MINIREVIEW

Molecular Actions of Prolactin in the Immune System (44111)

LI-YUAN YU-LEE[1]

Departments of Medicine, Cell Biology, and Microbiology and Immunology, Baylor College of Medicine, Houston, Texas 77030

Abstract. **The immunoregulatory properties of prolactin, a pituitary peptide hormone, have received renewed attention. The prolactin receptor, a member of the hematopoietin/cytokine receptor superfamily, is ubiquitously expressed by cells in the immune system. Certain subpopulations of lymphocytes synthesize and secrete biologically active prolactin, which suggests that prolactin can act as an autocrine and/or paracrine factor to modulate the activities of cells of the immune system. This review focuses on the molecular actions of prolactin in the immune system. Emphasis is given to recent information about the molecular mechanisms of prolactin receptor signal transduction, and the signaling molecules and prolactin-inducible target genes that participate in these responses. In particular, the prolactin-inducible interferon regulatory factor–1 gene and its roles in mediating diverse immune responses are highlighted.** [P.S.E.B.M. 1997, Vol 215]

Prolactin (PRL) is a peptide hormone that is synthesized and secreted primarily from the anterior pituitary. It belongs to a family of hormones that include growth hormone, placental lactogen, and proliferin (1, 2). In mammals, PRl regulates the growth and differentiation of several secretory glands such as the mammary gland (3), the ovary (4), and the male sex accessory organs (5). A role of PRL as a cytokine that modulates immune responses has received renewed and widespread attention, because of its synthesis and secretion from human and rodent lymphocytes (6–11), and the demonstration that its receptor (PRL-R), a member of the hematopoietin/cytokine receptor superfamily (12, 13), is expressed ubiquitously on hematopoietic cells. PRL may modulate the biology of immune function cells by regulating their potential for proliferation, differentiation, and/or apoptosis. These effects of PRL are accomplished in part by regulating the expression of a multifunctional transcription factor gene, interferon regulatory factor–1 (IRF-1). Understanding the various signals that regulate IRF-1 expression and potential IRF-1 functions in cells of the immune system should provide new insights into the immunoregulatory properties of PRL. Several excellent reviews have covered the potential role of PRL in immunity (14–21) and PRL regulation of gene expression in different target tissues (22–24). In this review, I will focus on the molecular aspects of PRL action in the immune system: (i) PRL and the immune response, including clinical, animal, and *in vitro* studies; (ii) lymphocyte PRL: (iii) lymphocyte PRL-R and signal transduction; and (iv) PRL-inducible IRF-1 gene and its potential functions.

PRL and Immune Response

Clinical Relevance. It is known that sex hormones such as estrogen contribute significantly to the patho-

[1] To whom requests for reprints should be addressed at Department of Medicine, Baylor College of Medicine, Houston, TX 77030.
This work received funding from the American Cancer Society (BE-49K), the National Institutes of Health (DK-44625), and the Linda and Ronald Finger Foundation for Lupus Research.

[P.S.E.B.M. 1997, Vol 215]

0037-9727/97/2151-0035$10.50/0

genesis of a number of autoimmune diseases, which are more common in women than in men (25). Estrogen is a prime regulator of both PRL and PRL-R expression (26). Recent clinical studies have focused on PRL and its possible involvement in immune dysfunction. PRL levels are higher in women than in men, and elevated PRL levels have been reported in patients with SLE (27, 28), multiple sclerosis (29), rheumatoid arthritis (30, 31), psoriatic arthritis (32), and AIDS (33), and in patients prior to transplant rejection (34). Furthermore, the bioactivity of serum PRL in some rheumatoid arthritis patients may be impaired (35). Bromocriptine (BRC), a dopamine agonist which inhibits PRL release from the pituitary, was shown to suppress autoimmune uveitis (36) and to correct T-cell and natural killer (NK)–cell abnormalities in patients with pathological hyperprolactinemia (37, 38). A reduction in the number $CD8^+$ T cells was observed in hyperprolactinemic patients, which suggests a susceptibility of this T-cytotoxic/suppressor-cell subpopulation to PRL (39). Though a clear causal relationship is still lacking, these clinical data suggest that elevated PRL levels are often found in patients with immunological diseases.

Animal Studies. A better correlation between PRL and immune regulation is observed in animal studies, where circulating PRL levels have been increased by administering exogenous PRL or decreased by either removal of the pituitary gland or by BRC treatment. Hypoprolactinemic animals were shown to be deficient in mounting various B and T cell–mediated immune responses (40, 41). Injections of PRL and other lactogenic hormones restored immunocompetence to these animals. In aged rats, implantation of PRL- and growth hormone (GH)–secreting pituitary tumor cells reconstituted thymic structure, increased $CD4^+$ thymocyte levels, and improved proliferative responses and interleukin-2 (IL-2) production in splenocytes (42, 43). A rise in PRL levels has been observed in rats with experimentally induced adjuvant arthritis (44) and encephalomyelitis (45), and lowering PRL with BRC treatment reduced disease symptoms. In the NZB/NZW mouse model of SLE, BRC was shown to delay mortality significantly, while PRL injection accelerated lupus-related death (46, 47). In DW/J dwarf mice, which lack PRL synthesis, PRL administration increased the number of circulating T cells and enhanced their antigen-specific proliferative responses (48). In Snell dwarf mice, PRL enhanced concanavalin (Con A)–stimulated IL-2 receptor expression on both the $CD4^+$ and the $CD8^+$ T-cell subsets in the popliteal lymph nodes (49). Finally, studies in athymic nude mice suggest that PRL influences thymic T cell development. Grafting of thymus (to provide thymocytes) and pituitary (to provide high levels of PRL) in nude mice decreased the number of $CD4^+CD8^+$ double-positive thymocytes, but increased $CD4^+$ T lymphocytes in the periphery (50). This suggests that PRL may enhance the development and exit of T lymphocytes from the thymus and influence functions of mature peripheral T cells. Whether PRL exerts immunostimulatory, inhibitory, or no effect on immune targets may be influenced by the dose of PRL employed (stimulatory at low but inhibitory at supraphysiological concentrations) (51–53) as well as by daily circadian rhythms in PRL secretion from the pituitary (54). Together, these animal studies support a role of PRL in promoting the growth and differentiation of immature lymphocytes as well as in stimulating mature lymphocyte functions.

***In Vitro* Studies.** A third line of studies to support a role of PRL in modulating immune responses comes from analysis of hematopoietic cells, including T cells, B cells, hematopoietic precursors, macrophages, and NK cells. By lowering serum PRL levels with BRC treatment in rats and mice, several studies have shown a decrease in T lymphocyte responsiveness to mitogens *in vitro* (55), in mixed lymphocyte reactions (56), in graft-versus-host reactions *in vivo* (56), and in macrophage activation, presumably by suppressing γ-interferon (IFN-γ) production by T lymphocytes (55). However, BRC may exert direct effects on lymphocytes, suppressing T and B lymphocyte activities (57) but inducing $CD8^+$ T suppressor cell functions (58). Another approach has been to add exogenous PRL to cells in culture, which demonstrated that PRL can act as a comitogen for Con A–stimulated splenocytes (59, 60), for thymocytes (6, 60), for peripheral blood lymphocytes (11, 49, 61), for IL-2–stimulated L2 T cells (62), and for antigen-specific proliferation of peripheral T cells (48), or as a direct mitogen for the rat Nb2 T cells (63, 64). These immunostimulatory effects of PRL may be due to its ability to directly induce the expression of growth-related genes (16, 63, 65) (see section entitled "PRL-Inducible Interferon Regulatory Factor–1 below) or indirectly by upregulating the expression of cytokine receptors, such as IL-2 receptor (IL-2R) on rat splenocytes (66), and mouse $CD4^+$ and $CD8^+$ T cells (67), erythropoietin receptor (EPO-R) expression on $CD34^+$ hematopoietic precursor cells (53), and its own PRL-R receptors in T cells (68, 69) and peripheral blood mononuclear cells (PBMC) (70). Recent studies have also shown that PRL inhibits glucocorticoid-induced apoptosis in T cells (71, 72). Further, PRL has also been suggested to enhance mature B-cell functions by increasing IgG and IgM antibody production (46, 73). Hence, the biological effects of PRL are determined by cell lineage and their stage of differentiation, and involve various aspects of lymphocyte activation, proliferation, differentiation, effector functions, and apoptosis.

Lymphocyte PRL

Understanding PRL action in the immune response has been complicated by the demonstration that lym-

phocytes themselves (6–11, 51, 74, 75) and other extrapituitary tissues (76) can synthesize and secrete biologically active PRL and PRL-like molecules. Blocking experiments that employed anti-PRL antibodies suggested that a PRL-like molecule is secreted by lymphocytes upon mitogenic stimulation and plays a critical role in T-cell proliferative responses (10). Endogenous PRL synthesis by lymphocytes may explain the inability of some studies to demonstrate any effect of exogenously added PRL on proliferation in lymphocyte cultures (10). Some controversy exists as to the exact molecular size (ranging from 22 to 60 kDa) and posttranslational modifications (glycosylation and phosphorylation) of lymphocyte PRL, and whether PRL from the serum of culture media can be "sequestered" and subsequently utilized by T cells (62, 77). A recent study also indicated that the PRL-like molecule secreted by mitogen-stimulated murine splenocytes may not be immunologically similar to pituitary PRL (78). Nevertheless, PRL has been cloned from human thymocytes (7) and PBL (8, 11). Interestingly, an alternative 5′ exon situated 6 kb upstream of the pituitary PRL exon I is used to express PRL in lymphoid and other nonpituitary cells (79, 80). The signals that regulate expression of the nonpituitary PRL RNA transcripts are different from those regulating the pituitary counterpart (80, 81) and remain to be further defined. It is not known which lymphocyte subsets are expressing PRL, nor is it clear how pituitary, extrapituitary, and lymphocyte PRL act in concert to modulate immune cell biology.

Lymphocyte PRL-R: A Member of the Hematopoietin/Cytokine Receptor Superfamily

PRL-R on Lympho-Hematopoietic Cells. PRL action on hematopoietic cells is presumably mediated by PRL-Rs, which have been demonstrated at both the mRNA (69, 82–84) and the cell surface protein levels (67, 68, 70, 85) in human, rat, and mouse hematopoietic cells. With the use of monoclonal or polyclonal antibodies directed against the external domain of the PRL-R (86), high-intensity PRL-R staining (PRL-R^{hi}) was observed in the immature cells in the thymus and bone marrow (68, 70), whereas the more mature cells in the periphery (PBL) and the secondary lymphoid organs (spleen, lymph nodes) showed low to medium PRL-R (PRL-R^{med}) staining (67, 85). Unexpectedly, over 90% of bone marrow precursors were PRL-R^{hi} (67, 68, 70), which suggests a potential PRL action on hematopoietic precursor cells. Interestingly, the bone marrow stromal cells, which provide the necessary microenvironment (cytokines and adhesion molecules) for hematopoietic cell differentiation (87), also express the PRL-R during lineage-specific differentiation (88). Within thymocyte subses, PRL-R^{hi} to PRL-R^{med} staining is found in this order: double negative (recently arrived from the PRL-R^{hi} bone marrow) > double positive > single positive, with $CD8^+ > CD4^+$ thymocytes in the mouse (68), and $CD4^+ > CD8^+$ thymocytes in humans (70). In the periphery, about 20% each of $CD4^+$ and $CD8^+$ T cells show PRL-R^{med} staining, while B220 B cells and macrophage are constitutively PRL-R^{hi}. In mature T cells, PRL-R levels can be induced by PRL (69), IL-2 (51, 89), and Con A (49, 68). Thus, lymphocyte PRL-R levels can be modulated by cytokines. These combined studies provide a broad basis for addressing the functional significance of PRL-R expression on various lineage as well as developmental stages of hematopoietic and stromal precursor cells.

PRL-R Structure/Function. PRL-R belongs to the hematopoietin/cytokine receptor superfamily, which now includes more than 20 members (12, 13, 16, 90). Other members include the receptors for IL-2β, IL-2γ, IL-3 to IL-7, IL-9, IL-12, IL-15, G-CSF, GM-CSF, EPO, GH, gp130, LIF, OSM, CNTF, MPL, and the more distantly related receptors for IFN-α/β and IFN-γ (13, 91). These receptors are broadly grouped by function, as they are all involved in the growth and differentiation of lympho-hematopoietic lineage cells, and by conserved extra- and intracellular structural motifs (13, 90–92). Three major PRL-R forms have been identified (93–95), which share identical extracellular ligand binding domain but differ in the length and sequence of the intracellular domains, suggesting potential differences in signaling capacities. The short (42-kDa) and long (80-kDa) PRL-Rs result from alternative splicing of 3′ end exons from a single gene (94, 96, 97), whereas the Nb2 T cell PRL-R (65 kDa) contains an in-frame deletion of a 198–amino acid cytoplasmic domain relative to the long PRL-R (97). Interestingly, a PRL-R containing a truncation near the site of the rat Nb2 PRL-R deletion has been cloned from human breast carcinoma tissues (98), but the functional activity of this PRL-R form is unknown. Although the long PRL-R predominates in lymphoid tissues (83, 99), the Nb2 T cell PRL-R shows the highest affinity for PRL (K_d of 3 nM) (96), followed by the long and short PRL-R (93). Whether the deletion in the Nb2 PRL-R is a basis for its greater mitogenic potential is not known. Only the Nb2 and long PRL-Rs mediate mitogenic activities in hematopoietic cells, whereas the short PRL-R is inactive (95, 100, 101). Indeed, transfected short/Nb2 PRL-R heterodimers are incapable of signaling for proliferation (101), which has led to the suggestion that the short PRL-R may serve to modify the signaling capacities of the long PRL-R complex (95, 101).

PRL-R Signal Transduction. Until recently, how cytokine receptors transduce signals had been a puzzle because these receptors do not possess intrinsic kinase or catalytic domains in their intracellular domains, and classical second messengers do not mimic their actions (93). Cytokine receptors signal *via* a group of receptor-associated protein tyrosine kinases (PTK), including

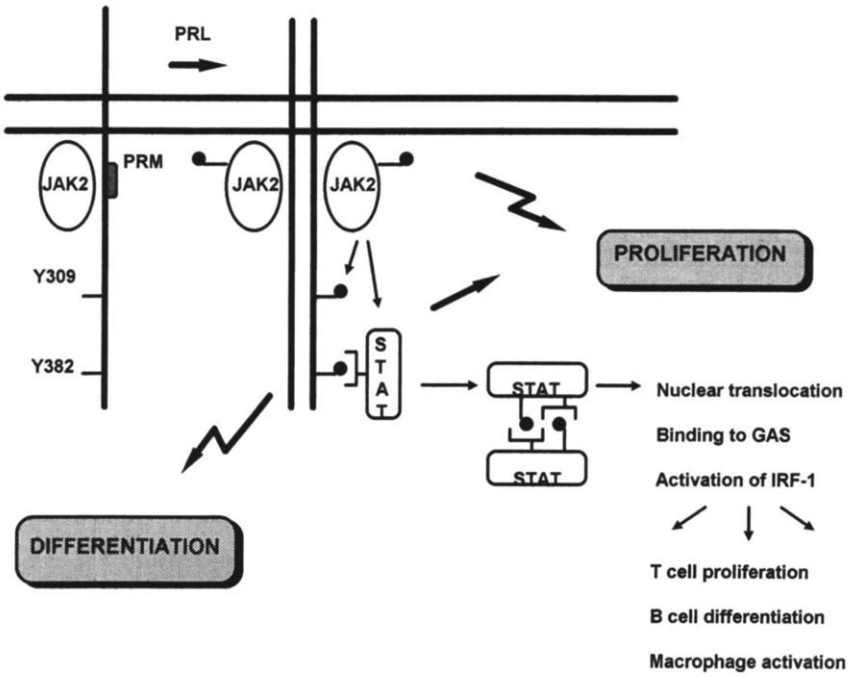

Figure 1. PRL-R signaling through the JAK/Stat pathway. See text for discussion.

Tyk2 (102), JAK1, JAK2 (103, 104), and JAK3 (105, 106), which are either bound to the unliganded receptor (107–110) or recruited (111) to the receptor upon ligand activation. Various combinations of activated JAK/Tyk2 then phosphorylate preexisting but latent cytoplasmic transcription factors, collectively called signal transducer and activator of transcription, or Stat (112) (see section of that name and Fig. 2 below). Tyrosine phosphorylated Stats translocate into the nucleus, bind to cognate DNA sequences, and activate different sets of target genes. This "JAK/Stat pathway" (112, 113) is the prototype signaling pathway used by all hematopoietin/cytokine receptors. In the case of the PRL-R, PRL binding to the PRL-R leads to receptor dimerization or oligomerization (114), which in turn leads to the activation of PRL-R–associated JAK2 kinases (109, 115, 116) (Fig. 1). Activated JAK2 then phosphorylates downstream targets, one of which is the PRL-R intracytoplasmic domain (ICD) itself (115–118) and another, the Stat proteins. Activated Stats form homo- or heterodimers, translocate into the nucleus, bind conserved DNA elements, and turn on target genes. Different parts of the PRL-R ICD regions are involved in signal transduction, including the proline-rich motif (PRM) or Box 1 (90, 117) and several tyrosine residues which become phosphorylated upon JAK2 activation of the PRL-R (118).

Proline rich motif or Box 1. An 8–amino acid (aa) proline-rich motif sequence, also called Box 1, is found close to the transmembrane domain not only in the PRL-R ICD (90) but also in the majority of the hematopoietic/cytokine receptors. The PRM sequence, aliphatic-aromatic-proline-X-aliphatic-proline-X-proline, is critical for receptor signaling (92, 117, 119–124), as it mediates interactions between the receptors and the tyrosine kinase JAK2 (100, 111, 124–127). Although this relationship has been determined by mutagenesis and transfection analyses, it is still unclear how JAK2 physically interacts with the PRM region of the PRL-R (100). Interestingly, a peptide corresponding to the 8 aa PRM of the PRL-R was found to exist as a *cis-trans* conformer, leading to the speculation that isomerization of the proline residues within the PRM by a peptidyl prolyl *cis-trans* isomerase or immunophilin (127a, 127b) may play a role in PRL-R structure/function (127c). This hypothesis remains to be tested. However, the PRM, though critical, is not sufficient for signaling (100, 117, 118, 128). Other regions including Box 2 (100, 128) in the PRL-R, Box 3 (120) and other motifs (129) in other cytokine receptors, as well as tyrosine residues, are also important for receptor function and interactions with signaling molecules, as described below.

Tyrosines residues in the PRL-R. Mutational analyses have shown that multiple tyrosines in the carboxy terminus of the long PRL-R ICD are important for mediating differentiated functions (118). In the context of the Nb2 PRL-R, only the last tyrosine residue, Y382, appears to be critical for mediating differentiation-specific functions. However, studies from our laboratory with the Nb2 PRL-R suggest that both Y309 and Y382 are needed for mediating a proliferative response (Y-F Wang, KD O'Neal, L-Y Yu-Lee, unpublished observation). One function for phosphorylated receptor tyrosine residues is to act as "docking sites" for the recruitment of Stat factors (130–137), as an intermediary step in signal transduction (Fig. 1). Specific se-

quences around these tyrosine residues provide some specificity as to which Stat factors are recruited (134). Although several Stat factors are activated upon PRL stimulation of T cells (see section entitled "PRL-Inducible Stats" below), it is not clear whether they are recruited by "docking" at tyrosine residues on the PRL-R and/or on JAK2 (134, 138). Recent studies of the GH-R indicate that phosphorylated tyrosine residues in the GH-R ICD may not be needed for selected GH-mediated functions (139, 140). This suggests that tyrosine residues on the JAK kinase may be involved in recruiting and activating Stat factors in the GH-R signaling pathway (138). In general, membrane proximal regions of cytokine receptors are involved in mediating proliferative responses (100, 121, 129, 141–145), while the membrane distal regions are important for mediating differentiated functions (117, 118, 123, 126, 128). Interestingly, data from our laboratory indicate that the PRL-R utilizes the PRM as well as both Y309 and Y382 residues, that is, both membrane proximal and distal regions, for signaling to a growth-related gene promoter, interferon regulatory factor–1 (IRF-1) (Y-F Wang, KD O'Neal, L-Y Yu-Lee, unpublished observation) (see section entitled "PRL-Inducible Interferon Regulatory Factor–1" below).

JAK Tyrosine Kinases and PTP Tyrosine Phosphatases. *Kinases.* Cytokine receptor activation is mediated by the activation of JAK PTKs. Up to five tyrosine kinases have been identified (104), including Tyk2, JAK1, JAK2, and JAK3, which is primarily found in hematopoietic cells (106, 146), and a Drosophila homolog *hopscotch* (147). These PTKs are either pre-bound (EPO-R, PRL-R, gp130) or recruited (IFN-α/β–R, IFN-γ–R, GH-R) to the cytokine receptors upon ligand binding (104). The kinases are about 120–140 kDa in size and contain an inactive kinase-like domain followed by a carboxy kinase domain (103, 148–150). Mutagenesis and *in vitro* binding analyses have shown that the amino-terminal portion of JAK2 is involved in binding to cytokine receptors (127, 151), while the carboxy domain is critical for JAK kinase activity (148, 151, 152). However, since the JAK kinases do not possess *src* homology (SH) SH2 or SH3 domains, two protein/protein interaction motifs widely used in signaling molecules (153), it is not clear how these kinases physically associate with either cytokine receptors or downstream substrate proteins. In PRL-R signaling, JAK2 phosphorylation and activation are associated with cell proliferation in T cells (109, 115, 116), and dominant negative mutants of JAK2 inhibit PRL-R signal transduction (152).

Phosphatases. Rapid and transient activation of JAK kinases is followed by downregulation of kinase activity and receptor signaling. Receptor downregulation involves the activities of at least two types of protein tyrosine phosphatases, PTP1C, which is found primarily in hematopoietic cells (154), and PTP1D, which is ubiquitous (155). These PTPases contain two SH2 domains (156), which are involved in binding to the phosphorylated tyrosines on receptor ICDs. Interestingly, PTPases are themselves activated by tyrosine phosphorylation, and upon activation they can dephosphorylate JAKs and/or the receptor itself to downregulate the signaling process (134, 154, 157). Different PTPases appear to inactivate different JAK kinases (158, 159). However, not all receptor-associated PTPases are involved in downregulation of signal transduction. A complex of PRL-R/JAK2/PTP1D appears to be necessary for initiating PRL-R signaling (160). Studies employing genetic mutants that are defective in the expression of JAKs (102, 112) as well as PTPases (154, 158) will help to determine the roles that these enzymes play in cytokine receptor signaling.

Signal Transducer and Activator of Transcription. In addition to receptor ICDs, another target of JAK kinase action is the small family of latent, cytoplasmic transcription factors collectively called Stat (113, 161–163). Seven distinct mammalian Stats have been identified, including Stat1α/β, Stat2, Stat3α/β, Stat4, and Stat5a, Stat5b, and Stat6, and a Drosophila D-Stat, which is most like Stat5 (164, 165). Stat proteins are about 800 aa in size and range from 89 to 97 kDa. Several Stats also contain carboxy-terminal truncations as a result of alternative splicing of 3′ sequences (166), which generates dominant negative β isoform variants (167–172) (Fig. 2). Stat proteins contain the following domains: (i) a helical domain in the amino terminus which appears to be involved in protein/protein interactions (174), (ii) a helix-β pleated sheet-helix domain important for DNA binding (136, 175), (iii) an SH2 domain which is critical for Stat/Stat and Stat/receptor interactions through phosphorylated tyrosines (168, 176), (iv) a critical tyrosine residue that is important for

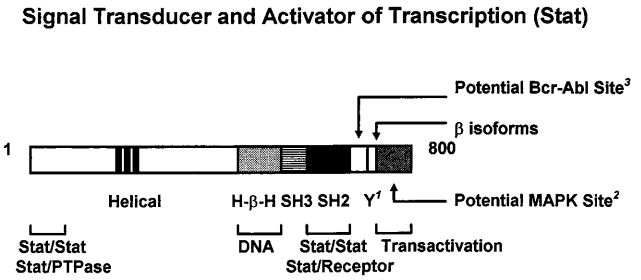

Figure 2. Structure conserved motifs in the Stats are shown, including helical region, a helix-β pleated sheet-helix region, an SH3-like domain that may not be functional, an SH2 domain, a critical tyrosine (Y) residue, and a carboxy-terminal transactivation domain. The β isoform of Stat, which lacks the carboxy-terminal transactivation domain, functions as a dominant negative mutant. See text for discussion. [1]Critical tyrosines in the Stat proteins are: Stat1, Y701; Stat2, Y690; Stat3, Y705; Stat4, Y694; Stat5a/b, Y699; and Stat6, Y641. [2]Potential MAPK consensus site, XPXSP, is observed in Stat1 and Stat3 (173). [3]Potential Bcr-Abl phosphorylation site, DEVYSKYYT, is observed in Stat5b (24).

Stat/Stat intermolecular interactions, nuclear translocation, and DNA binding (177, 178), and (v) a transactivation domain at the carboxy terminus (173, 178). The DNA binding, SH2, and carboxy transactivation domains function as modules, as demonstrated by studies with Stat1/Stat2 (176), Stat1/Stat3 (175), and Stat1/Stat6 (136) chimeric proteins. Additionally, Stat1 and Stat3 contain a potential MAPK phosphorylation site in the carboxy domain, and serine phosphorylation is thought to enhance Stat interaction with and transcriptional activation of target gene promoters (173, 179–181). Moreover, Stat5b contains a potential Bcr-Abl phosphorylation site (24) which may be targets of modification in certain leukemias (182, 183). Recent studies also showed that amino terminus of Stat proteins is also critical for dephosphorylation by PTPase (184). It is thought that the PTPases involved in dephosphorylating nuclear Stats are distinct from those dephosphorylating receptor and JAKs in the cytoplasm (158, 159, 185–187).

Stat binding sites. Two types of conserved DNA elements are targets for Stat factor binding. These sequences were determined initially from analyses of promoters of IFN-α/β–inducible genes (112, 113, 161, 163). The IFN-stimulated response element (ISRE) is comprised of TTTC NN TTTC (direct TTTC repeats) and binds Stat complexes containing Stat1, Stat2, and a non-Stat protein, p48, which stabilizes the Stat interactions at the ISRE (188, 189) (Fig. 2). Another conserved element determined from IFN-γ–inducible genes, called IFN-activation sequence (GAS), is comprised of TTTC NNN GAAA (inverted TTTC repeats or palindrome) and binds a host of Stat factors in response to numerous cytokines (162, 190). In addition, the spacing of the palindromic half sites within the GAS element affects which Stat complexes will preferentially interact at the GAS (163, 191). A GAS palindrome with 3-bp spacing has been shown to interact with Stat1, Stat3, Stat4, and Stat5, while that with 4 bp interacts preferentially with Stat6. Interestingly, in addition to Stat homodimers, Stat1/Stat2 (192) and Stat1/Stat3 (173, 179, 193, 194) heterodimers have been observed to bind at GAS elements and promote gene transcription (192). Further, non-Stat DNA binding factors, such as p48 (189), Sp1 (195), c-Jun (174), v-Src (196), and the glucocorticoid receptor (GR) (197), can interact with Stat complexes to modulate gene transcription. Recent studies of native tandem GAS-like sequences in the IFN-γ gene showed that Stats can interact cooperatively to form tetramers through amino-terminal domain interactions (198).

PRL-inducible Stats. Three PRL-inducible Stats have been described, and they differ in their relative abundance and functional importance depending on the target gene and cell type. The first PRL-inducible Stat was cloned by its ability to bind to a GAS-like element in the promoter region of a mammary gland milk protein gene, β-casein, and hence it was initially called mammary gland factor (MGF) (199–201). MGF, now known as Stat5a, has been shown to interact with GAS and GAS-like sequences in the promoter regions of several other PRL-inducible genes, including IRF-1 (202–205), β-lactoglobulin (206, 207), whey acidic protein (208), and α_2-macroglobulin (209). Indeed, Stat5a and Stat5b are two of the most widely used Stats in cytokine receptor signaling. A second and equally prominent Stat, Stat1, has also been shown to bind to the IRF-1 GAS in a PRL-inducible manner in hematopoietic cells (202, 203, 205, 210, 211) (see section entitled "Stats at the IRF-1 GAS" below). A third Stat, Stat3, has been shown by immunoblotting to become tyrosine phosphorylated upon PRL stimulation of premyeloid (212) and T cells (data not shown), but the molecular targets of PRL-inducible Stat3 activation have yet to be identified. Our transfection studies addressing the functional significance of Stat1 and Stat5 interactions at the IRF-1 promoter have shown that Stat1 is a positive regulator of IRF-1 gene expression, whereas, surprisingly, Stat5a and Stat5b both inhibit PRL-inducible IRF-1 promoter activity (G Luo, L-Y Yu-Lee, unpublished observations). The specific effects of PRL-inducible Stat complexes at either the IRF-1 or the β-casein gene promoters are influenced by the presence of other factors interacting at nearby sites (213–215), which provides another level of specficity of JAK/Stat signal transduction. Although Stat1, Stat3, and Stat5 are all activated by PRL stimulation, no Stat1/Stat5 (135, 216, 217) or Stat3/Stat5 (217) heterodimers have been convincingly observed in the same complex. PRL-inducible Stat5a/Stat5b heterodimer formation has been described (218).

Other Potential Signaling Molecules. In addition to the JAK/Stat signaling pathway, other PTKs such as Fyn (219), Raf (220), and Src (221), as well as serine/threonine kinases MAPK (222) and protein kinase C (77), have all been suggested to play a role in PRL-R signaling. How these parallel kinase cascades elicit coordinated gene transcription in PRL-stimulated target cells is currently still unclear (223). Since some of these molecules are pre-associated with the inactive monomeric PRL-R (219, 220), the possibility exists that other PRL-R signaling proteins may be interacting at the PRL-R and can therefore be cloned by genetic means. Using the PRL-R ICD as a bait, our laboratory has cloned 40 potential PRL-R interacting proteins by the yeast two-hybrid interacting cloning strategy (224). The significance of these proteins in PRL-R signal transduction is under investigation. The pleiotropic actions of PRL on cellular proliferation, differentiation, or apoptosis will be determined by the activation and interactions of these potential signaling proteins and parallel kinase cascades.

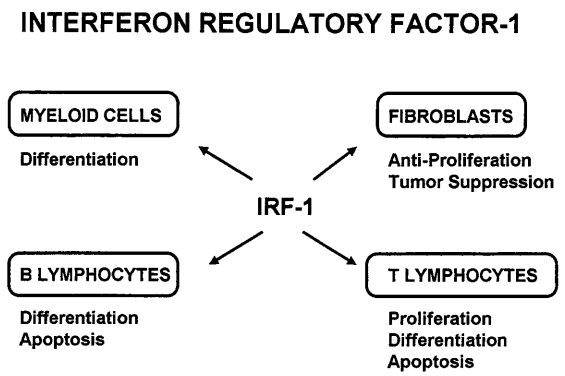

Figure 3. Potential IRF-1 functions. See text for discussion.

PRL-Inducible Interferon Regulatory Factor–1

Several years ago, our laboratory cloned a panel of novel PRL-inducible genes in activated T cells as one way of studying PRL-inducible proliferative responses in T lymphocytes. A PRL-sensitive T lymphoma cell line, Nb2, which was derived from the lymph node of an estrogen-treated male rat (64), was employed. The Nb2 T cells express a high number of high-affinity PRL-R (225), and can be synchronized in lactogen-deficient culture medium and restimulated to grow with 1–10 ng/ml PRL. A cDNA library from PRL-stimulated Nb2 T cells was generated and 26 PRL-responsive gene sequences were isolated by differential screening (65). The most PRL-inducible gene encoded a transcription factor called IRF-1. IRF-1 is currently used not only as a target gene for understanding PRL regulation of gene transcription, but also as a multifunctional factor for understanding how PRL modulates various immune responses.

A Multifunctional Transcription Factor. IRF-1 belongs to a small family of transcription factors, which includes IRF-2 (226), IFN consensus sequence binding protein (ICSBP) (227), IFN-α stimulated gene factor 3γ (ISGF3γ) (228), PU.1 interacting protein (PIP) (229), and IRF-3 (230). These factors act as either transcriptional activators or repressors. IRF-1 actions are antagonized by IRF-2 (226) and ICSBP (231), which have similar DNA binding but different transactivation domains. IRF-1 is inducible by PRL and IL-2 in rat Nb2 (65, 232) and murine L2 T cells (233), and by Con A in murine splenocytes (232), which suggests a role in T-cell activation. Originally cloned as a factor involved in IFN-β gene expression (234), IRF-1 turns out to be a more versatile factor which can be induced in a wide variety of tissues (65) in response to cytokine stimulation and host defense against viral infection (234–240). Thus, IRF-1 mediates diverse functions including tumor suppression (241, 242), myeloid differentiation (239), macrophage activation (243, 244), antigen presentation (245, 246), T- and B-cell differentiation (247, 248), and apoptosis (249–251) (Fig. 3) (see Table I below). A role for IRF-1 in hematopoietic cell differentiation, maturation, and function has been demonstrated by transgenic studies. Animals carrying a disrupted IRF-1 gene (IRF-$1^{-/-}$) show a 90% reduction in $CD8^+$ single-positive T lymphocytes within the thymus (248, 252), severely impaired macrophage functions (243, 252), and an inability to clear certain viruses (237, 248, 253). Further, overexpressing IRF-1 under a heavy chain μ enhancer resulted in a block in bone marrow precursor B-cell differentiation and an absence of mature B cells in the periphery (247). Thus, the transgenic studies showed that IRF-1, or an appropritate level of this transcription factor, is critical for T- and B-cell differentiation and maturation processes as well as macrophage functions. Recent studies have also revealed that IRF family members are expressed in activated T cells and in B cells of all developmental stages (254). Together, these studies agree with our cell culture studies, which show that IRF-1 modulates the activation (65) and proliferation of T cells (65, 255).

Signals That Regulate IRF-1. IRF-1 gene expression is regulated by many signals which are dependent on the particular cell type examined. In fibroblasts, IRF-1 expression is stimulated by virus, IFN, TNF, IL-1, and activators of the protein kinase C pathway such as phorbol esters and calcium ionophore (236, 238). In embryo carcinoma cells, IRF-1 is regulated by developmental cues (256). In myeloid lineage cells, LIF and IL-6 inhibit IRF-1 expression as part of the differentiation pathway (239, 240). In myeloid leukemia cells, IRF-1 is induced by all-*trans* retinoic acid, as part of the growth inhibition pathway (257). Studies from our laboratory show that IRF-1 mRNA is found in various normal proliferating and post-proliferating tissues (65). These combined data support the view that IRF-1 is a multifunctional transcription factor which can respond to a variety of external signals in different tissues and cell types (Fig. 3). In T cells, IRF-1 expression is exquisitely sensitive to PRL (65), IL-2, and Con A (232) stimulation, and is correlated with T-cell activation (G1 phase), cell cycle progression (S phase), and cell proliferation (233, 254, 255, 258).

Biphasic IRF-1 Gene Transcription. In most cell types, IRF-1 induction by cytokine stimulation is rapid and transient. In contrast, IRF-1 is induced twice in a single cell cycle in PRL-stimulated Nb2 T cells. In quiescent and synchronized Nb2 T cells, PRL stimulates IRF-1 gene transcription first at about 30–60 min during early G1, then again at 10–12 hr in early S phase (255, 258). This biphasic transcriptional response is mediated by multiple PRL responsive regions in the IRF-1 promoter and upstream 5′-flanking DNA (211, 258). By transfection analyses, a 1.7-kb IRF-1 5′-flanking DNA was shown to contain elements that promote not only G1- but also S-phase induction of IRF-1 gene transcription, while a 0.2-kb IRF-1 promoter mediates primarily

G1 induction (211). Within this minimal 0.2-kb promoter is a single copy of a GAS element (−123/−113), which can act as a PRL responsive enhancer element (211). Nearby 5′ to the GAS element are additional sites that interact with DNA binding proteins such as Sp1, a constitutive transcription factor (259), and other complexes containing cell cycle regulated proteins, such as Rb, cyclins, and CDKs (260, 261) (data not shown). The integration of cell cycle–regulated proteins and cytokine-regulated Stat factors at the IRF-1 promoter is likely to coordinate the biphasic expression of the IRF-1 gene over the cell cycle in PRL-stimulated Nb2 T cells.

Stats at the IRF-1 GAS. Gel shift analysis has been extensively used to examine which Stat proteins interact at the IRF-1 GAS in a PRL-inducible manner. Thus, Stat1 (202, 205, 210, 211) and Stat5 (203–205) have been detected at the IRF-1 GAS in a PRL-inducible manner in Nb2 T cells (see also section entitled "PRL-Inducible Stats" above). However, this approach is limited, as it does not address the functional significance of Stat interaction at the IRF-1 GAS. Transfection studies employing a PRL-R reconstituted COS cell system showed that Stat1 is a positive regulator of IRF-1 promoter activity, but both Stat5a and Stat5b inhibit PRL-inducible IRF-1 gene expression (G Luo, L-Y Yu-Lee, unpublished observations). Studies are underway to determine whether the different Stat proteins are competing for binding to the IRF-1 GAS element, or whether Stat5 may be "squelching" (262) or competing for factors interacting with Stat1 which are needed for transcriptional activation of the IRF-1 promoter. In this regard, it is possible that how Stat1, versus Stat5, interacts with nearby factors, such as Sp1 (195, 259), Sp1-associated TAF proteins (263), p300/CBP co-activators (263a, 263b, 263c), and cell cycle–regulated proteins (264), may explain their functional antagonism at the IRF-1 promoter. Understanding the kinetics of Stat1 and Stat5 translocation into the nucleus, their relative binding affinities, and their on/off rates at the IRF-1 GAS will elucidate how these different Stats are involved in the rapid up- and downregulation of IRF-1 gene transcription in response to PRL stimulation over the cell cycle. Other molecules, such as PTPases, may be involved in dephosphorylating Stats, JAKs, and the PRL-R to reset the system and to shut down IRF-1 transcription during G1 and early S phase of the cell cycle. Such tight regulation of the expression of a potent transcription factor is consistent with IRF-1's playing an important role in hematopoietic cell biology.

IRF-1 Target Genes. Understanding how IRF-1 modulates T-cell, B-cell, and macrophage biology is aided by studies of IRF-1 target genes in these cell types. Functional IRF-1 binding sites (IRE) with the consensus sequence GAAAGT (234, 265, 266) have been found in the promoter of numerous genes (Table I). These include the oncogene IRF-2 (281); genes involved in immune responses such as IFN-β (234), IL-6 (275), IL-7Rα (267), MHC class I (245, 246, 268, 284), transporter associated with peptide processing (TAP1) (270), LMP2 (270), inducible nitric oxide synthase (iNOS) (243, 244), 2′,5′-oligoadenylate synthetase (OAS) (244, 271), and double-stranded RNA-dependent protein kinase (PKR) (272–274); genes involved in cell growth such as ornithine decarboxylase (ODC) (276), angiotensin II Type 2 receptor (277), and GTPase guanylate binding protein (GBP) (278, 279); cell adhesion molecules V-CAM (283) and E-cadherin (265); and an IL-6–inducible, differentiation-specific gene MyD88 (280). In addition, IRF-1 has also been suggested to regulate the expression of cell cycle genes, such as the cyclin-dependent kinase inhibitor CKI or p21 (251) and the tumor suppressor gene p53 (282), cytokine genes IL-4, IL-5 (265), and IL-6 (275), and the apoptosis-related gene IL-1–converting enzyme (ICE) (250). Judging from the complex array and nature of these target genes, IRF-1 is indeed a multifunctional transcription factor which is involved in mediating diverse functions, ranging from immune responses, cell growth, and differentiation to apoptosis. With its potent transcriptional capabilities, it is not surprising that the IRF-1 protein has a short half-life, from 25 min during G1 phase to slightly over 60 min during S phase in T cells (255). Often, IRF-1 works in concert with Stats (270, 279, 280), other IRF members (231), and other DNA binding proteins (275, 285) in mediating

Table I. Potential IRF-1 Functions and Target Genes

Functions	Target Genes	References
Immune responses	IFNβ	234
	IL-7Rα	267
	MHC Class I	245, 268, 269
	TAP1	270
	LMP2	270
	iNOS	243, 244
	OAS	244, 271
	PKR	272–274
Cytokines	IL-4[a]	265
	IL-5[a]	265
	IL-6	275
Cell growth	ODC	276
	Angiotensin Type II receptor	277
	GTPase guanylate BP	278, 279
Cell cycle	CKI or P21[a]	251
Cell differentiation	MyD88	280
Oncogenesis/tumor suppression	IRF-2	281
	p53[a]	282
Apoptosis	ICE[a]	250
Cell adhesion	V-CAM	283
	E-cadherin[a]	265

Note. Functional IRF-1 binding site or IRF responsive element (IRE) has been demonstrated in these genes, whose expression contribute to diverse functions attributed to IRF-1.
[a] IRE not defined yet.

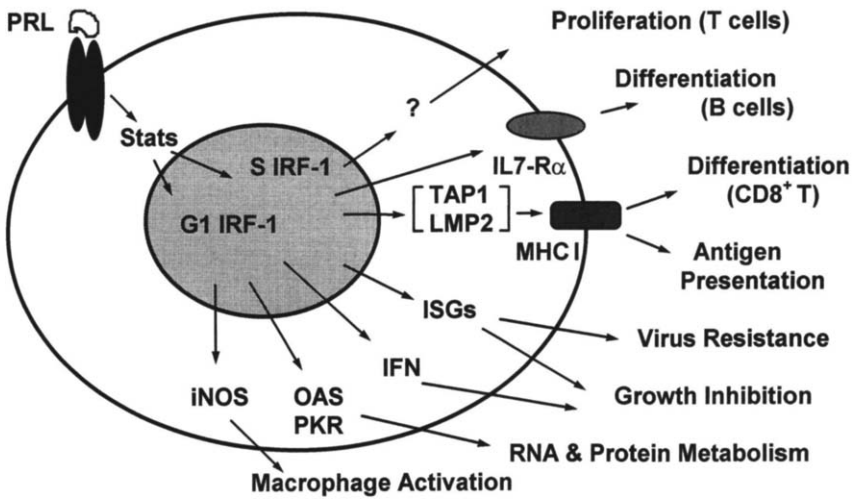

Figure 4. PRL and immune regulation through the transcription factor IRF-1. This cartoon represents a composite look at potential PRL actions in the immune system, through its regulation of the multifunctional IRF-1 gene. IRF-1 functions and IRF-1 target genes (see Table I) are determined in virus-infected fibroblasts (237, 248, 252, 253), cytokine-stimulated T cells (65, 232), B cells (267), and macrophages (243, 244), using cell lines as well as wild-type or IRF-$1^{-/-}$ animals.

rapid as well as sustained or delayed responses at these promoters. The recent demonstration that IRF-1 can interact directly with members of the Rel transcription factor family (246, 268, 283), a process known as cross-coupling (286, 287), broadens the transcriptional potentials of IRF-1 and vastly increases its pool of target genes.

Potential IRF-1 Functions. In fibroblasts, IRF-1 has been shown to be a tumor suppressor gene, involved in antioncogenic activities, suppression of oncogene-induced transformation (249, 288) and apoptosis (249, 250). However, IRF-1's functions in hematopoietic lineage cells are quite different (Fig. 4), as illustrated by studies with IRF-1 knock-out animals. In the IRF-$1^{-/-}$ mice, the three most remarkable phenotypes (i.e., reduction in $CD8^+$ thymocytes, impaired macrophage function, and weakened antiviral responses) may be explained in part by the loss of IRF-1 target gene expression in the affected tissues and by a general dampening of the IFNγ response pathway (253). The lack of $CD8^+$ thymocytes may be related to a loss of IRF-1–inducible IL-7Rα expression in the developing thymocytes (289, 290), resulting in an inability of $CD4^+CD8^+$ double-positive cells to switch to the IL-$7R\alpha^+CD8^+$ single-positive cells. Although no gross alterations in MHC class I were detected (248), MHC I complex stability and/or function may nevertheless be compromised, a problem compounded by the lack of TAPI expression, resulting in a block in MHC I–dependent $CD8^+$ T-cell differentiation in the thymus (291). These combined defects in IRF-1–regulated MHC class I, TAPI, and IL-7Rα expression may result in the drastic reduction in $CD8^+$ T-cell numbers and $CD8^+$ T-cell functions, such as the inability to clear certain viral infections in the IRF-$1^{-/-}$ mice. The loss of IFN-γ–inducible macrophage activation may be due to the lack expression of iNOS, OAS, PKR, and ODC, which are regulated by IRF-1. The lack of NO production underlies the inability of the IRF-$1^{-/-}$ macrophages to fight infections after challenge by bacteria, fungi, and viruses (237, 248, 253). In view of the central regulatory role played by IRF-1 and the ubiquitous expression of the PRL-R on various immune function cells, it is pertinent to reevaluate a potential role of PRL, through the JAK/Stat/IRF-1 pathway, in modulating some of these immune responses.

PRL and IRF-1 in Immune Regulation. It is becoming clear that an abnormal level of PRL is often associated with altered immune responses in animal models and autoimmune patients. In general, high PRL levels are immunostimulatory, and low PRL levels are immunoinhibitory. It is hypothesized that some of PRL's actions will be exerted through its activation of the IRF-1 gene. However, the functional outcome of IRF-1 expression is different, depending on the cell type and its stage of differentiation and development in the lymphoid tissues. This is best illustrated by IRF-1 functions in fibroblasts (antiproliferation, apoptosis, tumor suppression) versus T lymphocytes (proliferation, antiapoptosis) versus B lymphocytes (differentiation, apoptosis). The PRL/IRF-1 pathway may be involved in B-cell differentiation and apoptosis in the bone marrow, as overexpressed IRF-1 led to a depletion of B cells in the bone marrow (247). The PRL/IRF-1 pathway in developing thymocytes may promote TAPI/LMP2/MHC I expression and play a role in $CD8^+$ T-cell differentiation in the thymus, as IRF-$1^{-/-}$ animals have a 10-fold decrease in $CD8^+$ T cells (248). In T lymphocytes, PRL has been shown to protect against glucocorticoid-induced apoptosis (71, 72). One way in which PRL ex-

erts its antiapoptotic effects may be through the induction of Bcl-2 and Bax (292) gene expression. Another way in which PRL antagonizes glucocorticoid actions in lymphoid cells may be through modulating GR action *via* the formation of Stat/GR complexes (197). In this way, PRL-inducible Stat factors may antagonize the activities of GR and other members of the steroid receptor family at the promoters of either growth-related or apoptotic-related genes in immune function cells. A closer examination of PRL actions, through the JAK/Stat/IRF-1 pathway, in various cells of the immune system, will provide new insights into the role of PRL in immune regulation.

Summary

Several lines of research have pointed to a role for PRL and IRF-1 in modulating the immune response. The molecular analyses of PRL-R, its ligand PRL, and a PRL-inducible transcription factor, IRF-1, provide insights and generate tools with which to reexamine PRL's role in modulating immune function cell biology in both normal lymphocytes as well as in clinical diseases involving hyperprolactinemia. In the next few years, detailed analysis of the functional domains of the Stat and JAK proteins, cytokine receptors, and other signaling proteins will undoubtedly elucidate how these molecules regulate distinct patterns of gene transcription in various lymphoid cell types. As knock-out animals of PRL, PRL-R, JAKs, and Stats develop and become available for analysis along with IRF-1 knock-out mice, a better understanding of which immune targets and responses are affected or regulated by PRL will emerge. These transgenic animal studies, combined with comprehensive ongoing molecular, genetic, biochemical, and immunological studies in tissue culture, will help to elucidate the immunoregulatory properties of PRL and define its role as a cytokine.

I thank Drs. Jeff Rosen and Donald Marcus for their critical reading of this manuscript and members of my laboratory for their support.

1. Southard JN, Talamantes F. Placental prolactin-like proteins in rodents: Variations on a structural theme. Mol Cell Endocrinol **79:**133–140, 1991.
2. Soares MJ, Faria TN, Roby KF, Deb S. Pregnancy and the prolactin family of hormones: Coordination of anterior pituitary, uterine, and placental expression. Endocr Rev **12:**402–423, 1991.
3. Hobbs AA, Richards DA, Kessler DJ, Rosen JM. Complex hormonal regulation of rat casein gene expression. J Biol Chem **257:**3598–3605, 1982.
4. Krasnow JS, Hickey GJ, Richards JS. Regulation of aromatase mRNA and estradiol biosynthesis in rat ovarian granulosa and luteal cells by prolactin. Mol Endocrinol **4:**13–21, 1990.
5. Nevalainen MT, Valve EM, Makela SI, Blauer M, Tuohimaa PJ, Harkonen PL. Estrogen and prolactin regulation of rat dorsal and lateral prostate in organ culture. Endocrinology **129:**612–622, 1991.
6. Montgomery DW, Shen GK, Ulrich ED, Steiner LL, Parrish PR, Zukoski CF. Human thymocytes express a prolactin-like mRNA and synthesize bioactive prolactin-like proteins. Endocrinology **131:**3019–3026, 1992.
7. O'Neal KD, Montgomery DW, Truong TM, Yu-Lee L-Y. Prolactin gene expression in human thymocytes. Mol Cell Endocrinol **87:**R19–R23, 1992.
8. Pellegrini I, Lebrun J-J, Ali S, Kelly PA. Expression of prolactin and its receptor in human lymphoid cells. Mol Endocrinol **6:**1023–1031, 1992.
9. Jurcovicova J, Day RN, MacLeod RM. Expression of prolactin in rat lymphocytes. Prog Neuro Endocrin Immunol **5:**256–263, 1992.
10. Hartmann DP, Holaday JW, Bernton EW. Inhibition of lymphocyte proliferation by antibodies to prolactin. FASEB J **3:**2194–2202, 1989.
11. Sabharwal P, Glaser R, LaFuse W, Varma S, Liu Q, Arkins S, Kooijman R, Kutz L, Kelley KW, Malarkey WB. Prolactin synthesized and secreted by human peripheral blood mononuclear cells: An autocrine growth factor for lymphoproliferation. Proc Natl Acad Sci **89:**7713–7716, 1992.
12. Bazan JF. A novel family of growth factor receptors: A common binding domain in the growth hormone, prolactin, erythropoietin and IL-6 receptors, and the p75 IL-2 receptor beta-chain. Biochem Biophys Res Commun **164:**788–795, 1989.
13. Cosman D. The hematopoietin receptor superfamily. Cytokine **5:**95–106, 1993.
14. Bernton EW, Bryant HU, Holaday JW. Prolactin and Immune Function. Psychoneuroimmunology. New York: Academic Press, pp 403–428, 1991.
15. Hooghe R, Delhase M, Vergani P, Malur A, Hooghe-Peters EL. Growth hormone and prolactin are paracine growth and differentiation factors in the haemopoietic system. Immunol Today **14:**212–214, 1993.
16. Horseman ND, Yu-Lee L-Y. Transcriptional regulation by the helix bundle peptide hormones: Growth hormone, prolactin, and hematopoietic cytokines. Endocr Rev **15:**627–649, 1994.
17. Murphy WJ, Rui H, Longo DL. Effects of growth hormone and prolactin in immune development and function. Life Sci **57:**1–14, 1995.
18. Gala RR. Prolactin and growth hormone in the regulation of the immune system. Proc Soc Exp Biol Med **198:**513–527, 1991.
19. Savino W, Dardenne M. Immune-neuroendocrine interactions. Immunol Today **16:**318–322, 1995.
20. Weigent DA. Immunoregulatory properties of growth hormone and prolactin. Pharmacol Ther **69:**237–257, 1996.
21. Matera L. Endocrine, paracrine and autocrine actions of prolactin on immune cells. Life Sci **59:**599–614, 1996.
22. Doppler W. Regulation of gene expression by prolactin. Rev Physiol Biochem Pharmacol **124:**93–130, 1994.
23. Groner B, Gouillex F. Prolactin-mediated gene activation in mammary epithelial cells. Curr Opin Genet Dev **5:**587–594, 1995.
24. Rosen JM, Zahnow C, Kazansky AV, Raught B. Composite response elements mediate hormonal and developmental regulation of milk protein gene expression. In: Biochemical Society Annual Symposium. Rudland PS, Fernig D, Leinster S, Eds. London: Portland Press (in press), 1997.
25. Lahita RG. The importance of estrogens in systemic lupus erythematosus. Clin Immunol Immunopathol **63:**17–18, 1992.
26. Lieberman ME, Maurer RA, Gorski J. Estrogen control of prolactin synthesis in vitro. Proc Natl Acad Sci **75:**5946–5949, 1978.
27. Lavalle C, Loyo E, Paniagua R, Bermudez JA, Herrera J, Graef A, Gonzalez-Barcena D, Fraga A. Correlation study between prolactin and androgens in male patients with systemic lupus erythematosus. J Rheum **14:**268–272, 1987.
28. Jara LJ, Gomez-Sanchez C, Silveira LH, Martinez-Osuna P, Vasey FB, Espinoza LR. Hyperprolactinemia in systemic lupus

erythematosus: association with disease activity. Am J Med Sci **303:**222–226, 1992.
29. Kira JI, Harada M, Yamaguchi Y, Shida N, Goto I. Hyperprolactinemia in multiple sclerosis. J Neurol Sci **102:**61–66, 1991.
30. Chikanza IC, Petrou P, Chrousos G, Kingsley G, Panayi GS. Excessive and dysregulated secretion of prolactin in rheumatoid arthritis: Immunopathogenetic and therapeutic implications. Br J Rheumatol **32:**445–448, 1993.
31. McMurray RW, Allen SH, Pepmueller PH, Keisler D, Cassidy JT. Elevated serum prolactin levels in children with juvenile rheumatoid arthritis and antinuclear antibody seropositivity. J Rheum **22:**1577–1580, 1995.
32. Buskila D, Sukenik S, Holcberg G, Horowitz J. Improvement of psoriatic arthritis in a patient treated with bromocriptine for hyperprolactinemia. J Rheum **18:**611–612, 1991.
33. Graef AS, Gonzalez SS, Baca VR, Ramirez ML, Daza LB, Blanco FF, Ortiz OA, Lavalle CM. High serum prolactin levels in asymptomatic HIV-infected patients and in patients with acquired immunodeficiency syndrome. Clin Immunol Immunopathol **72:**390–393, 1994.
34. Carrier M, Emery RW, Wild-Mobley J, Perrotta NJ, Russell DH, Copeland JG. Prolactin as a marker of rejection in human heart transplantation. Transplant Proc **19:**3442–3443, 1987.
35. Berczi I, Cosby H, Hunter T, Baragar F, McNeilly AS, Friesen HG. Decreased bioactivity of circulating prolactin in patients with rheumatoid arthritis. Br J Rheumatol **26:**433–436, 1987.
36. Palestine AG, Nussenblatt RB. The effect of bromocriptine on anterior uveitis. Am J Ophthalmol **106:**488–489, 1988.
37. Geffner ME, Lippe BM. The role of immunotherapy in Type I diabetes mellitus. West J Med **146:**337–343, 1987.
38. Vidaller A, Guadarrama F, Llorente L, Mendez JP, Larrea F, Villa AR, Alarcon-Segovia D. Hyperprolactinemia inhibits natural killer (NK) cell function *in vivo* and its bromocriptine treatment not only corrects it but makes it more efficient. J Clin Immunol **12:**210–215, 1992.
39. Leite-de-Moraes MC, Touraine, P, Kelly PA, Kuttenn F, Dardenne M. Prolactin receptor expression in lymphocytes from patients with hyperprolactinemia or acromegaly. J Endocrinol **147:**353–359, 1995.
40. Nagy E, Berczi I, Friesen HG. Regulation of immunity in rats by lactogenic and growth hormones. Acta Endocrinol **102:**351–357, 1983.
41. Nagy E, Berczi I. Pituitary dependence of bone marrow function. Br J Haematol **71:**457–462, 1989.
42. Kelley KW, Brief S, Westly HJ, Novakofski J, Bechtel PJ, Simon J, Walker EB. GH_3 pituitary adenoma cells can reverse thymic aging in rats. Proc Natl Acad Sci **83:**5663–5667, 1986.
43. Li YM, Brunke DL, Dantzer R, Kelley KW. Pituitary epithelial cell implants reverse the accumulation of $CD4^-CD8^-$ lymphocytes in thymus glands of aged rats. Endocrinology **130:**2703–2709, 1992.
44. Neidhart M, Fluckiger EW. Hyperprolactinaemia in hypophysectomized or intact male rats and the development of adjuvant arthritis. Immunology **77:**449–455, 1992.
45. Riskind PN, Massacesi L, Doolittle TH, Hauser SL. The role of prolactin in autoimmune demyelination: Suppression of experimental allergic encephalomyelitis by bromocriptine. Ann Neurol **29:**542–547, 1991.
46. McMurray R, Keisler D, Kanuckel K, Izui S, Walker SE. Prolactin influences autoimmune disease activity in the female B/W mouse. J Immunol **147:**3780–3787, 1991.
47. Walker SE, Allen SH, McMurray RW. Prolactin and autoimmune disease. Trends Endocrinol Metab **4:**147–151, 1993.
48. Murphy WJ, Durum SK, Longo DL. Differential effects of growth hormone and prolactin on murine T cell development and function. J Exp Med **178:**231–236, 1993.
49. Gala RR, Shevach EM. Influence of prolactin and growth hormone on the activation of dwarf mouse lymphocytes in vivo. Proc Soc Exp Biol Med **204:**224–230, 1993.
50. Gaufo GO, Diamond MC. Prolactin increases CD4/CD8 cell ratio in thymus-grafted congenitally athymic nude mice. Proc Natl Acad Sci **93:**4165–4169, 1996.
51. Matera L, Bellone G, Cesano A. Prolactin and the neuroimmune network. Adv Neuroimmunol **1:**158–172, 1991.
52. Clodi M, Svoboda T, Kotzmann H, Deyssig R, Woloszczuk W, Zielinski CC, Luger A. Effect of elevated serum prolactin concentrations on cytokine production and natural killer cell activity. Neuroendocrinol **56:**775–779, 1992.
53. Bellone G, Geuna M, Carbone A, Silvestri S, Foa R, Emanuelli G, Matera L. Regulatory action of prolactin on the in vitro Growth of CD34+ve human hemopoietic progenitor cells. J Cell Physiol **163:**221–231, 1995.
54. Cincotta AH, Knisely TL, Landry RJ, Miers WR, Gutierrez PJA, Esperanza P, Meier AH. The immunoregulatory effects of prolactin in mice are time of day dependent. Endocrinology **136:**2163–2171, 1995.
55. Bernton EW, Meltzer MS, Holaday JW. Suppression of macrophage activation and T-lymphocyte function in hypoprolactinemic mice. Science **239:**401–404, 1989.
56. Hiestand PC, Mekler P, Nordmann R, Grieder A, Permmongkol C. Prolactin as a modulator of lymphocyte responsiveness provides a possible mechanism of action of cyclosporine. Proc Natl Acad Sci **83:**2599–2603, 1986.
57. Morikawa K, Oseko F, Morikawa S. Immunosuppressive activity of bromocriptine on human T lymphocyte function *in vitro*. Clin Exp Immunol **95:**514–518, 1994.
58. Blank M, Krause I, Buskila D, Teitelbaum D, Kopolovic J, Afek A, Goldberg I, Shoenfeld Y. Bromocriptine immunomodulation of experimental SLE and primary antiphospholipid syndrome via induction of nonspecific T suppressor cells. Cell Immunol **162:**114–122, 1995.
59. Montgomery DW, Zukoski, CF, Shah GN, Buckley AR, Pacholczyk T, Russell DH. Concanavalin A–stimulated murine splenocytes produce a factor with prolactin-like bioactivity and immunoreactivity. Biochem Biophys Res Commun **145:**692–698, 1987.
60. Skwarlo-Sonta K. Mitogenic effect of prolactin on chicken lymphocytes in vitro. Immunol Lett **24:**171–178, 1990.
61. Viselli SM, Stanek, EM, Mukherjee P, Hymer WC, Mastro AM. Prolactin-induced mitogenesis of lymphocytes from ovariectomized rats. Endocrinology **129:**983–989, 1991.
62. Clevenger CV, Sillman AL, Prystowsky MB. Interleukin-2 driven nuclear translocation of prolactin in cloned T-lymphocytes. Endocrinology **127:**3151–3159, 1990.
63. Yu-Lee L-Y. Prolactin stimulates transcription of growth-related genes in Nb2 T lymphoma cells. Mol Cell Endocrinol **68:**21–28, 1990.
64. Gout PW, Beer CT, Noble RL. Prolactin-stimulated growth of cell cultures established from malignant Nb rat lymphomas. Cancer Res **40:**2433–2436, 1980.
65. Yu-Lee L-Y, Hrachovy JA, Stevens AM, Schwarz LA. Interferon-regulatory factor 1 is an immediate-early gene under transcriptional regulation by prolactin in Nb2 T cells. Mol Cell Biol **10:**3087–3094, 1990.
66. Mukherjee P, Mastro AM, Hymer WC. Prolactin induction of interleukin-2 receptors on rat splenic lymphocytes. Endocrinology **126:**88–94, 1990.
67. Gala RR, Shevach EM. Identification by analytical flow cytometry of prolactin receptors on immunocompetent cell populations in the mouse. Endocrinology **133:**1617–1623, 1993.
68. Gagnerault M-C, Touraine P, Savino W, Kelly PA, Dardenne M. Expression of prolactin receptors in murine lymphoid cells in normal and autoimmune situations. J Immunol **150:**5673–5681, 1993.

69. O'Neal KD, Schwarz LA, Yu-Lee L-Y. Prolactin receptor gene expression in lymphoid cells. Mol Cell Endocrinol **82:**127–135, 1991.
70. Dardenne M, Leite de Moraes M, Kelly PA, Gagnerault M-C. Prolactin receptor expression in human hematopoietic tissues analysed by flow cytoflourometry. Endocrinology **134:**2108–2114, 1994.
71. Fletcher-Chiappini SE, Comptom MM, La Voie HA, Day EB, Witorsch RJ. Glucocorticoid-prolactin interactions in Nb2 lymphoma cells: antiproliferative versus anticytolytic effects. Proc Soc Exp Biol Med **202:**345–352, 1993.
72. LaVoie HA, Witorsch RJ. Investigation of intracellular signals mediating the anti-apoptotic action of prolactin in Nb2 lymphoma cells. Proc Soc Exp Biol Med **209:**257–269, 1995.
73. Lahat N, Miller A, Shtiller R, Touby E. Differential effects of prolactin upon activation and differentiation of human B lymphocytes. J Neuroimmunol **47:**35–40, 1993.
74. Kenner JR, Holaday JW, Bernton WEW. Prolactin-like protein in murine splenocytes: Morphological and biochemical evidence. Prog NeuroEndocrinImmunol **3:**188–195, 1990.
75. Wu H, Devi R, Malarkey WB. Expression and localization of prolactin messenger ribonucleic acid in the human immune system. Endocrinology **137:**349–353, 1996.
76. Ben-Jonathan N, Mershon J, Allen D, Steinmetz R. Extrapituitary prolactin: Distribution, regulation, functions and clinical aspects. Endocr Rev **19:**639–669, 1996.
77. Rao Y-P, Buckley DJ, Olson MD, Buckley AR. Nuclear translocation of prolactin: Collaboration of tyrosine kinase and protein kinase C activation in rat Nb2 node lymphoma cells. J Cell Physiol **163:**266–267, 1995.
78. Gala RR, Rillema JA. Evaluation of prolactin-like activity produced by concanavalin-A stimulated mouse splenocytes. Life Sci **57:**1683–1700, 1995.
79. DiMattia GE, Gellersen B, Duckworth ML, Friesen HG. Human prolactin gene expression: The use of an alternative noncoding exon in decidua and the IM-9-P3 lymphoblast cell line. J Biol Chem **265:**16412–16421, 1990.
80. Gellersen B, Kempf R, Telgmann R, DiMattia GE. Nonpituitary human prolactin gene transcriptin is independent of pit-1 and differentially controlled in lymphocytes and endometrial stroma. Mol Endocrinol **8:**356–373, 1994.
81. Gellersen B, Kempf R, Telgmann R, DiMattia GE. Pituitary-type transcription of the human prolactin gene in the absence of pit-1. Mol Endocrinol **9:**887–901, 1995.
82. Ali S, Edery M, Pellegrini I, Lesueur L, Paly J, Djiane J, Kelly PA. The Nb2 form of prolactin receptor is able to activate a milk protein gene promoter. Mol Endocrinol **6:**1242–1248, 1992.
83. Touraine P, Carmo Leite de Moraes M, Dardenne M, Kelly PA. Expression of short and long forms of prolactin receptor in murine lymphoid tissues. Mol Cell Endocrinol **104:**183–190, 1994.
84. Gunes H, Mastro AM. Prolactin receptor gene expression in rat splenocytes and thymocytes from birth to adulthood. Mol Cell Endocrinol **117:**41–52, 1996.
85. Viselli SM, Mastro AM. Prolactin receptors are found on heterogeneous subpopulations of rat splenocytes. Endocrinology **132:**571–576, 1993.
86. Okamura H, Zachwieja J, Raguet S, Kelly PA. Characterization and applications of monoclonal antibodies to the prolactin receptor. Endocrinology **124:**2499–2508, 1989.
87. Kincade PW, Lee G, Pietrangeli CE, Hayashi S-I, Gimble JM. Cells and molecules that regulate B lymphopoiesis in bone marrow. Annu Rev Immunol **7:**111–143, 1989.
88. McAveney KM, Gimble JM, Yu-Lee L-Y. Prolactin receptor expression during adipocyte differentiation of bone marrow stroma. Endocrinol **137:**5723–5726, 1996.
89. Clevenger CV, Russell DH, Appasamy PM, Prystowsky MB. Regulation of IL2-driven T-lymphocyte proliferation by prolactin. Proc Natl Acad Sci **87:** 6460–6464, 1990.
90. O'Neal KD, Yu-Lee L-Y. The proline-rich motif (PRM): A novel feature of the cytokine/hematopoietin receptor superfamily. Lymph Cyt Res **12:**309–312, 1993.
91. Bazan JF. Shared architecture of hormone binding domains in type I and II interferon receptors. Cell **61:**753–754, 1990.
92. Murakami M, Narazaki M, Hibi M, Yawata H, Yasukawa K, Hamaguchi M, Taga T, Kishimoto T. Critical cytoplasmic region of the interleukin 6 signal transducer gp130 is conserved in the cytokine receptor family. Proc Natl Acad Sci **88:** 11349–11353, 1991.
93. Kelly PA, Djiane J, Postel-Vinay M-C, Edery M. The prolactin/growth hormone receptor family. Endocr Rev **12:**235–251, 1991.
94. Davis JA, Linzer DIH. Expression of multiple forms of the prolactin receptor in mouse liver. Mol Endocrinol **3:**674–680, 1989.
95. O'Neal KD, Yu-Lee L-Y. Differential signal transduction of the short, Nb2, and long PRL receptor: Activation of IRF-1 and cell proliferation. J Biol Chem **269:**26076–26082, 1994.
96. Boutin J-M, Jolicoeur C, Okamura H, Gagnon J, Edery M, Shirota M, Banville D, Dusanter-Fourt I, Djiane J, Kelly PA. Cloning and expression of the rat prolactin receptor, a member of the growth hormone/prolactin receptor gene family. Cell **53:**69–77, 1988.
97. Ali S, Pellegrini I, Kelly PA. A prolactin-dependent immune cell line (Nb2) expresses a mutant form of prolactin receptor. J Biol Chem **266:**20110–20117, 1991.
98. Clevenger CV, Chang WP, Ngo W, Pasha TLM, Montone KT, Tomaszweski JE. Expression of prolactin and prolactin receptor in human breast carcinoma. Am J Pathol **146:**695–705, 1995.
99. Nagano M, Kelly PA. Tissue distribution and regulation of rat prolactin receptor gene expression. J Biol Chem **269:**13337–13345, 1994.
100. DaSilva L, Howard OMZ, Rui H, Kirken RA, Farrar WL. Growth signaling and JAK2 association mediated by membrane-proximal cytoplasmic regions of prolactin receptors. J Biol Chem **269:**18267–18270, 1994.
101. Chang W-P, Clevenger CV. Modulation of growth factor receptor function by isoform heterodimerization. Proc Natl Acad Sci **93:**5947–5952, 1996.
102. Velazquez L, Fellous M, Stark GR, Pellegrini S. A protein tyrosine kinase in the interferon alpha/beta signaling pathway. Cell **70:**313–322, 1992.
103. Wilks AF, Harpur AG, Kurban RR, Ralph SJ, Zurcher G, Ziemiecki A. Two novel protein-tyrosine kinases, each with a second phosphotransferase-related catalytic domain, define a new class of protein kinase. Mol Cell Biol **11:**2057–2065, 1991.
104. Ziemiecki A, Harpur AG, Wilks AF. JAK protein tyrosine kinases: Their role in cytokine signalling. Trends Cell Biol **4:**207–212, 1994.
105. Takahashi T, Shirasawa T. Molecular cloning of rat JAK3, a novel member of the JAK family of protein tyrosine kinases. FEBS lett **342:**124–128, 1994.
106. Kirken RA, Rui H, Malabarba MG, Farrar WL. Identification of interleukin-2 receptor-associated tyrosine kinase p116 as novel leukocyte-specific Janus kinase. J Biol Chem **269:**19136–19141, 1994.
107. Witthuhn BA, Quelle FW, Silvennoinen O, Yi T, Tang B, Miura O, Ihle JN. JAK2 associates with the erythropoietin receptor and is tyrosine phosphorylated and activated following stimulation with erythropoietin. Cell **74:**227–236, 1993.
108. Igarashi K, Garotta G, Ozmen L, Ziemiecki A, Wilks AF, Harpus AG, Larner AC, Finbloom DS. Interferon-gamma induces tyrosine phosphorylation of interferon-gamma receptor and regulated association of protein tyrosine kinases, Jak1 and Jak2, with its receptor. J Biol Chem **269:**14333–14336, 1994.
109. Rui H, Kirken RA, Farrar WL. Activation of receptor-associated tyrosine kinase JAK2 by prolactin. J Biol Chem **269:**5364–5368, 1994.

110. Stahl N, Boulton TG, Farruggella T, Ip NY, Davis S, Witthuhn BA, Quelle FW, Silvennoinen O, Barbieri G, Pellegrini S, Ihle JN, Yancopoulos GD. Association and activation of Jak-Tyk kinases by CNTF-LIF-OSM-IL-6 beta receptor components. Science **263:**92–95, 1994.
111. Argetsinger LS, Campbell GS, Yang X, Witthuhn BA, Silvennoinen O, Ihle JN, Carter-Su C. Identification of JAK2 as a growth hormone receptor-associated tyrosine kinase. Cell **74:**237–244, 1993.
112. Darnell JE Jr., Kerr IM, Stark GR. Jak-STAT pathways and transcriptional activation in response to IFNs and other extracellular signaling proteins. Science **264:**1415–1421, 1994.
113. Schindler C, Darnell JE Jr. Transcriptional responses to polypeptide ligands: The Jak-STAT pathway. Annu Rev Biochem **64:**621–651, 1995.
114. Elberg G, Kelly PA, Djiane J, Binder L, Gertler A. Mitogenic and binding properties of monoclonal antibodies to the prolactin receptor in Nb_2 rat lymphoma cells. J Biol Chem **265:**14770–14776, 1990.
115. Campbell GS, Argetsinger LS, Ihle JN, Kelly PA, Rillema JA, Carter-Su C. Activation of JAK2 tyrosine kinase by prolactin receptors in Nb2 cells and mouse mammary gland explants. Proc Natl Acad Sci **91:**5232–5236, 1994.
116. Lebrun J, Ali S, Sofer L, Ullrich A, Kelly PA. Prolactin-induced proliferation of Nb2 cells involves tyrosine phosphorylation of the prolactin receptor and its associated tyrosine kinase JAK2. J Biol Chem **269:**14021–14026, 1994.
117. Lebrun J, Ali S, Ullrich A, Kelly PA. Proline-rich sequence-mediated Jak2 association to the prolactin receptor is required but not sufficient for signal transduction. J Biol Chem **270:**10664–10670, 1995.
118. Lebrun J, Suhad A, Goffin V, Ullrich A, Kelly PA. A single phosphotyrosine residue of the prolactin receptor is responsible for activation of gene transcription. Proc Natl Acad Sci **92:**4031–4035, 1995.
119. O'Neal KD, Yu-Lee L-Y, Shearer WT. The proline-rich motif is necessary but not sufficient for prolactin receptor signal transduction. Ann NY Acad Sci **766:**282–284, 1995.
120. Fukunaga R, Ishizaka-Ikeda E, Pan C-X, Seto Y, Nagata S. Functional domains of the granulocyte colony-stimulating factor receptor. EMBO J **10:**2855–2865, 1991.
121. Takaki S, Kanazawa H, Shiiba M, Takatsu K. A critical cytoplasmic domain of the interluekin-5 (IL-5) receptor α chain and its function in IL-5–mediated growth signal transduction. Mol Cell Biol **14:**7404–7413, 1994.
122. Goujon L, Allevato G, Simmon G, Paquereau L, Le Cam A, Clark J, Nilesen JH, Djiane J, Postel-Vinay M-C, Edery M, Kelly PA. Cytoplasmic sequences of the growth hormone receptor necessary for signal transduction. Proc Natl Acad Sci **91:**957–961, 1994.
123. Billestrup N, Bouchelouche P, Allevato G, Ilondo M, Nielson JH. Growth hormone receptor C-terminal domains required for growth homrone-induced intracellular free Ca^{2+} oscillations and gene transcription. Proc Natl Acad Sci **92:**2725–2729, 1995.
124. Dinerstein H, Lago F, Goujon L, Ferrag F, Esposito N, Finidori J, Kelly PA, Postenl-Vinay M-C. The proline-rich region of the GH receptor is essential for JAK2 phosphorylation, activation of cell proliferation, and gene transcription. Mol Endocrinol **9:**1701–1707, 1995.
125. Quelle FW, Sato N, Witthuhn BA, Inhorn RC, Eder M, Miyajima A, Griffin JD, Ihle JN. JAK2 associates with the βc chain of the receptor for granulocyte-macrophage colony-stimulating factor, and its activation requires the membrane-proximal region. Mol Cell Biol **14:**4335–4341, 1994.
126. Sotiropoulos A, Perrot-Applanat M, Dinerstein H, Pallier A, Postel-Vinay M, Finidori J, Kelly PA. Distinct cytoplasmic regions of the growth hormone receptor are required for activation of JAK2, mitogen-activated protein kinase and transcription. Endocrinology **135:**1292–1297, 1994.
127. Zhao Y, Wagner F, Frank SJ, Kraft AS. The amino-terminal portion of the JAK2 protein kinase is necessary for binding and phosphorylation of the granulocyte-macrophage colony-stimulating factor receptor βc chain. J Biol Chem **270:**13814–13818, 1995.
127a. Sigal NH, Dumont FJ. Cyclosporin A, FK-506, and rapamycin: Pharmacologic probes of lymphocyte signal transduction. Annu Rev Immunol **10:**519–560, 1992.
127b. Galat A. Peptidylproline *cis-trans*-isomerases: Immunophilins. Eur J Biochem **216:**689–707, 1993.
127c. O'Neal KD, Chari MV, McDonald CH, Cook RG, Yu-Lee L-Y, Morrisett JD, Shearer WT. Multiple *cis-trans* conformers of the prolactin receptor proline-rich motif (PRM) peptide detected by reverse-phase HPLC, CD and NMR spectroscopy. Biochemical J **315:**833–844, 1996.
128. Edery M, Levi-Meyrueis C, Paly J, Kelly PA, Djian J. A limited cytoplasmic region of the prolactin receptor critical for signal transduction. Mol Cell Endocrinol **102:**39–44, 1994.
129. Miyazaki T, Liu Z-J, Kawahara A, Minami Y, Yamada K, Tsujimoto Y, Barsoumian EL, Perlmutter RM, Taniguchi T. Three distinct IL-2 signaling pathways mediated by bcl-2, c-myc, and lck cooperate in hematopoietic cell proliferation. Cell **81:**223–231, 1995.
130. Greenlund AC, Farrar MA, Viviano BL, Schreiber RD. Ligand-induced IFN-gamma receptor tyrosine phosphorylation couples the receptor to its signal transduction system (p91). EMBO J **13:**1591–1600, 1994.
131. Hou J, Schindler U, Henzel WJ, Ho TC, Brasseur M, McKnight SL. An interleukin-4–induced transcription factor: IL-4 Stat. Science **265:**1701–1706, 1994.
132. Gaffen SL, Lai SY, Xu W, Gouilleux F, Groner B, Goldsmith MA, Greene WC. Signaling through the interleukin-2 receptor beta chain activates a Stat5-like DNA-binding activity. Proc Natl Acad Sci **92:** 7192–7196, 1995.
133. Lin JX, Migone TS, Tsang M, Friedmann M, Weatherbee JA, Zhou L, Yamauchi A, Bloom ET, Mietz J, John S, Leonard WJ. The role of shared receptor motifs and common Stat proteins in the generation of cytokine pleiotrophy and redundancy by IL-2, IL-4, IL-7, IL-13, and IL-15. Immunity **2:**331–339, 1995.
134. Stahl N, Farruggella TJ, Boulton TG, Zhong, Z, Darnell JE, Yancopoulos GD. Choice of STATs and other substrates specified by modular tyrosine-based motifs in cytokine receptors. Science **267:**1349–1353, 1995.
135. Greenlund AC, Morales MO, Viviano BL, Yan H, Krolewski J, Schreiber RD. Stat recruitment by tyrosine-phosphorylated cytokine receptors: An ordered reversible affinity-driven process. Immunity **2:**677–687, 1995.
136. Schindler U, Wu P, Rothe M, Brasseur M, McKnight SL. Components of a Stat recognition code: Evidence for two layers of molecular selectivity. Immunity **2:**689–697, 1995.
137. Quelle FW, Wang D, Nosaka T, Thierfelder WE, Stravopodis D, Weistein Y, Ihle JN. Erythropoietin induces activation of Stat5 through association with specific tyrosines on the receptor that are not required for a mitogenic response. Mol Cell Biol **16:**1622–1631, 1996.
138. Smit LS, Meyer DJ, Billestrup N, Norstedt G, Schwartz J, Carter-Su C. The role of the growth hormone (GH) receptor and Jak1 and Jak2 kinases in the activation of Stats1, 3, and 5 by GH. Mol Endocrinol **10:**519–533, 1996.
139. Lobie PE, Allevato G, Nielsen JH, Norstedt G, Billestrup N. Requirement of tyrosine residues 333 and 338 of the growth hormone (GH) receptor for selected GH-stimulated function. J Biol Chem **270:**21745–21750, 1995.
140. Wang Y-D, Wong K, Wood WI. Intracellular tyrosine residues of the human growth hormone receptor are not required for the

signaling of proliferation of JAK-Stat activation. J Biol Chem **270:**7021–7024, 1995.
141. Colosi P, Wong K, Leong SR, Woor WI. Mutational analysis of the intracellular domain of the human growth hormone receptor. J Biol Chem **268:**12617–12623, 1993.
142. Fukunaga R, Ishizaka-Ikeda E, Nagata S. Growth and differentiation signals mediated by different regions in the cytoplasmic domain of granulocyte colony-stimulating factor receptor. Cell **74:**1079–1087, 1993.
143. D'Andrea A, Yoshimura A, Youssoufian H, Zon LI, Koo J, Lodish HF. The cytoplasmic region of the erythropoietin receptor contains nonoverlapping positive and negative growth regulatory domains. Mol Cell Biol **11:**1980–1987, 1991.
144. Sato N, Sakamaki K, Terada N, Arai K, Miyajima A. Signal transduction by the high-affinity GM-CSF receptor: Two distinct cytoplasmic regions of the common β subunit responsible for different signaling. EMBO J **12:**4181–4189, 1993.
145. Nelson BH, Lord JD, Greenberg PD. A membrane-proximal region of the interleukin-2 receptor gamma$_c$ chain sufficient for Jak kinase activation and induction of proliferation in T cells. Mol Cell Biol **16:**309–317, 1996.
146. Witthuhn BA, Silvennoinen O, Miura O, Lai KS, Cwik C, Liu ET, Ihle JN. Involvement of Jak3 Janus kinase in signaling by interleukins 2 and 4 in lynmphoid and myeloid cells. Nature **370:**153–157, 1994.
147. Luo H, Hanratty WH, Dearolf CR. An amino acid substitution in the *Drosophila hop*$^{Tum\text{-}l}$ Jak kinase causes leukemia-like hematopoietic defects. EMBO J **14:**1412–1420, 1995.
148. Zhuang HM, Patel SV, He TC, Sonsteby SK, Niu ZT, Wojchowski DM. Inhibition of erythropoietin-induced mitogenesis by a kinase-deficient form of Jak2. J Biol Chem **269:**21411–21414, 1994.
149. Duhe RJ, Farrar WL. Characterization of active and inactive forms of the JAK2 protein-tyrosine kinase produced via the baculovirus expression vector system. J Biol Chem **270:**23085–23090, 1995.
150. Velazquez L, Mogensen KE, Barbieri G, Fellous M, Uze G, Pellegrini S. Distinct domains of the protein tyrosine kinase tyk2 required for binding of interferon-α/β and for signal transduction. J Biol Chem **270:**3327–3334, 1995.
151. Frank SJ, Yi W, Zhao Y, Goldsmith JF, Gilliland G, Jiang J, Sakai K, Kraft AS. Regions of the JAK2 tyrosine kinase required for coupling to the growth hormone receptor. J Biol Chem **270:**14776–14785, 1995.
152. Gao J, Hughes JP, Auperin B, Buteau H, Edery M, Zhuang H, Woychowski DM, Horseman ND. Interactions among JANUS kinases and the prolactin (PRL) receptorin the regulation of a PRL response element. Mol Endocrinol **10:**847–856, 1996.
153. Pawson T, Gish GD. SH2 and SH3 domains: From structure to function. Cell **71:**359–362, 1992.
154. Yi T, Mui AL-F, Krystal G, Ihle JN. Hematopoietic cell phosphatase associates with the interleukin-3 (IL-3) receptor β chain and down-regulates IL-3–induced tyrosine phosphorylation and mitogenesis. Mol Cell Biol **13:**7577–7586, 1993.
155. Hunter T. Protein kinases and phosphatases: The yin and yang of protein phosphorylation and signaling. Cell **80:**225–236, 1995.
156. Pei D, Wang J, Walsh CT. Differential functions of the two Src homology 2 domains in protein tyrosine phosphatase SH-PTP1. Proc Natl Acad Sci **93:**1141–1145, 1996.
157. Klingmuller U, Lorenz U, Cantley LC, Neel BG, Lodish FH. Specific recruitment of SH-PTP1 to the erythropoietin receptor causes inactivation of JAK2 and termination of proliferative signals, Cell **80:**729–738, 1995.
158. David M, Chen HE, Goelz S, Larner AC, Neel BG. Differential regulation of the alpha/beta interferon-stimulated JAK/STAT pathway by the SH2 domain-containing tyrosine phosphatase SHPTP1. Mol Cell Biol **15:**7050–7058, 1995.
159. Petricoin E, David M, Igarashi K, Benjamin C, Ling L, Goelz S, Finbloom D, Larner AC. Inhibition of alpha interferon but not gamma interferon signal transduction by phorbol esters is mediated by a tyrosine phosphatase. Mol Cell Biol **16:**1419–1424, 1996.
160. Ali S, Chen Z, Lebrun J-J, Vogel W, Kharitonenkov A, Kelly PA, Ullrich A. PTP1D is a positive regulator of the prolactin signal leading to beta-casein promoter activation. EMBO J **15:**135–142, 1996.
161. Pellegrini S, Schindler C. Early events in signalling by interferons. Trends Biochem Sci **18:**338–342, 1993.
162. Rothman P, Kreider B, Azam M, Levy D, Wegenka U, Eilers A, Decker T, Forn F, Kashieva H, Ihle J, Schindler C. Cytokines and growth factors signal through tyrosine phosphorylation of a family of related transcription factors. Immunity **1:**457–468, 1994.
163. Ihle JN. STATs: Signal transducers and activators of transcription. Cell **43:**331–334, 1996.
164. Yan R, Small S, Desplan C, Dearolf CR, Darnell JE Jr. Identification of a *Stat* gene that functions in Drosphila development. Cell **84:**421–430, 1996.
165. Hou XS, Melnick MB, Berrimon N. *Marella* acts downstream of the Drosophila HOP/JAK kinase and encodes a protein similar to the mammalian STATs. Cell 84:411–419, 1996.
166. Yan R, Qureshi S, Zhong Z, Wen Z, Darnell JE. The genomic structure of the STAT genes: Multiple exons in coincident sites in Stat1 and Stat2. Nucleic Acids Res **23:**459–463, 1995.
167. Shuai K, Schindler C, Prezioso VR, Darnell JE Jr. Activation of transcription by IFN-gamma: Tyrosine phosphorylation of a 91-kD DNA binding protein. Science **258:**1808–1812, 1992.
168. Shuai K, Horvath CM, Huang LH, Qureshi SA, Cowburn D, Darnell JE Jr. Interferon activation of the transcription factor Stat91 involves dimerization through SH2-pohsphotyrosyl peptide interactions. Cell **76:**821–828, 1994.
169. Azam M, Erdjument-Bromage H, Kreider BL, Xia M, Quelle F, Basu R, Saris C, Tempst P, Ihle JN, Schindler C. Interleukin-3 signals through multiple isoforms of Stat5. EMBO J **14:**1402–1411, 1995.
170. Kazansky AV, Raught B, Lindsey SM, Wang Y-F, Rosen JM. Regulation of MGF/STAT5a during mammary gland development. Mol Endocrinol **9:**1598–1609, 1995.
171. Schaefer TS, Sanders LK, Nathans D. Cooperative transcriptional activity of Jun and Stat3β, a short form of Stat3. Proc Natl Acad Sci **92:**9097–9101, 1995.
172. Moriggl R, Fouilleux-Gruart V, Jahne R, Berchtold S, Gartmann C, Liu X, Hennighausen L, Sotiropoulos A, Groner B, Gouilleux F. Deletion of the carboxyl-terminal transactivation domain of MGF-Stat5 results in sustained DNA binding and a dominant negative phenotype. Mol Cell Biol **16:**5691–5700, 1996.
173. Wen ZL, Zhong Z, Darnell JE. Maximal activation of transcription by Stat1 and Stat3 requires both tyrosine and serine phosphorylation. Cell **82:**241–250, 1995.
174. Leung S, Li X, Stark GR. Stats find that hanging together can be stimulating. Science **273:**750–751, 1996.
175. Horvath CM, Wen Z, Darnell JE. A STAT protein domain that determines DNA sequence recognition suggests a novel DNA-binding domain. Genes Dev **9:**984–994, 1995.
176. Heim MH, Kerr IM, Stark GR, Darnell JE. Contribution of STAT SH2 groups to specific interferon signaling by the Jak-STAT pathway. Science **267:**1347–1349, 1995.
177. Schindler C, Shuai K, Prezioso VR, Darnell JE Jr. Interferon-dependent tyrosine phosphorylation of a latent cytoplasmic transcription factor. Science **257:**809–815, 1992.
178. Shuai K, Stark GR, Kerr IM, Darnell JE Jr. A single phosphotyrosine residue of Stat91 required for gene activation by interferon-gamma. Science **261:**1744–1746, 1993.
179. Zhang X, Blenis, J, Li HC, Schindler C, Chen-Kiang S. Requirement of serine phosphorylation for formation of STAT-promoter complexes. Science **267:**1990–1994, 1995.

180. Eilers A, Georgellis D, Klose B, Schindler C, Ziemiecki A, Harpur AG, Wilks AF, Decker T. Differentiation-regulated serine phosphorylation of STAT1 promotes GAF activation in macrophages. Mol Cell Biol **15:**3579–3586, 1995.
181. David M, Petricoin E, Benjamin C, Pine R, Weber MJ, Larner AC. Requirement for MAP kinase (ERK2) activity in interferon α and interferon β stimulated gene expression through STAT proteins. Science **269:**1721–1723, 1995.
182. Gouilleux-Gruart V, Gouilleux F, Desaint C, Claisse J-F, Capiod J-C, Delobel J, Weber-Nordt R, Dusanter-Fourt I, Dreyfus F, Groner B, Prin L. STAT-related transcription factors are constitutively activated in peripheral blood cells from acute leukemic patients. Blood **87:**1692–1697, 1996.
183. Carlesso N, Frank DA, Griffin JD. Tyrosyl phosphorylation and DNA binding activity of signal transducers and activators of transcription (STAT) proteins in hematopoietic cell lines transformed by Bcr/Abl. J Exp Med 183:811–820, 1996.
184. Shuai K, Liao J, Song MM. Enhancement of antiproliferative activity of gamma interferon by the specific inhibition of tyrosine dephosphorylation of Stat1. Mol Cell Biol **16:**4932–4941, 1996.
185. David M, Grimley PM, Finbloom DS, Larner AC. A nuclear tyrosine phosphatase downregulates interferon-induced gene expression. Mol Cell Biol **13:**7515–7521, 1993.
186. Igarashi K, David M, Finbloom DS, Larner AC. In vitro activation of the transcription factor gamma interferon activation factor by gamma interferon: evidence for a tyrosine phosphatase/kinase signaling cascade. Mol Cell Biol **13:**1634–1640, 1993.
187. Haque SJ, Flati V, Deb A, Williams BRG. Role of protein-tyrosine phosphatases in Stat1α-mediated cell signaling. J Biol Chem **270:**25709–25714, 1995.
188. Kessler DS, Veals SA, Fu X-Y, Levy DE. Interferon-alpha regulates nuclear translocation and DNA-binding affinity of ISGF3, a multimeric transcriptional activator. Genes Dev **4:**1753–1765, 1990.
189. Bluyssen HAR, Muzaffar R, Vlieststra RJ, Van der Made ACJ, Leung S, Stark GR, Kerr IM, Trapman J, Levy DE. Combinatorial association and abundance of components of interferon-stimulated gene factor 3 dictate the selectivity of interferon responses. Proc Natl Acad Sci **92:**5645–5649, 1995.
190. Decker T, Lew DJ, Mirkovitch J, Darnell JE Jr. Cytoplasmic activation of GAF, an IFN-gamma–regulated DNA-binding factor. EMBO J **10:**927–932, 1991.
191. Seidel HM, Milocco LH, Lamb P, Darnell JE Jr., Stein RB, Rosen J. Spacing of palindromic half sites as a determinant of selective STAT (signal transducers and activators of transcription) DNA binding and transcriptional activity. Proc Natl Acad Sci **92:**3041–3045, 1995.
192. Li X, Leung S, Qureshi S, Darnell JE Jr., Stark GR. Formation of Stat1-Stat2 heterodimers and their role in the activation of IRF-1 gene transcription by interferon-alpha. J Biol Chem **271:**5790–5794, 1996.
193. Sadowski HB, Shuai K, Darnell JE Jr., Gilman MZ. A common nuclear signal transduction pathway activated by growth factor and cytokine receptors. Science **261:**1739–1744, 1993.
194. Raz R, Durbin JE, Levy DE. Acute phase response factor and additional members of the interferon-stimulated gene factor 3 family integrate diverse signals from cytokines, interferons, and growth factors. J Biol Chem **269:**24391–24395, 1994.
195. Look DC, Pelletier MR, Tidwell RM, Roswit WT, Holtzman MJ. Stat1 depends on transcriptional synergy with Sp1. J Biol Chem **270:**30264–30267, 1995.
196. Cao X, Tay A, Guy G, Tan YH. Activation and association of Stat3 with Src in v-Src-transformed cell lines. Mol Cell Biol **16:**1595–1603, 1996.
197. Stocklin E, Wissler M, Gouilleux F, Groner B. Functional interactions between Stat5 and the glucocorticoid receptor. Nature **383:**726–728, 1996.
198. Xu X, Sun Y-L, Hoey T. Cooperative DNA binding and sequence-selective recognition conferred by the Stat amino-terminal domain. Science **273:**794–797, 1996.
199. Wakao H, Gouilleux F, Groner B. Mammary gland factor (MGF) is a novel member of the cytokine regulated transcription factor gene family and confers the prolactin response. EMBO J **13:**2182–2191, 1994.
200. Welte T, Garimorth K, Philipp S, Doppler W. Prolactin-dependent activation of a tyrosine phosphorylated DNA binding factor in mouse mammary epithelial cells. Mol Endocrinol **8:**1091–1102, 1994.
201. Gouilleux F, Wakao H, Mundt M, Groner B. Prolactin induces phosphorylation of Tyr694 of Stat5 (MGF), a prerequisite for DNA binding and induction of transcription. EMBO J **13:**4361–4369, 1994.
202. Gilmour KC, Reich NC. Receptor to nucleus signaling by prolactin and interleukin 2 via activation of latent DNA-binding factors. Proc Natl Acad Sci **91:**6850–6854, 1994.
203. Gilmour KC, Pine R, Riech NC. Interleukin 2 activates Stat5 transcription factor (mammary gland factor) and specific gene expression in T lymphocytes. Proc Natl Acad Sci **92:**10772–10776, 1995.
204. Gouilleux F, Moritz D, Humar M, Moriggl R, Berchtold S, Groner B. Prolactin and interleukin-2 receptors in T lymphocytes signal through a MGF-Stat5–like transcription factor. Endocrinology **136:**5700–5708, 1995.
205. Wang Y-F, Yu-Lee L-Y. Multiple Stat complexes interact at the IRF-1 GAS in prolactin-stimulated Nb2 T cells. Mol Cell Endocrinol **121:**19–28, 1996.
206. Burdon TG, Demmer J, Clark JA, Watson CJ. The mammary factor MPBF is a prolactin-induced transcriptional regulator which binds to STAT factor recognition sites. Fed Eur Biochem Soc **350:**177–182, 1994.
207. Demmer J, Burdon TG, Dijane J, Watson CJ, Clark AJ. The proximal milk protein binding factor binding site is required for the prolactin responsiveness of the sheep β-lactoglobulin promoter in Chinese hamster ovary cells. Mol Cell Endocrinol **107:**113–121, 1995.
208. Li S, Rosen JM. CTF/NF1 and mammary gland factor (Stat5) play a critical role in regulating rat whey acidic protein gene expression in transgenic mice. Mol Cell Biol **15:**2063–2070, 1995.
209. Dajee M, Kazansky AV, Raught B, Hocke G, Fey GH, Richards JS. Prolactin induction of the α_2-macroglobulin gene in rat ovarian granulosa cells: Stat5 activation and binding to the interleukin-6 response element. Mol Endocrinol **10:**171–184, 1996.
210. David M, Petricoin EF III, Igarashi K-I, Feldman GM, Finbloom DS, Larner AC. Prolactin activates the interferon-regulated p91 transcription factor and the Jak2 kinase by tyrosine phosphorylation. Proc Natl Acad Sci **91:**7174–7178, 1994.
211. Stevens AM, Wang Y-F, Sieger KA, Lu H-F, Yu-Lee L-Y. Biphasic transcriptional regulation of the interferon regulatory factor–1 gene by prolactin: Involvement of gamma-interferon activated sequence and Stat-related proteins. Mol Endocrinol **9:**513–525, 1995.
212. DaSilva L, Rui H, Erwin RA, Howard OMZ, Kriken RA, Malabarba MG, Hackett RH, Larner AC, Farrar WL. Prolactin recruits Stat1, Stat3, and Stat5 independent of conserved receptor tyrosines Tyr402, Tyr479, Tyr515 and Tyr580. Mol Cell Endocrinol **117:**131–140, 1996.
213. Meier VS, Groner B. The nuclear factor YY1 participates in repression of the β-casein gene promoter in mammary epithelial cells and is counteracted by mammary gland factor during lactogenic hormone induction. Mol Cell Biol **14:**128–137, 1994.
214. Raught B, Khursheed B, Kanzansky A, Rosen J. YY1 represses β-casein gene expression by preventing the formation of a lactation-associated complex. Mol Cell Biol **14:**1752–1763, 1994.
215. Raught B, Liao WS-L, Rosen JM. Developmentally and hormonally regulated CCAAT/enhancer-binding protein isoforms in-

fluence β-casein gene expression. Mol Endocrinol **9:**1223–1232, 1995.
216. Ruff-Jamison S, Chen K, Cohen S. Epidermal growth factor induces the tyrosine phosphorylation and nuclear translocation of Stat5 in liver. Proc Natl Acad Sci **92:**4215–4218, 1995.
217. Lai C-F, Ripperger J, Morella KK, Wang Y, Gearing DP, Horseman ND, Campos SP, Fey GH, Baumann H. STAT3 and STAT5B are targets of two different signal pathways activated by hematopoietin receptors and control transcription via separate cytokine response elements. J Biol Chem **270:**23254–23257, 1995.
218. Liu X, Robinson GW, Hennighausen L. Activation of Stat5a and Stat5b by tyrosine phosphorylation is tightly linked to mammary gland differentiation. Mol Endocrinol **10:**1496–1506, 1996.
219. Clevenger CV, Medaglia MV. The protein tyrosine kinase $p59^{fyn}$ is associated with prolactin (PRL) receptor and is activated by PRL stimulation of T-lymphocytes. Mol Endocrinol **8:**674–681, 1994.
220. Clevenger CV, Torigoe T, Reed JC. Prolactin induces rapid phosphorylation and activation of prolactin receptor-associated RAF-1 kinase in a T-cell line. J Biol Chem **269:**5559–5565, 1994.
221. Berlanga, JJ, Vara JAF, Martin-Perez J, Garcia-Ruiz JP. Prolactin receptor is associated with c-*src* kinase in rat liver. Mol Endocrinol **9:**1461–1467, 1995.
222. Rao Y-P, Buckley DJ, Buckley AR. Rapid activation of mitogen-activated protein kinase and p21ras by prolactin and interleukin-2 in rat Nb2 node lymphoma cells. Cell Growth Diff **6:**1235–1244, 1995.
223. Erwin RA, Kirken RA, Malabarba MG, Farrar WL, Rui H. Prolactin activates Ras via signaling proteins SHC, Growth Factor Receptor Bound 2, and Son of Sevenless. Endocrincology **136:**3512–3518, 1996.
224. Fields S, Song O-Y. A novel genetic system to detect protein-protein interactions. Nature **340:**245–246, 1989.
225. Tanaka T, Shiu RPC, Gout PW, Beer CT, Noble RL, Friesen HG. A new sensitive and specific bioassay for lactogenic hormones: measurement of prolactin and growth hormone in human serum. J Clin Endocrinol Metab **51:**1058–1063, 1980.
226. Harada H, Fujita T, Miyamoto M, Kimura Y, Maruyama M, Furia A, Miyata T, Taniguchi T. Structurally similar but functionally distinct factors, IRF-1 and IRF-2, bind to the same regulatory elements of IFN and IFN-inducible genes. Cell **58:**729–739, 1989.
227. Nelson N, Marks MS, Driggers PH, Ozato K. Interferon consensus sequence-binding protein, a member of the interferon regulatory factor family, suppresses interferon-induced gene transcription. Mol Cell Biol **13:**588–599, 1993.
228. Veals SA, Schinder C, Leonard D, Fu X-Y, Aebersold R, Darnell JE Jr., Levy DE. Subunit of an alpha-interferon–responsive transcription factor is related to interferon regulatory factor and Myb families of DNA-binding proteins. Mol Cell Biol **12:**3315–3324, 1992.
229. Eisenbeis CF, Singh H, Storb U. Pip, a novel IRF family member, is a lymphoid-specific, PU.1-dependent transcriptional activator. Genes Dev **9:**1377–1387, 1995.
230. Au W-C, Moore PA, Lowther W, Juang Y-T, Pitha PM. Identification of a member of the interferon regulatory factor family that binds to the interferon-stimulated response element and activates expression of interferon-induced genes. Proc Natl Acad Sci **92:**11657–11661, 1995.
231. Bovolenta C, Driggers PH, Marks MS, Medin JA, Politis AD, Vogel SN, Levy DE, Sakaguchi K, Appella E, Coligan JE, Ozato K. Molecular interactions between interferon consensus sequence binding protein and members of the interferon regulatory factor family. Proc Natl Acad Sci **91:**5046–5050, 1994.
232. Schwarz LA, Stevens AM, Hrachovy JA, Yu-Lee L-Y. Interferon regulatory factor–1 is inducible by prolactin, interleukin-2 and concanavalin A in T cells. Mol Cell Endocrinol **86:**103–110, 1992.
233. Clevenger CV, Sillman AL, Hanley-Hyde J, Prystowsky MB. Requirement for prolactin during cell cycle regulated gene expression in cloned T-lymphocytes. Endocrinology **130:**3216–3222, 1992.
234. Miyamoto M, Fujita T, Kimura Y, Maruyama M, Harada H, Sudo Y, Miyata T, Taniguchi T. Regulated expression of a gene encoding a nuclear factor, IRF-1, that specifically binds to IFN-beta gene regulatory elements. Cell **54:**903–913, 1988.
235. Ting JP-Y, Baldwin AS Jr. Regulation of MHC gene expression. Curr Opin Immunol **5:**8–16, 1993.
236. Sims SH, Cha Y, Romaine MF, Gao P-Q, Gottlieb K, Deisseroth AB. A novel interferon-inducible domain: Structural and functional analysis of the human interferon regulatory factor 1 gene promoter. Mol Cell Biol **13:**690–702, 1993.
237. Pine R. Constitutive expression of an ISGF2/IRF1 transgene leads to interferon-independent activation of interferon-inducible genes and resistance to virus infection. J Virol **66:**4470–4478, 1992.
238. Fujita T, Reis LF, Watanabe N, Kimura Y, Taniguchi T, Vilcek J. Induction of the transcription factor IRF-1 and interferon-beta mRNAs by cytokines and activators of second-messenger pathways. Proc Natl Acad Sci **86:**9936–9940, 1989.
239. Abdollahi A, Lord KA, Hoffman-Liebermann B, Liebermann DA. Interferon regulatory factor 1 is a myeloid differentiation primary response gene induced by interleukin 6 and leukemia inhibitory factor: Role in growth inhibition. Cell Growth Diff **2:**401–407, 1991.
240. Harroch S, Gothelf Y, Watanabe N, Revel M, Chebath J. Interleukin-6 activates and regulates transcription factors of the interferon regulatory factor family in M1 cells. J BiolChem **268:**9092–9097, 1993.
241. Willman CL, Sever CE, Pallavicini MG, Harada H, Tanaka N, Slovak ML, Yamamoto H, Harada K, Meeker TC, List AF, Taniguchi T. Deletion of IRF-1, mapping to chromosome 5q31.1, in human leukemia and preleukemic myelodysplasia. Science **259:**968–971, 1993.
242. Harada H, Kitagawa M, Tanaka N, Yamamoto H, Harada K, Ishihara M, Taniguchi T. Antioncogenic and oncogenic potentials of interferon regulator factors-1 and -2. Science **259:**971–974, 1993.
243. Kamijo R, Harada H, Matsuyama T, Bosland M, Gerecitano J, Shapiro D, Le J, Koh SI, Kimura T, Green SJ, Mak TW, Taniguchi T, Vilcek J. Requirement for transcription factor IRF-1 in NO synthase induction in macrophages. Science **263:**1612–1615, 1994.
244. Martin E. Nathan C, Xie Q-W. Role of interferon regulatory factor 1 in induction of nitric oxide synthase. J Exp Med **180:**977–984, 1994.
245. Chang C-H, Hammer J, Loh JE, Fodor WL, Flavel RA. The activation of major histocompatibility complex class I genes by interferon regulatory factor–1 (IRF-1). Immunogenetics **35:**378–384, 1992.
246. Johnson DR, Pober JS. HLA class I heavy-chain gene promote elements mediating synergy between tumor necrosis factor and interferons. Mol Cell Biol **14:**1322–1332, 1994.
247. Yamada G, Ogawa M, Akagi K, Miyamoto H, Nakano N, Itoh S, Miyazaki J-I, Nishikawa S-I, Yamamura K-I, Taniguchi T. Specific depletion of the B-cell population induced by aberrant expression of human interferon regulatory factor 1 gene in transgenic mice. Proc Natl Acad Sci **88:**532–536, 1991.
248. Matsuyama T, Kimura T, Kitagawa M, Pfeffer K, Kawakami T, Watanabe N, Kundig TM, Amakawa R, Kishihara K, Wakeham A, Potter J, Furlonger CL, Narendran A, Suzuki H, Ohashi PS, Paige CJ, Taniguichi T, Mak TW. Targeted disruption of IRF-1 or IRF-2 results in abnormal type I IFN gene induction and aberrant lymphocyte development. Cell **75:**83–97, 1993.
249. Tanaka N, Ishihara M, Kitagawa M, Harada H, Kimura T, Aizawa S, Mak TW, Taniguchi T. Cellular commitment to oncogene-induced transformation or apoptosis is dependent on the transcription factor IRF-1. Cell **77:**829–839, 1994.

250. Tamura T, Ishihara M, Lamphier MS, Tanaka N, Oishi I, Aizawa S, Matusyama T, Mak TW, Taki S, Taniguchi T. An IRF-1–dependent pathway of DBA damage-induced apoptosis in mitogen-activated T lymphocytes. Nature **376:**596–599, 1995.
251. Tanaka N, Ishihara M, Lamphier MS, Nozawa H, Matsuyama T, Mak TW, Aizawa S, Tokino T, Oren M, Taniguchi T. Cooperation of the tumour suppressor IRF-1 and p53 in response to DNA damage. Nature **382:**816–818, 1996.
252. Reis LFL, Ruffner H, Stark G, Aguet M, Weissmann C. Mice devoid of interferon regulatory factor 1 (IRF 1) show normal expression of type I interferon genes. EMBO J **13:**4798–4806, 1994.
253. Kimura T, Nakayama K, Penninger J, Kitagawa M, Harada H, Matsuyama T, Tanaka N, Kamijo R, Vilcek J, Mak TW, Taniguchi T. Involvement of IRF-1 transcription factor in antiviral responses to interferons. Science **264:**1921–1924, 1994.
254. Nelson N, Kanno Y, Hong C, Contursi C, Fujita T, Fowlkes BJ, O'Connell E, Hu-Li J, Paul WE, Jankovic, D, Sher AF, Coligan JE, Thornton A, Appella E, Yang Y, Ozato K. Expression of IFN regulatory factor family proteins in lymphocytes: Induction of Stat1 and IFN concensus sequence binding protein expression by T cell activation. J Immunol **156:**3711–3720, 1996.
255. Stevens AM, Yu-Lee L-Y. The transcription factor interferon regulatory factor–1 is expressed during both early G1 and the G1/S transition in the prolactin-induced lymphocyte cell cycle. Mol Endocrinol **6:**2236–2243, 1992.
256. Harada H, Willison K, Sakakibara J, Miyamoto M, Fujita T, Taniguchi T. Absence of the type I IFN system in EC cells: transcriptional activator (IRF-1) and repressor (IRF-2) genes are developmentally regulated. Cell **63:**303–312, 1990.
257. Matikainen S, Ronni T, Hurme M, Pine R, Julkunen I. Retinoic acid activates interferon regulatory factor–1 gene expression in myeloid cells. Blood **88:**114–123, 1996.
258. Stevens AM, Yu-Lee L-Y. Multiple prolactin-responsive elements mediate G1 and S phase expression of the interferon regulatory factor–1 gene. Mol Endocrinol **8:**345–355, 1994.
259. Dynan WS, Tjian R. The promoter-specific transcription factor Sp1 binds to upstream sequences in the SV40 early promoter. Cell **35:**79–87, 1983.
260. Dowdy SF, Hinds PW, Louie K, Reed SI, Arnold A, Weinberg RA. Physical interaction of the retinoblastoma protein with human D cyclins. Cell **73:**499–511, 1993.
261. Hu Q, Lees JA, Buchkovich KJ, Harlow E. The retinoblastoma protein physically associates with the human cdc2 kinase. Mol Cell Biol **12:**971–980, 1992.
262. Onate SA, Tsai SY, Tsai MJ, O'Malley BW. Sequence and characterization of a coactivator for the steroid hormone receptor superfamily. Science **270:**1354–1357, 1995.
263. Gill G, Pascal E, Tseng ZH, Tjian R. A glutamine-rich hydrophobic patchin transcription factor Sp1 contacts the dTAFII110 component of the Drosophila TFIID complex and mediates transcriptional activation. Proc Natl Acad Sci **91:**192–196, 1994.
263a. Bhattacharya S, Eckner R, Grossman S, Oldread E, Arany Z, D'Andrea A, Livingston DM. Cooperation of Stat2 and p300/CBP in signaling induced by interferon-α. Nature **383:**344–347, 1996.
263b. Zhang JJ, Vinkemeier U, Gu W, Chakravarti D, Horvath CM, Darnell JE, Jr. Two contact regions between Stat1 and CBP/p300 in interferon-γ signaling. Proc Natl Acad Sci **93:**15092–15096, 1996.
263c. Horvai A, Xu L, Korzus E, Brard D, Kalafus D, Mullen T-M, Rose DW, Rosenfeld MG, Glass CK. Nuclear integration of Jak/Stat and Ras/AP-1 signaling by CBP and p300. Proc Natl Acad Sci **94:**1074–1079, 1997.
264. Dou Q-P, Markell PJ, Pardee AB. Thymidine kinase transcription is regulated at G_1/S phase by a complex that contains retinoblastoma-like protein and a cdc2 kinase. Proc Natl Acad Sci **89:**3256–3260, 1992.
265. Tanaka N, Kawakami T, Taniguchi T. Recognition DNA sequences of interferon regulatory factor–1 (IRF-1) and IRF-2, regulators of cell growth and the interferon system. Mol Cell Biol **13:**4531–4538, 1993.
266. Sailer A, Nagata K, Naf D, Aebi M, Weissman C. Interferon regulatory factor–1 (IRF-1) activates the synthetic IRF-1–responsive sequence $(GAAAGT)_4$ in Saccharomyces cerevisiae. Gene Expression **2:**329–337, 1992.
267. Pleiman CM, Gimpel SD, Park LS, Harada H, Taniguchi T, Zeigler SF. Organization of the murine and human interleukin-7 receptor genes: Two mRNAs generated by differential splicing and presence of a type-I-interferon–inducible promoter. Mol Cell Biol **11:**3052–3059, 1991.
268. Girdlestone J, Ismat M, Gewert D, Milstein C. Transcriptional regulation of *HLA-A* and *-B:* Differential binding of members of Rel and IRF families of transcription factors. Proc Natl Acad Sci **90:**11568–11572, 1993.
269. Li-Weber M, Davydov IV, Krafft H, Krammer PH. The role of NF-Y and IRF-2 in the regulation of human IL-4 gene expression. J Immunol **153:**4122–4132, 1994.
270. Min W, Pober JS, Johnson DR. Kinetically coordinated induction of TAP1 and HLA Class I by IFN gamma. J Immunol **156:**3174–3183, 1996.
271. Coccia EM, Marziali G, Stellacci E, Perrotti E, Ilari R, Orsatti R, Battistini A. Cells resistant to interferon-β respond to interferon-gamma via the Stat1-IRF-1 pathway. Virology **211:**133–122, 1995.
272. Tanaka H, Samuel CE. Mechanism of interferon action: Structure of the mouse PKR gene encoding the interferon-inducible RNA-dependent protein kinase. Proc Natl Acad Sci **91:**7995–7999, 1996.
273. Kirchhoff S, Koromilas AE, Schaper F, Grashoff M, Sonenberg N, Hauser H. IRF-1 induced cell growth inhibition and interferon induction requires the activity of the protein kinase PKR. Oncogene **11:**439–445, 1995.
274. Beretta L, Gabbay M, Berger R, Hanash SM, Sonenberg N. Expression of the protein kinase PKR is modulated by IRF-1 and is reduced in 5q-associated leukemias. Oncogene **12:**1593–1596, 1996.
275. Sanceau J, Kaisho T, Hirano T, Wietzerbin J. Triggering of the human interleukin-6 gene by interferon gamma and tumor necrosis factor–alpha in monocytic cells involves cooperation between interferon regulatory factor–1, NFκB, and Sp1 transcription factors. J Biol Chem **270:**27920–27931, 1995.
276. Manzella L, Giuffrida MA, Pilastro MR, Girlando G, Picardi G, Malaguarnera L, Messina A. Possible role of the transcription factor interferon regulatory factor–1 (IRF-1) in the regulation of ornithine decarboxylase (ODC) gene expression during IFN gamma macrophage activation. FEBS Lett **348:**177–180, 1994.
277. Horiuchi M, Koike G, Yamada T, Mukoyama M, Nakajima M, Dzau VJ. The growth-dependent expression of angiotensin II type 2 receptor is regulation by transcription factors interferon regulatory factor–1 and –2. J Biol Chem **270:**20225–20230, 1995.
278. Decker T, Lew DJ, Darnell JE Jr. Two distinct alpha-interferon-dependent signal transduction pathways may contribute to activation of transcription of the guanylate-binding protein gene. Mol Cell Biol **11:**5147–5153, 1991.
279. Briken V, Ruffner H, Schultz U, Schwartz A, Reis LFL, Strehlow I, Decker T, Staeheli P. Interferon regulatory factor 1 is required for mouse Gbp gene activation by gamma interferon. Mol Cell Biol **15:**975–982, 1995.
280. Harroch S, Gothelf Y, Revel M, Chebath J. 5′ upstream sequences of MyD88, an IL-6 primary response gene in M1 cells:

Detection of functional IRF-1 and Stat factors binding sites. Nucleic Acids Res **23:**3539–3546, 1994.
281. Harada H, Takahashi E-I, Itoh S, Harada K, Hori T-A, Taniguchi T. Structure and regulation of the human interferon regulatory factor 1 (IRF-1) and IRF-2 genes: Implications for a gene network in the interferon system. Mol Cell Biol **14:**1500–1509, 1994.
282. Taniguchi T, Harada H, Lamphier M. Regulation of the interferon system and cell growth by the IRF transcription factors. J Cancer Res Clin Oncol **121:**516–520, 1995.
283. Neish AS, Read MA, Thanos D, Pine R, Maniatis T, Collins T. Endothelial interferon regulatory factor–1 cooperates with NF-(kappa)B as a transcriptional activator of vascular cell adhesion molecule 1. Mol Cell Biol **15:**2558–2569, 1995.
284. Massa PT, Wu HL. Interferon regulatory factor element and interferon regulatory factor–1 in the induction of major histocompatibility complex Class I genes in neural cells. J Interferon Cyto Res **15:**799–810, 1995.
285. Du W, Thanos D, Maniatis T. Mechanisms of transcriptional synergism between distinct virus-inducible enhancer elements. Cell **74:**887–898, 1993.
286. Stein B, Baldwin AS Jr., Ballard DW, Greene WC, Angel P, Herrlich P. Cross-coupling of the NFκB p65 and Fos/Jun transcription factors produces potentiated biological function. EMBO J **12:**3879–3891, 1993.
287. Stein B, Cogswell PC, Baldwin AS Jr. Functional and physical associations between NFκB and C/EBP family members: A Rel domain-bZIP interaction. Mol Cell Biol **13:**3946–3974, 1994.
288. Taneka N, Ishihara M, Taniguchi T. Suppression of c-myc or fosB-induced cell transformation by the transcription factor IRF-1. Cancer Lett **83:**191–196, 1994.
289. Peschon JJ, Morrissey PJ, Grabstein KH, Ramsdell FJ, Maraskovsky E, Gliniak BC, Park LS, Ziegler SF, Williams DE, Ware CB, Meyer JD, Davison BL. Early lymphocyte expansion is severely impaired in interleukin-7 receptor–deficient mice. J Exp Med **180:**1955–1960, 1994.
290. Sudo T, Nishikawa S, Ohno N, Akiyama N, Tamakoshi M, Yoshida H, Nishikawa S-I. Expression and function of the interleukin 7 receptor in murine lymphocytes. Proc Natl Acad Sci **90:**9125–9129, 1993.
291. van Santen HM, Woolsey A, Rickardt PA, Van Kaer L, Baas EJ, Berns A, Tonegawa S, Ploegh HL. Increase in positive selection of $CD8^+$ T cells in TAP1-mutant mice by human β2-microglobulin transgene. J Exp Med **181:**787–792, 1995.
292. Leff MA, Buckley DJ, Krumenacker J, Reed JC, Miyashita Y, Buckley AR. Rapid modulation of the apoptosis regulatory genes, bcl-2 and bax by prolactin in rat Nb2 lymphoma cells. Endocrinology **137:**5456–5462, 1996.