Cell therapy with intravascular administration of mesenchymal stromal cells continues to appear safe: An updated systematic review and meta-analysis

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Abstract

Background: Characterization of the mesenchymal stromal cell (MSC) safety profile is important as this novel therapy continues to be evaluated in clinical trials for various inflammatory conditions. Due to an increase in published randomized controlled trials (RCTs) from 2012–2019, we performed an updated systematic review to further characterize the MSC safety profile.

Methods: MEDLINE, EMBASE, Cochrane Central Register of Controlled Trials and Web of Science (to May 2018) were searched. RCTs that compared intravascular delivery of MSCs to controls in adult populations were included. Pre-specified adverse events were grouped according to: (1) immediate, (2) infection, (3) thrombotic/embolic, and (4) longer-term events (mortality, malignancy). Adverse events were pooled and meta-analyzed by fitting inverse-variance binary random effects models. Primary and secondary clinical efficacy endpoints were summarized descriptively.

Findings: 7473 citations were reviewed and 55 studies met inclusion criteria (n = 2696 patients). MSCs as compared to controls were associated with an increased risk of fever (Relative Risk (RR) = 2.48, 95% Confidence Interval (CI) = 1.27–4.86; I² = 0%), but not non-fever acute infusional toxicity, infection, thrombotic/embolic events, death, or malignancy (RR = 1.16, 0.99, 1.14, 0.78, 0.93; 95% CI = 0.70–1.91, 0.81–1.21, 0.67–1.95, 0.65–0.94, 0.60–1.45; I² = 0%, 0%, 0%, 0%, 0%). No included trials were ended prematurely due to safety concerns.

Interpretations: MSC therapy continues to exhibit a favourable safety profile. Future trials should continue to strengthen study rigor, reporting of MSC characterization, and adverse events.

Funding: Stem Cell Network, Ontario Institute for Regenerative Medicine and Ontario Research Fund

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1. Introduction

Mesenchymal stromal cells (mesenchymal stem cells; MSCs) are multipotent stem cells that can be isolated from many adult tissues (e.g. bone marrow, adipose tissue). First described in 1974 [1], they have recently received attention in a number of different clinical fields for their potential therapeutic effects. While often described as ‘adult stem cells’, MSCs have limited cellular differentiation ability as compared to other types of stem cells. Pre-clinical evidence suggests that MSCs exert their beneficial effects primarily through immunomodulatory and paracrine mechanisms. MSCs target sites of inflammation and secrete bioactive molecules [2] and there is a growing body of literature demonstrating the efficacy of MSC therapy in a
Research in context

Evidence before this study

Several small clinical trials have investigated the efficacy and safety of MSCs in diseases, including chronic heart failure, acute myocardial infarction, hematological malignancies, graft versus host disease and the acute respiratory distress syndrome, and found some benefit with MSC therapy compared to controls. A previous systematic review examined the safety of intravascular administration of MSC therapy in heterogeneous adult patient populations. The review included eight RCTs and identified fever as the only adverse event that was significantly associated with MSC therapy. Since that publication in 2012, several reviews of MSC efficacy and safety have included safety as part of the review objective. However, only one review included a detailed and systematic examination of the efficacy and safety of intravascular MSC administration that was limited to acute myocardial infarction and ischemic heart failure conditions and found no association between MSC therapy and adverse events. Another recent systematic review examined adverse events that occurred in RCTs and patients enrolled in MSC trials since that time, we decided to conduct and update our systematic review to further characterize the safety profile of MSC-based therapy and descriptively summarize primary and secondary efficacy outcomes in MSC RCTs.

2. Methods

The methods of this systematic review and meta-analysis are similar to our previously published review [8] with a few modifications; these are the addition of key words in our search strategy to capture placenta derived MSC trials, the inclusion of only randomized controlled trials, a focus on reporting adverse events that were pre-specified and that are potentially relevant to MSC administration, the addition of one additional pre-specified adverse event category (thrombotic and thromboembolic events) and one additional subgroup analysis comparing placental MSCs, documentation of all reported serious adverse events and their relatedness to study treatment (in the MSC or control group), pooling of pre-specified adverse event estimates according to relative risks and 95% confidence intervals, and a descriptive summary of primary and secondary efficacy outcomes in the included RCTs. This report follows the PRISMA guidelines (complete checklist can be found in Appendix 2) [9] and because our review is an update of a previously published review, no protocol was registered.

2.1. Search strategy and selection criteria

We conducted electronic searches of Ovid MEDLINE (1950 to April 2019), EMBASE (1980 to April 2019) and Cochrane Central Register of Controlled Trials (April 2019). Given the non-standard terminology associated with MSCs, a number of terms were used (Appendix I, search strategy). ClinicalTrials.gov was searched for ongoing or recently completed trials. Abstracts and proceedings from clinical conferences were identified and searched using Web of Science (April 2019). Bibliographies of retrieved articles and relevant reviews were manually searched. All searches were performed without any language restrictions; if included, any non-English studies were subsequently translated for data extraction.

We included RCTs that examined the intravascular (venous and arterial) administration of MSCs compared to a control group that did not receive MSCs in adult populations. We excluded studies that exclusively used non-intravascular routes of administration (e.g. injection into a joint), ex vivo differentiated MSCs, or MSCs co-administered with other experimental cells or treatments.

Study screening and selection, data extraction and risk of bias assessments were all performed in duplicate by three independent reviewers (DW, MT, ED) using standardized forms.

2.2. Data analysis

Data were extracted under the following subheadings using a standardized spreadsheet: RCT characteristics and patient populations, MSC preparation and administration, assessment of risk of bias, and primary (safety) and secondary (efficacy) outcome measures. We recorded primary and secondary efficacy endpoints as reported in the RCTs. We contacted authors via email correspondence when data relevant to our systematic review was not reported in the included studies.

Safety was examined according to pre-specified incident adverse events according to the following categories: (1) immediate events (i.e., fever and non-fever acute infusional toxicity that occurred within 24 h of study drug administration) that captured the potential for MSCs to embolize or cause hypersensitivity reactions, (2) infection events that occurred at any time post-infusion because MSCs are known to immune-modulate in pre-clinical models, (3) thrombotic

variety of pre-clinical models, including acute lung injury [3,4], sepsis [5] and acute myocardial infarction [6]. Indeed, evidence of the immune-modulatory ability of MSC therapy in pre-clinical models has led to interest in the possible therapeutic role for MSCs in a variety of acute and chronic inflammatory conditions.

To date, several small clinical trials have investigated the efficacy and safety of MSCs for a variety of conditions including chronic heart failure, acute myocardial infarction, hematological malignancies, graft versus host disease and acute respiratory distress syndrome. While the results of some trials suggest benefit, larger trials with clinically important endpoints are needed before more definitive conclusions can be drawn. Thus, as more and more patients are being asked to participate in the studies, the safety of MSC therapy is of increasing importance and any risk of adverse events could represent a significant barrier to their successful translation into clinical practice. These potential risks include neoplastic potential due to MSCs’ proliferative capacity, susceptibility to infection given their immunomodulatory effects, embolism of the cells, zoonoses associated with cell culture reagents, and acute or chronic immunogenicity of the cells themselves [7]. A previous systematic review published by our group in 2012 included eight randomized controlled trials (RCTs) (n = 369 patients) and identified fever as the only adverse event that was significantly associated with MSC therapy [8]. Given the increase in published RCTs and patients enrolled in MSC trials since that time, we decided to conduct and update our systematic review to further characterize the safety profile of MSC-based therapy and descriptively summarize primary and secondary efficacy outcomes in MSC RCTs.

Added value of this study

In our updated systematic review that now includes over 40 additional RCTs and over 2000 additional patients, aside from fever, we continue to detect no significant reported safety signals associated with MSC treatment.

Implications of all the available evidence

Our findings suggest that with the accumulation RCT evidence, the administration of MSCs continues to appear safe. The findings from our review should provide additional assurance to researchers, clinicians, health regulators and patients and families that, with this updated evidence, the administration of MSC therapy compared to controls was associated with delayed neurological events.

Our previous systematic review examined the safety of intravascular administration of MSC therapy in heterogeneous adult patient populations. The review included eight RCTs and identified fever as the only adverse event that was significantly associated with MSC therapy. Since that publication in 2012, several reviews of MSC efficacy and safety have included safety as part of the review objective. However, only one review included a detailed and systematic examination of the efficacy and safety of intravascular MSC administration that was limited to acute myocardial infarction and ischemic heart failure conditions and found no association between MSC therapy and adverse events. Thus, as more and more patients are being asked to participate in the studies, the safety of MSC therapy is of increasing importance and any risk of adverse events could represent a significant barrier to their successful translation into clinical practice. These potential risks include neoplastic potential due to MSCs’ proliferative capacity, susceptibility to infection given their immunomodulatory effects, embolism of the cells, zoonoses associated with cell culture reagents, and acute or chronic immunogenicity of the cells themselves [7]. A previous systematic review published by our group in 2012 included eight randomized controlled trials (RCTs) (n = 369 patients) and identified fever as the only adverse event that was significantly associated with MSC therapy [8]. Given the increase in published RCTs and patients enrolled in MSC trials since that time, we decided to conduct and update our systematic review to further characterize the safety profile of MSC-based therapy and descriptively summarize primary and secondary efficacy outcomes in MSC RCTs.
or thrombo-embolic events because MSCs can express or secrete tissue factor and other coagulation proteins [10–14] and therefore there is a theoretical risk of activation of coagulation and consequent adverse clinical events (i.e. deep venous thrombosis, pulmonary embolism, arterial thrombosis etc.), and (4) longer-term events including death and malignancy, the latter of which was captured due to the theoretical risk that MSCs could engrant.

Adverse event data were extracted based on the longest follow-up point. Adverse event data from RCTs with more than one MSC study arm (ex: dose escalation trial) were combined into one MSC study group. Meta-analyses for each pre-specified adverse event category was performed using OpenMetaAnalyst (for Windows 7). Data were analyzed using DerSimonian-Laird random effects models with a correction factor of 0.5 added to both arms for studies with 0 counts. Pooled events were described using Relative Risks (RR) and 95% confidence intervals (95% CI).

For all pre-specified adverse events we documented whether the events were reported as serious and if they were related to the study treatment (in either the MSC group or control group); we also captured other serious adverse events that were not pre-specified in our review and their reported relatedness to the study treatment. Finally, we captured the number of studies that were aborted pre-maturely due to safety concerns.

We used the CONSORT approach to harm reporting as a guide to capture the quality of adverse event reporting [15]. Specifically, we examined whether the reported approach to monitoring/recording adverse events (a priori plan to monitor events, types of events, frequency, and follow-up duration for events) were defined in the methods sections of the included studies.

Data related to MSC characterization as defined by the Dominici criteria were also recorded [16]. These included MSC cell source and origin, tri-lineage differentiation potential, cell surface markers, and cell morphology and adherence to plastic. We also described measures of MSC production (MSC viability, MSC potency, culture medium, and cryopreservation technique) because these measures could potentially impact both therapeutic efficacy and safety.

Heterogeneity between RCTs was evaluated using the I² as well as the P-value from X² test. Sub group analyses for each pre-specified adverse event category were planned according to the individual patient populations (cardiovascular, neurological, hematological/oncological, endocrine, renal, liver, respiratory, infectious, immune-deficient/inflammatory, other), MSC characteristics (type, origin, source), and MSC preparation (fresh versus cryopreserved, xenogeneic versus xeno-free culture media). No adjustments for multiple comparisons were made for these sub group analyses as they were considered exploratory. A post-hoc sensitivity analysis of the pre-specified adverse event pooled estimates that excluded studies published in abstract form only was also conducted to evaluate the robustness of the study findings. The secondary efficacy outcomes were not pooled but rather summarized descriptively in tabular form for the reader.

RCTs that met inclusion criteria were assessed for risk of bias according to the Cochrane Collaboration methods [17].

2.3. Role of funding source

The funders had no role in the analysis, interpretation of the study results, or drafting of the manuscript. The authors independently designed the study, collected data, had access to the raw data, did the statistical analysis, and were responsible for the decision to submit for publication.

3. Results

Our search identified 7473 unique titles and 55 RCTs met inclusion criteria (see Fig. 1 for PRISMA flow diagram). All 55 RCTs were included for review (n = 2696 patients) [18–66] (Table 1); six of the 55 RCTs were published in abstract form only [19,45,61,64,65,67]. Included RCTs were conducted in 12 different countries and 20 (36-4%) were multi-center [20–23,26,27,29,38,40,46,47,49,55,62–64,66,68–70]. Sample sizes ranged from nine to 135 patients (49 ± 31, mean ± standard deviation). The follow-up period ranged from one day to 60 months (14.2 ± 13.5, mean ± standard deviation). Thirteen (23%) reported funding from a for-profit manufacturer of MSCs (i.e. Osiris Therapeutics, Inc., FCB-Pharmancell Company Limited, Celgene Cellular Therapeutics, etc.) [20–23,27,29,49,52,55,56,65,69,70].

Patient populations were diverse and included cardiovascular (12 trials, n = 612 patients) [21,24–27,29,37,40,42,44,49,58], neurological (10 trials, n = 242 patients) [30–32,36,45,48,57,62,65,69,71], renal (three trials, n = 177 patients) [55,63,67], liver (seven trials, n = 404 patients) [35,43,47,53,54,59,66], respiratory (three trials, n = 134 patients) [18,23,68] and endocrine diseases (four trials, n = 169 patients) [22,28,39,50], hematological/oncological malignancies (five trials, n = 318 patients) [33,34,41,46,71], immune deficient or inflammatory conditions (nine trials, n = 544 patients) [20,38,51,52,60,61,64,70,72], general frailty (one trial, n = 30 patients) [56], and severe sepsis in severely neutropenic patients with hematologic malignancies (one trial, n = 30 patients) [19].

With respect to MSC preparation and administration, of the 55 included RCTs, 31 (56-4%) examined bone marrow [19,21–27,29–33,36,39,41–43,45,47,53–55,59,61,66,68,71], 16 (29-1%) umbilical cord [28,35,38,40,44,46–52,54,60,63,65,72], four (7-3%) adipose-derived MSCs [18,20,62,64], two (3-6%) placenta-derived cells [65,70]; and in two RCT (3-6%) the source of MSCs was unclear [37,67]. See Supplementary Table 1 for expanded detail. Twenty (36-4%) RCTs used autologous MSCs [24–26,29–32,36,37,39,42,43,45,47,57–59,61,62,66], 29 (52-7%) used third party unmatched allogeneic MSCs [18–23,27,28,35,38,40,44,46–56,60,63–65], and four (7-3%) used allogeneic MSCs from matched donors [33,34,41,67]. Two (3-6%) RCTs used placenta-derived mesenchymal-like cells [59,70] and one (1-8%) RCT used mesenchymal precursor cells (MPC) rather than MSCs [22]. Twelve (21-8%) RCTs cultured the MSCs in a xeno-free medium [21,27,28,39,41,49,50,52,55,58,59,71], whereas the remainder either used a xenogenic product (40-0%, n = 22) [18,22,23,25,26,29–35,42,44,46,47,53,56,57,60,62,66,68] or did not report the medium used (38-1%, n = 21) [19,20,24,36–38,40,43–45,48,51,54,61,63–65,67,68,70,72].

Fifteen (27-3%) RCTs cryopreserved MSCs prior to administration [19–23,27,30,39,52,53,55,56,60,69,70], 32 (58-2%) fresh used MSCs [18,24–26,28,29,31–35,38,40,42–44,46–51,54,57–59,62–64,66,68,72], two (3-6%) used both a fresh and cryopreserved product [41,71] and six (10-9%) it was unclear [36,37,45,61,65,67]. One trial that used both a fresh and cryopreserved product (1-8%) [71] and five of the 32 RCTs that used fresh MSCs (9-1%) used a cryopreserved cell product that was thawed and cultured prior to injection for a fresh cell product [18,47,49,68,72]. Of the 22 RCTs that reported cryopreserving their product, 14 (25-6%) used dimethyl sulfoxide as the cryoprotectant solution at a concentration of 10% or less [18,19,21–23,27,41,47,52,55,56,60,68,71]; the type cryoprotectant was unclear for the eight other RCTs (14-5%). Seven (12-7%) of the included RCTs reported all three Dominici criteria for MSC characterization [20,21,40,41,68,71,72]. Twenty-nine (52-7%) RCTs reported on cell viability [20–23,25,26,29–33,37,39,40,43–47–49,52,54–58,62,68,71,72] and eight (14-5%) reported on a measure of MSC potency [20,22,23,25,29,47,62,68].

A description and frequency of the pre-specified incident adverse events defined in our systematic review (infusional toxicity: fever and non-fever, infusion, thrombotic or thromboembolic events, death and malignancy) for each included RCT is provided in Supplementary Table 2 and a summary of pooled data presented as forest plots for each pre-specified adverse event category are summarized in Figs. 2A–F.

With respect to the occurrence immediate adverse events, a total of 19 RCTs (n = 880 patients) reported on fever infusional toxicity [20,31,32,35–38,43,47–51,57,59,60,63,71,72]. In the pooled analysis,
the risk of fever was significantly greater in the MSC group as compared to the control group (Relative Risk (RR) = 2.48, 95% Confidence Interval (CI) = 1.27–4.86, I² = 0%; see Fig. 2a). Pooled analysis of reported non-fever infusional toxicity events in a total of 32 RCTs (n = 1525 patients) however did not reveal any significant increase in risk for the MSC as compared to the control group (RR = 1.16, 95% CI = 0.70–1.91, I² = 0%; see Fig. 2a) [18, 21–23, 25–27, 29, 30, 33, 34, 37, 39, 40, 44, 46, 48–54, 56–60, 63, 68–70].

A total of 27 RCTs (n = 1315 patients) reported on infection [19–23, 27, 30–34, 38–40, 47, 50, 52–57, 62, 63, 69, 70, 72]. In the pooled analysis, there was no significant increase in the risk of infection for the MSC as compared to the control group (RR = 0.99, 95% CI = 0.81–1.21, I² = 0%; see Fig. 2c).

The occurrence of thrombotic or thrombo-embolic events were reported in a total of 24 RCTs (n = 1112 patients) [20, 21, 26, 29, 32, 33, 37, 40, 50, 56, 58, 60, 63, 66, 69, 70]. In the pooled analysis there was no significant increase in the risk of thrombotic/thrombo-embolic events for MSCs as compared to the control group (RR = 1.14, 95% CI = 0.67–1.95, I² = 0%; see Fig. 2d).

A total of 40 (n = 1991 patients) and 19 (n = 1015 patients) RCTs reported on death [18–21, 23–27, 29, 31–37, 40–42, 44–46, 48–53, 55, 56, 58, 59, 62, 63, 66, 68–71] and malignancy or and ectopic tissue formation respectively [20, 27, 31–34, 38–41, 44, 47, 49, 53, 55–57, 70, 71]. In the pooled analysis, the risk of death was significantly lower for the MSC group as compared to the control group (RR = 0.78, 95% CI = 0.65–0.94, I² = 0%; see Fig. 2e). There was no significant increase found in the risk of malignancy or ectopic tissue formation for the MSC as compared to the control group (RR = 0.93, 95% CI = 0.60–1.45, I² = 0%; see Fig. 2f).

The results of the risk of bias assessment found that only six (10%) RCTs fulfilled all six criteria for low risk of bias (Table 2) [23, 27, 30, 40, 49, 62]. Nine (16%) RCTs met five of six primary criteria [21, 28, 32, 46, 50, 52, 56, 68, 69]. The allocation lists were concealed in 24 (43%) [21–23, 27, 28–30–32, 39, 40, 46, 47, 49, 50, 52, 53, 55, 56, 59, 60, 62, 63, 68, 69]; 21 (38%) were double blinded [18, 21, 23, 27, 28, 30, 40, 46, 49–52, 55–57, 62, 64, 65, 68–70] and three (5%) had an open label intervention but blinded outcome measures [29, 32, 54]. In terms of other potential sources of bias, 35 (63%) of the RCTs were registered with either clinicaltrials.gov or their own regional registration program [18–23, 26, 29, 30, 35, 38–41, 43, 46–49, 51–57, 60, 62–66, 68, 70, 72]. Thirty-eight (69%) RCTs did not report an a priori sample size calculation or provide a rationale for the sample size [18, 19, 21, 24–28, 31–39, 41, 42, 44, 45, 48–52, 54, 57–59, 61, 64–67, 69, 71, 72].

Sub-groups were meta-analyzed for the six pre-specified adverse event outcome categories and are summarized in Supplementary Table 3. Briefly, the risk of fever related acute infusional toxicity in the MSC group was increased in the neurological and immune/inflammatory populations, when unmatched allogeneic and autologous, bone marrow, umbilical, or fresh MSCs were administered, and when the MSC culture medium was xenogenic or unclear. The risk of
| Source               | Country | Patient Population (Sample Size) | Source Country | Patient Population (Sample Size) |
|----------------------|---------|----------------------------------|----------------|----------------------------------|
| **Cardiovascular**   |         |                                  |                |                                  |
| Chen et al., 2004    | PRC     | Acute myocardial infarction (60) | Chen et al.,   | Ischemic heart failure (45)      |
|                     |         |                                  | 2006           |                                  |
| Follow Up Duration (months) | Single-center | 6 | Autologous BM-MSCs | Saline, IC | 34 (94) | 35 (97) | 58 ± 7 | 57 ± 5 |
|                      |         |                                  | Gao et al.,    | Acute ST-elevation myocardial infarction (43) | Multi-center (4) | 24 | Unmatched allogeneic UC-MSCs | Saline (Plasma-lyte A), IV | 38 (100) | 39 (95) | 62 ± 7 | 62 ± 6 |
|                      |         |                                  | Gao et al.,    | Acute ST-elevation myocardial infarction (53) | Multi-center (10) | 6 | Autologous BM-MSCs | Standard treatment | 30 (90) | 31 (86) | 59 ± 7 | 59 ± 6 |
|                      |         |                                  | Hare et al.,   | Acute myocardial infarction (53) | Multi-center (10) | 6 | Autologous BM-MSCs | Routine therapy | 21 (100) | 22 (96) | 61 ± 7 | 61 ± 6 |
|                      |         |                                  | Lee et al.,    | Acute myocardial infarction (58) | SR | 12 | Autologous BM-MSCs | Rehabilitation alone | 16 (50) | 17 (62) | 64 ± 9 | 64 ± 8 |
|                      |         |                                  | Zhao et al.,   | Acute myocardial infarction (58) | Single-center | 6 | Autologous BM-MSCs | Rehabilitation alone | 11 (81) | 12 (77) | 57 ± 3 | 57 ± 4 |
|                      |         |                                  | Bartolucci et al., 2016/2017 | Stable heart failure (30) | Multi-center (2) | 12 | Unmatched allogeneic UC-MSCs | Placebo | 15 (80) | 16 (93) | 57 ± 1 | 57 ± 2 |
|                      |         |                                  | Xiao et al.,   | Dilated cardiomyopathy (37) | Single-center | 12 | Autologous BM-MSCs | Placebo (saline) | 17 (70) | 18 (70) | 51 ± 6 | 51 ± 10 |
| **Neurological**     |         |                                  | Ibrahim et al., 2016* | Malaysia | Acute middle cerebral artery stroke (17) | NR | 12 | Autologous BM-MSCs | Standard treatment | NR | NR | NR | NR |
|                      |         |                                  | Lee et al., 2008 | Multiple system atrophy (28) | Single-center | 12 | Autologous BM-MSCs | Standard treatment | 11 (73) | 12 (67) | 57 ± 7 | 57 ± 7 |
|                      |         |                                  | Lee et al., 2010 | Ischemic stroke (52) | Single-center | 12 | Autologous BM-MSCs | Rehabilitation alone | 16 (50) | 17 (62) | 64 ± 12 | 65 ± 12 |
|                      |         |                                  | Lee et al., 2012 | Multiple system atrophy (31) | Single-center | 12 | Autologous BM-MSCs | Rehabilitation alone | 11 (81) | 12 (77) | 57 ± 3 | 57 ± 4 |
|                      |         |                                  | Xie et al., 2007* | Spinal cord injury (24) | Single-center | 3 | Autologous BM-MSCs | Rehabilitation alone | 11 (81) | 12 (77) | 57 ± 3 | 57 ± 4 |
|                      |         |                                  | Xie et al., 2016 | Encephalopathy (22) | Single-center | 6 | Autologous BM-MSCs | Rehabilitation alone | 11 (81) | 12 (77) | 57 ± 3 | 57 ± 4 |
|                      |         |                                  | Fernandez et al., 2018 | Secondary progressive multiple sclerosis (30) | Multi-center (2) | 12 | Autologous Adipose-MSCs | Placebo | 10 (40) | 11 (27) | 48 ± 8 | 47 ± 5 |
|                      |         |                                  | Tsang et al., 2017 | Chronic stroke/vegetative state (9) | Single-center | Day of or day after | Autologous BM-MSCs | Placebo (Ringer’s lactate) | 9 (22) | 10 (75) | 47 ± 9 | 47 ± 1 |
|                      |         |                                  | Kim et al., 2018* | Cerebral infarction (12) | Single-center | 6 | Unmatched allogeneic UC-MSCs | Placebo | 8 (NR) | 9 (NR) | NR | NR |
|                      |         |                                  | Lubin et al., 2014 | Multiple sclerosis (16) | Multi-center (8) | 12 | Placenta-derived mesenchymal-like cells | Placebo | 6 (33) | 7 (40) | 52 ± 5 | 52 ± 10 |
|                      |         |                                  |                     |                     |                     |                     |                     |                     |                     |                     |                     |                     |
| (continued on next page) |         |                                  |                     |                     |                     |                     |                     |                     |                     |                     |                     |                     |
Table 1 (Continued)

| Source                          | Country | Patient Population (Sample Size) | Single-center vs multi-center (Number of centers) | Follow Up Duration (months) | Intervention | Control Comparison | Patients Evaluated (n (% male)) | Age (years ± SD) |
|--------------------------------|---------|---------------------------------|-------------------------------------------------|-----------------------------|--------------|--------------------|--------------------------------|------------------|
| **Oncological/Hematological**  |         |                                 |                                                 |                             |              |                    | T C T C                         |                  |
| Gao et al., 2016               | PRC     | Stem cell transplantation for hematologic malignancy (124) | Multi-center (5)                                | 51 (24–70)                  | Unmatched allogeneic UC-MSCs | Saline, IV                      | 62 (47) 62 (48) NR NR            |                  |
| Liu et al., 2011               | PRC     | Stem cell transplantation for leukemia (55)                     | Single-center                                   | 24                           | Matched allogeneic BM-MSCs  | Stem cell transplant alone       | 27 (74) 28 (68) 30 (14–46) 31.5 (12–48) |                  |
| Ning et al., 2008              | PRC     | Stem cell transplantation for hematologic malignancy (25)      | Single-center                                   | 36                           | Matched BM-MSCs            | Stem cell transplant alone       | 10 (90) 15 (87) 36 ± 11 39 ± 12 |                  |
| Kuzmina et al., 2012           | Russia  | Recipients of allogeneic bone marrow transplants for hematologic malignancies (37) | Single-center                                   | 32                           | Unmatched BM-MSCs          | Standard aGVHD prophylaxis       | 19 (42) 18 (39) 34 (20–63) 29 (19–60) |                  |
| Shipounova et al., 2014        | Russia  | Recipients of allogeneic bone marrow transplants for hematologic malignancies (77) | Single-center                                   | 60                           | Matched BM-MSCs            | Standard aGVHD prophylaxis       | 39 (NR) 38 (NR) NR NR            |                  |
| **Endocrine**                  |         |                                 |                                                 |                             |              |                    | T C T C                         |                  |
| Carlsson et al., 2015          | Sweden  | Type 1 diabetes mellitus (18) | Single-center                                   | 12                           | Autologous BM-MSCs         | Insulin-only treatment           | 9 (89) 14 (57) 24 ± 2 17.6 ± 8.7 27 ± 2 |                  |
| Hu et al., 2013                | PRC     | Type 1 diabetes mellitus (29) | Single-center                                   | 24                           | Unmatched allogeneic UC-MSCs | Saline, IV                      | 9 (60) 14 (57) 19 ± 2 17 ± 8.7 18 ± 2 |                  |
| Hu et al., 2016                | PRC     | Type 2 diabetes mellitus (61) | Single-center                                   | 36                           | Unmatched allogeneic UC-MSCs | Saline, IV                      | 31 (55) 30 (53) 52.4 ± 4.8 53.21 ± 8.22 |                  |
| Skyler et al., 2015            | USA     | Type 2 diabetes mellitus (61) | Multi-center (18)                                | 3                            | Unmatched allogeneic BM-MPCs | Saline, IV                      | 15 (67) 16 (75) 57 ± 8.2 55.3 ± 11.4 57.2 ± 6.6 58.7 ± 7.3 |                  |
| **Renal disease**              |         |                                 |                                                 |                             |              |                    | T C T C                         |                  |
| Swaminathan et al., 2018       | USA     | Patients undergoing cardiac surgery using cardiopulmonary bypass, who developed acute kidney insufficiency (135) | Multi-center (27)                                | 3                            | Unmatched allogeneic BM-MSCs | Placebo                         | 67 (65.7) 68 (82.4) 65.6 ± 11.9 67.0 ± 9.9 |                  |
| Korotkov et al., 2018*         | Belarus | Renal transplantation (NR)   | Single-center                                   | 7 days                        | Matched allogeneic UC-MSCs | Standard treatment              | NR NR NR NR NR |                  |
| Sun et al., 2018               | PRC     | Renal allograft (42)          | Multi-center (3)                                 | 12                           | Unmatched allogeneic UC-MSCs | Standard treatment              | 21 (67) 21 (52) 40.8 ± 9.2 47.1 ± 10.2 |                  |
| **Liver disease**              |         |                                 |                                                 |                             |              |                    | T C T C                         |                  |
| Suk et al., 2016               | ROK     | Alcohol-related liver cirrhosis (58)                             | Multi-center (12)                                | 12                           | Autologous BM-MSCs         | Standard treatment              | 21 (83)*** 24 (94)*** 53.1 ± 8.7 *** 53.7 ± 8.2 *** |                  |
| Shi et al., 2012               | PRC     | Acute-on-chronic liver failure (43)                               | Single-center                                   | 18                           | Unmatched allogeneic UC-MSCs | Placebo (saline)                | 23 (89)*** 24 (83) 19 (79) 40 (24–59) 45 (26–62) |                  |
| Salama et al., 2014            | Egypt   | Post-HCV end-stage liver disease (40)                            | Multi-center (2)                                 | 6                            | Autologous BM-MSCs         | Antiviral therapy (no hepatic artery infusion) | 20 (85) 20 (80) 50.27 ± 6.05 50.90 ± 7.23 |                  |
| Xu et al., 2014                | PRC     | Hepatitis B virus-related liver cirrhosis (56)                   | Single-center                                   | 6                            | Autologous BM-MSCs         | Standard care                   | 27 (65)*** 29 (58)*** 44 ± 12 *** 45 ± 10 *** |                  |
| Lin et al., 2017               | PRC     | Hepatitis B virus-related acute-on-chronic liver failure (110)   | Single-center                                   | 6                            | Unmatched allogeneic BM-MSCs | Standard treatment              | 56 (91.1) 54 (98.2) 40 ± 9.9 42.8 ± 8.4 |                  |
| Zhang et al., 2017             | PRC     | Liver fibrosis induced by hepatolenticular degeneration (60)     | Single-center                                   | 3                            | Autologous BM-MSCs         | Standard treatment              | 30 (53.3) 30 (56.7) 30.98 ± 11.25 32.1 ± 10.36 |                  |
| Shi et al., 2017               | PRC     | First cadaveric liver transplantation (27)                      | Single-center                                   | 6                            | Unmatched allogeneic UC-MSCs | Standard treatment              | 14 (92.9) 13 (92.3) 57 ± 12 55 ± 11 |                  |

(continued on next page)
| Source                  | Country | Patient Population (Sample Size) | Single-center vs multi-center (Number of centers) | Follow Up Duration (months) | Intervention | Control Comparison | Patients Evaluated (n (% male)) | Age (years ± SD) |
|------------------------|---------|---------------------------------|--------------------------------------------------|-----------------------------|--------------|-------------------|-------------------------------|----------------|
| Weiss et al., 2013     | USA     | Moderate to severe chronic obstructive pulmonary disease (62) | Multi-center (6) | 24 | Unmatched allogeneic BM-MSCs | Vehicle solution, IV | 30 (60) | 32 (56) | 68.1 ± 7.54 | 64.1 ± 8.76 |
| Zheng et al., 2014     | PRC     | Acute respiratory distress syndrome (12) | Single-center | 1 | Unmatched allogeneic adipose-MSCs | Saline, IV | 6 (100) | 6 (83) | 66.7 ± 20.4 | 69.8 ± 9.1 |
| Matthay et al., 2018   | USA     | Acute respiratory distress syndrome (60) | Multi-center (5) | 2 | Unmatched allogeneic BM-MSCs | Plasma-Lyte A, IV | 40 (58) | 20 (30) | 55 (17) | 55 (20) |
| Galstian et al., 2015/2016* | Russia | Patients with severe neutropenia and severe sepsis (30) | Single-center | 3 | Unmatched allogeneic BM-MSCs | Standard treatment | 15 (43) | 15 (54) | 48 (30–75) | 55 (33–81) |
| Alvaro-Garcia et al., 2016 | Spain | Refractory rheumatoid arthritis (53) | Multi-center (18) | 6 | Unmatched allogeneic adipose-MSCs | Lactated Ringer’s solution, IV | 20 (10) | 20 (10) | 54.15 ± 7.79 | 57.40 ± 11.01 |
| Zhang et al., 2013     | PRC     | HIV-1 infection (13) | Multi-center (NR) | 12 | Unmatched allogeneic UC-MSCs | Saline, IV | 6 (0) | 7 (71) | 30 (26–49) | 38 (19–55) |
| Hu et al., 2016        | PRC     | Ulcerative colitis (70) | Single-center | 24 | Unmatched allogeneic UC-MSCs | Saline, IV and IA | 34 (62) | 36 (61) | 42.9 ± 23.1 | 43.7 ± 28.7 |
| Deng et al., 2017      | PRC     | Systemic lupus erythematosus (18) | Single-center | 12 | Unmatched allogeneic UC-MSCs | Placebo | 12 (8) | 6 (0) | 29 ± 10 | 29 ± 7 |
| Zhang et al., 2018     | PRC     | Crohn’s disease (82) | Single-center | 12 | Unmatched allogeneic UC-MSCs | Standard treatment | 41 (58.5) | 41 (63.4) | 34.3 (21–44) | 32.7 (20–41) |
| Arturo et al., 2017*   | Columbia | Crohn’s disease (26) | Single-center | 6 | Unmatched allogeneic UC-MSCs | Autologous BM-MSCs | Standard treatment | NR | NR | |
| Panes et al., 2017*    | Spain   | Crohn’s disease (131) | Multi-center (NR) | 12 | Unmatched allogeneic UC-MSCs | Placebo | 70 (NR) | 61 (NR) | NR | NR |
| Yang et al., 2018      | PRC     | Rheumatoid arthritis (105) | Single-center | 11 | Unmatched allogeneic UC-MSCs | Placebo | 28 (25) | 53 (19) | 50.7 | 49.8 |
| Melmed et al., 2015    | USA     | Crohn's disease (46) | Multi-center (NR) | 24 | Placenta-derived mesenchymal-like cells | Placebo | 24 (21) | 8 (53) | 51.2 | 35.3 ± 14.0 |
| Tompkins et al., 2017  | USA     | Frailty (30) | Single-center | 12 | Unmatched allogeneic BM-MSCs | Placebo | 10 (60) | 10 (60) | 75.0 ± 7.4 | 75.3 ± 6.8 |

PRC= People’s Republic of China; USA= United States of America; ROK= Republic of Korea; T= treatment; C= control; BM= bone marrow; UC= Umbilical cord; MSCs= mesenchymal stromal cells; MPCs= mesenchymal precursor cells; IA= intra-arterial; IC= intracoronary; IV= intravenous; NR= not reported.
* Abstract form only.
** Foreign language text only.
*** Age and gender demographic data reported only for patients that completed trial follow-up.
**** Age and gender demographic data reported for all patients randomized, not necessarily infused with study treatment.
***** Age and gender demographic data reported only in earlier abstract, not updated to current abstract.
### Studies and Estimates

| Studies          | Estimate (95% C.I.)  | MSCs | Control |
|------------------|----------------------|------|---------|
| Lee 2010         | 6.53 (0.28, 152.17)  | 1/16 | 0/36    |
| Lee 2008         | 20.58 (1.27, 333.17) | 6/11 | 0/18    |
| Shi 2012         | 4.00 (0.20, 78.66)   | 2/24 | 0/19    |
| Wang 2006        | 1.00 (0.02, 46.70)   | 0/12 | 0/12    |
| Xie 2007         | 17.50 (1.11, 275.65) | 7/11 | 0/13    |
| Zhang 2013       | 2.63 (0.13, 54.64)   | 1/7  | 0/6     |
| Xu 2014          | 9.64 (0.54, 171.09)  | 4/27 | 0/29    |
| Aviles-Garcia 2016 | 3.03 (0.21, 50.25)  | 9/46 | 0/7     |
| Suk 2016         | 5.00 (0.28, 89.12)   | 4/44 | 0/24    |
| Hu 2016a         | 1.06 (0.02, 51.84)   | 0/34 | 0/36    |
| Hu 2016b         | 0.97 (0.02, 47.32)   | 0/31 | 0/30    |
| Xie 2016         | 0.85 (0.02, 39.24)   | 0/12 | 0/10    |
| Tsang 2017       | 1.00 (0.02, 46.70)   | 0/5  | 0/4     |
| Zhang 2017       | 0.60 (0.16, 2.29)    | 3/30 | 5/30    |
| Zhang 2018       | 9.00 (0.50, 161.98)  | 4/41 | 0/41    |
| Bartolucci 2016/2017 | 1.00 (0.02, 47.38)  | 0/15 | 0/15    |
| Sun 2018         | 1.00 (0.02, 48.19)   | 0/21 | 0/21    |
| Yang 2018        | 7.13 (0.38, 134.75)  | 3/62 | 0/53    |
| Overall (I^2=0% , P=0.74) | 2.48 (1.27, 4.86)    | 44/439 | 5/404 |

### Figures

**Fig. 2.** a) Infusional toxicity: Fever. b) Infusional toxicity: Non-fever. c) Infection. d) Thrombotic/thrombo-embolic events. e) Mortality. f) Malignancy or ectopic tissue formation.

RR, relative risk; CI, confidence interval; MSC, mesenchymal stem cell.
Studies | Estimate (95% C.I.) | MSCs | Controls
--- | --- | --- | ---
Hare 2009 | 1.23 (0.50, 3.01) | 11/34 | 5/19
Lee 2008 | 1.58 (0.03, 74.61) | 0/11 | 0/18
Lee 2010 | 0.75 (0.23, 2.41) | 3/16 | 9/36
Lee 2012 | 0.81 (0.16, 4.19) | 2/14 | 3/17
Liu 2011 | 1.04 (0.71, 1.52) | 18/27 | 18/28
Ning 2008 | 1.20 (0.42, 3.41) | 4/10 | 5/15
Weiss 2013 | 1.23 (0.71, 2.14) | 15/30 | 13/32
Zhang 2013 | 0.87 (0.02, 38.59) | 0/7 | 0/6
Gao 2015 | 3.00 (0.12, 72.15) | 1/10 | 0/10
Chullikana 2015 | 3.00 (0.14, 65.90) | 1/10 | 0/10
Carlsson 2015 | 1.00 (0.02, 45.63) | 0/9 | 0/9
Suk 2016 | 2.45 (0.58, 10.46) | 9/44 | 2/24
Hu 2016b | 0.97 (0.02, 47.32) | 0/31 | 0/30
Galellan 2015/2016 | 1.38 (0.78, 2.41) | 11/15 | 8/15
Deng 2017 | 2.69 (0.15, 48.64) | 2/12 | 0/6
Lin 2017 | 0.56 (0.33, 0.97) | 14/56 | 24/54
Shi 2017 | 0.19 (0.01, 3.56) | 0/14 | 2/13
Swaminathan 2018 | 1.25 (0.61, 3.00) | 12/67 | 9/68
Tompkins 2017 | 0.17 (0.01, 3.94) | 0/20 | 1/20
Tsang 2017 | 0.83 (0.02, 34.94) | 0/5 | 0/4
Fernandez 2018 | 0.39 (0.14, 1.07) | 4/19 | 6/11
Sun 2018 | 0.50 (0.14, 1.74) | 3/21 | 6/21
Yang 2018 | 1.02 (0.02, 50.41) | 0/52 | 0/53

Overall (I^2=0%, P=0.75) | 0.99 (0.81, 1.21) | 110/582 | 111/557

Studies | Estimate (95% C.I.) | MSCs | Controls
--- | --- | --- | ---
Gao 2013 | 5.00 (0.25, 98.27) | 2/21 | 0/21
Lee 2008 | 1.58 (0.03, 74.61) | 0/11 | 0/18
Lee 2010 | 3.00 (0.76, 11.88) | 4/16 | 3/36
Lee 2012 | 1.01 (0.39, 2.62) | 5/14 | 6/17
Lee 2014 | 0.94 (0.02, 45.63) | 0/30 | 0/28
Liu 2011 | 1.04 (0.02, 50.42) | 0/27 | 0/28
Ning 2008 | 4.36 (0.20, 97.56) | 1/10 | 0/15
Wang 2006 | 1.00 (0.02, 46.70) | 0/12 | 0/12
Gao 2015 | 1.00 (0.06, 15.61) | 1/58 | 1/58
Chullikana 2015 | 0.33 (0.02, 7.32) | 0/10 | 1/10
Salama 2014 | 1.00 (0.02, 48.09) | 0/20 | 0/20
Alvaro-Garcia 2016 | 0.51 (0.02, 11.47) | 1/46 | 0/7
Skyler 2015 | 1.11 (0.05, 25.92) | 1/45 | 0/16
Hu 2016b | 0.97 (0.02, 47.32) | 0/31 | 0/30
Tsang 2017 | 0.83 (0.02, 34.94) | 0/5 | 0/4
Tompkins 2017 | 0.52 (0.01, 24.65) | 0/20 | 0/10
Xiao 2017 | 1.17 (0.02, 55.88) | 0/17 | 0/20
Swaminathan 2018 | 3.04 (0.13, 73.42) | 1/67 | 0/68
Zhang 2018 | 1.00 (0.02, 49.23) | 0/41 | 0/41
Sun 2018 | 0.33 (0.01, 7.74) | 0/21 | 1/21
Bartolucci 2016/2017 | 0.33 (0.01, 7.58) | 0/15 | 1/15
Deng 2017 | 0.18 (0.01, 3.85) | 0/12 | 1/6
Lublin 2014 | 1.15 (0.06, 23.88) | 1/12 | 0/4

Overall (I^2=0%, P=1.00) | 1.14 (0.67, 1.95) | 17/561 | 14/505

Fig. 2. Continued
### Table: Estimate (95% C.I.)

| Studies               | Estimate (95% C.I.) | MSCs    | Control |
|-----------------------|---------------------|---------|---------|
| Chen 2004             | 1.03 (0.02, 50.42)  | 0/34    | 0/35    |
| Chen 2006             | 0.52 (0.11, 2.57)   | 2/22    | 4/23    |
| Gao 2013              | 3.00 (0.13, 69.70)  | 1/01    | 0/21    |
| Hare 2009             | 0.57 (0.01, 27.73)  | 0/34    | 0/19    |
| Lee 2010              | 0.43 (0.18, 1.05)   | 4/16    | 21/36   |
| Lee 2008              | 1.58 (0.03, 74.61)  | 0/11    | 0/18    |
| Lee 2014              | 0.94 (0.02, 45.62)  | 0/30    | 0/28    |
| Liu 2011              | 0.92 (0.42, 2.04)   | 8/27    | 9/28    |
| Ning 2008             | 1.80 (0.75, 4.32)   | 6/10    | 5/15    |
| Shi 2012              | 0.36 (0.15, 0.86)   | 5/24    | 11/19   |
| Wang 2006             | 0.50 (0.05, 4.81)   | 1/12    | 2/12    |
| Weiss 2013            | 1.60 (0.29, 8.92)   | 3/30    | 2/32    |
| Xie 2007              | 1.17 (0.02, 84.46)  | 0/11    | 0/13    |
| Kuzmina 2012          | 0.24 (0.03, 1.92)   | 1/19    | 4/18    |
| Gao 2015              | 0.33 (0.01, 8.02)   | 0/58    | 1/58    |
| Chuklikova 2015       | 0.33 (0.02, 7.32)   | 0/10    | 1/10    |
| Shipounova 2014       | 0.55 (0.28, 1.09)   | 9/39    | 16/38   |
| Wang 2014             | 0.54 (0.05, 5.59)   | 1/28    | 2/30    |
| Salama 2014           | 0.09 (0.01, 1.54)   | 0/20    | 5/20    |
| Zhao 2015             | 0.28 (0.06, 1.22)   | 2/30    | 7/29    |
| Zheng 2014            | 0.50 (0.06, 4.15)   | 1/6     | 2/6     |
| Alvaro-Garcia 2016    | 0.17 (0.00, 7.98)   | 0/46    | 0/7     |
| Gao 2016              | 0.87 (0.55, 1.40)   | 21/62   | 24/62   |
| Ibrahim 2016          | 0.89 (0.07, 12.00)  | 1/9     | 1/6     |
| Hu 2016a              | 1.06 (0.02, 51.84)  | 0/34    | 0/36    |
| Hu 2016b              | 0.97 (0.02, 47.32)  | 0/31    | 0/30    |
| Xie 2016              | 0.85 (0.02, 39.24)  | 0/12    | 0/10    |
| Lin 2017              | 0.60 (0.36, 1.02)   | 15/56   | 24/54   |
| Swaminathan 2018      | 2.03 (0.81, 5.03)   | 12/67   | 6/68    |
| Tompkins 2017         | 1.57 (0.07, 35.46)  | 1/20    | 0/10    |
| Xiao 2017             | 0.23 (0.01, 4.55)   | 0/17    | 2/20    |
| Zhang 2017            | 1.00 (0.02, 48.82)  | 0/30    | 0/30    |
| Galtan 2015/2016      | 0.92 (0.62, 1.36)   | 11/15   | 12/15   |
| Bartolucci 2016/2017  | 1.00 (0.07, 14.55)  | 1/15    | 1/15    |
| Deng 2017             | 1.62 (0.08, 34.66)  | 1/12    | 0/6     |
| Fernandez 2018        | 0.12 (0.01, 2.29)   | 0/19    | 2/11    |
| Sun 2018              | 0.20 (0.01, 3.93)   | 0/21    | 2/21    |
| Mathay 2018           | 1.50 (0.64, 3.54)   | 15/40   | 5/20    |
| Lublin 2014           | 0.38 (0.01, 16.88)  | 0/12    | 0/4     |
| Melmed 2015           | 0.55 (0.01, 26.42)  | 0/30    | 0/16    |

Overall (I²=0%, P=0.85) 0.78 (0.65, 0.94) 122/1040 171/951

### Fig. 2, Continued
Table 2
Risk of bias assessments (expanded detail provided in Supplementary Table 2).

| Author/Year | Selection bias | Performance bias | Reporting bias | Other Bias |
|-------------|----------------|------------------|----------------|-----------|
|             | 1-Random sequence generation | 2-Allocation concealment | 3-ROB due to blinding: patients & personnel | 4-ROB due to blinding: outcome assessor | 5-Attrition: Incomplete outcome data | 6-Selective outcome reporting | Registered Protocol (Y/N) | A priori Sample Size Calculation for Primary Outcome in Methods | Industry or Biotech Sponsored |
| Cardiovascular |               |                  |                |           |           |         |                   |                          |                       |
| Chen et al., 2004 |       |                  |                |           |           |         |                   |                          |                       |
| Chen et al., 2006 |       |                  |                |           |           |         |                   |                          |                       |
| Chaulikena et al., 2015 |       |                  |                |           |           |         |                   |                          |                       |
| Gao et al., 2013 |       |                  |                |           |           |         |                   |                          |                       |
| Gao et al., 2015 |       |                  |                |           |           |         |                   |                          |                       |
| Hace et al., 2009 |       |                  |                |           |           |         |                   |                          |                       |
| Lee et al., 2014 |       |                  |                |           |           |         |                   |                          |                       |
| Wang et al., 2014 |       |                  |                |           |           |         |                   |                          |                       |
| Wang et al., 2014 |       |                  |                |           |           |         |                   |                          |                       |
| Zhao et al., 2015 |       |                  |                |           |           |         |                   |                          |                       |
| Barbouri et al., 2016/2017 |       |                  |                |           |           |         |                   |                          |                       |
| Xiao et al., 2017 |       |                  |                |           |           |         |                   |                          |                       |
| Neurological |               |                  |                |           |           |         |                   |                          |                       |
| Ibrahim et al., 2016* |       |                  |                |           |           |         |                   |                          |                       |
| Lee et al., 2010 |       |                  |                |           |           |         |                   |                          |                       |
| Lee et al., 2012 |       |                  |                |           |           |         |                   |                          |                       |
| Xie et al., 2007** |       |                  |                |           |           |         |                   |                          |                       |
| Xie et al., 2016 |       |                  |                |           |           |         |                   |                          |                       |
| Zhang et al., 2017 |       |                  |                |           |           |         |                   |                          |                       |
| Fernandez et al., 2018 |       |                  |                |           |           |         |                   |                          |                       |
| Kim et al., 2018* |       |                  |                |           |           |         |                   |                          |                       |
| Labrin et al., 2014 |       |                  |                |           |           |         |                   |                          |                       |
| Oncological/hematological |              |                  |                |           |           |         |                   |                          |                       |
| Gao et al., 2016 |       |                  |                |           |           |         |                   |                          |                       |
| Liu et al., 2014 |       |                  |                |           |           |         |                   |                          |                       |
| Ning et al., 2008 |       |                  |                |           |           |         |                   |                          |                       |
| Kaminina et al., 2012 |       |                  |                |           |           |         |                   |                          |                       |
| Stropanova et al., 2014 |       |                  |                |           |           |         |                   |                          |                       |
| Endocrine |               |                  |                |           |           |         |                   |                          |                       |
| Carlson et al., 2015 |       |                  |                |           |           |         |                   |                          |                       |
| Hu et al., 2013 |       |                  |                |           |           |         |                   |                          |                       |
| Skylar et al., 2015 |       |                  |                |           |           |         |                   |                          |                       |
| Hu et al., 2016 |       |                  |                |           |           |         |                   |                          |                       |
| Renal disease |               |                  |                |           |           |         |                   |                          |                       |
| Swaminathan et al., 2018 |       |                  |                |           |           |         |                   |                          |                       |
| Korotkov et al., 2018* |       |                  |                |           |           |         |                   |                          |                       |
| Sun et al., 2018 |       |                  |                |           |           |         |                   |                          |                       |

(continued)
death was significantly reduced in the MSC group in three clinical populations (cardiovascular, neurological, and liver disease), and with autologous, umbilical and freshly cultured MSCs. The sensitivity analysis which excluded RCTs published in abstract form only did not affect the strength or direction of the pre-specified pooled adverse event estimates (see Table 4).

A description of all reported serious adverse events (pre-specified and not pre-specified in our review) and their relatedness to study treatment in the MSC or control group is also provided in Supplementary Table 4. Of all reported serious adverse events (SAEs), a total of 1 SAE in the control group (ventricular tachycardia post-infusion in a trial that administered study drug intravenously) [21] and 3 SAEs in the MSC group (treatment related fever [47], in-stent thrombosis with death and acute coronary artery occlusion [26], the latter two of which were also associated with intra-coronary injection of study drug) were considered related to study treatment. Four other SAEs in the MSC group (grade 1 anaphylactoid reaction [69], gastric ulcer perforation [70], hypersensitivity reaction [70], and anal cancer [70]) were also judged to be possibly related to study treatment [70]. None of the included RCTs were ended prematurely due to safety concerns.

| Liver disease                  | Xu et al., 2014 | Salama et al., 2014 | Suk et al., 2016 | She et al., 2012 | Lim et al., 2017 | Zhang et al., 2017 | Shi et al., 2017 |
|-------------------------------|----------------|-------------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| Respiratory                   | Weiss et al., 2011 | Zheng et al., 2014 | Mattey et al., 2018 |
| Infectious                    | Galitani et al., 2016* |
| Immune-deficient/autoimmune/inflammatory | Zhang et al., 2013 | Alvaro-Garcia et al., 2016 | Hu et al., 2016 | Deng et al., 2017 | Zhang et al., 2018 | Arturo et al., 2017* | Paez et al., 2017* | Yang et al., 2018 | Menein et al., 2015 |
| Other                         | Tompkins et al., 2017 |

Green= low risk of bias; Yellow= unclear risk of bias; Red= high risk of bias; *Abstract form only; **Original review did not report on this outcome, but event rate is inserted for comparison purposes.

| Table 3                                                                 |
|-------------------------------------------------------------------------|
| Comparison of 2012 versus 2019 Safety Outcomes and Quality of Safety Reporting Findings. |

| Safety Outcomes | 2012 SafeCell SR | 2018 SafeCell SR Update |
|-----------------|------------------|-------------------------|
|                 | # of RCTs*       | Findings (RR, 95% CI)   | # of RCTs*       | Findings (RR, 95% CI)   |
| Infusional toxicity- non-fever | 4/8 | 2.01 (0.34 – 11.77) | 32/55 | 1.16 (0.70 – 1.91) |
| Infusional toxicity- fever | 4/8 | 9.28 (2.02 – 42.71) | 19/55 | 2.48 (1.27 – 4.86) |
| Infection       | 4/8 | 1.09 (0.61 – 1.94) | 27/55 | 0.99 (0.81 – 1.21) |
| Malignancy or ectopic tissue formation | 4/8 | 2.21 (0.85 – 5.74) | 19/55 | 0.93 (0.60 – 1.45) |
| Mortality       | 4/8 | 1.22 (0.71 – 2.10) | 40/55 | 0.78 (0.65 – 0.94) |
| Thrombotic or embolic events | 4/8** | 2.71 (0.86 – 8.48) | 24/55 | 1.14 (0.67 – 1.95) |
| Quality of Safety Reporting | # of RCTs* | Findings (%) | # of RCTs* | Findings (%) |
| A priori plan to monitor adverse events | 3/8 | 37.5% | 43/55 | 78.2% |

* That reported the adverse event.
** Original review did not report on this outcome, but event rate is inserted for comparison purposes.
A total of 43 (78.2%) of the 55 RCTs reported an *a priori* plan to monitor safety [18,20–23,26–35,38–40,42–44,46,47,49–51,53,55–58,63,68] (see Supplementary Table 4). Forty-five (81.8%) RCTs provided an *a priori* description of follow-up frequency for adverse events [18,20–35,38–40,42–44,47–53,55–60,62,63,65–70,72].

When comparing the *pre*-specified adverse event profile from 2012 compared to our updated systematic review, the risk of fever is the safety outcome that remains significantly associated with MSC administration (see Table 3). There now appears to be a reduction in the risk of death in association with MSC therapy. In comparison to 2012, our updated review found that more of the included RCTs reported an *a priori* plan to monitor for the occurrence of adverse events [18,20–35,38–40,42–44,47–53,55–60,62,63,65–70,72].

Thirty-five (63.6%) of the 55 RCTs included at least one efficacy outcome as a primary endpoint [21,24–26,28–31,35,36,38,39,41–47,49,52–55,57–61,63,64,66,67,70,71] where the remaining RCTs focused on safety alone. Of the 36 RCTs that reported efficacy outcomes, 23 (64.8%) found that MSCs were efficacious in at least one of the primary efficacy outcomes [24,28–31,35,38,43,44,46,47,49,52–54,58–60,64,66,67,70,71]. A more detailed description of each RCT’s primary and secondary endpoints and their respective findings is provided in Supplementary Table 5.

### 4. Discussion

In our updated systematic review that now includes over 40 additional RCTs and over 2000 additional patients, we continue to detect no associations between MSC treatment and the development of non-fever acute infusional toxicity, infection, or malignancy, nor did we detect associations between MSC treatment and the development of thrombotic or thrombo-embolic events. There does continue to be a significant association between MSC administration and reported fever. However of the 19 RCTs (n = 880 patients) that reported on fever, only six were reported as serious, albeit in all MSC treated patients. In contrast, with an increase in the number of RCTs and patients in our updated review, the risk of death is now significantly reduced in the MSC as compared to the control group. In our updated review we also found that the approach to safety reporting was improved as many more authors reported an *a priori* plan to monitor for safety (78.2% versus 35.5%) and none of the trials were ended pre-maturely due to safety concerns. The findings of our updated review should provide additional assurance to researchers, clinicians, regulators, and patients and families that the administration of MSCs continues to appear safe.

Our systematic review will require future updates as scientists continue to unravel the multitude of mechanisms of actions associated with the cells, as the sources and origins of MSCS expand, and the manufacturing process and the development of second generation MSC products evolve. To illustrate, recent in vitro, pre-clinical, and clinical data has found that MSCs can express or increase secretion of proteins associated with coagulation (ex: tissue factor, thrombin anti-thrombin complexes) and with reports of thromboses [10–14]. Depending on the clinical population this potential pro-coagulant effect could result in a beneficial or harmful clinical effect. In our updated review, we began to address this concern with the inclusion of thrombotic/thromboembolic events as a pre-specified adverse event category. Our findings suggest that these incident events reported in the included RCTs are rare (31 events in 24 RCTs and 1112 patients studied), were reported in both study groups (n = 17 and 14 in the MSC and control groups respectively) and were not significantly associated with administration of MSCs. Although a significant association was not detected, it is likely that these events will be rare and as such we encourage investigators to an *a priori* plan to monitor and report on these events to enable the detection of future thrombotic safety signals.

In contrast to our review from 2012, we found that safety reporting was improved in that more investigators reported an *a priori* plan to monitor for adverse events (78.2% versus 35.5% respectively). Serious adverse events that were reported as related to or as possibly related to study treatment (either in the MSC or control group) (n = 8 of 2634 patients studied) were very rare. This could be because these events are indeed rare or because it can be challenging if not impossible to attribute an event to study treatment, especially when the event does not occur during or shortly after completion of the infusion. To address this challenge in adverse event reporting, we sought to capture and synthesize pre-specified adverse events and any other SAEs, regardless of relatedness to study treatment in each of the RCTs. Even using this approach, safety signals other than fever generation were not detected.

A significant impediment to understanding whether MSCs are efficacious and safe relates to the quality of trial design and transparent reporting. Of the 55 included RCTs, only six trials met all six criteria for low risk of bias whereas none of the RCTs from the 2012 review met all six criteria. Although an improvement from 2012, it is important for investigators to address these risk of bias elements at the design phase of these clinical trials to maximize the internal validity of their research findings. With regard to MSC characterization, only seven trials reported on all three Dominici criteria [16] which aim to provide minimal and standardized criteria to define a MSC. Furthermore, only 29 of the included trials (52.7%) reported some measure of MSC viability during the manufacturing process and even fewer (n = 8, 14.5%) reported on a measure of MSC potency or functionality. We strongly assert that it is critical for investigators to transparently report on MSC characteristics, potency and viability in order to help readers, researchers, health regulators, and the community to better understand why a given trial may have succeeded or failed to meet study endpoints and with the ultimate aim to help move the field forward.

Our systematic review has several strengths. We included a transparent search strategy, pre-defined a set of adverse events that were clinically relevant to MSC administration, and reported on all SAEs that were and were not identified as part of our *a priori* event categories irregardless of relatedness to study treatment to provide the most comprehensive and up to date evaluation of the safety profile of MSC therapy. Our review also has limitations. Six of the RCTs were published in abstract form only and as such contained limited information to populate in our review. However, we included these trials.
so that the readership is aware of them and can further evaluate the efficacy and safety of study results when the full trials are published. Furthermore, the strength or direction of our pooled apriori adverse outcome estimates were not influenced by removal of studies that were published in abstract form only. As in our 2012 review, we pooled incident adverse events from RCTs from diverse adult clinical populations, MSC characteristics and MSC manufacturing in an effort to obtain signals for harm. However, to begin to address this diversity and due to the increased number of included RCTs in this review, we conducted several a priori derived sub group analyses to examine for heterogeneity in our a priori derived adverse event estimates and acknowledged that these analyses should be considered hypothesis generating. Only a few of the included trials (10.9%) met all six low risk of bias criteria which threatens the internal validity of the study findings from the perspectives of both safety and efficacy and we strongly encourage investigators to address these biases at the design stage and during the conduct of these RCTs. Finally, pooling efficacy outcomes for all of the included RCTs was not feasible within the scope of this safety review. However, in an attempt to provide some measure of efficacy information for the readership, we summarized the primary and secondary efficacy endpoints and associated results descriptively in Supplementary Table 5.

In conclusion, our review provides a systematic examination for incident adverse events related to the use of MSCs. Aside from fever, we did not identify any significant reported safety signals. Results from our systematic review provide further assurance to readers, investigators, health regulators, and our patients and communities that, with this updated evidence, MSC therapy continues to appear safe.

Declaration of Competing Interests

LM reports grants from CIHR, OIRM and SCN during the conduct of this study. DJS and SHJM reports affiliations with Northern Therapeutics Inc., outside the submitted work. BH reports prior honoraria from Cornerstone Research Group for the provision of methodologic advice related to systematic review and meta-analysis, outside the submitted work. KRW reported grants for CIHR, outside the submitted work. No other authors have any affiliations to report in relation to this study.

Acknowledgments

Our systematic review was funded by the Ontario Research Fund, the Ontario Institute for Regenerative Medicine and the Stem Cell Network; none of whom were involved in study design, in the collection and interpretation of data, in the writing of the report, nor in the decision to submit the paper for publication. Our research team would like to thank Ms. Risa Shorr (Medical Information Specialist) for her assistance with building and conducting the electronic search strategy. We would also like to thank Ms. Elham Sabri for her statistical expertise and Ms. Marnie Gordon for the administrative assistance that she provided.

Supplementary materials

Supplementary material associated with this article can be found in the online version at doi:10.1016/j.eclinm.2019.100249.

Appendix 1. Search strategy

**Medline**

Database: Ovid MEDLINE(R) ALL (1946 to September 23, 2019)

Search Strategy:

1. exp Mesenchymal Stem Cells/ (33,741)
2. exp Mesenchymal Stem Cell Transplantation/ (10,675)
3. exp Multipotent Stem Cells/ (36,140)
4. exp Mesenchymal Stromal Cells/ (33,741)
5. (mesenchymal adj3 (stem or stroma$1 or progenitor*)) and cell $1).tw. (47,264)
6. (mesenchymal adj2 (stem or stromal or progenitor or multipotent or bone marrow or adipose or placenta*)).tw. (47,000)
7. (MSC or MScs or ADMSC or ADMSCs or BM-MSC or BM-MSCs or BMD-MSC or BMD-MSCs or BMDMSC or BMDMSCs).tw. (29,267)
8. ((multipotent or multi-potent) adj3 (stroma$1 cell$1 or stem cell$1)).tw. (4521)
9. marrow stroma$1 cell$1.tw. (6975)
10. (colony-forming unit fibroblast* or CFU-F$1).tw. (844)
11. Mesoderm/cy (5710)
12. or/1-11 (71,482)
13. (ae or to or po or co).fs. (3,812,354)
14. (safe or safety).ti,ab. (723,468)
15. side efect$ti,ab. (236,090)
16. ((adverse or undesirable or harm* or serious or toxic) adj3 (effect* or reaction* or event* or outcome*)).ti,ab. (494,272)
17. exp product surveillance, postmarketing/ (14,736)
18. exp adverse drug reaction reporting systems/ (7274)
19. exp clinical trials, phase iv/ (289)
20. exp poisoning/ (154,008)
21. exp substance-related disorders/ (269,073)
22. exp drug toxicity/ (111,624)
23. exp abnormalities, drug induced/ (14,457)
24. exp drug monitoring/ (19,962)
25. exp drug hypersensitivity/ (44,888)
26. (toxicity or complication* or noxious or tolerability or hyper-sensitivity or abnormal*).ti,ab. (1,919,833)
27. exp Postoperative Complications/ (525,174)
28. exp Intraoperative Complications/ (51,079)
29. or/13-28 (6,186,659)
30. 12 and 29 (10,413)
31. randomized controlled trial.pt. (489,730)
32. controlled clinical trial.pt. (93,263)
33. randomized.ab. (454,901)
34. placebo.ab. (200,750)
35. drug therapy.fs. (2,140,942)
36. randomly.ab. (318,366)
37. trial.ab. (476,933)
38. groups.ab. (1,955,339)
39. (clinical trial* or multicenter study).pt. (747,085)
40. or/31-39 (4,746,525)
41. exp animals/ not humans/ (4,617,450)
42. 40 not 41 (4,135,721)
43. 30 and 42 (1815)
44. limit 43 to yr="2012 -Current" (1346)

Cochrane

Search Name: McIntyre-Lauralynn-MSCS-Safety_2019-09-25
Date Run: 25/09/2019 19:04:03
Comment: 1. MeSH descriptor: [Mesenchymal Stem Cells] explode all trees 97
2. MeSH descriptor: [Mesenchymal Stem Cell Transplantation] explode all trees 183
3. MeSH descriptor: [Multipotent Stem Cells] explode all trees 99
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