The brain as a source and a target of prolactin in mammals

https://doi.org/10.4103/1673-5374.332124

Date of submission: April 13, 2021

Date of decision: July 3, 2021

Date of acceptance: August 2, 2021

Date of web publication: January 7, 2022

Ana R. Costa-Brito[1], Isabel Gonçalves[1, 2], Cecília R. A. Santos[1, 2, *]

From the Contents

Abstract

Prolactin is a polypeptide hormone associated with an extensive variety of biological functions. Among the roles of prolactin in vertebrates, some were preserved throughout evolution. This is the case of its function in the brain, where prolactin receptors, are expressed in different structures of the central nervous system. In the brain, prolactin actions are principally associated with reproduction and parental behavior, and involves the modulation of adult neurogenesis, neuroprotection, and neuroplasticity, especially during pregnancy, thereby preparing the brain to parenthood. Prolactin is mainly produced by specialized cells in the anterior pituitary gland. However, during vertebrate evolution many other extrapituitary tissues do also produce prolactin, like the immune system, endothelial cells, reproductive structures and in several regions of the brain. This review summarizes the relevance of prolactin for brain function, the sources of prolactin in the central nervous system, as well as its local production and secretion. A highlight on the impact of prolactin in human neurological diseases is also provided.

Key Words: brain; brain disease; choroid plexus; neurogenesis; neuroplasticity; neuroprotection; prolactin; prolactin receptor

Introduction

Prolactin (PRL) is a pleiotropic hormone responsible for many biological functions that go far beyond its roles in reproduction and lactogenesis, like the modulation of the immune system, growth and metabolism, osmoregulation and regulation of brain function. These are well-documented roles of PRL in mammals, as well as in other vertebrates (Bernard et al., 2019; Dobolyi et al., 2020). The effects of PRL on the mammalian brain depend on factors such as age, gender, and reproductive status (Lajud et al., 2013; Salais-López et al., 2018; Phillipps et al., 2019). The functions of PRL in the brain are particularly relevant during pregnancy and lactation which are characterized by high levels of PRL (Phillipps et al., 2020). PRL induces neurogenesis during pregnancy in mice (Shingo et al., 2003; Larsen and Grattan, 2010; Wang et al., 2013) and parental behaviors, which have been conserved along evolution, from fish to mammals (Dobolyi et al., 2020), and modulates other brain functions (Leem et al., 2019; Vermani et al., 2020).

The main goal of this review is to highlight the most recent findings on the effects of PRL in the function of the mammalian brain. A brief overview of the main signaling cascades elicited by PRL in the brain and the distribution of extrapituitary sources of PRL in the central nervous system is initially provided followed by a thorough review on the role of PRL in the induction of neurogenesis, neuroprotection and neuroplasticity. The therapeutic potential of this neuropeptide in brain disorders is also discussed.

Search Strategy and Selection Criteria

Research articles published between January 2016 and May 2021 were retrieved from the PubMed database using the following terms: prolactin, brain, parental behavior, prolactin receptor signaling, neurogenesis, neuroprotection, neuroplasticity, and neurological disorder. Although bibliographic revision focused on the last 5 years, earlier mandatory references in the field were also considered.

Prolactin Structure and Prolactin Variants

PRL is a polypeptide hormone mainly produced in the lactotrophs of the anterior pituitary gland (Bernard et al., 2019). The mature PRL protein of pituitary origin is composed of 197–199 amino acids in mammals (Grattan, 2015). PRL appeared early in evolution and remain relatively conserved (Dobolyi et al., 2020), presenting high homology levels in mammals (**Table 1**). In vertebrates, PRL is encoded by a single gene composed of 5 exons and 4 introns (Bernard et al., 2015). However, despite sharing some common structural and regulatory features, the PRL gene is distinct between species. For instance, the human PRL gene contains an additional noncoding exon. This superdistal promoter region, firstly identified in the human decidua, is responsible for the regulation of its transcription at extrapituitary sites (Hiraoka et al., 1991).

PRL is part of a family of polypeptide hormones that have similar structural and biological characteristics, which includes the growth hormone and placental lactogen. Human PRL is a 23 kDa protein, structurally composed by four antiparallel α-helices (Molina-Salinas et al., 2021). Moreover, several other PRL isoforms, resulting from proteolytic cleavage, alternative splicing and post-translational modifications like glycosylation, dimerization and association with other circulating proteins, have also been described (Bernard et al.,

[1]CICS-UBI- Health Sciences Research Centre, Universidade da Beira Interior, Covilhã, Portugal; [2]C4-UBI-Cloud Computing Competence Centre, Universidade da Beira Interior, Covilhã, Portugal

*__Correspondence to:__ Cecília R. A. Santos, PhD, csantos@fcsaude.ubi.pt.

https://orcid.org/0000-0001-6074-7825 (Cecília R. A. Santos)

Funding: *This work was supported by Fundação para a Ciência e Tecnologia (FCT, I.P., Portugal, http://www.fct.p) project grants UID/Multi/00709/2013 and UID/Multi/00709/2019. ARCB is a recipient of a PhD fellowship funded by the FCT (SFRH/BD/121890/2016), supported by the State Budget of the Portuguese Ministry of Science, Technology and Higher Education, by the European Commission and by the European Social Fund through the "Programa Operacional Regional Centro". This work was also funded by the grant "Sociedade Portuguesa de Endocrinologia Diabetes e Metabolismovartis Oncology em Patologia Hipofisária 2019" and the European Regional Development Fund through the "Programa Operacional Regional do Centro (Centro 2020)—Sistema de Apoio à Investigação Científica e Tecnológica—Programas Integrados de IC&DT" (Project Centro-01-0145-FEDER-000019—C4—Centro de Competências em Cloud Computing).*

How to cite this article: *Costa-Brito AR, Gonçalves I, Santos CRA (2022) The brain as a source and a target of prolactin in mammals. Neural Regen Res 17(8):1695-1702.*

Table 1 | **Comparison of prolactin gene/protein in different species**

Species	Exon Number	Size (amino acids)	Homology*	Accession number†
Human (*Homo sapiens*)	6	227	–	NP_001157030.1
Chimpanzee (*Pan troglodytes*)	6	227	98.68%	XP_016810474.1
Rhesus monkey (*Macaca mulatta*)	6	227	97.80%	NP_001040593.1
Rabbit (*Oryctolagus cuniculus*)	5	227	78.85%	NP_001076144.1
Sheep (*Ovis aries*)	5	240	72.69%	NP_001009306.1
Cattle (*Bos taurus*)	5	229	73.13%	NP_776378.2
Chicken (*Gallus gallus*)	5	229	66.52%	NP_990797.2
Rat (*Rattus norvegicus*)	5	226	62.22%	NP_036761.1
Frog (*Xenopus laevis*)	5	230	61.23%	NP_001093699.1
Mouse (*Mus musculus*)	5	228	60.89%	NP_035294.2
Zebrafish (*Danio rerio*)	5	210	31.88%	NP_852102.2

* Homology (percent identity) between human prolactin precursor protein and prolactin precursor protein from different species was analyzed using the multiple sequence alignment Clustal Omega program (runed online in EBI web server, https://www.ebi.ac.uk/Tools/msa/clustalo/). † Input protein sequences were downloaded from NCBI protein database (https://www.ncbi.nlm.nih.gov/protein/).

2015). It is hypothesized that different PRL isoforms have distinct functions and biological activity, contributing in part to the plethora of PRL functions. For instance, in humans, macroprolactin (> 100 kDa) has reduced biological activity (Fahie-Wilson and Smith, 2013). On the other hand, vasohinibin (a 16 kDa PRL isoform) is a potent anti-angiogenic factor, while 23 kDa PRL seems to have angiogenic properties (Struman et al., 1999).

Prolactin Action Is Mediated by Its Receptors

PRL receptors (PRLR) belong to the type I cytokine receptor family and are composed by three domains: extracellular, transmembrane, and intracellular (Bugge et al., 2016). Similarly to other members of this family, the action of PRL is initiated when it binds to two PRLR units, forming a biological activated heterotrimeric complex (Bridges and Grattan, 2019). It is important to notice that other hormones, like growth hormone and placental lactogen, also bind PRLR (Bernard et al., 2015), and that in pregnancy, part of PRL signaling may be induced by placental lactogen (Phillipps et al., 2020). Alternative splicing of the PRLR gene results in several PRLR isoforms with distinct intracellular domain lengths that vary between species. While rats have three distinct PRLR isoforms, long, short and intermediate (Bernard et al., 2019; Molina-Salinas et al., 2021), several other additional PRLR isoforms, including two PRLR-short forms and a secreted soluble form, are present in humans (Trott et al., 2003). However, the differences between the intracellular domain of the PRLR do not have an impact in the elementary functions of the PRL, as observed in 84 mammalian species (Paré et al., 2021).

PRL binding to the long form of PRLR can elicit distinct signaling cascades mediated by tyrosine kinase activation (**Figure 1**), including Janus protein kinase 2 (JAK2) autophosphorylation and JAK2-mediated tyrosine phosphorylation of the receptors (Bernard et al., 2015). The JAK2 activation is the best characterized and one of the fastest mechanisms of action mediated by the PRLR-long form (Campbell et al., 1994). Binding of PRL to the PRLR-long form induces JAK2-mediated phosphorylation of signal transducer and activator of transcription (STAT) proteins that translocate to the nucleus. In the nucleus, STAT proteins bind to gene promoters that contain the γ-interferon activated sequence DNA-binding motif and regulate the transcription of PRL target genes (Gilmour et al., 1995), constituting the canonical JAK/STAT pathway. In turn, JAK/STAT signaling is inhibited by proteins of the suppressor of cytokine signaling (SOCS) family (particularly SOCS1 and SOCS3), by a negative-feedback loop promoted by the activation of JAK/STAT pathway. SOCS proteins contain a Src homology 2 domain and a kinase-inhibitory region (KIR) that inhibits the kinase activity of JAKs interfering with JAK/STAT signaling (Durham et al., 2019).

The mitogen-activated protein kinase (MAPK)/ extracellular signal regulated kinase (ERK) is another signaling pathway activated by PRL (**Figure 1**). Evidences from studies in human and mouse mammary epithelial cell lines, revealed that the activation of this pathway involves upstream intermediaries like SHC, growth factor receptor-bound protein 2, SOS, Ras and Raf (Das and Vonderhaar, 1996), that lead to the phosphorylation of MAPK and ultimately regulate cell proliferation (Piccoletti et al., 1994; Acosta et al., 2003). Another signaling cascade associated with the PRLR-long form signaling described in human breast cancer cell lines, is phosphatidylinositol 3-kinase (PI3K)/ protein kinase B (AKT) pathway (**Figure 1**). Briefly, PRL binding to PRLR activates Src, mediating the activation of PI3K and AKT that in turn modulates cell proliferation (Acosta et al., 2003). The exact mechanism of action of PRLR-short forms is unknown since the activation of JAK/STAT pathway is prevented. However, the PRLR-short isoform is able to activate the MAPK and PI3K/AKT pathways (Bridges and Grattan, 2019; Wen et al., 2020; Molina-Salinas et al., 2021). PRLR-short signaling was also associated with the inhibition of FOXO3 expression in the mice ovary, resulting in ovarian developmental defects (Halperin et al., 2008). On the other hand, the blockage of PRLR-short form with an antagonist in human uterine cancer cells lines was associated with a reduction in cell proliferation, possibly mediated by the reduction of PI3K/AKT activity and augmented FOXO3a nuclear translocation (Wen et al., 2020). Conversely, the activation of the PRLR-short form in pancreatic ductal adenocarcinoma mice and in human models reduced cell proliferation. In this case the reduced cell proliferation was associated with decreased expression of genes involved in the pentose phosphatase pathway, through the activation of the Hippo signaling pathway (Nie et al., 2021). Even though the available information regarding PRLR-short form signaling is conflicting, and that PRLR-short, like the PRLR-long signaling cascade, may be tissue and physiological state dependent, all data seem to reinforce that the PRLR-short form is involved in the modulation of relevant biological actions.

Despite sharing the same signaling cascade, the target genes regulated by PRLR activation are cell type dependent. For instance, while in many cells PRL was associated with antiapoptotic and proliferative effects, in rat lactotrophs, PRL elicited proapoptotic and antiproliferative effects instead (de Dios et al., 2019). In this case, the PRL-induced apoptotic and antiproliferative effects were mediated by the JAK2/STAT5 pathway and inhibition of both ERK and Akt phosphorylation (de Dios et al., 2019). It is important to recognize that most studies dedicated to the investigation of type I cytokine receptor signaling cascades were performed in cancer cells, and that information regarding the biological actions mediated by this receptor family in healthy cells is still lacking.

Prolactin Signaling in the Brain

In mammals, PRLR are expressed in brain regions (**Figure 2**), like the amygdala, the preoptic area, the thalamus, the hypothalamus, the epithalamus and the brainstem (Kokay et al., 2018; Voigt and Bennett, 2018; Szczesna et al., 2020). It is important to notice that most studies that investigated the expression of PRLR in the brain used probes that do not discriminate between the short and the long forms of PRLR. PRLR are also present in the subventricular zone (SVZ) and in the hippocampus in mice (Mak and Weiss, 2010; Anagnostou and Morales, 2019), as well as in the circumventricular organs, like the median eminence, the subfornical organ and in the choroid plexus (CP) in rodents (Kamesh et al., 2018; Kokay et al., 2018; Voigt and Bennett, 2018). In fact, the CP is the region of the brain with the highest expression of PRLR-long form (Kokay et al., 2018). This PRLR form is located in the cytoplasm and both the apical and the basal membrane of rat CP epithelial cells (Costa-Brito et al., 2021).

Phosphorylation of STAT5, mediated by activated PRLR-long form, is considered to be the main PRLR signaling pathway in the mice brain (Yip et al., 2012), especially during pregnancy (Salais-López et al., 2017; Gustafson et al., 2020). However, neuronal ablation of STAT5 in mice revealed that STAT5 phosphorylation is not mandatory for the expression of nursing behaviors. In fact, the induction of faster responses observed in the medial preoptic area neurons, indicate that PRL-mediated effects in this brain region do not always involve the activation of transcription factors but rather other unidentified mechanisms of action (Buonfiglio et al., 2015).

Increasing evidences support the hypothesis that PRL elicits fast and transient calcium channels/transporters responses in specific subsets of hippocampal and hypothalamic neurons in mice (Leem et al., 2019; Georgescu et al., 2020). Recently, PRL-mediated action was associated with the modulation of membrane excitability in the neurons of the subfornical organ (Kamesh et al., 2018) and the arcuate nucleus (Blum et al., 2019) in rodents, with a particular relevant role of the transient receptor potential cation channel Trpc5 in the later. Other PRL-induced responses in the brain include the activation of ERK1/2 MAPK pathway in the hypothalamus and in hippocampal progenitor cells in rats (Blume et al., 2009; Wagner et al., 2009), or AKT phosphorylation in the hippocampus of male mice (Anagnostou et al., 2021).

The expression of the PRLR-short form has been reported in hypothalamic areas, the amygdala, the thalamus and the CP of female rats (Pi et al., 2002; Bakowska and Morrell, 2003), and in the SVZ and the hippocampus of male mice (Mak and Weiss, 2010). Overall, PRLR-long and PRLR-short expression seem to overlap in several brain regions, but the expression of the PRLR-long form is usually higher than that of the PRLR-short (Bakowska and Morrell, 2003). Furthermore, the increase of both PRLR forms in the rat brain during lactation (Pi and Grattan, 1999) and the observation of PRL actions mediated by PRLR-short form in the mice nervous system (Belugin et al., 2013) have also been reported. Together, these evidences suggest that this PRLR-short isoform may be involved in PRL-mediated brain functions in rodents. In addition, PRLR isoforms can interact and regulate the action of each other in both mice and humans (Belugin et al., 2013; Kang et al., 2014). For instance, in mice, the transient action of PRL mediated by the PRLR-short form in sensory neurons of the trigeminal ganglia is negatively regulated by the presence of PRLR-long form in these neurons (Belugin et al., 2013). However, most studies involving the investigation of PRLR activation in the brain do not discriminate between isoforms of these receptors, PRL signaling in the brain may not always be mediated by the activation of the canonical PRLR-long form pathways.

Sources of Prolactin in the Brain

Based on its size, PRL should not cross the blood-brain barrier (BBB). However, PRL is present in the cerebrospinal fluid (CSF), mimicking the fluctuations found in the peripheral circulation, reinforcing that PRL transport into the brain must occur (Login and MacLeod, 1977). For instance, in mice, transport of PRL into the brain is increased during lactation (Brown et al., 2016), a physiological state characterized by high levels of serum PRL. Based on the high expression of PRLR in the CP, and on the evidence depicted from studies in primates, a saturable receptor-mediated mechanism of transport of PRL across the CP has been considered the likely route of entrance of PRL into the CNS (Walsh et al., 1987). The CP, located within the brain ventricles, is responsible for the production of CSF and several peptides, as well as for controlling the passage of molecules into and out of the brain (Talhada et al., 2019). The CP is composed by a single layer of epithelial cells bound together by tight junctions, forming the blood-CSF barrier (Marques et al., 2017; Talhada et al., 2019).

Recently, the observation that PRL transport into the CSF occurred at normal rates in CP PRLR knockout mice, contradicted the theory that PRL transport into the brain was mediated by PRLR. Intriguingly, the rapid activation of PRL signaling in the brain observed after the peripheral administration of exogenous PRL was not accompanied by a rapid increase in the levels of this hormone in the CSF, leading the authors to propose that PRL transport into the brain could be mediated at the cerebral vasculature level (Brown et al., 2016). The expression of PRLR and the observation of PRL-mediated action in circumventricular regions in mice, like the median eminence and subfornical organ (Kamesh et al., 2018; Kirk et al., 2019), that lack the conventional BBB (Ben-Zvi and Liebner, 2021), seem to strengthen the hypothesis that PRL transport into the brain may also occur at brain regions were the BBB is more leaky (Login and MacLeod, 1977). However, although the BBB becomes more permeable with age (Segarra et al., 2021), transport of PRL to the brain seems to be reduced by age in male mice (Barad et al., 2020). This suggests that PRL transport into the brain is not entirely explained by leakier BBB regions. Overall, the lack of evidence to support that PRL is transported via PRLR in the CP, does not exclude the CP as a relevant gateway for the entrance of this hormone into the brain. In fact, increased PRL transport, mediated by CP epithelial cells, was recently reported under specific physiological conditions (Tani et al., 2018). Notwithstanding, the saturable mechanism that is responsible for PRL entrance into the brain remains unidentified.

The first evidences of non-pituitary sources of PRL in the brain derived from studies demonstrating that PRL was still detectable in the CSF of rats that were subjected to hypophysectomy, contrasting with the observed reduction of plasma PRL levels (Barbanel et al., 1986). Since then, immunoreactive-PRL were reported in distinct rodent brain regions, like several hypothalamic regions, the amygdala, brainstem, hippocampus, cerebellum and cerebral cortex (Emanuele et al., 1987, 1992; Torner et al., 2004; Roselli et al., 2008) (**Figure 2**). Recently, our research group presented evidence that rather than being just a gateway for PRL, the CP itself constitutes an alternative source of PRL to the rat brain. Despite the expression of full-length PRL transcripts in the CP, identical to those expressed in the pituitary, the PRL detected in the CPs has a much higher molecular weight than pituitary PRL or PRL secreted to the conditioned media of pituitary cultures. This high molecular weight immunoreactive-PRL was observed in CP from pregnant female rats, in primary cultures of rat CP epithelial cells, and in CP-conditioned media and in the rat CSF (Costa-Brito et al., 2021). The existence of high molecular weight PRL isoforms similar to those found in the CP was previously described in other cells like human monocytes (López-Rincón et al., 2013). However, the biological activity and relevance of this higher molecular weight PRL remains controversial. The recognition of non-pituitary sources of PRL in the brain is not consensual either, with some investigators defending that the reported expression of PRL mRNA based in high sensitivity techniques like RT-PCR may be the result of sample contamination with other cells that express PRL (Bridges and Grattan, 2019). Overall, considering that the expression of PRL in brain regions other than the pituitary is very low, it is unlikely that PRL produced in extrapituitary tissues may have some impact in the circulating levels of PRL. Nevertheless, it is possible that brain PRL from extrapituitary sources could be produced in specific physiological circumstances, which require PRL autocrine and paracrine modulation of brain functions and account for the levels of PRL in the CSF.

Prolactin Actions in the Brain

The best documented actions of PRL in the mammalian brain, are associated with mechanisms that prepare the brain to motherhood/fatherhood (Larsen and Grattan, 2010; Mak and Weiss, 2010; Brown et al., 2017; Stagkourakis et al., 2020). In female mice, the action of PRL in specific areas of the brain, including the medial preoptic area region of the hypothalamus and SVZ, is essential for the development of maternal behaviors like retrieving and crouching over the pups (Larsen and Grattan, 2010; Brown et al., 2017). In turn, male mice, a biparental species, and rat males, a uniparental species, display different paternal behaviors, which may be explained by distinct release profiles of PRL between species (Stagkourakis et al., 2020). Yet, PRL administration is able to elicit fatherhood behaviors even in males of non-paternal species like rats (Stagkourakis et al., 2020). Furthermore, PRLR expression is relatively similar in the brain of male and female mice (Kokay et al., 2018), reinforcing the theory that PRL may also play a relevant role in male brain functions of biparental species. On the other hand, in sheep, the increased expression of PRLR in the arcuate nucleus, contrast with the reduction of PRLR levels in the median eminence and in the adenohypophysis, during gestation. This may suggest that, during sheep gestation, PRL may have a predominant role in the regulation of appetite (Szczesna et al., 2020).

In rodents, the fine-tuned balance of PRL levels during pregnancy and postpartum is fundamental to the development of healthy maternal behavior of the progenitors and of the offspring in their adult life. Abnormal levels of PRL during prenatal and early-life in mice, have a negative impact in the nursing behaviors of the pups (Sairenji et al., 2017; Mitani et al., 2018). More recently, the genes modulated by PRL in the hippocampus of female rats were examined by RNAseq. A

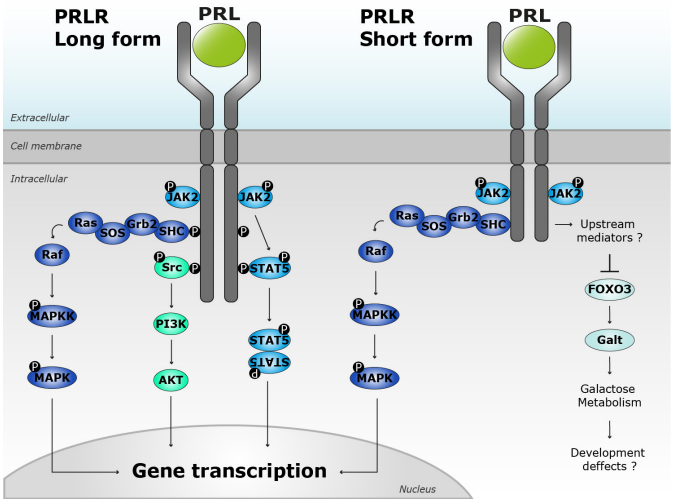

Figure 1 | **Main signaling pathways elicited by the activation of the long and short prolactin receptor isoforms.**
The action of prolactin (PRL) is initiated by its binding to a prolactin receptor (PRLR) homodimer, forming a heterotrimeric complex. PRL binding to the long form of PRLR triggers distinct signaling cascades that ultimately culminate in the regulation of gene transcription. The main activation pathway of PRL is the JAK/STAT pathway, but prolactin is also able to induce the PI3K/AKT pathway. The MAPK/ERK is another signaling pathway triggered by prolactin binding to both PRLR isoforms. The activation of this pathway involves the SHC, Grb2, SOS, Ras and Raf cascades. It is also believed that the activation of the PRLR-short form inhibits the expression of FOXO3 and Galt, involved in the metabolism of galactose to glucose, possibly leading to developmental defects. However, the exact mechanism of action of this pathway remains unidentified. AKT: Protein kinase B; ERK: extracellular signal regulated kinase; FOXO3: forkhead transcription factor 3; Galt: galactose-1-phosphate uridyltransferase; Grb2: growth factor receptor-bound protein 2; JAK2: Janus kinase 2; MAPK: mitogen-activated protein kinase; MAPKK: MAPK kinase; PI3K: phosphatidylinositol 3-kinase; Raf: rapidly accelerated fibrosarcoma; Ras: Rat sarcoma; SHC: SHC-transforming protein 1; SOS: son of sevenless; Src: proto-oncogene tyrosine-protein kinase Src; STAT5: signal transducer and activator of transcription 5.

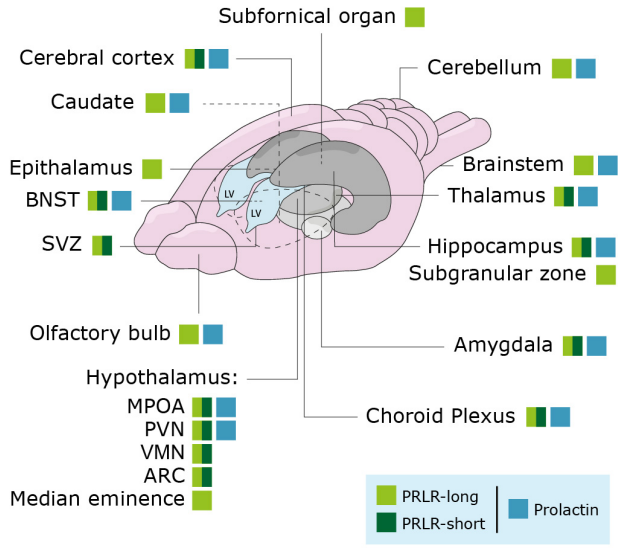

Figure 2 | **Schematic representation of the expression of prolactin and prolactin receptors in the brain.**
Reported sites of prolactin expression are signed by a blue square, while the expression of the long and the short form of the prolactin receptor (PRLR) are indicated in light green and dark green, respectively. Prolactin and PRLR expression are represented throughout several regions of the rat brain, since most of the available information is retrieved from studies conducted in rodents. ARC: Arcuate nucleus; BNST: bed nucleus of the stria terminalis; LV: lateral ventricle; MPOA: medial preoptic area; PVN: paraventricular nuclei; SVZ: subventricular zone; VMN: ventromedial hypothalamic nucleus.

total of 162 genes were differently expressed in the hippocampus of ovariectomized females exposed to PRL throughout 24 hours. Most differentially expressed genes were upregulated by PRL exposure and were correlated with cell cycle regulation and biological processes, like response to hypoxia, estradiol, or nutrients. Notably, PRL-modulated genes were associated with brain functions like learning, memory, plasticity, neuroprotection, neurogenesis and remodeling (Cabrera-Reyes et al., 2019).

Interestingly, in mice, the development of mild maternal behavior is not always associated with higher peripheral PRL levels. Nulliparous female mice presented enhanced PRL responsiveness in the medial preoptic nucleus after prolonged exposure to pups, despite the lack of serum PRL rise as observed in puerperia's. This observation may be a consequence of alterations in PRL signaling or could be explained by the existence of brain extrapituitary PRL, as suggested by the authors (Salais-López et al., 2020). However, further studies are necessary to understand if extrapituitary sources of PRL in the brain, play a relevant and direct role in the development of maternal behaviors.

Effects of prolactin in neurogenesis

In mice, PRL modulates neurogenesis in the SVZ, where progenitor cells differentiate into interneurons that migrate to the olfactory bulb (Shingo et al., 2003), and in the subgranular zone of the hippocampal dentate gyrus (**Figure 3**). These are the two brain neurogenic niches where progenitor cells continue to differentiate in new neurons and glial cells in adult life (Mak and Weiss, 2010; Walker et al., 2012).

In the SVZ, PRL plays an important role in the formation of the olfactory bulb by increasing the proliferation of neuronal stem cells during the early stage of pregnancy and early postpartum period in mice that further differentiate in olfactory neurons (Shingo et al., 2003). PRL also raises SVZ neurogenesis in male mice that interact with their pups (Mak and Weiss, 2010). In mice, PRL-induced adult SVZ neurogenesis is mediated by the ERK5 signaling pathway (Wang et al., 2013) and is fundamental for the development of healthy maternal behaviors (Larsen and Grattan, 2010), offspring recognition (Shingo et al., 2003; Mak and Weiss, 2010) and choice of mates by females (Mak et al., 2007). Additionally, exposure of females to male pheromones seems to be one of the factors that regulate PRL-induced neurogenesis. Besides increasing PRL levels in female mice, pheromone exposure mediates the generation of new neurons in the SVZ in a PRLR dependent manner (Mak et al., 2007; Larsen et al., 2008). Although olfactory bulb neurogenesis seemed to be fundamental for the development of maternal behavior and lamb recognition in sheep (Corona et al., 2018), no direct observation between PRL and olfactory bulb neurogenesis has been reported in this specie so far.

Considering that the CP is near the SVZ, the possible contribution of the CP PRL induction of neurogenesis should also be considered. The CP epithelia is a remarkable source of peptides that modulate brain function and neurogenesis (Marques et al., 2017). In that regard, in a recent RNAseq study conducted to examine transcriptomic differences induced by PRL, insulin-like growth factor 2 (Igf2) has emerged as a gene highly responsive to PRL in the adult mice CP. The upregulation of the Igf2 gene during lactation, suggest that this particular growth factor is possibly involved in the postpartum increase of SVZ neurogenesis (Phillipps et al., 2019). In fact, in the pioneer work of Shingo and colleagues, the authors suggested that due to the high expression of PRLR in the CP, that this structure could indirectly mediate PRL action in SVZ neurogenesis (Shingo et al., 2003). Local production of PRL in the rat CP (Costa-Brito et al., 2021), may explain the relevant role of the CP in PRL-induced neurogenesis in the SVZ.

The role of PRL in the neurogenesis in the dentate gyrus hippocampal progenitor cells is not consensual. Some studies describe that PRL enhances the proliferation of the dentate gyrus cells in both female and male mice (Mak and Weiss, 2010; Walker et al., 2012) and prevents the reduction of adult hippocampal neurogenesis promoted by chronic stress in male mice (Torner et al., 2009). Furthermore, PRL null mice display impaired learning and memory processes, and PRL infusion was able to restore the learning deficits observed in these mice. However, despite PRL-deficient mice presented lower generation of hippocampal precursor cells *in vitro*, the same was not observed *in vivo*, suggesting that in the absence of PRL, hippocampal neurogenesis may be sustained by other factors (Walker et al., 2012). Other studies report that PRL has either no effect on rodents and human neurogenesis (Shingo et al., 2003; Wagner et al., 2009;

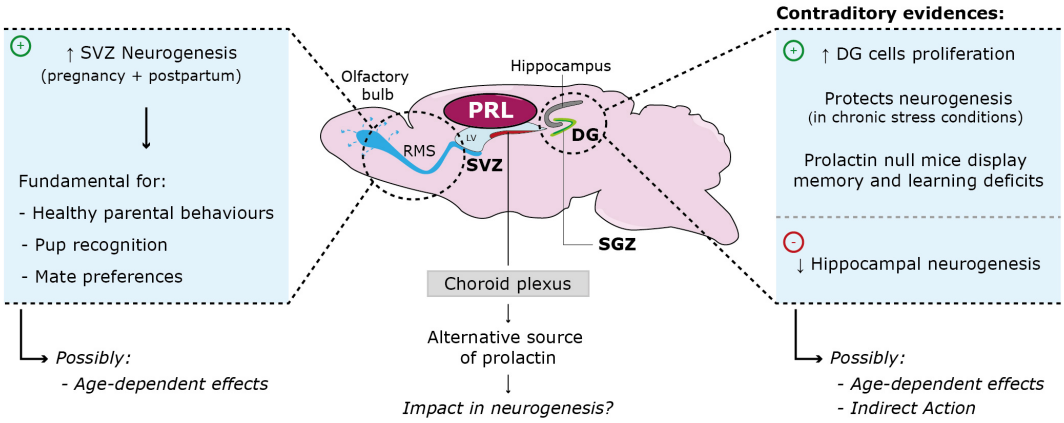

Figure 3 | **Summary of the main prolactin-mediated effects in the modulation of adult neurogenesis.**
Prolactin (PRL) has been associated with modulatory actions in the two major neurogenic niches in adult brain, the subventricular zone (SVZ) and subgranular zone (SGZ) of the dentate gyrus (DG). In the SVZ, progenitor cells differentiate into interneurons that migrate throughout the rostral migratory stream (RMS) to the olfactory bulb. During the early stage of gestation and lactation, PRL increases the number of proliferating cells in the SVZ and raises the amount of new interneurons in the olfactory bulb. The role of PRL in hippocampal progenitor cells neurogenesis is not as consensual. Some evidences support that PRL favors DG cells proliferation, while others suggest that PRL has either no effect or that negatively impacts hippocampal neurogenesis. In addition, recent evidence suggested that the choroid plexus may also be an alternative source of PRL in the rat brain. However, further studies are necessary to understand if choroid plexus derived PRL has any effects in SVZ neurogenesis. LV: Lateral ventricle.

Smeeth et al., 2021), or that in fact it has a negative impact on hippocampal neurogenesis in rats (Lajud et al., 2013).

More recently, a study conducted in an *in vitro* model of human hippocampal stem cells revealed that PRL treatment was able to increase neuronal differentiation. However, this increase was only transient and continued PRL exposure actually decreased the number of proliferating cells, suggesting that fluctuations of PRL levels in different reproductive stages may induce short-term alterations or not contribute at all to adult hippocampus neurogenesis (Smeeth et al., 2021). Interestingly, in an earlier study conducted with a distinct human neural stem cell *in vitro* model, distinct PRL levels were associated with different neuroblast and glial progenitor proliferation and migration rates (Pathipati et al., 2011), suggesting that the effects of PRL in neurogenesis may be dose dependent.

The lack of *in vitro* evidences that support the *in vivo* observations describing the valuable influence of PRL in hippocampal neurogenesis, may indicate that *in vivo*, the impact of PRL in this particular neurogenic niche may be mediated by indirect mechanisms (Wagner et al., 2009). As a matter of fact, the promoting of neurogenesis by PRL may be age- and species-dependent, since high levels of PRL during early-age were associated with a reduction in hippocampal neurogenesis in postnatal rats (Lajud et al., 2013), differing from the proliferative effect observed in adulthood mouse (Walker et al., 2012). Furthermore, the negative impact of PRL administration in both hippocampal and olfactory bulb neurogenesis in early life possibly contributes to the development of anxiety behaviors in males, but not females, during adulthood. However, the exact mechanism involved in such observations remained unknown (Lajud et al., 2013). A detailed review regarding the relation between motherhood, maternal experience and neurogenesis, not restricted to PRL-mediated effects, can be consulted elsewhere (Medina and Workman, 2020).

Effects of prolactin in neuroplasticity

Recently, analysis of the proteome of postpartum maternal preoptic area revealed that the common regulators and targets of the significantly altered proteins found between mother and pup-deprived female rats, like AKT, MAPK1 and STAT3, could be traced to PRL. Thus, PRL may be in part responsible for motherhood-associated neuroplasticity in this region (Udvari et al., 2019). In fact, PRL, together with estrogen and progesterone, were considered to be possible intermediaries of the medial preoptic area plasticity observed during pregnancy in rats (Uriarte et al., 2020). In addition, PRL may also be associated with brain plasticity modulation during fatherhood. In mandarin voles, experienced and first-time fathers showed increased spine density and greater dendrite length in the medial prefrontal cortex than non-fathers. This alteration may be explained by the observed increase in PRL levels in both experienced and new fathers (Wang et al., 2018).

Exercise ameliorates the PRL response in chronically stressed mice, improving memory consolidation, through stimulation of the hippocampus (Leem et al., 2019). At the synaptic level, it is believed that PRL modulates long- and short-term synaptic plasticity in the hippocampus of female mice at reproductive age. The enhancement of synaptic strength was not observed in immature females or male mice, suggesting that sex hormones are necessary to preserve long- and short-term plasticity response to PRL (Zamora-Moratalla and Martín, 2021). High serum PRL levels correlate with enhanced performance in learning and memory tasks possibly associated with the plasticity of the hippocampus in female mice (Moreno-Ruiz et al., 2021).

Prolactin in Neuroprotection and Neurological and Psychiatric Disorders

Neuroprotection

In rodents, PRL protects the hippocampus against glutamate excitotoxicity in the kainic acid model *in vivo* and *in vitro* (Rivero-Segura et al., 2017; Anagnostou and Morales, 2019; Ortiz-Pérez et al., 2019). Recently, the detailed mechanisms of PRL-mediated neuroprotection have been extensively reviewed (Molina-Salinas et al., 2021). Briefly, in rodents, parenthood seems to protect the hippocampus against neurodegenerative insults (Anagnostou and Morales, 2019; Cabrera et al., 2009). However, PRL neuroprotective properties were also observed in mice treated with PRL prior to kainic acid insult (Anagnostou et al., 2021). These protective effects were associated with increased AKT phosphorylation (Anagnostou et al., 2021) and ERK1/2 activation (Morales et al., 2014).

The pretreatment of rat hippocampal neuronal cultures with PRL prior to glutamate incubation prevented cell death and mitochondrial disfunction, and inhibited the increase of intracellular calcium levels, triggered by the excitotoxic insult (Rivero-Segura et al., 2017). On the other hand, vasohinibin, generated by enzymatic cleavage of PRL in the hippocampus, prompts neuronal cell dead in cultures of mouse hippocampal primary cells (Aroña et al., 2020). These effects were reversed by the addition of PRL which was able to block the negative effects of vasohinibin, suggesting that PRL action in the hippocampus is complex (Aroña et al., 2020).

Neuroprotective effects of PRL have also been reported in retinal cells. PRL reduces gliosis and favors the expression of survival factors in a rat model of light-induced retinal degeneration (Arnold et al., 2014). Furthermore, increased retinal dysfunction observed with age was correlated with the age-associated decrease of PRLR, and reduction of PRL signaling in the retina (Arnold et al., 2020).

Based on its functions as a neuropeptide, PRL may have an important impact in other neurological disorders. Aside from the potential role as a therapeutic agent in excitotoxicity-mediated diseases like neurodegenerative disorders, as discussed above, the number of studies on the relevance of PRL in neurological disorders is still rather limited. Anew, the relevance of PRL in brain health and disease seems

to be complex, possibly tissue-specific, and once more, apparently dependent on a combination of factors such as age or reproductive stage.

Brain injury

PRL treatment reduced the cerebral infarct area and edema in a rat cerebral ischemia model, and restored brain glutamate levels and intracellular calcium homeostasis (Vermani et al., 2020). Although PRL administration failed to rescue loss of cortical neurons in hypoxic ischemic juvenile rats, increased PRL immunoreactivity was observed in the injured parietal cortex, and this hormone was associated with glial responses that led to the formation of the glial scare (Möderscheim et al., 2007). In the human brain, hypoxia- and ischemia-related death cases were associated with increased levels of PRL in the CSF, suggesting that the selective passage of PRL to CSF, possibly mediated by the CP, is augmented in these conditions, possibly to adjust osmotic pressure in the brain (Tani et al., 2018) or by disruption of the BBB associated with stroke (Jiang et al., 2018). Notwithstanding, as there is production of PRL in the CP that can be released to the CSF, the increase of this hormone levels in the CSF found in hypoxia can not be ruled out. On the other hand, PRL was also associated with decrease of the BBB permeability by inducing the expression of tight junction proteins claudin-5 and occludin in primary cultures of bovine brain microvessel endothelial cells (Rosas-Hernandez et al., 2013).

In a prospective cohort study, patients with higher PRL levels during the first year after traumatic brain injury and aneurysmal subarachnoid hemorrhage, tended to perform worst in follow-up cognitive and behavioral tests. In this particular case, the higher PRL levels were considered a consequence of a major pituitary disfunction reported in these patients (Tölli et al., 2019) rather than a neuroprotective response mechanism. Many of the putative mechanisms involved in the beneficial actions of PRL in brain injury are linked with neuroprotective properties of PRL, as described above. Although some studies report an increase of PRL levels after brain injury, further research is necessary to investigate if this increase has any protective role, especially in humans.

Multiple sclerosis

Multiple sclerosis is a CNS autoimmune disorder associated with axonal degeneration and demyelination processes (Zhornitsky et al., 2013). In this regard, PRL is able to induce oligodendrocyte precursor cell proliferation and remyelination of in adult female mice brain during pregnancy (Gregg et al., 2007). Although PRL can be associated with modulation and proinflammatory profiles in immune cells, the combined administration of PRL and interferon-ß seems to be beneficial as a short-term treatment of experimental autoimmune encephalomyelitis, an animal model of multiple sclerosis (Zhornitsky et al., 2015). On the other hand, PRL produced by immune cells in the late phases of experimental autoimmune encephalomyelitis mice model is responsible for the development of persistent neuroinflammation in this pathology (Zhang et al., 2019), which may indicate that despite not being responsible for the onset of multiple sclerosis (Costanza et al., 2013), PRL contributes to the pathological outcome of this disease, as previously reviewed (Zhornitsky et al., 2013).

Psychiatric disorders and stress

Higher levels of PRL are often observed in patients with depression, stress, and psychopathologies. However, this so-called hyperprolactinemia state is probably a consequence of pharmacological treatment with D2 dopamine receptors blockers and not a cause of psychiatric disorders (Alosaimi et al., 2018). Nonetheless, it has been suggested that higher PRL levels, associated with higher stress rates, in some risk-prone and first-episode psychosis groups, that had never been treated with antipsychotics, could be associated with emerging psychosis (Ittig et al., 2017; Studerus et al., 2021). This hypothesis lacks the support of robust evidence that directly link higher circulating PRL levels with the onset of mental disorders.

It has been considered that PRL has anxiolytic properties, particularly relevant in the perinatal and postpartum periods (Torner et al., 2002; Larsen and Grattan, 2010; Golub et al., 2016). For instance, female mice exposed to trauma during gestation (exposure to contextual fear conditioning), presented lower levels of PRL during postpartum and decreased maternal behavior. Furthermore, pups of traumatized mothers, were born smaller and remained smaller throughout life when compared to non-traumatized animals and present anxiety-like behaviors (Golub et al., 2016). Similar results have been reported in humans, where the reduction of PRL levels was also observed in the plasma of mothers with depressive and anxiety symptoms during the perinatal phase, which was associated with reduced social interactive behavior and irritability in newborns (Zhang et al., 2018, 2017). Additionally, augmented brain PRL levels in rats exposed to stress and sleep-deprivation suggest that PRL is also associated with extended REM sleep periods and thus with stress coping mechanisms (Machado et al., 2017).

On the contrary, evidences that PRLR knockdown could be beneficial to the treatment of depression in mice subjected to chronic mild stress have also been reported (Tian et al., 2019). In addition, the administration of exogenous PRL to rats during early-life stages, elicits depressive-like behaviors (Lajud et al., 2013). Also in rats, the administration of bromocriptine, a dopamine receptor agonist widely used to inhibit PRL secretion, during early lactation, was correlated with lower peripheral PRL levels and the development of increased anxiety-like behaviors in adult life, whereas the administration of the same compound in the late phase of lactation had the opposite effect (Carvalho et al., 2016). Based on these findings, the effect of PRL in anxiety-like behaviors seems to be age and species dependent.

Glioblastoma

Some evidence suggest that PRL may also play an important role in the pathology of primary brain tumors. In the past, many studies focused on the pro-tumorigenic role of PRL signaling in breast and pancreatic cancers (Goffin, 2017). Recently, PRL and PRLR expression have been reported in human and rodents glioblastoma cells and induced PRLR overexpression was associated with increased proliferation, migration and chemoresistance of the tumors (Asad et al., 2019). PRLR was found in all samples of human grade II and III gliomas and in glioblastoma, while only 12% of grade II and III gliomas and approximately 30% and glioblastoma samples, expressed PRL mRNA. Furthermore, the long-term patient survival rate was reduced in grade II and III and glioblastoma that expressed PRL (Asad et al., 2019). In addition, within the group of glioblastoma patients with tumors that express PRL, higher levels of PRLR expression were correlated with decreased survival in men. Interestingly, the opposite was observed in women, in which higher levels of PRLR correlate with extended survival. Remarkably, low levels of PRLR expression were associated with reduced long-term survival rates in grade II and III glioma in men, while no differences were observed in women, which may suggest that PRL and PRLR roles may be dependent of gender and tumor grade (Asad et al., 2019). PRL treatment and PRLR expression in distinct glioblastoma cell lines, were positively correlated with increased invasion capacity of the cells (Alkharusi et al., 2016). Overall, the potential of PRL and PRLR as therapeutic targets and/or biomarkers is of particular interest in glioblastoma.

Concluding Remarks

PRL is a pleiotropic hormone that mediates a considerable diversity of endocrine, autocrine and paracrine actions. Apart from its fundamental in reproduction and lactation, PRL has remarkable actions in the brain, particularly in the development of parental behavior, adapting brain circuits to the necessities imposed by normal parental care that ultimately promote survival of the offspring. PRL brain actions are wide and mediated through the activation of its receptors. Considering the role of PRL in the regulation of neurogenesis, neuroplasticity as well as in neuroprotection, it is tempting to suggest that its potential as a therapeutic agent should be investigated for the treatment of neurological disorders, including neurodegenerative diseases. Nonetheless, this approach should be carefully studied since PRL functions are wide and not restricted to a single target.

Despite some developments over the past decade, there is still a considerable lack of scientific evidence regarding the functions of PRL in the brain, especially in humans. It is important to notice that most studies devoted to the investigation of PRL-mediated action in the brain were performed in rodents and may not be alike in humans. Although *in vivo* studies using rodents have boosted our knowledge about brain function, the organization and complexity of the brain itself is very distinct between human and rodents. In fact, PRL systems are distinct in rodents and humans. These differences include distinct PRLR isoforms, which could be associated with the activation of different signaling cascades, and the existence of an additional superdistal promoter that modulates the expression of extrapituitary PRL in humans. An effort in the development

of increasingly complex *in vitro* models that mimic human brain structures, like organoids, could be a practical tool for the study of PRL brain functions. Overall, several controversial issues like the biological relevance of brain extrapituitary PRL remain unanswered. Further studies are necessary to fully understand the dimension of PRL actions in the brain.

Author contributions: *Manuscript design and writing: ARCB. Manuscript revision: IG and CRAS. All authors approved the final version of the manuscript.*
Conflicts of interest: *The authors declare no conflicts of interest.*

References

Acosta JJ, Muñoz RM, González L, Subtil-Rodríguez A, Dominguez-Caceres MA, García-Martínez JM, Calcabrini A, Lazaro-Trueba I, Martín-Pérez J (2003) Src mediates prolactin-dependent proliferation of T47D and MCF7 cells via the activation of focal adhesion kinase/Erk1/2 and phosphatidylinositol 3-kinase pathways. Mol Endocrinol 17:2268-2282.
Alkharusi A, Yu S, Landázuri N, Zadjali F, Davodi B, Nyström T, Gräslund T, Rahbar A, Norstedt G (2016) Stimulation of prolactin receptor induces STAT-5 phosphorylation and cellular invasion in glioblastoma multiforme. Oncotarget 7:79572-79583.
Alosaimi FD, Fallata EO, Abalhassan M, Alhabbad A, Alzain N, Alhaddad B, Alassiry MZ (2018) Prevalence and risk factors of hyperprolactinemia among patients with various psychiatric diagnoses and medications. Int J Psychiatry Clin Pract 22:274-281.
Anagnostou I, Morales T (2019) Fatherhood diminishes the hippocampal damaging action of excitotoxic lesioning in mice. J Neuroendocrinol 31:e12783.
Anagnostou I, Muñoz-Mayorga D, Morales T (2021) Prolactin neuroprotective action against excitotoxic insult in the hippocampus of male mice. Peptides 135:170425.
Arnold E, Thébault S, Aroña RM, Martínez de la Escalera G, Clapp C (2020) Prolactin mitigates deficiencies of retinal function associated with aging. Neurobiol Aging 85:38-48.
Arnold E, Thebault S, Baeza-Cruz G, Arredondo Zamarripa D, Adán N, Quintanar-Stéphano A, Condés-Lara M, Rojas-Piloni G, Binart N, Martínez de la Escalera G, Clapp C (2014) The hormone prolactin is a novel, endogenous trophic factor able to regulate reactive glia and to limit retinal degeneration. J Neurosci 34:1868-1878.
Aroña RM, Arnold E, Macías F, López-Casillas F, Clapp C, Martínez de la Escalera G (2020) Vasoinhibin generation and effect on neuronal apoptosis in the hippocampus of late mouse embryos. Am J Physiol Regul Integr Comp Physiol 318:R760-771.
Asad AS, Nicola Candia AJ, Gonzalez N, Zuccato CF, Abt A, Orrillo SJ, Lastra Y, De Simone E, Boutillon F, Goffin V, Seilicovich A, Pisera DA, Ferraris MJ, Candolfi M (2019) Prolactin and its receptor as therapeutic targets in glioblastoma multiforme. Sci Rep 9:19578.
Bakowska JC, Morrell JI (2003) The distribution of mRNA for the short form of the prolactin receptor in the forebrain of the female rat. Brain Res Mol Brain Res 116:50-58.
Barad Z, Khant Aung Z, Grattan DR, Ladyman SR, Brown RSE (2020) Impaired prolactin transport into the brain and functional responses to prolactin in aged male mice. J Neuroendocrinol 32:e12889.
Barbanel G, Ixart G, Arancibia S, Assenmacher I (1986) Probable extrapituitary source of the immunoreactive prolactin measured in the cerebrospinal fluid of unanesthetized rats by push-pull cannulation of the 3rd ventricle. Neuroendocrinology 43:476-482.
Belugin S, Diogenes AR, Patil MJ, Ginsburg E, Henry MA, Akopian AN (2013) Mechanisms of transient signaling via short and long prolactin receptor isoforms in female and male sensory neurons. J Biol Chem 288:34943-34955.
Ben-Zvi A, Liebner S (2021) Developmental regulation of barrier- and non-barrier blood vessels in the CNS. J Intern Med doi: 10.1111/joim.13263.
Bernard V, Young J, Binart N (2019) Prolactin- a pleiotropic factor in health and disease. Nat Rev Endocrinol 15:356-365.
Bernard V, Young J, Chanson P, Binart N (2015) New insights in prolactin: pathological implications. Nat Rev Endocrinol 11:265-275.
Blum T, Moreno-Pérez A, Pyrski M, Bufe B, Arifovic A, Weissgerber P, Freichel M, Zufall F, Leinders-Zufall T (2019) Trpc5 deficiency causes hypoprolactinemia and altered function of oscillatory dopamine neurons in the arcuate nucleus. Proc Natl Acad Sci U S A 116:15236-15243.
Blume A, Torner L, Liu Y, Subburaju S, Aguilera G, Neumann ID (2009) Prolactin activates mitogen-activated protein kinase signaling and corticotropin releasing hormone transcription in rat hypothalamic neurons. Endocrinology 150:1841-1849.
Bridges RS, Grattan DR (2019) 30 years after: CNS actions of prolactin: Sources, mechanisms and physiological significance. J Neuroendocrinol 31:e12669.
Brown RS, Wyatt AK, Herbison RE, Knowles PJ, Ladyman SR, Binart N, Banks WA, Grattan DR (2016) Prolactin transport into mouse brain is independent of prolactin receptor. FASEB J 30:1002-1010.
Brown RSE, Aoki M, Ladyman SR, Phillipps HR, Wyatt A, Boehm U, Grattan DR (2017) Prolactin action in the medial preoptic area is necessary for postpartum maternal nursing behavior. Proc Natl Acad Sci U S A 114:10779-10784.
Bugge K, Papaleo E, Haxholm GW, Hopper JT, Robinson CV, Olsen JG, Lindorff-Larsen K, Kragelund BB (2016) A combined computational and structural model of the full-length human prolactin receptor. Nat Commun 7:11578.
Buonfiglio DC, Ramos-Lobo AM, Silveira MA, Furigo IC, Hennighausen L, Frazão R, Donato J Jr (2015) Neuronal STAT5 signaling is required for maintaining lactation but not for postpartum maternal behaviors in mice. Horm Behav 71:60-68.
Cabrera V, Cantú D, Ramos E, Vanoye-Carlo A, Cerbón M, Morales T (2009) Lactation is a natural model of hippocampus neuroprotection against excitotoxicity. Neurosci Lett 461:136-139.
Cabrera-Reyes EA, Vanoye-Carlo A, Rodríguez-Dorantes M, Vázquez-Martínez ER, Rivero-Segura NA, Collazo-Navarrete O, Cerbón M (2019) Transcriptomic analysis reveals new hippocampal gene networks induced by prolactin. Sci Rep 9:13765.
Campbell GS, Argetsinger LS, Ihle JN, Kelly PA, Rillema JA, Carter-Su C (1994) Activation of JAK2 tyrosine kinase by prolactin receptors in Nb2 cells and mouse mammary gland explants. Proc Natl Acad Sci U S A 91:5232-5236.
Carvalho JC, Lisboa PC, de Oliveira E, Peixoto-Silva N, Pinheiro CR, Fraga MC, Claudio-Neto S, Franci CR, Manhães AC, Moura EG (2016) Effects of early and late neonatal bromocriptine treatment on hypothalamic neuropeptides, dopaminergic reward system and behavior of adult rats. Neuroscience 325:175-187.
Corona R, Meurisse M, Cornilleau F, Moussu C, Keller M, Lévy F (2018) Disruption of adult olfactory neurogenesis induces deficits in maternal behavior in sheep. Behav Brain Res 347:124-131.
Costa-Brito AR, Quintela T, Gonçalves I, Duarte AC, Costa AR, Arosa FA, Cavaco JE, Lemos MC, Santos CRA (2021) The choroid plexus is an alternative source of prolactin to the rat brain. Mol Neurobiol 58:1846-1858.
Costanza M, Musio S, Abou-Hamdan M, Binart N, Pedotti R (2013) Prolactin is not required for the development of severe chronic experimental autoimmune encephalomyelitis. J Immunol 191:2082-2088.
Das R, Vonderhaar BK (1996) Involvement of SHC, GRB2, SOS and RAS in prolactin signal transduction in mammary epithelial cells. Oncogene 13:1139-1145.
de Dios N, Orrillo S, Irizarri M, Theas MS, Boutillon F, Candolfi M, Seilicovich A, Goffin V, Pisera D, Ferraris J (2019) JAK2/STAT5 pathway mediates prolactin-induced apoptosis of lactotropes. Neuroendocrinology 108:84-97.
Dobolyi A, Oláh S, Keller D, Kumari R, Fazekas EA, Csikós V, Renner É, Cservenák M (2020) Secretion and function of pituitary prolactin in evolutionary perspective. Front Neurosci 14:621.
Durham GA, Williams JJL, Nasim MT, Palmer TM (2019) Targeting SOCS proteins to control JAK-STAT signalling in disease. Trends Pharmacol Sci 40:298-308.
Emanuele NV, Jurgens JK, Halloran MM, Tentler JJ, Lawrence AM, Kelley MR (1992) The rat prolactin gene is expressed in brain tissue: detection of normal and alternatively spliced prolactin messenger RNA. Mol Endocrinol 6:35-42.
Emanuele NV, Metcalfe L, Wallock L, Tentler J, Hagen TC, Beer CT, Martinson D, Gout PW, Kirsteins L, Lawrence AM (1987) Extrahypothalamic brain prolactin: characterization and evidence for independence from pituitary prolactin. Brain Res 421:255-262.
Fahie-Wilson M, Smith TP (2013) Determination of prolactin: the macroprolactin problem. Best Pract Res Clin Endocrinol Metab 27:725-742.
Georgescu T, Ladyman SR, Brown RSE, Grattan DR (2020) Acute effects of prolactin on hypothalamic prolactin receptor expressing neurones in the mouse. J Neuroendocrinol 32:e12908.
Gilmour KC, Pine R, Reich NC (1995) Interleukin 2 activates STAT5 transcription factor (mammary gland factor) and specific gene expression in T lymphocytes. Proc Natl Acad Sci U S A 92:10772-10776.
Goffin V (2017) Prolactin receptor targeting in breast and prostate cancers: new insights into an old challenge. Pharmacol Ther 179:111-126.
Golub Y, Canneva F, Funke R, Frey S, Distler J, von Hörsten S, Freitag CM, Kratz O, Moll GH, Solati J (2016) Effects of In utero environment and maternal behavior on neuroendocrine and behavioral alterations in a mouse model of prenatal trauma. Dev Neurobiol 76:1254-1265.
Grattan DR (2015) 60 YEARS OF NEUROENDOCRINOLOGY: The hypothalamo-prolactin axis. J Endocrinol 226:T101-122.
Gregg C, Shikar V, Larsen P, Mak G, Chojnacki A, Yong VW, Weiss S (2007) White matter plasticity and enhanced remyelination in the maternal CNS. J Neurosci 27:1812-1823.
Gustafson P, Ladyman SR, McFadden S, Larsen C, Khant Aung Z, Brown RSE, Bunn SJ, Grattan DR (2020) Prolactin receptor-mediated activation of pSTAT5 in the pregnant mouse brain. J Neuroendocrinol 32:e12901.
Halperin J, Devi YS, Elizur S, Stocco C, Shehu A, Rebourcet D, Unterman TG, Leslie ND, Le J, Binart N, Gibori G (2008) Prolactin signaling through the short form of its receptor represses forkhead transcription factor FOXO3 and its target gene galt causing a severe ovarian defect. Mol Endocrinol 22:513-522.
Hiraoka Y, Tatsumi K, Shiozawa M, Aiso S, Fukasawa T, Yasuda K, Miyai K (1991) A placenta-specific 5′ non-coding exon of human prolactin. Mol Cell Endocrinol 75:71-80.
Ittig S, Studerus E, Heitz U, Menghini-Müller S, Beck K, Egloff L, Leanza L, Andreou C, Riecher-Rössler A (2017) Sex differences in prolactin levels in emerging psychosis: indication for enhanced stress reactivity in women. Schizophr Res 189:111-116.
Jiang X, Andjelkovic AV, Zhu L, Yang T, Bennett MVL, Chen J, Keep RF, Shi Y (2018) Blood-brain barrier dysfunction and recovery after ischemic stroke. Prog Neurobiol 163-164:144-171.
Kamesh A, Black EAE, Ferguson AV (2018) The subfornical organ: a novel site for prolactin action. J Neuroendocrinol 30:e12613.
Kang JH, Hassan SA, Zhao P, Tsai-Morris CH, Dufau ML (2014) Impact of subdomain D1 of the short form S1b of the human prolactin receptor on its inhibitory action on the function of the long form of the receptor induced by prolactin. Biochim Biophys Acta 1840:2272-2280.
Kirk SE, Grattan DR, Bunn SJ (2019) The median eminence detects and responds to circulating prolactin in the male mouse. J Neuroendocrinol 31:e12733.
Kokay IC, Wyatt A, Phillipps HR, Aoki M, Ectors F, Boehm U, Grattan DR (2018) Analysis of prolactin receptor expression in the murine brain using a novel prolactin receptor reporter mouse. J Neuroendocrinol 30:e12634.
Lajud N, Gonzalez-Zapien R, Roque A, Tinajero E, Valdez JJ, Clapp C, Torner L (2013) Prolactin administration during early postnatal life decreases hippocampal and olfactory bulb neurogenesis and results in depressive-like behavior in adulthood. Horm Behav 64:781-789.

Larsen CM, Grattan DR (2010) Prolactin-induced mitogenesis in the subventricular zone of the maternal brain during early pregnancy is essential for normal postpartum behavioral responses in the mother. Endocrinology 151:3805-3814.
Larsen CM, Kokay IC, Grattan DR (2008) Male pheromones initiate prolactin-induced neurogenesis and advance maternal behavior in female mice. Horm Behav 53:509-517.
Leem YH, Park JS, Chang H, Park J, Kim HS (2019) Exercise prevents memory consolidation defects via enhancing prolactin responsiveness of CA1 neurons in mice under chronic stress. Mol Neurobiol 56:6609-6625.
Login IS, MacLeod RM (1977) Prolactin in human and rat serum and cerebrospinal fluid. Brain Res 132:477-483.
López-Rincón G, Pereira-Suárez AL, Del Toro-Arreola S, Sánchez-Hernández PE, Ochoa-Zarzosa A, Muñoz-Valle JF, Estrada-Chávez C (2013) Lipopolysaccharide induces the expression of an autocrine prolactin loop enhancing inflammatory response in monocytes. J Inflamm (Lond) 10:24.
Machado RB, Rocha MR, Suchecki D (2017) Brain prolactin is involved in stress-induced REM sleep rebound. Horm Behav 89:38-47.
Mak GK, Enwere EK, Gregg C, Pakarainen T, Poutanen M, Huhtaniemi I, Weiss S (2007) Male pheromone-stimulated neurogenesis in the adult female brain: possible role in mating behavior. Nat Neurosci 10:1003-1011.
Mak GK, Weiss S (2010) Paternal recognition of adult offspring mediated by newly generated CNS neurons. Nat Neurosci 13:753-758.
Marques F, Sousa JC, Brito MA, Pahnke J, Santos C, Correia-Neves M, Palha JA (2017) The choroid plexus in health and in disease: dialogues into and out of the brain. Neurobiol Dis 107:32-40.
Medina J, Workman JL (2020) Maternal experience and adult neurogenesis in mammals: implications for maternal care, cognition, and mental health. J Neurosci Res 98:1293-1308.
Mitani S, Amano I, Takatsuru Y (2018) High prolactin concentration during lactation period induced disorders of maternal behavioral in offspring. Psychoneuroendocrinology 88:129-135.
Möderscheim TA, Gorba T, Pathipati P, Kokay IC, Grattan DR, Williams CE, Scheepens A (2007) Prolactin is involved in glial responses following a focal injury to the juvenile rat brain. Neuroscience 145:963-973.
Molina-Salinas G, Rivero-Segura NA, Cabrera-Reyes EA, Rodríguez-Chávez V, Langley E, Cerbon M (2021) Decoding signaling pathways involved in prolactin-induced neuroprotection: a review. Front Neuroendocrinol 61:100913.
Morales T, Lorenson M, Walker AM, Ramos E (2014) Both prolactin (PRL) and a molecular mimic of phosphorylated PRL, S179D-PRL, protect the hippocampus of female rats against excitotoxicity. Neuroscience 258:211-217.
Moreno-Ruiz B, Mellado S, Zamora-Moratalla A, Albarracín AL, Martín ED (2021) Increase in serum prolactin levels in females improves the performance of spatial learning by promoting changes in the circuital dynamics of the hippocampus. Psychoneuroendocrinology 124:105048.
Nie H, Huang PQ, Jiang SH, Yang Q, Hu LP, Yang XM, Li J, Wang YH, Li Q, Zhang YF, Zhu L, Zhang YL, Yu Y, Xiao GG, Sun YW, Ji J, Zhang ZG (2021) The short isoform of PRLR suppresses the pentose phosphate pathway and nucleotide synthesis through the NEK9-Hippo axis in pancreatic cancer. Theranostics 11:3898-3915.
Ortiz-Pérez A, Limón-Morales O, Rojas-Castañeda JC, Cerbón M, Picazo O (2019) Prolactin prevents the kainic acid-induced neuronal loss in the rat hippocampus by inducing prolactin receptor and putatively increasing the VGLUT1 overexpression. Neurosci Lett 694:116-123.
Pathipati P, Gorba T, Scheepens A, Goffin V, Sun Y, Fraser M (2011) Growth hormone and prolactin regulate human neural stem cell regenerative activity. Neuroscience 190:409-427.
Phillipps HR, Rand CJ, Brown RSE, Kokay IC, Stanton JA, Grattan DR (2019) Prolactin regulation of insulin-like growth factor 2 gene expression in the adult mouse choroid plexus. FASEB J 33:6115-6128.
Phillipps HR, Yip SH, Grattan DR (2020) Patterns of prolactin secretion. Mol Cell Endocrinol 502:110679.
Pi X, Voogt JL, Grattan DR (2002) Detection of prolactin receptor mRNA in the corpus striatum and substantia nigra of the rat. J Neurosci Res 67:551-558.
Pi XJ, Grattan DR (1999) Increased prolactin receptor immunoreactivity in the hypothalamus of lactating rats. J Neuroendocrinol 11:693-705.
Piccoletti R, Maroni P, Bendinelli P, Bernelli-Zazzera A (1994) Rapid stimulation of mitogen-activated protein kinase of rat liver by prolactin. Biochem J 303:429-433.
Rivero-Segura NA, Flores-Soto E, García de la Cadena S, Coronado-Mares I, Gomez-Verjan JC, Ferreira DG, Cabrera-Reyes EA, Lopes LV, Massieu L, Cerbón M (2017) Prolactin-induced neuroprotection against glutamate excitotoxicity is mediated by the reduction of [Ca2+]i overload and NF-κB activation. PLoS One 12:e0176910.
Rosas-Hernandez H, Cuevas E, Lantz SM, Hamilton WR, Ramirez-Lee MA, Ali SF, Gonzalez C (2013) Prolactin and blood-brain barrier permeability. Curr Neurovasc Res 10:278-286.
Roselli CE, Bocklandt S, Stadelman HL, Wadsworth T, Vilain E, Stormshak F (2008) Prolactin expression in the sheep brain. Neuroendocrinology 87:206-215.
Sairenji TJ, Ikezawa J, Kaneko R, Masuda S, Uchida K, Takanashi Y, Masuda H, Sairenji T, Amano I, Takatsuru Y, Sayama K, Haglund K, Dikic I, Koibuchi N, Shimokawa N (2017) Maternal prolactin during late pregnancy is important in generating nurturing behavior in the offspring. Proc Natl Acad Sci U S A 114:13042-13047.
Salais-López H, Abellan-Alvaro M, Bellés M, Lanuza E, Agustin-Pavon C, Martínez-García F (2020) Maternal motivation: exploring the roles of prolactin and pup stimuli. Neuroendocrinology doi: 10.1159/000510038.
Salais-López H, Agustín-Pavón C, Lanuza E, Martínez-García F (2018) The maternal hormone in the male brain: sexually dimorphic distribution of prolactin signalling in the mouse brain. PLoS One 13:e0208960.
Salais-López H, Lanuza E, Agustín-Pavón C, Martínez-García F (2017) Tuning the brain for motherhood: prolactin-like central signalling in virgin, pregnant, and lactating female mice. Brain Struct Funct 222:895-921.
Segarra M, Aburto MR, Acker-Palmer A (2021) Blood-brain barrier dynamics to maintain brain homeostasis. Trends Neurosci 44:393-405.
Shingo T, Gregg C, Enwere E, Fujikawa H, Hassam R, Geary C, Cross JC, Weiss S (2003) Pregnancy-stimulated neurogenesis in the adult female forebrain mediated by prolactin. Science 299:117-120.
Smeeth DM, Kourouzidou I, Duarte RRR, Powell TR, Thuret S (2021) Prolactin, estradiol and testosterone differentially impact human hippocampal neurogenesis in an in vitro model. Neuroscience 454:15-39.
Stagkourakis S, Smiley KO, Williams P, Kakadellis S, Ziegler K, Bakker J, Brown RSE, Harkany T, Grattan DR, Broberger C (2020) A neuro-hormonal circuit for paternal behavior controlled by a hypothalamic network oscillation. Cell 182:960-975.
Struman I, Bentzien F, Lee H, Mainfroid V, D'Angelo G, Goffin V, Weiner RI, Martial JA (1999) Opposing actions of intact and N-terminal fragments of the human prolactin/growth hormone family members on angiogenesis: an efficient mechanism for the regulation of angiogenesis. Proc Natl Acad Sci U S A 96:1246-1251.
Studerus E, Ittig S, Beck K, Del Cacho N, Vila-Badia R, Butjosa A, Usall J, Riecher-Rössler A (2021) Relation between self-perceived stress, psychopathological symptoms and the stress hormone prolactin in emerging psychosis. J Psychiatr Res 136:428-434.
Szczesna M, Kirsz K, Misztal T, Zieba DA (2020) Pregnancy-induced changes in the transcript levels of prolactin receptor and its suppressor in the ovine hypothalamus and adenohypophysis. Reprod Domest Anim 55:21-28.
Talhada D, Costa-Brito AR, Duarte AC, Costa AR, Quintela T, Tomás J, Gonçalves I, Santos CRA (2020) The choroid plexus: simple structure, complex functions. J Neurosci Res 98:751-753.
Tani N, Ikeda T, Watanabe M, Toyomura J, Ohyama A, Ishikawa T (2018) Prolactin selectively transported to cerebrospinal fluid from blood under hypoxic/ischemic conditions. PLoS One 13:e0198673.
Tian RH, Bai Y, Li JY, Guo KM (2019) Reducing PRLR expression and JAK2 activity results in an increase in BDNF expression and inhibits the apoptosis of CA3 hippocampal neurons in a chronic mild stress model of depression. Brain Res 1725:146472.
Tölli A, Höybye C, Bellander BM, Borg J (2019) Impact of pituitary dysfunction on cognitive and global outcome after traumatic brain injury and aneurysmal subarachnoid haemorrhage. J Rehabil Med 51:264-272.
Torner L, Karg S, Blume A, Kandasamy M, Kuhn HG, Winkler J, Aigner L, Neumann ID (2009) Prolactin prevents chronic stress-induced decrease of adult hippocampal neurogenesis and promotes neuronal fate. J Neurosci 29:1826-1833.
Torner L, Maloumby R, Nava G, Aranda J, Clapp C, Neumann ID (2004) In vivo release and gene upregulation of brain prolactin in response to physiological stimuli. Eur J Neurosci 19:1601-1608.
Torner L, Toschi N, Nava G, Clapp C, Neumann ID (2002) Increased hypothalamic expression of prolactin in lactation: involvement in behavioural and neuroendocrine stress responses. Eur J Neurosci 15:1381-1389.
Trott JF, Hovey RC, Koduri S, Vonderhaar BK (2003) Alternative splicing to exon 11 of human prolactin receptor gene results in multiple isoforms including a secreted prolactin-binding protein. J Mol Endocrinol 30:31-47.
Udvari EB, Völgyi K, Kékesi KA, Simon D, Hunyadi-Gulyás É, Dobolyi A (2019) Proteomic analysis of the maternal preoptic area in rats. Neurochem Res 44:2314-2324.
Uriarte N, Ferreño M, Méndez D, Nogueira J (2020) Reorganization of perineuronal nets in the medial preoptic area during the reproductive cycle in female rats. Sci Rep 10:5479.
Vermani B, Mukherjee S, Kumar G, Patnaik R (2020) Prolactin attenuates global cerebral ischemic injury in rat model by conferring neuroprotection. Brain Inj 34:685-693.
Voigt C, Bennett N (2018) Reproductive status affects the expression of prolactin receptor mRNA in the brain of female Damaraland mole-rats. J Chem Neuroanat 94:1-7.
Wagner K, Couillard-Despres S, Lehner B, Brockhoff G, Rivera FJ, Blume A, Neumann I, Aigner L (2009) Prolactin induces MAPK signaling in neural progenitors without alleviating glucocorticoid-induced inhibition of in vitro neurogenesis. Cell Physiol Biochem 24:397-406.
Walker TL, Vukovic J, Koudijs MM, Blackmore DG, Mackay EW, Sykes AM, Overall RW, Hamlin AS, Bartlett PF (2012) Prolactin stimulates precursor cells in the adult mouse hippocampus. PLoS One 7:e44371.
Walsh RJ, Slaby FJ, Posner BI (1987) A receptor-mediated mechanism for the transport of prolactin from blood to cerebrospinal fluid. Endocrinology 120:1846-1850.
Wang B, Li L, He Z, Wang L, Zhang S, Qiao H, Jia R, Tai F (2018) Effects of reproductive experience on paternal behavior, levels of testosterone, prolactin in serum and dendritic spines in medial prefrontal cortex of mandarin voles. Integr Zool 13:711-722.
Wang W, Pan YW, Wietecha T, Zou J, Abel GM, Kuo CT, Xia Z (2013) Extracellular signal-regulated kinase 5 (ERK5) mediates prolactin-stimulated adult neurogenesis in the subventricular zone and olfactory bulb. J Biol Chem 288:2623-2631.
Wen Y, Wang Y, Chelariu-Raicu A, Stur E, Liu Y, Corvigno S, Bartsch F, Redfern L, Zand B, Kang Y, Liu J, Baggerly K, Sood AK (2020) Blockade of the short form of prolactin receptor induces FOXO3a/EIF-4EBP1-mediated cell death in uterine cancer. Mol Cancer Ther 19:1943-1954.
Yip SH, Eguchi R, Grattan DR, Bunn SJ (2012) Prolactin signalling in the mouse hypothalamus is primarily mediated by signal transducer and activator of transcription factor 5b but not 5a. J Neuroendocrinol 24:1484-1491.
Zamora-Moratalla A, Martín ED (2021) Prolactin enhances hippocampal synaptic plasticity in female mice of reproductive age. Hippocampus 31:281-293.
Zhang C, Raveney BJE, Hohjoh H, Tomi C, Oki S, Yamamura T (2019) Extrapituitary prolactin promotes generation of eomes-positive helper t cells mediating neuroinflammation. Proc Natl Acad Sci U S A 116:21131-21139.
Zhang H, Shao S, Su Q, Yao D, Sun H, Ding D, Dang S, Wang S, Zhu Z, Li H (2018) Involvement of prolactin in newborn infant irritability following maternal perinatal anxiety symptoms. J Affect Disord 238:526-533.
Zhang H, Su Q, Yao D, Wang S, Dang S, Ding D, Zhu Z, Shao S, Li H (2017) Prolactin, a potential mediator of reduced social interactive behavior in newborn infants following maternal perinatal depressive symptoms. J Affect Disord 215:274-280.
Zhornitsky S, Johnson TA, Metz LM, Weiss S, Yong VW (2015) Prolactin in combination with interferon-β reduces disease severity in an animal model of multiple sclerosis. J Neuroinflammation 12:55.
Zhornitsky S, Yong VW, Weiss S, Metz LM (2013) Prolactin in multiple sclerosis. Mult Scler 19:15-23.

C-Editors: Zhao M, Liu WJ, Li JY; T-Editor: Jia Y