Erroneous diagnosis of COVID-19 mRNA vaccine–associated acute myocarditis due to false-positive high-sensitive troponin I assay: a case report

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Received 31 May 2022; first decision 4 July 2022; accepted 18 November 2022; online publish-ahead-of-print 22 November 2022

Background
Coronavirus disease 2019 (COVID-19) mRNA vaccine–associated acute myocarditis has been well described, and the demonstration of elevated high-sensitivity cardiac troponin (hs-cTn) is crucial for its diagnosis. However, falsely elevated hs-cTn can occasionally occur, leading to incorrect diagnosis. Here, we report the case of a patient who was given an erroneous diagnosis of COVID-19 mRNA vaccine–associated acute myocarditis due to falsely elevated hs-cTn, likely from assay interference.

Case summary
A 29-year-old Chinese male presented with 3 months of chest pain, dyspnoea, and palpitations starting a few days after his second dose of mRNA-1273 (Moderna) vaccine. High-sensitivity cardiac troponin I was elevated at presentation, which rose further 4 h later. The provisional diagnosis was acute myocarditis after a computed tomography coronary angiogram showed normal coronaries. Cardiac magnetic resonance was also negative for myocardial inflammation. The hs-cTn I levels fluctuated but remained elevated on outpatient serial testing, despite no new symptoms or clinical events. A paired serum sample showed elevated hs-cTn I but normal hs-cTn T, confirming a diagnosis of false-positive hs-cTn I. Further investigations, including blood tests before and after a subsequent uneventful mRNA-1273 booster vaccination, were performed to investigate for assay interference.

Discussion
Widespread COVID-19 mRNA vaccination has resulted in an awareness of vaccine-related acute myocarditis and a more thorough evaluation of post-vaccination cardiac symptoms. Although false-positive hs-cTn rarely occurs, extensive testing will inevitably result in a significant number of patients with falsely elevated hs-cTn. Clinicians should exclude this possibility and consider using alternative hs-cTn assay when investigation results and clinical presentation appear discordant.

Keywords
Troponin • False positive • COVID-19 vaccine • Myocarditis • Case report

ESC Curriculum
3.2 Acute coronary syndrome • 6.5 Cardiomyopathy

Introduction
Coronavirus disease 2019 (COVID-19) mRNA vaccine–related acute myocarditis has been well described. Using the US Centers for Disease Control and Prevention (CDC) working case definition, a probable case can be established when there is one new or worsening cardiac symptom and elevated troponin. However, false-positive troponin can occur. Here, we report the case of a patient erroneously diagnosed with COVID-19 mRNA vaccine–associated acute myocarditis due to falsely elevated high-sensitivity cardiac troponin (hs-cTn) I.
Learning points

- High-sensitivity cardiac troponins (hs-cTns) are generally specific for the diagnosis of cardiomyocyte injury, but there are limitations in their use.
- The possibility of falsely raised hs-cTn levels should be entertained when clinical presentation and investigation results are discordant.
- Assay interference, such as from heterophile antibodies, is an important cause of false-positive hs-cTn and can be rapidly excluded by repeating the sample using an alternative hs-cTn assay.

Timeline

| Date | Significant event |
|------|-------------------|
| 13 April 2021 | Received first dose of mRNA-1273 (Moderna) coronavirus disease 2019 (COVID-19) vaccine |
| 11 May 2021 | Received second dose of mRNA-1273 COVID-19 vaccine |
| 14 May 2021 | The patient developed chest pain a few days after his second dose of mRNA-1273 COVID-19 vaccine |
| 4 August 2021 | The patient presented to the emergency department due to persistent symptoms. The initial hs-cTn I value was 150 ng/L, rising to 171 ng/L (reference range: 0–18) 4 h later |
| 5 August 2021 | Computed tomography coronary angiogram showed non-obstructive coronaries |
| 6 August 2021 | Transthoracic echocardiogram showed a normal ejection fraction with no wall motion abnormalities or pericardial effusion |
| 19 August 2021 | Outpatient cardiac magnetic resonance imaging was negative for acute myocarditis |
| 9 November 2021 | Hs-cTn I remained elevated. Further tests sent to workup for falsely elevated troponin returned negative |
| 12 November 2021 | Hs-cTn I remained elevated. Paired sample sent for hs-cTn T returned normal |
| 19 January 2022 | Booster mRNA-1273 COVID-19 vaccine was administered. Blood samples were taken just before vaccination |
| 26 January 2022 | Blood taken 1 week after booster vaccination showed stably elevated hs-cTn I despite a sharp rise in COVID anti-S levels. Paired blood sample sent for hs-cTn T returned normal |

Case summary

A 29-year-old Chinese male with no medical history developed fever and chest pain a few days after his second dose of mRNA-1273 (Moderna) vaccination. He received the first dose uneventfully. The chest discomfort persisted after the vaccination and was associated with intermittent palpitations and exertional breathlessness. He presented to our hospital 85 days post-vaccination for persistent symptoms. There were no infective symptoms since the initial post-vaccine fever. Vital signs and physical examination were unremarkable with normal heart sounds and no pericardial rub. A 12-lead electrocardiogram (ECG) showed sinus rhythm with early repolarization changes and no pericarditis features (Figure 1). High-sensitivity cardiac troponin I (Beckman Coulter Access) was 150 ng/L (reference range: 0–18), rising to 171 ng/L 4 h later. Serum D-dimer was normal. He was admitted for suspected acute coronary syndrome (ACS), but a computed tomography coronary angiogram performed later showed normal coronaries with no pulmonary embolism or aortic dissection (Figure 2). Transthoracic echocardiography (TTE) showed normal ejection fraction with no wall motion or valvular abnormality. Given the temporal relationship between symptoms and vaccination, a diagnosis of COVID-19 mRNA vaccine–associated acute myocarditis was made. Cardiac magnetic resonance (CMR) scan 2 weeks later showed no myocardial oedema or delayed enhancement (Figure 3). Native T1 and T2 mapping values were normal.

These discordant findings prompted a repeat hs-cTn I 34 days after admission, which decreased to 34 ng/L. As it was unusual to have persistently elevated troponin, hs-cTn I was repeated 63 days later, which increased to 133 ng/L despite no new interval symptoms. The possibility of false-positive hs-cTn I was considered. Further history was negative for pet ownership or exposure to mice or rabbits. A basic biochemical workup was normal (Table 1). A separate blood test using hs-cTn T assay (Roche Diagnostics) returned normal (24 ng/L, reference range <30 ng/L). Serial dilution of the sample did not suggest the presence of assay interference.

As the initial hs-cTn I was likely false positive, the patient received a booster mRNA-1273 vaccine ~6 months after. Pre-vaccination blood tests showed an elevated hs-cTn I of 43.5 ng/L and an anti-SARS-CoV-2 S titre of 1384 U/mL. Blood taken 7 days after an uneventful booster vaccination showed a stable hs-cTn I value of 39.2 ng/L with a normal paired hs-cTn T. The anti-SARS-CoV-2 S titre had rose to 18057 U/mL. The patient was referred to the respiratory medicine department for further evaluation. He eventually underwent a cardiopulmonary exercise test that revealed normal exercise tolerance and aerobic capacity with normal cardiorespiratory and metabolic response to exercise. His symptoms gradually improved and resolved subsequently.

Discussion

Troponin elevation above the 99th percentile upper reference limit, together with a rise and/or fall in titre, indicates acute myocardial injury. Troponin comprises three single-chain polypeptides, namely troponins C, I, and T. There is 1 hs-cTn T and at least 18 hs-cTn I assays available commercially. We report a patient with new cardiac symptoms post-COVID-19 mRNA vaccination and an initial abnormal hs-cTn I (150 ng/L) that increased to 171 ng/L in 4 h. According to guidelines, this patient was initially ‘ruled-in’ as having ACS based on the elevated initial hs-cTn I and significant delta on repeat testing. Paired creatine kinase (CK) and CK-myocardial band (MB) testing is no longer recommended with the use of hs-cTn assays and was hence not performed during the index admission. A diagnosis of probable myocarditis was entertained subsequently as our patient had typical demographics for post-vaccination myocarditis and presented with new cardiac symptoms fulfilling the US CDC case definition, which does not mandate CMR for diagnosis. Similarly, CMR was not performed for most of
the reported COVID-19 vaccination–associated myocarditis cases. However, there were several clinical aspects inconsistent with this diagnosis. Firstly, although the symptoms started within days of the second mRNA vaccine, our patient had persistent symptoms lasting for months, whereas most reported cases had symptom resolution within a week. However, late presentation of acute myocarditis has been reported at 3 months after the second dose of mRNA vaccine. Secondly, our patient had normal ECG and TTE. Although abnormal TTE is uncommon amongst reported COVID-19 vaccine–associated acute myocarditis cases, most patients had abnormal ECG changes such as ST-segment elevation. Our patient had diffuse ST-segment elevation on presentation consistent with early repolarization, which remained unchanged 3 months after discharge (Figure 1).

When evaluating elevated hs-cTn, ACS and other cardiac causes should be excluded before considering non-cardiac aetiologies such as renal failure, sepsis, anaemia, hypotension, hypoxia, and infiltrative disease. High-sensitivity cardiac troponin can also be elevated from skeletal myopathies such as polymyositis/dermatomyositis or acute rhabdomyolysis even in the absence of cardiac involvement or skeletal myopathies.

Causes of false-positive hs-cTn may be divided into pre-analytical and analytical factors. Important causes of pre-analytical errors include
A major cause of false-positive hs-cTn is assay interference, most frequently from heterophile antibodies induced by autoimmune conditions (e.g. raised rheumatoid factor), vaccinations, blood transfusion, exposure to pets or mice, and intake of dairy products. High-sensitivity cardiac troponin I immunoassays contain two distinct antibodies (‘capture’ and ‘label’), with the capture antibody binding to troponin I and the label antibody providing a signal to determine the concentration. Heterophile antibodies crosslink to either of these antibodies, leading to a false-positive signal.

We attempted to exclude assay interference by reviewing animal exposure in our patient’s history and screening for autoimmune disease, although a heterophilic antibody interference should cause a constant and not a dynamic ‘rise and fall’ troponin trend seen in acute myocardial

**Figure 2** Coronary computed tomography angiography images showing unobstructed left anterior descending (left), circumflex (middle), and right coronary (right) arteries, respectively.

**Figure 3** Cardiac magnetic resonance images show no delayed gadolinium enhancement (top row) and no significant myocardial oedema on T2-weighted images (bottom row).
We also considered the possibility of assay interference from antibody response to COVID-19 vaccination; however, hs-cTn I levels remained stable despite a marked increase in anti-SARS-CoV-2 S levels a week after the booster vaccine. The use of heterophilic blocking antibodies or demonstration of a non-linear troponin result on serial dilution can also detect interfering antibodies, although failure to exhibit non-linearity does not exclude analytical interference. Elevated alkaline phosphatase (ALP) also causes interference leading to incorrect diagnosis of myopericarditis has been reported even with normal ALP.

**Table 1** Summary of serum investigations

| Variable               | Reference range | Admission (4–7 August 2021) | 7 September 2021 | 9–10 November 2021 | 19 January 2022 | 26 January 2022 |
|------------------------|-----------------|----------------------------|------------------|-------------------|----------------|-----------------|
| Troponin I (ng/L)      | 0–18            | 150 (initial)              | 34               | 133               | 43.5           | 39.2            |
|                        |                 | 171 (4 h later)            |                  |                   |                |                 |
| Troponin T (ng/L)      | <30             | 24                         | <13              |                   |                |                 |
| Anti-SARS-CoV-2 S (U/mL)|                |                            |                   |                   |                |                 |
| White blood cell (x10^9/L) | 4.0–9.6       | 4.8                        | 1384             | 18 057            |                |                 |
| Haemoglobin (g/dL)     | 13.6–16.6       | 14.3                       | 1384             | 18 057            |                |                 |
| Platelets (x10^9/L)    | 150–360         | 267                        | 1384             | 18 057            |                |                 |
| Sodium (mmol/L)        | 135–145         | 140                        | 1384             | 18 057            |                |                 |
| Potassium (mmol/L)     | 3.5–4.5         | 3.6                        | 1384             | 18 057            |                |                 |
| Creatinine (µmol/L)    | 60–105          | 86                         | 1384             | 18 057            |                |                 |
| Albumin (g/L)          | 38–48           | 43                         | 44.1             | 43                |                |                 |
| Bilirubin (µmol/L)     | 5–30            | 24                         |                   |                   |                |                 |
| ALT (U/L)              | 10–55           | 11                         |                   |                   |                |                 |
| AST (U/L)              | 20–45           | 18                         |                   |                   |                |                 |
| ALP (U/L)              | 40–120          | 64                         |                   |                   |                |                 |
| GGT (U/L)              | 15–90           | 19                         |                   |                   |                |                 |
| LDH (U/L)              | 140–280         | 441                        |                   |                   | 303            | 313.5           |
| D-dimer (FEU) (µg/mL)  | <0.50           | 0.40                       |                   |                   |                |                 |
| ESR (mm/h)             | 1–10            |                            |                   |                   | 2              |                 |
| C-reactive protein (mg/L) | 0.0–5.0       | 0.4                        |                   |                   | 0.9            |                 |
| Rheumatoid factor (RU/mL) | 0–20         |                            |                   | <2                |                |                 |
| Myoglobin (µg/L)       | 28–72           | 33.4                       |                   |                   |                |                 |
| Creatine kinase (U/L)  | 50–350          | 176                        | 137.2             | 123.1             |                |                 |
| CK-MB-mass (µg/L)      | 0.6–6.3         | 2.7                        | 1.78              | 1.7               |                |                 |

ALT, alanine aminotransferase; ALP, alkaline phosphatase; AST, aspartate aminotransferase; ESR, erythrocyte sedimentation rate; GGT, gamma-glutamyl transferase; LDH, lactic acid dehydrogenase.

Conclusions

Approaches to investigating false-positive troponin have been published.\(^9,11,14\) The simplest method is to repeat troponin using a different assay. To our knowledge, this is the first case report of a patient who was given a wrong diagnosis of COVID-19 mRNA vaccine–associated acute myocarditis due to a dynamically changing but falsely elevated hs-cTn I. Although abnormal troponin should not be uniformly viewed as false positives,\(^2\) in reality, 3.7% of healthy individuals have false-positive hs-cTn I, with a third attributable to heterophile antibodies.\(^14\) Although a strategy of routine CK-MB ‘reflex testing’ in all elevated troponin cases to exclude false positives\(^15\) has been criticized,\(^11\) there will inadvertently be many cases of false-positive troponins as patients present with and undergo investigations for post-vaccination cardiac symptoms amidst global vaccination efforts. A wrong diagnosis causes anxiety, unnecessary additional investigations, and other deleterious implications such as exclusion from further mRNA vaccines. We hope that this case report serves to remind clinicians of this potential pitfall.

**Lead author biography**

Dr Vivian Goh Yi Suan graduated from the National University of Singapore, Yong Loo Lin School of Medicine, in 2019. She is currently in her second year of Internal Medicine residency training with a keen interest in cardiology.
Supplementary material

Supplementary material is available at European Heart Journal – Case Reports.

Slide sets: A fully edited slide set detailing this case and suitable for local presentation is available online as Supplementary data.

Consent: The authors confirm that written consent for submission and publication of this case report including images and associated text has been obtained from the patient in accordance with COPE guidance.

Conflict of interest: None declared.

Funding: None declared.

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