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Efficacy and safety of stem cells for the treatment of patients infected with 2019 novel coronavirus (COVID-19): a systematic review and meta-analysis protocol

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Efficacy and safety of stem cells for the treatment of patients infected with 2019 novel coronavirus (COVID-19): a systematic review and meta-analysis protocol

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ABSTRACT

Introduction To date, no specific anti-virus drugs or vaccines have been available to prevent or treat the 2019 novel coronavirus disease (COVID-19) pandemic. Stem cell therapy has been considered as one of the most promising therapeutic approaches that may reduce the high mortality in critical cases. This protocol is proposed for a systematic review and meta-analysis that aims to evaluate the efficacy and safety of stem cell therapy on patients with COVID-19.

Methods and analysis Ten databases will be searched from inception to 1 December 2020, including PubMed, EMBASE, Cochrane Library, CIHAHL, Web of Science, Chinese National Knowledge Infrastructure (CNKI), Chinese Scientific Journals Database (VIP), Wanfang database, China Biomedical Literature Database(CBM) and Chinese Biomedical Literature Service System (SinoMed). All published randomized controlled trials (RCTs), clinical controlled trials (CCTs), case-control and case series that meet the pre-specified eligibility criteria will be included. The primary outcomes include mortality, clinical recovery rate, duration of fever, progression rate from mild or moderate to severe, improvement of symptoms, biomarkers of laboratory examination, and changes in computed tomography. The secondary outcomes include the dosage of hormonotherapy, incidence and severity of adverse events, and quality of life. Study selection, data extraction and assessment of bias risk will be conducted by two reviewers independently. RevMan software (V.5.3.5) will be used to perform data synthesis. Subgroup and sensitivity analysis will be performed when necessary. The strength of evidence will be assessed by the GRADE system.

Ethics and dissemination Ethical approval is unnecessary as no individual patient or privacy data is collected. The results of this study will be disseminated in a peer-reviewed scientific journal and/or conference presentation.

Trial registration number PROSPERO 2020 CRD42020190079.

Keywords Stem cells; COVID-19; systematic review; meta-analysis; protocol

Strengths and limitations of this study

- This systematic review protocol follows the Preferred Reporting Items for Systematic Review and Meta-Analysis Protocols guidelines.

- This systematic review, to the best of our knowledge, will be the first to explore the efficacy and safety of stem cell therapy for the treatment of patients infected with 2019 novel coronavirus (COVID-19).
- The study will systematically review qualitative data from various medical databases for an in-depth interpretation of the efficacy and safety of stem cell therapy on patients with COVID-19.
- The potential for low and inconsistent quality in the reporting of process evaluations, the publication bias, and the methodological quality of the grey literature found may be the limitations of the study.

INTRODUCTION

Description of the condition

Coronavirus disease 2019 (COVID-19), an infectious disease caused by a novel severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2, formerly termed as 2019-nCoV), has been sweeping the world.[1] However, to date, no efficient vaccines and specific antiviral medications are available to halt the pandemic. As of 24 June 2020, 9,110,186 confirmed cases of COVID-19 have been documented globally in 216 countries, areas or territories with 473,061 deaths.[2] Notably, approximately 15% of the infected patients develops severe pneumonia and 5% progresses to acute respiratory distress syndrome (ARDS), septic shock and multiorgan failure eventually.[3 4] High mortality rate was observed in critically ill patients and has prompted an urgent need for treatments that can address the critical cases and prevent fatal outcomes.[5 6]

Description of the intervention

Currently, mesenchymal stem cells (MSCs)-based treatment has been proposed as a promising therapeutic approach for patients with COVID-19. [7] MSCs are multipotent cells that can be obtained from various tissues including preferably bone marrow, adipose tissue, placenta, umbilical cord, and dental pulp. The safety and effectiveness of MSCs therapy have been well documented in several clinical studies including ARDS, bronchopulmonary dysplasia, and cardiovascular diseases. [8-12] In a recent case study, a 65-year-old critically ill ventilator ridden COVID-19 patient was treated with allogeneic human umbilical cord MSCs (three infusions of 5×10^7 cells at an interval of three days) and the patient was off the ventilator and able to walk after the second infusion. No obvious side effects were observed. [13] In another study, seven patients with COVID-19 (one critically severe, four severe, and two mild-to-moderate) received a

single intravenous administration of MSCs at a dose of 1 × 10^6 cells/kg body weight, all patients had significant improvements in clinical symptoms two days after the transplantation, one severe and two mild patients were discharged from the hospital by day 10, and a 14-day follow-up showed no adverse effects. [14] These completed clinical trials provided limited but promising evidence that the use of MSCs therapy might be efficient in the treatment of COVID-19. The US Food and Drug Administration authorized compassionate use of MSCs therapy in patients with an extremely dismal prognosis. Recently, 45 clinical trials of MSCs as a new treatment for COVID-19 have been registered on ChiCTR (http://www.chictr.org.cn) and clinicaltrials.gov (https://clinicaltrials.gov). Several trials are underway that may provide more evidence to evaluate the safety and effectiveness of MSCs therapy for COVID-19.

How the intervention might work

A growing body of research has interpreted the potential mechanism underlying the therapeutic effect of MSCs on COVID-19. SARS-CoV-2 cell entry depends on the angiotensin-converting enzyme-2 (ACE2) and the transmembrane serine protease 2 (TMPRSS2), gene expression profiling exhibited that transplanted MSCs were ACE2- and TMPRSS2- and had a natural immunity to the COVID-19 infection.[14 15] Virus-induced cytokine storm is considered as the hallmark of SARS-Cov-2 pathogenesis and has been reported to induce ARDS and multi-organ failure, leading to death in COVID-19 patients. [16] To prevent or attenuate the cytokine storm is pivotal to halt the pandemic. Compelling studies have demonstrated that MSCs have potent and broad immunomodulatory and anti-inflammatory effects. Such processes include the regulation of T cell function, proliferation and differentiation of B cells, influence of innate immune cells (such as macrophages and dendritic cells), decrease of pro-inflammatory cytokines (such as IL-1, IL-6, IFN, and TNF-α), increase of anti-inflammatory cytokines (such as IL-4, IL-5, and IL-10) and peripheral lymphocytes, and decline of over-activated cytokine-secreting immune cells (such as $CXCR3^+$ $CD4^+$ T cells, $CXCR3^+$ $CD8^+$ T cells, and $CXCR3^+$ NK cells).[14 17-23] Besides, MSCs inhibit bacterial growth, enhance the restoration of injured alveolar epithelial cells, improve pulmonary microenvironment, alleviate pulmonary fibrosis, and enhance pulmonary function.[24 25] Kyoto Encyclopedia of Genes and Genomes analysis implied that MSCs were involved in antiviral pathways.[14] Hence, MSCs therapy may improve the outcome of COVID-2019 patients through immunomodulation, regulating the inflammatory response, and promoting tissue repair.

Why it is important to perform this review

The pandemic COVID-19 caused by SARS-CoV-2 has been sweeping the world. MSCs therapy has been proposed as a safe, effective, and promising approach to treat COVID-19, especially for severe or critical

cases. It is important to perform a systematic review and meta-analysis to evaluate its efficacy and safety. In this systematic review, all potential RCTs, CCTs, case-control, and case series regarding MSCs for the treatment of COVID-19 will be fully considered and synthesized without language or publication restrictions. The findings of this study may yield helpful evidence for the patients, clinicians, investigators, and policymakers concerned about the efficacy and safety of MSCs therapy on COVID-19.

OBJECTIVES

This systematic review and meta-analysis aim to assess the evidence for the effectiveness and safety of MSCs therapy for COVID-19.

METHODS AND ANALYSIS

Criteria for including studies for this review

Types of studies

Randomized controlled trials (RCTs), clinical controlled trials (CCTs), case-control, and case series of stem cells treatment for COVID-19 will be included. Animal-based research and literature review will be excluded.

Types of participants

Patients diagnosed with COVID-19 using any recognized diagnostic criteria will be included regardless of the age, gender, and source of cases and the duration and severity of the disease. Patients infected with adenovirus, rhinovirus, human metapneumovirus, etc. will be excluded.

Types of interventions

The intervention group will be treated by stem cells and western conventional medicine. There will be no restriction regarding conventional western medical regimen (such as supportive treatment, IFN-α, lopinavir, or ritonavir).

Types of comparator(s)/control

The control group will be treated with the same conventional western medical regimen as the intervention group in the same original study. No restrictions are imposed regarding conventional western medicine treatment regimen.

Types of outcome measures

Primary outcomes

Primary outcome measures include mortality, clinical recovery rate, duration of fever, progression rate from mild or moderate to severe, improvement of symptoms, biomarkers of laboratory examination and changes in computed tomography.

Secondary outcomes

Secondary outcomes include the dosage of hormonotherapy, incidence and severity of adverse events, and quality of life.

Search methods for identification of studies

Electronics searches

The following ten databases will be searched from the inception to 1 December 2020: PubMed, EMBASE, Cochrane Library, CIHAHL, Web of Science, Chinese National Knowledge Infrastructure(CNKI), Chinese Scientific Journals Database (VIP), Wanfang database, China Biomedical Literature Database(CBM) and Chinese Biomedical Literature Service System (SinoMed). Two reviewers will search the literature independently. Any inconsistency will be solved by a third reviewer. Manual search will be performed for relevant studies found in the reference lists of included studies.

The electronic search will be conducted using a combination of following terms: novel coronavirus, Severe Acute Respiratory Syndrome Coronavirus 2, SARS-CoV-2, COVID-19, COVID19, 2019-nCoV, mesenchymal stem cells, mesenchymal stromal cells, MSCs, stem cells, SCs, randomized controlled trial, controlled clinical trial, randomized, randomly, trials, case-control, case series, CCT, and RCT. The search strategy for PubMed is presented in Table 1 and will be modified in other databases.

Searching other resources

Studies from Clinical Trials.gov (http://www.clinical trails.gov), Chinese Clinical Trial Registry (http://www.chictr. org/cn/), and WHO International Clinical Trial Registry Platform (https://www.who.int/ictrp/en/) will also be searched. The reference lists of the retrieved articles will be manually reviewed for further additional trials. The corresponding author will be contacted for incomplete data.

Data collection and analysis

Selection of studies

Two reviewers will perform screening, study selection, and data extraction independently. The literature obtained will be imported into EndnoteX9 to screen the title and abstract, the duplications and studies failing to meet the pre-specified inclusion criteria will be excluded. After reading the full text of the remaining studies, the final included studies will be determined. The corresponding author from any original study will be contacted when the full text is unavailable**.** Any disagreements will be arbitrated by a third reviewer. The entire process of study selection is presented in a PRISMA flow chart (figure 1).

Data extraction and management

The following data will be extracted by two reviewers independently from eligible studies and input into a pre-specified data acquisition form: reference ID, author information, year of publication, study type, study design, setting of study, sample size, participant characteristics (age, gender, duration and severity of illness, laboratory test, CT scan, etc.), stem cell intervention group and control group (details of randomization, blinding, allocation, intervention approach and duration), and primary and secondary outcomes at all reported time points. Inconsistency between two reviewers will be solved by a third reviewer. All data will be cross-checked and transferred to RevMan software (V.5.3).

Assessment of risk of bias

Two reviewers will assess the risk of publication bias for every included study using the Cochrane Risk of Bias Assessment Tool independently in terms of eight domains, namely randomization sequence generation, randomization allocation concealment, blinding of participants, blinding of personnel, blinding of outcome assessors, incomplete outcome data, selective reporting bias, and other bias. Each domain will be graded as high, unclear, or low risk of bias.[24] Corresponding authors will be contacted for unclear domains. Inconsistency will be solved by consultation with a third reviewer.

Measures of treatment effect

Efficacy data will be synthesized and statistically analyzed by two reviewers independently using RevMan 5.3.. A risk ratio or odd ration with 95% CIs will be adopted for dichotomous data, whereas a mean difference (MD) or standard mean difference (SMD) with 95% CIs will be utilized for continuous data. SMD will be employed if different assessment tools are used.

Dealing with missing data

If the required data is unclear or missing, reviewers will contact the corresponding author of the original study by E-mail or telephone. If data is still unattainable, the study concerned will be excluded from the analysis. A sensitivity analysis will be performed to address the potential impact of missing data.

Assessment of heterogeneity

Statistical heterogeneity will be investigated using χ^2 test and I^2 statistic. The fixed-effect model will be applied when heterogeneity is low (I^2<50%) and the random-effects model will be used for moderate heterogeneity (50% < I^2 <75%). When heterogeneity is considerably high, meta-analysis will not be performed.

Assessment of reporting biases

Funnel plots will be performed to assess potential reporting bias when more than 10 studies are included. In addition, the Egger regression and Begg correlation test will be conducted to identify the funnel plot asymmetry.

Data synthesis

In line with the Cochrane guideline, the fixed-effects model will be utilized for the pooled data if heterogeneity is deemed low and the random-effect model will be employed if heterogeneity is deemed moderate.[26] Subgroup analysis or meta-regression will be performed to assess the potential sources with reasonable explanations if heterogeneity is considerably high. The statistical significance is defined as p<0.05. If the meta-analysis is not feasible, a narrative description of the results will be provided.

Subgroup analysis and investigation of heterogeneity

If feasible, subgroup analyses will be performed in terms of the severity of included patients, duration of disease, routes of administration, dosage, and types of stem cells. Subgroup analyses will be conducted to interpret the heterogeneity.

Sensitivity analysis

If feasible, sensitivity analysis will be conducted to evaluate the robustness of the pooled effects of the included studies given the impact of such variables as sample size, methodological quality, missing data, or high risk of bias.

Summary of evidence

The Cochrane Collaboration Network GRADE (The Grading of Recommendations Assessment Development and Evaluation) will be utilized to grade the quality of evidence as very low, low, moderate or high.[27 28] The quality of evidence of a specific study will be assessed according to the risk of bias, imprecision, inconsistency, indirectness, publication bias, effect size or dose-response relation. The

findings will be presented with A Summary of Finding table. Any discrepancy will be arbitrated by discussion or a third reviewer.

Patient and public involvement

No patient involved.

Ethics and dissemination

Ethical approval is not necessary as no individual patient or privacy data will be collected. The results of this study will be disseminated as a publication in a peer-review journal or conference presentation.

DISCUSSION

This meta-analysis will provide a relatively convincing conclusion of whether MSCs therapy is effective and safe for treating patients with COVID-19. Conclusions drawn from this review may benefit patients, clinicians, investigators, and policymakers. The process of conducting this review will be divided into identification, study inclusion, data extraction, and data synthesis. If amendments to this protocol are necessary, the date of each amendment with a statement of the changes and the corresponding reasons will be provided.

Author Contributions Y-H C, Q Z, T-E Z and S-Q T conceived and designed the study. The protocol was drafted by Y-H C, Q Z, X-L L and S-Q T and revised by Y-H C, W P and T-E Z. Y-H C, W P, S-Q T and T-E Z designed the search strategies. X-L L and D L will perform searching, data curation and assessment independently. Q Z and Y-Y Y will analyze and interpret the data. S-Q T, T-E Z, X-L L and Y-H C will arbitrate disagreement if there is anything during the review. All authors have read and approved the publication of the protocol.

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Competing interests None declared.

Patient consent for publication Not required.

Ethics approval Ethics approval is not necessary because no individual patient data and privacy data will be collected.

Provenance and peer review Not commissioned; externally peer reviewed.

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REFERENCES

1. Lu R, Zhao X, Li J, et al. Genomic characterisation and epidemiology of 2019 novel coronavirus: implications for virus origins and receptor binding. *Lancet*. 2020;395(10224):565-574.
2. World Health Organization. Rolling updates on coronavirus disease (COVID-19). Retrieval June 24, 2020 on https://www.who.int/emergencies/diseases/novel-coronavirus-2019/events-as-they-happen.
3. Huang C, Wang Y, Li X, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet*. 2020;395(10223):497-506.
4. Xu Z, Shi L, Wang Y, et al. Pathological findings of COVID-19 associated with acute respiratory distress syndrome. *Lancet Respir Med*. 2020;8(4):420-422.
5. Bhatraju PK, Ghassemieh BJ, Nichols M, et al. Covid-19 in Critically Ill Patients in the Seattle Region - Case Series. *N Engl J Med*. 2020;382(21):2012-2022.
6. Zumla A, Hui DS, Azhar EI, Memish ZA, Maeurer M. Reducing mortality from 2019-nCoV: host-directed therapies should be an option. *Lancet*. 2020;395(10224):e35-e36.
7. Golchin A, Farahany TZ. Biological Products: Cellular Therapy and FDA Approved Products. *Stem Cell Rev Rep*. 2019;15(2):166-175.
8. Golchin A, Farahany TZ, Khojasteh A, Soleimanifar F, Ardeshirylajimi A. The Clinical Trials of Mesenchymal Stem Cell Therapy in Skin Diseases: An Update and Concise Review. *Curr Stem Cell Res Ther*. 2019;14(1):22-33.
9. Matthay MA, Calfee CS, Zhuo H, et al. Treatment with allogeneic mesenchymal stromal cells for moderate to severe acute respiratory distress syndrome (START study): a randomised phase 2a safety trial. *Lancet Respir Med*. 2019;7(2):154-162.
10. Chen J, Hu C, Chen L, et al. Clinical study of mesenchymal stem cell treating acute respiratory distress syndrome induced by epidemic Influenza A (H7N9) infection, a hint for COVID-19 treatment [published online ahead of print, 2020 Feb 28]. *Engineering (Beijing)*. 2020;10.1016/j.eng.2020.02.006. doi:10.1016/j.eng.2020.02.006
11. Namba F. Mesenchymal stem cells for the prevention of bronchopulmonary dysplasia. *Pediatr Int*. 2019;61(10):945-950.
12. Suvakov S, Richards C, Nikolic V, et al. Emerging Therapeutic Potential of Mesenchymal Stem/Stromal Cells in Preeclampsia. *Curr Hypertens Rep*. 2020;22(5):37.
13. Bing Liang, Junhui Chen, Tao Li, et al. Clinical remission of a critically ill COVID-19 patient treated by human umbilical cord. chinaXiv, 2020.
14. Leng Z, Zhu R, Hou W, et al. Transplantation of ACE2$^-$ Mesenchymal Stem Cells Improves the Outcome of Patients with COVID-19 Pneumonia. *Aging Dis*. 2020;11(2):216-228.
15. Hoffmann M, Kleine-Weber H, Schroeder S, et al. SARS-CoV-2 Cell Entry Depends on ACE2 and TMPRSS2 and Is Blocked by a Clinically Proven Protease Inhibitor. *Cell*. 2020;181(2):271-280.e8.
16. Soy M, Keser G, Atagündüz P, Tabak F, et al. Cytokine storm in COVID-19: pathogenesis and overview of anti-inflammatory agents used in treatment. *Clin Rheumatol*. 2020;39(7):2085-2094. doi:10.1007/s10067-020-05190-5

17. Chen QH, Wu F, Liu L, et al. Mesenchymal stem cells regulate the Th17/Treg cell balance partly through hepatocyte growth factor in vitro. *Stem Cell Res Ther*. 2020;11(1):91.
18. Luz-Crawford P, Hernandez J, Djouad F, et al. Mesenchymal stem cell repression of Th17 cells is triggered by mitochondrial transfer. *Stem Cell Res Ther*. 2019;10(1):232.
19. Cho KA, Lee JK, Kim YH, et al. Mesenchymal stem cells ameliorate B-cell-mediated immune responses and increase IL-10-expressing regulatory B cells in an EBI3-dependent manner. *Cell Mol Immunol*. 2017;14(11):895-908.
20. de Witte SFH, Luk F, Sierra Parraga JM, et al. Immunomodulation By Therapeutic Mesenchymal Stromal Cells (MSC) Is Triggered Through Phagocytosis of MSC By Monocytic Cells. *Stem Cells*. 2018;36(4):602-615.
21. Reis M, Mavin E, Nicholson L, et al. Mesenchymal Stromal Cell-Derived Extracellular Vesicles Attenuate Dendritic Cell Maturation and Function. *Front Immunol*. 2018;9:2538.
22. Shi Y, Wang Y, Li Q, et al. Immunoregulatory mechanisms of mesenchymal stem and stromal cells in inflammatory diseases. *Nat Rev Nephrol*. 2018;14(8):493-507.
23. Uccelli A, de Rosbo NK. The immunomodulatory function of mesenchymal stem cells: mode of action and pathways. *Ann N Y Acad Sci*. 2015;1351:114-126.
24. Krasnodembskaya A, Song Y, Fang X, et al. Antibacterial effect of human mesenchymal stem cells is mediated in part from secretion of the antimicrobial peptide LL-37. *Stem Cells*. 2010;28(12):2229-2238.
25. Rajarshi K, Chatterjee A, Ray S. Combating COVID-19 with Mesenchymal Stem Cell therapy [published online ahead of print, 2020 May 14]. *Biotechnol Rep (Amst)*. 2020;26:e00467.
26. Chandler J, Higgins JPT, Deeks JJ, et al. Cochrane handbook for systematic reviews of interventions version 5.2.0: Cochrane Community, 2017
27. Guyatt GH, Oxman AD, Vist GE, et al. GRADE: an emerging consensus on rating quality of evidence and strength of recommendations. *BMJ*. 2008;336(7650):924-926. doi:10.1136/bmj.39489.470347.AD
28. Guyatt GH, Oxman AD, Schünemann HJ, et al. GRADE guidelines: a new series of articles in the Journal of Clinical Epidemiology. *J Clin Epidemiol*. 2011;64(4):380-382. doi:10.1016/j.jclinepi.2010.09.011

Figure Legends

Figure 1. Flowchart of study selection

Table 1. Search strategy for the PubMed

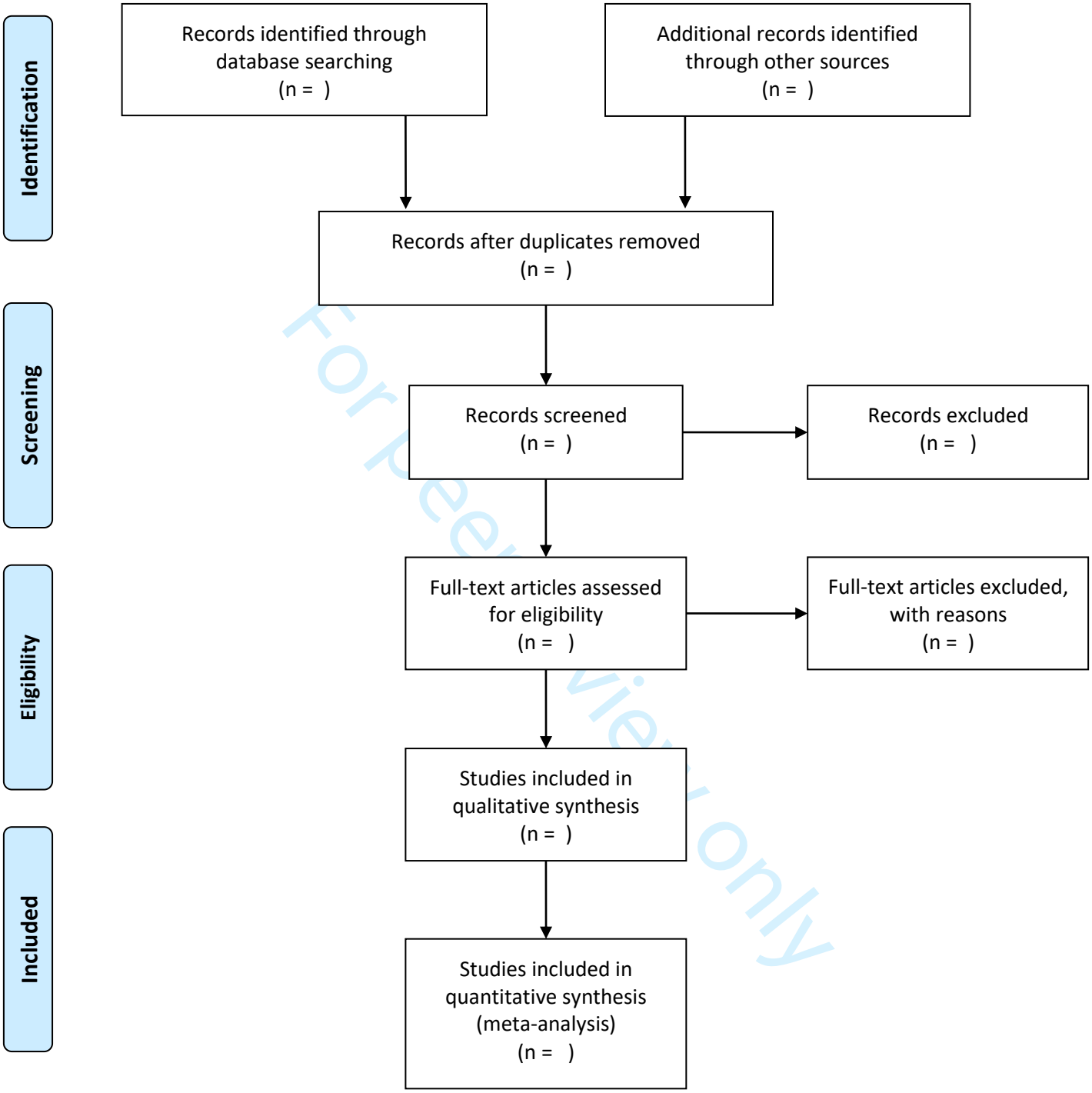

Table 1 Search strategy for the PubMed

No.	Search terms
#1	Novel coronavirus
#2	Severe Acute Respiratory Syndrome Coronavirus 2
#3	SARS-CoV-2
#4	COVID-19
#5	COVID19
#6	2019-nCoV
#7	OR/#1-#6
#8	Stem cells
#9	SC
#10	Mesenchymal stem cells
#11	mesenchymal stromal cells
#12	MSCs
#13	OR/#8-#12
#14	7# AND 13#
#15	Randomized controlled trial
#16	Controlled clinical trial
#17	Randomized*
#18	Randomly*
#19	Trails
#20	Case-control
#21	Case series
#22	CCT
#23	RCT
#24	OR/#14-#23
#25	#7AND#14AND#24

*Represent one or more characters of all characters.

PRISMA-P (Preferred Reporting Items for Systematic review and Meta-Analysis Protocols) 2015 checklist: recommended items to address in a systematic review protocol*

Section and topic	Item No	Checklist item	Reported on Page #
ADMINISTRATIVE INFORMATION			
Title:			
Identification	1a	Identify the report as a protocol of a systematic review	P1
Update	1b	If the protocol is for an update of a previous systematic review, identify as such	N/A
Registration	2	If registered, provide the name of the registry (such as PROSPERO) and registration number PROSPERO CRD42020190079	P2
Authors:			
Contact	3a	Provide name, institutional affiliation, e-mail address of all protocol authors; provide physical mailing address of corresponding author	P1
Contributions	3b	Describe contributions of protocol authors and identify the guarantor of the review	P9
Amendments	4	If the protocol represents an amendment of a previously completed or published protocol, identify as such and list changes; otherwise, state plan for documenting important protocol amendments	N/A
Support:			
Sources	5a	Indicate sources of financial or other support for the review	P9
Sponsor	5b	Provide name for the review funder and/or sponsor	N/A
Role of sponsor or funder	5c	Describe roles of funder(s), sponsor(s), and/or institution(s), if any, in developing the protocol	N/A
INTRODUCTION			
Rationale	6	Describe the rationale for the review in the context of what is already known	P3,4
Objectives	7	Provide an explicit statement of the question(s) the review will address with reference to participants, interventions, comparators, and outcomes (PICO)	P5
METHODS			
Eligibility criteria	8	Specify the study characteristics (such as PICO, study design, setting, time frame) and report characteristics (such as years considered, language, publication status) to be used as criteria for eligibility for the review	P5,6
Information sources	9	Describe all intended information sources (such as electronic databases, contact with study authors, trial registers or other grey literature sources) with planned dates of coverage	P6
Search strategy	10	Present draft of search strategy to be used for at least one electronic database, including planned limits, such that it could be repeated	P6

Study records:			
Data management	11a	Describe the mechanism(s) that will be used to manage records and data throughout the review	P6-8
Selection process	11b	State the process that will be used for selecting studies (such as two independent reviewers) through each phase of the review (that is, screening, eligibility and inclusion in meta-analysis)	P6,7
Data collection process	11c	Describe planned method of extracting data from reports (such as piloting forms, done independently, in duplicate), any processes for obtaining and confirming data from investigators	P7
Data items	12	List and define all variables for which data will be sought (such as PICO items, funding sources), any pre-planned data assumptions and simplifications	P7,8
Outcomes and prioritization	13	List and define all outcomes for which data will be sought, including prioritization of main and additional outcomes, with rationale	P5,6
Risk of bias in individual studies	14	Describe anticipated methods for assessing risk of bias of individual studies, including whether this will be done at the outcome or study level, or both; state how this information will be used in data synthesis	P7,8
Data synthesis	15a	Describe criteria under which study data will be quantitatively synthesised	P8
	15b	If data are appropriate for quantitative synthesis, describe planned summary measures, methods of handling data and methods of combining data from studies, including any planned exploration of consistency (such as I^2, Kendall's τ)	P7,8
	15c	Describe any proposed additional analyses (such as sensitivity or subgroup analyses, meta-regression)	P8
	15d	If quantitative synthesis is not appropriate, describe the type of summary planned	P8
Meta-bias(es)	16	Specify any planned assessment of meta-bias(es) (such as publication bias across studies, selective reporting within studies)	P8
Confidence in cumulative evidence	17	Describe how the strength of the body of evidence will be assessed (such as GRADE)	P8

*** It is strongly recommended that this checklist be read in conjunction with the PRISMA-P Explanation and Elaboration (cite when available) for important clarification on the items. Amendments to a review protocol should be tracked and dated. The copyright for PRISMA-P (including checklist) is held by the PRISMA-P Group and is distributed under a Creative Commons Attribution Licence 4.0.**

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Efficacy and safety of mesenchymal stem cells for the treatment of patients infected with 2019 novel coronavirus (COVID-19): a systematic review and meta-analysis protocol

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ABSTRACT

Introduction To date, no specific anti-virus drugs or vaccines have been available to prevent or treat the 2019 novel coronavirus disease (COVID-19) pandemic. Mesenchymal stem cells (MSCs) therapy may be a promising therapeutic approach that reduces the high mortality in critical cases. This protocol is proposed for a systematic review and meta-analysis that aims to evaluate the efficacy and safety of MSCs therapy on patients with COVID-19.

Methods and analysis Ten databases including PubMed, EMBASE, Cochrane Library, CIHAHL, Web of Science, Chinese National Knowledge Infrastructure (CNKI), Chinese Scientific Journals Database (VIP), Wanfang database, China Biomedical Literature Database(CBM) and Chinese Biomedical Literature Service System (SinoMed) will be searched from inception to 1 December 2020. All published randomized controlled trials (RCTs), clinical controlled trials (CCTs), and case series that meet the pre-specified eligibility criteria will be included. The primary outcomes include mortality, incidence and severity of adverse events, respiratory improvement, days from ventilator, duration of fever, progression rate from mild or moderate to severe, improvement of such serious symptoms as difficult breathing or shortness of breath, chest pain or pressure, and loss of speech or movement, biomarkers of laboratory examination, and changes in computed tomography. The secondary outcomes include dexamethasone doses and quality of life. Two reviewers will independently perform study selection, data extraction, and assessment of bias risk. Data synthesis will be conducted using RevMan software (V.5.3.5). If necessary, subgroup and sensitivity analysis will be performed. GRADE system will be utilized to assess the strength of evidence.

Ethics and dissemination Ethical approval is not necessary since no individual patient or privacy data has been collected. The results of this review will be disseminated in a peer-reviewed journal or an academic conference presentation.

Trial registration number PROSPERO 2020 CRD42020190079.

Keywords MSCs; COVID-19; systematic review; meta-analysis; protocol

Strengths and limitations of this study

- This systematic review protocol follows the Preferred Reporting Items for Systematic Review and Meta-Analysis Protocols guidelines.

- This systematic review, to the best of our knowledge, will be the first to explore the efficacy and safety of MSCs therapy for the treatment of patients infected with 2019 novel coronavirus (COVID-19).
- The study will systematically review quantitative data from various medical databases for an in-depth interpretation of the efficacy and safety of MSCs therapy on patients with COVID-19.
- The potential for low and inconsistent quality in the reporting of process evaluations, the publication bias, and the methodological quality of the grey literature found may be the limitations of the study. Other potential limitation might be whether a sufficient number of trials would be completed such that patient data is widely available to make interpretations or draw conclusions.

INTRODUCTION

Description of the condition

Coronavirus disease 2019 (COVID-19), an infectious disease caused by a novel severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2, formerly termed as 2019-nCoV), has been sweeping the world.[1] However, to date, no efficient vaccines and specific antiviral medications are available to halt the pandemic. As of 24 June 2020, 9,110,186 confirmed cases of COVID-19 have been documented globally in 216 countries, areas or territories with 473,061 deaths.[2] Notably, approximately 15% of the infected patients develops severe pneumonia and 5% progresses to acute respiratory distress syndrome (ARDS), septic shock and multiorgan failure eventually.[3 4] High mortality rate was observed in critically ill patients and has prompted an urgent need for treatments that can address the critical cases and prevent fatal outcomes.[5 6]

Description of the intervention

Currently, mesenchymal stem cells (MSCs)-based treatment has been proposed as a promising therapeutic approach for patients with COVID-19. [7] MSCs are multipotent cells that can be obtained from various tissues including preferably bone marrow, adipose tissue, placenta, umbilical cord, and dental pulp. The safety and effectiveness of MSCs therapy have been well documented in several clinical studies including ARDS, bronchopulmonary dysplasia, and cardiovascular diseases. [8-12] In a recent case study, a 65-year-old critically ill ventilator ridden COVID-19 patient was treated with allogeneic human umbilical cord MSCs (three infusions of 5×10^7 cells at an interval of three days) and the patient was off the ventilator

and able to walk after the second infusion. No obvious side effects were observed. [13] In another study, seven patients with COVID-19 (one critically severe, four severe, and two mild-to-moderate) received a single intravenous administration of MSCs at a dose of 1×10^6 cells/kg body weight, all patients had significant improvements in clinical symptoms two days after the transplantation, one severe and two mild patients were discharged from the hospital by day 10, and a 14-day follow-up showed no adverse effects. [14] These completed clinical trials provided limited but promising evidence that the use of MSCs therapy might be efficient in the treatment of COVID-19. The US Food and Drug Administration authorized compassionate use of MSCs therapy in patients with an extremely dismal prognosis. Recently, 45 clinical trials of MSCs as a new treatment for COVID-19 have been registered on ChiCTR (http://www.chictr.org.cn) and clinicaltrials.gov (https://clinicaltrials.gov). Several trials are underway that may provide more evidence to evaluate the safety and effectiveness of MSCs therapy for COVID-19.

How the intervention might work

A growing body of research has interpreted the potential mechanism underlying the therapeutic effect of MSCs on COVID-19. SARS-CoV-2 cell entry depends on the angiotensin-converting enzyme-2 (ACE2) and the transmembrane serine protease 2 (TMPRSS2), gene expression profiling exhibited that transplanted MSCs were ACE2- and TMPRSS2- and had a natural immunity to the COVID-19 infection.[14 15] Virus-induced cytokine storm is considered as the hallmark of SARS-Cov-2 pathogenesis and has been reported to induce ARDS and multi-organ failure, leading to death in COVID-19 patients. [16] To prevent or attenuate the cytokine storm is pivotal to halt the pandemic. Compelling studies have demonstrated that MSCs have potent and broad immunomodulatory and anti-inflammatory effects. Such processes include the regulation of T cell function, proliferation and differentiation of B cells, influence of innate immune cells (such as macrophages and dendritic cells), decrease of pro-inflammatory cytokines (such as IL-1, IL-6, IFN, and TNF-α), increase of anti-inflammatory cytokines (such as IL-4, IL-5, and IL-10) and peripheral lymphocytes, and decline of over-activated cytokine-secreting immune cells (such as $CXCR3^+$ $CD4^+$ T cells, $CXCR3^+$ $CD8^+$ T cells, and $CXCR3^+$ NK cells).[14 17-23] Besides, MSCs inhibit bacterial growth, enhance the restoration of injured alveolar epithelial cells, improve pulmonary microenvironment, alleviate pulmonary fibrosis, and enhance pulmonary function.[24 25] Kyoto Encyclopedia of Genes and Genomes analysis implied that MSCs were involved in antiviral pathways.[14] Hence, MSCs therapy may improve the outcome of COVID-19 patients through immunomodulation, regulating the inflammatory response, and promoting tissue repair.

Why it is important to perform this review

The pandemic COVID-19 caused by SARS-CoV-2 has been sweeping the world. MSCs therapy has been proposed as a safe, effective, and promising approach to treat COVID-19, especially for severe or critical cases. It is important to perform a systematic review and meta-analysis to evaluate its efficacy and safety. In this systematic review, all potential RCTs, CCTs, and case series regarding MSCs for the treatment of COVID-19 will be fully considered and synthesized without language or publication restrictions. The findings of this study may yield helpful evidence for the patients, clinicians, investigators, and policymakers concerned about the efficacy and safety of MSCs therapy on COVID-19.

OBJECTIVES

This systematic review and meta-analysis aim to assess the evidence for the effectiveness and safety of MSCs therapy for COVID-19.

METHODS AND ANALYSIS

A statement: this is to clarify that due to the similarity of the methods, there is some overlap with our previous publication. [26]

Criteria for including studies for this review

Types of studies

This review will include randomized controlled trials (RCTs), clinical controlled trials (CCTs), and case series of MSCs treatment for COVID-19. Snowballed papers from references will also be included. Animal-based research and literature review will be excluded.

Types of participants

This review will include patients diagnosed with COVID-19 using any recognized diagnostic criteria regardless of the age, gender, and source of cases and the duration and severity of the disease. Patients infected with adenovirus, rhinovirus, human metapneumovirus, etc. will be excluded.

Types of interventions

The intervention group will receive treatment of MSCs and standard care. This review will impose no restriction on standard care regimens (including supportive treatment, IFN-α , lopinavir, or ritonavir).

Types of comparator(s)/control

The control group will receive the same treatment of standard care regimen as the intervention group in the same original study. This review will impose no restrictions regarding standard care regimen.

Types of outcome measures

Primary outcomes

The primary outcomes include mortality, incidence and severity of adverse events, respiratory improvement, days from ventilator, duration of fever, progression rate from mild or moderate to severe, improvement of such serious symptoms as difficult breathing or shortness of breath, chest pain or pressure, and loss of speech or movement, biomarkers of laboratory examination, and changes in computed tomography.

Secondary outcomes

Secondary outcomes include dexamethasone doses and quality of life.

Search methods for identification of studies

Electronics searches

The following ten databases will be searched from the inception to 1 December 2020: PubMed, EMBASE, Cochrane Library, CIHAHL, Web of Science, Chinese National Knowledge Infrastructure(CNKI), Chinese Scientific Journals Database (VIP), Wanfang database, China Biomedical Literature Database(CBM) and Chinese Biomedical Literature Service System (SinoMed). The literature will be searched by two reviewers independently. Any discrepancies will be resolved by consultation with a third reviewer. Manual search will be performed on the reference lists of included studies for relevant publications.

The reviewers will use a combination of the following terms to conduct the electronic search: novel coronavirus, Severe Acute Respiratory Syndrome Coronavirus 2, SARS-CoV-2, COVID-19, COVID19, 2019-nCoV, mesenchymal stem cells, mesenchymal stromal cells, MSCs, stem cells, SCs, randomized controlled trial, controlled clinical trial, randomized, randomly, trials, case series, CCT, and RCT. Table 1 presents the search strategy for PubMed, and the reviewers will modify the strategy correspondingly upon the requirement of other databases.

Searching other resources

The reviewers will also search the studies from Clinical Trials.gov (http://www.clinical trails.gov), Chinese Clinical Trial Registry (http://www.chictr. org/cn/), and WHO International Clinical Trial Registry Platform (https://www.who.int/ictrp/en/). Grey literature such as guidelines, research and committee reports, government reports, and conference papers will be obtained from WHO, U.S National Library of Medicine, China Centre for Disease Control and Prevention, and online official news websites.

The reviewers will manually review the reference lists of the retrieved articles for further additional trials. For incomplete data, the corresponding author will be contacted.

Table 1 Search strategy for the PubMed

No.	Search terms
#1	Novel coronavirus
#2	Severe Acute Respiratory Syndrome Coronavirus 2
#3	SARS-CoV-2
#4	COVID-19
#5	COVID19
#6	2019-nCoV
#7	OR/#1-#6
#8	Stem cells
#9	SC
#10	Mesenchymal stem cells
#11	mesenchymal stromal cells
#12	MSCs
#13	OR/#8-#12
#14	7# AND 13#
#15	Randomized controlled trial
#16	Controlled clinical trial
#17	Randomized*
#18	Randomly*
#19	Trails
#20	Case series
#21	CCT
#22	RCT
#23	OR/#14-#22
#24	#7AND#14AND#23

*Represent one or more characters of all characters.

Data collection and analysis

Selection of studies

Two reviewers will independently conduct screening, study selection, and data extraction. The reviewers will import the literature obtained into EndnoteX9, screen the title and abstract, and exclude the duplications and studies that did not meet the inclusion criteria. The final included studies will be determined after reading the full text of the remaining studies. If the full text is not available, the corresponding author of the original study will be contacted**.** A third reviewer will be consulted for arbitrating any disagreements. A PRISMA flow chart is presented to summarize the entire process of the study selection (figure 1).

Data extraction and management

Two reviewers will independently extract the following data from eligible studies using a pre-specified data acquisition form: reference ID, author information, year of publication, study type, study design, setting of study, sample size, participant characteristics (age, gender, duration and severity of illness,

laboratory test, CT scan, etc.), MSCs intervention group and control group (details of randomization, blinding, allocation, intervention approach and duration), and primary and secondary outcomes at all reported time points. A third reviewer will be consulted to solve any inconsistency between the two reviewers. A cross-check will be performed on all data before transferring to RevMan software (V.5.3).

Assessment of risk of bias

The Cochrane Risk of Bias Assessment Tool will be used by two reviewers to independently evaluate the risk of publication bias for all the included studies in eight domains, namely randomization sequence generation, randomization allocation concealment, blinding of participants, blinding of personnel, blinding of outcome assessors, incomplete outcome data, selective reporting bias, and other bias. Each domain will be graded as high, unclear, or low risk of bias. For unclear domain, the reviewers will contact the corresponding authors of the original study. Any inconsistency will be solved by discussion with a third reviewer.

Measures of treatment effect

Two reviewers will independently synthesize and statistically analyze efficacy data using RevMan 5.3.. The reviewers will use a risk ratio or odds ratio with 95% CIs for dichotomous data and a mean difference (MD) or standard mean difference (SMD) with 95% CIs for continuous data. When different assessment tools are employed, SMD will be utilized.

Dealing with missing data

If the required data is unclear or unavailable, the original study's corresponding author will be contacted by E-mail or telephone. If data is still unattainable, the reviewers will exclude the study concerned from the analysis. To address the potential effects of missing data, a sensitivity analysis will be conducted.

Assessment of heterogeneity

χ^2 test and I^2 statistic will be employed to investigate statistical heterogeneity. The fixed-effect model will be utilized for a low heterogeneity (I^2<50%) and the random-effects model will be applied when the heterogeneity is moderate (50% < I^2 <75%). Meta-analysis will not be conducted if the heterogeneity is remarkably high (I^2 >75%).

Assessment of reporting biases

This review will perform funnel plots to assess potential reporting bias when more than ten eligible studies are included. Additionally, the Egger regression and Begg correlation test will be used to identify the funnel plot asymmetry.

Data synthesis

Following the Cochrane guideline, the reviewers will utilize the fixed-effect model for the pooled data when heterogeneity is deemed low and the random-effect model if heterogeneity is moderate.[27] This review will perform subgroup analysis or meta-regression to evaluate the potential sources and provide reasonable explanations when heterogeneity is considerably high. $P<0.05$ is deemed statistically significant. If the meta-analysis is unfeasible, the results will be described narratively.

Subgroup analysis and investigation of heterogeneity

If feasible, subgroup analyses will be performed in terms of the disease severity of included patients, duration of disease, routes of administration, dosage, and origin of MSCs. Subgroup analyses will be conducted to interpret the heterogeneity.

Sensitivity analysis

If feasible, this review will conduct sensitivity analysis to assess the robustness of the pooled effects of the included studies given the impact of such variables as sample size, methodological quality, missing data, or high risk of bias.

Summary of evidence

This review will utilize the Cochrane Collaboration Network GRADE (The Grading of Recommendations Assessment Development and Evaluation) to grade the quality of evidence as very low, low, moderate or high.[28] [29] The quality of evidence of a specific study will be evaluated based on the risk of bias, imprecision, inconsistency, indirectness, publication bias, effect size or dose-response relation. The findings will be presented in A Summary of Finding table. Any discrepancy will be resolved by discussion or arbitrated by a third reviewer.

Patient and public involvement

No patient involved.

Ethics and dissemination

Ethical approval is not necessary as no individual patient or privacy data will be collected. The results of this study will be disseminated in a peer-review journal or an academic conference presentation.

DISCUSSION

This meta-analysis will analyse the efficacy and safety of MSCs therapy for treatment of COVID-19 patients, using a structured and valid methodology. Conclusions drawn from this study may provide useful information and facilitate the decision-making process of patients, clinicians, investigators, and policymakers. The process of performing this review will include identification, study inclusion, data extraction, and data synthesis. If this protocol needs to be amended, we will provide the date of each amendment with a statement of the changes and corresponding reasons. For the ongoing incoming literature, this meta-analysis will be regularly updated with new incoming data from randomized studies.

Author Contributions Y-H C, Q Z, T-E Z and S-Q T conceived and designed the study. The protocol was drafted by Y-H C, Q Z, X-L L and S-Q T and revised by Y-H C, W P and T-E Z. Y-H C, W P, S-Q T and T-E Z designed the search strategies. X-L L and D L will perform searching, data curation and assessment independently. Q Z and Y-Y Y will analyze and interpret the data. S-Q T, T-E Z, X-L L and Y-H C will arbitrate disagreement if there is anything during the review. All authors have read and approved the publication of the protocol.

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Competing interests None declared.

Patient consent for publication Not required.

Ethics approval Ethics approval is not necessary because no individual patient data and privacy data will be collected.

Provenance and peer review Not commissioned; externally peer reviewed.

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REFERENCES

1. Lu R, Zhao X, Li J, et al. Genomic characterisation and epidemiology of 2019 novel coronavirus: implications for virus origins and receptor binding. *Lancet*. 2020;395(10224):565-574.
2. World Health Organization. Rolling updates on coronavirus disease (COVID-19). Retrieval June 24, 2020 on https://www.who.int/emergencies/diseases/novel-coronavirus-2019/events-as-they-happen.

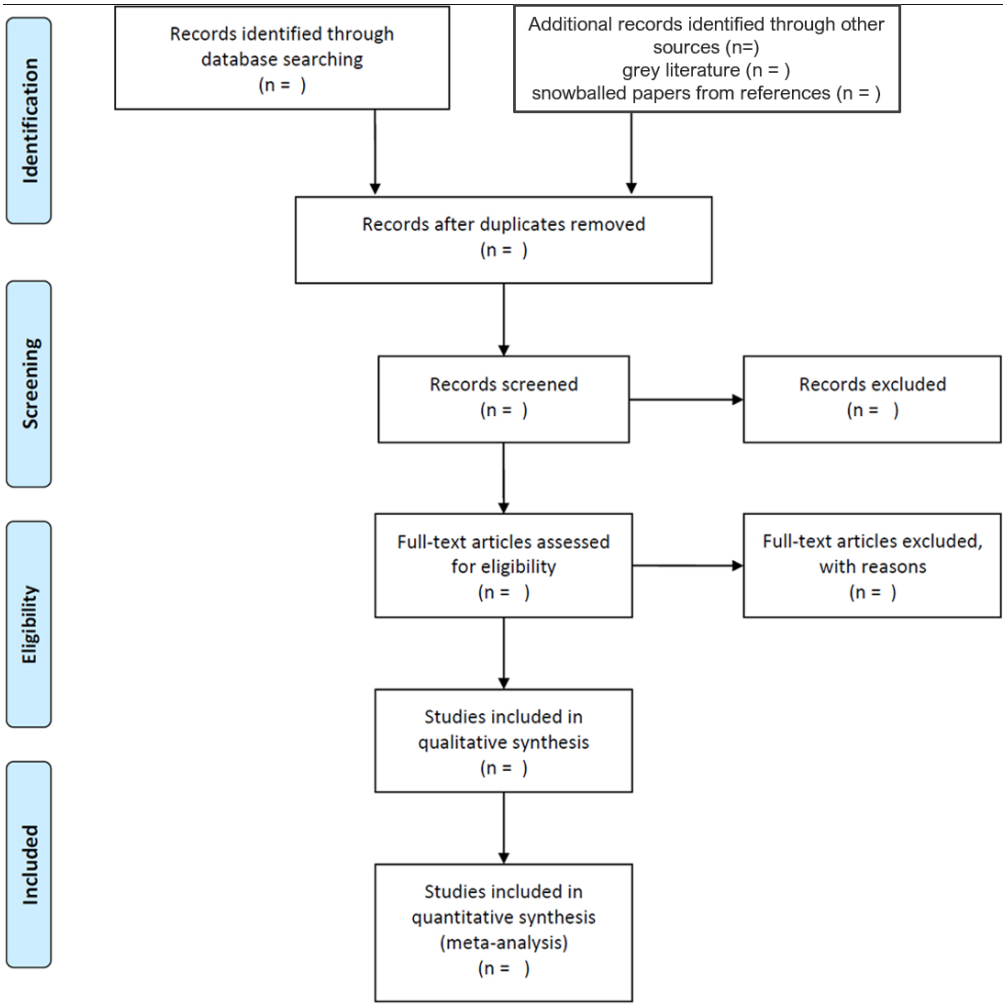
3. Huang C, Wang Y, Li X, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet*. 2020;395(10223):497-506.
4. Xu Z, Shi L, Wang Y, et al. Pathological findings of COVID-19 associated with acute respiratory distress syndrome. *Lancet Respir Med*. 2020;8(4):420-422.
5. Bhatraju PK, Ghassemieh BJ, Nichols M, et al. Covid-19 in Critically Ill Patients in the Seattle Region - Case Series. *N Engl J Med*. 2020;382(21):2012-2022.
6. Zumla A, Hui DS, Azhar EI, Memish ZA, Maeurer M. Reducing mortality from 2019-nCoV: host-directed therapies should be an option. *Lancet*. 2020;395(10224):e35-e36.
7. Golchin A, Farahany TZ. Biological Products: Cellular Therapy and FDA Approved Products. *Stem Cell Rev Rep*. 2019;15(2):166-175.
8. Golchin A, Farahany TZ, Khojasteh A, Soleimanifar F, Ardeshirylajimi A. The Clinical Trials of Mesenchymal Stem Cell Therapy in Skin Diseases: An Update and Concise Review. *Curr Stem Cell Res Ther*. 2019;14(1):22-33.
9. Matthay MA, Calfee CS, Zhuo H, et al. Treatment with allogeneic mesenchymal stromal cells for moderate to severe acute respiratory distress syndrome (START study): a randomised phase 2a safety trial. *Lancet Respir Med*. 2019;7(2):154-162.
10. Chen J, Hu C, Chen L, et al. Clinical study of mesenchymal stem cell treating acute respiratory distress syndrome induced by epidemic Influenza A (H7N9) infection, a hint for COVID-19 treatment [published online ahead of print, 2020 Feb 28]. *Engineering (Beijing)*. 2020;10.1016/j.eng.2020.02.006. doi:10.1016/j.eng.2020.02.006
11. Namba F. Mesenchymal stem cells for the prevention of bronchopulmonary dysplasia. *Pediatr Int*. 2019;61(10):945-950.
12. Suvakov S, Richards C, Nikolic V, et al. Emerging Therapeutic Potential of Mesenchymal Stem/Stromal Cells in Preeclampsia. *Curr Hypertens Rep*. 2020;22(5):37.
13. Bing Liang, Junhui Chen, Tao Li, et al. Clinical remission of a critically ill COVID-19 patient treated by human umbilical cord. chinaXiv, 2020.
14. Leng Z, Zhu R, Hou W, et al. Transplantation of ACE2- Mesenchymal Stem Cells Improves the Outcome of Patients with COVID-19 Pneumonia. *Aging Dis*. 2020;11(2):216-228.
15. Hoffmann M, Kleine-Weber H, Schroeder S, et al. SARS-CoV-2 Cell Entry Depends on ACE2 and TMPRSS2 and Is Blocked by a Clinically Proven Protease Inhibitor. *Cell*. 2020;181(2):271-280.e8.
16. Soy M, Keser G, Atagündüz P, Tabak F, et al. Cytokine storm in COVID-19: pathogenesis and overview of anti-inflammatory agents used in treatment. *Clin Rheumatol*. 2020;39(7):2085-2094. doi:10.1007/s10067-020-05190-5
17. Chen QH, Wu F, Liu L, et al. Mesenchymal stem cells regulate the Th17/Treg cell balance partly through hepatocyte growth factor in vitro. *Stem Cell Res Ther*. 2020;11(1):91.
18. Luz-Crawford P, Hernandez J, Djouad F, et al. Mesenchymal stem cell repression of Th17 cells is triggered by mitochondrial transfer. *Stem Cell Res Ther*. 2019;10(1):232.
19. Cho KA, Lee JK, Kim YH, et al. Mesenchymal stem cells ameliorate B-cell-mediated immune responses and increase IL-10-expressing regulatory B cells in an EBI3-dependent manner. *Cell Mol Immunol*. 2017;14(11):895-908.
20. de Witte SFH, Luk F, Sierra Parraga JM, et al. Immunomodulation By Therapeutic Mesenchymal Stromal Cells (MSC) Is Triggered Through Phagocytosis of MSC By Monocytic Cells. *Stem Cells*. 2018;36(4):602-615.
21. Reis M, Mavin E, Nicholson L, et al. Mesenchymal Stromal Cell-Derived Extracellular Vesicles Attenuate Dendritic Cell Maturation and Function. *Front Immunol*. 2018;9:2538.
22. Shi Y, Wang Y, Li Q, et al. Immunoregulatory mechanisms of mesenchymal stem and stromal cells in inflammatory diseases. *Nat Rev Nephrol*. 2018;14(8):493-507.
23. Uccelli A, de Rosbo NK. The immunomodulatory function of mesenchymal stem cells: mode of action and pathways. *Ann N Y Acad Sci*. 2015;1351:114-126.
24. Krasnodembskaya A, Song Y, Fang X, et al. Antibacterial effect of human mesenchymal stem cells is mediated in part from secretion of the antimicrobial peptide LL-37. *Stem Cells*. 2010;28(12):2229-2238.

25. Rajarshi K, Chatterjee A, Ray S. Combating COVID-19 with Mesenchymal Stem Cell therapy [published online ahead of print, 2020 May 14]. *Biotechnol Rep (Amst)*. 2020;26:e00467.
26. Liu D, You Y, Chen Y, et al. Efficacy of integrative Traditional Chinese and Western medicine for the treatment of patients infected with 2019 novel coronavirus (COVID-19): A protocol for systematic review and meta analysis. Medicine (Baltimore). 2020; 99(29):e20781.
27. Chandler J, Higgins JPT, Deeks JJ, et al. Cochrane handbook for systematic reviews of interventions version 5.2.0: Cochrane Community, 2017
28. Guyatt GH, Oxman AD, Vist GE, et al. GRADE: an emerging consensus on rating quality of evidence and strength of recommendations. *BMJ*. 2008;336(7650):924-926. doi:10.1136/bmj.39489.470347.AD
29. Guyatt GH, Oxman AD, Schünemann HJ, et al. GRADE guidelines: a new series of articles in the Journal of Clinical Epidemiology. *J Clin Epidemiol*. 2011;64(4):380-382. doi:10.1016/j.jclinepi.2010.09.011

Figure Legends

Figure 1. Flowchart of study selection

Table 1. Search strategy for the PubMed



PRISMA flowchart

PRISMA-P (Preferred Reporting Items for Systematic review and Meta-Analysis Protocols) 2015 checklist: recommended items to address in a systematic review protocol*

Section and topic	Item No	Checklist item	Reported on Page #
ADMINISTRATIVE INFORMATION			
Title:			
Identification	1a	Identify the report as a protocol of a systematic review	P1
Update	1b	If the protocol is for an update of a previous systematic review, identify as such	N/A
Registration	2	If registered, provide the name of the registry (such as PROSPERO) and registration number PROSPERO CRD42020190079	P2
Authors:			
Contact	3a	Provide name, institutional affiliation, e-mail address of all protocol authors; provide physical mailing address of corresponding author	P1
Contributions	3b	Describe contributions of protocol authors and identify the guarantor of the review	P9
Amendments	4	If the protocol represents an amendment of a previously completed or published protocol, identify as such and list changes; otherwise, state plan for documenting important protocol amendments	N/A
Support:			
Sources	5a	Indicate sources of financial or other support for the review	P9
Sponsor	5b	Provide name for the review funder and/or sponsor	N/A
Role of sponsor or funder	5c	Describe roles of funder(s), sponsor(s), and/or institution(s), if any, in developing the protocol	N/A
INTRODUCTION			
Rationale	6	Describe the rationale for the review in the context of what is already known	P3,4
Objectives	7	Provide an explicit statement of the question(s) the review will address with reference to participants, interventions, comparators, and outcomes (PICO)	P5
METHODS			
Eligibility criteria	8	Specify the study characteristics (such as PICO, study design, setting, time frame) and report characteristics (such as years considered, language, publication status) to be used as criteria for eligibility for the review	P5,6
Information sources	9	Describe all intended information sources (such as electronic databases, contact with study authors, trial registers or other grey literature sources) with planned dates of coverage	P6
Search strategy	10	Present draft of search strategy to be used for at least one electronic database, including planned limits, such that it could be repeated	P6

Study records:			
Data management	11a	Describe the mechanism(s) that will be used to manage records and data throughout the review	P6-8
Selection process	11b	State the process that will be used for selecting studies (such as two independent reviewers) through each phase of the review (that is, screening, eligibility and inclusion in meta-analysis)	P6,7
Data collection process	11c	Describe planned method of extracting data from reports (such as piloting forms, done independently, in duplicate), any processes for obtaining and confirming data from investigators	P7
Data items	12	List and define all variables for which data will be sought (such as PICO items, funding sources), any pre-planned data assumptions and simplifications	P7,8
Outcomes and prioritization	13	List and define all outcomes for which data will be sought, including prioritization of main and additional outcomes, with rationale	P5,6
Risk of bias in individual studies	14	Describe anticipated methods for assessing risk of bias of individual studies, including whether this will be done at the outcome or study level, or both; state how this information will be used in data synthesis	P7,8
Data synthesis	15a	Describe criteria under which study data will be quantitatively synthesised	P8
	15b	If data are appropriate for quantitative synthesis, describe planned summary measures, methods of handling data and methods of combining data from studies, including any planned exploration of consistency (such as I^2, Kendall's τ)	P7,8
	15c	Describe any proposed additional analyses (such as sensitivity or subgroup analyses, meta-regression)	P8
	15d	If quantitative synthesis is not appropriate, describe the type of summary planned	P8
Meta-bias(es)	16	Specify any planned assessment of meta-bias(es) (such as publication bias across studies, selective reporting within studies)	P8
Confidence in cumulative evidence	17	Describe how the strength of the body of evidence will be assessed (such as GRADE)	P8

*** It is strongly recommended that this checklist be read in conjunction with the PRISMA-P Explanation and Elaboration (cite when available) for important clarification on the items. Amendments to a review protocol should be tracked and dated. The copyright for PRISMA-P (including checklist) is held by the PRISMA-P Group and is distributed under a Creative Commons Attribution Licence 4.0.**

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