

## PEER REVIEW HISTORY

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### ARTICLE DETAILS

<b>TITLE (PROVISIONAL)</b>	Effect of treatments on skin microbiota in patients with atopic dermatitis: a protocol for a systematic review
<b>AUTHORS</b>	Guo, Yang; Dou, Xia; Jian, Xing-ling; Zhang, Kao-yuan; Zheng, Yingjie; Yu, Bo

### VERSION 1 – REVIEW

<b>REVIEWER</b>	Magnifico, Irene Department of Health and Medical Sciences “V. Tiberio” Università degli Studi del Molise
<b>REVIEW RETURNED</b>	09-Aug-2021

<b>GENERAL COMMENTS</b>	<p>The authors have conducted an interesting study protocol review on the effect of several treatments on skin microbiota among patients with Atopic Dermatitis.</p> <p>An extensive search of major electronic research databases (PubMed, EMBASE, Web of Science) assessing the diversity of the skin microbiota and treatment-related differential microbes differential microbes have been performed, microbiota functions, interactions, and immunological factors have been also taken in account.</p> <p>Nonetheless, some major revisions are needed prior to publication of this paper. Below are my comments to the authors.</p> <p>For a better review, the continuous numbering for the entire document is recommended.</p> <p>In the abstract section, authors chose to use future time. This choice is quite unusual, it is recommended to use the past time.</p> <p>INTRODUCTION, Lane 22 In AD skin dysbiosis, a central role is played by Staphylococcus aureus, but this is not the only microorganism with relative abundance variation. There are other bacterial and fungal species that change during AD dysbiosis. To better clarify this concept please take this reference in consideration Magnifico, I., Petronio Petronio, G., Venditti, N., Cutuli, M. A., Pietrangelo, L., Vergalito, F., ... &amp; Di Marco, R. (2020). Atopic dermatitis as a multifactorial skin disorder. Can the analysis of pathophysiological targets represent the winning therapeutic.</p> <p>METHODS- Search strategy Lane 17-18 It is not clear the reference time, the authors said “from their inception to April 2021” so this means all the articles published until April? Moreover the total number of paper found, how many included in the study, how many rejected is not reported. Should be reported.</p> <p>DISCUSSION Page.11 Line 36-45 References is needed</p> <p>From page 11 line 55 to Page.12 Line 16 in these two paragraphs the authors summarize the findings of this review. Although, “some baseline data can be presented, there should be no results or conclusions” , appropriate bibliographic references and findings</p>
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	<p>should be included for better clarify author findings. Perhaps it would be better to add a summary table that takes into account the bibliographies, the type of study performed and the main findings related to the study.</p> <p>Page 12 Lane 30 The authors conclude by stating that "the findings obtained may provide insights into future etiologic research and personalized therapy studies of AD" however, it is not clear what has been derived from the analyzed studies. For this reason, the Disussion paragraph should be revised and expanded upon.</p>
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<b>REVIEWER</b>	Zhu, Yuwei Vanderbilt University Medical Center, Biostatistics
<b>REVIEW RETURNED</b>	16-Sep-2021

<b>GENERAL COMMENTS</b>	<ol style="list-style-type: none"> <li>1. The inception of electronic databases including PubMed, EMBASE, and Web of Science etc. may be different, suggest to have a time frame i.e. 1991-2021 or 2000-2021.</li> <li>2. Page 10, MINORS, it lists the global score which ranges from 0 to 16 and authors may consider to categorize such score into similar categories that RoB2 or ROBINS-I has.</li> <li>3. Page 10, MINORS #12 an adequate statistical analyses. It's no clear how to make a judgment about it. Please provide more detail info to describe adequate statistical analyses.</li> <li>4. Please do a proofreading to make sure there are no grammar errors.</li> </ol>
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<b>REVIEWER</b>	Yang, Kun Xuanwu Hospital, Department of Evidence-based Medicine
<b>REVIEW RETURNED</b>	14-Nov-2021

<b>GENERAL COMMENTS</b>	<p>The protocol of systematic review will summarize studies evaluating the effect of treatments on skin microbiota among patients with AD. Authors need to pay attention to the following issues:</p> <ol style="list-style-type: none"> <li>1. Regarding the terms used in search strategy (in the Method section), atopic dermatitis related terms were "atopic dermatitis" and "atopic eczema". The term "eczematous dermatitis" may also be used. Therefore, the search strategy needs to be expanded to include "eczematous dermatitis".</li> <li>2. Please describe detailed methods used of high-throughput sequencing in the Eligibility criteria (in the Method section).</li> <li>3. Please consider deleting secondary outcomes on immunological factors, considering that they are not directly related to the skin microbial profile.</li> </ol>
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### VERSION 1 – AUTHOR RESPONSE

Reviewer: 1

Dr. Irene Magnifico, Department of Health and Medical Sciences "V. Tiberio" Università degli Studi del Molise

Comments to the Author:

The authors have conducted an interesting study protocol review on the effect of several treatments on skin microbiota among patients with Atopic Dermatitis.

An extensive search of major electronic research databases (PubMed, EMBASE, Web of Science) assessing the diversity of the skin microbiota and treatment-related differential microbes differential microbes have been performed, microbiota functions, interactions, and immunological factors have been also taken in account.

Nonetheless, some major revisions are needed prior to publication of this paper. Below are my comments to the authors.

A: We thank the reviewer for all the comments. Please see below our detailed responses to specific comments.

Q1: For a better review, the continuous numbering for the entire document is recommended. In the abstract section, authors chose to use future time. This choice is quite unusual, it is recommended to use the past time.

A: We thank the reviewer for the comment. For the formatting of BMJ Open, the subtitles of Introduction, Methods, Discussion, etc. are needed; the recent published papers in BMJ Open were also in such format. Thus, the style of continuous numbering for the entire document was not used in our paper. Regarding the future time used in Abstract, descriptions for protocols (our paper is a protocol for systematic review) usually use future time; also, the recent published protocols in BMJ Open were presented using future time. Accordingly, we chose to use future time, as well.

Q2: INTRODUCTION, Lane 22 In AD skin dysbiosis, a central role is played by *Staphylococcus aureus*, but this is not the only microorganism with relative abundance variation. There are other bacterial and fungal species that change during AD dysbiosis. To better clarify this concept please take this this reference in consideration Magnifico, I., Petronio Petronio, G., Venditti, N., Cutuli, M. A., Pietrangelo, L., Vergalito, F., ... & Di Marco, R. (2020). Atopic dermatitis as a multifactorial skin disorder. Can the analysis of pathophysiological targets represent the winning therapeutic.

A: We thank the reviewer for this insightful comment. Per this comment, we have further edited the Introduction section to clarify this concept that "In addition to *S. aureus*, the relative abundance of other species of the genus *Staphylococcus*, such as *S. haemolyticus*, also increased for AD cases.<sup>11</sup> Moreover, AD cases showed decreased relative abundance of multiple genera, including *Streptococcus* spp., *Propionibacterium* spp., *Acinetobacter* spp., etc.<sup>11</sup> In terms of fungal microbiota, which also play a critical role, a reduction in the relative abundance of *Malassezia* spp. and an increase of the *M. dermatis* etc. were observed for AD.<sup>11</sup>" (Page 5 Line 1-7). Also, the above-mentioned reference (Magnifico I, Petronio Petronio G, Venditti N, et al. Atopic Dermatitis as a Multifactorial Skin Disorder. Can the Analysis of Pathophysiological Targets Represent the Winning Therapeutic Strategy? *Pharmaceuticals*. 2020;13(11).) has been added, accordingly.

Q3: METHODS- Search strategy Lane 17-18 It is not clear the reference time, the authors said "from their inception to April 2021" so this means all the articles published until April? Moreover, the total number of paper found, how many included in the study, how many rejected is not reported. Should be reported.

A: We thank the reviewer for the comment. Similarly, Reviewer 2 also raised this point in the 1st question (Q1). Regarding the reference time, we believe that it could be better to set a time frame for searching these electronic databases; moreover, the microbiota studies began less than 20 years ago and the time frame "2000-2021" could be appropriate. Accordingly, we have revised the sentence that "The following electronic databases will be searched from November 2000 to November 2021: PubMed, EMBASE, Web of Science, ClinicalTrials.gov, and Chinese Clinical Trial Registry" (Page 6 Line 7-9). We also updated the time frames of search strategy in online supplemental file 2. In addition, we did preliminary literature search in November 2021 based on the updated search strategies; a total of 5192 records were found and 26 of them were included in the final analysis. Per the reviewer's suggestion, the numbers of papers found, included, excluded, etc. have been added in

the PRISMA flow diagram (the new Figure 1) and the manuscript that “The preliminary flow chart of study selection process is shown in the PRISMA flow diagram (Figure 1)” (Page 8 Line 13-14).

Q4: DISCUSSION Page.11 Line 36-45 References is needed

A: We thank the reviewer for the comment and we have added the references for these sentences “In recent years, multiple studies have demonstrated that skin microbiota dysbiosis plays a critical role in the development of AD, such as low microbial diversity, overabundant colonization of *S. aureus*, low abundance of other commensal bacteria, etc.<sup>9</sup> However, the impact of treatment on skin microbiota among patients with AD remained unclear and the results remained controversial across studies.<sup>12-15</sup>” (Page 12 Line 2-6).

Q5: From page 11 line 55 to Page.12 Line 16 in these two paragraphs the authors summarize the findings of this review. Although, “some baseline data can be presented, there should be no results or conclusions”, appropriate bibliographic references and findings should be included for better clarify author findings. Perhaps it would be better to add a summary table that takes into account the bibliographies, the type of study performed and the main findings related to the study.

A: We thank the reviewer for the comment. The above-mentioned paragraph summarized potential clinical implications of findings of the systematic review that the present protocol outlined. But it is not appropriate to present the findings of the systematic review as the present paper is the protocol of the systematic review. It is also mentioned in “Note from the Editors: Instructions for reviewers of study protocols”. Instead, the findings will be disseminated via another peer-reviewed publication or conference proceedings in the future. Additionally, we agree with the reviewer that it would be better to add a summary table. Per the reviewer’s suggestion, we have added the online supplemental file 5 and this file included four summary tables which will be presented in the systematic review. These tables were “I. Characteristics of the included studies”, “II. Major findings of the included studies on diversity (primary outcomes)”, “III. Major findings of the included studies on treatment-related differential microbes (primary outcomes)”, and “IV. Major findings of microbiota functions and microbial interactions (secondary outcomes)”. We also have added the related descriptions that “Major findings will be summarized as shown in online supplemental file 5.” (Page 11 Line 23-24) accordingly.

Q6: Page 12 Lane 30 The authors conclude by stating that “the findngs obtained may provide insights into future etiologic research and personalized therapy studies of AD” however, it is not clear what has been derived from the analyzed studies. For this reason, the Disussion paragraph should be revised and expanded upon.

A: We agree with the reviewer and have accordingly deleted the above-mentioned sentence. Per this comment, we have updated the paragraph of the Discussion section that “...this research protocol outlines a systematic review focusing on the effect of treatments on skin microbiota among patients with AD. The systematic review will provide a collective summary of impact of different types of treatment on skin microbiota for AD patients.” (Page 12 Line 26-28 and Page 13 Line 1).

Reviewer: 2

Dr. Yuwei Zhu, Vanderbilt University Medical Center

Comments to the Author:

Q1: The inception of electronic databases including PubMed, EMBASE, and Web of Science etc. may be different, suggest to have a time frame i.e. 1991-2021 or 2000-2021.

A: We agree with the reviewer. Similarly, Reviewer 1 also raised this point in the 3rd question (Q3). To respond to this comment and the Reviewer 1’s comment (Q3), the time frames for searching these electronic databases were modified to “2000-2021”. We have revised the sentence that “The following electronic databases will be searched from November 2000 to November 2021: PubMed, EMBASE, Web of Science, ClinicalTrials.gov, and Chinese Clinical Trial Registry” (Page 6 Line 7-9).

Additionally, we further updated the time frames of search strategy in online supplemental file 2.

Q2: Page 10, MINORS, it lists the global score which ranges from 0 to 16 and authors may consider to categorize such score into similar categories that RoB2 or ROBINS-I has.

A: We thank the reviewer for the comment and we have added the description of categories based on MINORS that “Scores of “13 ~ 16”, “7 ~ 12”, and “0 ~ 6” are classified as “low risk”, “moderate risk”, and “high risk” of bias” (Page 10 Line 28 and Page 11 Line 1).

Q3: Page 10, MINORS #12 an adequate statistical analyses. It's no clear how to make a judgment about it. Please provide more detail info to describe adequate statistical analyses.

A: We thank the reviewer for the comment. To respond to this comment, we have added the statistical analyses part and provided more details for statistical analyses (Page 11 Line 2-18).

Q4: Please do a proofreading to make sure there are no grammar errors.

A: We thank the reviewer for the comment. We have conducted very careful proofreading throughout the revised manuscript to avoid language issue (Page 3 Line 15, Page 8 Line 4, Page 12 Line 24, etc.). We hope the updated manuscript is now satisfactory and would like to make any further changes if necessary.

Reviewer: 3

Dr. Kun Yang, Xuanwu Hospital

Comments to the Author:

The protocol of systematic review will summarize studies evaluating the effect of treatments on skin microbiota among patients with AD. Authors need to pay attention to the following issues:

Q1: Regarding the terms used in search strategy (in the Method section), atopic dermatitis related terms were “atopic dermatitis” and “atopic eczema”. The term “eczematous dermatitis” may also be used. Therefore, the search strategy needs to be expanded to include “eczematous dermatitis”.

A: We thank the reviewer for raising this point. Per the reviewer’s suggestion, we have revised the search strategy in Method that “The search strategy is a combination of parameters “atopic dermatitis”, “atopic eczema”, “eczematous dermatitis”, ...” (Page 6 Line 12). Accordingly, full search strategy in online supplemental file 2 has been updated, as well.

Q2: Please describe detailed methods used of high-throughput sequencing in the Eligibility criteria (in the Method section).

A: We agree with the reviewer. To respond to this comment, we have revised the related sentences in the Method section, offering detailed methods that “...skin microbiota characteristics obtained using high-throughput sequencing, including 16S rRNA gene sequencing, metagenomic sequencing, and viral sequencing...” (Page 6 Line 23-24).

Q3: Please consider deleting secondary outcomes on immunological factors, considering that they are not directly related to the skin microbial profile.

A: We thank the reviewer for the comment and we have revised the secondary outcomes related sentences accordingly in Abstract (Page 2 Line 14-15), Methods (Page 7 Line 24 and Page 9 Line 17-18), and the data abstraction form (online supplemental file 3).

### VERSION 2 – REVIEW

<b>REVIEWER</b>	Zhu, Yuwei Vanderbilt University Medical Center, Biostatistics
<b>REVIEW RETURNED</b>	14-Dec-2021

<b>GENERAL COMMENTS</b>	Thanks for working on the revision and I don't have any additional questions.
<b>REVIEWER</b>	Yang, Kun Xuanwu Hospital, Department of Evidence-based Medicine
<b>REVIEW RETURNED</b>	05-Dec-2021
<b>GENERAL COMMENTS</b>	The authors of this manuscript have made additions and corrections in accordance with the suggestions I made last time, I have no other suggestions.