Pathologic Antibodies to Platelet Factor 4 after ChAdOx1 nCoV-19 Vaccination

Marie Scully, M.D., Deepak Singh, B.Sc., Robert Lown, M.D., Anthony Poles, M.D., Thomas Solomon, M.D., Marcel Levi, M.D., David Goldblatt, M.D., Ph.D., Pavel Kotoucek, M.D., William Thomas, M.D., and William Lester, M.D.

ABSTRACT

BACKGROUND
The mainstay of control of the coronavirus disease 2019 (Covid-19) pandemic is vaccination against severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Within a year, several vaccines have been developed and millions of doses delivered. Reporting of adverse events is a critical postmarketing activity.

METHODS
We report findings in 23 patients who presented with thrombosis and thrombocytopenia 6 to 24 days after receiving the first dose of the ChAdOx1 nCoV-19 vaccine (AstraZeneca). On the basis of their clinical and laboratory features, we identify a novel underlying mechanism and address the therapeutic implications.

RESULTS
In the absence of previous prothrombotic medical conditions, 22 patients presented with acute thrombocytopenia and thrombosis, primarily cerebral venous thrombosis, and 1 patient presented with isolated thrombocytopenia and a hemorrhagic phenotype. All the patients had low or normal fibrinogen levels and elevated D-dimer levels at presentation. No evidence of thrombophilia or causative precipitants was identified. Testing for antibodies to platelet factor 4 (PF4) was positive in 22 patients (with 1 equivocal result) and negative in 1 patient. On the basis of the pathophysiological features observed in these patients, we recommend that treatment with platelet transfusions be avoided because of the risk of progression in thrombotic symptoms and that the administration of a nonheparin anticoagulant agent and intravenous immune globulin be considered for the first occurrence of these symptoms.

CONCLUSIONS
Vaccination against SARS-CoV-2 remains critical for control of the Covid-19 pandemic. A pathogenic PF4-dependent syndrome, unrelated to the use of heparin therapy, can occur after the administration of the ChAdOx1 nCoV-19 vaccine. Rapid identification of this rare syndrome is important because of the therapeutic implications.
CORONAVIRUS DISEASE 2019 (COVID-19) has been associated with considerable morbidity and mortality.\textsuperscript{1,2} From the onset of the Covid-19 pandemic to March 2021, more than 126.8 million cases and 2.7 million deaths were documented worldwide.\textsuperscript{3}

With unprecedented speed, vaccines against severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) have been licensed and used worldwide.\textsuperscript{4,5} Rollout of the vaccines has been uneven, but in some countries, very high levels of coverage have been achieved. In Israel, more than half the population of 9 million has received a second dose; in the United Kingdom, more than 25 million people have received at least one dose. With such widespread and rapid uptake of the vaccines, safety signals should be documented.

A safety signal of particular concern temporally associated with the administration of the ChAdOx1 nCoV-19 vaccine (AstraZeneca) has recently been described,\textsuperscript{6,7} involving an unusual clinical constellation of abnormal clotting, particularly cerebral venous thrombosis, and thrombocytopenia that has resulted in death in some cases. Intensive reviews of the risk of venous thromboembolism associated with vaccines against SARS-CoV-2 were conducted by both the U.K. Medicines and Healthcare Products Regulatory Agency (MHRA) and the European Medicines Agency (EMA). After their initial reviews, both agencies confirmed that the risk of venous thromboembolism associated with the vaccines was not higher than the background risk in the general population and emphasized the overwhelmingly favorable risk–benefit ratio for vaccines against SARS-CoV-2. However, although a causal association has not yet been confirmed,\textsuperscript{8} they have acknowledged that vaccines against SARS-CoV-2 may be associated with a rare but serious adverse event related to thrombosis, primarily cerebral venous thrombosis, and thrombocytopenia.\textsuperscript{9}

To gain a better understanding of the described clinical syndrome of concern, we conducted a study involving 22 patients referred to a specialist hematologist for the evaluation of thrombosis and thrombocytopenia and 1 patient referred for the evaluation of isolated thrombocytopenia and a very high D-dimer level temporally associated with the administration of the first dose of the ChAdOx1 nCoV-19 vaccine. The thrombosis events were primarily cerebral venous thrombosis but also included arterial thrombosis and more common types of venous thromboembolism, such as pulmonary embolism. We identified a novel underlying mechanism associated with the presentation. On the basis of the clinical features and subsequent laboratory findings, we propose an altered therapeutic approach and give guidance on the assessment of patients who present with this rare syndrome.

### Methods

**Patient Identification**

Patients were identified for the investigation of suspected vaccine-induced thrombosis and thrombocytopenia (i.e., vaccine-induced immune thrombotic thrombocytopenia, or VITT). No evidence of hereditary or acquired thrombophilia had been identified at the referring hospitals. In the 3 index patients, the decision to test for antibodies to platelet factor 4 (PF4) was based on the presence of progressive thrombosis, thrombocytopenia, and clinical features similar to those seen in patients with heparin-induced thrombocytopenia (HIT), despite the absence of exposure to heparin. Within less than 7 days, a total of 23 patients were identified for testing, including new patients who were admitted with suggestive clinical features and 3 patients who had died after a clinical course consistent with this syndrome.

**Enzyme-Linked Immunosorbent Assays**

Testing for anti-PF4 antibodies was performed by means of enzyme-linked immunosorbent assays (ELISAs) at six reference laboratories in the United Kingdom. Additional testing for anti-PF4 antibodies was performed by means of various techniques used locally for HIT testing at individual centers. In many cases, HIT testing with the chemiluminescence HemosIL AcuStar HIT IgG assay (Werfen) was negative but testing with an ELISA was positive. ELISAs included the Life- codes PF4 IgG assay (Immucor) and the Assera-chrom HPIA IgG assay (Stago). These assays have a normal range (median plus standard deviation) of anti-PF4 antibodies for healthy controls and a range for patients who have received heparin. The positive thresholds were based on the normal ranges (≤0.238 optical density units
[OD] for the Asserachrom HPIA IgG assay and <0.40 OD for the Lifecodes PF4 IgG assay.10

FUNCTIONAL HIT ASSAY
In several cases, the ELISA results were confirmed by means of a functional HIT assay (HITAlert, Diapharma) performed at a reference laboratory in accordance with the manufacturer’s instructions.11 In brief, platelet-rich plasma was prepared from blood samples obtained from volunteer donors with group O blood. The platelet-rich plasma was incubated in five tubes that contained the following substances: calcium ionophore, heparin (0.3 U per milliliter), patient serum, patient serum plus heparin (0.3 U per milliliter), and patient serum plus an excess of heparin (100 U per milliliter). A previously tested serum sample from a patient with a confirmed diagnosis of HIT was used as a positive control (Fig. 1A). A positive threshold of more than 8% platelet activation was applied in accordance with the manufacturer’s recommendations, providing a sensitivity of 78% and specificity of 98% when used to confirm the diagnosis of HIT. Data were analyzed with the use of a flow cytometer (CytoFlex, Beckman Coulter).

ADDITIONAL SEROLOGIC AND ANTIBODY TESTS
A multiplexed electrochemiluminescence assay (Meso Scale Discovery) was used, as described previously,12 to measure serum levels of antibodies to SARS-CoV-2 antigens, including spike protein, receptor-binding domain (RBD), and nucleocapsid protein; levels of functional antibodies that inhibit the interaction of angiotension-converting–enzyme 2 (ACE2)–receptor protein with spike protein and RBD of SARS-CoV-2; and levels of antibodies to spike protein of seasonal coronaviruses HCoV-OC43, HCoV-HKU1, HCoV-HL63, and HCoV-229E.

RESULTS

PATIENT CHARACTERISTICS
Among the 23 patients included in this study, the median age was 46 years (range, 21 to 77), with 16 patients (70%) younger than 50 years. Fourteen patients (61%) were female. All the patients were reported as previously fit and well by referring hospitals, with no history of a medical condition or use of a medication likely to precipitate thrombosis, except for 1 patient who had a history of deep venous thrombosis and 1 patient who was known to be taking the combined oral contraceptive pill. All the patients had received the first dose of the ChAdOx1 nCoV-19 vaccine 6 to 24 days (median, 12 days) before presentation. Mild bruising and petechiae were evident in some patients. Secondary cerebral hemorrhage was noted in some patients who had cerebral venous thrombosis. The 1 patient who did not present with thrombosis had clinically significant bruising but no other hemorrhagic manifestations.

The clinical and laboratory features of the patients are summarized in Table 1. Of the 22 patients who presented with thrombosis, 13 had clinical features consistent with cerebral venous thrombosis (1 with concurrent acute portal vein thrombosis and pulmonary embolism), 4 had pulmonary embolism (1 with concurrent deep venous thrombosis), 1 had deep venous thrombosis and bilateral adrenal hemorrhage, 2 had an ischemic stroke affecting the middle cerebral artery territory, and 2 had portal vein thrombosis (1 with concurrent acute myocardial infarction and 1 with evidence of concurrent acute aortic thrombosis on imaging). Additional thrombosis events associated with progression occurred in patients who received platelet transfusions or heparin-based therapy at presentation. Within the entire cohort, 7 patients (30%) died. Results of a postmortem evaluation, available for 1 patient, showed evidence of thrombosis in many small vessels, especially vessels in the lungs and intestine, cerebral veins, and venous sinuses, as well as evidence of extensive intracerebral hemorrhage.

LABORATORY TESTING
All the patients had a negative SARS-CoV-2 polymerase-chain-reaction assay at presentation (data not shown). The 10 patients with samples available for testing had a negative SARS-CoV-2 serologic test for antibodies to nucleocapsid protein, a finding that ruled out recent exposure to SARS-CoV-2. In all 10 patients, levels of antibodies to spike protein and RBD of SARS-CoV-2 were within the range seen in recipients of one dose of the ChAdOx1 nCoV-19 vaccine, and levels of antibodies to seasonal coronaviruses were within the range seen in recipients of the ChAdOx1 nCoV-19 vaccine and in the general population. Levels of functional antibodies that
inhibit the interaction of ACE2-receptor protein with spike protein and RBD of SARS-CoV-2 were also within the range seen in recipients of one dose of the ChAdOx1 nCoV-19 vaccine (Goldblatt D; personal communication).

Thirteen patients had low fibrinogen levels as determined with the method of Clauss (range, 0.3 to 4.5 g per liter; normal range, 1.5 to 4.0). The d-dimer levels were much higher than would be expected in patients with acute venous thromboembolism (median, 31,301 fibrinogen-equivalent units [FEU]; range, 5000 to 80,000). No other relevant laboratory tests were positive, including tests for thrombophilia, antinuclear antibodies, extractable nuclear antigen, and antiphospholipid antibodies (data not shown). A test for lupus anticoagulant was positive in 5 of the 10 patients for whom results were available, but in the context of severe coagulopathy and negative tests for antiphospholipid antibodies and anti–β2-glycoprotein 1b antibodies, these results were considered to be unreliable.

In all 23 patients, the ELISA for anti-PF4 antibodies was performed on a sample obtained before the administration of heparin-based therapy. Although HIT testing with the HemosIL AcuStar HIT IgG assay was negative in all 9 patients who were tested, an ELISA for anti-PF4 antibodies was positive in 22 of the 23 patients. One patient — who presented 10 days after vac-
the ELISA result was positive in 5 of the 7 patients who were tested. These findings confirmed OD on the Asserachrom HPIA IgG assay). equivocal result on the ELISA (a level of 0.156 indication with deep venous thrombosis and bilateral adrenal hemorrhage but with a high D-dimer level and a low fibrinogen level, with no evidence of an alternative diagnosis — had an evidence of an alternative diagnosis — had an OD at presentation that were much higher than would be expected in patients with acute venous thromboembolism13 and are typically seen in patients with cancer.14 The very conservative D-dimer cutoff used in our algorithm, of 4000 FEU, was chosen to ensure that cases were not missed and were considered for further testing, because there is a possibility that a spectrum of severity in this syndrome could be missed otherwise. In all the patients, manifestations occurred 6 to 24 days after the administration of the first dose of the ChAdOx1 nCoV-19 vaccine.

HIT is a progressive thrombotic condition that can cause both venous and arterial thrombosis, typically 5 to 14 days after exposure to heparin. It is more common in female patients, particularly those who receive unfractionated heparin during cardiac surgery, as well as in patients who receive heparin after surgery, especially cardiac and orthopedic procedures.15 Diagnosis is confirmed by the presence of anti-PF4 antibodies.16,17
Table 1. Clinical and Laboratory Characteristics of the 23 Patients in the Study. *

| Patient Number | Time from Vaccination to Admission | Platelet Count | PT | APTT | Fibrinogen | d-Dimer | HemosIL AcuStar HIT IgG Assay | Asserachrom HPIA IgG Assay | Lifecodes PF4 IgG Assay | Functional HIT Assay | Clinical Features | Outcome |
|----------------|-----------------------------------|----------------|----|------|------------|---------|-------------------------------|---------------------------|----------------------|-------------------|-------------------|---------|---------|
|               | days                              | cells/µl       | sec| sec  | g/liter    | FEU     | OD                           | OD                        | OD                   | OD                |                   |         |
| Ref           | —                                 | 150–400        | 10.0–12.0 | 25.0–37.0 | 1.5–4.0 | 0–550 | Neg                          | ≤0.238                   | <0.40               | Neg               | —                 | —       |
| 1 (F, 30)     | 13                                | 27             | 1.1†| 35.0 | 2.5        | 16,280  | Neg                          | 0.776                    | ND                   | Pos               | CVT, PVT, PE, ischemic bowel with infarction | Alive   |
| 2 (F, 55)     | 6                                 | 11             | 13.1| 1.0† | 1.1        | 26,689  | ND                           | 1.310                    | ND                   | ND                | PVT, AAT, ICH    | Died   |
| 3 (F, 26)     | 12                                | 64             | 1.1†| 1.1† | 3.2        | >5,000  | ND                           | ND                       | 2.45                 | ND                | CVT               | Alive   |
| 4 (F, 52)     | 10                                | 31             | 15.0| 35.0 | 1.2        | 37,250  | ND                           | ND                       | 2.26                 | ND                | Post mortem: thrombosis in the lungs and intestine, CVT, ICH | Died    |
| 5 (M, 38)     | 14                                | 16             | 12.8| 30.8 | 1.2        | 45,229  | Neg                          | ND                       | 2.84                 | Neg               | Extensive bilateral PE with heart strain | Died    |
| 6 (F, 49)     | 15                                | 14             | 1.4†| 36.0 | 1.3        | 39,049  | Neg                          | 0.297                    | ND                   | ND                | CVT, IJVT, SAH   | Alive   |
| 7 (M, 25)     | 9                                 | 19             | 1.2†| 1.1† | 1.3        | ND      | ND                           | 0.297                    | ND                   | ND                | CVT               | Died    |
| 8 (M, 32)     | 19                                | 87             | 14.1| 26.7 | 1.7        | ND      | 1.440                        | ND                       | ND                   | ND                | CVT               | Alive   |
| 9 (F, 35)     | 9                                 | 65             | 13.2| 28.7 | 2.2        | 10,316  | Neg                          | 1.070                    | ND                   | ND                | CVT               | Alive   |
| 10 (M, 77)    | 8                                 | ND             | 13.1| 23.0 | 2.6        | 6,018   | Neg                          | 1.640                    | ND                   | ND                | PE                | Alive   |
| 11 (M, 66)    | 12                                | 34             | 2.1 | 10,388| ND          | Neg     | 0.156                        | ND                       | ND                   | ND                | DVT, adrenal hemorrhage | Alive   |
| 12 (M, 34)    | 14                                | 23             | 14.8| 22.0 | 0.7        | 37,000  | Neg                          | ND                       | Pos                  | ND                | CVT               | Alive   |
| 13 (M, 54)    | 10                                | 71             | 13.5| 32.7 | 1.2        | 80,000  | ND                           | 0.76                     | ND                   | ND                | PVT, MI          | Died    |
| 14 (F, 71)    | 14                                | 17             | 1.4†| 1.3† | 0.8        | >20,000 | Neg                          | ND                       | Pos                  | ND                | Hemorrhagic symptoms only | Alive   |
| 15 (F, 22)    | 10                                | 100            | 11.1| 23.6 | 3.0        | >10,000 | ND                           | ND                       | 1.40                 | ND                | CVT, ICH         | Died    |
| 16 (F, 39)    | 10                                | 57             | 1.2†| 0.9† | 4.4        | >5,000  | ND                           | ND                       | 1.40                 | ND                | MCA infarct       | Alive   |

* AAT denotes acute aortic thrombosis, APTT activated partial thromboplastin time, CVT cerebral venous thrombosis, DVT deep venous thrombosis, FEU fibrinogen-equivalent units, HIT heparin-induced thrombosis, ICH intracerebral hemorrhage, IJVT internal jugular vein thrombosis, MCA middle cerebral artery, ND not done, Neg negative, OD optical density units, PE pulmonary embolism, Pos positive, PT prothrombin time, PVT portal vein thrombosis, Ref reference range, and SAH subarachnoid hemorrhage.

† The patient’s value is expressed as a ratio of the mean normal value. The reference range for the PT ratio is 1.0 to 1.2 and for the APTT ratio is 0.8 to 1.2.
Data describing the rare detection of pathologic anti-PF4 antibodies unrelated to the use of heparin therapy are limited.\(^1\)\(^8\),\(^19\) Furthermore, the analysis of anti-PF4 antibodies appears to be specific to the given assay. In our study, confirmation of ELISA results for anti-PF4 antibodies was undertaken with the use of a functional HIT assay. The clinical features of this vaccine-induced syndrome are more typical of those seen in patients with HIT who have early reexposure to heparin, including severe thrombocytopenia, aggressive thrombosis, and disseminated intravascular coagulation.\(^20\)

The risk of thrombocytopenia and the risk of venous thromboembolism after vaccination against SARS-CoV-2 do not appear to be higher than the background risks in the general population, a finding consistent with the rare and sporadic nature of this syndrome. Furthermore, headaches, fevers, and muscle aches have occurred after vaccination for 48 to 72 hours in some patients. The events reported in this study appear to be rare, and until further analysis is performed, it is difficult to predict who may be affected.

The symptoms developed more than 5 days after the first vaccine dose, reflecting an immunologic pattern similar to that of HIT. We have identified a novel mechanism and pathophysiological basis that prompts careful consideration of treatment. Avoidance of platelet transfusions is critical, because such treatment would provide a substrate for further antibody-mediated platelet activation and coagulopathy. The exact nature of these pathologic antibodies has not been characterized, but they appear to be of the IgG subtype, and platelet activation can be completely abrogated with an excess of heparin, as seen in classic HIT. Identification of the mechanism through which the vaccine could trigger the formation of these pathologic antibodies would require further study. An understanding of the precise pathophysiological mechanism may allow for more targeted therapeutic interventions.

Although evidence does not yet suggest that the use of heparin will exacerbate this condition, pending further data, we would recommend considering anticoagulation with the use of a non-heparin anticoagulant agent, such as argatroban, danaparoid, fondaparinux, or direct oral anticoagulants. Intravenous immune globulin (IVIG) has been used successfully in the treat-

| Patient Number (Sex, Age in yr) | Time from Vaccination to Admission | Platelet Count | PT | APTT | Fibrinogen | d-Dimer | HemosIL | AcuStar | HIT IgG Assay | Functional HIT Assay | Clinical Features | Outcome |
|--------------------------------|-----------------------------------|----------------|-----|------|------------|---------|---------|---------|--------------|--------------------|--------------------|---------|
| 17 (F, 70)                     | 17                                | 22,903         | 1.1† | 1.4† | 3.8        | >5,000  | ND      | ND      | Pos          | ND                 | PE (saddle embolism) with cardiac arrest, DVT in the leg | Died    |
| 18 (M, 21)                     | 10                                | 31,301         | 1.1  | 1.1  | 1.1        | <0.4    | ND      | ND      | Pos          | MCA infarct        | DVT in the leg | Alive   |
| 19 (F, 46)                     | 14                                | 62,342         | 1.2  | 1.0  | ND         | 4.5     | ND      | ND      | ND           | CVT                | Alive             | Alive   |
| 20 (F, 32)                     | 12                                | 62,854         | 1.5† | 1.7† | <0.4       | 2.17    | ND      | ND      | Pos          | CVT                | CVT                | Died    |
| 21 (M, 48)                     | 24                                | 71,859         | 1.3† | 1.0† | ND         | 2.45    | ND      | ND      | Pos          | CVT                | Alive             | Alive   |
| 22 (F, 49)                     | 24                                | 71,859         | 1.3† | 1.0† | ND         | 2.45    | ND      | ND      | Pos          | CVT                | Alive             | Alive   |
| 23 (F, 46)                     | 10                                | 71,859         | 1.3† | 1.0† | ND         | 2.45    | ND      | ND      | Pos          | CVT                | Alive             | Alive   |

* AAT denotes acute aortic thrombosis, APTT activated partial thromboplastin time, CVT cerebral venous thrombosis, DVT deep venous thrombosis, FEU fibrinogen equivalent units, HIT heparin-induced thrombocytopenia, ICH intracerebral hemorrhage, IJVT internal jugular vein thrombosis, MCA middle cerebral artery, ND not done, Neg negative, OD optical density units, PE pulmonary embolism, Pos positive, PT prothrombin time, PVT portal vein thrombosis, Ref reference range, and SAH subarachnoid hemorrhage.

† The patient’s value is expressed as a ratio of the mean normal value. The reference range for the PT ratio is 1.0 to 1.2 and for the APTT ratio is 0.8 to 1.2.
ment of patients with “spontaneous” autoimmune HIT, which is the closest comparison to this vaccine-induced syndrome, and IVIG would be expected to have direct antibody-mediated toxic effects.21,23 Plasma exchange with plasma rather than albumin could also be effective in temporarily reducing levels of pathologic antibodies and providing some correction of the coagulopathy in terms of the hypofibrinogenemia.

A suggested algorithm for identification of vaccine-induced thrombosis and thrombocytopenia is presented and can be adapted as we generate further information. The combination of thrombosis and an apparent consumptive coagulopathy poses a dilemma with respect to the benefits and risks associated with aggressive anticoagulation. This dilemma is especially relevant in patients with cerebral venous thrombosis,

Figure 2. Suggested Algorithm for Testing and Treatment of Patients Presenting with Thrombosis and Thrombocytopenia 5 to 30 Days after Vaccination.
The HemosIL AcuStar HIT IgG assay is not recommended for the evaluation of suspected vaccine-induced thrombosis and thrombocytopenia. ELISA denotes enzyme-linked immunosorbent assay, FEU fibrinogen-equivalent units, HIT heparin-induced thrombosis, and IVIG intravenous immune globulin.

- Patient presents with acute thrombosis and thrombocytopenia
  - Refer to hematologist
  - Perform tests for prothrombin time, activated partial thromboplastin time, fibrinogen level, and D-dimer level
  - D-dimer level, <2000 FEU, normal results of coagulation tests and fibrinogen level
    - Vaccine-induced thrombosis and thrombocytopenia is unlikely
  - D-dimer level, >4000 FEU, low or normal fibrinogen level, no evidence of alternative diagnosis
    - Vaccine-induced thrombosis and thrombocytopenia is suspected
      - Avoid platelet transfusion
      - Administer IVIG
      - Consider use of glucocorticoids
      - Administer nonheparin anticoagulant
      - Consider treatment to increase fibrinogen level to >1.0 g/liter
      - Perform HIT ELISA
      - Positive
        - Continue treatment
      - Negative
        - Review diagnosis and treatment
          - Consider alternative or functional HIT assay

- Vaccine-induced thrombosis and thrombocytopenia is unlikely
  - Refer to hematologist

- Vaccine-induced thrombosis and thrombocytopenia is suspected
  - Perform tests for prothrombin time, activated partial thromboplastin time, fibrinogen level, and D-dimer level
  - D-dimer level, <2000 FEU, normal results of coagulation tests and fibrinogen level
    - Vaccine-induced thrombosis and thrombocytopenia is unlikely
  - D-dimer level, >4000 FEU, low or normal fibrinogen level, no evidence of alternative diagnosis
    - Vaccine-induced thrombosis and thrombocytopenia is suspected
      - Avoid platelet transfusion
      - Administer IVIG
      - Consider use of glucocorticoids
      - Administer nonheparin anticoagulant
      - Consider treatment to increase fibrinogen level to >1.0 g/liter
      - Perform HIT ELISA
      - Positive
        - Continue treatment
      - Negative
        - Review diagnosis and treatment
          - Consider alternative or functional HIT assay
in whom bleeding could be catastrophic but withholding anticoagulation could be equally harmful. It is unclear whether delaying aggressive anticoagulation until after initial disease control with IVIG or plasma exchange is warranted, but mortality among patients with cerebral venous thrombosis appears to be higher than expected, so early treatment decisions are likely to be critical. There is no evidence that heparin alternatives are required; however, in view of the similarity of this syndrome to conventional HIT, alternatives could be considered until further data are available.

In all cases reported to date, this syndrome of thrombocytopenia and venous thrombosis appears to be triggered by receipt of the first dose of the ChAdOx1 nCoV-19 vaccine. Although there have been a few reports of patients with symptoms consistent with this clinical syndrome after the receipt of other vaccines against SARS-CoV-2, none have yet been confirmed to fulfill the diagnostic criteria, specifically the presence of thrombocytopenia, thrombosis, a very high D-dimer level, and a low or normal fibrinogen level. Furthermore, in Israel, where two doses of the BNT162b2 vaccine (Pfizer–BioNTech) have been provided to more than 4 million people, no cases of this rare syndrome have been reported.

Although natural SARS-CoV-2 infection has been associated with thromboembolic phenomena, those events differ from the specific syndrome described in this study.

The risk of Covid-19 remains a serious public health consideration worldwide, and vaccination against SARS-CoV-2 provides critical protection.24 There is a substantial risk of ascertainment bias when associating adverse clinical events with vaccination; however, the syndrome described in this study has a combination of clinical and laboratory features that is exceptional and has not been previously observed by any of the authors who are specialist hematologists or neurologists. Ongoing data collection and studies could help to establish whether and how the development of pathologic platelet-activating anti-PF4 antibodies, unrelated to the use of heparin therapy, could be associated with vaccination against SARS-CoV-2.

Disclosure forms provided by the authors are available with the full text of this article at NEJM.org.

We thank the following colleagues for their input: Drs. Sue Pavord, Michael Makris, Beverley Hunt, Nichola Cooper, Quentin Hill, Catherine Bagot, Rachel Raymont, Gary Benson, and Nicola Curry of the U.K. Expert Group, as well as Drs. Clare Wykes, Pratima Chowdury, Roopen Arya, Rashid Kazmi, Oliver Lomas, Claire Davis, Smita Sinha, Sophie Lees, Gillian Lowe, and Saleem Shafeek.

REFERENCES

1. Price-Haywood EG, Burton J, Fort D, Seoane L. Hospitalization and mortality among Black patients and White patients with Covid-19. N Engl J Med 2020;382:2534-43.
2. Grasselli G, Greco M, Zanella A, et al. Risk factors associated with mortality among patients with COVID-19 in intensive care units in Lombardy, Italy. JAMA Intern Med 2020;180:1345-55.
3. Johns Hopkins Coronavirus Disease Resource Center. April 2021 (https://coronavirus.jhu.edu).
4. Hodgson SH, Mansatta K, Mallett G, Harris V, Emary KRW, Pollard AJ. What explains the similar and different clinical outcomes. Br J Haematol 2006;135:85-90.
5. Iida V, Jeffries MA, Sawalha AH. COVID-19: a review of therapeutic strategies and vaccine candidates. Clin Immunol 2021;222:108634.
6. Greinacher A, Thiele T, Warkentin TE, Weisser K, Kyrie PA, Eichinger S. Thrombotic thrombocytopenia after ChAdOx1 nCoV-19 vaccination. N Engl J Med. DOI: 10.1056/NEJMoa2104840.
7. Schultz NH, Servoll IH, Michelsen AE. Thrombosis and thrombocytopenia after ChAdOx1 nCoV-19 vaccination. N Engl J Med. DOI: 10.1056/NEJMoa2104882.
8. Government of the United Kingdom. UK regulator confirms that people continue to receive the COVID-19 vaccine AstraZeneca. March 18, 2021 (https://www.gov.uk/government/news/uk-regulator-confirms-that-people-should-continue-to-receive-the-covid-19-vaccine-astrazeneca).
9. European Medicines Agency. COVID-19 vaccine AstraZeneca: benefits still outweigh the risks despite possible link to rare blood clots with low blood platelets. March 18, 2021 (https://www.ema.europa.eu/en/news/covid-19-vaccine-astrazeneca-benefits-still-outweigh-risks-despite-possible-link-rare-blood-clots).
10. Liederman Z, Van Cott EM, Smock K, Meijer P, Selby R. Heparin-induced thrombocytopenia: an international assessment of the quality of laboratory testing. J Thromb Haemost 2019;17:2123-30.
11. Garritsen HS, Probst-Kepper M, LeGath N, et al. High sensitivity and specificity of a new functional flow cytometry assay for clinically significant heparin-induced thrombocytopenia antibodies. Int J Lab Hematol 2014;36:135-43.
12. Johnson M, Wagstaffe HR, Gilmour KC, et al. Evaluation of a novel multiplex assay for determining IgG levels and functional activity to SARS-CoV-2. J Clin Virol 2020;130:104572.
13. Bjørø E, Johnsen HS, Hansen JB, Braekkan SK. D-dimer at venous thrombosis diagnosis is associated with risk of recurrence. J Thromb Haemost 2017;15:917-24.
14. Paneesha S, Cheyne E, French K, Bacher S, Borg A, Rose P. High D-dimer levels at presentation in patients with venous thromboembolism is a marker of adverse clinical outcomes. Br J Haematol 2006;135:85-90.
15. Warkentin TE. Clinical picture of heparin-induced thrombocytopenia (HIT) and its differentiation from non-HIT thrombocytopenia. Thromb Haemost 2016;116:813-22.
16. Juhl D, Eichler P, Lubenow N, Strobel U, Wessel A, Greinacher A. Incidence and clinical significance of anti-PF4/heparin antibodies of the IgG, IgM, and IgA class in 755 consecutive patient samples referred for diagnostic testing for heparin-
induced thrombocytopenia. Eur J Haematol 2006;76:420-6.
17. Selleng S, Selleng K, Friesenke S, et al. Prevalence and clinical implications of anti-PF4/heparin antibodies in intensive care patients: a prospective observational study. J Thromb Thrombolysis 2015;39:60-7.
18. Hwang SR, Wang Y, Weil EL, Padmanabhan A, Warkenten TE, Pruthi RK. Cerebral venous sinus thrombosis associated with spontaneous heparin-induced thrombocytopenia syndrome after total knee arthroplasty. Platelets 2020 October 1 (Epub ahead of print).
19. Warkenten TE, Makris M, Jay RM, Kelton JG. A spontaneous prothrombotic disorder resembling heparin-induced thrombocytopenia. Am J Med 2008;121:632-6.
20. Rice L, Attisha WK, Drexler A, Francis JL. Delayed-onset heparin-induced thrombocytopenia. Ann Intern Med 2002;136:210-5.
21. Mohanty E, Nazir S, Sheppard J-Al, Forman DÀ, Warkenten TE. High-dose intravenous immunoglobulin to treat spontaneous heparin-induced thrombocytopenia syndrome. J Thromb Haemost 2019;17:841-4.
22. Warkenten TE. High-dose intravenous immunoglobulin for the treatment and prevention of heparin-induced thrombocytopenia: a review. Expert Rev Hematol 2019;12:685-98.
23. Irani M, Siegal E, Jella A, Aster R, Padmanabhan A. Use of intravenous immunoglobulin G to treat spontaneous heparin-induced thrombocytopenia. Transfusion 2019;59:931-4.
24. Voysey M, Clemens SÀ, Madhi SA, et al. Safety and efficacy of the ChAdOx1 nCoV-19 vaccine (AZD1222) against SARS-CoV-2: an interim analysis of four randomised controlled trials in Brazil, South Africa, and the UK. Lancet 2021;397:99-111.
Copyright © 2021 Massachusetts Medical Society.