Janus kinase (JAK)-signal transducer and activators of transcription (STAT) signaling pathways play crucial roles in lymphopoiesis. In particular, JAK3 has unique functions in the lymphoid system such that JAK3 ablation results in phenotypes resembling severe combined immunodeficiency syndrome. This review focuses on the biochemistry, immunological functions, and clinical significance of JAK3. Compared with other members of the JAK family, the biochemical properties of JAK3 are relatively less well characterized and thus largely inferred from studies of JAK2. Furthermore, new findings concerning the cross-talks between Notch and JAK signaling pathways through ubiquitin-mediated protein degradation are discussed in more detail.

Keywords JAK3; Notch; Asb2; JAK2

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Introduction

Janus kinase (JAK)-signal transducer and activators of transcription (STAT) signaling pathways are instrumental for the differentiation, proliferation, and survival of a variety of cell types including those of the hematopoietic lineages. Extensive investigations into these pathways have led to the accumulation of a wealth of information. In this review, we will focus on the role of JAK3 in lymphopoiesis and the clinical significance of JAK3 dysfunction. JAK3 belongs to a subfamily of non-receptor tyrosine kinases, along with JAK1, JAK2, and Tyk2. These proteins share extensive sequence homologies and thus have similar biochemical properties. Knowledge about the regulation of one of the JAK proteins is most likely applicable to the other and hence the regulation of JAK3 function will be discussed in relation to other JAKs. More significantly, we will highlight recent discoveries about the regulation of JAK protein turnover controlled by Notch signaling pathways. We believe that the cross-talks between JAK-STAT and Notch signaling pathways will emerge to be crucial for diverse developmental processes and disease states.

JAK Protein Structure

In mammals, the four members of the JAKs—JAK1, JAK2, JAK3, and Tyk2—constitute one subgroup of the intracellular non-receptor tyrosine kinases and transduce cytokine-mediated signals through the JAK-STAT pathway. These genes encoding the four JAK family members are located on three separate chromosomes. The JAK1 and JAK2 genes are located on human chromosomes 1p31.3 and 9p24, and mouse chromosomes 4 and 19, respectively [1]. Interestingly, the gene coding for the first identified JAK family member, Tyk2, is located on human chromosome 19p13.2 and in tandem with the JAK3 gene on 19p13.1 [2,3]. Tyk2 and JAK3 are also adjacent to each other on mouse chromosome 8. However, of the four highly homologous JAK proteins, Tyk2 and JAK3 are most distantly related, suggesting that an ancient gene duplication event gave rise to the two genes, which then evolved independently.

Although JAK1, JAK2, and Tyk2 are expressed ubiquitously, JAK3 expression is restricted to hematopoietic lineage cells [4,5]. JAKs are relatively large proteins of >1100 amino acids with masses ranging from 120 to 140 kDa. From the primary structure, seven different JAK homology regions (JHs) have been identified (JH1–JH7), numbered from the carboxyl to the amino terminus (Fig. 1). These JHs form the putative structural domains of the JAK family members, which are conserved between species. The enzymatically active kinase domain (JH1–JH7), numbered from the carboxyl to the amino terminus (Fig. 1). These JHs form the putative structural domains of the JAK family members, which are conserved between species. The enzymatically active kinase domain (JH1) is located at the carboxyl-terminus. The kinase activity of these proteins depends on their phosphorylation at tyrosine residues in the activation loop of the kinase domain (e.g. Y1007 and Y776 in mouse JAK2 and JAK3, respectively) [6–8]. N-terminal to JH1 is the catalytically inactive pseudo-kinase domain, which represents a unique feature of JAK proteins in contrast to other protein tyrosine kinases. This tandem architecture of kinase domains gives...
them the name, Janus, derived from the two-faced Roman god of doorways. Despite the lack of catalytic activity, the pseudo-kinase domain, designated JH2, is required for suppression of basal activity of tyrosine kinases and for cytokine-inducible activation of signal transduction. Some mutations in the JH2 domain have been shown to positively or negatively regulate JAK kinase activity, thus resulting in human diseases [9–11]. For example, a single-point mutation (V617F) within the JH2 pseudo-kinase domain of JAK2 leads to constitutive activation of JAK2 and has been shown to exist in the majority of patients with polycythemia vera (PV) [12–15]. Similarly, a number of mutations in this domain of JAK3 have been found to enhance the enzymatic activity [16,17]. However, other mutations in the JH2 domain of JAK3 have been shown to augment the inhibitory effect of JH2 and suppress the catalytic activity, resulting in abrogation of interleukin (IL)-2 signaling leading to severe combined immunodeficiency diseases (SCID) [11,18]. Similarly, mutations in the JH2 domain of Tyk2 have been shown to interfere with Tyk2 activity [19,20]. The association of JH2 mutations with these disorders suggests that the JH2 domain have a critical regulatory function in JAK-mediated signaling. The amino terminus of JAK proteins is composed of a SH2-like domain (including JH3–JH4) and a 4.1, ezrin, radixin, moesin (FERM) homology domain (consisting of JH6–JH7). The FERM domain, which is 300 amino acids in length can interact with the kinase domain and positively regulate the catalytic activity. Mutations within the FERM domain of JAK3 have been shown to dampen or augment kinase activity [21,22]. In addition, the FERM domain is shown to be essential in mediating interactions between JAKs and their cognate cytokine receptors. For example, artificial JAK3 mutants containing only the FERM domains but not the other JAK3 domains effectively bind to the common γ chain (γc) [23]. Mutation of hydrophobic residues within the FERM domain of JAK1 interfered with its binding to the interferon-γ (IFN-γ) receptor [24]. In addition to the well-known V617F mutation found in patients with PV, substitution of a tyrosine residue for glutamate (Y613E) in the pseudo-kinase domain led to constitutive JAK2 activation in the absence of erythropoietin (Epo) stimulation [25]. Interestingly, the Y613E mutant still required the presence of the receptor for activation, suggesting that negative charges brought by phosphorylation of the tyrosine residue or glutamate is important for maintaining unbound JAK2 in an inactive state. Receptor binding through the FERM domain unlocks the potential to activate the kinase. The role of the Src Homology 2 (SH2)-like domain (JH3 and JH4) in JAK proteins is not well defined. A mutation on SH2 domain of JAK1 did not interfere with either its kinase activity or its receptor association [26].

**JAK Signaling Pathways**

JAK proteins are essential mediators of cytokine-triggered signal transduction and interact with a variety of cytokine receptors (Fig. 2). The binding of cytokines to the homodimeric or heterodimeric receptors prompts their aggregation, thus bringing the associated JAK proteins to close proximity. Ligand binding most likely induces a conformational change in the cytoplasmic domains of the cognate receptors, which allows the formation of a multimeric receptor complex. The adjacent JAK proteins can then phosphorylate each other, leading to activation of the kinases. Subsequently, they are able to phosphorylate tyrosine residues on the receptor and generate sites for interaction with proteins that contain phosphotyrosine-binding SH2 domains. Since cytosolic DNA-binding STAT proteins possess SH2 domains, they are recruited to the receptors and phosphorylated at tyrosine residues by JAKs. Different STAT proteins can then phosphorylate each other, leading to activation of the kinases. In response to cytokine stimulation, each JAK protein can bind to many different cytokine receptors, but certain specificities exist [27,28]. A large number of cytokine receptors, including those for IL-2, IL-4, IL-7, IL-9, IL-15, and IL-21, share a receptor subunit called common γc and transmit signals through JAK1 and JAK3. Since JAK3 only binds to γc, it is specifically responsive to these cytokines. In contrast, JAK1 is also essential for another family of cytokine receptors that uses gp130 as a co-receptor. JAK1 is also indispensable for signaling triggered by granulocyte colony-stimulating factor, IFNs, and members of...
the IL-10 family. JAK2 is involved in signaling from receptors of hormone-like cytokines and also transduces signals through gp130-containing receptors and some IFN receptors. In general, receptors for cytokines supporting lymphopoiesis tend to utilize JAK1 and JAK3 with a few exceptions [29]. In contrast, myeloid or erythroid lineage-promoting cytokine receptors primarily bind to JAK2. Unlike JAK1-3, Tyk2 is dedicated to IL-12 and IL-23 signaling in T cells [30–33].

The downstream effectors of JAK proteins are STAT transcription factors, which are activated by JAK-mediated phosphorylation. Seven STAT proteins exist in mammals and they are selectively targeted by different JAK proteins with overlapping specificities. Therefore, a large number of cytokines rely on a small number of JAK and STAT proteins for their specific signaling transmission, which demands intricate control mechanisms of the signaling pathways. The dependence of a particular JAK may also be proportional to the levels of JAK expression in a given situation. For example, JAK3 is primarily expressed in the lymphoid lineages and hence its function there [34].

Regulation of JAK Signaling

JAK proteins are subjected to multiple regulatory mechanisms through phosphorylation/dephosphorylation, inhibitor binding, and ubiquitin-mediated degradation [35,36]. As other tyrosine kinases, JAK family members are regulated by protein phosphorylation. For example, IL-2 stimulation leads to autophosphorylation of Y785 of JAK3 and enables the binding of the SH2 domain-containing adaptor protein SH2-Bβ, which inhibits basal but enhances cytokine-stimulated kinase activity of JAK3. Similarly, autophosphorylation of Y813, in response to growth hormone, is required for SH2-Bβ binding to JAK2 [37]. For most kinases, autophosphorylation within the activation loop usually stimulates kinase activity. For example, two tyrosine phosphorylation sites, Y904 and Y939, were identified to positively regulate human JAK3 kinase activity [38]. Both of these tyrosine residues are located within the JH1 kinase domain and undergo phosphorylation to activate kinase activity responding to stimulation by cytokines that signal through the JAK3-associated γc-containing receptors. However, phosphorylation at Y913 in JH1 kinase domain of JAK2 is an instance of negative regulation of JAK kinase activity [8]. Epo stimulation induces phosphorylation of Y913, and in turn inhibits its kinase activity and downstream STAT5 activation [39]. Autophosphorylation of Y119 within the FERM domain triggers the dissociation of JAK2 from the Epo receptor followed by kinase degradation [40]. These examples illustrate that the distinct tyrosine residues within the JAK family members play opposing roles and thus confer the complexity of phosphorylation-mediated regulation of JAK activity. Furthermore, dephosphorylation of JAK, carried out by protein tyrosine phosphatases (PTPs), also plays an important role in the regulation of JAK kinase activity. For example, Src homology 2 domain-containing protein tyrosine phosphatase (SHP2), a ubiquitously expressed SH2 domain-containing PTP has been shown to positively regulate JAK2/STAT5 pathway by dephosphorylating Y1007 in JAK2 and thus preventing the proteasome-mediated degradation of JAK2 [41]. On the other hand, SHP2 was also found to negatively control JAK1/STAT1/2 pathway by dephosphorylating STAT proteins [42]. Besides tyrosine phosphorylation, JAK proteins are also phosphorylated at serine or threonine residues. However, the role of this type of phosphorylation remains less well-understood, except that phosphorylation of S523 of JAK2 inhibits its kinase activity [43,44].

A further regulatory mechanism of JAK signaling involves the suppressor of cytokine signaling (SOCS) family, consisting of eight cytoplasmic proteins capable of negative feedback regulation. Initiation of the JAK-STAT signaling pathways activates transcription of its downstream target genes including the SOCS genes, which encode SOCS1-7 and cytokine inducible SH2-domain-containing protein. These proteins all possess a carboxyl-terminal SOCS box, a central SH2 domain, and an amino-terminal variable domain. The SOCS proteins exert their negative
regulations by three distinct mechanisms: JAK kinase inhibition, competition with STATs to bind receptors, and degradation of JAK proteins. SOCS1 and SOCS3 contain an additional domain, called kinase inhibitory region (KIR). KIR is located immediately N-terminal of the SH2 domain. This 12 amino acid region seems to act as pseudo substrate to bind to the activation loop of tyrosine kinases and thus inhibit JAK activity [45,46]. SOCS1 is found to physically interact with JAK2 and JAK3 and directly inhibit the kinase activity of JAK2 and JAK3 [47]. Several reports have shown that SOCS proteins function as E3 ubiquitin ligases through binding to Elongin B/C/Cullin5 [48,49]. The SOCS box in SOCS1 has been demonstrated to be essential for the association of SOCS1 with the Elongin B/C complex as well as ubiquitination of JAK2 [50]. The SH2 region of SOCS1 is shown to be required for the binding to the kinase domain (JH1) of JAK2 [51]. Giordanetto and Kroemer [52] predicted the 3D structures of the complex between JAK2 and SOCS1 using computer modeling, and highlighted potential interacting residues, such as Y1007 and K1011 of JAK2 mediating the proposed interface between SOCS1-SH2 and JAK2 activation loop. Therefore, numerous regulatory mechanisms are integrated within JAK proteins to exquisitely control JAK activation.

Roles of JAK-mediated Cytokine Signaling in the Development of the Immune System

Cytokines play pivotal roles in regulating lymphopoiesis and immune responses. Although all four JAK family members are crucial for transmitting the signals from cytokines, JAK3 has the most discrete function, as it only associates with the γc-containing receptors including IL-2R, IL-4R, IL-7R, IL-9R, IL-15R, and IL-21R. Despite the involvement of all JAKs in transmitting the signals from cytokines responsible for T-cell development, proliferation, or differentiation, JAK3 plays an essential role. IL-7 signaling is absolutely indispensable for the development of mouse T and B lymphocytes from hematopoietic progenitor cells. Both patients and mice lacking the γc receptor suffer from SCID. In humans, SCID represents the absence of T and natural killer (NK) cells and the abnormal function of B cells. The phenotypes in mouse are similar except that B-cell development is severely blocked. Since JAK3 associates with the γc chain, it is not surprising that JAK3-deficient mice have SCID that resembles γc knock-out mice, in which both B- and T-cell development is blocked and lymph nodes are lacking [34,53,54]. Disruption of IL-7 signaling could primarily account for these defects. Another γc-containing receptor, IL-15R, is essential for directing progenitors to commit to the NK lineage. Consequently, the numbers of NK cells are dramatically reduced in JAK3-deficient mice and also in SCID patients with JAK3 mutations [55]. As expected, selective reconstitution of JAK3 in the thymus restored the development of both αβ and γδ T cells as well as NK cells in JAK3−/− mice [53].

JAK3-deficient peripheral T cells, generated by rescuing thymic T-cell development with a JAK3 transgene driven by the lck promoter, which functions in the thymus but not the periphery, had a significantly dampened cytokine responsiveness and proliferation, whereas they demonstrated an activated memory phenotype and failed to respond to further stimuli [53,56]. For example, although JAK3-deficient T cells can be activated in vitro by CD3 plus CD28 stimulation, they secreted greatly reduced amounts of IL-2, failed to proliferate, and had the propensity to undergo apoptosis [57]. Since IL-2 signaling and STAT5 activation are obstructed in the absence of JAK3, CD25 or forkhead box protein 3 (FoxP3) expression were severely blocked in these mice [58]. Lymphopenia coupled with the reduction in FoxP3-expressing regulatory T cells might explain the activated memory phenotype of CD4+ T helper (Th) cells in JAK3−/− mice. Several factors may account for the proliferation failure of JAK3-deficient CD4+ T cells. First, signaling is impaired from receptors for IL-2 and IL-7, two major cytokines essential for T-cell proliferation. JAK3-deficient Th cells also appear to be anergic since they express surface markers, such as programmed death-1 and lymphocyte activation gene-3. Finally, anti-proliferative cytokines, IL-10, and transforming growth factor-β, are elevated [58].

JAK3 signaling is not only critical for T-cell development and survival but also instrumental for Th cell differentiation. For example, JAK3-dependent Th2 differentiation of CD4+ T cells is likely mediated through signaling triggered by IL-4, which is known as the most important inducer for Th2-cell differentiation. The absence of JAK3 abrogates IL-4 signaling, which leads to failures in STAT6 activation and in turn GATA binding protein 3 induction [59,60]. In contrast to the effect of JAK3 on Th2 cytokine signaling, JAK3 influences Th1-cell differentiation via epigenetic mechanisms [61]. JAK3-mediated STAT5 activation plays a role to promote chromatin remodeling of the IFN-γ locus. Without JAK3, the binding of T-bet to the IFN-γ promoter is reduced, leading to impaired production of the Th1-skewing cytokine [61].

Comparison among JAK3 and γc-single or double knock-out mice displayed no distinguishable phenotypes, suggesting that JAK3 cannot be substituted by other JAKs to transduce signals originated from the γc receptor in the immune response [62]. Interestingly, a similar phenotype was observed in STAT5−/− mice [63]. On the other hand, a constitutively active JAK3 mutant (A572V) induced lymphoproliferative syndromes in murine bone marrow transplantation models [64]. Taken together, it appears that the
JAK3-STAT5 axis plays an essential and non-redundant role in the immune system.

**Notch-induced JAK Protein Degradation and Its Biological Significance**

Both JAK-STAT and Notch signaling pathways play crucial roles in lymphocyte differentiation and leukemogenesis. A direct cross-talk between these two pathways had not been well documented until the recent discoveries that Notch signaling stimulates ubiquitin-mediated degradation of JAK proteins [65]. It was initially found that activation of the Notch signaling pathways by exposure to Notch ligands or by expression of a constitutively active form of Notch1 accelerated the degradation of both JAK2 and JAK3 in a variety of hematopoietic lineage cells [65]. Interestingly, Notch-induced JAK degradation is ubiquitin-mediated and mitogen-activated protein (MAP) kinase dependent.

Since B and T lineage cells have very different levels of basal MAP kinase activities, Notch signaling promotes JAK turnover in B but not in T cells. Activation of MAP kinases in T cells enables Notch to stimulate JAK degradation, whereas inhibition of the kinase activities rescues JAK proteins in B lineage cells. These observations led to the postulation that the differential effects of Notch on protein turnover in B and T cells contribute to the B versus T lineage decision [65]. Notch is known to promote T-cell development while inhibiting B-cell differentiation. Notch1-deficient mice accumulate B cells in the thymus and fail to produce T cells [66,67]. Conversely, overexpression of activated Notch1 in the bone marrow results in extra thymic T-cell development [68]. Whether B and T cells are derived exclusively from the same population of common progenitors is a debatable issue, but it is plausible that Notch signaling exerts its suppressive effect on B-cell differentiation through MAP kinase-dependent degradation of proteins critical for B lymphopoiesis, such as E2A transcription factors and JAK proteins [65,69]. To test this hypothesis, Nie et al. created knock-in mice carrying the E2A gene with point mutations that render the encoded proteins resistant to Notch-induced degradation. To overcome Notch-mediated JAK degradation, a constitutively active form of STAT5 was co-expressed with the E2A mutants. Interestingly, the thymuses of these mice displayed a large number of B lineage cells, thus supporting the notion that Notch signaling suppresses B lymphopoiesis through enhancing the degradation of proteins involved in B-cell development [65]. Notch signaling has also been shown to be involved in numerous additional lineage decisions and whether its function in controlling protein stability play a role awaits verification.

It has been shown that the effects of Notch on protein turnover requires its DNA-binding partner, RBP-Jk, suggesting that transcriptional activation of downstream target genes is necessary [69]. Indeed, activation of Notch signaling stimulates transcription of the gene encoding ankyrin repeat SOCS box containing protein 2 (Asb2), which represents an 18-member SOCS family [70]. Asb2 has previously been reported to interact with Elongin B and Elongin C, as well as Cullin5. It has been demonstrated that Asb2 is able to interact with Skp2, an F-box containing protein that serves as a substrate-recruiting subunit of the Cullin1-based E3 ligase complexes, SCFskp2 [70–72]. In doing so, Asb2 facilitates the assembly of a non-canonical heterodimeric complexes consisting of both Cullin1 and Cullin5 plus their respective associated subunits and E2 enzymes (Fig. 2). Therefore, these dimeric complexes may be catalytically superior to conventional monomeric complexes, leading to enhanced ubiquitination of their substrates.

JAK2 has previously been shown to be ubiquitinated by Elongin-Cullin-SOCS box (ECS) E3 ligase complexes, which contain Elongin B, Elongin C, Cullin5, and SOCS1 [7,50]. SOCS1 is thought to bind to the activation loop of JAK2 through its SH2 domain [51,73]. JAK2 has been shown to associate with either SOCS1 or Asb2 in a mutually exclusive manner, which suggests that there exist two types of ECS complexes, ECS^SOCS1 and ECS^Asb2 [70]. Moreover, the ECS^Asb2 complex can join with SCF^skp2 to form dimeric complexes. Asb2 has strong affinities to both JAK2 and JAK3. In contrast, it associates with other proteins such as E2A transcription factors through Skp2. Several domains in JAK3 have been found to independently interact with Asb2 but the kinase domain of JAK3 appears to primarily associate with Skp2 [74]. Interestingly, a point mutation in the kinase domain diminishes the interaction between Skp2 and JAK3 and consequently renders JAK3 resistant to degradation induced by Notch, Asb2, or Skp2, which lends further support to the notion that Asb2 fosters the formation of the dimeric E3 ligase complexes by interacting with Skp2 and its associates like Skp1 and Cullin1, as well as by binding to Elongins and Cullin5. Additional evidence for the dimeric complexes comes from the data demonstrating the dependence of Notch or Asb2-induced JAK2 and JAK3 degradation on both Cullin1 and Cullin5 [70,74].

Considering the distinct cell differentiation processes controlled by Notch signaling pathways, it is possible that Notch employs a global regulatory mechanism through which it efficiently enhances the ubiquitination and degradation of a diverse array of substrates and thus exerts its biological effects. By activating the transcription of Asb2 as well as Skp2 [70,75], Notch signaling could facilitate the formation of the dimeric E3 ligase complexes, which are predicted to influence the ubiquitination of a large number of substrates, for example, those previously known to be
modified by SCFSkp2 or ECS complexes. The ability of Asb2 to interact with F-box proteins beside Skp2 may potentially lengthen the list of substrates targeted by Notch to a great extent.

**JAK3 in Human Diseases**

SCID indicates defects in both humoral and cellular immunities mediated by B and T cells, respectively. SCID is rare but fatal. Infants with SCID have very high mortality risks during the first 2 years of life. To survive in early life, patients with SCID typically lack almost all immune defenses, develop life-threatening infections, and require major treatment such as bone marrow transplantation. SCID has been shown to be due to the genetic defects in several different genes. Since SCID was initially observed mainly in boys, the disease was thought to be related to chromosome X (X-SCID). Indeed, in most cases, mutations of the IL-2 receptor γ (IL-2Rγ), encoded by the X-linked gene, was found in patients with SCID [76]. Phenotypically, patients with X-SCID had profound reduction in T and NK cell numbers as well as in B-cell function. In contrast, patients with IL-2 deficiency had normal T-cell counts, which suggested that IL-2Rγ has additional functions in addition to IL-2 signaling. The IL-2Rγ chain was subsequently described as a functional component in IL-7 and IL-4 receptor complexes [77–79]. IL-2Rγ is thus called the common γc. In the following years, γc was also found to be the indispensable subunit of receptor complexes for IL-9, IL-15, and IL-21. Although IL-2, IL-7, IL-9, and IL-15 are essential for B/T/NK-cell proliferation and survival, IL-4 and IL-21 mainly regulate B-cell function, including immunoglobulin production. The fact that the IL-2Rγ chain is shared by a wide variety of cytokines explained the severe consequences in T and NK-cell development as well as the impairment of B-cell function in patients with X-SCID.

Although mutations of the γc gene accounted for almost half of all known cases of SCID, a considerable proportion of SCID remained unexplained. Later, some patients were found to display non-X-linked autosomal-recessive forms of SCID, raising the possibility of other genetic causes of the disease. Given the selective association of JAK3 with γc chain, loss of function mutations in the JAK3 gene might lead to autosomal-recessive SCID. Indeed, analysis of patients with such diseases revealed homozygous mutations of the JAK3 gene in 7%–14% of patients with heritable SCID [80,81]. The majority of described JAK3 mutations either impairs protein expression and/or stability or disrupts JAK3 function. Since patients with JAK3-SCID have diseases limited to immune cells, transplantation of hematopoietic stem cells is by far the best treatment based on its high survival rates.

On the other hand, gain-of-function mutations of the JAK3 genes have been found in patients with acute megakaryoblastic leukemia, lymphoproliferative diseases, and adult T-cell leukemia [16,17,22]. Although most of the mutations occur in the JH2 or pseudo-kinase domain, mutations have also been found in the FERM domain. Taken together, it is clear that JAK3 activities must be carefully gauged within a narrow range to maintain healthy lymphoid functions.

**The Clinical Significance for Targeting JAK3**

In principle, all JAK family members could be therapeutic targets in different cases. However, given the very restricted function of JAK3, as exemplified by the situation found in JAK3-SCID patients, specifically interfering with JAK3 function would be a good strategy for developing a novel class of immunosuppressive agents or anti-cancer drugs. Among the members of JAK family, the expression level of JAK3 is the highest in hematopoietic cells and it associates only with the γc chain, a subunit of the receptors for IL-2, IL-4, IL-7, IL-9, IL-15, and IL-21. As a result, lack of JAK3 leads to immunodeficiency but not other abnormalities. Therefore, selectively targeting JAK3 has distinct advantages over the current immunosuppressive drugs such as calcineurin inhibitors, Cyclosporin A or tacrolimus. These drugs are effective and directed against ubiquitous targets. However, despite their efficacy, these drugs have many adverse effects such as renal and metabolic toxicities.

Several inhibitors of tyrosine kinases have been shown to be able to diminish JAK3 activities, but they are not selective and have unfavorable side effects [82–85]. An orally available, selective JAK3 antagonist, CP-690,550 (now known as tofacitinib), has recently been generated and shown to have nanomolar potency [86]. The in vivo effect of CP-690,550 was first tested in two animal models of organ graft rejection, a murine heterotrophic heart transplant model and a primate renal transplant model. The results showed that CP-690,550 efficaciously prevented heart or kidney rejection after transplantation and thus prolonged graft survival. In addition, metabolic abnormalities or severe side effects due to immune suppression were not observed [87]. Although transient and significant reduction in lymphocyte subsets was observed in a dose-dependent manner, only mild anemia but not granulocytopenia or thrombocytopenia was noted, suggesting that JAK2 antagonism is minimal for this drug. Besides, lymphocyte numbers began to normalize after dosing cessation [88]. In cynomolgus monkeys, a trend in the reduction of CD8⁺ T cells and a modest decline in NK cells were observed, but no significant decline in total T lymphocytes [89].
The decrease in the numbers of NK cells and T cells as well as lower levels of IFN-γ have been suggested to contribute to the prevention of graft rejection and prolonged survival of kidney allografts in CP-690,550-treated cynomolgus monkeys [90,91].

In view of these pre-clinical results, what is the clinical application of JAK3 inhibitors? Considering the restricted expression pattern and function of JAK3 in lymphoid tissues, these inhibitors are mostly suitable to diseases in which lymphocytes are key players. Undoubtedly, transplant rejection would be an important application. For example, these drugs have been shown to benefit renal transplant recipients who suffer from serious toxicities of calcineurin inhibitors [92]. In addition, JAK3 inhibitors may also be valuable in treating autoimmune diseases, because memory T cells are thought to be major contributors to the persistence of autoimmune disease and JAK3 inhibition has been shown to prevent cartilage damage in mice with collagen-induced arthritis and in rats with adjuvant-induced arthritis [93]. In the animals with experimental rheumatoid arthritis, the treatment with CP-690,550 was thought to decrease IL-6 production, a critical cytokine that drives inflammatory destruction in rheumatoid arthritis [94]. Promising clinical trials are also underway for rheumatoid arthritis [95–97].

JAK3 inhibitors can be a double-edged sword, because the use of JAK3 inhibitors is always associated with increased risk of infection as found in the study of renal transplantation [93]. Furthermore, it is also possible that additional unknown functions of JAK3 might exist and the inhibitors could have unexpected side effects. Therefore, a thorough understanding of the mode of JAK3 action is highly important for better utilizing JAK3 inhibitors like CP-690,550.

Conclusions

JAK-STAT signaling pathways are major players in controlling cellular differentiation and proliferation processes in almost all eukaryotic organisms. Likewise, Notch signaling pathways are also pivotal actors and their influence on the stability of JAK proteins adds a new dimension and complexity to the regulatory network. This is not only very important for the development of an organism, but also highly significant for the pathogenesis of various illnesses. For example, it has recently been shown that disabling Notch activation in mouse hematopoietic stem cells results in an aberrant expansion of granulocyte/monocyte progenitors, extramedullary hematopoiesis, and induction of chronic myelomonocytic leukemia (CMML)-like diseases [98]. Mutations potentially dampening Notch signaling have also been found in a fraction of CMML patients. It remains to be tested whether the tumor suppressor function of Notch involves alterations in JAK-STAT signaling, which plays critical roles in myeloid differentiation and expansion. The availability of Notch-resistant mutants of JAK2 and JAK3 will greatly aid this line of investigation.

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References


41 Waihobi LW, Ahmed CM, Mujtaba MG, Flowers LO, Martin JP, Haider MI and Johnson HM. Both the suppressor of cytokine signaling 1...


57 Thomis DC, Lee W and Berg LJ. T cells from JAK3-deficient mice have intact TCR signaling, but increased apoptosis. J Immunol 1997, 159: 4708–4719.


65 Nie L, Xue M, Vladimirova A and Sun XH. Notch-induced E2A ubiquitination and degradation are controlled by MAP kinase activities. EMBO J 2003, 22: 5780–5792.


69 Nie L, Xu M, Vladimirova A and Sun XH. Notch-induced E2A ubiquitination and degradation are controlled by MAP kinase activities. EMBO J 2003, 22: 5780–5792.


