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Australian New Zealand approach to diagnosis and management of vaccine induced immune thrombosis and thrombocytopenia

Vivien Chen
Haematologist
Concord Hospital
Department of Haematology
Concord, New South Wales, Australia

Associate Professor
The University of Sydney
ANZAC Research Institute
Concord, New South Wales, Australia

Jennifer L Curnow
Director of Clinical Haematology
Westmead Hospital
Haematology and Westmead Haemophilia Treatment Centre
Westmead Hospital
Westmead, New South Wales, Australia

Huyen Tran
Head, Haemostasis Thrombosis Unit
Alfred Hospital
Department of Clinical Haematology
Melbourne, Victoria, Australia

Professor
Monash University
Central Clinical School
Clinical Haematology
Australian Centre for Blood Diseases
Melbourne, Victoria, Australia

Philip Young-Ill Choi
Staff Specialist
The Canberra Hospital
Department of Haematology
Garran, Australian Capital Territory, Australia

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Vivien Chen (Concord Hospital, Sydney)
Huyen Tran (Alfred Hospital, Melbourne)
Philip Choi (The Canberra Hospital, ACT)
Jennifer Curnow (Westmead Hospital, Sydney)
Sanjeev Chunilal (Monash Medical Centre, Melbourne)
Christopher Ward (Royal North Shore Hospital, Sydney)
Freda Passam (Royal Prince Alfred Hospital, Sydney)
Timothy Brighton (Prince of Wales Hospital, Sydney)
Beng Chong (St George Hospital, Sydney)
Robert Bird, (Princess Alexandra Hospital, Brisbane)
Anoop Enjeti (John Hunter Hospital, Newcastle)
Leonardo Pasalic (ICPMR, Sydney)
Emmanuel Favaloro (ICPMR, Sydney)
Chee Wee Tan (Royal Adelaide Hospital, Adelaide)
Ross Baker (Perth Blood Institute, Murdoch University, WA)
Simon McCrae (Launceston Hospital, Tasmania)
Ibrahim Tohidi-Esfahani (ANZAC Research Institute. Sydney)
Elizabeth Gardiner (Australian National University, Canberra)
Joanne Joseph (St Vincent's hospital, Sydney)
Danny Hsu (Liverpool hospital, Sydney)
Laura Young (Auckland City Hospital, NZ)
Claire McClintock (Auckland City Hospital, NZ)
Eileen Merriman (Waitemata, NZ)

HSANZ contributers:
Steven Lane
Leanne Berkhan

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Introductory line:

VITT is a potential complication of ChAdOx1-nCov-19 vaccination. Early recognition is key to improved outcomes.

Abstract:
Vaccinations against coronavirus 2019 with ChAdOx1 nCov-19 are a key component of the Australian government response to the pandemic. However, a syndrome of thrombocytopenia and potentially severe thrombosis has been described in a small proportion of patients receiving this vaccine. We present the current recommendations from the Thrombosis and Haemostasis Society of Australia and New Zealand (THANZ) on the diagnosis and management of this novel complication of COVID-19 vaccination.

Background

A syndrome of thrombosis and thrombocytopenia has been described in a small proportion of patients vaccinated against severe acute respiratory distress syndrome coronavirus 2 (SARSCoV2). The thrombosis can be severe and on occasion, fatal. The syndrome has been described in patients receiving adenovirus vector ChAdOx1 nCov-19 (AstraZeneca) or Ad26.COV2.S (Johnson & Johnson/Janssen) encoding SARSCoV2 Spike protein(1-4).

Reporting rates of this syndrome currently vary. A Norwegian case series identified five patients from a population of 132,686 receiving ChAdOx1 nCov-19; Medicines & Healthcare Products Regulatory Agency (MHRA) received 209 reports after 22 million first doses of ChAdOx1 nCoV-19 (2, 4, 5) and at time of writing, 24 cases have been confirmed in Australia with 2.1 million ChAdOx1 nCov-19 vaccines given.

This novel thrombosis syndrome is known by several names: vaccine-induced immune thrombotic thrombocytopenia (VITT); vaccine-induced prothrombotic immune thrombocytopenia (VIPIT); and thrombosis with thrombocytopenia syndrome (TTS). For consistency with the prevailing literature, we refer to this syndrome as VITT.

Antibodies against platelet factor 4 (PF4) or PF4/polyanion complexes have been identified in this syndrome. Serum/plasma from these patients directly and strongly activates platelets ("functional assays"). An immunological basis for this syndrome is further supported by the ability of both pooled human immunoglobulins (IVIg) and a monoclonal antibody called "IV.3" which blocks the immune complex receptors on platelets (FcγRIIa), to abrogate platelet activation(1, 3).

The syndrome is analogous to, but distinct from another thrombotic thrombocytopenic syndrome – "spontaneous" or "autoimmune" heparin induced thrombocytopenia (HIT). Similar to autoimmune HIT, in VITT the serological activation of platelets is abolished with high concentrations of heparin and enhanced by PF4(6). In contrast to HIT, VITT cases are not associated with antecedent exposure to heparin and a heparin independent hyperactive platelet response is seen in *in vitro* assays. VITT is a distinct syndrome from HIT and standard HIT diagnostic pathways are NOT appropriate for the diagnostic work-up.

Thrombosis driven by classical factors of Virchow's triad is not uncommonly coincidental to vaccination. VITT is rare, however it requires an alternate, immediately instituted management pathway. Understanding of this syndrome is rapidly evolving and currently only observation studies are available to formulate expert consensus guidance (evidence grading: low). Against this background, we present our current strategy for the Australian context.

Methodology:
Australia and New Zealand experts in thromboembolic disorders and/or laboratory haemostasis drafted an approach to VITT investigation and management appropriate for the Australian context in March 2021. All publications and pre-publications on VITT, information from regulatory authorities and available guidance from international societies were reviewed and discussed in a series of online meetings, revisions made via email and consensus published in a living online document on April 1, 2021. Additional published evidence continues to be curated and circulated prior to a weekly update meeting in which clinical and laboratory data of all confirmed Australasian VITT cases are reviewed. Updates are uploaded after consensus. https://www.thanz.org.au/documents/item/591

When to suspect VITT
Patients who present with symptom onset suggestive of thrombosis or thrombocytopenia 4 to 30 days following vaccination with ChAdOx1 nCov-19 or Ad26.COV2.S merit urgent clinical assessment to exclude this syndrome.
While cases of well controlled thrombosis have been encountered, the tempo of disease can be catastrophic within hours and we strongly advise careful clinical review of persistent symptoms with repeat screening blood tests in patients with high index of suspicion. Thrombosis in the cerebral venous sinus system (CVST); splanchnic circulation (portal, mesenteric, hepatic), deep vein, pulmonary and arterial circulation have all occurred.

While early reports demonstrated an over-representation with females (80%) aged between 22–54 years (1, 2), the Australian experience does not demonstrate a strong gender bias and vigilance irrespective of age and gender is strongly recommended.

How to investigate for VITT?
THANZ criteria for VITT diagnosis is summarised in Box 1, the diagnostic algorithm in Figure 1.

In patients presenting with symptoms suggestive of either thrombosis or thrombocytopenia within 4-28 days of COVID-19 vaccination, appropriate investigations should always be initiated based on the severity of presenting symptoms after clinical assessment. Neurological symptoms of CVST can include visual changes, seizures, focal neurological deficits, and symptoms of encephalopathy. Symptoms of splanchnic thrombosis may be subtle. It may be

necessary to transfer urgently to a facility where laboratory investigations and appropriate radiology are readily available.

While the suspicion of VITT is being explored:

- avoid platelet transfusions,
- do not begin heparin-based anticoagulation.

Investigation for VITT in Australia will occur in two stages – Screen and Confirm.

Screen

Blood samples marked "urgent" to assess the full blood count, D-dimer, and fibrinogen levels.

VITT is *suspected* in patients who present in the appropriate timeframe from ChAdOx1 nCov-19 vaccination with symptoms of thrombosis if (1) the platelet count is $<150x10^9$/L AND either (2) D-dimers are elevated (5x upper limit of normal; ULN) OR (3) fibrinogen is reduced. Further serum and plasma samples must be taken (at least 4x citrate tubes and 4 serum clot tubes), specialist haemostasis haematologist consultation obtained, and radiology performed and reported urgently to investigate for relevant organ-specific thrombosis (for example CT brain +/- venogram for CVST, abdominal CT for splanchnic vein thrombosis).

- If thrombosis is found, VITT is *probable*, and treatment urgently initiated with non-heparin anticoagulation and intravenous immunoglobulin (IVIg) (7).
- If no thrombosis is found, VITT remains *possible*.
- VITT is *less likely* if the platelet count is >150x109/L, but D-dimers are elevated or fibrinogen is reduced.
- VITT is *much less likely* if the platelet count is stable and >150x109/L, D-dimers are not elevated AND fibrinogen is normal. Patients can be treated as non-VITT thrombosis.

It is important to recognise that not all thrombocytopenia following vaccination is VITT. Secondary immune thrombocytopenia (ITP) from immunisation has been seen with BNT162b2 (Pfizer-BioNTech), mRNA-1273 (Moderna) vaccines and ChAdOx1 nCov-19 (AstraZeneca). An alternative diagnosis of ITP should always be considered in patients with thrombocytopenia, with or without raised D-dimers and normal fibrinogen, without evidence of thrombosis. ITP may manifest a bleeding phenotype and patients are managed with usual first-line therapies including corticosteroids and IVIg.(8)

Likewise, the majority of DVT/PE post vaccination are statistically unlikely to be VITT. Once VITT is excluded, these patients can be treated as per standard DVT/PE.

Confirm

Patients with thrombosis (*probable VITT*) or no thrombosis (*possible VITT*) or thrombosis with D-Dimer > 5xULN should be further investigated for:

- presence of PF4 or PF4/polyanion antibodies using an ELISA platform,

- the ability for serum/plasma to activate platelets *in vitro*. Platelet activating antibodies on functional testing are considered pathological, and requisite for confirming the diagnosis of VITT.

Antigen-based "VITT" immune assay: Antibodies against PF4 or PF4/polyanion complexes using ELISA methods are present in the majority of VITT. Other platforms used for HIT antibody detection such as HemosIL® AcuStar, Stago STiC Expert®, and particle gel immunoassays (PaGIA) do not reliably detect VITT antibodies and are not appropriate for use in this setting.

Functional antibody testing: *in vitro* assessment of platelet activating antibodies are available in centralised laboratories (serotonin release assay, flow cytometry procoagulant assay and whole blood aggregation assays). They should be performed in all "probable" or "possible" VITT or in "less likely" cases with a positive ELISA to confirm the diagnosis (See Box 2 for case vignettes).

Currently, there is a co-ordinated national effort to ensure timely investigation. Complete the specific request form in the following link, contact the nearest local haemostasis expert, and send samples to the appropriate listed location. https://www.thanz.org.au/documents/item/579 .

How do I treat suspected VITT?

Suspected VITT will require treatment before results of PF4/polyanion ELISA testing are available. Specialist consultation with a haemostasis haematologist is recommended.

Probable VITT(7)

- We recommend *probable* VITT (suspected WITH thrombosis) to be treated with non-heparin anticoagulation.
- IVIg (1-2g/kg a day for two consecutive days) is recommended upfront or high dose steroids if IVIg is unavailable. This is particularly important in cases at high risk from deterioration (including presentation platelets < 30 x 10^9/L, fibrinogen <1.5g/L, severe thrombosis). The haematologist may consider the addition of high dose methylprednisone and/or plasma exchange in the appropriate context (for example, progressive thrombosis or rapid deterioration).
- Anticoagulant treatment options are as per local therapeutic practice for HIT: bivalirudin, argatroban, danaparoid, fondaparinux, rivaroxaban, apixaban, dabigatran, and (after initial treatment with another agent) warfarin.
- Avoid platelet transfusion.
- Hospitalisation is considered safest until there is a reduction of *in vivo* platelet activation and thrombin generation (normalised platelet count, falling D-dimers, normal fibrinogen). We currently suggest retesting for presence of VITT antibody in confirmed cases at 6 weeks, 3 months and 6 months, and prior to cessation of anticoagulation.

Possible VITT

- We recommend *possible* VITT (suspected WITHOUT thrombosis) be monitored closely with repeat FBC, D-dimer, fibrinogen approximately every three days.
- Anticoagulation with a non-heparin anticoagulant should be considered – particularly with very high D-dimers and a positive immunoassay. We currently recommend consideration of fondaparinux or DOAC at prophylactic dosing until normalised platelet count, falling D-dimers, normal fibrinogen. IVIg may be considered if there are any signs to suggest progression (9). Anticoagulation duration should be time limited or until HIT ELISA and functional testing is negative.

We recommend against second dose ChAdOx1 nCov-19 in patients with confirmed or strongly suspected VITT (10, 11).

Conclusions

ChAdOx1 nCov-19 is a key component of the Australian government vaccination strategy against the coronavirus 2019 pandemic. We present our current perspectives on this novel complication of ChAdOx1 nCov-19 (Box 3).

Diagnostic algorithms and treatment strategies for VITT are available and continue to be refined as local experience increases. We provide a link to the THANZ and HSANZ endorsed THANZ living guidance document and refer the reader to online resources as referenced in the text. https://www.thanz.org.au/documents/item/591

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Box 1: THANZ Diagnostic criteria for VITT:

1. Exposure to ChAdOx1 nCov-19, AstraZeneca) within 4-30 days of symptom onset
2. Thrombocytopenia or falling platelet count AND elevated D-Dimer (>5 times upper limit normal) or reduced fibrinogen
3. Thrombosis: Any deep vein thrombosis, pulmonary embolism, or arterial thrombosis. Thrombosis in uncommon sites such as CVST and splanchnic vein thrombosis are strongly suggestive..
4. Antibodies detected against PF4/polyanion and/or
5. Functional assay indicating patient derived plasma/serum induction of prothrombotic phenotype in healthy donor platelets.

* Further details available at: https://www.thanz.org.au/documents/item/591

Box 2: Case Vignettes:

Case 1:
44 year old male presented day 8 post ChAdOx1 nCov-19 with fevers, fatigue, head "fogginess" with abdominal discomfort. The platelet count was 70 $x10^9$/L (reduction to 17 $x10^9$/L within 12 hours), D-dimer 114 mg/L (ULN, 0.5 mg/L) and normal fibrinogen. Radiology demonstrated portal splenic and mesenteric thrombosis. Patient was treated as PROBABLE VITT. Treatment was immediately initiated with IVIg and anticoagulation with fondaparinux. Anticoagulation changed to bivalirudin when surgery was required. Confirmatory investigations supported a diagnosis of VITT with a positive ELISA immunoassay and serum/plasma induced platelet activation on functional assay testing (12). VITT confirmed.

Case 2:
46 year old woman with a history of quiescent SLE presents day 4 post ChAdOx1 nCov-19 (AZ) with investigations consistent with proximal DVT and bilateral pulmonary emboli without evidence of right heart strain. The platelet count was 97 $x10^9$/L and D-dimer 1.35 mg/L (ULN, 0.25 mg/L), fibrinogen was normal. Patient was treated as PROBABLE VITT, immediately anticoagulated with fondaparinux and received one dose of IVIg 1mg/kg. Platelet count was stable during hospitalisation. Confirmatory investigations returned a negative ELISA, negative functional assays and a positive lupus anticoagulant. Patient was deemed to have VTE secondary to anti-phospholipid syndrome, not VITT, and was changed to standard low molecular weight heparin (LMWH) anticoagulation. VITT not supported.

Case 3:
60 year old male smoker presented day 15 post ChAdOx1 nCov-19 (AZ) with chest pain, dyspnoea and found to have bilateral pulmonary emboli without haemodynamic compromise or right heart strain. The platelet count was 228 $x10^9$/L and D-dimer 0.6 mg/L (ULN, 0.5 mg/L), fibrinogen was normal at 3.0g/Lwith known baseline platelet count of 235 x 10^9/L. Patient was treated as MUCH LESS LIKELY to be VITT (normal platelet count), commenced apixaban at standard dosing. Repeat platelet count 3 days later was stable and confirmatory testing for VITT did not proceed. VITT not supported.

Case 4:
50 year old female presents day 4 post ChAdOx1 nCov-19 (AZ) with extensive petechiae and oral mucosal wet purpura. No symptoms of thrombosis are present on system review. Patient had no previous history of ITP or autoimmune disease. The platelet count was 3$x10^9$/L and D-dimer 1.25 mg/L (ULN, 0.25 mg/L), fibrinogen was normal at 2.2 g/L. Patient was treated as LESS likely VITT. Patient was diagnosed with ITP, likely vaccine associated and commenced IVIg 2g/kg divided over 2 days with dexamethasone 40mg daily x 4. Platelet count improved to 50$x10^9$/L day 2. Non-urgent VITT testing supported a diagnosis of NOT VITT with negative ELISA result. VITT not supported.

Case 5:
82 year old male with prostate cancer presents day 17 post ChAdOx1 nCov-19 (AZ) with dyspnoea, with background of inflammatory bowel disease was found to have sub-massive pulmonary emboli. Platelet count was 198 x 10^9/L, D-Dimer was 9.8,mg/L (ULN 0.25mg/L) and fibrinogen was normal. And 2.8g/L. Patient was treated as LESS LIKELY to be VITT (normal platelet count, markedly raised D-Dimer), commenced therapeutic fondaparinux. Confirmatory investigations did not support a diagnosis of VITT with a negative ELISA immunoassay and patient did not progress to functional testing. Patient was changed to LMWH then discharged on DOAC when stable. VITT not supported.

** Case 1 reflects a published case (Hocking J, et al. The first known ChAdOx1 nCoV-19 vaccine-induced thrombotic thrombocytopenia in Australia. Med J Aust 2021; https://www.mja.com.au/journal/2021/first-known-chadox1-ncov-19-vaccine-induced-thrombotic-thrombocytopenia-australia [Preprint, 7 May 2021]). Other vignettes do not refer to specific individuals.

Abbreviations: VITT (vaccine induced thrombocytopenia and thrombosis); ULN (Upper limit of normal); LMWH (low molecular weight heparin); DOAC (direct oral anticoagulant).

Box 3: Key points

- Patients who present with symptoms suggestive of thrombosis from days 4 to 30 after ChAdOx1 nCov-19 (AstraZeneca) vaccination need urgent blood tests looking for thrombocytopenia, markedly elevated D-dimers, and hypofibrinogenemia.
- Radiological investigation should be expedited to look for thrombosis.
- When the diagnosis of VITT is *probable*, treatment should be initiated urgently with IVIg, and non-heparin anticoagulation.
- Platelet transfusions should be avoided.
- VITT is a distinct syndrome that is separate to HIT. Standard HIT diagnostic pathways are NOT appropriate for the diagnostic work-up and specific VITT tests are required. For all suspected cases, samples (4 citrate and 4 serum) are to be collected **prior** to treatment. https://www.thanz.org.au/documents/item/579
- Readers are strongly encouraged to access the latest updated guidelines available online (link). https://www.thanz.org.au/documents/item/591

Figure 1: Diagnostic and management pathway for VITT