Even more receptor families

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JAK/STAT Cytokine Receptors

Discovery of interferons

Virus interference. I. The interferon

By A. Isaacs and J. Lindenmann*

National Institute for Medical Research, London

(*Communicated by C. H. Andrewes, F.R.S.—Received 7 March* 1957)

During a study of the interference produced by heat-inactivated influenza virus with the growth of live virus in fragments of chick chorio-allantoic membrane it was found that following incubation of heated virus with membrane a new factor was released. This factor, recognized by its ability to induce interference in fresh pieces of chorio-allantoic membrane, was called interferon. Following a lag phase interferon was first detected in the membranes after 3 h incubation and thereafter it was released into the surrounding fluid.

Proceedings of the Royal Society of London. Series B, Biological Sciences. **147** (927): 258–67.

IFN-induced gene expression

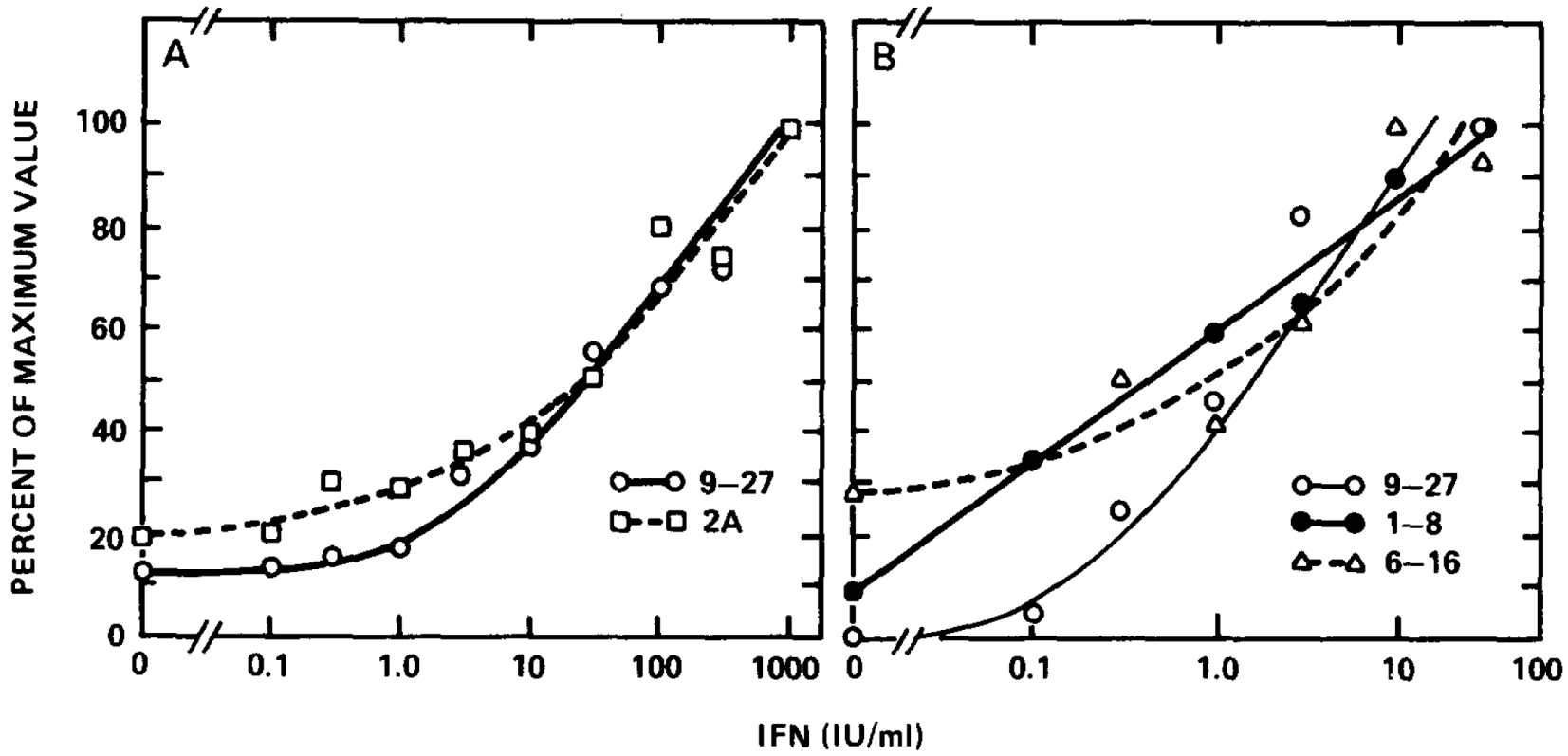


Figure 3. Induction of mRNAs by Different Amounts of IFN

Cells were treated for 8 hr with the indicated concentrations of IFN. Total RNA (200 μg/slot) was bound to nitrocellulose and probed with each cDNA. The experiments shown in A and B were carried out on different days. As noted in the text, levels of the mRNAs in untreated cells varied from day to day.

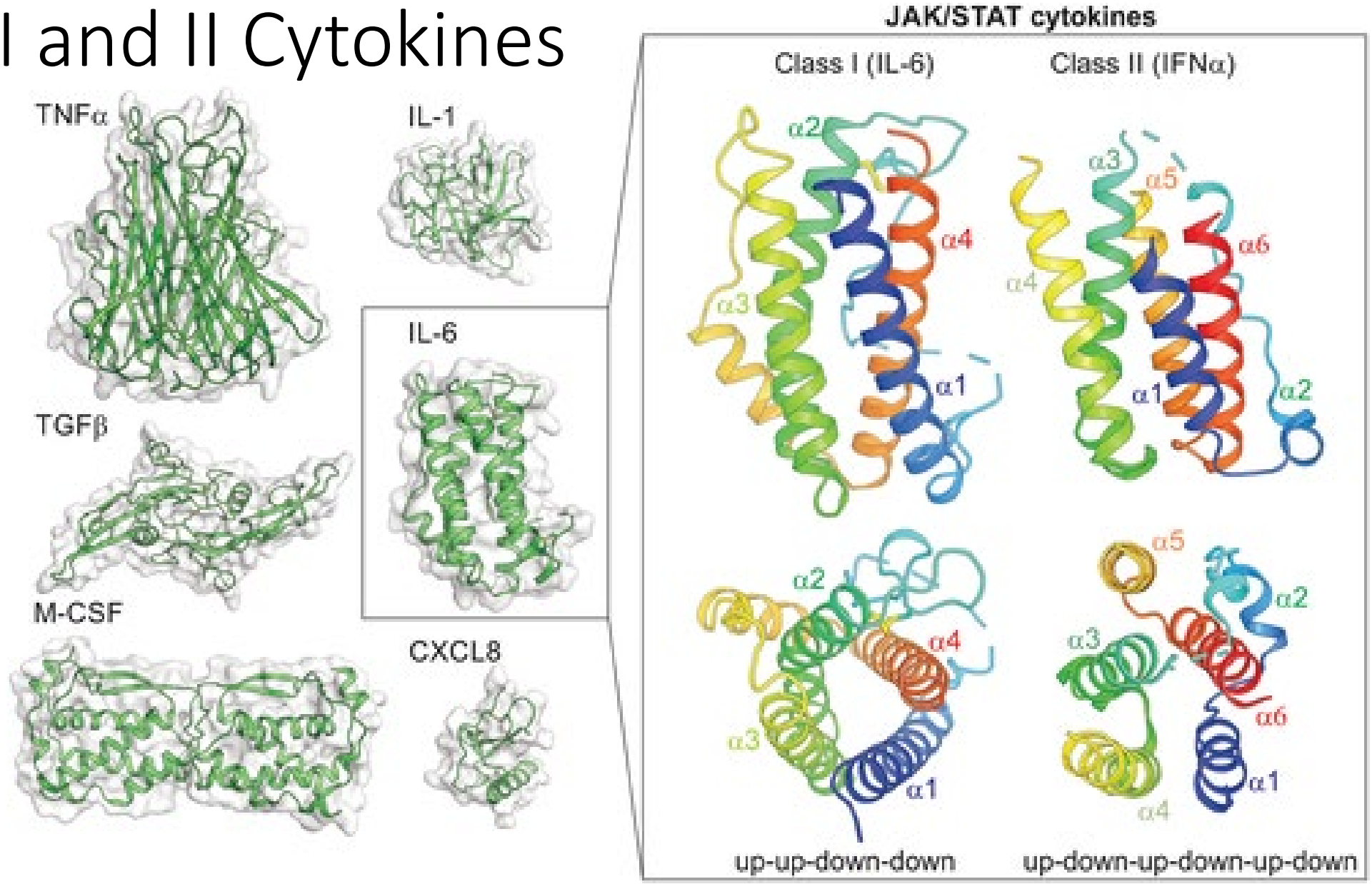
Cell, Vol. 38, 745-755, October 1984

Table 1. Sequence of the Discoveries that Have Revealed the JAK-STAT Pathways

Year	Milestone
1957	Isaacs and Lindenmann describe interferon
1975–1977	Oligonucleotide [2′-5′ oligoadenylates(s)] inhibitors of protein synthesis induced by IFN found
1979	Actinomycin-sensitive IFN-β-dependent new protein induction shown
1984	IFN-α-induced transcriptional stimulation of specific genes (ISGs) demonstrated; no new protein synthesis required
1986–1988	IFN-dependent promoters identified (ISREs, interferon-stimulated response elements)
1988–1989	IFN-α-induced ISRE binding protein complexes (ISGF3; E complex) in cytoplasm in 1–2 min; in nucleus in 5 min
1989	Genetic selection system for defective IFN-induced transcription described and first cell mutant selected
	IFN-γ-dependent promoters (GAS, gamma IFN-activated sequences, and GAF, gamma IFN-activated factor) identified
1989–1991	JAKs 1 and 2 and TYK2 identified
1990	ISGF-3 partially purified; identified subunits 113, 91, 84, 48
1991	Noncomplementing mutant cells unresponsive to both IFN-α and IFN-γ described
1992	cDNA clones sequenced later called STAT1 (a and b) and STAT2; RNA for IRF9 completing make up of ISGF3, establishing STAT family of proteins
	First IFN response mutant identified as Tyk2 by molecular complementation
	Upon IFN activation by IFN-α STAT1 and STAT2 are tyrosine phosphorylated; STAT1 also tyrosine phosphorylated after IFN-γ treatment
1993–1994	Major signaling events driven by IFN and IL-6 pinpointed by molecular complementation of mutant cells
1994	JAK3 described and sequenced
1994–1995	STAT3, 4, 5A, 5B, and 6 all described and sequenced
1995–1998	Functional and structural domains of STATs described
1996	*Drosophila* STAT (dSTAT92E) first described; later studied extensively genetically
	Mouse genetics identifies physiological functions for all STATs in various specific cells
1997	Negative regulation of pathways initially characterized
1998	First crystal structures of STATs
2000	Initial information that human mutations in JAKs and STATs and persistent activation of STATs cause disease
	First posttranslational modifications of STATs in addition to phosphorylations noted (methylation, acetylation, etc.)
2001	Comprehensive gene target sets identified

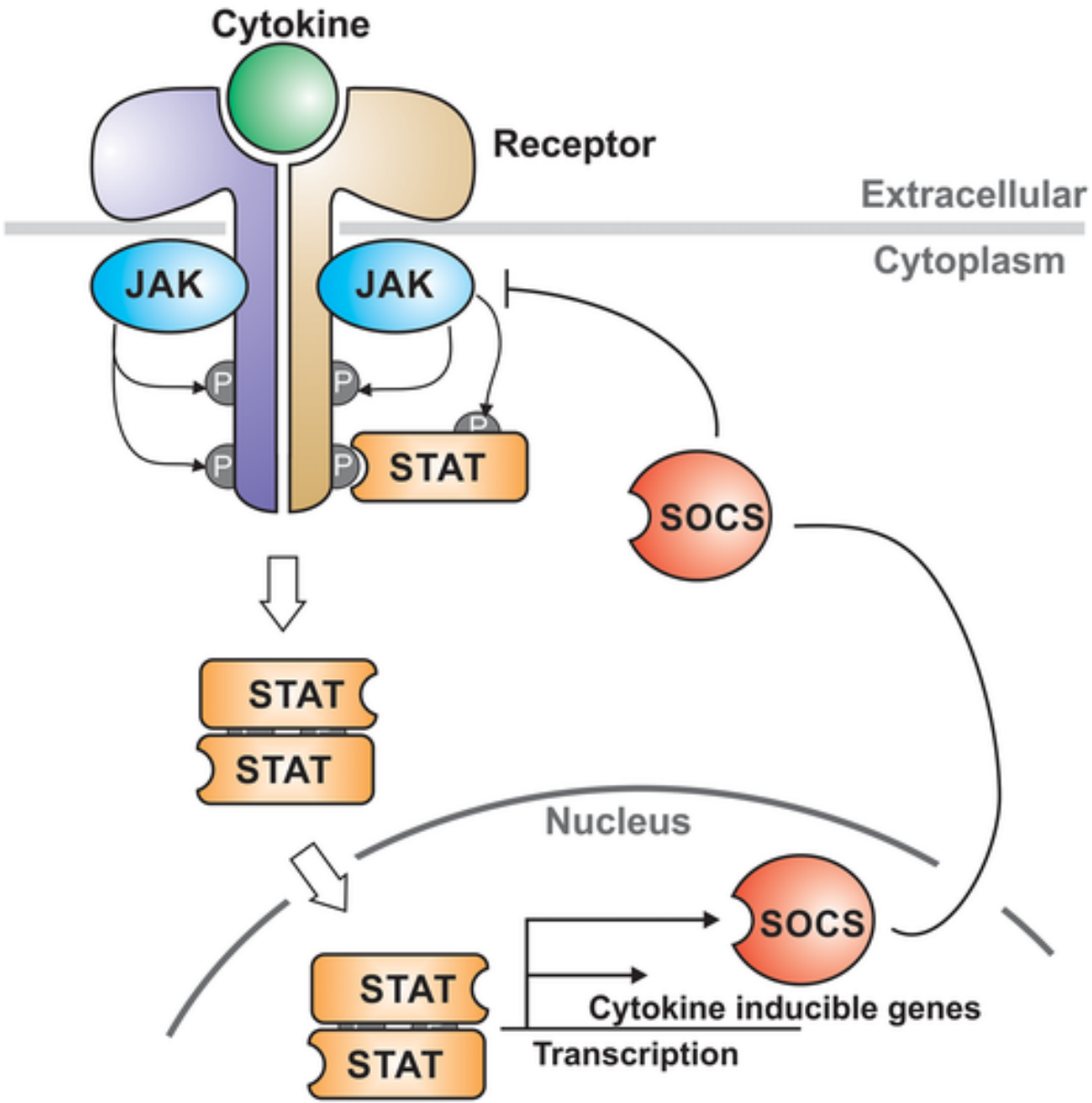
Immunity 36, April 20, 2012

Class I and II Cytokines



Protein Science, Volume: 27, Issue: 12, Pages: 1984-2009, First published: 29 September 2018, DOI: (10.1002/pro.3519)

The molecular details of cytokine signaling via the JAK/STAT pathway



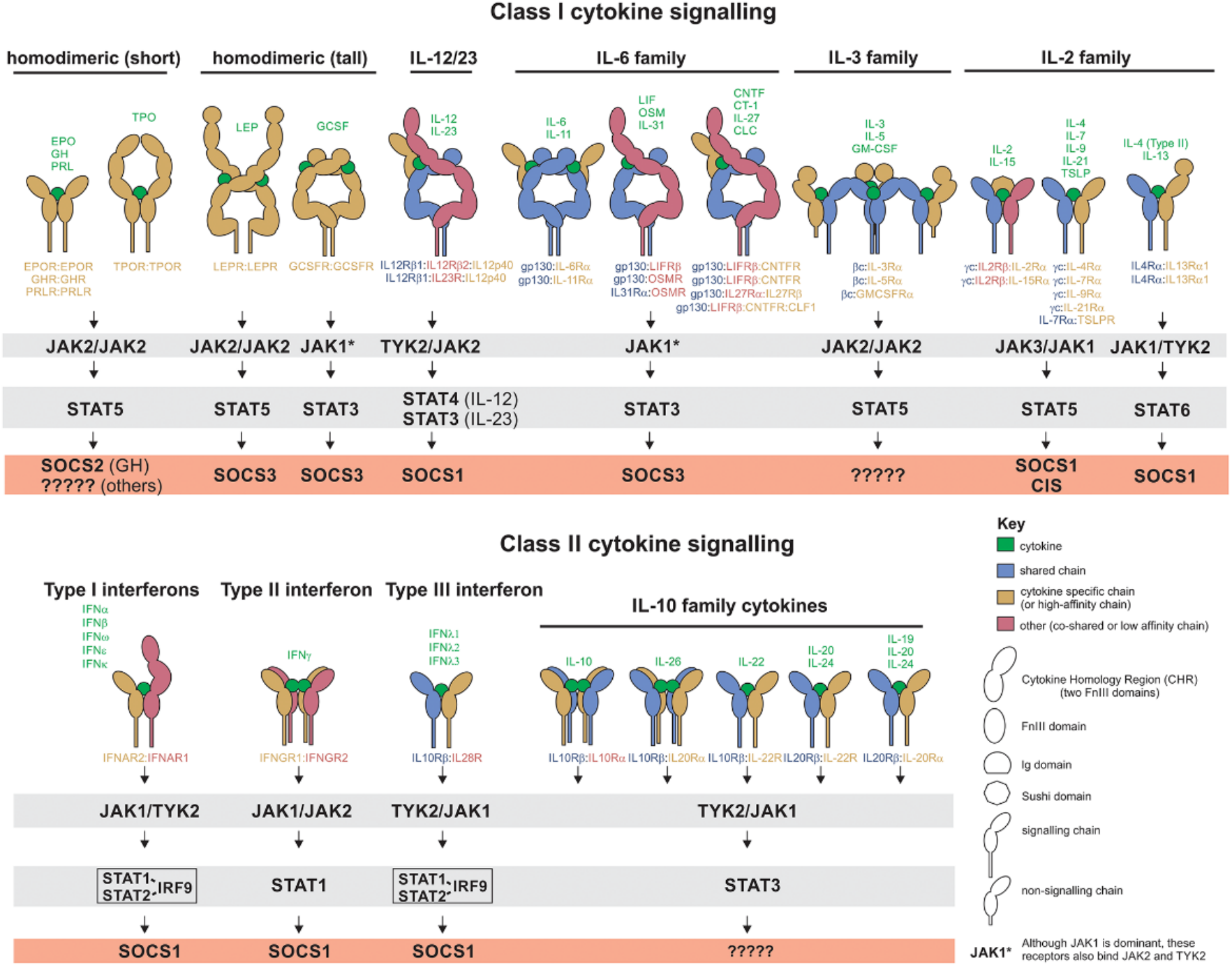
Components for response to cytokine:

- Receptor
- Receptor Kinase (JAK)
- Transcription Factor (STAT)
- SOCS – suppressor of cytokine signaling

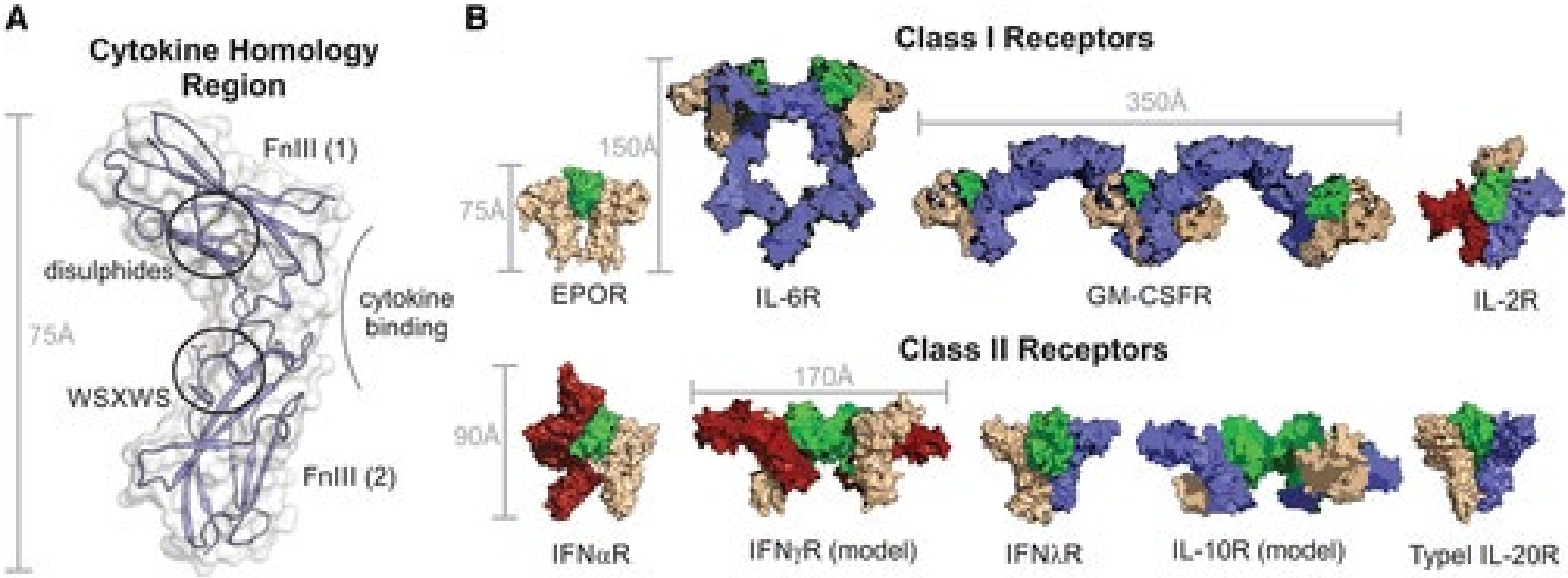
Protein Science, Volume: 27, Issue: 12, Pages: 1984-2009, First published: 29 September 2018, DOI: (10.1002/pro.3519)

CHR – cytokine receptor homology region – composed of FnIII domains

Cytokines bind at the junction between two FnIII domains

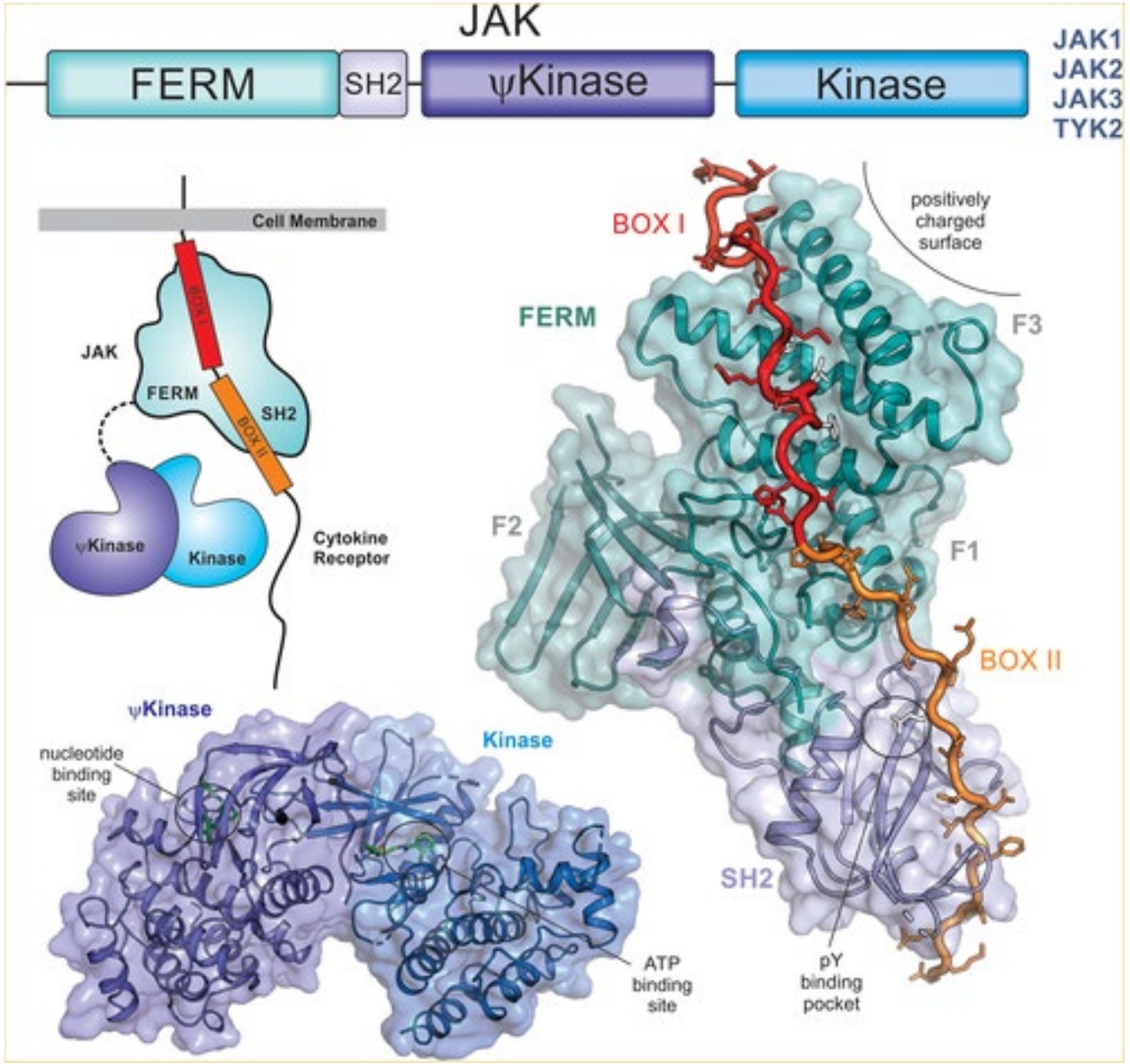


Protein Science, Volume: 27, Issue: 12, Pages: 1984-2009, First published: 29 September 2018, DOI: (10.1002/pro.3519)



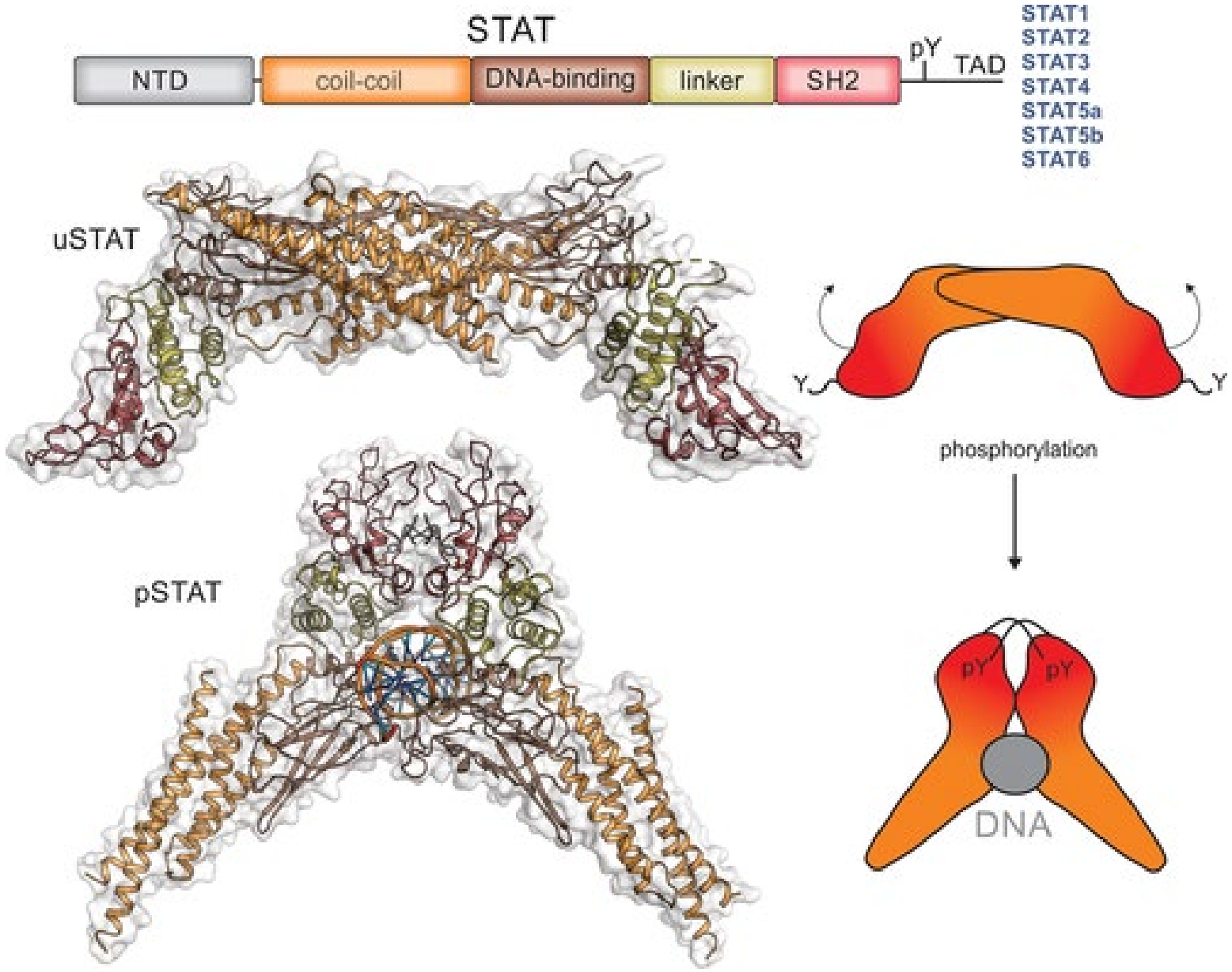
Protein Science, Volume: 27, Issue: 12, Pages: 1984-2009, First published: 29 September 2018, DOI: (10.1002/pro.3519)

- Four members of JAK family: JAK1-3 and Tyk2
- FERM and SH2 domains tether JAK to the receptor
  - FERM binds Box I
  - SH2 binds Box II (through a Glu, not a pTyr)
- Pseudokinase (ψK) regulates kinase activity
  - V617F mutation of JAK2 results in a hyperactive kinase associated with myeloproliferative diseases
- JAKs phosphorylate activation loops in *trans* like RTKs



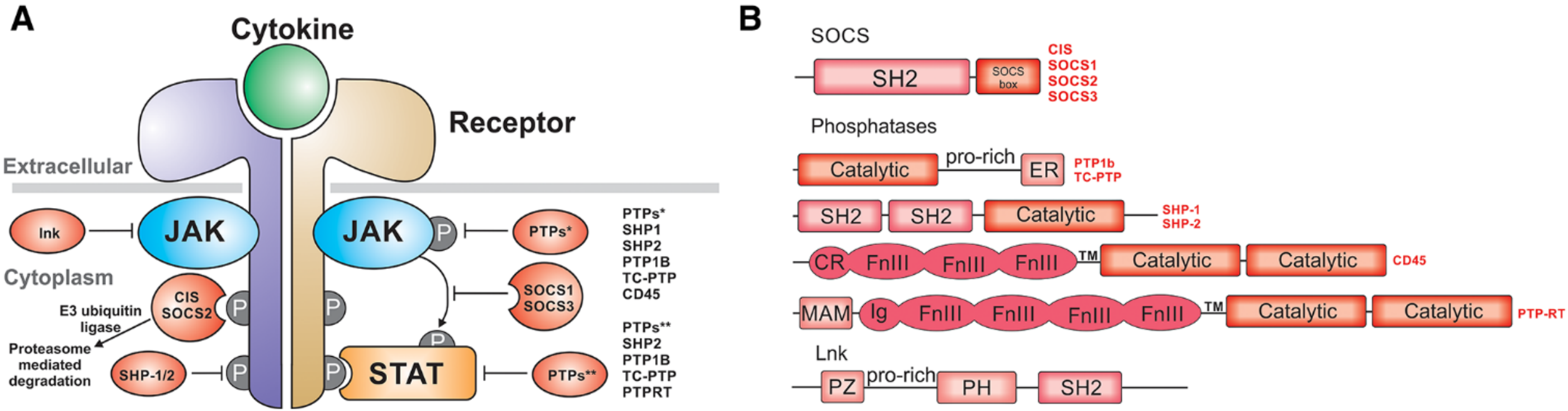
Protein Science, Volume: 27, Issue: 12, Pages: 1984-2009, First published: 29 September 2018, DOI: (10.1002/pro.3519)

- Activated JAK phosphorylates the receptor, promoting STAT binding through the SH2 domain
- JAKs also phosphorylate Tyr residue between the SH2 and transactivation domain
  - SH2 domain then binds to the pTyr of the partner in the dimer



Protein Science, Volume: 27, Issue: 12, Pages: 1984-2009, First published: 29 September 2018, DOI: (10.1002/pro.3519)

Negative regulation of cytokine receptor signaling

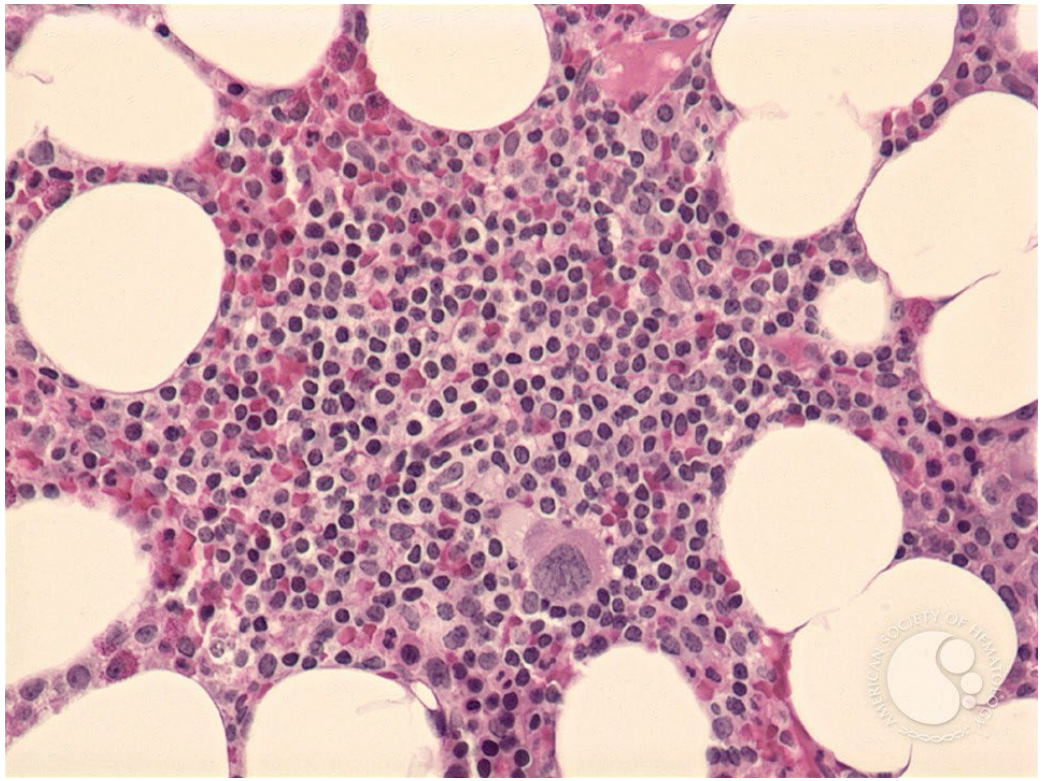


SOCS proteins can inhibit signaling by: 1) binding the receptor and promoting its degradation by ubiquitination; and 2) directly inhibiting JAKs (for SOCS1 and SOCS3)

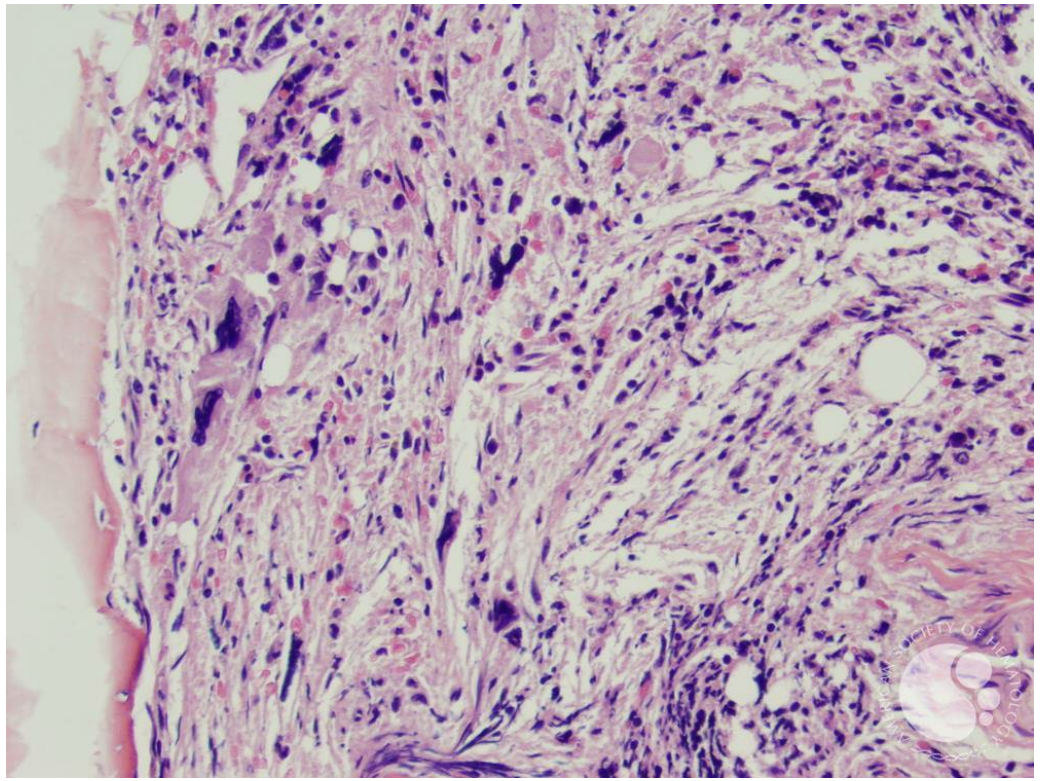
Protein Science, Volume: 27, Issue: 12, Pages: 1984-2009, First published: 29 September 2018, DOI: (10.1002/pro.3519)

Myelofibrosis – scarring of the bone marrow

Normal

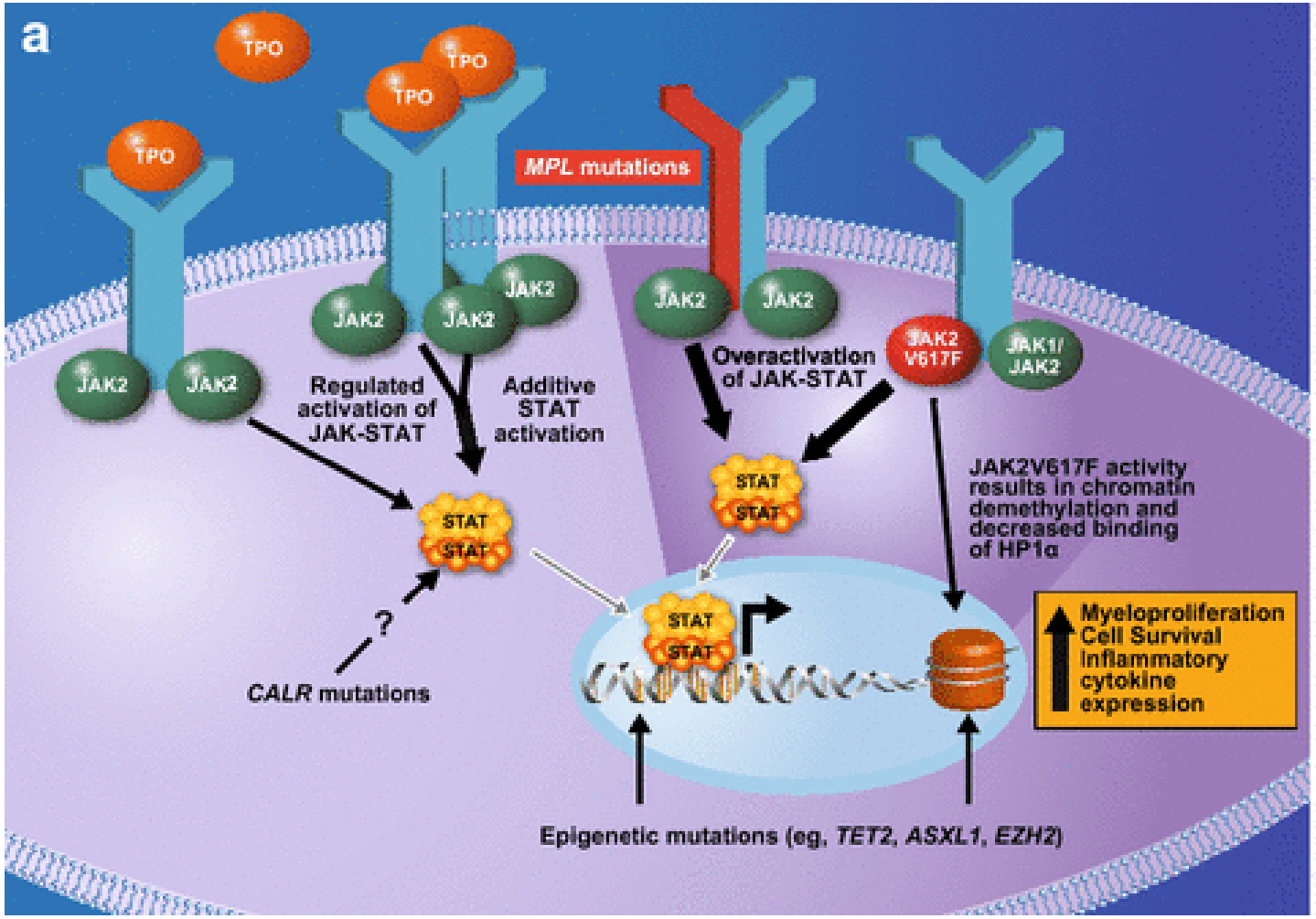


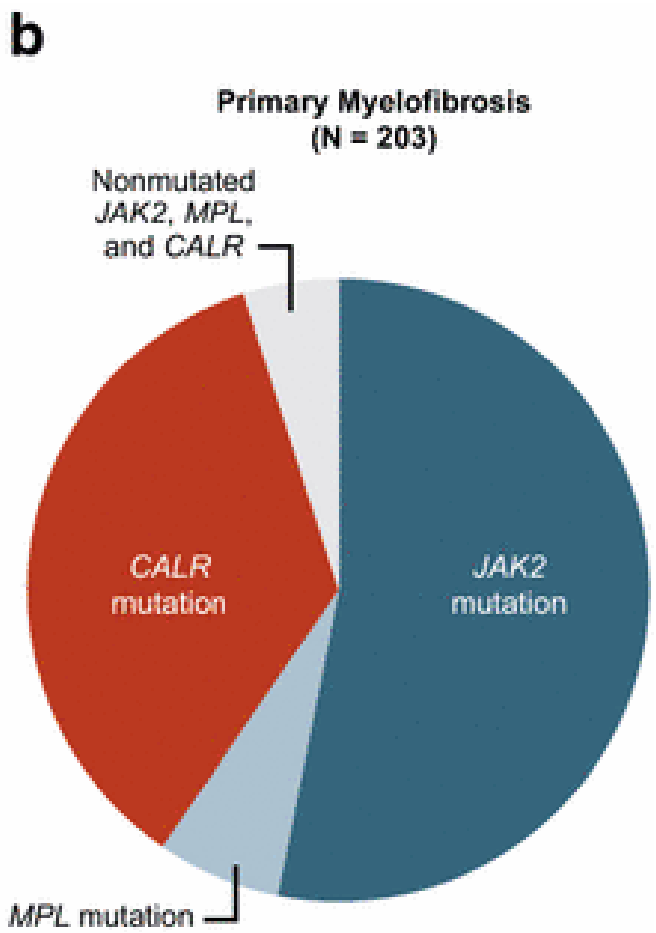
Myelofibrosis



https://imagebank.hematology.org/

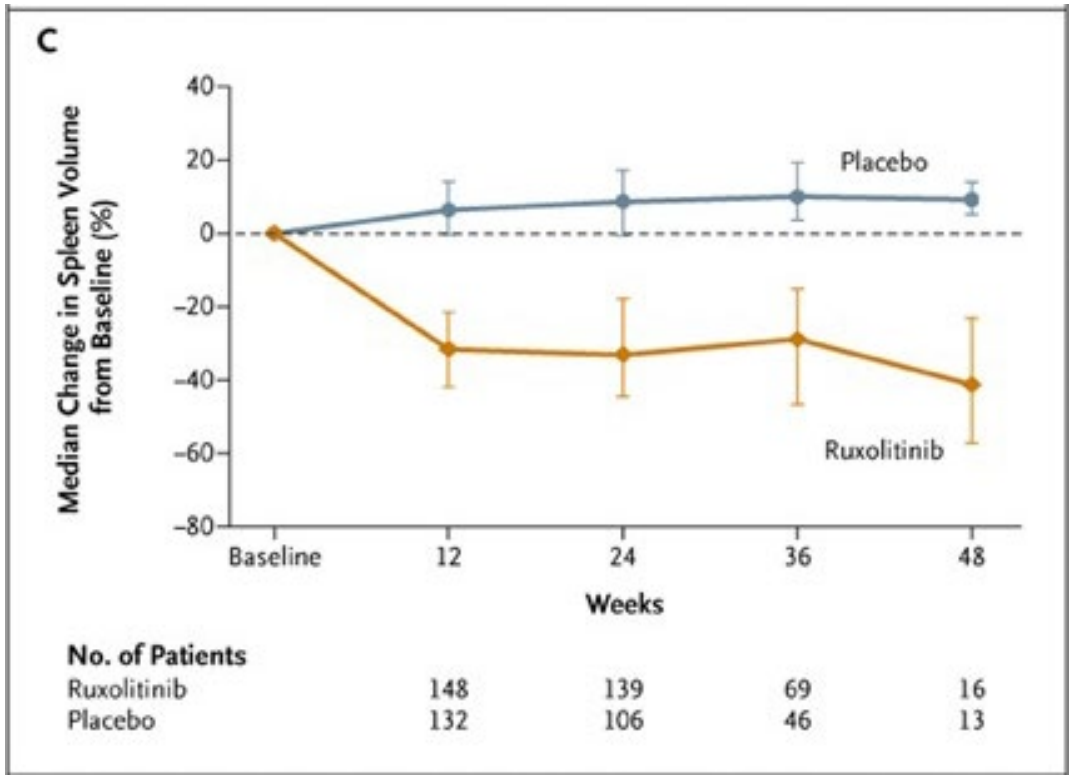
JAK2 and Growth Factor Receptor Mutations in Myelofibrosis

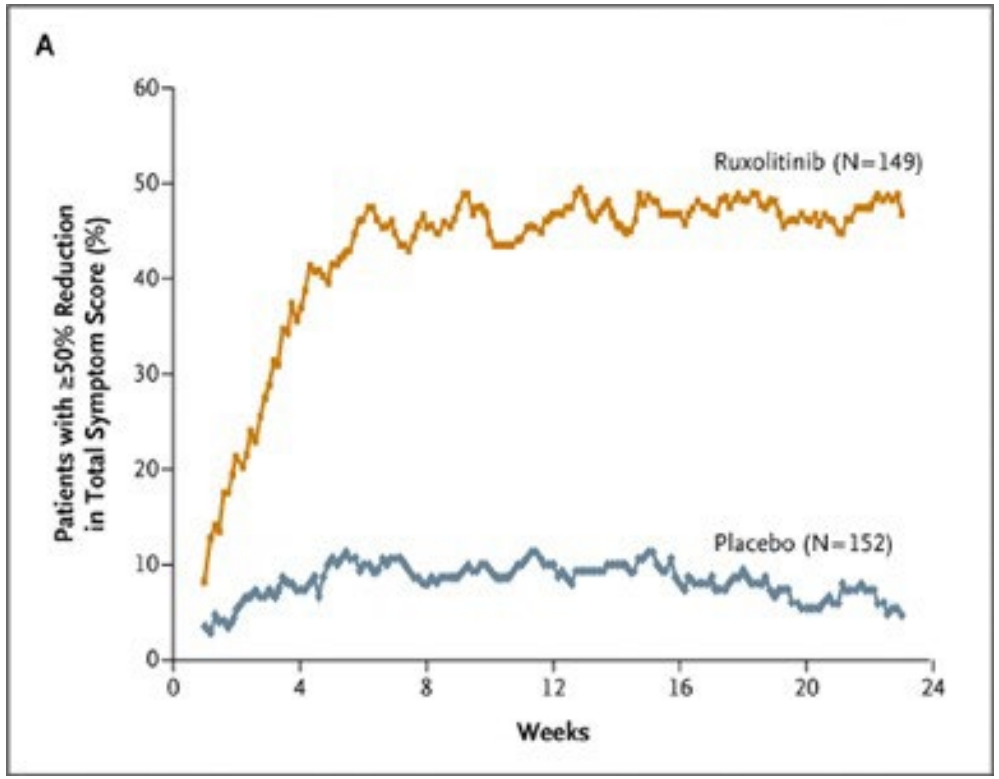




Cancer Chemotherapy and Pharmacology **volume 77**, pages1125–1142(2016)

Treatment of Myelofibrosis with a JAK1/2 inhibitor: Ruxolitinib





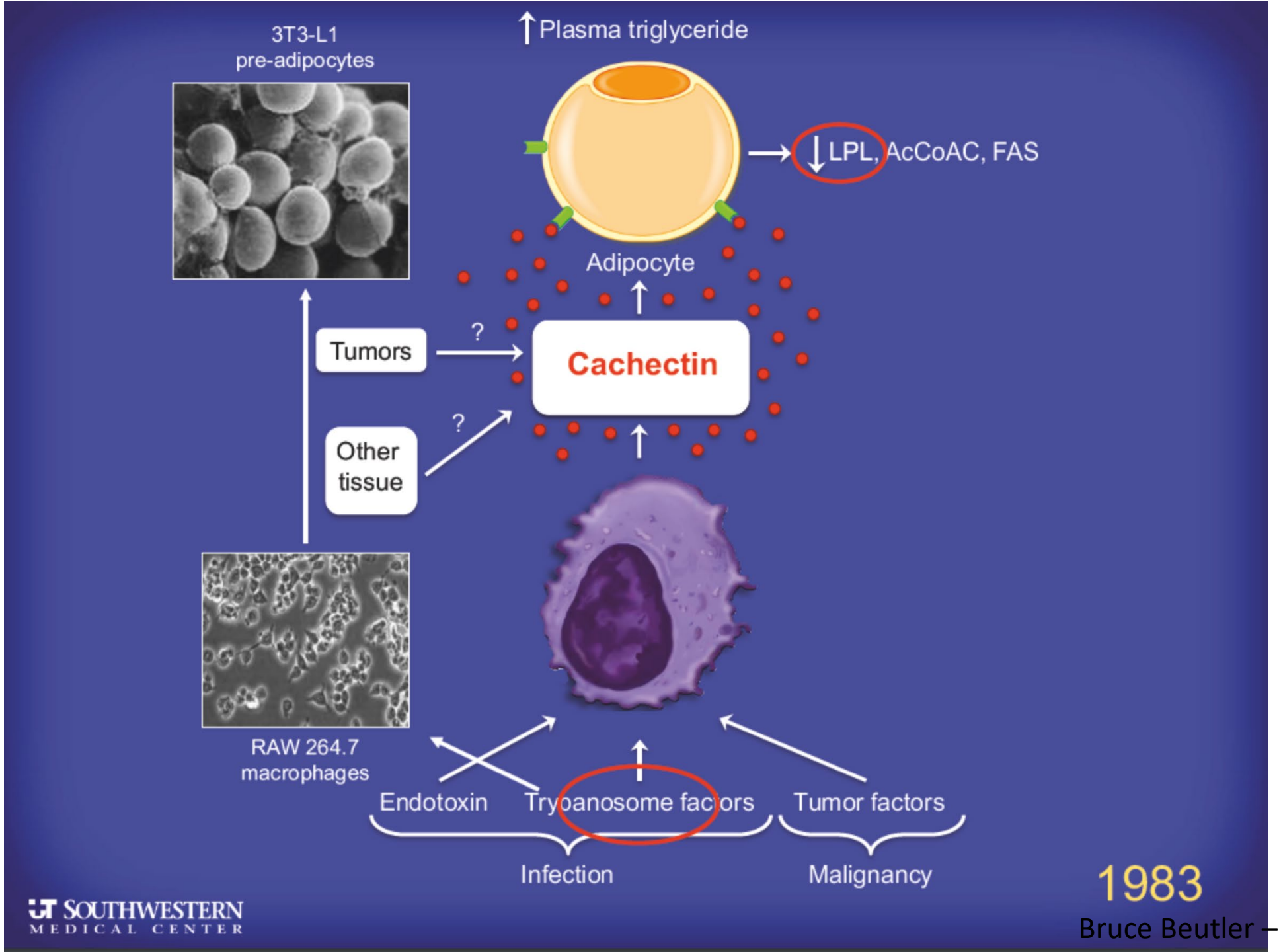
Verstovsek S et al. N Engl J Med 2012;366:799-807.

TNF-α receptor superfamily

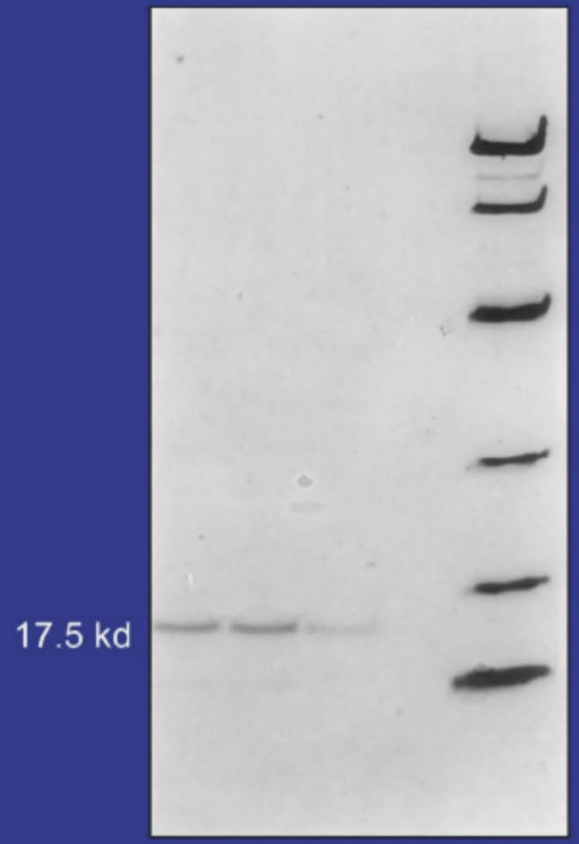
TNFs

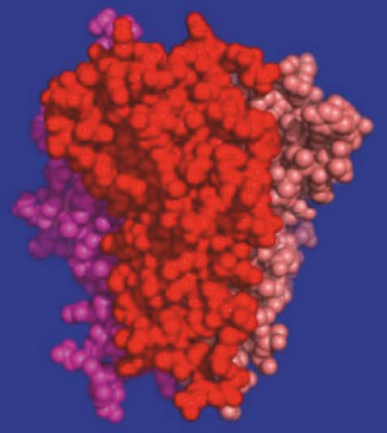
- 1968: Gale Granger and Nancy Ruddle – Cytotoxic factor produced by lymphocytes (lymphotoxin)
- 1975: Lloyd Old – Cytotoxic factor produced by macrophages and named it tumor necrosis factor (TNF). Factors could kill fibrosarcoma cells.
- 1984: cDNAs encoding LT and TNF were cloned in 1984 – similar structure
- 1985: Bruce Beutler and Anthony Cerami – cachectin was actually TNF

Wasting disease (cachexia) in a cow with African trypanosomiasis



Bruce Beutler – Nobel Lecture





TNF

Cachectin = Mouse tumor necrosis factor

(mouse CACH)

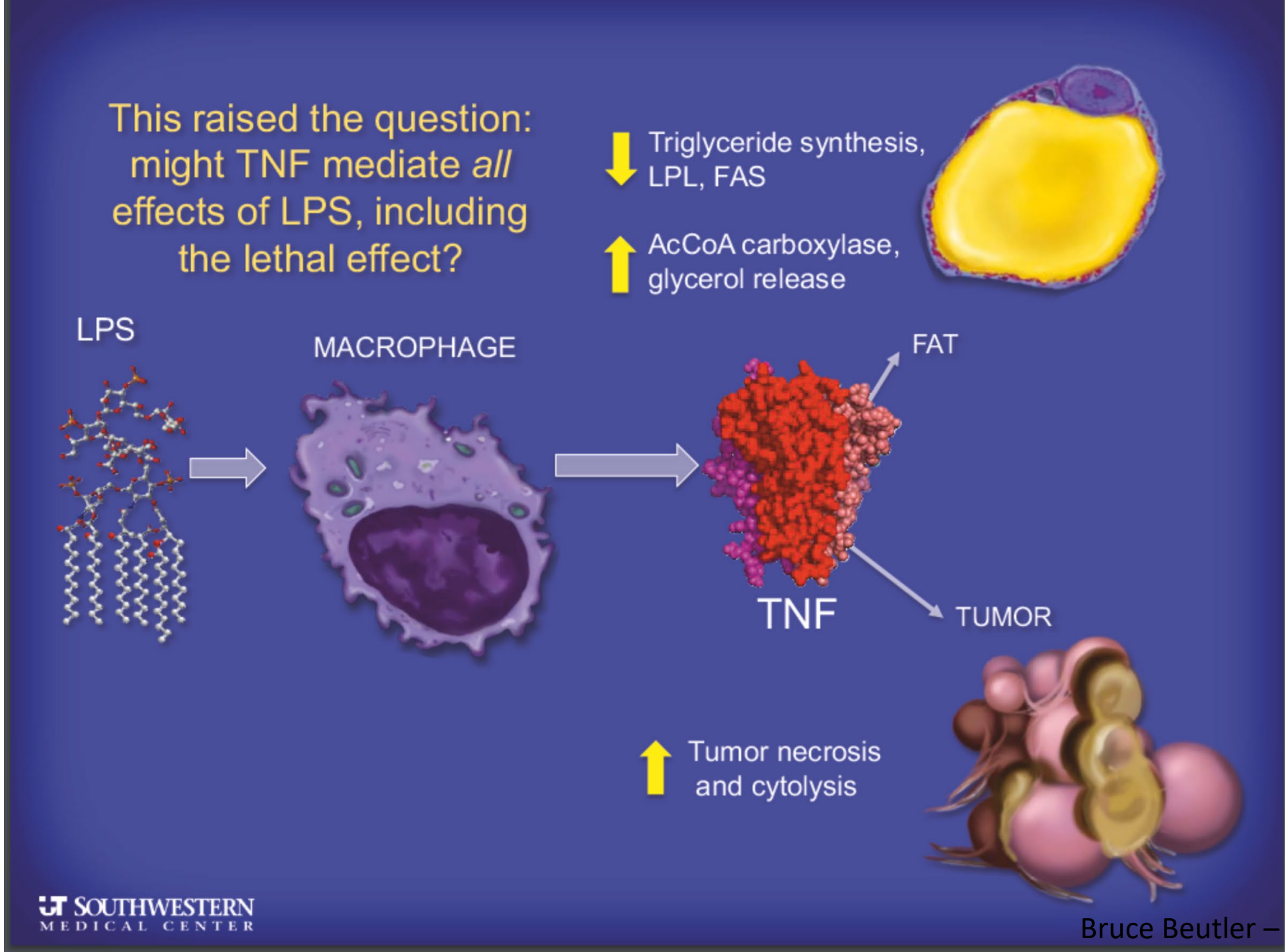
H_2N LEU-ARG-SER-SER-SER-GLU-ASN-SER-SER-ASP-PRO-PRO-VAL-ALA- ? -VAL-VAL-ALA-ASN…

H_2N VAL-ARG-SER-SER-SER-ARG-THR-PRO-SER-ASP-LYS-PRO-VAL-ALA-HIS-VAL-VAL-ALA-ASN…

(human TNF)

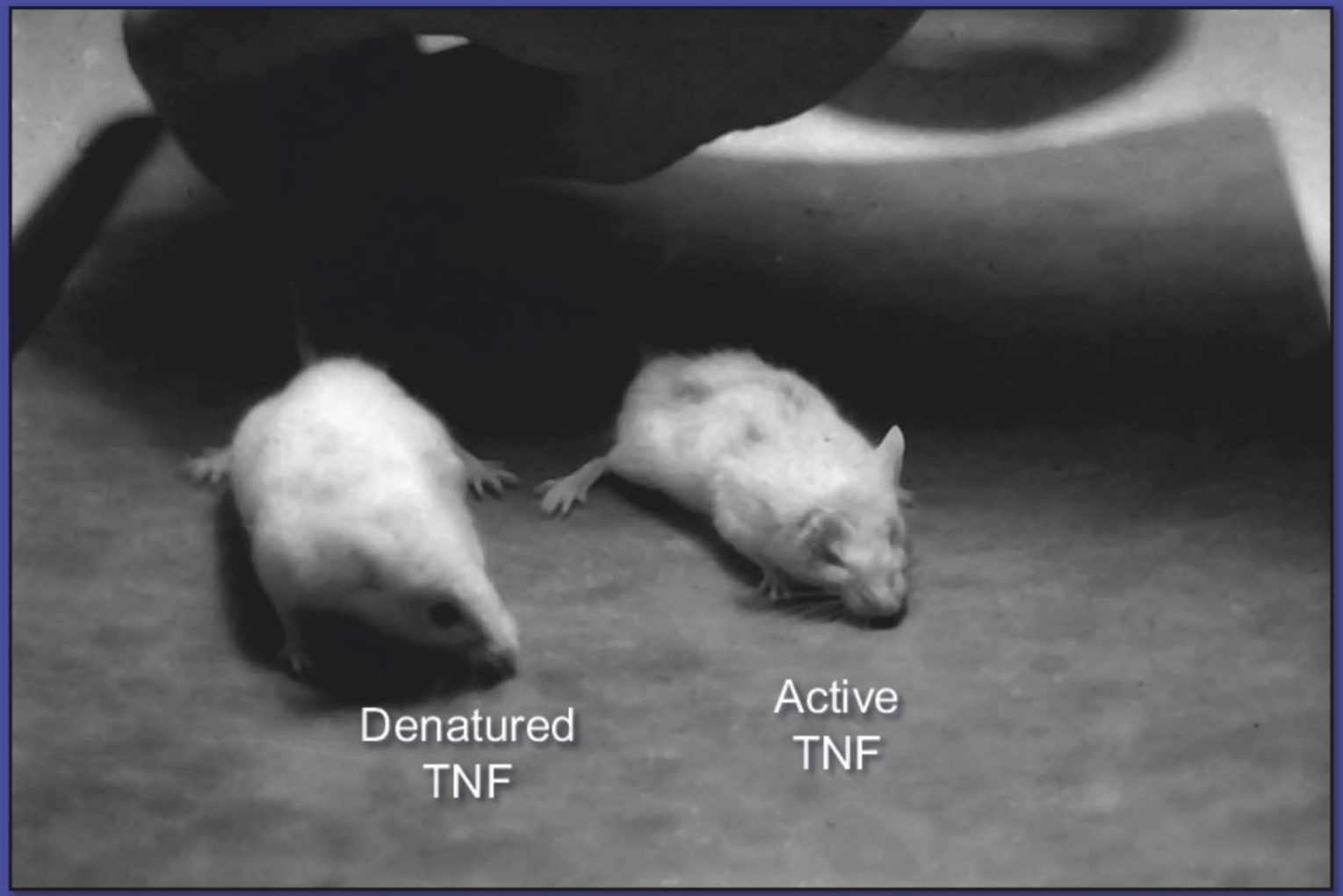
1 µg of cachectin had 10^8 U of TNF activity

UT SOUTHWESTERN MEDICAL CENTER



Bruce Beutler – Nobel Lecture

Purified TNF mimics LPS toxicity

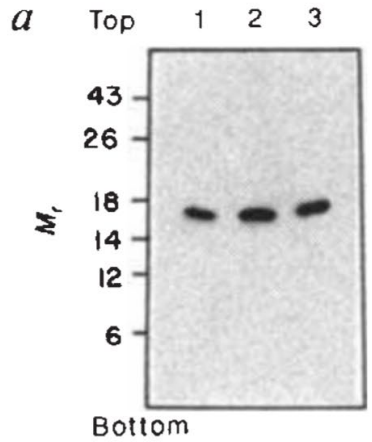


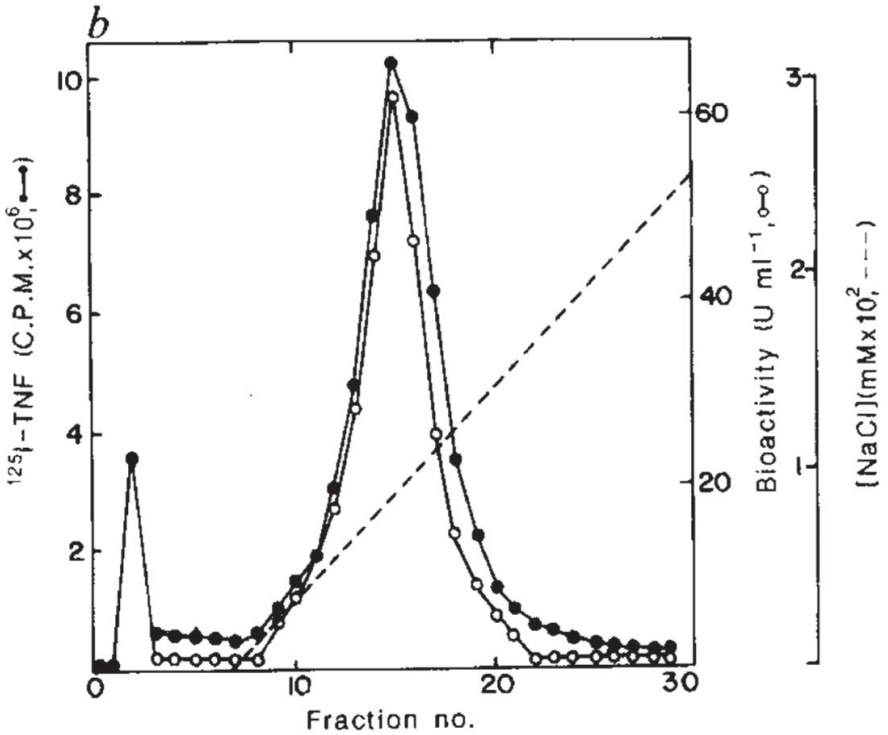
1984

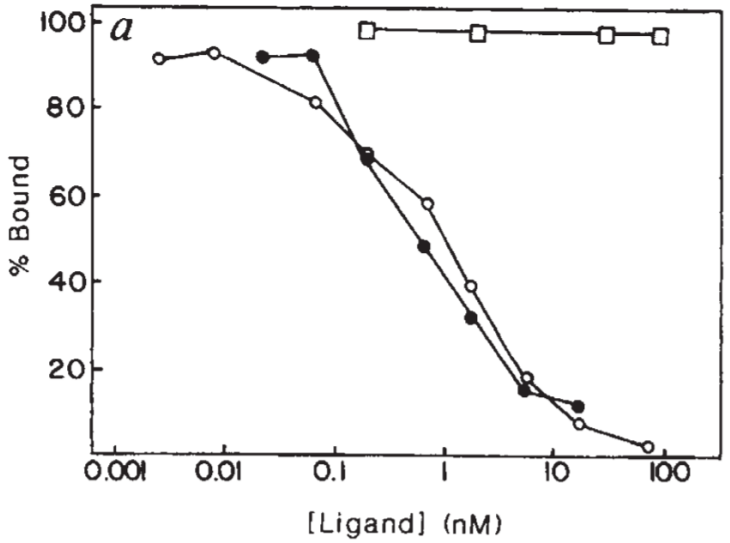
UT SOUTHWESTERN MEDICAL CENTER

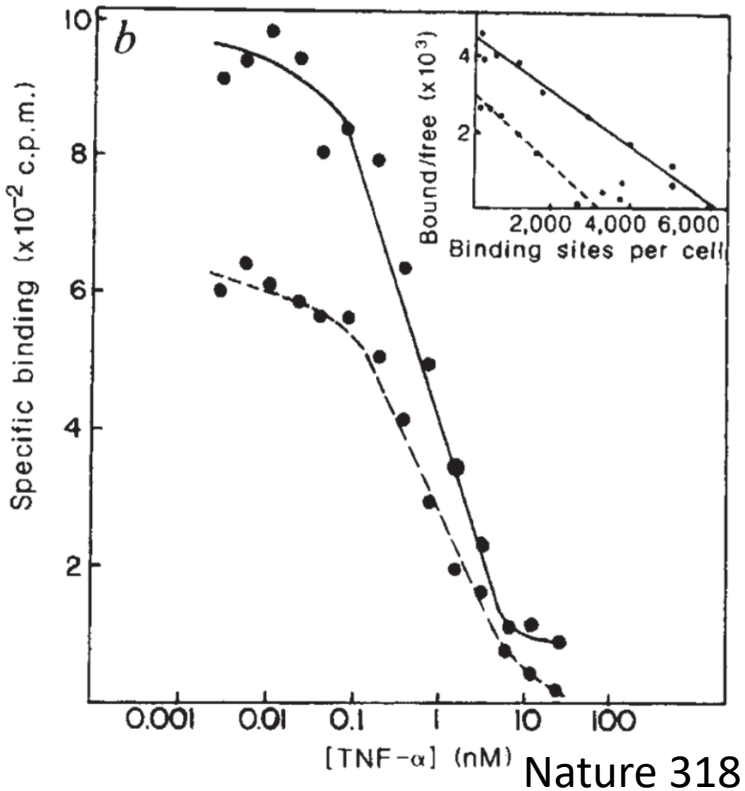
Bruce Beutler – Nobel Lecture

TNFR1



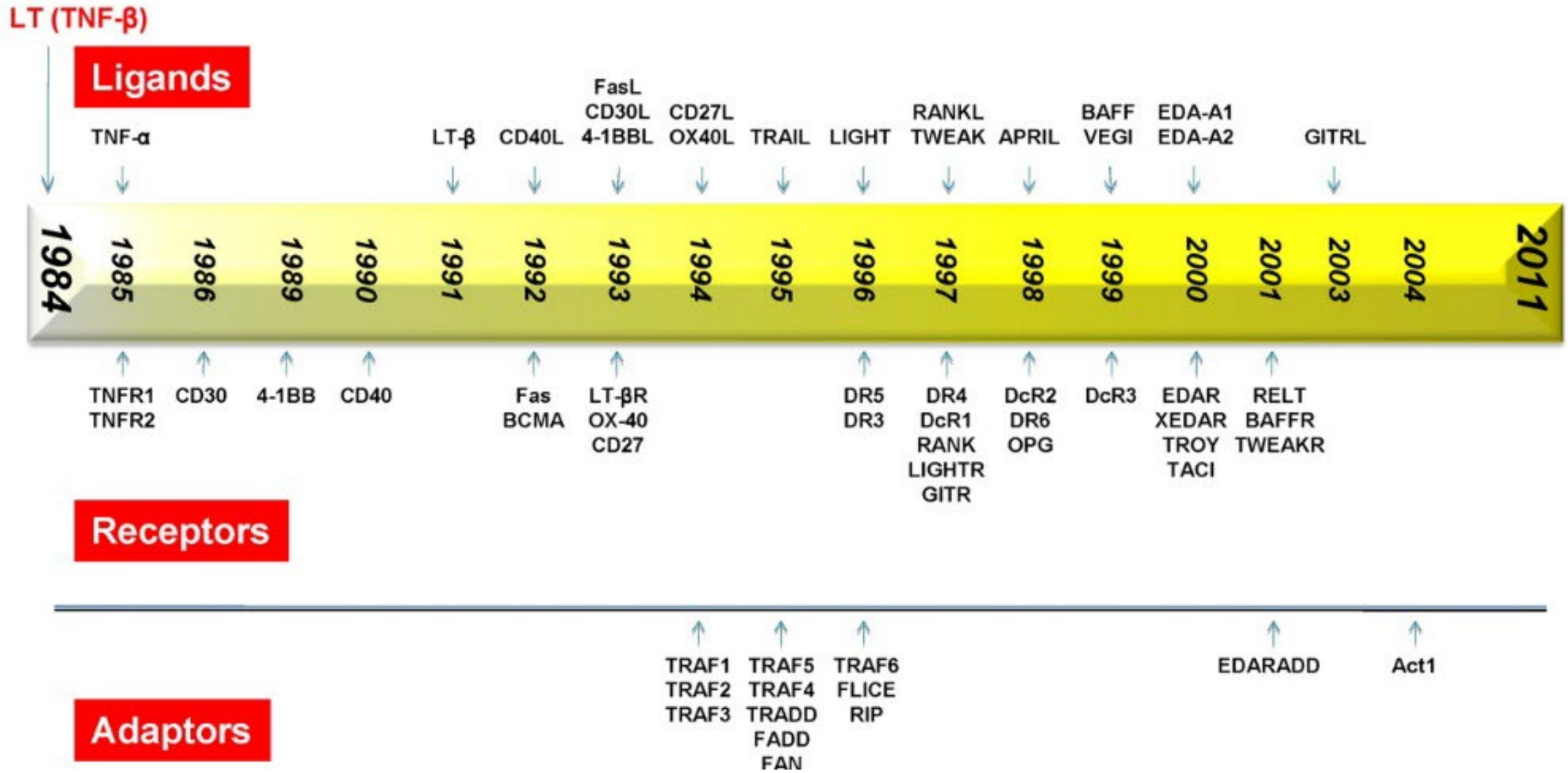






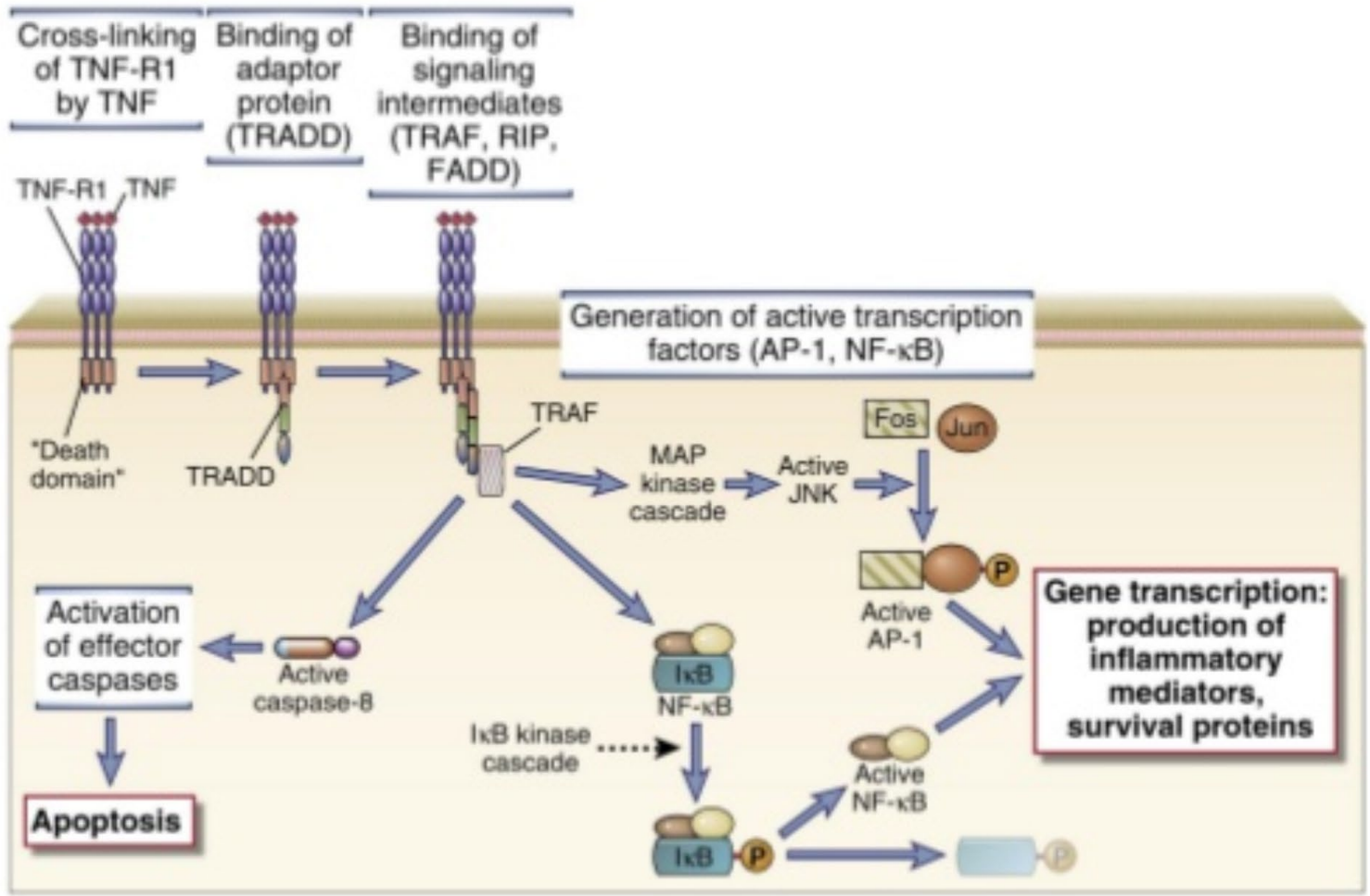
Nature 318 (6047), 665-7 1985

Historical perspectives on tumor necrosis factor and its superfamily: 25 years later, a golden journey



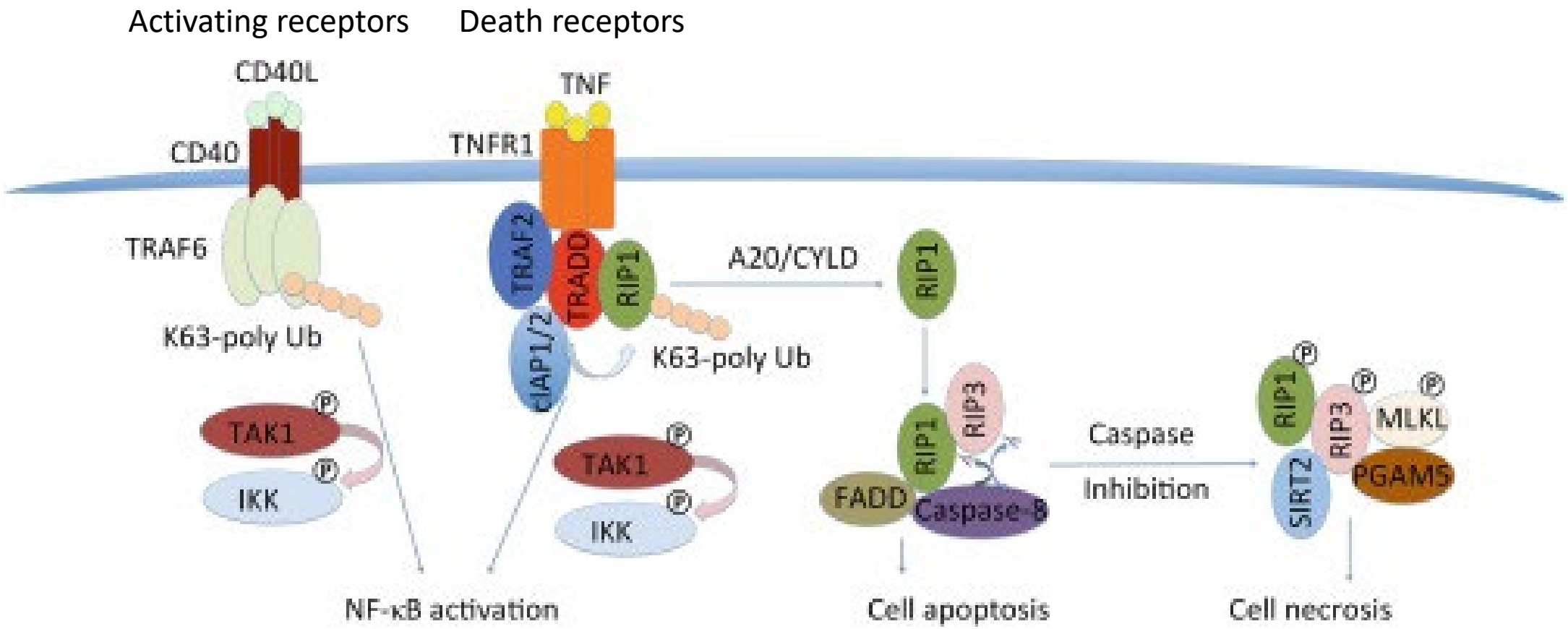
Bharat B. Aggarwal,Subash C. Gupta,Ji Hye Kim, Historical perspectives on tumor necrosis factor and its superfamily: 25 years later, a golden journey, Blood, 2012, Figure 2

TNF Receptor Family



Abbas AK, et al. Cellular and molecular immunology Ed 8th

TNF receptor superfamily



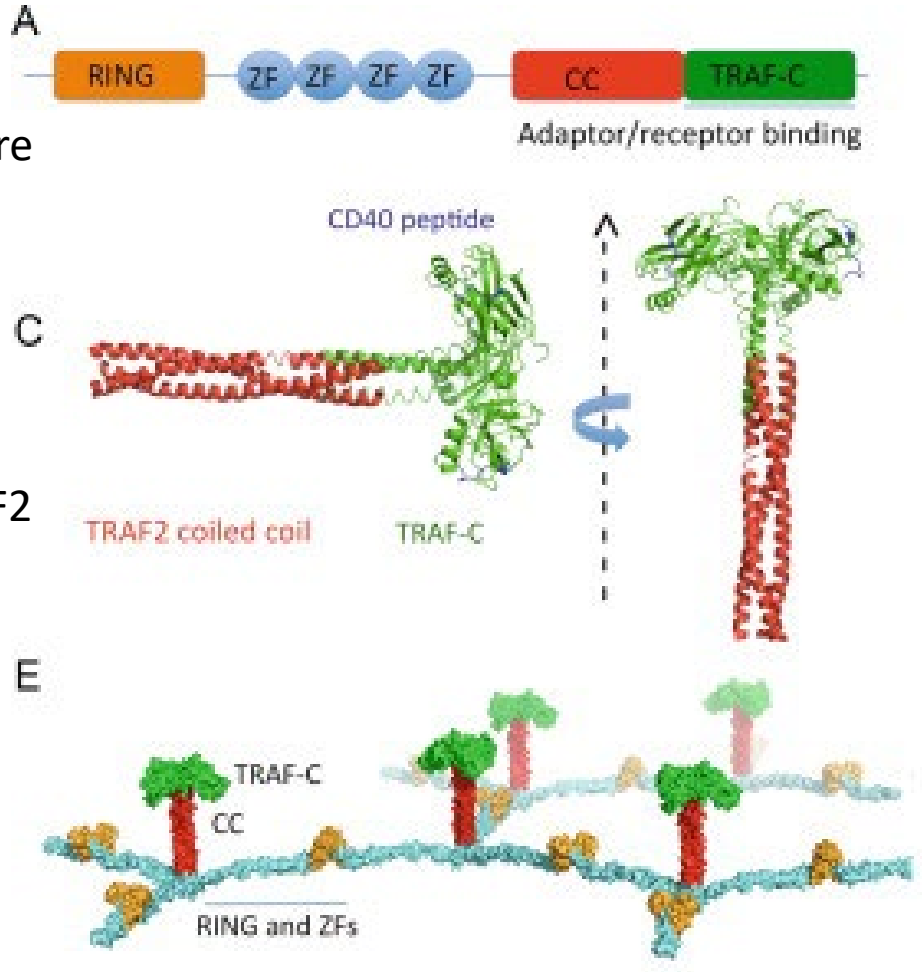
http://dx.doi.org/10.1016/B978-0-12-407707-2.00005-9

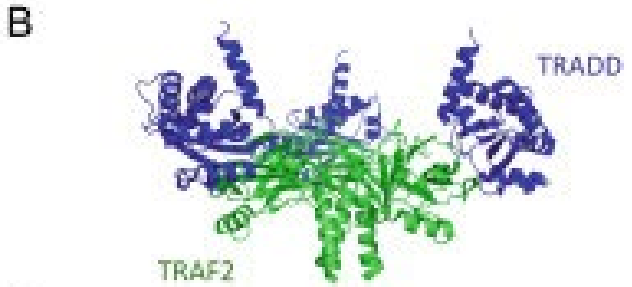
Downstream activation of TRAFs and TAK1

TRAF2 and 6 structure

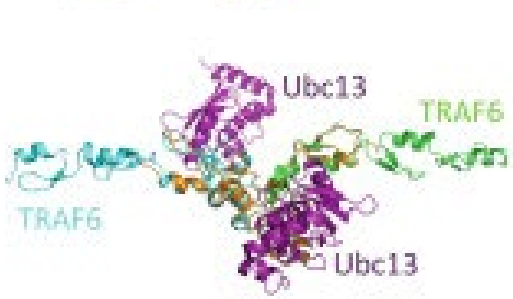
Activating receptor interaction with TRAF2

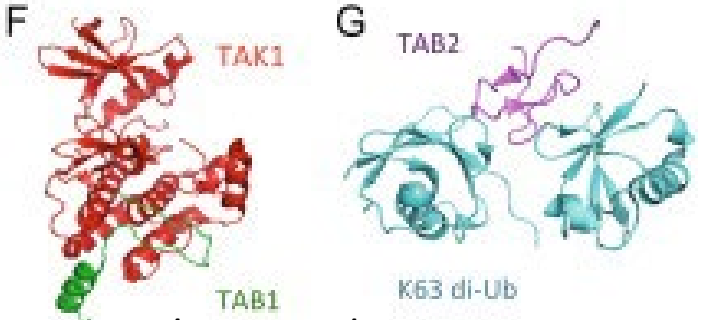
RING domains dimerize while the coiled coil trimerizes





Linking death domains to TRAF2

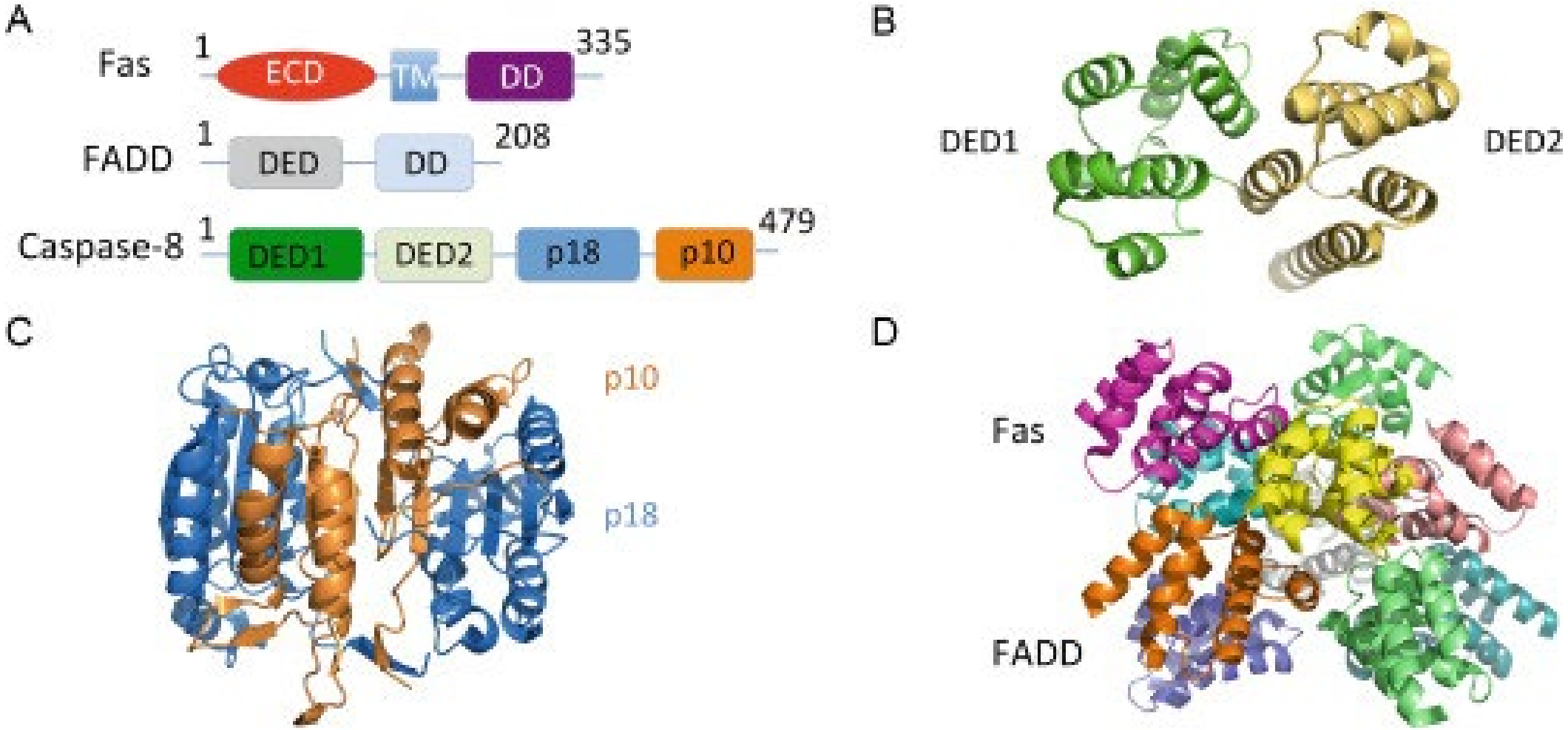




TAB2 interacting with K63-linked di-Ub complex

http://dx.doi.org/10.1016/B978-0-12-407707-2.00005-9

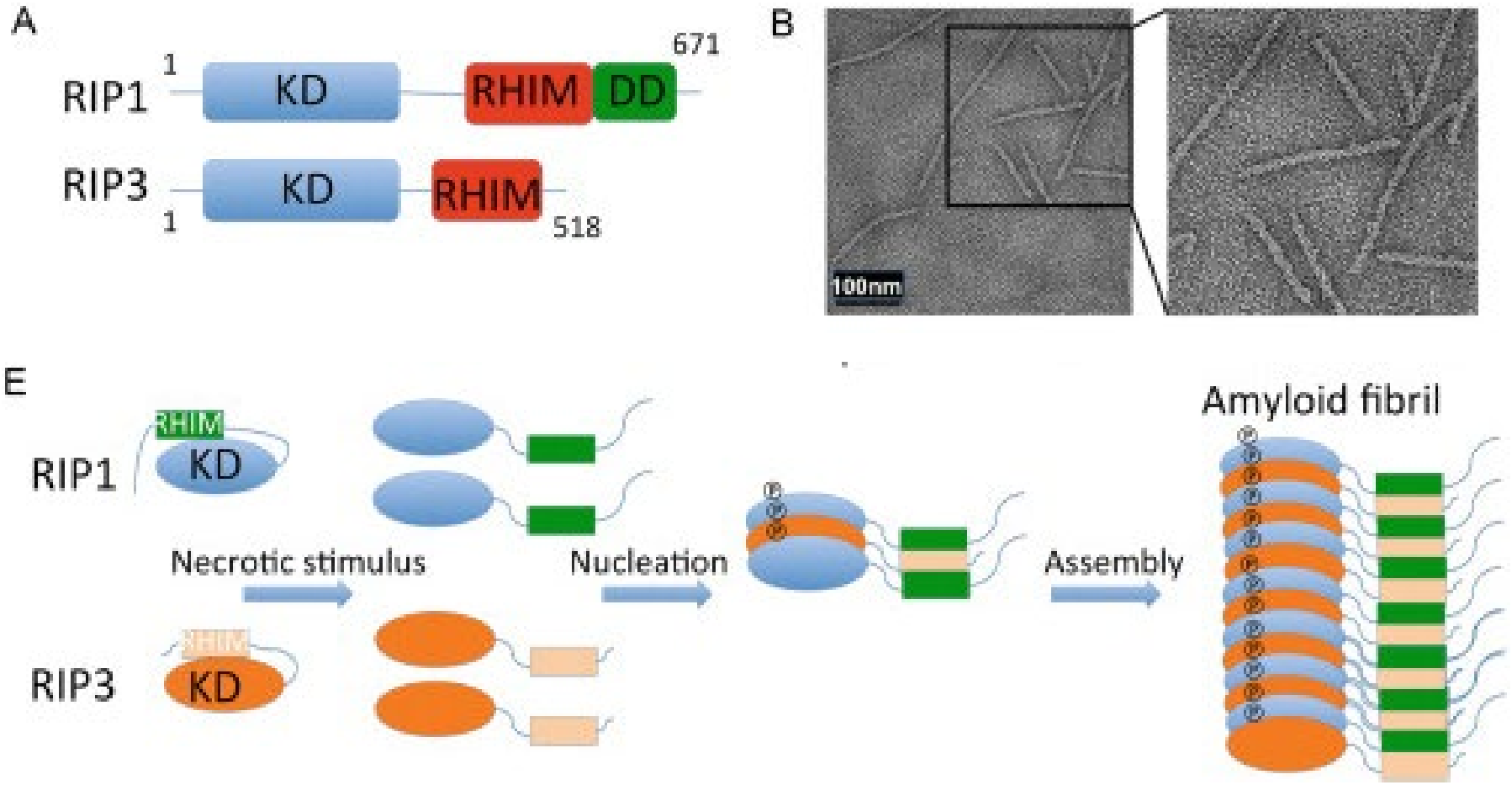
Death-induced signaling complex



Catalytic domain structure of caspase-8

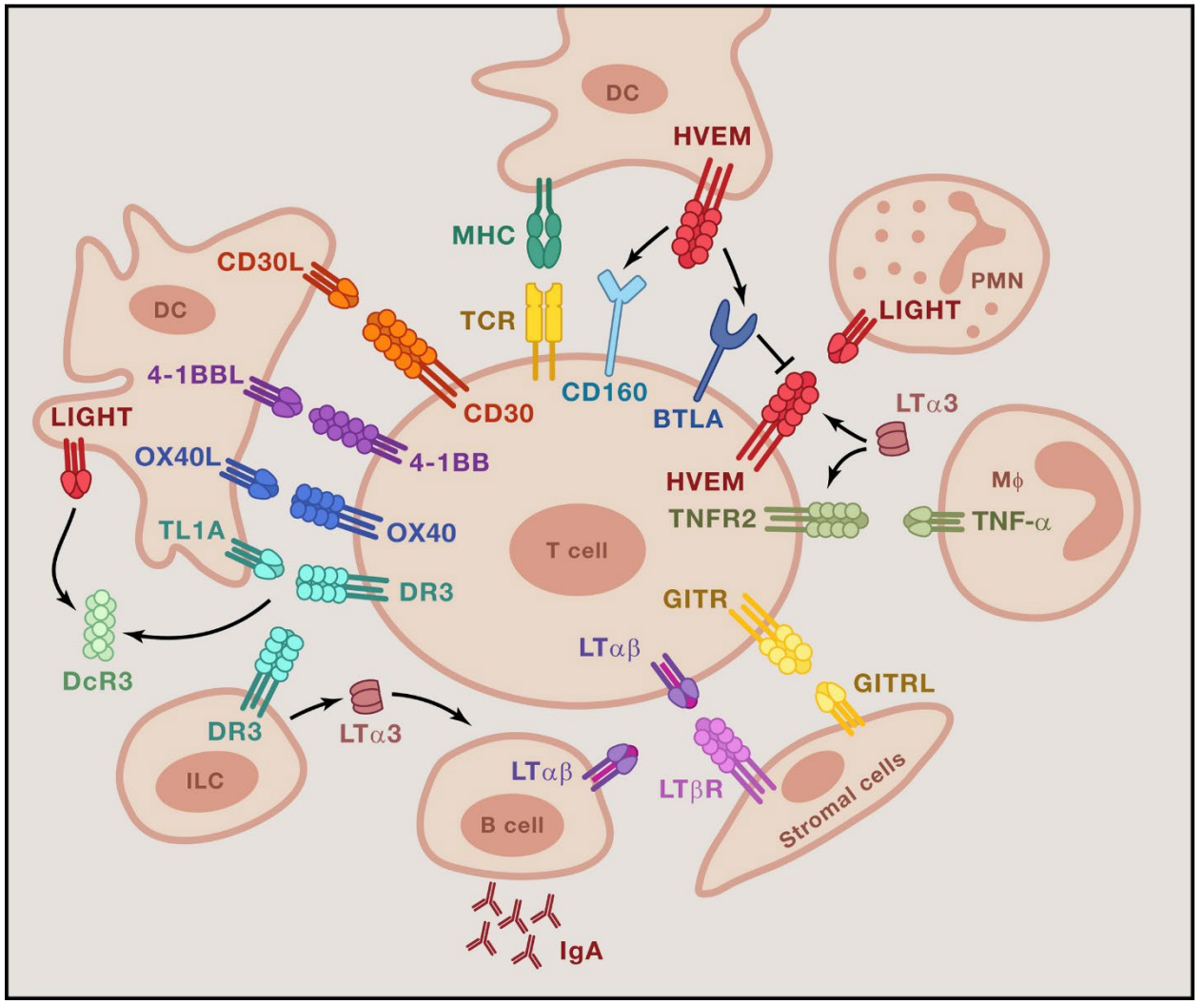
http://dx.doi.org/10.1016/B978-0-12-407707-2.00005-9

RIP1/3 programmed necrosis - “necroptosis”



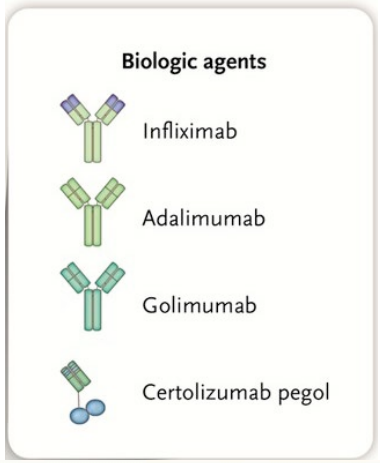
http://dx.doi.org/10.1016/B978-0-12-407707-2.00005-9

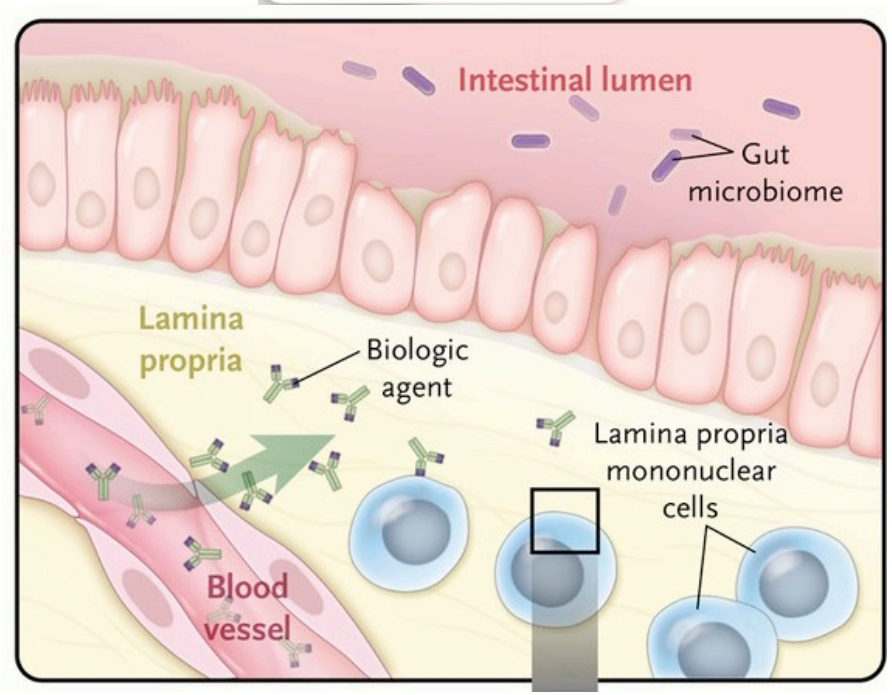
Signaling networks in immunity

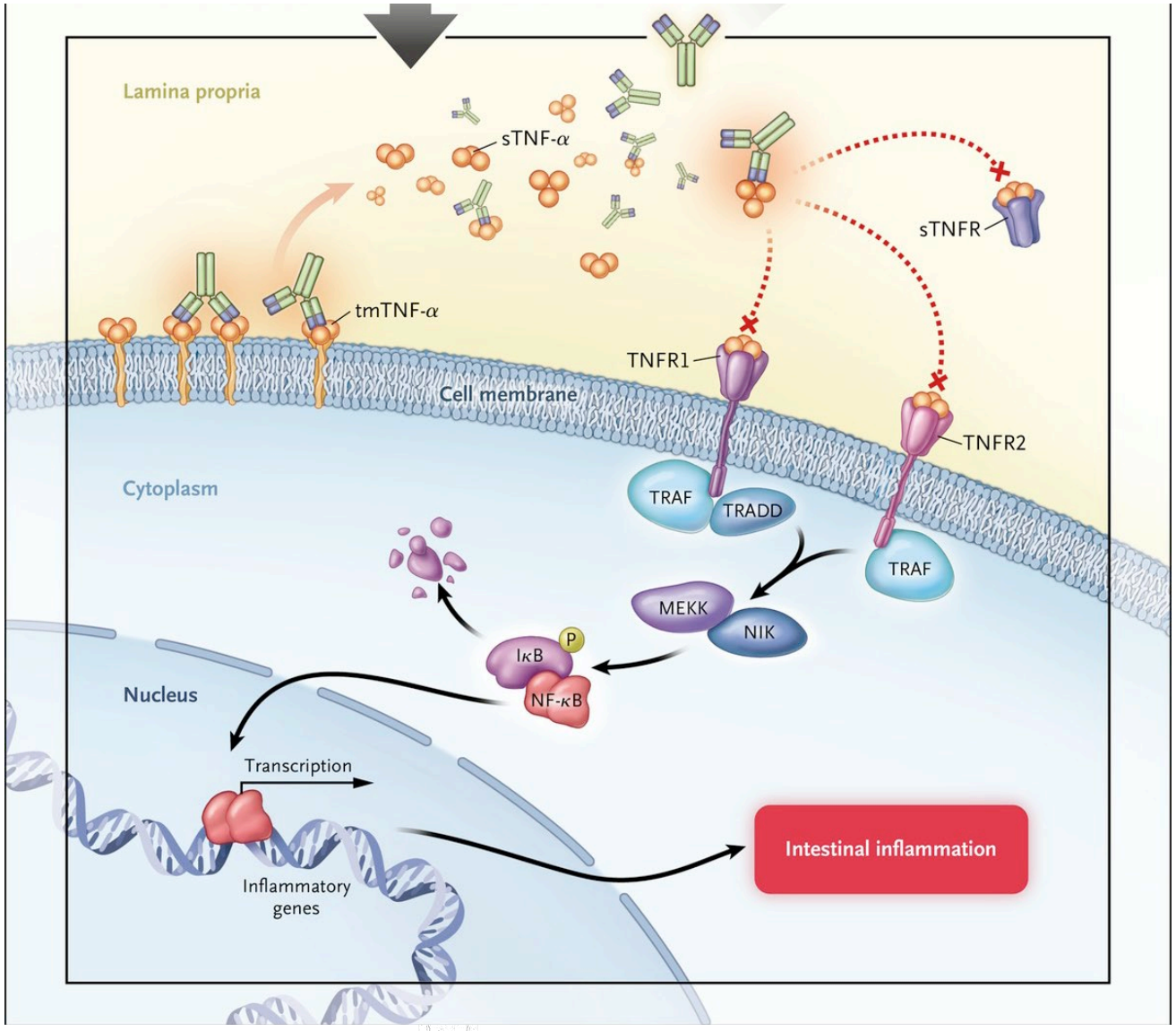


Immunity 2016 44, 1005-1019DOI: (10.1016/j.immuni.2016.04.019)

Anti–Tumor Necrosis Factor α (TNF-α) Antibodies in Inflammatory Bowel Disease.

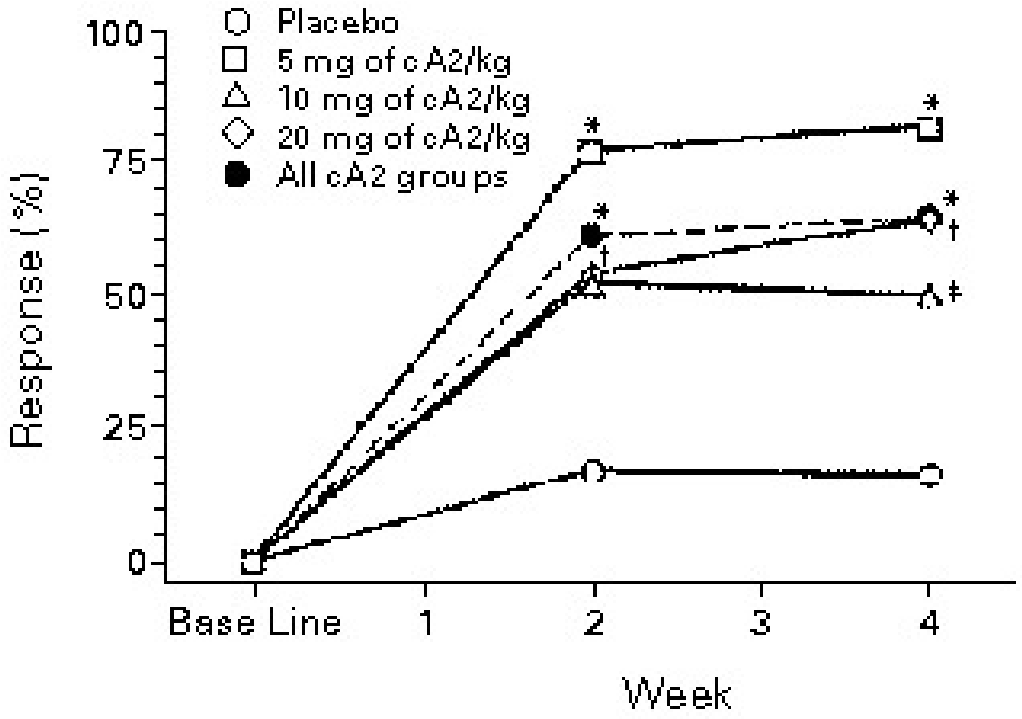


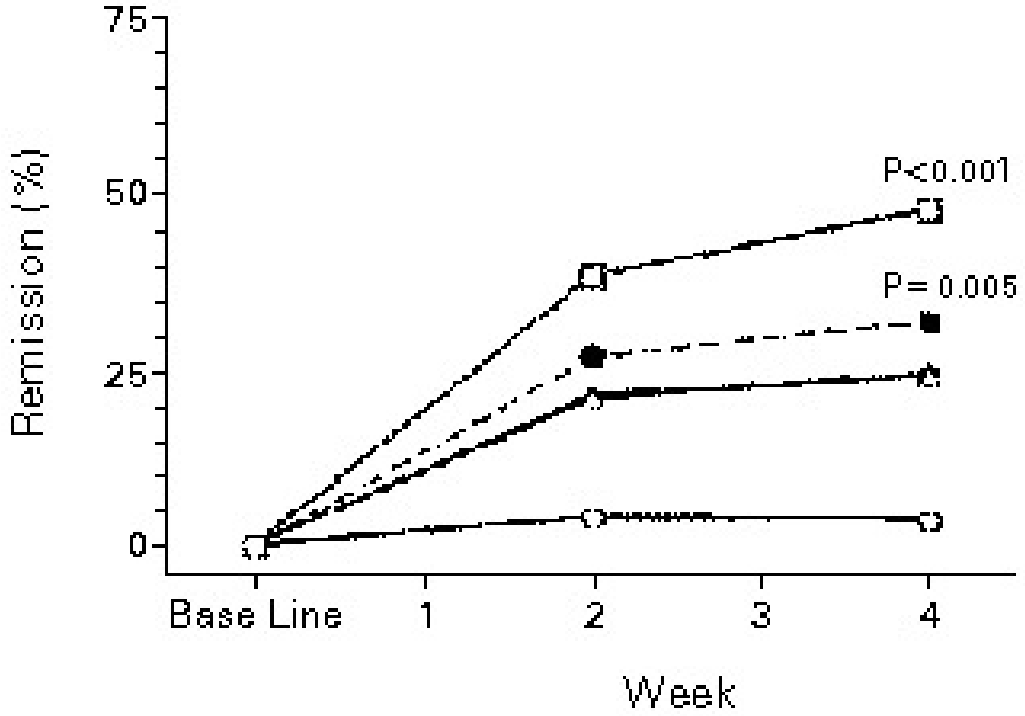




Nielsen OH, Ainsworth MA. N Engl J Med 2013;369:754-762.

Rates of Clinical Response and Remission after a Single Infusion of cA2 or Placebo.





No. of Patients Evaluated			
Placebo	25	24	24
5 mg of cA2/kg	27	26	27
10 mg of cA2/kg	28	23	28
20 mg of cA2/kg	28	28	28
All cA2 groups	83	77	83

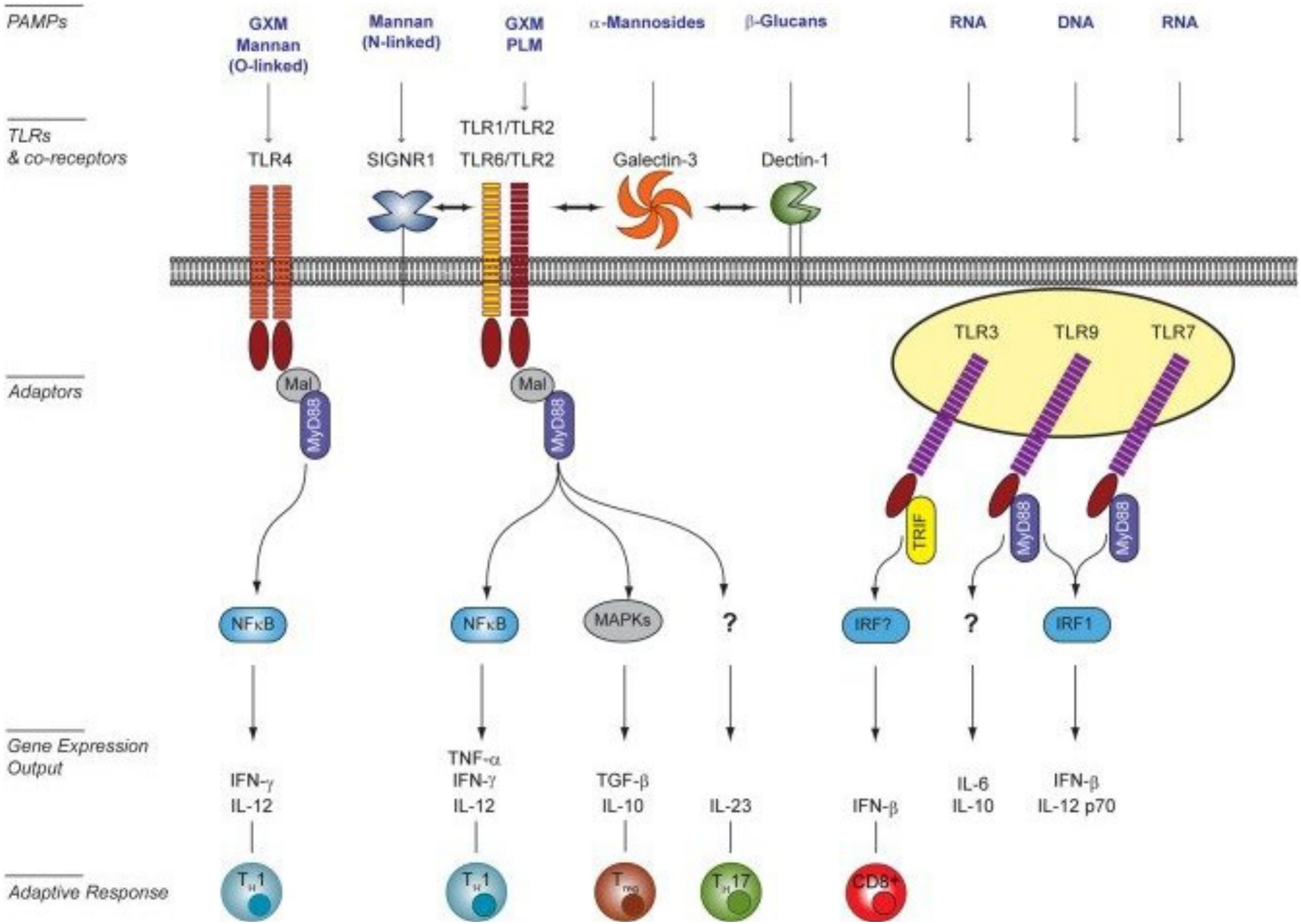
Targan SR et al. N Engl J Med 1997;337:1029-1036.

Pattern recognition receptors

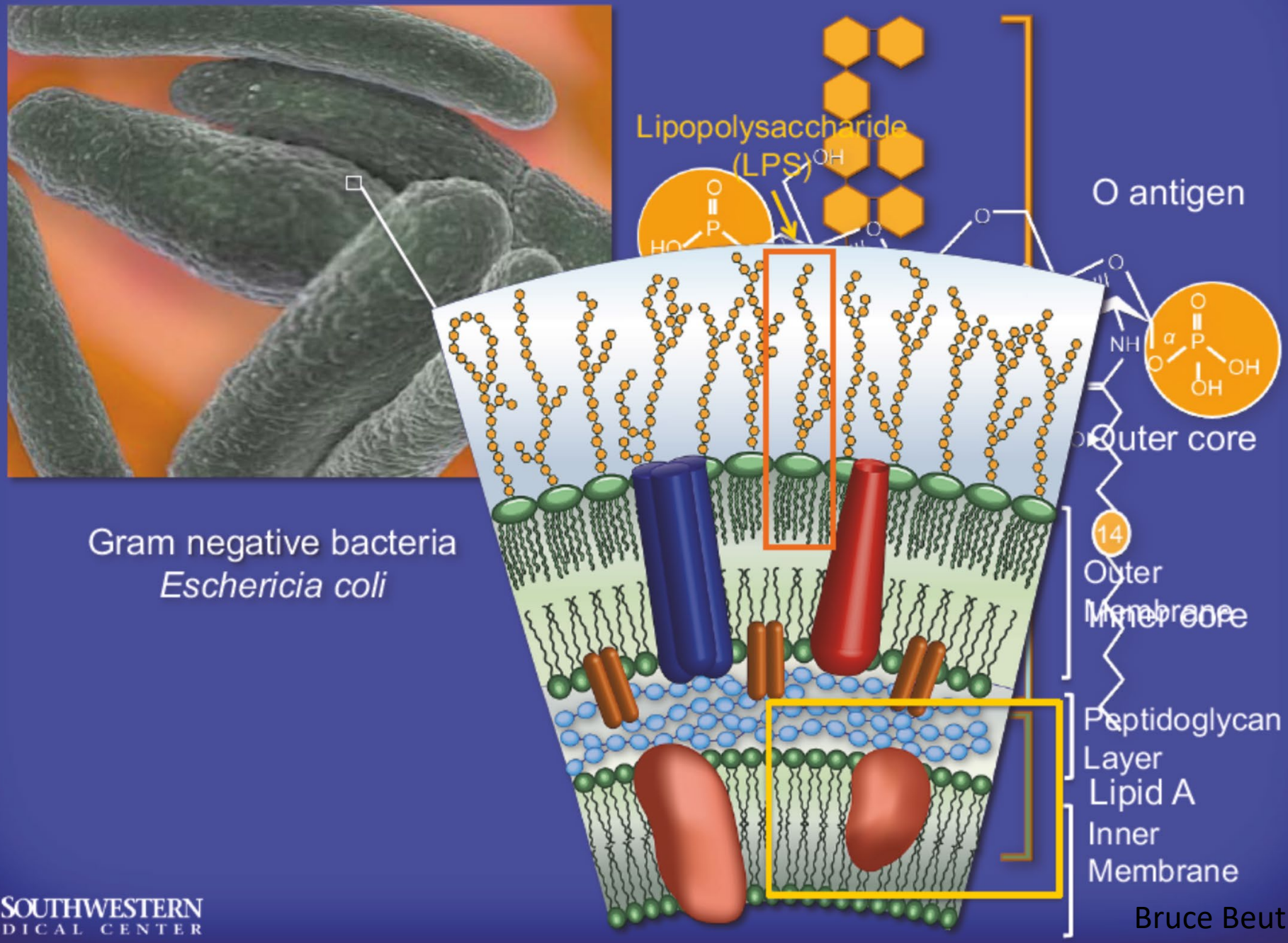
Toll-like receptors

NOD-like receptors

RIG-I-like receptors

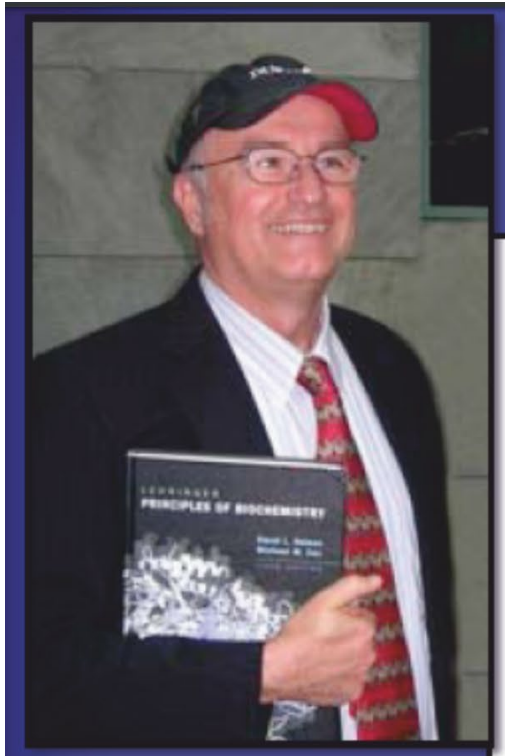


Frontiers in Cellular and Infection Microbiology 2(142):142

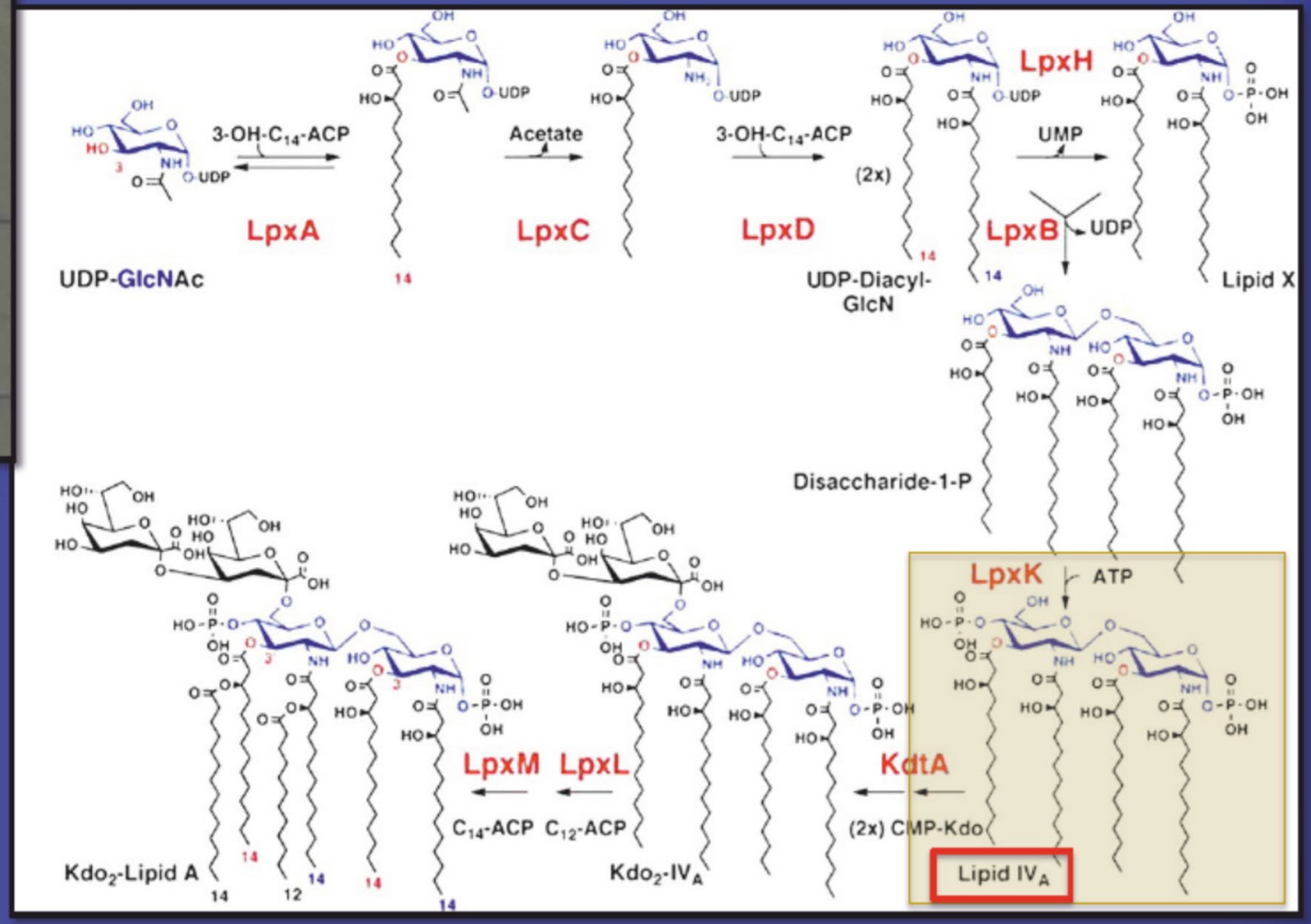


Bruce Beutler – Nobel Lecture

Biosynthesis of Lipid A



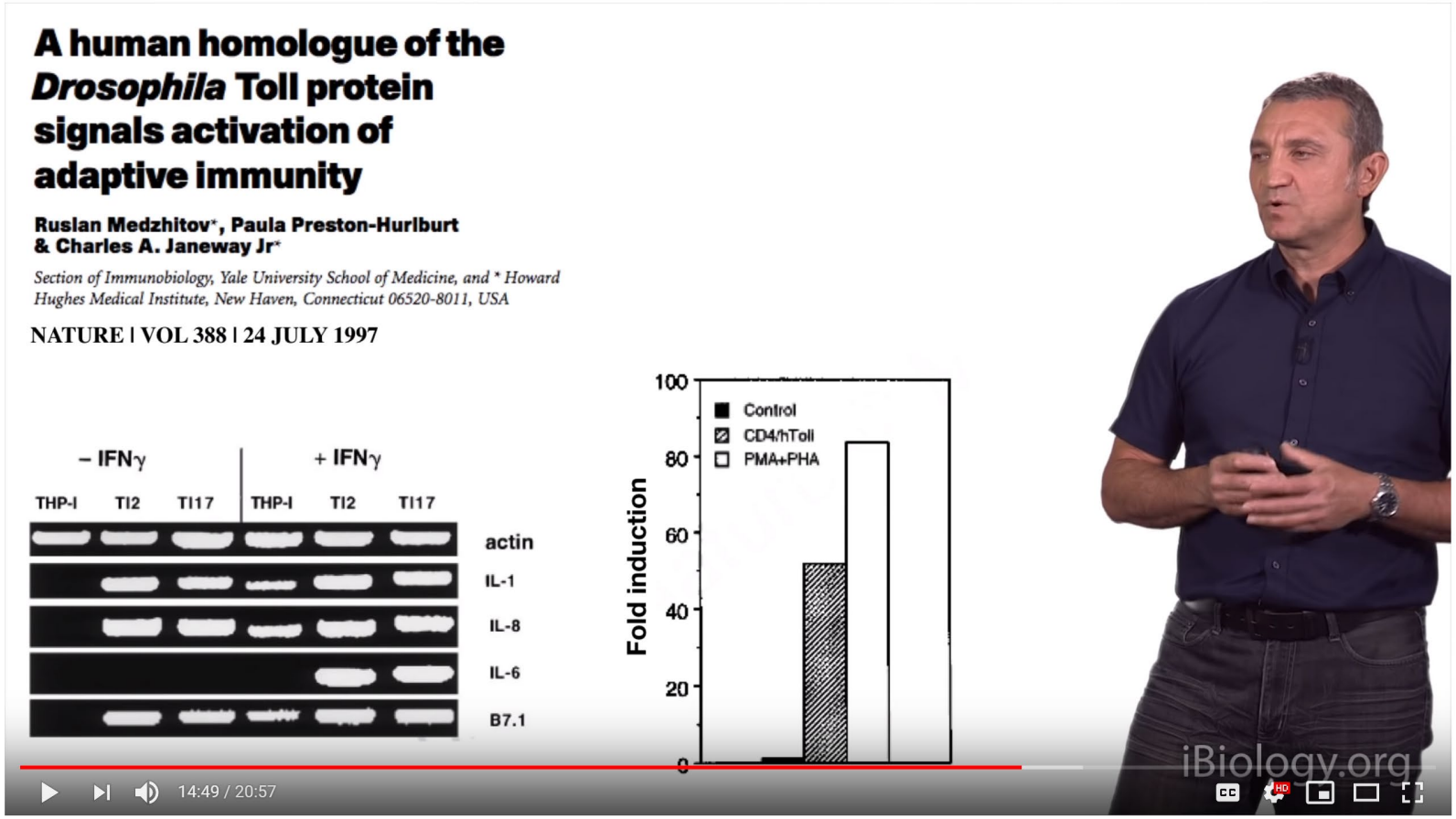
Christian R.H. Raetz
1946 - 2011



Discover of TLRs

YouTube

ruslan medzhitov



Ruslan Medzhitov (Yale / HHMI): The Role of Toll-Like Receptors in the Control of Adaptive Immunity

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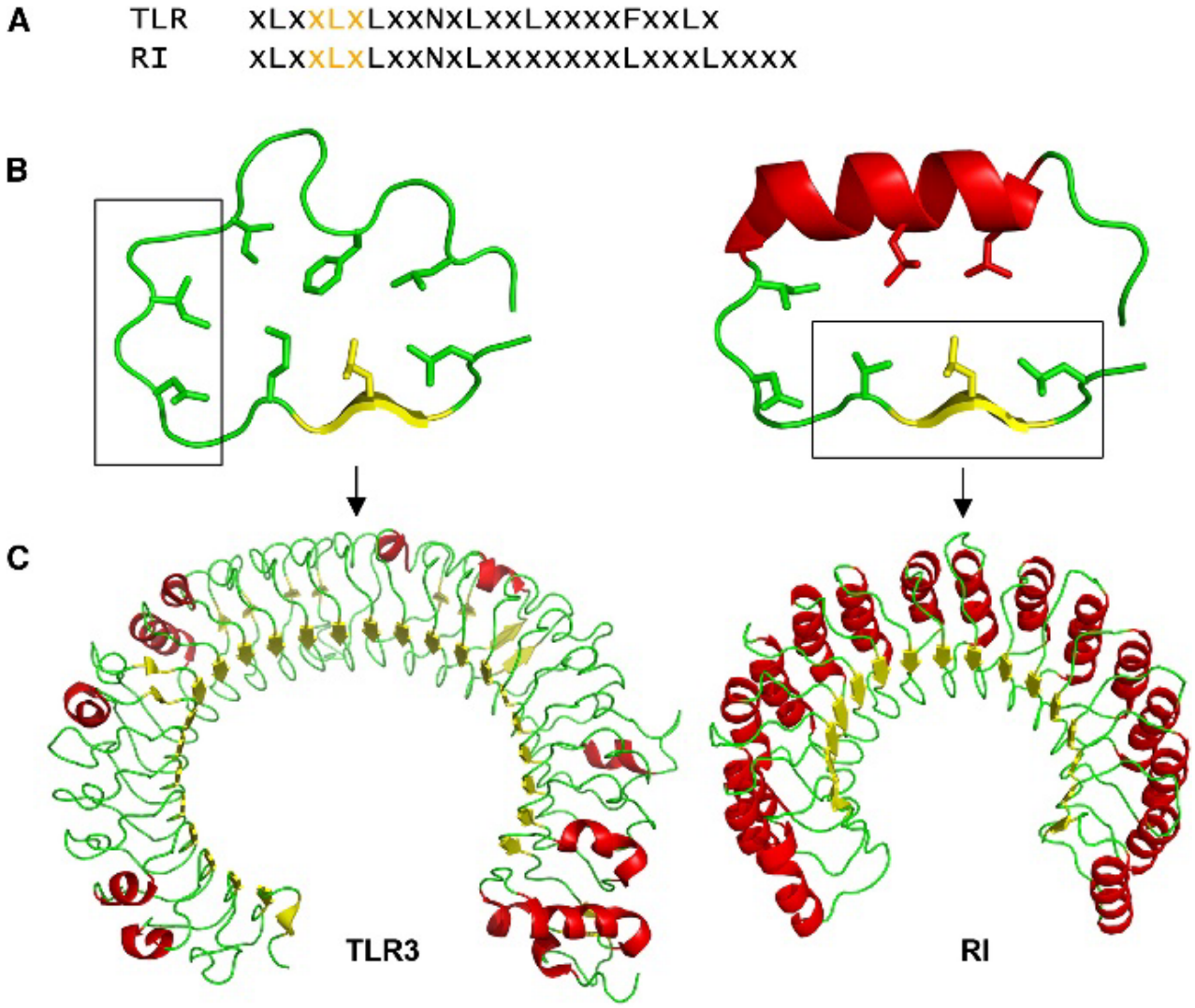
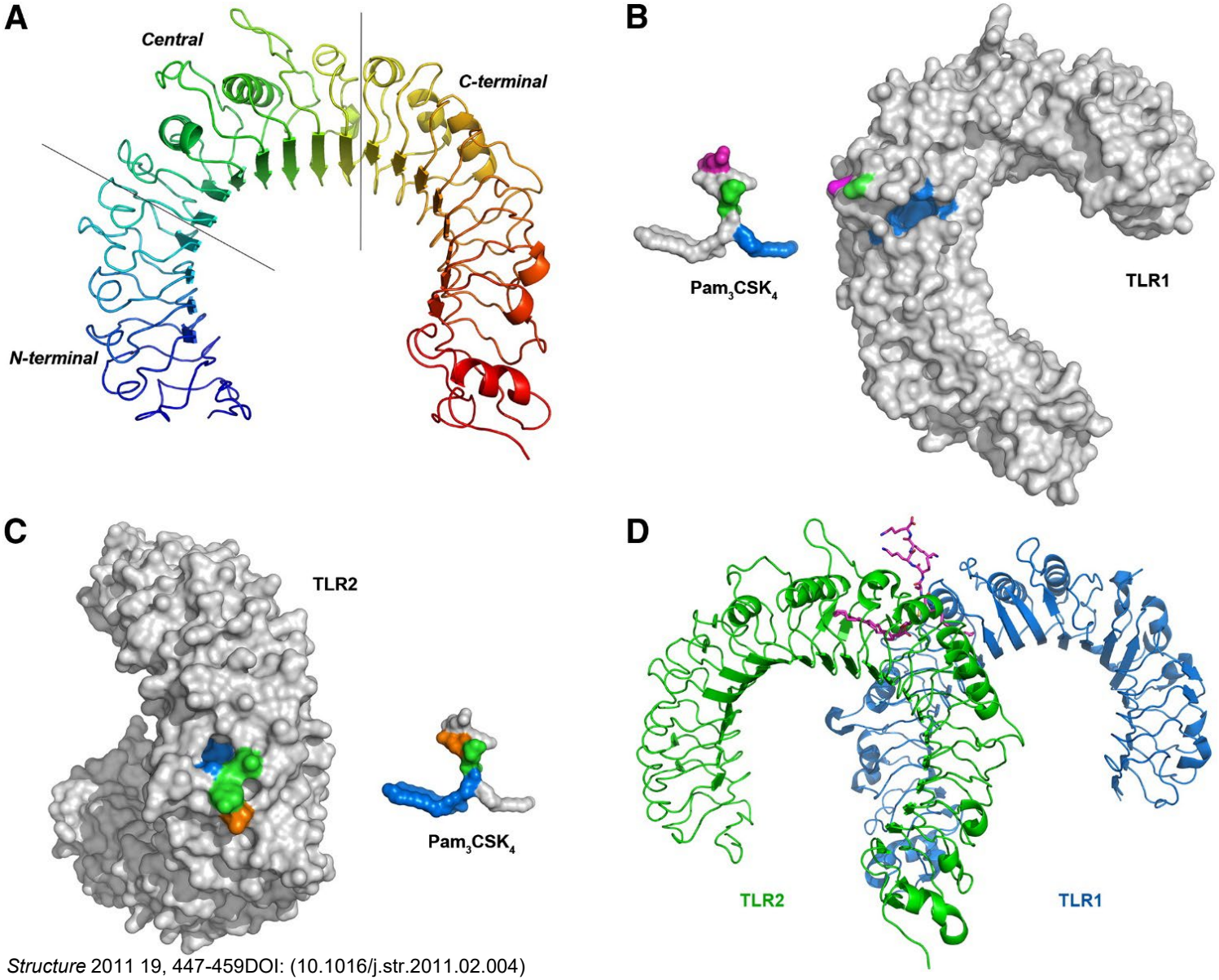
Structure of Leucine-Rich Repeats in the ECD

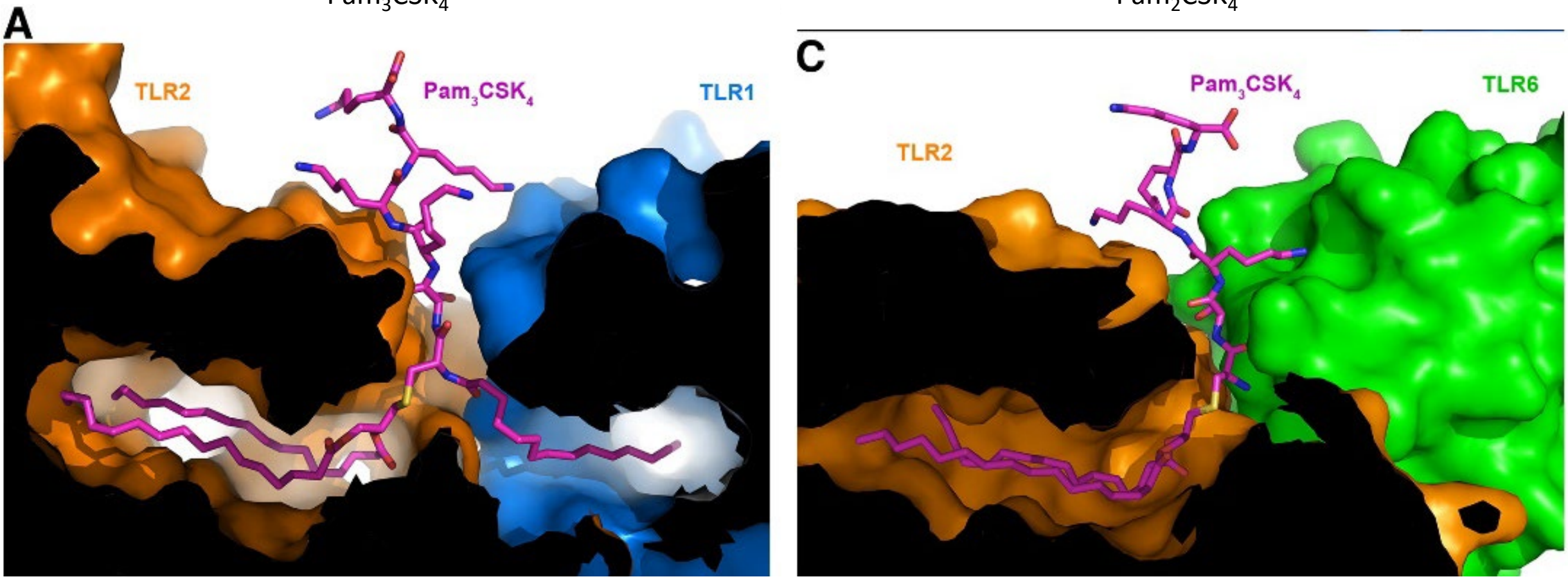


Structure 2011 19, 447-459DOI: (10.1016/j.str.2011.02.004)

Ligand-binding promotes dimerization

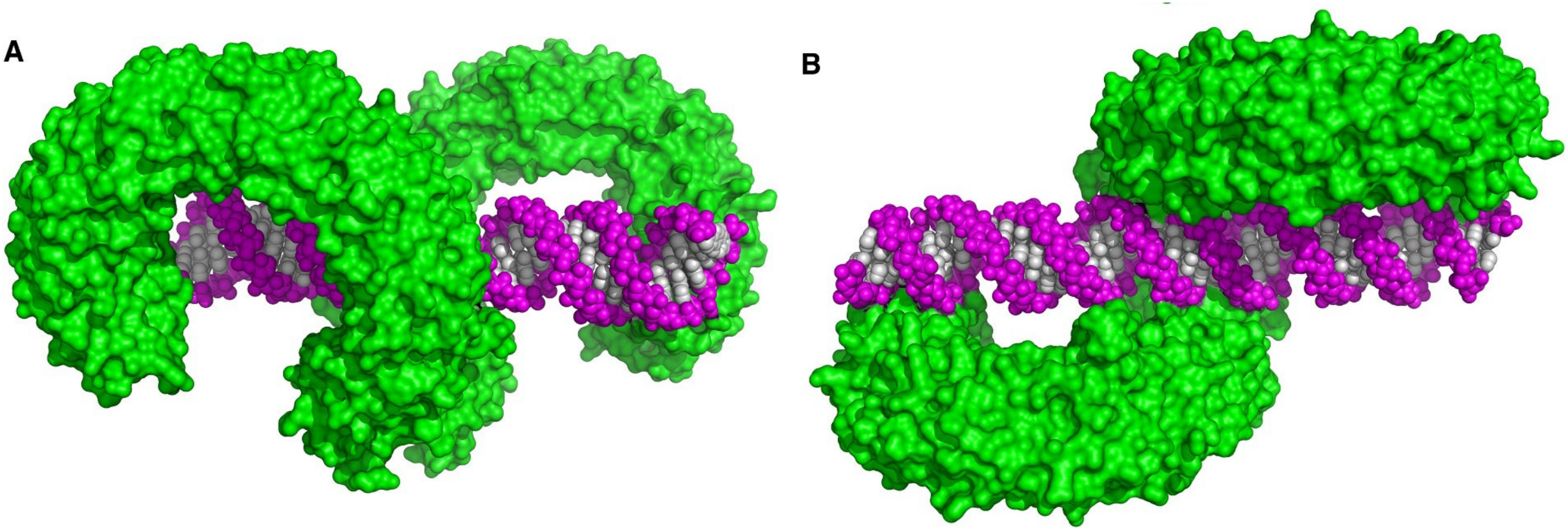


Structure 2011 19, 447-459DOI: (10.1016/j.str.2011.02.004)



Structure 2011 19, 447-459DOI: (10.1016/j.str.2011.02.004)

TLR3/dsRNA complex



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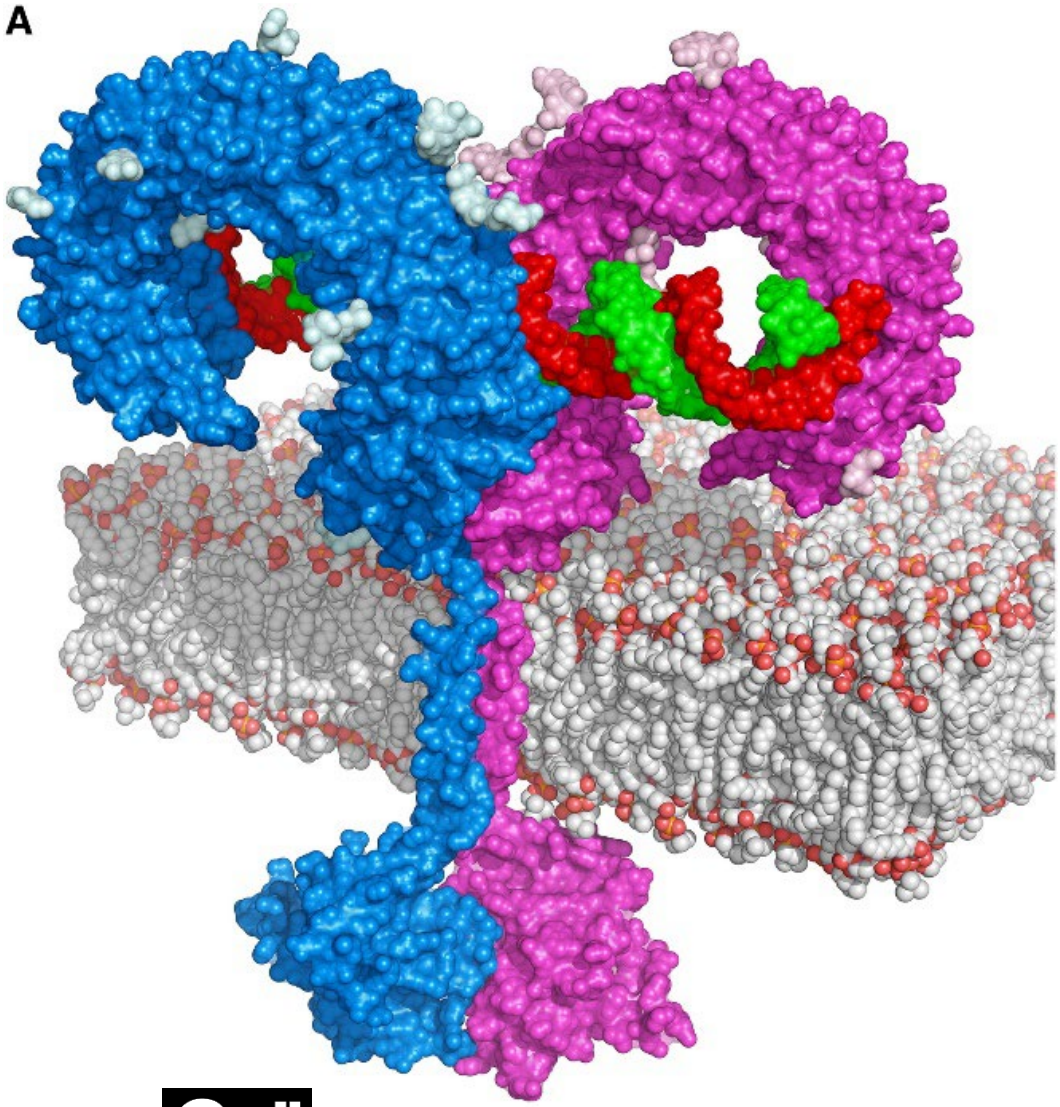
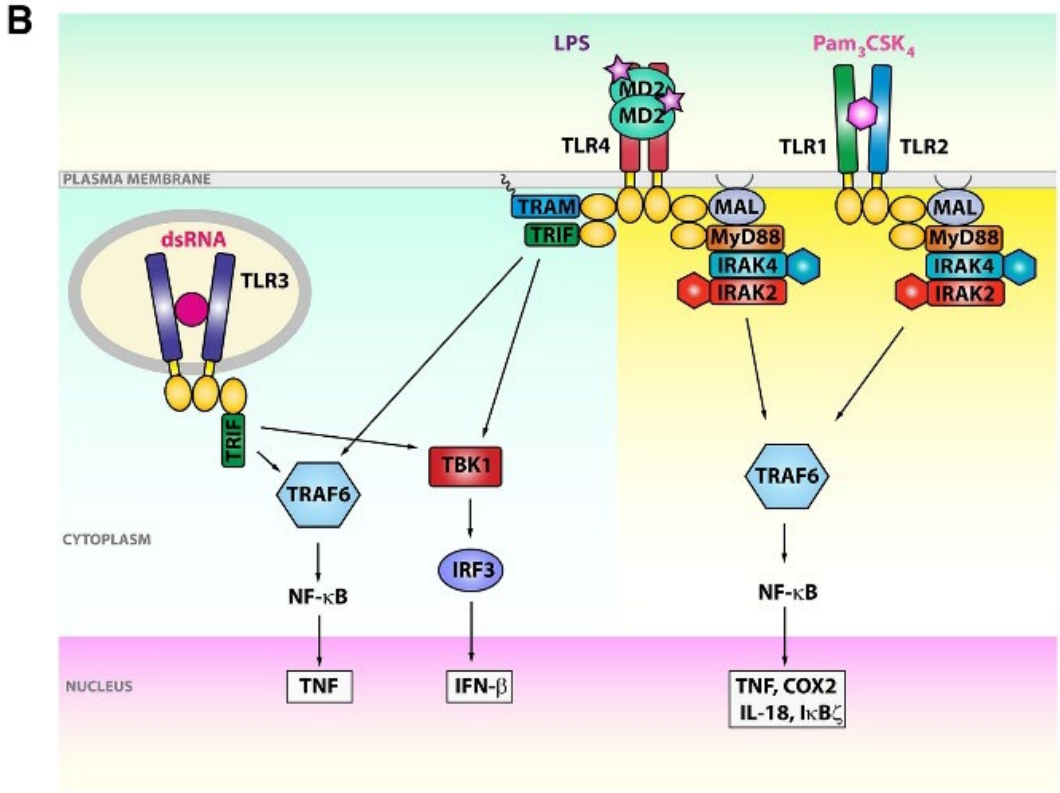
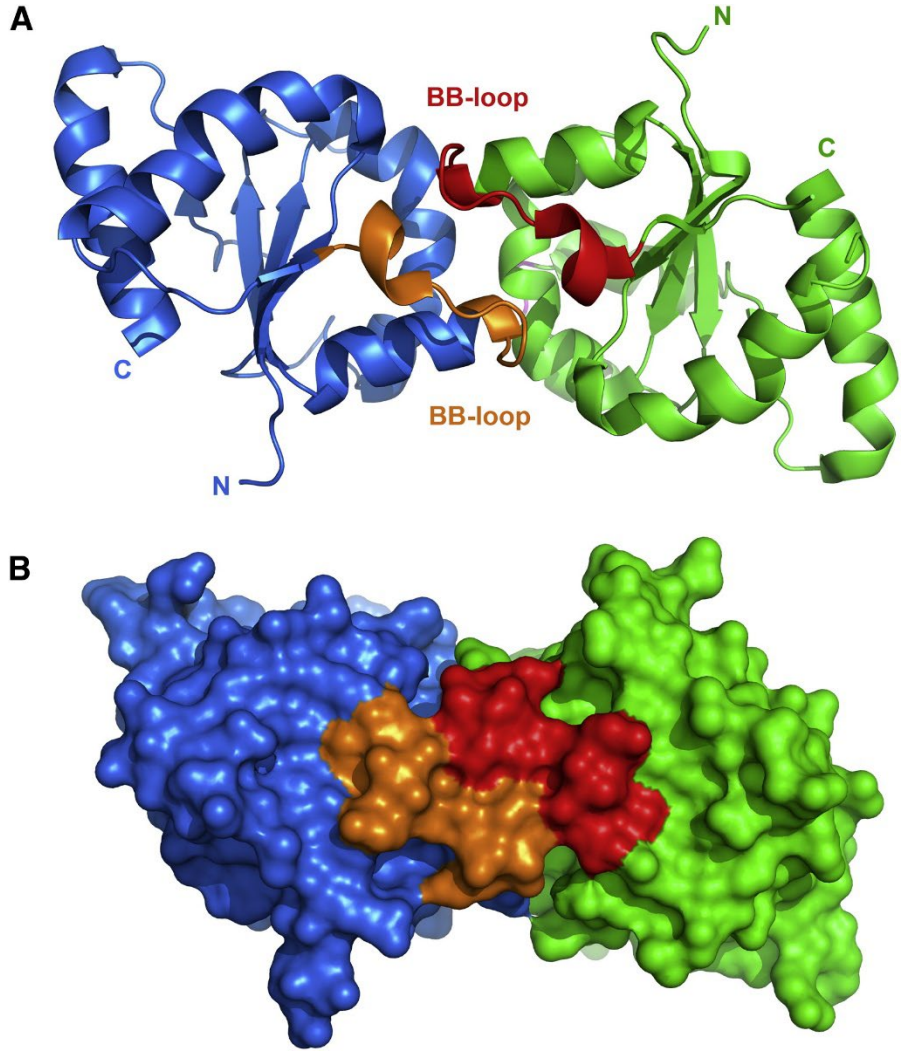




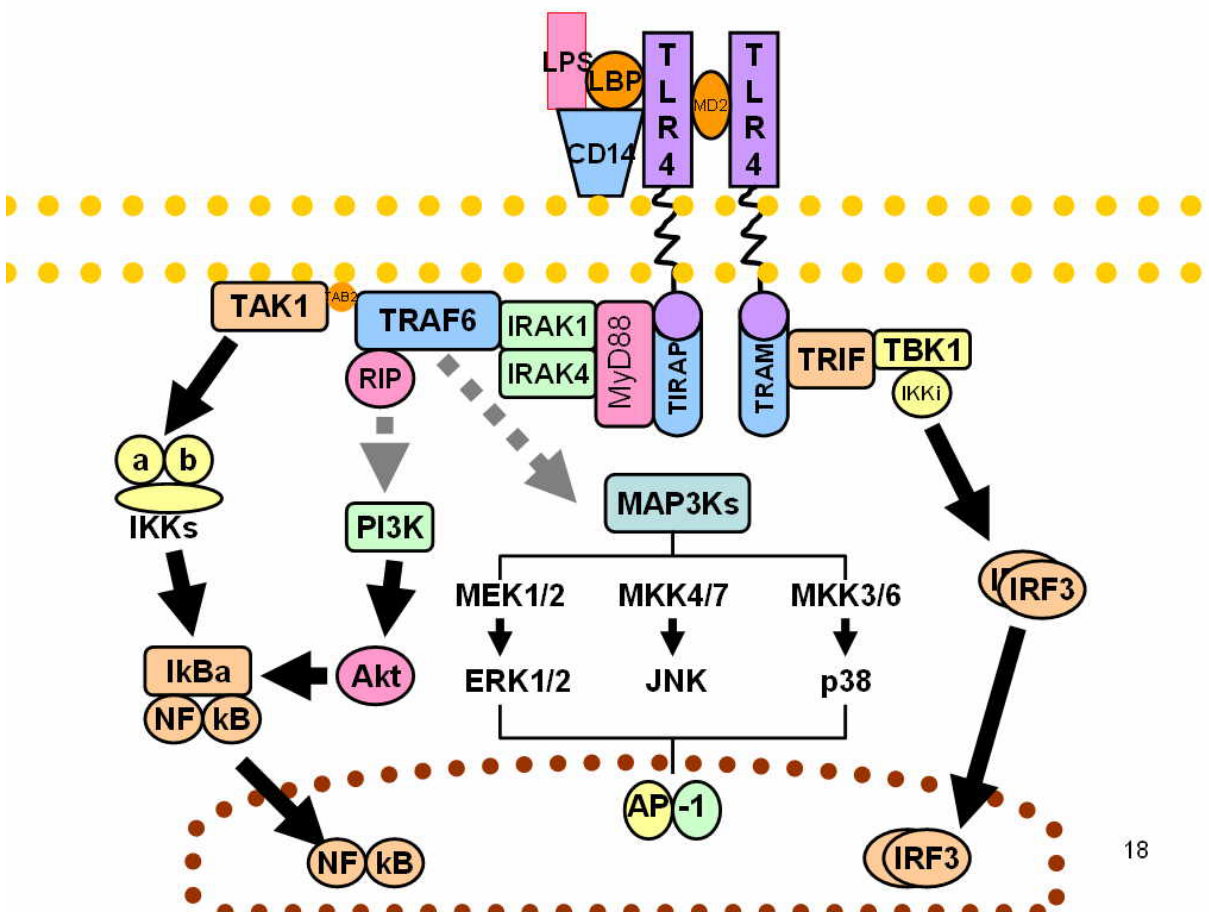
Structure 2011 19, 447-459DOI: (10.1016/j.str.2011.02.004)

Toll IL-1 receptor (TIR) domains heterodimerize with effector TIR domains



Structure 2011 19, 447-459DOI: (10.1016/j.str.2011.02.004)

MyD88



Wikipedia

Development of TLR agonists as drugs

Application of TLR Immunomodulator	Examples	Mechanism	Potential Therapeutic Outcome	Potential Adverse Consequences
Resistance to infection	TLR2 agonist Pam_2Cys, TLR4 agonists MPLA & PHAD, TLR3 agonist poly I:C, TLR9 agonist CpG	Increased leukocyte recruitment and antimicrobial functions	Improved survival; reduced risk of nosocomial infections; reduced reliance on antibiotics	Chronic inflammation; autoimmune disease
Vaccine adjuvant	TLR4 agonist MPLA as an approved adjuvant in malaria (AS01), human papillomavirus (HPV), and hepatitis B (AS04) vaccines	Immune stimulation for increased antibody titers	Improved efficacy of vaccines and reduced dosing strategies	Discomfort at injection site; transient malaise
Cancer immunotherapy	TLR3 agonist poly I:C & derivatives; TLR7 agonist 1V270	T-cell activation and DC maturation	Antitumor immunity	Dose-limiting side effects (fatigue, malaise, fever)
Chronic infections & inflammatory diseases	TLR4 antagonist Eritoran to treat sepsis; TLR9 agonist Lefitolimod for reduction of HIV-1 viral reservoir	Antagonize TLR to prevent activation and downstream inflammation	Reduced inflammation and associated organ injury	Immune tolerance

Front. Immunol., 18 February 2021 | https://doi.org/10.3389/fimmu.2020.622614

Nuclear receptor superfamily

Steroid hormone receptors

Thyroid receptor

Dr. Charles Huggins

INOVA
Schar Cancer Institute

Nobel Prize 1966

for “his discoveries concerning hormonal treatment of prostatic cancer"

- Huggins and Hodges first treated men with prostate cancer with either orchiectomy or estrogen
- Huggins and Bergenstal used adrenalectomy for the “immediate and persistent relief of crippling bone pain”

1. Huggins C, Hodges. J Urol. 2002 Jul; 168(1):9-12
2. Huggins C, Bergenstal DM. Proc Natl Acad Sci U S A. 1952 Jan; 38(1):73-6.

https://slideplayer.com/slide/17626307/

Dr. Charles Huggins

INOVA
Schar Cancer Institute

Nobel Prize 1966

for "his discoveries concerning hormonal treatment of prostatic cancer"

Studies on Prostatic Cancer

I. The Effect of Castration, of Estrogen and of Androgen Injection on Serum Phosphatases in Metastatic Carcinoma of the Prostate*

Charles Huggins, M.D., and Clarence V. Hodges, M.D.

(*From the Department of Surgery, the University of Chicago, Chicago, Illinois*)

(Received for publication March 22, 1941)

1. Huggins C, Hodges. J Urol. 2002 Jul; 168(1):9-12
2. Huggins C, Bergenstal DM. Proc Natl Acad Sci U S A. 1952 Jan; 38(1):73-6.

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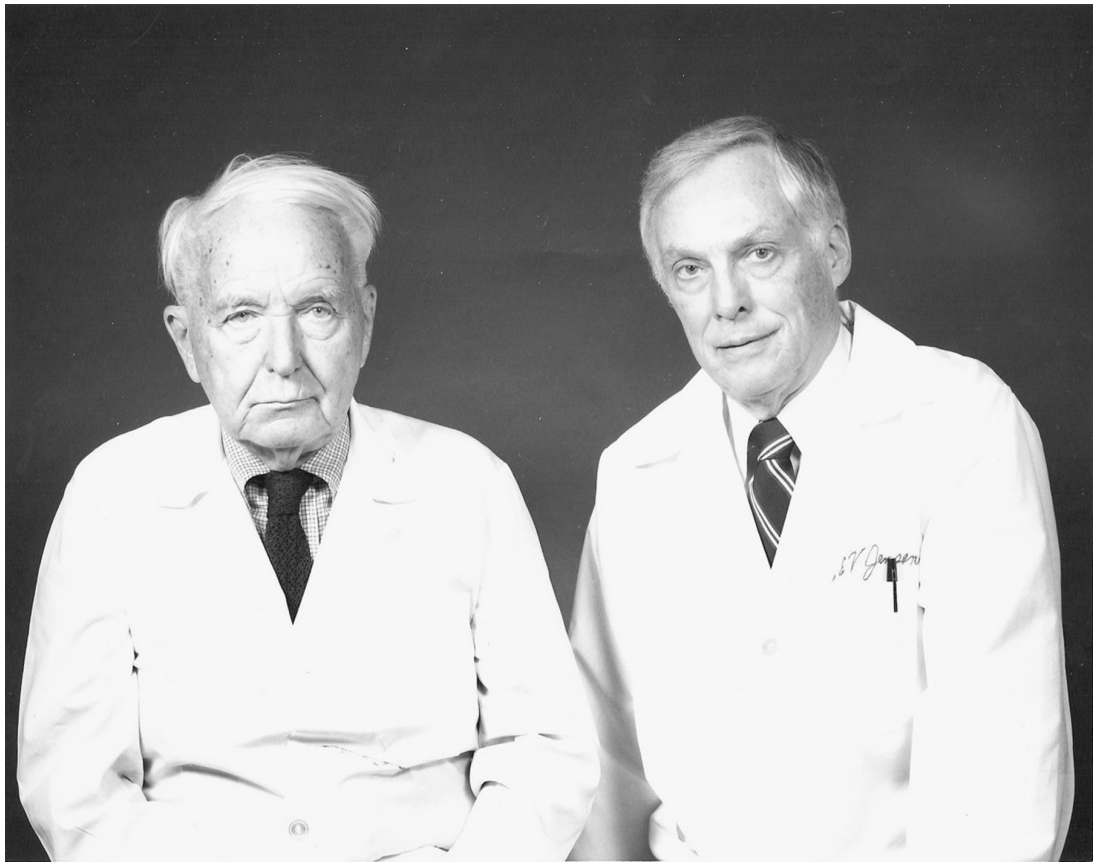
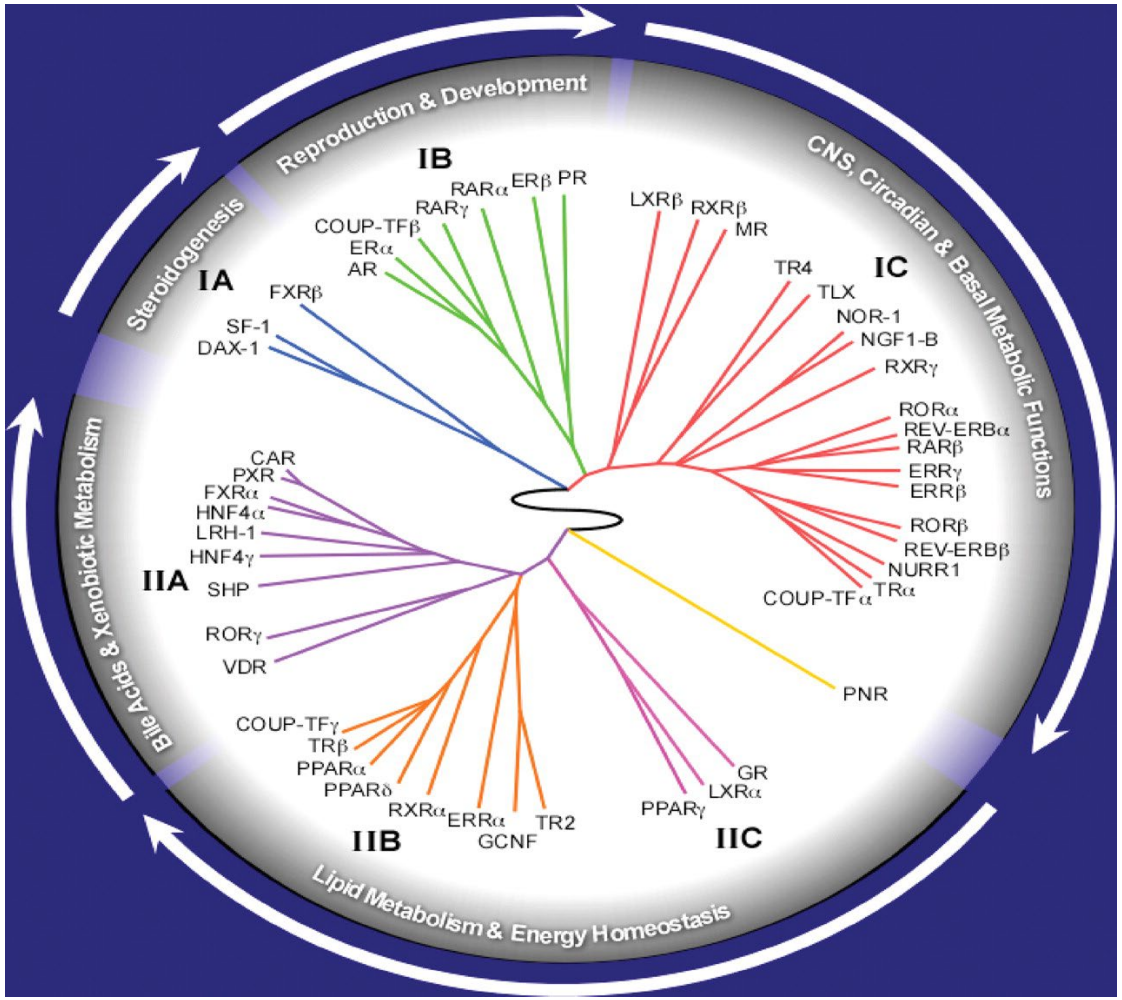
Discovery of the estrogen receptor



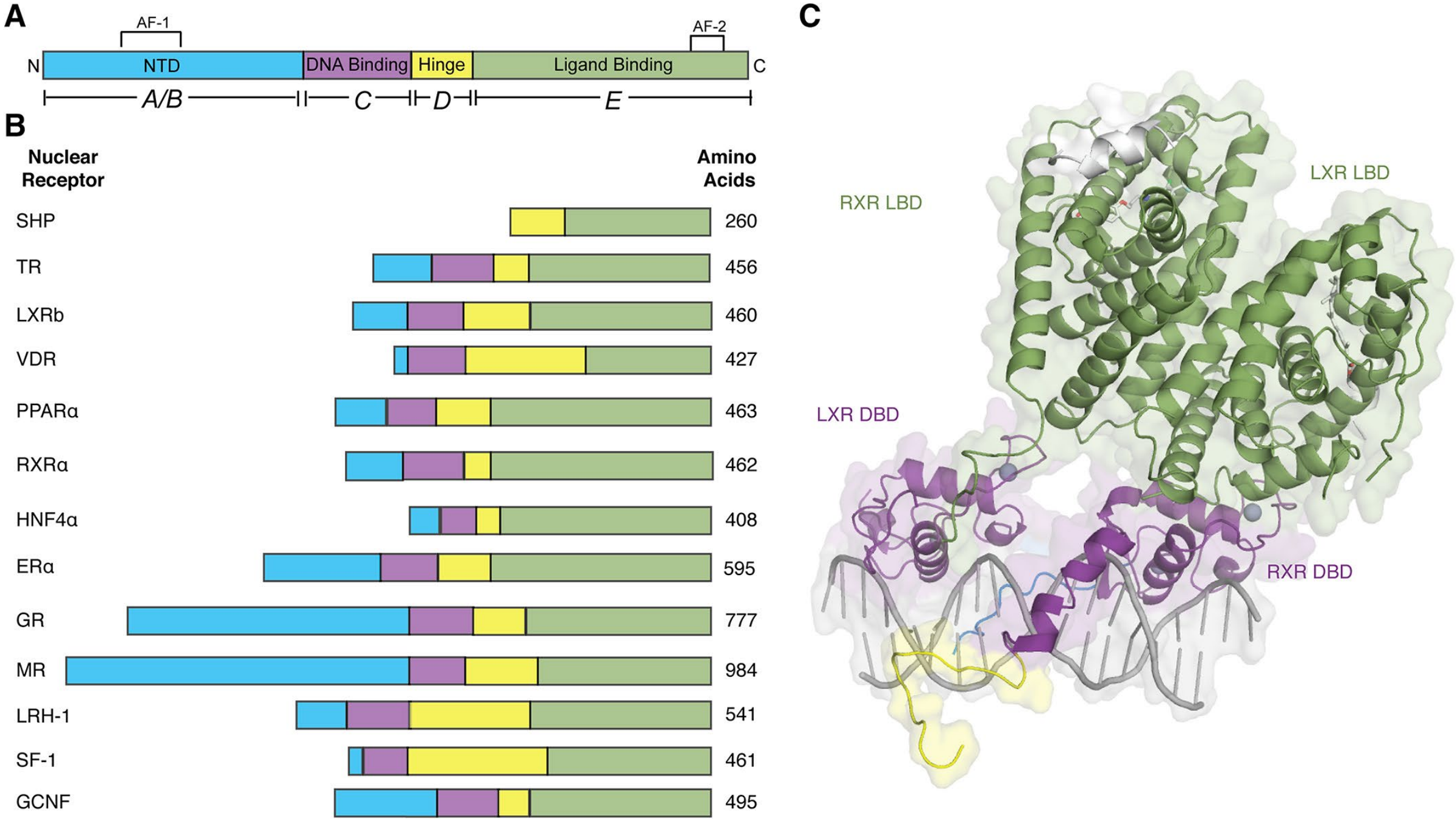
- Steroids were thought to act via enzyme activation, etc.
- 1950s (Jensen): radioactive estrogen to ovariectomized rats – localized to reproductive tissues. ER hypothesized.
- 1960s: O'Malley suggested nuclear concept based on mRNA induction by estrogen
- 1985 – Ron Evans cloned the glucocorticoid receptor

https://doi.org/10.1073/pnas.1301566110

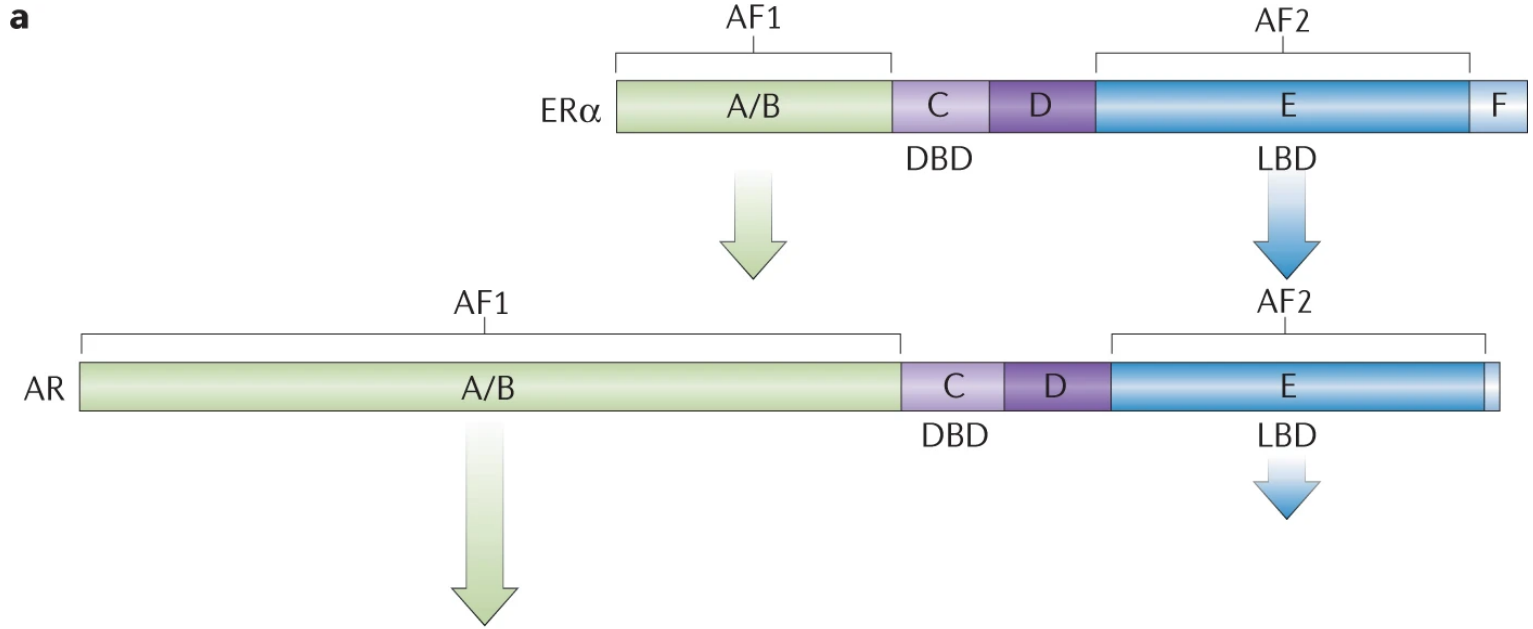
Nuclear receptor physiology

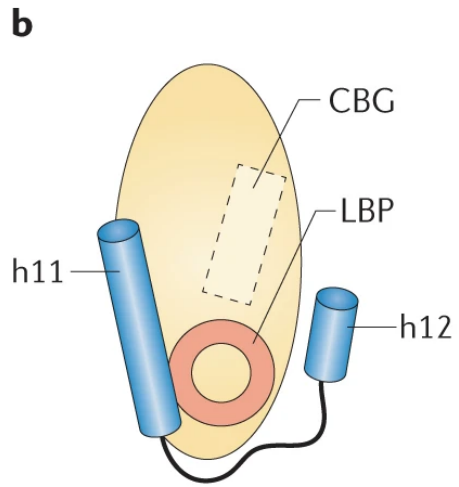


Mol Endocrinol, Volume 23, Issue 6, 1 June 2009, Pages 740–746, https://doi.org/10.1210/me.2009-0135



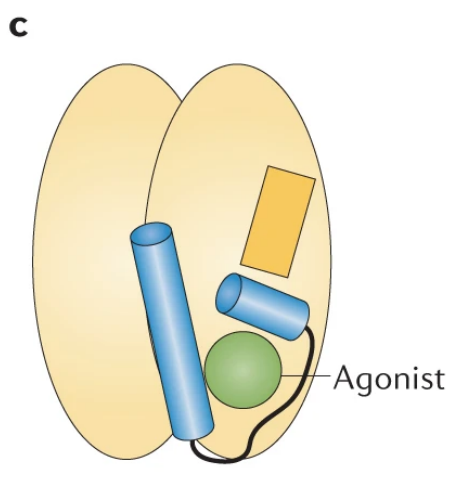
Protein Science, Volume: 27, Issue: 11, Pages: 1876-1892, First published: 15 August 2018, DOI: (10.1002/pro.3496)





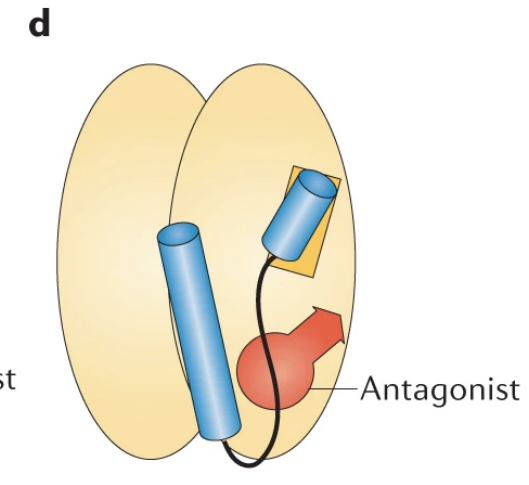
Apo-receptor

- No ligand
- h12 disordered
- Bound to HSP
- Monomer



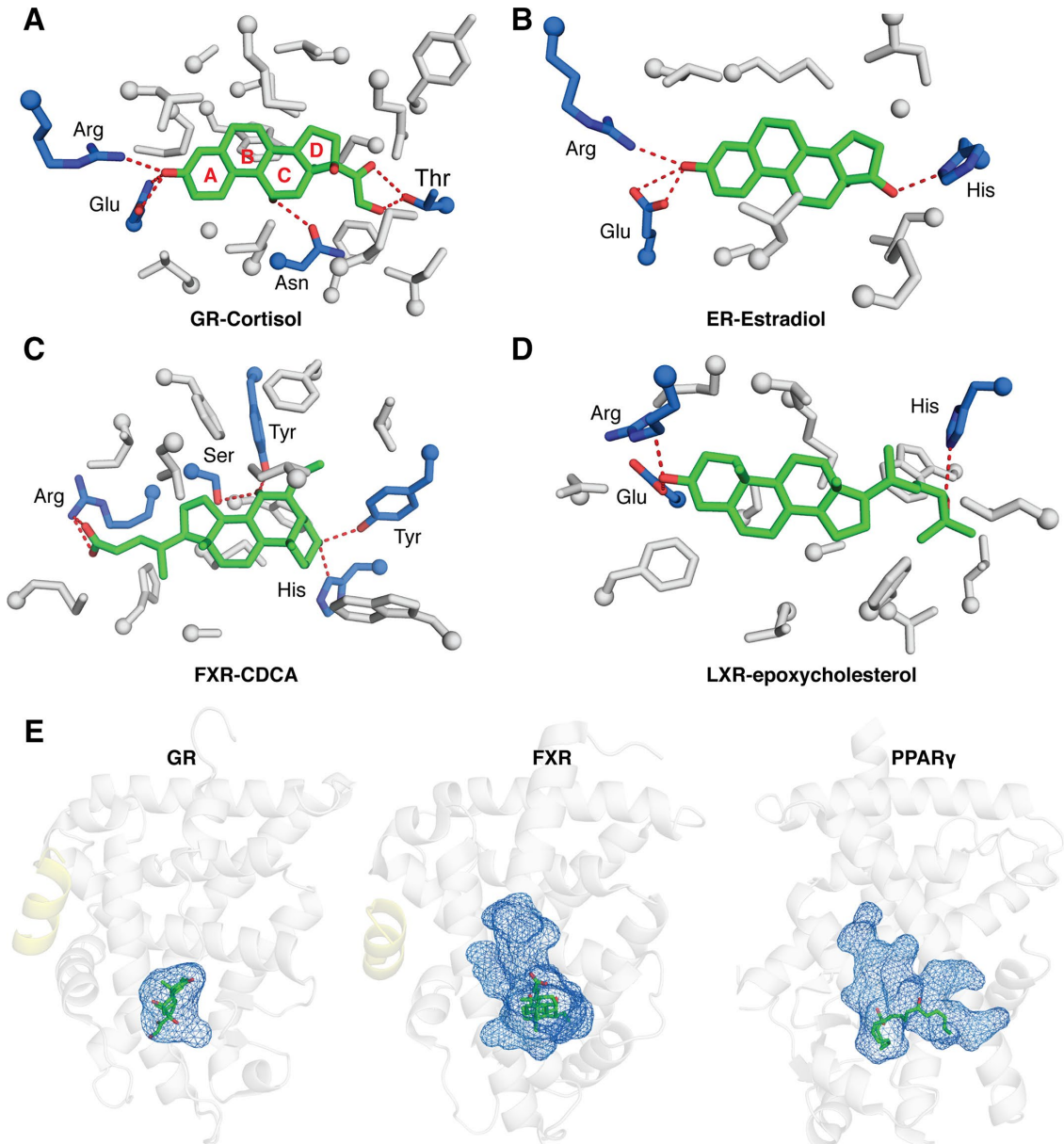
Agonist bound

- h12 stabilized
- CBG formed
- Dimer



Antagonist bound

- h12 shifted to block groove
- Dimer



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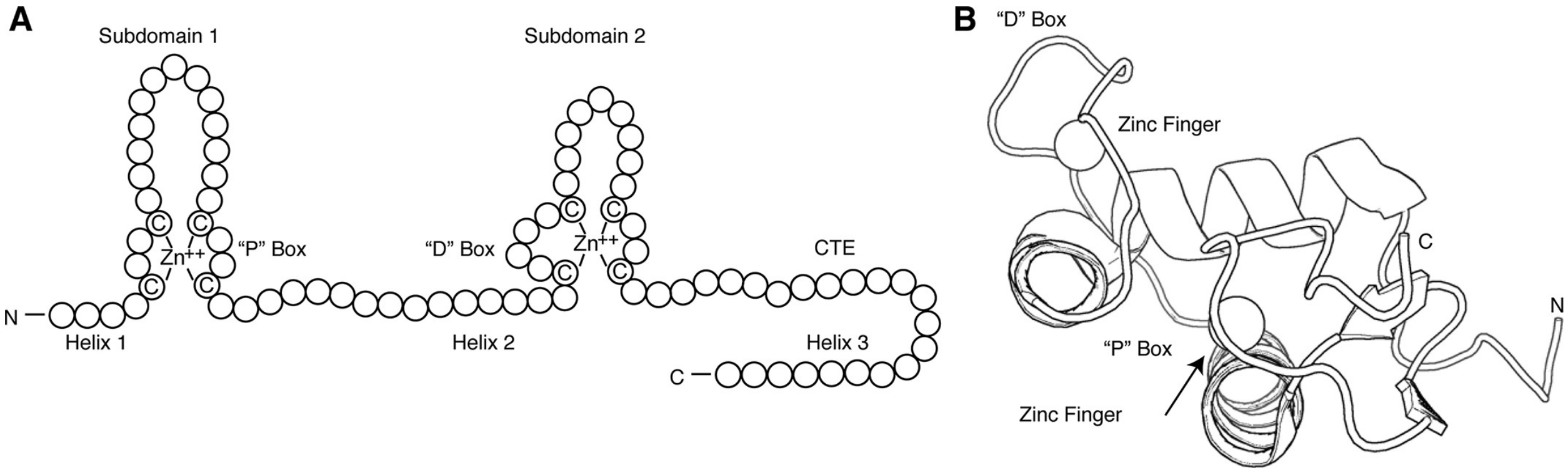

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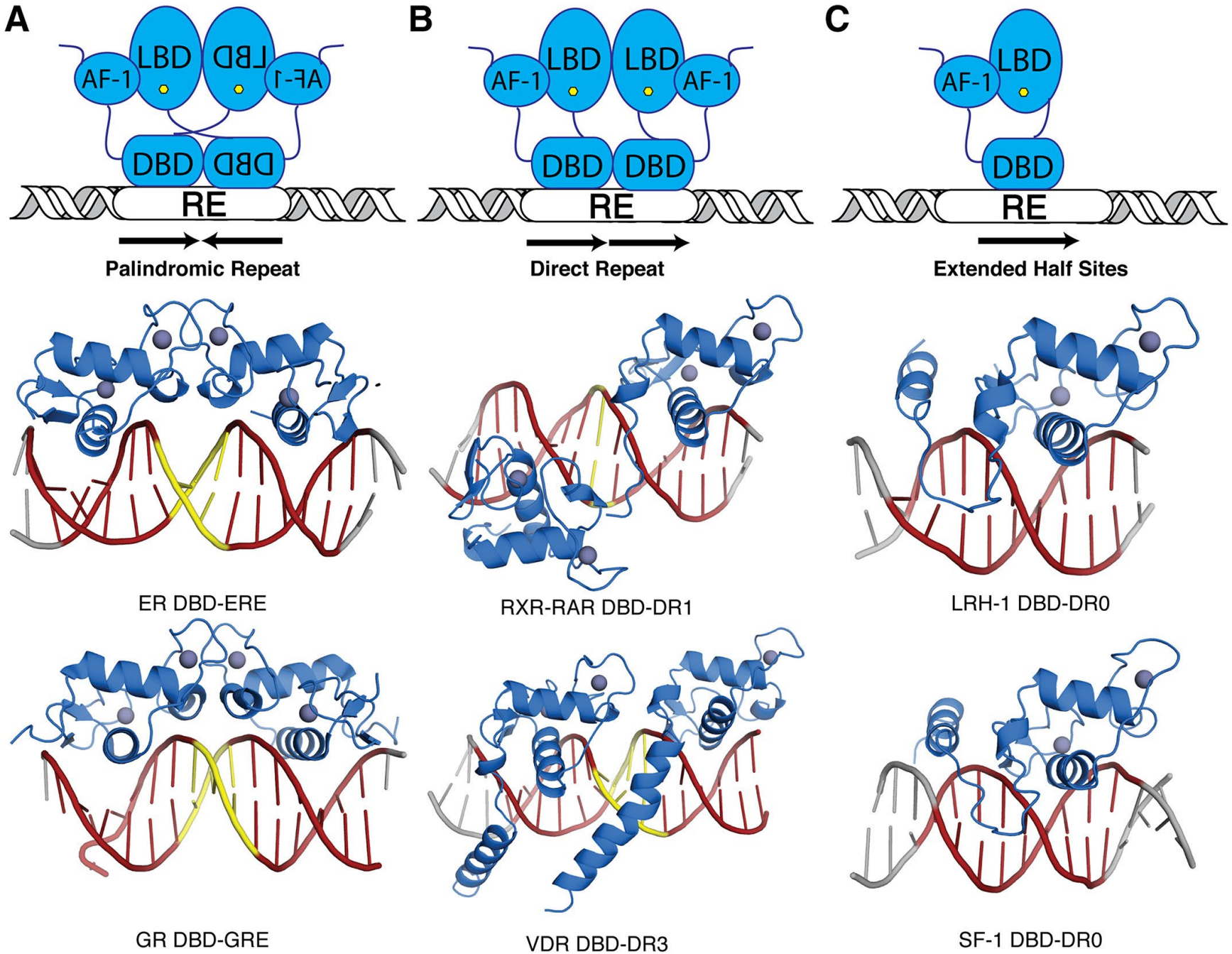

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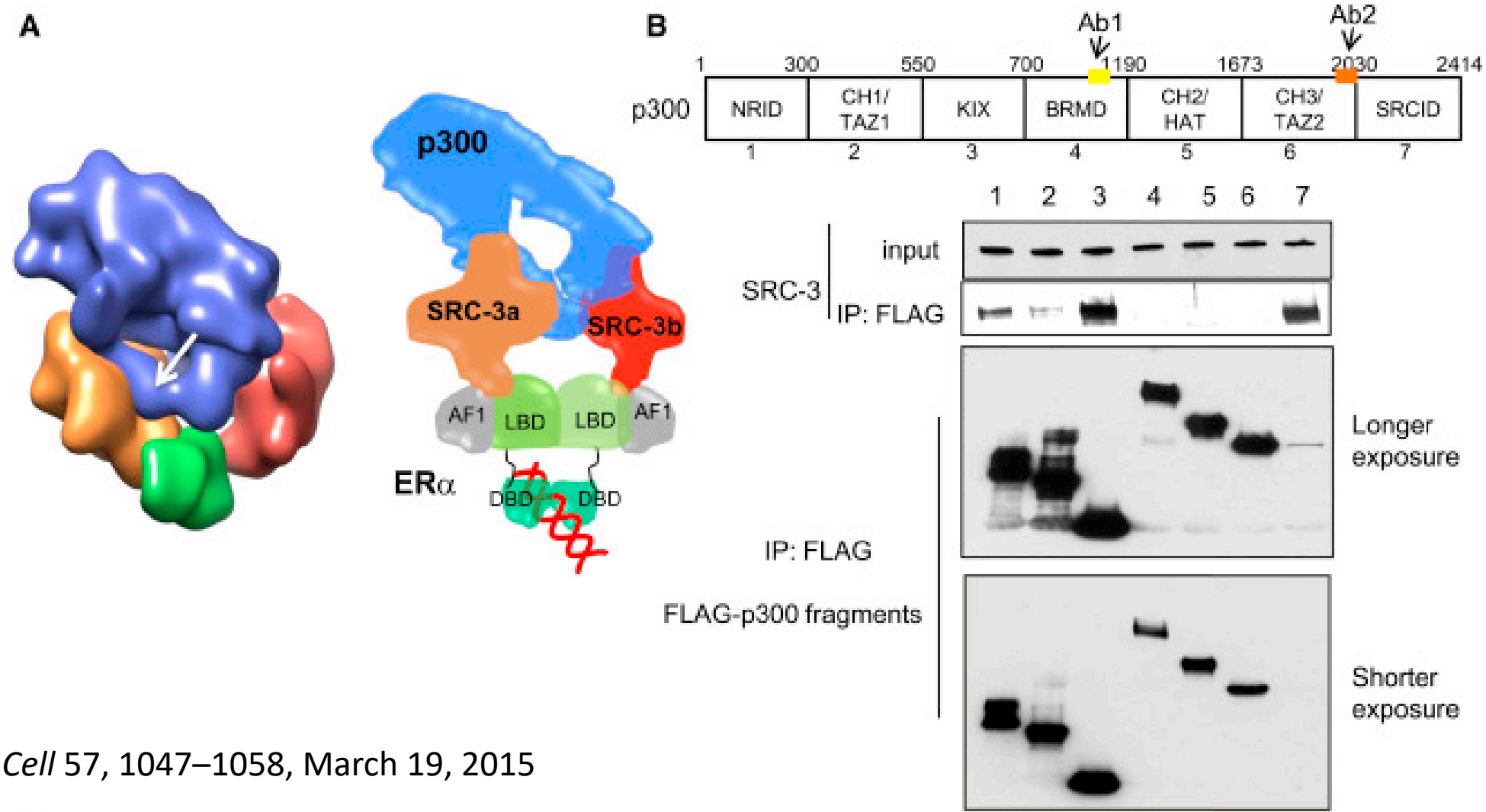
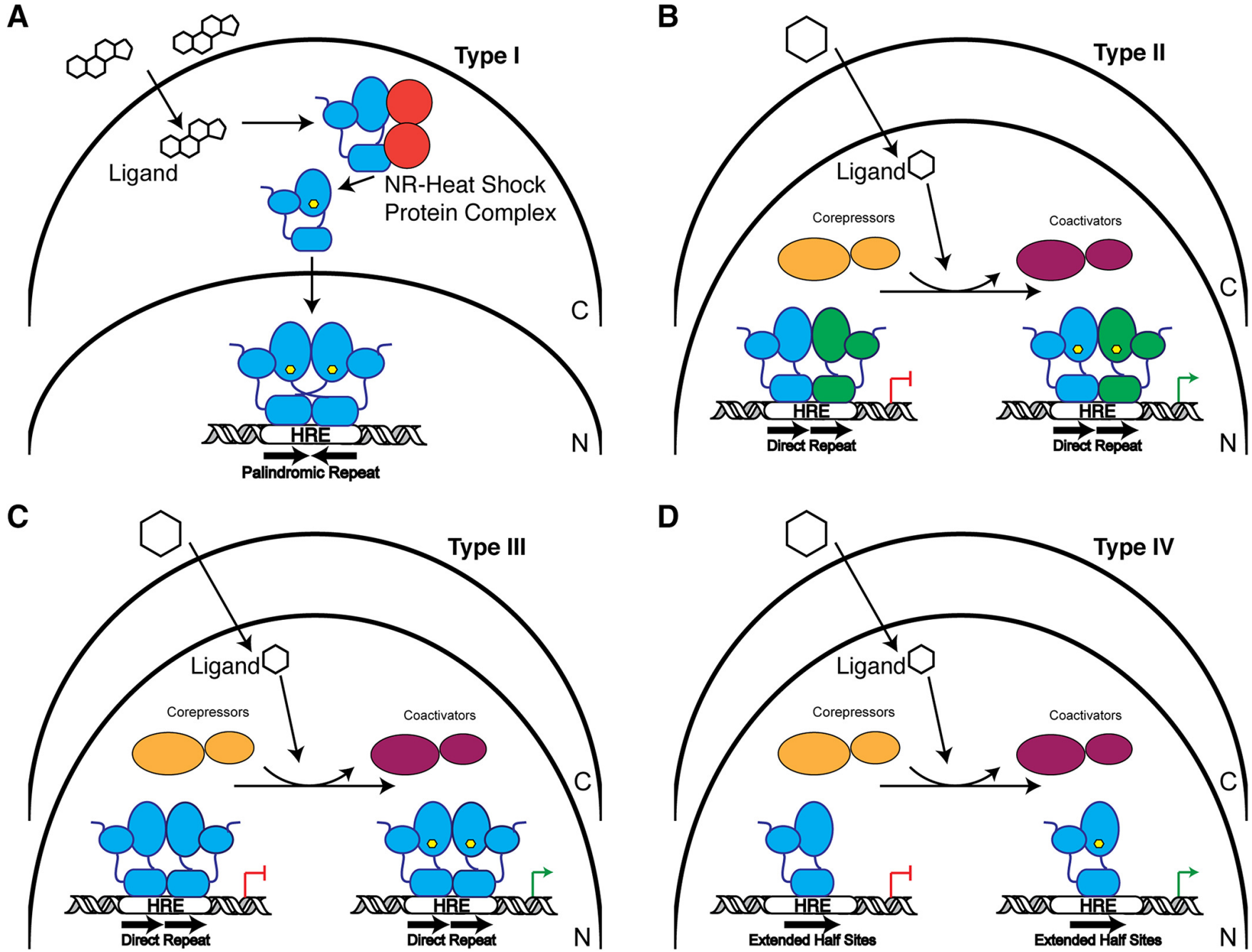
Structure of the ERE-DNA/ERa/SRC-3/p300 Complex



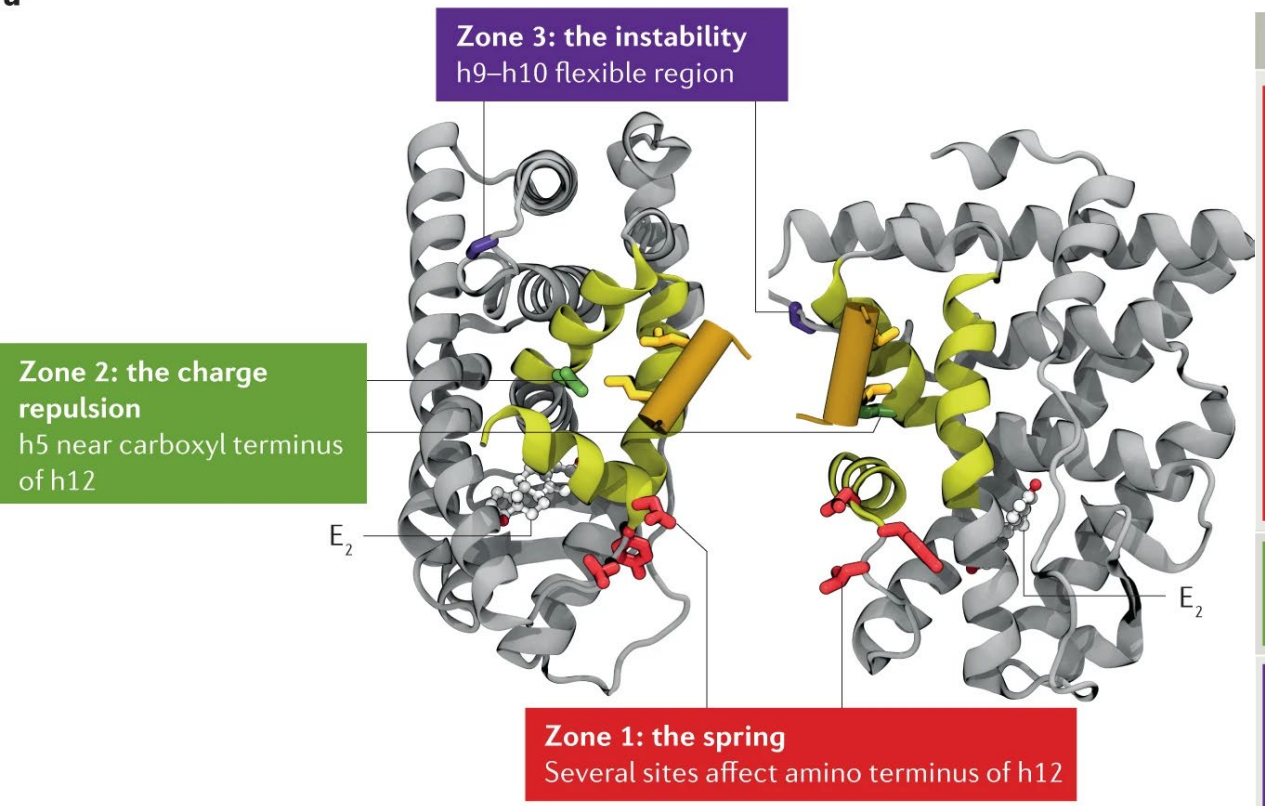
Molecular Cell 57, 1047–1058, March 19, 2015



Protein Science, Volume: 27, Issue: 11, Pages: 1876-1892, First published: 15 August 2018, DOI: (10.1002/pro.3496)

Activating mutations in the ERα ligand- binding domain

a



Zone	Mutation site	Pharmacological phenotype	Likely mechanism
1. h11–h12 loop, near the amino terminus of h12	Y537S, Y537N, Y537C and Y537D	Strong constitutive activity and AE resistance Y537S > Y537N ~ Y537C	Strong hydrogen bond to D351 stabilizes AF2 conformation and allows loop to pack better
	D538G	Moderate constitutive activity and more easily reversed by AEs	Flexibility in linkage to h12 allows loop to pack better
	L536R, L536H, L536P and L536Q	Modest constitutive activity but harder to reverse with AEs	Replacing leucine with hydrophilic residue eliminates hydrophobic penalty from water exposure
2. h5, near the carboxyl terminus of h12	E380Q	Weak constitutive activity and easily reversed by AEs	Relieves coulombic repulsion with two acidic residues in h12
3. h9–h10 loop	S463P	Moderate constitutive activity and easily reversed by AEs	Potentially stabilizes the dimer interface and/or a flexible loop and possibly enables further intra-domain interactions

If we have time