PEER REVIEW HISTORY

BMJ Open publishes all reviews undertaken for accepted manuscripts. Reviewers are asked to complete a checklist review form (**http://bmjopen.bmj.com/site/about/resources/checklist.pdf**) and are provided with free text boxes to elaborate on their assessment. These free text comments are reproduced below.

ARTICLE DETAILS

TITLE (PROVISIONAL)	Negative predictive value of the FebriDx host response point-of-care test in patients presenting to a single Australian Emergency Department with suspected COVID-19: An observational diagnostic accuracy study
AUTHORS	Buntine, Paul; Miller, Joseph; Pope, Alun; Guy, Stephen; Wong, Fang Qi (Alex); McDonald, Hannah; Ahmed, Mania; Teow, Kang Hui; Roney, Morgan; Mohammadi, Farzaneh; Aldridge, Emogene; Hackett, Liam; Jenner, Susanna; Davis, Belinda

VERSION 1 – REVIEW

REVIEWER	Pandey , Santosh Iowa State University, Electrical and Computer Engineering
REVIEW RETURNED	20-Jul-2022

GENERAL COMMENTS	Thank you for your submission. Overall the manuscript is well written and I have minor revisions. Below are some suggestions for the authors: 1) On page 11, line 31-35, it is suggested that the vaccinated patients had less host response proteins which reduced the sensitivity. Are there any articles or references that corroborate with this? 2) One page 11, line 45-48, the topic seems to change from "dual biomarkers" to "reduced sensitivity". I wonder if the last sentence belongs to the previous paragraph. 3) On page 11, line 51-56, this paragraph could be revisited. In my view, the Discussion should stay within the realm of low sensitivity of FebriDx and not suggest a potential strategy for a commercial product (unless there is value). 4) Some references could be added related to the commercialized technologies of antigen tests to help readers understand where FebriDx fits in the marketplace. One recent article is: Benda et al, "COVID-19 Testing and Diagnostics: A Review of Commercialized Technologies for Cost, Convenience and Quality of Tests", 2021 5) Is there evidence of reduced NPV in other rapid antigen tests within immunized populations as reported in the literature? If so, this would help the argument here.

REVIEWER	Brendish, Nathan University of Southampton, Clinical & Experimental Sciences, Faculty of Medicine
REVIEW RETURNED	24-Jul-2022

GENERAL COMMENTS	Thank you for asking me to review this manuscript on the diagnostic accuracy of the FebriDx point-of-care test in identifying COVID-19 vs PCR in an Australian ED.

	The manuscript is comprehensive, concise, and very well-written, and broadly conforms the STARD reporting guidelines. I have essentially nothing to suggest to improve the study manuscript itself as it stands. I believe it is the first study to report on FebriDx use since COVID-19 immunisations has become widespread and this is therefore novel. However, the overwhelming issue with this study is that is contains too few patients (<100) from which to draw definitive conclusions. Therefore, I recommend that publication in a good journal such as BMJopen is unwarranted. I encourage the authors to still attempt to publish their findings in another journal. If the study had continued to the originally-planned 300 patients I would have been fascinated to see the results. I have a few minor suggestions for improvement: - Fig 1 appears to be distorted in pdf format - suggest revising the image to a different file format to prevent this. - There are further FebriDx articles now available that you may wish to incorporate into your references as they are relevant to use of the FebriDx in the acute care of hospitalised patients: Mansbridge et al Infect Control Hosp Epi 2022 and Brendish et al Infect Dis and Therapy 2022. This reviewer is an author on these papers and so there may be a conflict of interest in this suggestion.

VERSION 1 – AUTHOR RESPONSE

Reviewer: 1
Dr. Santosh Pandey, Iowa State University
Comments to the Author:
Thank you for your submission. Overall the manuscript is well written and I have minor revisions. Below are some suggestions for the authors:

1) On page 11, line 31-35, it is suggested that the vaccinated patients had less host response proteins which reduced the sensitivity. Are there any articles or references that corroborate with this?

There is very little published literature on this topic. However, we were able to find a single pre-print article from 2021 suggesting reduced host response during post-vaccination COVID-19 infection. We have included this as a relevant citation in our subsequent revision (page 15, line 42).

2) On page 11, line 45-48, the topic seems to change from "dual biomarkers" to "reduced sensitivity". I wonder if the last sentence belongs to the previous paragraph.

We believe this can been clarified by replacing the current wording ("The reduced sensitivity found in our study…") with "The reduced sensitivity found in *the vaccinated participants* in our study…" (page 15, line 55 onwards).

3) On page 11, line 51-56, this paragraph could be revisited. In my view, the Discussion should stay within the realm of low sensitivity of FebriDx and not suggest a potential strategy for a commercial product (unless there is value).

We are thankful for this comment and have deleted the relevant paragraph (page 16, line 8 onwards).

4) Some references could be added related to the commercialized technologies of antigen tests to help readers understand where FebriDx fits in the marketplace. One recent article is: Benda et al, "COVID-19 Testing and Diagnostics: A Review of Commercialized Technologies for Cost, Convenience and Quality of Tests", 2021

We thank the reviewer for drawing our attention to this paper; however, the primary source from which the FebriDx data was obtained for this secondary paper was Clark et al. We have already referred extensively to this paper in our manuscript.

5) Is there evidence of reduced NPV in other rapid antigen tests within immunized populations as reported in the literature? If so, this would help the argument here.

This comment seems to relate to rapid antigen testing, but we did not assess this. In our study, a reduction in NPV was seen with FebriDx, which was a host response test. To our knowledge, ours is the only paper to examine this.

Reviewer: 2
Dr. Nathan Brendish, University of Southampton
Comments to the Author:
Thank you for asking me to review this manuscript on the diagnostic accuracy of the FebriDx point-of-care test in identifying COVID-19 vs PCR in an Australian ED.

The manuscript is comprehensive, concise, and very well-written, and broadly conforms the STARD reporting guidelines. I have essentially nothing to suggest to improve the study manuscript itself as it stands. I believe it is the first study to report on FebriDx use since COVID-19 immunisations has become widespread and this is therefore novel.

We thank the reviewer for these encouraging comments.

However, the overwhelming issue with this study is that is contains too few patients (<100) from which to draw definitive conclusions. Therefore, I recommend that publication in a good journal such as BMJopen is unwarranted. I encourage the authors to still attempt to publish their findings in another journal. If the study had continued to the originally-planned 300 patients I would have been fascinated to see the results.

We were also disappointed to terminate our study prior to reaching our projected sample size. However, we would like to stress that in hindsight, the fundamental assumption of our sampling calculations – that the sensitivity and specificity of a diagnostic test would be functions of the test and not the host – was flawed. This failed to account for possible changes in host response from vaccination and ongoing mutations to the COVID-19 virus. With this in mind, we feel that our findings are still valid, while acknowledging that a larger sample size would have assisted by reducing the confidence intervals that we presented. We have provided additional commentary around this point in our revised manuscript.

I have a few minor suggestions for improvement:

Fig 1 appears to be distorted in pdf format - suggest revising the image to a different file format to prevent this.

We appreciate this finding and have resubmitted the image as a .PNG file.

There are further FebriDx articles now available that you may wish to incorporate into your references as they are relevant to use of the FebriDx in the acute care of hospitalised patients: Mansbridge et al Infect Control Hosp Epi 2022 and Brendish et al Infect Dis and Therapy 2022. This reviewer is an author on these papers and so there may be a conflict of interest in this suggestion.

We thank the reviewer for highlighting these additional recent publications and for acknowledging the potential conflict of interest. We have now included citations to both additional references in our revised manuscript (page 10, line 41).

Reviewer: 1
Competing interests of Reviewer: none

Reviewer: 2
Competing interests of Reviewer: No competing interests beyond that I have published in this field.

VERSION 2 – REVIEW

REVIEWER	Pandey , Santosh Iowa State University, Electrical and Computer Engineering
REVIEW RETURNED	17-Oct-2022

GENERAL COMMENTS	Overall the manuscript is well written and balanced in the review and results. The comments have been addressed. I have some minor points to consider: 1) On page 9 first line, COVID-19 is misspelt. 2) Most of the limitations and issues with the study are covered nicely in your Discussion section. It may be worthwhile to see if there are other studies that have reported a lower NPV for this assay since your work.

REVIEWER	Brendish, Nathan University of Southampton, Clinical & Experimental Sciences, Faculty of Medicine
REVIEW RETURNED	13-Oct-2022

GENERAL COMMENTS	As I noted in my previous review, the manuscript is well written, and comprehensive yet concise. I have no changes to suggest to the manuscript.