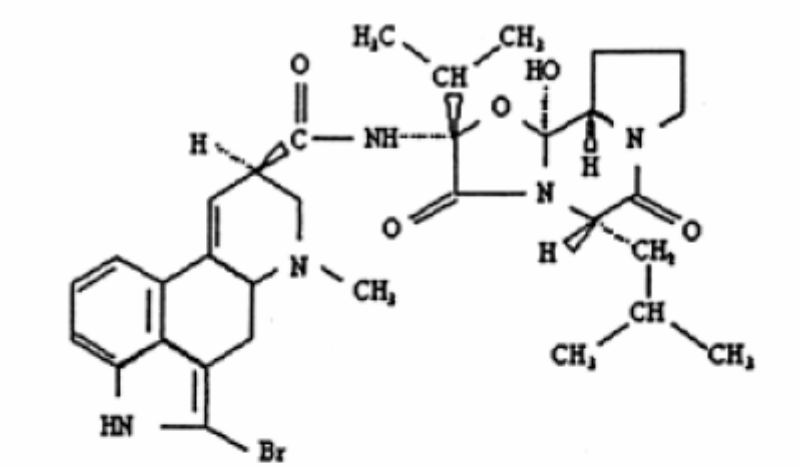
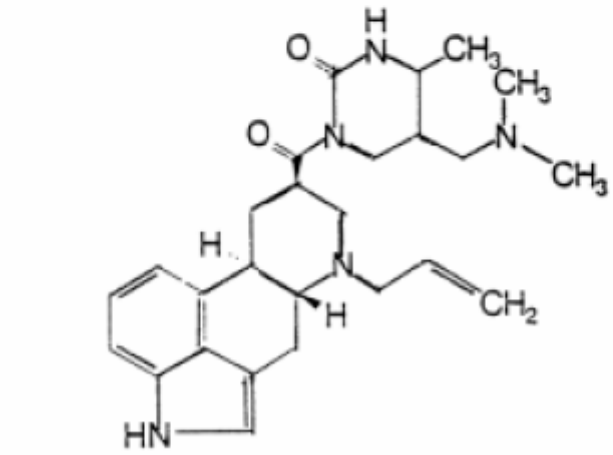
BROMOCRIPTINA - CABERGOLINA

IN

MALATTIE LINFOPROLIFERATIVE



Bromocriptina



Cabergolina

Introduzione

La logica ispiratrice del protocollo Di Bella realizza l'effetto antiblastico attraverso l'eliminazione o almeno il contrasto dei più importanti fattori di crescita, tra cui l'ormone prolattina.

La prolattina fa parte di una famiglia di ormoni che comprende anche l'ormone della crescita (GH) e l'ormone lattogeno placentare (PL) (Goffin, Binart et al. 2002; Harris, Stanford et al. 2004). La prolattina esercita i suoi effetti sulle cellule bersaglio mediante il legame al proprio recettore di membrana, di cui ne esistono tre varianti: recettore breve, intermedio e lungo. Il recettore di membrana della prolattina è un membro della super-famiglia di recettori di membrana di Classe I (Bole-Feysot, Goffin et al. 1998; Kossiakoff 2004). A questa super-famiglia appartengono anche i seguenti recettori per:

- GH (ormone della crescita)
- EPO (eritropoietina)
- TPO (trombopoietina)
- G-CSF (fattore che stimola la colonia granulocitica)
- GM-CSF (fattore che stimola la colonia granulocito-macrofagica)
- Interleuchine

I recettori della prolattina sono espressi in molti tipi di cellule diverse indicando che tale ormone esercita molti effetti fisiologici diversi. Infatti, ad oggi sono state descritte oltre 300 differenti funzioni della prolattina (Bole-Feysot, Goffin et al. 1998; Harris, Stanford et al. 2004).

La prolattina è stata associata con differenti forme di cancro (Ben-Jonathan, Liby et al. 2002). Per esempio, la prolattina può:

- aumentare l'aggressività del tumore colorettale (Bhatavdekar, Patel et al. 1994; Bhatavdekar, Patel et al. 1995)
- indurre la proliferazione di diverse linee di cancro del seno umano (Vonderhaar 1998; Vonderhaar 1999)
- indurre la proliferazione di cellule di cancro della prostata (Janssen, Darro et al. 1996)
- indurre la proliferazione di cellule di leucemia acuta mieloide (Nishiguchi, Hibasami et al. 1993)
- indurre la proliferazione di cellule di leucemia acuta linfoide (Matera, Cutufia et al. 1997)
- indurre la proliferazione di linfociti B maligni (Walker, Montgomery et al. 1995)
- indurre la proliferazione di cellule di linfoma (Gout, Beer et al. 1980; Yu-Lee 1990)
- favorire il processo di epatocarcinogenesi (Buckley, Putnam et al. 1988)

È stato anche mostrato che tumori miometrali fibromuscolari benigni (leiomiomi) producono più prolattina rispetto al normale miometrio, indicando che la prolattina prodotta localmente possa esercitare un'azione mitogenica sulla crescita di questi tumori (Nowak, Rein et al. 1993).

La secrezione di prolattina è inibita dalla dopamina, che appartiene alla classe di neurotrasmettitori conosciuti come catecolamine. La dopamina dopo il legame ai propri recettori di tipo 2 (D_2R), situati sulla membrana cellulare, sopprime la produzione ed il rilascio di prolattina, dalle cellule che producono tale ormone (Ben-Jonathan and Hnasko 2001). Sono stati sintetizzati degli agonisti della dopamina, che sono già utilizzati in clinica, come la bromocriptina (Mehta and Tolis 1979) e la cabergolina (Rains, Bryson et al. 1995). Bromocriptina e cabergolina, come tutte le molecole dopamino-agoniste, inibiscono l'increzione prolattinica interagendo con i recettori D2, la cui attivazione riduce l'attività dell'adenilato-ciclasi e la concentrazione di AMPc intracellulare (Ben-Jonathan and Hnasko 2001).
L'emivita della bromocriptina si aggira sulle 4-5 ore. La cabergolina ha un'emivita di circa 63-68 ore, con sintomi collaterali a livello dell'apparato gastro-enterico più lievi nella maggior parte dei pazienti ed una risposta terapeutica migliore della bromocriptina secondo vari studi farmacologici (Rains, Bryson et al. 1995; Colao, di Sarno et al. 2002).

Bromocriptina in malattie linfoproliferative

Diversi dati sperimentali indicano il potenziale per un'azione autocrina/paracrina della prolattina nelle cellule emopoietiche (Matera 1996; Ben-Jonathan, Liby et al. 2002). Il recettore della prolattina è espresso dalla maggior parte delle cellule del sistema immunitario, sia normali che maligne (O'Neal, Schwarz et al. 1991; Dardenne, de Moraes Mdo et al. 1994; Matera, Geuna et al. 2000).
Mentre il recettore della prolattina è espresso da molte cellule, la prolattina è prodotta principalmente dalle cellule T, anche se altre cellule immuni possono produrre prolattina (DiMattia, Gellersen et al. 1988; Pellegrini, Lebrun et al. 1992).
Anche cellule emopoietiche maligne possono produrre prolattina. È stato riportato che cellule leucemiche mieloidi, così come mieloblasti isolati da pazienti con leucemia acuta producono la prolattina (Kooijman, Gerlo et al. 2000). Un altro lavoro ha mostrato che anche diverse linee cellulari di linfoma non-Hodgkin producono la prolattina (Matera, Geuna et al. 2000).

La linea cellulare di linfoma di ratto, detta Nb2, dipende dalla prolattina per la crescita. Inoltre, tale linea cellulare è stata ampiamente utilizzata per studiare le vie regolative indotte dalla prolattina (Davis and Linzer 1988; LaVoie and Witorsch 1995; Ganguli, Hu et al. 1996; Camarillo, Linebaugh et al. 1997; Camarillo and Rillema 1998; Krumenacker, Buckley et al. 1998; Al-Sakkaf, Mooney et al. 2000; Yu and Rillema 2000).

Sebbene le cellule Nb2 siano prolattina dipendenti per la crescita, nella maggior parte delle cellule immuni umane non è stato dimostrato un simile ruolo obbligato per la prolattina. Tuttavia, è stato riportato che la prolattina agisce da co-mitogeno, specialmente attraverso l'induzione del recettore per Interleuchina-2 (IL2), promuovendo così la proliferazione cellulare stimolata da IL2 (Ben-Jonathan, Liby et al. 2002).

Un'eccezione è rappresentata dalla linea cellulare di leucemia T umana, detta Jurkat, che non esprime costitutivamente IL-2 o il recettore per IL2; per cui in tali cellule la prolattina agisce come un mitogeno autocrino (Matera, Cutufia et al. 1997).

Nelle cellule immuni, la prolattina inibisce il processo apoptotico, stimolando la produzione di diverse proteine antiapoptotiche, come Pim-1, Bax e Bcl-2 (Krumenacker, Buckley et al. 1998; Buckley and Buckley 2000).

Sia la prolattina che il GH, entrambi appartenenti alla stessa famiglia di ormoni, possono partecipare allo sviluppo e/o alla progressione di certe neoplasie ematologiche (Hooghe, Merchav et al. 1998).

Per cui, l'uso di inibitori del rilascio di questi due ormoni, come somatostatina/octreotide e bromocriptina/cabergolina, utilizzati nel Metodo Di Bella, possono essere di grande utilità nel trattamento di leucemie e linfomi.

Uno studio clinico ha valutato la somatostatina e la bromocriptina in combinazione con ciclofosfamide, retinoidi, melatonina e ACTH nel trattamento di linfomi non-Hodgkin a basso grado in fase avanzata (Todisco, Casaccia et al. 2001). Su 20 pazienti valutabili per risposta e tossicità il 70% (14 su 20) ebbe una risposta parziale; il 20% (4 su 20) ebbe una malattia stabile ed il 10% (2 su 20) ebbe progressione di malattia. Continuando con la terapia, nessuno dei 14 pazienti con una risposta parziale ebbe una progressione della malattia (tempo di controllo medio di 21 mesi, intervallo da 7 a 25) ed il 50% di questi pazienti ebbe una risposta completa. Dei 4 pazienti con malattia stabile, il 25% (1 di 4) ebbe una risposta parziale ed il 75% (3 di 4) progredirono con la terapia (tempo medio di progressione 14.3 mesi, intervallo da 7 a 21). La tossicità era molto modesta, gli effetti collaterali più comuni furono: sonnolenza, diarrea e iperglicemia.
Questo studio conferma che in neoplasie ematologiche, l'utilizzo di inibitori della prolattina, associati ad altre molecole efficaci in tali neoplasie, può essere di utilità terapeutica e supporta il razionale scientifico del Metodo Di Bella.

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J Endocrinol. 2000 Oct;167(1):85-92.

Possible role for protein kinase B in the anti-apoptotic effect of prolactin in rat Nb2 lymphoma cells.

Al-Sakkaf KA, Mooney LM, Dobson PR, Brown BL.

Prolactin (PRL) is a mitogen for a number of cell types and its action as a survival factor has recently been demonstrated in Nb2 lymphoma cells. However, the intracellular signalling pathways by which PRL promotes the survival of Nb2 cells is unknown. In previous studies, we have shown that PRL caused the activation of phosphatidylinositol 3-kinase (PI3-kinase) and its association with tyrosine phosphorylated fyn. Protein kinase B (PKB), a serine/threonine kinase, is now known to be a downstream component of the PI3-kinase pathway. The aim of the present study was to examine the effect of PRL on the activation of PKB and to find out whether this has any role on the PRL-induced survival of Nb2 cells. Our studies have revealed the phosphorylation and activation of PKB in PRL-stimulated Nb2 cells. We have also observed, using confocal microscopy, translocation of PKB to the membrane of Nb2 cells in response to PRL. These effects were blocked by the PI3-kinase inhibitor, LY294002 (10 microgram/ml). Apoptosis was induced by the general protein kinase inhibitor, staurosporine (STS; 0.1-1 microM), the synthetic glucocorticoid, dexamethasone (Dex; 100 nM) or ionising radiation by exposing Nb2 cells to X-irradiation (IR; 10 Gy). PRL had no effect on STS-induced apoptosis. On the other hand, PRL (100 ng/ml) inhibited apoptosis induced by Dex or IR; this effect of PRL was reversed by the addition of LY294002 (10 microgram/ml). Furthermore, Western blot analysis using phosphospecific PKB antibody on lysates from PRL-treated Nb2 cells showed that phosphorylation of PKB in response to PRL was inhibited by STS (0.5 microM), but not by Dex (100 nM). These results suggest that the PI3-kinase/PKB pathway may mediate the anti-apoptotic effect of PRL in Nb2 cells.

Endocr Rev. 2001 Dec;22(6):724-63.

Dopamine as a prolactin (PRL) inhibitor.

Ben-Jonathan N, Hnasko R.

Dopamine is a small and relatively simple molecule that fulfills diverse functions. Within the brain, it acts as a classical neurotransmitter whose attenuation or overactivity can result in disorders such as Parkinson's disease and schizophrenia. Major advances in the cloning and characterization of biosynthetic enzymes, transporters, and receptors have increased our knowledge regarding the metabolism, release, reuptake, and mechanism of action of dopamine. Dopamine reaches the pituitary via hypophysial portal blood from several hypothalamic nerve tracts that are regulated by PRL itself, estrogens, and several neuropeptides and neurotransmitters. Dopamine binds to type-2 dopamine receptors that are functionally linked to membrane channels and G proteins and suppresses the high intrinsic secretory activity of the pituitary lactotrophs. In addition to inhibiting PRL release by controlling calcium fluxes, dopamine activates several interacting intracellular signaling pathways and suppresses PRL gene expression and lactotroph proliferation. Thus, PRL homeostasis should be viewed in the context of a fine balance between the action of dopamine as an inhibitor and the many hypothalamic, systemic, and local factors acting as stimulators, none of which has yet emerged as a primary PRL releasing factor. The generation of transgenic animals with overexpressed or mutated genes expanded our understanding of dopamine-PRL interactions and the physiological consequences of their perturbations. PRL release in humans, which differs in many respects from that in laboratory animals, is affected by several drugs used in clinical practice. Hyperprolactinemia is a major neuroendocrine-related cause of reproductive disturbances in both men and women. The treatment of hyperprolactinemia has greatly benefited from the generation of progressively more effective and selective dopaminergic drugs.

Trends Endocrinol Metab. 2002 Aug;13(6):245-50.

Prolactin as an autocrine/paracrine growth factor in human cancer.

Ben-Jonathan N, Liby K, McFarland M, Zinger M.

Prolactin (PRL) has a dual function -- as a circulating hormone and as a cytokine. This understanding is based on PRL production and distinct regulation in extrapituitary sites, its binding to membrane receptors of the cytokine receptor superfamily, and activation of signaling pathways that promote cell growth and survival. There is increasing evidence that PRL plays a role in several types of cancer in reproductive and non-reproductive tissues via local production or accumulation. The expression of both PRL and its receptor in human cancer cell lines of diverse origin lends further support to its action as an autocrine/paracrine growth factor. Establishment of PRL as an active participant in tumorigenesis should inspire the development of novel therapies aimed at reducing tumor growth by suppressing PRL production or by blocking its receptors.

J Surg Oncol. 1994 Apr;55(4):246-9.

Interrelationship of prolactin and its receptor in carcinoma of colon and rectum: a preliminary report.

Bhatavdekar J, Patel D, Ghosh N, Vora H, Shah N, Karelia N, Balar D, Chikhlikar P, Dave R.

The prolactin receptors (PRLR) were correlated with circulating prolactin and various clinicopathologic parameters to investigate its prognostic value in patients with colorectal cancer. The prolactin (by radioimmunoassay) and its receptors (by radioligand method) were estimated in a total of 71 male patients with colorectal cancer enrolled at the Gujarat Cancer and Research Institute, Ahmedabad. The patients were followed for a period of 3 years. We have observed that 51% colorectal tumors were PRLR+. Significant correlation was not observed between presence/absence of PRLR and clinicopathologic variables. Dukes' D patients were lost to follow-up after 2-3 months; therefore, the results of prognostic significance were analysed only in patients with Dukes' A, B, and C (N = 64). Statistically significant difference in overall survival was not observed when the patients were subgrouped according to the presence/absence of PRLR and according to the cutoff level (i.e., 2%). PRLR+ hyperprolactinemic (Prolactin > 20.0 ng/ml plasma) patients had better overall survival than that of patients with PRLR- hyperprolactinemia, although the difference was statistically nonsignificant. However, PRLR- hyperprolactinemia patients had a more unfavourable prognosis than that of their counterparts. A similar trend was observed in patients with Dukes' B and C disease. Our preliminary study suggests an unequivocal finding, that PRLR- with concomitant hyperprolactinemia probably characterises a subgroup of patients with aggressive colorectal cancer.

Eur J Surg Oncol. 1995 Feb;21(1):23-6.

Prognostic value of insulin-like growth factor-1 receptors in patients with colon/rectal cancer: correlation with plasma prolactin.

Bhatavdekar JM, Patel DD, Shah NG, Karelia NH, Vora HH, Ghosh N, Suthar TP, Balar DB.

Prognostic value of IGF-1 receptors (IGF-1R) was evaluated and compared with circulating prolactin (PRL) in 59 patients with Dukes B or C colon/rectal cancer. IGF-1R was estimated by radioligand binding assay and PRL was estimated by immunoradiometric assay. Eighty-five percent (50/59) of patients had IGF-1R- tumours while IGF-1R positivity was observed in only 15% (9/59) of patients. None of the clinicopathological parameters showed any association with IGF-1R status. No significant difference was observed in overall survival period between patients with IGF-1R+ tumours and those with IGF-1R- tumours. However, a significant difference in overall survival time was observed between patients with PRL < 20.0 and > 20.0 ng/ml plasma ($X2 = 4.70$, $df = 1$, $P < 0.05$). In bivariate analysis, patients with IGF-1R- tumours and concomitant hyperprolactinemia had unfavourable prognosis compared to their counterpart ($X2 = 4.21$, $df = 1$, $P < 0.05$). We conclude that there was a trend of better overall survival in patients with IGF-1R+ tumours, and PRL < 20.0 ng/ml plasma when compared to patients with IGF-1R- tumours, and PRL > 20.0 ng/ml plasma. Further, IGF-1R negativity in conjunction with hyperprolactinemia could be used as an indicator of unfavourable prognosis in patients with Dukes B or C colon/rectal cancer.

Endocr Rev. 1998 Jun;19(3):225-68.

Prolactin (PRL) and its receptor: actions, signal transduction pathways and phenotypes observed in PRL receptor knockout mice.

Bole-Feysot C, Goffin V, Edery M, Binart N, Kelly PA.

PRL is an anterior pituitary hormone that, along with GH and PLs, forms a family of hormones that probably resulted from the duplication of an ancestral gene. The PRLR is also a member of a larger family, known as the cytokine class-1 receptor superfamily, which currently has more than 20 different members. PRLRs or binding sites are widely distributed throughout the body. In fact, it is difficult to find a tissue that does not express any PRLR mRNA or protein. In agreement with this wide distribution of receptors is the fact that now more than 300 separate actions of PRL have been reported in various vertebrates, including effects on water and salt balance, growth and development, endocrinology and metabolism, brain and behavior, reproduction, and immune regulation and protection. Clearly, a large proportion of these actions are directly or indirectly associated with the process of reproduction, including many behavioral effects. PRL is also becoming well known as an important regulator of immune function. A number of disease states, including the growth of different forms of cancer as well as various autoimmune diseases, appear to be related to an overproduction of PRL, which may act in an endocrine, autocrine, or paracrine manner, or via an increased sensitivity to the hormone. The first step in the mechanism of action of PRL is the binding to a cell surface receptor. The ligand binds in a two-step process in which site 1 on PRL binds to one receptor molecule, after which a second receptor molecule binds to site 2 on the hormone, forming a homodimer consisting of one molecule of PRL and two molecules of receptor. The PRLR contains no intrinsic tyrosine kinase cytoplasmic domain but associates with a cytoplasmic tyrosine kinase, JAK2. Dimerization of the receptor induces tyrosine phosphorylation and activation of the JAK kinase followed by phosphorylation of the receptor. Other receptor-associated kinases of the Src family have also been shown to be activated by PRL. One major pathway of signaling involves phosphorylation of cytoplasmic State proteins, which themselves dimerize and translocate to nucleus and bind to specific promoter elements on PRL-responsive genes. In addition, the Ras/Raf/MAP kinase pathway is also activated by PRL and may be involved in the proliferative effects of the hormone. Finally, a number of other potential mediators have been identified, including IRS-1, PI-3 kinase, SHP-2, PLC gamma, PKC, and intracellular Ca2+. The technique of gene targeting in mice has been used to develop the first experimental model in which the effect of the complete absence of any lactogen or PRL-mediated effects can be studied. Heterozygous (+/-)

females show almost complete failure to lactate after the first, but not subsequent, pregnancies. Homozygous (-/-) females are infertile due to multiple reproductive abnormalities, including ovulation of premeiotic oocytes, reduced fertilization of oocytes, reduced preimplantation oocyte development, lack of embryo implantation, and the absence of pseudopregnancy. Twenty per cent of the homozygous males showed delayed fertility. Other phenotypes, including effects on the immune system and bone, are currently being examined. It is clear that there are multiple actions associated with PRL. It will be important to correlate known effects with local production of PRL to differentiate classic endocrine from autocrine/paracrine effects. The fact that extrapituitary PRL can, under some circumstances, compensate for pituitary PRL raises the interesting possibility that there may be effects of PRL other than those originally observed in hypophysectomized rats. The PRLR knockout mouse model should be an interesting system by which to look for effects activated only by PRL or other lactogenic hormones. On the other hand, many of the effects reported in this review may be shared with other hormones, cytokines, or growth factors and thus will be more difficult to study. (ABSTRACT TRUNCATED)

Adv Enzyme Regul. 1988;27:371-91.

Prolactin as a mammalian mitogen and tumor promoter.

Buckley AR, Putnam CW, Russell DH.

Cellular proliferation and differentiation of the mammalian mammary gland requires a medley of hormones including the anterior pituitary hormone, PRL. Recent evidence extends the role of PRL as a mammalian mitogen to cells in several physiological systems not directly involved in reproductive functions, such as liver and lymphocytes. PRL administration induces biochemical markers expressed during the G1 phase of cell cycle and activates DNA synthesis in rat liver. Chronic PRL treatment causes hepatomegaly, reflecting its stimulation of the proliferative process. In vitro, a lactogen-dependent cell line, the Nb2 rat node lymphoma cell, serves as a useful paradigm to study PRL action on mitogenesis. These cells, when cultured in the presence of lactogens, proliferate in a dose-dependent manner. The effects of various pharmacological agents on discrete phases of the cell cycle may be readily assessed in these cells since PRL-stimulated entry into cycle is signalled by an elevation of ODC activity at 6 hr and entry into S-phase at 6-12 hr. The parallel effects of phorbol ester tumor promoters and PRL on cell cycle progression in Nb2 lymphoma cells and in hepatic proliferation suggest that PRL may likewise mediate proliferation in aberrant growth conditions such as neoplasia. The data presented support the hypothesis that PRL is capable of promoting hepatocarcinogenesis. Its chronic administration after a hepatic initiating agent stimulated the development of histochemical and biochemical markers characteristic of preneoplasia. Further, the effect of PRL was comparable to that of the hepatocarcinogen when either was administered alone. Thus, hyperprolactinemia may serve to promote the development of hepatic tumors. Phorbol esters are thought to promote tumorigenesis by directly activating PKC. In the Nb2 lymphoma cell model, tumor promoting phorbol esters mimic the effects of PRL. Similarly, PRL-stimulated enzyme induction in liver is mirrored by phorbol ester treatment, and inhibitors of PKC block PRL-stimulated mitogenesis in Nb2 cells. Further, PRL or TPA administration to rats causes translocation of PKC activity from the hepatic cytosol to the membrane fraction, reflecting kinase activation. Therefore, PRL activation of PKC appears to be a physiological phenomenon of general significance, occurring as the result of lactogen receptor stimulation and serving to transmit intracellular signals linked to the regulation of mitogenesis. Further study is required to more fully define the scope of PRL-mediated mitogenic actions as well as its effects on the expression of differentiated products in tissues and cells.

Ann N Y Acad Sci. 2000;917:522-33.

Prolactin regulation of apoptosis-associated gene expression in T cells.

Buckley AR, Buckley DJ.

Evidence accumulated over the last two decades indicates important actions for prolactin (PRL) in regulation of several functions of the immune system. That PRL can serve to facilitate immune cell proliferation is well established. In addition, PRL appears to play a salient role in the genesis and/or potentiation of certain autoimmune diseases. Recent evidence from several laboratories has extended the spectrum of PRL actions in immunological systems to include regulation of lymphocyte pool size through the process of apoptosis. Experimental results obtained using lactogen-dependent rat pre-T cell lines, the Nb2 lymphoma, have demonstrated that PRL suppresses cell death mechanisms activated by cytokine/hormone deprivation and cytotoxic drugs such as glucocorticoids. In this paper, we review results from studies conducted to investigate the mechanism(s) underlying PRL-regulated apoptosis suppression. Effects of the hormone on expression of apoptosis-associated genes of the Bcl-2 family as well as the protooncogene pim-1 in proliferating Nb2 sublines and in cells exposed to apoptotic stimuli are presented. It is concluded that PRL-mediated apoptosis suppression in immune cells reflects a complex interaction among several gene products.

Proc Soc Exp Biol Med. 1997 Jun;215(2):198-202.

Differential tyrosyl-phosphorylation of multiple mitogen-activated protein kinase isoforms in response to prolactin in Nb2 lymphoma cells.

Camarillo IG, Linebaugh BE, Rillema JA.

Prolactin (PRL) stimulates mitogenesis and differentiative processes in a variety of cell types. Not all of the molecules involved in PRL signaling, which follows an initial PRL-receptor interaction, have been identified. In the present studies, PRL is shown to stimulate the differential tyrosyl phosphorylation of three isoforms (ERK-1, 2, and 4) of mitogen-activated protein kinases (MAP kinase) in a rat pre-T lymphoma cell line (Nb2). Evidence also suggests that PRL stimulates the tyrosyl phosphorylation of ERK-3, a MAP kinase isoform recently identified. When G1-arrested Nb2 cells are treated with 50 ng/ml oPRL, ERK-1 through 3 become tyrosyl phosphorylated within minutes (an indication of enzyme activation) and then become dephosphorylated within 30 min. Conversely, ERK-4 is rapidly tyrosyl phosphorylated by 5 min, and remains in this state for at least 1 hr.

J Endocrinol. 1998 Oct;159(1):R1-4.

Changes in numbers of prolactin receptors during the cell cycle of Nb2 cells.

Camarillo IG, Rillema JA.

Lactogenic hormones including prolactin (PRL) have mitogenic effects on Nb2 cells, a pre-T lymphoma cell line. Previous studies have characterized the PRL stimulation of cellular processes such as RNA/DNA synthesis, signalling molecule activation, and the expression of specific genes. The data presented here explores the fluctuations in plasma membrane PRL receptor (PRLR) number that occur in the Nb2 cells during the course of a 24 h cell cycle. PRLR abundance was determined by measuring specific binding of [I125] oPRL to G1 arrested-intact Nb2 cells in which the cell cycle was initiated by addition of nonradioactive oPRL. Preliminary studies revealed that 1 ng/ml oPRL was the minimum PRL concentration that causes a maximal stimulation of mitogenesis, without interfering with [I125] oPRL binding measurements. Subsequent experiments revealed that upon cell cycle initiation of G1 arrested Nb2 cells with 1 ng/ml oPRL, PRLR number remained constant for the initial 6 h. After 8 h PRLR numbers decreased and at 12 h, the PRLR number was less than 25% of the initial value. After 12 hr, PRLR numbers increased and reach initial values by 18 hr. These studies show that the expression of cell surface PRL receptors is modulated in a sequential fashion during the cell cycle of Nb2 cells.

Expert Opin Investig Drugs. 2002 Jun;11(6):787-800.

Dopamine receptor agonists for treating prolactinomas.

Colao A, di Sarno A, Pivonello R, di Somma C, Lombardi G.

Prolactinomas are the most common hormone-secreting pituitary tumours and cause infertility and gonadal and sexual dysfunction in both sexes. The approach to prolactinomas has changed in the last 25 years thanks to the availability of dopaminergic drugs characterised by a potent prolactin-inhibitory effect, a tumour shrinking effect associated with a satisfactory tolerability. In more recent years, cabergoline 1-[(6-allelylergolin-8beta-yl)carbonyl]-1-[3-(dimethylamino) propyl]-3-ethyl-urea an ergoline derivative with potent, selective and long-lasting inhibitory activity on prolactin release, has been used to suppress prolactin secretion in women with hyperprolactinaemia. Cabergoline was shown to be significantly more effective than bromocriptine in inducing a complete biochemical response and clinical efficacy and was better tolerated than bromocriptine in the majority of patients. Notable tumour shrinkage until tumour disappearance was observed during cabergoline treatment in most patients with macroprolactinoma and it was also proven effective in patients resistant to or with a poor response to bromocriptine. In view of the limited data on cabergoline-associated pregnancies and the long half-life of the drug, it is currently recommended that women hoping to become pregnant, once ovulatory cycles have been established, should discontinue cabergoline therapy 1 month before they intend to conceive. However, no data concerning negative effects on pregnancy or offspring have been reported. The great efficacy of this compound together with its excellent tolerability makes this drug the current treatment of choice for the majority of patients with hyperprolactinaemic disorders.

Endocrinology. 1994 May;134(5):2108-14.

Prolactin receptor expression in human hematopoietic tissues analyzed by flow cytofluorometry.

Dardenne M, de Moraes Mdo C, Kelly PA, Gagnerault MC.

PRL receptor (PRL-R) expression has been analyzed in human hematopoietic tissues using flow cytofluorometric analysis with a series of biotinylated monoclonal antibodies (mAbs) directed against the extracellular domain of the rat liver PRL-R. In the thymus, more than 75% of cells were labeled by the anti-PRL-R mAb. Regarding PRL-R expression in the four T-cell subsets defined by CD4/CD8 expression, the majority of cells expressed low receptor levels, whereas a minority of double negative (CD4-CD8-) and single positive CD4+ cells were strongly labeled by the anti-PRL-R mAb. In the peripheral blood, an average of 80% of lymphoid cells, comprising all B-cells, all monocytes, and 75% of T-cells, were consistently PRL-R positive. Regarding T-cell subsets, similar percentages of PRL-R+ cells were observed in CD4+ and CD8+ peripheral lymphocytes (70-75%), and the density of labeling per cell was significantly lower than that occurring in B-cells or monocytes. Interestingly, the intensity of labeling significantly increased in peripheral T-cells after T-cell activation. The ubiquitous distribution of PRL-R in bone marrow stem cells, B-cells, monocytes, and T-cells was confirmed by the positive staining obtained in a set of human lymphoid cell lines. These data along with those showing that the PRL gene is specifically expressed in human T-cells suggest that lymphocyte PRL may act in a paracrine or autocrine fashion in both central and peripheral lymphoid organs.

Mol Endocrinol. 1988 Aug;2(8):740-6.

Autocrine stimulation of Nb2 cell proliferation by secreted, but not intracellular, prolactin.

Davis JA, Linzer DI.

We have introduced expression constructs for mouse PRL (mPRL) or a nonsecreted form of mPRL into the PRL-responsive Nb2 rat lymphoma cell line. Cell lines resulting from transfection of Nb2 cells with the wild type mPRL construct synthesize and secrete mPRL. These cells are able to grow independently of added lactogens, and conditioned media and cell extracts from these cultures stimulate the growth of Nb2 cells. In contrast, cells synthesizing the nonsecreted mPRL do not proliferate in the absence of added lactogenic hormones, and conditioned media from these cell cultures do not have PRL-like activity in the Nb2 cell growth assay. PRL protein is detected in these nonsecreting cell lines; however, extracts from these lines are generally unable to stimulate Nb2 cell proliferation. These results indicate that cells can respond in an autocrine fashion to PRL, but that an intracellular form of PRL is unable to activate Nb2 cell growth.

Endocrinology. 1988 Jun;122(6):2508-17.

A human B-lymphoblastoid cell line produces prolactin.

DiMattia GE, Gellersen B, Bohnet HG, Friesen HG.

A variety of cell lines were examined by Northern blot hybridization for the expression of PRL or PRL-related mRNAs. We found that a human B-lymphoblast cell line transcribed a mRNA which hybridized to human PRL cDNA under high stringency conditions. The human lymphoblast cell line of interest is a variant subline of the IM-9 line that we have designated IM-9-P. The lymphoblast-derived PRL mRNA is approximately 150 bases longer than that produced by the human pituitary as determined by Northern blot analysis. IM-9-P PRL was immunoaffinity purified from conditioned medium and found to be identical in mol wt by sodium dodecyl sulfate-polyacrylamide gel electrophoresis to human pituitary PRL. Moreover, IM-9-P PRL is biologically active in the rat Nb2 lymphoma mitogenic assay. Ribonuclease-H digestion of mRNA poly(A) tracts indicated that the size difference between pituitary and IM-9-P PRL transcripts was not due to an elongated poly(A) tail on the lymphoid PRL mRNA. Genomic Southern blot analysis showed no major rearrangements of the PRL gene in IM-9-P cells compared to the parent IM-9 line and human placenta DNA. Thus, it is highly likely that an elongation of the 5' and/or 3' untranslated regions of IM-9-P PRL mRNA account for the size difference with pituitary PRL mRNA. The PRL-producing IM-9-P line was cloned by limiting dilution, and a high PRL-producing clone IM-9-P3 and a non-PRL producer IM-9-P6 were isolated for further analysis. IM-9-P3 cells were found to secrete 40-50 ng PRL/10(6) cells.24 h regardless of cell density. The level of PRL mRNA also remained constant during exponential growth of IM-9-P3 cells. The existence of the PRL-producing IM-9-P3 clone and the IM-9-P6 clone which does not produce PRL as well as the IM-9 progenitor line provides a unique system with which to analyze the molecular mechanism of ectopic human PRL expression.

J Cell Physiol. 1996 May;167(2):251-60.

Nuclear accumulation of multiple protein kinases during prolactin-induced proliferation of Nb2 rat lymphoma cells.

Ganguli S, Hu L, Menke P, Collier RJ, Gertler A.

Intracellular kinases play important roles in signal transduction and are involved in the surface receptor-mediated regulation of cellular functions, including mitogenesis. In the present study, we examined the possible involvement of various protein kinases in the passage of a mitogenic signal from the cell surface to the nucleus of Nb2 cells, a rat nodal lymphoma cell line in which prolactin is a mitogen. Following a prolactin challenge, various kinase activities were monitored at short intervals in different cellular fractions over a 60 min period. Protein kinase C (PKC) activity in the cytosolic fraction rapidly declined to 50% of its original activity within the first 30 min, while PKC activity in the nuclear fractions increased sharply, reaching its highest level by 30 min following a prolactin challenge. There were also increases in both casein kinase and protein tyrosine kinase (PTK) activities in the nuclear fractions during the first 30 min following a prolactin challenge that paralleled PKC activity. The activities of all three kinases declined thereafter, reaching levels close to their respective basal values by 60 min following initiation of prolactin treatment. These observations suggest the possibility that multiple protein kinases may be involved in mitogenic signal transduction for prolactin in Nb2 cells.

Annu Rev Physiol. 2002;64:47-67.

Prolactin: the new biology of an old hormone.

Goffin V, Binart N, Touraine P, Kelly PA.

Prolactin (PRL) is a paradoxical hormone. Historically known as the pituitary hormone of lactation, it has had attributed to it more than 300 separate actions, which can be correlated to the quasi-ubiquitous distribution of its receptor. Meanwhile, PRL-related knockout models have mainly highlighted its irreplaceable role in functions of lactation and reproduction, which suggests that most of its other reported target tissues are presumably modulated by, rather than strictly dependent on, PRL. The multiplicity of PRL actions in animals is in direct opposition to the paucity of arguments that suggest its involvement in human pathophysiology other than effects on reproduction. Although many experimental data argue for a role of PRL in the progression of some tumors, such as breast and prostate cancers, drugs lowering circulating PRL levels are ineffective. This observation opens new avenues for research into the understanding of whether local production of PRL is involved in tumor growth and, if so, how extrapituitary PRL synthesis is regulated. Finally, the physiological relevance of PRL variants, such as the antiangiogenic 16K-like PRL fragments, needs to be elucidated. This review is aimed at critically discussing how these recent findings have renewed the manner in which PRL should be considered as a multifunctional hormone.

Cancer Res. 1980 Jul;40(7):2433-6.

Prolactin-stimulated growth of cell cultures established from malignant Nb rat lymphomas.

Gout PW, Beer CT, Noble RL.

A malignant Nb rat lymphoma which in vivo is stimulated by estrogens has been established in suspension culture. The cultured cells grew readily in Fischer's medium supplemented with fetal calf serum (10%) and 2-mercaptoethanol (10(-4) M). If horse serum was substituted for fetal calf serum, population growth ceased; i.e., cultures became "stationary." Such stationary cultures could be induced to resume active growth by the addition of a pituitary hormone, prolactin (ovine, rat); concentrations as low as 10 pg/ml had a detectable effect. In contrast, other pituitary hormones or estrogens had little or no effect. The evidence in this and an accompanying paper suggests that prolactin (or related substances) has a role in the growth of some cancers of lymphoid origin in rats.

Ann Med. 2004;36(6):414-25.

Prolactin and the prolactin receptor: new targets of an old hormone.

Harris J, Stanford PM, Oakes SR, Ormandy CJ.

Prolactin (PRL) is one of a family of related hormones including growth hormone (GH) and placental lactogen (PL) that are hypothesized to have arisen from a common ancestral gene about 500 million years ago. Over 300 different functions of PRL have been reported, highlighting the importance of this pituitary hormone. PRL is also synthesized by a number of extra-pituitary tissues including the mammary gland and the uterus. Most of PRL's actions are mediated by the unmodified 23 kDa peptide, however, PRL may be modified post-translation, thereby altering its biological effects. PRL exerts these effects by binding to its receptor, a member of the class I cytokine receptor super-family. This activates a number of signaling pathways resulting in the transcription of genes necessary for the tissue specific changes induced by PRL. Mouse knockout models of the major forms of the PRL receptor have confirmed the importance of PRLs role in reproduction. Further knockout models have provided insight into the importance of PRL signaling intermediates and the advent of transcript profiling has allowed the elucidation of a number of PRL target genes.

Cell Mol Life Sci. 1998 Oct;54(10):1095-101.

A role for growth hormone and prolactin in leukaemia and lymphoma?

Hooghe R, Merchav S, Gaidano G, Naessens F, Matera L.

Growth hormone (GH) and prolactin (PRL) quality as lymphohaemopoietic growth and differentiation factors, and so does insulin-like growth factor (IGF)-I, which mediates many of GH activities. Although there is only limited evidence that endocrine, paracrine or autocrine GH or PRL play a role in human leukaemia and lymphoma, the expression of these factors or their receptors may have diagnostic or therapeutic implications. Indeed, the participation of GH, PRL or IGF-I in the development or progression of certain haematological malignancies or to the antitumour immune response has been documented. Examples discussed in this review include a rat lymphoma in which the PRL receptor acts as an oncogene; the rat Nb2 lymphoma, which is dependent on PRL for growth; and experiments showing that PRL stimulates natural killer cell activity and the development of lymphokine-activated killer cells.

Cancer. 1996 Jan 1;77(1):144-9.

In vitro characterization of prolactin-induced effects on proliferation in the neoplastic LNCaP, DU145, and PC3 models of the human prostate.

Janssen T, Darro F, Petein M, Raviv G, Pasteels JL, Kiss R, Schulman CC.

BACKGROUND. Proliferation of normal and tumoral prostate tissue is regulated by androgens and various growth factors. We characterized the in vitro proliferative influence of prolactin (PRL) in androgen-sensitive and androgen-insensitive human prostate cancers. METHODS. The biologic models employed included the androgen-sensitive LNCaP and the androgen-insensitive DU145 and PC3 cell lines. PRL-induced influences (0.1-10 mIU/ml of medium) on proliferation were assessed using the colorimetric methylthiotetrazole assay. Androgen sensitivity in the three cell lines was determined by assessing the proliferative influence of dihydrotestosterone (DHT) (0.1-10 nM). PRL-induced modifications in PC3 cell kinetics were assessed using Feulgen-stained nuclear image cytometry. RESULTS. Although DHT markedly stimulated LNCaP proliferation, it had no proliferative effect on the DU145 and PC3 cell lines. By contrast, PRL significantly modulated the proliferation of the DU145 and PC3 lines, but exerted weak, if any, effect on the proliferation of the LNCaP cell line. PRL increased the percentage of PC3 proliferating cells (i.e., cells in the S/G2 phases of the cell cycle) at low doses (0.1 mIU/mL) and decreased this percentage at high doses (10 mIU/ml). CONCLUSIONS. Proliferation of androgen-insensitive human prostate cell lines can be significantly modulated by prolactin.

J Neuroimmunol. 2000 Oct 2;110(1-2):252-8.

Myeloid leukemic cells express and secrete bioactive pituitary-sized 23 kDa prolactin.

Kooijman R, Gerlo S, Coppens A, Hooghe-Peters EL.

Prolactin (PRL) is a 23 kDa polypeptide hormone of pituitary origin which is of major importance for reproduction. In addition, PRL has immunomodulatory effects and can be produced in small quantities in nonpituitary tissues. To address possible autocrine or paracrine functions of PRL in leukemia, we characterized immunoreactive PRL from the culture medium of leukemic cells. The myeloid cell line Eol-1 expresses the long extrapituitary type mRNA for PRL and synthesizes immunoreactive PRL with a molecular weight of 23 kDa. The biological activity in Eol-1 culture medium was determined using the Nb2 bioassay. This activity co-eluted with recombinant human (rh) PRL on an S-200 Sephacryl gel filtration column and could be blocked by anti-PRL antiserum. Western blot analysis and Nb2 bioassays also suggest that acute myelogenous leukemic blasts secrete bioactive 23 kDa PRL in one out of three tested patients.

Adv Protein Chem. 2004;68:147-69.

The structural basis for biological signaling, regulation, and specificity in the growth hormone-prolactin system of hormones and receptors.

Kossiakoff AA.

The pituitary hormones growth hormone (GH), prolactin (PRL) and placental lactogen (PL), are members of an extensive cytokine superfamily of hormones and receptors that share many of the same general structure-function relationships in expressing their biological activities. The biology of the pituitary hormones involves a very sophisticated interplay of cross-reactivity and specificity. Biological activity is triggered via a hormone-induced receptor homodimerization process that is regulated by tertiary features of the hormone. These hormones have an asymmetric four alpha-helical bundle structure that gives rise to two receptor binding sites that have distinctly different topographies and electrostatic character. This feature plays an important role in the regulation of these systems by producing binding surfaces with dramatically different binding affinities to the receptor extracellular domains (ECD). As a consequence, the signaling complexes for systems that activate through receptor homodimerization are formed in a controlled sequential step-wise manner. Extensive biochemical and biophysical characterization of the two hormone-receptor interfaces indicate that the energetic properties of the two binding sites are fundamentally different and that the receptor shows extraordinary conformational plasticity to mate with the topographically dissimilar sites on the hormone. An unexpected finding has been that the two hormone binding sites are allosterically coupled; a certain set of mutations in the higher affinity site can produce both conformational and energetic effects in the lower affinity site. These effects are so large that at some level they must have played some role in the evolution of the molecule.

Endocrine. 1998 Oct;9(2):163-70.

Prolactin-regulated apoptosis of Nb2 lymphoma cells: pim-1, bcl-2, and bax expression.

Krumenacker JS, Buckley DJ, Leff MA, McCormack JT, de Jong G, Gout PW, Reed JC, Miyashita T, Magnuson NS, Buckley AR.

Lactogen-dependent Nb2 lymphoma cells, widely employed for studying prolactin (PRL) mitogenic mechanisms, are also useful for investigations of apoptosis in T-lineage lymphocytes. Utilizing PRL-dependent Nb2-11 cultures, apoptosis-regulatory genes were evaluated for participation in dexamethasone- (DEX) provoked cell death or its inhibition by PRL. Treatment of lactogen-starved, G1-arrested Nb2-11 cells with DEX (100 nM) activated apoptosis within 12 h evaluated by flow cytometric analysis of fragmented DNA. This effect was not associated with altered expression of bcl-2, bax, or pim-1. PRL (10 ng/mL), coincubated with DEX-treated cells, completely blocked DEX-induced apoptosis. This inhibition was associated with increased expression of bcl-2 and pim-1 mRNAs, genes reported to suppress apoptosis, within 2-6 h after addition of the hormone. Moreover, the increased transcription of bcl-2 and pim-1 was coupled to increases in their protein levels. The results suggest that bcl-2, bax, and pim-1 do not play a critical role in DEX-induced apoptosis in Nb2 cells. However, expression of bcl-2, together with pim-1, may have a role in mediating the antiapoptotic actions of PRL.

Proc Soc Exp Biol Med. 1995 Jul;209(3):257-69.

Investigation of intracellular signals mediating the anti-apoptotic action of prolactin in Nb2 lymphoma cells.

LaVoie HA, Witorsch RJ.

Studies were undertaken to identify intracellular mediators of prolactin inhibition of glucocorticoid-induced apoptosis in Nb2 lymphoma cells. A short-term assay was implemented that quantitates fragmented DNA released from the genome by reaction with diphenylamine. Induction and inhibition of internucleosomal DNA cleavage (indicative of apoptosis) was verified by agarose gel electrophoresis of extracted cellular DNA. Synchronized Nb2 cells (G0/G1) exhibited increased DNA fragmentation after 4-hr incubation with dexamethasone (DEX) (25-100 nM) which was inhibited by ovine prolactin (oPRL) (0.1-1 ng/ml), the glucocorticoid receptor antagonist, RU486 (500 nM), and the nuclease inhibitor, aurintricarboxylic acid (100 microM). Signals previously implicated in prolactin induction of mitogenesis in Nb2 cells were investigated for their role in prolactin inhibition of apoptosis including: protein kinase C activation, arachidonic acid metabolism, polyamine production, tyrosine phosphorylation, and extracellular calcium. Protein kinase C agonists, phorbol-12-myristate-13-acetate, and 1,2-dioctanoyl-sn-glycerol, +/- the calcium ionophore, A23187 (200 nM), did not mimic oPRL inhibition of DEX-induced DNA fragmentation. Protein kinase C inhibitors, gossypol and quercetin, did not block prolactin action. Arachidonic acid did not mimic prolactin protection against DEX-induced DNA fragmentation. Inhibitors of arachidonic acid metabolism, 5,8,11,14-eicosatetraynoic acid, nordihydroguaiaretic acid, and indomethacin did not block prolactin action. The polyamine, spermine, inhibited DEX-induced DNA fragmentation at 1.5 to 2.5 mM. However, inhibition of polyamine synthesis with alpha-difluoromethyl ornithine or methylglyoxal bis(guanylhydrazone) did not inhibit prolactin action. Prolactin action was not blocked by inhibitors of tyrosine kinase activation, genistein and tyrphostin-47. On the other hand, pervanadate, a potent tyrosine phosphatase inhibitor, consistently inhibited DEX-induced DNA fragmentation. Prolactin action and DEX-induced apoptosis both occurred in calcium-free PBS. In summary, protein kinase C activation and eicosanoid production do not appear to mediate this prolactin action. Although spermine could block DNA fragmentation, blockade of the polyamine cascade did not inhibit prolactin action, suggesting that polyamines do not mediate this prolactin effect. While inhibitors of tyrosine kinase activation did not block prolactin action, tyrosine phosphatase inhibition in the presence of basal tyrosine kinase activity mimicked prolactin action, suggesting

tyrosine phosphorylation participation in the anti-apoptotic effect. Extra-cellular calcium was not required for prolactin or DEX action.

Life Sci. 1996;59(8):599-614.

Endocrine, paracrine and autocrine actions of prolactin on immune cells.

Matera L.

The immune response is regulated by locally released factors, collectively referred to as cytokines. Data on the human immune system have convincingly demonstrated that the hormone prolactin (PRL), in addition to exerting its endocrine control on the immune system, acts as a cytokine in that it is released within the immune system and regulates the lymphocyte response by paracrine and autocrine mechanisms. Both lymphocyte and pituitary PRLs are under the control of immune factors. Synthesis of human PRL by lymphocytes is induced by T-cell stimuli, while increased release of PRL by the pituitary, observed in vivo after immune challenge, may be mediated by cytokines produced by monocyte-macrophages. Since hyperprolactinemia and hypoprolactinemia are both immunosuppressive, physiological levels of circulating PRL must be necessary to maintain basal immunocompetence. The effects of Cyclosporin (CsA) on IL-2 and PRL gene activation and the analysis of the intracellular signaling events downstream IL-2 and PRL receptors suggest coordinate actions of these two cytokines during T cell activation.

J Neuroimmunol. 1997 Oct;79(1):12-21.

Prolactin is an autocrine growth factor for the Jurkat human T-leukemic cell line.

Matera L, Cutufia M, Geuna M, Contarini M, Buttiglieri S, Galin S, Fazzari A, Cavaliere C.

Despite convincing evidence of cooperation between IL-2 and endogenous prolactin (PRL) during T cell activation, the individual role of PRL as a T-cell lineage cytokine remains to be defined. We have examined the production and function of PRL on the Jurkat human T-leukemic cell line, which does not constitutively produce IL-2. The majority of Jurkat cells expressed PRL receptor (R) under standard culture conditions, whereas appearance of the alpha chain of the IL-2-R required PHA-PMA stimulation, as did IL-2 synthesis. Western blotting revealed a predominant band at 23.5 kDa and a weaker band at 25.5 kDa in both Jurkat cell lysates and human (h) pituitary PRL. Metabolic labeling of the cell lysates with 35S-methionine and immunoprecipitation with an antiserum against hPRL showed that both forms of PRL are actively synthesized by the Jurkat cell line. PRL released in the medium was biologically active in the rat Nb2 lymphoma mitogenic assay. Depletion of medium PRL with two polyclonal anti-hPRL antisera inhibited the growth of Jurkat cells in a dose-dependent manner, as evaluated by cell number and 3H-TdR uptake. Purified pituitary or recombinant hPRL at a wide range of concentrations had no significant effect on their growth, but reversed the blocking activity of the anti-hPRL antibody. Recombinant IL-2 had no effect on the antibody-induced growth inhibition. Taken as a whole, these results demonstrate that PRL can act as an autocrine T cell growth factor independently of IL-2 and are the first evidence of its involvement in human leukemic growth and possibly in leukemic transformation.

Int J Cancer. 2000 Jan 1;85(1):124-30.

Expression of prolactin and prolactin receptors by non-Hodgkin's lymphoma cells.

Matera L, Geuna M, Pastore C, Buttiglieri S, Gaidano G, Savarino A, Marengo S, Vonderhaar BK.

Prolactin (PRL) interacts with lymphocyte-signaling molecules and cytokines. Previous work has shown independent and synergistic effects of PRL on the generation of IL-2-driven anti-tumor lymphokine activated killer (LAK) activity by peripheral blood mononuclear cells (PBMC). The potential importance of PRL as a biological immunomodifier, however, is challenged by its ability to influence normal lymphocyte mitogenesis and hence lymphoid tumor growth. Since non-Hodgkin's lymphoma (NHL) cell lines were efficiently killed by LAK generated with native (n) or recombinant (r) human PRL combined with low, per se ineffective doses of IL-2, we have addressed here the question of whether PRL acts as a growth factor for LAK targets. NHL cells were analyzed for: 1. expression of the PRL receptor (PRL-R); 2. responsiveness to nPRL or rPRL; 3. constitutive expression and release of PRL; 4. existence of a PRL autocrine loop. PRL-R, defined by multiple antibodies, was detected in 3 of 12 NHL cell lines. However, nPRL or rPRL, in a wide range of concentrations (0.75-50 ng/ml), were not mitogenic for growth-arrested, PRL-R positive NHL cell lines. PRL mRNA was detected by RT-PCR in 10 of the 12 cell lines examined with a higher frequency among AIDS-related NHL cell lines. PRL protein in the immunoprecipitate of (35)S-methionine-labeled cell lysates and supernatants paralleled mRNA expression, and Western blotting analysis showed the presence of the pituitary/lymphocyte non-glycosylated (23.5 kDa) and glycosylated (25 kDa) isoforms. Experiments with blocking antibodies showed the independence from endogenous PRL for NHL cell growth.

Drugs. 1979 May;17(5):313-25.

Pharmacology of bromocriptine in health and disease.

Mehta AE, Tolis G.

Bromocriptine, a lysergic acid derivative with a bromine atom at position 2, has been found to have unique effects on the dopamine receptors in the pituitary and central nervous system and peripherally. It is rapidly and completely absorbed from the gut and is mainly excreted in the bile and faeces. It seems to have a particular specificity for the pituitary prolactinotrophe although it does have other effects in different diseases states. In spite of the fact that it is an ergot derivative, it is remarkably free of ergot vascular side effects in the doses needed for therapeutic benefit. The most common adverse effect are nausea, vomiting and postural symptoms. These can be overcome by starting at low doses and increasing the therapeutic levels. Its major use is in the suppression of prolactin in states where this hormone is elevated irrespective of cause. It has also been used in the treatment of acromegaly and is under investigation for use in other disease states probably linked with prolactin system or dopaminergic receptors.

Leuk Res. 1993 Aug;17(8):633-7.

Human promyelocytic cell line HL60 has the specific binding sites for prolactin and its ornithine decarboxylase, DNA synthesis and cellular proliferation are induced by prolactin.

Nishiguchi Y, Hibasami H, Komada Y, Sakurai M, Nakashima K.

Human prolactin (hPRL) induced ornithine decarboxylase (ODC) activity, subsequently DNA synthesis and cellular proliferation on human promyelocytic cells, HL60, cultured in a serum-free medium. HL60 cells had 2100 specific binding sites for hPRL per cell, showing a dissociation constant of 1.1 x 10(-10) M. Binding of 125I-PRL to the cells was not blocked by simultaneous addition of human growth hormone. ODC activity and DNA synthesis were activated maximally at 5 and 20 h, respectively, after the addition of 0.05 nM hPRL. These effects of PRL on cellular proliferation, ODC activity and DNA synthesis were abolished by the simultaneous addition of anti-hPRL antibody. Simultaneous addition of an irreversible inhibitor of ODC, difluoromethyl ornithine (DFMO), also abolished the inductions of ODC and DNA synthesis by hPRL. The inhibitory effect of DFMO on hPRL-induced DNA synthesis was reversed by the addition of putrescine to the culture medium. These results suggest that hPRL binds to the prolactin receptor on HL60 cells and induces ODC activity to increase cellular polyamine levels, which eventually stimulates DNA synthesis and cellular proliferation.

J Clin Endocrinol Metab. 1993 May;76(5):1308-13.

Production of prolactin by smooth muscle cells cultured from human uterine fibroid tumors.

Nowak RA, Rein MS, Heffner LJ, Friedman AJ, Tashjian AH Jr.

Uterine leiomyomas, which are myometrial smooth muscle tumors, secrete PRL. We investigated the actions of several hormones known to stimulate PRL secretion by the pituitary gland or decidua on PRL secretion by leiomyoma-derived smooth muscle cells (SMC) in monolayer culture. Cultures were verified to be SMC by immunostaining for smooth muscle alpha-action and desmin. Hormone treatments were performed in serum-free medium for 72 h. Medium was harvested every 24 h and assayed for PRL. 17 beta-Estradiol, progesterone, TRH, insulin-like growth factor-I, epidermal growth factor, and the GnRH agonist leuprolide did not affect PRL secretion by these SMC. Insulin caused a significant suppression of PRL secretion by 72 h, and this was accompanied by a 64% increase in total cell protein per well, which represented an increase in cell number. Cells were also plated at various densities to determine the effects of cell number on PRL secretion. The amount of PRL secreted per 1000 cells decreased significantly as cell number per well increased. Northern blot analysis identified PRL mRNA in fresh leiomyoma tissue. PRL mRNA in three independent cultures of SMC was then detected by reverse transcription and the polymerase chain reaction. Hybridization occurred only with the expected band of approximately 423 basepairs in size. We conclude that leiomyomas express PRL mRNA in vivo and that leiomyoma-derived SMC in culture continue to express the PRL mRNA and secrete PRL in the absence of ovarian steroids. PRL secretion by SMC in culture appears to be modulated primarily by changes in cell density.

Mol Cell Endocrinol. 1991 Dec;82(2-3):127-35.

Prolactin receptor gene expression in lymphoid cells.

O'Neal KD, Schwarz LA, Yu-Lee LY.

To understand the role of pituitary prolactin (PRL) and its receptor (PRL-R) in the growth and differentiation of lymphoid cells, PRL-R gene expression was analyzed in various lymphoid tissues and in a rat T lymphoma cell line, Nb2, which requires PRL for growth. The technique of reverse transcription coupled to polymerase chain reaction (RT-PCR) was used to detect the low abundance PRL-R transcripts. Within 30 min to 1 h, PRL stimulates a rapid but transient increase in PRL-R mRNA levels in Nb2 T cells. By 4 h, PRL-R mRNA returned to near basal levels and then gradually declined to a new steady-state level by 12 h. Significant increases in receptor RNA levels were observed in the presence of protein synthesis inhibitors, which suggests that PRL-R mRNA levels are under negative regulation. PRL-R gene expression was also demonstrated in normal mouse thymocytes, splenocytes, and in several lymphoid cell lines. The expression of the PRL-R gene in stimulated lymphoid cells provides additional evidence for the role of PRL as an immunomodulatory molecule.

Mol Endocrinol. 1992 Jul;6(7):1023-31.

Expression of prolactin and its receptor in human lymphoid cells.

Pellegrini I, Lebrun JJ, Ali S, Kelly PA.

We have investigated whether human lymphoid cells are able to synthesize and secrete human PRL (hPRL) and to express PRL receptors. Metabolic labeling with [35S]methionine and immunoprecipitation of cell extracts from human mononuclear cells (MNC) and a human T lymphocyte cell line with an antiserum against hPRL revealed protein of M(r) 23,000, identical in size to pituitary hPRL. Dilution curves of lymphocyte immunoreactive hPRL were parallel to those obtained with pituitary hPRL in an immunoradiometric assay using two monoclonal antibodies against hPRL. Polymerase chain reaction experiments with primers located in the coding sequence of hPRL showed that the hPRL gene was expressed in MNC. Furthermore, cDNA cloning and sequence analysis indicated the presence of an extra 5' noncoding exon previously described for decidual hPRL. When MNCs were further separated into B cells, T cells, and monocytes, the expression of hPRL appeared to be mainly associated with the T lymphocyte fraction. The hPRL transcript was also detected in thymocytes and in a set of human lymphoid cell lines. Finally, polymerase chain reaction experiments revealed a ubiquitous distribution of PRL receptor gene expression in B cells, T cells, and monocytes. The presence of the receptor for PRL and production of PRL by T lymphocytes suggest a possible autocrine or paracrine effect of PRL in immune cell function.

Drugs. 1995 Feb;49(2):255-79.

Cabergoline. A review of its pharmacological properties and therapeutic potential in the treatment of hyperprolactinaemia and inhibition of lactation.

Rains CP, Bryson HM, Fitton A.

Cabergoline is a synthetic ergoline which shows high specificity and affinity for the dopamine D2 receptor. It is a potent and very long-acting inhibitor of prolactin secretion. Prolactin-lowering effects occur rapidly and, after a single dose, were evident at the end of follow up (21 days) in puerperal women, and up to 14 days in patients with hyperprolactinaemia. In the only comparative study to date, cabergoline 0.5 to 1.0 mg twice weekly was more effective than bromocriptine 2.5 to 5.0 mg twice daily in the treatment of hyperprolactinaemic amenorrhoea, restoring ovulatory cycles in 72% of women and normalising plasma prolactin levels in 83%, compared with 52 and 58%, respectively, for bromocriptine. In the prevention of puerperal lactation, a single dose of cabergoline 1.0mg was as effective as bromocriptine 2.5mg twice daily for 14 days. A significantly lower incidence of rebound lactation in the third postpartum week was seen with cabergoline. Unpublished data suggest cabergoline 0.25mg twice daily for 2 days is effective in suppressing established puerperal lactation in about 85% of women. Nausea, vomiting, headache and dizziness are characteristic adverse events of the dopaminergic ergot derivatives. Cabergoline appears to be better tolerated than bromocriptine in both patients with hyperprolactinaemia and postpartum women. Most patients intolerant of other ergot derivatives can tolerate cabergoline. Bromocriptine use in the puerperium has been associated with an increased risk of serious thromboembolic events. However, there are no such reports with cabergoline and whether these events will become associated with other dopaminergic agents is unknown. The teratogenic potential of cabergoline has not been extensively investigated in humans. Ten congenital abnormalities have been reported in 199 cabergoline-associated pregnancies. Although there is no pattern to these abnormalities, the limited experience with cabergoline in pregnancy means the drug cannot be considered as a first-line therapy for the treatment of infertility associated with hyperprolactinaemia. At this stage of its development, cabergoline will prove useful in patients with hyperprolactinaemia who have failed treatment with, or are intolerant of, other dopamine agonists such as bromocriptine. If drug treatment is required for the prevention or suppression of puerperal lactation, cabergoline offers significant advantages over bromocriptine and should become the drug treatment of first choice for this indication.

Cancer Biother Radiopharm. 2001 Apr;16(2):171-7.

Cyclophosphamide plus somatostatin, bromocriptin, retinoids, melatonin and ACTH in the treatment of low-grade non-Hodgkin's lymphomas at advanced stage: results of a phase II trial.

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PURPOSE: Somatostatin, prolactin, retinoids, melatonin and ACTH have been shown to influence the lymphatic growth, and the action of the cyclophosphamide in lymphoproliferative disorders is well known. This provided the rationale to conduct, in patients with low-grade non-Hodgkin's lymphomas (NHL), a phase II trial of a combined association of cyclophosphamide, somatostatin, bromocriptin, retinoids, melatonin and ACTH. PATIENTS AND METHODS: Twenty patients with a diagnosis of low-grade NHL, stage III or IV, were included in this study. Patients received for one month the following treatment: cyclophosphamide, somatostatin, bromocriptin, retinoids, melatonin, and ACTH. The therapy was continued for two additional months in patients with stable or responding disease. After three months, the responding patients continued the therapy for three months and more. RESULTS: Twenty patients were assessable for toxicity and response; 70% (14 of 20 patients; 95% confidence interval [CI], 50% to 90%) had a partial response; 20% (4 of 20) had stable disease, and 10% (2 of 20) progressed on therapy. Going on with the treatment, none of the 14 patients with partial response had a disease progression (average follow-up time of 21 months, range, 7 to 25), and 50% of these patients had a complete response; among 4 patients with stable disease, 25% (1 of 4) had a partial response and 75% (3 of 4) progressed on therapy (mean time to progression [TTP] 14.3 months, range, 7 to 21). The toxicity was very mild, the most common side effects being drowsiness, diarrhea and hyperglycemia. CONCLUSIONS: The association of cyclophosphamide, somatostatin, bromocriptin, retinoids, melatonin, and ACTH is well tolerated and effective in treatment of low-grade NHL at advanced stage.

Pharmacol Ther. 1998 Aug;79(2):169-78.

Prolactin: the forgotten hormone of human breast cancer.

Vonderhaar BK.

Prolactin (PRL) is both a mitogen and a differentiating agent in the mammary gland. It has been shown to be involved in mammary cancer development in rodents, but in human breast cancer, its role has long been overlooked. Three criteria are applied to demonstrate PRL's involvement in this disease: (1) PRL receptors are present in human breast cancer cells, (2) human breast cancer cells in culture respond to PRL as a mitogen, and (3) PRL is synthesized by human breast cancer cells and inhibition of the binding of PRL to its receptors inhibits cell growth.

Endocr Relat Cancer. 1999 Sep;6(3):389-404.

Prolactin involvement in breast cancer.

Vonderhaar BK.

Normal development and differentiation of the mammary gland are profoundly influenced by prolactin (PRL). In rodent mammary cancer PRL plays a well defined role, but its role, in human breast cancer has not been appreciated until recently. It is now clear that breast tissue, both normal and malignant, is a significant source of extrapituitary PRL. Thus an autocrine/paracrine role of PRL in human breast cancer may be invoked. Both PRL and PRL receptor mRNA are expressed in the vast majority of breast cancer biopsies independent of estrogen and progesterone receptor status. An autocrine/paracrine PRL acting in human breast cancer requires that this hormone's action be blocked at the cellular level, as opposed to suppressing the synthesis and secretion of pituitary PRL. Mutants of PRL or human growth hormone are being explored which act as selective PRL antagonists. In addition, tamoxifen has been shown to act locally at the target tissue by binding directly to the PRL receptor and thus inhibiting PRL's action. These strategies may have clinical relevance in treating PRL-responsive human breast cancer.

Proc Natl Acad Sci U S A. 1995 Apr 11;92(8):3278-82.

Prolactin-immunoglobulin G complexes from human serum act as costimulatory ligands causing proliferation of malignant B lymphocytes.

Walker AM, Montgomery DW, Saraiya S, Ho TW, Garewal HS, Wilson J, Lorand L.

Several lines of evidence indicate that immunoglobulin-bound prolactin found in human serum is not a conventional complex between an anti-prolactin antibody and prolactin but a different type of association of prolactin with the Fab portion of IgG heavy chains. The complex of prolactin with IgG was purified from serum by anti-human prolactin affinity chromatography and was shown to contain close to 1 mole of N epsilon-(gamma-glutamyl)lysine crosslinks per mole of complex, a characteristic feature in structures crosslinked by transglutaminase. Interestingly, the complex caused a proliferation of cells from a subset of patients with chronic lymphocytic leukemia, while it was inactive in a cell proliferation prolactin bioassay. By contrast, human prolactin stimulated the proliferation of cells in the bioassay but had no effect on the complex-responsive cells from the patients. Competition studies with prolactin and free Fc fragment of IgG demonstrated a necessity for engaging both the prolactin and the immunoglobulin receptors for proliferation. More importantly, competition for the growth response by free prolactin and IgG suggests both possible reasons for the slow growth of this neoplasm as well as avenues for control of the disease.

Mol Cell Endocrinol. 1990 Jan 2;68(1):21-8.

Prolactin stimulates transcription of growth-related genes in Nb2 T lymphoma cells.

Yu-Lee LY.

The pituitary peptide hormone prolactin exerts a profound effect on various physiological processes involving both cellular proliferation and differentiation. The rat Nb2 T lymphoma cell line has been used as a model system for studying prolactin regulation of cell proliferation. Several genes associated with cell growth (c-myc, ornithine decarboxylase (ODC), heat shock protein 70 (hsp 70)-homologue, and beta-actin) are induced rapidly within 4 h after prolactin addition. Nuclear run-on transcription assays indicate that prolactin induction of these growth-related genes occurs primarily at the transcriptional level. According to the different kinetics of transcriptional response to prolactin, these growth-related genes can be divided into immediate-early (actin, c-myc), early (ODC) and mid-G1 (hsp 70-homologue) genes. Thus, prolactin may regulate Nb2 T cell-proliferative responses by modulating the transcriptional induction of various growth-related genes. These studies also represent a first report of a transcriptional cascade set off in rapid response to prolactin in cultured T cells.

Biochim Biophys Acta. 2000 Jun 2;1497(1):89-93.

Prolactin stimulation of tyrosyl phosphorylation of Shc proteins in Nb(2) lymphoma cells, but not mammary tissues.

Yu TX, Rillema JA.

Prolactin (PRL) stimulates lactogenesis in mammary cells and mitogenesis in a variety of cell types including Nb(2) cells. Studies indicate that a different composite of signaling pathways is involved in the PRL stimulation of mitogenesis as compared to lactogenesis. In the present studies, PRL is shown to stimulate the tyrosyl phosphorylation of all three isoforms of Shc proteins in Nb(2) cells (mitogenesis), but not in the mammary gland. Maximal phosphorylation of the Shc proteins is expressed between 10 and 15 min after a 50-ng/ml PRL treatment. In addition, there is an increased association between the Grb2 protein and Shc proteins upon PRL stimulation. However, no increased association between JAK2 and Shc proteins was observed in either the Nb(2) cells or mammary tissues.