Evidence-based Guidelines for the use of Stem Cell Therapy

Neurological Conditions Supplement



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ABBREVIATIONS

AIS	:	ASIA Impairment Scale
ADL	:	Activities of Daily Living
ALD	:	Aldehyde Dehydrogenase
ADMSCs	:	Adipose tissue-Derived Mesenchymal Stem Cells
AEs	:	Adverse Events
AHSCT	:	Autologous Hematopoietic Stem Cell Transplantation
ALS	:	Amyotrophic Lateral Sclerosis
ALSFRS-R	:	Amyotrophic Lateral Sclerosis Functional Rating Scale Revised
ASIA	:	American Spinal Injury Association
BI	:	Barthel Index
BM	:	Bone Marrow
BMA	:	Bone Marrow Aspiration
BMAC	:	Bone Marrow Aspirate Concentrate
BM-MNCs	:	Bone Marrow Mononuclear Cells
BM-MSCs	:	Bone marrow derived Mesenchymal Stem/Stromal Cells
BMSCs	:	Bone Marrow Stromal Cells
CBSCs	:	Cord Blood Stem Cells
CD	:	Cluster of Differentiation
CI	:	Confidence Interval
CoI	:	Conflict of Interest
CSF	:	Cerebrospinal Fluid
DMT	:	Disease Modifying Therapy
EDSS	:	Expanded Disability Status Scale
ERG	:	External Review Group
EPCs	:	Epithelial Progenitor Cells
EQ-5D	:	Euro-QoL- 5D
ESCs	:	Embryonic Stem Cells
F/U	:	Follow-Up
FVC	:	Forced Vital Capacity
GDG	:	Guideline Development Group
GRADE	:	Grading of Recommendations Assessment, Development and Evaluation
haMPCs	:	Human Autologous Adipose-Derived Mesenchymal Progenitor Cells
hESCs	:	Human Embryonic Stem Cells
HHD	:	Hand-Held Dynamometry
HIV	:	Human Immunodeficiency Virus
HMSC	:	Human Mesenchymal Stem Cell
HSCs	:	Hematopoietic Stem Cells
hUC	:	Human Umbilical Cord
HuCNS-SCs	:	Human Central Nervous System Neural Stem Cells
IA	:	Intra Arterial
ICA	:	Internal Carotid Artery
ICH	:	Intracerebral Haemorrhage
IM	:	Intramuscular

iPSCs	:	Induced Pluripotent Stem Cells
IS	:	Ischemic Stroke
IT	:	Intrathecal
IV	:	Intravenous
MCA	:	Middle Cerebral Artery
MCID	:	Minimal Clinical Important Difference
MD	:	Mean Difference
Med	:	Median
MNCs	:	Mononuclear Cells
MPCs	:	Mesenchymal Progenitor Cells
mRS	:	Modified Rankin scale
MS	:	Multiple Sclerosis
MSC	:	Mesenchymal Stem Cell
MSC-NTFs	:	Mesenchymal Stem Cell Induced to Secrete High Levels of Neurotrophic Factors
MSCs	:	Mesenchymal Stem/Stromal Cells
MSFC	:	Multiple Sclerosis Functional Composite
MTX	:	Methotrexate
NIHSS	:	National Institute of Health Stroke Scale
NR	:	Not Reported
NSAID	:	Non-Steroid Anti-Inflammatory Drug
NSCs	:	Neural Stem Cells
PBSCs	:	Peripheral Hematopoietic Stem Cells
PET	:	Positron Emission Tomography
PRISMA	:	Preferred Reporting Items for Systematic Reviews and Meta-Analysis
PT	:	Physical Therapy
RCT	:	Randomized Controlled Trial
RoB 2	:	Risk of Bias 2
RPMS	:	Rapidly Progressive Multiple Sclerosis
RR	:	Relative Risk
RRMS	:	Relapsing Remitting Multiple Sclerosis
SAEs	:	Severe Adverse Events
SCI	:	Spinal Cord Injury
SCIM	:	Spinal Cord Independence Measure
SCLs	:	Spinal Cord Lesions
SD	:	Standard Deviation
SE	:	Standard Error
SF-36	:	36-Item Short-form Health Survey
SMD	:	Standardized Mean Difference
SPMS	:	Secondary Progressive Multiple Sclerosis
SR/MA	:	Systematic Review/Meta-Analysis
SVC	:	Slow Vital Capacity
TEAEs	:	Treatment Emergent Adverse Events
UCMSCs	:	Umbilical Cord Mesenchymal Stem Cells
VC	:	Vital Capacity

1. STROKE

i. **Key question in PICO format**

ii. **Search strategy**

iii. **PRISMA flow diagram**

iv. **Summary of included studies**

v. **Classification of studies based on level of stem cell manipulation**

vi. **Evidence to decision framework**

vii. **Data extraction**

viii. **Publication bias**

ix. **List of excluded studies**

i. Key Question in PICO format:

In patients with stroke, what is the efficacy and safety of stem cell therapy as compared to usual care?

Population: Patients with Ischemic stroke/ Hemorrhagic stroke
Subgroup analysis: Acute vs chronic; First/recurrent stroke
Intervention: Any Stem cell and product derived from stem cells
Comparator: Usual Care/ Conventional Care
Critical Outcomes: Efficacy: any measure of Disability and Handicap;
Safety outcomes: mortality, serious adverse events, any tumour formation;
Important: quality of life, other adverse events

ii. Search Strategy

Search 1: **Medline**:

Line #	Query terms
1	Exp Cerebrovascular disease/
2	Exp Ischemic stroke/
3	Exp hemorrhagic stroke/
4	Cerebral hemorrhage/or exp stroke/or Hemorrhagic stroke/
5	Stroke/ or Brain Ischemia/
6	Brain infarction/
7	Cerebral arterial diseases/
8	Brain ischemia/
9	Intracranial bleed/
10	(Stroke* or Ischemic stroke* or Ischaemic stroke* or Intracranial hemorrhage* or Intracranial haemorrhage* or Cerebrovascular disorders* or Brain infarct or cerebral arterial disease or Intracranial embolism or Intracranial thrombosis or CVA or CVD).m_titl.
11	Limit 10 to abstracts
12	(isch?emi$ adj5 (stroke$ or apoplex$ or cerebrovasc$ or cerebrovasc$ or cva$ or attack$)).tw.
13	((Brain or cerebr$ or cerebell$ or vertebrobasil$ or hemisphere$ or intracra$ or basal gang$ or putami$ or intracerebral or infratentorial or supratentorial or middle cerebr$ or mca$ or anterior circulation) adj5 (isch?emi$ or infract$ or thrombo$ or emboli$ or occlus$ or hypoxi$ or h?emorrhag$)).tw.
14	Or/1-13
15	Exp stem cell transplantation/
16	Cell transplantation/
17	Stem cell therapy.mp.
18	Cord blood stem cell transplantation/

19	Hematopoietic stem cell transplantation/
20	Mesenchymal stem cell transplantation/
21	Peripheral blood stem cell transplantation/
22	(Stem cell* or adult stem cell* or mesenchymal stem cell* or embryonic stem cell* or fetal stem cell* or hematopietic stem cell* or peripheral blood stem cell* or fibroblasts or myeloid progenitor cell or erythroid progenitor cell or multipotent stem cell or myoblast or pluripotent stem cell or totipotent stem cells or tumor stem cells).m_titl.
23	limit 22 to abstracts
24	((stem or progenitor or embry$ or fetal or foetal or umbilical or bone marrow or cord blood adj5 cell or cells)).tw.
25	(cell adj5 (transplant$ or graft$)).tw.
26	(fibroblast$ or myoblast$).tw.
27	(fibroblast$ or myoblast$).tw.
28	Or 15-27
29	Randomized controlled trial/
30	Random allocation/
31	Controlled clinical Trial/
32	Control groups/
33	Clinical trials as topic/or clinical trials, phase I as topic/ or clinical trials, phase II as topic/ or clinical trials., phase III as topic/ or clinical trials, phase IV as topic/
34	Double-blind method/
35	Single-blind method/
36	Placebos/
37	Placebo effect/
38	Drug evaluation/
39	Research design/
40	Randomized controlled trial.pt
41	Controlled clinical trial.pt
42	(clinical trial or clinical trial Phase I or clinical trial phase II or clinical trial phase III or clinical trial phase IV).pt
43	(random$ or RCT or RCTs).tw.
44	(Controlled adj5 (trial$ or stud$)).tw.
45	(Clinical$ adj5 trial$).tw.
46	((Control or treatment or experiment$ or intervention)adj5 (group$ or subjects$ or patients$).tw.
47	(quasi-random$ or quasi random$ or pseudo-random$ or pseud or random$).tw.
48	((Singl$ or doubl$ or tripl$ or trebl$) adj5 (blind$ or mask$)).tw.
49	Placebo$.tw.
50	Controls.tw.
51	Or/29-50
52	Exp animals/not humans.sh

53	12 and 25 and 49
54	50 not 49

Search 2: **Pubmed:**

Line #	Query terms
1	Exp Cerebrovascular disease/
2	Exp Ischemic stroke/
3	Exp hemorrhagic stroke/
4	Cerebral hemorrhage /or exp stroke/or Hemorrhagic stroke/
5	Stroke/ or Brain Ischemia/
6	Brain infarction/
7	Cerebral arterial diseases/
8	Brain ischemia/
9	Intracranial bleed/
10	(Stroke* or Ischemic stroke* or Ischaemic stroke* or Intracranial hemorrhage* or Intracranial haemorrhage* or Cerebrovascular disorders* or Brain infarct or cerebral arterial disease or Intracranial embolisim or Intracranial thrombosis or CVA or CVD).m_titl.
11	Limit 10 to abstracts
12	(isch?emi$ adj5 (stroke$ or apoplex$ or cerebrovasc$ or cerebrovasc$ or cva$ or attack$)).tw.
13	((Brain or cerebr$ or cerebell$ or vertebrobasil$ or hemisphere$ or intracra$ or basal gang$ or putami$ or intracerebral or infratentorial or supratentorial or middle cerebr$ or mca$ or anterior circulation) adj5 (isch?emi$ or infract$ or thrombo$ or emboli$ or occlus$ or hypoxi$ or h?emorrhag$)).tw.
14	Or/1-13
15	Exp stem cell transplantation/
16	Cell transplantation/
17	Stem cell therapy.mp.
18	Cord blood stem cell transplantation/
19	Hematopoietic stem cell transplantation/
20	Mesenchymal stem cell transplantation/
21	Peripheral blood stem cell transplantation/
22	(Stem cell* or adult stem cell* or mesenchymal stem cell* or embryonic stem cell* or fetal stem cell* or hematopietic stem cell* or peripheral blood stem cell* or fibroblasts or myeloid progenitor cell or erythroid progenitor cell or multipotent stem cell or myoblast or pluripotent stem cell or totipotent stem cells or tumor stem cells).m_titl.
23	limit 22 to abstracts
24	((stem or progenitor or embry$ or fetal or foetal or umbilical or bone marrow or cord blood) adj5(cell or cells)).tw.
25	(cell adj5 (transplant$ or graft$)).tw.

26	(fibroblast$ or myoblast$).tw.
28	Or 15-27
29	Randomized controlled trial/
30	Random allocation/
31	Controlled clinical Trial/
32	Control groups/
33	Clinical trials as topic/or clinical trials, phase I as topic/ or clinical trials, phase II as topic/ or clinical trials., phase III as topic/ or clinical trials, phase IV as topic/
34	Double-blind method/
35	Single-blind method/
36	Placebos/
37	Placebo effect/
38	Drug evaluation/
39	Research design/
40	Randomized controlled trial.pt
41	Controlled clinical trial.pt
42	(clinical trial or clinical trial Phase I or clinical trial phase II or clinical trial phase III or clinical trial phase IV).pt
43	(random$ or RCT or RCTs).tw.
44	(Controlled adj5 (trial$ or stud$)).tw.
45	(Clinical$ adj5 trial$).tw.
46	((Control or treatment or experiment$ or intervention)adj5 (group$ or subjects$ or patients$).tw.
47	(quasi-random$ or quasi random$ or pseudo-random$ or pseud or random$).tw.
48	((Singl$ or doubl$ or tripl$ or trebl$) adj5 (blind$ or mask$)).tw.
49	Placebo$.tw.
50	Controls.tw.
51	Or/29-50
52	Exp animals/not humans.sh
53	12 and 25 and 49
54	50 not 49

Search 3: **Cochrane central:**

Search Hits

#1 MeSH descriptor: [Ischemic Stroke] explode all trees: 994

#2 MeSH descriptor: [Cerebrovascular Disorders] explode all trees and with qualifier(s): [drug therapy- DT, mortality - MO] 5167

#3 MeSH descriptor: [Hemorrhagic Stroke] explode all trees 40

#4 MeSH descriptor: [Stroke] 1 tree(s) exploded 15152

#5 (Brain ischemia):ab 1059

#6 (Intracranial bleed):ab 95

#7 (Stroke* or Ischemic stroke* or Ischaemic stroke* or Intracranial hemorrhage* or Intracranial haemorrhage* or Cerebrovascular disorders* or Brain infarct or cerebral arterial disease or Intracranial embolism or Intracranial thrombosis or CVA or CVD):kw 30036

#8 #1 or #2 or #3 or #4 or #5 or #6 or #7 33122

#9 (stem cell transplantation):ti,ab,kw (Word variations have been searched) 11284

#10 (Stem cell therapy):ti,ab,kw 9841

#11 (Cord blood stem cell transplantation):ti,ab,kw (Word variations have been searched) 386

#12 ("hematopoietic stem cell transplantation"):ti,ab,kw (Word variations have been searched) 4630

#13 (Mesenchymal stem cell transplantation):ti,ab,kw (Word variations have been searched) 972

#14 (Peripheral blood stem cell transplantation):ti,ab,kw (Word variations have been searched) 1977

#15 (Stem cell* or adult stem cell* or mesenchymal stem cell* or embryonic stem cell* or fetal stem cell* or hematopietic stem cell* or peripheral blood stem cell* or fibroblasts or myeloid progenitor cell or erythroid progenitor cell or multipotent stem cell or myoblast or pluripotent stem cell or totipotent stem cells or tumor stem cells):kw (Word variations have been searched) 10372

#16 (stem or progenitor or embry$ or fetal or foetal or umbilical or bone marrow or cord blood adj5 cell or cells):ab (Word variations have been searched) 139685

#17 (fibroblast$ or myoblast$):ti,ab,kw (Word variations have been searched) 3474

#18 #9 #11 or #12 or #13 or #14 or #15 or #16 or #17 143419

#19 ("randomized controlled trial"):ti,ab,kw (Word variations have been searched) 660973

#20 ("random allocation"):ab (Word variations have been searched) 49593

#21 ("controlled clinical trial"):ti (Word variations have been searched) 14165

#22 (Control groups):ab (Word variations have been searched) 555759

#23 (Clinical trials as topic or clinical trials, phase I as topic or clinical trials, phase II as topic or clinical trials, phase III as topic or clinical trials, phase IV as topic):ab (Word variations have been searched) 5754

#24 ("double blind method"):ti,ab,kw (Word variations have been searched) 155836

#25 ("single blind method"):ti,ab,kw (Word variations have been searched) 24786

#26 ("placebo"):ab (Word variations have been searched) 339632

#27 ("placebo effect"):ti,ab,kw (Word variations have been searched) 5570

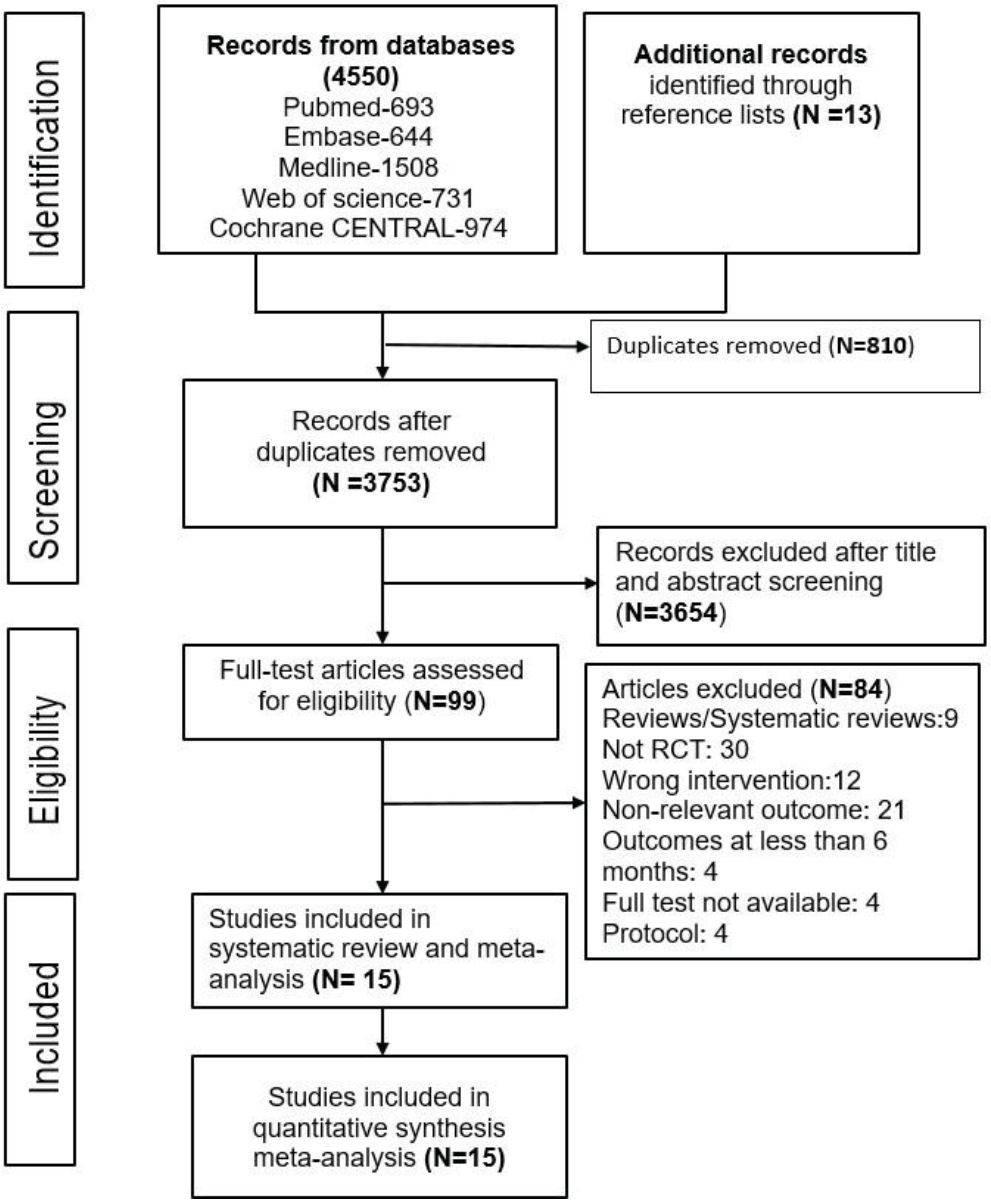
#28 (Drug evaluation):ab (Word variations have been searched) 76217

#29 ("research design"):ab (Word variations have been searched)6733

#30 #19 or #20 or #21 or #22 or #23 or #24 or #25 or #26 or #27 or #28 or #29 1194911

#31 #8 AND #18 AND #30 974

iii. PRISMA Flow Diagram



Out of a total of 15 RCTs in ischemic stroke, 11 trials met the inclusion criteria as specified by the GDG and were used for synthesizing evidence.

iv. Summary of included studies:

Study	Population	Intervention	Comparator	Outcome	Comments
Bang et al. 2005[1]	30 participants with cerebral infarction in the MCA territory and severe neurological deficit Treatment 10, Control 20 After randomization 5 patients refused and were considered under control Treated between 32 and 61 days from stroke onset	Intravenous infusion of 2 boosts of 50 million culture-expanded autologous bone marrow mesenchymal stem cells	Standard care (Detail not specified)	NIHSS score as an index of neurological deficit and BI and mRS as indices of functional recovery at the end of the 12-month follow-up	Trial excluded after loss of 30% post randomization in treatment arm (as per terms of reference given by committee)
Bhatia et al. 2018[2]	Twenty participants with subacute ischemic stroke in the MCA territory and NIHSS > 7 10 Intervention and 10 control Treated between 8 and 15 days from stroke onset	Intra-arterial infusion of bone marrow‐derived mononuclear cells (mean 6.1 million) into the ipsilateral MCA	Not specified	Primary safety outcomes and secondary efficacy endpoints as combined mRS, BI, and NIHSS score at the end of the 6-month follow-up	
Celis-Ruiz et al. 2022[3]	Nineteen participants with acute or subacute ischemic stroke, and NIHSS between 8 and 20. Nine participants randomized to Intervention arm 9 and 10 to placebo arm. Five participants in intervention arm and 1 in placebo excluded prior to	Intravenous administrationof allogeneic mesenchymal stem cells from adipose tissue (1 million/kg) within the first 2 weeks after the onset of stroke symptoms.	Placebo (composition not specified):0.1 ml per kilogram of patient's weight administered IV	Safety and efficacy analysis (mRS, NIHSS, size of infarct, and biochemical markers)	Trial excluded after loss of 30% post randomization in treatment arm (as per terms of reference given by committee)

	treatment administration Median time for treatment administered was 12 days in interventional arm and 13 days in placebo arm				
Chen et al. 2014[4]	Thirty participants with chronic MCA infarction and neurological deficits of intermediate severity. Treatment arm 15, Control arm 15 Treated between 6 months to 5 years of stroke onset	Subcutaneous granulocyte-colony stimulating factor injections (15 μg/kg/day) for 5 consecutive days, followed by stereotaxic implantation of autologous 3 to 8 million $CD34^+$ immunosorted peripheral blood stem cells	Conventional treatment with antiplatelet and antihypertensive	Improvements in stroke scales (NIHSS, European Stroke Scale, and European Stroke Scale Motor sub scale) and functional outcomes measure (mRS) from baseline to the end of the 12-month follow-up	
Fang et al. 2018[5]	Adult (18-80 years) with acute ischemic stroke within 7 days from onset of symptoms, with NIHSS >7 at days 7 after onset. Three arm study. 6 randomised to Epithelial progenitor cell (EPC) arm, 6 to Bone marrow stromal cell (BMSCs) arm and 6 to placebo arm One in EPC and 2 in BMSCs arm did not receive the intervention	Two cycles 2.5 million cells/kg of autologous EPCs or BMSCs were transplanted intravenously, 1 week apart	Underwent a bone marrow aspiration with saline infusion	Safety (adverse events, neurological detoriation, mortality) and efficacy by NIHSS, Scandinavian stroke scale, functional scales, Barthel Index and mRS at 48 months	The bone marrow arm of the study was excluded as there was >30% loss post randomization in treatment arm

Hess et al. 2017[6]	Participants with acute ischemic stroke involving MCA territory, with NIHSS score 8 to 20, and infarct size between 5 and 100 cc^3, were enrolled Treatment arm 67, Control arm 62 Treated between 24-48 hours of stroke	Intravenous infusion of 1200 million multipotent progenitor cells (in a solution containing PlasmaLyte-A, dimethyl sulfoxide, and human serum albumin)	Placebo solution (containing PlasmaLyte-A, dimethyl sulfoxide, and human serum albumin, without the progenitor cell)	The primary efficacy endpoint combined the mRS, the BI, and change in NIHSS score from baseline, and was evaluated at day 90, and at the end of the 12-month follow-up	
Houkin et al. 2024[7]	Ischemic stroke with NIHSS 8-20, between 20-84 years. Treatment arm 105, Control 102 Treated within 18-36 hours of stroke onset	Single intravenous infusion of Multi Stem over 30 to 60 min within 18- 36 hours of stroke onset	Placebo (Contained only cryopreservation medium)	Primary: Proportion of patients with excellent outcome at 90 days. (mRS ≤1, NIHSS ≤1, BI ≥95). Secondary: Excellent outcome at 365 days., distribution of mRS at 90 and 365, Proportion of patients with mRS ≤ 1 and ≤ 2 at 90 days. Safety outcomes	
Jaillard et al. 2020[8]	Ischemic stroke within 2 weeks of onset of stroke with NIHSS >7. Treatment arm 20, Control 11 Four patients did not receive the intervention after randomization as were included in control. Median days to receive the treatment was 32 days (IQR 28-40)	Two different doses via Intravenous infusion. The first ten patients assigned to treatment received low-dose MSCs (100 million) and the next ten patients received high-dose MSCs (300 million)	Not specified (Except for rehabilitation which was provided in both groups)	Safety outcome (Adverse events) and efficacy outcomes (NIHSS, mRS and Barthel index) at 6 months and 24 months	

Jin et al. 2017[9]	Participants with subacute cerebral infarction and NIHSS score between 5 and 30. Treatment arm 10, Control arm 10 Treated between 3 weeks to 3 months of stroke onset	Subarachnoid infusion of a cell suspension containing 10 million autologous bone marrow mononuclear cells	Routine medical management including antiplatelet and statin	Various safety and efficacy outcomes (Barthel Index, mRS, Functional measure and Fugl-Meyer motor score) through a 7-year follow-up	
Law et al. 2021[10]	Participants with sub-acute infarct (within 2 months) with NIHSS between 10 and 35. Treatment arm 9, Control arm 8 Median days to intervention was 63 (IQR 61-122)	Autologous bone marrow derived mesenchymal stem cells administered intravenously at a mean dosage of 2.05 million cells per kg	Standard medical care	Efficacy outcomes by NIHSS, mRS and BI scores at 12 months	
Lee et al. 2010[11]	Participants with MCA territory infarct and severe neurological deficit (modified NIHSS >7) Treatment 16, Control 36 Treated between 4 and 9 weeks of stroke onset	Intravenous infusion of 2 boosts of 50 million culture-expanded autologous bone marrow mesenchymal stem cells	Not specified	Functional outcome (mRS) at the end of the last follow up	Trial excluded after loss of 30% post randomization in treatment arm (as per terms of reference given by committee)
Monich e et al. 2023[12]	MCA territory stroke within 7days with NIHSS scale 6-20. Treatment:39 (Low dose BMMNCs 20, High dose BMMNCs 19), Control 38 Median time for intervention 6 (IQR 5-7)	Autologous Bone marrow mononuclear cells transfused via intra-arterial route in two different dose (2million cells/kg and 5 million cells/kg)	Standard stroke treatment	Efficacy (mRS, NIHSS, and Barthel Index) and safety at 6 months	

Niizuma et al. 2023[13]	Ischemic stroke patients with NIHSS ≥6 and mRS ≥3 within 14-28 days of onset stroke Treatment 27, Control 10 Treated between 14-28 days of stroke	Allogenic muse cell-based product (CL2020), 15 million cells/15 ml were administered intravenously	In control arm 15 ml of placebo was administered. Placebo was of same composition without the cells	Safety and efficacy (mRS, NIHSS, Barthel Index, EU-5D-5L) at 52 weeks	
Prasad et al. 2014[14]	Participants with anterior circulation ischemic stroke, onset within 7-30 days NIHSS ≥7 Treatment 60, Control 60 Treated within 7-30 days of stroke onset	Intravenous infusion of a mean of 280 million autologous bone marrow mononuclear stem cells	Conventional treatment	Primary efficacy measures e.g. mRS and BI at 6 months and Secondary outcome of NIHSS at 1 year. Safety outcome included death, adverse events, epileptiform discharges and any new growth in PET scan	
Savitz et al. 2019[15]	First time MCA territory stroke with mRS>/3 Treatment 29, Placebo 19 Treated between 14-20 days	Single internal carotid infusion of 3.08 million autologous Bone-marrow derived ALD-401 cells	Sham infusion given in the placebo arm	Safety of delivery of ALD-401 cells and efficacy as measured by mRS, Secondary outcomes were NIHSS, Barthel Index and EQ-5D at 3month sand 12 months	

v. Classification of studies on the basis of level of manipulation of the stem cell/ Stem cell derived product used (*interpreted by the Secretariat as defined by CDSCO* and the information provided in the trial itself*):

S. No.	Trial Details	Intervention	Level of manipulation	Information extracted from trial
1.	Bang et al. 2005	Autologous MSCs- Intravenous infusion of 2 boosts of 50 million culture-expanded autologous bone marrow mesenchymal stem cells	More	*"Culture expanded MSCs… Cells checked for SH-2 and SH-4 using flow cytometry"*
2.	Bhatia et al. 2018	Autologous BMMNC- Intra-arterial infusion of bone marrow–derived mononuclear cells (mean 6.1 million) into the ipsilateral MCA	Less	*"The BMMNC were separated using a ficoll density gradient centrifugation procedure… Quality control of the final cell product was performed to determine the total number of cells, total BMMNC, total CD34+ cells, CD34+ cell percentage, sterility, and viability*."
3.	Chen et al. 2014	Autologous PBSCs- Subcutaneous granulocyte-colony stimulating factor injections (15μg/kg/day) for 5 consecutive days, followed by stereotaxic implantation of autologous 3 to 8 million CD34+ immune-sorted peripheral blood stem cells	Less	*"Collected through leukapheresis, Immune sorting using CD34 antibody conjugated with nano-iron beads (CD34-nano-iron complex); percentage of CD34+ cells by flow cytometry using CD34+ antibody"*
4.	Fang et al. 2018	Autologous ex vivo expanded endothelial progenitor cells (EPCs)/ bone marrow stromal cells (BMSCs)- Two cycles of 2.5 million cells/kg of culture-expanded autologous EPCs or BMSCs were transplanted intravenously, 1 week apart	More	*"BMMNC isolated and cultured; EPC identified using UEA-1 letin and endocytosis of Dil-Ac-LDL."*
5.	Hess et al. 2017	Multipotent adult progenitor cells (MAPC)- Intravenous infusion of 1200 million multipotent progenitor cells (in a solution containing PlasmaLyte-A, dimethyl sulfoxide, and human serum albumin)	More	*"MultiStem multipotent adult progenitor cells (Lonza, Walkersville, MD, USA; under contract with Athersys, Inc) were provided to the clinical sites in standard units; commercially available MAPC provided; thawed and combined to reach appropriate dose"*

6.	Houkin et al. 2024	Allogeneic multipotent adult progenitor cells- Single intravenous infusion of Multi Stem (HLCM051) of 1.2 billion cells	More	*"Bone marrow derived; commercially available cryopreserved product"*
7.	Jaillard et al. 2020	Autologous mesenchymal stem cells (MSCs)- Two different doses via Intravenous infusion. The first ten patients assigned to treatment received low-dose MSCs (100 million) and the next ten patients received high-dose MSCs (300 million)	More	*"Autologous cell expansion done for 3–4 weeks"*
8.	Jin et al. 2017	Autologous bone marrow mononuclear cells- Subarachnoid infusion of a cell suspension containing 10 million BMMNCs	Less	*"Bone marrow mononuclear cells were collected the treatment group using the density gradient method. A cell suspension containing 1×10^7 bone marrow mononuclear cells was then injected into the subarachnoid space,"*
9.	Law et al. 2021	Autologous BM-MSCs- culture-expanded bone marrow derived mesenchymal stromal cells administered intravenously at a mean dosage of 2 million cells per kg	more	*"BMMSCs were isolated based on density gradient centrifugation and adherence to a plastic surface… BMMSCs were cultured with proprietary BMMSC culture medium and maintained in a 5% carbon dioxide incubator at 37°C."*
10.	Lee et al. 2010	Autologous Mesenchymal Stem Cell- Intravenous infusion of 2 boosts of 50 million culture-expanded autologous bone marrow mesenchymal stem cells	More	*"Bone marrow mononuclear cells were separated by Ficoll density centrifugation. Mononuclear cells were cultured in a 175 cm^2 flask (Falcon, Franklin Lakes, NJ, http://www.bd.com) with low-glucose Dulbecco's modified Eagle's medium."*
11.	Moniche et al. 2023	Autologous BMMNC- Autologous Bone marrow mononuclear cells transfused via intraarterial route in two different doses (2 million cells/kg and 5 million cells/kg) The transplantation procedure was done within 1–7 days of stroke onset, as previously described.	Less	*"The harvested bone marrow was centrifuged on a Ficoll density gradient to isolate the mononuclear cells and obtain the low and high doses (2×10^6 BMMNCs/kg or 5×10^6 BMMNCs/kg), which were washed and resuspended in 20 mL of Ringer lactate solution supplemented with 1% heparin and 2·5% glucose and packaged in two sterile syringes, with 10 mL each, for infusion into the patient."*

12.	Niizuma et al. 2023	Allogenic muse cell-based product (CL2020)- 15 million cells/15 ml were administered intravenously administered intravenously over 10–15 minutes.	More	*"Clinical-grade CL2020 ($1.5x10^7$ cells/15 mL) was manufactured from human (allogenic) MSC by Life Science, Institute, Inc, Japan. Commercially available; derived from mesenchymal stem cells"*
13.	Prasad et al. 2014	Autologous BMMNC- Intravenous infusion of a mean of 280 million autologous bone marrow mononuclear stem cells	Less	*"BMSCs were separated by ficoll density separation method. Immunophenotyping to count CD34+ cells was done using flow cytometry. The cells suspended in phosphate-buffered saline were infused into antecubital vein of the patients in BMSC arm."*
14.	Savitz et al. 2019	Autologous Bone-marrow derived ALD-401 cells- Single internal carotid infusion of 3.08 million	Less	*"Cell isolation and manufacturing of ALD-401 cells. ALD-401 is an enriched population of aldehyde dehydrogenase- bright stem cells… Cells were administered a mean and median of 2 days, and in no case >3 days from harvest."*

* as defined by CDSCO in Annexure- I of the guideline document.

vi. Evidence to Decision Framework:

QUESTION

In patients with Ischemic stroke, what is the efficacy and safety of stem cell therapy compared to usual care?	
POPULATION:	Ischemic stroke
INTERVENTION:	Stem cell transplantation
COMPARISON:	Usual Care
MAIN OUTCOMES:	Disability, Dependency, All-cause mortality, Serious Adverse Events
SETTING:	Hospital
PERSPECTIVE:	Health Systems/Population
BACKGROUND:	Stroke is a leading cause of morbidity and mortality worldwide, with very large healthcare and social costs resulting in a major economic burden on the patient, family and society. The Global Burden of Disease study found that globally, stroke remained the second-leading cause of death (11·6% [10·8–12·2] of total deaths) and the third-leading cause of death and disability combined (5·7% [5·1–6·2] of total DALYs) in 2019. Despite the availability of numerous medical innovations, interventions and therapeutic approaches, it continues to be one of the leading causes of disability worldwide.[16-18]
CONFLICT OF INTERESTS:	None

ASSESSMENT

Problem Is the problem a priority?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**

○ No ○ Probably no ○ Probably yes ● Yes ○ Varies ○ Don't know	Stroke is a leading cause of morbidity and mortality worldwide, with very large healthcare and social costs. Majority of the strokes are ischemic in nature caused by interruption of blood flow leading to irreversible damage to the brain. The use of therapeutic options like thrombolysis and thrombectomy is limited due to narrow time window and strict selection criteria for patients. Therefore, there is a strong demand for alternative therapeutic approaches.[16,18]	
Desirable Effects How substantial are the desirable anticipated effects?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
● Trivial ○ Small ○ Moderate ○ Large ○ Varies ○ Don't know	**Disability:** Eleven trials, with a total of 697 participants, reported the Modified Rankin Scale (mRS) score at the end of follow up (ranged from 6 months to 7 years). The mean difference of mRS was -0.35 (95% CI: -0.51 to -0.19) in the stem cell arm as comparison to usual care. There seems to be a trivial reduction in the disability with the use of stem cell therapy i.e. less than half of the MCID of one (dotted line). Therefore, the effect observed is statistically significant but unimportant clinically. **Dependency:** Five trials, with a total of 197 participants, reported the Barthel Index (BI) score as a continuous variable at the end of follow up. (Follow-up period ranged from 1 year to 7 years) The mean difference in BI was 12.1 (95% CI: -2.19 to 26.38) in stem cell arm compared to usual care. The difference was statistically non-significant in pooled analysis. **All-cause mortality:** Twelve studies with a total of 745 participants and 67 events reported mortality. Pooled analysis yielded a risk ratio of 0.83 (95% CI: 0.54 to 1.28) in the stem cell armas compared to usual care, which was statistically non-significant.	The GDG decided the following MCID for the critical outcomes. MCID of mRS SCALE: 1 level change is considered clinically important. MCID of Barthel Index: The most conservative estimate of the MCID is 10 points.

Undesirable Effects

How substantial are the undesirable anticipated effects?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
○ Trivial ○ Small ○ Moderate ○ Large ● Varies ○ Don't know	**Serious Adverse Events:** Below is a tabulated description of the serious adverse events reported by the trials:	

Outcomes	No of studies reporting	Total events in stem cell arm	Total events in conventional arm	Risk ratio (95 % CI)
Any cause mortality	12	32	35	0.83 (0.54 to 1.28)
Recurrent stroke	7	11	7	0.95 (0.42 to 2.14)
Infection	7	52	43	0.89 (0.64 to 1.24)
Seizure	5	11	7	0.84 (0.39 to 1.81)
Worsening of neurological deficits	4	11	5	2.09 (0.80 to 5.46)
Development of any neoplasm	5	0	4	0.20 (0.03 to 1.11)
Recurrent vascular events	4	13	4	1.85 (0.67 to 5.08)

Certainty of evidence

What is the overall certainty of the evidence of effects?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
● Very low ○ Low ○ Moderate ○ High ○ No included studies	The evidence in this review is of very low certainty due to high risk of bias, inconsistency and imprecision in the effect shown by stem cell therapy.	

Values Is there important uncertainty about or variability in how much people value the main outcomes?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Important uncertainty or variability ○ Possibly important uncertainty or variability ● Probably no important uncertainty or variability ○ No important uncertainty or variability	Main outcome is clinical improvement in the form of increased activity of daily living and decreased disability which is likely to be highly valued by most persons.[19]	
Balance of effects Does the balance between desirable and undesirable effects favor the intervention or the comparison?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Favors the comparison ○ Probably favors the comparison ● Does not favor either the intervention or the comparison ○ Probably favors the intervention ○ Favors the intervention ○ Varies ○ Don't know	It is less clear if benefits of stem cell intervention outweigh the harms, based on limited evidence.	

Resources required How large are the resource requirements (costs)?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
● Large costs ○ Moderate costs ○ Negligible costs and savings ○ Moderate savings ○ Large savings ○ Varies ○ Don't know	No direct evidence of the resources required in stem cell transplantation in patients with stroke was identified. The information provided by the websites of Insurance Agencies and third party agencies is as follows:[20,21,22] • 3.2-4.8 lacs (Medaccess) • 5000-50,000 USD However, indirect evidence shows that the median cost of hematopoietic stem cell transplantation for hematological diseases was USD, $ 12,500 (range $ 10,331–39,367) - Rs. 10,00,000.[23]	
Certainty of evidence of required resources What is the certainty of the evidence of resource requirements (costs)?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Very low ○ Low ● Moderate ○ High ○ No included studies	No research evidence was identified.	The GDG members were fairly certain that stem cell therapy requires large resources.
Cost effectiveness Does the cost-effectiveness of the intervention favor the intervention or the comparison?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**

○ Favors the comparison ● Probably favors the comparison ○ Does not favor either the intervention or the comparison ○ Probably favors the intervention ○ Favors the intervention ○ Varies ○ No included studies	Despite significant progress in clinical research in the use of SCT in neurological diseases, economic evaluation of these therapies is still at a nascent stage.[24]	The intervention was not found to be effective and hence the committee deferred to comment on cost effectiveness.
Equity What would be the impact on health equity?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Reduced ● Probably reduced ○ Probably no impact ○ Probably increased ○ Increased ○ Varies ○ Don't know	No research evidence was identified.	As stem cell therapy is an expensive treatment offered only at tertiary centers, it is likely to reduce equity.
Acceptability Is the intervention acceptable to key stakeholders?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**

○ No ○ Probably no ● Probably yes ○ Yes ○ Varies ○ Don't know	Despite uncertainty regarding the medical risks and benefits associated with stem cell injections, stroke survivors may still consider undergoing treatment in private, unregulated clinics.[25]	

Feasibility

Is the intervention feasible to implement?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
○ No ○ Probably no ● Probably yes ○ Yes ○ Varies ○ Don't know	No research evidence was identified.	Feasible to implement in tertiary care centers.

SUMMARY OF JUDGEMENTS

	JUDGEMENT						
PROBLEM	No	Probably no	Probably yes	**Yes**		Varies	Don't know
DESIRABLE EFFECTS	**Trivial**	Small	Moderate	Large		Varies	Don't know
UNDESIRABLE EFFECTS	Trivial	Small	Moderate	Large		**Varies**	Don't know
CERTAINTY OF EVIDENCE	**Very low**	Low	Moderate	High			No included studies

VALUES	Important uncertainty or variability	Possibly important uncertainty or variability	**Probably no important uncertainty or variability**	No important uncertainty or variability			
BALANCE OF EFFECTS	Favors the comparison	Probably favors the comparison	**Does not favor either the intervention or the comparison**	Probably favors the intervention	Favors the intervention	Varies	Don't know
RESOURCES REQUIRED	**Large costs**	Moderate costs	Negligible costs and savings	Moderate savings	Large savings	Varies	Don't know
CERTAINTY OF EVIDENCE OF REQUIRED RESOURCES	**Very low**	Low	**Moderate**	High			No included studies
COST EFFECTIVENESS	Favors the comparison	**Probably favors the comparison**	Does not favor either the intervention or the comparison	Probably favors the intervention	Favors the intervention	Varies	No included studies
EQUITY	Reduced	**Probably reduced**	Probably no impact	Probably increased	Increased	Varies	Don't know
ACCEPTABILITY	No	Probably no	**Probably yes**	Yes		Varies	Don't know
FEASIBILITY	No	Probably no	**Probably yes**	Yes		Varies	Don't know

TYPE OF RECOMMENDATION

Strong recommendation against the intervention ○	Conditional recommendation against the intervention ○	**Use only in the context of rigorously conducted RCTs** ○	Conditional recommendation for either the intervention or the comparison ○	Conditional recommendation for the intervention ○	Strong recommendation for the intervention ○

CONCLUSIONS

Recommendation

Stem cell therapy is **not recommended** in routine clinical practice for the treatment of stroke*. It may be used only in the context of rigorously conducted randomized controlled trials.

* The evidence comes from RCTs that included patients with ischemic stroke only. Whether stem cell therapy can be used in patients with haemorrhagic stroke is not known as there are no RCTs in patients with haemorrhagic stroke.

Justification

Overall justification

This recommendation has been made as there is very low certainty evidence of trivial reduction in mortality and trivial improvement in functional and disability scale. The undesirable effects are variable and heterogenous. In addition, there is uncertainty on the long-term safety of stem cell therapy in patients with stroke. Results should be interpreted with caution, in view of various study limitations like high risk of bias, small number of participants and/or events in the included studies and different sources of stem cell use.

Detailed justification

Desirable Effects

This judgment was made as there is very low certainty evidence of trivial reduction in mortality and trivial improvement in function and disability.

Undesirable Effects

This judgment was made as the undesirable effects are variable and heterogenous. Also, the follow up period of one year is too small to comment on the side effect profile and long-term safety is not known.

Certainty of evidence

Very Low certainty of Evidence

Resources required

The committee opined that stem cell treatment is associated with large costs.

vii. Data Extraction:

To ensure accuracy in data extraction, a standardized data extraction sheet was developed using Microsoft Excel. This sheet captured the essential information from each eligible study, including the first author's name, year of publication, study site, study design, study population, sample size, participant characteristics (age, gender), type of stroke, type of stem cell, route of stem cell administration, the dose of stem cell, time for stem cell administration (Post-stroke), follow up duration, outcomes related to neurological function, and safety profile. For the continuous variable, mean and standard deviation was extracted. In case, the data is provided in the standard error, the same was converted to standard deviation using the formula SD= SE x $\sqrt{n}$. For the SD was calculated from confidence interval (CI) using the formula SD = $\sqrt{N}$ x (Upper limit- lower limit)/ 3.92. Data from graphs were extracted using Plot digitizer. For the modified Rankin scale (mRS), the data were extracted from the shift analysis figures of mRS outcomes in relevant studies. The values were analyzed with Microsoft excel to calculate the mean and SD. Two authors independently extracted the data from the included studies. In cases of disagreement, a consensus was reached through discussion. For incomplete data, the corresponding authors of the articles were contacted.

For continuous outcomes, the mean difference (MD) and 95% confidence interval (CI) were calculated for studies using same scales and the standardized mean difference (SMD) with 95% CI was used if different scales were used. For dichotomous outcomes, RR and 95% CI was calculated. The unit of analysis was the participants. I^2 statistics was used to measure the heterogeneity. Fixed effect model was used if heterogeneity was less than 50%, otherwise random effects model was used. Meta-regression and sub-group analysis were conducted to examine the source of heterogeneity. For the primary outcomes, the pre specified subgroup analysis considered were: 1) phase of the disease: acute (within 7 days of onset of stroke), subacute (between 7-30 days) or chronic (more than 30 days of onset of stroke), 2) Types of treatment (types of stem cells) and 3) route of administration (Intravenous, intrarterial, intracerebral). Sensitivity analysis was planned to assess the robustness of the pooled effects of the included studies. Additionally, funnel plot was applied to evaluate potential publication bias of the main results of the included studies. All statistical analyses were conducted using RevMan Version 5.4 with a significance level set at $p < 0.05$ (two-sided).

Study	**1- Bang et al. 2005[1]**	**2-Bhatia et al. 2018[2]**	**3-Celis-Ruiz et al. 2022[3]**	**4-Chen et al. 2014[4]**
Study type	Single- blind, randomized, controlled Phase I/II clinical trial	Prospective, randomized, open-label, blinded–end point study	Phase IIa, pilot, prospective, double blind, placebo controlled RCT	randomized, single blind controlled phase II clinical trial
Number of participants	30	20	19	30
Countries and setting:	Korea, Hospital	India, Hospital	Spain, Hospital	Taiwan, Hospital

Duration of study Follow up (post intervention):	12 months	6 Months	24 months	12 months
Method of assessment of disease condition:	Diffusion weighted imaging	Clinically and on imaging conforming tothe MCA territory stroke	CT/MRI	Ischemic stroke confirmed by MRI
Subgroup analysis within study	No	No	No	No
Inclusion criteria:	• Observed within 7 days of the onset of symptoms; • Relevant lesions within the MCA territory as assessed using DWI and • NIHSS score of 7 or more points after 7 days of admission	• Age 20 to 80 years • Symptoms and signs of clinically definite MCA stroke (0 –14 days postictus), • NIHSS score >7, • MCA territory stroke, • Recanalization/patency of the involved M1segment of the MCA on imaging, • Patency of the carotid arteries for intra-arterial access of cerebral circulation.	• Moderate to severe ischemic stroke (NIHSS 8-20) • Presenting within 24 hours • Ct/MRI compatible with ischemic stroke in MCA	• Age 35 to 75, • Onset of stroke between 6 months to 5 years, • NIHSs 9-20 with motor deficits stable for 3 months
Exclusion criteria:	• Lacunar syndrome, hematological causes of stroke, malignant diseases, severe comorbidity, hepatic or renal dysfunction, or unwillingness toparticipate.	• Cerebral hemorrhage on CT/MR imaging, imaging evidence of M1 MCA segment complete occlusion, hemodynamic instability, known defect of clotting or platelet function, severe comorbidity precluding intra-	• Comatose patient, • evidence of brain tumor, primary intraventricular, intracerebral, or subarachnoid hemorrhage on neuroimaging;cerebral edema with midline shift; or cerebellar or	Patients with cancer, infection, increased bleeding tendency, leukopenia, thrombocytopenia, hepatic/renal dysfunction, or pregnancy

		arterial intervention, hepatic dysfunction, renal dysfunction, pregnancy, patients likely to be unavailable for follow-up, patients with evidence of chronic illness or advanced cancer, patients already dependent in activities of daily living before the present acute stroke (i.e., prestrike mRS of >3), and refusal to give informed consent.	brainstem infarction, • Clinical or laboratory signs suggestive of active infection, including human immunodeficiency virus and hepatitis B and C, were also considered exclusion criteria, along with a previous diagnosis of dementia or any health condition that could preclude appropriate diagnosis, treatment, or follow-up	
Recruitment/selection of patients:	Random allocation by randomization table	Randomization done, but details not available	Computer-generated, simple randomization	Participants were assigned randomly (1:1) via SAS software
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Intravenous infusion of 2 boosts of 50 million culture-expanded autologous bone marrow mesenchymal stem cells. Cells had high expression of SH antigens	Intra-arterial infusion of bone marrow–derived mononuclear cells (mean 6.1 million) into the ipsilateral MCA	Intravenous administration of allogeneic mesenchymal stem cells from adipose tissue (1 million / kg) within the first 2 weeks after the onset of stroke symptoms	Subcutaneous granulocyte-colony stimulating factor injections (15 μg/kg/day) for 5 consecutive days, followed by stereotaxic implantation of autologous 3 to 8 million $CD34^+$ immunosorted peripheral blood stem cells
Outcomes reported with time points	Functional outcome assessed at 7 days, 3, 6	The primary safety end point was clinically severe	Primary outcomes: safety parameters.	Improvements in stroke scales (NIHSS, European

	and 12 months Imaging (MRI): at 12 months	procedural complications (increase in the NIHSS score of >4 points), symptomatic intracerebral hemorrhage, new ischemic lesions on imaging in the intervened territory, or death. Secondary end point (good clinical outcome) was defined as an mRS score of 0 –1 in patients with an NIHSS score of 8 –14 at admission and an mRS score of 0 –2 in patients with an NIHSS score of >14 at admission	Secondary outcomes: efficacy analysis mRS, NIHSS, size of infarct, and biochemical markers. Time points: Day 7, 3, 6, 12, 18 and 24 months	Stroke Scale, and European Stroke Scale Motor sub scale) and functional outcomes measure (mRS) from baseline Timepoints: 6-month sand 12 months
Funding	Korea Research Foundation and Korea Health 21 R&D	Not available	Spanish ministry of Health and social policy	Taiwan department of health clinical trial center of excellence and Taiwan national science council
RoB2 Assessment: Specify separately for each domain	D1-High Risk D2-High risk D3- Some concerns D4-Low risk D5-Some concerns Overall-High risk	D1-Some concerns D2-Low risk D3-Low risk D4-Low risk D5-Some concerns Overall-Some concerns	D1- Low risk D2-High risk D3-Low risk D4-Low risk D5-High Risk Overall-High risk	D1-Low risk D2-Low risk D3-Low risk D4-Low risk D5-Low risk Overall-Low risk

Study	**Fang et al. 2018[5]**	**Hess et al. 2017[6]**	**Houkin et al. 2024[7]**	**Jaillard et al. 2020[8]**
Study type	Single-blinded, randomized, parallel,	Double-blind, randomized, placebo-controlled, phase II	Multicenter, double-blind, parallel group,	Single center, Open label randomised control trial

	placebo-controlled trial	multicenter dose-escalation clinical trial	placebo controlled RCT (Phase 2/3)	with blinded outcome evaluation
Number of participants	18	129	207	31
Countries and setting:	China, Hospital	UK and USA, Hospital	Japan, Hospital	France, Hospital
Duration of study Follow up (post intervention):	24 months	12 months	12 months	24 months
Method of assessment of disease condition:	Ischemic stroke assessed with diffusion weighted imaging	Hemispheric cortical infarct in the anterior circulation with brain MRI including diffusion weighted imaging	Acute infarct determined by DW-MRI	MRI (DWI and FLAIR)
Subgroup analysis within study	No	No	No	Yes (Low dose and high dose)- for safety outcomes)
Inclusion criteria	• Patients aged 18–80 years, within 7 days of onset of symptoms; • Ischemic lesion withinthe middle cerebral artery territory as assessed usingdiffusion-weighted imaging; 3- NIHSS score ≥7 at day 7 after onset; and 4-signed informed consent	• Age 18-83 years, with NIHSS score 8-20 at baseline, 2-hemispheric cortical infarct in the anterior circulation with brain MRI including diffusion weighted imaging showing an acute lesion measuring more than 5 mL and less than 100 mL., 3-Treatemnt window within 24-48 hours	• Age >20 ye, 2- NIHSS 8-20 at baseline, 3-acute infract involving cerebral cortex and measuring more than 2 cm, 4-mRS score of 0-1 before stroke onset.	• Age 18-70 years, 2-NIHSS >7, 3- Ischemic stroke of less than 2 weeks, 4- mRS score of 0-1 before stroke onset, 5- Wiling and able to follow a rehabilitation programe.
Exclusion criteria:	• Lacunar syndrome; • Diagnosis other than ischemic	• If there was a change in NIHSS score of four or more points during at	• Lacunar or brainstem infarction,	• Very severe stroke with possible fatal outcome, • NIHSS>24,

	stroke (e.g., intracranial hemorrhage or intracranial tumor); • Hematological causes of stroke; • Severe respiratory, hepatic, or renal disorders; • presence of severe febrile illness or viral diseases; • Malignant diseases; • Presence of autoimmune diseases; • Positive response of penicillin skin test, or multiple drug allergies; and • Breast-feeding or pregnant women	least a 6 h period between screening and randomization • Brainstem or lacunar infarct; • Substantial comorbid disease such as severe congestive heart failure, chronic obstructive pulmonary disease, or renal or hepatic failure; • An inability to undergo an MRI scan, and • With a history of splenectomy	• Change in NIHSS score of 4 or greater during a minimum period of 6 hours between screening and randomization, and • Patients who received combined reperfusion therapy (intravenous thrombolysis plus mechanical thrombectomy)	• Baseline mRS>2, • History of myocardial infract in less than 3 months, • Severe psychiatric illness, • History of malignant diseases, chemotherapy, thromboembolic disorder within 6 months, severe kidney and liver disease, • Contraindication to MRI, • Life expectancy less than 3 months
Recruitment/selection of patients:	Randomization was done using layered segment randomization.	Computer-generated randomization, interactive voice and web-response system	Computer-generated randomization process	Randomization done with 'Clininfo' program
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Two cycles 2.5 million cells/kg of autologous EPCs or BMSCs were transplanted intravenously, 1 week	Intravenous infusion of 1200 million multipotent progenitor cells (in a solution containing PlasmaLyte-A, dimethyl	Single intravenous infusion of MultiStem over 30 to 60 min within 18- 36 hours of stroke onset.	Two different doses via Intravenous infusion. The first ten patients assigned to treatment received low-dose MSCs (100 million)

	apart	sulfoxide, and human serum albumin) over 1 hour	Single unit (6ml vials) of MultiStem contained 1.2 billion cells (±20%), cryopreserved in a medium of Plasma-Lyte A, dimethylsulfoxide and human albumin.	and the next ten patients received high-dose MSCs (300 million)
Outcomes reported with time points	Safety (adverse events, neurological detoriation, mortality) and efficacy by NIHSS, Scandinavian stroke scale, functional scales, Barthel Index and mRS Time points: 3,6,12 and 24 months	The primary efficacy endpoint combined the mRS, the BI, and change in NIHSS score from baseline. Time points: Day 7, 30, 90 and 12 months	Primary: Proportion of patients with excellent outcome at 90 days. (mRS ≤1, NIHSS ≤1, BI ≥95) Secondary: Excellent outcome at 365 days, distribution of mRS at 90 and 365, Proportion of patients with mRS ≤ 1 and ≤ 2 at 90 days. And Safety outcomes Timepoints: 3 and 12 months	Safety outcome (Adverse events) and efficacy outcomes (NIHSS, mRS and Barthel index) Time points: 6 months and 24 months
Funding	Special Foundation for Science and Technology Key Plan of Guangdong Province, Special Foundation for Science and Technology Key Plan of Guangzhou City; Clinical Research	Athersys, Inc (Cleveland, OH, USA)	Healios KK. Investigational products were provided by Athersys Inc	French health Ministry. Data analysis by RESSTORE project, funded by European commission under H2020 program.

	Project of Southern Medical University, Project of Outstanding Young Talent Pool of Zhujiang Hospital.			
RoB2 Assessment: Specify separately for each domain	D1-Low risk D2-Some concerns D3-Low risk D4-Low risk D5-High risk Overall- High risk	D1-Low risk D2-Low risk D3-Low risk D4-Low risk D5-High risk Overall- High risk	D1-Low risk D2-Low risk D3-Low risk D4-Low risk D5-Low risk Overall- Low risk	D1-Low risk D2-Some concerns D3-Low risk D4-Low risk D5-High risk Overall- High risk

Study	**Jin et al. 2017[9]**	**Law et al. 2021[10]**	**Lee et al. 2010[11]**	**Moniche et al. 2023[12]**
Study type	Randomized controlled clinical trial	Single center, assessor-blinded randomized controlled, Phase 2 trial	Single-blind, randomized, controlled clinical trial	Multicenter, open-label, phase 2 randomized controlled trial
Number of participants	20	17	85	77
Countries and setting:	China, Hospital	Malaysia, Hospital	South Korea, Hospital	Spain, Hospital
Duration of study Follow up (post intervention):	7 years	12 months	5 years	6 months
Method of assessment of disease condition:	MRI	Infarct on MRI or CT scan	MRI-DWI (Diffusion weighted Imaging)	MRI-DWI (Diffusion weighted Imaging)
Subgroup analysis within study	No	No	No	No
Inclusion criteria:	• Diagnosed ischemic stroke between 3 weeks and 3	• Age 30-75years, • Stroke onset within2 months,	• Age 30-75 years, • Observed within 7 days of the onset of	• Age 18–80 years, • NIHSS score of 6–20 at inclusion with at least 2

	months prior, • NIHSS score between 5 and 30	• Did not receive or failed intravenous thrombolytic therapy, • NIHSS score between 10-35. • Evidence of unilateral MCA infarct	symptoms, • Lesions within the middle cerebral artery territory as assessed using diffusion-weighted imaging (DWI); • Modified NIH Stroke Scale [mNIHSS]≥ 7 pointson the seventh admission day with stable course; and • No hematologic disorders or bone marrow suppression	points on motor items in the NIHSS or aphasia, and • Treatment window within 7 days of stroke onset.
Exclusion criteria:	• History of malignancy, serious cardiopulmonary insufficiency, or liver or kidney dysfunction, 2-patients who refuse to be treated	• Patients who were medically unfit, with worsening consciousness; • Brain tumor or other space-occupying lesion on brain MRI; • Transient ischemic attacks or lacunar infarct; • Suffered from infections, malignancy and/or primary hematological disorders; • Renal impairment, with serum creatinine >200 µmol/L or creatinine	• Lacunar infarction, • Premorbid dependence; • Hematologic cause of stroke; • Presence of severe medical illness; • Presence of severe febrile illness; • Hepatic or renal dysfunction; • Positive response of penicillin skin test; and • Unwillingness to participate	• Patients with lacunar or haemorrhagic stroke, • Pregnancy, • Childbearing potential, • Malignant disease during the past 5 years, • Life-threatening illness, • Stroke of haematological cause, • Significant previous disability (prestroke modified Rankin scale [mRS] score ≥3), and • Severe comorbidity that would prevent follow-up

		clearance<30 ml/min, • Liver impairment, with serum aspartate transaminase and alanine transaminase four times greater than the normal upper limit; • Other comorbidities that would be deemed contraindications to BMMSC transplantation therapy or bone marrow aspiration (BMA), • Comorbidities that affected the ability to obtain adequate stem cells, such as chronic debilitating diseases, frailty or known osteoporosis; or • Had contraindications to brain MRI.		
Recruitment/selection of patients:	Patients were randomized but the method mentioned	Permuted block randomization	Participants were randomly allocated by a blinded, independent coordinator, using a randomization table	Randomization done by a central computerbased procedure
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Subarachnoid infusion of a cell suspension containing 10 million autologous bone marrow mononuclear cells	Autologous bone marrow derived mesenchymal stem cells administered intravenously at a mean dosage of 2.05 million cells per kg	Intravenous infusion of 2 boosts of 50 million culture-expanded autologous bone marrow mesenchymal stem cells	Autologous Bone marrow mononuclear cells transfused via intra-arterial route in two different dose (2 million cells/kg and 5 million cells/kg)

Outcomes reported with time points	Various safety and efficacy outcomes (Barthel Index, mRS, Functional measure and Fugl-Meyer motor score) Time points: 3, 6, 12 months, 2, 3, 4, 5, 6, 7th year	Efficacy outcomes by NIHSS, mRS and BI scores. Timepoints: 45 days, 90 days, 6, 9 and 12 months	Primary: Long term safety outcome, mortality and serious adverse effects. Secondary: Functional outcome (mRS) at last visit Time points: Till last visit (Median 3.5 year, IQR 2.9-4.6 years)	Efficacy (mRS, NIHSS, and Barthel Index) and safety outcomes Time points: 3 and 6 months
Funding	Not available	National University of Malaysia and Cytopeutics Sdn Bhd.	Korea Health 21 R&D Project, Ministry of Health and Welfare, Republic of Korea and SBRI C-A9-216-1	Instituto de Salud Carlos III co-funded by the European Regional Development Fund/European Social Fund, Mutua Madrileña, and the Regional Ministry of Health of Andalusia
RoB2 Assessment: Specify separately for each domain	D1-Some concerns D2-Low risk D3-Low risk D4-Some concerns D5-High risk Overall- High risk	D1- Some concerns D2-Low risk D3-Low risk D4- Low risk D5-Some concerns Overall- Some concerns	D1-High risk D2-Some concerns D3-Low risk D4- Low risk D5-Some concerns Overall- High risk	D1-Low risk D2-Low risk D3-Low risk D4- Low risk D5-Low risk Overall- Low risk

Study	**Niizuma et al. 2023[13]**	**Prasad et al. 2014[14]**	**Savitz et al. 2019[15]**
Study type	Single center, Phase 2, randomized, double blind controlled trial	Single-blind, randomized, controlled clinical trial	Randomized, sham controlled, Parallel group, multicenter Phase 2 trial
Number of participants	37	60	48
Countries and setting:	Japan, Hospital	India, Hospital	USA, Hospital

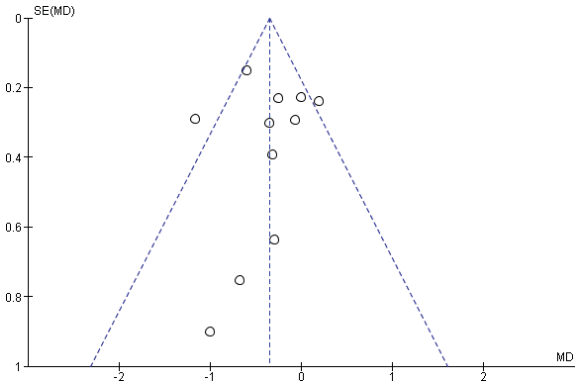
Duration of study Follow up (post intervention):	52 weeks	1 year	1 year
Method of assessment of disease condition:	MRI	CT or MRI	CT or MRI
Subgroup analysis within study	No	Yes	No
Inclusion criteria:	• Age 20-80 years, • NIHSS scale >6, • mRS score ≥ 3 at enrollment, • An mRS score of 0 or 1 before ischemic stroke (patient/family-reported), and • Who could receive study treatment 14–28 days after stroke onset	• Age 18-75 years, • CT or MRI scan showing relevant infarct within the middle cerebral artery or anterior cerebral artery territory and excluding a hematoma, • Onset of stroke between 7 and 8, • BI score of ≤50, 5-NIHSS score of ≥7 and inability to walk unaided or raise upper limb by 90° and • Clinically stable condition ≥48 hours	• Age 30-83 years, • First time MCA ischemic stroke, • Persistent neurological deficits and mRS ≥3 at randomization, which occurred between 9 and 15 days after the stroke. • Demonstrated vessel patency with flow in the ipsilateral carotid artery (proximal internal carotid artery [ICA]<50% stenosis or proximal MCA occlusions if there was collateral circulation to the injured area
Exclusion criteria:	• Decreased consciousness, • Clinically important hemorrhagic transformation by CT or MRI; • Cognitive impairment that might interfere with evaluation; • History of thromboembolism (excluding previous stroke), • Systolic blood pressure >200 mm Hg or diastolic	• Lacunar syndrome, • Intubation, 3 • Posterior circulation stroke, • Co-morbidity likely to limit survival to less than 3 years, • Pre-stroke disability leading to dependence on others for activity of daily living, • In accessibility for follow up, • Allergy to local anaesthetic, • Unwillingness to provide	• Patients unable to undergo the bone marrow harvest, return • Days after harvest for intracarotid infusion, • Unable to undergo angiography • Patients with clinically significant intracranial hemorrhage, • Patients with previous disability with mRS ≥1

	blood pressure >120 mm Hg, • Current or previous malignancy, • Collagen disease or related diseases, • Diabetes with glycated hemoglobin A1c >10%, • Active infections requiring treatment, • Severe complications, • Current use of systemic steroids or immunosuppressants, • Positive for hepatitis B virus surface antigen, hepatitis C virus antibody, human immunodeficiency virus antibody, human Tcell leukemia virus type 1 antibody, or syphilis; • Pregnant or breastfeeding; or • History of hypersensitivity to aminoglycoside antibiotics, human serum albumin preparations, or proteins of bovine or porcine origin	written informed consent, • Symptoms suggestive of acute cardiac, • Hepatic or renal disease, • Pregnancy, • HIV positivity, or • Participation in any other trial.	before stroke. • Patients who could not be temporally discontinued from systemic anticoagulation for the bone marrow harvest
Recruitment/selection of patients:	Patients were randomized by Web response system was used for sequence allocation method by minimization method.	Participants were randomly assigned by a central computer, using permuted block randomization	Central randomization scheme conducted by PharPoint Inc
Intervention: Type of stem cells with method of their	Allogenic muse cell-based product (CL2020), 15 million	Intravenous infusion of a mean of 280 million autologous bone	Single internal carotid infusion of 3.08 million of autologous

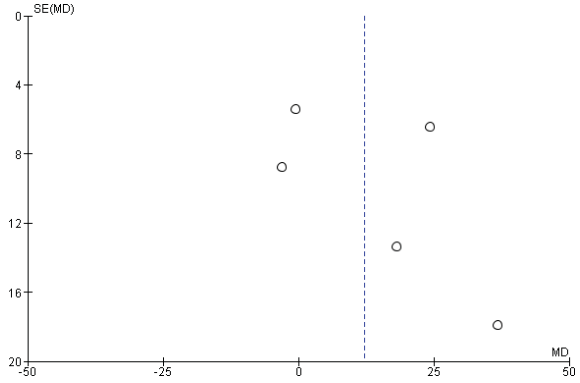
characterization, Route of administration, Dose	cells/15 ml were administered intravenously	marrow mononuclear stem cells	Bone-marrow derived ALD-401 cells.
Outcomes reported with time points	Safety and efficacy (mRS, NIHSS, Barthel Index, EU- 5D-5L) Time points: Outcomes assessed every 4 weeks till 52 weeks	Primary efficacy measurese.gmRS and BI at 6 months and Secondary outcome of NIHSS at 1 year. Safety outcome included death, adverse events, epileptiform discharges and any new growth in PET scan. Time points: 6 months and 1 year	Safety of delivery of ALD-401 cells and efficacy as measured by mRS, Secondary outcomes were NIHSS, Barthel Index and EQ-5D at 3 months
Funding	Life Science Institute, Inc., (manufacturer of CL2020)	Department of Biotechnology, Ministry of Science and Technology, Government of India	Aldagen, Inc, a wholly owned subsidiary of Cytomedix, Inc. Partial Funding by a grant from the Robertson Foundation to Duke University.
RoB2 Assessment: Specify separately for each domain	D1-Low risk D2-Some concerns D3-Low risk D4-Low risk D5-Low risk Overall-Some concerns	D1-Low risk D2-Low risk D3-Low risk D4-Low risk D5-Low risk Overall-Low risk	D1-Low risk D2-Low risk D3-Low risk D4-Low risk D5-High risk Overall-High risk

viii. Publication bias

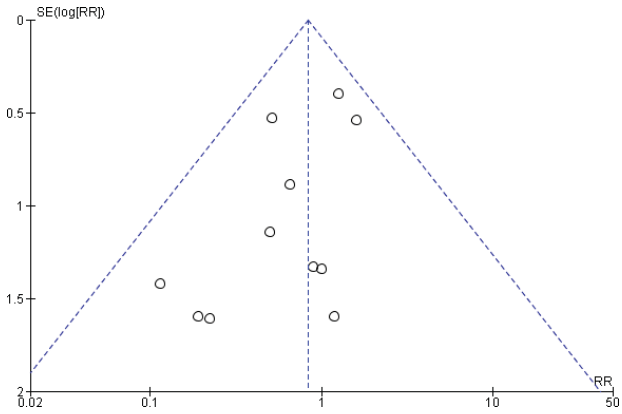
Funnel plot: Modified Rankin Scale



Funnel plot: Barthel Index



Funnel plot: All-cause mortality



ix. List of Excluded Studies:

Reference No	Study ID	Brief summary	Reason for exclusion
Studies on Intracerebral haemorrhage (ICH)			
1	Sobrino et al. 2011	CD341 progenitor cells in the outcome of intracerebral haemorrhage at 3 months	Not RCT
2	Li et al. 2013	Efficacy of mesenchymal stem cell (MSCs) in patients with intracerebral haemorrhage. Outcomes assessed at 6 months	Not RCT
3	Zhu et al. 2015	Surgery Combined with autologous Bone Marrow Stromal Cell Transplantation in Intracerebral Hemorrhage. Efficacy outcomes assessed at 12 months	Not RCT
4	Tsang et al. 2017	Autologous BM derived mesenchymal stem cell in chronic stroke (ICH), phase I/II RCT. Outcome assessed at 60 weeks	Efficacy parameters of two groups not published.
5	Khalil et al. 2023	Intravenous mesenchymal stem cell in chronic ICH patients. RCT. Outcome assessed at 2 years.	Outcomes of control arm not published.
Studies with both intracerebral haemorrhage and Ischemic stroke			
6	Kondoziolka et al. 2005	Stereotactic implantation of cultured neuronal cells in chronic ischemic and haemorrhagic stroke. Outcome assessed at 6 months Phase 2 RCT.	Separate results for ischemic and haemorrhagic patients not available.
Studies in Ischemic stroke			
7	Meng et al. 2009	Autologous marrow mesenchymal cell transplantation in ischemic stroke. RCT	Full test not available. Article in Chinese
8	Battistella et al. 2011	Intra-arterial transplantation of autologous BM mononuclear cells in subacute stroke. Outcome assessed at 6months	Not RCT
9	Prasad et al. 2012	Intravenous autologous BM mononuclear cell in ischemic stroke. Phase 1 study	Not RCT

10	Moniche et al. 2012	Intra-Arterial Bone Marrow Mononuclear Cells in Ischemic Stroke. Efficacy outcome assessed at 6 months	Not RCT
11	Bhasin et al. 2012	Autologous intravenous mononuclear stem cell in chronic ischemic stroke. Outcome assessed at 6 months	Not RCT
12	Jiang et al. 2013	Mesenchymal stem cell delivery via catheter in MCA territory stroke. Outcome assessed at 6 months	Not RCT
13	Qiao et al. 2014	Co-transplantation With Neural Stem/Progenitor Cells and Mesenchymal Stromal Cells in Ischemic Stroke Patients, outcome assessed at 2 years.	Not RCT
14	Liu et al. 2014	Autologous mesenchymal nerve stem cells in subacute stroke. RCT. Outcomes assessed at 3 months.	Outcomes assessed at less than 6 months (3 months)
15	Banerjee et al. 2014	Intra-arterial Immunoselected CD 34 + stem cells in acute ischemic stroke.	Not RCT
16	Wang et al. 2014	Intravenous autologous marrow mesenchymal cells in stroke. RCT	Full text not available. Article in Chinese.
17	Wanamaker et al. 2015	Surgical mesenchymal stem cell implantation in patients with MCA stroke. Outcomes asses at 1 year.	Not RCT, Radiological outcome
18	Taguchi et al. 2015	Intravenous BM Mononuclear stem cell transplantation in acute stroke. Phase 1.2b trial	Not RCT
19	Nakazaki et al. 2015	Mesenchymal stem cell in cerebral ischemia. Phase 3 study	Full test not available. Article in Japanese.
20	Ghali et al. 2016	Intra-arterial infusion of autologous BM Mononuclear stem cells in subacute stroke. Outcomes assessed at 12 months.	Not RCT
21	Bhasin et al. 2016	Intravenous BM derived mononuclear stem cells in chronic ischemic stroke. Single blind RCT. Outcome assessed at 8 weeks.	Outcome assessed at less than 6 months. (8 weeks)

22	Kalladka et al. 2016	Human neural stem cell line product CTX-DP, in chronic ischemic stroke. Phase 1 study.	Not RCT
23	Honmou et al. 2016	Intravenous autologous mesenchymal cells in subacute stroke patients.	Full test not available. Article in Japanese
24	Bhasin et al. 2017	Autologous mesenchymal stem cell in chronic stroke. Outcome at 4 years.	Not RCT
25	Steinberg et al. 2018	Modified BM derived mesenchymal stem cells (SB 623) in chronic stroke. Phase 1/2a study.	Not RCT
26	Laskowitz et al. 2018	Allogenic umbilical cord blood in ischemic stroke, Phase 1 study	Not RCT
27	Levy et al. 2019	Intravenous allogenic mesenchymal stem cell in chronic stroke, Phase I/II study.	Not RCT
28	Zhang et al. 2019	Intracerebral transplantation of neural stem cell in chronic stroke. Outcome assessed at 12 months.	Not RCT
29	Vahidy et al. 2019	Intravenous BM mononuclear cell in acute ischemic stroke. Phase 1 trial	Not RCT
30	Muir et al. 2020	Intracerebral implantation of human neural stem cells in chronic stroke. Outcomes assessed at 12 months.	Not RCT
31	Chung et al. 2021	Intravenous Mesenchymal stem cells in chronic stroke. Open labelled RCT. Outcomes assessed at 3 months	Outcomes assessed at less than 6 months (3 months)
32	Lee et al. 2021	Intravenous mesenchymal stem cells in subacute ischemic stroke. Open labelled RCT. Outcome assessed at 3 months.	Outcomes assessed at less than 6 months (3 months)

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2. SPINAL CORD INJURY

i. **Key question in PICO format**

ii. **Search Strategy**

iii. **PRISMA Flow Diagram**

iv. **Summary of included studies**

v. **Classification of studies based on level of stem cell manipulation**

vi. **Evidence to Decision Framework**

vii. **Data Extraction**

viii. **List of excluded studies**

i. Key Question in PICO format:

In patients with Spinal cord injury, what is the efficacy and safety of stem cell therapy as compared to usual care?

Population: Patients with spinal cord injury- subgroups-acute/subacute/chronic
Intervention: Any Stem cell and product derived from stem cells
Comparator: Usual Care/ Conventional Care
Critical Outcomes: Efficacy: Any measure of Disability or Handicap, Quality of life, Bladder and bowel function;
Safety outcomes: mortality, serious adverse events, Any Tumour formation;
Important: other adverse events

ii. Search Strategy

PubMed

No.	Query
#6	((stem AND cell AND therapy OR 'stem cell therapy'/exp/mj OR 'mesenchymal stem cells':lnk OR 'bone marrow stem cells':lnk OR 'mesenchymal stem cell' OR 'bone marrow stem cell' OR 'stem cell transplantation') AND 'chronic spinal cord injury' OR 'chronic spinal cord injury'/exp) AND [randomized controlled trial]/lim AND [<1966-2023]/py AND [embase]/lim AND [humans]/lim AND [abstracts]/lim AND [clinical study]/lim AND [01-01-1900]/sd NOT [18-11-2023]/sd
#5	((stem AND cell AND therapy OR 'stem cell therapy'/exp/mj OR 'mesenchymal stem cells':lnk OR 'bone marrow stem cells':lnk OR 'mesenchymal stem cell' OR 'bone marrow stem cell' OR 'stem cell transplantation') AND 'acute spinal cord injury' OR 'acute spinal cord injury'/exp) AND [randomized controlled trial]/lim AND [<1966-2023]/py AND [embase]/lim AND [humans]/lim AND [abstracts]/lim AND [clinical study]/lim AND [01-01-1900]/sd NOT [18-11-2023]/sd
#4	((stem AND cell AND therapy OR 'stem cell therapy'/exp/mj OR 'mesenchymal stem cells':lnk OR 'bone marrow stem cells':lnk OR 'mesenchymal stem cell' OR 'bone marrow stem cell' OR 'stem cell transplantation') AND 'spinal cord injury' OR 'spinal cord injury'/exp) AND [randomized controlled trial]/lim AND [<1966-2023]/py AND [embase]/lim AND [humans]/lim AND [abstracts]/lim AND [clinical study]/lim AND [01-01-1900]/sd NOT [18-11-2023]/sd
#3	((stem AND cell AND therapy OR 'stem cell therapy'/exp/mj OR 'mesenchymal stem cells':lnk OR 'bone marrow stem cells':lnk OR 'mesenchymal stem cell' OR 'bone marrow stem cell' OR 'stem cell transplantation') AND 'spinal cord injury' OR 'spinal cord injury'/exp) AND [randomized controlled trial]/lim AND [<1966-2023]/py AND [embase]/lim AND [humans]/lim AND [abstracts]/lim AND [clinical study]/lim
#2	((stem AND cell AND therapy OR 'stem cell therapy'/exp/mj OR 'mesenchymal stem cells':lnk OR 'bone marrow stem cells':lnk OR 'mesenchymal stem cell' OR 'bone marrow

	stem cell' OR 'stem cell transplantation') AND 'spinal cord injury' OR 'spinal cord injury'/exp) AND [randomized controlled trial]/lim AND [<1966-2023]/py AND [embase]/lim
#1	((stem AND cell AND therapy OR 'stem cell therapy'/exp/mj OR 'mesenchymal stem cells':lnk OR 'bone marrow stem cells':lnk OR 'mesenchymal stem cell' OR 'bone marrow stem cell' OR 'stem cell transplantation') AND 'spinal cord injury' OR 'spinal cord injury'/exp) AND [randomized controlled trial]/lim AND [<1966-2023]/py

Embase

No	Query
#6	((stem AND cell AND therapy OR 'stem cell therapy'/exp/mj OR 'mesenchymal stem cells':lnk OR 'bone marrow stem cells':lnk OR 'mesenchymal stem cell' OR 'bone marrow stem cell' OR 'stem cell transplantation') AND 'chronic spinal cord injury' OR 'chronic spinal cord injury'/exp) AND [randomized controlled trial]/lim AND [<1966-2023]/py AND [embase]/lim AND [humans]/lim AND [abstracts]/lim AND [clinical study]/lim AND [01-01-1900]/sd NOT [18-11-2023]/sd
#5	((stem AND cell AND therapy OR 'stem cell therapy'/exp/mj OR 'mesenchymal stem cells':lnk OR 'bone marrow stem cells':lnk OR 'mesenchymal stem cell' OR 'bone marrow stem cell' OR 'stem cell transplantation') AND 'acute spinal cord injury' OR 'acute spinal cord injury'/exp) AND [randomized controlled trial]/lim AND [<1966-2023]/py AND [embase]/lim AND [humans]/lim AND [abstracts]/lim AND [clinical study]/lim AND [01-01-1900]/sd NOT [18-11-2023]/sd
#4	((stem AND cell AND therapy OR 'stem cell therapy'/exp/mj OR 'mesenchymal stem cells':lnk OR 'bone marrow stem cells':lnk OR 'mesenchymal stem cell' OR 'bone marrow stem cell' OR 'stem cell transplantation') AND 'spinal cord injury' OR 'spinal cord injury'/exp) AND [randomized controlled trial]/lim AND [<1966-2023]/py AND [embase]/lim AND [humans]/lim AND [abstracts]/lim AND [clinical study]/lim AND [01-01-1900]/sd NOT [18-11-2023]/sd
#3	((stem AND cell AND therapy OR 'stem cell therapy'/exp/mj OR 'mesenchymal stem cells':lnk OR 'bone marrow stem cells':lnk OR 'mesenchymal stem cell' OR 'bone marrow stem cell' OR 'stem cell transplantation') AND 'spinal cord injury' OR 'spinal cord injury'/exp) AND [randomized controlled trial]/lim AND [<1966-2023]/py AND [embase]/lim AND [humans]/lim AND [abstracts]/lim AND [clinical study]/lim
#2	((stem AND cell AND therapy OR 'stem cell therapy'/exp/mj OR 'mesenchymal stem cells':lnk OR 'bone marrow stem cells':lnk OR 'mesenchymal stem cell' OR 'bone marrow stem cell' OR 'stem cell transplantation') AND 'spinal cord injury' OR 'spinal cord injury'/exp) AND [randomized controlled trial]/lim AND [<1966-2023]/py AND [embase]/lim
#1	((stem AND cell AND therapy OR 'stem cell therapy'/exp/mj OR 'mesenchymal stem cells':lnk OR 'bone marrow stem cells':lnk OR 'mesenchymal stem cell' OR 'bone marrow stem cell' OR 'stem cell transplantation') AND 'spinal cord injury' OR 'spinal cord injury'/exp) AND [randomized controlled trial]/lim AND [<1966-2023]/py

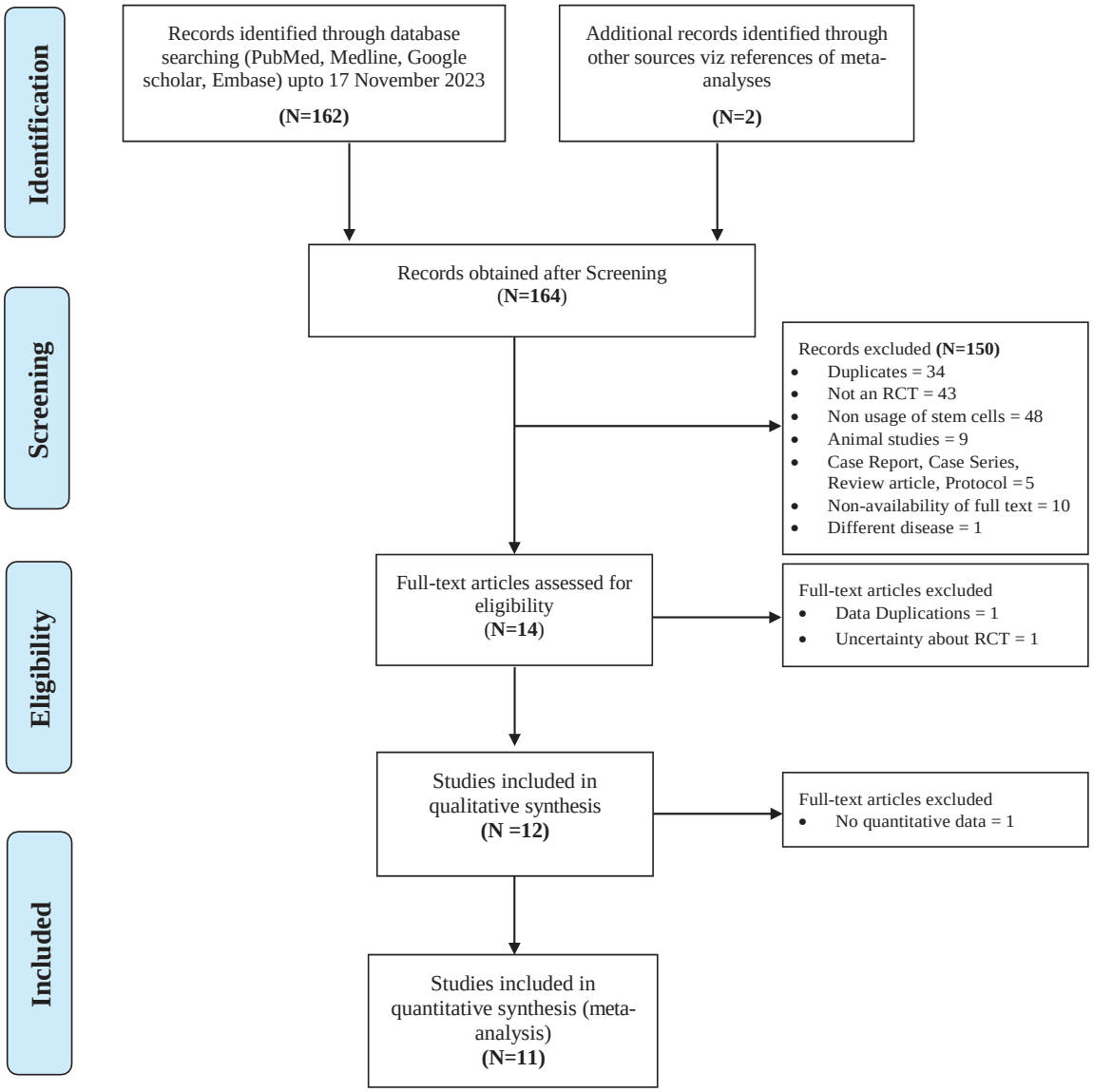
Web of Science

#	Search Query
1	(((((TS=(Stem cell therapy)) OR TS=(Mesenchymal stem cell)) OR TS=(Bone marrow stem cell)) AND TS=(Spinal Cord Injury)) OR TS=(Acute spinal cord injury)) OR TS=(Chronic spinal cord injury) and Preprint Citation Index (Exclude – Database) Timespan: 1900-01-01 to 2023-11-17
2	((((((TS=(Stem cell therapy)) OR TS=(Mesenchymal stem cell)) OR TS=(Bone marrow stem cell)) AND TS=(Spinal Cord Injury)) OR TS=(Acute spinal cord injury)) OR TS=(Chronic spinal cord injury)) Timespan: 1900-01-01 to 2023-11-17
3	(((((((TS=(Stem cell therapy)) OR TS=(Mesenchymal stem cell)) OR TS=(Bone marrow stem cell)) AND TS=(Spinal Cord Injury)) OR TS=(Acute spinal cord injury)) OR TS=(Chronic spinal cord injury))) AND TS=(randomized controlled trial) and Preprint Citation Index (Exclude – Database) Timespan: 1900-01-01 to 2023-11-17
4	(((((((TS=(Stem cell therapy)) OR TS=(Mesenchymal stem cell)) OR TS=(Bone marrow stem cell)) AND TS=(Spinal Cord Injury)) OR TS=(Acute spinal cord injury)) OR TS=(Chronic spinal cord injury))) AND TS=(randomized controlled trial) and Preprint Citation Index (Exclude – Database) and Clinical Trial (Document Types) Timespan: 1900-01-01 to 2023-11-17

Cochrane

ID	Search
#1	(Stem Cell Therapy) OR (Mesenchymal Stem Cell) OR (Bone Marrow Stem Cell) AND (Spinal Cord Injury) with Publication Year from 1900 to 2023, in Trials
#2	MeSH descriptor: [Stem Cell Transplantation] explode all trees
#3	MeSH descriptor: [Mesenchymal Stem Cell Transplantation] explode all trees
#4	MeSH descriptor: [Spinal Cord Injuries] explode all trees
#5	((Stem cell therapy) AND (Spinal cord injury)):ti,ab,kw (Word variations have been searched) with Publication Year from 1900 to 2023, in Trials
#6	((Stem cell therapy)) OR ("mesenchymal stem cell") OR ("bone marrow stem cell") AND ("spinal cord injury") OR (Acute Spinal Cord Injury):ti,ab,kw with Publication Year from 1900 to 2023, in Trials
#7	((Stem cell therapy)) OR ("mesenchymal stem cell") OR ("bone marrow stem cell") AND ("spinal cord injury") OR (Chronic Spinal Cord Injury) (Word variations have been searched) with Publication Year from 1900 to 2023, in Trials

iii. PRISMA Flow Diagram



Out of a total of 11 RCTs, 5 trials met the inclusion criteria as specified by the GDG and were used for synthesizing evidence.

iv. Summary of included studies

	Population	Intervention	Comparison	Outcomes	Comments
Abdelaziz et al.[1]	Patients having chronic traumatic dorsal spinal cord injury with durations of at least 6 months were included in the study	20 - They received autologous adult bone marrow stem cells transplantation through open surgical intraparenchymal and intralesional injection into the site of cord injury. The treatment was continued with monthly intrathecal injection of stem cells through lumbar or cisternal punctures	10 - Patients with complete or incomplete spinal injury who will not have stem cell transplantation	AIS Grade Improvement	No proper mention of stem cell characterization
Albu et al.[2]	Participants were 10 patients (7 males, 3 females, age range, 25-47 years) with chronic complete SCI (American Spinal Injury Association A) at dorsal level (T3-11)	Patients were randomly assigned to receive a single dose of intrathecal ex vivo-expanded WJ-MSCs (10 X 10^6 cells) from human umbilical cord or placebo and were then switched to the other arm at 6 months.	Patients were randomly assigned to receive a single dose of intrathecal ex vivo-expanded WJ-MSCs (10X 10^6cells) from human umbilical cord or placebo and were then switched to the other arm at 6 months.	AIS Motor, AIS Sensory, SCIM	
Cheng et al.[3]	Patients of thoracolumbar spinal cord injury that were graded 'A' by the AIS grading system	10 - The stem cell transplantation group was given CT-guided UCMSC transplantation twice	10 - Patients were left at home for self healing with no specific treatment.	AIS Motor, AIS Sensory, Barthel Index	No proper mention of stem cell characterization
Dai et al.[4]	Patients with complete and chronic cervical SCI. The treatment group consisted of 20 patients, 14 males and six females with average age of 34.77±8.9 years (range: 22–54 years) and the	Bone marrow mesenchymal stem cells (BMMSC)transplantation to the area surrounding injury	control group was not treated with cell transplantation	AIS Motor, AIS Sensory	

	average time interval between the injury and stem cell therapy was 51.97±18.3 months (range: 18–74 months).				
El-Kheir et al.[5]	Patients with traumatic chronic SCI of 10–36 months in duration with completion of at least 6 months of physiotherapy postinjury with no spontaneous recovery or neurological improvement. Baseline assessment of American Spinal Injury Association (ASIA) Impairment Scale (AIS) A and B.	15 AIS A and 35 AIS B chronic SCI patients for physiotherapy and Autologous Bone Marrow-Derived Cell Therapy	9 AIS A and 11 AIS B Chronic SCI patients only treated with physiotherapy	AIS Grade improvement, AIS Motor	There are two intervention groups in study El-Kheir et al. on the basis of AIS Grade (A and B). Therefore, the data has been incorporated separately.
Ghobrial et al.[6]	Subjects with traumatic, nonpenetrating SCI were enrolled and given intramedullary injections of HuCNS-SC R (Stem cells, Inc) rostral and caudal to the C5-C7 injury level with AIS grade A or B.	It consisted of two cohorts. Cohort 1 consisted of an open label, dose-escalation protocol (15, 30, and 40 million cells), while cohort 2 was randomized, controlled, single blinded, with a treatment dose of 40 million cells.	Not mentioned	GRASSP Scoring	Insufficient data not included in any analysis
Levi et al.[7]	Participants enrolled had a sequential investigation involving transplantation of allogeneic HuCNS-SC cells into three cohorts with chronic SCI. With C5-7 and AIS A, B or C.	6 - It consisted of participants with C5-7 motor complete (AIS A or B) SCI, randomized, single blinded and received the transplant of HuCNS-SC.	4 - Randomized, single blinded, did not receive transplant.	Baseline Upper Extremity Motor Score, GRASSP Scoring	
Saini et al.[8]	Patients aged 18–50 years presenting with the	Bone marrow derived stem cells (BMSCs) (intramedullary route)	Placebo (intramedullary route) intraoperatively	SCIM, AIS motor, AIS	More than 30% of patients in

	complete motor (ASIA grade A) spinal cord injury (quadriplegia/paraplegia) requiring surgical intervention 21 days of injury were included in the study.	intraoperatively		sensory	each arm died. Their data was incorporated in the analysis assuming the worst outcome.
Song et al.[9]	Enrolled=Thirty-six patients with acute spinal cord injury	Eighteen control group's treatment + autologous bone marrow mesenchymal stem cells injection in the subarachnoid space	Eighteen treated with decompression+ internalfixation+ conventional medical treatments	ASIA Sensory Score, ASIA Exercise Score, Botsford Score, SCIM Total	No proper mention of stem cell characterization
Srivastava et al.[10]	Participants of ASCI having complete lesion (ASIA Impairment Scale [AIS]-A grade) with thoracolumbar injury severity score (≥4) (unstable injury requiring stabilization by surgery). Thoracolumbar spine injury level between T4 and L2 vertebra with age between 18 and 65 years of either gender. Duration of injury<6 weeks.	ASCI participants managed by posterior instrumentation (titanium pedicle screw and rod devices) followed by infusion of autologous BM-derived stem cells as an adjuvant	ASCI participants managed nonoperatively.	AIS Grade Improvement, AIS Motor, AIS sensory	
Yang et al.[11]	Sixty SCI patients were included in the study with chronic SCI	Autologous BMSCs transplantation therapy	Standard rehabilitation therapy	AIS Grade improvement	No proper mention of stem cell characterization

v. Classification of studies on the basis of level of manipulation of the stem cell/ Stem cell derived product used (*interpreted by the Secretariat as defined by CDSCO* and the information provided in the trial itself*):

S. No.	Trial Details	Intervention	Level of manipulation	Information extracted from trial
1.	Albu et al. 2021	Wharton Jelly mesenchymal stromal cells (WJ-MSCs)- single dose of intrathecal ex vivo-expanded WJ-MSCs from human umbilical cord	More	*"WJ-MSCs were derived following current Good Manufacturing Practice-compliant procedures using donated umbilical cord with appropriate donor informed consent approved by the local ethics committee and the competent tissue and organ donation authority."*
2.	Dai et al. 2013	Bone marrow mesenchymal stem cells (BMMSC)- transplantation to the area surrounding injury; intrathecally	More	*"A volume of 90 ml bone marrow was collected from unilateral or bilateral posterior iliac spine. The BMMSC was separated, allowed for adhesion and proliferation in GMP laboratory. After passaging every 5–7 days, the fourth generation cells were collected for transplantation."*
3.	El-Kheir et al. 2014	Autologous Bone Marrow-Derived Stem Cell (ABMC) Therapy- Intrathecal transplantation through lumbar puncture	Less	*"autologous BMCs are allowed to adhere for 72 h to form "minimally manipulated" adherent BMCs (ABMCs)"*
4.	Levi et al. 2019	Allogenic Human neural stem cell transplantation (HuCNS-SC)	More	*"HuCNS-SC cells were prepared and released by Stem Cells Inc. according to product manufacturing specifications and regulatory standards established with the FDA."*
5.	Saini R et al. 2022	Mononuclear Bone marrow derived stem cells (BMSCs)- intramedullary route, intraoperatively	Less	*"Mononuclear BMSCs were separated under good laboratory practice conditions by SmartPReP2 Bone Marrow Aspirate Concentrate (BMACTM) System (Harvest Technologies GmbH, Munich, Germany) in the sterile zone at Operation theater."*

* as defined by CDSCO in Annexure- I of the guideline document

vi. Evidence to Decision Framework:

QUESTION

In patients with spinal cord injury, what is the efficacy and safety of stem cell therapy compared to usual care?	
POPULATION:	Spinal Cord Injury
INTERVENTION:	Stem cell transplantation
COMPARISON:	Usual care
MAIN OUTCOMES:	Independence and Disability Scales - Spinal Cord Independence Measure Scale, ASIA Motor Score, AIS Grade Improvement
SETTING:	Hospital
PERSPECTIVE:	Health Systems/Population
BACKGROUND:	Spinal cord injury (SCI) is a chronic condition leading to physical and psychological problems along with numerous economic burdens for patients. SCI interrupts the neural connections between the upper center and the spinal cord, leading to devastating and permanent neurological impairment, including sensory and motor disability, abnormal reflexes, and autonomic disorders. SCI not only seriously affects the quality of life and life span of patients, but it also leads to psychological and emotional disorders and social phobia, with enormous impact and heavy economic burden on the family as well as the social health care system. Therefore, effective treatments are urgently needed to cure individuals with SCI.[12,13]
CONFLICT OF INTERESTS:	None

ASSESSMENT

Problem Is the problem a priority?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ No ○ Probably no ○ Probably yes ● Yes ○ Varies ○ Don't know	Incidence of SCI in India is estimated to be around 20 per million with 2500 fresh cases added every year. In addition to physical, social and psychological suffering for these patients, taking care of these patients creates a significant burden for the healthcare system. The current clinical treatment for SCI mainly includes early surgical decompression and stabilization, augmentation of spinal cord perfusion, intravenous administration of high-dose corticosteroids, anti-inflammatory therapy in the acute phase, and neurological rehabilitation training during the chronic phase. Unfortunately, the clinical benefit achieved using current approaches is limited and there are no effective strategies currently existing to repair SCI, thus patients suffer long-term dysfunction and lifelong disability.[12,13]	
Desirable Effects How substantial are the desirable anticipated effects?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Trivial ○ Small ○ Moderate ○ Large ○ Varies ● Don't know	There is insufficient evidence to draw firm conclusions regarding the desirable effects of stem cell therapy in patients with spinal cord injury. **Dependency:** Evidence from one RCT with 21 participants of acute complete spinal cord injury reported a mean difference of 9.76 (95% CI: -2.14 to 21.66) in the SCIM Score in the stem cell therapy arm (intramedullary route) as compared to usual care at the end of six months. The difference was statistically non-significant.	The GDG considered an MCID of 10 for the SCIM scale. A change in Wexner score by 2 points was considered as MCID.

	Bowel Function: Evidence from one RCT with 10 participants of chronic complete spinal cord injury reported a reduction in the Wexner Score with a mean difference of -1.87 (95% CI: -3.50 to - 0.24) in the stem cell therapy arm (intrathecal route) as compared to usual care at the end of six months. The difference was statistically significant but unimportant clinically as it was less than the MCID of 2. **Bladder Function:** Evidence from one RCT with 10 participants of chronic complete spinal cord injury reporting the Qualiven questionnaire (subscale-specific impact of urinary symptoms on quality of life) observed a mean difference of 0.30 (95% CI: -0.45 to 1.05) in the stem cell arm (intrathecal route) as compared to usual care at the end of six months. The difference was statistically non-significant. **Quality of Life:** Evidence from one RCT with 10 participants of chronic complete spinal cord injury reporting WHOQOL-BREF observed a mean difference of 0.70 (95% CI: -22.06 to 23.46) in the stem cell therapy arm (intrathecal route) as compared to usual care at the end of six months. The difference was statistically non-significant.	

Undesirable Effects

How substantial are the undesirable anticipated effects?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
○ Trivial ○ Small ○ Moderate ○ Large ● Varies ○ Don't know	**Serious Adverse Events:** Albu et al[2], Dai et al[4] and El Kheir et al[5] did not report any SAEs in either of the arms. The SAEs reported by Levi et al[7] included sepsis, posterior reversible encephalopathy syndrome, seizure, wound hematoma and autonomic dysreflexia in the stem cell arm and urinary tract infection in the usual care arm. **All-cause mortality:** Saini et al[8] reported all-cause mortality, 5 patients in the usual care arm and 3 patients in the stem cell arm expired during the follow up period due to ventilation associated pneumonia. This difference was statistically not significant (p = 0.31).	

Certainty of evidence What is the overall certainty of the evidence of effects?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
● Very low ○ Low ○ Moderate ○ High ○ No included studies	The evidence in this review is of very low quality due to high risk of bias, inconsistency and imprecision in the effect shown by stem cell therapy.	
Values Is there important uncertainty about or variability in how much people value the main outcomes?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Important uncertainty or variability ○ Possibly important uncertainty or variability ● Probably no important uncertainty or variability ○ No important uncertainty or variability	Main outcome is clinical improvement in the form of increased independence and increase in motor activity which is likely to be highly valued by most persons.[13,14]	
Balance of effects Does the balance between desirable and undesirable effects favor the intervention or the comparison?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**

○ Favors the comparison ○ Probably favors the comparison ● Does not favor either the intervention or the comparison ○ Probably favors the intervention ○ Favors the intervention ○Varies ○ Don't know	It is less clear if benefits of stem cell intervention outweigh the harms due to small to trivial benefit, based on limited evidence.	
Resources required How large are the resource requirements (costs)?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
● Large costs ○ Moderate costs ○ Negligible costs and savings ○ Moderate savings ○ Large savings ○ Varies ○ Don't know	No direct evidence of the resources required in stem cell transplantation in patients with stroke was identified. The information provided by the websites of Insurance Agencies and third party agencies is as follows[15]: • 5.0-5.5 lacs (TATA AIG) • 5000-50,000 USD However, indirect evidence shows that the median cost of hematopoietic stem cell transplantation for hematological diseases were USD, $ 12,500 (range $ 10,331–39,367) - Rs. 10,00,000.[17]	
Certainty of evidence of required resources What is the certainty of the evidence of resource requirements (costs)?		

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
○ Very low ○ Low ● Moderate ○ High ○ No included studies	No research evidence was identified.	The GDG members were fairly certain that stem cell therapy requires large resources.
Cost effectiveness Does the cost-effectiveness of the intervention favor the intervention or the comparison?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Favors the comparison ● Probably favors the comparison ○ Does not favor either the intervention or the comparison ○ Probably favors the intervention ○ Favors the intervention ○ Varies ○ No included studies	No research evidence for stem cell therapy was identified.	The intervention was not found to be effective and hence the committee deferred to comment on cost effectiveness.
Equity What would be the impact on health equity?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**

○ Reduced ● Probably reduced ○ Probably no impact ○ Probably increased ○ Increased ○ Varies ○ Don't know	No research evidence was identified.	As stem cell therapy is an expensive treatment offered only at tertiary care centers, it is likely to increase inequity as it would not be affordable by all.
Acceptability Is the intervention acceptable to key stakeholders?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ No ○ Probably no ● Probably yes ○ Yes ○ Varies ○ Don't know	Despite uncertainty regarding the medical risks and benefits associated with stem cell injections, patients with spinal cord injury have very high expectations from stem, cell therapy.[17]	
Feasibility Is the intervention feasible to implement?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**

○ No ○ Probably no ● Probably yes ○ Yes ○ Varies ○ Don't know	No research evidence was identified.	Stem Cell transplantation can be performed only in tertiary care centers so feasibility in lesser equipped hospitals and clinics is not possible

SUMMARY OF JUDGEMENTS

	JUDGEMENT						
PROBLEM	No	Probably no	Probably yes	**Yes**		Varies	Don't know
DESIRABLE EFFECTS	Trivial	**Small**	Moderate	Large		Varies	**Don't know**
UNDESIRABLE EFFECTS	Trivial	Small	Moderate	Large		**Varies**	Don't know
CERTAINTY OF EVIDENCE	**Very low**	Low	Moderate	High			No included studies
VALUES	Important uncertainty or variability	Possibly important uncertainty or variability	**Probably no important uncertainty or variability**	No important uncertainty or variability			
BALANCE OF EFFECTS	Favors the comparison	Probably favors the comparison	**Does not favor either the intervention or the comparison**	Probably favors the intervention	Favors the intervention	Varies	Don't know

RESOURCES REQUIRED	**Large costs**	Moderate costs	Negligible costs and savings	Moderate savings	Large savings	Varies	Don't know
CERTAINTY OF EVIDENCE OF REQUIRED RESOURCES	Very low	Low	**Moderate**	High			No included studies
COST EFFECTIVENESS	Favors the comparison	**Probably favors the comparison**	Does not favor either the intervention or the comparison	Probably favors the intervention	Favors the intervention	Varies	No included studies
EQUITY	Reduced	**Probably reduced**	Probably no impact	Probably increased	Increased	Varies	Don't know
ACCEPTABILITY	No	Probably no	**Probably yes**	Yes		Varies	Don't know
FEASIBILITY	No	Probably no	**Probably yes**	Yes		Varies	Don't know

TYPE OF RECOMMENDATION

Strong recommendation against the intervention	Conditional recommendation against the intervention	**Use only in the context of rigorously conducted RCTs**	Conditional recommendation for either the intervention or the comparison	Conditional recommendation for the intervention	Strong recommendation for the intervention
○	○	●	○	○	○

CONCLUSIONS

Recommendation

Stem Cell Therapy is **not recommended** in routine clinical practice for the treatment of spinal cord injury. It may be used only in the context of rigorously conducted RCTs.

Justification

Overall justification

This recommendation has been made as the evidence is inadequate in quantity and quality to determine the efficacy of stem cell therapy in patients with spinal cord injury. The incidence of undesirable effects including mortality are variable. In addition, the reported follow up period is too small to comment on the side effect profile and long-term safety is not known.

Detailed justification

Desirable Effects

This judgment was made as the evidence is inadequate in quantity and quality to determine the efficacy of stem cell therapy in patients with spinal cord injury.

Undesirable Effects

This judgment was made as the incidence of undesirable effects including mortality are variable. Also, the follow up period of one year is too small to comment on the side effect profile and long-term safety is not known.

Certainty of evidence

Very low certainty of Evidence

Resources required

The committee opined that stem cell treatment is associated with large costs.

vii. Data Extraction:

Two reviewers independently extracted the data from the electronic databases mentioned above. Data for the following variables were extracted: author name, study year, country, study setting, study duration, phase of study, sample size at beginning, sample size at analysis, treatment and control groups, sex, age, injury type, stem cell type, cell count, volume, dosage, follow-up duration, ASIA motor score, sensory score, pin prick score, light touch score, AIS grade and Spinal Cord Independence Measure (SCIM) scale, adverse events and safety outcomes. Disagreements were resolved through the consensus among all the authors. Analysis was carried out using Review Manager version 5.4.1, (Cochrane Collaboration, Syracuse, NY, USA). Dichotomous values were assessed using Odds Ratios (ORs) with 95% confidence intervals while, continuous variables were measured using standard mean deviations of the represented cumulative data (Mean $\pm$ Standard Deviation). Cochrane's I^2 statistic was used for assessing the heterogeneity. Studies with I^2 values exceeding 50% had higher heterogeneity, while those with I^2<50% had lower heterogeneity. For value of I^2 <50% fixed effects model was used and for I^2>50% random effects model was used. Risk of Bias for the included studies was performed using Cochrane's risk of bias tool in RevMan software. The following parameters were used for assessing the risk of bias: random sequence generation, allocation concealment, blinding of participants and personnel, blinding of outcome assessment, incomplete outcome data, selective reporting and other bias. The quality of evidence was graded as high, some concerns and low risk of bias. Publication bias was assessed using Begg's funnel plot.

Study-Efficacy and outcome of bone marrow derived stem cells transplanted via intramedullary route in acute complete spinal cord injury – A randomized placebo-controlled trial	**Saini et al. 2022**[8]
Study type:	RCT
Number of participants:	13
Countries and setting:	India, Hospital
Duration of study Follow up (post intervention):	6 months
Method of assessment of disease condition:	Patients of spinal cord injury (quadriplegia/paraplegia) with AIS grade A.
Subgroup analysis within study:	-
Inclusion criteria:	Patients aged 18–50 years presenting with the complete motor (ASIA grade A) spinal cord injury (quadriplegia/paraplegia) requiring surgical intervention 21 days of injury were included in the study.
Exclusion criteria:	Patients were excluded if there was anatomical transaction of the cord

	visualized by on MRI; history of myocardial infarction and coronary artery disease; chronic infections: HIV, HbSAg; history of liver & renal insufficiency or of any malignancy; pregnant or nursing women; inability to communicate effectively with neurological examiner and active participation in another experimental procedure/intervention
Recruitment/selection of patients:	180 patients were eligible out of which 27 randomized, 11 patients were lost to follow up. In total, there were 6 patients in the placebo group and 7 in stem cell group.
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Bone Marrow Mesenchymal Stem Cells, flow cytometry, intramedullary, 1.8mL
Outcomes reported with time points	SCIM (3 months and 6 months), AIS Motor (3 months and 6 months), AIS Sensory (3 months and 6 months)
Funding	None.
RoB2 Assessment: Specify separately for each domain	Randomization Process– Low risk Deviation from intended intervention– Low risk Missing outcome data– High risk Measurement of the outcome– Low risk Selection of the reported result– High risk

Study-Transplantation of autologous bone marrow mesenchymal stem cells in the treatment of complete and chronic cervical spinal cord injury	**Dai et al. 2013[4]**
Study type:	RCT
Number of participants:	40
Countries and setting:	China, Hospital
Duration of study Follow up (post intervention):	6 months
Method of assessment of disease condition:	Through physical examination and cervical spinal cord magnetic resonance imaging (MRI), the patients diagnosed with cervical SCI were selected.
Subgroup analysis within study:	-
Inclusion criteria:	• Complete cervical SCI with post-operative or post-traumatic period

	more than one year and • The site of injury within the cervical spinal cord C3–C7 excluding C3 and C7.
Exclusion criteria:	• Incomplete spinal cord injury; • Neurological function with tendency to recover; • Adhesion, syringomyelia,dural vascular malformations or significant compression in spinal cord, requiring surgical intervention; • Unexplained nerve dysfunction; • Intended surgical site with inflammation or skin ulceration; • With bleeding tendency or coagulation disorders; • Poor general condition or other organ dysfunction intolerant of surgery and Disagreement with the research protocol.
Recruitment/selection of patients:	Forty patients with complete and chronic cervical SCI were selected and randomly assigned to one of the two experimental groups, treatment group and control group.
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Bone Marrow Mesenchymal Stem Cells, flow cytometry, intrathecally, 25μL
Outcomes reported with time points	AIS Motor (6 months), AIS Sensory (6 months)
Funding	Not mentioned
RoB2 Assessment: Specify separately for each domain	Randomization Process– High risk Deviation from intended intervention– High risk Missing outcome data– Low risk Measurement of the outcome– Low risk Selection of the reported result– High risk

Study: Autologous Bone Marrow-Derived Cell Therapy Combined with Physical Therapy Induces Functional Improvement in Chronic Spinal Cord Injury Patients	**El-Kheir et al. 2014**[5]
Study type:	Randomized Controlled Trial
Number of participants:	70

Countries and setting:	Egypt, University Setting
Duration of study Follow up (post intervention):	18 months
Method of assessment of disease condition:	Patients with traumatic chronic SCI of 10–36 months in duration were taken into consideration
Subgroup analysis within study:	-
Inclusion criteria:	• Traumatic chronic SCI of 10–36 months in duration. • Completion of at least 6 months of physiotherapy postinjury with no spontaneous recovery or neurological improvement • No concomitant systemic disease. • Baseline assessment of American Spinal Injury Association (ASIA) Impairment Scale (AIS) A and B.
Exclusion criteria:	• Nontraumatic SCI, including patients with either transverse myelitis or demyelination, or lumbar SCI. • Acute SCI or duration of injury less than 10 months or more than 36 months. • Evidence for clinical and/or electrophysiological improvement in spinal cord functions over the previous 6 months. 4. Baseline assessment of AIS C or D. • Major impairment of respiratory, cardiac, hepatic, or other systemic functions. • Unfitness for physical therapy, such as patients with muscle fibrosis.
Recruitment/selection of patients:	Patients were recruited from the outpatient clinics of Physical Medicine at Al-Azhar, Cairo, and Alexandria University Hospitals. During the enrollment period, 159 patients were evaluated, and 70 patients were enrolled.
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Human Autologous Bone Marrow-Derived Stem Cell (ABMCs) were isolated from Bone marrow aspirates from SCI patients. ABMCs were analyzed by flow cytometry with cell surface markers: cluster of differentiation 13-phycoerythrin-cyanine 7 (CD13-PC7), CD29-PC7, CD34-phycoerythrin (PE), CD44-fluorescein isothiocyanate (FITC), CD45-PC7, CD73-PE, CD90-PE, CD105-PE, CD166-PE, CD271-PE, and c-kit-PE. Intrathecal transplantation through lumbar puncture.

	2×10^6 cells/kg
Outcomes reported with time points	AIS Grade Improvement (18 months), Total Motor Score (18 months)
Funding	New Jersey Health Foundation
RoB2 Assessment: Specify separately for each domain	Randomization Process– High risk Deviation from intended intervention– Low risk Missing outcome data– Low risk Measurement of the outcome– Low risk Selection of the reported result– High risk

Study: Effectiveness of bone marrow-derived mononuclear stem cells for neurological recovery in participants with spinal cord injury: A randomized controlled trial	**Srivastava et al. 2019[10]**
Study type:	Randomized Controlled Trial
Number of participants:	138
Countries and setting:	India, King George's Medical University
Duration of study Follow up (post intervention):	One year (12 months)
Method of assessment of disease condition:	Participants of ASCI having complete lesion (ASIA Impairment Scale [AIS]-A grade) with thoracolumbar injury severity score (≥4)
Subgroup analysis within study:	-
Inclusion criteria:	•Participants of ASCI having complete lesion (ASIA Impairment Scale [AIS]-A grade) with thoracolumbar injury severity score (≥4) (unstable injury requiring stabilization by surgery) • Thoracolumbar spine injury level between T4 and L2 vertebra • Age between 18 and 65 years of either gender • Duration of injury
Exclusion criteria:	• Polytrauma patients with injury spine and associated thoraco-abdominal injuries and/or head injury • Medically unfit patients not suitable for surgery • Patients with other comorbid conditions such as osteoporosis, pressure sores (Grade III–IV), psychiatric illness and those on steroids or other

	immune suppressants • Patients who did not give their consent for participating in the study.
Recruitment/selection of patients:	Group 1 had 70 participants who underwent conventional surgery along with BM-MNSCs as an adjuvant. In Group 2, there were 68 participants who were treated by conventional surgery
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Bone marrow-derived mono-nuclear stem cells (BM-MNSCs) Flowcytometry (MNCs and CD34+ count) Intrathecal 10 ml of precipitate was used for infusion
Outcomes reported with time points	AIS Grade Improvement (12 months), Total Motor Score (12 months), Total Sensory Score (12 months)
Funding	No external source of funding
RoB2 Assessment: Specify separately for each domain	Randomization Process – High risk Deviation from intended intervention – Low risk Missing outcome data – Low risk Measurement of the outcome – High risk Selection of the reported result – High risk

Study: Clinical effects of intrathecal administration of expanded Wharton jelly mesenchymal stromal cells in patients with chronic complete spinal cord injury: a randomized controlled study	**Albu et al. 2021**[2]
Study type:	phase 1/2a clinical trial with a randomized, double-blind, crossover, placebo-controlled design
Number of participants:	10
Countries and setting:	Spain, Hospital
Duration of study Follow up (post intervention):	6 months
Method of assessment of disease condition:	Patients with spinal cord injury of traumatic etiology, with complete SCI (American Spinal Injury Association [ASIA] A), thoracic injury level (T1-12)

	and chronic evolution (1-5 years after injury)
Subgroup analysis within study	-
Inclusion criteria:	• spinal cord injury of traumatic etiology • adults (18-65 years old) with complete SCI (American Spinal Injury Association [ASIA] A) • thoracic injury level (T1-12) • chronic evolution (1-5 years after injury)
Exclusion criteria:	• Patients who presented non-traumatic SCI • Cord lesion at the cervical or lumbosacral segments, extended (>3 segments) or multiple lesioned segments • Associated brain injury or peripheral nerve injuries • Currently enrolled in other clinical trials or had scheduled surgery within the subsequent 24 months after entering the trial • Received intrathecal medication or immunosuppressive drugs 2 months before the trial • Pregnant or breastfeeding
Recruitment/selection of patients:	Patients were enrolled by health professionals from the Guttmann Institute. After signing informed consent, participants were randomly assigned to WJ-MSC or placebo infusion using simple randomization and admitted to the hospital.
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Wharton Jelly mesenchymal stromal cells (WJ-MSCs) Intrathecal 4±1 mL, resulting in approximately $2.5x10^6$ live WJ-MSCs/mL
Outcomes reported with time points	AIS Grade Improvement (6 months), ASIA Sensory Light Touch Score (6 months), ASIA Sensory Pin Prick Score (6 months), Total Motor Score (6 months), Total Sensory Score (6 months), SCIM Total
Funding	FundacioMarato TV3 (grant no. 616/ 2012), AES 2019 ISCIII (PI19/01680)
RoB2 Assessment: Specify separately for each domain	Randomization Process– Low risk Deviation from intended intervention– Low risk Missing outcome data– Low risk Measurement of the outcome– Low risk

	Selection of the reported result– High risk

Study: Clinical Outcomes from a Multi-Center Study of Human Neural Stem Cell Transplantation in Chronic Cervical Spinal Cord Injury	**Levi et al. 2019**[7]
Study type:	Randomized controlled trial
Number of participants:	10
Countries and setting:	USA, Hospital
Duration of study Follow up (post intervention):	3, 6 and 9 months
Method of assessment of disease condition:	Participants should be 4–24 months post-injury with C5-C7 ISNCSCI motor levels, with a single traumatic and non-penetrating SCI, based on magnetic resonance imaging (MRI)
Subgroup analysis within study	-
Inclusion criteria:	• Participants were 4–24 months post-injury • C5-C7 ISNCSCI motor levels • Male and female • 18–60 years of age • With a single traumatic and non-penetrating SCI • Based on magnetic resonance imaging (MRI) • AIS Grade A or B
Exclusion criteria:	• Patients not in generally good medical condition other than their injury • Patients to have no contraindications for systemic immunosuppression, MRIs, or safe surgical exposure of the lesion area
Recruitment/selection of patients:	Patients were enrolled by health professionals from the Guttmann Institute. After signing informed consent, participants were randomly assigned to WJ-MSC or placebo infusion using simple randomization and admitted to the hospital.
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Human neural stem cell transplantation (HuCNS-SC) Intramedullary

	$40x10^6$ cells
Outcomes reported with time points	GRASSP strength (3 months), Upper Motor Extremity Score (3 months)
Funding	The work was supported by StemCells, Inc. and the respective Academic Institutions. Bright Oceans Corporation (BOCO) provided funds for post-termination data collection.
RoB2 Assessment: Specify separately for each domain	Randomization Process –High risk Deviation from intended intervention –Low risk Missing outcome data –Low risk Measurement of the outcome –Low risk Selection of the reported result –High risk

viii. List of Excluded studies:

1	Associations Between Insufficient Sleep Syndrome and Health Outcomes for Care Partners	Li C-Y	2022	No use of stem cells in spinal cord injury
2	The development of lived experience-centered word clouds to support research uncertainty gathering in degenerative cervical myelopathy: results from an engagement process and protocol for their evaluation, via a nested randomized controlled trial	Davies BM	2021	No use of stem cells in spinal cord injury
3	Safety, tolerability, and activity of mesenchymal stem cells versus placebo in multiple sclerosis (MESEMS): a phase 2, randomised, double-blind crossover trial	Uccelli A	2021	Stem cells used in Multiple Sclerosis
4	Randomized, Placebo-Controlled Analysis of the Knee Synovial Environment Following Platelet-Rich Plasma Treatment for Knee Osteoarthritis	Tucker JD	2021	No use of stem cells in spinal cord injury
5	Spine Treatment Appraisal Report (STAR): Bone Marrow-Derived Stem Cells Improve Neurological Recovery in Participants with Spinal Cord Injury	Dettori JR	2021	Full text not available, only abstract
6	Denosumab Versus Zoledronic Acid in Bone Disease Treatment of Newly Diagnosed Multiple Myeloma: An International, Double-Blind, Randomized Controlled Phase 3 Study—Asian Subgroup Analysis	Huang S-Y	2020	No use of stem cells in spinal cord injury
7	Two year follow up of acute complete spinal cord injury patients receiving autologous bone marrow-derived mononuclear stem cell therapy	Rani R	2020	Only the abstract is published
8	Intrathecal enzyme replacement for cognitive decline in mucopolysaccharidosis type I, a randomized, open-label, controlled pilot study	Chen AH	2020	No use of stem cells in spinal cord injury
9	Human umbilical cord mesenchymal stem cells to treat spinal cord injury in the early chronic phase: Study protocol for a prospective, multicenter, randomized, placebo-controlled, single-blinded clinical trial	Yang Y	2020	Only study protocol
10	Pain inhibition through transplantation of fetal neuronal progenitors into the injured spinal cord in rats	Batista CM	2019	Animal study
11	Clinical trial for spinal cord injury with neural lineage stem cell	Kim K-N	2019	Not a randomised controlled trial
12	Therapeutic application of autologous bone marrow mononuclear stem cell in complete spinal cord injury in human	Saini R	2018	Only abstract available

13	Global Spine Congress 2018, GSC 2018	Global Spine Journal	2018	Conference abstract, no use of stem cells in spinal cord injury
14	Safety of Autologous Human Schwann Cell Transplantation in Subacute Thoracic Spinal Cord Injury	Anderson KD	2017	Non-randomised trial
15	Effects of single-agent bortezomib as post-transplant consolidation therapy on multiple myeloma-related bone disease: a randomized phase II study	Sezer O	2017	No use of stem cells in spinal cord injury
16	Dendritic cell vaccination in combination with docetaxel for patients with metastatic castration-resistant prostate cancer: A randomized phase II study	Kongsted P	2017	No use of stem cells in spinal cord injury
17	Autologous mesenchymal stromal cell transplantation for spinal cord injury: A phase I pilot study	Satti HS	2017	Non-randomised trial
18	Daratumumab monotherapy in patients with treatment-refractory multiple myeloma (SIRIUS): An open-label, randomised, phase 2 trial	Lonial S	2016	No use of stem cells in spinal cord injury
19	Core decompression and autologous bone marrow concentrate for treatment of femoral head osteonecrosis: A randomized prospective study	Pepke W	2016	No use of stem cells in spinal cord injury
20	Developing clinical-grade cell therapy for spinal cord regeneration	Sabaawy H	2016	Review of trials conducted, only abstract available
21	Impact of chemokines on the properties of spinal cord-derived neural progenitor cells in a rat spinal cord lesion model	Knerlich-Lukoschus F	2015	Animal study
22	Phase I, multicentre, dose-escalation trial of monotherapy with milatuzumab (humanized anti-CD74 monoclonal antibody) in relapsed or refractory multiple myeloma	Kaufman JL	2013	No use of stem cells in spinal cord injury
23	Tracking induced pluripotent stem cells-derived neural stem cells in the central nervous system of rats and monkeys	Tang H	2013	Animal study
24	Safety profile, feasibility, and early clinical outcome of co-transplantation of olfactory mucosa and bone marrow stem cells in spinal cord injury-a novel implantation with two-year follow-up	Kantharajanna SB	2013	Prospective case series

25	Dose-dependent facilitation of peripheral nerve regeneration by bone marrow-derived mononuclear cells: A randomized controlled study. Laboratory investigation	Raheja A	2012	Animal Study
26	Combined transplantation of human neural stem cells and human mesenchymal stem cells following spinal cord injury	Cheng I	2011	Animal Study
27	Motor and somatosensory evoked potential evaluation in rat spinal cord transection models following bone marrow mesenchymal stem cell transplantation	Li K	2011	Animal study
28	High-dose methylprednisolone in acute spinal cord injuries: Proceed with caution	Pandya KA	2010	No use of stem cells in spinal cord injury
29	Multidisciplinary management of Hunter syndrome	Muenzer J	2009	Focused on Hunter Syndrome
30	Functional recovery and neural differentiation after transplantation of allogenic adipose-derived stem cells in a canine model of acute spinal cord injury.	Ryu HH	2009	Animal study
31	Clinical studies in spinal cord injury: Moving towards successful trials	Knafo S	2008	Review article
32	New thinking on olfactory ensheathing cell transplantation in the repair of spinal cord injury	Sun L-Q	2007	Not a randomised controlled trial
33	Curative effect of autologous mesenchymal stem cell transplantation on spinal cord injury	Xie Z-W	2007	From non indexed journal
34	Cellular replacement therapy for Parkinson's disease - Where we are today?	Redmond Jr. DE	2002	No use of stem cells in spinal cord injury
35	Systemic Administration of Allogeneic Cord Blood Mononuclear Cells in Adults with Severe Acute Contusion Spinal Cord Injury: Phase 1/2a Pilot Clinical Study-Safety and Primary Efficacy Evaluation	Smirnov VA	2022	Not a randomised controlled trial
36	Safety and neurological assessments after autologous transplantation of bone marrow mesenchymal stem cells in subjects with chronic spinal cord injury	Mendonça, MVP	2014	Not a randomised controlled trial
37	Effects of Hematopoietic Autologous Stem Cell Transplantation to the Chronically Injured Human Spinal Cord Evaluated by Motor and Somatosensory Evoked Potentials Methods	Frolov, AA	2012	Not a randomised controlled trial

38	Delivery of autologous bone marrow precursor cells into the spinal cord via lumbar puncture technique in patients with spinal cord injury: A preliminary safety study	Callera F	2006	Not a randomised controlled trial
39	Application of autologous bone marrow stem cells in the therapy of spinal cord injury patients	Chernykh ER	2007	Not a randomised controlled trial
40	Spinal cord injury treatment with intrathecal autologous bone marrow stromal cell transplantation: The first clinical trial case report	Saito F	2008	Case report
41	A First-in-Human, Phase I Study of Neural Stem Cell Transplantation for Chronic Spinal Cord Injury	Curtis E	2018	Not a randomised controlled trial
42	Significant Improvement of Acute Complete Spinal Cord Injury Patients Diagnosed by a Combined Criteria Implanted with Neuro Regen Scaffolds and Mesenchymal Stem Cells	Xiao ZF	2018	Not a randomised controlled trial
43	Autologous Bone Marrow Derived Mononuclear Cell Therapy for Spinal Cord Injury: A Phase I/II Clinical Safety and Primary Efficacy Data	Kumar AA	2009	Not a randomised controlled trial
44	Stem cells in the treatment of chronic spinal cord injury: evaluation of somatosensitive evoked potentials in 39 patients	Cristante AF	2009	Not a randomised controlled trial
45	The Damaged Spinal Cord Is a Suitable Target for Stem Cell Transplantation	Curt A	2020	Not a randomised controlled trial
46	Clinical Trial of Human Fetal Brain-Derived Neural Stem/Progenitor Cell Transplantation in Patients with Traumatic Cervical Spinal Cord Injury	Shin JC	2015	Not a randomised controlled trial
47	Mobilization of CD133+ CD34- Cells in Healthy Individuals Following Whole-Body Acupuncture for Spinal Cord Injuries	Moldenhauer S	2010	Not a randomised controlled trial
48	A Phase III Clinical Trial Showing Limited Efficacy of Autologous Mesenchymal Stem Cell Therapy for Spinal Cord Injury	Oh SK	2016	Not a randomisedcontrolled trial
49	Emerging Safety of Intramedullary Transplantation of Human Neural Stem Cells in Chronic Cervical and Thoracic Spinal Cord Injury	Levi AD	2018	Not a randomised controlled trial
50	Safety and feasibility of autologous olfactory ensheathing cell and bone marrow mesenchymal stem cell co-transplantation in chronic human spinal cord injury: a clinical trial	Zamani H	2021	Not a randomised controlled trial

51	Combined protocol of cell therapy for chronic spinal cord injury.: Report on the electrical and functional recovery of two patients	Moviglia GA	2006	Not a randomised controlled trial
52	Intraspinal Stem Cell Transplantation in Amyotrophic Lateral Sclerosis: A Phase I Trial, Cervical Microinjection, and Final Surgical Safety Outcomes	Riley J	2014	No use of stem cells in spinal cord injury
53	Clinical safety and primary efficacy of bone marrow mesenchymal cell transplantation in subacute spinal cord injured patients	Karamouzian S	2012	Not a randomised controlled trial
54	Intrathecal administration of autologous mesenchymal stromal cells for spinal cord injury: Safety and efficacy of the 100/3 guideline	Vaquero J	2018	Not a randomised controlled trial
55	Transplantation of Purified Autologous Leukapheresis-Derived $CD34^+$ and $CD133^+$ Stem Cells for Patients With Chronic Spinal Cord Injuries: Long-Term Evaluation of Safety and Efficacy	Al-Zoubi A	2014	Not a randomised controlled trial
56	Intravenous infusion of auto serum-expanded autologous mesenchymal stem cells in spinal cord injury patients: 13 case series	Honmou O	2021	Not a randomised controlled trial
57	Administration of cultured autologous bone marrow stromal cells into cerebrospinal fluid in spinal injury patients: A pilot study	Saito F	2012	Not a randomised controlled trial
58	Intrathecal transplantation of autologous adipose-derived mesenchymal stem cells for treating spinal cord injury: A human trial	Hur JW	2016	Not a randomised controlled trial
59	Treatment of pressure ulcers with autologous bone marrow nuclear cells in patients with spinal cord injury	Sarasúa JG	2011	Not a randomised controlled trial
60	Autologous Bone Marrow Mesenchymal Stem Cell Therapy in the Subacute Stage of Traumatic Brain Injury by Lumbar Puncture	Tian CL	2013	No use of stem cells in spinal cord injury
61	Assessing fetal human neural stem cells tumorigenicity potential in athymic rats with penetrating traumatic brain injury (pTBI)	Andreu M	2022	No use of stem cells in spinal cord injury
62	An approach to personalized cell therapy in chronic complete paraplegia: The Puerta de Hierro phase I/II clinical trial	Vaquero J	2016	
63	Administration of Autologous Bone Marrow-Derived Mononuclear Cells in Children With Incurable Neurological Disorders and Injury Is Safe and Improves Their Quality of Life	Sharma A	2012	No use of stem cells in spinal cord injury

64	Complete spinal cord injury treatment using autologous bone marrow cell transplantation and bone marrow stimulation with granulocyte macrophage-colony stimulating factor: Phase I/II clinical trial	Yoon SH	2007	Not a randomised controlled trial
65	An attempt to treat patients who have injured spinal cords with intralesional implantation of concentrated autologous bone marrow cells	Attar A	2011	Not a randomised controlled trial
66	Safety of Intravenous Infusion of Human Adipose Tissue-Derived Mesenchymal Stem Cells in Animals and Humans	Ra JC	2011	Not a randomised controlled trial
67	A Prospective Randomized Double-Blind Clinical Trial Using a Combination of Olfactory Ensheathing Cells and Schwann Cells for the Treatment of Chronic Complete Spinal Cord Injuries	Chen L	2014	No use of stem cells in spinal cord injury
68	Cell therapy with autologous mesenchymal stromal cells in post-traumatic syringomyelia	Vaquero J	2018	No use of stem cells in spinal cord injury
69	Phase I-II Clinical Trial Assessing Safety and Efficacy of Umbilical Cord Blood Mononuclear Cell Transplant Therapy of Chronic Complete Spinal Cord Injury	Zhu H	2016	Not a randomised controlled trial
70	Treatment of chronic thoracic spinal cord injury patients with autologous Schwann cell transplantation: An interim report on safety considerations and possible outcomes	Saberi H	2008	Not a randomised controlled trial
71	Ex vivo-expanded autologous bone marrow-derived mesenchymal stromal cells in human spinal cord injury/paraplegia: a pilot clinical study	Pal R	2009	Not a randomised controlled trial
72	Effectiveness of intense, activity-based physical therapy for individuals with spinal cord injury in promoting motor and sensory recovery: Is olfactory mucosa autograft a factor?	Larson CA	2013	No use of stem cells in spinal cord injury
73	Safety of Autologous Bone Marrow Stromal Cell Transplantation in Dogs with Acute Spinal Cord Injury	Nishida H	2012	Animal Study
74	Repeated subarachnoid administrations of autologous mesenchymal stromal cells supported in autologous plasma improve quality of life in patients suffering incomplete spinal cord injury	Vaquero J	2017	Not a randomised controlled trial
75	Olfactory Mucosal Autografts and Rehabilitation for Chronic Traumatic Spinal Cord Injury	Lima C	2010	Not a randomised controlled trial

76	Safety of Granulocyte Colony-Stimulating Factor (G-CSF) Administration for Postrehabilitated Motor Complete Spinal Cord Injury Patients: An Open-Label, Phase I Study	Derakhshanrad N	2013	No use of stem cells in spinal cord injury
77	Human intracerebroventricular (ICV) injection of autologous, non-engineered, adipose-derived stromal vascular fraction (ADSVF) for neurodegenerative disorders: results of a 3-year phase 1 study of 113 injections in 31 patients	Duma C	2019	No use of stem cells in spinal cord injury
78	Multicenter Prospective Nonrandomized Controlled Clinical Trial to Prove Neurotherapeutic Effects of Granulocyte Colony-Stimulating Factor for Acute Spinal Cord Injury	Inada T	2014	Not a randomised controlled trial
79	Neuroprotective therapy using granulocyte colony-stimulating factor for acute spinal cord injury: a phase I/IIa clinical trial	Takahashi H	2012	Not a randomised controlled trial
80	A phase II clinical trial with repeated intrathecal injections of autologous mesenchymal stem cells in patients with amyotrophic lateral sclerosis	Petrou P	2021	No use of stem cells in spinal cord injury
81	Neuroprotective therapy using granulocyte colony-stimulating factor for patients with worsening symptoms of compression myelopathy, part 1: a phase I and IIa clinical trial	Sakuma T	2012	No use of stem cells in spinal cord injury
82	Nerve repair using acidic fibroblast growth factor in human cervical spinal cord injury: a preliminary Phase I clinical study	Wu JC	2008	Not a randomised controlled trial
83	Efficacy of a cultured conditioned medium of exfoliated deciduous dental pulp stem cells in erectile dysfunction patients	Koga S	2022	No use of stem cells in spinal cord injury
84	Stem-cell transplantation into the frontal motor cortex in amyotrophic lateral sclerosis patients	Martinez HR	2009	No use of stem cells in spinal cord injury
85	Cell transplantation therapy in reanimating severely head-injured patients	Seledtsov VI	2005	No use of stem cells in spinal cord injury
86	Stem Cell Transplantation in Amyotrophic Lateral Sclerosis Patients: Methodological Approach, Safety, and Feasibility	Martínez HR	2012	No use of stem cells in spinal cord injury
87	Safety and Feasibility of Autologous Bone Marrow Cellular Therapy in Relapsing-Progressive Multiple Sclerosis	Rice CM	2010	No use of stem cells in spinal cord injury

88	Transplantation of Mesenchymal Stromal Cells in Patients With Amyotrophic Lateral Sclerosis: Results of Phase I/IIa Clinical Trial	Syková E	2017	No use of stem cells in spinal cord injury
89	Safety and tolerability of intrathecal delivery of autologous bone marrow nucleated cells in children with cerebral palsy: an open-label phase I trial	Mancías-Guerra C	2014	No use of stem cells in spinal cord injury
90	Forskolin Enhances In Vivo Bone Formation by Human Mesenchymal Stromal Cells	Doorn J	2012	No use of stem cells in spinal cord injury
91	Renal shielding and dosimetry for patients with severe systemic sclerosis receiving immunoablation with total body irradiation in the scleroderma: cyclophosphamide or transplantation trial	Craciunescu OI	2011	No use of stem cells in spinal cord injury
92	Clinical Experience With Autologous M2 Macrophages in Children With Severe Cerebral Palsy	Chernykh ER	2014	No use of stem cells in spinal cord injury
93	Early loss of pericytes and perivascular stromal cell-induced scar formation after stroke	Fernández-Klett F	2013	No use of stem cells in spinal cord injury
94	Safety and efficacy of Wharton's jelly-derived mesenchymal stem cells with teriparatide for osteoporotic vertebral fractures: A phase I/IIa study	Shim J	2021	No use of stem cells in spinal cord injury
95	Mucosal Immune Responses to Treadmill Exercise in Elite Wheelchair Athletes	Leicht CA	2011	No use of stem cells in spinal cord injury
96	Safety and Therapeutic Potential of M2 Macrophages in Stroke Treatment	Chernykh ER	2016	No use of stem cells in spinal cord injury
97	Effects of Intravesical Lactobacillus Rhamnosus GGon Urinary Symptom Burden in People with Neurogenic Lower Urinary Tract Dysfunction	Tucker JD	2021	No use of stem cells in spinal cord injury
98	Olfactory Ensheathing Cell Neurorestorotherapy for Amyotrophic Lateral Sclerosis Patients: Benefits From Multiple Transplantations	Chen L	2012	No use of stem cells in spinal cord injury
99	Assessing fetal human neural stem cells tumorigenicity potential in athymic rats with penetrating traumatic brain injury (pTBI)	Mary Lourdes Andreu	2022	Animal Study
100	Difference Between Rehabilitation Therapy and Stem Cells Transplantation in Patients With Spinal Cord Injury in China	An Yihua doctor	2011	Registered Trial (NCT01393977), no publication

				available
101	Autologous Bone Marrow Stem Cell Transplantation in Patients With Subacute Spinal Cord Injury	Carolina Macedo PhD	2023	Registered Trial (NCT05671796), no publication available
102	Cell therapy for CNS diseases	Honmou O	2016	
103	Stem cell therapy for neurological disorders	Alessandrini M	2019	No use of stem cells in spinal cord injury
104	Phase 1 trial of autologous bone marrow stem cell transplantation in patients with spinal cord injury	Kakabadze Z	2016	Not a randomised controlled trial
105	Efficacy and safety of local stem cell transplantation for sequelae of spinal cord injury		2014	Not a randomised controlled trial
106	Collagen scaffold combined with human umbilical cord-mesenchymal stem cells transplantation for acute complete spinal cord injury	Wu-Sheng Deng	2020	Not a randomised controlled trial
107	Clinical study of human umbilical cord mesenchymal stem cells for repairing spinal cord injury by local combined intrathecal transplantation	Feng Shiqing	2022	Registered Trial (ChiCTR2200061962), no publication available
108	Different Efficacy Between Rehabilitation Therapy and Stem Cells Transplantation in Patients With SCI in China (SCI-III)		2012	Registered Trial (NCT01873547), no publication available
109	Granulocyte colony-stimulating factor-mediated neuroprotective therapy for acute spinal cord injury: from bench to bedside	Koda M	2015	No use of stem cells in spinal cord injury
110	Studying the effect of bone marrow mesenchymal stem cell injection into the subarachnoid space on improvement and reduction of complications in spinal cord injured patients	Farshad Homayouni Moghadam	2014	Registered Trial (IRCT2013050813267N1), no publication available

111	A Clinical Trial to study the safety & effectiveness of Adult Stem Cells derived from Bone Marrow via different routes of administration in the treatment of patients with complete Spinal Cord Injury (SCI)	Sreedhar Singamala	2011	Registered Trial (CTRI/2010/091/001469), no publication available
112	Role of Stem Cells in Improving Implantation Rates in ICSI Patients	Nawara Mohamed Hashish	2015	No use of stem cells in spinal cord injury
113	The Effect of Peer Education on Stem Cell Donation in Nursing Students	Dilek YILDIRIM	2023	No use of stem cells in spinal cord injury
114	NTCP model validation method for DAHANCA patient selection of protons versus photons in head and neck cancer radiotherapy	CR Hansen	2019	No use of stem cells in spinal cord injury
115	Six weeks of oral Echinacea purpurea supplementation does not enhance the production of serum erythropoietin or erythropoietic status in recreationally active males with above-average aerobic fitness	TD Martin	2019	No use of stem cells in spinal cord injury
116	Autologous Cell-derived Tissue Engineered Cartilage for Repairing Articular Cartilage Lesions	Quanyi Guo	2016	No use of stem cells in spinal cord injury
117	Valproate and Levocarnitine in Children with Spinal Muscular Atrophy	Sheffali Gulati	2012	No use of stem cells in spinal cord injury

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3. AMYOTROPHIC LATERAL SCLEROSIS

i. **Key question in PICO format**

ii. **Search Strategy**

iii. **PRISMA Flow Diagram**

iv. **Summary of included studies**

v. **Classification of studies based on level of stem cell manipulation**

vi. **Evidence to Decision Framework**

vii. **Data Extraction Sheet**

viii. **List of excluded studies**

i. Key question in PICO format

In patients with amyotrophic lateral sclerosis, what is the efficacy and safety of stem cell therapy as compared to usual care?

Population: Patients with Amyotrophic Lateral Sclerosis (ALS). Subgroup analysis: Duration, age, comorbidities, clinical subtypes of ALS, Familial ALS
Intervention: Any Stem cell and product derived from stem cells
Comparator: Usual Care/ Conventional Care
Critical Outcomes: Efficacy outcomes: Any measure of progressive functional disability and/or handicap-Pulmonary Function, tracheostomy-free Life; tube-free feeding;
Safety outcomes: Mortality, serious adverse events;
Important: Quality of Life, adverse events

ii. Search Strategy

The following search strategy was adopted to search various databases and obtain eligible reports for inclusion in the present meta-analysis.

PubMed

#1 "Amyotrophic Lateral Sclerosis"[MeSH Terms] OR "Amyotrophic Lateral Sclerosis" OR "Motor Neuron Disease" OR "MND"
#2 "Stem Cells"[MeSH Terms] OR "Stem Cell Transplantation"[MeSH Terms] OR "Stem Cells" OR "Stem Cell Therapy" OR "Cell Therapy" OR "Cell Transplantation" OR "Regenerative Medicine"
#3 "Randomized Controlled Trial"[Publication Type] OR "Randomized Controlled Trials as Topic"[MeSH Terms] OR "RCT" OR "Random Allocation" OR "Double-Blind Method" OR "Single-Blind Method"
#4 #1 AND #2 AND #3

Scopus

("Amyotrophic Lateral Sclerosis"[MeSH Terms] OR "Amyotrophic Lateral Sclerosis" OR "ALS" OR "Motor Neuron Disease" OR "MND")
AND
("Stem Cells"[MeSH Terms] OR "Stem Cell Transplantation"[MeSH Terms] OR "Stem Cells" OR "Stem Cell Therapy" OR "Cell Therapy" OR "Cell Transplantation" OR "Regenerative Medicine")
AND
("Randomized Controlled Trial"[Publication Type] OR "Randomized Controlled Trials as Topic"[MeSH Terms] OR "RCT" OR "Random Allocation" OR "Double-Blind Method" OR "Single-Blind Method")

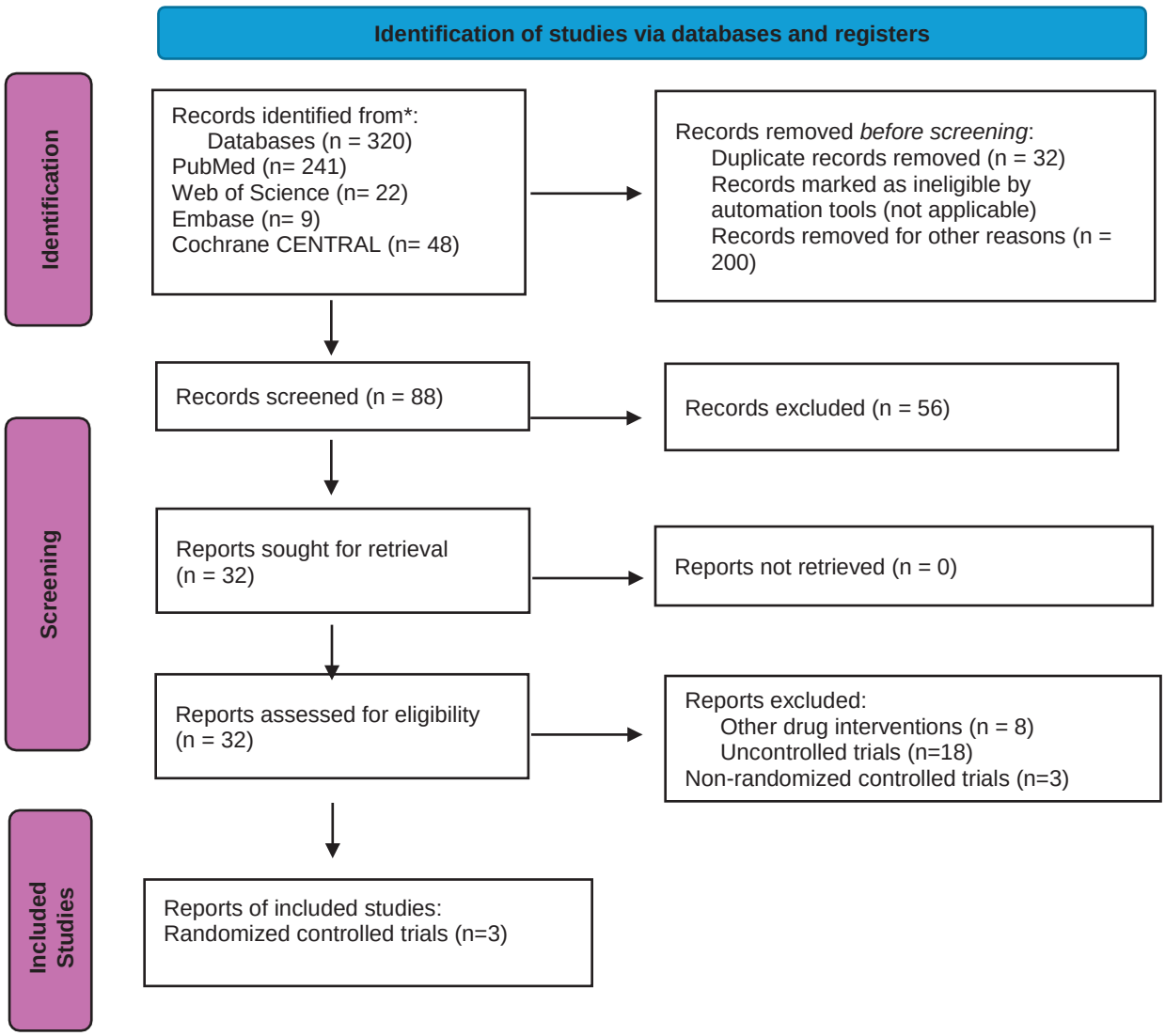
Embase

('amyotrophic lateral sclerosis'/exp OR 'amyotrophic lateral sclerosis' OR 'ALS' OR 'motor neuron disease' OR 'MND')
AND

('stem cell'/exp OR 'stem cell transplantation'/exp OR 'stem cell therapy' OR 'cell therapy' OR 'cell transplantation' OR 'regenerative medicine')
AND
('randomized controlled trial'/exp OR 'RCT' OR 'random allocation' OR 'double-blind method' OR 'single-blind method')

Cochrane Library
#1 [MeSH] "Amyotrophic Lateral Sclerosis" OR "Amyotrophic Lateral Sclerosis" OR "ALS" OR "Motor Neuron Disease" OR "MND"
#2 [MeSH] "Stem Cells" OR [MeSH] "Stem Cell Transplantation" OR "Stem Cell Therapy" OR "Cell Therapy" OR "Cell Transplantation" OR "Regenerative Medicine"
#3 [MeSH] "Randomized Controlled Trial" OR "RCT" OR "Random Allocation" OR "Double-Blind Method" OR "Single-Blind Method"
#4 #1 AND #2 AND #3

iii. PRISMA Flow Diagram



iv. Summary of included studies

Study	Population	Intervention	Comparison	Outcomes
Oh et al. 2018[1]	Participants (age 25–75 years) were diagnosed with clinically probable or definite ALS according to the revised El Escorial criteria	Received 2 injections of bone marrow–derived mesenchymal stem cells (BM-MSCs) at a 26-day interval, 1 × 10^6 cells/kg Plus Riluzole (100mg/day), and symptomatic treatments	Regular treatment with riluzole (100 mg/day), and symptomatic treatments	**Primary Outcome:** The mean change of ALSFRS-R score from Visit 5 (baseline, 0 months) to Visit 9 (+4 months) **Secondary Outcomes:** • The changes in slope (monthly rate of decline) of ALSFRS between the lead-in and follow-up periods • Responder analyses at 4 and 6 months after treatment **Additional secondary outcomes:** • The mean change of AALS (30 [normal] to 164 [maximally impaired]) scores at 4 months • mean changes of FVC at 4 months • changes in slope of AALS score and FVC between the lead-in and follow-up periods
Cudkowicz et al. 2022[2]	Eligible participants were 18–60 Y old with possible, laboratory supported probable, probable, or definite ALS per the revised-ElEscorial criteria,7 and revised ALS Functional Rating Scale8 (ALSFRS-R) total score ≥ 25 at screening	Autologous transplantation of bone marrow derived mesenchymal stem cells propagated ex vivo and induced to secrete neurotrophic factors.	Patients with three intrathecal administrations of Placebo at bi-monthly intervals	**Primary Outcome:** A responder analysis based on change in disease progression determined by the rate of change in ALSFRS-R per month using linear regression, with a separate line fit to pre-treatment and post-treatment data through week 28. **Secondary Outcomes:** • The percentage of participants with post-treatment rate of decline improved by ≥100% as measured by ALSFRS-R slope • change from baseline to week 28 in ALSFRS-R, • The combined analysis of function and survival (CAFS) and SVC. **Safety objective:** • Adverse events (AEs) • The Columbia-Suicide Severity Rating Scale (CSSRS) physical examination, vital signs, and electrocardiogram (ECG) assessments

Berry et al. 2019[3]	At screening, eligible participants aged 18–75 years had a diagnosisof possible, probable, laboratory-supported, probable, or definite ALS by El Escorial Criteria,6 ALSFRS-R ≥30, vital capacity (VC) ≥65% of the predicted normal value and symptoms duration of between 1 and 2 years.	Administered 125×10^6 MSC-NTF cells on a single occasion intrathecal and 48×10^6 MSC-NTF cells at 24 separate sites on the right upper arm biceps and triceps muscles by intramuscular injection	Excipient administration by combined intramuscular and intrathecal administration	**Primary Outcomes:** Safety, assessed with respect to the incidence of treatment-emergent AEs (TEAEs) and serious AEs (SAEs), laboratory abnormalities, vital signs, ECGs, and physical examinations. **Secondary Outcomes:** • The change in slopes (post-treatment slope through 24 weeks as compared to the pretreatment slope) in ALSFRS-R and SVC between the treatment and placebo groups. • The post-treatment slope of decline in the ALSFRS-R and SVC at 24 weeks following transplantation relative to the 12- to 16-week baseline period before transplantation (pretreatment slope) in all patients (within each treatment group) **Exploratory Outcomes:** • Hand-held dynamometry (HHD), compared muscle strength in the IM transplanted arm to the noninjected arm. CSF for changes in NTF, biomarker and microRNA expression between treated and placebo groups and between pre-transplant and post-transplant time points.

v. Classification of studies on the basis of level of manipulation of the stem cell/ Stem cell derived product used (*interpreted by the Secretariat as defined by CDSCO* and the information provided in the trial itself*):

S. No.	Trial Details	Intervention	Level of manipulation	Information extracted from trial
1.	Oh et al. 2018	Bone marrow–derived mesenchymal stem cells (BM-MSCs)- 2 injections at a 26-day interval, 1 × 10^6 cells/kg, Plus Riluzole (100mg/day), and symptomatic treatments	More	*"BM-MSCs were isolated, expanded, and analyzed under Good Manufacturing Practice (GMP) conditions at CORESTEM Inc. (Seoul, Korea), based on the International Society of Cellular Therapy guidelines."*
2.	Cudkowicz et al. 2022	Autologous bone marrow derived mesenchymal stem cells-propagated ex vivo and induced to secrete neurotrophic factors	More	*MSC-NTF (NurOwn®, manufactured by Dana-Farber Cancer Institute, Boston, MA or City of Hope, Duarte, CA; Mesenchymal stem cells (MSC) derived from adult bone marrow were induced under proprietary ex vivo conditions to secrete high levels of neurotrophic factors (MSC-NTF).*
3.	Berry et al. 2019	Mesenchymal stem cell (MSC)-neurotrophic factor (NTF) cells-Administered 125×10^6 MSC-NTF cells on a single occasion intrathecal and 48×10^6 MSC-NTF cells at 24 separate sites on the right upper arm biceps and triceps muscles by intramuscular injection	More	*The cell therapy product was manufactured at 2 central FDA-* *cleared cGMP-compliant manufacturing facilities using standard operating procedures. MSCs were isolated from the bone marrow, expanded, and differentiated to secrete NTFs using a culture-based approach.*

* as defined by CDSCO in Annexure- I of the guideline document.

vi. Evidence to Decision Framework

QUESTION

What is efficacy and safety of stem cell therapy as compared to usual care in Amyotrophic Lateral Sclerosis?	
POPULATION:	Amyotrophic Lateral Sclerosis (ALS)
INTERVENTION:	Stem cell therapy
COMPARISON:	Usual care
MAIN OUTCOMES:	Functional status (ALS Function Rating Scale-R), All-cause mortality, Forced Vital Capacity, Slow Vital Capacity, SAEs
SETTING:	Tertiary care centers
PERSPECTIVE:	Health System/Population
BACKGROUND:	Amyotrophic lateral sclerosis (ALS) is a fatal neurodegenerative disorder. It is a type of motor neuron disease characterized by progressive degeneration of neurons in the brain and spinal cord and is more common in men. The illness is relentlessly progressive, leading to death from respiratory paralysis and the median survival is between 3–5 years.
CONFLICT OF INTERESTS:	None

ASSESSMENT

Problem

Is the problem a priority?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
○ No ○ Probably no ○ Probably yes ● Yes ○ Varies ○ Don't know	The incidence of ALS is between 0.6 and 3.8 per 100 000 person-years.[4] None of the current disease modifying therapies reverse ALS progression and clinical care is associated with high costs for the patients and their families. Currently there is no cure for ALS and the treatment is mainly supportive.	

Desirable Effects How substantial are the desirable anticipated effects?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○Trivial ○Small ○Moderate ○Large ○ Varies ● Don't know	**Revised Amyotrophic Lateral Sclerosis Functional Rating Scale (ALSFRS-R) score:** Two trials with 248 participants reported the ALSFRS-R score. The mean difference for change from baseline in ALSFRS-R score between the stem cell therapy arm as compared to usual care at 6 months follow up was 1.82 (95% CI: -1.14 to 4.77), which was statistically non-significant. **Vital Capacity:** Oh et al[1] reported the mean difference of change from baseline in FVC between the stem cell arm and the usual care to be-0.53 (95 % CI: -5.37 to 4.31) at the end of four months, which was statistically not significant. Cudkowicz et al[2] reported the mean difference of change in SVC to be -1.39 (95% CI: -6.39 to 3.61), which was statistically non-significant.	A change of ALSFRS score by 3.24 was considered as MCID. A change in FVC by 2-6% was considered as MCID.
Undesirable Effects How substantial are the undesirable anticipated effects?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○Trivial ○Small ○Moderate ○Large ○Varies ● Don't know	**Serious adverse events:** Three RCTs with 301 participants reported SAEs and the pooled analysis yielded a risk ratio of 1.15 (95% CI: 0.72 to 1.85) in the stem cell group as compared to usual care, which was statistically non-significant. Three RCTs with 301 participants reported all-cause mortality and the pooled analysis yielded a risk ratio of 1.20 (95% CI: 0.51 to 2.79) in the stem cell group as compared to usual care, which was statistically non-significant.	
Certainty of evidence What is the overall certainty of the evidence of effects?		

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
● Very low ○ Low ○ Moderate ○ High ○ No included studies	The quality of the evidence is very low quality due to the high risk of bias, inconsistency, and imprecision in the reported studies.	
Values Is there important uncertainty about or variability in how much people value the main outcomes?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Important uncertainty or variability ○ Possibly important uncertainty or variability ● Probably no important uncertainty or variability ○ No important uncertainty or variability	ALS patients in Locked in state (LIS) maintain a high sense of well-being despite severe physical restrictions. They are content with their life-sustaining treatments and have a strong will to live.[5]	Main outcome is functional status and mortality which are likely to be valued by most patients
Balance of effects Does the balance between desirable and undesirable effects favor the intervention or the comparison?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**

○Favors the comparison ○Probably favors the comparison ●Does not favor either the intervention or the comparison ○Probably favors the intervention ○Favors the intervention ○Varies ○Don't know	It is less clear if the benefits of stem cell therapy outweigh the harms due to unclear benefits and harms.	
Resources required How large are the resource requirements (costs)?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
●Large costs ○ Moderate costs ○Negligible costs and savings ○Moderate savings ○Large savings ○Varies ○Don't know	No direct evidence on the resources required for stem cell transplantation in patients with ALS was identified. However, there is indirect evidence in the form of Hematopoietic stem cell transplantation done for hematological conditions. The median cost of autologous transplant was USD, $ 12,500 (approx. Rs 10.5 lakhs) and the median cost of allogeneic transplant was $ 17,914 (approx. 15 lakhs) as reported by a private center in New Delhi, India.[6]	
Certainty of evidence of required resources What is the certainty of the evidence of resource requirements (costs)?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL**

		CONSIDERATIONS
○Very low ○Low ●Moderate ○High ○No included studies	No research evidence was identified.	The GDG was fairly confident that the certainty of evidence of resources required is moderate.
Cost effectiveness Does the cost-effectiveness of the intervention favor the intervention or the comparison?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○Favors the comparison ●Probably favors the comparison ○ Does not favor either the intervention or the comparison ○Probably favors the intervention ○Favors the intervention ○Varies ○No included studies	No research evidence was identified	Because of no clear benefit and large costs associated with stem cell therapy, the committee opined that the cost effectiveness probably favors the comparison
Equity What would be the impact on health equity?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**

○Reduced ●Probably reduced ○Probably no impact ○Probably increased ○Increased ○Varies ○Don't know	No research evidence was identified	As stem cell therapy is an expensive treatment offered only at tertiary centers, it is likely to reduce equity.
Acceptability Is the intervention acceptable to key stakeholders?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○No ○Probably no ●Probably yes ○Yes ○Varies ○Don't know	No research evidence was identified.	Despite uncertainty regarding the risks and benefits associated with stem cell therapy, patients may still consider undergoing treatment in private clinics.
Feasibility Is the intervention feasible to implement?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○No ○Probably no ●Probably yes ○Yes ○Varies ○Don't know	No research evidence was identified	Feasible to implement in tertiary care centers.

SUMMARY OF JUDGEMENTS

	JUDGEMENT						
PROBLEM	No	Probably no	Probably yes	**Yes**		Varies	Don't know
DESIRABLE EFFECTS	Trivial	Small	Moderate	Large		Varies	**Don't know**
UNDESIRABLE EFFECTS	Trivial	Small	Moderate	Large		Varies	**Don't know**
CERTAINTY OF EVIDENCE	**Very low**	Low	Moderate	High			No included studies
VALUES	Important uncertainty or variability	Possibly important uncertainty or variability	**Probably no important uncertainty or variability**	No important uncertainty or variability			
BALANCE OF EFFECTS	Favors the comparison	Probably favors the comparison	**Does not favor either the intervention or the comparison**	Probably favors the intervention	Favors the intervention	Varies	Don't know
RESOURCES REQUIRED	**Large costs**	Moderate costs	Negligible costs and savings	Moderate savings	Large savings	Varies	Don't know
CERTAINTY OF EVIDENCE OF REQUIRED RESOURCES	Very low	Low	**Moderate**	High			No included studies
COST EFFECTIVENESS	Favors the comparison	**Probably favors the comparison**	Does not favor either the intervention or the comparison	Probably favors the intervention	Favors the intervention	Varies	No included studies

	JUDGEMENT						
EQUITY	Reduced	**Probably reduced**	Probably no impact	Probably increased	Increased	Varies	Don't know
ACCEPTABILITY	No	Probably no	**Probably yes**	Yes		Varies	Don't know
FEASIBILITY	No	Probably no	**Probably yes**	Yes		Varies	Don't know

TYPE OF RECOMMENDATION

Strong recommendation against the intervention	Conditional recommendation against the intervention	**Use in the context of rigorously conducted RCTs.**	Conditional recommendation for either the intervention or the comparison	Conditional recommendation for the intervention	Strong recommendation for the intervention
○	○	●	○	○	○

CONCLUSIONS

Recommendation

Stem Cell Therapy is **not recommended** in routine clinical practice for the treatment of amyotrophic lateral sclerosis. It may be used only in the context of rigorously conducted randomized controlled trials.

Justification

Overall justification

This recommendation has been made as the evidence is inadequate in quantity and quality to determine the safety and efficacy of stem cell therapy in patients with ALS. The difference in the Amyotrophic Lateral Sclerosis Functional Rating Scale (ALSFRS) score between the two arms was statistically non-significant. The difference in the forced vital capacity and slow vital capacity between both arms was also statistically non-significant. The difference in all-cause mortality and serious adverse events in the stem cell arm as compared to usual care was also statistically non-significant. In addition, the follow up period of one year is too small to comment on the side effect profile and long-term safety is not known. Results should be interpreted with caution, in view of very few studies with small number of participants and/or events.

Detailed justification

Desirable effects

This recommendation has been made as the evidence is inadequate in quantity and quality to determine the safety and efficacy of stem cell therapy in patients with ALS. The difference in the Amyotrophic Lateral Sclerosis Functional Rating Scale (ALSFRS) score between the two arms was statistically non-significant. The difference in the forced vital capacity and slow vital capacity between both arms was also statistically non-significant.

Undesirable effects

The difference in all-cause mortality and serious adverse events in the stem cell arm as compared to usual care was also statistically non-significant. In addition, the follow up period of one year is too small to comment on the side effect profile and long term safety is not known.

Certainty of evidence

Very low certainty of Evidence

Resources required

The committee opined that stem cell treatment is associated with large costs.

vii. Data Extraction:

For this systematic review, studies were selected based on the predefined criteria for inclusion and exclusion. Two investigators independently collected data from eligible articles, which included information such as the name of the authors and the year of publication, details of trials with registration numbers, the number of subjects in the group receiving treatment and those receiving usual care, the mean age and gender ratio of patients in different groups, the type of stem cells used, the dose and method of administration, the site and duration of the disease onset, ALS functional rating scores (ALSFRS-R), respiratory function forced or slow vital capacity (F/SVC) before and after intervention, as well as any adverse events that occurred.

For conducting statistical analyses related to meta-analysis the Comprehensive Meta-analysis v4 software (CMA) was used to assess the heterogeneity among the included studies, Q statistics, probability values of heterogeneity, and I^2 values (Melsen et al. 2014) were used. This allowed us to choose an appropriate effect model for the meta-analysis. For homogeneous outcomes (probability heterogeneity $p > 0.05$ and $I^2 < 50$), a fixed effect model was used, while for heterogeneous outcomes (probability heterogeneity $p < 0.05$ and $I^2 > 50$), a random effect model (Hedges and Vevea, 1998) was used. Tests with probability values less than 0.05 were deemed significant. To grade the evidence and prepare the summary finding table, GRADEpro GDT (Guyatt et al. 2011) software was used.

Study	**Oh et al. 2018[1]**	**Cudkowicz et al. 2022[2]**	**Berry et al. 2019[3]**
Study type:	RCT	RCT	RCT
Number of participants:	59	189	48
Countries and setting:	Korea	USA	USA
Duration of study Follow up (post intervention):	24 weeks	28 weeks	24 weeks
Method of assessment of disease condition:	ALSFRS-R score, SVC	ALSFRS-R score, FVC	ALSFRS-R score, FVC
Subgroup analysis within study	Subgroup analysis is done for responder's analysis (<50%, ≥50%, ≥75% and ≥75% improvement in ALSFRS-R slope)	Subgroup analysis is done for Baseline ALSFRS-R (≤25 and >25)	Subgroup analysis is not done
Inclusion criteria:	• Age: 25-75, • Amyotrophic Lateral Sclerosis Functional Rating Scale–Revised (ALSFRS-R) score between 31 and 46,	• Patients diagnosed with ALS as defined by revised El Escorial criteria as following groups: possible, laboratory-supported probable, probable, or definite.	• Participants aged 18–75 years had a diagnosis of possible, probable, laboratory-supported, probable, or definite ALS by El Escorial

	• Stable riluzole treatment (50mg, twice daily) for at least 3 months before screening, • disease duration no longer than 5 years after the onset of the first symptom	• Having onset of ALS disease symptoms, including limb weakness within 24 months at the Screening Visit. • ALSFRS-R ≥ 25 at the screening Visit. • Upright slow vital capacity (SVC) measure ≥ 65% of predicted for gender, height, and age at the screening Visit. • Rapid progressors • Participants taking a stable dose of Riluzole are permitted in the study • Citizen or permanent resident of the United States or Canadian citizen able to travel to a US site for all follow-up study visits	Criteria • ALSFRS-R ≥30 • Vital capacity (VC) ≥65% of the predicted normal value for height • Symptom duration of between 1 and 2 years • Participants were either not receiving riluzole or were on a stable dose for ≥30 days • Had the ability to lie flat for the IT cell transplant procedure, and had at least some limb weakness due to ALS
Exclusion criteria:	• Participation in other clinical trials within the past 12 months, • Forced vital capacity (FVC) of <40% of the predicted value, • Presence of any comorbidity that might interfere with the outcome • Tracheostomy or noninvasive ventilation, • Any hemorrhagic tendency • Administration of any drug that could affect the bone marrow	• Prior stem cell therapy of any kind • History of autoimmune or other serious disease (including malignancy and immune deficiency) that may confound study results • Current use of immunosuppressant medication or anticoagulants (per Investigator discretion) • Exposure to any other experimental agent or participation in an ALS clinical trial within 30 days prior to Screening Visit	• Use of mechanical ventilation • Presence of feeding tube; prior treatment with stem cells • Pregnancy • Exposure to investigational agents or immunosuppressive • therapy within 4 weeks of screening • Active autoimmune disease or infection (including hepatitis B, hepatitis C, or HIV) • Cancer within the previous 5 years • Unstable psychiatric or medical condition • Clinically significant abnormal

		• Use of RADICAVA (edaravone injection) within 30 days of screening or intent to use edaravone at any time during the course of the study including the follow up period • Use of non-invasive ventilation (BIPAP), diaphragm pacing system or invasive ventilation (tracheostomy) • Feeding tube • Pregnant women or women currently breastfeeding	safety laboratory values.
Recruitment/selection of patients:	A total of 71 participants with ALS were screened, of whom 7 were exclude. Of the remaining 64 participants, 33 were allocated to receive BM-MSCs with riluzole treatment (MSC group) and 31 received riluzole alone (control group). A total of 59 participants at 4 months and 56 participants at 6 months were include in the primary efficacy analysis	A total of 263 persons with ALS were screened for eligibility from whom 196 were randomly assigned to treatment group and 189 received at least one treatment. 7 participants did not receive treatment. A total of 45 participants discontinued the study prematurely.	Fifty-nine participants were screened in this trial. There were 8 screen failures, and during the 3-month run-in period, 3 Participants discontinued the study prior to randomization. Forty-eight participants were randomized, of whom 43 completed follow-up.
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Regular treatment with riluzole (100 mg/day), and symptomatic treatments	Autologous transplantation of bone marrow derived mesenchymal stem cells propagated ex vivo and induced to secrete neurotrophic factors.	Administered 125 × 10^6 MSC-NTF cells on a single occasion and 48 × 10^6 MSC-NTF cells at 24 separate sites on the right upper arm biceps and triceps muscles

Outcomes reported with time points

First Author &Year	**Oh et al. 2018[1]**		**Cudkowicz et al. 2022[2]**		**Berry et al. 2019[3]**	
Time Points	**24 weeks follow up**		**28 weeks follow up**		**24 weeks follow up**	
	MSC	**Control**	**MSC**	**Placebo**	**MSC**	**Placebo**

	Sample Size, n (M/F)	32 (18/14)	27 (11/16)	95 (68/27)	94 (59/35)	36 (25/11)	12 (10/2)
	Age, Yrs (Mean±SD)	53.7±7.7	52.5±9.4	48.1±9.71	49.1±8.38	50.3±11.9	53.5±9.11
	Sample Size (n)	32	27	95	94	36	12
	ALSFRS-R Score Baseline (Mean ± SD)	35.5 ± 4.2	34.7 ± 5.5	30.3 ± 6.5	31.4 ± 6.1	38.1 ± 3.6	38.6 ±3.85
	Sample Size (n)	31	25	95	94	36	12
	ALSFRS-R Score Change from lead-in to follow up (Mean ± SD)	32.4 ± 3.51	28.22 ± 4.53	24.78 ± 6.52	25.52 ± 6.49	Not Reported	Not Reported
	Sample Size (n)	32	27	95	94	36	12
	≥100% improvement in ALSFRS-R slope, n(%)	8 (25)	1 (4)	13 (13.7)	13 (13.8)	Not Reported	Not Reported
	Sample Size (n)	32	27	—	—	—	—
	FVC at baseline% (Mean ± SD)	73.1 ± 21.1	71.3±16.1	—	—	—	—
	FVC % change from baseline to 4 months (Mean ± SD)	-11.28 ± 10.06	-10.75 ± 8.40	—	—	—	—
	Sample Size (n)	–	–	95	94	36	12
	SVC Baseline % (Mean ± SD)	–	–	76.2 ± 20.9	75.0 ± 19.8	90.6 ± 18.1	88.7 ± 8.91
	SVC % predicted, LS mean change from baseline	–	–	-12.9 ± 1.80	-11.55 ± 1.81	Not Reported	Not Reported

	(SE)						
	Sample Size (n)	33	31	95	94	36	12
	Adverse drug reactions (n)	3	0	76	66	Not Reported	Not Reported
	Serious Adverse events (n)	3	6	23	17	1	0
	Death (n)	1	3	10	6	0	0
Funding	Supported by Ministry for Health and Welfare Affairs, Republic of Korea (HI10C1673 and HI15C0876) and CORESTEM Inc.		Funded by Brainstorm Cell Therapeutics. Supported by the California Institute for Regenerate Medicine (CIRM, CLIN2-09894) and a grant from the ALS Association and I AM ALS.		Brainstorm Cell Therapeutics		
RoB2 Assessment: Specify separately for each domain	D1: Low D2: Low D3: Low D4: Low D5: Low Overall: Low		D1: Some Concern D2: Low D3: Low D4: Low D5: Low Overall: Some Concern		D1: Low D2: Low D3: Some Concern D4: Low D5: Some Concern Overall: Some Concern		

viii. List of excluded studies:

Details of the studies not included in the present analysis and the reason for their exclusion:

Sl. No.	Study Name	Reason of exclusion
1	Petrou et al. 2021	Non-Randomised trial
2	Martinez et al. 2009	Non-Randomised trial
3	Sharma et al. 2015	Non-Randomised trial
4	Kim et al. 2014	Uncontrolled Trial
5	Oh et al. 2015	Uncontrolled Trial
6	Petrou et al. 2016	Uncontrolled Trial
7	Sykova et al. 2017	Uncontrolled Trial
8	Nabavi et al. 2019	Uncontrolled Trial
9	Mazzini et al. 2010	Uncontrolled Trial
10	Feldman et al. 2014	Uncontrolled Trial
11	Glass et al. 2016	Uncontrolled Trial
12	Mazzini et al. 2019	Uncontrolled Trial
13	Blanquer et al. 2012	Uncontrolled Trial
14	Deda et al. 2009	Uncontrolled Trial
15	Gamez et al. 2010	Uncontrolled Trial
16	Karussis et al. 2010	Uncontrolled Trial
17	Mazzine et al. 2012	Uncontrolled Trial
18	Prabhakar et al. 2012	Uncontrolled Trial
19	Riley et al. 2012	Uncontrolled Trial
20	Tarella et al. 2010	Uncontrolled Trial
21	Riley et al. 2014	Uncontrolled Trial
22	Wong et al. 2022	Other drugs
23	Pawlukowska et al. 2019	Other drugs
24	Pagsnoni et al. 2022	Other drugs
25	Oh et al. 2018	Other drugs
26	Miaer et al. 2015	Other drugs
27	Gotkine et al. 2023	Other drugs
28	Baloh et al. 2020	Other drugs
29	Fan Dong Sheng et al. 2019	Other drugs

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4. MULTIPLE SCLEROSIS

1). AUTOLOGOUS HEMATOPOIETIC STEM CELL TRANSPLANTATION

i. **Key question in PICO format**

ii. **Search Strategy**

iii. **PRISMA Flow Diagram**

iv. **Summary of included studies**

v. **Classification of studies based on level of stem cell manipulation**

vi. **Evidence to Decision Framework**

vii. **Data Extraction**

viii. **List of excluded studies**

i. Key question in PICO format

In patients with multiple sclerosis, what is the efficacy and safety of stem cell therapy as compared to usual care?

Population: Patients with Multiple Sclerosis. Subgroups: Primary Progressive Multiple Sclerosis (PPMS), Relapsing Remitting Multiple Sclerosis (RRMS), Secondary Progressive Multiple Sclerosis (SPMS), Highly Active Multiple Sclerosis.

Intervention: Any Stem cell and product derived from stem cells

Comparator: Usual Care/ Conventional Care

Critical Outcome: Efficacy outcomes: any measure of disability, any measure of relapse
Safety outcomes: Serious Adverse Events: mortality, any tumor formation
Important: Quality of life, Adverse Events

ii. Search Strategy

EMBASE
('stem cell transplantation' OR 'stem cell' OR 'mesenchymal stem cell' OR 'hematopoietic stem cell transplantation' OR 'hematopoietic stem cell transplantation' OR 'hematopoietic stem cell' OR 'stem cell mobilization' OR 'embryonic stem cell' OR 'bone marrow transplantation' OR 'allogenic bone marrow transplantation' OR 'neural stem cell' OR 'neural stem cell transplantation' OR 'cell transplantation' OR 'multipotent stem cell' OR 'induced pluripotent stem cell' OR 'induced pluripotent stem cell transplantation'):ab,ti AND (('multiple sclerosis' OR 'relapsing remitting multiple sclerosis' OR 'progressive multiple sclerosis' OR 'Highly Active Multiple Sclerosis' OR 'relapsing remitting multiple sclerosis'):ab,ti) filter Randomized controlled trial

Cochrane Library
Stem cell transplantation OR Hematopoietic stem cell mobilization OR Hematopoietic stem cell OR stem cells OR Cell transplantation OR Bone marrow transplantation OR Bone marrow mesenchymal OR Mesenchymal OR Stromal OR bone marrow mesenchymal stromal cell OR Neural stem cell OR Embryonic Stem cell in Title Abstract Keyword AND MULTIPLE SCLEROSIS OR MS OR Relapsing-remitting MS OR RRMS OR Primary progressive MS OR PPMS OR Highly Active Multiple Sclerosis OR Secondary progressive MS OR SPMS in Title Abstract Keyword AND Randomized controlled OR Randomized OR randomised OR RCT OR Randomised controlled trial in Title Abstract Keyword - (Word variations have been searched)

PubMed
{Stem cell transplantation} OR {Hematopoietic stem cell mobilization} OR {Hematopoietic stem cell} OR {stem cells} OR {Cell transplantation} OR {Bone marrow transplantation} OR {Bone marrow mesenchymal} OR {Mesenchymal} OR {Stromal} OR {bone marrow mesenchymal stromal cell} OR {Neural stem cell} OR {Embryonic Stem cell}) AND (({MULTIPLE SCLEROSIS} OR {MS} OR

{Relapsing-remitting MS} OR {RRMS} OR {Primary progressive MS} OR {PPMS} OR {Highly Active Multiple Sclerosis} OR {Secondary progressive MS} OR {SPMS}))Filters:Clinical Study, Clinical Trial, Meta-Analysis, Randomized Controlled Trial, Systematic Review, Humans

Web of Science

Web of Science Search Strategy (v0.1)
Database: Web of Science Core Collection
Entitlements:
- WOS.SCI: 1952 to 2023

Searches:
1: ((((((((((((((((TI=(stem cell transplantation)) OR TI=(stem cell)) OR TI=(mesenchymal stem cell))) OR TS=(hematopoietic stem cell transplantation)) OR TS=(hematopoietic stem cell transplantation)) OR TS=(hematopoietic stem cell)) OR TS=(stem cell mobilization)) OR TS=(embryonic stem cell)) OR TS=(bone marrow transplantation)) OR TS=(allogenic bone marrow transplantation)) OR TS=(neural stem cell)) OR TS=(neural stem cell transplantation)) OR TS=(cell transplantation)) OR TS=(multipotent stem cell)) OR TS=(induced pluripotent stem cell)) OR ALL=(induced pluripotent stem cell transplantation). Date Run: Fri Oct 20 2023 21:30:10 GMT+0530 (India Standard Time). Results: 532276

2: ((((TS=(multiple sclerosis)) OR TS=(relapsing remitting multiple sclerosis)) OR TS=(progressive multiple sclerosis)) OR TS=(Highly Active Multiple Sclerosis)) OR TS=(relapsing remitting multiple sclerosis). Date Run: Fri Oct 20 2023 21:36:30 GMT+0530 (India Standard Time). Results: 144591

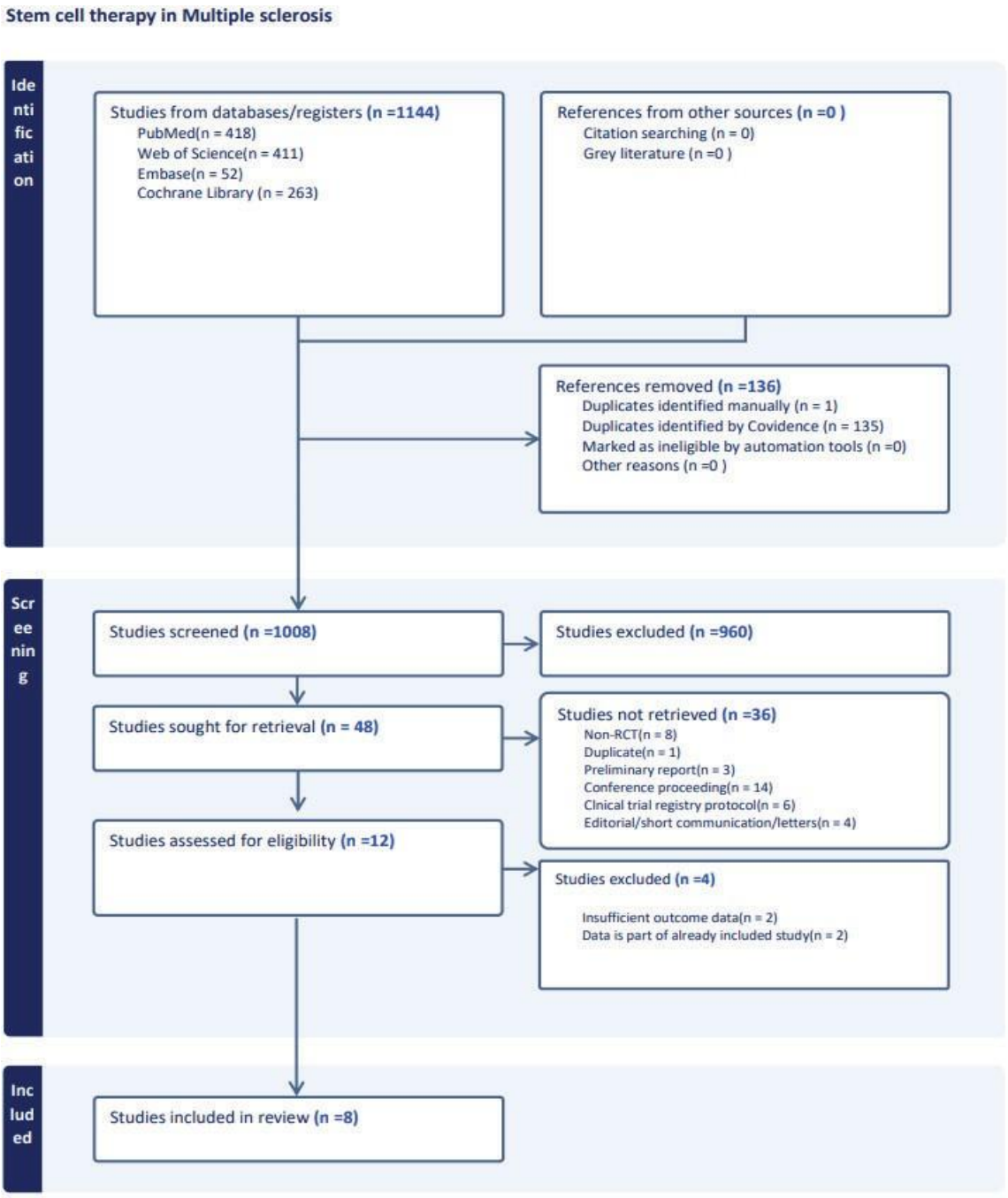
3: #2 AND #1. Date Run: Fri Oct 20 2023 21:36:41 GMT+0530 (India Standard Time). Results: 4114

4: (((((((((TS=(Randomized controlled trial)) OR TS=(Controlled trial)) OR TS=(Random Allocation)) OR TS=(Clinical trial Phase I)) OR TS=(Clinical trial Phase II)) OR TS=(Clinical trail Phase III)) OR TS=(Clinical Trail Phase IV)) OR TS=(Double blind method)) OR TS=(Single blind method)) OR TS=(Placebo). Date Run: Fri Oct 20 2023 22:32:38 GMT+0530 (India Standard Time).
Results: 963645

5: #3 AND #4 Date Run: Fri Oct 20 2023 22:32:58 GMT+0530 (India Standard Time). Results: 411

Last date of search strategy: Oct 2023

iii. PRISMA Flow Diagram



iv. Summary of included studies

Study	Population	Intervention	Comparison	Outcomes
Mancardi GL et al. 2015[1]	Twenty-one patients with confirmed MS AHSCT, n = 9 MTX, n = 12	Autologous hematopoietic stem cells transplantation (AHSCT)	mitoxantrone (MTX)	Primary endpoints: To determine the effect of AHSCT vs MTX on disease activity measured by the cumulative number of new T2 MRI lesions in the 4 years following randomization. Secondary endpoints: Activity of AHSCT vs MTX on the cumulative number of Gd-enhancing lesions on T1-weighted MRI from baseline to year 4. Secondary clinical endpoints: The cumulative number of relapses from baseline to year 4 and the time to disability progression confirmed after 6 and 12 months. AHSCT-related mortality, safety, side effects, early adverse events, and serious adverse events (SAEs) in the 2 arms.
Burt RK et al. 2019[2]	110 patients with relapsing-remitting MS, at least 2 relapses while receiving DMT in the prior year, and an Expanded Disability Status Scale (EDSS; score range, 0-10 [10 = worst neurologic disability]) score of 2.0 to 6.0	Nonmyeloablative Hematopoietic Stem Cell	Continued Disease-Modifying Therapy	The primary end point was time to disease progression, defined as an increase (worsening) in EDSS score of at least 1 point on 2 evaluations 6 months apart after at least 1 year of treatment, and not due to a non-MS disease process. Secondary end points survival; relapses; Neurologic Rating Scale (NRS) (range, 0-100 in 1-point increments from worst [0] to no [100] disability; minimal clinically important difference, 10) MRI T2-weighted lesion volume; Short Form 36 quality-of-life score (range, 1-100; higher scores indicate more favorable health state); Multiple Sclerosis Functional Composite (MSFC) score, which incorporates a timed 25-ft walk test; the 9-Hole Peg Test; and the Paced Auditory Serial Addition Test

v. Classification of studies on the basis of level of manipulation of the stem cell/ Stem cell derived product used (*interpreted by the Secretariat as defined by CDSCO* and the information provided in the trial itself*):

S. No.	Trial Details	Intervention	Level of manipulation	Information extracted from trial
1.	Mancardi GL et al[1] 2015	Autologous hematopoietic stem cells transplantation (AHSCT)- Peripheral hematopoietic stem cells (PBSCs) collected with a leukapheresis procedure and an unmanipulated graft containing between 3 and 8 x 10^6 CD34+/kg cells was cryopreserved. PBSC were then thawed and infused through the central venous catheter. Route: Intravenous	Less	*In the transplant arm, peripheral hematopoietic stem cells (PBSC) were mobilized by cyclophosphamide (4 g/m^2) in 1 day. After 5 days, filgrastim was administered daily (5 mg/kg body weight SC) until the completion of the stem cells harvest. Hematopoietic stem cells were then collected with a leukapheresis procedure and an unmanipulated graft containing between 3 and 8 x 10^6 CD34+/kg cells was cryopreserved.*
2.	Burt RK et al. 2019[2]	Non-myeloablative Hematopoietic Stem Cells- Peripheral blood stem cells were collected. Route: Intravenous	Less	*Peripheral blood stem cells were collected 10 days after intravenous cyclophosphamide (2 g/m^2) and 5 to 10 µg/kg per day of subcutaneous filgrastim beginning 5 days after cyclophosphamide.*

* as defined by CDSCO in Annexure- I of the guideline document.

vi. Evidence to Decision Framework

QUESTION

What is the efficacy and safety of hematopoietic stem cell transplantation compared to usual care in Multiple Sclerosis?	
POPULATION:	Patients with Multiple Sclerosis
INTERVENTION:	Stem cell therapy (AHSCT- Autologous Hematopoietic Stem Cell Transplantation)
COMPARISON:	Usual care
MAIN OUTCOMES:	EDSS at Six Months (EDSS 0-10, 0 good 10 worse)- EDSS at one year, Average or Annual relapse rate; Proportion free of relapse, Disease progression, Serious adverse events
SETTING:	Tertiary care centers
PERSPECTIVE:	Health system/Population
BACKGROUND:	Multiple sclerosis (MS) is one of the leading causes of neurological disability in young adults with symptom onset generally occurring between the ages of 20 to 40 years. It is an autoimmune inflammatory disorder characterized by demyelination of neurons in the central nervous system and affects women more commonly than men. Initially the episodes are reversible, that are followed by progressive neurological deterioration over time.
CONFLICT OF INTERESTS:	None

ASSESSMENT

Problem Is the problem a priority?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ No ○ Probably no ○ Probably yes ● Yes ○ Varies ○ Don't know	The prevalence of MS in our country has increased from 1.33/100,000 in 1985 to 8.35/100,000 in 2014.[3] There is no cure for MS and the current disease modifying therapies provide mainly symptomatic relief or shorten the duration of symptoms.	
Desirable Effects How substantial are the desirable anticipated effects?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Trivial ○ Small ○ Moderate ● Large ○ Varies ○ Don't know	**AHSCT:** **Disability:** One trial with 103 participants reported EDSS score at six months and at one year of follow up. The mean difference in EDSS score was -1.20 (95% CI: -1.76 to -0.64) at six months and -1.60 (95% CI: -2.20 to -1.00) at one year in the HSCT arm as compared to usual care. There is a statistically significant improvement in EDSS score both at six months (two times the MCID-dotted line) and at one year (three times the MCID-dotted line), which is important clinically. **Disease progression:** Two trials with 123 participants reported the disease progression to be lower at one year in the HSCT group as compared to the usual care group. The risk ratio for disease progression was 0.43 (95% CI: 0.02 to 10.32) in the	A change of EDSS score by 0.5 was considered as MCID. A difference of 0.6 for Annualized relapse rate (ARR) was considered as MCID. A difference of 20/100 (20%) was considered as MCID for Proportion free of relapse.

	HSCT arm as compared to usual care which was statistically non-significant. **Proportion free from relapse:** One trial with 103 participants reported that proportion of patients free of relapse at one year was higher in the HSCT group as compared to usual care with a risk ratio of 3.13 (95% CI: 2.08 to 4.70) and the results were statistically significant and important clinically. **Annualized relapse rate:** One trial with 20 participants reported a lower annualized relapse rate in the patients with HSCT as compared to usual care. The mean difference was -0.41 (95% CI: -0.69 to -0.13), which is statistically significant but unimportant clinically (less than MCID of 0.6).	
Undesirable Effects How substantial are the undesirable anticipated effects?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Trivial ○ Small ● Moderate ○ Large ○ Varies ○ Don't know	**AHSCT:** **Serious adverse events:** No deaths were reported. Two trials with 123 participants reported serious adverse events. Pooled analysis revealed a risk ratio of 21.46 (95% CI: 2.99 to 154.08) in the AHSCT arm as compared to the usual care. There is a increase in serious adverse events with AHSCT therapy as compared to usual care but the results had very serious imprecision. Burt et al[2], mentioned that there were no deaths and no non-hematopoietic grade 4 toxicities (such as myocardial infarction, sepsis, or other disabling or potential life-threatening events or transfer to intensive care unit). However, the following Grade 3 toxicities reported by Burt et al[2] were taken as serious adverse events in this analysis: febrile neutropenia (n=13), atrial fibrillation (n=1), Infection (n=4), engraftment bone pain (n=1), serum sickness (n=1), seizure (n=1).	

Certainty of evidence What is the overall certainty of the evidence of effects?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
● Very low ○ Low ○ Moderate ○ High ○ No included studies	The certainty of evidence is very low.	
Values Is there important uncertainty about or variability in how much people value the main outcomes?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Important uncertainty or variability ○ Possibly important uncertainty or variability ● Probably no important uncertainty or variability ○ No important uncertainty or variability	A nationwide survey in UK in patients with relapsing remitting multiple sclerosis (RRMS) reported that most respondents (73%) had personal goals related to lifestyle and many (69%) were concerned about maintaining independence.[4] In RRMS Patient preference focused on symptoms and prevention of progression but not on relapse prevention.[5] Pain contributed the most to multiple sclerosis outpatients' perception of health, followed by gait dysfunction and fatigue.[6]	The GDG also opined that an improvement in disability is likely to be highly valued by the patients.

Balance of effects Does the balance between desirable and undesirable effects favor the intervention or the comparison?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Favors the comparison ○ Probably favors the comparison ○ Does not favor either the intervention or the comparison ● Probably favors the intervention ○ Favors the intervention ○ Varies ○ Don't know	In the hematopoietic stem cell group, there is evidence for benefit along with a concomitant increase in harms.	
Resources required How large are the resource requirements (costs)?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
● Large costs ○ Moderate costs ○ Negligible costs and savings ○ Moderate savings ○ Large savings ○ Varies ○ Don't know	For Hematopoietic stem cell transplantation done for hematological conditions the median cost of autologous transplant was USD, $ 12,500 (approx. Rs 10.5 lakhs) and the median cost of allogeneic transplant was $ 17,914 (approx. Rs 15 lakhs) as reported by a private center in New Delhi.[7]	The cost in the government hospitals may be slightly lower

Certainty of evidence of required resources What is the certainty of the evidence of resource requirements (costs)?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Very low ○ Low ● Moderate ○ High ○ No included studies	One study was identified for HSCT therapy for hematological conditions.	
Cost effectiveness Does the cost-effectiveness of the intervention favor the intervention or the comparison?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Favors the comparison ○ Probably favors the comparison ● Does not favor either the intervention or the comparison ○ Probably favors the intervention ○ Favors the intervention ○ Varies ○ No included studies	A study on cost effectiveness reported meaningful cost savings for the use of HSCT in secondary Progressive Multiple sclerosis in the base case scenario, but also suggested that further research is needed to enable meaningful assessment of cost effectiveness.[8]	

Equity What would be the impact on health equity?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Reduced ● Probably reduced ○ Probably no impact ○ Probably increased ○ Increased ○ Varies ○ Don't know	No research evidence available.	Probably reduced as the treatment is costly and is available in tertiary care centres/ big private hospitals in major cities only.
Acceptability Is the intervention acceptable to key stakeholders?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ No ○ Probably no ● Probably yes ○ Yes ○ Varies ○ Don't know	No research evidence was identified on the acceptability on the use of stem cell therapy in multiple sclerosis.	Despite uncertainty regarding the medical risks and benefits associated with stem cell therapy, patients may still consider undergoing treatment in private, unregulated clinics

Feasibility Is the intervention feasible to implement?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ No ○ Probably no ● Probably yes ○ Yes ○ Varies ○ Don't know	No research evidence was identified on the feasibility on the use of stem cell therapy in multiple sclerosis.	Feasible to implement in tertiary care centres.

SUMMARY OF JUDGEMENTS (HSCT)

	JUDGEMENT						
PROBLEM	No	Probably no	Probably yes	**Yes**		Varies	Don't know
DESIRABLE EFFECTS	Trivial	Small	Moderate	**Large**		Varies	Don't know
UNDESIRABLE EFFECTS	Trivial	Small	**Moderate**	Large		Varies	Don't know
CERTAINTY OF EVIDENCE	**Very low**	Low	Moderate	High			No included studies
VALUES	Important uncertainty or variability	Possibly important uncertainty or variability	**Probably no important uncertainty or variability**	No important uncertainty or variability			
BALANCE OF EFFECTS	Favors the comparison	Probably favors the	Does not favor either the	**Probably favors the**	Favors the intervention	Varies	Don't know

	JUDGEMENT						
		comparison	intervention or the comparison	**intervention**			
RESOURCES REQUIRED	**Large costs**	Moderate costs	Negligible costs and savings	Moderate savings	Large savings	Varies	Don't know
CERTAINTY OF EVIDENCE OF REQUIRED RESOURCES	Very low	Low	**Moderate**	High			No included studies
COST EFFECTIVENESS	Favors the comparison	Probably favors the comparison	**Does not favor either the intervention or the comparison**	Probably favors the intervention	Favors the intervention	Varies	No included studies
EQUITY	Reduced	**Probably reduced**	Probably no impact	Probably increased	Increased	Varies	Don't know
ACCEPTABILITY	No	Probably no	**Probably yes**	Yes		Varies	Don't know
FEASIBILITY	No	Probably no	**Probably yes**	Yes		Varies	Don't know

TYPE OF RECOMMENDATION

Strong recommendation against the intervention	Conditional recommendation against the intervention	Conditional recommendation for either the intervention or the comparison	**Conditional recommendation for the intervention**	Strong recommendation for the intervention
○	○	○	●	○

CONCLUSIONS

Recommendation

a) Autologous Hematopoietic Stem Cell Transplantation is **recommended** for the treatment of highly active relapsing remitting multiple sclerosis*, if there is no satisfactory improvement with disease modifying therapies. (Conditional#).

*The evidence overwhelmingly comes from Relapsing Remitting Multiple Sclerosis. It is not known whether a HSCT is effective in other forms of MS (relapsing progressive, secondary progressive).

#

A. Highly active treatment-resistant relapsing MS, defined as ≥ 2 episodes of disease activity in the 36 months prior to the assessment for AHSCT. The two disease activity episodes will be a clinical MS relapse or MRI evidence of MS disease activity and must meet all the criteria described below:

1. At least one episode of disease activity must occur following ≥ 1 month of treatment with one of the following: (i) a DMT approved for the treatment of relapsing MS, or (ii) a monoclonal antibody approved for the treatment of relapsing MS, or (iii) rituximab. Qualifying DMTs include: dimethyl fumarate, diroximel fumarate, monomethyl fumarate, teriflunomide, cladribine, daclizumab, ponesimod, siponimod, ozanimod, fingolimod, rituximab, ocrelizumab, natalizumab, alemtuzumab, ublituximab, and ofatumumab, and
2. At least one episode of disease activity must have occurred within the 12 months prior to the AHSCT, and
3. At least one episode of disease activity must be a clinical MS relapse confirmed by a neurologist. The other episode(s) must occur at least one month before or after the onset of the clinical MS relapse, and must be either another clinical MS relapse or MRI evidence of disease activity in the form of a gadolinium-enhancing lesion, or a new non-enhancing T2 lesion compared to a reference scan obtained not more than 36 months prior to the time of evaluation.

B. Expanded Disability Status Scale (EDSS) ≤ 6

C. No contraindications to AHSCT

Justification

Overall justification

This recommendation has been made as there is very low certainty evidence of a large benefit and known harms associated with autologous HSCT. The committee decided that benefits clearly outweigh harms. There seems to be a clinically important improvement in EDSS score at 6 months (greater than two times of MCID) and at one year (greater than three times of MCID) that was statistically significant. The proportion of patients free of relapse was higher in the HSCT group as compared to usual care and the results were statistically significant. There was a statistically non-significant difference in disease progression between the stem cell arm as compared to usual care. Serious adverse events were higher in the HSCT group, but the results were highly imprecise. No deaths were reported in either group.

Detailed justification

Desirable Effects

One trial with 103 participants reported EDSS score at six months and at one year of follow up. The mean difference in EDSS score was -1.20 (95% CI: -1.76 to -0.64) at six months and -1.60 (95% CI: -2.20 to -1.00) at one year in the HSCT arm as compared to usual care. There seems to be a clinically important and statistically significant improvement in EDSS score both at six months (two times the MCID) and at one year (three times the MCID).

Two trials with 123 participants reported the disease progression to be lower at one year in the HSCT group as compared to the usual

care group. The risk ratio for disease progression was 0.43 (0.02 to 10.32) in the HSCT arm as compared to usual care but the results were not significant statistically. One trial with 103 participants reported that proportion of patients free of relapse was higher in the HSCT group as compared to usual care with a risk ratio of 3.13 (95% CI: 2.08 to 4.70) and the results were statistically significant.
One trial with 20 participants reported a lower annualized relapse rate in the patients with HSCT as compared to usual care. The mean difference was -0.41 (95% CI: -0.69 to -0.13) but was not important clinically (less than MCID of 0.6).

Undesirable Effects

No deaths were reported. Two trials with 123 participants reported serious adverse events. Pooled analysis revealed a risk ratio of 21.46 (95% CI: 2.99 to 154.08) in the HSCT arm as compared to the usual care. There seems to be an increase in serious adverse events with HSCT therapy as compared to controls but the results were highly imprecise.

Certainty of evidence

Very low certainty of Evidence

Resources required

The committee opined that stem cell treatment is associated with large costs.

Subgroup considerations

None

Implementation considerations

The autologous hematopoietic stem cell transplantation should be performed at centers that are well equipped and have substantial experience in carrying out hematopoietic stem cell transplantation.

Monitoring and evaluation

Not applicable

vii. Data Extraction:

Data Extraction Methods:

Three independent reviewers extracted the relevant data from the included studies and any discrepancies were resolved by consensus among all the reviewers. The following key data *viz* author(s)/year of publication, country, duration of enrollment of subjects, Male female ratio, mean age, total sample size, sample size in the intervention arm, sample size in the control arm, dose of stem cell infusion, route of infusion, types of stem cells, mean duration of disease, types of MS, follow up duration/ data taken for the time point at outcome assessment (before the cross over period), for the presentation of characteristic of included studies were extracted. The outcome data was extracted by the same researchers for pooled analysis. The data in which values were not reported in text and represented in the graphical format, then web plot digitizer (WebPlotDigitizer - Copyright 2010-2024 Ankit Rohatgi (automeris.io) was used for the extracting the possible data. The corresponding author was also contacted for providing the relevant data through email in case the needed data were not reported in the published paper.

Heterogeneity was assessed using the I2 values. If I^2 is less than 50%, then fixed-effect model for pooling the results was used. In the case of I2 greater than 50%, the source of heterogeneity was examined by reviewing the PICO of each study. We conducted a subgroup analysis in the case of heterogeneity, explained by the differences in the PICO, and results were provided for the same. In the absence of critical differences in the PICO, we used the random-effects model to pool the results of all the included studies. Continuous outcome data were pooled using a mean difference with a 95% confidence interval in cases of similar units or a standardized mean difference in cases of variation of units in the outcome data. A risk ratio with a 95% confidence interval was used to pool the data for a categorical outcome. Publication bias was assessed using a funnel plot or statistical tests (Begg's or Egger's) in cases where the number of studies was equal to or more than 10 for any outcome data. In the event of the unavailability of data reported in the number in the text or table and data is presented in the graph, a graph reader (http://www.graphreader.com/) was used to extract the data, which might have an effect on a little variation from the original values. If the IQR data was reported, then the SD was estimated using the formula IQR/1.35. In cases of cross-over trials, outcome data was collected only for the period of no cross-over. In case of outcome data could not retrieved then we sent email to corresponding author for providing the relevant data with two reminders. All the statistical analyses were conducted using the software RevMan version 5.4.1.

As pre-specified the sensitivity analyses was conducted considering high risk of bias study to explore impact to the excluding and including studies in the pooled effect size. As pre-specified the subgroup analysis had to be conducted based in cases of heterogeneity could be explained by the differences in the PICO. However, due to no significant heterogeneity was observed in the primary outcome EDSS, we did not conduct the subgroup analysis for heterogeneity exploration.

Study	**Mancardi et al. 2015[1]**
Study type:	RCT
Number of participants:	21
Countries and setting:	Italy, Spain, France Setting: Hospital
Duration of study Follow up (post intervention)	4 years
Method of assessment of disease condition:	Active disease was defined as follows, as detailed elsewhere: clinically defined MS,23 a secondary progressive (SP) or relapsing remitting (RR) form that accumulates disability between relapses, with a documented worsening during the last year (1 step of Expanded Disability Status Scale [EDSS], or 0.5 when EDSS is between 5.5 and 6.5), in spite of conventional therapy (interferon-b or glatiramer acetate or immunosuppressive therapy), and presence of one or more gadolinium (Gd)-enhancing areas on MRI. The EDSS score had to be between 3.5 and 6.5
Subgroup analysis within study	No sub-group analysis was conducted.
Inclusion criteria:	Patients eligible for the study had clinically defined MS, a secondary progressive (SP) or relapsing remitting (RR) form that accumulates disability between relapses, with a documented worsening during the last year (1 step of Expanded Disability Status Scale [EDSS], or 0.5 when EDSS is between 5.5 and 6.5), despite conventional therapy (interferon-b or glatiramer acetate or immunosuppressive therapy), Presence of one or more gadolinium (Gd)-enhancing areas on MRI. The EDSS score between 3.5 and 6.5.
Exclusion criteria:	Not available, Trial protocol could not be downloaded
Recruitment/selection of patients:	Twenty-one patients with clinically defined MS, 23 a secondary progressive (SP) or relapsing remitting (RR) form that accumulates disability between relapses, with a documented worsening during the last year (1 step of Expanded Disability Status Scale [EDSS], or 0.5 when EDSS is between 5.5 and 6.5), in spite of conventional therapy (interferon-b or glatiramer acetate or immunosuppressive therapy), and presence of one or more gadolinium (Gd)-enhancing areas on MRI. The EDSS score had to be between 3.5 and 6.5
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Peripheral hematopoietic stem cells (PBSCs), PBSCs were mobilized by cyclophosphamide (4g/m^2) in 1 day. After 5 days, filgrastim was administered daily (5 mg/kg body weight SC) until the completion of the stem cells harvest. Hematopoietic stem cells were then collected with a leukapheresis procedure and an unmanipulated graft containing between 3 and 8 3 106 CD341/kg cells was cryopreserved. The conditioning regimen was BEAM, which includes BCNU (carmustine, 300 mg/m2 at day 26); cytosine-arabinoside (200 mg/m^2); etoposide (200 mg/m^2 from day 25 to day 22); and melphalan (140 mg/m^2 at day 21). PBSC were then thawed and infused through the central venous catheter. Rabbit ATG was added at a total dose of 3.75 mg/kg/d at day 11 and 12. Acyclovir and sulfamethoxazole/trimethoprim from discharge until day 1180 were recommended.

		Route: Intravenous
Outcomes reported with time points		The primary endpoint of ASTIMS was to determine the effect of AHSCT vs MTX on disease activity measured by the cumulative number of new T2 MRI lesions in the 4 years following randomization. Secondary endpoints were the activity of AHSCT vs MTX on the cumulative number of Gd-enhancing lesions on T1-weighted MRI from baseline to year 4. Secondary clinical endpoints also included the cumulative number of relapses from baseline to year 4 and the time to disability progression confirmed after 6 and 12 months. Progression was defined as an increase of 1 or more EDSS points when baseline EDSS is between 3.5 and 5.5 or of 0.5 EDSS points when baseline EDSS is between 5.5 and 6.5. Other secondary endpoints were AHSCT-related mortality, safety, side effects, early adverse events, and serious adverse events (SAEs) in the 2 arms. Additional analyses included comparisons between treatment arms in the cumulative number of new T2 lesions over the first, second, third, and fourth year after therapy, as well as in the time of appearance of the first new T2 MRI lesion.
Funding		Supported by the Italian Multiple Sclerosis Foundation, grants 2001-R-38 and 2002-R-36.
RoB2 Assessment: Specify separately for each domain		
RoB-2 Assessment: Outcome EDSS at follow up		
Domain	**Risk assessment**	**Support for judgment (including statements from the trial)**
D1 (Randomization process)	Some concern	Randomized consort diagram is available which indicate that randomization done however no information about concealment process although the study is multicentre no mentioned of allocation concealment, we judged no information of 1.2. Baseline EDSS was little higher in the treatment group 6.5 and 6 in control which indicate no issue with baseline differences.
D2 (deviation from intended intervention)	Low	No information of blinding of carers and people delivering intervention and participants however, due to nature trial setting blinding is not possible therefore we judged it as Probably yes Clinical data was available for the 20 patients out of 21 patients. One patient left after the randomization. Yes the appropriate analysis was used.
D3 (missing outcome data)	Low	EDSS outcome data is available for all the patient randomized
D4 (Measurement of outcome)	Low	EDSS is a well-established valid outcome measure, and in this study, it was measured by treating a neurologist or examining a physician; therefore, there is probably no risk of inappropriate outcome measurement.

D5 (selection of reported results	Low	Protocol could not be retrieved, although the EUDRCT number (2007-000-64-24) is provided in the article for registration but could not be downloaded. Since EDSS is commonly used to measure outcomes, we judged this domain with a probable yes for outcomes analyzed with a pre-specified plan. EDSS is a commonly used outcome, and no other similar outcome was assessed; therefore, we judged there was no risk for multiple eligible outcome assessments. The analysis was done properly; therefore, we judged there was no issue with multiple eligible analyses of the data.
Overall		Some concern
RoB-2 Assessment: Annualized relapse rate		
Domain	**Risk assessment**	**Support for judgment (including statements from the trial)**
D1 (Randomization process)	Some concern	Randomized consort diagram is available which indicate that randomization done however no information about concealment process although the study is multicentre no mentioned of allocation concealment, we judged no information of 1.2. Baseline EDSS was little higher in the treatment group 6.5 and 6 in control which indicate no issue with baseline differences.
D2 (deviation from intended intervention)	Low	No information of blinding of carers and people delivering intervention and participants however, due to nature trial setting blinding is not possible therefore we judged it as Probably yes Clinical data was available for the 20 patients out of 21 patients. One patient left after the randomization. Yes the appropriate analysis was used.
D3 (missing outcome data)	Low	ARR data is available for all the patient randomized
D4 (Measurement of outcome)	Low	Although protocol could not retrieved and ARR AS important outcome Examination was done by expert neurologist MRI was examined by Central operator blinded by treatment status
D5 (selection of reported results	Low	Protocol could not retrieved although EUDRCT number (2007-000-64-24) is provided in the article for registration but could not be downloaded. MRI outcome is an important outcome No concern Analysis was done properly
Overall		Some Concern

Study	**Burt et al. 2019[2]**
Study type:	RCT
Number of participants:	110
Countries and setting:	• Northwestern University (Chicago, Illinois)

	• Sheffield Teaching Hospitals NHS Foundation Trust & University of Sheffield (Sheffield, England) • University of Uppsala (Uppsala, Sweden) • University of São Paulo (Ribeirão Preto, Brazil) Setting: Hospital
Duration of study Follow up (post intervention):	103 remained in the trial, with 98 evaluated at 1 year and 23 evaluated yearly for 5 years (median follow-up, 2 years; mean, 2.8 years) Data extraction done at 1 year follow-up before cross-over
Method of assessment of disease condition:	relapsing-remitting MS according to McDonald criteria,11 age 18 to 55 years, 2 or more clinical relapses or 1 relapse and MRI gadolinium-enhancing lesion(s) at a separate time within the previous 12 months despite receiving treatment with DMT, and an Expanded Disability Status Scale (EDSS) score between 2.0 and 6.0
Subgroup analysis within study	None
Inclusion criteria:	• Relapsing-remitting MS according to McDonald criteria, • Age 18 to 55 years, 2 or more clinical relapses or 1 relapse and MRI gadolinium-enhancing lesion(s) at a separate time within the previous 12 months despite receiving treatment with DMT, • Expanded Disability Status Scale (EDSS) score between 2.0 and 6.0.
Exclusion criteria:	• primary or secondary progressive multiple sclerosis; hereditary neurologic diseases; pregnancy; pulmonary, cardiac, renal, or liver dysfunction; abnormal platelet or white blood cell counts; active infection; prior treatment with alemtuzumab or mitoxantrone; or use of natalizumab within the prior 6 months, fingolimod within 3 months, or, for teriflunomide (which undergoes extensive enterohepatic recycling), failure of oral cholestyramine to decrease teriflunomide to a plasma concentration of less than 0.02 μg/mL
Recruitment/selection of patients:	Conducted at 4 centers, Patients were enrolled between 2005 and 2016. Northwestern University (Chicago, Illinois) enrollment began in 2005, Sheffield Teaching Hospitals NHS Foundation Trust & University of Sheffield (Sheffield, England) enrollment began in 2014, University of Uppsala (Uppsala, Sweden) enrollment began in 2011, and University of São Paulo (Ribeirão Preto, Brazil) enrollment began in 2009.
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Non-myeloablative Hematopoietic Stem Cells Peripheral blood stem cells were collected 10 days after intravenous cyclophosphamide (2g/m2) and 5 to 10 μg/kg per day of subcutaneous filgrastim beginning 5 days after cyclophosphamide. The immune ablative regimen was intravenous cyclophosphamide, 50 mg/kg per day on days −5 to day −2 before stem cell infusion (day 0) and rabbit antithymocyte globulin, 0.5 mg/kg on day −5, 1.0 mg/kg on day −4, and 1.5 mg/kg on days −3, −2, and −1. Methylprednisolone (1000 mg) was infused 30 minutes prior to rabbit antithymocyte globulin infusion. Beginning on day 0, daily oral prednisone was dosed at 60 mg for 3 days, 40 mg for 2 days, 20 mg for 2 days, and 10 mg for 2 days.

		Blood products were irradiated, cytomegalovirus safe, and leukocyte depleted. Filgrastim (5-10 μg/kg per day) was started on day +4 and continued until engraftment. Hydration (125- 150 mL normal saline per hour), diuretics, and intravenous mesna were continued until 24 hours after the last dose of cyclophosphamide. A Foley catheter was placed in patients with greater than 60 mL of postvoid urinary residual. Intravenous cephalosporin was started on day 0. Intravenous vancomycin was added for a febrile episode. Methylprednisolone (250 mg) was infused for rabbit antithymocyte globulin–related fever. Oral acyclovir was started on admission and continued for 1 year. Oral fluconazole was started on day +2, and oral trimethoprim-sulfamethoxazole or monthly nebulized pentamidine was started after platelet engraftment and continued for 3 months. Cytomegalovirus viral load was monitored for 90 days and was treated pre-emptively by switching from acyclovir to oral valganciclovir (900 mg twice daily) until testing negative by quantitative polymerase chain reaction CD34+ cells/kg recipient weight will ensure engraftment Route of administration: Intravenous Dose: $CD34^{+}$ cells/kg recipient weight will ensure engraftment
Outcomes reported with time points		The primary end point was time to disease progression, defined as an increase (worsening) in EDSS score of at least 1 point on 2 evaluations 6 months apart after at least 1 year of treatment, and not due to a non-MS disease process. Secondary end points Survival; relapses; Neurologic Rating Scale (NRS) (range, 0-100 in 1-point increments from worst [0] to no [100] disability; minimal clinically important difference, 10) MRI T2-weighted lesion volume; Short Form 36 quality-of-life score (range, 1-100; higher scores indicate more favorable health state); Multiple Sclerosis Functional Composite (MSFC) score, which incorporates a timed 25-ft walk test; the 9-Hole Peg Test; and the Paced Auditory Serial Addition Test
Funding		The Danhakl family, the Cumming Foundation, the McNamara Purcell Foundation, Morgan Stanley, and the National Institute for Health Research Sheffield Clinical Research Facility
RoB2 Assessment: Specify separately for each domain		
Outcome EDSS at follow up (one year, second, third and fourth year)		
Domain	**Risk assessment**	**Support for judgment (including statements from the trial)**
D1 (Randomization process)	Low	Randomization by computer generated block sequence by central team, center team coordinates notified by email Baseline EDSS seems to be similar between both groups

D2 (deviation from intended intervention)	Low	Open label Study Due to trial setting blinding of patient and caregiver is not possible. Out of 55 patients in experimental group, three patients have not received intervention may to effect significantly Appropriate analysis was used.
D3 (missing outcome data)	Low	52 subjects out of 55 in the intervention group and 51 out of 55 in the control group were available for analysis. The lost to follow up was judged as not influential to outcome
D4 (Measurement of outcome)	Low	Neurologist and experts assessed the outcome Neurologist and experts assessed the outcome A neurologist masked to treatment group assessed the outcome.
D5 (selection of reported results	Low	Protocol was available NCT00273364. Protocol specified that EDSS was primary outcome. Multiple eligible outcome neurology rating scale (NRS) for disability was used as secondary endpoint however both reported analyzed results does not seems to have issue with multiple eligible analysis
Overall		Overall Low Risk of Bias
RoB-2 Assessment: Outcome Relapse free events		
Domain	**Risk assessment**	**Support for judgment (including statements from the trial)**
D1 (Randomization process)	Low	Randomization by computer generated block sequence by central team, center team coordinates notified by email Baseline EDSS seems to be similar between both groups
D2 (deviation from intended intervention)	Low	Open label Study Due to trial setting blinding of patient and caregiver is not possible. Out of 55 patients in experimental group, three patients did not receive intervention may to effect significantly Appropriate analysis was used.
D3 (missing outcome data)	Low	52 subjects out of 55 in the intervention group and 51 out of 55 in the control group were available for analysis. The lost to follow up was judged as not influential to outcome
D4 (Measurement of outcome)	Low	Neurologist and experts assessed the outcome Neurologist and experts assessed the outcome A neurologist masked to treatment group assessed the outcome.
D5 (selection of reported results	Low	Protocol was available NCT00273364. Protocol specified that EDSS was primary outcome Multiple eligible outcome neurology rating scale (NRS) for disability was used as secondary endpoint however both reported Analyzed results does not seems to have issue with multiple eligible analysis

viii. List of excluded studies

Studies	Reason for exclusion
Tremblay et al. 2022	Extension of earlier published study Ucceli et al. 2021
Berard et al. 2022	Outcome data considered in the study was not reported
Petrou et al. 2020	Relevant data was not reported

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4. MULTIPLE SCLEROSIS

2) MESENCHYMAL STEM CELL THERAPY

i. **Key question in PICO format**

ii. **Search Strategy**

iii. **PRISMA Flow Diagram**

iv. **Summary of included studies**

v. **Classification of studies based on level of stem cell manipulation**

vi. **Evidence to Decision Framework**

vii. **Data Extraction Sheet**

viii. **List of excluded studies**

For i. to iii., refer to the subsection Hematopoitic stem cell transplantation explained in previous section

iv. Summary of included studies

Study	Population	Intervention	Comparison	Outcomes
Nabavi et al. 2023[1]	Twenty-one patients with confirmed MS according to Mac Donald criteria 2010 (Wen et al., 2010) and disease duration of 2-10 years, Expanded Disability Status Scale (EDSS) equal to 3.0-6.5, and 18-55 years of age from both genders were enrolled.	Bone marrow-derived mesenchymal stem cells	Placebo	Adverse events Number of Relapse between both the groups EDSS Change Cognitive assessment by Rao test Number of GAD+ lesion Number of new T2 lesion
Li et al. 2014[2]	RRMS or SPMS	Human Umbilical Cord Derived Mesenchymal stem Cells	Anti-inflammatory treatment and immuno suppressive therapy	The EDSS was used to assess the status of neurological function. Scheduled clinical evaluations were performed before hUC-MSC treatment (baseline) and at 1, 2, 3, 6, 9 and 12 months after treatment.
Liufriu et al. 2014.[3]	RRMS	Bone-marrow-derived-MSCs/kg	Placebo	**The coprimary endpoints** 1. Safety of IV MSCs in RRMS patients and efficacy in terms of cumulative number of gadolinium-enhancing lesions (GEL) between groups of treatment during the first 6 months 2. The reduction in the mean number of GEL (MSCs vs placebo period) at the end of the study. **Secondary endpoints** Clinical outcomes (number of relapses, change in the EDSS and MSFC z- score), MRI-based measures (listed in the MRI protocol) and OCT measures between groups of treatment during the first 6 months and at the end of the study. Exploratory analysis included the immunological evaluation.

Lublin et al. 2014[4]	RRMS or SPMS as per the 2005 Mc Donald criteria	Human placenta-derived cells (PDA-001) a preparation of mesenchymal-like cells derived from full term human placenta	placebo, administered by intravenous infusion	**Primary end point** Ruling out the possibility of paradoxical worsening of MS disease activity. This was monitored using Cutter's rule (Z5 new gadolinium lesions on 2 consecutive scans) by brain magnetic resonance imaging on a monthly basis for six months and also the frequency of multiple sclerosis relapse. **Secondary end points** included changes in MS-related disability and quality of life
Ucelli et al. 2021[5]	Patients with different forms of active multiple sclerosis (relapsing remitting multiple sclerosis, primary progressive multiple sclerosis, and secondary progressive multiple sclerosis), not responding to at least 1 year of attempted therapy with one or more approved therapies	Bone marrow-derived mesenchymal stem cells	Placebo	**Coprimary outcomes were** The safety evaluation was based on the number and severity of adverse events that occurred within each treatment group. Activity was evaluated by the total number of GELs at week 24 (sum of number of GELs at weeks 4, 12, and 24) after MSCs treatment compared with placebo (suspension media). Secondary outcomes assessed The activity of the cell therapy by analyzing MRI activity defined as: number of GELs counted over week 28, 36, and 48 (crossover retreatment) compared with the number of GELs counted over 4, 12, and 24 weeks (placebo vs. active treatment periods) within each group; Clinical measures assessed by the following secondary endpoint: Number of relapses in the MSC treatment group vs placebo group in the first 24 weeks and after crossover re-treatment in the two groups; time to sustained progression of disability and proportion of progression-free patients compared between treatment groups during the first 24 weeks; proportion of disease-free patients defined as patients without relapses, with no evidence of sustained progression of disability and new MRI activity compared between treatment groups during the first 24 weeks changes in Multiple Sclerosis Functional Composite (MSFC) score An additional safety evaluation was ensured by gathering the

				number and severity of adverse events from the second infusion (crossover) until 48 weeks.
Fernande et al. 2018[6]	Thirty-four patients underwent lipectomy for AdMSCs collection, were randomized and thirty were infused (11 placebo, 10 low-dose and 9 high-dose).	Adipose-derived mesenchymal stem cells (AdMSCs)	placebo product consisted of Ringer's lactate packaged in identical opaque 50 mL-syringes	Safety was assessed primarily by monitoring for AEs and standard laboratory measures. Possible treatment effects were assessed by changes in baseline, 6 and 12 months after infusion: clinical variables (number of exacerbations, EDSS), immunologic assessments, Analysis of cerebrospinal fluid (CSF), MRI (T2-weighted lesions, T1 lesions, T1 gadolinium-enhanced lesions, volume, and magnetization transfer ratio-MTR), evoked potentials (EP) (visual-VEP, acoustic-BAEP, somatosensory-SEP and motor-MEP), optical coherence tomography (OCT) of the retina (retinal nerve fiber layer- RNFL), Cognition measured with Paced Auditory Serial Addition Test (PASAT) and quality-of-life by questionnaire SF-36, EuroqoL-5D and MusiQoL

v. Classification of studies on the basis of level of manipulation of the stem cell/ Stem cell derived product used (*interpreted by the Secretariat as defined by CDSCO* and the information provided in the trial itself*):

S. No.	Trial Details	Intervention	Level of manipulation	Information extracted from trial
1.	Nabavi et al. 2023	Bone marrow-derived mesenchymal stem cells- Intravenous infusion with dosage of 2×10^6 cells per kg body weight	More	*MSC isolation, expansion and characterization performed.*
2.	Li et al. 2014	Human Umbilical Cord Derived Mesenchymal Stem Cells- intravenous infusion of culture expanded hUC-MSCs at a dosage of 4×10^6 cells/kg once every 2 weeks for three consecutive times	More	*The cultured cells were harvested within two to three passages and assessed for lineage markers via flow cytometry.*

3.	Liufriu et al. 2014	Autologous Mesenchymal Stem Cells- Bone-marrow derived-culture expanded MSCs with dosage of 1–2 x 10^6 MSCs/Kg body weight	More	*A number between 20–60 millions of mononuclear cells were seeded per flask (175 cm^2) with growth medium… About 10–15 days later, colonies were formed.*
4.	Lublin et al. 2014	Human placenta-derived cells (PDA-001)- a preparation of mesenchymal-like cells derived from full term human placenta; 150 x10^6 cells & 600 x 10^6 cells	More	*Infusion of PDA-001, a preparation of mesenchymal-like cells derived from full-term human placenta*
5.	Ucelli et al. 2025	Bone marrow-derived mesenchymal stem cells- Intravenous infusion with dosage of 1–2×10^6 cells per kg bodyweight	More	*At the local cell factories, MSCs were isolated from the bone marrow and expanded up to a number of 1–2×10^6 cells per kg bodyweight and cryopreserved until use.*
6.	Fernande et al. 2018	Adipose-derived mesenchymal stem cells (AdMSCs)- Intravenous infusion with dosage of 1x10^6 cells/kg (Low dose) or 4x10^6 cells/kg (High dose)	More	*After 24–72 hours of incubation, the medium was changed and non-adherent cells were removed. For AdMSC cultures, a complete change of medium every 2–3 days until reaching 80% confluence was applied.*

* as defined by CDSCO in Annexure- I of the guideline document.

vi. Evidence to Decision Framework

QUESTION

What is the efficacy and safety of mesenchymal stem cell therapy as compared to usual care in patients with multiple sclerosis?	
POPULATION:	Patients with Multiple Sclerosis (MS)
INTERVENTION:	Stem cell therapy (MSC-Mesenchymal stem cell therapy)
COMPARISON:	Usual care
MAIN OUTCOMES:	EDSS at Six Months (EDSS 0-10, 0 good 10 worse)- EDSS at one year, Average or Annual relapse rate; Proportion free of relapse, Disease progression, Serious adverse events
SETTING:	Tertiary care centers
PERSPECTIVE:	Health system/Population
BACKGROUND:	Multiple sclerosis (MS) is one of the leading causes of neurological disability in young adults with symptom onset

	generally occurring between the ages of 20 to 40 years. It is an autoimmune inflammatory disorder characterized by demyelination of neurons in the central nervous system and affects women more commonly than men. Initially the episodes are reversible, that are followed by progressive neurological deterioration over time.
CONFLICT OF INTERESTS:	None

ASSESSMENT

Problem

Is the problem a priority?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
○ No ○ Probably no ○ Probably yes ● Yes ○ Varies ○ Don't know	The prevalence of MS in our country has increased from 1.33/100,000 in 1985 to 8.35/100,000 in 2014.[7] There is no cure for MS and the current disease modifying therapies provide mainly symptomatic relief or shorten the duration of symptoms.	

Desirable Effects

How substantial are the desirable anticipated effects?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
○ Trivial ● Small ○ Moderate ○ Large ○ Varies	**Mesenchymal stem cell:** **EDSS score:** Six trials with 237 participants reported the EDSS score at 6 months. There appears to be no improvement in EDSS score at 6 months in the MSC therapy group as compared to the usual care group. The mean difference reported was -0.05 (95% CI: -0.37 to 0.28), which was not statistically significant. Two trials with 52 participants reported the EDSS score at 12	A change of EDSS score by 0.5 was considered as MCID. A difference of 0.6 for Annualized relapse rate (ARR) was considered as MCID.

○Don't know	months. The mean difference reported was -0.55 (95% CI: -2.38 to 1.27), which was statistically non-significant. **Annual relapse rate:** Two trials with 162 participants reported annual relapse rate. The mean difference for annual relapse rate was -0.85 (95% CI: -1.44 to -0.26) in the MSC therapy arm as compared to the usual care. There seems to be a small clinically important reduction in average or annual relapse rate in the MSC therapy arm, which was crossing the MCID of 0.6. **Proportion free from relapse:** Three trials with 53 participants reported the proportion free from relapse. Pooled analysis yielded a risk ratio of 1.76 (95% CI: 0.44 to 7.06) in the stem cell arm as compared to usual care, which was statistically non-significant.	A difference of 20/100 (20%) was considered as MCID for Proportion free of relapse.
Undesirable Effects How substantial are the undesirable anticipated effects?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
●Trivial ○Small ○Moderate ○Large ○Varies ○Don't know	**Mesenchymal stem cell:** **Serious adverse events:** Three studies with a total of 54 participants reported serious adverse events. Pooled analysis revealed a risk ratio of 0.51 (95% CI: 0.15 to 1.78) in the mesenchymal stem cell arm as compared to usual care, which was statistically non-significant. The type of SAEs reported by Lublin et al[4] in the above analysis were choking, respiratory infection, urinary infection, Grade I anaphylactoid reaction and Grade 2 superficial thrombophelebitis.	
Certainty of evidence What is the overall certainty of the evidence of effects?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL**

		CONSIDERATIONS
● Very low ○ Low ○ Moderate ○ High ○ No included studies	The certainty of evidence is very low.	
Values Is there important uncertainty about or variability in how much people value the main outcomes?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○ Important uncertainty or variability ○ Possibly important uncertainty or variability ● Probably no important uncertainty or variability ○ No important uncertainty or variability	A nationwide survey in UK in patients with relapsing remitting multiple sclerosis (RRMS) reported that most respondents (73%) had personal goals related to lifestyle and many (69%) were concerned about maintaining independence.[8] In RRMS Patient preference focused on symptoms and prevention of progression but not on relapse prevention.[9] Pain contributed the most to multiple sclerosis outpatients' perception of health, followed by gait dysfunction and fatigue.[10]	The GDG also opined that an improvement in disability is likely to be highly valued by the patients.
Balance of effects Does the balance between desirable and undesirable effects favor the intervention or the comparison?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**

○ Favors the comparison ○ Probably favors the comparison ● Does not favor either the intervention or the comparison ○ Probably favors the intervention ○ Favors the intervention ○ Varies ○ Don't know	It is not clear whether benefits outweigh the harms or not in the mesenchymal stem cell group for Multiple Sclerosis.	
Resources required How large are the resource requirements (costs)?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
● Large costs ○ Moderate costs ○ Negligible costs and savings ○ Moderate savings ○ Large savings ○ Varies ○ Don't know	No research evidence was identified for resource use for mesenchymal stem cell therapy in Multiple sclerosis.	The cost in the government hospitals may be slightly lower
Certainty of evidence of required resources What is the certainty of the evidence of resource requirements (costs)?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**

○Very low ○Low ●Moderate ○High ○No included studies	No research evidence was identified.	The GDG members were fairly certain that stem cell therapy requires large resources.
Cost effectiveness Does the cost-effectiveness of the intervention favor the intervention or the comparison?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○Favors the comparison ●Probably favors the comparison ○ Does not favor either the intervention or the comparison ○Probably favors the intervention ○Favors the intervention ○Varies ○No included studies	No studies were identified for mesenchymal stem cell therapy for Multiple Sclerosis.	
Equity What would be the impact on health equity?		
JUDGEMENT	**RESEARCH EVIDENCE**	**ADDITIONAL CONSIDERATIONS**
○Reduced ●Probably reduced ○Probably no impact	No research evidence available.	Probably reduced as the treatment is costly and is available in tertiary care

○Probably increased ○Increased ○Varies ○Don't know		centres/ big private hospitals in major cities only.

Acceptability

Is the intervention acceptable to key stakeholders?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
○No ○Probably no ●Probably yes ○Yes ○Varies ○Don't know	No research evidence was identified on the acceptability on the use of stem cell therapy in multiple sclerosis.	Despite uncertainty regarding the medical risks and benefits associated with stem cell therapy, patients may still consider undergoing treatment in private, unregulated clinics

Feasibility

Is the intervention feasible to implement?

JUDGEMENT	RESEARCH EVIDENCE	ADDITIONAL CONSIDERATIONS
○No ○Probably no ●Probably yes ○Yes ○Varies ○Don't know	No research evidence was identified on the feasibility on the use of stem cell therapy in multiple sclerosis.	Feasible to implement in tertiary care centres.

SUMMARY OF JUDGEMENTS MSCT

	JUDGEMENT						
PROBLEM	No	Probably no	Probably yes	**Yes**		Varies	Don't know
DESIRABLE EFFECTS	Trivial	**Small**	Moderate	Large		Varies	Don't know
UNDESIRABLE EFFECTS	**Trivial**	Small		Large		Varies	Don't know
CERTAINTY OF EVIDENCE	**Very low**	Low	Moderate	High			No included studies
VALUES	Important uncertainty or variability	Possibly important uncertainty or variability	**Probably no important uncertainty or variability**	No important uncertainty or variability			
BALANCE OF EFFECTS	Favors the comparison	Probably favors the comparison	**Does not favor either the intervention or the comparison**	Probably favors the intervention	Favors the intervention	Varies	Don't know
RESOURCES REQUIRED	**Large costs**	Moderate costs	Negligible costs and savings	Moderate savings	Large savings	Varies	Don't know
CERTAINTY OF EVIDENCE OF REQUIRED RESOURCES	Very low	Low	**Moderate**	High			No included studies
COST EFFECTIVENESS	Favors the comparison	**Probably favors the comparison**	Does not favor either the intervention or the comparison	Probably favors the intervention	Favors the intervention	Varies	No included studies

	JUDGEMENT						
EQUITY	Reduced	**Probably reduced**	Probably no impact	Probably increased	Increased	Varies	Don't know
ACCEPTABILITY	No	Probably no	**Probably yes**	Yes		Varies	Don't know
FEASIBILITY	No	Probably no	**Probably yes**	Yes		Varies	Don't know

TYPE OF RECOMMENDATION

Strong recommendation against the intervention	Conditional recommendation against the intervention	**Use in the context of rigorously conducted RCTs**	Conditional recommendation for either the intervention or the comparison	Conditional recommendation for the intervention	Strong recommendation for the intervention
○	○	●	○	○	○

CONCLUSIONS

Recommendation

Mesenchymal Stem Cell Therapy is **not recommended** in routine clinical practice for the treatment of multiple sclerosis. It may be used only in the context of rigorously conducted randomized controlled trials.

Justification

Overall justification

This recommendation has been made as there is very low certainty evidence of small benefit in terms of disability and relapse rate. There seems to be statistically non-significant change in EDSS score at 6 months and at one year. There seems to be a small improvement in annual relapse rate (just crossing the MCID of 0.6), which is important clinically. There is little to no difference in undesirable effects between stem cell therapy and usual care. In addition, the follow up period of one year is too small to comment on the side effect profile and long-term safety is not known.

Detailed justification
Desirable Effects

EDSS score
Six trials with 237 participants reported the EDSS score at 6 months. There appears to be no improvement in EDSS score at 6 months in the MSC therapy group as compared to the usual care group. The mean difference reported was -0.05 (95% CI: -0.37 to 0.28), which was not statistically significant. Two trials with 52 participants reported the EDSS score at 12 months. The mean difference reported was -0.55(95% CI: -2.38 to 1.27), which was not significant statistically.

Average or annual relapse rate
Two trials with 162 participants reported average or annual relapse rate. The mean difference for annual relapse rate was -0.85(95% CI: -1.44 to -0.26) in the MSC therapy arm as compared to the usual care. There seems to be a small clinically important reduction in average or annual relapse rate in the MSC therapy arm, which was crossing the MCID of 0.6.

Proportion free from relapse

Three trials with 53 participants reported the proportion free from relapse. Pooled analysis yielded a risk ratio of 1.76 (95% CI: 0.44 to 7.06) in the stem cell arm as compared to usual care, which was statistically non-significant.

Undesirable Effects

Three studies with a total of 54 participants reported serious adverse events. Pooled analysis revealed a risk ratio of 0.51 (95% CI: 0.15 to 1.78) in the mesenchymal stem cell arm as compared to usual care, which was statistically non-significant. The type of SAEs reported by Lublin et al[41] in the above analysis were choking, respiratory infection, urinary infection, Grade I anaphylactoid reaction and Grade 2 superficial thrombophelebitis.

Certainty of evidence
Very low certainty of Evidence

Resources required
The committee opined that stem cell treatment is associated with large costs.

vii. Data Extraction:

Study	**Nabavi et al. 2023[1]**
Study type:	RCT (crossover)
Number of participants:	21
Countries and setting:	Iran Setting: Hospital
Duration of study Follow up (post intervention):	18 months
Method of assessment of disease condition:	Active disease was defined as follows, as detailed elsewhere: confirmed MS according to Mac Donald criteria 2010 (Wen et al. 2010) and disease duration of 2-10 years; diagnosis of relapsing-remitting MS (RRMS) which had not responded to one or two DMDs (i.e., at least one severe relapse on first-line or second-line DMDs recently). Primary progressive MS (PPMS) patients with oligodonal bands (OCB) positivity in the CSF or GAD* enhancement on MRI accompanied by secondary progressive MS (SPMS) with or without relapse but one point increasing in EDSS within the last 18 months
Subgroup analysis within study	No sub-group analysis was conducted
Inclusion criteria:	• Confirmed MS according to Mac Donald criteria 2010 • Disease duration of 2-10 years • Expanded Disability Status Scale equal to 3 to 6.5, and 18-55 years of age from both genders • approved diagnosis of RRMS which had not responded to one to two DMDs (i.e. at least one severe relapse on first-line or second line DMDs recently) • Primary progressive MS (PPMS), patients with oligoclonal bands positivity in CSF or GAD enhancement on MRI accompanied by secondary progressive MS (SPMS) with or without relapse
Exclusion criteria:	• Treatment with cytotoxins within the recent three months before trial treatment with immunomodulators within one month before the trial, treatment with immunomodulators within one month before the trial, having relapses within 30 days prior to the beginning of the trial, and primary progressive MS (PPMS) without OCB in the CSF or GAD+ enhancement on MRI • Pregnant women
Recruitment/selection of patients:	Twenty-one patients with confirmed MS according to Mac Donald criteria 2010 (Wen et al., 2010) and disease duration of 2-10 years, Expanded Disability Status Scale (EDSS) equal to 3.0-6.5, and 18-55years of age from both genders were enrolled. They had approved diagnosis of relapsing-remitting MS (RRMS) which had not responded to one or two DMDs (i.e., at least one severe relapse on first-line or second-line DMDs recently).
Intervention: Type	fresh autologous hBM-MSCs,

of stem cells with method of their characterization, Route of administration, Dose	The characterization panel consisted of monoclonal antibodies for mesenchymal lineages markers CD90-FITC (BD, PharmingenTM, USA), CD105- PE (Endoglin, BD PharmingenTM, USA), CD73-PE (BD PharmingenTM, USA), CD44-FITC (BD PharmingenTM, USA), CD45 FITC-CD34 PE (BD PharmingenTM, USA) and CD11b (BD PharmingenTM, USA Route: Intravenous Dose: 2×10^6 cells per kg body weight	
Outcomes reported with time points	All patients were clinically evaluated for any adverse events, number of relapses, disability status measured by EDSS (Freedman et al. 2010), and the cognitive assessment by Rao's brief repeatable battery test (Cognitive screening tools in multiple sclerosis revisited: sensitivity and specificity of a short version of Rao's Brief Repeatable Battery). The clinical follow-up assessments were done at 24 h, 1, 3, 6, 12, and 18 months after baseline. Rao cognitive tests were done at 6, 12, and 18 months. Laboratory assessments including blood tests and serologic profiles were performed at 24 h, 1, 6, 12, and 18 months after transplantation. The overall follow-up durations were 18 months for both groups.	
Funding	Royan institute by the grant number of 90210900.	
RoB2 Assessment: Specify separately for each outcome		
EDSS Score		
Domain	**Risk assessment**	**Support for judgment (including statements from the trial)**
D1 (Randomization process)	Some concern	Protocol mentioned randomized allocation; however, sequence generation was not available, and placebo-containing cell-free media suggest no issue with allocation concealment. There was no significant baseline difference in the EDSS outcome; however, the age difference was significant-32 years in the treatment group and 39 in the control group. The proportion of RRMS was approximately 50% higher in the treatment group compared to the control group.
D2 (deviation from intended intervention)	Low	Participants and clinicians were blinded in a placebo-controlled trial. Since no loss to follow up therefore, we judged it as low risk of bias

D3 (missing outcome data)	Low	Outcome available in 12 patients in MSC group and 9 patients in placebo group, therefore we judged yes for outcome data available for nearly all participants
D4 (Measurement of outcome)	Some Concern	No information about that who assessed the EDSS outcome No information was provided that who assessed the outcome No information is available for outcome assessor blinded Since no difference in the outcome EDSS was observed between two arms in the follow up period, we judged probably no concern for outcome have been influenced by knowledge of intervention.
D5 (selection of reported results	Low	Protocol mention EDSS outcome measure pre specified No similar outcome was assessed therefore we judged no concern for the multiple eligible outcome No multiple analysis was performed for this outcome
Overall		Overall some concern for risk of bias
Proportion free from Relapse		
Domain	**Risk assessment**	**Support for judgment (including statements from the trial)**
D1 (Randomization process)	Some concern	Protocol mentioned randomized allocation; however, sequence generation was not available, and placebo-containing cell-free media suggest no issue with allocation concealment. There was no significant baseline difference in the EDSS outcome; however, the age difference was significant—32 years in the treatment group and 39 in the control group. The proportion of RRMS was approximately 50% higher in the treatment group compared to the control group.
D2 (deviation from intended intervention)	Low	Participants and clinicians were blinded in a placebo-controlled trial. Since no loss to follow up therefore, we judged it as low risk of bias
D3 (missing outcome data)	Low	Outcome available in 12 patients in MSC group and 9 patients in placebo group, therefore we judged yes for outcome data available for nearly all participants

D4 (Measurement of outcome)	Some Concern	No information that who assessed the outcome No information was provided that who assessed the outcome No information is available for outcome assessor blinding Since no difference in the outcome relapse free events was observed between two arms in the follow up period
D5 (selection of reported results	Low	No information for relapse event outcome measure pre specified in the Protocol No similar outcome was assessed No multiple analysis was performed for this outcome
Overall		Overall, some concern for Risk of Bias.

Study	**Li et al. 2014[2]**
Study type:	RCT
Number of participants:	**23**
Countries and settings:	**China** **Setting: Hospital**
Duration of study Follow up (post intervention):	**1,2,3,6,9 and12 month after treatment**
Method of assessment of disease condition:	The EDSS was used to assess the status of neurological function. The scores range from o to 10, representing normal to death, respectively. Patient scores ranged from 5 to 8 at enrollment, indicating impairment of daily activities and degrees of disability ranging from inability to walk longer than 200m to restriction to a wheelchair. Scheduled clinical evaluations were performed before hUC-MSC treatment (baseline) and at 1, 2, 3, 6, 9 and 12 months after treatment. Two neurologist who had no knowledge of the study evaluated every case independently to minimize bias**.**
Subgroup analysis within study	
Inclusion criteria	• RRMS or SPMS with an average Expanded Disability Status Scale (EDSS) of 5.0 (range 4.0–8.0) at the time of enrollment and clinical deterioration at 1.0 point or higher on EDSS over the past 12 months; • Males or nonpregnant females with an age range from 25 to 55; • Had two or more attacks in the last 2 years with a disease course of more than 2 years; • Characteristic abnormalities on magnetic resonance imaging (MRI) scan.
Exclusion criteria	Patients were excluded from the study if they • were treated with cytotoxic medications (i.e., cyclophosphamide, mitoxantrone, and azathioprine) in the past 3

	months before the trial; • had significant cardiac, renal, or hepatic failure or any other severe diseases that may interfere with result interpretation; • had an active infection; • showed severe cognitive decline; and • unable to understand and sign the informed consent. The patients did not pay for their inclusion or treatment in this study.
Recruitment/selection of patients	All patients were enrolled into the study from January 2010 to December 2012. The experimental group included 13 patients (four males and nine females), with an average age of 41.7±5.6 years, and had either RRMS or SPMS with mean disease duration of 2.90±0.9 years and EDSS score of 6.98±1.2 (range 4.0–8.0). The control group included 10 patients (three males and seven females), with average age of 39.4±3.8 years. The control patients had either RRMS or SPMS with mean disease duration of 2.57±0.8 years and EDSS score of 6.02±1.6 (range 4.0–8.0).
Intervention	hUC-MSCs were isolated from human UCs and cultured. The cultured cells were harvested within two to three passages and assessed for lineage markers via flow cytometry. Fixed cells were washed twice with PBS and incubated for 30 min with fluorescein isothiocyanate (FITC) or phycoerythrin (PE)- conjugated antibodies against cluster of differentiation 44 (CD44), CD29, CD105, CD31, CD45, and human leukocyte antigen (HLA)-DR (Abcam, Cambridge, UK; antibody concentration in accordance with the instructions) before being analyzed by flow cytometry (BD, Franklin Lakes, NJ, USA). Baseline data were collected from each patient prior to any treatment, including serological samples. At acute relapsing phase, all patients were given anti-inflammatory and immunomodulation reagent methylprednisolone (Jin-yao Pharmaceutical Co., Ltd, Tianjin, China) intravenously 1,000 mg/kg daily for 3 days, 500 mg/kg for 2 days, followed by oral prednisone (Xian Ju Pharmaceutical Co., Ltd, Zhejiang, China) 1 mg/kg/day for 10 days. Subsequent dosage of prednisone was reduced by 5 mg every 2 weeks until reaching a 5-mg/day maintenance dosage. Serum g-globulin (KangBao Biological Products Co., Ltd, Shanxi, China) was simultaneously supplemented at 400 mg/kg/ day for 5 days. Thirteen patients were randomly selected for hUC-MSC therapy (intravenous infusion, hUC-MSCs suspended in 100 ml normal saline) at a dosage of 4×10^6 cells/kg once every 2 weeks for three consecutive times. The control group did not receive a saline infusion. All patients continued the maintenance dosage of anti-inflammatory and immunosuppressive therapy during the follow-up stage.
Outcomes reported with time point	• The EDSS was used to assess the status of neurological function. • Scheduled clinical evaluations were performed before hUC-MSC treatment (baseline) and at 1, 2, 3, 6, 9, and 12 months after treatment.
Funding	This project was supported, in part, by the Chinese National Science Foundation (No. 30500208) and IAEA Research Project (No. CPR-13305). The authors declare no conflicts of interest.
RoB2 Assessment:	

specify separately for each domain		
EDSS		
Domain	**Risk assessment**	**Support for judgment (including statements from the trial)**
D1 (Randomization process)	Some concern	The patient was randomly selected; the process of concealment of the random sequence is not mentioned. The protocol could not be retrieved. Baseline mean EDSS scores were 6.98 and 6.02 in the treatment and control groups, respectively.
D2 (deviation from intended intervention)	Some concern	No mentioned of blinding of patients and caregiver. control group did not receive the saline infusion indicate that study was not double blinded Authors have no mention that deviation raised due to trial context. All the patients data was used for analysis
D3 (missing outcome data)	Low	There was no loss in follow-up; therefore, we judged the outcome available for nearly all participants.
D4 (Measurement of outcome)	Low	EDSS was measured by a neurologist. EDSS was assessed in similar ways. Two neurologists who had no knowledge of the study evaluated every case independently to minimize bias.
D5 (selection of reported results	Some concern	The protocol could not be retrieved. Non-such concern for multiple outcome assessment There is no such concern for multiple eligible analyses.
Overall		Overall, we judged, High Risk of Bias
Annual Relapse Rate		
Domain	**Risk assessment**	**Support for judgment (including statements from the trial)**
D1 (Randomization process)	Some concern	The patient was randomly selected; the process of concealment of the random sequence is not mentioned. The protocol could not be retrieved. Baseline mean EDSS scores were 6.98 and 6.02 in the treatment and control groups, respectively.

D2 (deviation from intended intervention)	Some concern	No mentioned of blinding of patients and caregiver. control group did not receive the saline infusion indicate that study was not double blinded Authors have no mention that deviation raised due to trial context. All the patient's data was used for analysis
D3 (missing outcome data)	Low	There was no loss in follow-up; therefore, we judged the outcome available for nearly all participants.
D4 (Measurement of outcome)	Low	Assessed by neurologist No such concern about differed ascertainment method between both groups the outcome was assessed by two neurologists who had no knowledge of the study evaluated every case independently to minimize bias
D5 (selection of reported results	Some concern	Protocol could not be retrieved Non such concern No such concern
Overall		Overall, we judged, High Risk of Bias
Proportion free of relapse		
Domain	**Risk assessment**	**Support for judgment (including statements from the trial)**
D1 (Randomization process)	Some concern	The patient was randomly selected; the process of concealment of the random sequence is not mentioned. The protocol could not be retrieved. Baseline mean EDSS scores were 6.98 and 6.02 in the treatment and control groups, respectively.
D2 (deviation from intended intervention)	Some concern	No mentioned of blinding of patients and caregiver. control group did not receive the saline infusion indicate that study was not double blinded Authors have no mention that deviation raised due to trial context. All the patient's data was used for analysis
D3 (missing outcome data)	Low	There was no loss in follow-up; therefore, we judged the outcome available for nearly all participants.

D4 (Measurement of outcome)	Low	Assessed by neurologist No such concern about differed ascertainment method between both groups the outcome was assessed by two neurologists who had no knowledge of the study evaluated every case independently to minimize bias
D5 (selection of reported results	Some concern	Protocol could not be retrieved Non such concern No such concern
Overall		Overall, we judged, High Risk of Bias

Study	**Liufriu et al. 2014[3]**
Study type	RCT
Number of participants	9
Countries and settings	Spain Setting: Hospital
Duration of study Follow up (post intervention)	6 months
Method of assessment of disease condition	Clinical assessments were performed at screening, and at randomization (baseline), and study visits, including safety assessments, were scheduled at 1, 3, 6, 7, 9 and 12 months after randomization. Relapses were defined by one of the participant neurologists as the development of new neurologic symptoms and confirmed signs at least 30 days after onset of last relapse. In case of corticosteroids treatment the MRI was delayed 1 month. The EDSS and the Multiple Sclerosis Scale Functional Composite (MSFC) z- score [10] were evaluated every 3 months
Subgroup analysis within study	None
Inclusion criteria	• Relapsing-remitting MS not responding to at least a year of approved therapy, defined by at least 1 clinically documented relapse and/or at least 1 gadolinium-enhancing lesion (GEL) on MRI within the last 12 months, • Age 18 to 50 years • disease duration of 2 to 10 years • Expanded Disability Status Scale (EDSS) score between 3.0 to 6.5.
Exclusion criteria	If patients had • Any active or chronic infection, • Treatment with any immunosuppressive therapy within the previous 3 months or interferon-beta,

	• Glatiramer acetate or corticosteroids within 30 days prior to randomization.
Recruitment/selection of patients	Eligible participants were those with relapsing-remitting MS not responding to at least a year of approved therapy, defined by at least 1 clinically documented relapse and/or at least 1 gadolinium-enhancing lesion (GEL) on MRI within the last 12 months, aged 18 to 50 years, disease duration of 2 to 10 years and Expanded Disability Status Scale (EDSS) [9] score between 3.0 to 6.5.
Intervention	Briefly, the mononuclear cell fraction was isolated by Ficoll density gradient centrifugation (Ficoll-Paque, GE Healthcare BioSciences, AB). A number between 20–60 millions of mononuclear cells were seeded per flask (175 cm^2) with growth medium. The flasks were maintained in culture at 37°C/5% CO^2.the cells were analyzed by flow cytometry to confirm expression of CD90, CD73 y CD44 and absence of CD34 and CD45 surface markers. The cells were administrated in the first 24 hours postproduction (baseline) or cryopreserved for reversed administration at 6 months. A dose of 1–26106 MSCs/Kg body weight or suspension media were slowly infused over 2–4 min through a peripheral venous cannula at baseline. At 6 months, treatment was reversed compared to baseline and all patients received premedication with 2 mg dexchlorpheniramine, 1 g paracetamol, and 100 mg methylprednisolone to prevent infusion reactions.
Outcomes reported with time points	**The coprimary endpoints** 1. Safety of IV MSCs in RRMS patients and efficacy in terms of cumulative number of gadolinium-enhancing lesions (GEL) between groups of treatment during the first 6 months 2. The reduction in the mean number of GEL (MSCs vs placebo period) at the end of the study. **Secondary endpoints** Clinical outcomes (number of relapses, change in the EDSS and MSFC z- score), MRI-based measures (listed in the MRI protocol) and OCT measures between groups of treatment during the first 6 months and at the end of the study. Exploratory analysis included the immunological evaluation.
Funding	**None**
RoB2 Assessment: Specify separately for each domain	

EDSS Score

Domain	Risk assessment	Support for judgment (including statements from the trial)
D1 (Randomization process)	Low	Randomized by computer generated assignment list. Placebo controlled trial in which control group suspension media, therefore, we judged no concern for allocation sequence random and allocation sequence concealed. No significant difference between both group regarding demographics and imaging parameters

D2 (deviation from intended intervention)	Low	All patients and study personal, except for the hematologist, therefore, we judged no concern with devotion from intended intervention. Analysis was performed based on the intention to treat with last observation carried forward (LOCF) to impute missing values.
D3 (missing outcome data)	Low	No significant loss to follow up
D4 (Measurement of outcome)	Low	No information was available who assessed the outcome Similar way outcome assessed in the both the groups No information for the blinding of outcome assessor in the protocol and published study As the EDSS difference was not significant between the groups we assessed it as No influence of knowledge of intervention.
D5 (selection of reported results	Low	EDSS was pre-specified No concern No concern
Overall		We judged low concern for overall as low risk in all domain
Proportion free of relapse		
Domain	Risk assessment	Support for judgment (including statements from the trial)
D1 (Randomization process)	Low	Randomized by a computer-generated assignment list. Placebo-controlled trial in which control group suspension media There is no significant difference between the two groups regarding demographics and imaging parameters.
D2 (deviation from intended intervention)	Low	All patients and study personal, except for the haematologist An analysis was performed based on the intention to treat the last observation carried forward (LOCF) to impute missing values.
D3 (missing outcome data)	Low	No significant loss to follow up

D4 (Measurement of outcome)	Low	No information who assessed the outcome No such concern for ascertaiment of outcome differed between groups No information about outcome assessor were blinded It may differ depending on intervention knowledge. Since outcomes did not differ in both groups, we judged this domain as probable for the assessment of outcome influence.
D5 (selection of reported results	Low	Relapse event was pre-specified No concern No concern
Overall		Low Risk for Bias

Study	**Lublin et al. 2014[4]**
Study type	RCT
Number of participants	16
Countries and setting	USA and Canada Setting: Hospital
Duration of study Follow up (post intervention)	12 months
Method of assessment of disease condition	**The primary end point** was ruling out the possibility of paradoxical worsening of MS disease activity. This was monitored using Cutter's rule (Z5 new gadolinium lesions on 2 consecutive scans) by brain magnetic resonance imaging on a monthly basis for six months and also the frequency of multiple sclerosis relapse. **Secondary end points** included changes in MS-related disability and quality of life
Subgroup analysis within study	**Not done**
Inclusion criteria	• Patients between 18 and 65 years of age with a pre-existing diagnosis of RRMS or SPMS per the 2005 McDonald Criteria (Polman et al., 2005) • Disease duration of at least 2 years • Patients had to have evidence of active disease as evidenced by clinical progression, continued relapses, or worsening magnetic resonance imaging (MRI) scans after at least 1 year of attempted therapy.
Exclusion criteria	If patients were treated with systemic immunosuppressive medications (eg, infliximab, cyclophosphamide, mitoxantrone, azathioprine, methotrexate, linomide, cyclosporine, cladribine, deoxyspergualin) or

	natalizumab within 6 months before screening. Adequate cardiac, pulmonary, liver, and renal function was required.
Recruitment/selection of patients	Patients between 18 and 65 years of age with a pre-existing diagnosis of RRMS or SPMS per the 2005 McDonald Criteria (Polman et al., 2005) and disease duration of at least 2 years were eligible to participate in the study. Patients had to have evidence of active disease as evidenced by clinical progression, continued relapses, or worsening magnetic resonance imaging (MRI) scans after at least 1 year of attempted therapy. The MRI worsening is evidenced by an increase of at least 1 point in Expanded Disability Status Scale (EDSS) score (if screening EDSS was r5.0) or 0.5 EDSS score (if baseline EDSS was≥ 5.5).
Intervention	PDA-001 and placebo units were supplied in a cryopreserved state. Immediately before administration, units were thawed, reconstituted with sufficient diluent (infusion grade dextran 40) for a total of 240 mL per infusion, and transferred to an infusion bag through the port using sterile technique. Each PDA-001 unit contained approximately 150 $x10^6$ cells. Placebo contained all of the excipients and at the same concentrations as the PDA-001 product, but did not contain the PDA-001 cells.
Outcomes reported with time points	**Primary end points** Ruling out the possibility of paradoxical worsening of MS disease activity. This was monitored using Cutter's rule (Z5 new gadolinium lesions on 2 consecutive scans) by brain magnetic resonance imaging on a monthly basis for six months and also the frequency of multiple sclerosis relapse. **Secondary end points** included changes in MS-related disability and quality of life
Funding	This study was supported by Celgene Cellular Therapeutics, Warren, NJ. The authors received editorial support in the preparation of this report from Christine H. Blood, PhD, of Peloton Advantage, funded by Celgene Cellular Therapeutics. The authors, however, directed and are fully responsible for all content and editorial decisions for this report. The sponsor was jointly responsible for the study design, along with the other authors. It was also responsible for the study execution, and the data collection and analysis. It was jointly responsible for the writing of the report and the decision to publish this manuscript along with the other authors.
RoB2 Assessment: Specify separately for each domain	

EDSS Score		
Domain	Risk assessment	Support for judgment (including statements from the trial)

D1 (Randomization process)	Low	Randomization was done by independent statistical who was masked to treatment allocation and trial is double blind placebo-controlled study Baseline EDSS 5.0 in low dose 6 in high dose and 4.0 in the placebo group was observed. Since the sample size was small (16), these differences may be possible, therefore we judged probably no risk for baseline difference in the randomization process.
D2 (deviation from intended intervention)	Low	Placebo controlled all patients received bags containing drugs identical in appearance. All were masked except pharmacist, Therefore, we judged no risk for deviation from intended intervention EDSS data were extracted from the graph and numerical values not reported in the article.
D3 (missing outcome data)	Low	16 patients enrolled and all the patients completed the follow up and safety profile.
D4 (Measurement of outcome)	Low	No information who assessed the EDSS score It is not mentioned about the blinding of outcome assessor however due to No difference in the EDSS score at follow, we judged the study with probably no for this domain. No information for blinding for outcome assessor, therefore we judged no information for outcome assessors aware of the intervention. no difference in the EDSS score at follow up,
D5 (selection of reported results	Low	The study protocol could not be retrieved; however, the trial was conducted under the investigational new drug license (US-FDA and clinical trial application for Health Canada. EDSS is a commonly used outcome for this disease. No multiple outcome assessment concern No multiple analysis was done; therefore, we judged there was no concern.
Overall		Overall Low Risk of Bias

Study	**Ucelli et al. 2021[5]**
Study type:	RCT
Number of participants:	144
Countries and setting:	Austria, Canada, Denmark, France, Italy, Iran, Spain, Sweden, UK Setting: Hospital
Duration of study Follow up (post	12 months

intervention):	
Method of assessment of disease condition:	Active disease was defined as follows, as detailed elsewhere:17 for patients with relapsing remitting multiple sclerosis, by the occurrence of a recent relapse or relapses with or without MRI activity on a scan done within the past 12 months; for secondary progressive multiple sclerosis, by progression of disease in the previous year, in presence of relapses or MRI activity; and for primary progressive multiple sclerosis, by disease progression together with evidence of MRI activity in the year before transplantation.
Subgroup analysis within study	
Inclusion criteria:	• To be eligible for the study, adult patients of either sex had to have a diagnosis of SPMS, with an Expanded Disability Status Score (EDSS) between 5.5 and 9 and have failed previous therapies and have activity or progression of the disease (relapse in the previous year or progression of at least 0.5 points on the EDSS despite therapy).
Exclusion criteria:	• Patients were excluded if they had experienced a relapse or had received steroid treatment in the month prior to inclusion. Patients positive for HIV, hepatitis B, or hepatitis C, history of malignant neoplasms, participation in an interventional trial in the previous 3 months, contraindications for magnetic resonance imaging (MRI), history of liver, kidney, cardiac, or psychiatric disease that may have impacted the study procedures were also excluded
Recruitment/selection of patients:	
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Bone marrow mesenchymal stem cell, CD105 expression ≥ 70% CD73 expression ≥ 70% CD90 expression ≥ 70% CD14 expression ≤ 10% CD45 expression ≤ 10% CD34 expression ≤ 10% Viability ≥ 80% at the time of freezing and in an aliquot thawed 2–3 weeks after freezing Detection of ≥ 70% expression of CD73, CD90, and CD105 in an aliquot thawed 2–3 weeks after freezing Lack of colony forming capacity in methylcellulose Lack of abnormal karyotype detected in at least 20 metaphases analyzed (unless an insufficient number of metaphases is obtained due to low cell growth as evidenced by the mitotic index Route: Intravenous Dose: $1–2\times10^6$ cells per kg bodyweight

Outcomes reported with time points	Coprimary outcomes were • The safety evaluation was based on the number and severity of adverse events that occurred within each treatment group. • Activity was evaluated by the total number of GELs at week 24(sum of number of GELs at weeks 4, 12, and 24) after MSC treatment as compared with placebo (suspension media). Secondary outcomes assessed • The activity of the cell therapy by analyzing MRI activity defined as: number of GELs counted over week 28, 36, and 48 (cross over retreatment) compared with the number of GELs counted over 4,12, and 24 weeks (placebo vs. active treatment periods) within each group; • Clinical measures assessed by the following secondary endpoint: • Number of relapses in the MSC treatment group vs placebo group in the first 24 weeks and after crossover re-treatment in the two groups; • time to sustained progression of disability and proportion of progression-free patients compared between treatment groups during the first 24 weeks; • proportion of disease-free patients defined as patients without relapses, with no evidence of sustained progression of disability and new MRI activity compared between treatment groups during the first 24 weeks • Changes in Multiple Sclerosis Functional Composite (MSFC) score • An additional safety evaluation was ensured by gathering the number and severity of adverse events from the second infusion(crossover) until 48 weeks	
Funding	s Fondazione Italiana Sclerosi Multipla, the European Committee for Treatment and Research in Multiple Sclerosis, the Multiple Sclerosis International Federation, Compagnia di San Paolo, the Danish Multiple Sclerosis Society, Toyota Foundation, the Danish Blood Donors' Research Foundation, Spanish Health Research Institute Carlos 3, Foundacion Progreso y Salud, the Royan Institute, the Spinal Cord Injury and Tissue Regeneration Centre, Fondation pour l'aide à la recherche sur la scléroseen plaques (ARSEP), French Muscular Dystrophy Association (AFM)-Telethon, the UK Multiple Sclerosis Society, the UK Stem Cell Foundation, the Multiple Sclerosis Society of Canada, the Multiple Sclerosis Scientific Research Foundation, and Research Manitoba.	
RoB2 Assessment: Specify separately for each domain		
EDSS Score		
Domain	Risk assessment	Support for judgment (including statements from the trial)

D1 (Randomization process)	Low	A randomization list was generated by a centralized clinical trial center and loaded on the eCRF.
D2 (deviation from intended intervention)	Low	Double blind phase I/II placebo controlled randomized trial Multicentre study Analysis seems to be appropriate. Baseline EDSS did not differ significantly as per the data provided throughout the published study. The early stem cell group had a mean of 4.6 (SD1.38) and the delayed group had a mean of 4.5 (SD 1.40).
D3 (missing outcome data)	Low	Out of 69 experimental and 75 control participants, data was available for 68 and 71 participants, respectively.
D4 (Measurement of outcome)	Low	EDSS was assessed by examining neurologist Outcome was assessed in the same way in both the groups Treating physicians and examining neurologists were different.
D5 (selection of reported results	Low	The mean EDSS score comparison was not the primary or secondary objective mentioned in the protocol; however, data for EDSS were sent through email by the authors of the published paper; therefore, we consider it probably yes in accordance with the
Overall		We judged low concern for overall as low risk in all domain
Annual Relapse Rate		
Domain	**Risk assessment**	**Support for judgment (including statements from the trial)**
D1 (Randomization process)	Low	A randomization list was generated by a centralized clinical trial center and loaded on the eCRF.
D2 (deviation from intended intervention)	Low	Double blind phase I/II placebo controlled randomized trial Multicentre study Analysis seems to be appropriate. Baseline EDSS did not differ significantly as per the data provided throughout the published study. The early stem cell group had a mean of 4.6 (SD1.38) and the delayed group had a mean of 4.5 (SD 1.40).
D3 (missing outcome data)	Low	Out of 69 experimental and 75 control participants, data was available for 68 and 71 participants, respectively.

D4 (Measurement of outcome)	Low	Examined by neurologist Outcome was assessed in the same way in both the groups Treating physician and examining neurologist were different
D5 (selection of reported results	Low	ARR was pre-specified
Overall		We judged low concern for overall as low risk in all domain

Study	**Fernandez et al 2018[6]**
Study type:	RCT
Number of participants:	21
Countries and setting:	Spain (two-centers - Hospital Regional Universitario de Ma´laga and Hospital Virgen Macarena in Seville) Setting: Hospital
Duration of study Follow up (post intervention):	12 months
Method of assessment of disease condition:	Active disease was defined as follows: diagnosis of SPMS, with an Expanded Disability Status Score (EDSS) between 5.5 and 9 and have failed previous therapies and have activity or progression of the disease (relapse in the previous year or progression of at least 0.5 points on the EDSS despite therapy)
Subgroup analysis within study	Sub-group analysis was conducted to compare between the high dose and low dose groups
Inclusion criteria:	To be eligible for the study, adult patients of either sex had to have a diagnosis of SPMS, with an Expanded Disability Status Score (EDSS) between 5.5 and 9 and have failed previous therapies and have activity or progression of the disease (relapse in the previous year or progression of at least 0.5 points on the EDSS despite therapy).
Exclusion criteria:	**Exclusion Criteria** • Patients were excluded if they had experienced a relapse, • Steroid treatment in the month prior to inclusion, • Patients positive for HIV, hepatitis B, or hepatitis C, • History of malignant neoplasms, • Participation in an interventional trial in the previous 3 months, • Contraindications for magnetic resonance imaging (MRI), history of liver, kidney, cardiac, or psychiatric disease that may have impacted the study procedures
Recruitment/selection of patients:	Thirty-four patients underwent lipectomy for AdMSCs collection, were randomized and thirty were

	infused (11 placebo, 10 low-dose and 9 high-dose)	
Intervention: Type of stem cells with method of their characterization, Route of administration, Dose	Adipose-derived mesenchymal stem cells (AdMSCs) quality controls: immunophenotype (>90% positivity for CD90, CD73, CD105, CD13 and CD29, and <10%positivity for CD14/CD20/CD34/CD45/CD31 and HLA class II), negativity for mycoplasms, negativity for endotoxins, and no numeric or structural karyotype abnormalities. Results from the sterility test were obtained prior to infusion and 14 days after the release of the finished product. Route: Intravenous Dose: Low dose: $1x10^6$ cells/kg or High dose: $4x10^6$ cells/kg	
Outcomes reported with time points	Neurological assessments were performed by experienced neurologists at the participating centers. Safety was assessed primarily by monitoring for AEs and standard laboratory measures. Vital signs and spirometric parameters were also monitored. Possible treatment effects were assessed by changes in baseline, 6 and 12 months after infusion: clinical variables (number of exacerbations, EDSS), immunologic assessments, analysis of cerebrospinal fluid (CSF), MRI (T2-weighted lesions, T1 lesions, T1 gadolinium-enhanced lesions, volume, and magnetization transfer ratio-MTR), evoked potentials (EP) (visual-VEP, acoustic-BAEP, somatosensory-SEP and motor-MEP), optical coherence tomography (OCT) of the retina (retinal nerve fiber layer- RNFL), cognition measured with Paced Auditory Serial Addition Test (PASAT) and quality-of-life by questionnaire SF-36, EuroqoL-5D and MusiQoL.	
Funding	This work was supported by Instituto de Salud Carlos III, Ministry of Health – Spain – Grant number: EC08/00224-ISCIII; Grants from ISCIII Co-funded by Fondos FEDER, FIS PI14/01015; RD/oo19/0028 and RD16/0011/0034 to B Soria and PI16/00259 to A Hmadcha. The sponsor was the Andalusian Initiative for Advanced Therapies, supported by the Andalusian Health and Progress Foundation	
RoB2 Assessment: Specify separately for each domain		
Domain	**Risk assessment**	**Support for judgment (including statements from the trial)**
D1 (Randomization process)	Low	Randomization was performed at CABIMER with a random allocation. sequence using sequentially numbered containers stratified by treatment center. Placebo was used in the control group. Baseline EDSS score of placebo 7.64±0.98, low dose 7.50±0.71, and high dose 7.78±0.44 seems to be no problem with the randomization process. The age distribution was similar between groups.

D2 (deviation from intended intervention)	Low	Placebo-controlled study. The study was triple blinded; treating physicians, patients, and statisticians were unaware of treatment assignment. The intention-to- treat population comprised all patients who were randomized.
D3 (missing outcome data)	Low	34 patients were randomized, out of which 4 patients did not receive infusions due to karyotyping annomalies in the cell project, which is an automatic process. Out of the remaining 30, 29 had outcome assessments.
D4 (Measurement of outcome)	Low	Neurological assessments were performed by experienced neurologists at the participating sites. There is a minimal chance that the measurement of outcomes would have differed between the groups. In the protocol, the outcome assessor was blinded. In the articles, neurologists were blinded, and outcomes were assessed by neurologists.
D5 (selection of reported results	Low	Protocols indicate that EDSS was an outcome measure pre-specified. There were no possibilities as no similar outcome was assessed and EDSS was pre-specified. There were no possibilities as the data was analyzed as per protocol.
Overall		Overall Low Risk of Bias

viii. List of excluded studies:

Studies	**Reason for exclusion**
Tremblay et al. 2022	Extension of earlier published study Ucceli et al.
Berard et al. 2022	Outcome data considered in the study was not reported
Petrou et al. 2020	Relevant data was not reported

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