Οι ακάλυπτες θεραπευτικές ανάγκες στη Ρευματοειδή αρθρίτιδα και το ενδοκυττάριο μονοπάτι JAK/STAT ως νέος θεραπευτικός στόχος

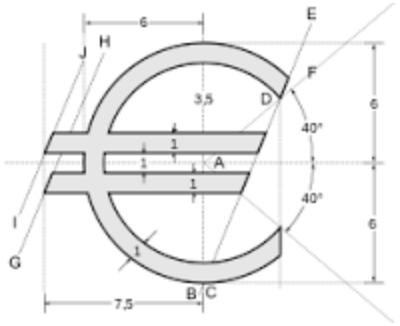
Ανδρέας Γ. Μπούνας
ΡΕΥΜΑΤΟΛΟΓΟΣ

ΠΟΡΤΟ ΧΕΛΙ 28/04 - 01/05/2018

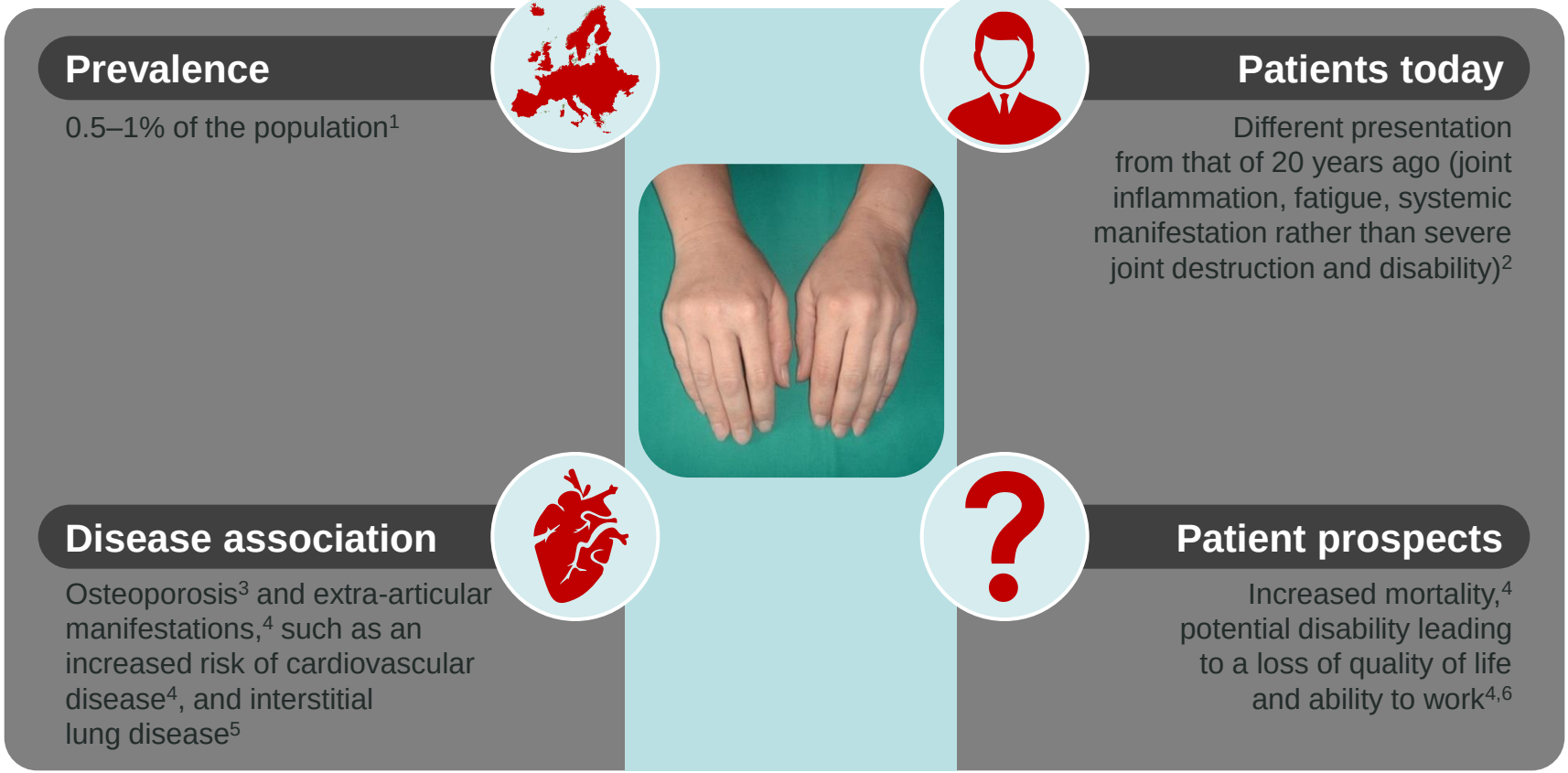
ΔΗΛΩΣΗ ΣΥΜΦΕΡΟΝΤΩΝ

Συμμετοχή την τελευταία 15ετία ως ομιλητής ή παρέχοντας συμβουλευτικές υπηρεσίες σε συναντήσεις στις κάτωθι φαρμακευτικές εταιρείες

- AbbVie (Abbott)
- Bristol-Mayer Squibb
- MSD
- Novartis Hellas
- Pfizer Hellas
- Roche Hellas



RA



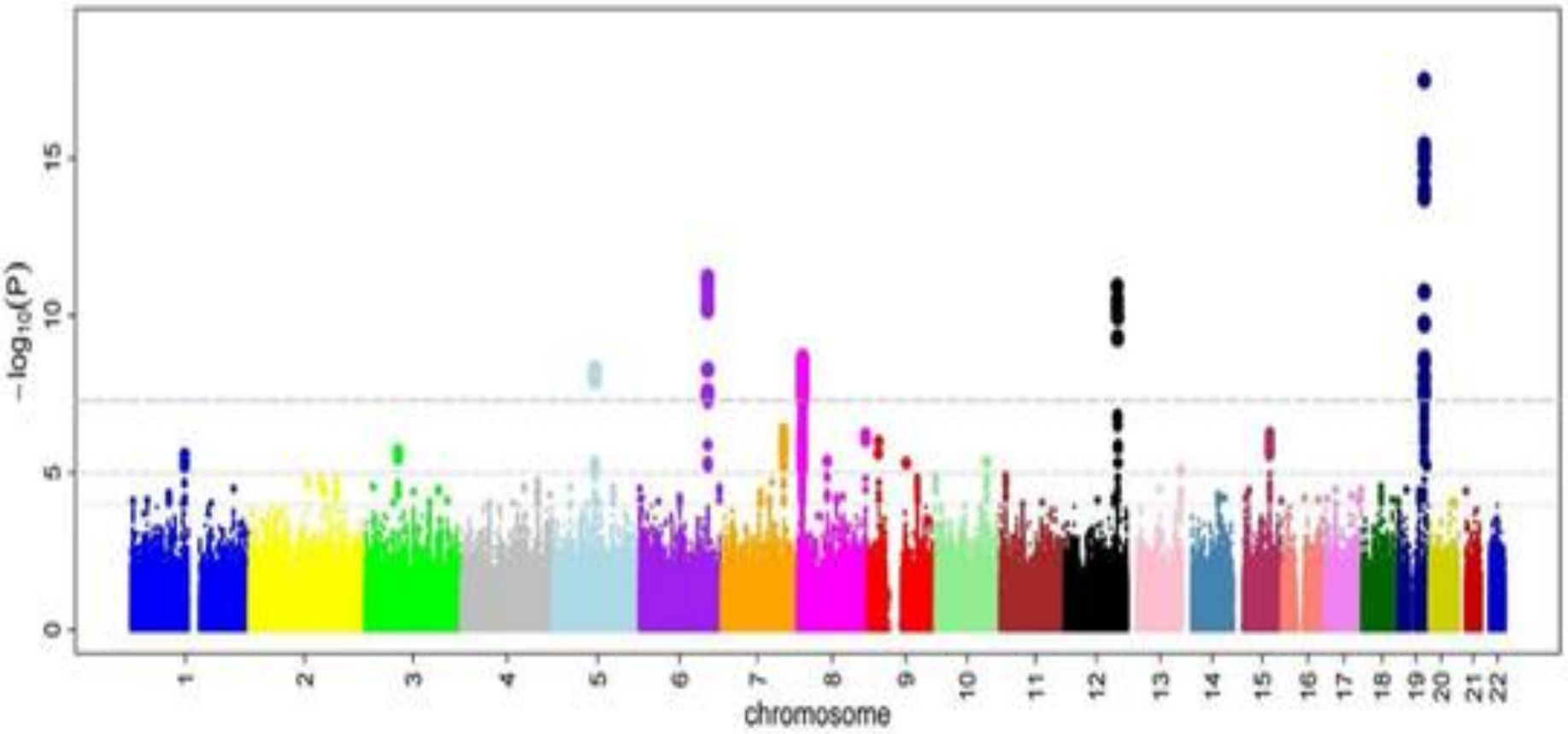
1. Scott DL, et al. *Lancet* 2010;376:1094–1108; 2. Overman CL, et al. *Arthritis Care Res* 2014;5:671–782016 Apr;55:607–14; ; 3. Takahashi K, et al. *BMC Musculoskel Dis* 2015;16:269;
4. Gerlag DM, et al. Rheumatology

5

- Τις τελευταίες λίγες 10ετίες…..

- Μεγάλη πρόοδος σε
  - κατανόηση νόσου
  - θεραπεία

GWAS


−log10(P)
0
5
10
15
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
chromosome

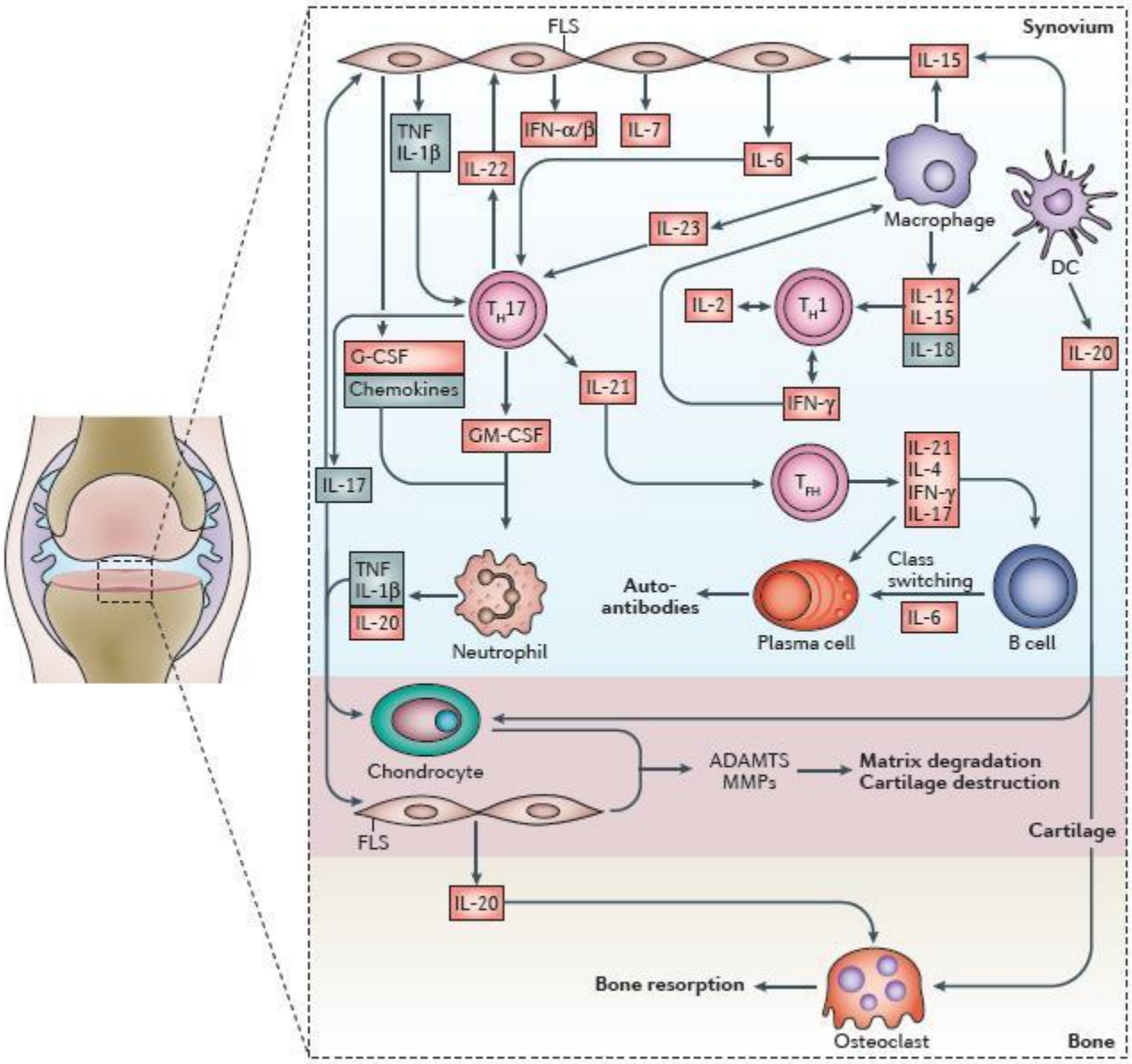
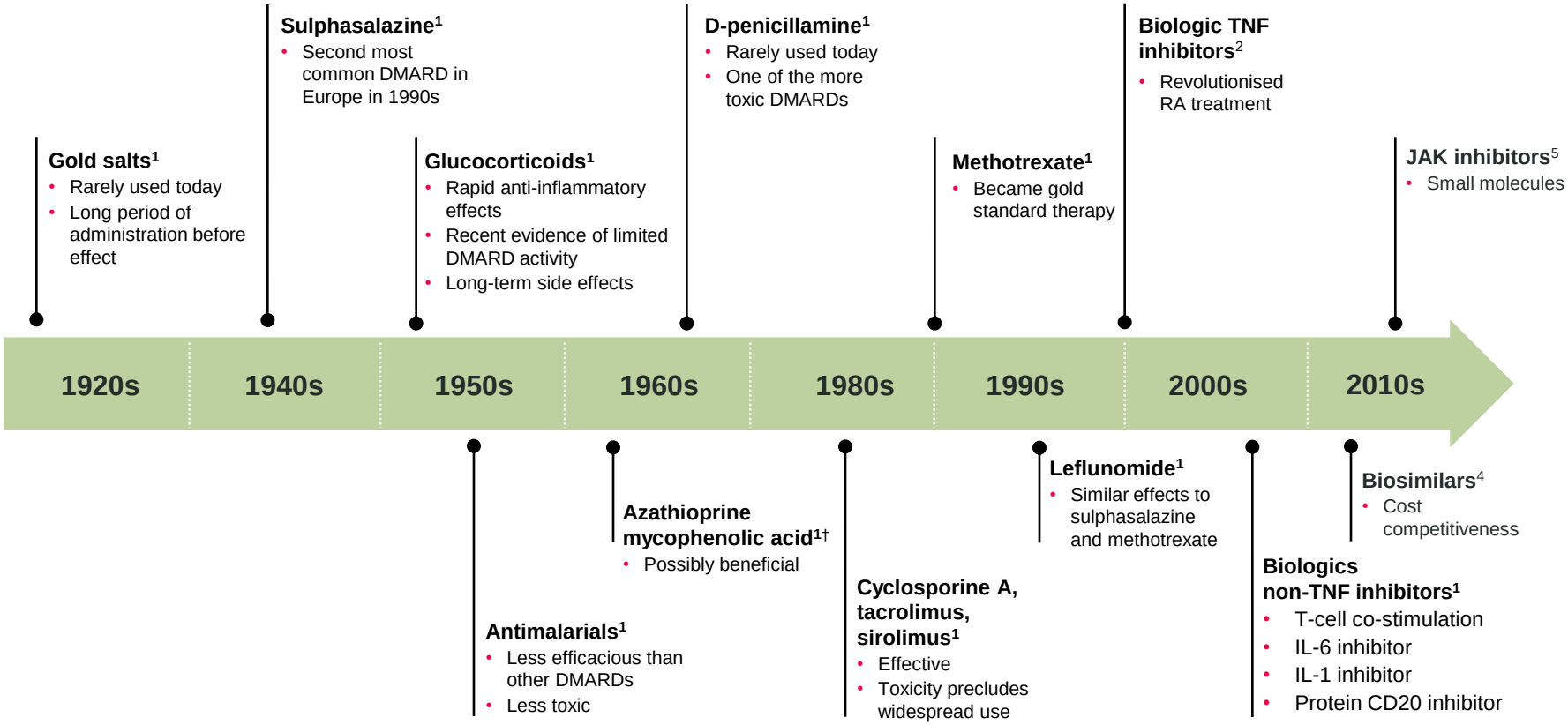


Figure 2 | **Type I and II cytokines (red), which are blocked by Jakinibs, are major drivers of autoimmune diseases such as rheumatoid arthritis.** Within the rheumatoid joint, fibroblast-like synoviocytes (FLSs) are major sources of

Pharmacotherapy for RA has evolved

- Therapies for RA have come a long way in the last 100 years
- However, new agents and treatment strategies are still needed

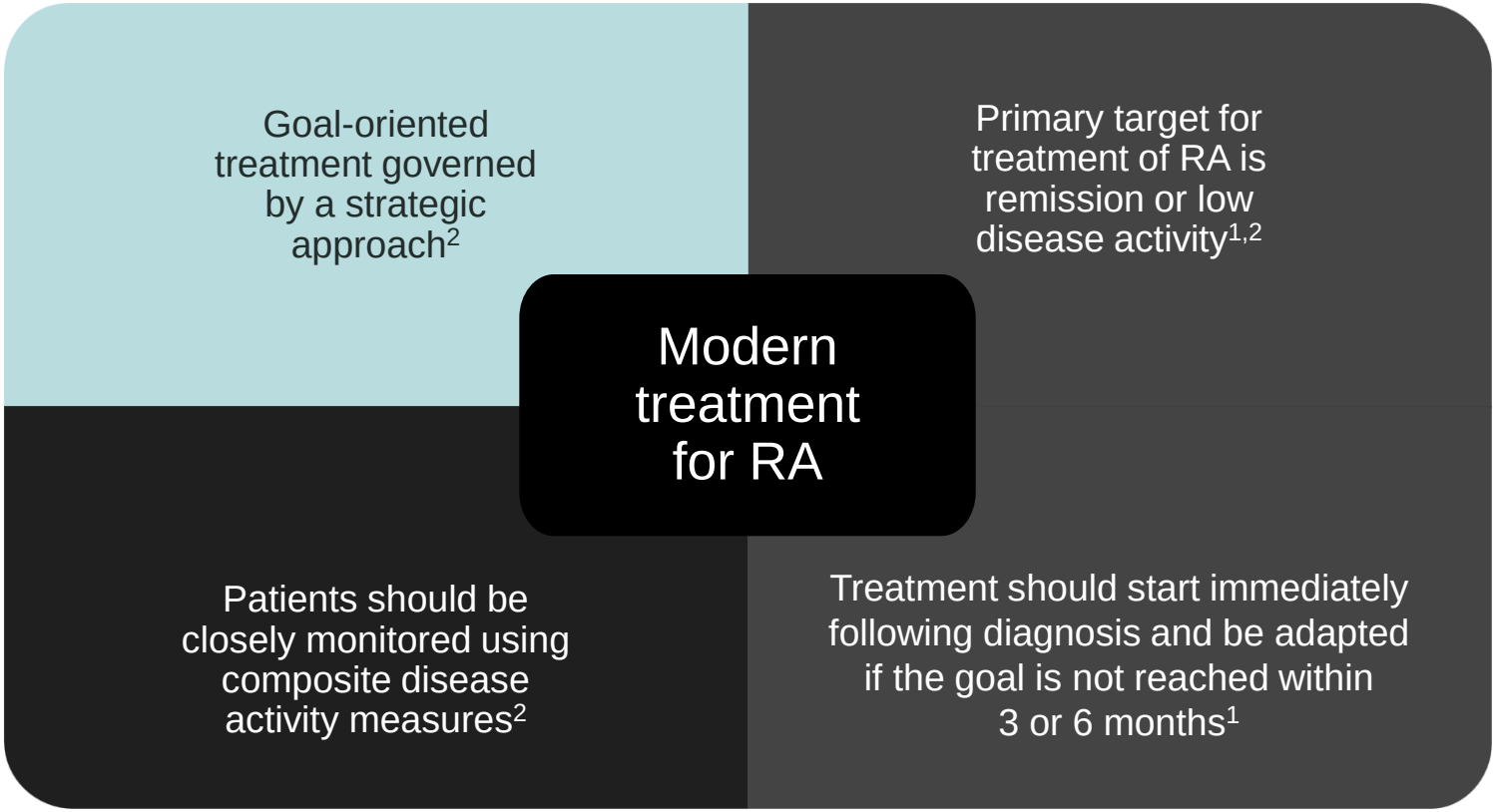


Biologics

TREAT TO TARGET

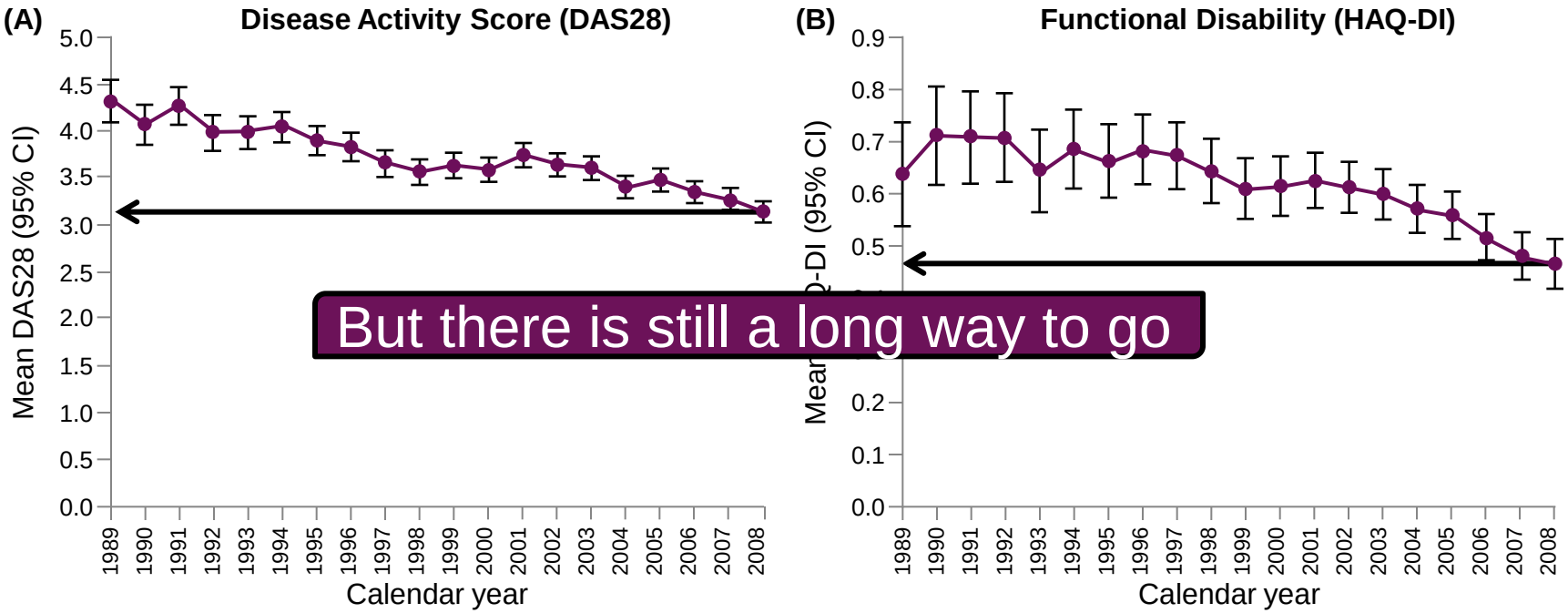
T2T

Defined treatment goals in RA



1. Smolen JS, et al. *Ann Rheum Dis* 2014;73:492–509.
2. Smolen JS, et al. *Ann Rheum Dis* 2010;69:631–37.

Over time clinical outcomes have improved for patients with RA



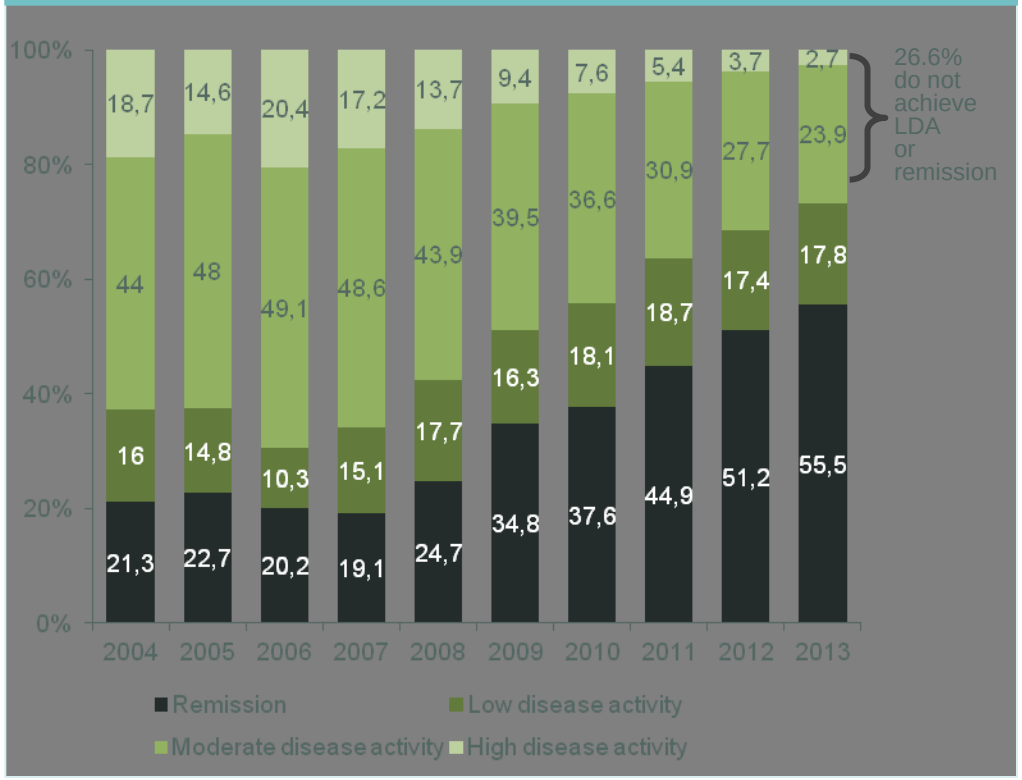
Year	1989	1990	1991	1992	1993	1994	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006	2007	2008
N observed	164	172	193	236	271	332	368	398	443	478	517	538	551	571	595	634	654	686	724	780

HAQ-DI, Health Assessment Questionnaire-Disability Index.

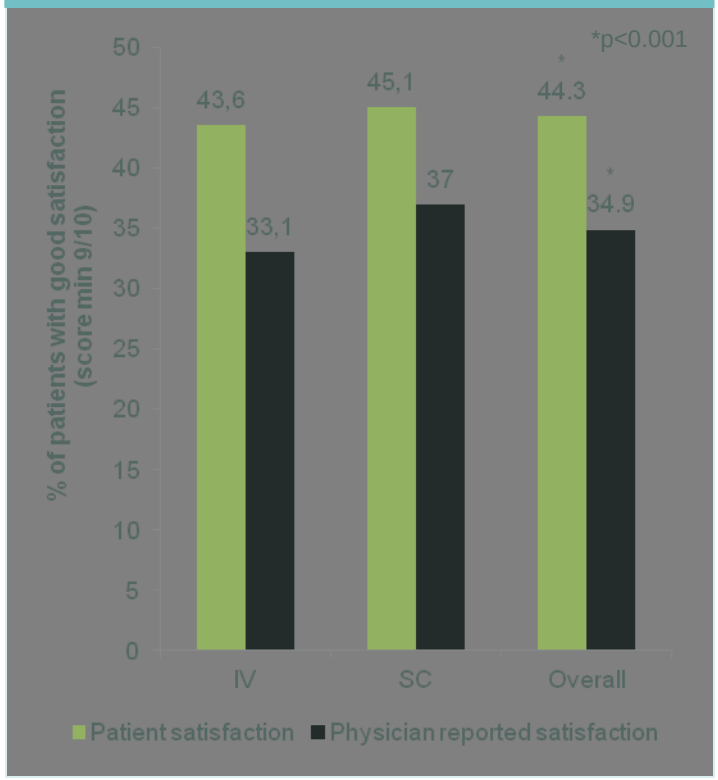
1. Kievit W, et al. *Rheumatology* 2013;52:1500–1508.

Not enough patients are achieving treatment goals with existing biologics

Percentage of patients with RA according to disease activity measured by DAS28 criteria[1]



Satisfaction of control of RA symptoms[2]



1. Haugeberg G, et al. *Arthritis Res Ther.* 2015;17:219.
2. De Mits S, et al. *PLoS One* 2016 Nov 28;11(11).

DAS28, disease activity score in 28 joints.

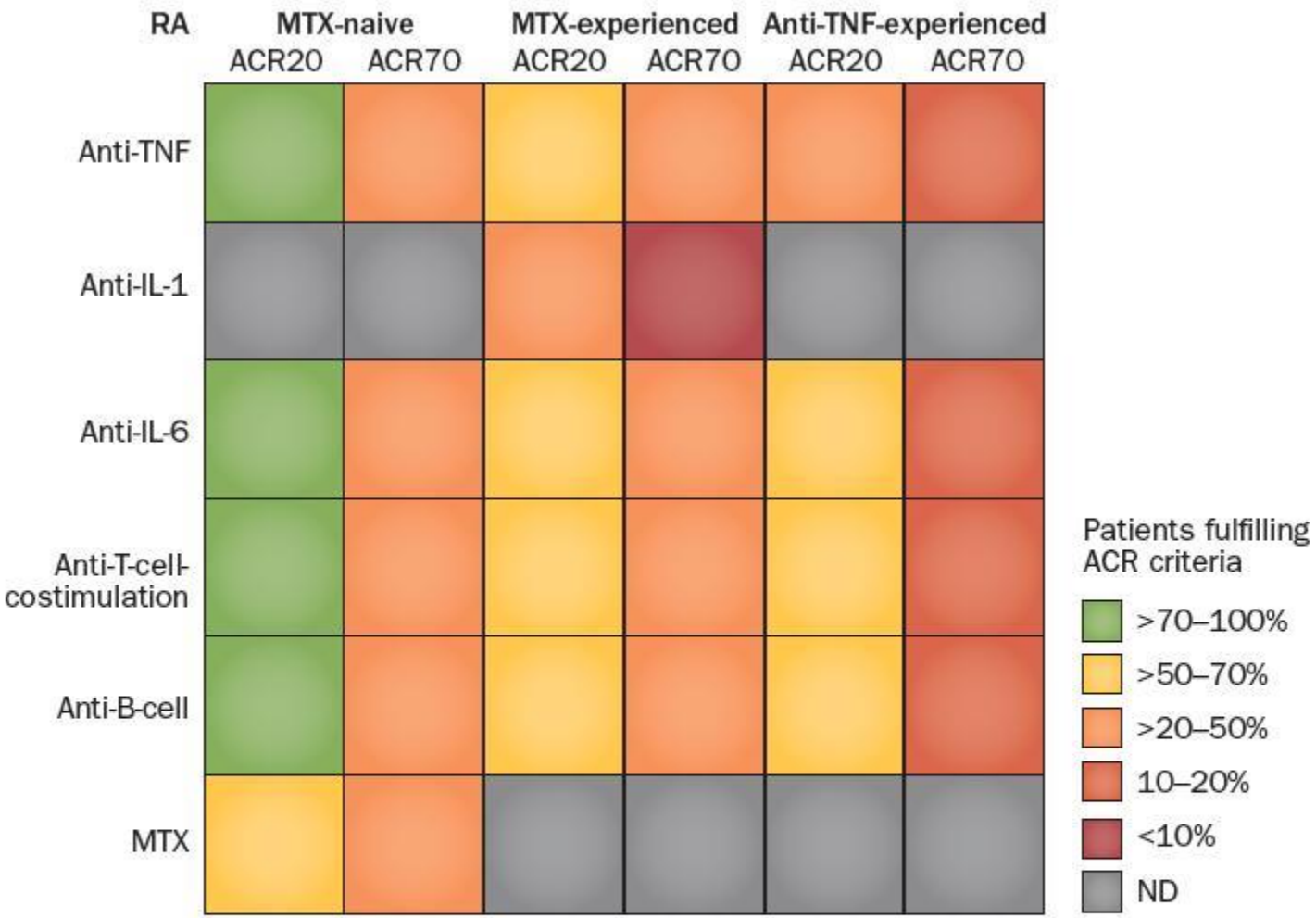
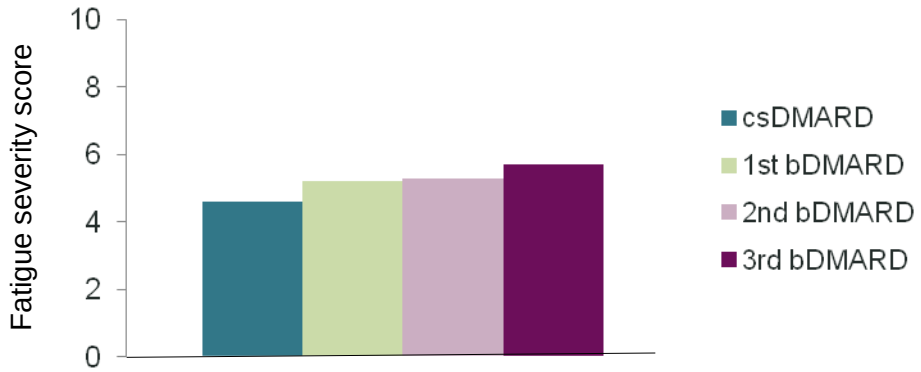


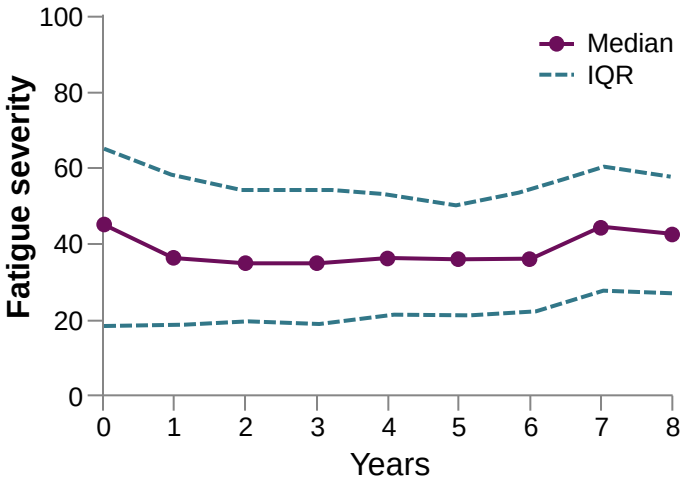
Figure 1 | Generalized ACR20 and ACR70 response rates to different therapies in patients with RA. The data are based on trials of anti-TNF agents,[12,39,46,47,49,51–55,89] anakinra (anti-IL-1),[147] tocilizumab (anti-IL-6),[42,64,93] abatacept (anti-T-cell-costimulation),[23,42,44] rituximab (anti-B-cell),[22,40,48] and MTX (MTX-naive control arms of biologic agent trials). ACR20 and ACR70 indicate improvement, according to ACR criteria, of ≥20% and ≥70%, respectively. Abbreviations: MTX, methotrexate; ND, not done; RA, rheumatoid arthritis.

Fatigue remains a significant problem for patients with RA

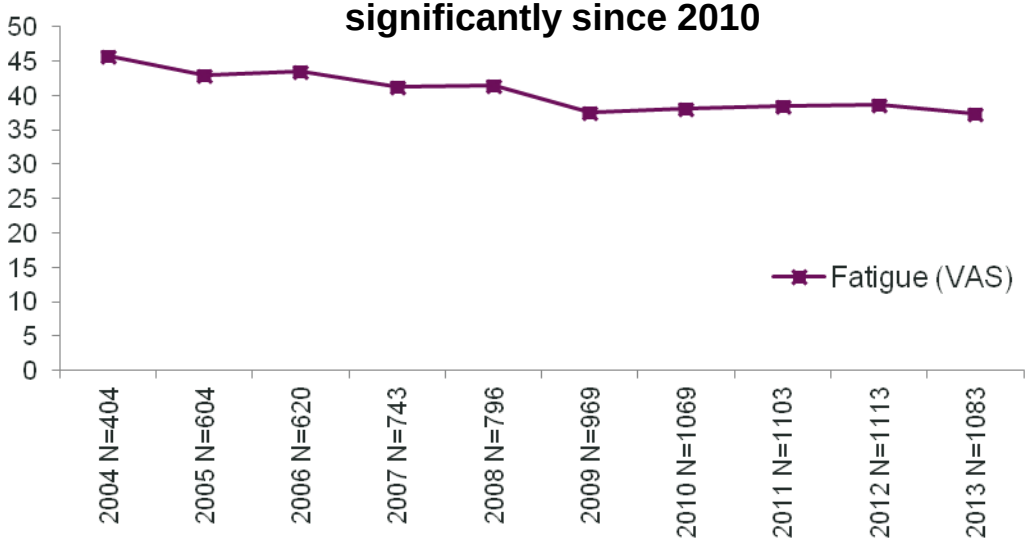
RABBIT registry: Severity of fatigue in patients with RA (N=3875)



Severity of fatigue in 8 years of RA (N=626)

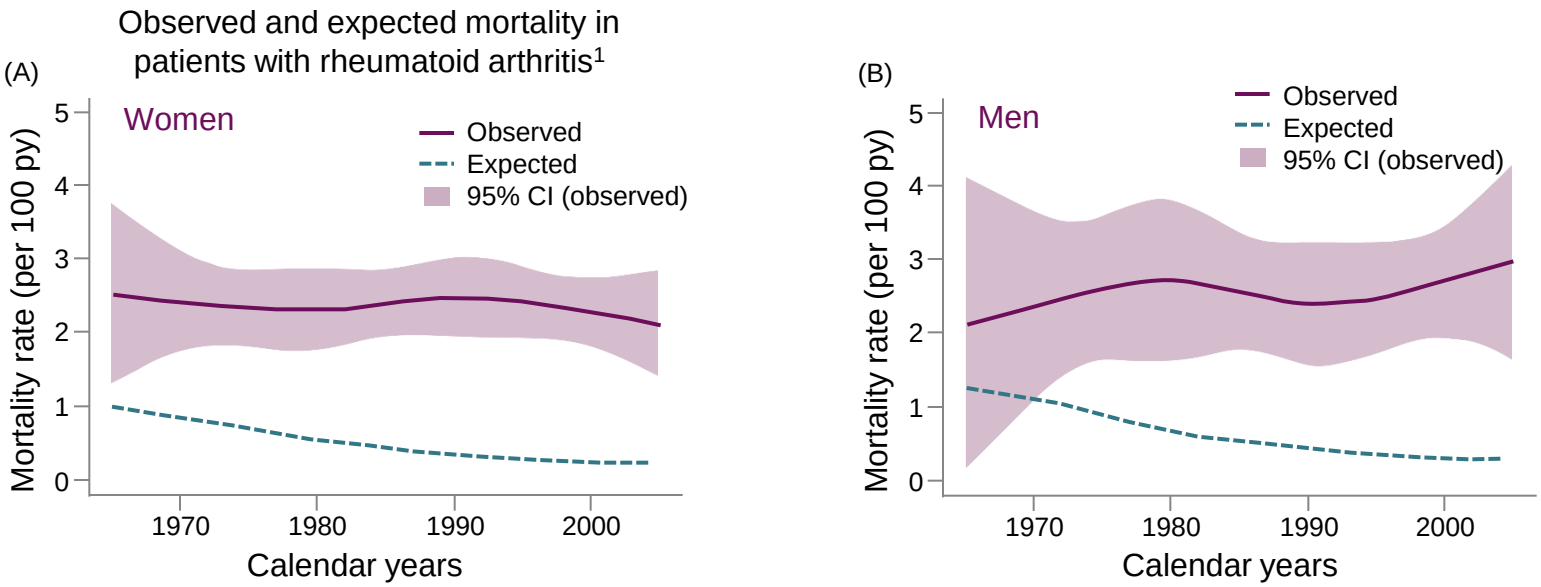


Fatigue scores in patients with RA have not improved significantly since 2010



No significant change in mortality

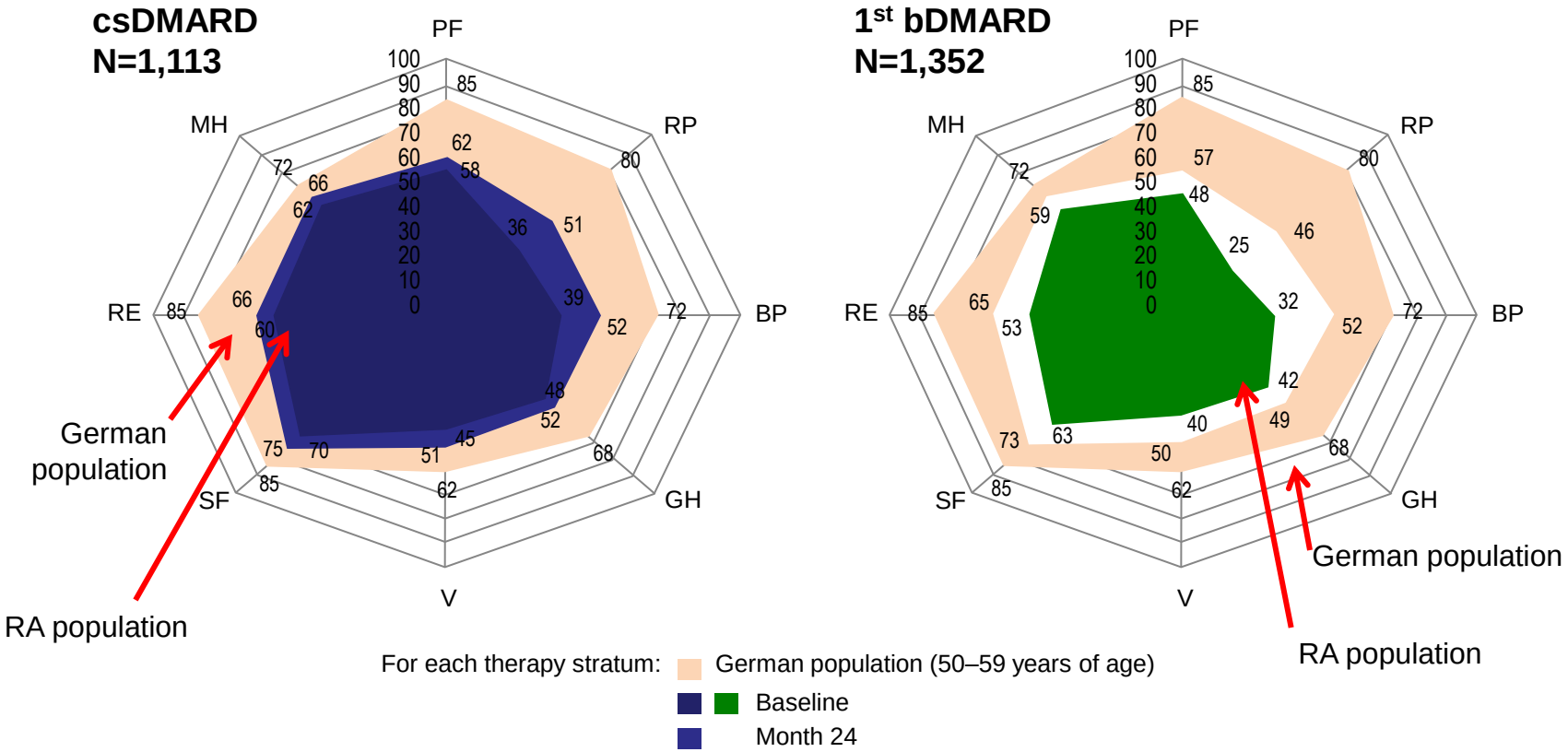
- Standardized mortality ratio 1.28–2.98*



*Differences: diagnosis methods, geographical location, demographic characteristics, study design, follow-up, disease status in the past three decades.

1. Gabriel SE, Michaud K. *Arthritis Research & Therapy* 2009;11:229; 2. Humphreys JH. *Arthritis Care Res (Hoboken)* 2014;66:1296–1301.

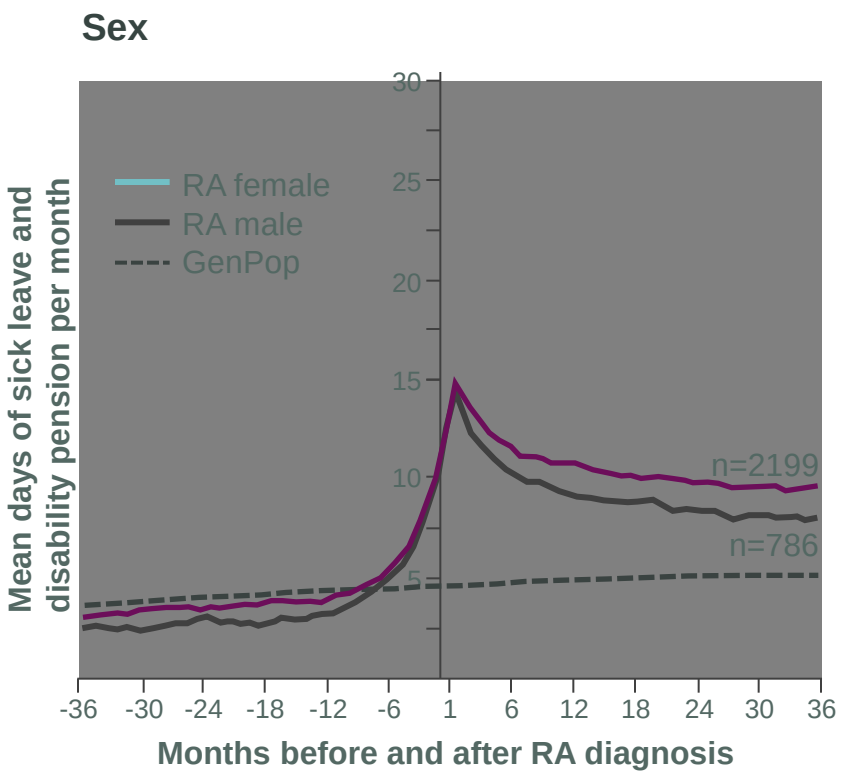
HR-QoL in patients with RA compared with the general population

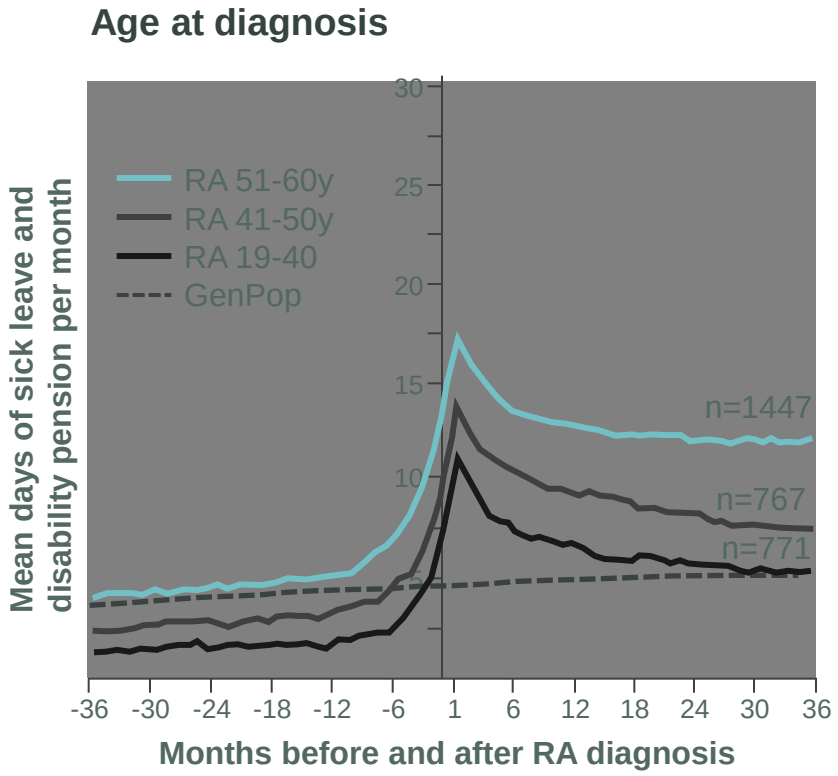


bDMARD, biological disease modifying antirheumatic drug; BP, bodily pain; csDMARD, conventional synthetic disease modifying antirheumatic drug; GH, global health; HR-QoL, health-related quality of life; MH, mental health; PF, physical functioning; RE, role emotional; RP, role physical; SF, social functioning; V, vitality.

1. Gerhold K et al. *Rheumatology (Oxford)* 2015;54(10):1858–1866.

The capacity to work in patients with RA compared with the general population

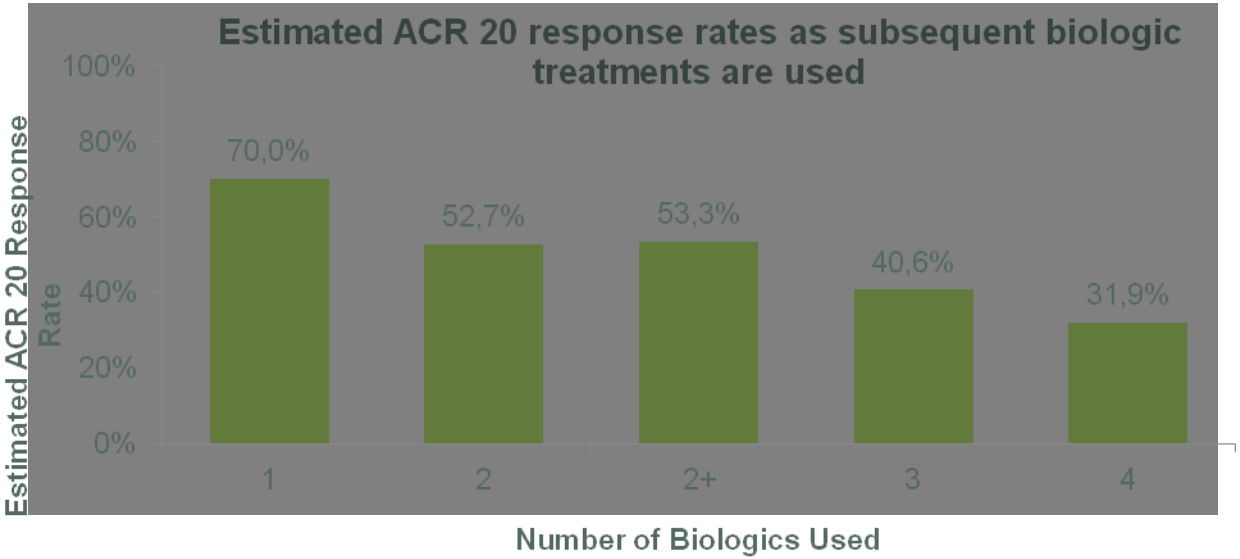




Olofsson T, et al. Ann Rheum Dis 2014;73:845--53.

Patients are less likely to respond to subsequent biologics

As the number of previous TNF-inhibitors increase, **patients are less likely to respond to subsequent biologics**,
as demonstrated by a recent literature review[4]



4. Rendas-Braun R, et al. *Arthritis Res Ther* 2011;13:R25.

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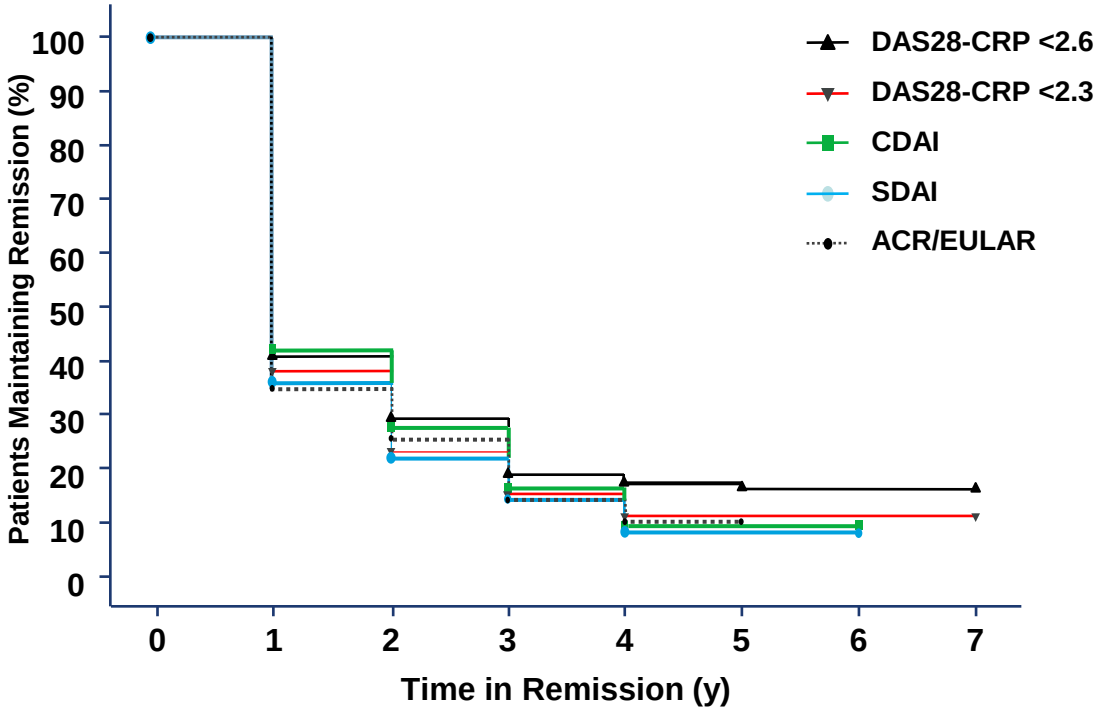
Sustained Remission Is Uncommon in Clinical Practice in Patients With RA

Approximately 40% of patients maintain remission after 1 year



Kaplan-Meier survival curves for subjects maintaining remission (assessed according to 5 remission definitions)

Prince FHM et al. *Arthritis Res Ther.* 2012;14(2):R68.

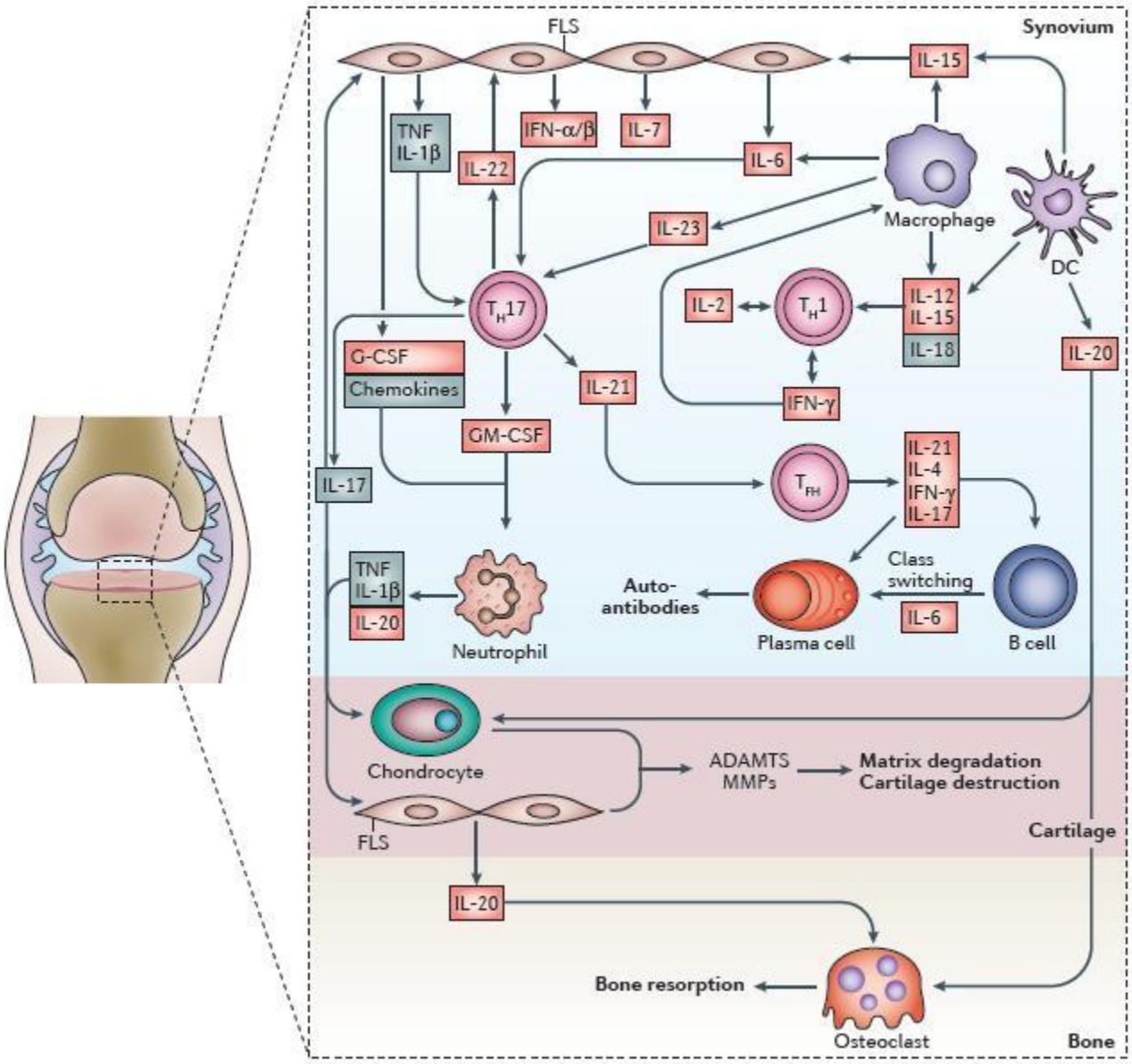
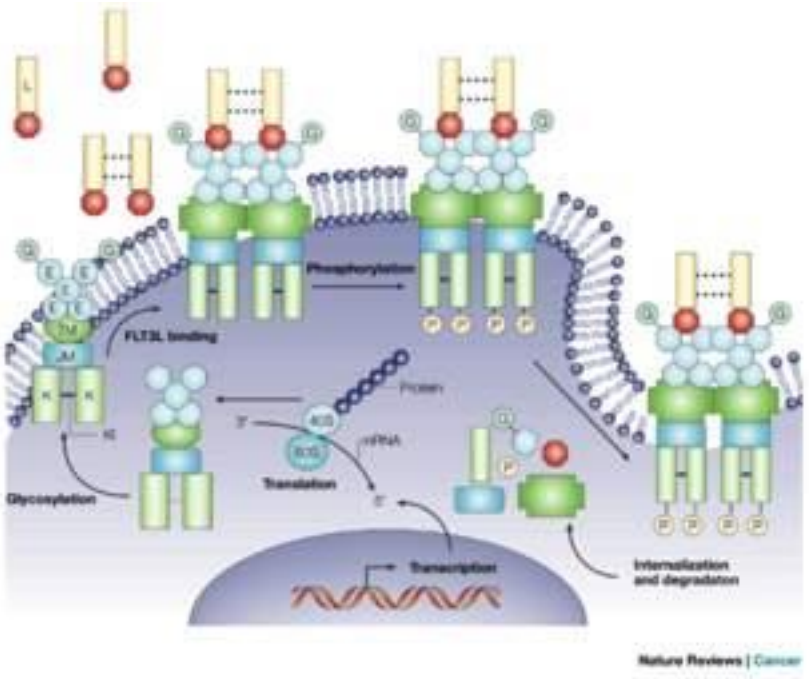
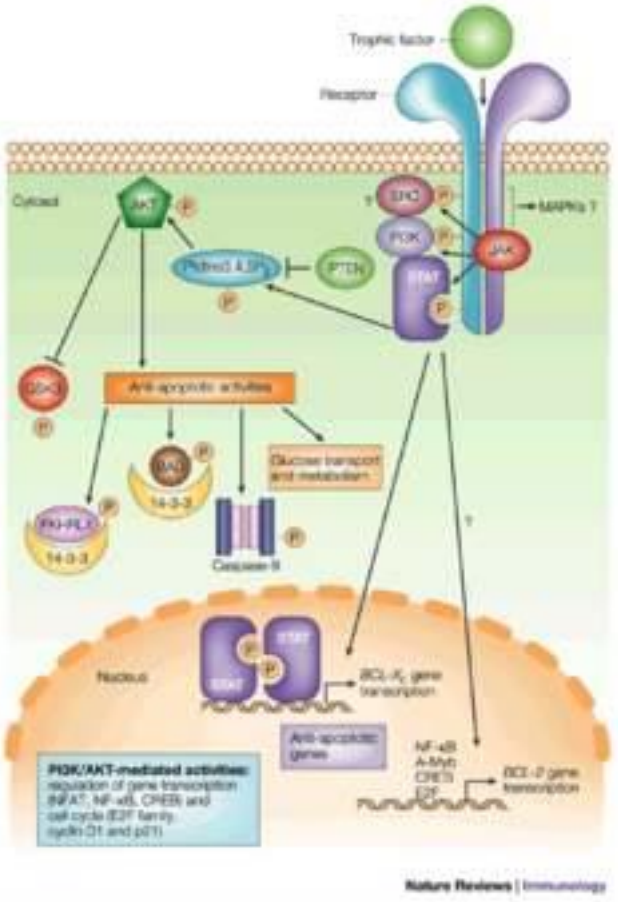


Figure 2 | **Type I and II cytokines (red), which are blocked by Jakinibs, are major drivers of autoimmune diseases such as rheumatoid arthritis.** Within the rheumatoid joint, fibroblast-like synoviocytes (FLSs) are major sources of

Signaling

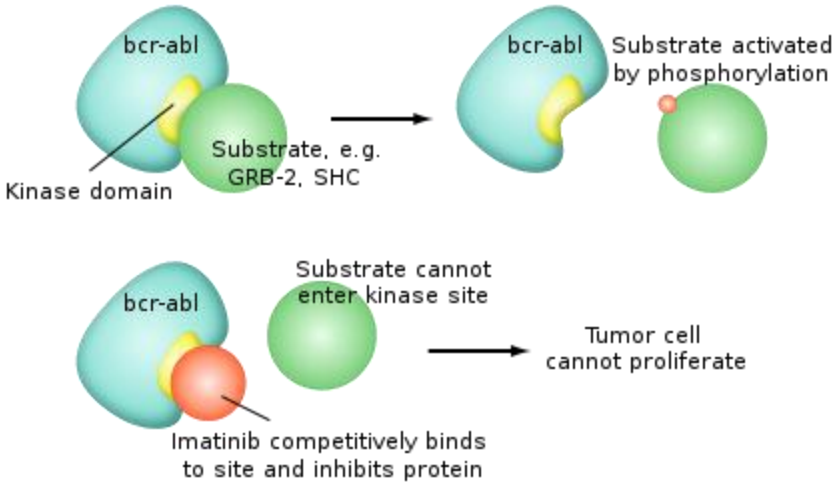

FLT3L binding
Phosphorylation
Protein
Translation
Glycosylation
Transcription
Internalization and degradation
Nature Reviews | Cancer


Trophic factor
Receptor
Cytosol
PTEN
Anti-apoptotic activities
Glucose transport and metabolism
Nucleus
Anti-apoptotic genes
PI3K/AKT-mediated activities:
Nature Reviews | Immunology

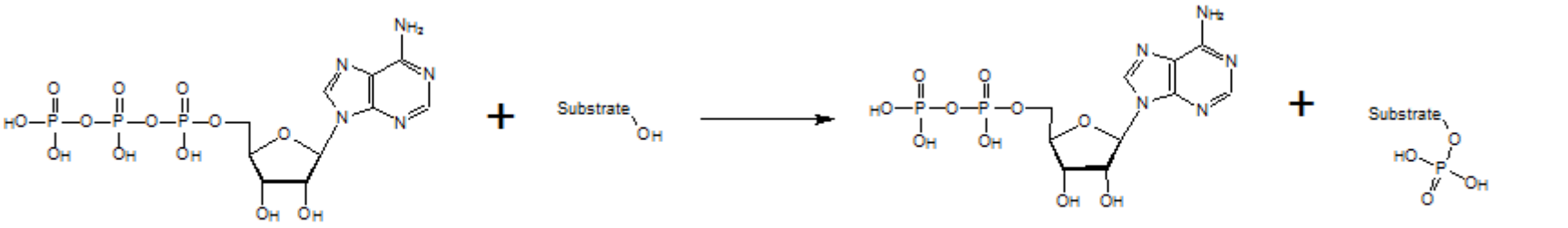
Protein kinases

(ΠΡΩΤΕΪΝΙΚΕΣ ΚΙΝΑΣΕΣ)

Imatinib


bcr-abl
Kinase domain
Substrate, e.g.
GRB-2, SHC
bcr-abl
Substrate activated
by phosphorylation
bcr-abl
Substrate cannot
enter kinase site
Tumor cell
cannot proliferate
Imatinib competitively binds
to site and inhibits protein

ΚΙΝΑΣΕΣ



- ΚΙΝΑΣΕΣ (Φωσφοτρανσφεράσες)
- (#) Φωσφορυλάσες
- (⇔) Φωσφατάσες

ΚΙΝΑΣΕΣ

- …σε Πλείστες (κυτταρικές) διεργασίες **(signaling,…)**
- Παντού σε Ζωϊκό & Φυτικό βασίλειο(& βακτήρια) !!!
- 2% γονιδιώματος ευκαρυωτικών

History

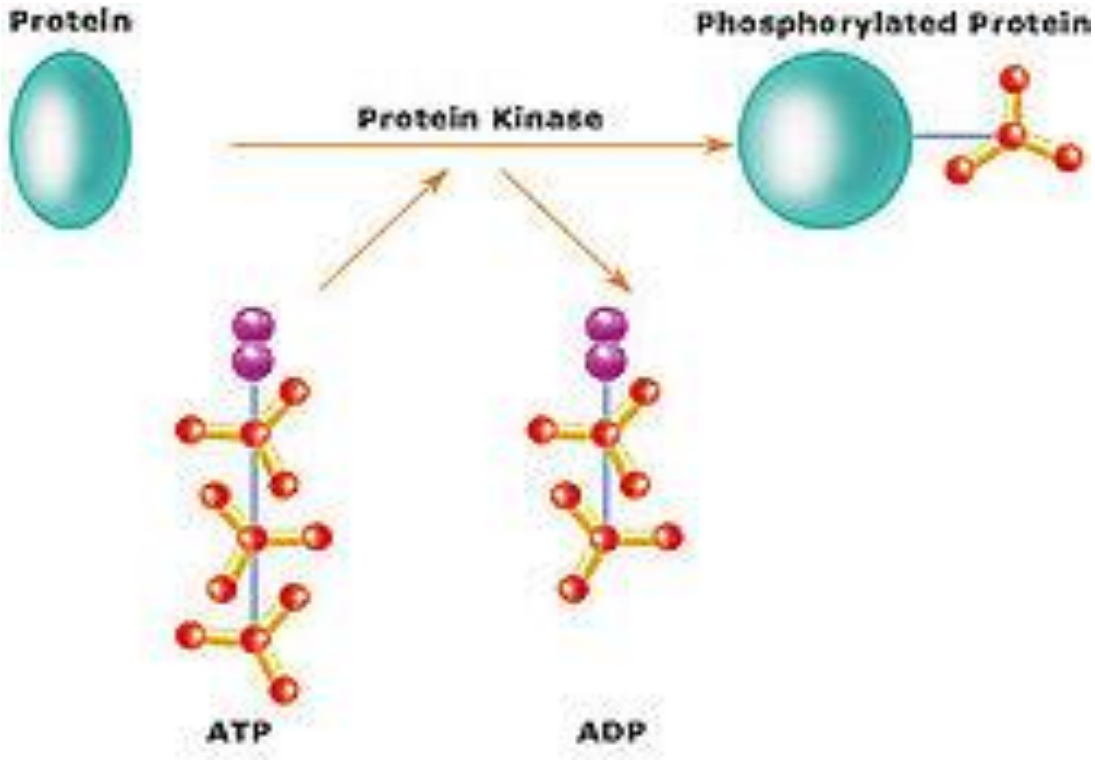
- In 1956, Edmond H. Fischer and Edwin G. Krebs

- The 1990s may be described as the ‘'decade of protein kinase cascades’’

Kinases Classification

- Protein kinases (MAPK,JAKs)
- Lipid kinases (PI3K)
- Carbohydrate kinases
- Other kinases

ΠΡΩΤΕΪΝΙΚΕΣ ΚΙΝΑΣΕΣ



Protein kinases groups

- AGC kinases - containing PKA, PKC and PKG.
- CaM kinases - containing the calcium/calmodulin-dependent protein kinases
.
- CK1 - containing the casein kinase 1 group
.
- CMGC - containing CDK, MAPK, GSK3 and CLK kinases.
- STE - containing the homologs of yeast Sterile 7, Sterile 11, and Sterile 20 kinases
.
- TK - containing the tyrosine kinases.
- TKL - containing the tyrosine-kinase like group of kinases.

Protein kinases (ταξινόμηση)

- Serine/threonine- directed protein kinases
- **Tyrosine-** directed protein kinases (PTKs)

Tyrosine- directed protein kinases (PTKs)

- **Janus family** (JAK1, JAK2, JAK3, Tyk2)
- Syk family
- Src family

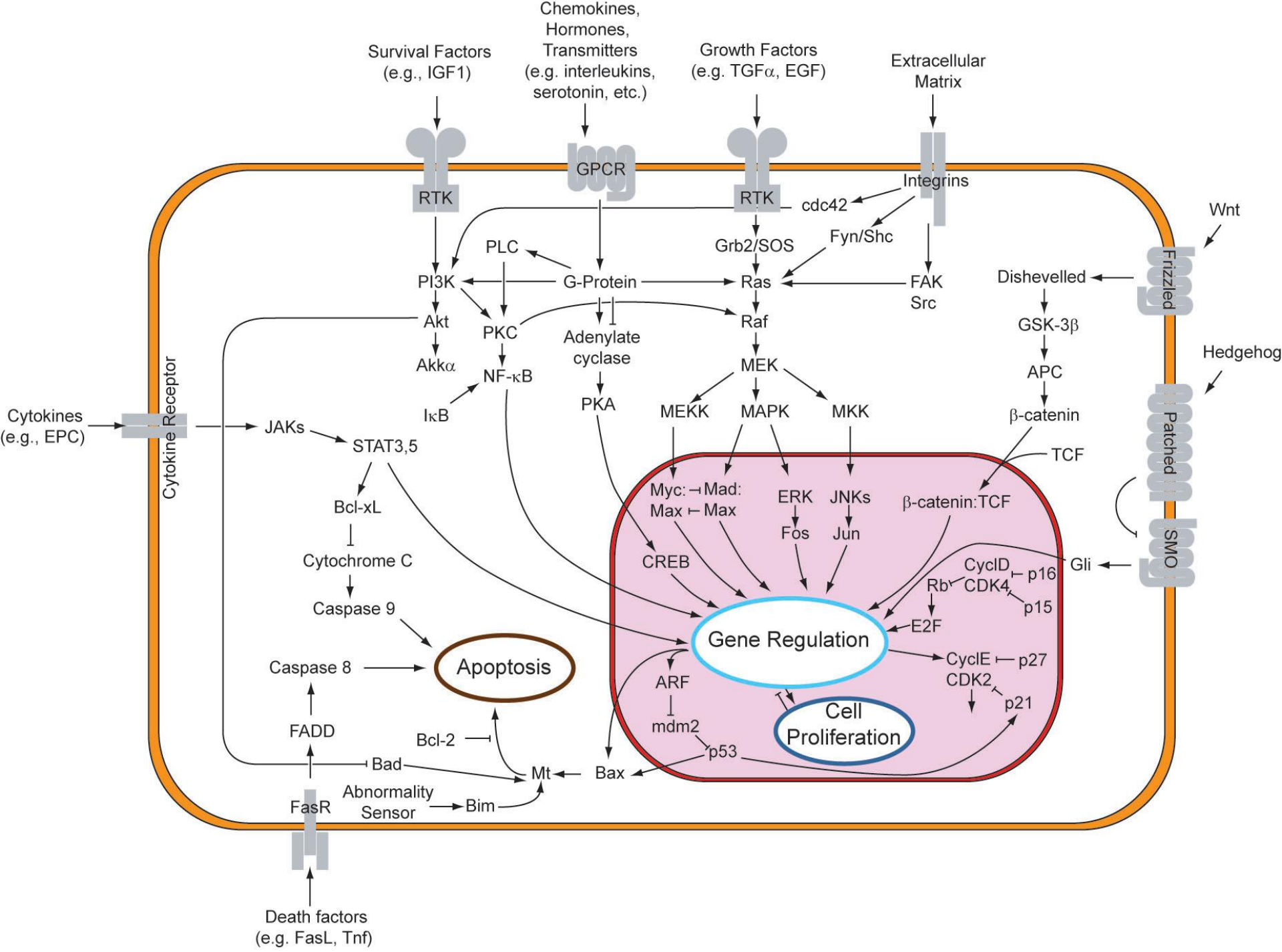
Janus family kinases (JAKs)

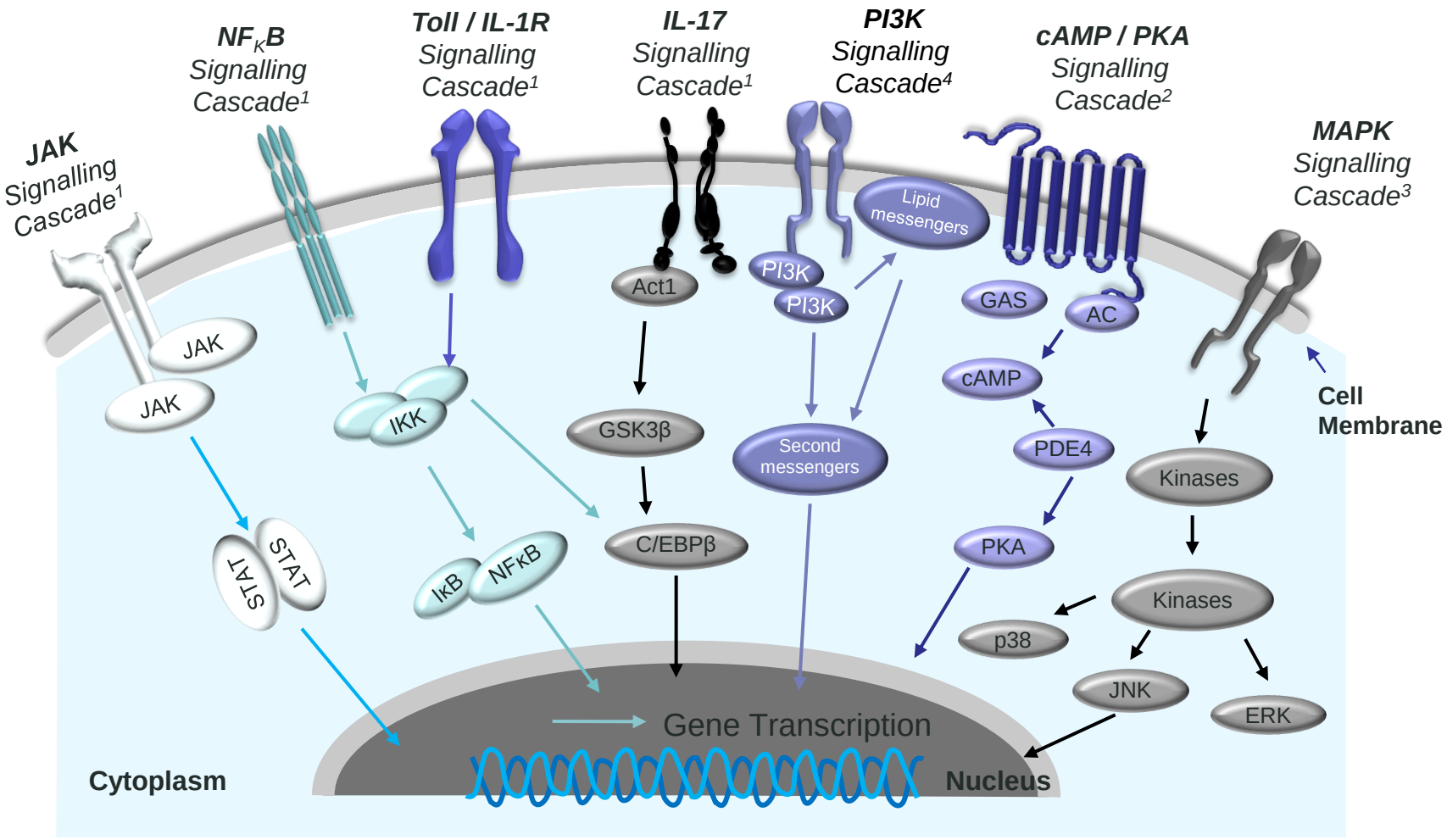
Janus

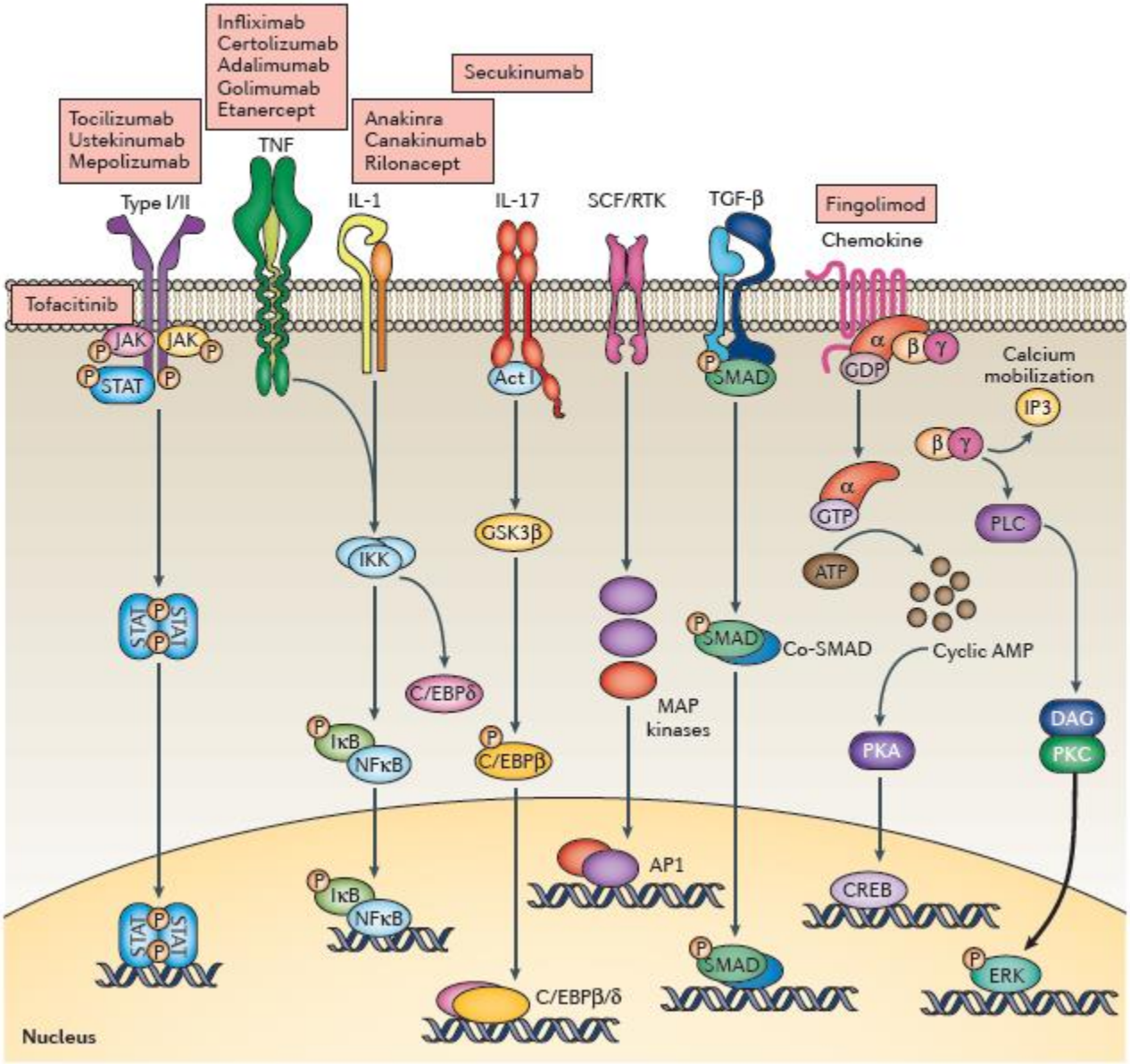
"The two-faced Roman God of gates, doors and passages"

- A subgroup of Intracellular Proteins, non-receptor protein tyrosine kinases
  - **Four JAK family members: JAK1, JAK2, JAK3 and TYK2**
  - Seven STAT family members: STAT1, 2, 3, 4, 5a, 5b and 6 activate transcription[3]

- Characterised by two adjacent kinase domains (JH1 and JH2)
- Implicated in cell growth, survival, development and cell differentiation
- Essential for immune and hematopoietic cells


Survival Factors
(e.g., IGF1)
Chemokines,
Hormones,
Transmitters
(e.g. interleukins,
serotonin, etc.)
Growth Factors
(e.g. TGFα, EGF)
Extracellular
Matrix
RTK
GPCR
RTK
Integrins
cdc42
Fyn/Shc
Wnt
PLC
Grb2/SOS
PI3K
G-Protein
Ras
FAK
Src
Dishevelled
Frizzled
Akt
PKC
Adenylate
cyclase
Raf
GSK-3β
Akkα
NF-κB
MEK
APC
Hedgehog
IκB
PKA
MEKK
MAPK
MKK
β-catenin
Cytokines
(e.g., EPC)
Cytokine Receptor
JAKs
STAT3,5
TCF
Patched
Myc: ⊣ Mad:
Max ⊢ Max
ERK
JNKs
β-catenin:TCF
Bcl-xL
Fos
Jun
SMO
CREB
CyclD
p16
Gli
Rb
CDK4
p15
Cytochrome C
Caspase 9
E2F
Gene Regulation
Caspase 8
Apoptosis
CyclE
p27
ARF
CDK2
p21
Cell
Proliferation
FADD
mdm2
Bcl-2
p53
Bad
Mt
Bax
FasR
Abnormality
Sensor
Bim
Death factors
(e.g. FasL, Tnf)


JAK
Signalling
Cascade1
NFKB
Signalling
Cascade1
Toll / IL-1R
Signalling
Cascade1
IL-17
Signalling
Cascade1
PI3K
Signalling
Cascade4
cAMP / PKA
Signalling
Cascade2
MAPK
Signalling
Cascade3
Lipid
messengers
JAK
JAK
IKK
Act1
PI3K
PI3K
GAS
AC
cAMP
PDE4
PKA
Cell
Membrane
GSK3β
Second
messengers
Kinases
STAT
STAT
IκB
NFκB
C/EBPβ
Kinases
p38
JNK
ERK
Gene Transcription
Cytoplasm
Nucleus


Infliximab
Certolizumab
Adalimumab
Golimumab
Etanercept
Secukinumab
Tocilizumab
Ustekinumab
Mepolizumab
Anakinra
Canakinumab
Rilonacept
Fingolimod
TNF
Type I/II
IL-1
IL-17
SCF/RTK
TGF-β
Chemokine
Tofacitinib
JAK
STAT
Act I
SMAD
GDP
Calcium mobilization
IP3
GTP
ATP
PLC
IKK
GSK3β
Co-SMAD
Cyclic AMP
C/EBPδ
MAP kinases
IκB
NFκB
C/EBPβ
DAG
PKC
PKA
AP1
CREB
ERK
C/EBPβ/δ
Nucleus

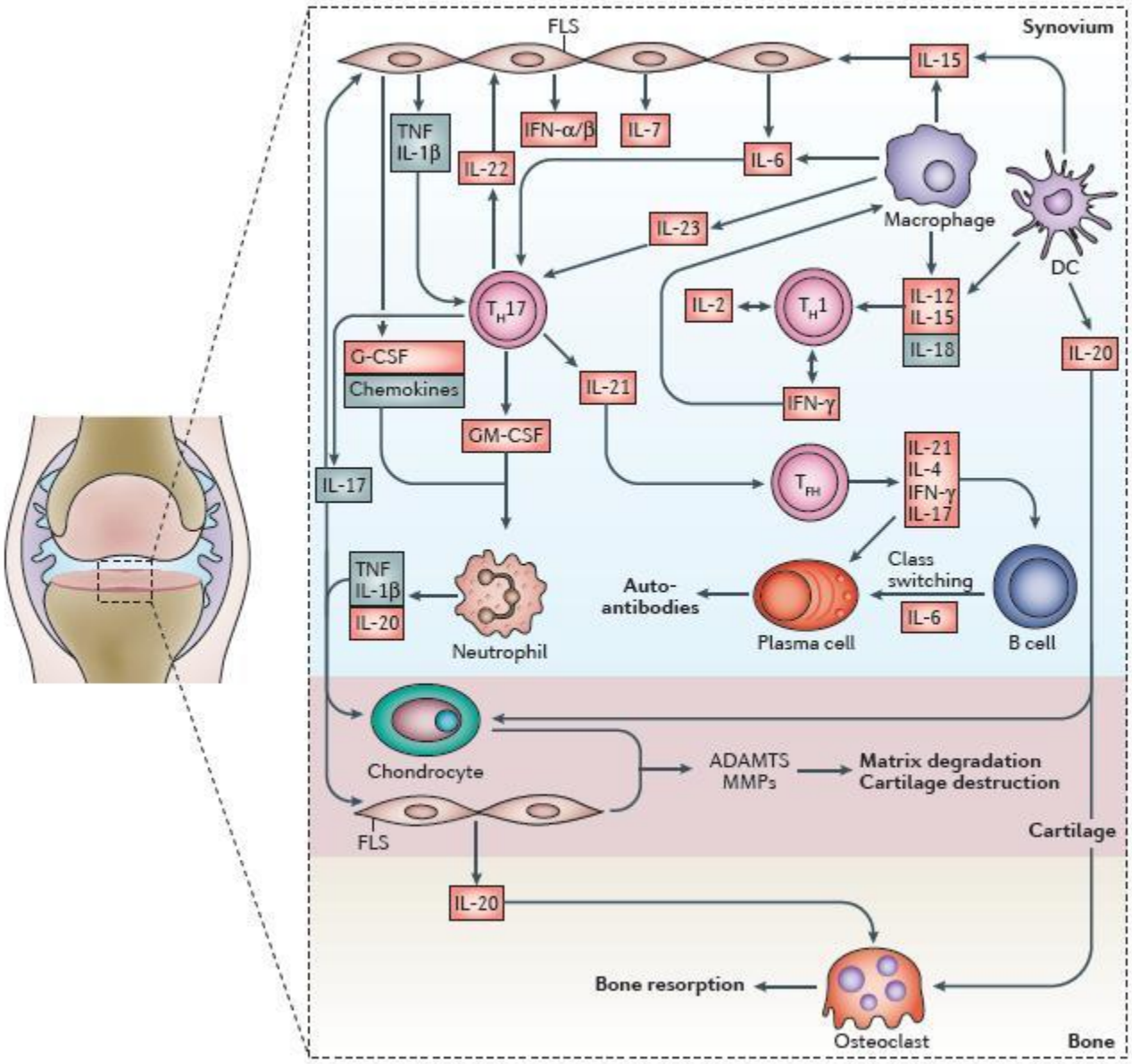


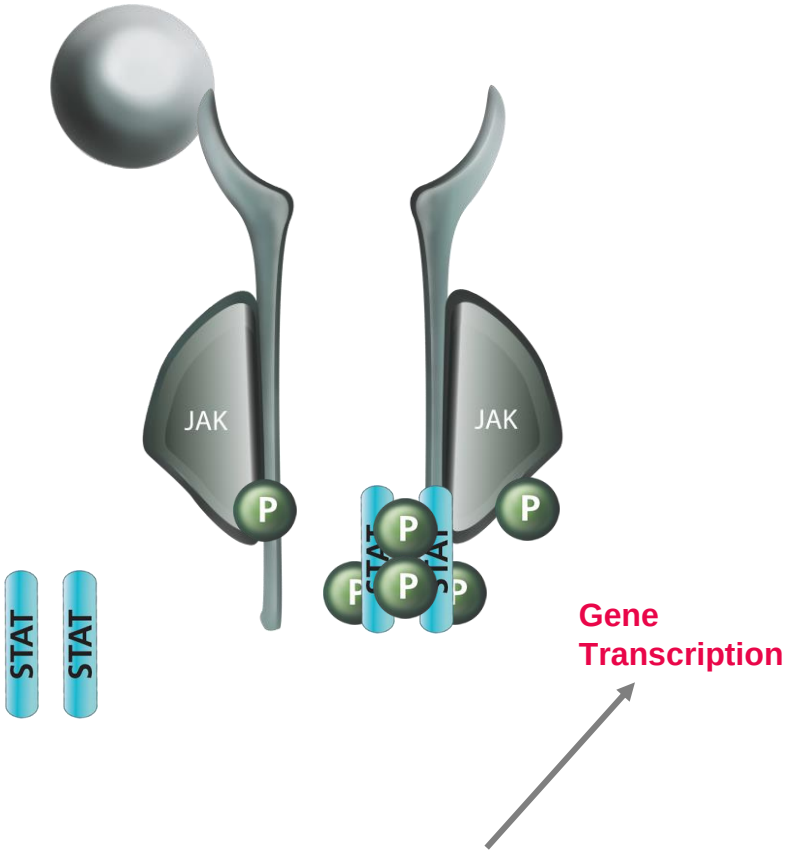
Figure 2 | **Type I and II cytokines (red), which are blocked by Jakinibs, are major drivers of autoimmune diseases such as rheumatoid arthritis.** Within the rheumatoid joint, fibroblast-like synoviocytes (FLSs) are major sources of

Binding of cytokine receptors activates JAK signalling pathways

1. Cytokine binding to its cell surface receptor leads to receptor polymerisation and autophosphorylation of associated JAKs

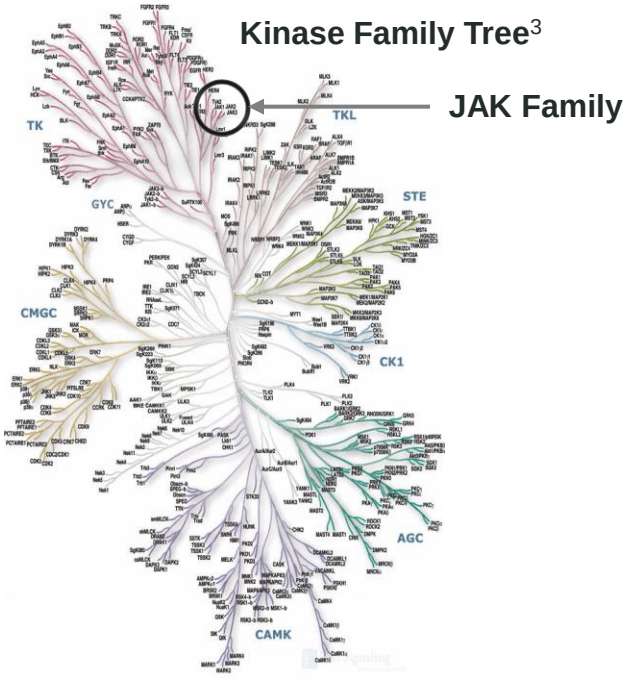
2. Activated JAKs phosphorylate the receptors that dock STATs

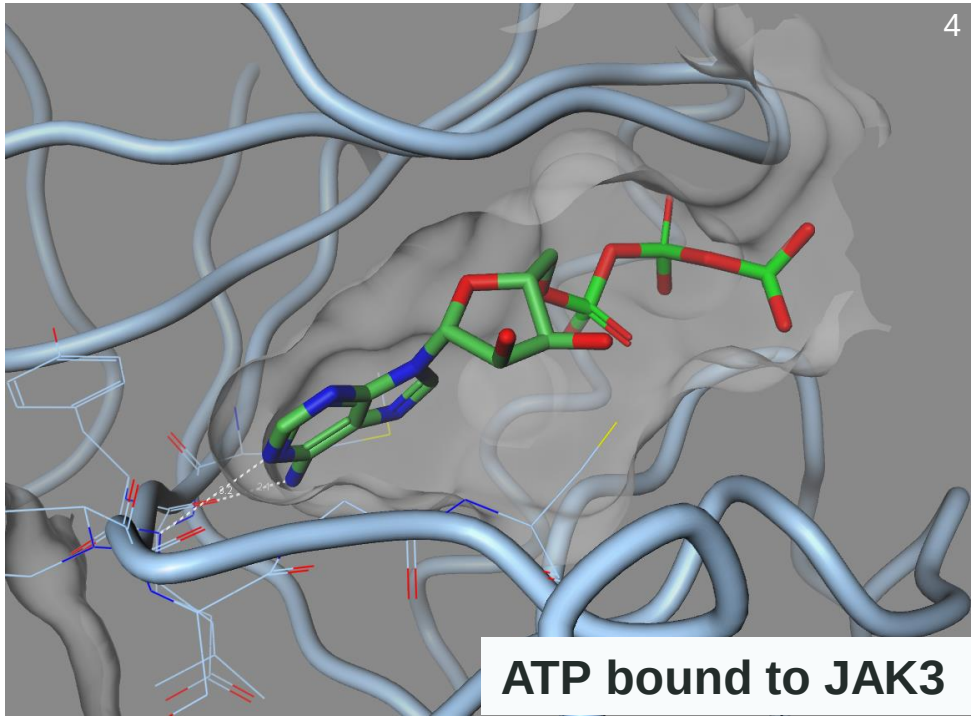
3. Activated JAKs phosphorylate STATs, which dimerise and move to the nucleus to activate new gene transcription



- More than 500 kinases in the human kinome[1]
- All have highly conserved active sites and use ATP as a cofactor[2]
- Competitive inhibition of ATP most common approach[1]

Kinase Family Tree[3]





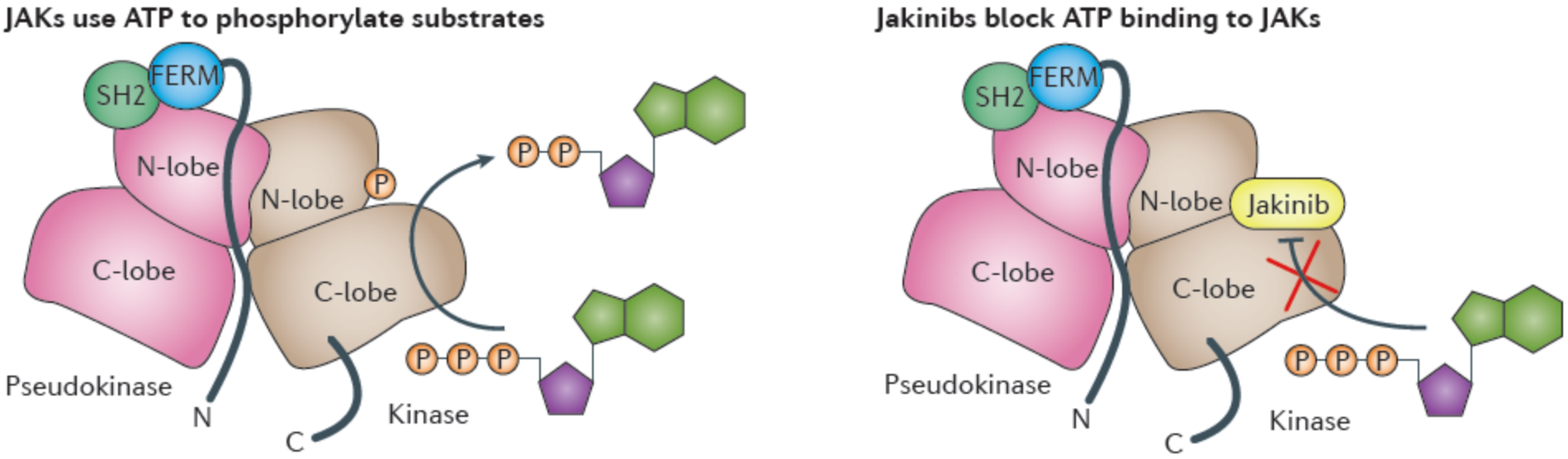
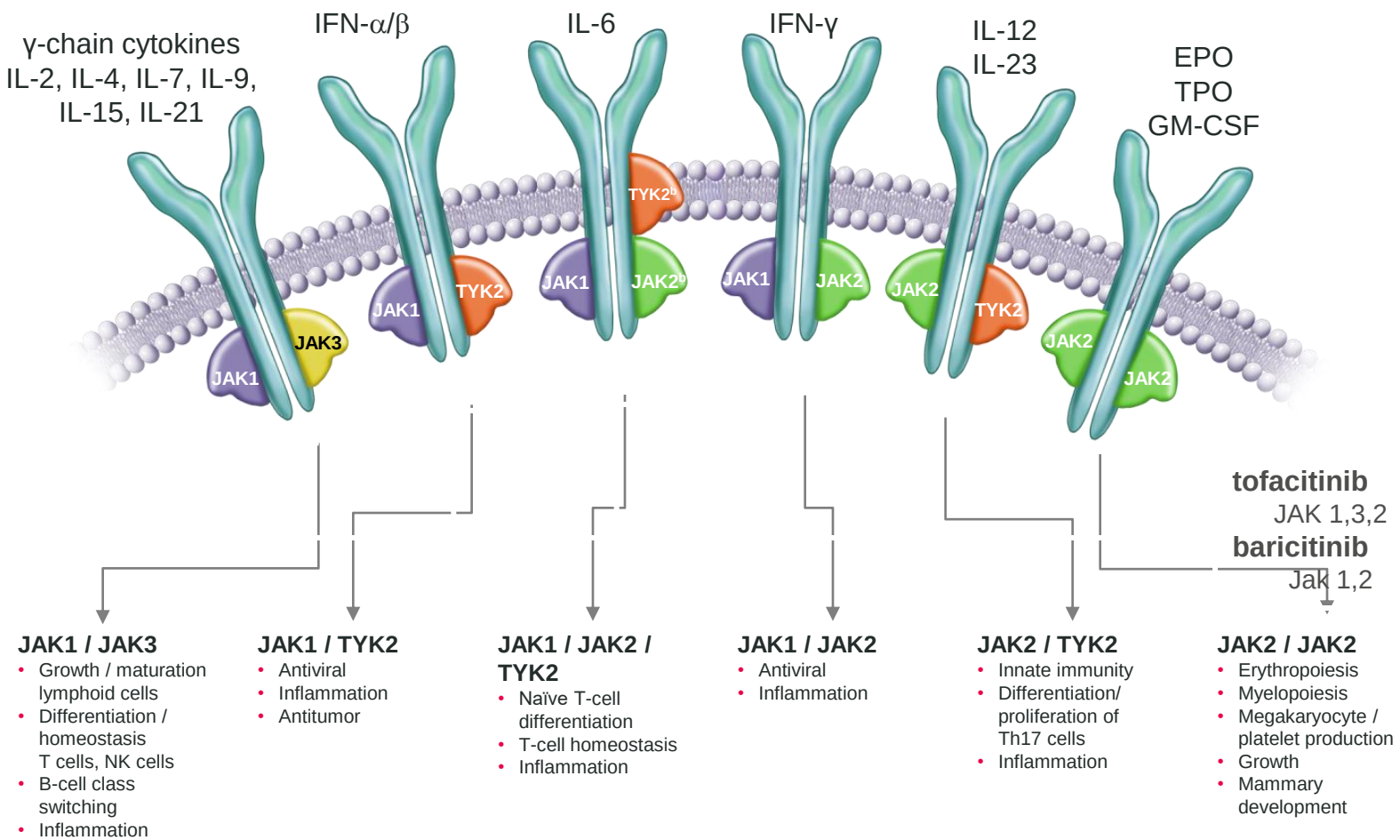


Figure 3 | **JAKs are composed of several key domains including a tyrosine kinase domain, pseudokinase domain, FERM (band four-point-one, ezrin, radixin, moesin) domain, and SH2 (Src homology 2) domain.** The tyrosine kinase domain binds ATP hydrolysing it to ADP and catalyses both autophosphorylation and other substrates including signal transducers and activators of transcription (STATs). JAK, Janus kinase (JAK) inhibitors (Jakinibs) bind to the pocket ordinarily occupied by ATP, thereby preventing JAKs from using ATP and phosphorylating their substrates.

joint-space narrowing, particularly at the higher dose of 4 mg twice daily, and to a lesser degree with 2 mg twice daily[74–77].

latter being a key cytokine for NK-cell homeostasis[85]. However, NK cell counts are only moderately reduced in tofacitinib-treated patients and do not correlate with infection risk[86,87]. Tofacitinib-treated patients also have

Current JAK inhibitors target different receptors based on their selectivity



	IC_{50}, nM[5]			
Inhibitor	JAK1	JAK2	JAK3	TYK2
Tofacitinib	15	77	55	489
Baricitinib	4	7	787	61

SCIENCE * VOL. 270 * 3 NOVEMBER 1995

Mutation of Jak3 in a Patient with SCID: Essential Role of Jak3 in Lymphoid Development

Sarah M. Russell, Nahid Tayebi,* Hiroshi Nakajima,*
Mary C. Riedy,† Joseph L. Roberts,† M. Javad Aman,†
Thi-Sau Migone, Masayuki Noguchi, M. Louise Markert,
Rebecca H. Buckley, John J. O'Shea, Warren J. Leonard‡

Males with X-linked severe combined immunodeficiency (XSCID) have defects in the common cytokine receptor γ chain (γ_c) gene that encodes a shared, essential component of the receptors for interleukin-2 (IL-2), IL-4, IL-7, IL-9, and IL-15. The Janus family tyrosine kinase Jak3 is the only signaling molecule known to be associated with γ_c, so it was hypothesized that defects in Jak3 might cause an XSCID-like phenotype. A girl with immunological features indistinguishable from those of XSCID was therefore selected for analysis. An Epstein-Barr virus (EBV)–transformed cell line derived from her lymphocytes had normal γ_c expression but lacked Jak3 protein and had greatly diminished Jak3 messenger RNA. Sequencing revealed a different mutation on each allele: a single nucleotide insertion resulting in a frame shift and premature termination in the Jak3 JH4 domain and a nonsense mutation in the Jak3 JH2 domain. The lack of Jak3 expression correlated with impaired B cell signaling, as demonstrated by the inability of IL-4 to activate Stat6 in the EBV-transformed cell line from the patient. These observations indicate that the functions of γ_c are dependent on Jak3 and that Jak3 is essential for lymphoid development and signaling.

XSCID

(X-linked severe combined immunodeficieny)

- Χαμηλά ποσοστά immunoglobulin G (IgG)
- Διαταραχή λειτουργίας Β λεμφοκυττάρων
- Σχεδόν πλήρης έλλειψη Τ λεμφοκυττάρων

Cell type	Normal lymphocyte count average (range)	X-SCID count average (range)
T-cells	3,680 (2,500-5,500)	200 (0-800)
B-cells	730 (300-2,000) [15]	1,300 (44 - >3,000)
NK cells	420 (170-1,100)	<100
Total	0–3 months: 5,400 (3,400-7,300)	<2,000

XSCID

- Σοβαρές λοιμώξεις (νωρίς)
- Χαμηλό ΒΣ
- Έκζεμα
- Διάρροια
- Μυϊκή υποτονία

XSCID

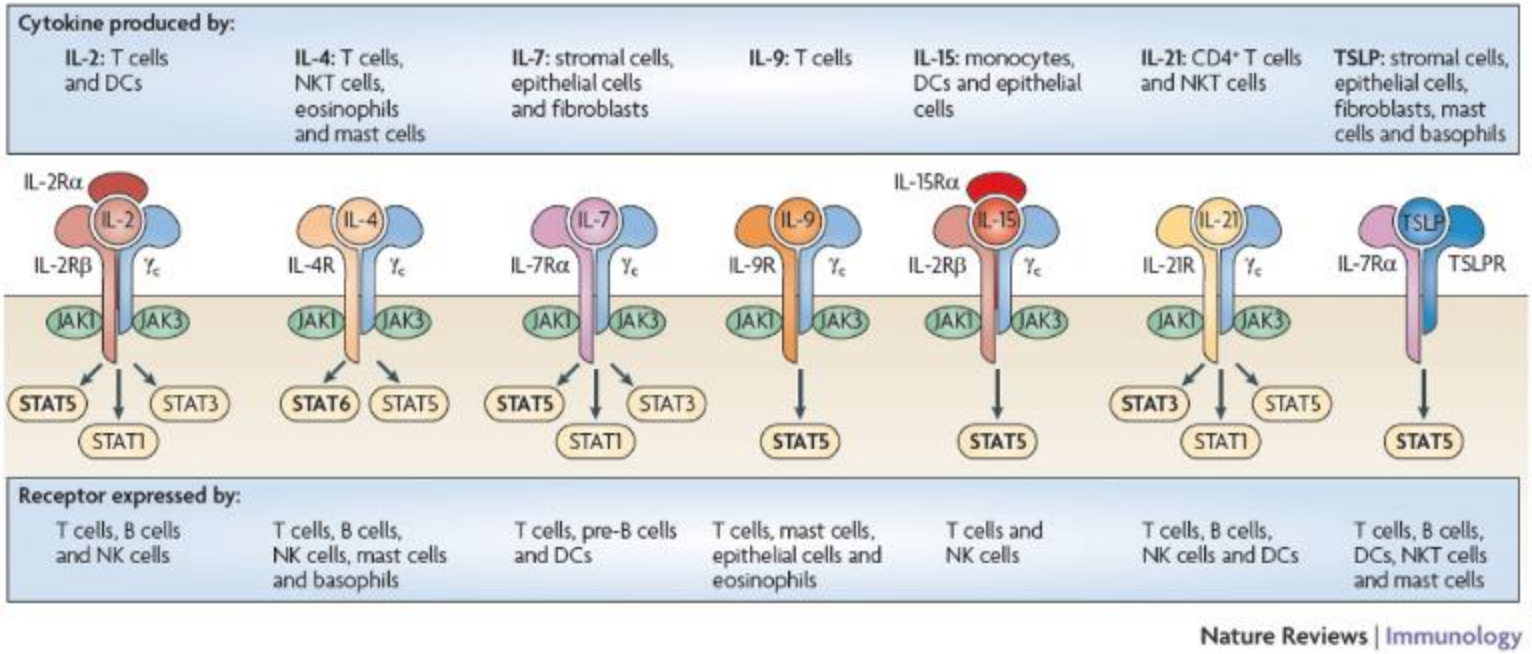
λόγω ελαττωματικών γ-chain
(gamma common chain)

- ***IL2RG*** => **IL-2RG** (γ-chain)

γ-chain (γc)

- IL-2
- IL- 4
- IL-7
- IL-9
- IL-15
- IL- 21

The common gamma chain and X-SCID

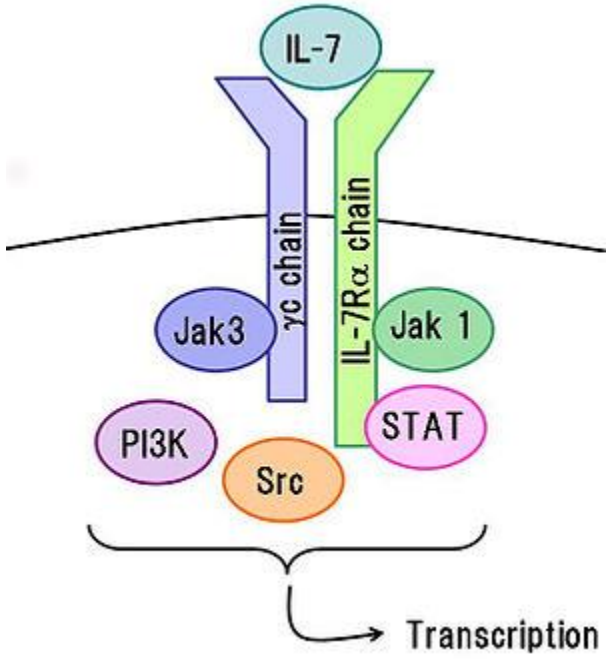


Rochman Y, Spolski R, & Leonard WJ (2009)

- Το γονίδιο που κωδικοποιεί για την γ-chain βρίσκεται στο χρωμόσωμα Χ

- Γ γ-chain (Χ/Χ) (+/+) , (-/+)

- Α γ-chain (Χ/Υ) (+/Υ) , (-/Υ)


IL-7
γc chain
IL-7Rα chain
Jak3
Jak 1
PI3K
Src
STAT
Transcription

Αποκλειστικά!!!

η **Jak3** συνδέεται με την γ-chain

S. M. Russell et al., ibid. 266, 1042 (1994

Το γονίδιο για την Jak3 δεν βρίσκεται στο χρωμόσωμα Χ

16. J. N. Ihle et al., Annu. Rev. Immunol. 13, 369 (1995)

SCIENCE * VOL. 270 * 3 NOVEMBER 1995

Mutation of Jak3 in a Patient with SCID: Essential Role of Jak3 in Lymphoid Development

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Males with X-linked severe combined immunodeficiency (XSCID) have defects in the common cytokine receptor γ chain (γ_c) gene that encodes a shared, essential component of the receptors for interleukin-2 (IL-2), IL-4, IL-7, IL-9, and IL-15. The Janus family tyrosine kinase Jak3 is the only signaling molecule known to be associated with γ_c, so it was hypothesized that defects in Jak3 might cause an XSCID-like phenotype. A girl with immunological features indistinguishable from those of XSCID was therefore selected for analysis. An Epstein-Barr virus (EBV)–transformed cell line derived from her lymphocytes had normal γ_c expression but lacked Jak3 protein and had greatly diminished Jak3 messenger RNA. Sequencing revealed a different mutation on each allele: a single nucleotide insertion resulting in a frame shift and premature termination in the Jak3 JH4 domain and a nonsense mutation in the Jak3 JH2 domain. The lack of Jak3 expression correlated with impaired B cell signaling, as demonstrated by the inability of IL-4 to activate Stat6 in the EBV-transformed cell line from the patient. These observations indicate that the functions of γ_c are dependent on Jak3 and that Jak3 is essential for lymphoid development and signaling.

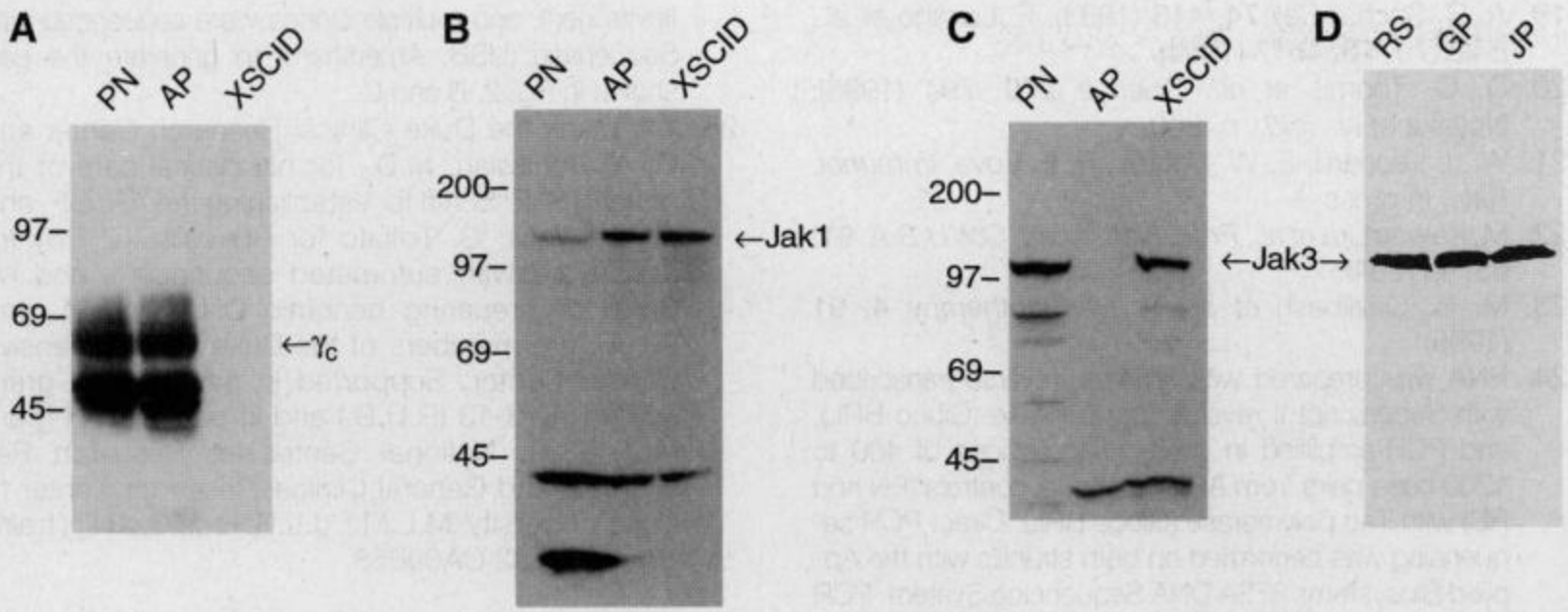
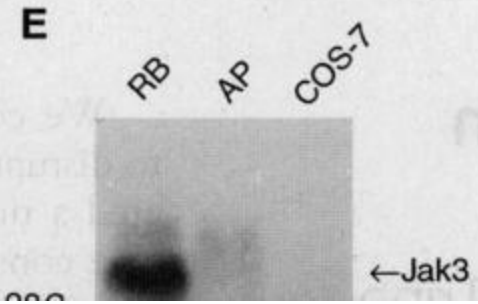


Fig. 1. Defective expression of Jak3 in an EBV-transformed B cell line from AP. Lysates from 10^6 EBV-transformed cells from a normal control (PN), the patient (AP), and a patient with XSCID [Pt. 2 from (9)] were immunoblot-



probe (F). RNA was isolated with RNAzol (Tel-Test, Friendswood, Texas).

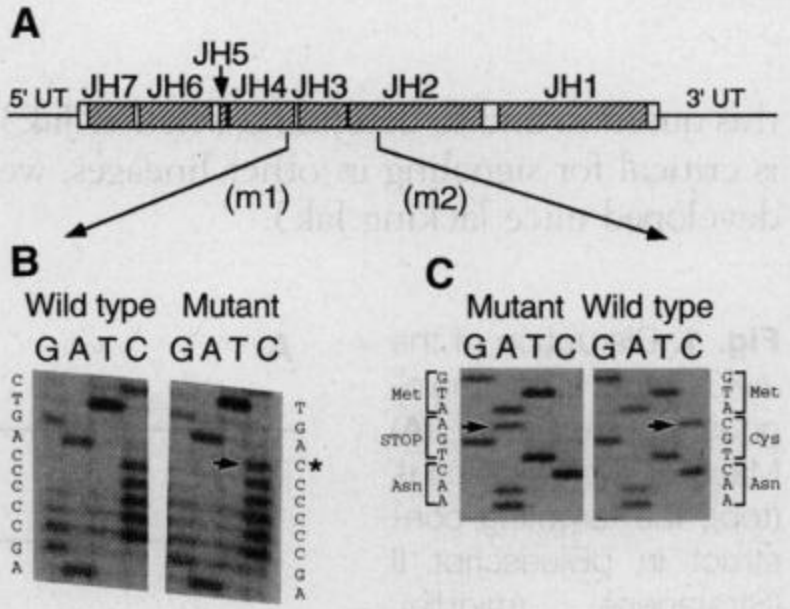


Fig. 2. Analysis of the Jak3 defects in AP. (**A**) Schematic of the coding region for Jak3 (boxed) and 5′ and 3′ untranslated (UT) regions. The seven Jak homology (JH) domains are indicated by hatched boxes, and the positions of the two mutations (m1, insertion at nucleotide 1172; m2, nonsense mutation at nucleotide 1695) identified in AP are shown. (**B**) Sequencing of the m1 insertion (sequenced on the reverse strand). Compare the AP mutation on the right [the band corresponding to the sixth C (asterisk) is identified by an arrow] to the wild type on the left. (**C**) Sequencing of the m2 nonsense mutation that resulted from a C-to-A transversion. The bands corresponding to the normal (wild-type, at right) and mutated (AP, at left) nucleotides are identified by arrows. See (*24*) for methods.

The identification of defective Jak3 expression in a case of SCID allows a number of conclusions to be drawn. First, the diminished

required for signaling through γ_c-containing receptors, is in contrast to other Jak family kinases, which are ubiquitously expressed and

rmed cell lines
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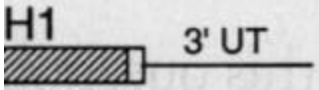
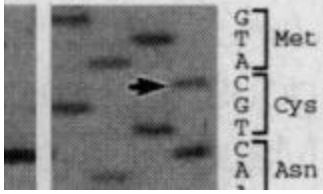
mber U09607)
y DNA (cDNA)



Wild type
C G A T C



agents that block γ_c-Jak3 association as immunosuppressants has been suggested (6). The current study further suggests that any agents that inactivate Jak3 function may be potent immunosuppressants. Moreover, the identification of Jak3 deficiency as the molecular basis for some autosomal recessive cases of SCID will allow the development of diagnostic procedures for identification of Jak3-deficient SCID patients and carriers and investigation into the possibility of gene therapy for Jak3-deficient SCID patients, analogous to that being developed for XSCID (23).

REFERENCES AND NOTES

1. T. Takeshita *et al.*, *Science.* **257**, 379 (1992).

31 OCTOBER *2003* VOL 302 *SCIENCE*

Prevention of Organ Allograft Rejection by a Specific Janus Kinase 3 Inhibitor

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Because of its requirement for signaling by multiple cytokines, Janus kinase 3 (JAK3) is an excellent target for clinical immunosuppression. We report the

with current therapies. Cytokine receptors, which use the common gamma chain, or γc [interleukin (IL)-2, -4, -7, -9, -15, -21], are critical for the development and homeostasis of immune cells, and patients with mutations in γc suffer from severe combined immunodeficiency (SCID) (*3*, *4*). These receptors all require the cytoplasmic tyrosine kinase JAK3 for signaling, and patients lacking expression of JAK3 also display a SCID phenotype (*5*, *6*). On the basis of the critical but selective role for JAK3 kinase in lymphocyte biology, we searched for inhibitors of this enzyme as potential immunosuppressive therapy.

The Pfizer chemical library was screened for inhibitors of in vitro JAK3 kinase activity, providing the lead compound, CP-352,664. Extensive chemical modification led to CP-690,550 (*7*) (Fig. 1A). Although CP-690,550 was highly potent for JAK3 inhibition [enzyme inhibitory potency (*8*) of 1 nM], it was 20- to 100-fold less potent for JAK2 and JAK1, respectively (Table 1). Because JAK2 mediates signaling via many hematopoietic cytokines [e.g., erythropoietin, thrombopoietin, and colony-stimulating factor receptors (*3*)], potent JAK2 inhibition could result in anemia, thrombocytopenia, and leukopenia in vivo. In addition, CP-690,550 did not have potent activity against 30 other kinases [all median inhibitory concentration (IC_{50}) > 3000 nM]. This included Lck, a key T lymphocyte–signaling molecule downstream of the T cell

Ming-Sing Si,[3] Ricardo Paniagua,[3] John Higgins,[6] Bari Holm,[3] Bruce Reitz,[5] Yong-Jie Zhou,[7] Randall E. Morris,[5] John J. O'Shea,[7] Dominic C. Borie[5]*

Because of its requirement for signaling by multiple cytokines, Janus kinase 3 (JAK3) is an excellent target for clinical immunosuppression. We report the development of a specific, orally active inhibitor of JAK3, CP-690,550, that significantly prolonged survival in a murine model of heart transplantation and in cynomolgus monkeys receiving kidney transplants. CP-690,550 treatment was not associated with hypertension, hyperlipidemia, or lymphoproliferative disease. On the basis of these preclinical results, we believe JAK3 blockade by CP-690,550 has potential for therapeutically desirable immunosuppression in human organ transplantation and in other clinical settings.

In spite of numerous treatment options for organ transplant and autoimmune disease patients (*1*), there remains a need for effective and safe immunosuppressive agents. The molecular targets for all currently used transplant drugs (cyclosporin A, tacrolimus, mycophenolate mofetil, and sirolimus) are ubiquitously expressed (*2*). In this respect, a

Research article

Open Access

Cartilage preservation by inhibition of Janus kinase 3 in two rodent models of rheumatoid arthritis

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This article is online at: http://arthritis-research.com/content/10/1/R14

Abstract

Introduction CP-690550 is a small molecule inhibitor of Janus kinase 3 (JAK3), a critical enzyme in the signaling pathway of multiple cytokines (interleukin (IL)-2, -7, -15 and -21) that are important in various T cell functions including development, activation and homeostasis. The purpose of this study was to evaluate CP-690550 in murine collagen-induced (CIA) and rat

Results CP-690550 dose-dependently decreased endpoints of disease in both RA models with greater than 90% reduction observed at the highest administered dose. An approximate ED_{50} of approximately 1.5 mg/kg/day was determined for the compound based upon disease endpoints in both RA models examined and corresponds to CP-690550 serum levels of 5.8

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Abstract

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Methods CIA and AA were induced using standard protocols and animals received the JAK3 inhibitor via osmotic mini-pump infusion at doses ranging from 1.5–15 mg/kg/day following disease induction. Arthritis was assessed by clinical scores in the CIA models and paw swelling monitored using a plethysmometer in the AA model until study conclusion, at which time animals were killed and evaluated histologically.

Results CP-690550 dose-dependently decreased endpoints of disease in both RA models with greater than 90% reduction observed at the highest administered dose. An approximate ED_{50} of approximately 1.5 mg/kg/day was determined for the compound based upon disease endpoints in both RA models examined and corresponds to CP-690550 serum levels of 5.8 ng/ml in mice (day 28) and 24 ng/ml in rats (day 24). The compound also reduced inflammatory cell influx and joint damage as measured histologically. Animals receiving a CP-690550 dose of 15 mg/k/d showed no histological evidence of disease.

Conclusion The efficacy observed with CP-690550 in CIA and AA suggests JAK3 inhibition may represent a novel therapeutic target for the treatment of RA.

Modulation of Innate and Adaptive Immune Responses by Tofacitinib (CP-690,550)

Kamran Ghoreschi,*[,1] Michael I. Jesson,[†,1] Xiong Li,[†] Jamie L. Lee,[†] Sarbani Ghosh,[†] Jason W. Alsup,[†] James D. Warner,[†] Masao Tanaka,[‡] Scott M. Steward-Tharp,* Massimo Gadina,[‡] Craig J. Thomas,[§] John C. Minnerly,[†] Chad E. Storer,[†] Timothy P. LaBranche,[†] Zaher A. Radi,[†] Martin E. Dowty,[†] Richard D. Head,[†] Debra M. Meyer,[†] Nandini Kishore,[†] and John J. O'Shea*

Inhibitors of the JAK family of nonreceptor tyrosine kinases have demonstrated clinical efficacy in rheumatoid arthritis and other inflammatory disorders; however, the precise mechanisms by which JAK inhibition improves inflammatory immune responses remain unclear. In this study, we examined the mode of action of tofacitinib (CP-690,550) on JAK/STAT signaling pathways involved in adaptive and innate immune responses. To determine the extent of inhibition of specific JAK/STAT-dependent pathways, we analyzed cytokine stimulation of mouse and human T cells in vitro. We also investigated the consequences of CP-690,550 treatment on Th cell differentiation of naive murine CD4$^+$ T cells. CP-690,550 inhibited IL-4–dependent Th2 cell differentiation and interestingly also interfered with Th17 cell differentiation. Expression of IL-23 receptor and the Th17 cytokines IL-17A, IL-17F, and IL-22 were blocked when naive Th cells were stimulated with IL-6 and IL-23. In contrast, IL-17A production was enhanced when Th17 cells were differentiated in the presence of TGF-β. Moreover, CP-690,550 also prevented the activation of STAT1, induction of T-bet, and subsequent generation of Th1 cells. In a model of established arthritis, CP-690,550 rapidly improved disease by inhibiting the production of inflammatory mediators and suppressing STAT1-dependent genes in joint tissue. Furthermore, efficacy in this disease model correlated with the inhibition of both JAK1 and JAK3 signaling pathways. CP-690,550 also modulated innate responses to LPS in vivo through a mechanism likely involving the inhibition of STAT1 signaling. Thus, CP-690,550 may improve autoimmune diseases and prevent transplant rejection by suppressing the differentiation of pathogenic Th1 and Th17 cells as well as innate immune cell signaling. *The Journal of Immunology*, **2011, 186: 4234–4243.**

Clinical Immunology (2013) 148, 66–78

available at www.sciencedirect.com

Clinical Immunology

www.elsevier.com/locate/yclim

REVIEW

Kinase inhibitors: A new tool for the treatment of rheumatoid arthritis

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T.K. Sengupta, A. Chen, Z. Zhong, J.E. Darnell Jr., L.B. Ivashkiv, Activation of monocyte effector genes and STAT family transcription factors by inflammatory synovial fluid is independent of interferon gamma, J. Exp. Med. 181 (3) (1995) 1015–1025.

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JAK Inhibition With Tofacitinib Suppresses Arthritic Joint Structural Damage Through Decreased RANKL Production

Timothy P. LaBranche, Michael I. Jesson, Zaher A. Radi, Chad E. Storer, Julia A. Guzova, Sheri L. Bonar, Janice M. Thompson, Fernando A. Happa, Zachary S. Stewart, Yutian Zhan, Chris S. Bollinger, Prashant N. Bansal, Jeremy W. Wellen, Dean P. Wilkie, Steven A. Bailey, Peter T. Symanowicz, Martin Hegen, Richard D. Head, Nandini Kishore, Gabriel Mbalaviele, and Debra M. Meyer

Objective. **The mechanistic link between Janus kinase (JAK) signaling and structural damage to arthritic joints in rheumatoid arthritis (RA) is poorly understood. This study was undertaken to investigate how selective inhibition of JAK with tofacitinib (CP-690,550) affects osteoclast-mediated bone resorption in a rat adjuvant-induced arthritis (AIA) model, as well as assessed via quantitative tartrate-resistant acid phosphatase staining and degradation of human bone collagen, respectively.**

Results. **Edema, inflammation, and osteoclast-mediated bone resorption in rats with AIA were dramatically reduced after 7 days of treatment with the JAK inhibitor, which correlated with reduced numbers**

Tofacitinib σε ασθενείς με RA

- 5 μελέτες φάσεως II
- 5 μελέτες φάσεως III
- Σαν β΄ γραμμής φάρμακο ή
- Μονοθεραπεία (σε δυσανεξία στη MTX)


96
Σεπτέμβριος
September 2011
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THANKS !!!!

Ευχαριστώ !!!