001 compared with vehicle (veh) control by one-way ANOVA and Dunnett’s multiple comparison test. d Comparison of the effects of exogenous B. cerus sphingomyelinase (SMase, 0.5 and 1.0 U/ml) and 17,20β-dihydroxy-4-pregene-3-one (DHP, 20 and 50 nM) on percent GVBD of denuded zebrafish oocytes. Large, fully grown zebrafish oocytes (diameter >550 μm) were denuded and the oocytes (30–40 oocytes/1 ml Leibovitz L-15 medium/well, 4 wells/treatment) were incubated with the various treatments for 3 h and scored at the end of the incubation period for GVBD as described previously [91]. Data represent mean percent oocytes completing GVBD ± SEM, n = 4. * p < 0.05 compared with vehicle control by one-way ANOVA and Dunnett’s multiple comparison test.
pathways, including the hydrolysis of sphingomyelin by sphingomyelinases (SMases) [78]. Moussatche and Lyons [40] have recently discussed evidence supporting an involvement of sphingolipids in nonnongenic signaling through mPRs. These authors point out that ceramide and exogenous SMase can mimic the actions of progesterone in inducing maturation of amphibian oocytes in vitro [79–81]. However, seasonal differences in the sensitivity of frog oocytes to induction of meiosis by progesterone and ceramide as well as different effects of them on membrane microdomain integrity toad oocytes have been observed [79, 81, 82]. Moreover, the importance of mPRs in mediating the nongenonic action of progesterone to induce oocyte meiotic maturation in amphibians remains unclear because nuclear PRs or PR-like forms have also been implicated [83, 84]. In contrast, extensive evidence has been obtained that mPRs are the primary intermediaries in progestin induction of oocyte maturation in teleosts [5]. The demonstration that microinjection of antisense oligonucleotides to mPRα into zebrafish and goldfish oocytes blocks progestin-induced oocyte maturation in these species [15, 33] provides the clearest evidence to date of a specific function of mPRs in any vertebrate model. Previous studies have shown that treatment of denuded Xenopus (X. laevis), frog (R. pipiens) or toad (R. arenarum) oocytes with 0.25–1.0 units/ml neutral SMase induces germinal vesicle breakdown (GVBD) with a similar time course to that of 0.25–1.0 units/ml neutral SMase induces germinal vesicle breakdown (GVBD) with a similar time course to that of progesterone [79, 81, 82]. Therefore, we investigated the effects of neutral SMase in the well-characterized teleost mPR model of progestin induction of oocyte maturation. Removal of the ovarian follicle layers surrounding zebrafish oocytes results in spontaneous meiotic maturation (GVBD) which is significantly increased in the presence of 17,20β-dihydroxy-4-pregene-3-one (DHP), the maturation inducing steroid in this species (fig. 2d; [5]). The percent of zebrafish oocytes completing GVBD after treatment with 0.5 and 1.0 U/ml Bacillus cereus neutral SMase was not significantly different from that in the vehicle-control group, which suggests mPR signaling to induce oocyte maturation does not involve sphingolipids (fig. 2d). To the best of our knowledge there are no reports of stimulatory effects of SMase or ceramide on oocyte meiotic maturation in any teleost species amongst the extensive literature on hormonal regulation of this process in fish. The absence of any evidence for an involvement of sphingolipid signaling in mPR induction of oocyte maturation in teleosts suggests that it is not an intrinsic characteristic of mPRs in homologous vertebrate cell models.

Finally, the role of mPRs in mediating the antiapoptotic effects of progestins in croaker granulosa and human breast cancer cells is inconsistent with their involvement in sphingolipid signaling because SMases and ceramide have been shown to be primary mediators of an opposite effect, apoptosis, in a wide range of tissues and cell types, including granulosa and neuronal cells [85–87]. For example, ceramide suppresses prosurvival signaling pathways such as Akt [88]. SMases induce ceramide and cytokine production after ischemia and neurons lacking the enzyme have increased survival and decreased vulnerability to ischemia [89]. In addition, SMase inhibitors have proven useful for treatment of neuronal apoptosis [90]. It is concluded that the evidence obtained to date in vertebrate cell models does not support the suggestion that mPRs are alkaline ceramidases involved in sphingolipid signaling. However, the ability of other vertebrate PAQRs such as the adiponectin receptors to have this function has not been extensively investigated and remains a possibility.

Concluding Remarks

Recent studies showing that mPRs are widely expressed in the central nervous system and that one mPR isoform, mPRδ (PAQR6), is exclusively expressed in the brain suggests that mPRs are likely intermediates in a variety of nonclassical progesterone actions in neural tissues. The finding that allopregnanolone can act as an mPR agonist at low physiologically relevant concentrations indicates an additional receptor mechanism by which neurosteroids can potentially modulate neural functions. Our preliminary results in GT1–7 cells suggesting that neuroprotective effects of progesterone and allopregnanolone are partially mediated through mPRs need to be confirmed in additional neuronal models. The receptors are also potential intermediaries in progesterone modulation of GnRH secretion under certain conditions, but direct evidence is lacking. More comprehensive studies using a combination of in vivo and in vitro models, additional experimental approaches such as the use of specific mPR agonists and siRNA technology, and investigations of additional targets such as mPRδ will be required to obtain a clearer understanding of the physiological functions of mPRs in the brain.

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References


Neural Functions of mPRs

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25 Thomas P: Characteristics of membrane progesterone receptor alpha (mPRa) and progesterone membrane component 1 (PGRMC1) and their roles in mediating rapid steroid actions. Front Neuroendocrinol 2008;29:292–312.
34 Intlekofer KA, Petersen SL: Distribution of mRNAs encoding classical progestin receptor, progesterone membrane components 1 and 2, serpine mRNA binding protein 1, and progestin and ADIPOQ receptor family members 7 and 8 in rat forebrain. Neuroscience 2011;172:55–65.


Neuroendocrinology 2012;96:162–171

Thomas/Pang
71 Turgeon JL, Waring DW: Luteinizing hormone secretion from wild-type and progesterone receptor knockout mouse anterior pituitary cells. Endocrinology 2001;142:3108–3115.


