Cytokine 118 (2019) 48–63

Contents lists available at ScienceDirect

Cytokine

journal homepage: www.elsevier.com/locate/cytokine

Review article

The regulation of JAKs in cytokine signaling and its breakdown in disease

Henrik M. Hammarén[a,*], Anniina T. Virtanen[a], Juuli Raivola[a], Olli Silvennoinen[a,b,c]

[a] Faculty of Medicine and Life Sciences, University of Tampere, Arvo Ylpön katu 34, FI-33014 Tampere, Finland
[b] Clinical Hematology, Department of Internal Medicine, Tampere University Hospital, Medisiinarinkatu 3, FI-33520 Tampere, Finland
[c] Fimlab Laboratories, Pirkanmaa Hospital District, Tampere, Finland

ARTICLE INFO

Keywords:
Janus kinase (JAK)
Cytokine signaling
Myeloproliferative neoplasm (MPN)
Cancer
Mutation

ABSTRACT

The JAK–STAT signal transduction pathway is responsible for mediating signals of over fifty cytokines, growth factors and hormones. Signaling through the JAK–STAT pathway is regulated on multiple levels, including intramolecular regulation by the JAK pseudokinase domain, and intermolecular regulation by a host of regulatory proteins. The advent of accessible genomic tools have provided a wealth of information on disease-associated mutations in the JAK–STAT pathway and its regulatory components. The vast number of these mutations in diseases ranging from immunodeficiencies and obesity to many cancers highlight the importance of correct regulation of JAK–STAT signaling for biological processes such as hematopoiesis, regulation of the immune system, metabolism, and growth. Simultaneously, JAK inhibitors are gaining traction in clinical use, both for treatment of diseases driven by JAK mutations, and for a host of inflammatory disorders, in which proinflammatory cytokine signaling through the JAK–STAT pathway is an integral part of pathogenesis. The elucidation of molecular mechanisms in the pathogenesis of complex diseases has also, however, brought the limitations of our current understanding on the regulation of cytokine signaling to the foreground. Indeed, deeper understanding of these regulatory mechanisms are a prerequisite for the development of the next generation of pharmacological modulators of the JAK–STAT pathway. In this review we discuss the current state of knowledge of the intra- and intermolecular regulation of the JAK–STAT pathway, with a focus on diseases arising from disruptions in the regulatory apparatus.

1. Introduction

Hematopoietic cytokines play a critical role in orchestrating fundamental processes such as the immune response and hematopoiesis, but also cell differentiation and growth. Thereby it is not surprising that the central cytokine signaling pathway through Janus kinases (JAKs) and signal transducer and activator of transcription (STAT) transcription factors has been shown to either cause or participate in

Abbreviations: AL, Activation loop; ALL, Acute lymphoblastic leukemia; AML, Acute myeloid leukemia; AMKL, Acute megakaryoblastic leukemia; βc, Beta common; B-ALL, B-cell acute lymphoblastic leukemia; BCR, B-cell receptor; BSF-3, B-cell stimulating factor-3; CALR, Calreticulin; CLC, Cardiotrophin-like cytokine; CLCF1, Cardiotrophin-like cytokine factor 1; CLF, Colony-stimulating factor; CML, Chronic myeloid leukemia; CNTF, Ciliary neurotrophic factor; CNTFRα, Ciliary neurotrophic factor receptor α subunit; CRLF2, Cytokine receptor-like factor 2; CSF3R, Granulocyte colony-stimulating factor receptor; CT-1, Cardiotrophin 1; del, Deletion; DS-AMKL, Down syndrome AMKL; EBI3, Epstein-Barr virus induced gene 3; EGFR, Epidermal growth factor; EPO(R), Erythropoietin (receptor); ET, Essential thrombocythemia; ETV6, Ets variant 6 (TEL); ETP-ALL, Early T-cell precursor acute lymphoblastic leukemia/lymphoma; FERM, Band 4.1 protein, ezrin, radixin and moesin; fs, Frame-shift; γc, Gamma common; GCSFR, Granulocyte colony-stimulating factor receptor; GH(R), Growth hormone (receptor); GLMR, GP130-like monocyte receptor; GM-CSF-Rα, Granulocyte-macrophage colony-stimulating factor receptor α; GOF, Gain-of-function; gp130, Glycoprotein 130; HSE, Herpes simplex encephalitis; IFN, Interferon; IFNAR1, Interferon alpha/beta receptor 1; IFNGR1, Interferon gamma receptor 1; IFNLR1, Interferon lambda receptor 1; IL, Interleukin; IMF, Idiopathic myelofibrosis; JAK, Janus kinase; JH1/2, JAK homology 1/2; JMML, Juvenile myelomonocytic leukemia; LEPR, Leptin receptor; LIF, Leukemia inhibitory factor; LEPR, Leptin receptor; LIFRβ, Leukemia inhibitory factor receptor β; LOF, Loss-of-function; mda7, Melanoma differentiation associated gene-7; MDS, Myelodysplastic syndromes; MPL, Myeloproliferative leukemia protein, a.k.a. TPOR; MPN, Myeloproliferative neoplasms; MS, Multiple sclerosis; NF1-MPNSTs, Neurofibromatosis type 1-associated malignant peripheral nerve sheath tumors; NF-E2, Nuclear factor, erythroid 2; NKTCL, NK/T-cell lymphoma; NNT-1, Novel neurothrophin-1; NSCLC, Non-small cell lung cancer; OPR, OB-receptor; OSMRβ, Oncostatin M receptor β; PCM1, Pericentriolar Material 1; PH, Pleckstrin homology; PIAS, Protein inhibitor of activated STAT; PID, Primary immune deficiency; PMF, Primary myelofibrosis; PRL(R), Prolactin (receptor); PTK, Protein tyrosine kinase; PTP, Protein tyrosine phosphatase; PTPRC/T, Protein tyrosine phosphatase receptor type C/T; PTPBL, Basophil-like PTP; PV, Polycythemia vera; RA, Rheumatoid arthritis; RPN1, Ribophorin 1; RTK, Receptor tyrosine kinase; RUNX1, Runt-related transcription factor 1; SCID, Severe combined immunodeficiency; SH2, Src homology 2; SHP2, Protein tyrosine phosphatase, non-receptor type 11; SLE, Systemic lupus erythematosus; SOCS, Suppressors if cytokine signaling; SPAG9, Sperm associated antigen 9; SSBP2, Single-stranded DNA-binding protein 2; STAT, Signal transducer and activator of transcription; STRN3, Striatin 3; T-ALL, T-cell acute lymphoblastic leukemia; TCCR, T-cell cytokine receptor; TCPTP, T-cell protein tyrosine phosphatase; TEL, Ets variant 6 (ETV6); TM, Transmembrane; T-PLL, T-cell prolymphocytic leukemia; TPO(R), Thrombopoietin (receptor); TSLP(R), Thymic stromal lymphopoietin (receptor); TYK2, Tyrosine kinase 2

* Corresponding author.
E-mail address: henrik.hammaren@uta.fi (H.M. Hammarén).

https://doi.org/10.1016/j.cyto.2018.03.041
Received 30 November 2017; Received in revised form 29 March 2018; Accepted 30 March 2018
Available online 21 May 2018

Table 1
Type I and II cytokines, their receptor chain configurations, as well as the corresponding JAKs and STATs are shown. JAKs and STATs critical for signaling identified with the highest confidence are shown in **bold**, associated JAKs and STATs for which the data is weaker are shown in parentheses. Cytokines that signal through the same receptor-JAK–STAT configuration are separated by commas. Synonyms are separated by a slash. In case of three or more often-used synonyms and for additional explanations, see footnotes.

		Cytokine	Receptor chain(s)			JAKs	STATs	References
TYPE II CYTOKINE RECEPTORS	IFN family	IFN-I (typeI)*	IFNAR1		IFNAR2	**JAK1**, TYK2	**STAT1, STAT2**, STAT3, STAT4 (STAT5, STAT6)	[154,155]
		IFN-γ (typeII)	IFNGR1		IFNGR2	**JAK1, JAK2**	**STAT1**	[150,154,155]
		IL-28a, IL-28b, IL-29‡	IL-28R/ IFNLR1		IL-10Rβ	**JAK1**, TYK2	**STAT1, STAT2,** STAT3, STAT5	[154,156–158]
		IL-10	IL-10Rα		IL-10Rβ	**JAK1, TYK2**	**STAT3**, STAT1	[154–156]
		IL-19	IL-20Rα		IL-20Rβ	JAK1, JAK2	**STAT3,** STAT1	[150,154,156]
		IL-20, IL-24/mda7	IL-20Rα or IL-22R		IL-20Rβ	JAK1, JAK2	**STAT3,** STAT1	[150,151,154,156]
		IL-22/IL-TIF†	IL-22R		IL-10Rβ	JAK1, TYK2	**STAT3,** STAT1, (STAT5)	[150,154]
		IL-26/AK155	IL-20Rα		IL-10Rβ	JAK1, TYK2	**STAT3,** STAT1	[151,154,156]
TYPE I CYTOKINE RECEPTORS	gp130 family	IL-6	IL-6Rα		gp130	**JAK1,** JAK2, TYK2	**STAT3,** STAT1	[90,154,155]
		IL-11	IL-11Rα		gp130	**JAK1,** JAK2, TYK2	**STAT3,** STAT1	[90,154,155]
		LIF	LIFRβ		gp130	**JAK1,** JAK2, TYK2	**STAT3,** STAT1	[90,154,155,159]
		CNTF	CNTFRα	LIFRβ	gp130	JAK1, (JAK2, TYK2)	**STAT3,** (STAT1)	[90,154,155,160]
		CLCF1§, NP	CNTFRα	LIFRβ	gp130	JAK1, (JAK2)	**STAT3,** STAT1	[90,154,160]
		CT-1	CNTFRα	LIFRβ	gp130	JAK1, (JAK2, TYK2)	**STAT3**	[90,154,155]
		OSM	OSMRβ or LIFRβ		gp130	JAK1, (JAK2, TYK2)	**STAT3,** STAT1	[90,154,155,159]
		IL-31	IL-31Rα/ GLMR		OSMRβ	JAK1, (JAK2)	STAT3, STAT5, STAT1	[154,161]
		G-CSF		GCSFR/ CSF3R		JAK1, (JAK2)	**STAT3**	[90,154,155]
		Leptin		LEPR/OBR		JAK2	**STAT3**	[154]
		IL-12 (p35 + p40)	IL-12Rβ2		IL-12Rβ1	**TYK2,** JAK2	**STAT4**	[153–155]
		IL-23 (p19 + p40)	IL-23R		IL-12Rβ1	**TYK2,** JAK2	**STAT3, STAT4,** STAT1	[153,154]
		IL-27 (p28 + EBI3)‖	IL-27Rα¶		gp130	JAK1, JAK2, TYK2	**STAT1, STAT3**, STAT4, (STAT5)	[90,153,154]
		IL-35 (p35 + EBI3)#	IL-12Rβ2		gp130	JAK1, JAK2	STAT1, STAT4	[153,154]
	γ_c family	IL-2	IL-2Rα	IL-2Rβ	γ_c	**JAK1, JAK3,** (JAK2)	**STAT5,** (STAT3)	[90,154,155,162]
		IL-4	IL-4Rα		γ_c	**JAK1, JAK3**	**STAT6**	[90,154,155,162]
		IL-7	IL-7Rα		γ_c	**JAK1, JAK3**	**STAT5,** (STAT3)	[90,154,155,162]
		IL-9	IL-9Rα		γ_c	JAK1, JAK3	**STAT5,** STAT3	[90,154,155,162]
		IL-15	IL-15Rα	IL-2Rβ	γ_c	**JAK1, JAK3**	**STAT5,** (STAT3)	[90,154,162]
		IL-21	IL-21R		γ_c	JAK1, JAK3	**STAT3,** STAT5, (STAT1)	[90,154,162]
		TSLP	IL-7Rα		TSLPR/ CRLF2	JAK1, JAK2	STAT1, STAT3, STAT4, STAT5, STAT6	[154,162,163]
		IL-13	IL-4Rα		IL-13R	JAK1, JAK2, TYK2	**STAT6,** (STAT3)	[154,155,162]
	IL-3/β_c	IL-3	IL-3Rα		β_c (gp140)	JAK2, JAK1	**STAT5**, STAT3	[90,154,155,164]
		IL-5	IL-5Rα		β_c (gp140)	JAK2	**STAT5**, STAT1, STAT3	[90,154,155]
		GM-CSF	GM-CSF-Rα		β_c (gp140)	JAK2	**STAT5**	[90,154,155]
	Single chain	EPO	EPOR			**JAK2**	**STAT5**	
		GH	GHR			**JAK2**	**STAT5,** (STAT3)	
		PRL	PRLR			**JAK2**	**STAT5**	
		TPO	TPOR/MPL			**JAK2**	**STAT5**	

* In humans this family consists of 12 IFN-αs, IFN-ω and Limitin (a.k.a. IFN-ζ).
† IL-22 also has a soluble receptor IL-22BP, which probably works as an agonist *in vivo* [150,151].
‡ Interleukins 28 and 29 are also called Type III IFNs, or IFN-λs, as follows: IL-29/IFN-λ1, IL-28a/IFN-λ2, and IL-28b/IFN-λ3.
§ a.k.a. CLC, CLF, NNT-1, BSF-3; CLCF1 is secreted either with sCNTFR or CRLF1.
‖ IL-27 p28 subunit is also called IL-30, which might signal through IL-6Rα [152].
¶ a.k.a. WSX-1, TCCR.
# IL-35 has also been reported to signal through IL-12Rβ2 or gp130 homodimers [153].

tumorigenesis. The first demonstration of this were the identification of rare oncogenic fusions involving the JAK kinase domain, e.g., the TEL/ETV6-JAK2 translocation resulting in leukemia driven by constitutively dimeric and active JAK2 kinase domain [1]. Subsequently, several other JAK2 fusions have been identified [2] as well as JAK2 gene amplifications in lymphomas [3] and triple-negative breast cancer [4]. Nevertheless, somatic JAK driver mutations are most commonly found in hematological malignancies, where a single oncogenic JAK2 mutation, JAK2 V617F, underlies more than half of all classical myeloproliferative neoplasm (MPN) cases (reviewed in [5,6]). JAK mutations are also common in leukemia, where, e.g., somatic JAK1 mutations are found in 10–20% of T-cell acute lymphoblastic leukemia (T-ALL) and JAK2 mutations in ~ 20% of down syndrome-associated B-ALL [5].

Recently, (epi)genetic changes in JAKs, STATs, as well as regulatory components of the pathway have also been increasingly linked to other

cancers. Examples include hepatitis B-associated hepatocellular carcinoma (JAK1 mutations in ~9% of patients), large granular lymphocytic leukemia (STAT3 mutations in 40% of patients), prostate cancer (amplification of *STAT5A/B* locus) and non-small cell lung cancer (NSCLC, hypermethylation of suppressor of cytokine signaling (SOCS) 3 promoter) (reviewed in [7]). Furthermore, several cytokines, particularly interleukin (IL-)6 mediate critical signals for the growth of solid tumors, and inhibition of JAK kinases by small-molecular weight inhibitors has been shown to efficiently abrogate tumor formation or restore sensitivity to other protein tyrosine kinase (PTK) inhibitors in xenograft models [8,9]. Currently, clinical JAK inhibition is focused on the treatment of MPNs [10] and rheumatoid arthritis (RA) [11], and clinical trials with JAK inhibitors are ongoing also in other autoimmune and inflammatory diseases [12]. Mounting evidence of the involvement of the JAK–STAT pathway in cancer, however, has already led to clinical trials with JAK inhibitors in, e.g., pancreatic cancer [13], advanced solid tumors (ClinicalTrials identifier: NCT02646748), NSCLC (NCT02917993), and triple-negative breast cancer (NCT02876302).

The majority of clinical JAK mutations concentrate in the pseudokinase domain of the protein and highlight the importance of understanding the molecular mechanisms of normal and pathogenic JAK signaling. Given the prevalence of JAK mutations as well as the broad involvement of JAK-mediated signaling in disease, this understanding is a vital prerequisite for developing better future therapies with potential for wide-ranging implications. In this review, we discuss the current views on the molecular basis of regulation of JAKs, as well as mechanisms of JAK deregulation in disease.

2. Jaks and the JAK–STAT pathway

JAKs are intracellular, non-receptor PTKs, but constitutively associated with their cognate receptors [14–16]. There are four JAKs in mammals, birds, and fish (JAK1–3, TYK2), with JAK1, JAK2 and TYK2 being ubiquitously expressed, while expression of JAK3 being mostly restricted to cells of hematopoietic origin [17]. JAKs are multi-domain proteins consisting of an N-terminal FERM-domain, a SH2-like domain, a so-called pseudokinase domain (JAK homology 2, JH2) and the catalytically active, signaling PTK domain (JH1). The FERM and SH2-like domains mediate the interaction of JAKs with their receptors [18,19], as well as regulating JAK kinase activity by as-of-yet unknown mechanisms [20,21]. Recent crystal structures have revealed that the FERM and SH2 domains form a structurally tightly-linked continuous unit with the receptor peptide running over, and intimately contacting both domains [22–25].

The most defining JAK domain is probably the pseudokinase domain, JH2, which constitutes the second kinase domain, and thus the second face of its two-faced namesake ancient Roman god Janus. JH2 strongly resembles eukaryotic protein kinases, but shows a distinct pattern of deviations from active kinases including lacking the catalytic aspartate, which is replaced by an asparagine in all JAK JH2s, thus gaining them the classification of 'pseudokinase' [26–28]. JH2 has critical regulatory functions, and is a veritable hotspot for clinically relevant mutations, including driver mutations underlying hematopoietic malignancies (JAK2 mutations), leukemia and lymphomas (all JAKs), and cancer (JAK1, JAK3), discussed in more detail below [5]. Lastly, JAKs consist of a C-terminal active PTK domain, JH1 , which is closely related to the kinase domain of receptor tyrosine kinases (RTKs), like epidermal growth factor receptor (EGFR), and seems to be evolutionarily distinct from JH2 [29].

JAKs mediate signaling of around sixty different cytokines, hormones, and growth factors ranging from regulators of the immune system and hematopoiesis, like interleukins (IL), interferons (IFN), erythropoietin (EPO), and thrombopoietin (TPO), to regulators of development and metabolism, like prolactin (PRL) and growth hormone (GH) (see Table 1) [30]. Signaling through the JAK–STAT pathway (Fig. 1A) is initiated by binding of a cytokine to the extracellular

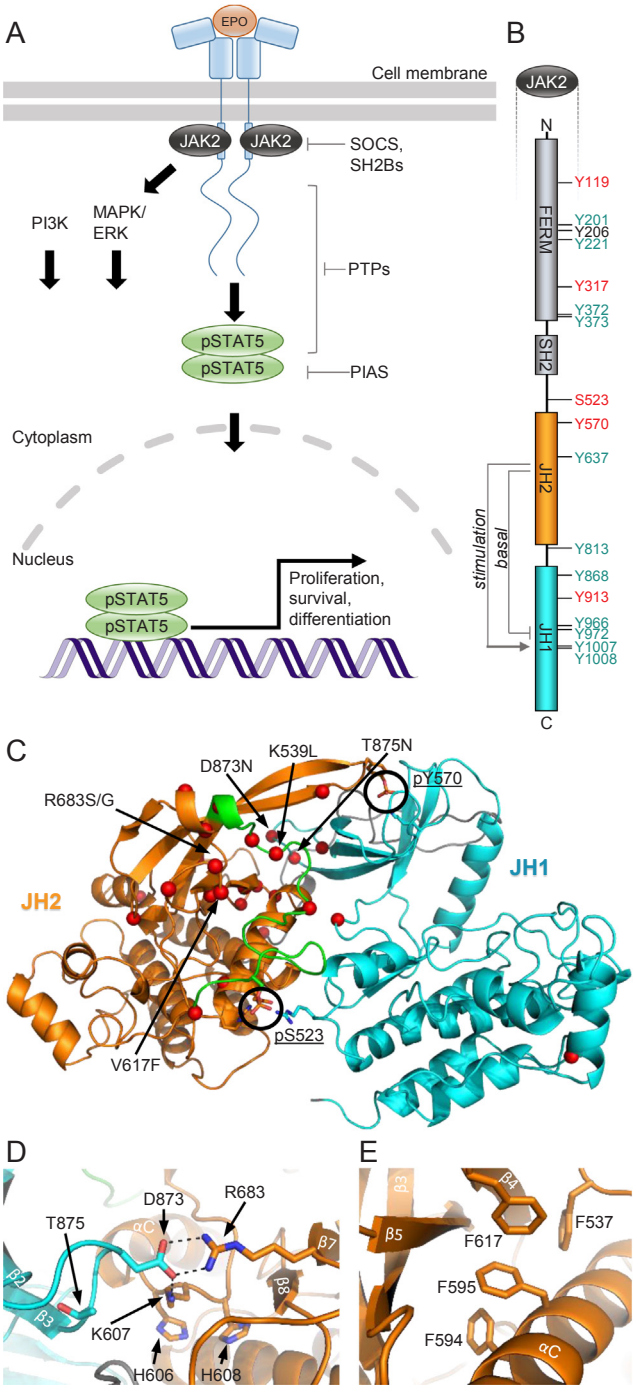

Fig. 1. The JAK–STAT signaling pathway and its regulation. A) Overview of the JAK–STAT signaling pathway using erythropoietin (EPO) signaling as an example. The most important intermolecular regulators are depicted along with their point of action along the signaling pathway. B) Characterized phosphorylation sites along JAK2. Activating phosphorylation sites are shown in green and inhibiting in red. The effect of Y206 phosphorylation on kinase activity is uncertain. See also Table 3. C) The JH2–JH1 inhibitory interaction using JAK2 as an example [44]. Sites of known clinical activating JAK2 mutations are shown with red spheres (α carbon), and most important mutations are highlighted. The two inhibitory phosphorylation sites, pS523 and pY570, are encircled. D) Close-up of the D873–R683 interaction, along with surrounding residues, all of which are known sites of activating mutations (Table 2). E) The phenylalanine stack around F617 in JAK2 V617F crucial for V617F-induced JAK2 activation.

portions of its cytokine receptor chain(s). This induces dimerization, oligomerization, or a conformational change of the receptor complex [31], which activates associated JAKs inducing trans-autophosphorylation of the activation loop (AL) of JAK JH1s [32] and a subsequent increase in catalytic activity [33]. The molecular details of this activation mechanism are still unknown with multiple competing models currently under investigation (see below) [34]. Activated JAKs next phosphorylate specific tyrosines on the receptor chains, which serve as docking sites for SH2 domain-containing signaling molecules like STATs [35]. Receptor-bound STATs are next phosphorylated by JAKs on a specific tyrosine in the C-terminal tail, enabling SH2-mediated dimerization of STATs, and subsequent translocation into the nucleus [35–37], where they act as transcription factors with far-reaching effects on regulation of transcription and epigenetics (reviewed in [38]). This simple signal transduction pathway of cytokine receptors, associated JAKs, downstream STATs, along with key protein regulators is evolutionarily conserved throughout bilateria [39].

3. Regulation of JAK activity

Activity of the JAK–STAT pathway is tightly regulated on multiple levels to ensure suppression of signaling in the absence of cytokine stimulation, but also to allow rapid, transient activation upon stimulation (Fig. 1). Regulation relies primarily on control of JH1 tyrosine kinase activity by intra- and intermolecular mechanisms, as well as on controlling the availability of parts of the signaling machinery. Many of these levels of regulation are initiated or modulated by phosphorylation of regulatory residues on JAKs, receptors, and STATs

3.1. Intramolecular regulation of JAK activity

The most basic level of regulation of JAK tyrosine kinase activity is by the AL of JH1. Isolated JH1, when produced using recombinant protein technology, is constitutively active and able to autophosphorylate its AL in solution leading to full activation of the domain's enzymatic activity [33,40,41]. In PTKs, this phosphorylation is usually necessary to relieve inhibition caused by the insertion of the unphosphorylated AL into the active site of the domain [42]. However, no crystal structures of JH1 currently exist of this presumed inactive form [43], and recent molecular dynamics data have suggested, that activation of JH1 via phosphorylation of its AL is not (only) due to vacation of the active site, but rather, due to destabilization of the autoinhibitory JH2–JH1 interaction [44].

3.2. Inhibition and activation of JH1 by JH2

Early domain deletion experiments indicated that both JH2 and JH1 were needed for JAK-mediated signaling [45–48]. Subsequently, work on JAK2 and JAK3 found that loss of JH2 was also associated with increased basal JAK activity (i.e. in the absence of cytokine stimulation) [47,48]. Furthermore, adding JH2 to isolated JH1 in cells or in recombinant constructs has been shown to significantly decrease kinase activity of JH1 [40,41,47,49]. Thus, JH2 is a regulatory domain with two roles: firstly, inhibition of the kinase activity of JH1 in the absence of stimulation and, secondly, mediation of the stimulatory signal from the cytokine receptor to JH1 [48]. Both of these functions are strikingly apparent in the effects of various known clinical and experimental JAK mutations, as JH2 harbors both gain-of-function and loss-of-function mutations [46,50] (see Table 2).

Recently, virtually identical structures of a JH2–JH1 interaction were proposed by long time scale molecular dynamics simulations for JAK2 [44] and a crystal structure for TYK2 [41], which provide explanation for JH2-mediated inhibition of JH1 (Fig. 1C). In the interaction, JH2 binds to the hinge-side of JH1 in a front-to-back orientation, loosely analogous to the interaction seen in the autoinhibited structure of Src kinase, in which SH2-SH3 domains bind to the hinge-side of the kinase domain [51]. In the JH2–JH1 interaction, JH1 activity is probably inhibited by an opening of the catalytic site and conformational restriction of the JH1 lobes preventing the conformational dynamics of JH1 needed for the kinase reaction.

The simulation-derived, autoinhibitory JAK2 JH2–JH1 interaction includes two previously known, JAK2-specific inhibitory phosphorylation sites: JAK2 S523 and Y570 [52–55] (Fig. 1B, Table 3), phosphorylation of both of which is expected to strengthen the interaction (Fig. 1C) [44].

3.3. Activation of JAKs by mutation

The inhibitory JH2–JH1 interaction interface harbors the majority of known clinical and experimental activating JAK mutations, and disruption of the inhibitory interaction thus provides an explanation for the hyperactive phenotype. Striking examples are mutations in JAK2 JH2 β7-β8 and αC-β4, and JH1 β2-β3 loops, which contain JAK2 R683 and D873 that form an ionic interaction over the JAK2 JH2–JH1 interface (Fig. 1D). Mutations in these loops (including mutations of R683 and D873 themselves) are expected to weaken the JH2–JH1 interaction by disrupting the R683–D873 link. Similarly, these residues are also known mutation sites in other JAKs (Table 2), indicating that the identified mode is likely to be conserved across all four JAKs. The multitude of known *JAK2* exon 12 mutations (Table 2), on the other hand, fall into the SH2-JH2 linker region, which probably makes extensive contacts along the JH2–JH1 interface [44], and has previously been shown to be important in suppression and regulation of basal activity [19,56]. Similarly, mutations such as JAK2 V617 are expected to disrupt the conformation of the SH2-JH2 linker, which is probably (at least part of) the activation mechanism of, e.g., JAK2 V617F.

However, it is currently unclear, whether the high activating potency of, e.g., JAK2 V617F is explained by SH2-JH2 linker-mediated disruption of the JH2–JH1 interaction alone, or whether a distinct activating intra- or intermolecular interaction is involved. Indeed, analysis of recombinant JAK2 fragments has shown that introduction of V617F to JH2-JH1 constructs only increases catalytic activity by ~3-fold [40], suggesting that another mechanism beyond disruption of the JH2–JH1 might be needed to explain the abnormally high activity of JAK2 V617F *in vivo*. CoIP experiments with full-length JAK2 have suggested that addition of the V617F mutation leads to a JAK2–JAK2 interaction *in vitro*, which cannot be detected with wild-type JAK2 [57]. The high density of activating mutations in the N lobe of JH2 as well as the SH2-JH2 linker has also been suggested to be circumstantial evidence for an activating interaction, probably involving these regions [58]. Transformation by practically all activating JAK2 mutations (including JAK2 V617F) requires the presence of cytokine receptors [59–61], which could be due to requirements for an activating trans-interaction. Alternatively, the receptor could function as a simple scaffold allowing co-localization of activated JAKs with their substrate (s). Data showing that correct orientation of the receptor chains are not needed for activation by JAK2 V617F [62] support the latter model.

Analysis of experimental mutations capable of inhibiting activation by clinical disease mutations (e.g., JAK2 V617F) has identified regions in JH2 required for mutational activation. These regions include the SH2-JH2 linker [63], a functional ATP-binding pocket in JH2 (at least in JAK1, JAK2 [64], and JAK3 (Raivola, Hammarén, Silvennoinen et al, manuscript in preparation)), as well as JH2 αC [62,65,66]. Indeed, molecular dynamics simulations of JAK2 JH2 suggest that V617F induces stabilization of JH2 αC, which can be reversed by addition of the inhibiting αC mutation JAK2 F595A thus breaking the phenylalanine stack around F617 (Fig. 1E) [67]. Similarly, loss of JH2 ATP-binding is likely to suppress mutational activation by destabilizing αC [64]. Interestingly, these mutations have been reported to not strongly inhibit cytokine-dependent JAK activation, suggesting that activation by cytokine could be mechanistically distinct from activation by mutations like JAK2 V617F [57,62,64–66]. Whether this is due to a distinct

Table 2
Clinical mutations in JAKs. Data in the table were expanded from similar compilations in [41,165–167]. Kinase activity – measured as basal JAK activation loop phosphorylation (e.g., JAK2 Y1007/1008), STAT phosphorylation, or transcriptional activity in reporter assay: +/− increase/decrease in activity, respectively. An increase in activity (+) refers to basal (unstimulated) activity (e.g., ligand-independent activation by JAK2 V617F); for decreased activity (−) signaling upon stimulation is included (e.g., cytokine-irresponsive JAK3 SCID mutations); NE – no appreciable effect. fs – frame-shift. Due to the increased pace of genetic analysis of patient samples, experimentally uncharacterized mutations are only shown, if of special interest.

Exon #	JAK domain	Mutation	Kinase activity	Effect	Associated disease	Reference
JAK1						
3	FERM	I62V			B-ALL and T-ALL	[168]
3	FERM	S71C			ETP-ALL	[169]
4	FERM	D82A		Might lower receptor association (shown together with JAK1 Y81A)	Gynecologic tumors	[167,170]
5	FERM	K142fs		LOF by frame-shift	Gynecologic tumors	[167]
6	FERM	K204M			B-ALL	[167]
8	FERM	N339fs		LOF by frame-shift	Gynecologic tumors	[167]
8	FERM	R360W			T-ALL	[168]
8	FERM	I377K			ETP-ALL	[169]
9	Linker	V427M			Pediatric T-ALL	[171]
9	Linker	N451S	+	Increased pSTAT3/5	Hepatocellular carcinoma (xenograft)	[172]
9	Linker	P430fs		LOF by frame-shift	Gynecologic tumors	[167]
10	SH2	W467*			Breast Cancer (Triple Neg)	[173]
10	SH2	T478S	+	Increased pSTAT1/3/5	AML	[41]
10	SH2	E483D	+	Increased pSTAT3/5	Hepatocellular carcinoma (xenograft)	[172]
11	SH2	G511D			Gynecologic tumors	[167]
11	SH2	S512L		Overexpression of ALL (and JAK)-related genes	T-ALL	[168]
12	Linker	R577Q			Gynecologic tumors	[167]
13	JH2	T593M			Gynecologic tumors	[167]
13	JH2	D604Y			T-ALL	[174]
13	JH2	V623A	+	Increased pSTAT1/3/5	AML	[41]
13	JH2	L624-629W			ALL	[175]
13	JH2	I631R/G/I			ALL	[176]
14	JH2	A634D	+		B-ALL and T-ALL	[168]
14	JH2	E637K			Breast Cancer (HER2+)	[173]
14	JH2	Q644H			Hepatocellular carcinoma	[123]
14	JH2	V645F	+	Constitutive pJAK1/STAT5	Hepatocellular carcinoma	[123]
14	JH2	S646F/P	+	Increased pJAK1/STAT5	ALL	[175,177]
14	JH2	H647Y			Invasive ductal carcinoma	[178]
14	JH2	K648N			T-ALL	[179]
14	JH2	V651M			T-ALL	[41]
14	JH2	Y652H	+	Constitutive STAT5 activity	T-ALL	[41,177]
14	JH2	L653F	+		Childhood ALL	[168]
14	JH2	C657R			Gynecologic tumors	[167]
14	JH2	V658F/L	+	Constitutive STAT5 activity	ALL	[175,177,178,180]
15	JH2	E668Q	+		Pediatric T-ALL	[171]
15	JH2	S703I	+	High basal pSTAT3/5	ALL & hepatocellular carcinoma	[172,176,177]
16	JH2	R724H/Q/S	+	Increased STAT1 activity	B-ALL and T-ALL	[168]
16	JH2	S729C	+	Increased pSTAT3/5	Hepatocellular carcinoma (xenograft)	[172]
16	JH2	P733L	−		Immunosuppression	[181]
16	JH2	F734L			T-ALL	[177]
17	JH2	T782M			Adenocarcinoma	[178]
17	JH2	L783F	+	Increased STAT5 activity	T-ALL	[177,178]
17	JH2	L799P			Gynecologic tumors	[167]
18	Linker	P832S	−		Immunosuppression	[181]
19	Linker	K860fs	−	LOF by frame-shift	Gynecologic tumors	[167]
19	Linker	G871E			SKN tumour cell line	[182]
19	JH1	R879S/C/H	+	Increased STAT1 activity	T-ALL	[168]
19	JH1	G882E		Loss of JH1 ATP-binding	SKN tumour cell line	[182]
20	JH1	T901G	+		T-ALL	[171]
20	JH1	K908T			Pediatric T-ALL	[171]
20	JH1	K924fs		LOF by frame-shift	Gynecologic tumors	[167]
22	JH1	G990_splice			Gynecologic tumors	[167]
23	JH1	E1051Q/P			Gynecologic tumors	[167]
24	JH1	A1086S	+	Increased pSTAT3/5	Hepatocellular carcinoma (xenograft)	[172]
24	JH1	R1113H			Gynecologic tumors	[167]
JAK2						
3	FERM	E61K			Putative primary erythrocytosis	[183]
4	FERM	T108A	NE	Slightly EPO hypersensitive	Found as germline mutation in V617F-positive PV patient	[184]
6	FERM	E177V			Putative primary erythrocytosis	[183]
7	FERM	G276A			Putative primary erythrocytosis	[183]
8	FERM	R340Q			PV	[185]
9	Linker	L393V	NE	Might be weakly EPO hypersensitive	Found as germline mutation in V617F-positive PV patient	[184]
SH2						
12	Linker	T514M			MPNs	[186]

(*continued on next page*)

Table 2 (*continued*)

Exon #	JAK domain	Mutation	Kinase activity	Effect	Associated disease	Reference
12	Linker	N533I/Y			PV (together with K539L)	[166]
12	Linker	M353I	+		AMKL	[19]
12	JH2	K539L	+		MPNs	[187]
12	JH2	I540T			PV	[41]
12	JH2	538–547		(Various deletions and insertions)	MPNs	[41,188]
12	JH2	D544G			PV	[41]
12	JH2	L545S			PV	[41]
12	JH2	F547L			PV	[41]
13	JH2	F556L			MPNs	[166]
13	JH2	R564Q	+		Hereditary ET	[189]
13	JH2	V567A			MPNs	[166]
13	JH2	G571S			Unknown (non-affected parent of MPN patient has this variant)	[183]
13	JH2	H587N			MPNs	[166]
13	JH2	S591L			MPNs	[166]
14	JH2	H606Q			MPNs	[166]
14	JH2	K607N	+		AML	[166]
14	JH2	H608Y			MPNs	[166]
14	JH2	L611S	+		ALL	[166]
14	JH2	V617F	+		MPNs	[116–119]
14	JH2	V617I	(+)	Cytokine hyperresponsiveness	Hereditary thrombocythemia	[190,191]
14	JH2	C618R	+		MPNs	[192]
14	JH2	D620E			MPS	[193]
15	JH2	L624P			MPNs	[166]
15	JH2	I645V			MPNs	[166]
16	JH2	I682F	+		ALL	[194]
16	JH2	R683S/G	+		MPNs	[194,195]
17	JH2	S755R			Hereditary thrombocythemia (together with R938Q *in cis*)	[196]
19	Linker	Y813D			IMF	[41]
19	Linker	E846D	(+)	Increases JH1 activity, when EPOR is present, EPO hypersensitivity	Germ-line mutation found in erythrocytosis and megakaryocytic atypia	[197]
20	JH1	R867Q	+		ALL, hereditary thrombocythemia	[175,196]
20	JH1	D873N	+		ALL	[194]
20	JH1	T875N	+		AMKL	[41]
21	JH1	P933R	+		ALL	[194]
21	JH1	R938Q	+		Hereditary thrombocythemia (together with S755R *in cis*)	[196]
24	JH1	R1063H	(+)	Increases JH1 activity, when EPOR is present, EPO hypersensitivity	Germ-line mutation found in erythrocytosis and megakaryocytic atypia	[197]
25	JH1	N1108S			PV	[41]
JAK3						
2	FERM	M1V			SCID	[165]
2	FERM	G36fs	−	LOF by frame-shift	SCID	[198]
2	FERM	A58P/del	−	Lack of JAK3 mRNA/protein	SCID	[165,198]
3	FERM	G62S	+	Does not transform BaF3, kinase activity higher than WT	AML	[199]
3	FERM	I87T	+	Increased basal pJAK3/pSTAT5	DS-TMD	[198,200]
3	FERM	L98/99A		Weakens receptor binding	SCID	[201]
3	FERM	Y100C	−	Weakens receptor binding	SCID	[201,202]
4	FERM	P132T/A	+	P132A oncogenic in mouse xenotransplants	AMKL	[203,204]
5	FERM	P151R			SCID, DS-TMD, AMLK	[165,205]
5	FERM	L156P	+	No significant increase in pSTAT5 in 293T cells, but does transform BaF3 cells	ATLL	[206]
5	FERM	D169E	−	No IL-2 responsive pSTAT5	SCID	[24]
5	FERM	R172Q	+	No significant increase in pSTAT5 in 293T cells, but does transform BaF3 cells	ATLL	[206]
5	FERM	E183G	+		ATLL	[206]
6	FERM	R223H			Small cell carcinoma/neuroendocrine prostate cancer (NePC)	[207]
6	FERM	R272H		Nontransforming passenger	T-ALL	[208]
6	FERM	Q283H		Nontransforming passenger	T-ALL	[209]
8	FERM	T391fs		LOF by frame-shift	SCID	[198]
9	SH2	R403H		Nontransforming passenger	T-ALL	[208,210]
10	SH2	R445X	−		SCID	[211]
11	SH2	E481G	−		SCID	[212]
11	Linker	K482S			SCID	[165]
	Linker	fsX483		LOF by frame-shift	SCID	[165]
11	Linker	del482-596	−		SCID	[212]
11	Linker	Q501H	+		AMKL	[41,200]
11	Linker	Q507P			T-PLL	[213]
11	Linker	M511I	+	Slight kinase activity increase, transforms Ba/F3	T-PLL, AML, JMML, NKTCL	[208,213]
12	JH2	C565X	−		SCID	[108,198]

(*continued on next page*)

Table 2 (*continued*)

Exon #	JAK domain	Mutation	Kinase activity	Effect	Associated disease	Reference
13	JH2	A572V/T	+	Increased pSTAT3/5; features of megakaryoblastic leukemia and transforms murine lymphoid cells *in vivo*	AMKL	[41,208]
13	JH2	A573V	+	T-ALL-like disease in mice	DS-ALL,DS AMKL, NTCL	[41,208,214–216]
13	JH2	M576L			Adult non-DS AMKL	[217]
13	JH2	del586-592	−	No pSTAT5, but constitutive pJAK3	SCID	[49]
13	JH2	R582W	−	Leads to two products, insufficient for signal transduction	SCID	[165]
13	JH2	H583Y	+		NTCL	[198,216]
13	JH2	G589S	−		SCID	[198]
13	JH2	G589D	+		NTCL	[216]
13	JH2	delV590-S596			SCID	[165]
13	JH2	del592			AMKL/TMD	[205]
13	JH2	A593R/T	+		DS-AMKL	[41,215]
15	JH2	R657Q(/H/W)	+	High basal pSTAT5, causes B-cell leukemia in mice	T-PLL, AML, JMML, NKTCL	[41,213]
15	JH2	R651W			SCID	[218]
15	JH2	V674A/F	+	Increased STAT5 activation, T-ALL-like disease in mice	T-ALL	[213,219,220]
15	JH2	V678L/M			T-ALL	[208,213]
16	JH2	P689S			SCID	[218]
16	JH2	R694K			SCID	[218]
16	JH2	E698X			SCID	[165]
16	JH2	V715I			Breast cancer	[178]
16	JH2	V722I	+		AMKL, SCID,NKTCL	[203,211]
16	JH2	K733fs		LOF by frame-shift	SCID	[221]
17	JH2	Del734-784			SCID	[165]
17	JH2	C759R	−	No pSTAT5, but constitutive pJAK3	SCID	[49]
17	JH2	V765D			JMML	[222]
17	JH2	Q766X			SCID	[165]
17	Linker	R771X			SCID	[165]
18	Linker	S789P	+	Increased kinase activity, but only weakly transforms BaF3	Childhood ALL	[194,215]
18	JH1	Y824D			T-PLL	[223]
19	JH1	L857Q/P	+	Receptor independent, L857Q causes severe thymus hyperplasia in mice	T-ALL/T-PLL/JMML	[208,220]
19	JH1	Q865E		Nontransforming passenger	T-ALL	[209]
19	JH1	fsX844		LOF by frame-shift	SCID	[165]
20	JH1	Y904X			SCID	[224]
20	JH1	P906S	+		ALL	[220]
20	JH1	L910S	−		SCID	[218]
20	JH1	R918C	+	Increased kinase activity, but only weakly transforms BaF3	AML	[199,215]
20	JH1	R925S		Nontransforming passenger	T-ALL	[208]
20	JH1	Y929X			SCID	[198]
21	JH1	E958K	+		T-ALL, JMML	[209,220,222,225]
21	JH1	G987fs	−	LOF by frame-shift	SCID	[198]
21	JH1	Q988P			T-ALL	[226]
22	JH1	L1017M	+	Increased kinase activity, but only weakly transforms BaF3	CML	[199]
22	JH1	Y1023X	−		SCID	[218]
23	JH1	1024fs	−	LOF by frame-shift	SCID	[165]
24	JH1	E1106G		Nontransforming passenger	T-ALL	[208]
TYK2						
3	FERM	G36D			T-ALL (MOLT-16 cell line)	[134]
3	FERM	S47N			T-ALL (MOLT-16 cell line)	[134]
3	FERM	A53T	NE	PBMCs show decreased CXCL10 response	Increased MS risk, found in herpes simplex encephalitis (HSE) patient	[227–229]
8	FERM	V362F	NE		Increased SLE and SSc risk	[230]
8	FERM	G363S	NE		AML	[228]
9	FERM Linker SH2 Linker	R425H			T-ALL (MOLT-16 cell line)	[134]
15	JH2	I684S	−	Impairs TYK2 kinase activity, but still enables signaling through JAK1/TYK2 or JAK2/TYK2	Protect against RA and Autoimmunity. Also found in T-ALL.	[134,231,232]
15	JH2	R703W	NE		AML	[228]
16	JH2	V731I	+		ALL (RPMI-8402 cell line)	[134]
16	JH2	P760L	+		ALL (germline)	[135]
16	JH2	G761V	+		ALL (germline)	[135]
Linker						
20	JH1	M926V	NE		Germline mutation in ALL, but probably not deleterious	[135]
20	JH1	A928V	NE		Protect against RA and Autoimmunity	[228,231]
20	JH1	E957D	+		ALL (MOLT-4 cell line)	[134,135]
22	JH1	A1016S	NE		AML	[228]

(*continued on next page*)

Table 2 (*continued*)

Exon #	JAK domain	Mutation	Kinase activity	Effect	Associated disease	Reference
22	JH1	R1027H	+		ALL (MOLT-16 and CCRF-CEM cell lines)	[134]
23	JH1	P1104A	−	Impairs TYK2 kinase activity, but still enables signaling through JAK1/TYK2 or JAK2/TYK2	Decreased susceptibility to RA and Autoimmunity; Found in NF1-MPNSTs	[228,231–233]

Table 3
Regulatory phosphorylation sites, their effect on basal JAK2 activity (measured as pJAK2 (pY1007/Y1008) or a downstream measure like STAT phosphorylation (pSTAT) or transcriptional reporter activity), the probable phosphorylating kinase, and assumed effects of phosphorylation (mostly based on mutagenesis experiments) are shown. Cytokines, which have been experimentally tested for inducing specific phosphorylation, are shown in parenthesis. Sites for which no functional follow-up data has been published, have been omitted for clarity (see, e.g. [234,235]). NE – no effect. Autophos. – autophosphorylation by full-length JAK2. ? – no data available.

Domain	JAK2 Residue	Kinase activity	Phosphorylated upon	Phosphorylated by	Effect of phosphorylation/mutation of site	References
FERM	Y119	−	Stimulation (EPO)	Probably JH1*	Phosphorylation likely to mimic Y119E, which induces dissociation from EPOR, GHR, PRLR, but not IFNGR2	[70]
FERM	Y201	+§	?	Autophos. (*in vitro*)	Y201F inhibits Ang II-mediated JAK2-signalling§	[69,237]
FERM	Y206	NE	?	?	Y206F has no appreciable effect on EPO signaling	[69,237]
FERM	Y221	+	Stimulation (GH, IL-3)	Autophos. (*in vitro* and in cells)	Y221F decreases basal pJAK2	[52,53]
FERM	Y317	−	Stimulation (EPO)	Probably mostly JH1*	Y317F causes ligand-independent pJAK2	[69]
FERM	Y372	+	?	?	Y372F decreases basal and stimulated (IFN-γ, EGF) pJAK2, pSTAT1 and JAK2-STAT1 interaction	[69,238]
FERM	Y373	+	?	?	Y373F decreases basal pJAK2	[238]
SH2						
Linker	S523	−	Constitutive	JH2 (*in vitro*)	S523A slightly increases basal pJAK2	[54,55,68]
JH2	Y570	−	Stimulation (GH, EPO, IL-3)	Autophos. (*in vitro*), JH2 (*in vitro*)	Y570F increases basal pJAK2, pSTAT3 and prolongs EPO-induced activity. pY570 likely to strengthen autoinhibitory JH2–JH1 interaction	[41,44,52,53]
JH2	Y637	+	Stimulation (EPO)	Probably mostly JH1*	Y637F lessens and shortens JAK2 activation upon EPO stimulation and partially inhibits V617F	[69]
Linker	Y813	+	Stimulation (GH)	Autophos. (*in vitro*)	Y813F reduces SH2-Bβ induced pJAK2 and pSTAT5. pY813 putative binding site for activator protein SH2-Bβ	[239]
JH1	Y868	+	Stimulation (GH)	JH1 (*in vitro*)	Y868F decreases basal and GH-induced pJAK2, pSTAT3/5	[240]
JH1	Y913	−	Upon and after stimulation (EPO)	Probably JH1*	Y913F increases EPO-induced pJAK2/pSTAT5. Y913E removes EPO-induced JAK2 activation	[241]
JH1	Y966	+	Stimulation (GH)	JH1 (*in vitro*)	Y966F decreases basal and GH-induced pJAK2, pSTAT3/5	[240]
JH1	Y972	+	Stimulation (GH)	JH1 (*in vitro*)	Y972F decreases basal and GH-induced pJAK2, pSTAT3/5	[240]
JH1	Y1007	+	Stimulation	JH1	Y1007F removes basal and cytokine-induced JAK2 activation and downstream signaling. pY1007 likely to weaken JH2–JH1 interaction	[32,33,44]
JH1	Y1008	NE	Stimulation	JH1	Y1008F has little effect on JAK2 activation	[32]

* Dependent on JAK2 Y1007/Y1008 phosphorylation and/or JH1 kinase activity.
§ Y201F has no effect on EPO-induced pJAK2, but inhibits Ang II-mediated pJAK2, pSTAT1/3. Phosphorylation probably enables binding of SHP2 and subsequent binding to AT1 receptor. Y201F has also been shown to inhibit activation by JAK2 V617F [236].

activating interaction employed by, e.g., JAK2 V617F, is still unknown.

3.4. Phosphorylation as mediator of regulation

Much of the regulation of JAK–STAT signaling is controlled by phosphorylation (Fig. 1B). In the basal state, JAK2 (for which there is the most data available) is phosphorylated only on S523 (potentially by JH2 itself [54,68]), which strengthens the JH2–JH1 interaction described above [44]. Upon activation of receptor by cytokine, JAK2 is activated by (trans-)autophosphorylation not only on the JH1 AL (Y1007/Y1008), but also on multiple other residues (Y637, Y868, Y966, Y972), which are needed for full activation of kinase activity by as-of-yet unknown mechanisms (Table 3) [69]. Termination of signaling is subsequently initiated by phosphorylation of numerous inhibitory residues along JAK2 (Fig. 1B, Table 3), which may cause dissociation of JAK2 from its receptors (Y119 [70]), strengthen the JH2–JH1 inhibitory interaction (Y570, Fig. 2C [44,68,69]), or act as binding sites for regulatory proteins.

Likewise, dephosphorylation of both JAK and receptor activating tyrosines is an important part of regulation of signaling, and is induced by multiple protein tyrosine phosphatases (PTP), including SH2 domain-containing phosphatases 1 and 2 (SHP1, SHP2), PTP1B, T-cell-PTP (TCPTP), Receptor-type PTPs C (PTPRC/CD45) and T (PTPRT), as well as basophil-like PTP (PTPBL) (reviewed in [71]). Notably, SHP2, which is a known oncoprotein and increasingly interesting drug target itself [72], has been reported to be both an activator and suppressor of JAK–STAT signaling depending on the context [71]. Other phosphatases associated with regulation of JAK–STAT signaling function as classical negative regulators of signaling in termination or basal suppression of signaling (reviewed in [34,71]). Most recorded phosphorylation events on JAK2 have been shown to be dependent on JAK2 JH1 kinase activity (or AL phosphorylation), but the exact sequence of phosphorylation events or their mechanism of action during receptor-mediated activation is yet to be fully elucidated. Potential differences in phosphorylation-mediated regulation in different cytokine receptor–JAK contexts also remain mostly unknown.

3.5. Intermolecular regulation of JAK activity

Most known intermolecular modes of JAK–STAT signaling regulation are involved in termination of signaling after cytokine stimulation, and are thus initiated by phosphorylation during cytokine-mediated

activation. Mechanisms of termination involve (de)phosphorylation, production of inhibitory proteins, and removal of signaling complexes through internalization and lysosomal or ubiquitination-mediated proteasomal degradation of receptors [5,34].

One family of direct inhibitory proteins are suppressors of cytokine signaling (SOCS1–7 and CIS), transcription of which is upregulated by activated STATs, thus forming a negative feedback loop [73]. The mechanism for SOCS-mediated inhibition has been revealed for SOCS3, which binds to both JH1 and the associated cytokine receptor, leading to inhibition of JH1 activity by direct interaction with the JH1 AL, in addition to initiation of ubiquitination of JAKs [74,75].

Active JAK–STAT signaling also induces transcription of SH2B family proteins (consisting of SH2B1/SH2-B, SH2B2/APS, and SH2B3/LNK), which each consist of a dimerization domain (DD), Pleckstrin homology (PH) domain and a C-terminal SH2 domain. SH2-B and APS bind to phosphorylated receptor or JAK tyrosines (e.g. JAK2 pY613 and pY813) via their SH2 domain [76–79], and are able to either activate or inhibit JAK activation, potentially by acting as dimerization scaffolds for JAKs [80]. The activating role of SH2-B has been most comprehensively demonstrated in leptin–JAK2 signaling [78,81], and mutations or deletions in *SH2B1* have been linked to severe obesity (see Table 4). Interestingly, SH2-B has also been reported to bind to EPOR and putatively act as a negative regulator of EPOR signaling [82]. The third SH2B family member, LNK is a well-defined inhibitor of JAK2, as well as potentially JAK3 (reviewed in [79]). How LNK negatively regulates JAK activity is not clear, but it is likely to involve a direct interaction with the LNK N-terminal domains and JAK kinase domains, and/or ubiquitination via recruitment of E3 ligases [79].

JAK–STAT signaling is also regulated at the level of STATs by protein inhibitor of activated STATs (PIAS) proteins, which are one of only few known small ubiquitin-like modifier (SUMO) E3 ligases (see [83] for a review). As their name suggests, PIAS proteins (consisting of PIAS1, PIAS3, PIASx, and PIASy) were first identified as negative regulators of STATs [84,85], but they have since been shown to also be involved in regulation of multiple other cellular processes [86]. Following cytokine stimulation, PIAS proteins have been shown to inhibit STAT signaling activity through inhibition of their DNA-binding activity by directly interacting with STATs and and/or by inducing STAT SUMOylation (reviewed in [87]).

3.6. Activation of JAKs by cytokines – The role of the cytokine receptor

Despite long-continuing research into the mechanisms of JAK activation by mutation and intermolecular regulation by various negative regulator proteins, the molecular mechanisms of JAK activation on cytokine receptors following cytokine stimulation have mostly remained elusive. One complicating factor is the sheer number of different JAK-associated receptors (Table 1) and their variable architectures. These range from 'short receptors' (e.g., GHR, EPOR, γ_c family, IFNGR) where ligand-binding domains are proximal to the membrane, and 'tall receptors' (e.g., gp130, TCCR, LIFRβ) with ligand-binding domains up to 10 nm removed from the membrane [88–91]. Numerous crystal structures of receptor extracellular domains have been generated showing cytokine-bound and unbound states [90], but how these changes are translated into intracellular activation of JAKs is not known.

The simplest mode of cytokine receptor activation is an induced dimer/oligomer model, where ligand binding induces dimerization/oligomerization of receptor chains and associated kinases, which are subsequently activated by transphosphorylation due to proximity. This mode is widely seen in RTKs [92,93], with exceptions like insulin receptor kinase (IRK), which seems to exist as preformed dimers rearranged by ligand binding [94]. Studies with engineered artificial chimeric receptors have shown that simple dimerization is sufficient to activate JAKs [95] suggesting an induced-dimer activation model. Nevertheless, preformed dimers have been reported for many JAK-associated receptors, including EPOR [96], GHR [97], gp130 [98], LEPR [99], as well as IL-2Rβ/IL-9Rα and γ_c [100]. However, some of these data may be confounded by the use of artificial overexpression systems and/or dimerization-prone fusion proteins. A current lack of methods to simultaneously measure receptor oligomerization and activation states also leaves it unclear, how many of the observed preformed dimers are actually actively signaling and thus contributing to basal signaling activity. Still, studies with forced EPOR or TPOR dimers have indicated that correct orientation of receptor chains is needed for activation of wild-type JAK2, thus potentially enabling non-signaling preformed dimers *in vivo* [101–103]. Additionally, FRET-studies with forced preformed GHR dimers suggested that the intracellular portions of GHR would move apart during stimulation, leading the study authors to hypothesize that JH2-mediated inhibition of JH1 could occur *in trans* at least for JAK2 [104]. How, or whether this architecture would apply to heteromeric JAK configurations, is unclear.

Despite these earlier findings, recent single-molecule imaging studies have shown a distinct lack of preformed dimers for EPOR at least, and approaches using engineered protein ligands have suggested that activation of EPOR–JAK2 is mainly determined by the distance between receptor chains [105], thus again arguing for a ligand-induced dimer model [91]. Further research using less invasive methodology with (at least close to) native expression levels of receptors is thus sorely needed to settle the question of receptor-mediated activation of JAKs.

4. Failure of JAK regulation leads to disease

Failure of JAK–STAT pathway regulation is a common cause of myeloid and lymphoid disorders but also plays a role in multiple cancers (Table 4) [6,7,106]. The first JAK disease-link was discovered in 1995 when JAK3 loss-of-function mutations were reported causative of severe combined immune deficiency (SCID) [107,108]. Shortly later, inhibition of acute lymphoblastic leukemia (ALL) with JAK-inhibitor AG-490 lead to the discovery of constitutive JAK2 activity in patient-derived ALL cells [109]. Subsequently constitutively active JAK2 fusion protein TEL/ETV6-JAK2 was identified in early pre-B cell ALL and T-cell childhood ALL [1,110].

Oncogenic JAK2 rearrangements to multiple fusion gene partners (TEL, PCM1, BCR, SSPB2, STRN3, RPN1, NF-E2, RUNX1, SEC31A, SPAG9) have since been identified in ALL, acute myeloid leukemia (AML), atypical chronic myeloid leukemia (CML), MPN and/or Hodgkin lymphoma [2,110–114], and similar oncogenic fusions have also been reported for TYK2 [106]. The chimeras comprise of JH1, with or without (pieces of) JH2, fused to dimerization domain(s) of the fusion partner, leading to constitutive tyrosine kinase activity presumably via dimerization/oligomerization [1,112,115]. These translocations are rare, however: a cytogenetic study of 24 262 unique patients, for example, revealed these kinds of JAK2 abnormalities in only 0.06% of hematopoietic neoplasms [113].

In contrast, somatic JAK2 V617F is highly prevalent in MPN and causes approximately 95% of polycythemia vera (PV) and 50–60% essential thrombocythemia (ET) and primary myelofibrosis (PMF) cases [116–119]. Allelic burden probably plays a role in defining the clinical phenotype of JAK2 V617F as the burden increases from ET over PV to PMF [120]. Furthermore, the pSTAT profiles discriminate among the MPN diseases and are independent of JAK2 V617F: high pSTAT3 and pSTAT5 are typical in PV, high pSTAT3 and low pSTAT5 in ET, and low pSTAT3 and pSTAT5 in PMF [121], but the mechanism behind these differences have remained largely unknown. PV, ET and PMF can evolve into AML and therefore it is not surprising that approximately 5% of AML patients are also V617F-positive [122]. Numerous gain-of-function mutations identified in JAK1, JAK2 and JAK3 (Table 2) are drivers in ~20% of T-ALL cases (mainly JAK1, also JAK2, JAK3) and to lesser extent in B-ALL (JAK1, JAK2) or hepatocellular adenoma (JAK1) [5,123].

V617F-negative MPN often show mutations in other components or

Table 4
Diseases caused by failure in JAK regulation. See Table 2 for a full list of disease-associated JAK mutations. See list of abbreviations for explanations of abbreviations.

Mode of regulation disrupted		Example(s)	Associated disease / clinical phenotype	Reference
All levels of regulation		Activating JAK2 chimeras (e.g. TEL-, PCM1-, BCR-, SSBP2-, STRN3-, SEC31A-JAK2)	T-ALL; B-ALL; atypical CML; Hodgkin lymphoma (SEC31A-JAK2); MPNs	[2,110–113,115]
		JAK3 deficiency (truncations, destabilizing mutations)	SCID	[131]
		TYK2 deficiency (truncations)	PID	[132,133]
		Catalytically inactive TYK2 JH1 variant P1104A	Decreased susceptibility to autoimmunity and RA	[231,232]
Intramolecular	Inhibition of JH1 by JH2	Activating JAK JH2 mutations (e.g. JAK2 V617F, JAK2 R683G, JAK1 R724H, JAK3 M511I) presumably disrupting the JH2–JH1 inhibitory interaction	MPNs; lymphoid and myeloid leukaemia: T-ALL, B-ALL, AML, AMKL, T-PLL, JMML, NTCL, NKTCL; hepatocellular carcinoma (JAK1)	[41,116,123,166,168,213,216]
	Inhibition of JH1 by FERM	Activating JAK3 FERM mutations (E183G, L156P, R172Q)	ATLL	[206]
	Activation of JH1 by JH2	Inactivating JAK3 JH2 mutations (e.g. C759R)	SCID	[49]
		Inactivating TYK2 JH2 variant I684S	Decreased susceptibility to autoimmunity and RA	[231,232]
Intermolecular	Inhibition by SOCS	Activation by decreased SOCS levels (DNA hypermethylation)	Multiple cancers: hepatocellular carcinoma, cholangiocarcinoma, cervical cancer, esophageal squamous cell carcinoma, Barrett's adenocarcinoma, non-small-cell lung cancer, prostate cancer, breast cancer, and ovarian cancer	[136,242]
		Inhibition by elevated SOCS levels	Asthma (SOCS1, 3); atopic dermatis (SOCS3); RA (SOCS1, 3); ulcerative colitis (SOCS3); Crohn's disease (SOCS3); enhanced disease severity in tuberculosis (SOCS1)	[136,139–141]
	Activation by SH2B1	SH2B1 mutations (SNPs, deletions) leading presumably to defects in leptin–JAK2 signaling	Severe obesity	[243–245]
	Inhibition by LNK	LNK mutations/genetic variants (e.g. 603_607delGCGCT, E208Q, R262W) leading to activation	V617F-negative MPNs; AML; CML; idiopathic erythrocytosis; increased risk for chronic inflammatory, autoimmune and vascular diseases (LNK R262W): celiac disease, asthma, type I diabetes, RA, multiple sclerosis and systemic lupus erythematosus	[79]
	Inhibition by PTP	EpoR truncations leading to loss of cytoplasmic negative regulatory domain and thus preventing binding of SHP1	Ph-like ALL; familial polycythemia	[71,246,247]
		TCPTP deficiency (deletions)	T-ALL	[248]
		LOF mutation in PTPRC	T-ALL	[249]
	Activation by PTP (SHP2)	GOF mutations in SHP2 presumably resulting in increased signaling[†]	Noonan syndrome; Lymphoid and myeloid leukemias: JMML, AML, B-ALL; Solid cancers: Breast, lung, gastric, and neuroblastoma; MDS	[250,251]
Receptor-mediated	Regulation of receptor conformation	Activating TpoR receptor mutations MPL S505N, MPL W515 (L/K/A)	Hereditary thrombocythemia; V617F-negative MPN	[252,253]
		Activating IL7R mutations (S185C, TM-domain)	T-ALL; B-ALL; SCID	[176,254,255]
	JAK–receptor interaction	Inactivating JAK3 receptor-binding deficient mutant Y100C	SCID	[201]
	Intermolecular	CALR mutations (e.g. p.L367fs*46, a 52-bp deletion) enabling direct binding and activation of TPOR	V617F-negative MPN	[128,256,257]

[†] Suggested to activate Ras/ERK1/2 pathway, but SHP2 has also been shown to regulate PI3K/AKT, JAK–STAT, JNK, and NF-κB signaling [72].

regulatory elements of the JAK–STAT pathway. For example, 1–5% PMF and ET patients carry point mutations (e.g., S505N, W515L/K/R) in the thrombopoietin receptor (TPOR) gene *MPL* [124], which enable ligand-independent activation of TPOR–JAK2 signaling [125,126]. Relatively common are also calreticulin (CALR) frameshift mutations causing TPO-independent TPOR–JAK2 signaling in up to ~13% of MPNs. CALR is a multifunctional endoplasmic reticulum protein, and its common frameshifts result in the formation of a highly-charged protein structure at the C-terminus of mutated CALR, which has been shown to be able to directly bind to the ligand-binding domain of TPOR, thus activating the receptor [127,128]. Another MPN-linked activation mechanism through TPOR is caused by mutations in the inhibitory adaptor protein LNK, which lead to mislocalization of LNK from the plasma membrane into the cytoplasm and result in TPO hyper-responsiveness [129]. LNK mutations are found in 5–7% of MPNs [130].

Besides disease-causing mutations hyperactivating JAK–STAT signaling, also deleterious loss-of-function mutations in the JAK–STAT regulatory apparatus have been described. Probably most famously, loss-of-function (LOF) germline mutations in JAK3 or JAK3-associated receptors IL-7R and common gamma chain (γ_c; also known as CD320 or IL-2RG) cause autosomal recessive SCID [131]. In fact, JAK3/IL-7R/γ_c mutations cover the majority of all SCID cases: JAK3 mutations account for approximately 10–18% of heritable SCID, IL-7R < 10%, and γ_c 25–46% [131]. Although JAK3 SCID-associated mutations are found in all JAK domains, the majority is located either in JH2 or the FERM domain (Table 2). Known JAK3 mutations include frameshifts and nonsense mutations (readily explaining LOF), but interestingly also point mutations, for all of which the mechanism action has not been conclusively determined (Table 2). Some JAK3 mutations are, however, known to affect protein expression and structural stability, or less commonly hinder receptor binding (e.g. JAK3 Y100C) [131].

Knowledge about diseases caused by TYK2 mutations has only lately started to emerge. TYK2 deficiency caused by frameshift mutations has been reported in two primary immunodeficiency (PID) patients, one with and one without hyper-IgE syndrome [132,133]. Activating driver TYK2 mutations have recently been found in multiple ALL cell lines [134] and patient samples, including two gain-of-function germline TYK2 mutations [135]. Additionally, multiple TYK2 polymorphisms have been described, some of which alter the activity of the protein and are associated with increased or decreased risks for autoimmune or inflammatory diseases (Table 2).

SOCS as regulators of JAK-STAT pathways have critical functions in pathogenesis of inflammatory and other immune disorders, susceptibility to infectious diseases and development and progression of cancer [136,137]. SOCS are suggested to have tumor suppressive role and decreased SOCS levels due to DNA hypermethylation of one or more *SOCS* genes have been observed in multiple cancers, such as hepatocellular carcinoma, cholangiocarcinoma, Hodgkin lymphoma, and primary mediastinal B-cell lymphoma. Furthermore, reduced expression of SOCS3 was shown to associate closely with lymph node metastasis in breast cancer [138]. Elevated SOCS levels possibly caused by certain SOCS promoter variants have been found in asthma (SOCS1, 3), RA (SOCS1, SOCS3), and atopic dermatitis (SOCS3) [136,139–141].

Although outside of the scope of this review, the role of various STAT mutations in development of immunological diseases cannot be underestimated as they lead to similar clinical phenotypes as deregulation of the upstream components. For recent reviews see [142,143].

5. Future outlook and perspectives

Current clinical and pharmacological interest in JAKs is at an all-time high. JAK inhibition has proven to be effective not only in diseases directly caused by activating deregulation of the JAK–STAT pathway (e.g., JAK2 mutation-driven MPNs), but also in diseases, in which JAKs act as mediators and amplifiers of proinflammatory signals, even though the disease might not be caused by deleterious changes in the JAK–STAT pathway itself. This is the case for RA for which two JAK inhibitors are already FDA and EMA-approved (tofacitinib, baricitinib), and potentially many other inflammatory diseases currently under investigation.

Similarly, understanding of the regulatory mechanisms governing the JAK–STAT pathway has grown steadily. Nevertheless, molecular understanding of JAK activation and inhibition is still far from complete. Detailed elucidation of the mechanisms of JAK inhibition and activation on receptors especially will be key for the development of modulators of JAK activity with novel modes of actions. The need for these kinds of novel inhibitors is great, and the fields of application wide. All current JAK inhibitors ('Jakinibs') target the catalytic ATP-binding site of JH1, which are highly similar between JAKs, making specificity within the JAK family problematic for most inhibitors. Furthermore, current inhibitors cannot distinguish between mutated JAKs (e.g. JAK2 V617F in MPNs) and wild-type JAKs leading to potential side-effects and an inability to eradicate diseases caused by somatic JAK mutations.

Encouragingly development of multiple novel JAK activity-modulator types is already underway. These include covalent inhibitors against JAK3 JH1 [144], compounds targeting the ATP-binding site of JH2 [145–148], protein-protein interaction interfaces within JAK [149], and the extracellular parts of JAK-associated receptors [91]. These targets have the potential to deliver more specificity (including mutant-specificity over wild-type JAK), as well as safety.

Acknowledgements

Funding: This work was supported by the Medical Research Council of Academy of Finland, Sigrid Juselius Foundation, Jane and Aatos Erkko Foundation, Finnish Cancer Foundation, Competitive Research Funding of Tampere University Hospital and Tampere Tuberculosis Foundation.

Declarations of interest

None.

References

[1] V. Lacronique, A. Boureux, V.D. Valle, H. Poirel, C.T. Quang, M. Mauchauffe, et al., A TEL-JAK2 fusion protein with constitutive kinase activity in human leukemia, Science 278 (5341) (1997) 1309–1312.
[2] K. Van Roosbroeck, L. Cox, T. Tousseyn, I. Lahortiga, O. Gielen, B. Cauwelier, et al., JAK2 rearrangements, including the novel SEC31A-JAK2 fusion, are recurrent in classical hodgkin lymphoma, Blood 117 (15) (2011) 4056–4064.
[3] K. Van Roosbroeck, J.F. Ferreiro, T. Tousseyn, J. van der Krogt, L. Michaux, B. Pienkowska-Grela, et al., Genomic alterations of the JAK2 and PDL loci occur in a broad spectrum of lymphoid malignancies, Genes Chromosomes Cancer 55 (5) (2016) 428–441.
[4] J.M. Balko, L.J. Schwarz, N. Luo, M.V. Estrada, J.M. Giltnane, D. Davila-Gonzalez, et al., Triple-negative breast cancers with amplification of JAK2 at the 9p24 locus demonstrate JAK2-specific dependence, Sci. Transl. Med. 8 (334) (2016) 334ra53.
[5] W. Vainchenker, S. Constantinescu, JAK/STAT signaling in hematological malignancies, Oncogene 32 (2013) 2601–2613.
[6] R.C. Skoda, A. Duek, J. Grisouard, Pathogenesis of myeloproliferative neoplasms, Exp. Hematol. 43 (8) (2015) 599–608.
[7] S. Thomas, J. Snowden, M. Zeidler, S. Danson, The role of JAK/STAT signalling in the pathogenesis, prognosis and treatment of solid tumours, Br. J. Cancer 113 (3) (2015) 365–371.
[8] M. Hedvat, D. Huszar, A. Herrmann, J.M. Gozgit, A. Schroeder, A. Sheehy, et al., The JAK2 inhibitor AZD1480 potently blocks Stat3 signaling and oncogenesis in solid tumors, Cancer Cell 16 (6) (2009) 487–497.
[9] S.P. Gao, Q. Chang, N. Mao, L.A. Daly, R. Vogel, T. Chan, et al., JAK2 inhibition sensitizes resistant EGFR-mutant lung adenocarcinoma to tyrosine kinase inhibitors, Sci. Signal 9 (421) (2016) ra33.
[10] J.M. O'Sullivan, C.N. Harrison, JAK-STAT signaling in the therapeutic landscape of myeloproliferative neoplasms, Mol. Cell Endocrinol. 451 (2017) 71–79.
[11] K. Yamaoka, Janus kinase inhibitors for rheumatoid arthritis, Curr. Opin. Chem. Biol. 32 (2016) 29–33.
[12] S. Banerjee, A. Biehl, M. Gadina, S. Hasni, D.M. Schwartz, JAK–STAT signaling as a target for inflammatory and autoimmune diseases: Current and future prospects, Drugs 77 (5) (2017) 521–546.

[13] H.I. Hurwitz, N. Uppal, S.A. Wagner, J.C. Bendell, J.T. Beck, S.M. Wade IIIet al., Randomized, double-blind, phase II study of ruxolitinib or placebo in combination with capecitabine in patients with metastatic pancreatic cancer for whom therapy with gemcitabine has failed, J. Clin. Oncol. 33 (34) (2015) 4039–4047.
[14] L.J. Huang, S.N. Constantinescu, H.F. Lodish, The N-terminal domain of janus kinase 2 is required for golgi processing and cell surface expression of erythropoietin receptor, Mol. Cell. 8 (6) (2001) 1327–1338.
[15] B. Giese, C. Au-Yeung, A. Herrmann, S. Diefenbach, C. Haan, A. Küster, et al., Long term association of the cytokine receptor gp130 and the janus kinase Jak1 revealed by FRAP analysis, J. Biol. Chem. 278 (40) (2003) 39205–39213.
[16] C. Haan, S. Kreis, C. Margue, I. Behrmann, Jaks and cytokine receptors—an intimate relationship, Biochem. Pharmacol. 72 (11) (2006) 1538–1546.
[17] K. Yamaoka, P. Saharinen, M. Pesu, V. Holt 3rd, O. Silvennoinen, J.J. O'Shea, The janus kinases (jaks), Genome Biol.. 5 (12) (2004) 253.
[18] S. Radtke, S. Haan, A. Jörissen, H.M. Hermanns, S. Diefenbach, T. Smyczek, et al., The Jak1 SH2 domain does not fulfill a classical SH2 function in jak/STAT signaling but plays a structural role for receptor interaction and up-regulation of receptor surface expression, J. Biol. Chem. 280 (27) (2005) 25760–25768.
[19] L. Zhao, H. Dong, C.C. Zhang, L. Kinch, M. Osawa, M. Iacovino, et al., A JAK2 interdomain linker relays epo receptor engagement signals to kinase activation, J. Biol. Chem. 284 (39) (2009) 26988.
[20] Y.J. Zhou, M. Chen, N.A. Cusack, L.H. Kimmel, K.S. Magnuson, J.G. Boyd, et al., Unexpected effects of FERM domain mutations on catalytic activity of Jak3: Structural implication for janus kinases, Mol. Cell. 8 (5) (2001) 959–969.
[21] L. Zhao, Y. Ma, J. Seemann, L. Huang, A regulating role of the JAK2 FERM domain in hyperactivation of JAK2 (V617F), Biochem. J. 426 (2010) 91–98.
[22] H.J. Wallweber, C. Tam, Y. Franke, M.A. Starovasnik, P.J. Lupardus, Structural basis of recognition of interferon-α receptor by tyrosine kinase 2, Nat. Struct. Mol. Biol. 21 (5) (2014) 443–448.
[23] R. Ferrao, H.J. Wallweber, H. Ho, C. Tam, Y. Franke, J. Quinn, et al., The structural basis for class II cytokine receptor recognition by JAK1, Structure 24 (6) (2016) 897–905.
[24] R. McNally, A.V. Toms, M.J. Eck, Crystal structure of the FERM-SH2 module of human Jak2, PLoS One 11 (5) (2016) e0156218.
[25] D. Zhang, A. Wlodawer, J. Lubkowski, Crystal structure of a complex of the intracellular domain of interferon λ receptor 1 (IFNLR1) and the FERM/SH2 domains of human JAK1, J. Mol. Biol. 428 (23) (2016) 4651–4668.
[26] G. Manning, D.B. Whyte, R. Martinez, T. Hunter, S. Sudarsanam, The protein kinase complement of the human genome, Sci. Signal 298 (5600) (2002) 1912–1934.
[27] J. Boudeau, D. Miranda-Saavedra, G.J. Barton, D.R. Alessi, Emerging roles of pseudokinases, Trends Cell. Biol. 16 (9) (2006) 443–452.
[28] H.M. Hammarén, A.T. Virtanen, O. Silvennoinen, Nucleotide-binding mechanisms in pseudokinases, Biosci. Rep. 36 (1) (2015) e00282.
[29] J. Gu, Y. Wang, X. Gu, Evolutionary analysis for functional divergence of jak protein kinase domains and tissue-specific genes, J. Mol. Evol. 54 (6) (2002) 725–733.
[30] J.J. O'Shea, R. Plenge, JAK and STAT signaling molecules in immunoregulation and immune-mediated disease, Immunity 36 (4) (2012) 542–550.
[31] J.N. Ihle, B.A. Witthuhn, F.W. Quelle, K. Yamamoto, O. Silvennoinen, Signaling through the hematopoietic cytokine receptors, Annu. Rev. Immunol. 13 (1) (1995) 369–398.
[32] J. Feng, B.A. Witthuhn, T. Matsuda, F. Kohlhuber, I.M. Kerr, J.N. Ihle, Activation of Jak2 catalytic activity requires phosphorylation of Y1007 in the kinase activation loop, Mol. Cell. Biol. 17 (5) (1997) 2497–2501.
[33] K. Chatti, W.L. Farrar, R.J. Duhé, Tyrosine phosphorylation of the janus kinase 2 activation loop is essential for a high-activity catalytic state but dispensable for a basal catalytic state, Biochemistry (N Y) 43 (14) (2004) 4272–4283.
[34] J.J. Babon, I.S. Lucet, J.M. Murphy, N.A. Nicola, L.N. Varghese, The molecular regulation of janus kinase (JAK) activation, Biochem. J. 462 (1) (2014) 1–13.
[35] J.E. Darnell Jr., STATs and gene regulation, Science 277 (5332) (1997) 1630–1635.
[36] S. Becker, B. Groner, C.W. Müller, Three-dimensional structure of the Stat3β homodimer bound to DNA, Nature 394 (6689) (1998) 145–151.
[37] X. Chen, U. Vinkemeier, Y. Zhao, D. Jeruzalmi, J.E. Darnell, J. Kuriyan, Crystal structure of a tyrosine phosphorylated STAT-1 dimer bound to DNA, Cell 93 (5) (1998) 827–839.
[38] A.V. Villarino, Y. Kanno, J.J. O'Shea, Mechanisms and consequences of jak-STAT signaling in the immune system, Nat. Immunol. 18 (4) (2017) 374–384.
[39] C. Liongue, R. Sertori, A.C. Ward, Evolution of cytokine receptor signaling, J. Immunol. 197 (1) (2016) 11–18.
[40] A. Sanz, D. Ungureanu, T. Pekkala, R. Ruijtenbeek, I.P. Touw, R. Hilhorst, et al., Analysis of Jak2 catalytic function by peptide microarrays: the role of the JH2 domain and V617F mutation, PLoS One 6 (4) (2011) e18522.
[41] P.J. Lupardus, M. Ultsch, H. Wallweber, P. Bir Kohli, A.R. Johnson, C. Eigenbrot, Structure of the pseudokinase–kinase domains from protein kinase TYK2 reveals a mechanism for janus kinase (JAK) autoinhibition, Proc. Natl. Acad. Sci. U. S. A. 111 (22) (2014) 8025–8030.
[42] J.A. Endicott, M.E. Noble, L.N. Johnson, The structural basis for control of eukaryotic protein kinases, Annu. Rev. Biochem. 81 (2012) 587–613.
[43] I.S. Lucet, R. Bamert, Production and crystallization of recombinant JAK proteins, JAK-STAT Signalling, Springer, 2013, pp. 275–300.
[44] Y. Shan, K. Gnanasambandan, D. Ungureanu, E.T. Kim, H. Hammaren, K. Yamashita, et al., Molecular basis for pseudokinase-dependent autoinhibition of JAK2 tyrosine kinase, Nat. Struct. Mol. Biol. 21 (2014) 579–584.
[45] L. Velazquez, K.E. Mogensen, G. Barbieri, M. Fellous, G. Uzé, S. Pellegrini, Distinct domains of the protein tyrosine kinase tyk2 required for binding of interferon-/ and for signal transduction, J. Biol. Chem. 270 (7) (1995) 3327–3334.
[46] H. Luo, P. Rose, D. Barber, W.P. Hanratty, S. Lee, T.M. Roberts, et al., Mutation in the jak kinase JH2 domain hyperactivates drosophila and mammalian jak-stat pathways, Mol. Cell. Biol. 17 (3) (1997) 1562–1571.
[47] P. Saharinen, K. Takaluoma, O. Silvennoinen, Regulation of the Jak2 tyrosine kinase by its pseudokinase domain, Mol. Cell. Biol. 20 (10) (2000) 3387–3395.
[48] P. Saharinen, O. Silvennoinen, The pseudokinase domain is required for suppression of basal activity of Jak2 and Jak3 tyrosine kinases and for cytokine-inducible activation of signal transduction, J. Biol. Chem. 277 (49) (2002) 47954–47963.
[49] M. Chen, A. Cheng, F. Candotti, Y.J. Zhou, A. Hymel, A. Fasth, et al., Complex effects of naturally occurring mutations in the JAK3 pseudokinase domain: Evidence for interactions between the kinase and pseudokinase domains, Mol. Cell. Biol. 20 (3) (2000) 947–956.
[50] T.C. Yeh, E. Dondi, G. Uzé, S. Pellegrini, A dual role for the kinase-like domain of the tyrosine kinase Tyk2 in interferon-α signaling, Proc. Natl. Acad. Sci. U. S. A. 97 (16) (2000) 8991–8996.
[51] J.M. Bradshaw, The src, syk, and tec family kinases: Distinct types of molecular switches, Cell. Signal 22 (8) (2010) 1175–1184.
[52] L.S. Argetsinger, J.L.K. Kouadio, H. Steen, A. Stensballe, O.N. Jensen, C. Carter-Su, Autophosphorylation of JAK2 on tyrosines 221 and 570 regulates its activity, Mol. Cell. Biol. 24 (11) (2004) 4955–4967.
[53] E.P. Feener, F. Rosario, S.L. Dunn, Z. Stancheva, M.G. Myers Jr, Tyrosine phosphorylation of Jak2 in the JH2 domain inhibits cytokine signaling, Mol. Cell. Biol. 24 (11) (2004) 4968–4978.
[54] R. Ishida-Takahashi, F. Rosario, Y. Gong, K. Kopp, Z. Stancheva, X. Chen, et al., Phosphorylation of Jak2 on Ser523 inhibits Jak2-dependent leptin receptor signaling, Mol. Cell. Biol. 26 (11) (2006) 4063–4073.
[55] A.M. Mazurkiewicz-Munoz, L.S. Argetsinger, J.K. Kouadio, A. Stensballe, O.N. Jensen, J.M. Cline, et al., Phosphorylation of JAK2 at serine 523: A negative regulator of JAK2 that is stimulated by growth hormone and epidermal growth factor, Mol. Cell. Biol. 26 (11) (2006) 4052–4062.
[56] A.S. Sanz, Y. Niranjan, H. Hammarén, D. Ungureanu, R. Ruijtenbeek, I.P. Touw, et al., The JH2 domain and SH2-JH2 linker regulate JAK2 activity: A detailed kinetic analysis of wild type and V617F mutant kinase domains, BBA Proteins Proteomics 1844 (10) (2014) 1835–1841.
[57] S.P. Gorantla, T.N. Dechow, R. Grundler, A.L. Illert zum, C.M. Büschenfelde, M. Kremer, et al., Oncogenic JAK2V617F requires an intact SH2-like domain for constitutive activation and induction of a myeloproliferative disease in mice, Blood 116 (22) (2010) 4600–4611.
[58] O. Silvennoinen, S.R. Hubbard, Molecular insights into regulation of JAK2 in myeloproliferative neoplasms, Blood 125 (22) (2015) 3388–3392.
[59] X. Lu, L.J.S. Huang, H.F. Lodish, Dimerization by a cytokine receptor is necessary for constitutive activation of JAK2V617F, J. Biol. Chem. 283 (9) (2008) 5258–5266.
[60] G. Wernig, J.R. Gonneville, B.J. Crowley, M.S. Rodrigues, M.M. Reddy, H.E. Hudon, et al., The Jak2V617F oncogene associated with myeloproliferative diseases requires a functional FERM domain for transformation and for expression of the myc and pim proto-oncogenes, Blood 111 (7) (2008) 3751–3759.
[61] H. Yao, Y. Ma, Z. Hong, L. Zhao, S.A. Monaghan, M.C. Hu, et al., Activating JAK2 mutants reveal cytokine receptor coupling differences that impact outcomes in myeloproliferative neoplasm, Leukemia 31 (10) (2017) 2122–2131.
[62] A. Dusa, C. Mouton, C. Pecquet, M. Herman, S.N. Constantinescu, JAK2 V617F constitutive activation requires JH2 residue F595: a pseudokinase domain target for specific inhibitors, PLoS One 5 (6) (2010) 207–212.
[63] A.V. Toms, A. Deshpande, R. McNally, Y. Jeong, J.M. Rogers, C.U. Kim, et al., Structure of a pseudokinase-domain switch that controls oncogenic activation of jak kinases, Nat. Struct. Mol. Biol. 20 (10) (2013) 1221–1223.
[64] H.M. Hammaren, D. Ungureanu, J. Grisouard, R.C. Skoda, S.R. Hubbard, O. Silvennoinen, ATP binding to the pseudokinase domain of JAK2 is critical for pathogenic activation, Proc. Natl. Acad. Sci. U. S. A. 112 (15) (2015) 4642–4647.
[65] K. Gnanasambandan, A. Magis, P.P. Sayeski, The constitutive activation of Jak2-V617F is mediated by a π stacking mechanism involving phenylalanines 595 and 617, Biochemistry 49 (46) (2010) 9972–9984.
[66] E. Leroy, A. Dusa, D. Colau, A. Motamedi, X. Cahu, C. Mouton, et al., Uncoupling JAK2 V617F activation from cytokine-induced signalling by modulation of JH2 alphaC helix, Biochem. J. 473 (11) (2016) 1579–1591.
[67] R.M. Bandaranayake, D. Ungureanu, Y. Shan, D.E. Shaw, O. Silvennoinen, S.R. Hubbard, Crystal structures of the JAK2 pseudokinase domain and the pathogenic mutant V617F, Nat. Struct. Mol. Biol. 19 (2012) 754–759.
[68] D. Ungureanu, J. Wu, T. Pekkala, Y. Niranjan, C. Young, O.N. Jensen, et al., The pseudokinase domain of JAK2 is a dual-specificity protein kinase that negatively regulates cytokine signaling, Nat. Struct. Mol. Biol. 18 (9) (2011) 971–976.
[69] S.A. Robertson, R.I. Koleva, L.S. Argetsinger, C. Carter-Su, J.A. Marto, E.P. Feener, et al., Regulation of Jak2 function by phosphorylation of Tyr317 and Tyr637 during cytokine signaling, Mol. Cell. Biol. 29 (12) (2009) 3367–3378.
[70] M. Funakoshi-Tago, S. Pelletier, T. Matsuda, E. Parganas, J.N. Ihle, Receptor specific downregulation of cytokine signaling by autophosphorylation in the FERM domain of Jak2, EMBO J. 25 (20) (2006) 4763–4772.
[71] D. Xu, C.K. Qu, Protein tyrosine phosphatases in the JAK/STAT pathway, Front Biosci. 13 (2008) 4925–4932.
[72] R. Frankson, Z.H. Yu, Y. Bai, Q. Li, R.Y. Zhang, Z.Y. Zhang, Therapeutic targeting of oncogenic tyrosine phosphatases, Cancer Res. 77 (21) (2017) 5701–5705.
[73] R. Starr, T.A. Willson, E.M. Viney, L.J. Murray, J.R. Rayner, B.J. Jenkins, et al., A family of cytokine-inducible inhibitors of signalling, Nature 387 (6636) (1997)

917–921.
[74] J.J. Babon, N.J. Kershaw, J.M. Murphy, L.N. Varghese, A. Laktyushin, S.N. Young, et al., Suppression of cytokine signaling by SOCS3: Characterization of the mode of inhibition and the basis of its specificity, Immunity 36 (2) (2012) 239–250.
[75] N.J. Kershaw, J.M. Murphy, N.P. Liau, L.N. Varghese, A. Laktyushin, E.L. Whitlock, et al., SOCS3 binds specific receptor–JAK complexes to control cytokine signaling by direct kinase inhibition, Nat. Struct. Mol. Biol. 20 (4) (2013) 469–476.
[76] J.H. Kurzer, P. Saharinen, O. Silvennoinen, C. Carter-Su, Binding of SH2-B family members within a potential negative regulatory region maintains JAK2 in an active state, Mol. Cell. Biol. 26 (17) (2006) 6381–6394.
[77] Z. Li, Y. Zhou, C. Carter-Su, M.G. Myers Jr, L. Rui, SH2B1 enhances leptin signaling by both janus kinase 2 Tyr813 phosphorylation-dependent and-independent mechanisms, Mol. Endocrinol. 21 (9) (2007) 2270–2281.
[78] D.L. Morris, K.W. Cho, L. Rui, Critical role of the src homology 2 (SH2) domain of neuronal SH2B1 in the regulation of body weight and glucose homeostasis in mice, Endocrinology 151 (8) (2010) 3643–3651.
[79] N. Maslah, B. Cassinat, E. Verger, J. Kiladjian, L. Velazquez, The role of LNK/SH2B3 genetic alterations in myeloproliferative neoplasms and other hematological disorders, Leukemia (2017).
[80] M. Nishi, E.D. Werner, B.C. Oh, J.D. Frantz, S. Dhe-Paganon, L. Hansen, et al., Kinase activation through dimerization by human SH2-B, Mol. Cell. Biol. 25 (7) (2005) 2607–2621.
[81] D. Ren, M. Li, C. Duan, L. Rui, Identification of SH2-B as a key regulator of leptin sensitivity, energy balance, and body weight in mice, Cell Metab. 2 (2) (2005) 95–104.
[82] M. Javadi, E. Hofstatter, N. Stickle, B.K. Beattie, R. Jaster, C. Carter-Su, et al., The SH2B1 adaptor protein associates with a proximal region of the erythropoietin receptor, J. Biol. Chem. 287 (31) (2012) 26223–26234.
[83] A. Flotho, F. Melchior, Sumoylation: A regulatory protein modification in health and disease, Annu. Rev. Biochem. 82 (2013) 357–385.
[84] C.D. Chung, J. Liao, B. Liu, X. Rao, P. Jay, P. Berta, et al., Specific inhibition of Stat3 signal transduction by PIAS3, Science 278 (5344) (1997) 1803–1805.
[85] B. Liu, J. Liao, X. Rao, S.A. Kushner, C.D. Chung, D.D. Chang, et al., Inhibition of Stat1-mediated gene activation by PIAS1, Proc. Natl. Acad. Sci. U. S. A. 95 (18) (1998) 10626–10631.
[86] A. Rabellino, C. Andreani, P.P. Scaglioni, The role of PIAS SUMO E3-ligases in cancer, Cancer Res. 77 (7) (2017) 1542–1547.
[87] K. Shuai, Regulation of cytokine signaling pathways by PIAS proteins, Cell Res. 16 (2) (2006) 196–202.
[88] G. Skiniotis, M.J. Boulanger, K.C. Garcia, T. Walz, Signaling conformations of the tall cytokine receptor gp130 when in complex with IL-6 and IL-6 receptor, Nat. Struct. Mol. Biol. 12 (6) (2005) 545–551.
[89] R. Matadeen, W. Hon, J.K. Heath, E.Y. Jones, S. Fuller, The dynamics of signal triggering in a gp130-receptor complex, Structure 15 (4) (2007) 441–448.
[90] X. Wang, P. Lupardus, S.L. LaPorte, K.C. Garcia, Structural biology of shared cytokine receptors, Annu. Rev. Immunol. 27 (2009) 29–60.
[91] J.B. Spangler, I. Moraga, J.L. Mendoza, K.C. Garcia, Insights into cytokine–receptor interactions from cytokine engineering, Annu. Rev. Immunol. 33 (2015) 139–167.
[92] S.R. Hubbard, W.T. Miller, Receptor tyrosine kinases: Mechanisms of activation and signaling, Curr. Opin. Cell Biol. 19 (2) (2007) 117–123.
[93] M.A. Lemmon, J. Schlessinger, Cell signaling by receptor-tyrosine kinases, Cell 141 (7) (2010) 1117.
[94] J.G. Menting, J. Whittaker, M.B. Margetts, L.J. Whittaker, G.K.W. Kong, B.J. Smith, et al., How insulin engages its primary binding site on the insulin receptor, Nature 493 (7431) (2013) 241–245.
[95] K. Saka, M. Kawahara, H. Ueda, T. Nagamune, Activation of target signal transducers utilizing chimeric receptors with signaling-molecule binding motifs, Biotechnol. Bioeng. 109 (6) (2012) 1528–1537.
[96] S.N. Constantinescu, T. Keren, M. Socolovsky, H. Nam, Y.I. Henis, H.F. Lodish, Ligand-independent oligomerization of cell-surface erythropoietin receptor is mediated by the transmembrane domain, Proc. Natl. Acad. Sci. U. S. A. 98 (8) (2001) 4379–4384.
[97] R.J. Brown, J.J. Adams, R.A. Pelekanos, Y. Wan, W.J. McKinstry, K. Palethorpe, et al., Model for growth hormone receptor activation based on subunit rotation within a receptor dimer, Nat. Struct. Mol. Biol. 12 (9) (2005) 814–821.
[98] P.J. Lupardus, G. Skiniotis, A.J. Rice, C. Thomas, S. Fischer, T. Walz, et al., Structural snapshots of full-length Jak1, a transmembrane gp130/IL-6/IL-6Rα cytokine receptor complex, and the receptor-Jak1 holocomplex, Structure 19 (1) (2011) 45–55.
[99] C. Couturier, R. Jockers, Activation of the leptin receptor by a ligand-induced conformational change of constitutive receptor dimers, J. Biol. Chem. 278 (29) (2003) 26604–26611.
[100] Y. Malka, T. Hornakova, Y. Royer, L. Knoops, J.C. Renauld, S.N. Constantinescu, et al., Ligand-independent homomeric and heteromeric complexes between interleukin-2 or -9 receptor subunits and the gamma chain, J. Biol. Chem. 283 (48) (2008) 33569–33577.
[101] N. Seubert, Y. Royer, J. Staerk, K.F. Kubatzky, V. Moucadel, S. Krishnakumar, et al., Active and inactive orientations of the transmembrane and cytosolic domains of the erythropoietin receptor dimer, Mol. Cell 12 (5) (2003) 1239–1250.
[102] X. Lu, A.W. Gross, H.F. Lodish, Active conformation of the erythropoietin receptor: Random and cysteine-scanning mutagenesis of the extracellular juxtamembrane and transmembrane domains, J. Biol. Chem. 281 (11) (2006) 7002–7011.
[103] J. Staerk, J. Defour, C. Pecquet, E. Leroy, H. Antoine-Poirel, I. Brett, et al., Orientation-specific signalling by thrombopoietin receptor dimers, EMBO J. 30 (21) (2011) 4398–4413.
[104] A.J. Brooks, W. Dai, M.L. O'Mara, D. Abankwa, Y. Chhabra, R.A. Pelekanos, et al., Mechanism of activation of protein kinase JAK2 by the growth hormone receptor, Science 344 (6185) (2014) 1249783.
[105] I. Moraga, G. Wernig, S. Wilmes, V. Gryshkova, C.P. Richter, W. Hong, et al., Tuning cytokine receptor signaling by re-orienting dimer geometry with surrogate ligands, Cell 160 (6) (2015) 1196–1208.
[106] N.R. Leitner, A. Witalisz-Siepracka, B. Strobl, M. Müller, Tyrosine kinase 2–Surveillant of tumours and bona fide oncogene, Cytokine 89 (2017) 209–218.
[107] P. Macchi, A. Villa, S. Giliani, M.G. Sacco, A. Frattini, F. Porta, et al., Mutations of jak-3 gene in patients with autosomal severe combined immune deficiency (SCID), Nature 377 (6544) (1995) 65–68.
[108] S.M. Russell, N. Tayebi, H. Nakajima, M.C. Riedy, J.L. Roberts, M.J. Aman, et al., Mutation of Jak3 in a patient with SCID: Essential role of Jak3 in lymphoid development, Science 270 (5237) (1995) 797–800.
[109] N. Meydan, T. Grunberger, H. Dadi, M. Shahar, E. Arpaia, Z. Lapidot, et al., Inhibition of acute lymphoblastic leukaemia by a jak-2 inhibitor, Nature 379 (6566) (1996) 645–648.
[110] P. Peeters, S.D. Raynaud, J. Cools, I. Wlodarska, J. Grosgeorge, P. Philip, et al., Fusion of TEL, the ETS-variant gene 6 (ETV6), to the receptor-associated kinase JAK2 as a result of t(9;12) in a lymphoid and t(9;15;12) in a myeloid leukemia, Blood 90 (7) (1997) 2535–2540.
[111] M. Bousquet, C. Quelen, V. De Mas, E. Duchayne, B. Roquefeuil, G. Delsol, et al., The t (8; 9)(p22; p24) translocation in atypical chronic myeloid leukaemia yields a new PCM1-JAK2 fusion gene, Oncogene 24 (48) (2005) 7248–7252.
[112] A. Reiter, C. Walz, A. Watmore, C. Schoch, I. Blau, B. Schlegelberger, et al., The t (8;9)(p22;p24) is a recurrent abnormality in chronic and acute leukemia that fuses PCM1 to JAK2, Cancer Res. 65 (7) (2005) 2662–2667.
[113] M.M. Patnaik, R.A. Knudson, N. Gangat, C.A. Hanson, A. Pardanani, A. Tefferi, et al., Chromosome 9p24 abnormalities: Prevalence, description of novel JAK2 translocations, JAK2V617F mutation analysis and clinicopathologic correlates, Eur. J. Haematol. 84 (6) (2010) 518–524.
[114] M. Kawamura, T. Taki, H. Kaku, K. Ohki, Y. Hayashi, Identification of SPAG9 as a novel JAK2 fusion partner gene in pediatric acute lymphoblastic leukemia with t (9; 17)(p24; q21), Genes Chromosomes Cancer 54 (7) (2015) 401–408.
[115] F. Griesinger, H. Hennig, F. Hillmer, M. Podleschny, R. Steffens, A. Pies, et al., A BCR–JAK2 fusion gene as the result of at (9; 22)(p24; q11. 2) translocation in a patient with a clinically typical chronic myeloid leukemia, Genes Chromosomes Cancer 44 (3) (2005) 329–333.
[116] E.J. Baxter, L.M. Scott, P.J. Campbell, C. East, N. Fourouclas, S. Swanton, et al., Acquired mutation of the tyrosine kinase JAK2 in human myeloproliferative disorders, Lancet 365 (9464) (2005) 1054–1061.
[117] C. James, V. Ugo, J.P. Le Couédic, J. Staerk, F. Delhommeau, C. Lacout, et al., A unique clonal JAK2 mutation leading to constitutive signalling causes polycythaemia vera, Nature 434 (7037) (2005) 1144–1148.
[118] R. Kralovics, F. Passamonti, A.S. Buser, S.S. Teo, R. Tiedt, J.R. Passweg, et al., A gain-of-function mutation of JAK2 in myeloproliferative disorders, N. Engl. J. Med. 352 (17) (2005) 1779–1790.
[119] R.L. Levine, M. Wadleigh, J. Cools, B.L. Ebert, G. Wernig, B.J.P. Huntly, et al., Activating mutation in the tyrosine kinase JAK2 in polycythemia vera, essential thrombocythemia, and myeloid metaplasia with myelofibrosis, Cancer Cell 7 (4) (2005) 387–397.
[120] T.S. Larsen, N. Pallisgaard, M.B. Møller, H.C. Hasselbalch, The JAK2 V617F allele burden in essential thrombocythemia, polycythemia vera and primary myelofibrosis–impact on disease phenotype, Eur. J. Haematol. 79 (6) (2007) 508–515.
[121] L. Teofili, M. Martini, T. Cenci, G. Petrucci, L. Torti, S. Storti, et al., Different STAT-3 and STAT-5 phosphorylation discriminates among ph-negative chronic myeloproliferative diseases and is independent of the V617F JAK-2 mutation, Blood 110 (1) (2007) 354–359.
[122] C. Vicente, I. Vázquez, N. Marcotegui, A. Conchillo, C. Carranza, G. Rivell, et al., JAK2-V617F activating mutation in acute myeloid leukemia: Prognostic impact and association with other molecular markers, Leukemia 21 (11) (2007) 2386–2391.
[123] H.J. Xie, H.J. Bae, J.H. Noh, J.W. Eun, J.K. Kim, K.H. Jung, et al., Mutational analysis of JAK1 gene in human hepatocellular carcinoma, Neoplasma 56 (2) (2009) 136–140.
[124] A.M. Vannucchi, E. Antonioli, P. Guglielmelli, A. Pancrazzi, V. Guerini, G. Barosi, et al., Characteristics and clinical correlates of MPL 515W > L/K mutation in essential thrombocythemia, Blood 112 (3) (2008) 844–847.
[125] J. Defour, M. Itaya, V. Gryshkova, I.C. Brett, C. Pecquet, T. Sato, et al., Tryptophan at the transmembrane–cytosolic junction modulates thrombopoietin receptor dimerization and activation, Proc. Natl. Acad. Sci. U. S. A. (2013).
[126] E. Leroy, J. Defour, T. Sato, S. Dass, V. Gryshkova, S.M. Marlar, et al., His499 regulates dimerization and prevents oncogenic activation by asparagine mutations of the human thrombopoietin receptor, J. Biol. Chem. (2015) jbc. M115. 696534.
[127] M. Araki, Y. Yang, N. Masubuchi, Y. Hironaka, H. Takei, S. Morishita, et al., Activation of the thrombopoietin receptor by mutant calreticulin in CALR-mutant myeloproliferative neoplasms, Blood 127 (10) (2016) 1307–1316.
[128] S. Elf, N.S. Abdelfattah, E. Chen, J. Perales-Paton, E.A. Rosen, A. Ko, et al., Mutant calreticulin requires both its mutant C-terminus and the thrombopoietin receptor for oncogenic transformation, Cancer Discov. 6 (4) (2016) 368–381.
[129] S.T. Oh, J. Gotlib, JAK2 V617F and beyond: Role of genetics and aberrant signaling in the pathogenesis of myeloproliferative neoplasms, Expert. Rev. Hematol. 3 (3) (2010) 323–337.
[130] M.F. McMullin, H. Cario, LNK mutations and myeloproliferative disorders, Am. J. Hematol. 91 (2) (2016) 248–251.

[131] J.J. O'Shea, M. Husa, D. Li, S.R. Hofmann, W. Watford, J.L. Roberts, et al., Jak3 and the pathogenesis of severe combined immunodeficiency, Mol. Immunol. 41 (6) (2004) 727–737.
[132] Y. Minegishi, M. Saito, T. Morio, K. Watanabe, K. Agematsu, S. Tsuchiya, et al., Human tyrosine kinase 2 deficiency reveals its requisite roles in multiple cytokine signals involved in innate and acquired immunity, Immunity 25 (5) (2006) 745–755.
[133] S.S. Kilic, M. Hacimustafaoglu, S. Boisson-Dupuis, A.Y. Kreins, A.V. Grant, L. Abel, et al., A patient with tyrosine kinase 2 deficiency without hyper-IgE syndrome, J. Pediatr. 160 (6) (2012) 1055–1057.
[134] T. Sanda, J.W. Tyner, A. Gutierrez, V.N. Ngo, J. Glover, B.H. Chang, et al., TYK2-STAT1-BCL2 pathway dependence in T-cell acute lymphoblastic leukemia, Cancer Discov 3 (5) (2013) 564–577.
[135] E. Waanders, B. Scheijen, M. Jongmans, H. Venselaar, S. van Reijmersdal, A. van Dijk, et al., Germline activating TYK2 mutations in pediatric patients with two primary acute lymphoblastic leukemia occurrences, Leukemia 31 (4) (2017) 821.
[136] M.C. Trengove, A.C. Ward, SOCS proteins in development and disease, Am J Clin Exp Immunol 2 (1) (2013) 1–29.
[137] Y. Liang, W. Xu, H. Peng, H. Pan, D. Ye, SOCS signaling in autoimmune diseases: Molecular mechanisms and therapeutic implications, Eur. J. Immunol. 44 (5) (2014) 1265–1275.
[138] T. Nakagawa, S. Iida, T. Osanai, H. Uetake, T. Aruga, Y. Toriya, et al., Decreased expression of SOCS-3 mRNA in breast cancer with lymph node metastasis, Oncol. Rep. 19 (1) (2008) 33–39.
[139] M. Harada, K. Nakashima, T. Hirota, M. Shimizu, S. Doi, K. Fujita, et al., Functional polymorphism in the suppressor of cytokine signaling 1 gene associated with adult asthma, Am. J. Respir. Cell Mol. Biol. 36 (4) (2007) 491–496.
[140] P. Isomäki, T. Alanärä, P. Isohanni, A. Lagerstedt, M. Korpela, T. Moilanen, et al., The expression of SOCS is altered in rheumatoid arthritis, Rheumatology 46 (10) (2007) 1538–1546.
[141] C. Daegelmann, G. Herberth, S. Röder, O. Herbarth, T. Giese, U. Krämer, et al., Association between suppressors of cytokine signalling, t-helper type 1/t-helper type 2 balance and allergic sensitization in children, Clin. Exp. Allergy 38 (3) (2008) 438–448.
[142] J.J. O'Shea, D.M. Schwartz, A.V. Villarino, M. Gadina, I.B. McInnes, A. Laurence, The JAK-STAT pathway: Impact on human disease and therapeutic intervention, Annu. Rev. Med. 66 (2015) 311–328.
[143] A.V. Villarino, Y. Kanno, J.R. Ferdinand, J.J. O'Shea, Mechanisms of jak/STAT signaling in immunity and disease, J. Immunol. 194 (1) (2015) 21–27.
[144] L. Tan, K. Akahane, R. McNally, K.M.S.E. Reyskens, S.B. Ficarro, S. Liu, et al., Development of selective covalent janus kinase 3 inhibitors, J. Med. Chem. 58 (16) (2015) 6589–6606.
[145] J.S. Tokarski, A. Zupa-Fernandez, J.A. Tredup, K. Pike, C. Chang, D. Xie, et al., Tyrosine kinase 2-mediated signal transduction in T lymphocytes is blocked by pharmacological stabilization of its pseudokinase domain, J. Biol. Chem. 290 (17) (2015) 11061–11074.
[146] R. Moslin, D. Gardner, J. Santella, Y. Zhang, J. Duncia, C. Liu, et al., Identification of imidazo [1, 2-b] pyridazine TYK2 pseudokinase ligands as potent and selective allosteric inhibitors of TYK2 signalling, Med. Chem. Commun. 8 (4) (2017) 700–712.
[147] A.S. Newton, L. Deiana, D. Puleo, J.A. Cisneros, K.J. Cutrona, J. Schlessinger, et al., JAK2 JH2 fluorescence polarization assay and crystal structures for complexes with three small molecules, ACS Med. Chem. Lett. 8 (6) (2017) 614–617.
[148] D. Puleo, K. Kucera, H. Hammaren, D. Ungureanu, A. Newton, O. Silvennoinen, et al., Identification and characterization of JAK2 pseudokinase domain small molecule binders, ACS Med. Chem. Lett. 8 (6) (2017) 618–621.
[149] E. Leroy, S.N. Constantinescu, Rethinking JAK2 inhibition: Towards novel strategies of more specific and versatile janus kinase inhibition, Leukemia 31 (5) (2017) 1023–1038.
[150] J. Renauld, Class II cytokine receptors and their ligands: Key antiviral and inflammatory modulators, Nat. Rev. Immunol. 3 (8) (2003) 667–676.
[151] S. Rutz, X. Wang, W. Ouyang, The IL-20 subfamily of cytokines—from host defence to tissue homeostasis, Nat. Rev. Immunol. 14 (12) (2014) 783–795.
[152] S. Aparicio-Siegmund, C. Garbers, The biology of interleukin-27 reveals unique pro-and anti-inflammatory functions in immunity, Cytokine Growth Factor Rev. 26 (5) (2015) 579–586.
[153] D.A. Vignali, V.K. Kuchroo, IL-12 family cytokines: Immunological playmakers, Nat. Immunol. 13 (8) (2012) 722–728.
[154] C. Schindler, C. Plumlee, Inteferons pen the JAK–STAT pathway, Semin. Cell Dev. Biol. 19 (4) (2008) 311–318.
[155] S. Baker, S. Rane, E. Reddy, Hematopoietic cytokine receptor signaling, Oncogene 26 (47) (2007) 6724–6737.
[156] S. Commins, J.W. Steinke, L. Borish, The extended IL-10 superfamily: IL-10, IL-19, IL-20, IL-22, IL-24, IL-26, IL-28, and IL-29, J. Allergy Clin. Immunol. 121 (5) (2008) 1108–1111.
[157] A. Quintás-Cardama, H. Kantarjian, J. Cortes, S. Verstovsek, Janus kinase inhibitors for the treatment of myeloproliferative neoplasias and beyond, Nat. Rev. Drug Discov. 10 (2) (2011) 127–140.
[158] S.V. Kotenko, IFN-λs, Curr. Opin. Immunol. 23 (5) (2011) 583–590.
[159] G. Dey, A. Radhakrishnan, N. Syed, J.K. Thomas, A. Nadig, K. Srikumar, et al., Signaling network of oncostatin M pathway, J. Cell Commun. Signal 7 (2013) 103–108.
[160] N.A. Sims, Cardiotrophin-like cytokine factor 1 (CLCF1) and neuropoietin (NP) signalling and their roles in development, adulthood, cancer and degenerative disorders, Cytokine Growth Factor Rev. 26 (5) (2015) 517–522.
[161] Q. Zhang, P. Putheti, Q. Zhou, Q. Liu, W. Gao, Structures and biological functions of IL-31 and IL-31 receptors, Cytokine Growth Factor Rev. 19 (5) (2008) 347–356.
[162] P.E. Kovanen, W.J. Leonard, Cytokines and immunodeficiency diseases: Critical roles of the γc-dependent cytokines interleukins 2, 4, 7, 9, 15, and 21, and their signaling pathways, Immunol. Rev. 202 (1) (2004) 67–83.
[163] J. Zhong, J. Sharma, R. Raju, S.M. Palapetta, T. Prasad, T. Huang, et al., TSLP signaling pathway map: A platform for analysis of TSLP-mediated signaling, Database (Oxford) 2014 (2014) bau007.
[164] S.J. Rodig, M.A. Meraz, J.M. White, P.A. Lampe, J.K. Riley, C.D. Arthur, et al., Disruption of the Jak1 gene demonstrates obligatory and nonredundant roles of the jaks in cytokine-induced biologic responses, Cell 93 (3) (1998) 373–383.
[165] L.D. Notarangelo, P. Mella, A. Jones, Basile G de Saint, G. Savoldi, T. Cranston, et al., Mutations in severe combined immune deficiency (SCID) due to JAK3 deficiency, Hum. Mutat. 18 (4) (2001) 255–263.
[166] K. Gnanasambandan, P. Sayeski, A structure-function perspective of jak2 mutations and implications for alternate drug design strategies: The road not taken, Curr. Med. Chem. 18 (30) (2011) 4659–4673.
[167] Y. Ren, Y. Zhang, R.Z. Liu, D.A. Fenstermacher, K.L. Wright, J.K. Teer, et al., JAK1 truncating mutations in gynecologic cancer define new role of cancer-associated protein tyrosine kinase aberrations, Sci. Rep. 3 (2013) 3042.
[168] E. Flex, V. Petrangeli, L. Stella, S. Chiaretti, T. Hornakova, L. Knoops, et al., Somatically acquired JAK1 mutations in adult acute lymphoblastic leukemia, J. Exp. Med. 205 (4) (2008) 751–758.
[169] M. Neumann, S. Heesch, C. Schlee, S. Schwartz, N. Gokbuget, D. Hoelzer, et al., Whole-exome sequencing in adult ETP-ALL reveals a high rate of DNMT3A mutations, Blood 121 (23) (2013) 4749–4752.
[170] C. Haan, H. Is'harc, H.M. Hermanns, H. Schmitz-Van De Leur, I.M. Kerr, P.C. Heinrich, et al., Mapping of a region within the N terminus of Jak1 involved in cytokine receptor interaction, J. Biol. Chem. 276 (40) (2001) 37451–37458.
[171] K. Cante-Barrett, J.C. Uitdehaag, J.P. Meijerink, Structural modeling of JAK1 mutations in T-cell acute lymphoblastic leukemia reveals a second contact site between pseudokinase and kinase domains, Haematologica 101 (5) (2016) e189–e191.
[172] S. Yang, C. Luo, Q. Gu, Q. Xu, G. Wang, H. Sun, et al., Activating JAK1 mutation may predict the sensitivity of JAK-STAT inhibition in hepatocellular carcinoma, Oncotarget 7 (5) (2016) 5461–5469.
[173] Z. Kan, B.S. Jaiswal, J. Stinson, V. Janakiraman, D. Bhatt, H.M. Stern, et al., Diverse somatic mutation patterns and pathway alterations in human cancers, Nature 466 (7308) (2010) 869–873.
[174] Q. Wang, H. Qiu, H. Jiang, L. Wu, S. Dong, J. Pan, et al., Mutations of PHF6 are associated with mutations of NOTCH1, JAK1 and rearrangement of SET-NUP214 in T-cell acute lymphoblastic leukemia, Haematologica 96 (12) (2011) 1808–1814.
[175] S. Suryani, L.S. Bracken, R.C. Harvey, K.C. Sia, H. Carol, I. Chen, et al., Evaluation of the in vitro and in vivo efficacy of the JAK inhibitor AZD1480 against JAK-mutated acute lymphoblastic leukemia, Mol. Cancer Ther. 14 (2) (2014) 364–374.
[176] J. Zhang, L. Ding, L. Holmfeldt, G. Wu, S.L. Heatley, D. Payne-Turner, et al., The genetic basis of early T-cell precursor acute lymphoblastic leukaemia, Nature 481 (7380) (2012) 157–163.
[177] T. Hornakova, L. Springuel, J. Devreux, A. Dusa, S.N. Constantinescu, L. Knoops, et al., Oncogenic JAK1 and JAK2-activating mutations resistant to ATP-competitive inhibitors, Haematologica 96 (6) (2011) 845–853.
[178] E.G. Jeong, M.S. Kim, H.K. Nam, C.K. Min, S. Lee, Y.J. Chung, et al., Somatic mutations of JAK1 and JAK3 in acute leukemias and solid cancers, Clin. Cancer Res. 14 (12) (2008) 3716–3721.
[179] M. Kleppe, J. Soulier, V. Asnafi, N. Mentens, T. Hornakova, L. Knoops, et al., PTPN2 negatively regulates oncogenic JAK1 in T-cell acute lymphoblastic leukemia, Blood 117 (26) (2011) 7090–7098.
[180] J. Staerk, A. Kallin, J. Demoulin, W. Vainchenker, S.N. Constantinescu, JAK1 and Tyk2 activation by the homologous polycythemia vera JAK2 V617F mutation cross-talk with IGF1 receptor, J. Biol. Chem. 280 (51) (2005) 41893–41899.
[181] D. Eletto, S.O. Burns, I. Angulo, V. Plagnol, K.C. Gilmour, F. Henriquez, et al., Biallelic JAK1 mutations in immunodeficient patient with mycobacterial infection, Nat. Commun. 7 (2016) 13992.
[182] T. Hayashi, Y. Kobayashi, S. Kohsaka, K. Sano, The mutation in the ATP-binding region of JAK1, identified in human uterine leiomyosarcomas, results in defective interferon-γ inducibility of TAP1 and LMP2, Oncogene 25 (29) (2006) 4016–4026.
[183] C.D. Camps, N. Petousi, C. Bento, H. Cario, R.R. Copley, M.F. McMullin, et al., Gene panel sequencing improves the diagnostic work-up of patients with idiopathic erythrocytosis and identifies new mutations, Haematologica 101 (11) (2016) 1306–1318.
[184] L. Lanikova, O. Babosova, S. Swierczek, L. Wang, D.A. Wheeler, V. Divoky, et al., Coexistence of gain-of-function JAK2 germline mutations with JAK2V617F in polycythemia vera, Blood 128 (18) (2016) 2266–2270.
[185] P. Aranaz, C. Ormazábal, C. Hurtado, I. Erquiaga, M.J. Calasanz, M. García-Delgado, et al., A new potential oncogenic mutation in the FERM domain of JAK2 in BCR/ABL1-negative and V617F-negative chronic myeloproliferative neoplasms revealed by a comprehensive screening of 17 tyrosine kinase coding genes, Cancer Genet. Cytogenet. 199 (1) (2010) 1–8.
[186] W. Ma, H. Kantarjian, X. Zhang, C.H. Yeh, Z.J. Zhang, S. Verstovsek, et al., Mutation profile of JAK2 transcripts in patients with chronic myeloproliferative neoplasias, J. Mol. Diagn. 11 (1) (2009) 49–53.
[187] L.M. Scott, W. Tong, R.L. Levine, M.A. Scott, P.A. Beer, M.R. Stratton, et al., JAK2 exon 12 mutations in polycythemia vera and idiopathic erythrocytosis, N. Engl. J. Med. 356 (5) (2007) 459–468.
[188] S. Delic, D. Rose, W. Kern, N. Nadarajah, C. Haferlach, T. Haferlach, et al., Application of an NGS-based 28-gene panel in myeloproliferative neoplasms

reveals distinct mutation patterns in essential thrombocythaemia, primary myelofibrosis and polycythaemia vera, Br. J. Haematol. 175 (3) (2016) 419–426.
[189] S.L. Etheridge, M.E. Cosgrove, V. Sangkhae, L.M. Corbo, M.E. Roh, M.A. Seeliger, et al., A novel activating, germline JAK2 mutation, JAK2R564Q, causes familial essential thrombocytosis, Blood 123 (7) (2014) 1059–1068.
[190] A.J. Mead, M.J. Rugless, S.E.W. Jacobsen, A. Schuh, Germline JAK2 mutation in a family with hereditary thrombocytosis, N. Engl. J. Med. 366 (10) (2012) 967–969.
[191] A.J. Mead, O. Chowdhury, C. Pecquet, A. Dusa, P. Woll, D. Atkinson, et al., Impact of isolated germline JAK2V617I mutation on human hematopoiesis, Blood 121 (20) (2013) 4156–4165.
[192] Q.Y. Wu, F. Li, H.Y. Guo, J. Cao, C. Chen, W. Chen, et al., Amino acid residue E543 in JAK2 C618R is a potential therapeutic target for myeloproliferative disorders caused by JAK2 C618R mutation, Arch. Biochem. Biophys. 528 (1) (2012) 57–66.
[193] S. Schnittger, U. Bacher, W. Kern, M. Schröder, T. Haferlach, C. Schoch, Report on two novel nucleotide exchanges in the JAK2 pseudokinase domain: D620E and E627E, Leukemia 20 (12) (2006) 2195–2197.
[194] C.G. Mullighan, J. Zhang, R.C. Harvey, J.R. Collins-Underwood, B.A. Schulman, L.A. Phillips, et al., JAK mutations in high-risk childhood acute lymphoblastic leukemia, Proc Natl Acad Sci U S A 106 (23) (2009) 9414–9418.
[195] D. Bercovich, I. Ganmore, L.M. Scott, G. Wainreb, Y. Birger, A. Elimelech, et al., Mutations of JAK2 in acute lymphoblastic leukaemias associated with down's syndrome, Lancet 372 (9648) (2008) 1484–1492.
[196] C. Marty, C. Saint-Martin, C. Pecquet, S. Grosjean, J. Saliba, C. Mouton, et al., Germ-line JAK2 mutations in the kinase domain are responsible for hereditary thrombocytosis and are resistant to JAK2 and HSP90 inhibitors, Blood 123 (9) (2014) 1372–1383.
[197] K. Kapralova, M. Horvathova, C. Pecquet, J. Fialova Kucerova, D. Pospisilova, E. Leroy, et al., Cooperation of germ line JAK2 mutations E846D and R1063H in hereditary erythrocytosis with megakaryocytic atypia, Blood 128 (10) (2016) 1418–1423.
[198] J.L. Roberts, A. Lengi, S.M. Brown, M. Chen, Y.J. Zhou, J.J. O'Shea, et al., Janus kinase 3 (JAK3) deficiency: Clinical, immunologic, and molecular analyses of 10 patients and outcomes of stem cell transplantation, Blood 103 (6) (2004) 2009–2018.
[199] Y. Yamashita, J. Yuan, I. Suetake, H. Suzuki, Y. Ishikawa, Y. Choi, et al., Array-based genomic resequencing of human leukemia, Oncogene 29 (25) (2010) 3723.
[200] T. Sato, T. Toki, R. Kanezaki, G. Xu, K. Terui, H. Kanegane, et al., Functional analysis of JAK3 mutations in transient myeloproliferative disorder and acute megakaryoblastic leukaemia accompanying down syndrome, Br. J. Haematol. 141 (5) (2008) 681–688.
[201] N.A. Cacalano, T.S. Migone, F. Bazan, E.P. Hanson, M. Chen, F. Candotti, et al., Autosomal SCID caused by a point mutation in the N-terminus of Jak3: Mapping of the Jak3-receptor interaction domain, EMBO J. 18 (6) (1999) 1549–1558.
[202] W. Tang, H. Huo, J. Zhu, H. Ji, W. Zou, L. Xu, et al., Critical sites for the interaction between IL-2Rγ and JAK3 and the following signaling, Biochem. Biophys. Res. Commun. 283 (3) (2001) 598–605.
[203] D.K. Walters, T. Mercher, T. Gu, T. O'Hare, J.W. Tyner, M. Loriaux, et al., Activating alleles of JAK3 in acute megakaryoblastic leukemia, Cancer Cell 10 (1) (2006) 65–75.
[204] L. Riera, E. Lasorsa, L. Bonello, F. Sismondi, F. Tondat, C. Di Bello, et al., Description of a novel janus kinase 3 P132A mutation in acute megakaryoblastic leukemia and demonstration of previously reported janus kinase 3 mutations in normal subjects, Leuk Lymphoma 52 (9) (2011) 1742–1750.
[205] S. De Vita, C. Mulligan, S. McElwaine, F. Dagna-Bricarelli, M. Spinelli, G. Basso, et al., Loss-of-function JAK3 mutations in TMD and AMKL of down syndrome, Br. J. Haematol. 137 (4) (2007) 337–341.
[206] N.E. Elliott, S.M. Cleveland, V. Grann, J. Janik, T.A. Waldmann, U.P. Davé, FERM domain mutations induce gain of function in JAK3 in adult T-cell leukemia/lymphoma, Blood 118 (14) (2011) 3911–3921.
[207] K.C. Kadakia, S.A. Tomlins, S.K. Sanghvi, A.K. Cani, K. Omata, D.H. Hovelson, et al., Comprehensive serial molecular profiling of an "N of 1" exceptional non-responder with metastatic prostate cancer progressing to small cell carcinoma on treatment, J. Hematol. Oncol. 8 (1) (2015) 109.
[208] S. Degryse, C.E. de Bock, L. Cox, S. Demeyer, O. Gielen, N. Mentens, et al., JAK3 mutants transform hematopoietic cells through JAK1 activation, causing T-cell acute lymphoblastic leukemia in a mouse model, Blood 124 (20) (2014) 3092–3100.
[209] C. Vicente, C. Schwab, M. Broux, E. Geerdens, S. Degryse, S. Demeyer, et al., Targeted sequencing identifies associations between IL7R-JAK mutations and epigenetic modulators in T-cell acute lymphoblastic leukemia, Haematologica 100 (10) (2015) 1301–1310.
[210] F. Candotti, S.A. Oakes, J.A. Johnston, S. Giliani, R.F. Schumacher, P. Mella, et al., Structural and functional basis for JAK3-deficient severe combined immunodeficiency, Blood 90 (10) (1997) 3996–4003.
[211] R.F. Schumacher, P. Mella, R. Badolato, M. Fiorini, G. Savoldi, S. Giliani, et al., Complete genomic organization of the human JAK3 gene and mutation analysis in severe combined immunodeficiency by single-strand conformation polymorphism, Hum. Genet. 106 (1) (2000) 73–79.
[212] J. Li, H. Nara, M. Rahman, F.M. Juliana, A. Araki, H. Asao, Impaired IL-7 signaling may explain a case of atypical JAK3-SCID, Cytokine 49 (2) (2010) 221–228.
[213] A.K. Bergmann, S. Schneppenheim, M. Seifert, M.J. Betts, A. Haake, C. Lopez, et al., Recurrent mutation of JAK3 in t-cell prolymphocytic leukemia, Genes Chromosomes Cancer 53 (4) (2014) 309–316.
[214] M.G. Cornejo, M.G. Kharas, M.B. Werneck, S. Le Bras, S.A. Moore, B. Ball, et al., Constitutive JAK3 activation induces lymphoproliferative syndromes in murine bone marrow transplantation models, Blood 113 (12) (2009) 2746–2754.
[215] G.S. Martinez, J.A. Ross, R.A. Kirken, Transforming mutations of Jak3 (A573V and M511I) show differential sensitivity to selective Jak3 inhibitors, Clin. Cancer Drugs 3 (2) (2016) 131–137.
[216] S.H. Sim, S. Kim, T.M. Kim, Y.K. Jeon, S.J. Nam, Y. Ahn, et al., Novel JAK3-activating mutations in extranodal NK/T-cell lymphoma, nasal type, Am. J. Pathol. 187 (5) (2017) 980–986.
[217] H. Kiyoi, S. Yamaji, S. Kojima, T. Naoe, JAK3 mutations occur in acute megakaryoblastic leukemia both in down syndrome children and non-down syndrome adults, Leukemia 21 (3) (2007) 574–576.
[218] P. Mella, R.F. Schumacher, T. Cranston, Basile G de Saint, G. Savoldi, L.D. Notarangelo, Eleven novel JAK3 mutations in patients with severe combined immunodeficiency—including the first patients with mutations in the kinase domain, Hum. Mutat. 18 (4) (2001) 355–356.
[219] T. Bains, M. Heinrich, M. Loriaux, C. Beadling, D. Nelson, A. Warrick, et al., Newly described activating JAK3 mutations in T-cell acute lymphoblastic leukemia, Leukemia 26 (9) (2012) 2144–2146.
[220] E. Losdyck, T. Hornakova, L. Springuel, S. Degryse, O. Gielen, J. Cools, et al., Distinct acute lymphoblastic leukemia (ALL)-associated janus kinase 3 (JAK3) mutants exhibit different cytokine-receptor requirements and JAK inhibitor specificities, J. Biol. Chem. 290 (48) (2015) 29022–29034.
[221] C.M. Mjaanes, R.W. Hendershot, R.R. Quinones, E.W. Gelfand, A novel mutation of intron 22 in janus kinase 3–deficient severe combined immunodeficiency, J. Allergy Clin. Immunol. 119 (6) (2007) 1542–1545.
[222] H. Sakaguchi, Y. Okuno, H. Muramatsu, K. Yoshida, Y. Shiraishi, M. Takahashi, et al., Exome sequencing identifies secondary mutations of SETBP1 and JAK3 in juvenile myelomonocytic leukemia, Nat. Genet. 45 (8) (2013) 937–941.
[223] D. Bellanger, V. Jacquemin, M. Chopin, G. Pierron, O. Bernard, J. Ghysdael, et al., Recurrent JAK1 and JAK3 somatic mutations in T-cell prolymphocytic leukemia, Leukemia 28 (2) (2014) 417–419.
[224] H.D. Ochs, C.I.E. Smith, J.M. Puck, Primary Immunodeficiency Diseases: A Molecular and Cellular Approach, OUP, USA, 2013.
[225] S. Bresolin, P. De Filippi, F. Vendemini, M. D'Alia, M. Zecca, L.H. Meyer, et al., Mutations of SETBP1 and JAK3 in juvenile myelomonocytic leukemia: A report from the italian AIEOP study group, Oncotarget 7 (20) (2016) 28914–28919.
[226] Z.K. Atak, V. Gianfelici, G. Hulselmans, K. De Keersmaecker, A.G. Devasia, E. Geerdens, et al., Comprehensive analysis of transcriptome variation uncovers known and novel driver events in T-cell acute lymphoblastic leukemia, PLoS Genet 9 (12) (2013) e1003997.
[227] D.A. Dyment, M.Z. Cader, M.J. Chao, M.R. Lincoln, K.M. Morrison, G. Disanto, et al., Exome sequencing identifies a novel multiple sclerosis susceptibility variant in the TYK2 gene, Neurology 79 (5) (2012) 406–411.
[228] M.H. Tomasson, Z. Xiang, R. Walgren, Y. Zhao, Y. Kasai, T. Miner, et al., Somatic mutations and germline sequence variants in the expressed tyrosine kinase genes of patients with de novo acute myeloid leukemia, Blood 111 (9) (2008) 4797–4808.
[229] N. Mørk, E. Kofod-Olsen, K. Sørensen, E. Bach, T. Ørntoft, L. Østergaard, et al., Mutations in the TLR3 signaling pathway and beyond in adult patients with herpes simplex encephalitis, Genes Immun. 16 (8) (2015) 552–566.
[230] E. Lopez-Isac, D. Campillo-Davo, L. Bossini-Castillo, S.G. Guerra, S. Assassi, C.P. Simeon, et al., Influence of TYK2 in systemic sclerosis susceptibility: A new locus in the IL-12 pathway, Ann. Rheum. Dis. 75 (8) (2016) 1521–1526.
[231] D. Diogo, L. Bastarache, K.P. Liao, R.R. Graham, R.S. Fulton, J.D. Greenberg, et al., TYK2 protein-coding variants protect against rheumatoid arthritis and autoimmunity, with no evidence of major pleiotropic effects on non-autoimmune complex traits, PLoS One 10 (4) (2015) e1022271.
[232] Z. Li, M. Gakovic, J. Ragimbeau, M. Eloranta, L. Rönnblom, F. Michel, et al., Two rare disease-associated Tyk2 variants are catalytically impaired but signaling competent, J. Immunol. 190 (5) (2013) 2335–2344.
[233] A.C. Hirbe, M. Kaushal, M.K. Sharma, S. Dahiya, M. Pekmezci, A. Perry, et al., Clinical genomic profiling identifies TYK2 mutation and overexpression in patients with neurofibromatosis type 1-associated malignant peripheral nerve sheath tumors, Cancer 123 (7) (2017) 1194–1201.
[234] T. Matsuda, J. Feng, B.A. Witthuhn, Y. Sekine, J.N. Ihle, Determination of the transphosphorylation sites of Jak2 kinase, Biochem. Biophys. Res. Commun. 325 (2) (2004) 586–594.
[235] K. Rikova, A. Guo, Q. Zeng, A. Possemato, J. Yu, H. Haack, et al., Global survey of phosphotyrosine signaling identifies oncogenic kinases in lung cancer, Cell 131 (6) (2007) 1190–1203.
[236] D. Yan, R.E. Hutchison, G. Mohi, Tyrosine 201 is required for constitutive activation of JAK2V617F and efficient induction of myeloproliferative disease in mice, Blood 120 (9) (2012) 1888–1898.
[237] M.D. Godeny, J. Sayyah, D. VonDerLinden, M. Johns, D.A. Ostrov, J. Caldwell-Busby, et al., The N-terminal SH2 domain of the tyrosine phosphatase, SHP-2, is essential for Jak2-dependent signaling via the angiotensin II type AT1 receptor, Cell. Signal. 19 (3) (2007) 600–609.
[238] J. Sayyah, K. Gnanasambandan, S. Kamarajugadda, S. Tsuda, J. Caldwell-Busby, P.P. Sayeski, Phosphorylation of Y372 is critical for Jak2 tyrosine kinase activation, Cell. Signal. 23 (11) (2011) 1806–1815.
[239] J.H. Kurzer, L.S. Argetsinger, Y.J. Zhou, J.L. Kouadio, J.J. O'Shea, C. Carter-Su, Tyrosine 813 is a site of JAK2 autophosphorylation critical for activation of JAK2 by SH2-B beta, Mol. Cell. Biol. 24 (10) (2004) 4557–4570.
[240] L.S. Argetsinger, J.A. Stuckey, S.A. Robertson, R.I. Koleva, J.M. Cline, J.A. Marto, et al., Tyrosines 868, 966, and 972 in the kinase domain of JAK2 are autophosphorylated and required for maximal JAK2 kinase activity, Mol. Endocrinol. 24 (5) (2010) 1062–1076.
[241] M. Funakoshi-Tago, K. Tago, T. Kasahara, E. Parganas, J.N. Ihle, Negative

regulation of Jak2 by its auto-phosphorylation at tyrosine 913 via the epo signaling pathway, Cell. Signal. 20 (11) (2008) 1995–2001.
[242] B.A. Croker, H. Kiu, S.E. Nicholson, SOCS regulation of the JAK/STAT signalling pathway, Semin. Cell Dev. Biol. 19 (4) (2008) 414–422.
[243] E.G. Bochukova, N. Huang, J. Keogh, E. Henning, C. Purmann, K. Blaszczyk, et al., Large, rare chromosomal deletions associated with severe early-onset obesity, Nature 463 (7281) (2010) 666–670.
[244] R. Walters, S. Jacquemont, A. Valsesia, A. De Smith, D. Martinet, J. Andersson, et al., A new highly penetrant form of obesity due to deletions on chromosome 16p11. 2, Nature 463 (7281) (2010) 671–675.
[245] A. Volckmar, F. Bolze, I. Jarick, N. Knoll, A. Scherag, T. Reinehr, et al., Mutation screen in the GWAS derived obesity gene SH2B1 including functional analyses of detected variants, BMC Med. Genomics 5 (1) (2012) 65.
[246] A. de la Chapelle, A.L. Traskelin, E. Juvonen, Truncated erythropoietin receptor causes dominantly inherited benign human erythrocytosis, Proc. Natl. Acad. Sci. U. S. A. 90 (10) (1993) 4495–4499.
[247] I. Iacobucci, Y. Li, K.G. Roberts, S.M. Dobson, J.C. Kim, D. Payne-Turner, et al., Truncating erythropoietin receptor rearrangements in acute lymphoblastic leukemia, Cancer Cell 29 (2) (2016) 186–200.
[248] M. Kleppe, I. Lahortiga, T. El Chaar, K. De Keersmaecker, N. Mentens, C. Graux, et al., Deletion of the protein tyrosine phosphatase gene PTPN2 in T-cell acute lymphoblastic leukemia, Nat. Genet. 42 (6) (2010) 530–535.
[249] M. Porcu, M. Kleppe, V. Gianfelici, E. Geerdens, K. De Keersmaecker, M. Tartaglia, et al., Mutation of the receptor tyrosine phosphatase PTPRC (CD45) in T-cell acute lymphoblastic leukemia, Blood 119 (19) (2012) 4476–4479.
[250] M. Tartaglia, E.L. Mehler, R. Goldberg, G. Zampino, H.G. Brunner, H. Kremer, et al., Mutations in PTPN11, encoding the protein tyrosine phosphatase SHP-2, cause noonan syndrome, Nat. Genet. 29 (4) (2001).
[251] M. Bentires-Alj, J.G. Paez, F.S. David, H. Keilhack, B. Halmos, K. Naoki, et al., Activating mutations of the noonan syndrome-associated SHP2/PTPN11 gene in human solid tumors and adult acute myelogenous leukemia, Cancer Res. 64 (24) (2004) 8816–8820.
[252] J. Ding, H. Komatsu, A. Wakita, M. Kato-Uranishi, M. Ito, A. Satoh, et al., Familial essential thrombocythemia associated with a dominant-positive activating mutation of the c-MPL gene, which encodes for the receptor for thrombopoietin, Blood 103 (11) (2004) 4198–4200.
[253] A.D. Pardanani, R.L. Levine, T. Lasho, Y. Pikman, R.A. Mesa, M. Wadleigh, et al., MPL515 mutations in myeloproliferative and other myeloid disorders: A study of 1182 patients, Blood 108 (10) (2006) 3472–3476.
[254] P.P. Zenatti, D. Ribeiro, W. Li, L. Zuurbier, M.C. Silva, M. Paganin, et al., Oncogenic IL7R gain-of-function mutations in childhood T-cell acute lymphoblastic leukemia, Nat. Genet. 43 (10) (2011) 932–939.
[255] S. Degryse, J. Cools, JAK kinase inhibitors for the treatment of acute lymphoblastic leukemia, J. Hematol. Oncol. 8 (1) (2015) 91.
[256] M. Cazzola, R. Kralovics, From janus kinase 2 to calreticulin: The clinically relevant genomic landscape of myeloproliferative neoplasms, Blood 123 (24) (2014) 3714–3719.
[257] M. Araki, N. Komatsu, Novel molecular mechanism of cellular transformation by a mutant molecular chaperone in myeloproliferative neoplasms, Cancer Sci. 108 (10) (2017) 1907–1912.