

OPEN PEER REVIEW

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Peer Review Report for "Preparation and Characterization of a Chitosan-Based Nanoformulation Containing *Prangos acaulis* Extract and Evaluation of Its Effect on the Viability of HT-29 and MCF-7 Cancer Cells"

Author(s): Ahmed Dhahir Latif, Ahmed Jawad, Murtadha Hamzah Hashim Al-Isawi, Hyder Kareem Abbood, Hassan Al-Karagoly, Mahmoud Gharbavi

Review Timeline:

Submit Date:	14 Apr 2025
Revised Date:	1 Jul 2025
Accepted Date:	4 Jul 2025

Revision (0)

Here, you can see the **Reviewers**, **Associate Editors** and **EICs'** comments from the beginning to the end of the revision process.

Esmaeil Mohammadian : Reviewer

16 Apr 2025

Dear Editor

Have a nice day.

The author investigated the Fabrication and characterization of a chitosan-based nanoformulation of *Prangos acaulis* (DC.) Bornm extract with improved anti-cancer activities against HT-29 and MCF-7 cancer cell lines.

I would suggest reconsidering the manuscript for publication only after the following

major revision.

1. Typing errors need to be corrected.
2. The peak changes in Figure 1 are further interpreted.
3. What is the reason for low extraction efficiency?
4. Was the effect of DMSO investigated as a negative effect or a solvent effect?

REFEREE : Associate Editor

12 May 2025

Dear Authors,

Thank you for submitting your manuscript, "Fabrication and characterization of a chitosan-based nanoformulation of Prangos acaulis (DC.) Bornm extract with improved anti-cancer activities against HT-29 and MCF-7 cancer cell lines", to Jundishapur Journal of Natural Pharmaceutical Products. We have completed the review process, and I am pleased to inform you that your work has been assessed as suitable for publication pending minor revisions.

The reviewers have provided constructive feedback to enhance the clarity, presentation, and accuracy of your manuscript.

Please address all reviewer comments point-by-point in a response letter, highlight changes in the revised manuscript, and submit the revised manuscript and response.

We appreciate your commitment to improving this work and look forward to receiving your revision.

Kind Regards | Publisher Supervisor

REFEREE : EIC

12 May 2025

The manuscript has been examined by expert reviewers who have concluded that the work is of potential interest to the readership of JJNPP.

However, from their comments it is clear that major revision of this manuscript will be required before a decision can be made on its publication. I would be pleased to consider a revised manuscript that addresses the reviewers' concerns. Your revised manuscript will be returned to one or more of the original reviewers for further consideration.

OPEN PEER REVIEW

Revision (1)

Reply to Reviewers

Ideally, the reviewing process can significantly improve the submitted manuscripts by allowing the authors to take into account the advice of reviewers. Author(s) must reply to all reviewers' comments in a separate Word file, point by point. A "**Reply to Reviewers**" document is submitted along with revised manuscript during submission of revised files, summarizing the changes that the authors made in response to the reviewers' comments. The responses to reviewers' comments specifies how the authors addressed each comment the reviewers made.

You can read the authors' responses to the reviewers' comments in the next page.

24 June 2025

Dear editorial team and reviewers,

We appreciate your valuable comments regarding our manuscript submission entitled **“Fabrication and characterization of a chitosan-based nanoformulation of Prangos acaulis (DC.) Bornm extract with improved anti-cancer activities against HT-29 and MCF-7 cancer cell lines”**. We strongly believe that all of the positive comments and suggestions given by the reviewers have increased the scientific value of revised manuscript by many folds. In this context, we would like to thank the reviewers and editors for their time spent on reviewing our manuscript and their comments helping us improving the quality of current work. Here is the point by point responses to your valuable and constructive comments.

We are looking forward to having your decision at your earliest convenience.

Yours sincerely,

Reviewer(s)' Comments to Author:

Response to Reviewer 1:

Dear Editor

Have a nice day.

The author investigated the Fabrication and characterization of a chitosan-based nanoformulation of Prangos acaulis (DC.) Bornm extract with improved anti-cancer activities against HT-29 and MCF-7 cancer cell lines.

I would suggest reconsidering the manuscript for publication only after the following major revision.

1. Typing errors need to be corrected.

Response: Thank you for available comment, all grammatical errors within the article were corrected and highlighted in the text.

2. The peak changes in Figure 1 are further interpreted.

Response: Thank you for good comment, the FT-IR results were further interpreted as follows in manuscript:

The FT-IR spectra shown in the above image compare the methanolic extract and contrast it with the blank chitosan nanoparticle (Chitosan-NPs) and chitosan-extract nanoparticles (Chitosan-Extract-NPs). The high peaks shown on the Chitosan-NPs indicate that there are characteristics that define the chitosan itself because of the broad absorptions within the region of $3500\text{--}3000\text{ cm}^{-1}$ attributed to the O–H and N–H stretching vibrations and a peak around $1400\text{--}1480\text{ cm}^{-1}$ relating to the C–H bending arising from polymerization. The extract differentiates by having distinct, but especially with comparison within the regions associated with functional groups such as phenolic O–H, C=C, and C–O stretching. The broadness, shifts in peak positions, and changes in intensity found in the $3500\text{--}3000$ and the $1100\text{--}1500\text{ cm}^{-1}$ regions occur when considering the Chitosan-Extract-NPs. This is an indication that the phytochemicals from the extract associate with the chitosan matrix through hydrogen bonding and intermolecular forces. An indication that these amino groups of chitosan apparently got involved in the aforementioned interactions is decreased intensity of the NH_2 peak in chitosan-extract nanoparticles over the blank chitosan nanoparticles. New peaks appear or broaden within the $1100\text{--}1500\text{ cm}^{-1}$ region to indicate incorporation of extract functional groups such as P=O stretching and phenolic C–O stretching into the nanoparticle platform. In general, the FTIR spectra show that chitosan-extract nanoparticles were not only physical mixtures of chitosan and extract but also from effective chemical interaction between them. Such evidence includes the shifts and change in the so-called characteristic peaks proving successful encapsulation and establishment of a very stable nanoparticle system.

3. What is the reason for low extraction efficiency?

Response: Thank you for good comment, the reported extraction efficiency of 14% is calculated based on the amount of quercetin equivalents used as a standard for total flavonoid content, meaning this value reflects the relative yield of flavonoid compounds rather than the total extractable phytochemicals. This measurement inherently focuses on flavonoid-associated compounds that absorb at specific wavelengths and does not account for other bioactive

constituents present in the extract. Additionally, factors such as the complex plant matrix of *Prangos acaulis*, which includes cell wall components that can limit solvent penetration, and the selectivity of the 70% ethanol solvent for mid-polar flavonoids, contribute to this moderate extraction efficiency. Therefore, the 14% value is consistent with the method and standard used and should be interpreted as an estimate of flavonoid extraction relative to quercetin rather than an absolute measure of total extract yield.

4. Was the effect of DMSO investigated as a negative effect or a solvent effect?

Response: Thank you for good comment, since 1% DMSO was used as the solvent to dissolve the samples, and this concentration is known to be non-toxic to the cells, its potential effect was carefully controlled. To ensure that any observed effects were due to the samples themselves and not the solvent, 1% DMSO was also included in the control groups. This approach eliminates the possibility of solvent-induced toxicity influencing the viability results and prevents errors in data interpretation.

Best Regards,

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Revision (1)

Esmail Mohammadian : Reviewer

3 Jul 2025

Dear Editor

Have a nice day.

The addressed issues and concerns have been effectively resolved. Consequently, the reviewer concurs to proceed with the publication of this paper in its current state.

REFEREE : Associate Editor

4 Jul 2025

The Ms can be accepted in its current format

REFEREE : EIC

4 Jul 2025

I am pleased to inform you that your manuscript has been accepted for publication.