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OPEN PEER REVIEW

Performance Comparison and Clinical Evaluation of Three Extraction-Free SARS-CoV-2 RT-PCR Assays

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1 Original submission

1 comment · 18 Nov 2024

Referee 1 Reviewer

MAJOR REVISION 18 Nov 2024

Dear Editor,
The authors of the following study, titled "Performance comparison and clinical evaluation of three extraction-free SARS-CoV-2 RT-PCR assays", evaluate three extraction-free RT-PCR kits for SARS-CoV-2 detection and compare their results with those obtained using a standard magnetic bead extraction kit. The manuscript uses different tables and graphs to evaluate the accuracy and reliability of the kits and analyzes the results using different statistical methods. In my view, the manuscript is of acceptable quality and can be accepted for publication after some corrections.
1- In the introduction section, please explain more about the extraction-free methods used during the COVID pandemic and provide references.
2- In the method section, why did the authors not use a column-based extraction method as a standard method? As everybody knows, column-based extraction is a very common method for nucleic acid extraction compared to the magnetic bead method and yields much purer nucleic acid than the magnetic bead method. It is recommended that you add the results of the column-based extraction method to your manuscript.
3- In the discussion section, please compare your results with those of other studies that use extraction-free methods. You can find many studies in this field conducted during the COVID pandemic on the web.

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Revision 1

AUTHORS' RESPONSE TO REVIEWERS

Authors' reply to reviewers

The next pages contain the authors' point-by-point reply to the comments of this revision round, as submitted with the revised manuscript. It is published as the authors wrote it.

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Title: Performance comparison and clinical evaluation of three extraction-free SARS-CoV-2 RT-PCR assays

Here are my replies to the reviewers' comments.

Replies to Reviewer 1

The authors in following study with the title "Performance comparison and clinical evaluation of three extraction-free SARS-CoV-2 RT-PCR assays" evaluate three extraction free RT-PCR kit for SARS-CoV-2 detection and compare their results with a standard magnetic bead extraction kit. The manuscript uses different tables and graphs to evaluate accuracy and reliability of kits and analyzed the results using different statistical methods. in my view the manuscript has acceptable quality and can be accepted for publication after some correction.

1. in introduction section please more explain about extraction free methods which used in COVID pandemic by reference.

Reply: We thank the reviewer's suggestion. In introduction section, we have added some descriptions of the nucleic acid extraction-free methods (see Page 3, lines 53-60 and Page 21, lines 427-444).

2. in method section, why the author did not use column base extraction method as a standard method? as everybody know, column base extraction method is very current and usual method for nucleic acid extraction compare to magnetic bead method and yielded much pure nucleic acid than magnetic bead method. It is recommended to add the column base extraction method result to your manuscript.

Reply: Although the column based extraction method has some advantages, it is relatively time-consuming and unsuitable for large-scale nucleic acid testing applications. Magnetic bead-based nucleic acid extraction technology is currently the most widely used automated nucleic acid extraction technology in clinical applications on the market, playing a crucial role in the prevention and control of COVID-19. Therefore, we compared the results with the magnetic bead-based nucleic acid extraction method.

3. in discussion section please compare your results with other studies which use extraction free methods, you can find a lot of studies in this field in period of COVID pandemic through the web.

Reply: We thank the reviewer's suggestion. In discussion section, We have added some descriptions that compare our results with those of other studies using extraction-free methods (see Page 12, lines 255-260).

Replies to Reviewer 2

1. In the abstract, the type of samples should be specified in the methods section.

Reply: We thank the reviewer's suggestion. In the revised manuscript, the type of samples has been clearly specified (nasopharyngeal swab) in the methods section (see Page 1, line 17).

2. Were the 4 kits used in this study homemade or commercial?

Reply: The four kits used in this study were commercial in period of COVID-19 pandemic.

3. If the kits are commercial, the name of the company and country of manufacture must be stated.

Reply: The name of the company and country of manufacture for the four kits have been stated (see Page 5, lines 89-91).

4. Why the reaction system of all 4 kits was amplified using different instruments?

Reply: Because each kit was designed based on a different technological platform, which required specific instruments for amplification. Table S1 (**Supplementary Material**) provides basic information about these kits.

5. The sequences of the RdRp, E, and N genes of the phage pseudovirus particles of SARS-CoV-2 obtained from BDS Company should be identical in sequence to the genes of the Omicron variants.

Reply: The Omicron variant has become the predominant strain worldwide since 2022. As the virus continues to mutate, the full genome sequence shows a slight decrease in identity with the original strain, but it remains over 98% identical.

Reply to Associate Editor 1

The manuscript entitled "Performance Comparison and Clinical Evaluation of three Extraction-free SARS-CoV-2 RT-PCR Assays" needs major revision.

Reply: We have studied all comments carefully and have made correction which we hope meet with approval.

2 Revision 1

0 comments

No public comments in this round.