Round 1

Reviewer #1:

Scientific Quality: Grade C (Good)

Language Quality: Grade B (Minor language polishing)

Conclusion: Minor revision

Specific Comments to Authors: Authors have reported the molecular mechanism of the Gansong-Rhubarb drug pair（GRDP）for the treatment of diabetic kidney disease. The receptor for advanced glycation end products (RAGEs), which is mediated by the mitogen-activated protein kinase (MAPK), plays a critical role in the severity of chronic inflammatory disease diabetes mellitus. Authors have addressed role of AGE-RAGE pathway in DKD. Study is well designed and has translational potential applications. Few issues which need to be addressed are as below.

1. Molecular docking has provided good score. But Molecular dynamic simulations of those targets would validate stable binding kinetics of protein-target interactions. MDS needs to be performed for confirming stability and affinity.

First of all, thank you for your valuable comments. I think molecular docking is sufficient in our current research, but we will continue to learn about MDS.

2. Cell cycle kit and apoptosis links to be rectified and verified. Check other link details provided for correct hyperlink.

We reconfirmed the information on the cycle and apoptosis kits and made modifications, as detailed in manuscript 213 and 224.

3. Flow meter term should be changed to Flow Cytometer and details of instrument used for sample acquisition and flow analysis should be provided.

We have made additions and amendments to this section, as detailed in manuscript 215 and 225.

4. If Chemidoc instrument was used for acquiring chemiluminescence, then machine details need to be provided.

Thank you for your corrections to our manuscript. I have supplemented the specific information of this machine from the text, see line 236 for details.

5. For every technique, any positive and negative control used, need to be mentioned in methids section.

Thank you very much for your comments. I checked the Materials and Methods section and the groupings for each experiment are listed.

6. IL1b and TNF which are proinflammatory cytokines shown, they if validated in culture supernatants by ELISA , then it adds value to data.

We sincerely thank you for your valuable comments, but based on the current research, IL1b and TNF were not selected as the core proteins, so this experiment was not added this time, but it will be added in related studies in the future to enhance the value of the data.

7. Any experiments conducted to check expression of RAGE at genomic or protein levels may be reported.

We feel great thanks for your professional review work on our article. But the proteins selected in this experiment are based on network pharmacology and molecular docking results. We will add research on RAGE in future experiments.

8. Unit microlitre ul should be shown using symbol for micro and not use ‘u’.

We apologize for our carelessness. In our resubmitted manuscript, the wrong words have been corrected. Thanks for your correction.

9. Minor : Gansong word may be spell checked for typographical errors within manuscript.

Sincerely thank you for your comments, we have revised this to change Gansong to Nardostachyos Radix et Rhizoma.

10. ‘Graphd Prism 8.0 software’ should be rectified with correct software name.

We are very sorry for our careless mistake. We have corrected it to 'GraphPad Prism 8.0.2 '.

Reviewer #2:

Scientific Quality: Grade D (Fair)

Language Quality: Grade C (A great deal of language polishing)

Conclusion: Major revision

Specific Comments to Authors: 1.The purpose of the study is to discover the molecular mechanism of the Gansong-Rhubarb drug pair（GRDP） for the treatment of diabetic kidney disease. ‘‘Gansong’’ and “Rhubarb” are two different ways of naming, which is not standardized.

Thank you for your professional review, we have changed "Gansong" to "Nardostachyos Radix et Rhizoma" in the manuscript.

2. This study showed that GRDP regulated the expression of p-STAT3, BAX, CASPASE3, and CASPASE9 proteins of TCMK-1 cells, which did not match the mechanism shown in Figure 7.

We sincerely thank you for your valuable comments. We have modified and supplemented Figure 7, as shown in Figure 7 for details.

3. Several of the English language is not properly expressed, English writing skill needs to be improved. Check the accuracy of presentation and format in the whole manuscript is needed.

Thank you for your suggestion. We have done our best to revise and polish the article. We hope the revised manuscript will be accepted by you.

Reviewer #3:

Scientific Quality: Grade B (Very good)

Language Quality: Grade C (A great deal of language polishing)

Conclusion: Minor revision

Specific Comments to Authors: The aim and methodology of this manuscript are well-presented. The results are clear and correlate well with the methods used. However, I would suggest the following revisions to improve manuscript quality:

1- Authors are advised to improve integrity and cohesiveness of writing and also the flow of ideas.

We did our best to improve the manuscript and invited a native English speaker to help polish our article, hoping that the revised article will be accepted by you.

2- Authors should state what each abbreviation stands for in the manuscript.

For the revision of this section, I added the abbreviations mentioned in the article to the section before the reference.

3- Some paragraphs were left with no citation.

We sincerely appreciate your valuable comments. We have carefully reviewed the literature and supplemented and corrected the citations.

4- I would argue against the following sentences as these claims are controversial "Clinical medications are mostly Angiotensin Converting Enzyme Inhibitors (ACEI) and sulfonylureas to improve renal blood circulation. Such drugs can damage other functions of the human body, so we need a natural drug and compound with few side effects."

Thank you for your professional comments. We have made changes in the text, as detailed in lines 81-85 of the manuscript.

5- Authors should write the genus and species name in the correct style.

Thank you for your careful review. We have corrected the text.

6- Figure 7 is missing the required caption.

A description of Figure 7 has been added, and specific modifications are shown in Figure 7.

7- Authors should mention the version of Autodock and Pymol. Also, they should mention the PDB code for each target crystal.

We have made changes in the text and supplemented the PDB code for each target crystal, as detailed in lines 180 and 303-305.

Reviewer #4:

Scientific Quality: Grade B (Very good)

Language Quality: Grade C (A great deal of language polishing)

Conclusion: Major revision

Specific Comments to Authors:

1. Are there controversies in this field? What are the most recent and important achievements in the field? In my opinion, answers to these questions should be emphasized. Perhaps, in some cases, novelty of the recent achievements should be highlighted by indicating the year of publication in the text of the manuscript.

We are very grateful for your professional comments on our article. As you have asked, we have made amendments to the previous manuscript, specifically in the INTRODUCTION and DISCUSSION sections.

2. The results and discussion section is very weak and no emphasis is given on the discussion of the results like why certain effects are coming in to existence and what could be the possible reason behind them?

First of all, we sincerely thank you for your comments. We have revised and supplemented the results and discussion section. Under the stimulation of high glucose state, the level of AGE increases and the combination of RAGE leads to the aggravation of diabetes and its complications. Activation of downstream pathways increases oxidative stress, inflammation and apoptosis. Through this study, it was found that NRDP can reduce apoptosis in DKD state, thereby delaying the development of DKD disease course.

3. Conclusion: not properly written.

Thank you for your valuable comments. We have included the conclusion at the end of the article, as detailed in lines 420-430.

4. Results and conclusion: The section devoted to the explanation of the results suffers from the same problems revealed so far. Your storyline in the results section (and conclusion) is hard to follow. Moreover, the conclusions reached are really far from what one can infer from the empirical results.

We sincerely appreciate your valuable feedback, which we use to improve the quality of the manuscript. Specific revisions can be found on lines 350-419.

5. The discussion should be rather organized around arguments avoiding simply describing details without providing much meaning. A real discussion should also link the findings of the study to theory and/or literature.

We sincerely appreciate your comments. We have made every effort to modify the manuscript and hope that the corrections will be approved by you.

Round 2

1. Are there controversies in this field? What are the most recent and important achievements in the field? In my opinion, answers to these questions should be emphasized. Perhaps, in some cases, novelty of the recent achievements should be highlighted by indicating the year of publication in the text of the manuscript.

We appreciate your expert advice on our articles. Based on your suggestions, we have added relevant content to the discussion section. DKD is one of the serious complications of DM, and many experts and scholars have been committed to finding effective treatments over the years. According to the literature in the last three years, in addition to some commonly used methods to control the course of diabetic nephropathy (e.g., controlling blood glucose and controlling blood pressure, etc.), many new kinds of research have never stopped. With the in-depth study of pathogenesis, it brings new thinking to the clinical treatment of diabetic nephropathy. For example, combining ERA and SGLT2 inhibitors with combination therapy.

2. The results and discussion section is very weak and no emphasis is given on the discussion of the results like why certain effects are coming in to existence and what could be the possible reason behind them?

We have made changes and rewrites based on the suggestions you made. See the Discussion section for details. We have added to the Discussion section the experimental results, explaining the purpose of the experiment and verifying the pharmacological effects of NRDP on DKD.

3. Conclusion: not properly written.

Thank you for your careful reading, and we have made changes to the conclusion, as described in lines 427-436 of the manuscript.

4. Results and conclusion: The section devoted to the explanation of the results suffers from the same problems revealed so far. Your storyline in the results section (and conclusion) is hard to follow. Moreover, the conclusions reached are really far from what one can infer from the empirical results.

We sincerely thank you for your valuable feedback, based on which we have made changes to improve the quality of the manuscript. The role of the AGE-RAGE signaling pathway in the pathogenesis of diabetic nephropathy is known from reading the extensive literature. Based on the results of network pharmacology and molecular docking, we chose to study the effects of NRDP on STAT3 and apoptosis-related proteins after NRDP intervention. In this way, we demonstrated the therapeutic effect of NRDP on DKD and laid the foundation for future related studies.

5. The discussion should be rather organized around arguments avoiding simply describing details without providing much meaning. A real discussion should also link the findings of the study to theory and/or literature.

Thank you very much for your suggestion, we think it's a good one. In response, we have partially rewritten the discussion section, which can be found in the yellow section of the discussion. Other sections not highlighted in yellow have also been revised.

6. Spacing, punctuation marks, grammar, and spelling errors should be reviewed thoroughly. I found so many typos throughout the manuscript.

We apologize for our carelessness. The typos have been corrected in our resubmitted manuscript. Thank you for correcting them.

7. English is modest. Therefore, the authors need to improve their writing style. In addition, the whole manuscript needs to be checked by native English speakers.

Thank you sincerely for your suggestions. In response, we have done our best to revise and embellish the manuscript and have invited a native English speaker to check it. We hope that the revised manuscript will be acceptable to you.

Journal editor-in-chief review

An interesting work, and also did significant changes based on 4 reviewers' comments. However, the title needs to be changed before acceptation. Sincer the study was done based on database search and cell culture only, which does not fit into the current title: "Nardostachyos Radix et Rhizoma-Rhubarb in the treatment of diabetic kidney disease based on network pharmacology and experimental verification", should be "Potential application of Nardostachyos Radix et Rhizoma-Rhubarb for the treatment of diabetic kidney disease based on network pharmacology and cell culture experimental verification"

Response: Thank you for your suggestion, I have revised the title.

Dear Authors, Your manuscript: "Nardostachyos Radix et Rhizoma-rhubarb, Diabetic kidney disease, Molecular docking, Network pharmacology, and Experimental validation" after implementation of Reviewers suggestions and commenst looks good. I have no remarks.

Response: Thank you for your valuable advice.

The research in the field of chronic microvascular complications of diabetes is of great importance and finding the treatment options targeting specific molecular pathways should be a prerogative. I have revised the improved version of the Manuscript, all the reviewers' comments and authors replies which have substantially improved the Manuscript quality. I suggest its publication.

Response: Thank you for your valuable advice.