Clinical Lessons of MSC Therapy Over the Past 15 Years: A Systematic Review and Meta-Analysis

Yang Wang ( wangyang5@mail2.sysu.edu.cn )
Third affiliated hospital of Sun-Yat sen university  https://orcid.org/0000-0003-2809-2827

Hanxiao Yi
Sun Yat-Sen Memorial Hospital

Yancheng Song
The First Affiliated Hospital of Guangdong Pharmaceutical University

Research

Keywords: MSC therapy, randomized clinical trials, all populations, safety, meta-analysis

DOI: https://doi.org/10.21203/rs.3.rs-587655/v1

License: © This work is licensed under a Creative Commons Attribution 4.0 International License. Read Full License
Abstract

Background: Despite increasing clinical investigations in emphasizing the safety of MSC therapy in different populations with different diseases, recently, no article overall reviewed the side events in all populations.

Aim: To evaluate the safety of MSC therapy in all populations receiving MSC therapy and explore the potential heterogeneities influencing the clinical application of MSC.

Methods: The databases of PubMed, Embase, Web of Science and Scopus were searched from onset until March 1st, 2021.

Results: All side events were displayed as odds ratio (OR) and 95% CI (confidence intervals). Totally, 62 randomized clinical trials (RCTs) that enrolled 3546 participants diagnosed with various diseases (about 20 kinds of diseases) treated with intravenous or local implantation vs placebo, or no treatment were included. All studies were high quality, neither serious publication bias nor serious adverse events (such as death and infection) were discovered across included studies. The pooled analysis demonstrated that MSC administration was extremely associated with transient fever [OR, 3.65, 95% CI: 2.05 to 6.49, \(p<0.01\)], administration site conditions [OR, 1.98, 95% CI: 1.01 to 3.87, \(p=0.05\)], constipation [OR, 2.45, 95% CI: 1.01 to 5.97, \(p=0.05\)], fatigue [OR, 2.99, 95% CI: 1.06 to 8.44, \(p=0.04\)], and sleeplessness [OR, 5.90, 95% CI: 1.04 to 33.47, \(p=0.05\)]. Interestingly, MSC administration trended to lower rather than boost the incidence rate of arrhythmia [OR, 0.62, 95% CI: 0.36 to 1.07, \(p=0.09\)].

Conclusions: Conclusively, MSC administration was safe in different populations compared with the other placebo modalities.

Introduction

Mesenchymal stromal cells (MSCs), a class of highly heterogeneous cells, that can be isolated from bone marrow, adipose, umbilical cord, and placenta, are primarily discovered in 1974 by Friedenstein[1]. Over these years, exogenous MSCs are amazingly found to have a therapeutic effect on many diseases (e.g., myocardial infarction, liver cirrhosis, limb ischemia and spinal cord injury)[2-5].

Deferring from multipotent stem cells, the potency of MSCs is restricted but MSCs can be induced into osteoblasts, chondrocytes and adipocytes in vitro. Universally, MSCs exert their favourable effects by immunomodulatory regulation and paracrine manners[6, 7]. Clinically, MSCs have been applied in many refractory diseases, such as cerebral palsy[8], spinal cord injury[9] and systemic lupus erythematosus[10]. However, MSCs easily flock together forming the core of the clot and leading to vascular disorders. Additional, MSCs are tumorigenic as a result of their reproductive capacity and potentially cause acute or chronic immunogenicity of the cells themselves as foreign matters[11-13]. A large number of studies, most of which are small samples, have been investigating the safety of MSCs transplantation, but no article overall reviews these studies to characterize the side events closely associated with MSCs administration over the past 9 years.

We performed this systematic review and meta-analysis to identify all treatment-related side events concerning MSCs administration and explore the safety of MSCs in clinical utilization.

Methods And Materials

Search results

This meta-analysis was limited to published articles assessing the safety of MSC administration and was performed by searching PubMed, EMBASE, Web of Science, Scopus, and the Cochrane Library databases (from inception to 1st March 2021). The search strategy is as follows: ((MSC [title/abstract]) OR (mesenchymal stem cell [title/abstract]) OR (Wharton's jelly [title/abstract])) AND ((safety [title/abstract]) OR (side event [title/abstract]) OR (side effect [title/abstract]) OR (adverse event [title/abstract]) OR (adverse effect [title/abstract])). The reference lists of the included articles were also browsed to identify potential studies. To perform a comprehensive search, we did not limit the "study type"; retrospective studies were excluded during the study selection process. The detailed database search strategy is provided in Additional file 1.

Article selection

Primarily, duplicates of all articles were excluded. Two participants initially screened all titles and abstracts to preclude articles that were irrelated to our research objectives. Then, we carefully read the full manuscripts and selected the eligible ones.

Eligibility criteria

The selection process strictly obeyed the PICOS (participants, interventions, comparison, outcome and study) principles and these principles were listed in Table 1.

Data extraction

Two skilled reviewers (YW and HX Y) independently extracted data from all articles according to pre-set criteria. We retrieved 12 characteristic entries from the original articles, including author, year, and study type, location, disease, and cell type, administration method, study phrase, and language, dose, follow-up day, and the NO. of the patient in each group. Conflicts were resolved in consultation with a third referee.

Quality assessment
Risk of bias in individual study and across studies were performed by using the Cochrane Collaboration’s tool for assessing the risk of bias.

Outcome definition

Totally, we reported 17 side events appeared during MSC therapy, and of which 9 events (death, infection, diarrhea, central nervous system disorders, arrhythmia, urticaria/dermatitis, vascular disorders) were classified into major events, and 8 events (anemia, constipation, metabolism disorders, fatigue, nausea, seizure, sleepless, vomiting) were classified into minor events. One event would be considered as a major event if it was reported by more than 5 studies or life-threatening judged by our clinician; otherwise, it should be sorted into a minor event. Among these events, some events were not specifically clinical symptoms but referred to a series of correlated symptoms, such as central nervous system disorders, vascular disorders, infection, arrhythmia, administration site conditions, metabolism and nutrition disorders. These side events were redefined in Table 2. Other entries were retrieved from the original definitions.

Statistical analysis

All data were synthesized by using R software version 4.0.3 (University of Auckland, New Zealand). All results presented in this article were presented as odds ratio (OR) with 95% CIs for outcomes. A random-effect model was used to analyze the data when heterogeneity was significant (p<0.05 or I²>50%); otherwise, a fixed-effect model was used. Publication bias was tested by Egger’s and Begg’s tests were utilized to analyze the publication bias of the included articles with R software version 4.0.3 (meta package). Subgroup analysis was also conducted to seek potential heterogeneous factors.

Results

The items of this meta-analysis were reported according to the Preferred Reporting Items for Systematic Review and Meta-Analysis (PRISMA) (Additional file 2).

Article selection process

Approximately, 2078 articles were identified after the initial search. 1898 irrelevant articles were eliminated through browsing titles and abstracts, and 118 articles were excluded due to unexpected outcomes and interventions. Finally, 62 clinical trials, including 2 trials from the reference list, were taken into the analysis even if the elimination of 2 systematic reviews (Figure 1).

Baseline of included studies

The data were purified from studies performed over the past 11 years. Only 2 studies were prospective and the rest were randomized controlled trials (RCTs) ranging from study phrase 1/2 to study phrase 3. Asia was ranked first in the No. of studies, and then North America and Europe. The MSC used in these studies were mainly isolated from bone marrow, adipose and umbilical cord. The injection dose ranged from 4×10⁷ to 1.2×10⁹ cells. The follow-up day was from 6 months to 2 years.

Pooled analysis of all studies

Totally, 62 clinical trials, containing populations with different characteristics, were included into analysis (Figure 2A). We discovered that MSC administration would not induce major side events, such as vascular disorders (1.17, 95% CI: 0.52-2.62, p=0.70), urticaria/dermatitis (0.93, 95% CI: 0.93-1.07, p=0.70), central nervous system disorders (1.13, 95% CI: 0.61-2.12, p=0.69), diarrhea (0.90, 95% CI: 0.49-1.63, p=0.73), death (0.99, 95% CI: 0.66-1.49, p=0.96), infection (1.03, 95% CI: 0.70-1.53, p=0.87). However, our analysis demonstrated that transient fever (3.65, 95% CI: 2.05-6.49) will occur within 48 hours if people receive MSC administration. Meanwhile, MSC injections also potentially caused administration site conditions (1.98. 95% CI: 1.01-3.87, p<0.01). Populations tended to benefit from receiving MSC therapy as they were like to have a lower rate of arrhythmia (0.62, 95% CI: 0.36-1.07, p=0.05).

As for minor side events, MSC presumably caused sleeplessness (5.90, 95% CI: 1.04-33.47, p=0.05), constipation (2.45, 94% CI:1.01- 5.97, p=0.05), and fatigue (2.99, 1.06-8.44, p=0.05). Other minor side events, including, anemia (1.25, 95% CI: 0.39-4.07, p=0.71), metabolism and nutrition disorders (0.69, 95% CI: 0.20-2.43, p=0.56), nausea (2.00, 95% CI: 0.81-4.93, p=0.13), seizure (2.27, 94% CI: 0.79-6.56, p=0.13), vomiting (1.87,95% CI: 0.22-7.94, p=0.40), were non-significantly intimate to MSC treatment (Figure 2B).

Subgroup analysis of all studies

Subsequently, we dissected potential factors, including administration (method), age, methodology (analysis) of the article, cell type, population (disease), gender proportion, location, study phrase and publication date (year), influencing the major side events (Figure 3A). We identified that the non-significance of death, infection and diarrhea, which were not treatment-related side events of MSC therapy, were not altered in the slightest by any of the analyzed factors. MSC therapy was demonstrated to reduce the incidence of arrhythmia in the population with the age <60 years (p<0.01), PP analysis (p=0.01) and beyond 5 years (p<0.01). Despite the non-significant central nervous system disorders (head and dizziness) proved by pooled analysis, AD-MSC (p<0.01), placenta MSC (p<0.01) and uc-MSC (p<0.01) were more easily to cause headache and dizziness. Meanwhile, a population with degenerative joint diseases (p<0.01) and gastrointestinal diseases (p<0.01) could have headache and dizziness symptoms while receiving MSC implantation. Urticaria significantly occurred when the data were analyzed by PP analysis exclusively (p<0.01). As for vascular disorders, Asian people more easily had vascular disorders (p<0.01) after MSC treatment. Administration site conditions preferred to occur in populations with the age < 60 years (p=0.02), heart related diseases (p=0.01), the male proportion >60% (p=0.08), in study phrase 1/2 (p=0.01), and within 5 years (p=0.05). Even though transient fever was conspicuously associated with MSC treatment, populations with the age > 60 years (p=0.86), the male proportion < 60% (p=0.7), receiving local implantation (p=0.76), in North America (p=0.82) and study phrase 1 (p=0.15), had a lower risk of transient fever over the period MSC of therapy.
In terms of the minor side events, only five side events, including anemia, constipation, metabolism and nutrition disorders and nausea, were analyzed (Figure 3B). Similarly, the interactions between the 9 predicted factors and seldomly reported side events were dissected. Contrary to pooled analysis, neither constipation nor fatigue was a significant side event in these subgroup analyses. Similar to pooled analysis, both metabolism and nutrition disorders and nausea were non-impacted by these factors and were non-significant side events. Interestingly, we found that populations with the age < 60 years trended to have transient anemia (p=0.07) post-MSC treatment.

**Pooled analysis of high-quality studies**

After the elimination of low-quality articles (Kim 2018; Koh 2012; Lee 2017; Lin 2012; Oh 2018; Shi 2012; Sponer 2018; Wang 2006; Wang 2014; Wang 2016; Xie 2007; Zeng 2015; Xiao 2012; Skyler 2015), only seven major side events and one minor side event left (Figure 4). We merely found a close relationship between transient fever (3.08, 95% CI: 1.67-1.48, p=0.01) and MSC administration. Other side events, such as metabolism and nutrition disorders (0.49, 95% CI: 0.11-2.10, p=0.33), infection (1.05, 95% CI: 0.59-1.61, p=0.83), death (0.99, 95% CI: 0.66-1.48, p=0.96), arrhythmia (0.58, 95% CI: 0.33-1.03, p=0.06), central nervous system disorders (0.96, 95% CI: 0.49-1.88, p=0.91), vascular disorders (0.85, 95% CI: 0.30-2.45, p=0.77), and administration site conditions (2.15, 95% CI: 0.98-4.73, p=0.06) were not significantly impacted by MSC administration.

**Subgroup analysis of high-quality studies**

We examined whether the potential factors significantly influenced the terminal outcomes (7 major side events) reported by high-quality studies (Figure 5). MSC administration would not directly lead to death, death, central nervous disorders (headache and dizziness), or vascular disorders. Populations with the age <60 years (p<0.01), receiving BMSC injection (p=0.04), in study phrase 3 (p=0.04), and beyond 5 years (p<0.01) seemed to have a lower incidence of arrhythmia and benefit from MSC administration. When it came to transient fever, MSC administration would not trigger fever in populations with the age > 60 years (p=0.86), the male proportion <60% (p=0.70), from Europe (p=0.82), in study phrase 2 (p=0.15), beyond 5 years (p=0.11), and receiving local implantation (p=0.76).

**Sensitivity analysis**

Leave-one meta-analysis was performed for administration site conditions, arrhythmia, death, dermatitis, diarrhea, transient fever, infection, central nervous system disorders, and vascular disorders, and fatigue, metabolism and nutrition disorders, anemia, constipation, and nausea from all studies (Additional file 3-16), and for administration site conditions, arrhythmia, death, transient fever, infection, central nervous system disorders, and vascular disorders from high-quality studies (Additional file 17-23).

**Publication bias and article quality**

We assessed the article quality by using the Cochrane Collaboration’s tool for assessing the risk of bias (Figure 6). We concluded that most studies’ design was suitable and high quality. Only 14 studies were considered as low quality because they had more than two entries marked as high risk and less than four entries evaluated as low risk. There was performance bias, selection bias, detection bias, and attrition bias potentially lowering the integral quality of included studies. Furtherly, we tested the publication bias for administration site conditions, arrhythmia, death, dermatitis, diarrhea, transient fever, infection, central nervous system disorders, and vascular disorders (Additional file 24-32) from all studies. Publication bias for administration site conditions, arrhythmia, death, fever, infection, central nervous system disorders, and vascular disorders (Additional file 33-39) from high-quality studies were conducted as well.

**Discussion**

**Summary of evidence**

The association between the side events and MSC administration is first reported by M. Lalu et al.[1] and the association between MSC administration and the infusional toxicity, organ system complications, infection, death was not explored due to limited clinical researches. However, aside from these side events above, which were analyzed in this systematic review, more side events are described in recent trials with the expansion of population. In addition to transient fever, which is the most frequently reported by researchers, other side events such as constipation, fatigue, administration site conditions and sleeplessness can be induced by MSC administration as well. As for arrhythmia, MSC seems to benefit patients with cardiac diseases.

We were unable to discover the conspicuous association between MSC administration and the rest side events (vascular disorders, urticaria/dermatitis, dizziness/headache, diarrhea, infection, death, anemia, metabolism and nutrition disorders, nausea, seizure, and vomiting). Neither, there was direct proof suggesting that the MSC administration is tumorigenic. Up to date, the malignance of MSC was merely reported by Ning et al.[5] despite the potential of tumorigenesis of MSC.

After the elimination of the low-quality studies, eight side events were actually analyzed, including metabolism and nutrition disorders, infection, fever, and death, arrhythmia, dizziness/headache, vascular disorders, and administrations. Among these side events, transient fever was exclusively associated with MSC administration. Arrhythmia and administration site conditions trended to be significant after MSC administration. Other side events had no relevance to MSC administration.

Furtherly, we analyzed each side events in various sub-populations to dissect by which side event was determined. We discovered that age, gender proportion, location, and year, analysis, disease, study phrase, cell type, and administration method were the main factors impacting the final side event. Take the definite side event fever as an instance, the aged were not susceptible to MSC administration and this may be because of blunt reactions of the organism to acute
inflammation triggered by MSC\textsuperscript{[16]}. The female more easily suffers from transient fever and the estrogen level is under serious doubt\textsuperscript{[17]}. The population in North America less undergo transient fever compared to other regional populations and this may suggest the racial discrepancy of MSC administration.

**Strengths and weakness**

This meta-analysis removes studies of low-grade evidence (retrospective study, single-arm study, and case) and included 62 prospective studies. All results suggest the strong association between MSC administration and transient fever, and administration site conditions. Moreover, more side events that were not reported before (e.g., anemia constipation and vomiting) are gradually being discovered\textsuperscript{[18-20]}. Theoretically, the side events of MSC administration should be under stringent surveillance in case of occurrence of other side events that were not reported before along with the expansion of clinical trials. We also notice that the longest follow-up is 5 years, which may be a shorter time considering the fact that we are using cell products. We should be cautious that longer term events in the farther future possibly impend.

Our research has limitations. First, we synthesized the data across heterogeneous disease states. Despite subgroup analysis of disease, it was difficult to distinguish whether one side event was specifically disease-related owing to the limited number of studies. Second, some studies presented their data in the form of abstract prior to formal publication which may impose an unknown effect on the interpretation of the outcomes. These data are difficult for us to obtain because many ongoing trials are in the middle stage and the performers would not like to release these data. Third, several side events are merely comprehensive conceptions rather than specific clinic symptoms and we contend that it is important to record these obscure descriptions (e.g., metabolism and nutrition disorders and gastrointestinal dysfunction). Fourth, we are not informed of whether cell dose is closely associated with these side events as a result of the lack of dose-dependent trials. If possible, a Bayesian network meta-analysis should be conducted to explore the puzzlement. Finally, tumorigenesis, which theoretically exists in MSC therapy, is rarely reported by researchers. And this interesting point should raise our attention.

**Conclusions**

We summarized all side events potentially related to the application of MSC and no serious safety signals other than transient fever, administration site conditions, sleeplessness, and constipation were discovered. Many population characteristics, including age, analysis, cell type, disease, gender, location, study phrase, year, and administration method possibly impacted the occurrence of one side event. The safety of MSC administration should be under sustained observation despite the innovative therapy appears safe.

**Abbreviations**

OR, odds ratio; CI; confidential intervals; RCTs, randomized clinical trials; MSCs, mesenchymal stromal cells; PICOS, participants, interventions, comparison, outcome and study; NO., number; PRISMA, Systematic Review and Meta-Analysis; pp, per-protocol; ESC, embryonic stem cell; NSC, neuronal stem cell; h-IPS, human induced pluripotent stem cell; PSVT, paroxysmal supraventricular tachycardia; VT, ventricular tachycardia; COPD, chronic obstructive pulmonary disease; BMSC, bone marrow-derived human umbilical cord mesenchymal stromal cell; AD-MSC, adipose-derived human umbilical cord mesenchymal stromal cell; NA, not available.

**Declarations**

**Acknowledgement**

Not applicable.

**Authors' contributions**

Conceptualization, YC S and Y W; Methodology, Y W and HX Y; Investigation, Y W and HX Y; Software, Y W; Formal analysis, Y W and HX Y; Writing—original draft, Y W and YC S; Writing—review & editing, YC S; Funding acquisition, HX Y; Supervision, Y W and YC S. All authors read and approved the final manuscript.

**Funding**

Not applicable.

**Availability of data and materials**

Not applicable.

**Ethics approval and consent to participate**

Not applicable.

**Consent for publication**

The publication of this manuscript is approved by all authors.

**Competing interests**

No conflicts of interests are declared.
Consent to participate
Not applicable.

References

1. Friedenstein AJ, Chailakhyan RK, Latsnik NV, Panasyuk AF, Keiliss-Borok IV. Stromal cells responsible for transferring the microenvironment of the hemopoietic tissues. Cloning in vitro and retransplantation in vivo. Transplantation. 1974;17(4):331–40.

2. Lalu MM, Mazzarello S, Zlepign J, Dong YYR, Montroy J, McIntyre L, et al. Safety and Efficacy of Adult Stem Cell Therapy for Acute Myocardial Infarction and Ischemic Heart Failure (SafeCell Heart): A Systematic Review and Meta-Analysis. Stem Cells Transl Med. 2018;7(12):857–66.

3. Eom YW, Shim KY, Baik SK. Mesenchymal stem cell therapy for liver fibrosis. Korean J Intern Med. 2015;30(5):580–9.

4. Jin MC, Medress ZA, Azad TD, Doulames VM, Veeravagu A. Stem cell therapies for acute spinal cord injury in humans: a review. Neurosurg Focus. 2019;46(3):E10.

5. Cortez-Toledo E, Rose M, Agu E, Dahlenburg H, Yao W, Nolts JA, et al. Enhancing Retention of Human Bone Marrow Mesenchymal Stem Cells with Prosurvival Factors Promotes Angiogenesis in a Mouse Model of Limb Ischemia. Stem Cells Dev. 2019;28(2):114–9.

6. Liu F, Qiu H, Xue M, Zhang S, Zhang X, Xu J, et al. MSC-secreted TGF-β regulates lipopolysaccharide-stimulated macrophage M2-like polarization via the Akt/FoxO1 pathway. Stem Cell Res Ther. 2019;10(1):345.

7. Zhang S, Teo KYW, Chuah SJ, Lai RC, Lim SK, Toh WS. MSC exosomes alleviate temporomandibular joint osteoarthritis by attenuating inflammation and restoring matrix homeostasis. Biomaterials. 2019;20035–47.

8. Simon-Martinez C, Mailleux L, Ortibus E, Feihrenbach A, Sgandurra G, Cioni G, et al. Combining constraint-induced movement therapy and action-observation training in children with unilateral cerebral palsy: a randomized controlled trial. BMC Pediatr. 2018;18(1):250.

9. Vaquero J, Zurita M, Rico MA, Aguayo C, Bonilla C, Marin E, et al. Intrathecal administration of autologous mesenchymal stromal cells for spinal cord injury: Safety and efficacy of the 100/3 guideline. Cytotherapy. 2018;20(6):806–19.

10. Wang D, Li J, Zhang Y, Zhang M, Chen J, Li X, et al. Umbilical cord mesenchymal stem cell transplantation in active and refractory systemic lupus erythematosus: a multicenter clinical study. Arthritis Res Ther. 2014;16(2):R79.

11. Fukumitsu M, Suzuki K. Mesenchymal stem/stromal cell therapy for pulmonary arterial hypertension: Comprehensive review of preclinical studies. J Cardiol. 2019;74(4):304–12.

12. Li P, Gong Z, Shultz LD, Ren G. Mesenchymal stem cells: From regeneration to cancer. Pharmacol Ther. 2019;20042–54.

13. Le Blanc K, Davies LC. Mesenchymal stromal cells and the innate immune response. Immunol Lett. 2015;168(2):140–6.

14. Lalu MM, McIntyre L, Pugliese C, Fergusson D, Winston BW, Marshall JC, et al. Safety of cell therapy with mesenchymal stromal cells (SafeCell): a systematic review and meta-analysis of clinical trials. PLoS One. 2012;7(10):e47559.

15. Ning H, Yang F, Jiang M, Hu L, Feng K, Zhang J, et al. The correlation between cotransplantation of mesenchymal stem cells and higher recurrence rate in hematologic malignancy patients: outcome of a pilot clinical study. Leukemia. 2008;22(3):593–9.

16. Neves J, Sousa-Victor P. Regulation of inflammation as an anti-aging intervention. Febs j. 2020;287(1):43–52.

17. Coelho LCM, Cruz JV, Maba IK, Zampronio AR. Fever Induced by Zymosan A and Polyinosinic-Polycytidylic Acid in Female Rats: Influence of Sex Hormones and the Participation of Endothelin-1. Immunochemistry. 2021;44(1):321–33.

18. Zollino I, Campioni D, Sibilla MG, Tessari M, Malagoni AM, Zamboni P A phase II randomized clinical trial for the treatment of recalcitrant chronic leg ulcers using centrifuged adipose tissue containing progenitor cells. Cytotherapy. 2019;21(2):200–11.

19. Huang L, Zhang C, Gu J, Wu W, Shen Z, Zhou X, et al. A Randomized, Placebo-Controlled Trial of Human Umbilical Cord Blood Mesenchymal Stem Cell Infusion for Children With Cerebral Palsy. Cell Transplant. 2018;27(2):325–34.

20. Molendijk I, Bonsing BA, Roelofs H, Peeters KC, Wasser MN, Dijkstra G, et al. Allogeneic Bone Marrow-Derived Mesenchymal Stromal Cells Promote Healing of Refractory Perianal Fistulas in Patients With Crohn’s Disease. Gastroenterology. 2015;149(4):918 – 27.e6.

Tables

Table 1. Inclusion and exclusion principles.
### Principle

| Population | Inclusion criteria | Exclusion criteria |
|------------|--------------------|--------------------|
| Any populations including the healthy people and the diseased people | NA | |

| Intervention | Inclusion criteria | Exclusion criteria |
|--------------|--------------------|--------------------|
| Using MSC as treatment, regardless of the administration methods (e.g. local implantation and injection) and sources of MSC (e.g. from the adipose, bone marrow and gum) | Using NSC, ESC, olfactory neuron, schwann cell, h-IPS and stem cell from body fluids (e.g. saliva, urine, serum and tears) etc but MSC as interventions | |

| Comparison | Inclusion criteria | Exclusion criteria |
|------------|--------------------|--------------------|
| Placebo treatment, non-treatment or basic treatment both utilized in the control and the intervention groups | Merely using traditional treatment (surgery and drug) in the control group but not in the intervention group | |

| Outcome | Inclusion criteria | Exclusion criteria |
|---------|--------------------|--------------------|
| 1) Any side events associated with MSC treatment; 2) one side event reported by more than one study; 3) regardless of the efficacy of MSC therapy for any diseases | No side events reported | |

| Study | Inclusion criteria | Exclusion criteria |
|-------|--------------------|--------------------|
| 1) RCT; 2) prospective controlled study | 1) Case report (series); 2) single arm study; 3) retrospective controlled study; 4) cross controlled study; 5) study protocol | |

ESC, embryonic stem cell; MSC, mesenchymal stromal cells ; NSC, neuronal stem cell; h-IPS, human induced pluripotent stem cell; NA, not available.

**Table 2. Outcome definition.**

| Side event                 | Definition                                                                 |
|----------------------------|---------------------------------------------------------------------------|
| **Vascular disorders**     | 1) Vascular thrombosis including venous and arterial thrombosis;          |
|                            | 2) vasculitis                                                             |
| **Arrhythmia**             | 1) PSVT; 2) VT;                                                          |
|                            | 3) atrial fibrillation; 4) ventricular fibrillation                       |
| **Central nervous disorders** | 1) dizzy; 2) headache                                                      |
| **Diarrhea**               | Non-infectious diarrhea                                                   |
| **Infection**              | 1) non-injection site infection; 2) respiratory system infection;        |
|                            | 3) urinary system infection; 4) biliary tract; 5) digestive tract and spontaneous peritonitis |
| **Fever**                  | Transient fever (low-grade, 37.3-38°C) within 48 hours                   |
| **Administration site conditions** | 1) injection site bleeding; 2) injection site swelling;                    |
|                            | 3) injection site pain; 4) injection site itchy;                         |
|                            | 5) injection site infection                                               |
| **Anemia**                 | Defined by Hb<110g/L                                                       |
| **Metabolism and nutrition disorders** | Mainly refer to malnutrition                                             |

PSVT, paroxysmal supraventricular tachycardia; VT, ventricular tachycardia.

**Table 3. Study characteristics**
| Author   | Year | Study type | Location       | Disease                        | Cell   | Administration       | Analysis | Study type | Language | Dose                                |
|----------|------|------------|----------------|--------------------------------|--------|----------------------|----------|------------|----------|------------------------------------|
| Emadedin | 2018 | RCT        | Iran           | Knee osteoarthritis            | BMSC   | Local implantation   | pp       | 1/2        | English  | 40×10^6 cells                      |
| Gao      | 2013 | RCT        | China          | Acute myocardial infarction    | BMSC   | Intracoronary injection | PP       | 3          | English  | (3.08 ± 0.52) × 10^6 cell          |
| Gupta    | 2013 | RCT        | India          | Critical limb ischemia         | BMSC   | Local injection      | ITT      | 1/2        | English  | 2×10^6 cells/kg                    |
| Lee      | 2010 | RCT        | South Korea    | Ischemic stroke                | BMSC   | Intracoronary injection | PP       | 3          | English  | 1×10^8 cells                        |
| Zollino  | 2018 | RCT        | Italy          | Chronic leg ulcers             | BMSC   | Local injection      | PP       | 2          | English  | NA                                  |
| Weiss    | 2013 | RCT        | USA            | COPD                           | BMSC   | Intravenous injection | PP       | 3          | English  | 100×1 cells                         |
| Xiao     | 2017 | RCT        | China          | Dilated cardiomyopathy        | BMSC   | Intracoronary injection | PP       | 3          | English  | (4.9 ± 1×10^6) cells               |
| Hare     | 2009 | RCT        | USA            | Acute myocardial infarction    | BMSC   | Intravenous injection | PP       | 3          | English  | 0.5, 1.6, and 5 × 10^6 cells/kg     |
| Huang    | 2018 | RCT        | China          | Cerebral palsy                 | uc-MSC | Intravenous injection | PP       | 3          | English  | 5×10^7 cells                        |
| Centeno  | 2014 | RCT        | USA            | Knee osteoarthritis            | BMSC   | Local implantation   | PP       | 3          | English  | 75%, BMSC, 12.5% PRP, 12.5% PBS    |
| FernaÁndez | 2018 | RCT        | Spain          | Multiple sclerosis             | AD-MSC | Intravenous injection | ITT      | 1/2        | English  | 1×10^6 cells/kg, 4×10^6 cells/kg   |
| Vangsness| 2014 | RCT        | USA            | Partial medial meniscectomy    | BMSC   | Local implantation   | PP       | 3          | English  | 50×10 cells                         |
| Lin      | 2017 | RCT        | China          | Liver failure                  | BMSC   | Intravenous injection | PP       | 3          | English  | 1.0-10×10^5 cells/kg               |
| Molendijk| 2015 | RCT        | USA            | Crohn's disease                | BMSC   | Local implantation   | PP       | 3          | English  | 1.9×10^7 cells                     |
| Tompkins | 2017 | RCT        | USA            | Aging frailty                  | BMSC   | Intravenous injection | PP       | 2          | English  | 100-200×10 cells                   |
| Li       | 2013 | RCT        | China          | Leg ischemia                   | BMSC   | Local implantation   | PP       | 3          | English  | 1×10^7 cells/m                      |
| Lee      | 2007 | RCT        | South Korea    | Multiple system atrophy        | BMSC   | Intravenous injection | PP       | 3          | English  | 4×10^7 cells                        |
| Jaillard | 2020 | RCT        | France         | Ischemic stroke                | BMSC   | Intravenous injection | ITT      | 3          | English  | 100-300×10 cells                   |
| Mathiasen| 2019 | RCT        | Denmark        | Ischaemic heart failure        | BMSC   | Intramyocardial injection | ITT      | 3          | English  | 0.2 mL                              |
| Bartunek | 2016 | RCT        | USA            | Ischaemic heart failure        | BMSC   | Intramyocardial injection | ITT      | 3          | English  | 24×10^6 cells                      |
| Bartunek | 2013 | RCT        | Belgium        | Heart failure                  | BMSC   | Intramyocardial injection | PP       | 3          | English  | 600-12×10^6 cells                 |
| Powell   | 2012 | RCT        | USA            | Critical limb ischemia         | BMSC   | Local implantation   | PP       | 3          | English  | 0.5 ml                              |
| Olivera  | 2017 | RCT        | Brazil         | Pulmonary emphysema            | BMSC   | Intravenous injection | ITT      | 1          | English  | 10^6 cells                          |
| Study | Year | Design | Country | Condition | Cell Type | Treatment | No. | Cell Type | Unit | Location |
|-------|------|--------|---------|-----------|-----------|-----------|-----|-----------|------|----------|
| Sper  | 2019 | RCT    | China   | Knee OA   | BMSC      | Local implantation | PP  | 3         | English |
| Wang  | 2016 | RCT    | Russia  | Limb ischemia | BMSC | Local implantation | PP  | 3         | Chinese |
| Chuna | 2018 | RCT    | Korea   | Myocardial infarction | BMSC | Intravenous injection | PP  | 3         | English |
| Tan   | 2019 | RCT    | China   | Kidney transplants | BMSC | Intravenous injection | PP  | 3         | English |
| Hauze | 2017 | RCT    | Belgium | Osteonecrosis | BMSC | Local implantation | PP  | 3         | English |
| Norie | 2018 | RCT    | Spain   | Intervertebral disc regeneration | BMSC | Local implantation | PP  | 3         | English |
| Gao   | 2019 | RCT    | China   | Myocardial infarction | BMSC | Intracoronary injection | PP  | 3         | English |
| Asche | 2018 | RCT    | USA     | Left ventricular assist device patients | BMSC | Intramyocardial injection | PP  | 3         | English |
| Koh   | 2018 | RCT    | South Korea | Knee osteoarthritis | BMSC | Local implantation | PP  | 3         | English |
| Berry | 2019 | RCT    | USA     | Amyotrophic lateral sclerosis | BMSC | Local implantation | PP  | 2         | English |
| Chulli | 2014 | RCT    | India   | Acute myocardial infarction | BMSC | Intracoronary injection | PP  | 1/2       | English |
| Oh    | 2018 | RCT    | Republic of Korea | Amyotrophic lateral sclerosis | BMSC | Intramuscular injection | ITT | 3         | English |
| Hess  | 2017 | RCT    | USA     | Acute ischaemic stroke | BMSC | Intracerebral injection | ITT | 2         | English |
| Bartol | 2017 | RCT    | Chile   | Heart failure | BMSC | Intravenous injection | PP  | 1/2       | English |
| Wang  | 2017 | RCT    | Australia | Anterior cruciate ligament reconstruction patients | BMSC | Local implantation | ITT | 3         | English |
| Wang  | 2014 | RCT    | China   | Myocardial infarction | BMSC | Intracoronary injection | PP  | 3         | English |
| Gu    | 2020 | RCT    | China   | Cerebral palsy | BMSC | Intravenous injection | PP  | 3         | English |
| Name     | Year | Study Type | Country   | Condition                        | Cell Source               | Delivery Method           | Protocol | Language | Dose                  |
|----------|------|------------|-----------|----------------------------------|---------------------------|---------------------------|----------|----------|------------------------|
| Zhang    | 2016 | RCT        | China     | Liver transplantation            | uc-MSC                    | Intravenous infusion      | PP       | English | $1.0 \times 10^6$ cells/kg |
| Suk      | 2016 | RCT        | South Korea | Alcoholic cirrhosis               | BMSC                      | Hepatic arterial injection | PP       | English | $5 \times 10^7$ cells/ kg |
| Zheng    | 2014 | RCT        | China     | Acute respiratory distress syndrome | AD-MSC                    | Intravenous infusion      | PP       | English | $1 \times 10^6$ cells/ kg |
| Matas    | 2019 | RCT        | Chile     | Knee Osteoarthritis              | AD-MSC                    | Local implantation        | PP       | English | $2 \times 10^7$ cells/ kg |
| Wang     | 2006 | RCT        | China     | Dilated cardiomyopathy           | BMSC                      | Intracoronary injection   | ITT      | Chinese | $8 \times 10^7$ cells |
| Lin      | 2012 | RCT        | China     | Liver fibrosis                   | uc-MSC                    | Intravenous infusion      | ITT      | Chinese | $0.5-1 \times 10^7$ cells/ kg |
| Xie      | 2007 | RCT        | China     | Spinal cord injury               | BMSC                      | Intrathecal injection     | ITT      | Chinese | $20.56-58.87 \times 10^6$ cells |
| Xiao     | 2012 | RCT        | China     | Myocardial infarction            | BMSC                      | Intracoronary injection   | ITT      | Chinese | $1-10 \times 10^6$ cells/ kg |
| Erpicum  | 2018 | RCT        | Belgium   | Kidney transplantation           | BMSC                      | Intravenous injection     | PP       | English | $2 \times 10^6$ cells/ kg |
| Shadmafar| 2017 | Prospective study | China | Liver fibrosis                   | uc-MSC                    | Hepatic artery injection  | ITT      | English | $(42 \pm 4) \times 10^6$ cells |
| Zeng     | 2015 | RCT        | Iran      | Rheumatoid arthritis             | BMSC                      | Local implantation        | PP       | Chinese | $6.0-7.0 \times 10^7$ cells/ kg |
| Ning     | 2008 | RCT        | China     | Hematologic malignancy           | BMSC                      | Intravenous injection     | NA       | NA       | $1.0-2.0 \times 10^6$ cells/ kg |
| José     | 2020 | RCT        | Spain     | Knee osteoarthritis              | BMSC                      | Local implantation        | ITT      | English | $100 \times 10^6$ cells |

uc-MSC, human umbilical cord mesenchymal stromal cell; BMSC, bone marrow-derived human umbilical cord mesenchymal stromal cell; AD-MSC, adipose-derived human umbilical cord mesenchymal stromal cell; PP, per-protocol; ITT, intention to treat; RCT, randomized clinical trial; COPD, chronic obstructive pulmonary disease.

**Figures**
Figure 1

Article selection process

Figure 2

Bar plot for events of all articles. This figure depicted the significance of major events (A) and minor events (B) of all included articles. The OR value of each pooled event is presented as mean and 95% confidential intervals. The significance of each event is marked by different colors. The more the color approaches the bottom of the p value bar, the occurrence of the event is significant. Scarcely reported event (reported by single one article) was not collected and considered as minor event.
Figure 3

Bar plot for events of high-quality articles. This figure depicted the significance of major events (A) and minor events (B) of high-quality articles. The OR value of each pooled event is presented as mean and 95% confidence intervals. The significance of each event is marked by different colors. The more the color approaches the bottom of the p value bar, the occurrence of the event is significant. Scarcely reported event (reported by single one article) was not collected and considered as minor events.
Circular net-work map for events of all articles. This figure depicted potential factors impacting major events (A) and minor events (B) of included articles. Each line connecting 2 color blocks indicates a potential interaction between the side affair and the factor. If no interaction exists between the factor and the side affair, the value was denoted as 1 by default. The area of the connecting line is proportional to the value of 1 minus p. The larger of the connecting line area, the likely the side affair is to be impacted by the factor.

Figure 5

Circular net-work map for events of high qualities articles. This figure depicted potential factors impacting major events (A) and minor events (B) of high-quality articles. Each line connecting 2 color blocks indicates a potential interaction between the side affair and the factor. If no interaction exists between the factor and the side affair, the value was denoted as 1 by default. The area of the connecting line is proportional to the value of 1 minus p. The larger of the connecting line area, the likely the side affair is to be impacted by the factor.
Figure 6

Quality assessment of included articles. A. Quality assessment of each article. B. Pooled result of quality assessment.

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- Additionalfile1.searchstrategy.docx
- Additionalfile2PRISMA2009Checklist.docx
- additionalfile24.tif
- additionalfile25.tif
- additionalfile26.tif
- additionalfile27.tif
- additionalfile28.tif
- additionalfile29.tif
- additionalfile3.tif
- additionalfile30.tif
- additionalfile31.tif
- additionalfile32.tif
- additionalfile33.tif
- additionalfile34.tif
- additionalfile35.tif
- additionalfile36.tif
- additionalfile37.tif
