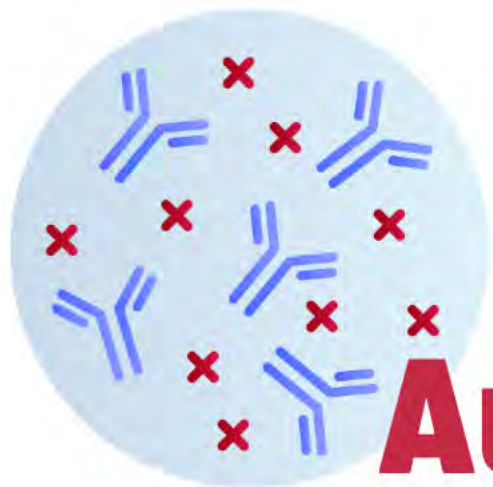
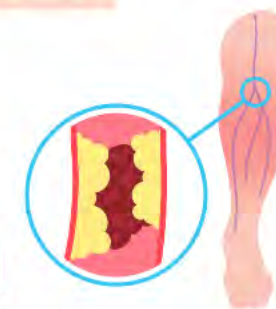


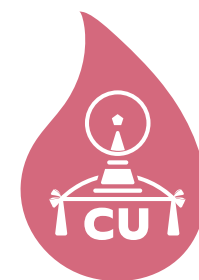


Autoimmune Anti-PF4 Thrombotic Syndrome



Thita Chiasakul, MD

Division of Hematology, Department of Medicine,
King Chulalongkorn Memorial Hospital and
Chulalongkorn University



Hematology

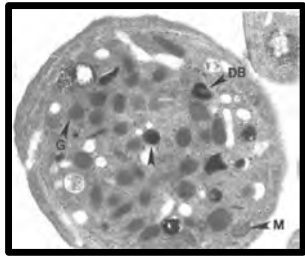
Outline

- **PF4 and anti-PF4 antibodies**
- **Laboratory testing for anti-PF4 antibodies**
- **Clinical spectrum of anti-PF4 disorders: Diagnosis & treatment**

Outline

- **PF4 and anti-PF4 antibodies**
- Laboratory testing for anti-PF4 antibodies
- Clinical spectrum of anti-PF4 disorders: Diagnosis & treatment

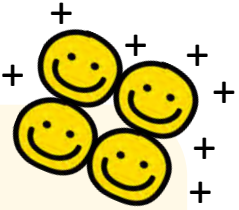
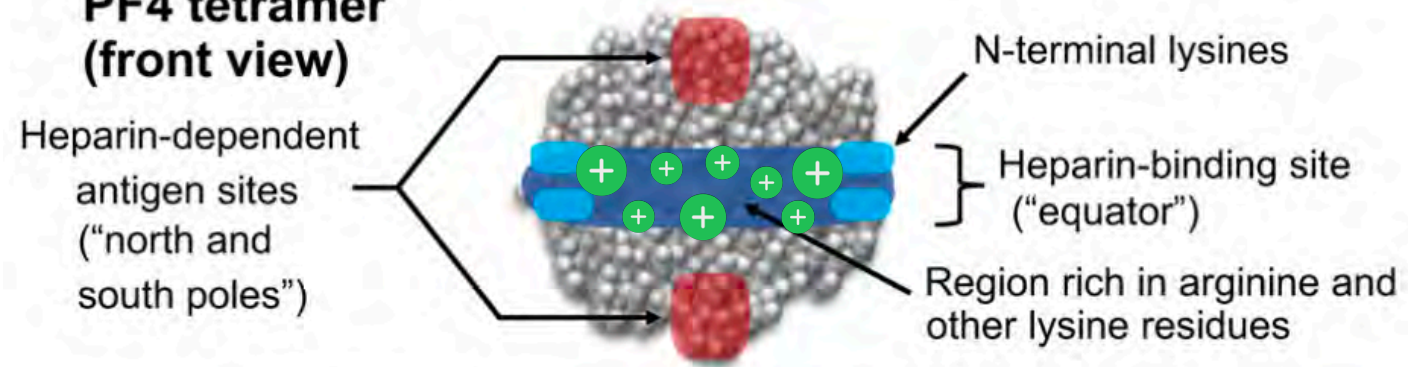
Platelet Factor4 (PF4, CXCL4)



Platelet α -granules



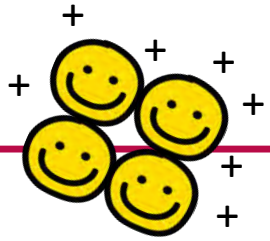
PF4 tetramer (front view)



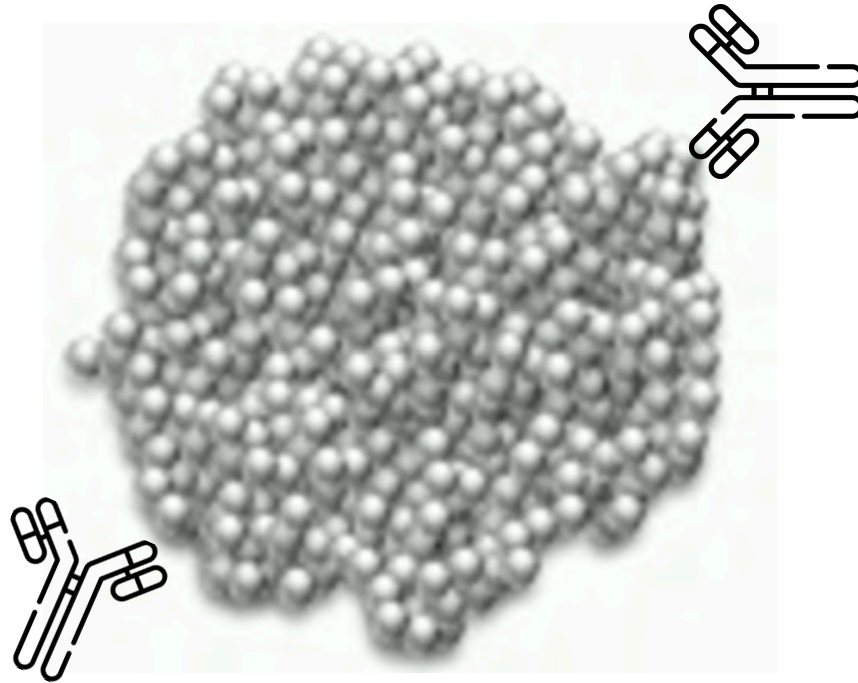
- Mainly exists as **tetramers**
- Highly **positively charged**
- Can **pair with another protein** to form heterodimers

• **Function:**

- Coagulation (Heparan sulfate)
- Inflammation and wound repair (IL8)
- Immune defense (CXCR4, pathogen)
- Anti-angiogenesis (VEGF)

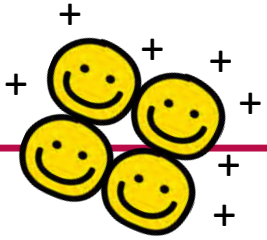


PF4 tetramer

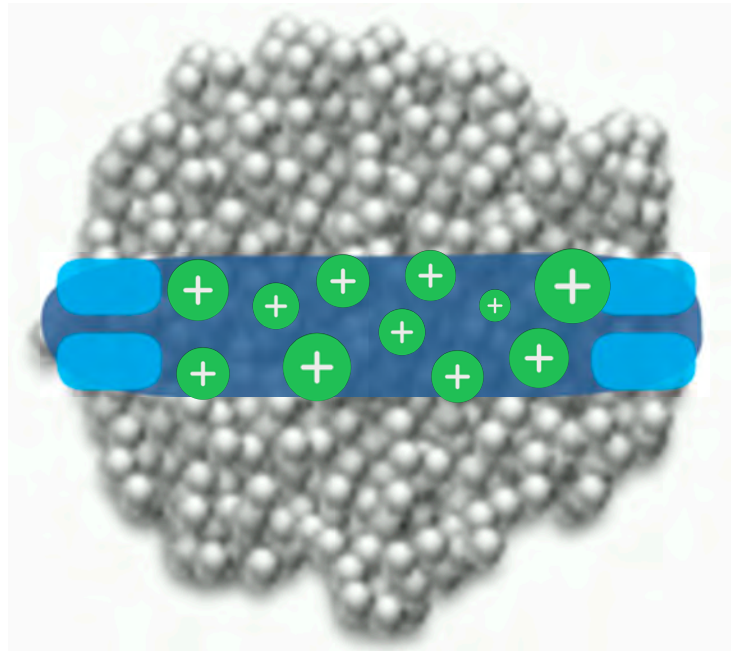


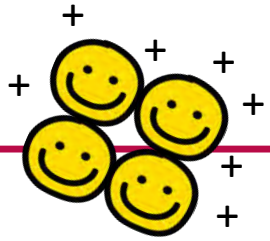
Not all anti-PF4 antibodies cause HIT: Type 1 Anti-PF4 antibodies (non-pathogenic, non-platelet activating)

PF4 tetramer

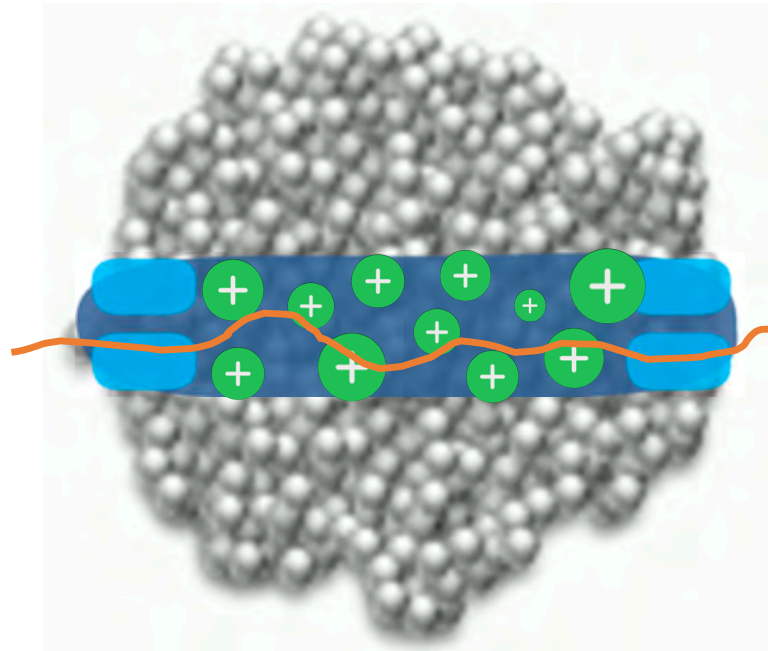


 **Heparin**

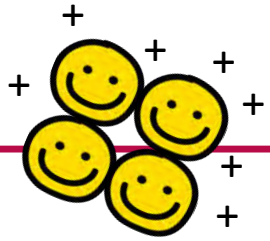




PF4/heparin Complex

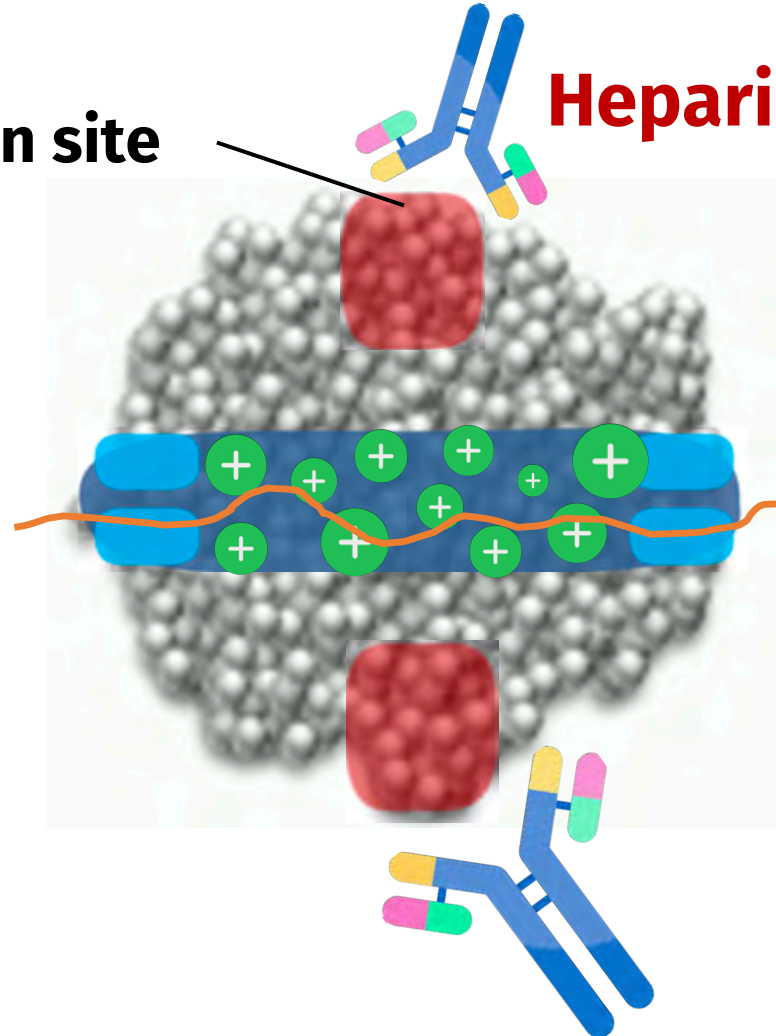


Heparin



PF4/heparin/IgG Immune Complex

Heparin-dependent antigen site



Heparin-dependent antibody

“HIT antibodies”

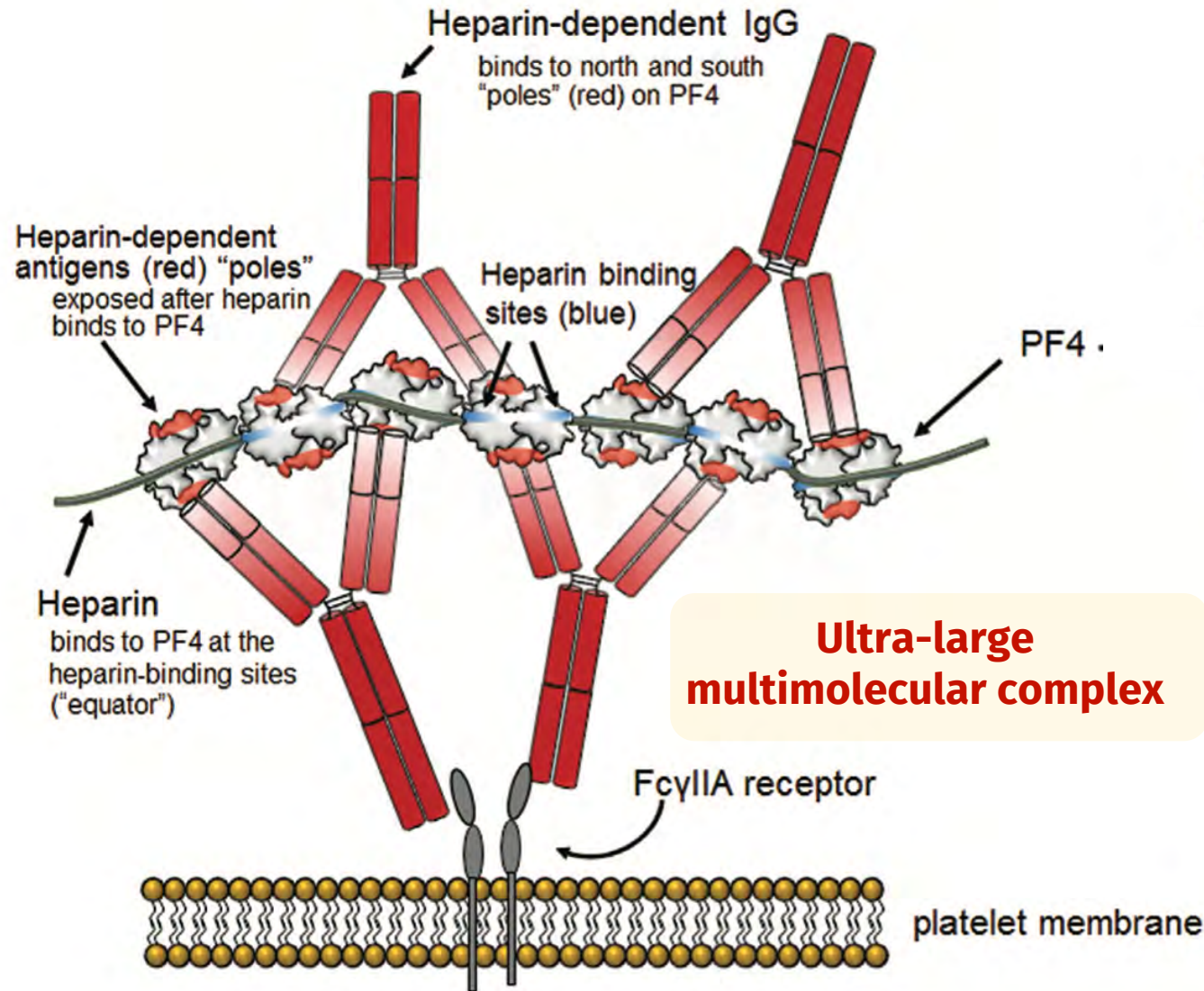
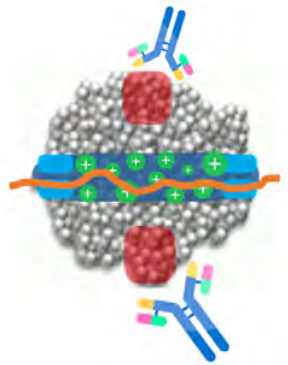
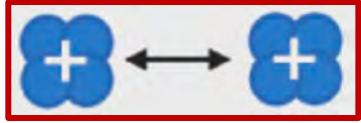
“HIT-like antibodies”

“Type 2 Anti-PF4 antibodies”

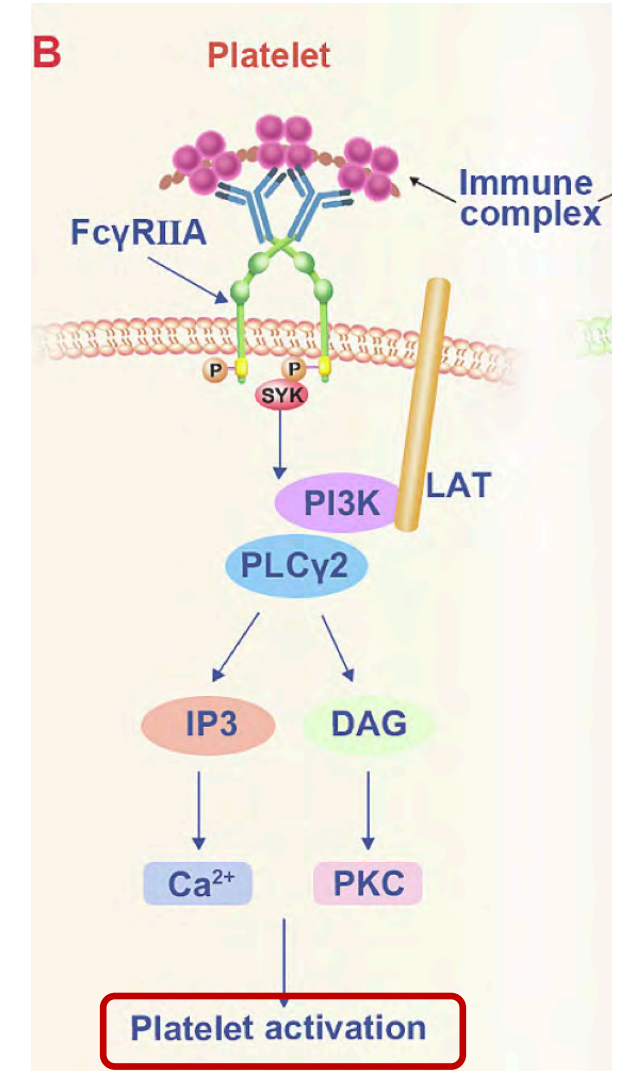
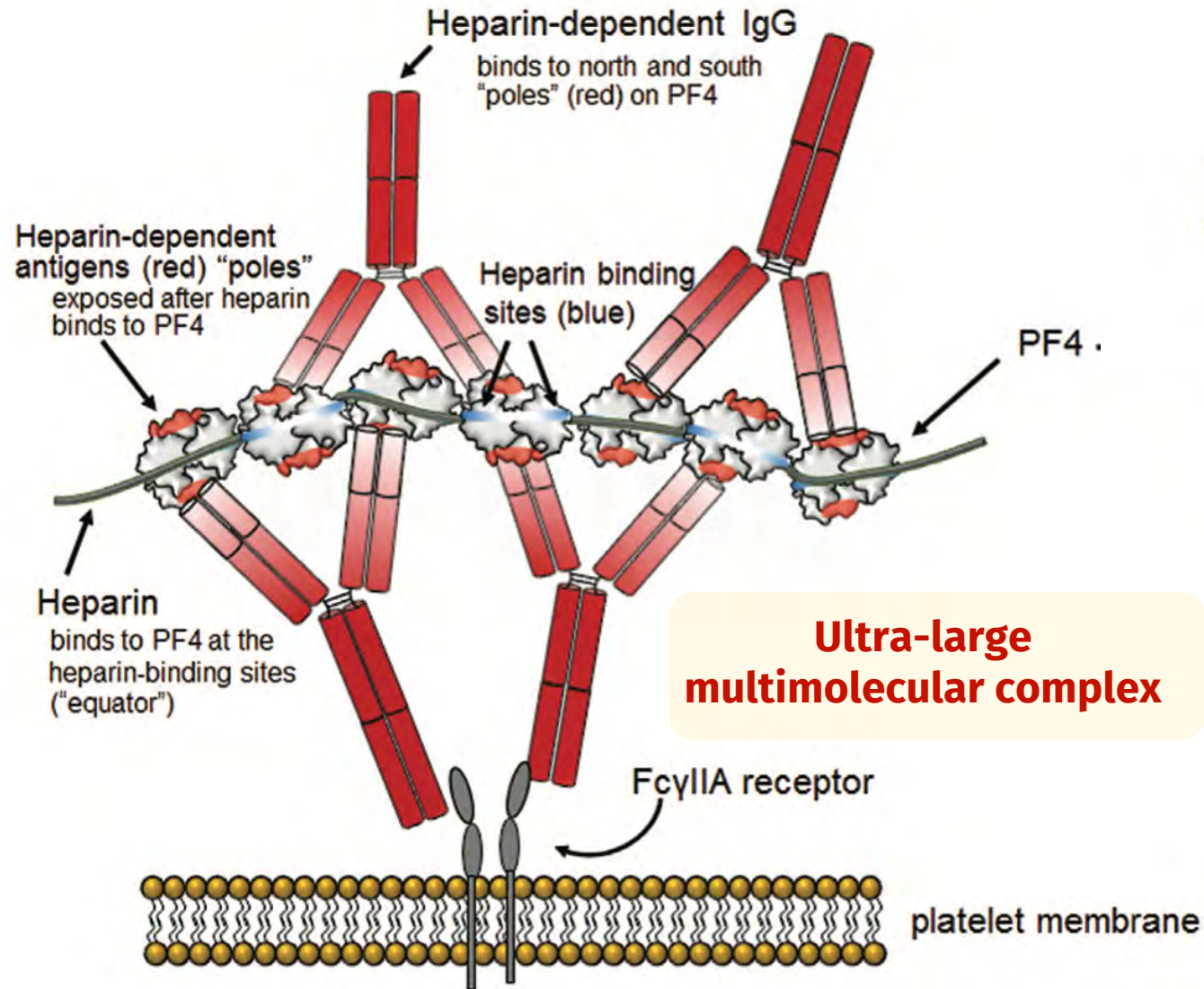
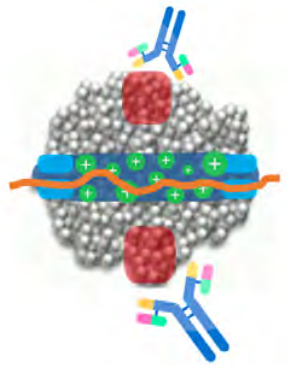
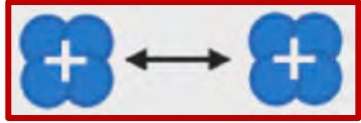
Heparin

❖ Classic HIT

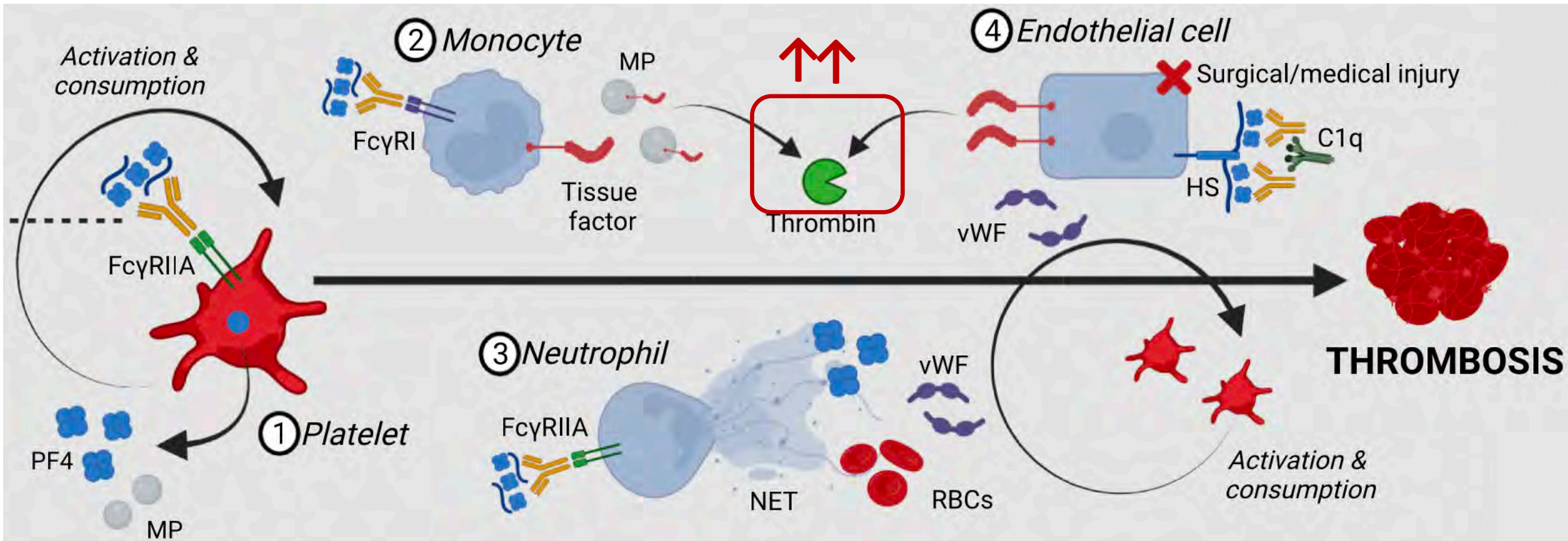
PF4/heparin/IgG Immune Complex



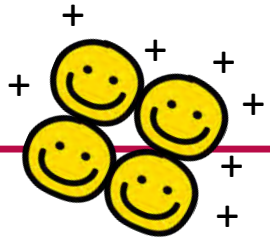
PF4/heparin/IgG Immune Complex



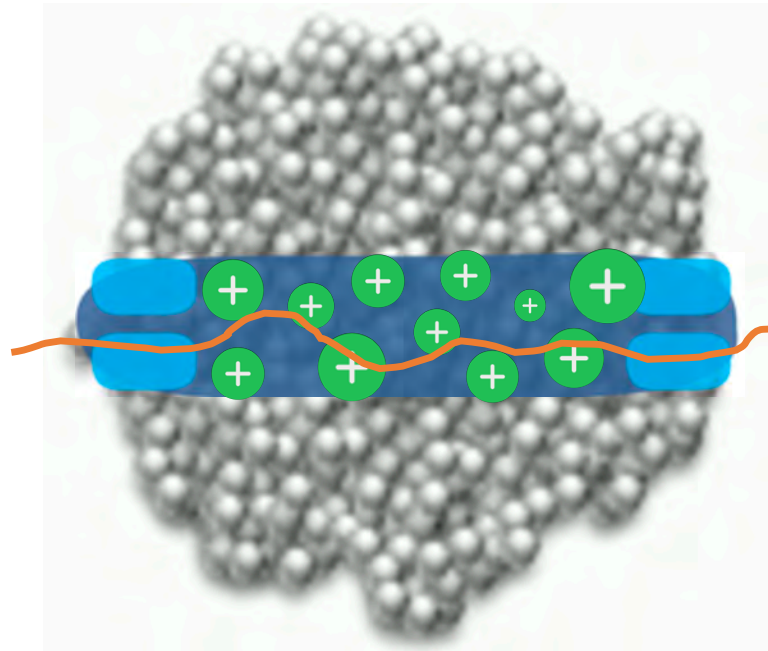
FcγRIIa Binding and Pancellular Activation



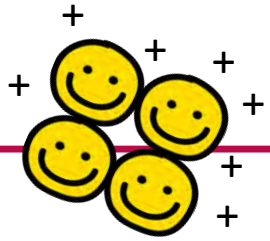
❖ **Classic HIT**



PF4/heparin Complex



Heparin



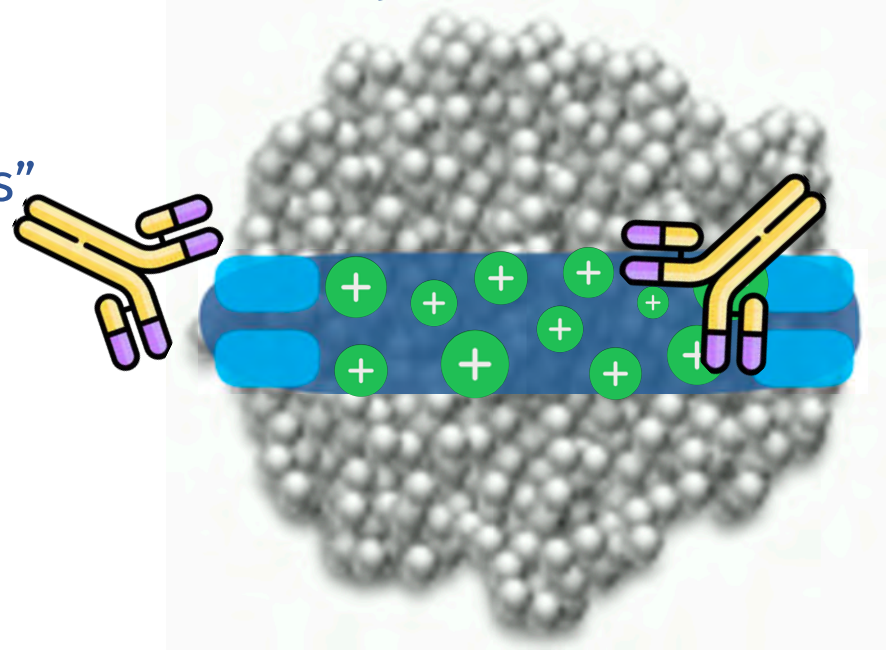
PF4/IgG Immune Complex

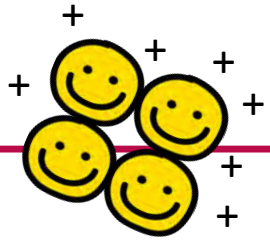
Heparin-independent antibody

“VITT antibodies”

“VITT-like antibodies”

“Type 3 Anti-PF4 antibodies”





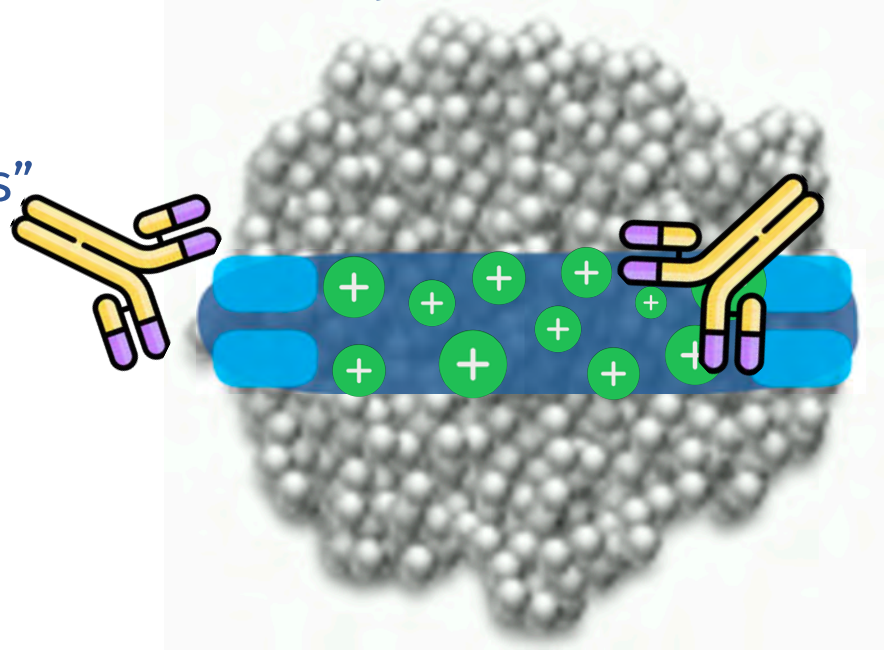
PF4/IgG Immune Complex

Heparin-independent antibody

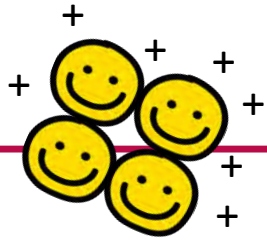
“VITT antibodies”

“VITT-like antibodies”

“Type 3 Anti-PF4 antibodies”



- ❖ Vaccine-induced Immune Thrombocytopenia with Thrombosis (VITT)
- ❖ Spontaneous VITT



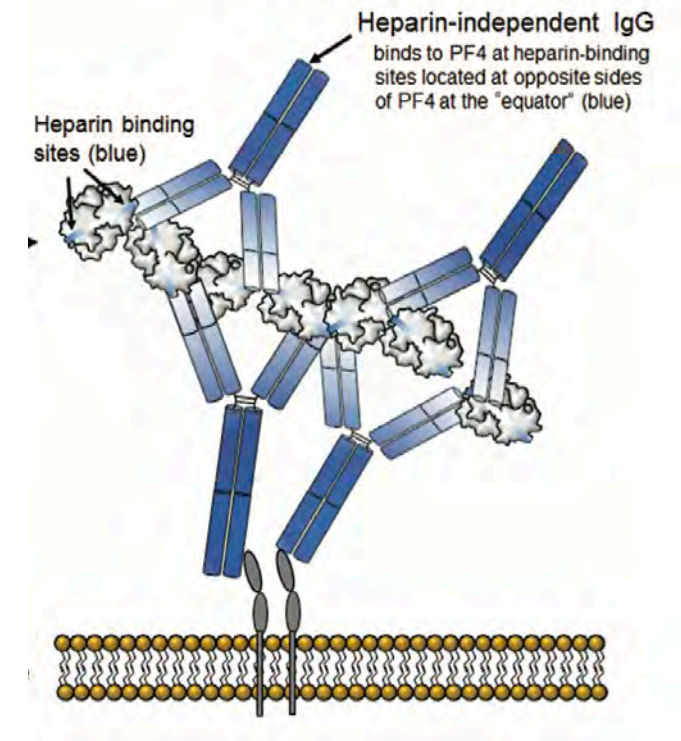
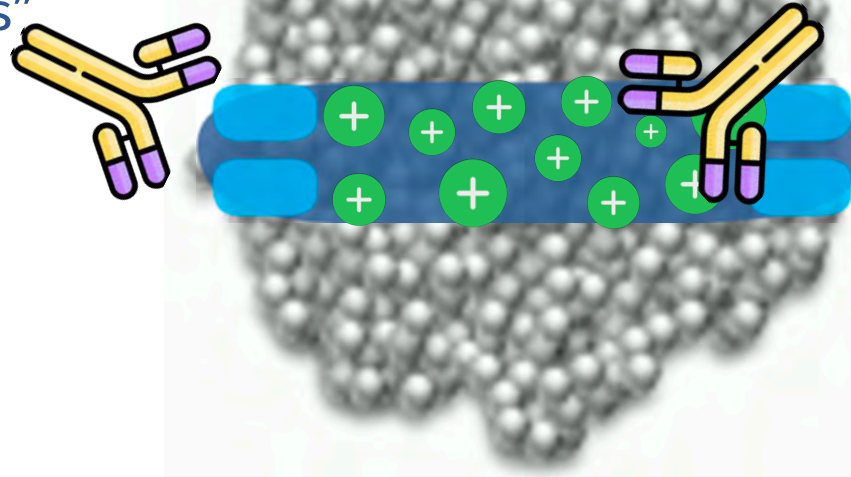
PF4/IgG Immune Complex

Heparin-independent antibody

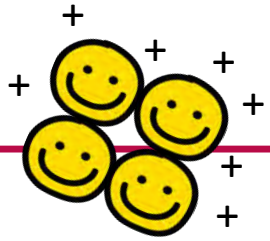
“VITT antibodies”

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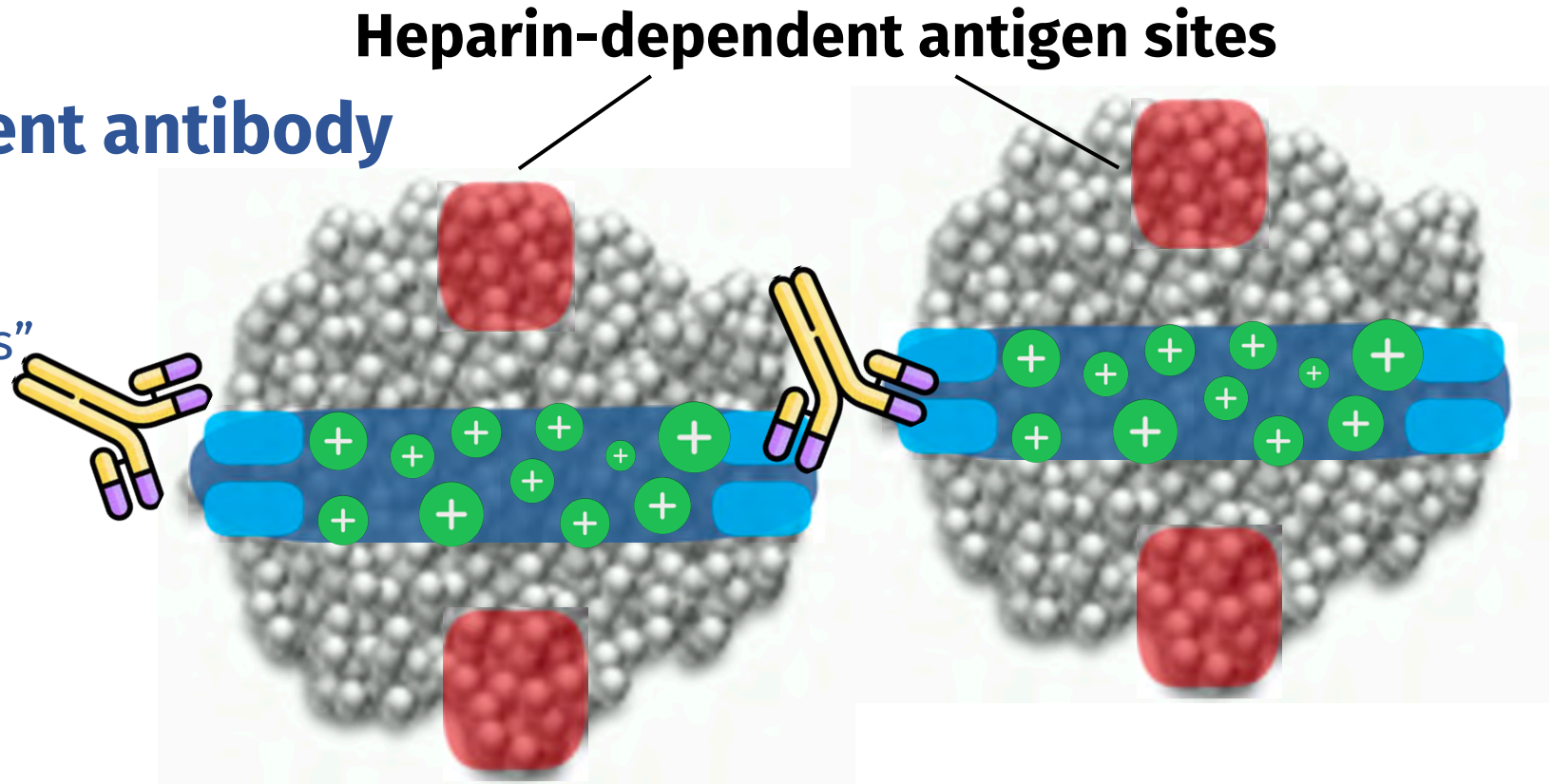
PF4/IgG Immune Complex

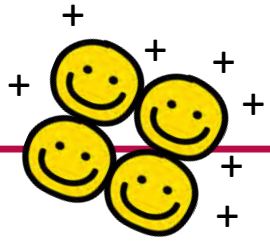
Heparin-independent antibody

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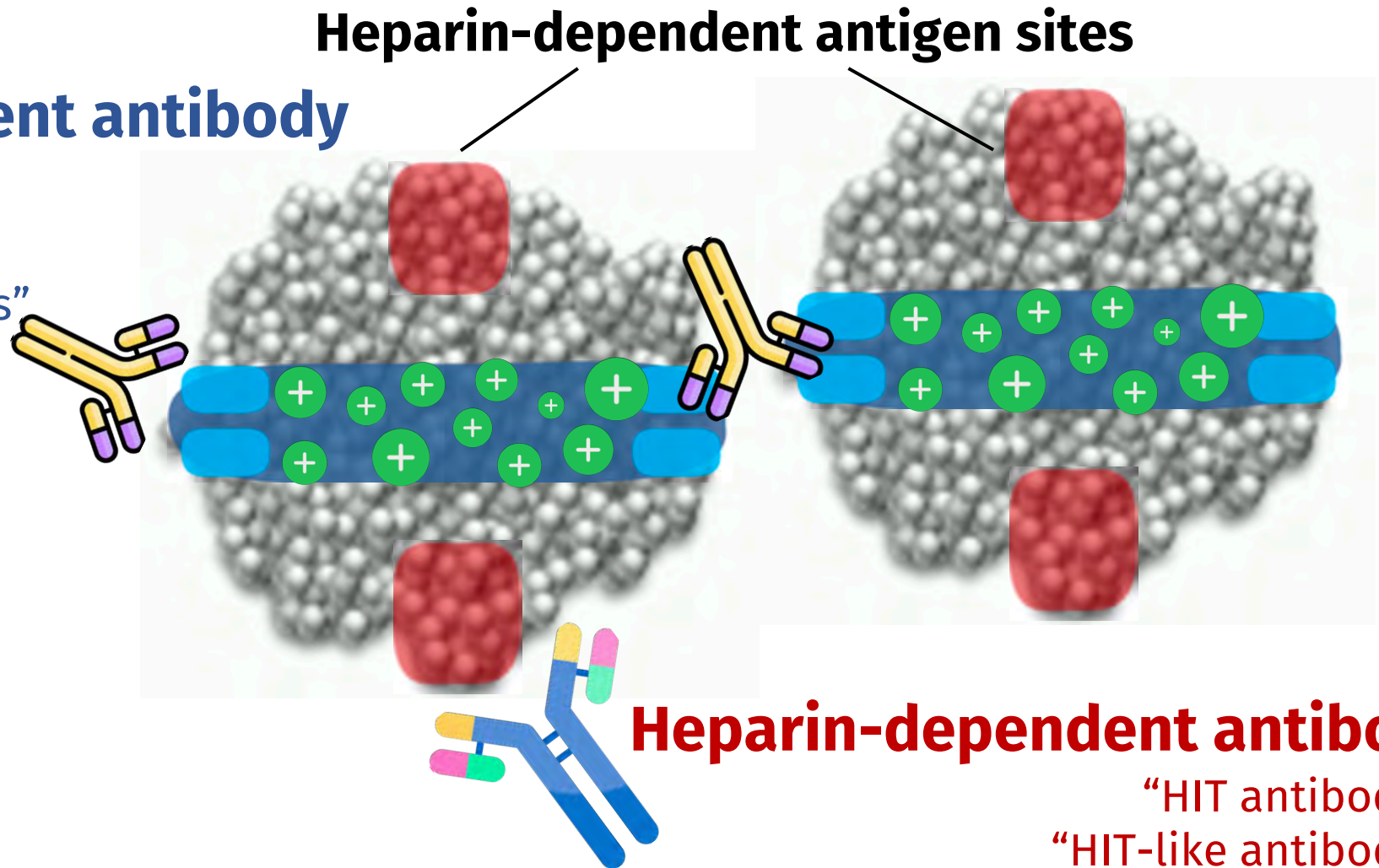
PF4/IgG Immune Complex

Heparin-independent antibody

“VITT antibodies”

“VITT-like antibodies”

“Type 3 Anti-PF4 antibodies”



- ❖ Autoimmune HIT
- ❖ Spontaneous HIT

Heparin-dependent antibody

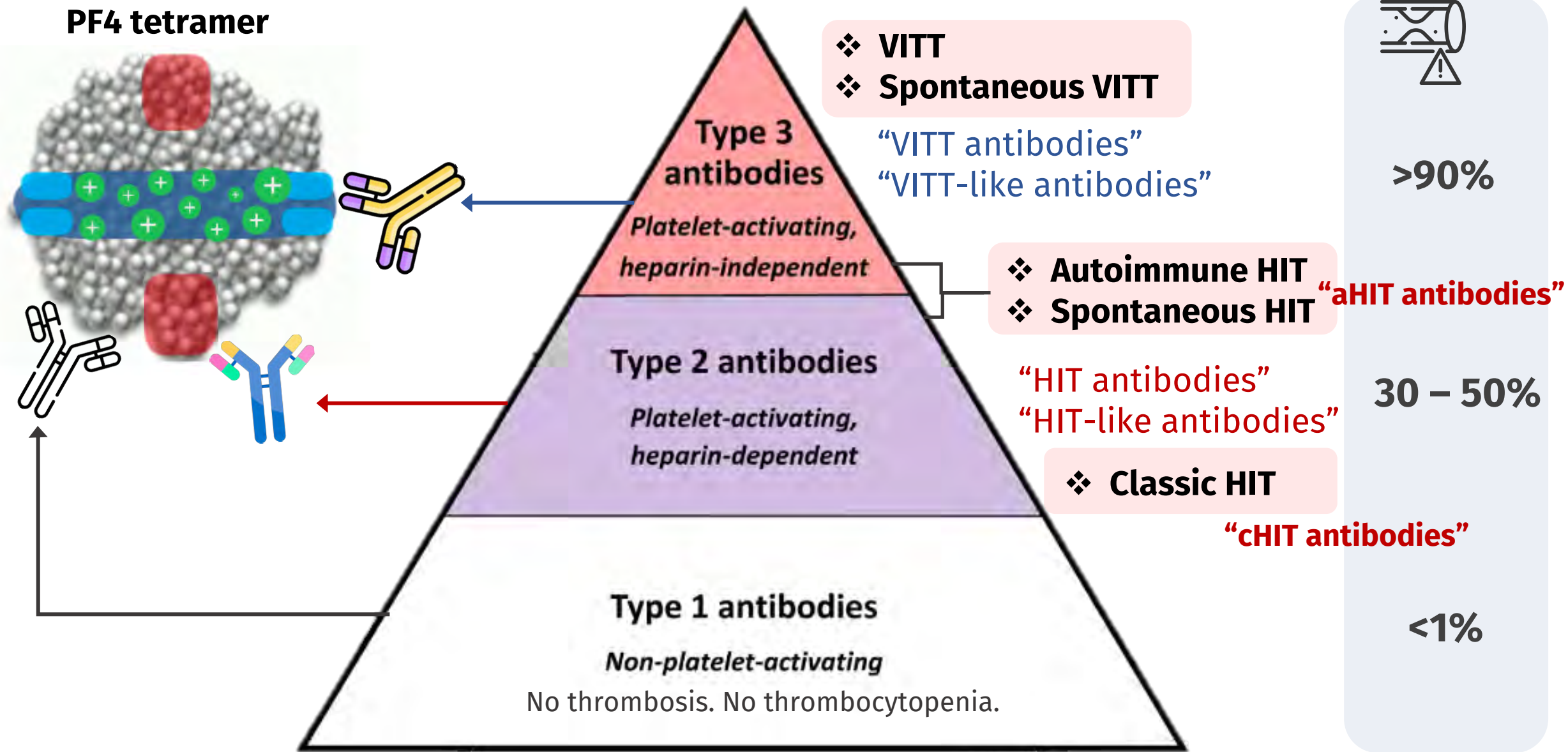
“HIT antibodies”

“HIT-like antibodies”

“Type 2 Anti-PF4 antibodies”

Anti-PF4 Antibodies

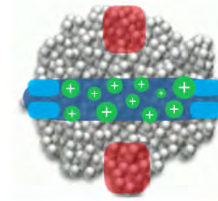
Thrombosis risk



HIT antibodies vs. VITT antibodies: Similarities and Differences



HIT antibodies
Heparin dependent



VITT antibodies
Heparin independent

Pathogenicity	- Binds to FcγR1a on platelets → platelet activation and clearance - Pan cellular activation (monocytes, neutrophils, endothelium)	
Type of immune response	Secondary immune response, IgG class	
Persistence	Transient (Platelet-activating: HIT Ab 6 wk, VITT Ab 18 wk)	
Trigger	Heparin or polyanion	Vaccines, adenovirus, paraprotein
Binding site on PF4	North and south poles	Equator (heparin binding site)
Binding forces to PF4	Lower (60 – 100 pN)	Stronger (>100 pN)
Clonality	Polyclonal	Monoclonal or oligoclonal
Frequency	Rare (0.1 – 10%)	Ultra-rare (0.0005% - 0.002%)
Thrombosis risk	High (30-50%) VTE, arterial	Ultra high (>90%) CVST, splanchnic, multiple sites VTE, arterial
Disorders	Classic HIT , aHIT, spHIT	VITT , SpVITT , aHIT, spHIT

Outline

- PF4 and anti-PF4 antibodies
- **Laboratory testing for anti-PF4 antibodies**
- Clinical spectrum of anti-PF4 disorders: Diagnosis & treatment

Laboratory Testing for Anti-PF4 Antibodies

Screening/ Immunoassays:

Detection of anti-PF4/heparin antibodies

Confirmatory/ Functional assays:

Determine if antibodies are pathogenic (platelet activating)

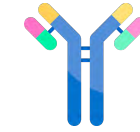
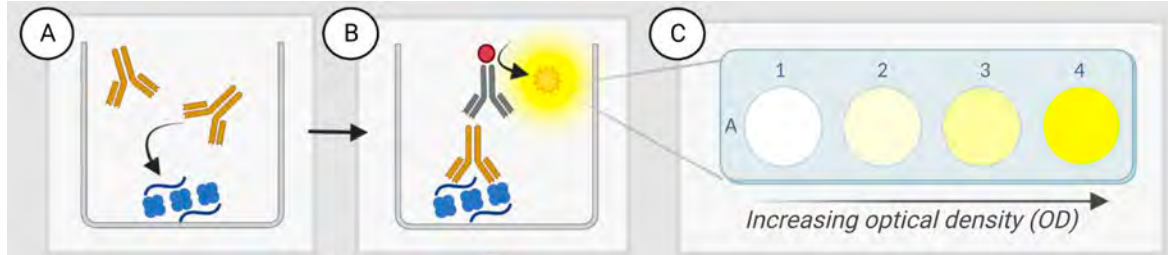
Laboratory Testing for Anti-PF4 Antibodies: Screening

Screening/ Immunoassays:

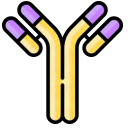
Detection of anti-PF4/heparin antibodies

EIA (ELISA)

IgG-specific or
poly-specific
(IgGAM)



HIT antibodies



VITT antibodies

Sensitivity

>99%



>99%

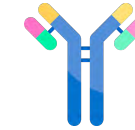
Strongly positive



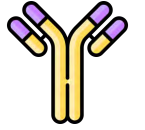
Laboratory Testing for Anti-PF4 Antibodies: Screening

Screening/ Immunoassays:

Detection of anti-PF4/heparin antibodies



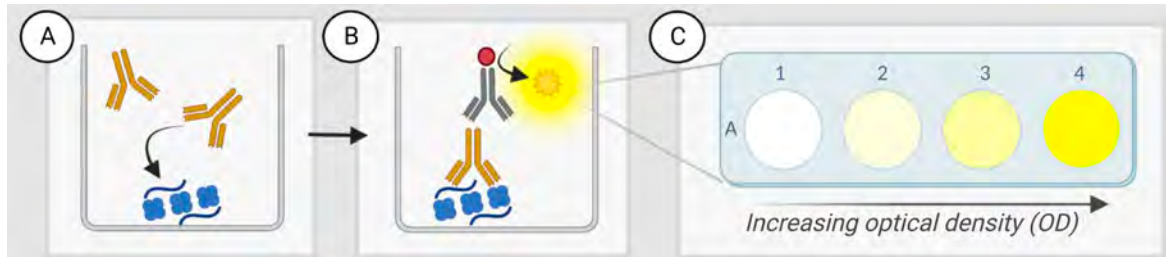
HIT antibodies



VITT antibodies

EIA (ELISA)

IgG-specific or
poly-specific
(IgGAM)



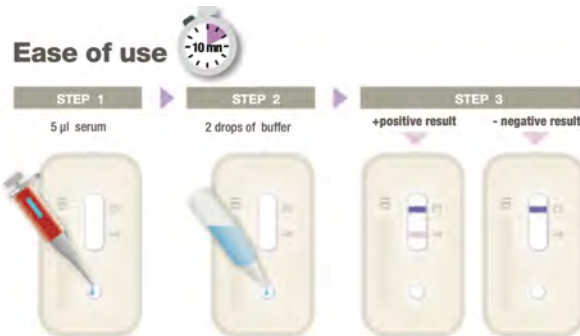
Sensitivity

>99%

>99%

Strongly positive

Rapid immunoassays



Particle gel immunoassay ³²	PaGIA		Polymers coated with PF4/heparin complexes
IgG-specific chemiluminescence assay ³²	IgG CIA		Magnetic particles coated with PF4/polyvinyl sulfonate
Lateral flow immunoassay ³²	LFIA		PF4-polyanion complexes
Latex immunoturbidimetric assay ³³	LIA		Competitive inhibition of nanoparticles coated with HIT-like antibodies

>95%

~45%

~95%

<5%

~90%

~10%

~95%



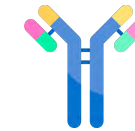
<5%

**Not sensitive for
VITT antibodies!**

Laboratory Testing for Anti-PF4 Antibodies: Screening

Screening/ Immunoassays:

Detection of anti-PF4/heparin antibodies



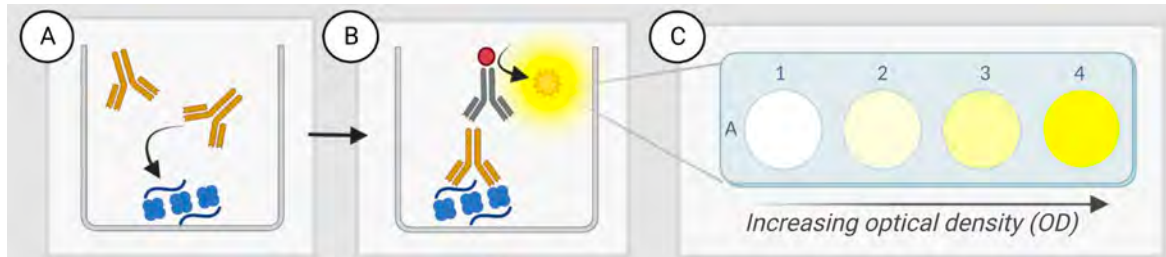
HIT antibodies



VITT antibodies

EIA (ELISA)

IgG-specific or
poly-specific
(IgGAM)



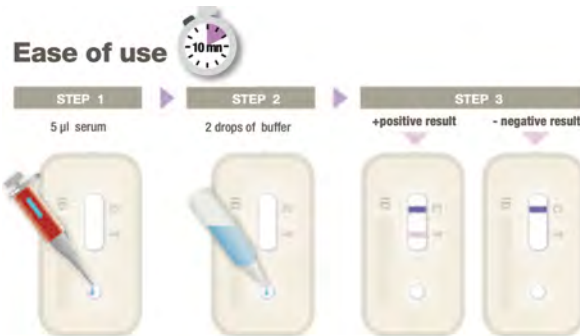
Sensitivity

>99%

>99%

Strongly positive

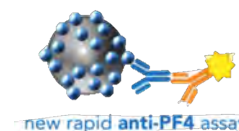
Rapid immunoassays



Particle gel immunoassay ³²	PaGIA		Polymers coated with PF4/heparin complexes	>95%	~45%
IgG-specific chemiluminescence assay ³²	IgG CIA		Magnetic particles coated with PF4/polyvinyl sulfonate	~95%	<5%
Lateral flow immunoassay ³²	LFIA		PF4-polyanion complexes	~90%	~10%
Latex immunoturbidimetric assay ³³	LIA		Competitive inhibition of nanoparticles coated with HIT-like antibodies	~95%	<5%



CLIA – Anti-PF4 (2023)



~30%

>95%

Laboratory Testing for Anti-PF4 Antibodies: Confirm

Confirmatory/ Functional assays:

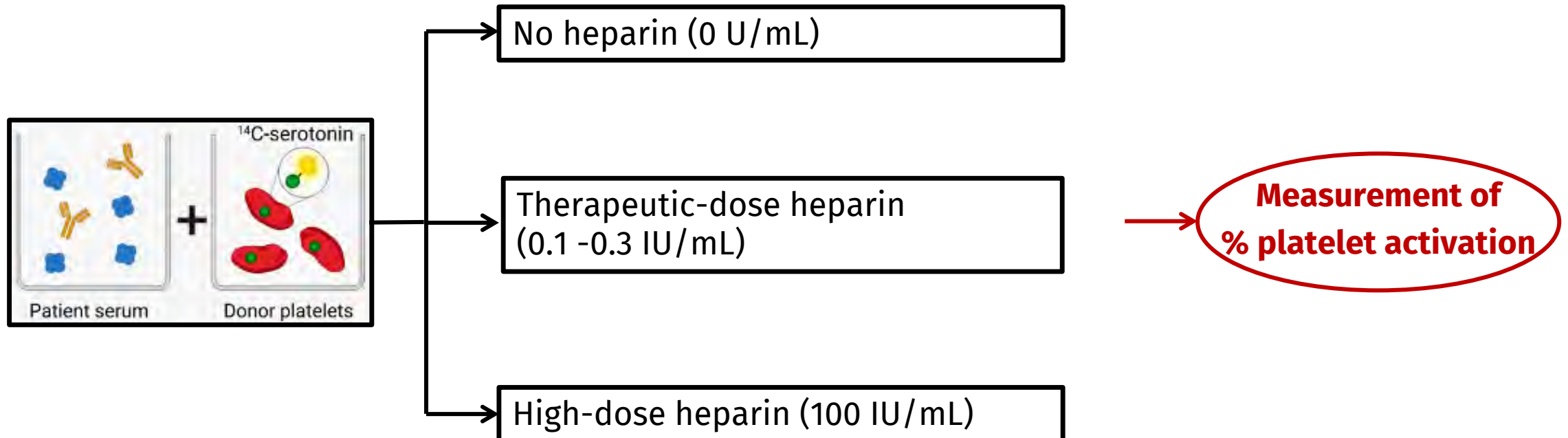
Determine if antibodies are pathogenic (platelet activating)

Serotonin release assay (SRA)

Heparin-induced platelet activation (HIPA)

Platelet aggregometry

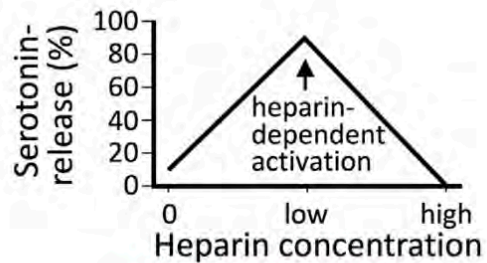
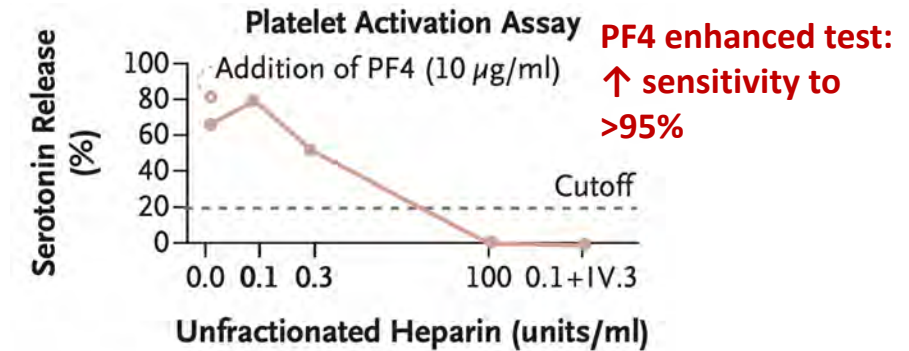
P-selectin expression assay (PEA)



Laboratory Testing for Anti-PF4 Antibodies: Confirm

Confirmatory/ Functional assays:

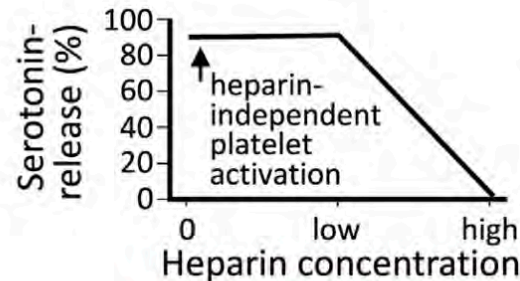
Determine if antibodies are pathogenic (platelet activating)



Classic HIT

HIT antibodies

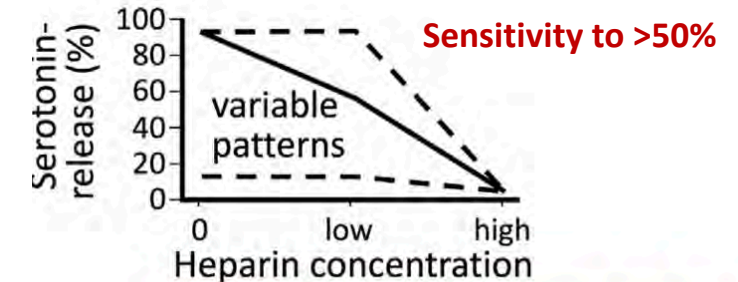
Sensitivity to >95%



Autoimmune HIT

Spontaneous HIT

HIT antibodies + VITT antibodies



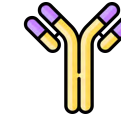
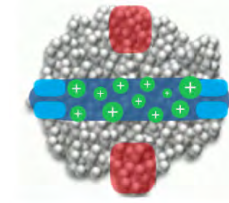
VITT / Spontaneous VITT

VITT antibodies

HIT antibodies vs. VITT antibodies: Laboratory tests



HIT antibodies
Heparin dependent



VITT antibodies
Heparin independent

Rapid immunoassay	PF4/H + (New rapid anti-PF4 assays -/+)	PF4/H -/ + (New rapid anti-PF4 assays +)
EIA (ELISA)	+, moderate to strong (Fluid phase PF4/H+ , PF4 -)	+, strong ODs Fluid phase PF/H-, PF4 ++)
Platelet activation tests		
No heparin	-	-/+ (increased with PF4 addition)
Therapeutic dose	+	-/+ (may show inhibiting effect of heparin)
High dose	-	-

Table 2. Getting to know your institution's HIT assays

- ☒ HIT antibody (screening)
- ☒ HIT antibody (confirm)

Assay type	What questions should you ask your coagulation laboratory?	Why is it important to know?
Screening assay(s) 	Which assay(s) do we use to screen for HIT?	Many options exist. EIAs are the prototypical immunoassay. Rapid immunoassays also exist.
	What is the sensitivity and specificity of our assay(s)?	Every assay has unique performance characteristics. Understanding the performance characteristics of an assay is essential in clinical risk prediction.
	If we have access to an EIA, tell me more about it: Do we report the OD? Is it IgG specific or not?	The likelihood of HIT increases with a higher OD. Specificity is higher with an IgG-only EIA.
	What is the turnaround time of our assay(s)?	Some assays take minutes to run; some take hours. Some labs batch samples so results can take days to return. Knowing when to expect results can help inform management.
Functional assay(s)	Which assay(s) do we use?	Options exist, although fewer than screening assays. SRA is considered the "gold standard," but others exist, including HIPA and PEA.
	Do we use laboratory techniques to remove any residual heparin from the sample prior to performing the assay?	Residual heparin can interfere with assay performance. If not removed, additional care must be taken to ensure the sample is collected at least 1 hour after heparin is stopped.
	What is the sensitivity and specificity of our assay(s)?	Every assay has unique performance characteristics. Even within assay classes, performance can vary by lab. Understanding the performance characteristics of an assay is essential in clinical risk prediction.
	What is the turnaround time of our assay(s)?	Most institutions do not run functional assays in-house. Knowing when to expect results can help inform management.

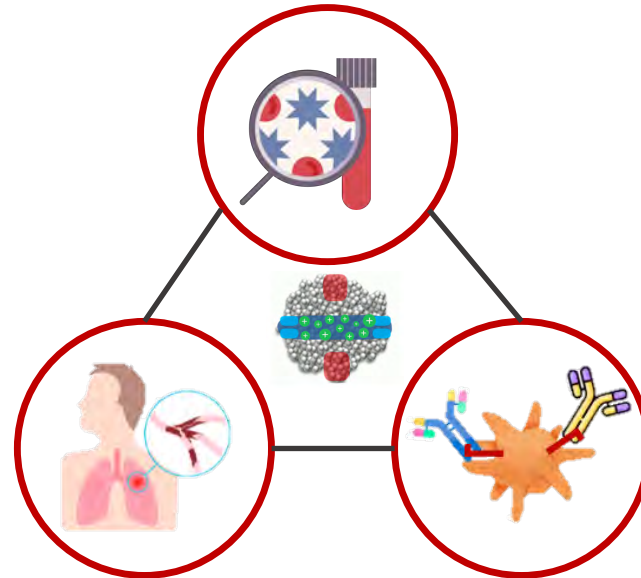
Outline

- PF4 and anti-PF4 antibodies
- Laboratory testing for anti-PF4 antibodies
- **Clinical spectrum of anti-PF4 disorders: Diagnosis & treatment**

The Spectrum of Anti-PF4 Disorders

Anti-PF4 Disorders

Thrombocytopenia



Thrombosis

Platelet-activating
anti-PF4 antibodies
(via FcγRIIa)

The Spectrum of Anti-PF4 Disorders

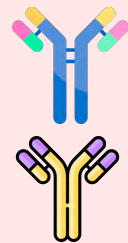
Heparin triggered



1973

Classic HIT

- **Triggers:** heparin, LMWH, polyanions



2001

Autoimmune HIT

- **Triggers:** heparin, LMWH, fondaparinux



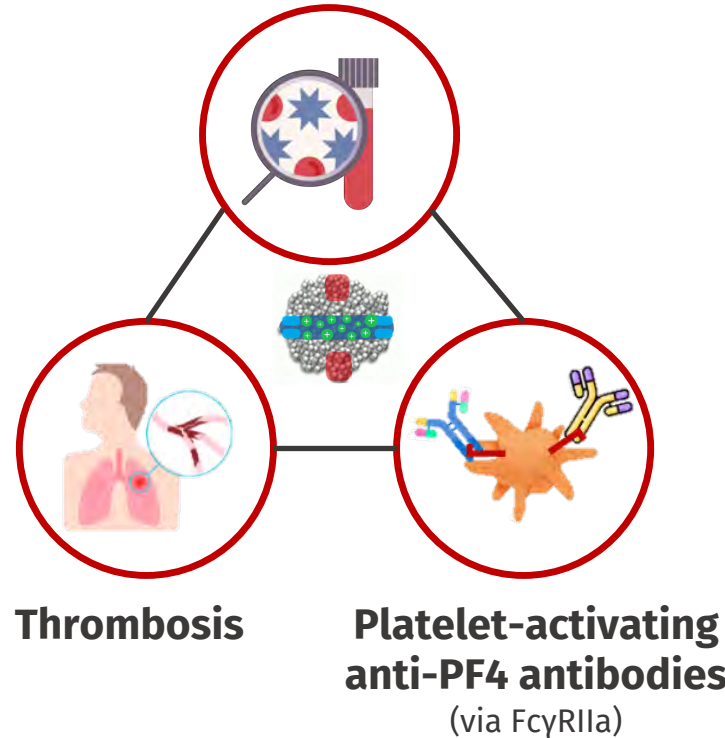
= Heparin dependent



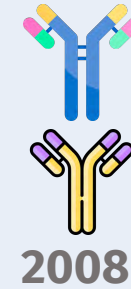
= Heparin independent

Anti-PF4 Disorders

Thrombocytopenia



Non-heparin triggered



2008

Spontaneous HIT

- **Triggers:** total knee arthroplasty, infection



2021

VITT

- **Trigger:** adenovirus vector vaccines



2022

Spontaneous VITT

- **Triggers:** adenovirus infection, chronic autoimmune disorder, MGTS

The Spectrum of Anti-PF4 Disorders

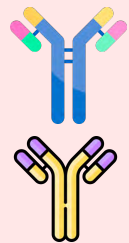
Heparin triggered



1973

Classic HIT

- **Triggers:** heparin, LMWH, polyanions



2001

Autoimmune HIT

- **Triggers:** heparin, LMWH, fondaparinux



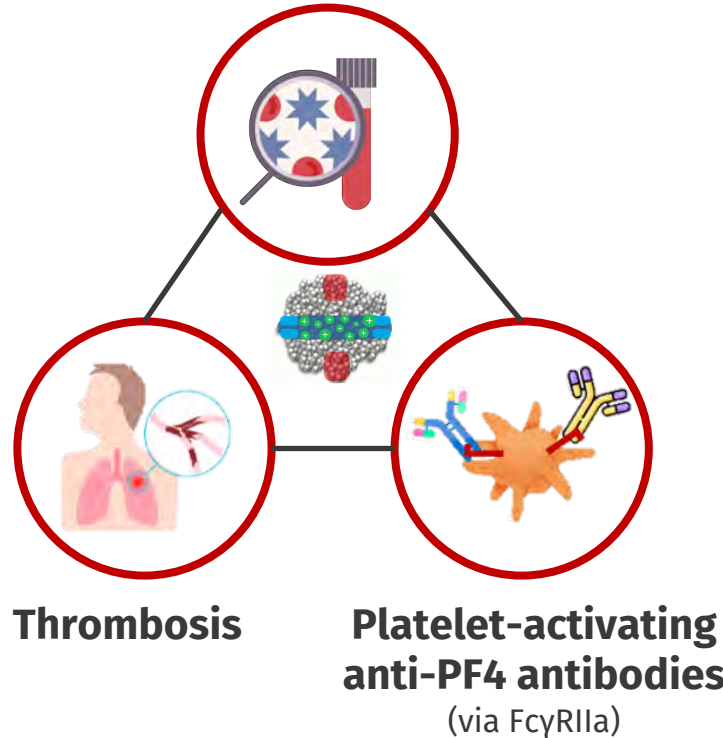
= Heparin dependent



= Heparin independent

Anti-PF4 Disorders

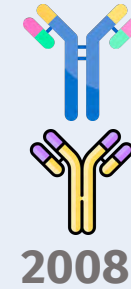
Thrombocytopenia



Thrombosis

Platelet-activating
anti-PF4 antibodies
(via FcγRIIa)

Non-heparin triggered



2008

Spontaneous HIT

- **Triggers:** total knee arthroplasty, infection



2021

VITT

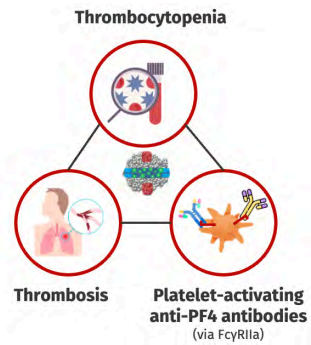
- **Trigger:** adenovirus vector vaccines



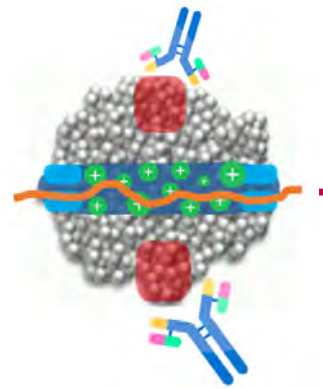
2022

Spontaneous VITT

- **Triggers:** adenovirus infection, chronic autoimmune disorder, MGTS

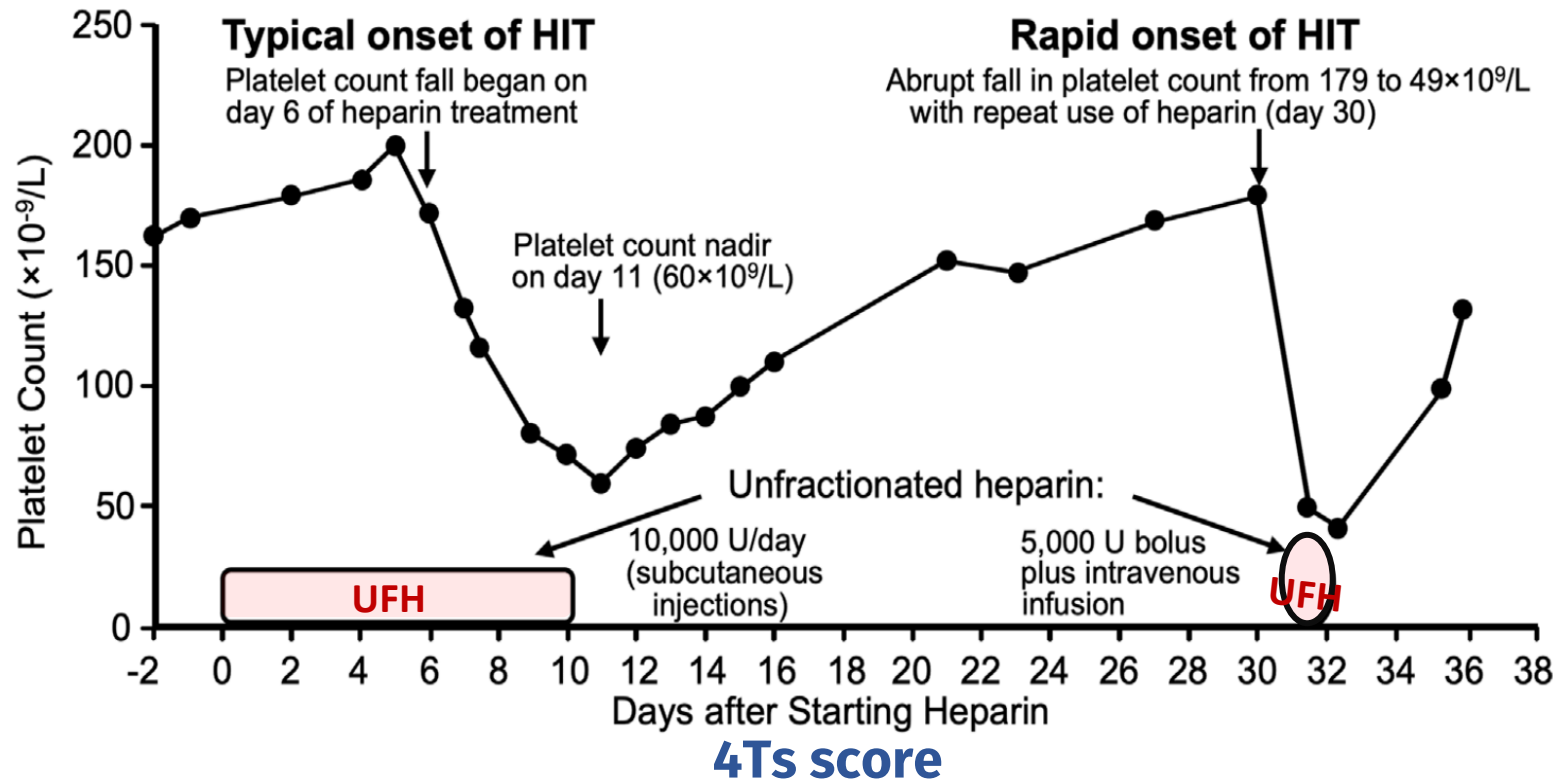


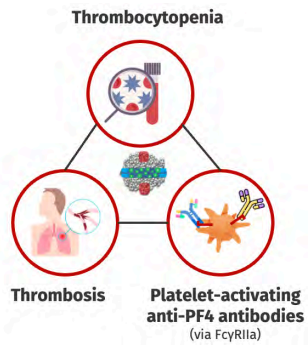
Classic HIT



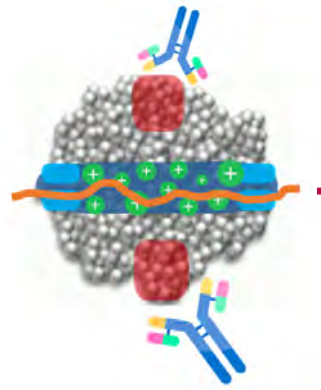
- Antibody pathogenicity **requires the presence of heparin**
- Thrombocytopenia should **only occur if the patient continues to receive heparin and should resolve quickly upon stopping heparin.**

Trigger: heparin or polyanions



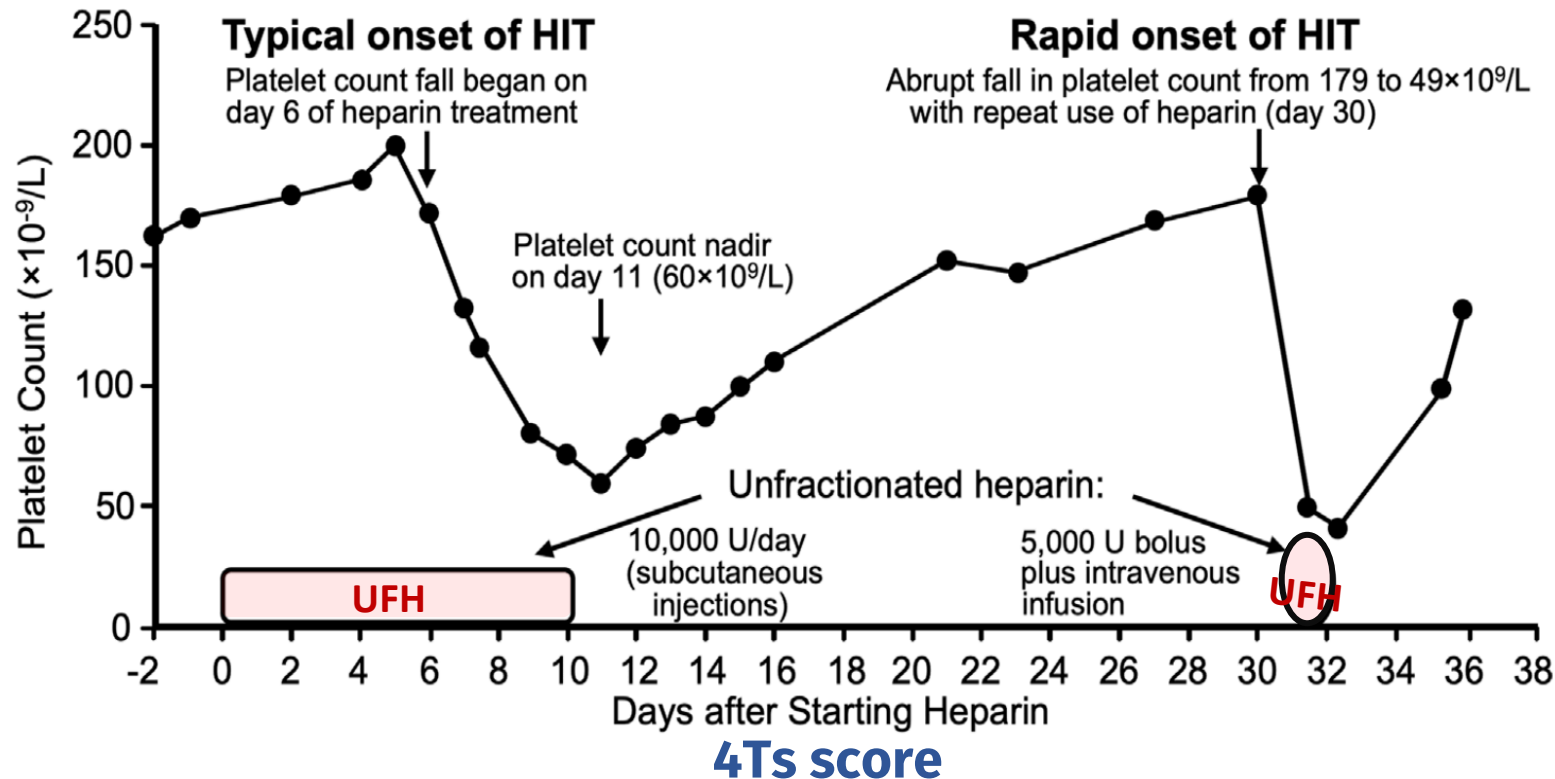


Classic HIT



- Antibody pathogenicity **requires the presence of heparin**
- Thrombocytopenia should **only occur if the patient continues to receive heparin and should resolve quickly upon stopping heparin.**

Trigger: heparin or polyanions



Thrombosis rate



30 – 50%

Venous > arterial (4:1)

DVT (lower) 40-50%

PE 25%

Arterial 10-15% (Limb > stroke > MI)

Adrenal 1-2%

Splachnic 1-2%

CVST 1%

Injection site necrosis 1%

Multiple <10%

The Spectrum of Anti-PF4 Disorders

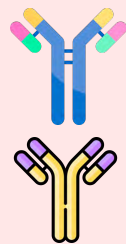
Heparin triggered



1973

Classic HIT

- **Triggers:** heparin, LMWH, polyanions



2001

Autoimmune HIT

- **Triggers:** heparin, LMWH, fondaparinux



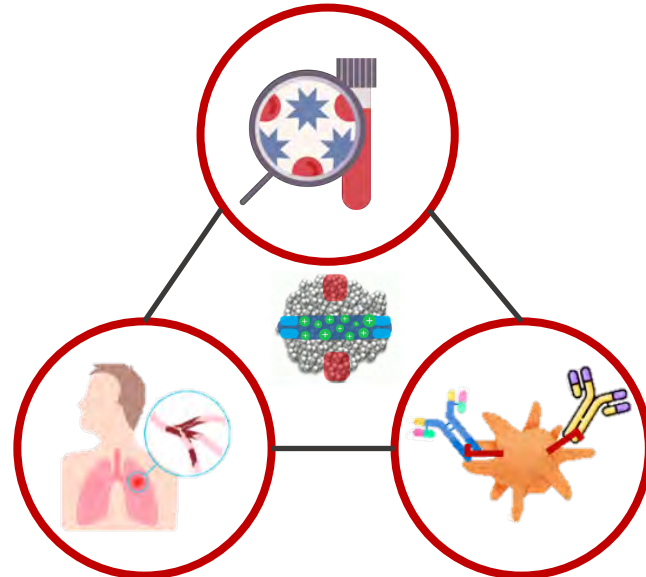
= Heparin dependent



= Heparin independent

Anti-PF4 Disorders

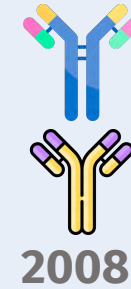
Thrombocytopenia



Thrombosis

Platelet-activating
anti-PF4 antibodies
(via FcγRIIa)

Non-heparin triggered



2008

Spontaneous HIT

- **Triggers:** total knee arthroplasty, infection



2021

VITT

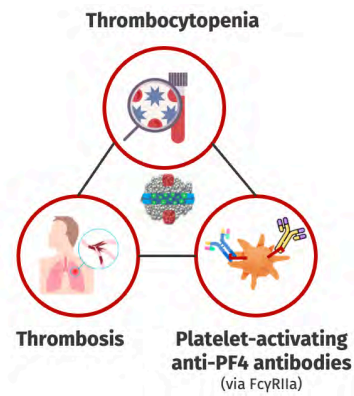
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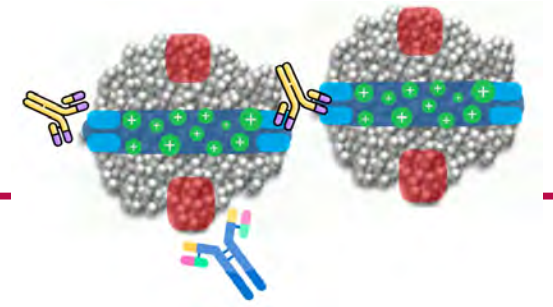
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Spontaneous VITT

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Autoimmune HIT (aHIT)

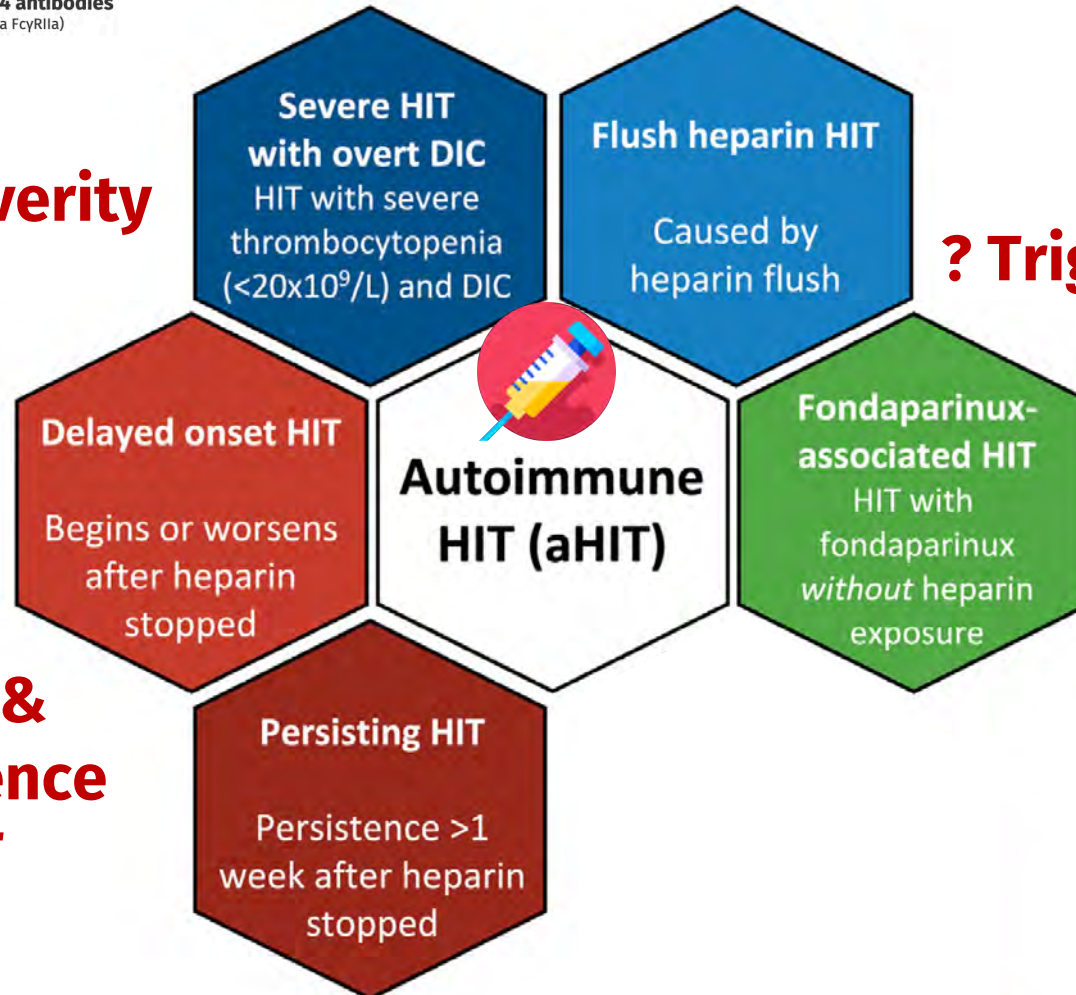


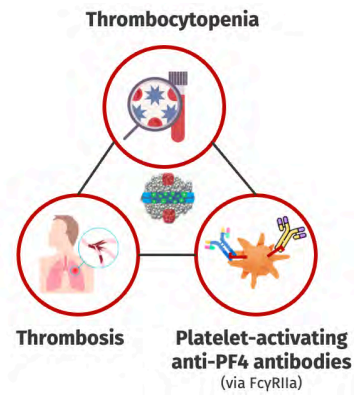
Proximate heparin exposure but....

? Severity

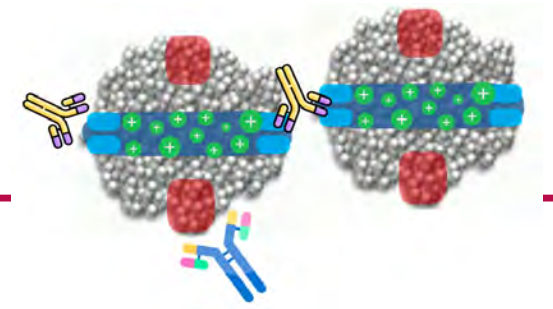
? Trigger

? Onset & persistence of ↓PLT

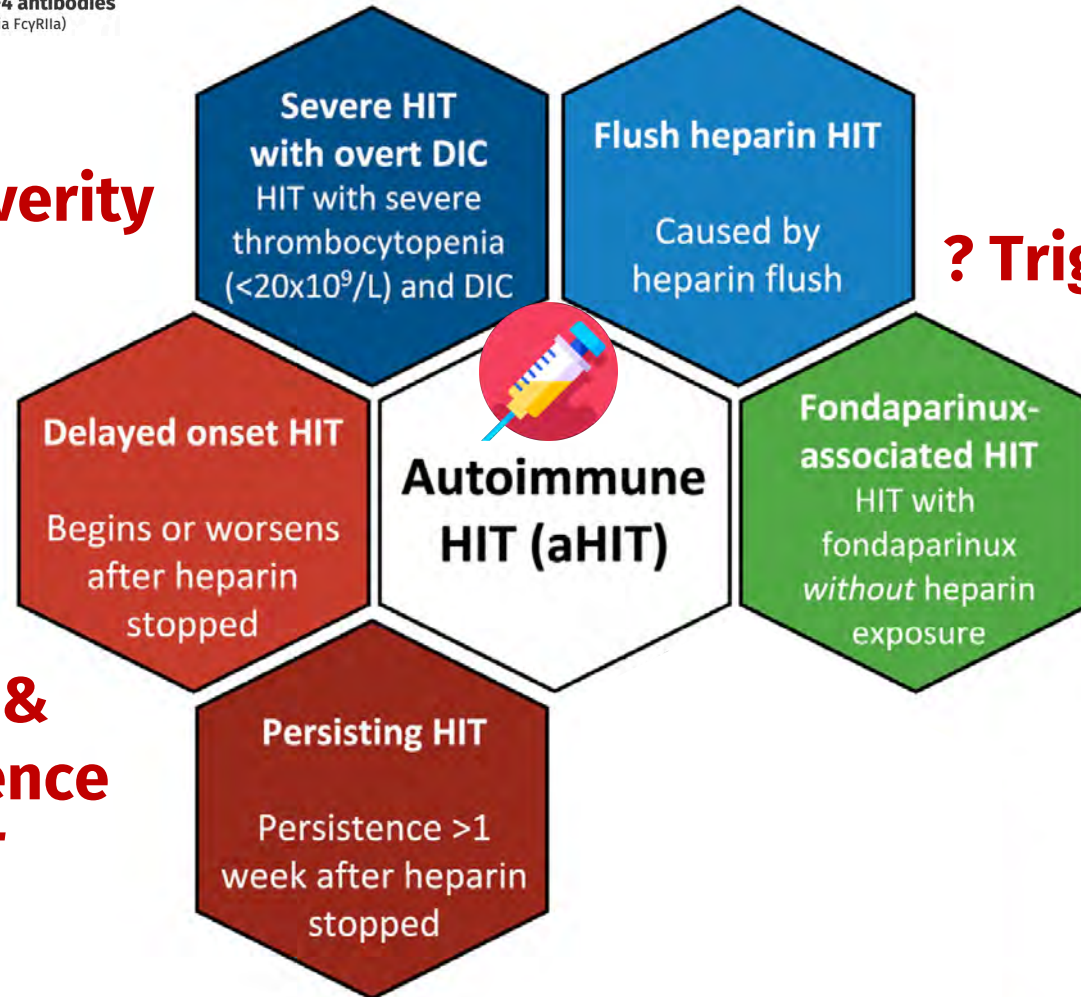




Autoimmune HIT (aHIT)



Proximate heparin exposure but....



- High frequency of thrombosis (~90%)
 - DVT/PE
 - Arterial
 - **Microvascular thrombosis & DIC**
- Variable, often severe thrombocytopenia ($<20 \times 10^9/L$)

Clinical Picture of Autoimmune HIT

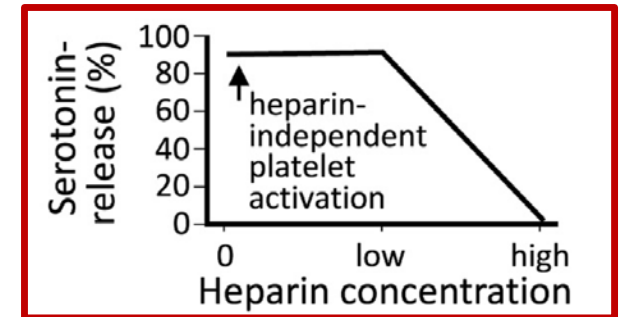
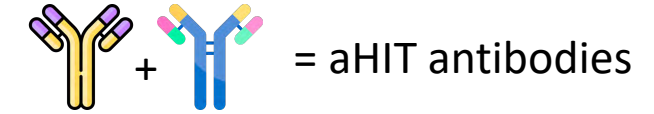
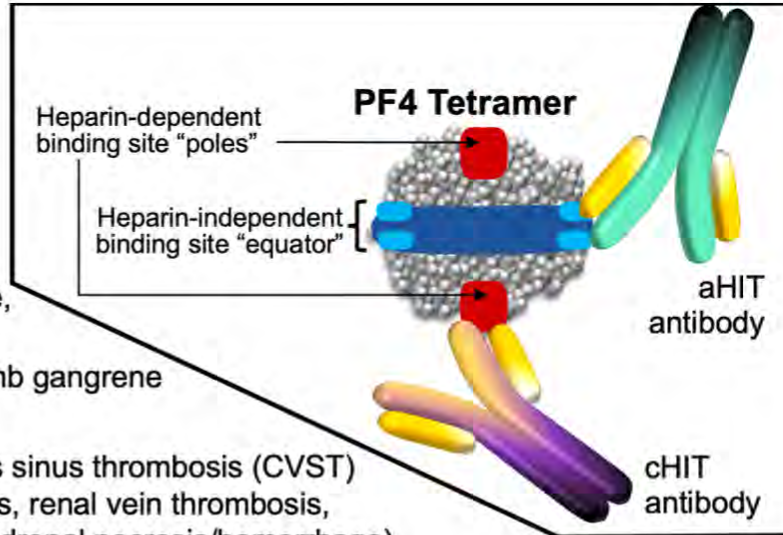
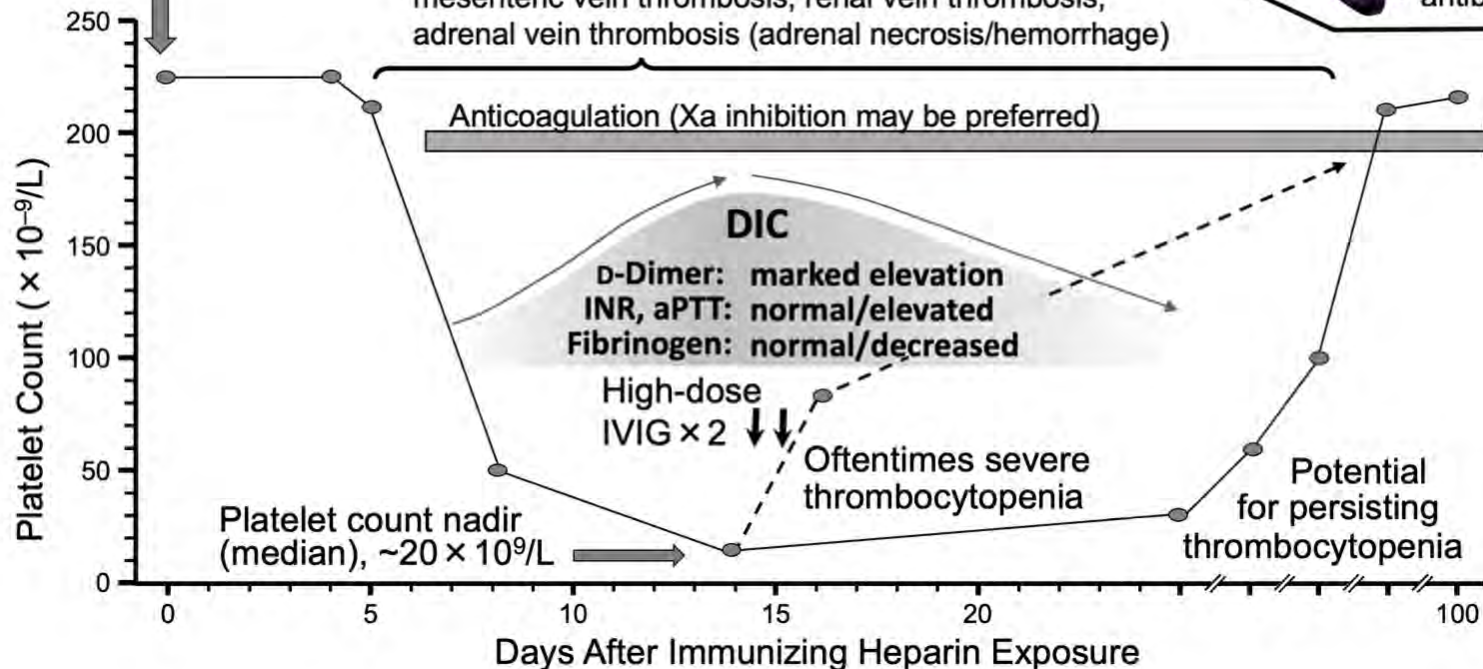
Anti-PF4 test positive:

PF4/polyanion ELISA
Rapid assays: e.g., CLIA, LIA
Serotonin-release assay (SRA)
(Positive at 0 and 0.1 – 0.5 U/mL)

Thrombotic event(s):

Venous, e.g., DVT, PE
Arterial, e.g., limb artery, stroke, myocardial infarction, etc.
Microvascular, e.g., venous limb gangrene
Multi-site thrombosis
Unusual, e.g., cerebral venous sinus thrombosis (CVST), mesenteric vein thrombosis, renal vein thrombosis, adrenal vein thrombosis (adrenal necrosis/hemorrhage)

Heparin or LMWH or fondaparinux



Serotonin-release assay (SRA)

UFH (U/mL)	0	0.1	0.3	100
Percent release	78	87	88	0

- Heparin cessation is not sufficient to interrupt aHIT-induced platelet activation
- High-dose IVIg is indicated**
- Plasma exchange if IVIg ineffective**

The Spectrum of Anti-PF4 Disorders

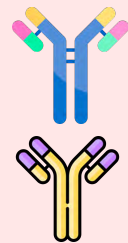
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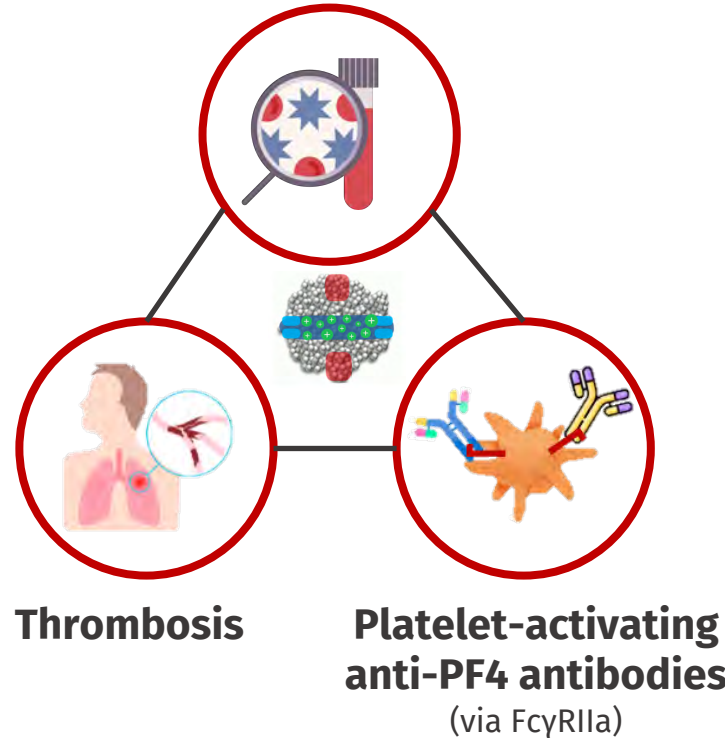
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Anti-PF4 Disorders

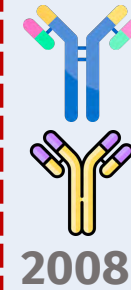
Thrombocytopenia



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(via FcγRIIa)

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- **Triggers:** total knee arthroplasty, infection



2021

VITT

- **Trigger:** adenovirus vector vaccines

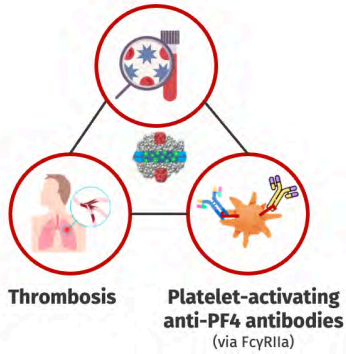
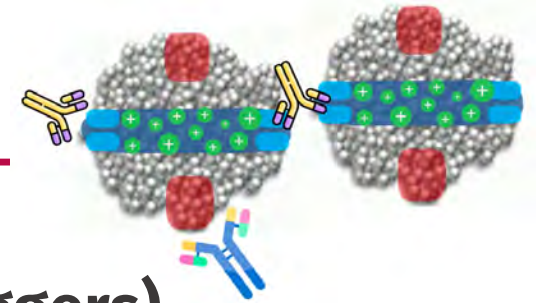


2022

Spontaneous VITT

- **Triggers:** adenovirus infection, chronic autoimmune disorder, MGTS

Spontaneous HIT (SpHIT)



- First reported in 2008
- **No proximate heparin exposure** (non-heparin triggers)

Surgical patients

Post-knee arthroplasty

- 24 cases reported (Frequency ~ 1 in 300,000)
- Onset 5-10 days after TKA
- Female predominance (70%)
- 100% thrombosis
 - DVT/PE 80%
 - **Adrenal vein thrombosis (hemorrhage/necrosis) 50%**
 - Arterial 13% (limb > stroke)
- **Chondroitin sulfate, polyanions (RNA, DNA) released in bolus after tourniquet use**

Thrombosis rate



95%

Severe
thrombocytopenia
<20 x 10⁹/L

Medical patients

Post-infection

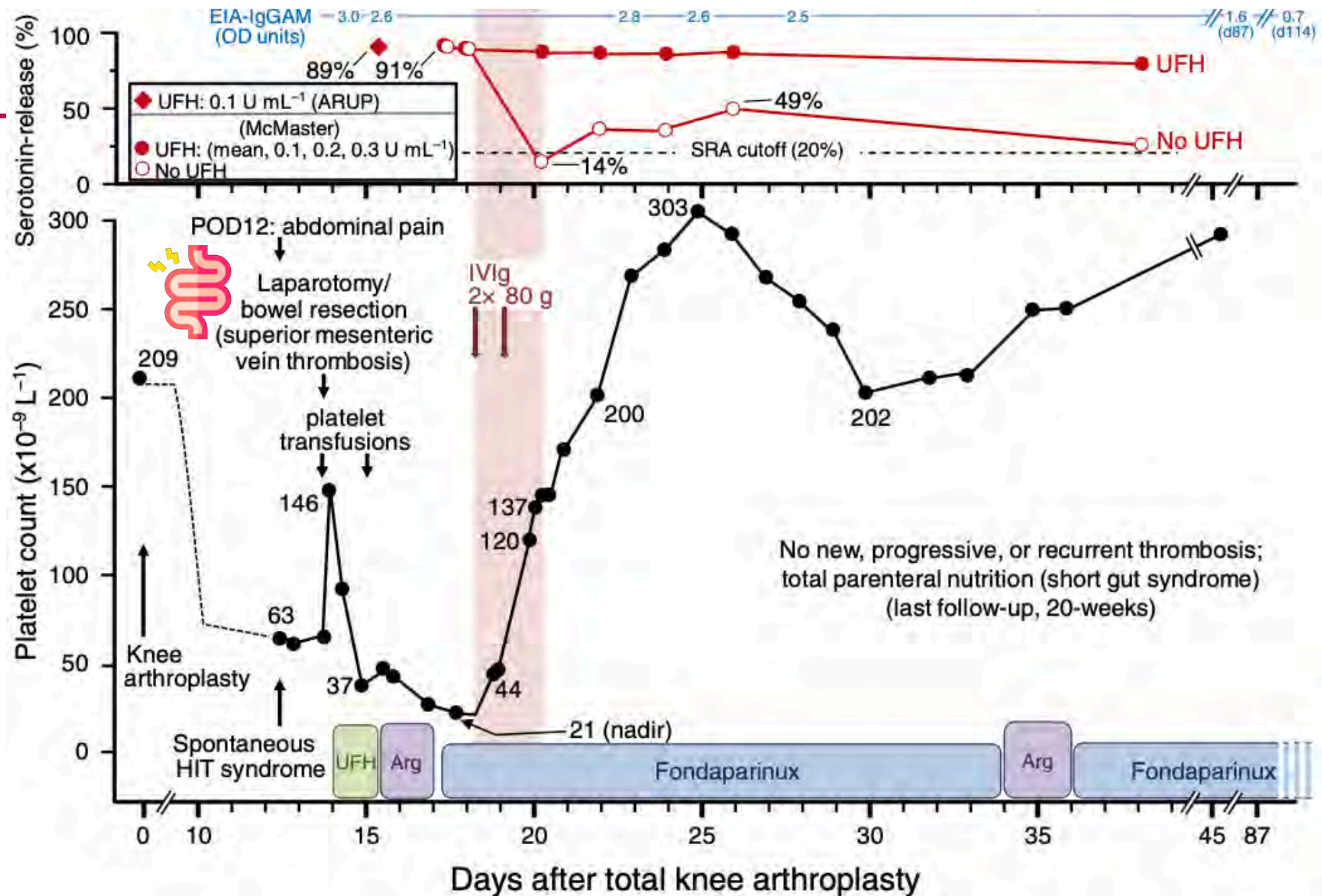


- 15 cases reported (virus & gram negative bacteria infection)
- **High frequency of stroke (ischemic 20% and CVST 40%)**
- Rapid-onset spontaneous HIT syndrome
- **DNA, RNA, lipopolysaccharide, GAGs**

Idiopathic



- No identifiable trigger
- Associated with **IgG monoclonal gammopathy**

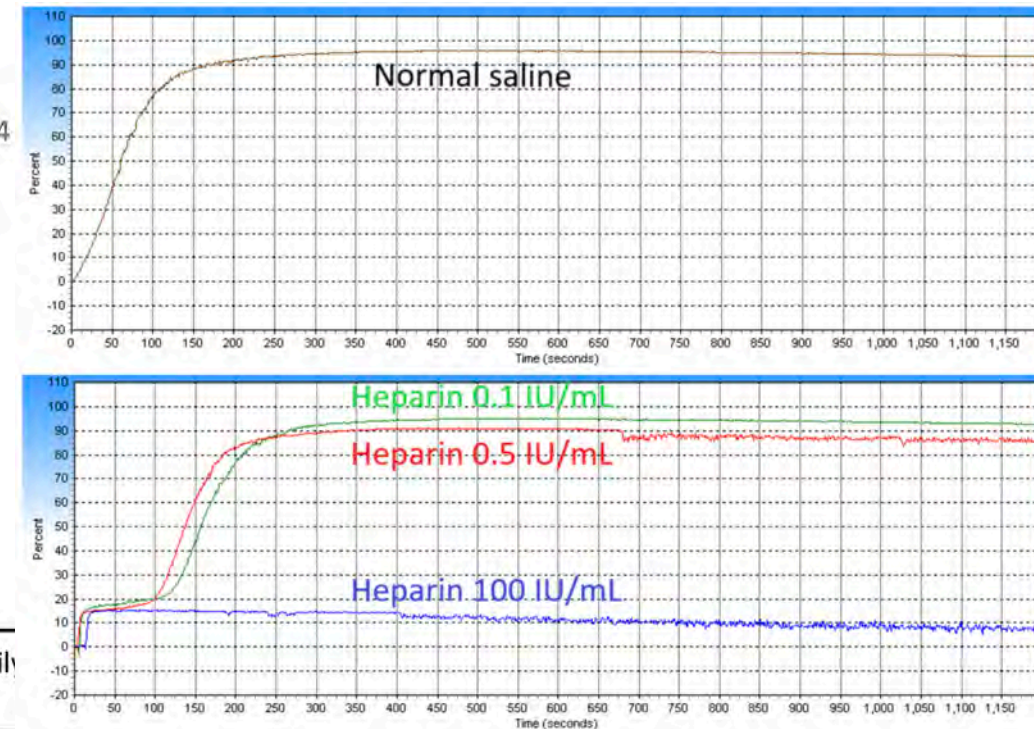
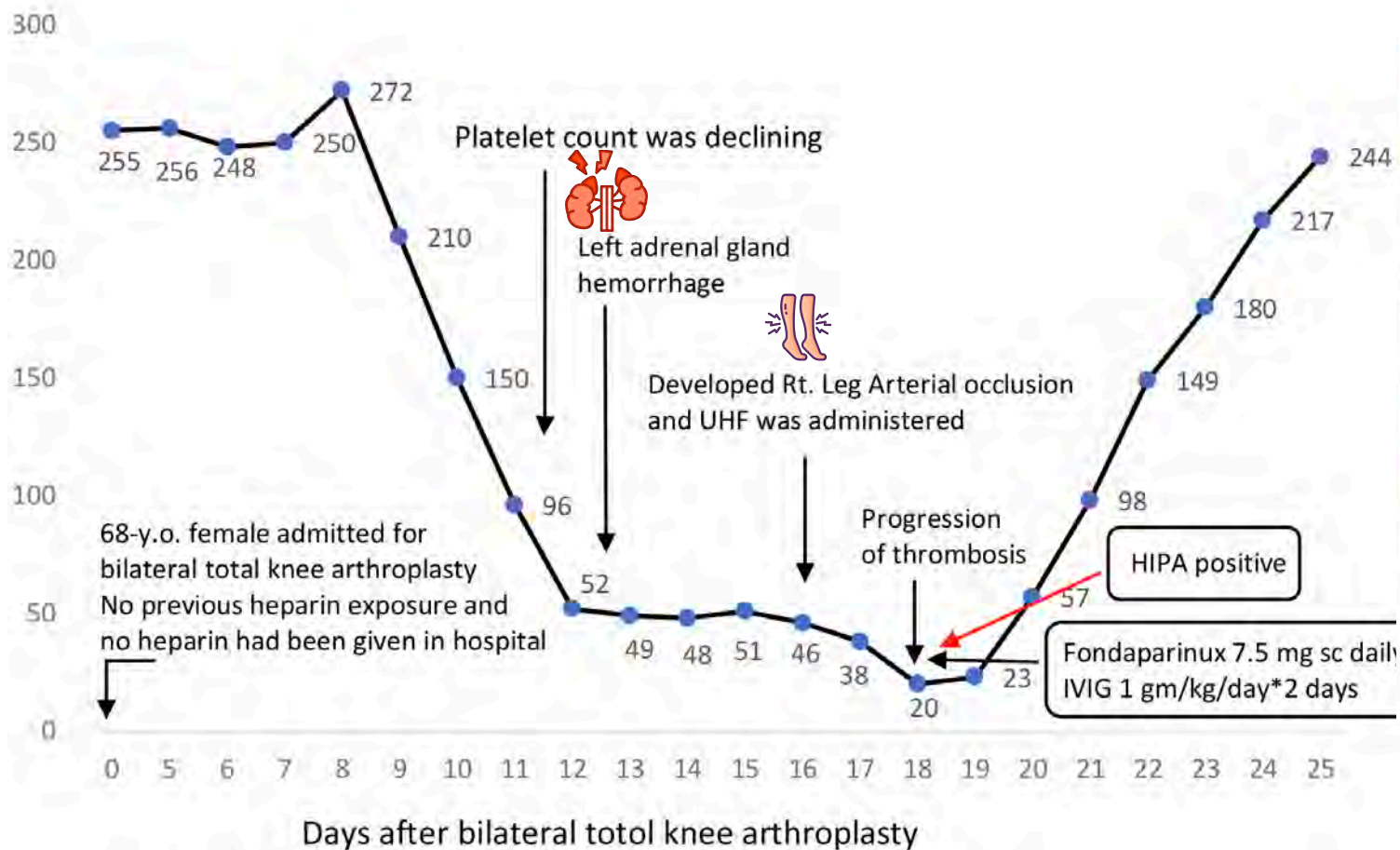


Spontaneous heparin-induced thrombocytopenia with adrenal haemorrhage following orthopaedic surgery: a case report and literature review

Supawee Saengboon ¹, Yingyong Chinthammitr,² Nonglak Kanitsap¹

October 2021

- A 68-year-old woman
- POD 12 after TKA
- No heparin exposure



The Spectrum of Anti-PF4 Disorders

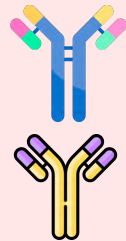
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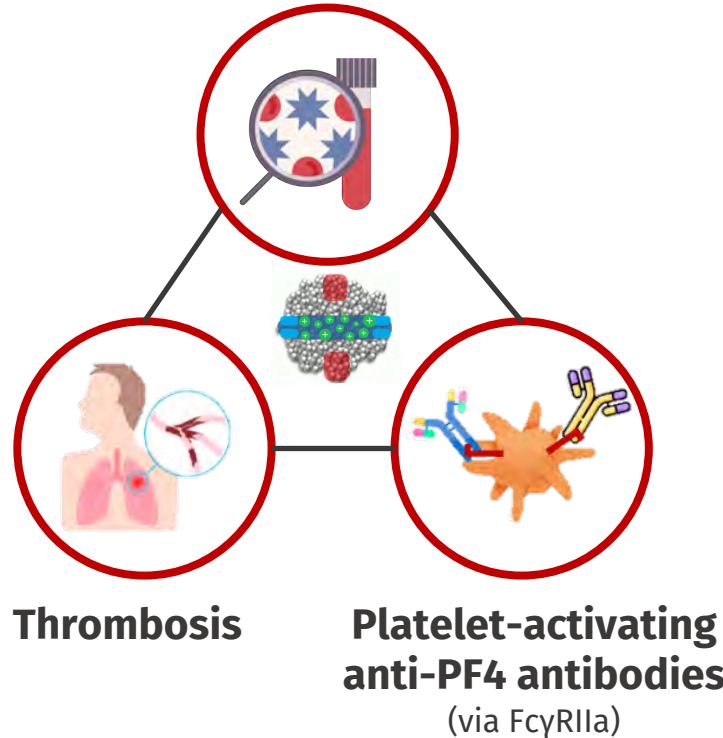
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Anti-PF4 Disorders

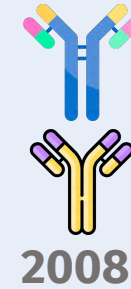
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VITT

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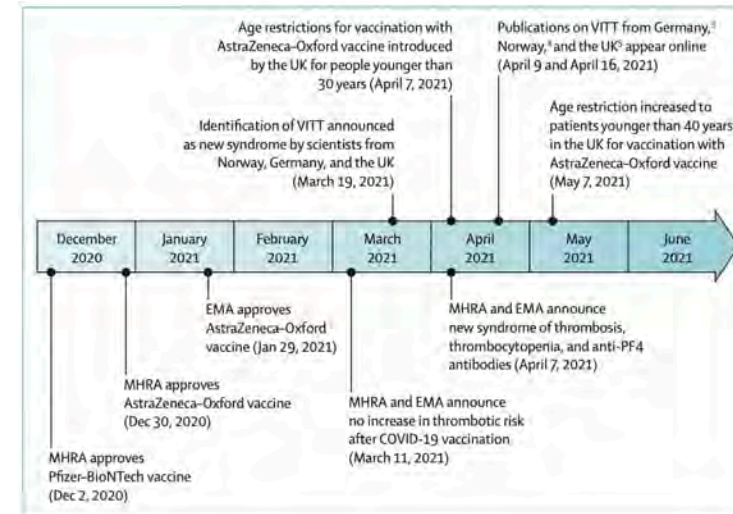


2022

Spontaneous VITT

- **Triggers:** adenovirus infection, chronic autoimmune disorder, MGTS

March 2021



The NEW ENGLAND JOURNAL of MEDICINE

ORIGINAL ARTICLE

Thrombotic Thrombocytopenia after ChAdOx1 nCov-19 Vaccination

Andreas Greinacher, M.D., Thomas Thiele, M.D., Theodore E. Warkentin, M.D., Karin Weisser, Ph.D., Paul A. Kyrle, M.D., and Sabine Eichinger, M.D.

The NEW ENGLAND JOURNAL of MEDICINE

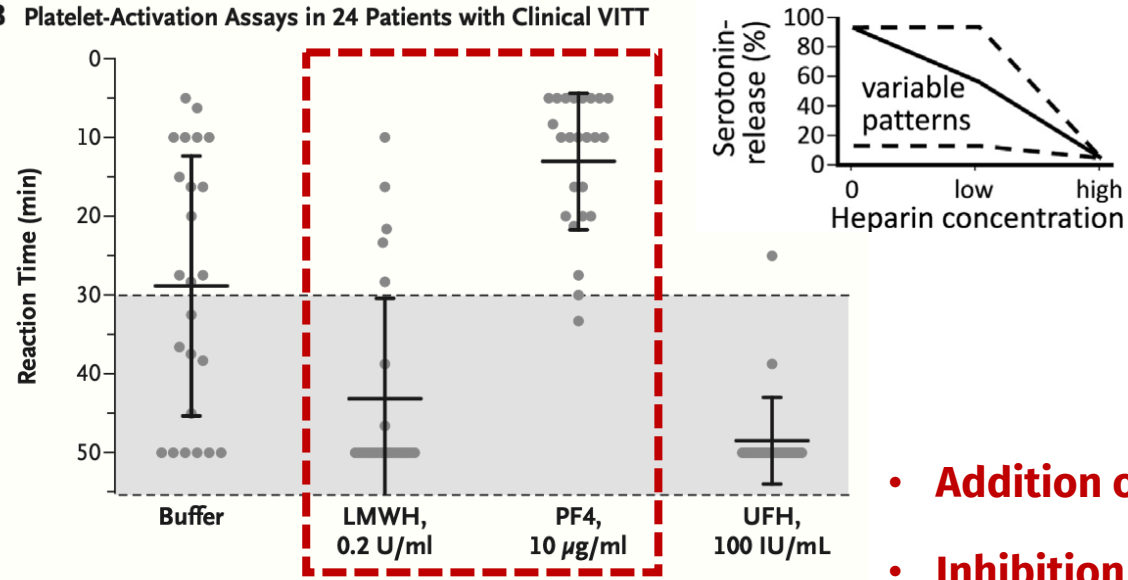
BRIEF REPORT

Thrombosis and Thrombocytopenia after ChAdOx1 nCoV-19 Vaccination

Nina H. Schultz, M.D., Ph.D., Ingvild H. Sørvoll, M.D., Annika E. Michelsen, Ph.D., Ludvig A. Munthe, M.D., Ph.D., Fridtjof Lund-Johansen, M.D., Ph.D., Maria T. Ahlen, Ph.D., Markus Wiedmann, M.D., Ph.D., Anne-Hege Aamodt, M.D., Ph.D., Thor H. Skattør, M.D., Geir E. Tjønnfjord, M.D., Ph.D., and Pål A. Holme, M.D., Ph.D.

March 2021 Two case series published in NEJM, N =16

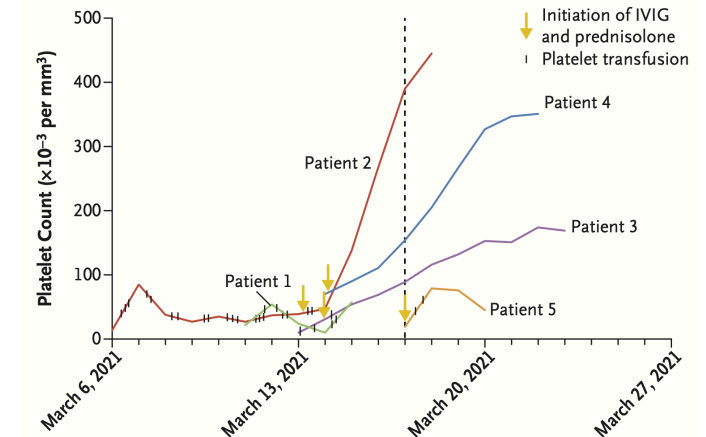
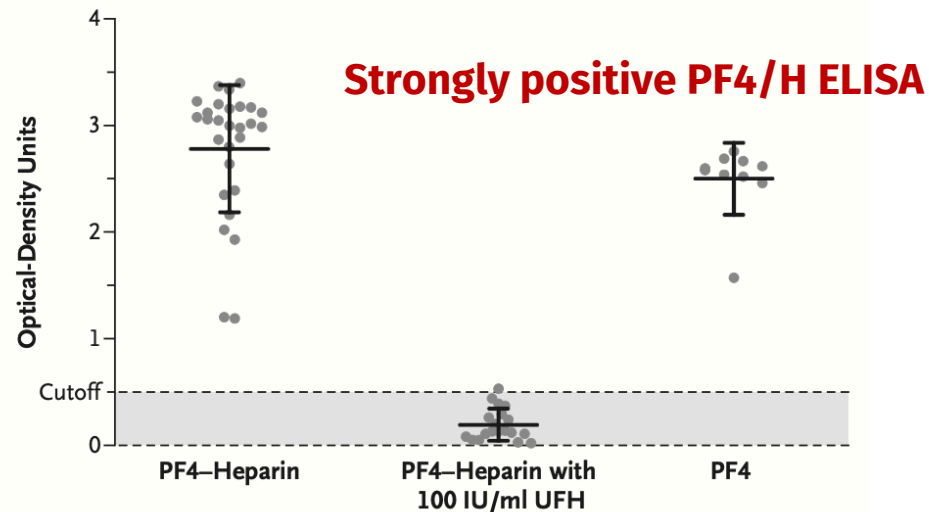
B Platelet-Activation Assays in 24 Patients with Clinical VITT



- Previously healthy, young individuals
- Severe thrombosis in unusual sites
- 7 -10 days after vaccination
- Thrombocytopenia and high D-dimer
- **High level of PF4/H antibodies**

- Addition of PF4 enhanced the detectability in platelet activation tests
- Inhibition of platelet activation with heparin

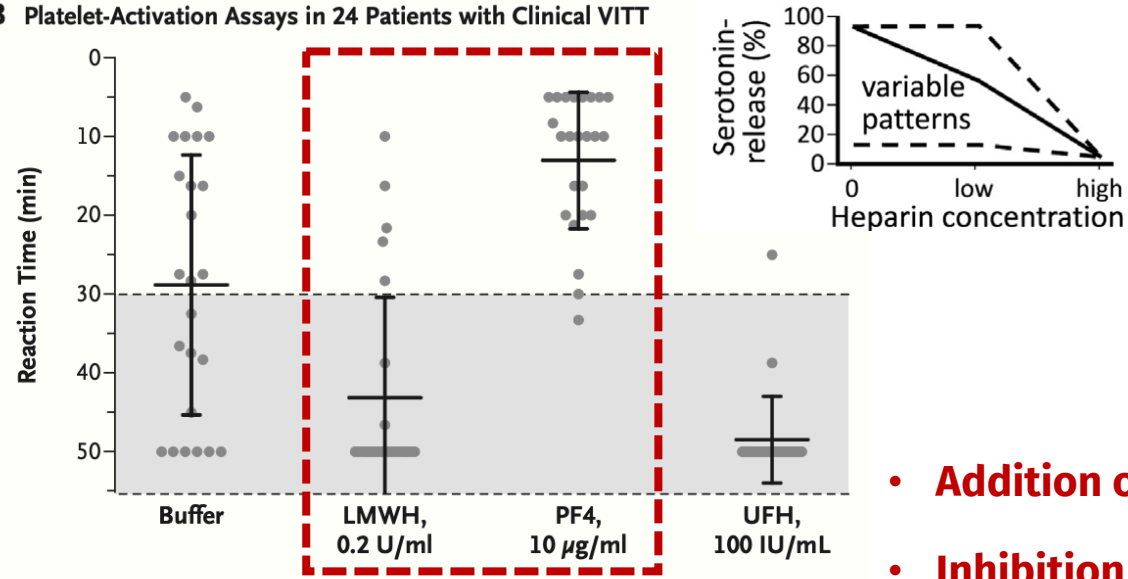
C ELISA Results for Combined Serum Samples from 28 Patients with VITT



- Platelet counts increased with IVIg

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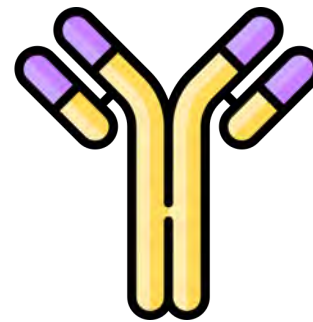
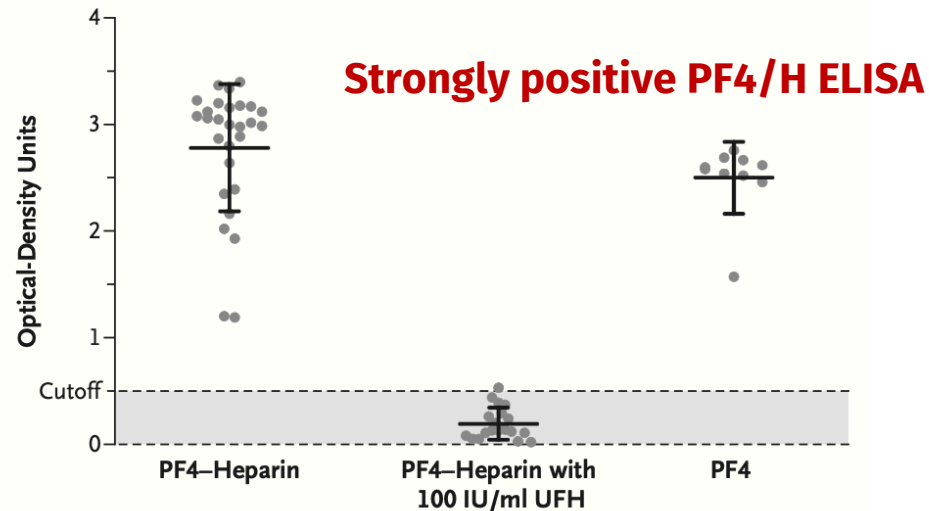
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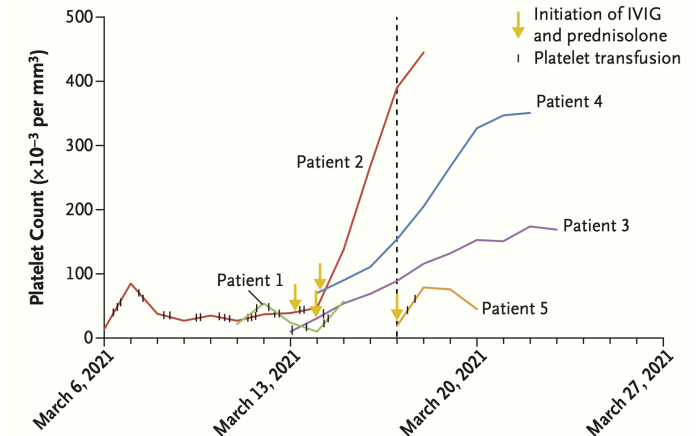
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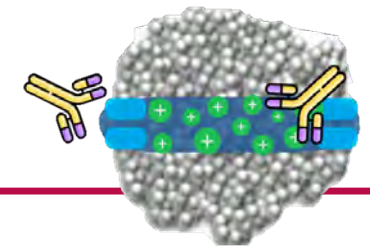


VITT antibodies
Heparin independent



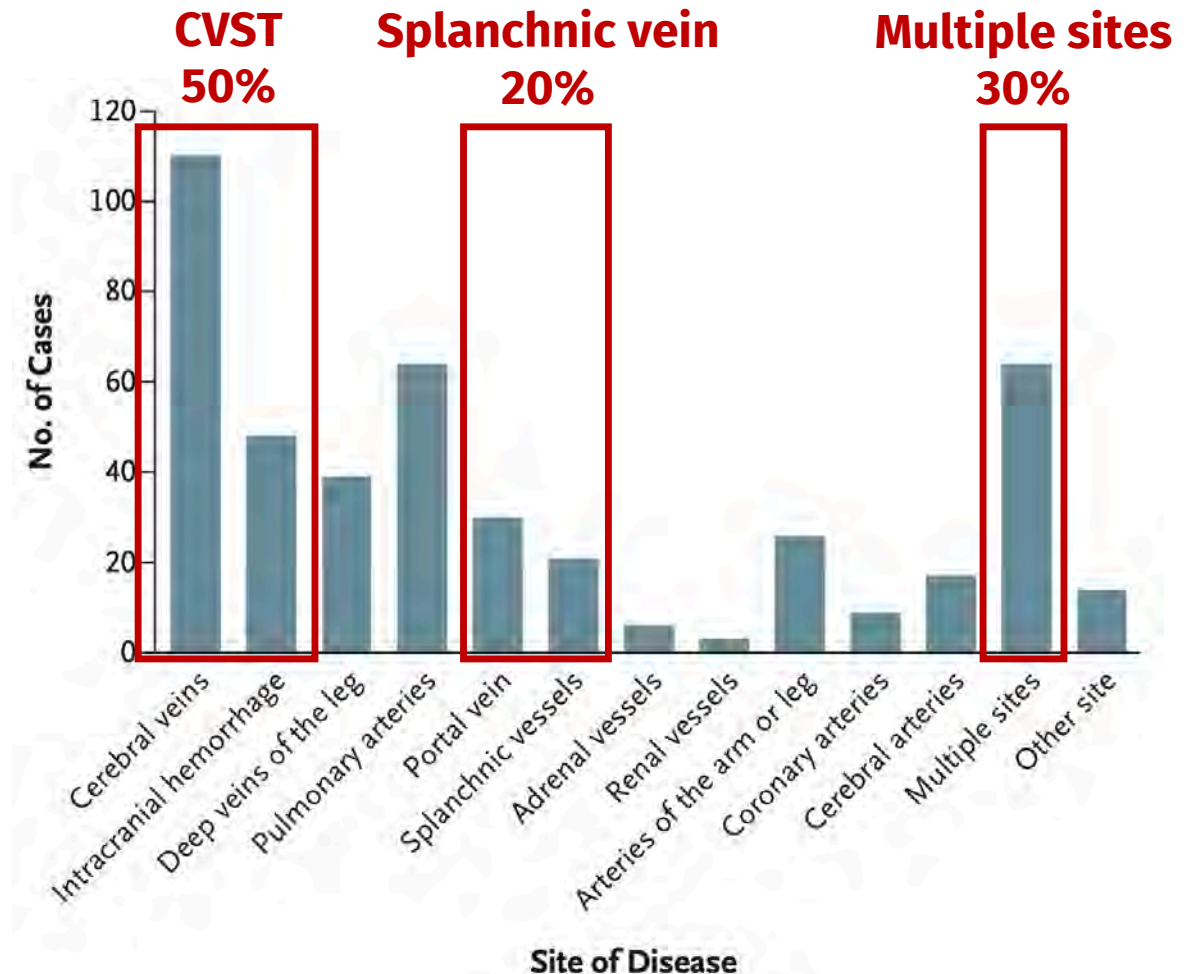
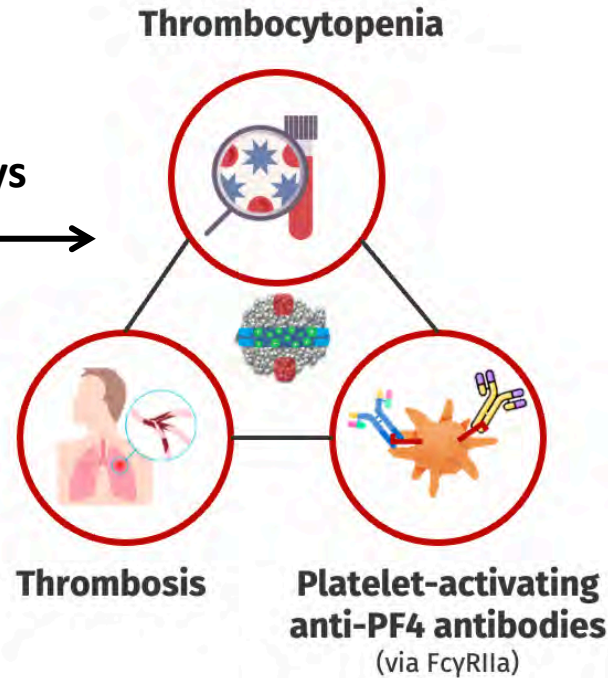
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Vaccine-induced Immune Thrombocytopenia with Thrombosis (VITT)

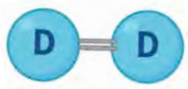


5 – 30 days

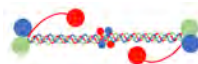
Trigger: Adenoviral vector vaccine
ChAdOx1 nCoV-19 (1: 25,000)
Ad26.COV2.S (1: 250,000)



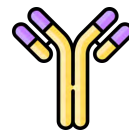
- Median platelet $47 \times 10^9/L$ (range 6 – 344)
- 1/3 had platelet $<30 \times 10^9/L$ at nadir



D-dimer
↑↑↑



Fibrinogen ↓



VITT antibodies
Heparin independent

Mortality rate 20 - 40%

The Spectrum of Anti-PF4 Disorders

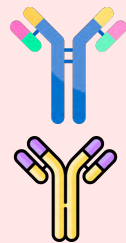
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1973

Classic HIT

- **Triggers:** heparin, LMWH, polyanions



2001

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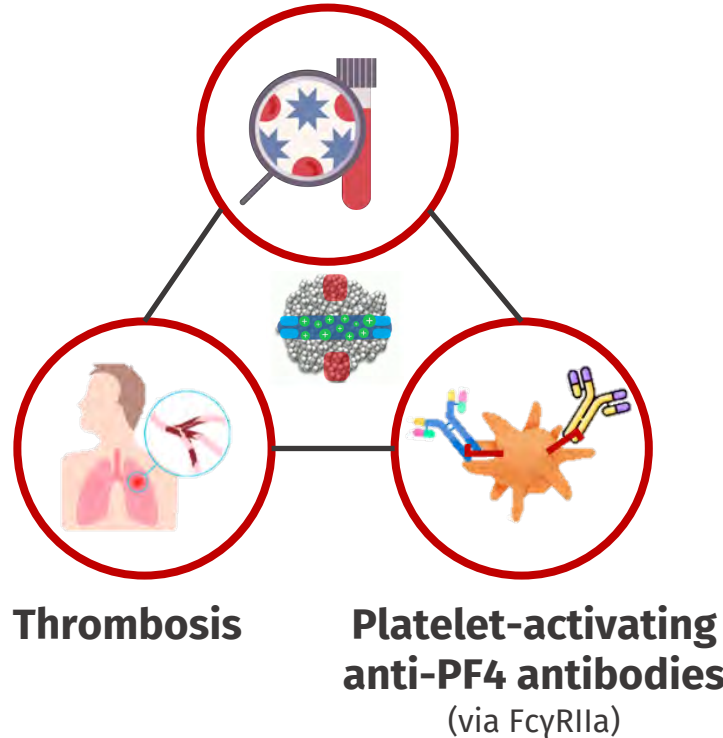
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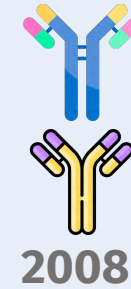
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Anti-PF4 Disorders

Thrombocytopenia



Non-heparin triggered



2008

Spontaneous HIT

- **Triggers:** total knee arthroplasty, infection



2021

VITT

- **Trigger:** adenovirus vector vaccines



2022

Spontaneous VITT

- **Triggers:** adenovirus infection, chronic autoimmune disorder, MGTS

December 2023



THROMBOSIS AND HEMOSTASIS

Anti-PF4 immunothrombosis without proximate heparin or adenovirus vector vaccine exposure

VITT-like disorder HITs the headlines

Marie Scully | University College London Hospitals Biomedical Research Centre and National Institute for Health Research

Antibodies to platelet factor 4 (PF4) are central to the pathology of heparin-induced thrombocytopenia (HIT) and vaccine-induced thrombocytopenia and thrombosis (VITT). However, in this issue of *Blood*, Schönborn and colleagues present a new scenario, VITT-like disorder.¹

Schonborn L, Esteban O, Wesche J, et al. *Blood*. 2023;142(26):2305-2314.

Anti-PF4 Immunothrombosis Without Proximate Heparin or Adenovirus Vector Vaccine Exposure: A Novel Clinical-Pathological Anti-PF4 Disorder

Context of research

Platelet-activating anti-PF4/heparin antibodies and anti-PF4 antibodies cause

- heparin-induced thrombocytopenia (HIT)
- vaccine-induced immune thrombocytopenia and thrombosis (VITT), respectively.

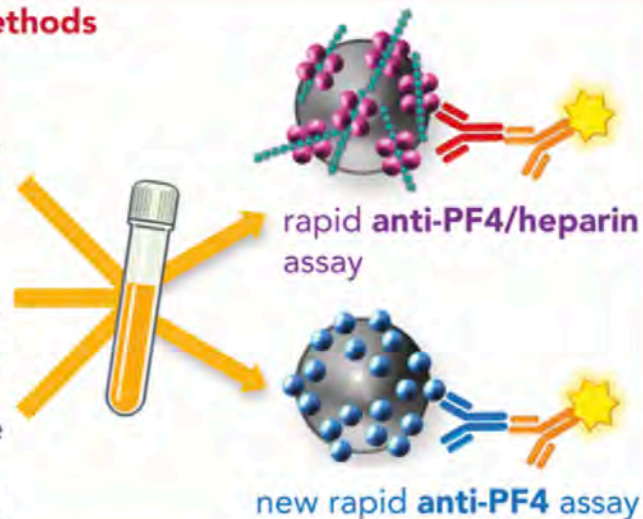
It is currently unclear whether other clinical-pathological anti-PF4 disorders exist.

Patients and Methods

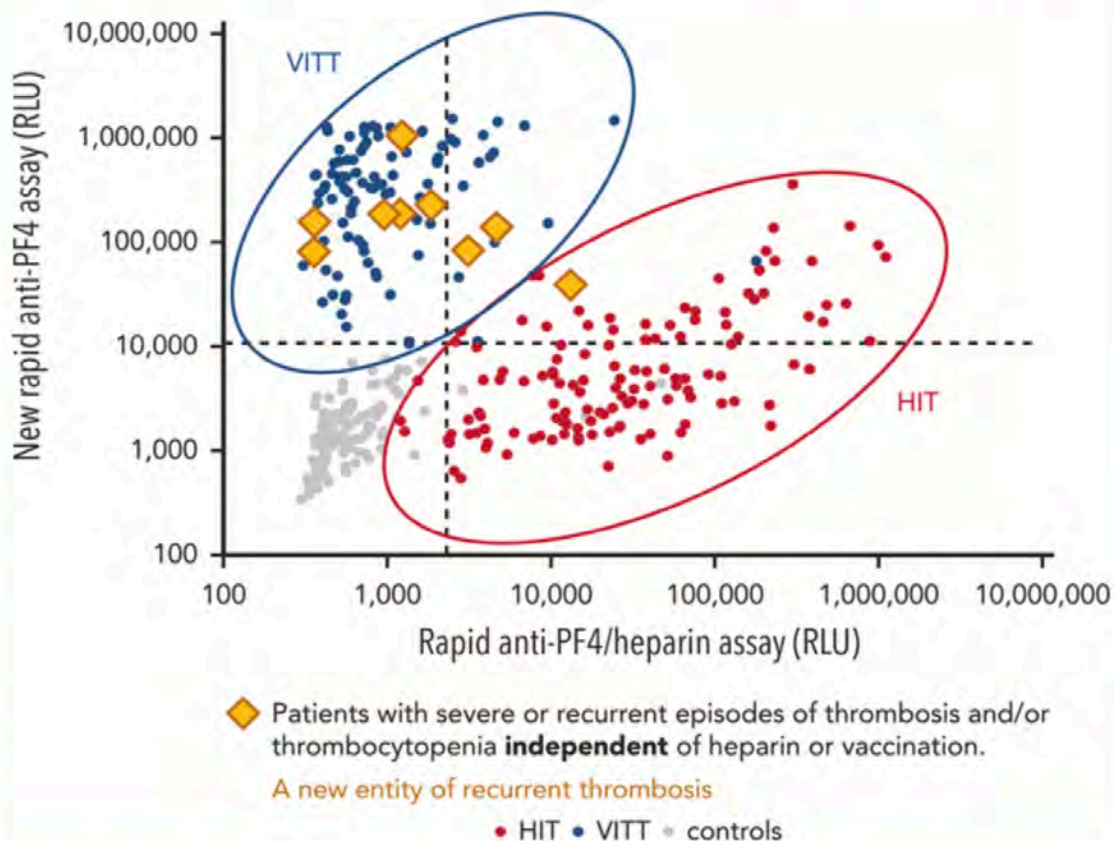
HIT
patients
N = 131

VITT
patients
N = 103

Negative
controls
N = 155



Findings of the combination of the rapid anti-PF4 and the anti-PF4/heparin assay



Conclusions: Highly prothrombotic, platelet-activating VITT-like anti-PF4 antibodies can occur without preceding heparin or Covid-19 vaccination, defining a novel anti-PF4 disorder.

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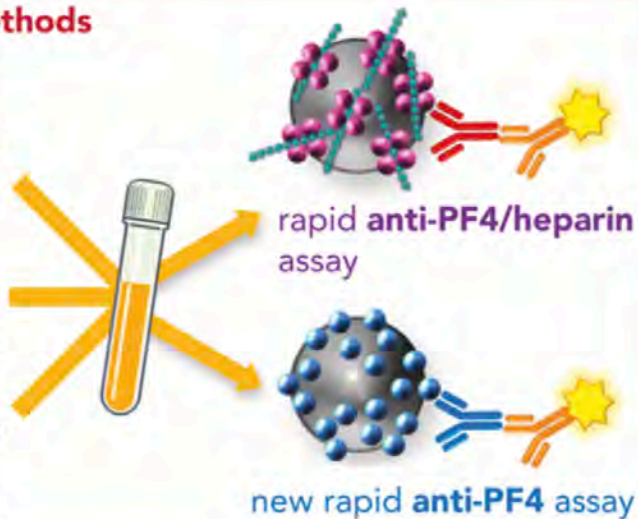
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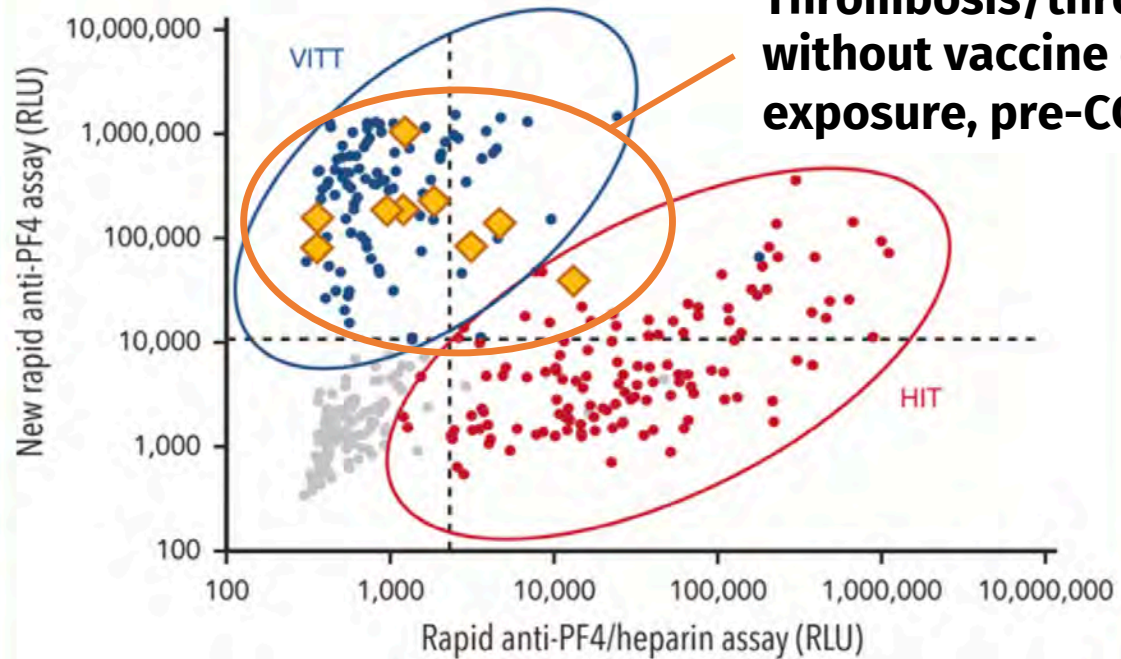
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♦ Patients with severe or recurrent episodes of thrombosis and/or thrombocytopenia **independent** of heparin or vaccination.

A new entity of recurrent thrombosis

• HIT • VITT • controls

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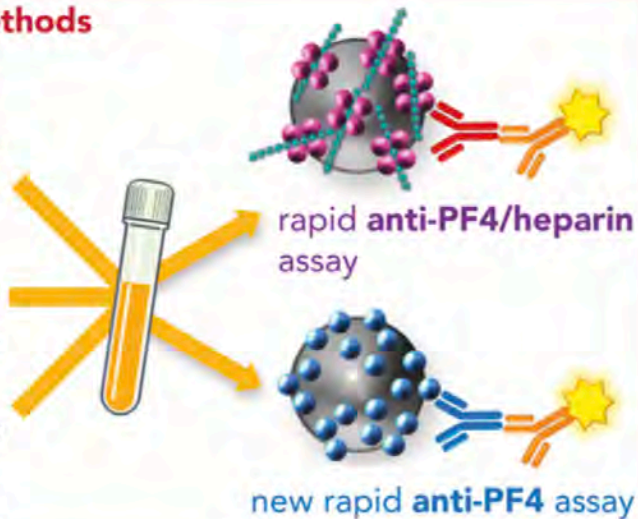
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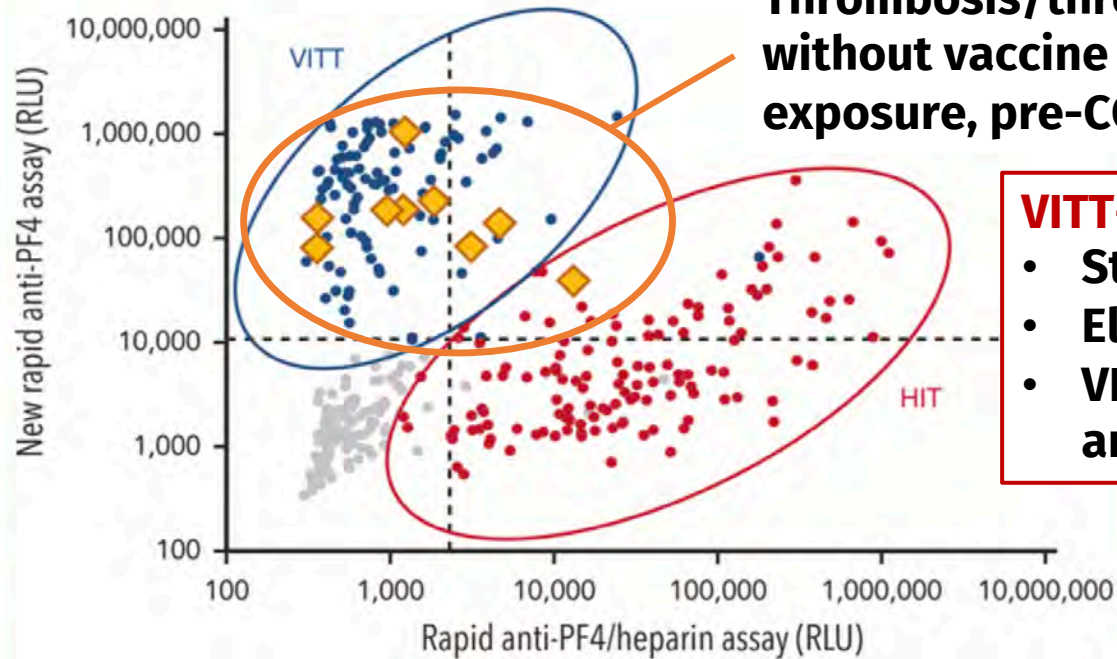
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Findings of the combination of the rapid anti-PF4 and the anti-PF4/heparin assay



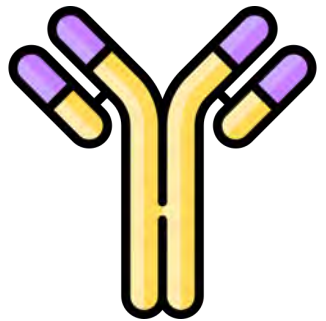
◆ Patients with severe or recurrent episodes of thrombosis and/or thrombocytopenia **independent** of heparin or vaccination.

A new entity of recurrent thrombosis

• HIT • VITT • controls

VITT-like disorders

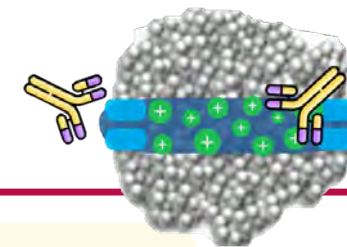
- Stroke/CVST
- Elevated D-dimer
- VITT-like antibodies



VITT antibodies
Heparin independent

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Spontaneous VITT syndromes



New clinic-pathological entity in 2023

VITT-like syndromes (acute & chronic)

VITT-like disorders

Anti-PF4 disorders, VITT-like

Reported triggers

- Infection
 - Adenovirus (~50%), CMV
- Vaccines: HPV
- Paraproteinemia (MGTS)
- Chronic autoimmune disease

No proximate heparin/vaccine exposures

D-dimer

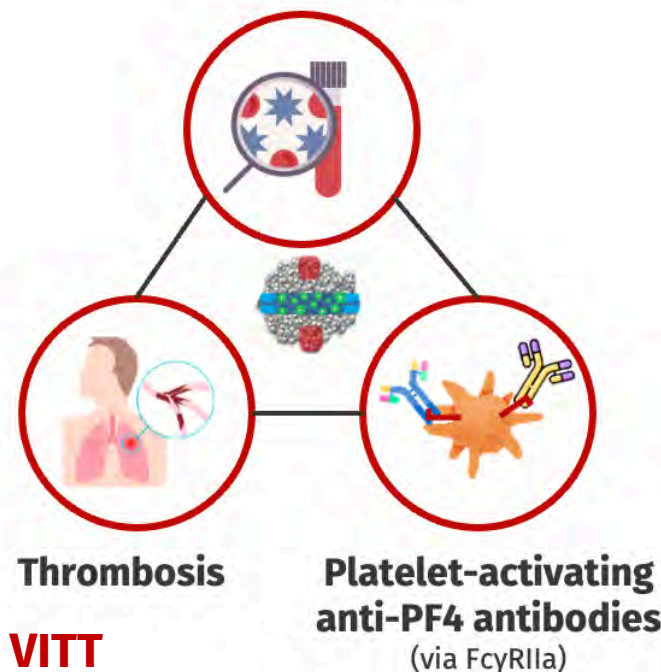
↑ ↑ ↑



Fibrinogen

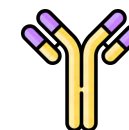
↓

Thrombocytopenia



Similar to VITT

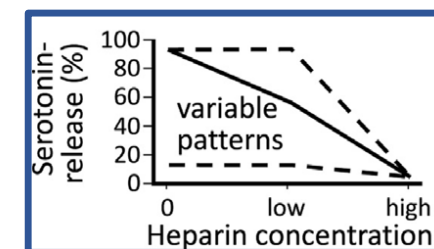
- Life-threatening
- Unusual site (CVST, SpVT)
- Multiple sites
- DIC



VITT antibodies

Heparin independent

- **ELISA Strongly +**
- **HIPA -/+**
- **PIPA +**



Adenovirus-associated VITT-like Syndromes

CORRESPONDENCE



**NEJM, August 2023
(N=2)**

Adenovirus-Associated Thrombocytopenia, Thrombosis, and VITT-like Antibodies

CORRESPONDENCE VOL. 389 NO. 18 NOV 02, 2023

More on Adenovirus-Associated Thrombocytopenia, Thrombosis, and VITT-like Antibodies

**NEJM, November 2023
(N=1)**

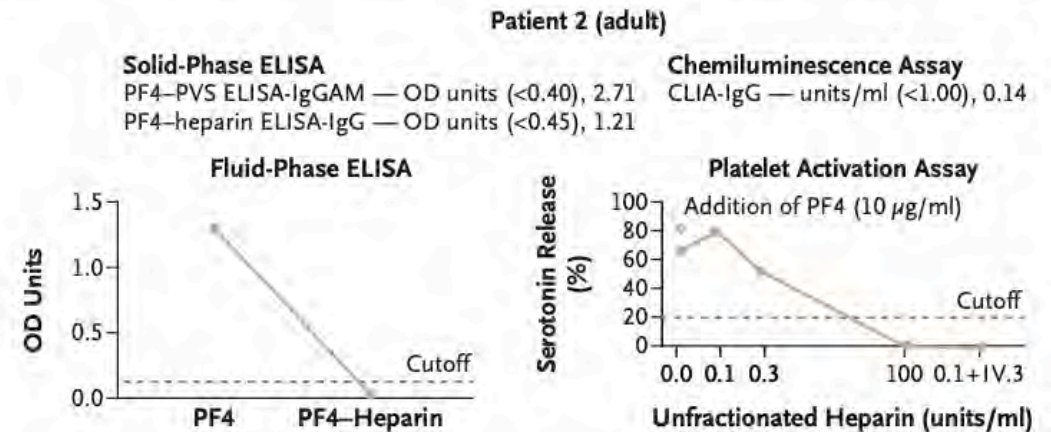
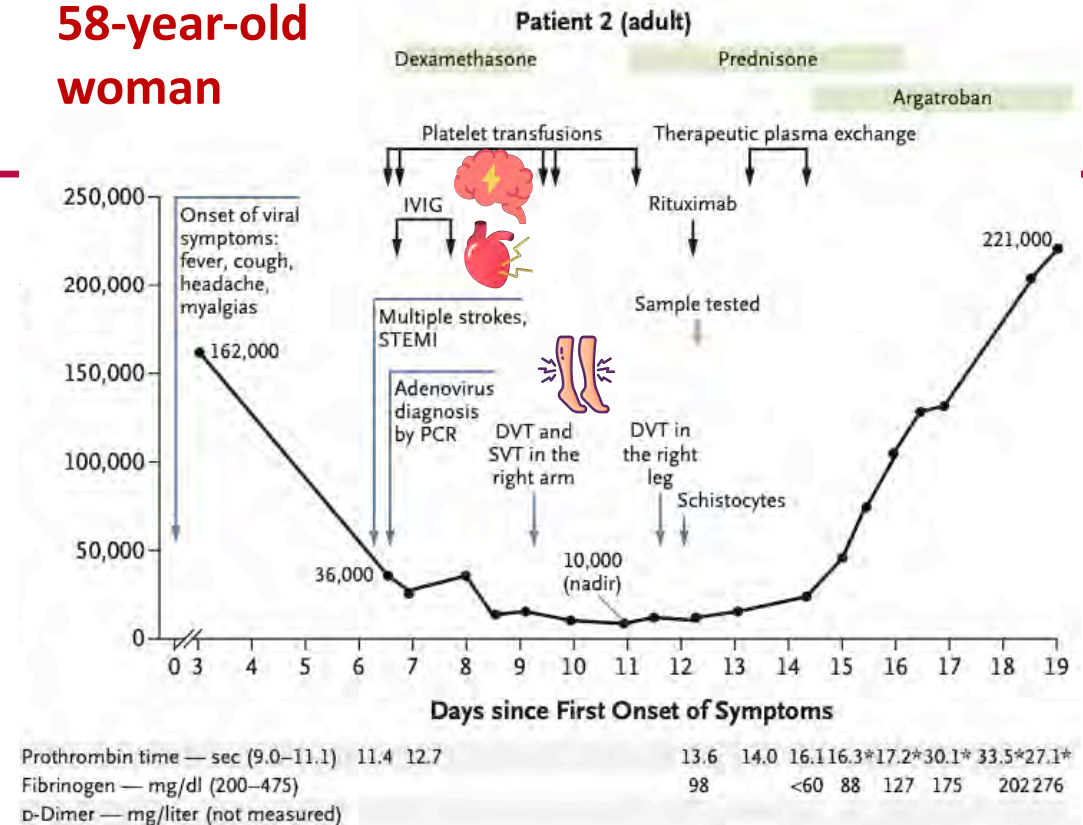
N Engl J Med 2023;389:1729-1731

CASE REPORT

Cerebral venous sinus thrombosis and thrombocytopenia due to heparin-independent anti-PF4 antibodies after adenovirus infection

**Haematologica, 2024
(N=1)**

58-year-old woman



Clusters of post-adenoviral infection VITT-like syndromes in children

Received: 13 July 2024 | Revised: 29 September 2024 | Accepted: 1 October 2024

<https://doi.org/10.1016/j.rpth.2024.102589>

CASE REPORT

November 2024
(N=6)



A cluster of pediatric VITT-like cases with thrombosis and thrombocytopenia following respiratory infections-Case series

Dimitra Dimopoulou¹ | Lida Mentesidou² | Athina Dettoraki³ |
Christina Karastathi⁴ | Maria Berikopoulou¹ | Panagiota Katsouli⁵ |
Ioanna Anastasopoulou⁶ | Iason G. Stamatakis⁷ | Theodora Bachou² | Flora Tzifi⁴ |
Aikaterini Michalopoulou³ | Anna Messaritaki¹ | Vana Spoulou⁵ | Helen Pergantou³

Received: 28 October 2024 | Revised: 14 November 2024 | Accepted: 14 November 2024

<https://doi.org/10.1016/j.rpth.2024.102641>

COMMENTARY



Mini-clusters of postadenovirus VITT

Michele P. Lambert^{1,2} | Theodore E. Warkentin³

- Greece: 3 cases in 3 months (2023)
- USA: 3 cases in 7 months (2015 – 2016)
- Previously healthy children (age 3 – 8)
- Presentation: acute symptomatic CVST, multiple site DVT
- Thrombocytopenia, elevated D-dimer, low fibrinogen
- Recent respiratory infection (3/6 adenovirus)
- 4/6 heterozygous FVL/FII mutations
- Positive anti-PF4 antibodies (ELISA)
- Treatment included IVIg, fondaparinux, and enoxaparin

Chronic Prothrombotic Anti-PF-4 Disorders

9 Reported Cases

Platelet-activating anti-PF4 antibodies mimic VITT antibodies in an unvaccinated patient with monoclonal gammopathy

Monoclonal gammopathy of thrombotic/thrombocytopenic significance

Adam J. Kanack,^{1,*} Jordan K. Schaefer,^{2,*} Meera Sridharan,^{3,*} Noah P. Splinter,¹ Mindy C. Kohlhausen,¹ Bandana Singh,¹ Silvana B. De Lorenzo,¹ Emily E. Mauch,¹ Maen A. Hussein,⁴ Marwan Shaikh,⁵ Shaji Kumar,³ Renren Wen,⁶ Demin Wang,⁶ David Murray,¹ and Anand Padmanabhan¹

The NEW ENGLAND JOURNAL of MEDICINE

Myeloma Therapy for Monoclonal Gammopathy of Thrombotic Significance

The NEW ENGLAND JOURNAL of MEDICINE

BRIEF REPORT

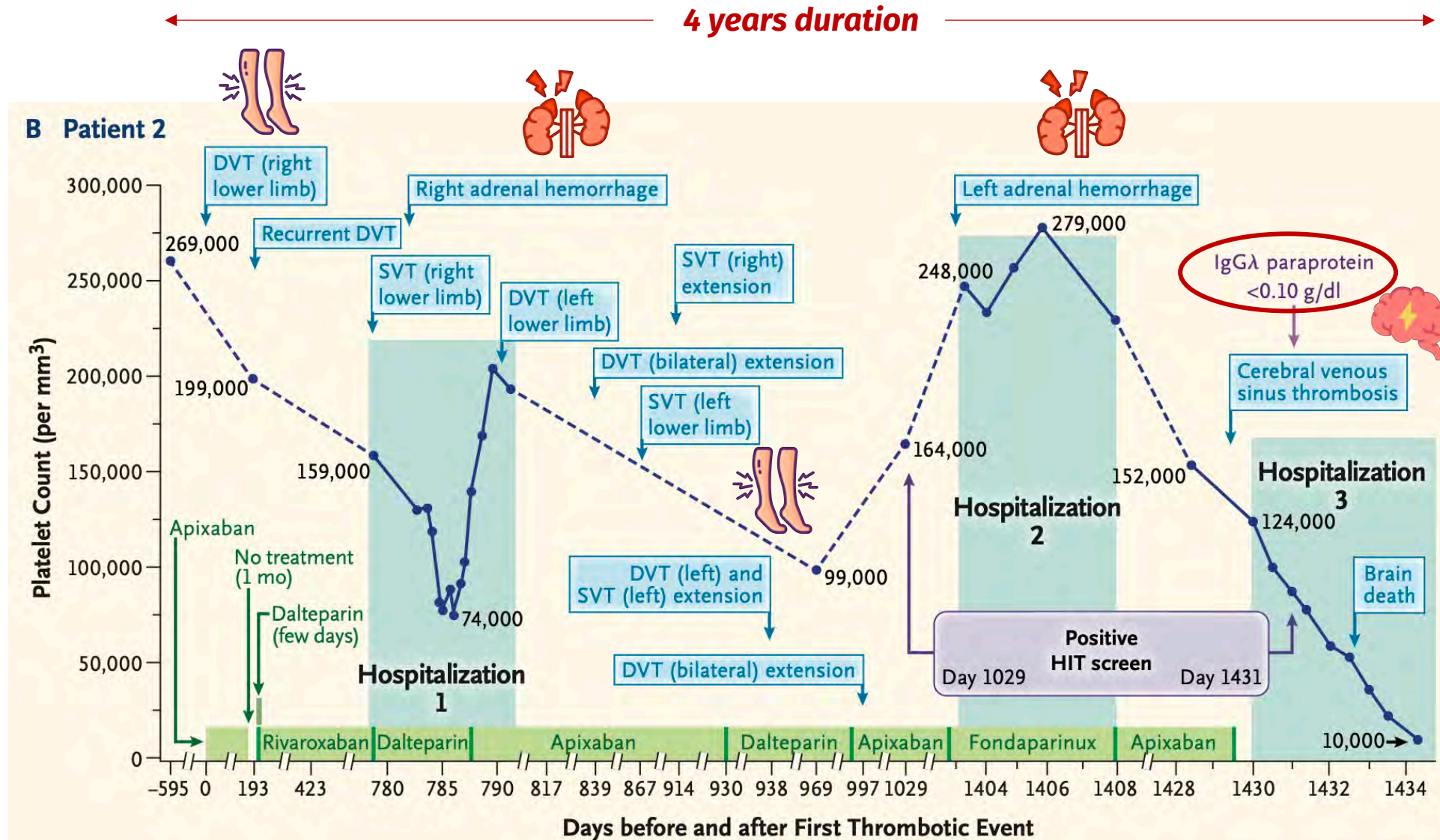
VITT-like Monoclonal Gammopathy of Thrombotic Significance

- **Chronic** prothrombotic disorders
 - Anticoagulant-refractory thromboses
 - Intermittent thrombocytopenia
- Persistent **VITT-like antibodies** (> 3 mo)
- 8/9 reported cases: M-protein detected
- **M-protein = VITT-like antibodies**
- Low levels of M-protein (<0.1 – 0.3 g/dL)
- Potential treatments
 - IVIg
 - Inhibition of FcγRIIa pathway: Ibrutinib
 - Plasma cell-directed therapy: Daratumumab, DVd

Anti-PF4 antibodies and testing for M proteins (SPEP/IF) may be indicated in unexplained chronic prothrombotic disorders

VITT-like Monoclonal Gammopathy with Thrombotic Significance (MGTS)

A 46-year-old man



Chronic Autoimmune Anti-PF4 Disorder

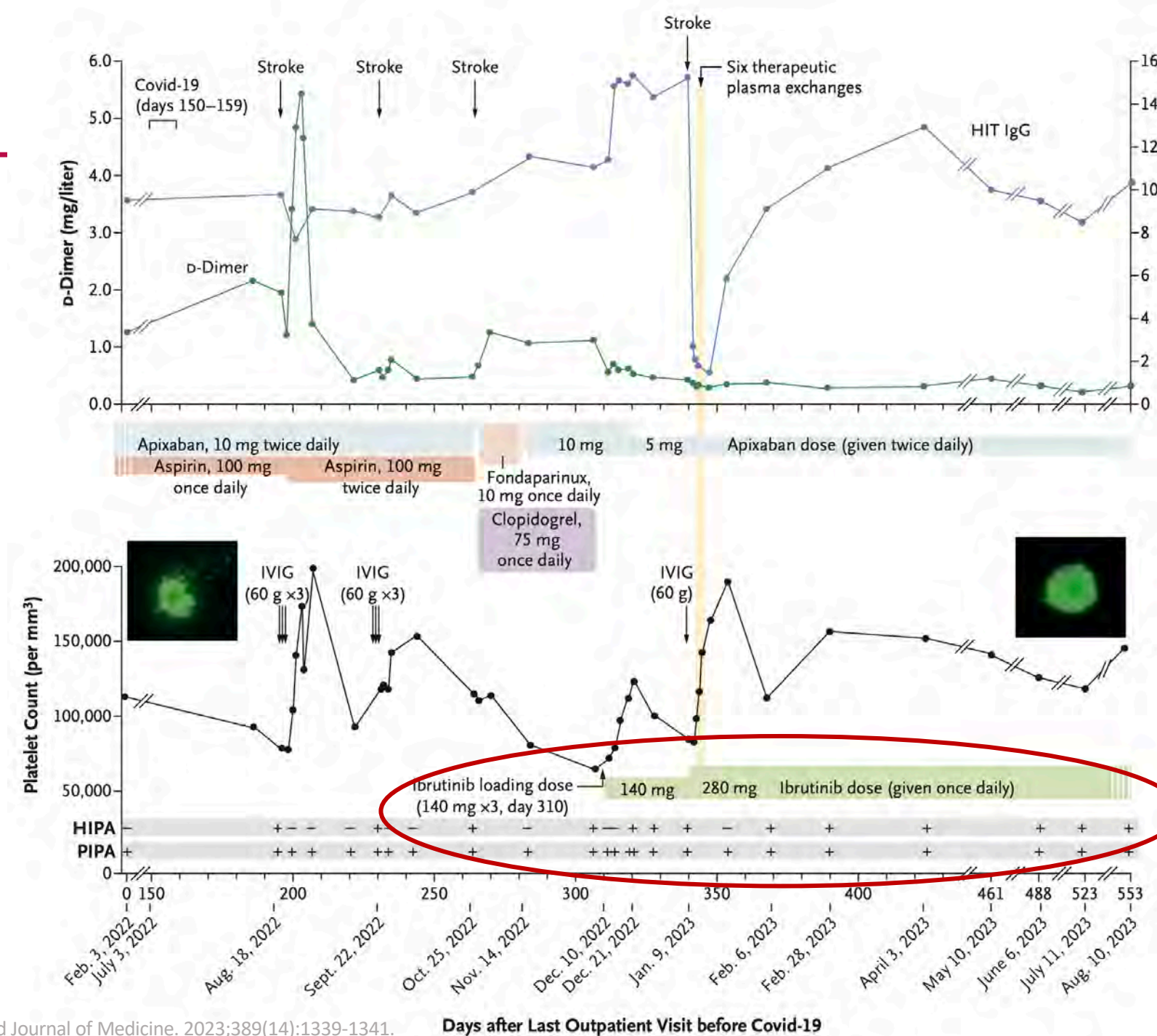
The NEW ENGLAND JOURNAL of MEDICINE

CORRESPONDENCE



Rescue Therapy in Chronic Prothrombotic Autoimmune Anti-PF4 Disorder

- A 37 year-old female
- Hx of recurrent DVTs & PE
- Multiple microembolic strokes
- Heterozygous FVL
- **No detectable M-protein or APS-Ab**
- **Persistent-platelet activating anti-PF4 antibodies**
- Rescue therapy with ibrutinib with clinical and laboratory responses, but still persistent anti-PF4 antibodies

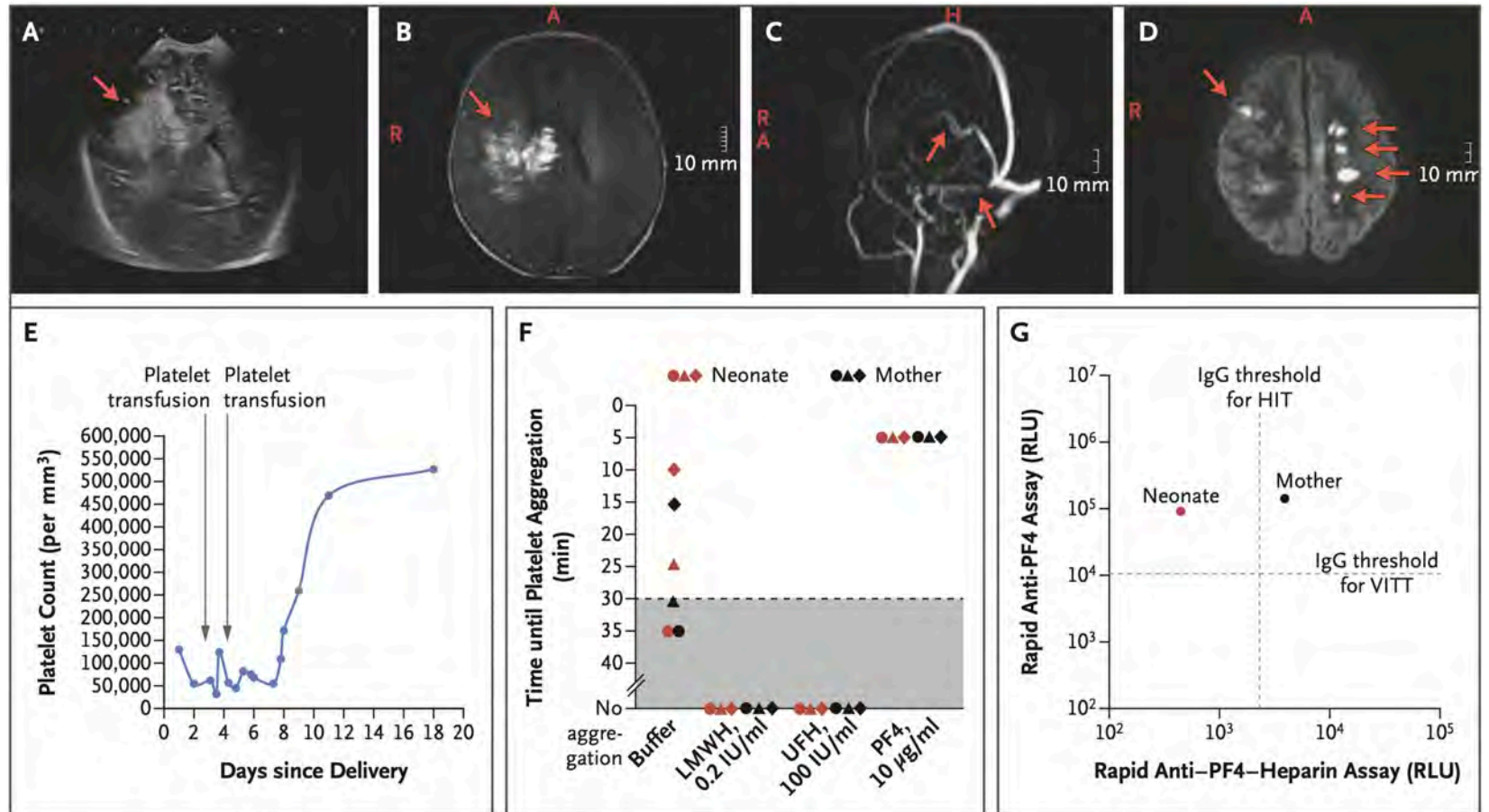


Maternal Anti-PF4 Antibodies as Cause of Neonatal Stroke

- A female neonate born to a 35-yo mother with history of low normal platelet (150k), history of PE receiving LMWH prophylaxis, and IgGk MGUS

- CVST with hemorrhagic infarction
- ↓ Plt 32,000 /mL
- ↓ Fibrinogen 1.08 mg/dL
- ↑↑ D-dimer 12900 ng/L

Transplacental transfer of anti-PF4 antibodies



Treatment of Anti-PF4 Disorders

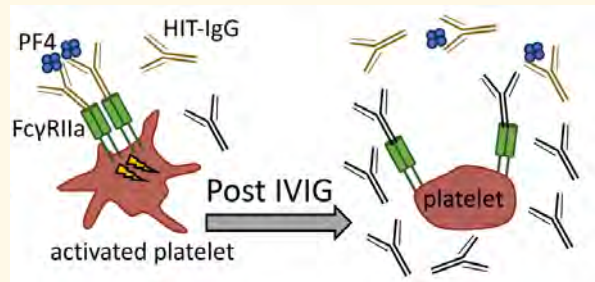
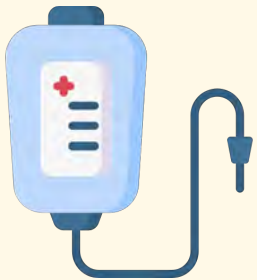
Management of Thrombosis

Anticoagulants: Non-heparin anticoagulants

- Fondaparinux, FXa inhibitors
- Heparin in VITT-like disorders?

- Organ-based management & consultation
- Cryoprecipitate if low fibrinogen
- Avoid platelet transfusion (unless critical bleeding)
- Vitamin K antagonists are contraindicated

Inhibition of anti-PF4-induced platelet activation via FcγRIIa



- **High-dose IVIg**
1 g/kg x 2 days, with the 3rd dose if suboptimal response
- **Plasma exchange** in IVIg-refractory
- Steroids (esp CVST), rituximab?
- Ibrutinib (as rescue therapy)
- Plasma cell-directed therapies in MGTS



Monitoring of response

- Platelet count
- D-dimer, fibrinogen
- EIA ODs
- Platelet activation (% at UFH 0 IU/mL)

	Classic HIT	Autoimmune HIT	Spontaneous HIT	VITT	Spontaneous VITT
Trigger	UFH, LMWH, polyanions	UFH, LMWH, polyanions, fondaparinux	Orthopedic surgery, infection <i>No proximate heparin/vaccine exposures</i>	Adenoviral vector vaccines	Adenovirus infection, Non-adenoviral vector vaccine, MGTS, chronic <i>No proximate heparin/vaccine exposures</i>
Anti-PF4 antibody	Type 2	Type 2 & 3	Type 2 & 3	Type 3	Type 3
Timing after exposure	5 – 10 days, still on heparin	Onset, persistence, or worsen at 5- 10 days after stopping heparin, heparin flush, or fondaparinux. Severe HIT with DIC	5 – 10 days	5 – 30 days	1 -4 days after infection
Thrombocytopenia	Mild to moderate, rapid recovery after stopping heparin	Variable, often severe (<20 x 10 ⁹) or persistent	Variable, often severe (<20 x 10 ⁹) or persistent	Variable, often severe (<20 x 10 ⁹) or persistent	Variable, often severe (<20 x 10 ⁹) or persistent

	Classic HIT	Autoimmune HIT	Spontaneous HIT	VITT	Spontaneous VITT
Thrombosis risk	30-50%	95%	95%	95%	95%
Thrombosis	Venous : arterial = 4:1 DVT, PE	DVT, PE Arterial Microvascular	DVT, PE Unusual sites Adrenal vein (Post-TKA) CVST/stroke (Med)	DVT, PE Arterial Unusual sites Multiple sites	DVT, PE Arterial Unusual sites Multiple sites
DIC	Rare	Often			
D-dimer	Increased	Markedly increased			
Rapid immunoassays (PF4/H)	Positive	Positive	Positive	Negative	Negative
Rapid immunoassays (PF4)	Negative (mostly)	Positive	Positive	Positive	Positive
EIA	Positive, moderate to strong	Strongly positive			
Platelet-activation test	Positive	Positive	Positive	Positive Enhanced with PF4	Positive Enhanced with PF4
		Positive in the absence of heparin			
Cessation of heparin		IVIg, plasma exchange, immunomodulation			

The Spectrum of Anti-PF4 Disorders: The Future



1973

Heparin triggered

Classic HIT

- **Triggers:** heparin, LMWH, polyanions

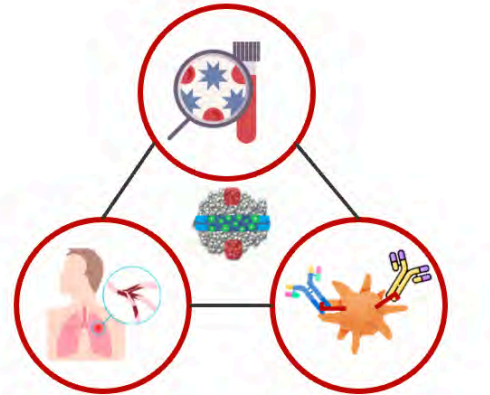


2001

Autoimmune HIT

- **Triggers:** heparin, LMWH, fondaparinux

Thrombocytopenia

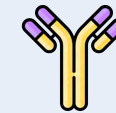


Thrombosis

Platelet-activating
anti-PF4 antibodies
(via FcγRIIa)



Adenoviral vaccine triggered



2021

VITT

- **Trigger:** adenovirus vector vaccines

Anti-PF4 Disorders

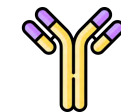
With no proximate heparin or vaccine exposure

Acute

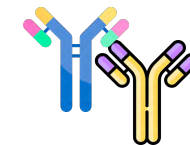
- **HIT-like:** post-TKA
- **VITT-like:** adenovirus infection

Chronic

- **MGTS** (HIT-like & VITT-like)
- **Chronic autoimmune** (HIT-like & VITT-like)



VITT-like Ab



HIT-like Ab



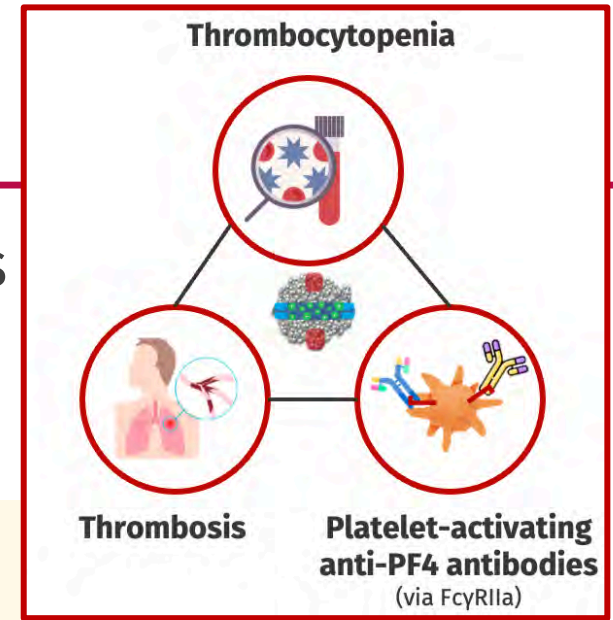
= Heparin dependent



= Heparin independent

Summary

- Anti-PF4 disorders = highly prothrombotic and fatal disorders
 - **Can occur in the absence of heparin/vaccine exposure**
- Awareness and early detection is key
- **Clinical clues:**
 - **Thrombosis + thrombocytopenia** (with no alternative explanation)
 - Especially severe, unusual sites, multiple sites, arterial + venous, microvascular thrombosis & DIC (but not necessarily)
 - Severe and persistent thrombocytopenia (but not necessarily)
 - Thrombotic storm (DDx with TTP, CAPS, cancer-associated thrombosis)
 - Markedly elevated **D-dimer**, decreased fibrinogen
- Laboratory testing have variable sensitivity for detecting different anti-PF4 antibodies (**rapid HIT screening tests may be negative!**)
- **High-dose IVIg is an important life-saving treatment**



INTERNATIONAL SCIENTIFIC SYMPOSIUM

“Scientific Forum on Myeloid and Lymphoid Malignancies :
Advancing Knowledge and Innovation ”

Registration: 8.00-8.20 | Opening Remark: 8.20-8.30

•••• Session I BONE MARROW FAILURE SYNDROMES

8.30 - 8.55	Deciphering the Genetic Landscape of Myelodysplastic Syndromes in the Modern Era	Prof. Seishi Ogawa (Japan)	
8.55 - 9.20	Unraveling the Impact of <i>DDX41</i> Mutations in Myeloid Malignancies: Genetic Insights and Clinical Relevance	Prof. Hideki Makishima (Japan)	
9.20 - 9.45	The Role of HLA in Aplastic Anemia: Uncovering Pathogenic Mechanisms	Assist. Prof. Sunisa Kongkiatkamon (Thailand)	

•••• Session II MYELOPROLIFERATIVE DISORDERS

Moderator	Asso. Prof. Noppacharn Uaprasert		
9.45 - 10.10	Investigating MPN in the iPSC model: Insights into the pathogenesis and screening for novel therapy	Prof. Ponlapat Rojnuckarin (Thailand)	
10.10 - 10.35	Chronic Myelomonocytic Leukemia (CMML): Bridging the Gap from Bench to Bedside	Prof. Mrinal S. Patnaik (USA)	

Break 10.35 - 10.50

•••• Session III ACUTE MYELOID LEUKEMIA

10.50 - 11.15	<i>FLT3</i> Inhibition in AML: Targeting Pathways for Therapeutic Advancement	Assist. Prof. Naoko Hosono (Japan)	
11.15 - 11.40	Prognostic significance and therapeutic implication of TIM-3 in AML	Assist. Prof. Chantiya Chanswangphuwana (Thailand)	

LUNCH | Symposium I 11.40 - 12.20 | Break 12.20 - 13.10 | Symposium II 13.10 - 13.50



Professor
Seishi
Ogawa

Professor
Mrinal
S. Patnaik



Professor
Ponlapat
Rojnuckarin

Asso. Prof.
Chantana
Polprasert

Asso. Prof.
Kitsada
Wudhikarn

•••• Session IV LYMPHOID MALIGNANCIES

Moderator	Prof. Tanin Intragumtornchai		
13.50 - 14.15	The Genomic Landscape of CLL: Unveiling Clinical Implications in Asian Population	Asso. Prof. Ja min Byun (South Korea)	
14.15 - 14.50	Personalized Medicine in DLBCL: Advancing from Traditional to Innovative Diagnostic Strategies	Dr. Narittee Suksawai (Thailand)	
14.50 - 15.15	Germline <i>HAVCR2</i> Mutation in SPTCL and HLH: Implications for Pathogenesis and Clinical Outcomes	Asso. Prof. Chantana Polprasert (Thailand)	

Break 15.15 - 15.30

•••• Session V CELLULAR THERAPY

Moderator	Prof. Nattiya Hirankarn		
15.30 - 15.55	Integrating CAR T-Cell Therapy into the Evolving Landscape of R/R DLBCL Treatment	Asso. Prof. Kitsada Wudhikarn (Thailand)	
15.55 - 16.20	Harnessing CAR T-Cell Therapy in AML: A New Era in Therapeutic Innovation	Assist. Prof. Shoji Saito (Japan)	
16.20 - 16.45	Navigating the Management of Complications in CAR T-Cell Therapy	Asso. Prof. Jakrawadee Julamanee (Thailand)	

SATURDAY **20.09.25**

Venue : Bhumisirimangalanusorn Building, Room 1210,
King Chulalongkorn Memorial Hospital (370 seats)

Main Sponsor :



Special Thank |