Mechanisms of Thrombosis in Heparin-Induced Thrombocytopenia and Vaccine-Induced Immune Thrombotic Thrombocytopenia

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Abstract

Heparin-induced thrombocytopenia (HIT) and vaccine-induced immune thrombotic thrombocytopenia (VITT) are rare, iatrogenic immune-mediated conditions with high rates of thrombosis-related morbidity and mortality. HIT is a long-recognized reaction to the administration of the common parenterally administered anticoagulant heparin (or its derivatives), while VITT is a new, distinct syndrome occurring in response to adenovirus-based vaccines against coronavirus disease 2019 and potentially other types of vaccines. A feature of both HIT and VITT is paradoxical thrombosis despite a characteristic low platelet count, mediated by the presence of platelet-activating antibodies to platelet factor 4. Several additional factors have also been suggested to contribute to clot formation in HIT and/or VITT, including monocytes, tissue factor, microparticles, endothelium, the formation of neutrophil extracellular traps, complement, procoagulant platelets, and vaccine components. In this review, we discuss the literature to date regarding mechanisms contributing to thrombosis in both HIT and VITT and explore the pathophysiological similarities and differences between the two conditions.

Keywords
- heparin-induced thrombocytopenia
- vaccine-induced thrombotic thrombocytopenia
- thrombosis
- platelet factor 4

Both heparin-induced thrombocytopenia (HIT) and vaccine-induced immune thrombotic thrombocytopenia (VITT) are prothrombotic, immune-mediated conditions associated with high rates of morbidity and mortality secondary to thrombotic complications. The anticoagulant heparin was first used in humans in 1937 and was in widespread use by the late 1940s.[1] Thrombosis occurring as a result of heparin therapy was first highlighted in 1958 by Weissman and Tobin, who described a series of 10 patients with peripheral arterial embolism following therapeutic heparin

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administration.[2] While thrombocytopenia associated with heparin was also described in the literature in the 1960s,[3,4] it was not until 1973 that HIT as we know it today—immune-mediated thrombocytopenia with thrombosis—was defined.[5]

In contrast, the first reports of VITT were published in early 2021, following the rapid development and widespread introduction of vaccines against coronavirus disease 2019 (COVID-19), a life-threatening infectious disease caused by SARS CoV-2 (severe acute respiratory syndrome coronavirus 2). The variably approved formulations of these vaccines now include the adenovirus-based vaccines ChAdOx1 nCoV-19 produced by Oxford-AstraZeneca,[6–8] Ad26.COV2.S produced by Johnson & Johnson,[9,10] and Sputnik-V (Gam-COVID-Vac) developed by the Gamaleya Research Institute of Epidemiology and Microbiology in Russia.[11]

Prevalence, Morbidity, and Mortality

HIT is a rare condition, with a prevalence of up to 3% in patients receiving unfractionated heparin[12,13] and 0.2% for those administered low molecular weight heparin (LMWH).[12] Comparatively, VITT is exceedingly uncommon, with published incidences ranging between 0.87 per million in central Europe to 8.6 per million in the United Kingdom.[14] The incidence of VITT appears to be 3 to 10 times higher following a dose of ChAdOx nCoV-19 compared with post-Ad26.COV2.S vaccination.[15,16] VITT generally occurs following the first, but not subsequent, dose of the vaccine,[17] with cases after the second dose being rare.[18]

HIT is associated with high rates of morbidity and mortality, with thrombotic complications occurring in over 50% of patients.[19] Rapid diagnosis and treatment, which involves cessation of heparin administration, and commencement of a therapeutic dose of an alternative anticoagulant,[20] can improve outcomes. However, these nonheparin anticoagulants are associated with an increased bleeding risk—the risk of major bleeding has been found to be 7% with argatroban[21] and 2.5% with danaparoid.[22] Furthermore, these drugs are more difficult to source and manage and do not prevent thrombosis in all cases—in one study, 14.6% of patients developed new thrombosis despite argatroban treatment, and the mortality rate remained significant at 18.1%, compared with 28.3% with no treatment.[23]

Documented rates of morbidity and mortality following a VITT diagnosis vary given the small studies to date and evolution of case definitions over time. In a composite series of around 100 early reported cases, the case fatality rate was 39.2%.[24] In a separate series of 220 patients with VITT, the overall mortality rate was 22%.[17] Within the Australian series, with a system for early recognition and early adoption of upfront immunomodulation intervention, the mortality rate was 5%.[25] Finally, a recent systematic review has reported mortality as 32%.[26] Current treatment recommendations for VITT include the commencement of a nonheparin anticoagulant as in HIT, as well as a course of intravenous immunoglobulin (IVIG).[27] Plasma exchange, high-dose corticosteroids, rituximab, and eculizumab are further treatment options in high-risk patients or those who do not respond to initial therapies.[28,29]

Clinical Presentation

The classical clinical presentation of HIT is moderate thrombocytopenia that occurs 5 to 10 days after the commencement of therapeutic heparin.[30] The fall in platelet count in HIT generally occurs over a period of 1 to 3 days, with a nadir of 50 to 80 $\times 10^9$/L, although sometimes lower.[31] The risk of thrombosis may be present even before this time—in the majority of patients, a thrombotic complication occurs either before or on the same day as a decline in platelet count is noted.[19] Furthermore, the risk of thrombosis persists after heparin cessation, and patients can present with delayed thrombosis up to weeks later.[32,33]

The time course from vaccine to presentation with thrombotic complications in VITT is reported as ranging from 3 to 48 days,[6,7,17,25] with a median of 14 days, with 75% of patients presenting by day 16 in the largest clinical case series.[17] The platelet nadir is variable, with a systematic review reporting the range as 5 to 127 $\times 10^9$/L,[34] although some patients may not present with thrombocytopenia.[25]

Sites of Thrombosis

The thrombotic complications of HIT can occur in both the arterial (limb artery occlusion, myocardial infarction, stroke) and venous (deep vein thrombosis [DVT], pulmonary embolism [PE]) systems, with the incidence of venous thrombosis predominating 4:1 over that of arterial thrombosis.[35,36]

Interestingly, unlike in HIT, the most common sites of thrombosis in VITT appear to be the normally atypical locations of the cerebral venous sinus and splanchnic veins.[6–8] Thrombosis in more typical locations such as lower limb DVT and PE also occurs[37] and may not have been well captured in early case series. Arterial thrombosis is less common, although it has been described.[7] Notably, a recent case series found that in 83% of patients with confirmed VITT who underwent whole-body imaging, additional sites of occult thrombosis (most commonly cerebral venous sinus thrombosis, PE, and portal vein thrombosis) were present that were not identified on initial focal imaging targeted to symptoms.[38]

Mechanisms of Thrombosis

Both HIT and VITT are platelet factor 4 (PF4)-mediated thrombotic conditions. The pathogenic agent in HIT is the presence of platelet-activating IgG antibodies against PF4-heparin complexes.[39,40] These IgG antibodies bind to PF4-heparin complexes to form immune complexes, which are able to bind and cross-link the FcγRIIa receptor on platelets, leading to platelet activation and aggregation[41] and the clinical manifestations of thrombocytopenia and thrombosis. Several other hematological cells, including neutrophils, monocytes, and endothelial cells, are also activated by HIT antibodies.

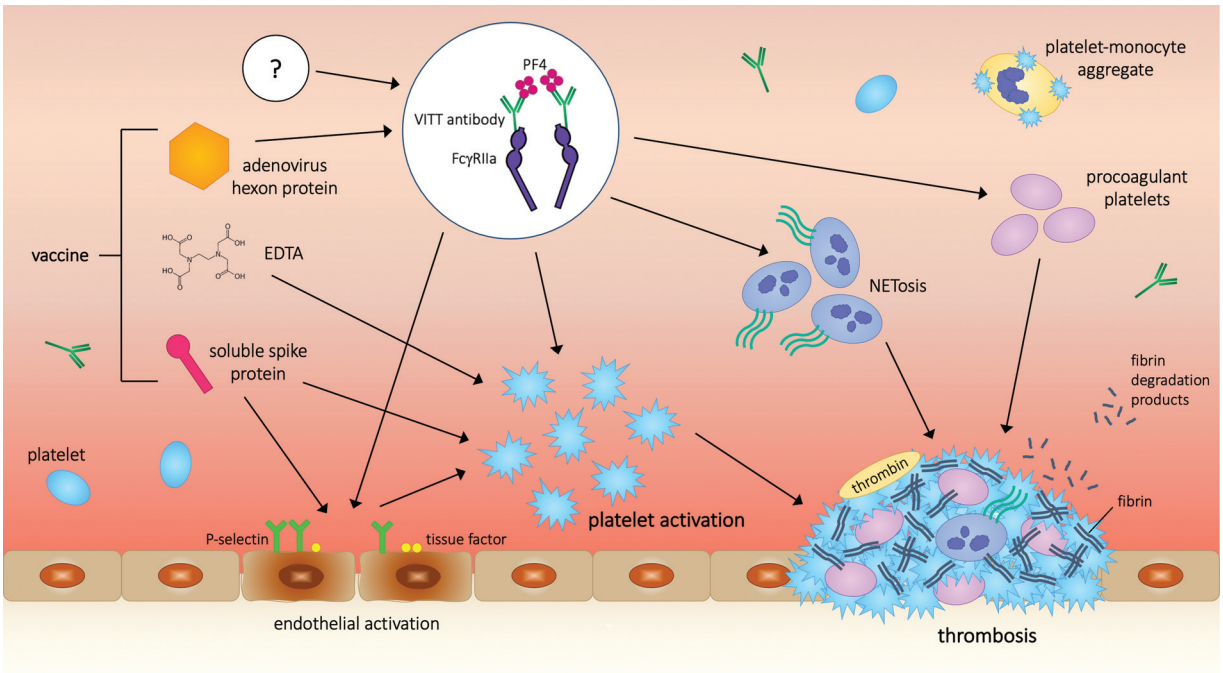

Fig. 1 Summary of potential mechanisms of thrombosis in VITT. Thrombosis in VITT occurs due to the formation of IgG antibodies to PF4, which activate platelets via FcγRIIa. These antibodies may bind to complexes of PF4 with vaccine components such as adenovirus hexon protein. Other factors that may contribute to thrombosis in VITT include the production of neutrophil extracellular traps (NETosis) by activated neutrophils, the generation of procoagulant platelets, endothelial activation with generation of activated P-selectin and tissue factor, the direct activating effect of vaccine components on platelets, and activated monocytes.

A particular feature of HIT is that PF4 and heparin must be present in the correct stoichiometric ratio to generate immune complexes—if either is in excess, complexes do not form, and thus immune complexes cannot activate FcγRIIa.[42] This concept is the basis of the gold standard diagnostic test for HIT, the serotonin release assay (SRA), a platelet functional test that measures serotonin release from donor platelets in the presence of patient serum or plasma at both therapeutic (0.1–0.3 U/mL) and high (100 U/mL) doses of heparin, with a positive test occurring if serotonin release occurs at therapeutic, but not high, heparin concentrations.[43] In HIT, donor platelet activation in the SRA classically does not occur in the absence of heparin. However, a variant of HIT exists where patients demonstrate clinical and laboratory features of HIT without heparin exposure, known as autoimmune or spontaneous HIT (aHIT). Antibodies from these patients are able to activate platelets even in the absence of heparin.[44]

Similarly to HIT, serum from patients with VITT contains platelet-activating IgG antibodies to PF4 and PF4-heparin,[6] or PF4-polyanion,[8] with platelet activation inhibited at high heparin concentrations.[6] However, in the SRA, plasma from patients with VITT do not show the heparin dependence classically seen in HIT. Rather, VITT plasma shows platelet activation in the absence of heparin, with variable inhibition of activation seen in the presence of therapeutic-dose heparin, and complete abrogation of activation with high-dose heparin.[24,45,46] It is for this reason that VITT is thought to most closely resemble the clinical entity of aHIT and may in fact be a clinical variant of this disease.[47] Indeed, treatment recommendations for VITT were based on those previously in place for the treatment of aHIT.[48]

Several recent studies have provided important insight into the mechanism of VITT (►**Fig. 1**). Huynh et al have shown that PF4 is required for platelet activation by VITT antibodies and that these antibodies bind to a site on PF4 that overlaps with the heparin-binding site, leading to FcγRIIa-dependent platelet activation.[46] Greinacher et al found that anti-PF4 antibodies from patients with VITT do not bind to the SARS-CoV-2 spike protein, suggesting that VITT is not caused by vaccine-induced antibodies cross-reacting with PF4, but rather represents a distinct process.[49] A very recent study of five patients with VITT has found that these anti-PF4 antibodies are highly stereotyped, with antibodies in all patients composed of a single IgG H-chain species, and a single lambda L-chain species encoded by the IGLV3–21*02 gene subfamily.[50] Interestingly, it appears that anti-PF4 antibodies may persist for several months postdiagnosis and treatment of VITT, and in most cases, this is not associated with any relapse of thrombosis.[51,52]

In addition to the PF4-dependent platelet activation described above, several contributory mechanisms for the thrombotic complications of HIT have been described, with thrombosis appearing multifactorial in origin. Several of these have also been suggested to contribute to thrombosis in VITT, although given VITT is a relatively recent entity, the literature remains in its early stages. These contributors to thrombosis will be discussed individually below.

Monocytes and Tissue Factor

Monocytes have been shown to play a role in HIT thrombosis. It has been shown that HIT immune complexes trigger monocyte expression of tissue factor,[53,54] a transmembrane

glycoprotein receptor and cofactor for factor (F) VII, and a major initiator of coagulation which in pathological settings can provoke both venous and arterial thrombosis.[55] HIT immune complexes also cause the release of tissue factor-positive microparticles by monocytes.[56] The pathway by which this occurs remains unclear—while one study has suggested that tissue factor expression occurs via the FcγRI receptor on monocytes, and not FcγRIIa,[56] a subsequent study reported that this process was mediated by FcγRIIa and not either of FcγRI or FcγRIII.[57]

In addition to tissue factor expression, monocytes also play other roles in the pathogenesis of HIT thrombosis. Rauova et al found that monocytes bind PF4 with greater affinity than platelets and form HIT-like complexes as a result. Furthermore, in a mouse model of HIT, monocyte depletion reduced thrombus formation following photochemical vascular injury and monocytes were found to be incorporated in developing arterial, although not venous, thrombi.[58] Tutwiler et al subsequently built on these studies to show that in a humanized microfluidic model of HIT thrombosis, monocytes are required for both platelet accretion and fibrin deposition, with monocyte depletion significantly decreasing, and repletion largely restoring, both outcomes.[57]

There have been very few studies examining the contribution of monocyte activity to COVID-19 vaccine-induced thrombosis. In a case study of a patient with VITT, McFadyen et al found an increased level of platelet-monocyte aggregates, likely reflecting monocyte, in addition to platelet, activation, and that this was returned to normal levels following administration of IVIG.[59] To date, there have not been any published studies examining the contribution of tissue factor dysregulation in the pathogenesis of VITT.

Microparticles

Microparticles are tiny vesicles that are derived from cell membranes and released in response to stimuli or cell death and which have procoagulant activity likely due to the presence of phosphatidylserine and tissue factor.[60,61] They too appear to play a role in HIT thrombosis. The role of microparticles in HIT was first identified in 1994 by Warkentin et al, who found that sera from patients with HIT are able to generate platelet-derived microparticles in a heparin-dependent manner and that these microparticles are found in plasma from patients with acute HIT at higher levels than in patients with non-HIT thrombocytopenia or thrombosis.[62] These microparticles were subsequently morphologically confirmed to be a distinct population of vesicles that likely originate from the pseudopodia of activated platelets.[63] More recently, Campello et al showed that sera from patients with HIT contain significantly higher levels of PF4-bearing microparticles compared with non-HIT patients. However, there was no statistically significant difference in PF4-bearing or overall microparticle levels in HIT-positive patients with thrombotic sequelae compared with HIT-positive patients with no thrombosis, suggesting that these may not in fact contribute to thrombotic outcomes.[64]

It has been postulated that tissue factor- and phosphatidylserine-positive procoagulant microparticles may contribute to the occurrence of thrombosis in VITT given the similarities in pathogenesis between HIT and VITT, and the high rates of atypical cerebral venous thrombosis in VITT,[65] although this has not yet been shown experimentally. Very recently, Petito et al have found increased platelet-derived microvesicles in patients following the administration of ChAdOx1, but not Ad26.COV2.S or the mRNA Pfizer BNT162b2 vaccines, although the clinical relevance of this is unclear given that none of the patients in the study developed thrombotic complications.[66]

Endothelial Cells

The endothelial surface itself also appears to contribute to thrombosis in HIT. A handful of studies have shown that sera from HIT patients deposit increased amounts of immunoglobulins on endothelial cells[67] and that HIT antibodies bind to endothelial cells in the presence of PF4.[68,69] Hayes et al showed that a contributor to clot enhancement in HIT was PF4 expression on endothelium surrounding the forming thrombus, allowing binding of HIT antibodies, formation of immune complexes, and subsequent thrombus propagation.[70] Furthermore, Davidson et al have more recently found that markers of endothelial cell injury are higher in HIT patients compared with both healthy controls and postcardiac surgery non-HIT patients, suggesting that endothelial dysfunction may play a role in HIT pathogenesis.[71] Recently, Johnston et al have found that PF4 binds to von Willebrand factor (VWF) strings released upon endothelial cell damage and that HIT antibodies bind to these PF4-VWF complexes, promoting thrombus propagation.[72]

While there have been no published studies to date directly linking endothelial cell activation or dysfunction to the pathogenesis of VITT, one study has found that the SARS-CoV-2 spike protein can cause vascular endothelial damage in an animal model.[73] Both adenovirus-based vaccines linked to VITT and mRNA-based COVID-19 vaccines which are not associated with VITT encode the spike protein. However, Kowarz et al have found that adenovirus-based, but not mRNA-based, COVID-19 vaccines can generate soluble spike protein splice variants and proposed that these secreted splice variants may bind to endothelial cells ultimately provoking localized inflammatory reactions and predisposing to thrombosis.[74] De Michele et al have shown in a small case series that soluble spike protein in VITT serum induces the activation of healthy donor platelets as measured by integrin $\alpha_{IIb}\beta_3$ activation, with serum-induced activation significantly reduced by the addition of an antibody against the S1 subunit of the spike protein.[75] Furthermore, published abstracts from two different groups implicate endothelial activation as a potential additional or even alternative driver to platelet activation for thrombus formation in VITT.[76,77] Interestingly, sera from patients that fit the clinical syndrome in which PF4 antibodies are not detectable appear to demonstrate endothelial-associated ex vivo thrombus formation.[77]

Neutrophils and NETosis

Neutrophils are the most abundant white blood cell and play an important role in immunity. Activated neutrophils release

neutrophil extracellular traps (NETs) in a process termed NETosis, which contributes to the host defense against infection. NETs are made up of DNA together with granule proteins such as elastase, myeloperoxidase, and histones.[78] In addition to their role in immunity, they have been identified as components of pathological arterial as well as venous thrombi.[79]

Recently, several studies have identified a role for activated neutrophils and the formation of NETs in the pathogenesis of HIT-associated thrombosis. Gollomp et al found that neutrophils adhere to venous endothelium in HIT. NETs produced by these activated cells bind PF4, which confers DNase resistance, and these PF4-NET complexes subsequently bind HIT antibodies.[80] Perdomo et al showed that HIT immune complexes can activate neutrophils enabling NETosis—both directly via neutrophil FcγRIIa, and indirectly via activated platelets—and that this NET generation is required for thrombus formation in ex vivo and in vivo models of HIT.[81] More recent work from Lelliott et al indicates that heparin can directly stimulate the formation of NETs, with unfractionated heparin inducing NET formation to a significantly greater extent than LMWH.[82] Finally, a study by Leung et al has found a role for reactive oxygen species (ROS) in NETosis and subsequent thrombosis in HIT, showing that HIT antibodies promote the generation of ROS by neutrophils and that this is required for the formation of NETs and thrombosis in HIT. In the KKO/PF4 mouse model of HIT, inhibition of ROS was able to significantly reduce the occurrence of pulmonary thromboembolism, although did not have an impact on thrombocytopenia.[83]

NETosis may also play a role in thrombus formation in VITT. Greinacher et al found that sera from patients with VITT were able to induce NET formation in donor neutrophils in a PF4-dependent manner.[84] Two independent studies have found that levels of markers of NETosis in plasma from patients with VITT are significantly increased compared with healthy controls and that extensive neutrophil activation and NET formation are present in thrombi from patients with VITT.[84,85] Using a murine model of VITT thrombosis, Leung et al provided a mechanistic link demonstrating inhibition of NETosis through pharmacological or genetic means, prevents VITT IgG-induced thrombosis. PAD4 is the enzyme responsible for the citrullination of histones and is requisite for NETosis. Interestingly, VITT IgG-induced NET deposition and in vivo thrombosis were dramatically reduced in the VITT mouse model deficient in PAD4 (FcγRIIa^{+}/hPF4^{+}/PAD4$^{-/-}$); however, there was no effect on the thrombocytopenia, suggesting that the thrombotic and thrombocytopenic processes may have overlapping but different drivers.[86]

Complement

The complement system, primarily a component of the innate immune system, comprises several plasma proteins and membrane receptors that contribute to host defenses. The system can be activated through one of three pathways, which all converge at a set of effector molecules that induce pathogen opsonization, inflammatory responses, and cell lysis.[87] Complement is linked to coagulation and has been implicated in the pathogenesis of the prothrombotic antiphospholipid syndrome[88] and may also play a role in the thrombotic complications of HIT and VITT.

The potential role of complement pathways in the pathogenesis of HIT was first highlighted by Cines et al, who found that platelets from patients with HIT had higher levels of IgG and C3 than controls and that plasma from HIT patients was able to induce complement-mediated platelet activation in healthy platelets in the presence of therapeutic heparin concentrations.[89] The same group subsequently built on these studies to show that serum from most patients with HIT was associated with complement activation and deposition of complement on endothelial cells.[67] However, it would be several decades before the literature on complement in HIT was further expanded. A more recent study has provided more detail on the role of complement in HIT thrombosis. Khandelwal et al have shown that HIT immune complexes activate complement via the classical pathway, resulting in the deposition of IgG and complement on multiple cell types including monocytes. This subsequently increases monocyte procoagulant function and platelet adhesion to endothelium.[90]

Complement has also been identified as a potential contributor to thrombosis in VITT. Mastellos et al proposed that complement pathway activation by anti-PF4 antibodies and immune complexes following adenovirus-based COVID vaccination, particularly C3 activation, promotes thrombogenic responses across a variety of cells—platelets, neutrophils, monocytes, and endothelial cells.[91] In a case study of a patient with VITT, Cugno et al identified several alterations to complement pathways, including the absence of classical and lectin pathway activity, consumption of C2, and increased levels of the terminal complex sC5b-9 on admission. These changes were normalized following IVIG therapy.[92]

Procoagulant Platelets

Procoagulant platelets are a subpopulation of platelets that promote coagulation by allowing the assembly and propagation of coagulation factors leading to thrombin generation.[93] This cell population has also recently been identified as a potential contributor to thrombotic outcomes in HIT. Tutwiler et al found that thrombin generation that occurs as a result of monocyte activation provides a second step in platelet activation, by enabling the formation of prothrombotic "coated" platelets that express P-selectin and bind annexin V, in response to KKO and PF4 stimulation.[57] Furthermore, our own studies have shown that the combination of HIT plasma and platelet agonists in the presence of therapeutic concentration of heparin promotes a marked increase in procoagulant platelets, which may contribute to thrombotic risk in HIT, with the level of procoagulant platelet induction correlating with the presence or absence of thrombosis.[94]

Procoagulant platelets may also play a role in VITT. Althaus et al found that sera from patients with VITT were able to induce the formation of P-selectin- and phosphatidylserine-positive procoagulant platelets in a PF4-dependent manner, which was inhibited in the presence of high-dose heparin or IV.3.[95] Singh et al confirmed these findings

Table 1 Comparison of clinical and pathophysiological features of HIT and VITT

	HIT	VITT
Incidence	2.6% (unfractionated heparin); 0.2% (LMWH)[12] Dependent on clinical situation[19]	0.000087–0.00086% adenoviral vaccine[14] Dependent on vaccine type[16] Regional variation[14]
Onset	5–10 d postcommencement of heparin[30,31]	5–42 d postvaccination[7,17,25]
Platelet nadir	>50% fall in platelet count $50–80 \times 10^9$/L Nadir $<30 \times 10^9$/L uncommon[19]	$2–144 \times 10^9$/L[25] Some patients do not present with thrombocytopenia[25]
Primary cause of mortality	Thrombosis	Thrombosis (primarily venous)
Most common locations of thrombosis	Deep vein thrombosis (DVT) Pulmonary embolism (PE)	Cerebral venous thrombosis DVT/PE Splanchnic vein thrombosis
Treatment	Cessation of heparin, nonheparin anticoagulant[20]	Intravenous immunoglobulin, nonheparin anticoagulant[27] Heparin possibly safe in selected individuals
Performance in serotonin release assay	Serotonin release at therapeutic-concentration heparin; inhibition with high-dose heparin[43]	Serotonin release in absence of heparin; variable inhibition at therapeutic-dose heparin; inhibited at high-dose heparin[24,45]
Key mechanism of thrombosis	Platelet-activating IgG antibodies against PF4-heparin bind FcγRIIa leading to platelet activation and aggregation[39–41]	Platelet-activating IgG antibodies against PF4/PF4 in complex bind FcγRIIa leading to platelet activation and aggregation[46] Non-PF4 antigens possible
Other contributors to thrombosis	NETosis[80–83] Endothelium[67–70,72] Procoagulant platelets[57,94] Monocytes[53,54,56–58] Microparticles[62,63] Complement[67,89,90]	NETosis[84–86] Endothelium[75–77] Procoagulant platelets[95–97] Monocytes[59] Vaccine components[84]

Abbreviations: HIT, heparin-induced thrombocytopenia; LMWH, low molecular weight heparin; NETosis, the formation of neutrophil extracellular traps; PF4, platelet factor 4; VITT, vaccine-induced immune thrombotic thrombocytopenia.

and showed that VITT sera-induced procoagulant platelet formation ex vivo is impacted differently by different anticoagulants—inhibited by heparin and danaparoid, but not by fondaparinux or argatroban.[96] Lee et al found that priming platelets with low-dose PAR1 agonist instead of addition of exogenous PF4 induced a FcγRIIa-dependent procoagulant platelet response to VITT plasma and utilized this in the establishment of a diagnostic platform.[97] In contrast to HIT, however, the induction of a procoagulant platelet response from VITT plasma is reduced rather than enhanced in the presence of heparin in the majority of, though not all, patients.[97] This may reflect Huynh et al.'s characterization of the restricted target of the VITT antibody, located within the heparin-binding site of PF4.[46] We speculate that this allows antibody to displace heparin from PF4, disrupting the PF4-polyanion complex. Thus, despite the similarities between HIT and VITT pathophysiology, it is possible that differences in antibody binding sites may result in differing therapeutic responses to anticoagulants from VITT to HIT,[96] though this has not been tested in a clinical setting.

Vaccine

Finally, there are several components of adenovirus-based vaccines that may trigger or contribute to VITT. Greinacher et al have found that in vitro ChAdOx1 nCoV-19 vaccine constituent particles, including adenovirus hexon protein, form complexes with PF4 which are recognized by anti-PF4 antibodies in plasma from patients with VITT, leading to both platelet activation and induction of an inflammatory response.[84] Furthermore, the vaccine component ethylenediaminetetraacetic acid (EDTA) induced vascular leakage and dissemination of vaccine particles in mice, which may contribute to the systemic inflammatory response.[84] Importantly, EDTA can be per se a trigger of platelet activation, as seen in samples with EDTA-dependent pseudothrombocytopenia.[98] In addition, Baker et al have shown that the ChAdOx1 viral vector itself can bind PF4 and that this complex can bind anti-PF4 antibodies in vitro.[99] Furthermore, Nicolai et al found that intravenous (IV) injection of ChAdOx1 nCov-19 vaccine in mice triggers the formation of platelet-adenovirus aggregates, which are subsequently taken up and processed by splenic macrophages. This is followed by a splenic B cell response leading to the formation of circulating antiplatelet antibodies. Thrombocytopenia was seen post-IV but not postintramuscular vaccination.[100] Although almost a third of VITT patients studied were found to have antiplatelet antibodies, it remains to be seen whether inadvertent IV administration of adenovirus-based vaccines contributes to VITT in humans.[100]

Conclusions

The pathogenesis of thrombosis in both HIT and VITT is multifactorial, and the complex interplay of the various components is not yet fully understood. Early studies have suggested that many of the same mechanisms that have been identified as contributing to thrombosis in HIT, also play a role in VITT; however, distinct differences have also emerged that may give additional insights into our understanding of both syndromes (►**Table 1**). Given VITT is a relatively new phenomenon with extremely low incidence, the literature on underlying mechanisms continues to evolve. Further research into the contributors to thrombotic outcomes in VITT, as well as continued research into HIT, would be useful in reducing the significant morbidity and mortality associated with these rare conditions, even if the administration of adenoviral COVID-19 vaccines has now ceased in several countries worldwide. Moreover, thrombosis with thrombocytopenia with clinical course and laboratory features consistent with VITT has also been described in a patient receiving the protein-based Gardasil 9 human papillomavirus vaccine,[101] suggesting that VITT may not in fact be a complication unique to COVID-19 or adenovirus-based vaccinations and that understanding of the underlying mechanisms may be more widely applicable than previously recognized.

Conflict of Interest
None declared.

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