Vaccine-induced immune thrombotic thrombocytopenia – overview

Immunologiczna małopłytkowość zakrzepowa indukowana szczepieniem – przegląd

Jakub Sleziak¹, Antoni Gawor¹, Krzysztof Gomułka²

¹Student Research Group of Adult Allergology, Wroclaw Medical University, Wroclaw, Poland
Head of the Group: Dr. Krzysztof Gomułka

²Department of Internal Medicine, Pneumology and Allergology, Wroclaw Medical University, Wroclaw, Poland
Head of the Department: Dr. Robert Pawłowicz

Key words: COVID-19, thrombosis, thrombocytopaenia, vaccines, platelet factor 4.

Abstract
In response to the ongoing COVID-19 pandemic, pharmaceutical companies have been able to rapidly develop and distribute effective vaccines. Despite some common, minor, local, and systemic adverse effects following vaccination, there is also, in rare cases, a potential for the development of anti-platelet factor 4 antibodies, resulting in platelet activation and aggregation leading to potentially life-threatening thrombosis. Vaccine-induced immune thrombocytopaenia (VITT) is a rare immune response following administration of adenoviral vector vaccines against severe acute respiratory coronavirus 2 (SARS-CoV-2) such as ChAdOx1 nCoV-19 AstraZeneca – Oxford and Ad26.COV2. S Johnson & Johnson. Since the discovery of the syndrome, the mortality rate has decreased by 90%. Therefore, in this paper, data including epidemiology and pathophysiology of the syndrome, followed by the diagnosis criteria and management options, were collected. The authors believe that spreading knowledge further among primary care physicians and other healthcare professionals will lead to better VITT treatment outcomes.

Streszczenie
W odpowiedzi na wciąż trwającą pandemię COVID-19 firmom farmaceutycznym udało się w krótkim czasie opracować i rozprowadzić skuteczne szczepionki. Chociaż po szczepieniu występują pewne powszechne, niewielkie miejscowe i ogólnoustrojowe działania niepożądane, w rzadkich przypadkach możliwe jest wytworzenie przeciwko czynnikiowi płynkowemu 4, co powoduje aktywację płytek krwi i ich agregację prowadzącą do potencjalnie zagrażającej życiu zakrzepicy. Opisana powyżej immunologiczna małopłytkowość zakrzepowa indukowana szczepionką (VITT) jest rzadko występującą reakcją immunologiczną po podaniu adenowirusowych szczepionek wektorowych przeciwko ciężkiemu ostromu koronawirusowi układu oddechowego 2 (SARS-CoV-2), takim jak ChAdOx1 nCoV-19 AstraZeneca – Oxford i Ad26.COV2. S Johnsona & Johnsona. Od czasu odkrycia zespołu, dzięki wzrostowi świadomości pracowników ochrony zdrowia, śmiertelność zmniejszyła się o 90%. W niniejszej pracy przedstawiono dane dotyczące epidemiologii i patofizjologii zespołu, a następnie kryteria rozpoznania i opcje postępowania. Autorzy uważają, że rozpowszechnianie wiedzy wśród lekarzy podstawowej opieki zdrowotnej i innych pracowników ochrony zdrowia prowadzi do lepszych wyników leczenia VITT.

Introduction
Vaccine-induced thrombotic thrombocytopenia (VITT) was described for the first time in Germany in March 2021 and referred to as vaccine-induced prothrombotic immune thrombocytopenia (VIPIT) [1]. Later, the name thrombosis with thrombocytopaenia syndrome (TTS) was given to this disease by the Brighton Collaboration and is still used by the American Society of Haematology [2]. TTS is a broad, general term comprising all cases of thrombosis and thrombocytopaenia following COVID-19 vaccination regardless of the pathomechanism [3]. Many clinicians use the term VITT, which refers to the occurrence of thrombosis and thrombocytopaenia caused by particular autoimmune mechanisms following vaccination against COVID-19 [2–4].

It is estimated that due to the rise in both public and clinical awareness of the syndrome, the mortality rate of VITT has been reduced by almost 90% [5]; therefore, it is vital to spread knowledge on this topic further.

The syndrome has been reported almost entirely after vaccinations with ChAdOx1 nCoV-19 AstraZen-
eadoviral vaccines [2, 6]. Although there have been certain reports of VITT cases after mRNA COVID-19 vaccination, those patients presented different demographic characteristics or different laboratory testing results, which suggest that these represent background cases or that the pathogenesis was different [4]. Moreover, before the COVID-19 pandemic, the incidence of thrombocytopenia with cerebral venous sinus thrombosis per million was 0.062. The incidence of TTS after mRNA COVID-19 vaccines reaches 0.00855 per million [4], which is far less and suggests that the TTS cases following mRNA COVID-19 vaccination should be considered irrelevant [3].

**Methodology**

Electronic database searches focused on original articles, case reports, reviews, and guidelines regarding adverse events following vaccinations with ChAdOx1 nCoV-19 AstraZeneca–Oxford and Ad26.COV2.S Johnson & Johnson vaccines. The search process was carried out in PubMed and Google Scholar and comprised the period from 1 June 2021 to 20 February 2022. The keywords included “VITT”; “VIPIT”; “Vaccine-Induced Immune Thrombotic Thrombocytopenia”; “thrombosis”; “COVID-19”; “SARS-CoV-2”; “Vaccine” and were used both in combinations and separately. Moreover, another additional search was performed with the keywords: “heparin-induced thrombocytopenia”; “HIT”; “platelet factor 4”; “PF4”; “Thrombotic thrombocytopenic purpura”; and “Hemolytic–uremic syndrome”.

**Epidemiology**

The first reports on VITT indicate that the majority (80%) of patients with VITT are females ages between 20 and 55 years [7]. However, the data that support this thesis come from observations from countries in which the AstraZeneca Oxford vaccine was initially used for vaccination of frontline healthcare workers who were mostly female. Other sources claim that the sex bias among VITT cases may be much less prominent [3, 6]. According to a series of 220 patients with definite or probable VITT, 55% of patients were female [8].

So far, the causal relationship between either the ChAdOx1 nCoV-19 or Ad26.COV2 vaccination and VITT has not been proven [7]; however, there is supporting evidence [4].

The reported incidence of VITT varies depending on the source of the data.

Rates of TTS following ChAdOx1 nCoV-19 vaccination range from 13 (UK) to 39 (Norway) cases per million doses of vaccine administered [4, 6, 9].

According to the World Health Organization (WHO), the syndrome's incidence among patients vaccinated with adenovirus vector-based vaccine ranges from 5 to 68 cases per million vaccinees [10].

In the United States during the period between 14 December 2020 and 31 August 2021 a total of 57 cases of VITT were reported. Fifty-four cases were associated with vaccination with Ad26.COV2.S, which leads to reporting rate of 3.83 per million. Three cases were connected with mRNA-based COVID-19 vaccines, which equals a reporting rate of 0.00855 per million vaccine doses. All of the patients who presented symptoms of VITT after being vaccinated with Ad26.COV2.S required hospitalization, 67% of them in an Intensive Care Unit (ICU) [4].

The highest reporting rates after receiving Ad26.COV2.S vaccine were among women aged 30 to 39 years (10.6 per million doses) and 40 to 49 years (9.02 per million doses) [4].

The median age also varies depending on the source, from 44.5 years [4] to 48 years [3, 6], and the age range is from 18 to 79 years.

In terms of different age groups, it is estimated that the risk of VITT for people aged 49 years and less is 1 : 50,000, and for people older than 50 years the risk is 1 : 100,000.

It is claimed that approximately 15% of the VITT cases that followed vaccination with Ad26.COV2.S were lethal [11], which is less than 22% of the mortality rate of VITT after ChAdOx1 nCoV-19 vaccination [12]. According to initial reports from Norway, Germany, and Austria, the mortality rates were 60% and 55%, respectively. The significant differences may be a result of either the better therapeutic methods or the fact that the first reports included more severe cases [6].

**Pathophysiology**

Even though the pathophysiology of VITT has still not been fully understood, it has been established that the essence of the pathomechanism is the formation of antiplatelet factor 4 (PF4) IgG antibodies, which directly activate platelets. That leads to the formation of thrombosis both in veins and arteries [6] followed by thrombocytopenia, hypofibrinogenemia, and consumptive coagulopathy with elevated D-dimer levels [3, 6]. The antiPF4 antibodies bind to platelets’ FvIIIa receptors, which contributes to platelet clearance and results in release of procoagulant microparticles containing PF4 as well as phosphatidylserine and tissue factor, which promote formation of thrombi. Moreover, the anti-PF4 antibodies present in VITT cause activation of monocytes, neutrophils, and endothelial cells, which further contribute to tissue factor expression [3]. Tissue factor typically contributes to the formation of venous thrombosis and is vital for cerebral microvascular thrombogenesis, which explains the atypical sites of thrombosis formation in VITT [6, 13].

The binding site of immunoglobulins binding to the PF4 is an 8 amino acid region and is contained within the binding site of antibodies found in heparin-induced thrombocytopenia (HIT) [2–4]. That
explains the inhibition of antibody binding in PF4 enzyme immunoassays caused by heparin [6]. The antibodies against platelet factor 4, which are found in serum of patients affected by HIT, correspond to the antibodies present in VITT patients and are detected by the same enzyme-linked immunosorbent assays. However, other rapid assays that are used to identify HIT PF4/H antibodies tend to be negative in VITT [3]. This suggests that despite the similarities between those two diseases [4, 14] the pathogenesis is different [2].

It has been proven that adenoviruses have the ability to directly activate platelets and induce formation of platelet-leukocyte aggregates [15]. Viral infections, in general, can contribute to complications such as suppression of thrombocyte production or destruction of platelets caused by immune complexes which may cause thrombocytopaenia [6]. It has been reported that the hexon proteins of adenoviruses used in ChAdOx1 nCoV-19 can bind to PF4 and therefore a new antigen is created which, after being phagocytosed by monocytes and transported to lymph nodes, stimulates proliferation of anti-PF4 memory B cells [3, 6]. That results in massive production of anti-PF4 antibodies. However, it is worth mentioning that neither adenoviral infections nor adenoviral vector vaccines have been associated with thrombosis, which is a strong argument against this theory [6].

Clinical presentation

The onset of VITT symptoms occurs 4 or 5 to 30 days (median time 14 days) after vaccination with either AstraZeneca–Oxford or Johnson & Johnson vaccine [3, 6, 7]. Other guidelines suggest a timeframe of 4–42 days with the latest manifestations of the disease being pulmonary embolism and deep vein thrombosis [2, 3]. Headaches are the most frequent symptom of VITT [5] and are typically accompanied by red flags including resistance to standard symptomatic treatment and progressive aggravation [5]. Typical symptoms of VITT include backache, chest pain, abdominal pain, leg pain, seizures, double vision, aphasia, focal weakness, shortness of breath, and leg swelling. On the patient’s skin purpura, bruising, or petechiae may be observed [3, 5, 7]. Patients with VITT often develop decompensated disseminated intravascular coagulation with all its manifestations in laboratory tests results [3].

Thromboses are formed at atypical sites, with cerebral vein thrombosis being the most frequent and constituting about 50% of thrombotic complications [6]. Other common locations are deep vein thrombosis of the leg, pulmonary embolism, and splanchic vein thrombosis [2, 6, 7]. Arterial thrombosis or thrombosis in adrenal veins, although less frequent, may also occur [3, 6]. In the case of bilateral adrenal vein thrombosis, acute adrenal failure may follow [3]. Formation of pulmonary embolism, intracerebral haemorrhage, or coronary thrombosis in VITT patients may result in sudden death [3].

Diagnosis

Five criteria for the diagnosis of the VITT have been distinguished by the UK Expert Haematology Panel, and all 5 must be present for the definite diagnosis (Table 1). Those criteria are as follows:

1. The onset of symptoms occurring 5–30 days after vaccination.
2. Presence of thrombosis.
4. D dimer > 4000 µg/l fibrinogen-equivalent units (FEU).
5. Positive anti-PF4 Abs ELISA assay.

The diagnosis is considered to be probable when the D-dimer levels are > 4000 FEU but one of the other criteria is not met or when the D-dimer levels are within the 2000–4000 FEU range and all of the other criteria are present [16].

It is necessary to obtain information on the patient’s exposure to heparin because HIT can present similar symptoms to VITT [7].

Laboratory assessment in VITT should contain the following tests [7]:

1. Complete blood count (CBC) with platelet count and peripheral blood smear.
2. The reports for median platelet count range from 32.5 × 10^9 cells/l to 47 × 10^9 cells depending on the source (range: 5–127 × 10^9 cells/l) [4, 6, 8].

<table>
<thead>
<tr>
<th>Criteria for the diagnosis of the VITT</th>
<th>Onset of symptoms</th>
<th>Presence of thrombosis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>5–30 days after vaccination</td>
<td>&lt; 150 × 10^9/l (thrombocytopaenia)</td>
</tr>
<tr>
<td>Platelet count</td>
<td></td>
<td>&gt; 4000 µg/l FEU</td>
</tr>
<tr>
<td>D-dimer level</td>
<td></td>
<td>Positive</td>
</tr>
<tr>
<td>Anti-PF4 Abs ELISA assay</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>VITT definitive</th>
<th>All 5 criteria are present</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>VITT probable</th>
<th>D-dimer level × 4000 FEU + 3/4 other criteria are met</th>
<th>OR D-dimer level 2000–4000 FEU + all 4/4 other criteria are met</th>
</tr>
</thead>
</table>

Platelet counts correlate with the severity of the syndrome as well as the speed of progression of symptoms [4]. Patients whose conditions do not require admission to the ICU have higher platelet counts than those in more severe conditions that required admission to the ICU [4].

For every 50% decrease in the baseline platelet count, the odds of death increase by a factor of 1.7 [12].

According to the American Society of Hematology, normal platelet count with simultaneous presence of thrombosis may indicate an early stage of VITT [17].

A lack of thrombocytopenia should not be used to exclude VITT, because certain individuals have physiologically higher baseline platelet counts [2, 3].

The presence of small aggregates of 5–10 platelets visible in a blood smear allows the distinction of VITT from immune thrombocytopenic purpura (ITP) because they are absent in the latter [5].

2. D-dimer levels
   It is typical for the D-dimer concentrations to be significantly elevated above 4000 FEU [6]. It is claimed that highly raised D-dimer levels provide more consistent proof of VITT than thrombocytopenia [2]. The mortality rate increases by a factor of 1.7 for every 10,000 FEU increase [12].

3. Fibrinogen
   Despite their minor importance in diagnosing VITT, fibrinogen levels are significant when assessing bleeding risk and monitoring results of IVIG and anticoagulation treatment [2]. The odds of death in VITT are increased by a factor of 1.7 for every 50% decrease in the baseline fibrinogen level [12].

4. Coagulation panel (prothrombin time (PT), international normalized ratio (INR), and activated partial thromboplastin time (aPTT))
   It is advised that the blood sample be collected before anticoagulation therapy because it affects the INR/aPTT results [5].

5. Platelet factor 4 - enzyme-linked immunosorbent assay (PF4-ELISA)
   Due to the low incidence of ELISA false-negative results, it is recommended that the test be either run on another ELISA assay or a functional assay be performed before excluding VITT [2, 5].

It has been proven that anti-PF4 antibody ELISA assays, when modified for VITT, increase both sensitivity and specificity [2].

A positive result of this test is not alone sufficient to make diagnosis because some studies report individuals who test positive for anti-PF4 antibodies even before vaccination [3].

6. PF4-dependent platelet activation assay
   The standard serotonin release assay (SRAs) with inclusion of heparin allows the differentiation between HIT and VITT, because the inclusion of heparin in SRAs testing of VITT tends to inhibit platelet activation [2]. Although not necessary, the SRA test results may prove helpful when ELISA tests turn negative despite strong arguments for VITT diagnosis [3]. If the result is negative, VITT or HIT are excluded and treatment with heparin is possible [2]. It is important to keep in mind that this test should be performed before treatment with intravenous immunoglobulins, to obtain valuable results [2, 5, 7].

Due to the observable tendency in VITT to form thrombus in cerebral sinuses, splanchnic thrombosis, pulmonary emboli, or deep vein thrombosis, intracranial imaging (computed tomography (CT) or magnetic resonance imaging (MRI)), transabdominal imaging, thoracic imaging, and duplex ultrasonography of lower extremities should be performed, respectively [7]. It has been established that the mortality rate of VITT increases by a factor of 2.7 among patients with cerebral venous sinus thrombosis [12].

Despite the significant importance of radiological examination of VITT, it must be remembered that negative results do not guarantee that thrombosis in the same location will not develop later [4, 5]. The inability to document thrombosis should not be a reason to abstain from treatment for VITT [3]; hence, the necessity of repeated imaging for patients who present persistent and/or progressive symptoms of TTS. Repeat imaging should be performed 1 week after the beginning of the anticoagulation therapy. What is more, post-mortem examinations have proven that the thrombosis turns out to be more widespread and significant than imaging tests usually suggest [6].

The following diseases should be taken into consideration as a differential diagnoses:

1. Thrombotic thrombocytopenic purpura (TTP)/haemolytic–uremic syndrome (HUS) – Schistocytes are present on the blood smear. Microangiopathic haemolytic anaemia (MAHA) occurs due to mechanical fragmentation of erythrocytes within occluded vessels. Haemolytic anaemia is present with LDH activity increased and unconjugated hyperbilirubinemia. In addition to neurological signs, fever and renal impairment are observed. In TTP ADAMTS13 activity is severely reduced (< 5%), and in HUS ADAMTS13 activity is > 10% [3, 18, 19].

2. Catastrophic antiphospholipid syndrome – antiphospholipid antibodies are present.

3. Immune thrombocytopenic purpura – typically APTT and INR are within the normal range, D-dimers are only mildly elevated or are at normal levels, the antiplatelet antibodies found in ITP do not activate platelets and do not cause thrombosis, and therefore anticoagulation treatment in this syndrome can only contribute to greater risk of bleeding [3].

4. Heparin-induced thrombocytopenia – connected with heparin usage, the PF4 antibodies are heparin dependent [3].

5. COVID-19 – can contribute to the formation of thrombosis and impact coagulation; however, positive PF4 assay results are not typically present.
Treatment

VITT treatment consists of 3 key concepts: anticoagulation, modulation of the autoimmune processes, and management of complications [6] (Figure 1).

During the treatment of VITT, assistance of a haematologist and neurologist should be available [3, 5, 16]. Because the patient’s condition may deteriorate rapidly, the treatment should take place in the inpatient setting and should not be delayed [3, 5]. It is estimated that around 5% of VITT patients do not present overt thrombosis. However, in these cases, the treatment should not differ from the one described below [5].

The anticoagulation therapy should be based on non-heparin anticoagulants such as fondaparinux, bivalirudin, argatroban, danaparoid, or direct oral anticoagulants [6, 7]. The doses used should correspond to the dosage used to treat uncomplicated venous thromboembolism (VTE), with appropriate adjustments for kidney functioning [3, 5]. It is recommended that the treatment begin with parenteral agents, which can be switched to direct oral anticoagulants (e.g. dabigatran, rivaroxaban) in the subacute and chronic phase of the disease [6]. It is claimed that thrombocytopaenia is not a contraindication to the anticoagulation treatment, and neither is cerebral venous thrombosis [6]. If the fibrinogen concentration decreases below 1.5 g/l, fibrinogen concentrate is required [5, 17]. Conversely, if the fibrinogen concentration decreases below 1.5 g/l, fibrinogen concentrate or cryoprecipitate should be applied [3].

Very severe cases with platelet counts less than 30 × 10^9/l may be treated with plasma exchange [3, 5]. It is worth mentioning that the VITT treatment methods described above have not yet been approved by drug regulatory agencies around the world, and hence it is off-label treatment. Therefore, patients should be well informed of potential adverse effects and give informed consent for the treatment.

It is safe and indicated for pregnant or lactating patients with VITT to undergo CT scans, MRIs, IVIG, danaparoid, or fondaparinux treatment; however, direct oral inhibitors of factor Xa are not recommended. Treatment with short courses of steroids should be consulted with obstetricians and maternal foetal medicine specialists [5].

The patients should have platelet counts monitored daily when hospitalized [3]. Although no clear requirements for discharge have yet been published, it is recommended that inpatient management continue until the following occur [3]:
1. The platelet count is > 50 × 10^9/l and improving for at least 2–3 days.
2. Stable anticoagulation is administered and no new or progressive thrombosis arises.
3. There is no bleeding for at least 2–3 days.
4. Appropriate follow-up has been assured.

Since VITT is an autoimmune disease, it is vital to modulate the autoimmune phenomenon. Urgent treatment with intravenous immunoglobulin (1 g/kg) for 2 days is strongly recommended [5–7]. This form of treatment not only neutralizes antibodies but also inhibits VITT-mediated platelet activation [6]. Treatment of cerebral venous sinus thrombosis (CVST) and splanchnic thrombosis should comprise adding another dose of 1 g/kg body weight of IVIC [5]. A second dose of IVIG should be applied on the 3rd and 4th day if the platelet count and D-dimer levels fail to respond to initial treatment or if the patient’s condition deteriorates [5].

The American Society of Hematology recommends treatment with intravenous immunoglobulin and non-heparin anticoagulation for patients vaccinated with adenoviral vaccine 5–30 days prior to the symptoms and who suffer from headache, presence with thrombocytopaenia and high D-dimer levels, even if the results for PF4 ELISA testing is not yet available [3, 7, 17].

In general, platelet transfusions are contraindicated; however, if platelet count decreases below 30 × 10^9/l, it should be considered as a form of prevention from major bleeding [6], especially if surgery is required [5, 17]. Conversely, if the fibrinogen concentration decreases below 1.5 g/l, fibrinogen concentrate or cryoprecipitate should be applied [3].

Supportive care daily platelet count monitoring in case of major bleeding – platelet transfusion

Anticoagulation for minimum of 3 months

Non-heparin anticoagulants:
- fondaparinux
- bivalirudin
- argatroban
- danaparoid
- direct oral anticoagulants

Management of complications

Treatment

Modulation of the autoimmune processes

- Urgent treatment with intravenous immunoglobulin (1 g/kg) for 2 days
- In severe cases – plasma exchange
- Platelet transfusion contraindicated
- Heparin contraindicated

Figure 1. Treatment options for vaccine-induced immune thrombotic thrombocytopenia

Medical Studies/Studia Medyczne 2022; 38/3
It is recommended that the patient’s platelet count, D-dimer levels, and fibrinogen concentration are measured as often as every 2–3 days for the first 2 weeks following hospital discharge [6]. Occurrence of VITT syndrome disqualifies individual from receiving any dose of any adenoviral vector vaccine but not mRNA-based vaccines, which are safe and recommended [3, 5, 6]. It is advised that VITT management pathways be placed in medical facilities prior to the start of massive vaccination campaigns [5].

**Conclusions**

Since the discovery of VITT, knowledge of its pathophysiology has been considerably widened, although it is yet to be fully understood. Along with multiple case studies, it has allowed organizations such as the WHO, ASH, and BSH to provide guidelines referring to diagnosis criteria and management options. According to those, patients with suspected VITT should undergo the following diagnostic tests: CBC with platelet count and peripheral blood smear, D-dimer level, fibrinogen coagulation panel, PF4-ELISA, and PF4-dependent platelet activation assay. Treatment should comprise anticoagulation, modulation of the autoimmune processes, and management of complications. These guidelines empowered healthcare professionals to identify and treat VITT more effectively. Further investigation of the pathophysiology and long-term management options would allow for safer administration of adenoviral-vector vaccines, which are a viable prevention method against COVID-19.

**Conflict of interest**

The authors declare no conflict of interest.

**References**


Address for correspondence:
Krzysztof Gomułka MD, PhD
Department of Internal Medicine, Pneumology and Allergology
Wrocław Medical University
Wrocław, Poland
E-mail: kgomulka@wp.pl