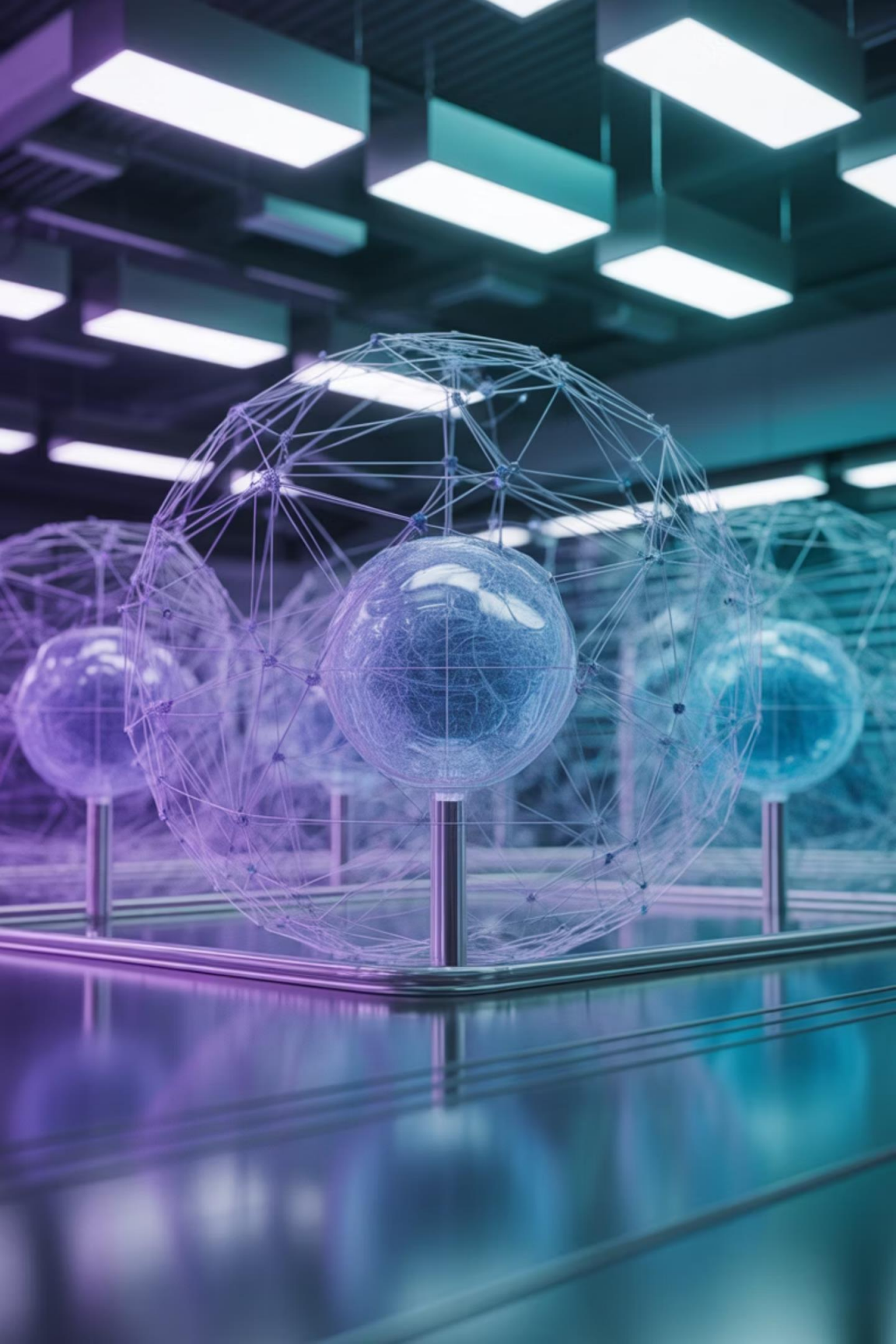




SYK in Immune Thrombocytopenia: Mechanistic Foundations and Network Biology

A translational exploration of immune, inflammatory and thrombotic crosstalk

Alessandro Luchesi, MD, PhD



Objectives of the talk

- Reframe ITP as a systemic, network-driven disorder where immunity, inflammation, and vascular biology intersect.
- Examine SYK as a high-connectivity signalling hub linking B-cell activation, macrophage-mediated clearance, platelet immunoreactivity, and cytokine amplification.
- Highlight how SYK-centred biology enhances our understanding of ITP far beyond thrombocytopenia.

Reference: Provan et al., EBioMedicine 2022, doi:10.1016/j.ebiom.2021.103841

The sickening consequences of too much SYK signaling

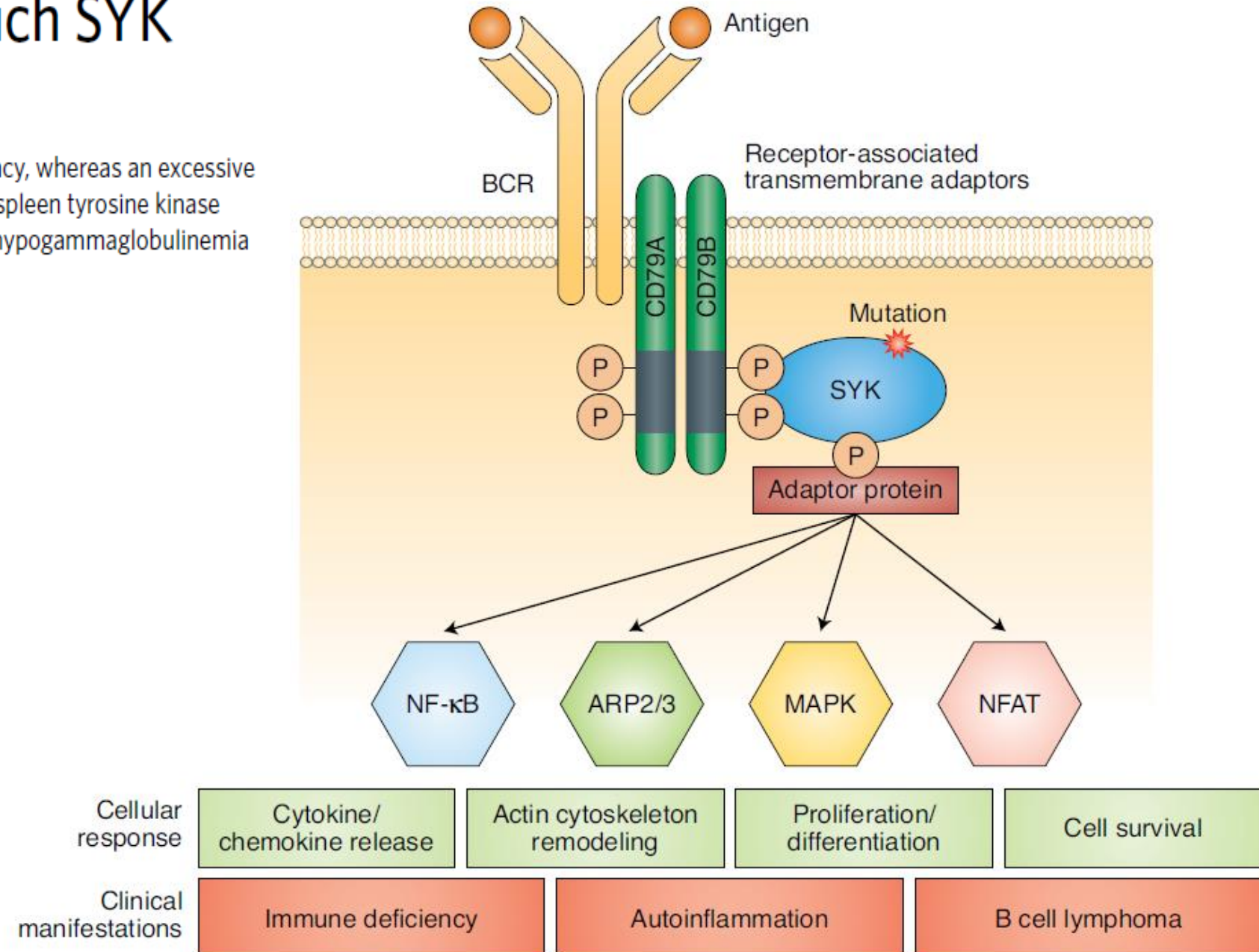
Immune responses require a delicate balance: a weak response can cause immunodeficiency, whereas an excessive response can lead to hyperinflammatory disease and hematological malignancy. Because spleen tyrosine kinase has roles in multiple signaling pathways, its gain-of-function alterations in humans cause hypogammaglobulinemia as well as autoinflammation and predisposition to B cell lymphoma.

Ivona Aksentijevich

NATURE GENETICS | VOL 53 | APRIL 2021 | 426-434

Increased BCR signaling and autoreactive B cells have been implicated in the pathogenesis of autoimmune diseases, leukemias, and B-line lymphomas

SYK with gain of function mutations, paradoxical features of immunodeficiency and hyperinflammatory responses



Clinical Implications: SYK Beyond ITP

SYK's central role in immune signalling extends to various chronic inflammatory and autoimmune diseases, highlighting its broad therapeutic potential.



Airway Inflammation

Exacerbates asthma & COPD via epithelial cell phosphorylation.



Liver Fibrosis

Promotes disease progression through TLR4 interaction on hepatic stellate cells.



Dermatological Conditions

Drives IL-17A secretion in psoriasis and atopic dermatitis.



Intestinal Inflammation

Involved in colitis pathogenesis through β -glucan and Mincle pathways.



Neurodegeneration

Regulates mTOR, contributing to increased Tau phosphorylation and accumulation.



Kidney Injury

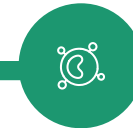
Activates Mincle-driven M1 macrophage phenotype, linked to inflammation.

Reference: Zhou et al, Immun Inflamm Dis. 2023; doi:10.1002/iid3.934

ITP: a *Systemic Disorder*, Not a Numerical Diagnosis



ITP is not defined by platelet count alone.



Chronic immune stimulation.



Dysregulated B- and T-cell responses.



Inflammatory splenic–hepatic niches.



Endothelial activation.



Coexistence of bleeding and thrombosis reflects a profound disturbance in immune–vascular homeostasis.

SYK: A Molecular Convergence Point

Spleen tyrosine kinase (SYK) is a 72-kDa non-receptor tyrosine kinase, widely expressed in haematopoietic cells like B lymphocytes, macrophages, and platelets. Its SH2 domains and kinase domain enable signal transduction from ITAM-bearing receptors.

ITAM receptors

FcγR, BCR, GPVI, CLEC-2

Integrin pathways

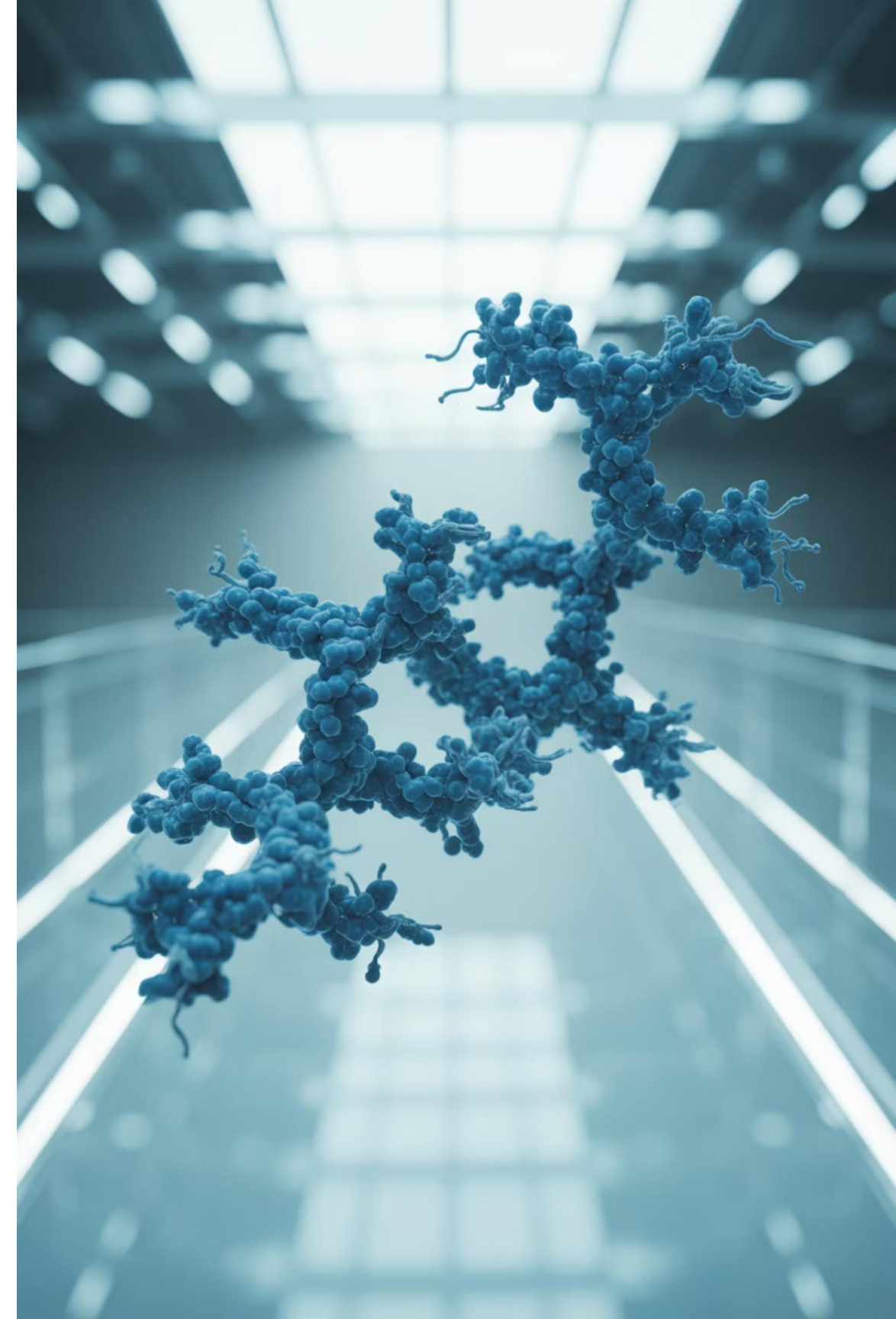
Outside-in signalling modulation

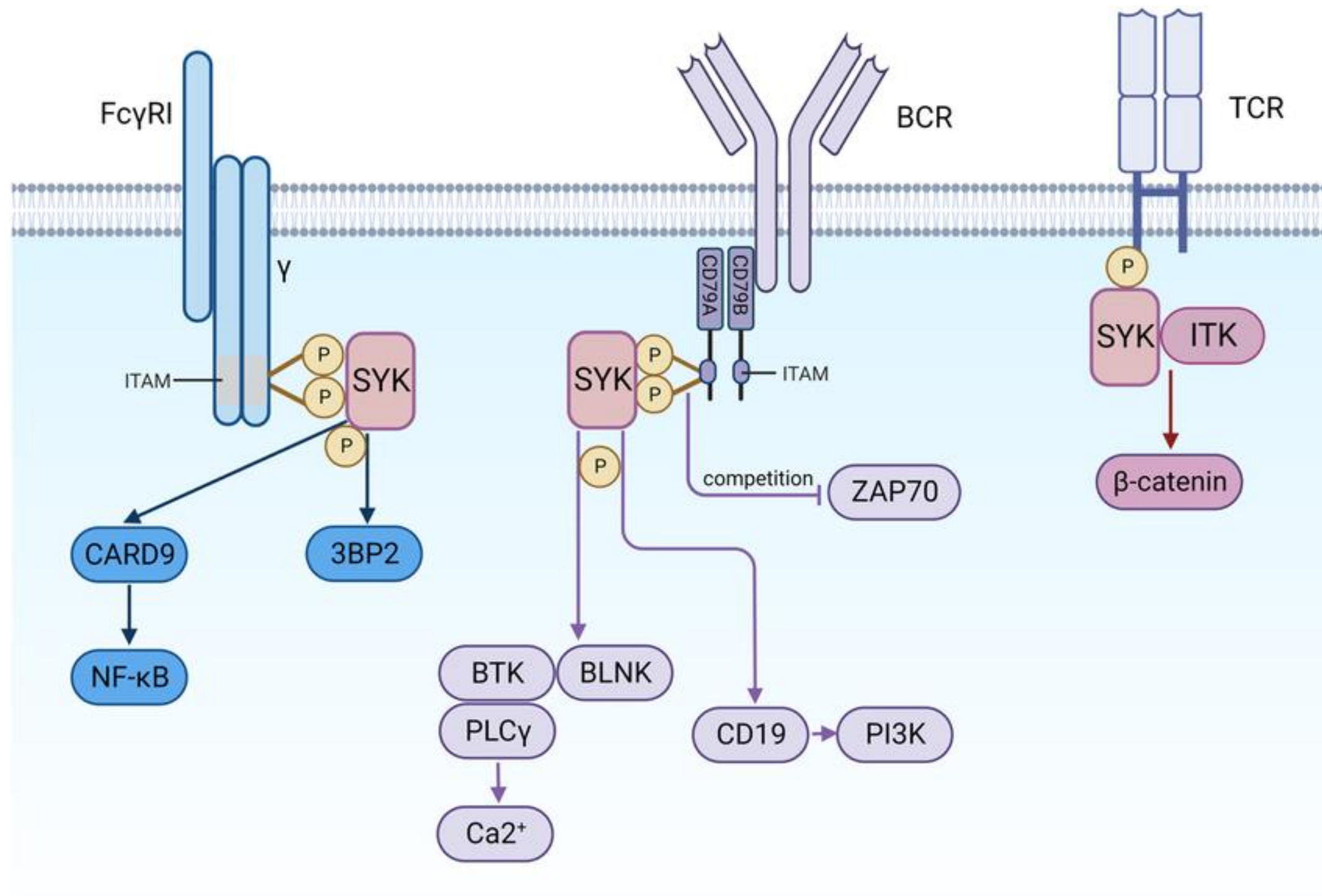
Pattern recognition

Dectin-1, TLR-associated signalling

This broad distribution positions SYK as a **central integrator** of innate immunity, adaptive responses, and platelet immunoreactivity.

Reference: Cooper et al., Platelets 2023, doi:10.1080/09537104.2022.2131751





ITAM–SYK Signalling: Translating Recognition into Action



ITAM Phosphorylation

Antigen or immune-complex binding



SYK Recruitment

Kinase activation



Cascade Initiation

PI3K–AKT, PLCγ2, MAPK



Cellular Response

Phagocytosis, cytokines, activation

ITAM phosphorylation upon antigen or immune-complex binding recruits and activates SYK. Activated SYK initiates PI3K–AKT, PLCγ2, and MAPK cascades that drive phagocytosis, cytokine release and platelet activation. In ITP, this architecture connects autoantibody binding with multifaceted downstream immune consequences.

Reference: Swinkels et al., Front Immunol 2018, doi:10.3389/fimmu.2018.00880

The Macrophage–Platelet Synapse



IgG opsonisation

Autoantibodies coat platelet surface glycoproteins.



Fcγ receptor engagement

Splenic macrophages recognise IgG-opsonised platelets.



SYK activation

ITAM phosphorylation triggers signalling cascade.

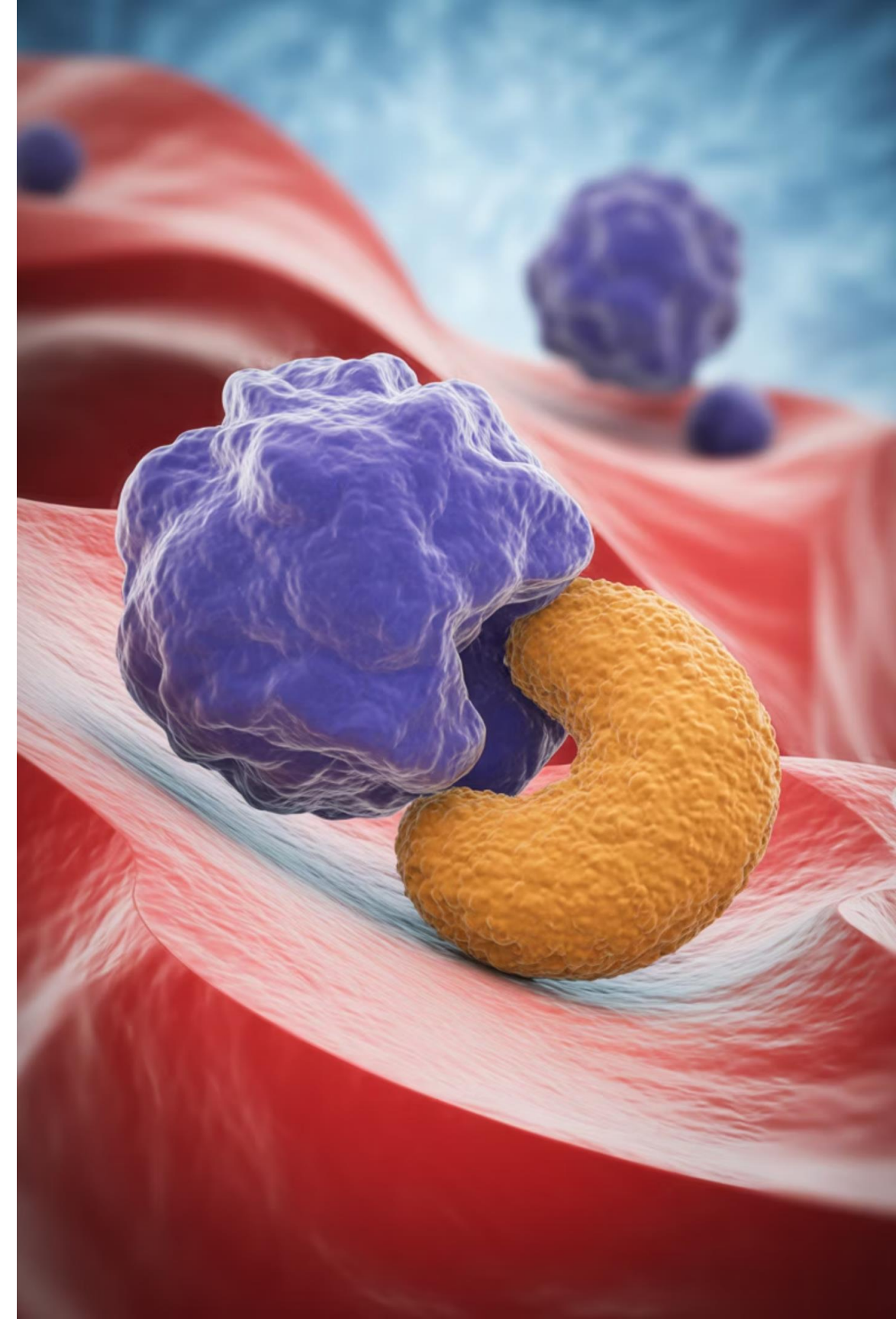


Phagocytosis

Cytoskeletal remodelling leads to platelet destruction.

This immunological synapse in the splenic red pulp represents the **primary anatomical site** of accelerated peripheral platelet clearance in ITP pathophysiology. SYK orchestrates the transition from immune recognition to cellular destruction.

Reference: Swinkels et al., Front Immunol 2018, doi:10.3389/fimmu.2018.00880



SYK as the Engine of B-Cell Autoimmunity

SYK occupies an indispensable position in B-cell receptor (BCR) signalling, serving as the principal transducer of antigen recognition. Following BCR engagement and CD19 co-receptor activation, SYK drives critical B-cell developmental checkpoints:

O1

Initial activation

Transition from naive to activated state

O2

Germinal centre reactions

Somatic hypermutation and affinity maturation

O3

Class-switch recombination

IgG isotype generation

O4

Plasma cell differentiation

Sustained autoantibody secretion

The persistence, affinity, and isotype profile of anti-platelet autoantibodies—and consequently the chronicity of ITP—are fundamentally linked to the strength and duration of SYK-dependent signalling.

Reference: Cooper et al., Platelets 2023, doi:10.1080/09537104.2022.2131751

Crosstalk in the Marrow: T Cells, Stromal Niches and Megakaryocytes

CD8+ cytotoxic T lymphocytes contribute to megakaryocyte apoptosis through perforin-granzyme and FasL-mediated pathways.

SYK participates in complex bidirectional communication between immune cells and stromal elements that shapes a chronically pro-inflammatory microenvironment.

Impaired Thrombopoiesis

Reduced megakaryocyte ploidy, maturation defects, and diminished proplatelet formation.

Altered Platelet Quality

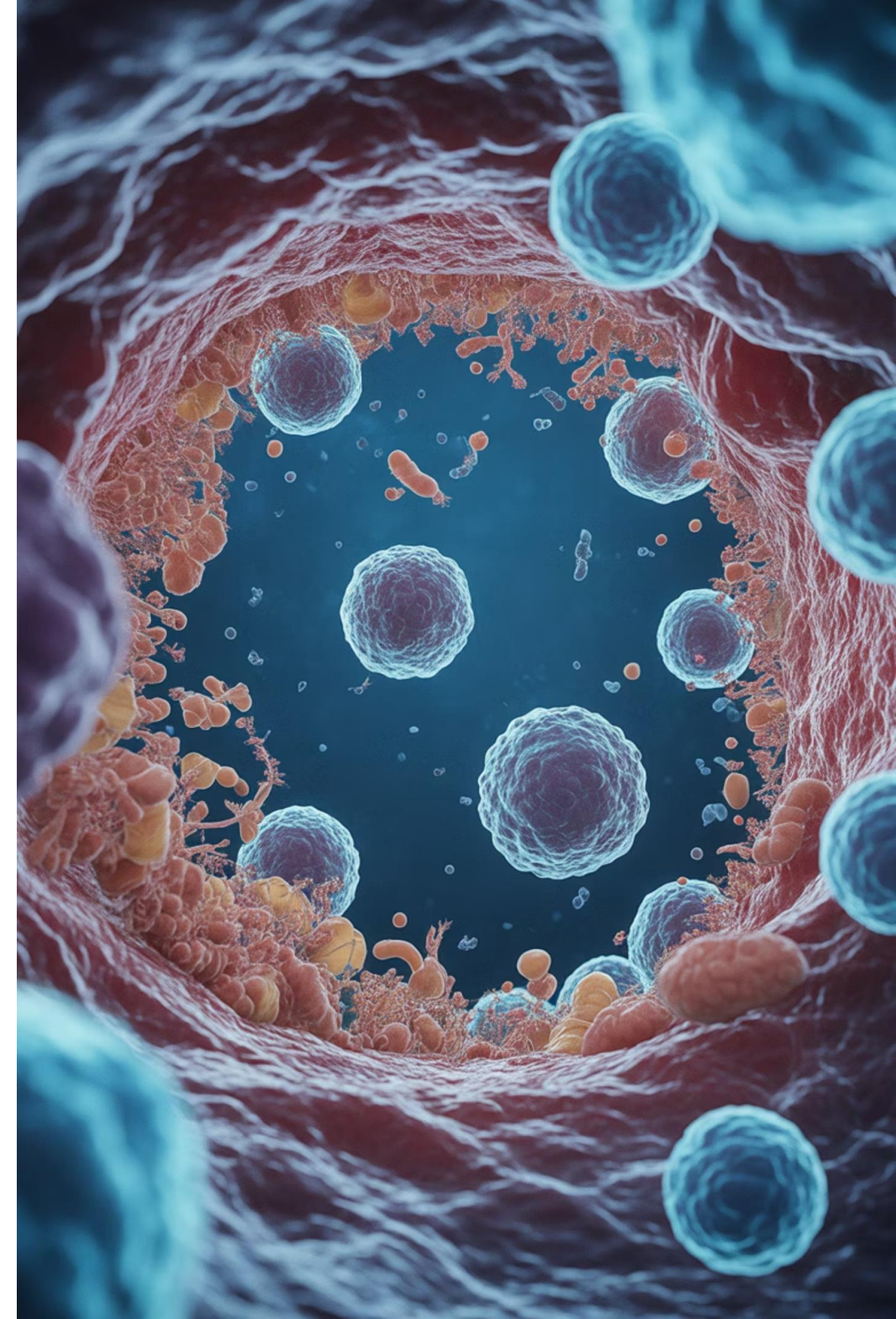
Dysregulated cytokine milieu affects platelet RNA content and functional capacity.

Stromal Dysfunction

Mesenchymal stem cell abnormalities and niche disruption.

ITP is therefore best understood as a **dual-compartment disorder**: impaired platelet production coupled with accelerated peripheral clearance.

Reference: Swinkels et al., Front Immunol 2018, doi:10.3389/fimmu.2018.00880



A Multi-Compartment Disease: A Rationale for Network Interpretation



Bone marrow

Impaired megakaryopoiesis



Spleen

Principal site of clearance



Liver

Alternative clearance pathway



Circulation

Immune complexes, NETs



Lymphoid tissues

Autoantibody generation



Endothelium

Vascular activation, thrombosis

ITP pathophysiology unfolds across anatomically and functionally distinct compartments: bone marrow, spleen, liver, peripheral blood, lymphoid organs, and the vascular endothelium. SYK represents one of the rare molecular nodes with demonstrated functional relevance across **all these compartments simultaneously**. Consequently, understanding ITP necessitates a system-level, network-oriented perspective rather than reductionist linear pathway models.

The **Pleiotropic Reach** of SYK

Phagocytosis

Autoantibody-mediated clearance

B-Cell Activation

Autoantibody production

Innate Immunity

Pattern recognition signalling

Platelet Activation

ITAM-dependent responses

Cytokine Amplification

Inflammatory cascade enhancement

Neutrophil Interactions

NET formation and aggregation

Inflammasome

NLRP3 activation

SYK regulates: • autoantibody-mediated phagocytosis • B-cell activation • innate immune signalling • platelet ITAM-dependent activation • cytokine amplification • neutrophil–platelet interactions • NET formation • inflammasome activation. Such pleiotropy explains its disproportionate influence on ITP biology.

Reference: Cooper et al., Platelets 2023, doi:10.1080/09537104.2022.2131751

ITP as a Network Disturbance, Not a Linear Pathway

Classical Model

Autoantibody → clearance

Lacks explanatory power

Network Model

ITP arises from a distributed network of immune, inflammatory, stromal and vascular interactions. SYK is a high-connectivity node whose activation state reshapes the entire network.

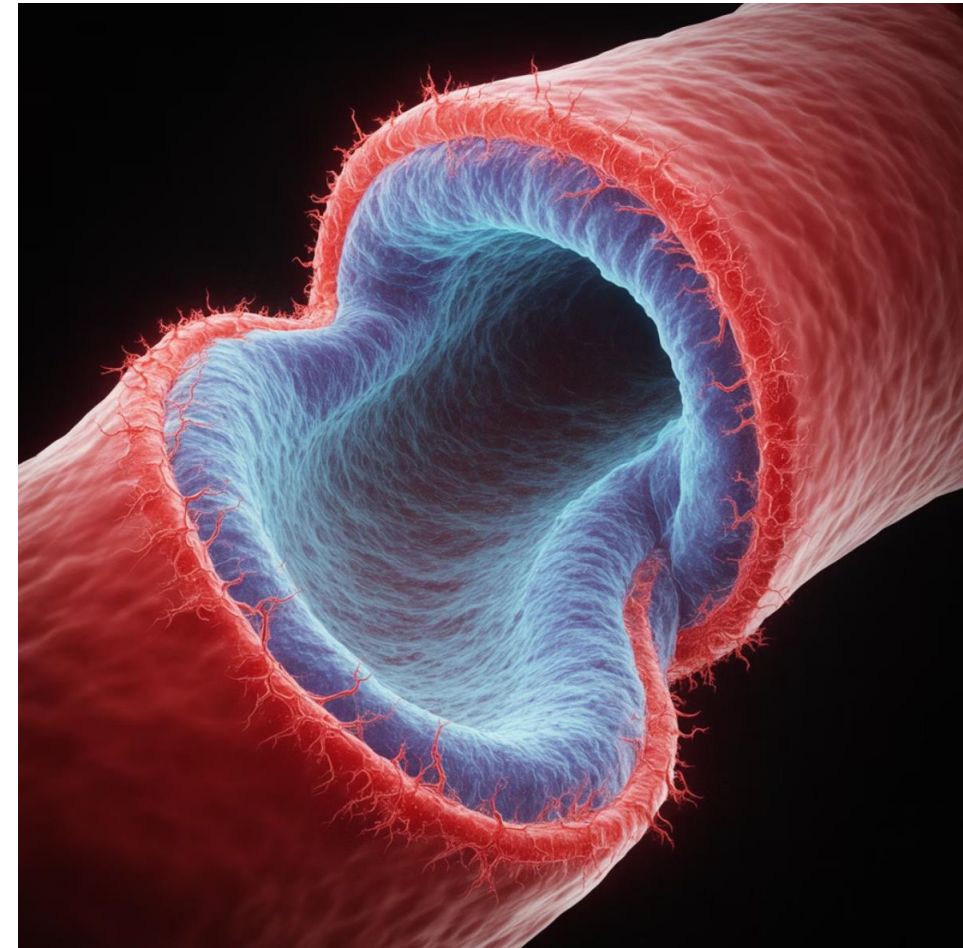
Reference: Provan et al., EBioMedicine 2022, doi:10.1016/j.ebiom.2021.103841

SYK and Thrombo-Inflammation: Insights from Vascular Disease

Translational models of haemolytic and inflammatory vasculopathies have illuminated SYK's role beyond classical haemostasis. In murine models of sickle cell disease, thrombotic microangiopathy, and complement-mediated injury, SYK emerges as a critical coordinator of:

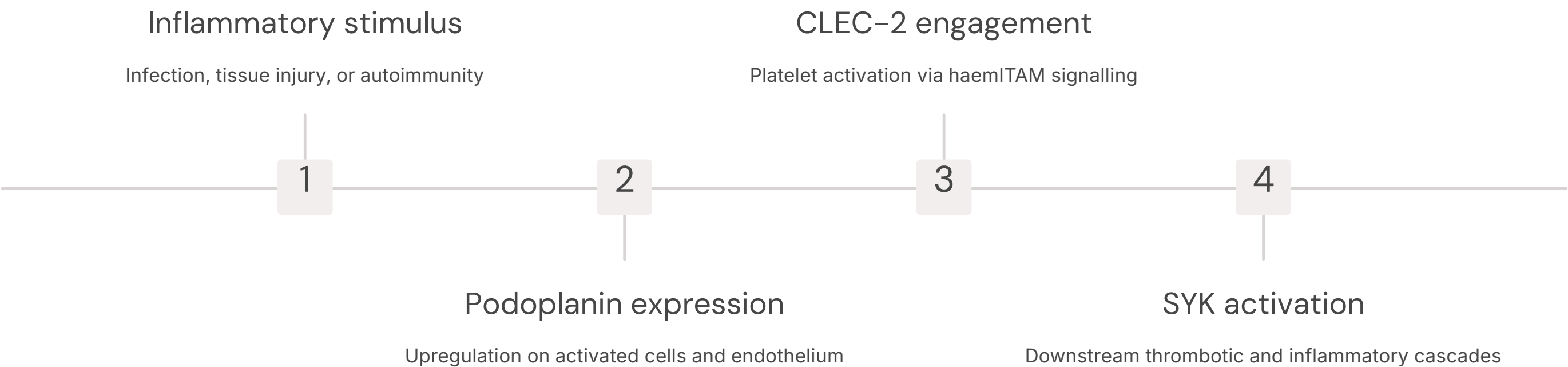
- Platelet activation via GPVI and CLEC-2
- Neutrophil recruitment to inflamed endothelium
- Endothelial-leukocyte interactions facilitating vaso-occlusion
- Microvascular injury propagation

Pharmacological or genetic SYK inhibition in these experimental systems reduces neutrophil-platelet aggregate formation, diminishes vaso-occlusive crises, and improves tissue perfusion—establishing SYK as a viable target for **microvascular protection** in thrombo-inflammatory conditions.



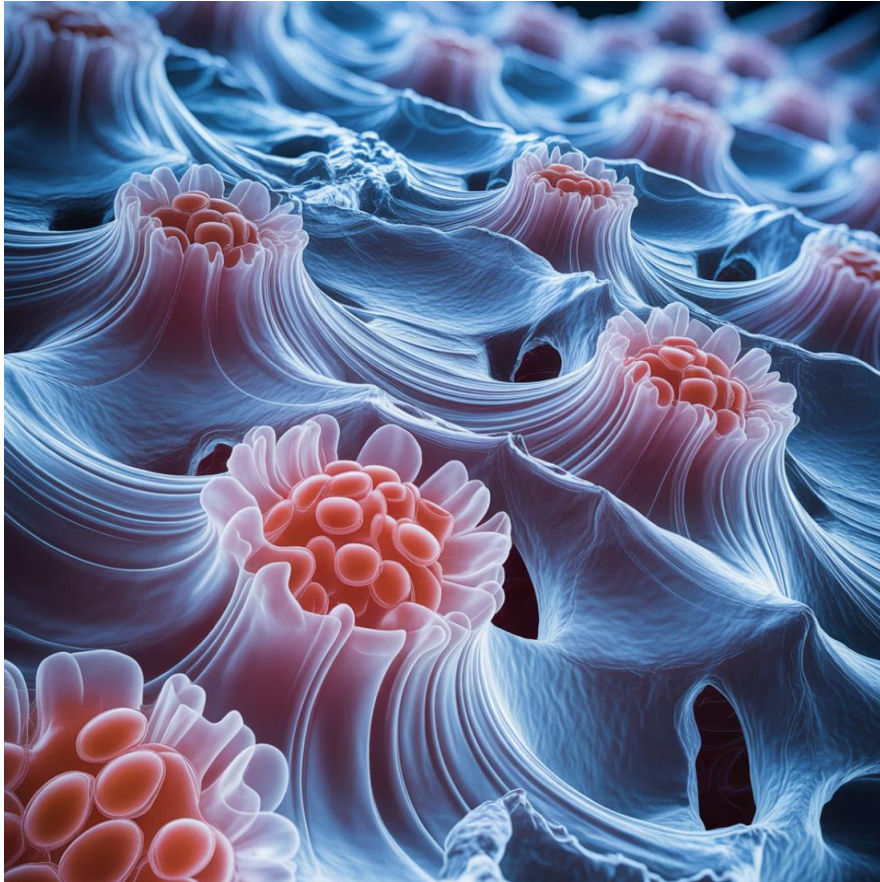
The CLEC-2/SYK Axis in Immune-Driven Platelet Activation

C-type lectin-like receptor 2 (CLEC-2) represents a haemITAM-bearing receptor that triggers robust SYK-dependent signalling during inflammation, infection, and endothelial stress. Its endogenous ligand, podoplanin, is upregulated on inflamed lymphatic endothelium and certain tumour cells.



In SARS-CoV-2 infection models, genetic or pharmacological disruption of the CLEC-2/SYK axis substantially reduces pulmonary immunothrombosis and acute lung injury. These findings reinforce the concept of platelets as **immune effector cells** rather than merely haemostatic elements, with direct relevance to ITP's inflammatory phenotype.

GPVI–SYK: Collagen Recognition and Vascular Integrity



Glycoprotein VI (GPVI) functions as the principal platelet collagen receptor, initiating ITAM-dependent SYK signalling upon exposure to subendothelial matrix proteins. This pathway ensures appropriate platelet responses at sites of vascular injury.

In the context of ITP, several observations suggest dysregulated GPVI–SYK signalling contributes to clinical complexity:

- Reticulated platelets

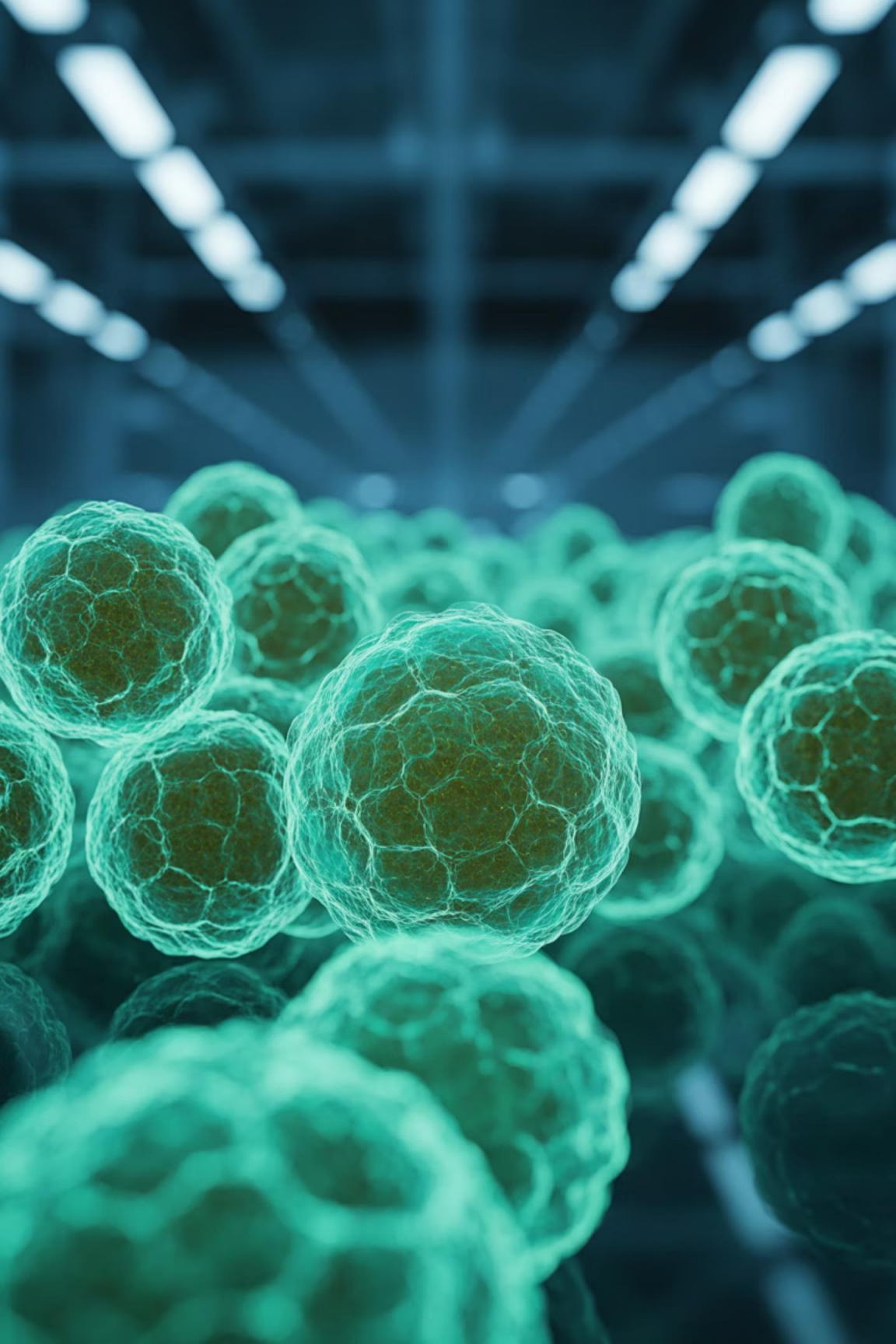
Young, stress-induced platelets exhibit heightened GPVI expression and SYK-dependent reactivity.

- Inflammatory priming

Circulating cytokines (IL-1 β , TNF- α) enhance GPVI-mediated activation thresholds.

- Thrombotic paradox

Hyperreactive residual platelets may contribute to thrombotic events despite thrombocytopenia.

A microscopic image showing several neutrophils (leukocytes) with their characteristic multi-lobed nuclei. Fine, thread-like structures called neutrophil extracellular traps (NETs) are visible extending from the cells. The image is tinted with a blue and green color scheme.

SYK as a Gatekeeper of NETosis

Neutrophil extracellular traps (NETs) are crucial for host defence but can drive immunothrombosis when dysregulated. SYK activation is essential for NET formation, including pathways triggered by:



Immune complexes

FcγR-mediated SYK activation drives NADPH oxidase-dependent NETosis.



Uric acid crystals

DAMP-induced SYK signalling promotes NET release.



Platelet–neutrophil interactions

Contact-dependent & paracrine mechanisms require SYK.

NETs amplify immunothrombosis by scaffolding platelet adhesion, complement activation, and coagulation. In chronic inflammatory states like ITP, persistent immune activation may enhance this **SYK-dependent neutrophil–platelet axis**, increasing vascular inflammation and thrombotic risk.

Reference: Markovtsov et al., Blood 2022, doi:10.1182/blood-2022-164455

SYK and the NLRP3 Inflammasome



DAMP Recognition

Danger signals detected



Lyn/SYK Signalling

Kinase cascade activation



NLRP3 Activation

Inflammasome assembly



IL-1 β and IL-18

Pro-inflammatory release

Several DAMPs activate NLRP3 through Lyn/SYK signalling, yielding IL-1 β and IL-18 release. Inflammasome activation contributes to systemic inflammation, marrow suppression and endothelial dysfunction—features increasingly recognised in ITP physiology.

Reference: Shio et al., PLoS Pathog 2009, doi:10.1371/journal.ppat.1000559

ITP as a "Hot" Immune State

Chronic Inflammation

Accumulating evidence characterises chronic ITP as a systemically "hot" immunological state, distinguished by multiple converging inflammatory pathways:



IL-1-driven inflammation

Sustained inflammasome activity and pyrogenic cytokine release



NET formation

Chronic neutrophil activation and extracellular trap release



Endothelial activation

Upregulation of adhesion molecules and procoagulant surface expression



Platelet priming

Enhanced responsiveness to agonists despite reduced platelet counts

SYK occupies a central position within this inflammatory network, integrating signals from FcγR, BCR, ITAM receptors, and pattern-recognition pathways. Its activation state thereby amplifies disease activity across anatomical and cellular compartments, perpetuating the chronic inflammatory milieu.

Reference: Vogel et al., Thromb Res 2024, doi:10.1016/j.thromres.2024.03.020

SYK as a Selective Immune Modulator



Network Modulation, Not Global Suppression

- Reduces autoantibody-mediated clearance
- Attenuates cytokine cascades
- Decreases ITAM-dependent platelet activation

This represents selective **network modulation** rather than global immunosuppression.

Reference: Cooper et al., Platelets 2023,
doi:10.1080/09537104.2022.2131751

The Concept of Network Dampening

SYK functions as a "gain regulator" for multiple immune and haemostatic signalling pathways, including FcγR, BCR, GPVI, CLEC-2, and inflammasome activation cascades.

By modulating SYK activity, one effectively lowers the **amplitude of interconnected immune and thrombo-inflammatory responses** propagating through the system. This approach differs fundamentally from pathway-specific blockade:

Pathway-specific inhibition

Targets single mechanism; compensatory pathways remain active; limited clinical benefit

Network dampening via SYK

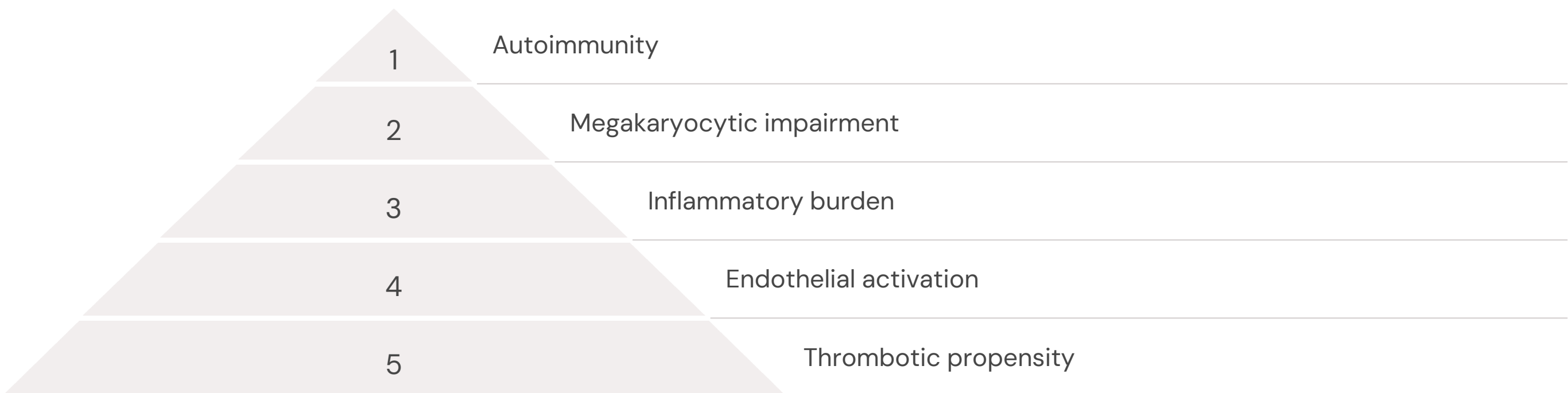
Modulates multiple convergent pathways; reduces system-wide hyperreactivity; broader therapeutic potential

This systems-level intervention aligns with ITP's network pathophysiology, addressing the distributed nature of immune dysregulation rather than attempting to sequentially block individual effector mechanisms.

Reference: Provan et al., EBioMedicine 2022, doi:10.1016/j.ebiom.2021.103841

Geometric Variability: Why Patients Behave So Differently

ITP patients show highly variable clinical outcomes and treatment responses, which complicates therapeutic prediction and personalized management.



A network-biology framework explains this heterogeneity: patients exhibit unique combinations of autoimmune activation, megakaryocytic impairment, inflammatory burden, and endothelial dysfunction. Variability in SYK-dependent pathway engagement (due to genetics, environment, and immune events) provides a **mechanistic explanation** for diverse clinical behaviors in ITP.

Future precision medicine may use SYK pathway biomarkers to stratify patients and guide treatment, enabling mechanism-informed personalized care.

Reference: Cooper et al., Platelets 2023, doi:10.1080/09537104.2022.2131751

Why SYK Is Mechanistically Compelling

Spleen tyrosine kinase (SYK) represents a critical nexus in immune thrombocytopenic purpura (ITP) pathophysiology, exerting influence across multiple disease-relevant domains simultaneously. Its functional breadth positions SYK as a mechanistically compelling therapeutic target and a conceptual framework for interpreting disease complexity.

Peripheral Clearance

Regulates macrophage-mediated platelet phagocytosis via FcγR signalling cascades

Antibody Generation

Modulates B-cell receptor signalling and autoantibody production

Inflammatory Tone

Influences cytokine networks and inflammasome activation

Platelet Immunoreactivity

Mediates platelet activation and immune-mediated dysfunction

Microvascular Integrity

Affects endothelial function and thromboinflammatory responses

Reference: Cooper et al., Platelets 2023, doi:10.1080/09537104.2022.2131751

A Disease of Nodes, Not Events

"You cannot understand a system by isolating its parts; you must study the patterns of connection." —
Gregory Bateson

ITP is best understood as a perturbation of patterns—immune, inflammatory, vascular. **SYK is a central connector within these patterns.**

Reference: Bateson, Steps to an Ecology of Mind 1972

A Network-Based Interpretation of ITP

1

From platelet numbers
to trajectories

2

From autoantibodies
to system behaviour

3

From haemostasis
to immunothrombosis

A network perspective shifts clinical thinking. **SYK bridges these interpretative domains.**

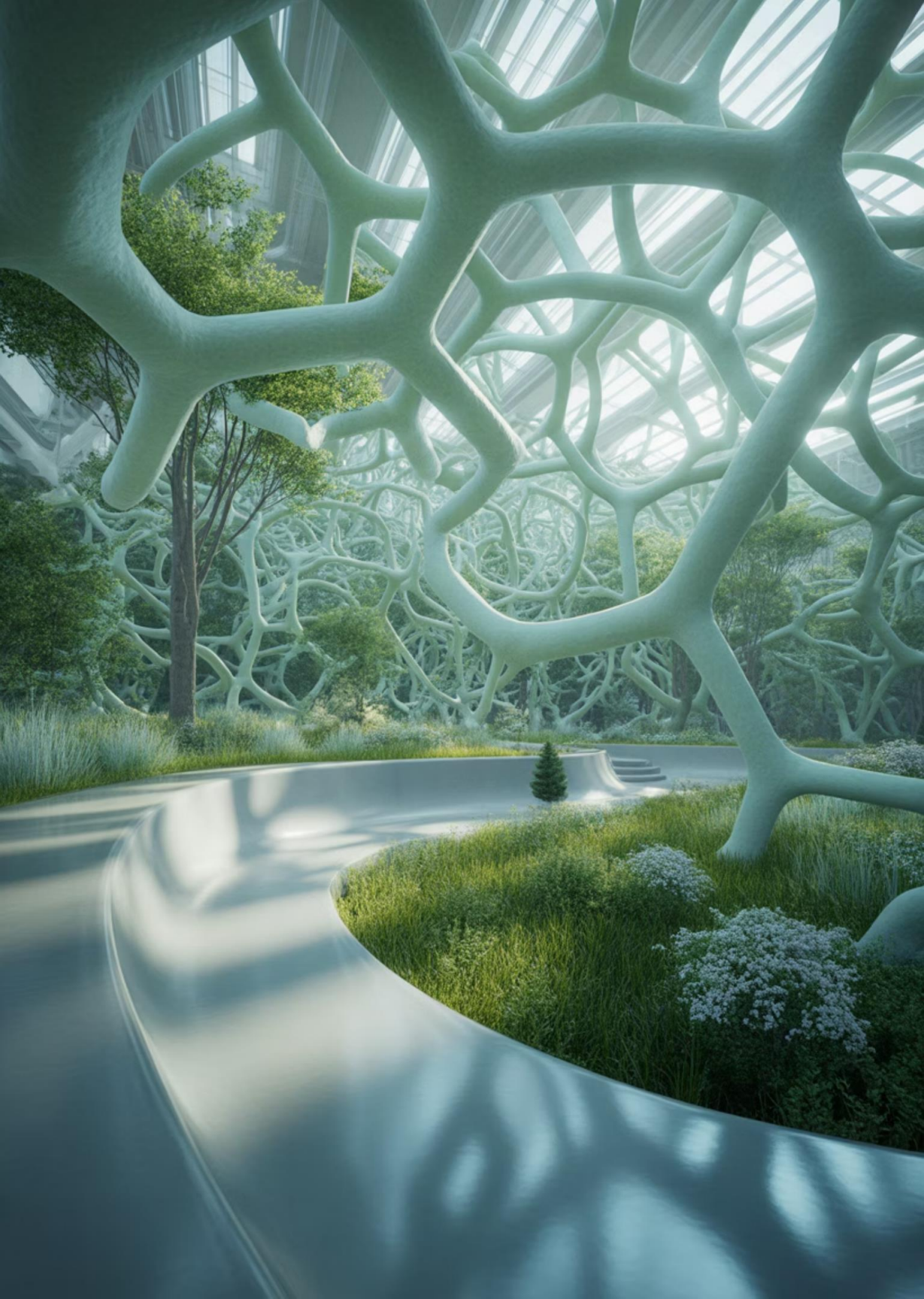
Reference: Provan et al., EBioMedicine 2022, doi:10.1016/j.ebiom.2021.103841

Risk as a Continuum: Beyond Threshold Thinking

Immune activation, thrombo-inflammation and platelet dysfunction exist on a continuum. SYK signalling intensity correlates with graded biological outputs rather than binary on/off states.

Translational medicine must adopt this continuum-based logic.

Reference: Cooper et al., Platelets 2023, doi:10.1080/09537104.2022.2131751



A Philosophical Interlude

"The major problems in the world are the difference between how nature works and how people think."

— Gregory Bateson

Biology works as an interconnected network. **ITP must be approached with the same epistemological humility.**

Reference: Bateson, Mind and Nature 1979



Clinical Implications of Understanding SYK

Recognising SYK-centred biology transforms clinical practice by providing a mechanistic framework for interpreting ITP's heterogeneous presentations and paradoxical features. This understanding enables more sophisticated phenotyping and individualised therapeutic strategies.

Fluctuating Phenotypes

SYK activity variability explains temporal changes in platelet counts, bleeding risk, and treatment responsiveness across disease course

Bleeding–Thrombosis Paradox

Concurrent thrombocytopenia and prothrombotic states reflect differential SYK signalling in distinct cellular compartments

Systemic Immune Derangement

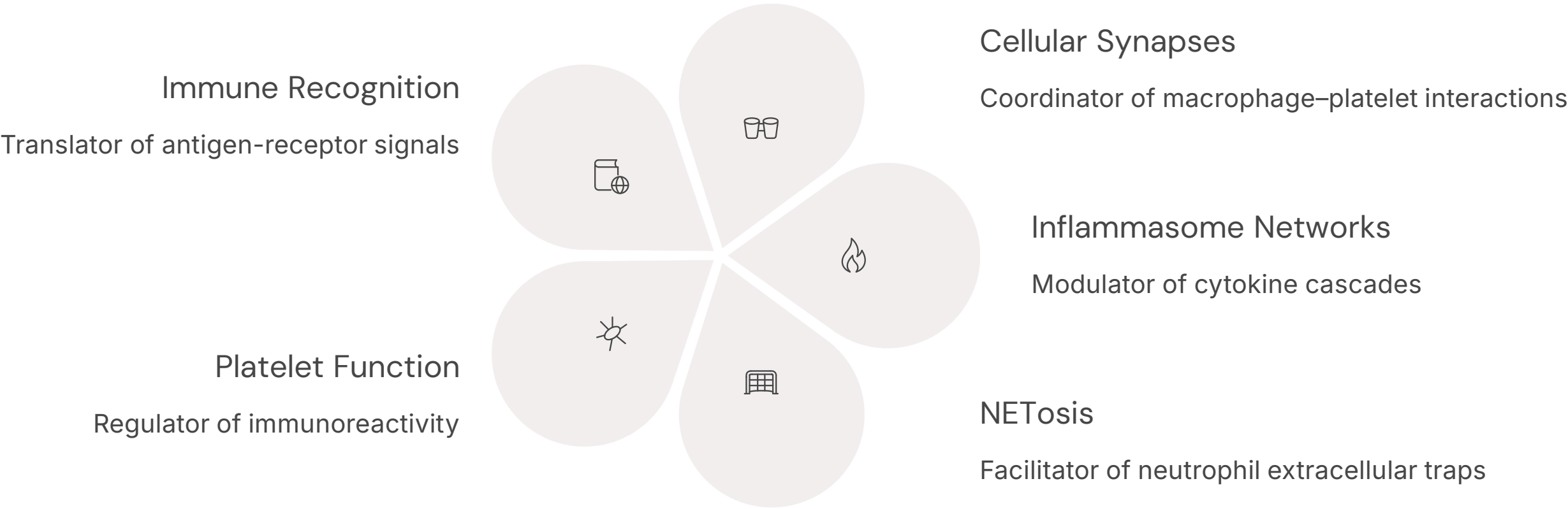
ITP represents more than numerical cytopenia—it embodies widespread immune dysregulation with multi-organ implications

Reference: Provan et al., EBioMedicine 2022, doi:10.1016/j.ebiom.2021.103841

Synthesis

SYK's Constellation of Functions in ITP

The centrality of SYK in ITP pathophysiology emerges from its multifaceted role across immune, inflammatory, and haemostatic domains. This functional constellation defines its mechanistic importance and therapeutic potential.



Reference: Cooper et al., Platelets 2023, doi:10.1080/09537104.2022.2131751



Thank you

Happy to take questions.