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Study Protocol for the G-SPIRIT trial: A Randomized, Placebo-Controlled, Double-blinded Phase 3 Trial of Granulocyte Colony Stimulating Factor-Mediated Neuroprotection for Acute Spinal Cord Injury

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Abstract

Introduction: Granulocyte Colony Stimulating Factor (G-CSF) is generally used for neutropenia. Previous experimental studies revealed that G-CSF promoted neurological recovery after spinal cord injury (SCI). Next we moved to early phase of clinical trials. In a phase 1/2a trial, no adverse events were observed. Next, we conducted a non-randomized, non-blinded, comparative trial, which suggested the efficacy of G-CSF for promoting neurological recovery. Based on those results, we are now performing a phase 3 trial.
Methods and Analysis: The objective of this study is to prove the efficacy of G-CSF for acute SCI. The study design is a prospective, multicenter, randomized, double-blinded, placebo-controlled comparative study. The current trial includes cervical SCI (severity of American Spinal Injury Association (ASIA) Impairment Scale B/C) within 48 hours after injury. Patients are randomly assigned to G-CSF and placebo groups. The G-CSF group is administered 400 μg/m^2/d×5d of G-CSF in normal saline via intravenous infusion for 5 consecutive days. The placebo group is similarly administered a placebo. Our primary endpoint is changes in ASIA motor scores from baseline to 3 months. Each group includes 44 patients (88 total patients).This trial is funded by the Center for Clinical Trials, Japan Medical Association.
Ethics and Dissemination: The study was approved by the Institutional Review Board of each institution and will be conducted according to the principles of the World Medical Association Declaration of Helsinki and in accordance with the Japanese Medical Research Involving Human Subjects Act and other guidelines, regulations and Acts. Results of the clinical study will be submitted to the head of the respective clinical study site as a report after conclusion of the clinical study by the sponsor-investigator. Even if the results are not favorable despite conducting the clinical study properly, the data will be published as a paper.
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Strengths and Limitations of This Study

Strengths

- ✓ Novel drug therapy for acute spinal cord injury is much-needed.
- ✓ Randomized, placebo-controlled, double-blinded design can eliminate bias.

Limitations

- ✓ Acute spinal cord injury patients are difficult to recruit to the trial.
- ✓ Patient's neurological status in acute phase is unstable, possibly resulting in dispersion of the patient's background.
- ✓ Concealment must be performed very strictly because G-CSF apparently increases

white blood cell count.

Introduction

Spinal cord injury (SCI) is a devastating injury by which the patient can suffer from long-lasting severe sequelae including palsy of extremities, sensory disturbance, bowel/bladder/sexual dysfunction, and neuropathic pain. Conceptually, SCI is divided into two chronological phases: a primary and a secondary phase. Primary injury is mechanical damage to spinal cord tissue itself caused by fracture and/or dislocation or compression. Secondary injury is triggered by the primary injury and is a biological reaction of the spinal cord, which includes ischemia, hemorrhage, excitotoxicity, hyperpermeability, and inflammation[1]. Because secondary injury can be the main target of treatment, extensive laboratory and clinical investigation of neuroprotection is needed to manage secondary injury[2].

To date, the only approved neuroprotective therapy for SCI is massive methylprednisolone sodium succinate (MPSS) therapy based on the NASCIS 2 study[3]. However, recent reports revealed that MPSS shows only a modest effect for SCI. In addition, several reports have described adverse events induced by MPSS for SCI including infections (pneumonia, urinary tract infection) and gastrointestinal disorders (gastric ulcer, etc.)[4]. Therefore, the use of MPSS for SCI has become controversial[5]. Accordingly, a new therapeutic drug for SCI is desirable.

Granulocyte colony-stimulating factor (G-CSF, generic name: filgrastim) is a growth factor that affects the hematopoietic system, promoting differentiation, proliferation, and survival of granulocytes[6]. Clinically, in Japan, G-CSF is administered to patients with leukopenia, and to peripheral stem cell transplantation donors, G-CSF is administered to mobilize hematopoietic stem cells into the peripheral blood[7]. In the central nervous system, G-CSF has properties to mobilize the movement of bone marrow cells into the brain[8] and spine, and in a stroke model, has shown neuroprotective properties[9]. In other countries, clinical studies of the effects of G-CSF in cerebral infarction have been reported[10].

To prove the hypothesis that G-CSF has neuroprotective properties against SCI, G-CSF was administered to rat and mouse animal models of spinal cord injury, and hind limb

function improved significantly after administration of G-CSF. Further investigations into the mechanism of action of G-CSF in SCI were conducted. Data obtained to date identify the following properties of G-CSF: [1] mobilization of bone marrow-derived stem cells causing their biointegration at the site of SCI[11], [2] direct inhibition of nerve cell death[12], [3] protection of the myelin sheath by inhibiting oligodendrocyte cell death[13], [4] inhibition of inflammatory cytokine expression (TNF-α, IL-1β)[13], and [5] promotion of neovascularization[14]. These properties suggest that G-CSF has a neuroprotective effect in acute SCI.

Based on these properties, a phase 1/2a clinical study was conducted where the main objective was to confirm the safety and feasibility of G-CSF for treatment of patients with acute SCI[15]. This study was an open-label, dose-titrating study with no control group. As the initial step, 5 patients were given 5 μg/kg/day of G-CSF for 5 consecutive days by i.v. infusion, and as the second step, 11 patients were given 10 μg/kg/day of G-CSF for 5 consecutive days by i.v. infusion. No serious adverse events were noted, and the safety of G-CSF administration in patients with acute SCI was confirmed[15].

As a next step, to validate the efficacy of G-CSF in neuroprotective treatment, a multicenter, prospective, nonrandomized, nonblinded, comparative control study (phase 2b clinical trial) was conducted[16]. Based on the results of the previous phase 1/2a clinical trial, the dosage and duration of treatment with G-CSF in this study was 10 μg/kg/day for 5 consecutive days. Patients with acute cervical SCI (within 48 h of injury) were registered in the clinical trial and allocated to either the G-CSF group (G-CSF 10 μg/kg/day × 5 days i.v. infusion) or control group (no G-CSF administration) at each treatment facility. A total of 19 patients in the G-CSF group and 26 patients in the control group were observed for 3 months or longer. American Spinal Injury Association (ASIA) motor score (motor: 0 to 100 points) was compared between the groups. ASIA motor scores were 26.1 ± 18.9 in the G-CSF group and 12.2 ± 14.7 in the control group showing a significant improvement of motor paralysis in the G-CSF group ($p < 0.01$). In addition, in cases that could be followed for 1 year or longer, a significant improvement of ASIA motor score was observed in the G-CSF group[16].

Based on results of these preclinical and early phase clinical trials, we are now conducting a phase 3 confirmative trial.

Specific Objective

The objective of this study is to prove the efficacy of G-CSF for improving motor paralysis in acute SCI.

Methods and Analysis

Design of the study

The study design of the current trial is a prospective, multicenter, randomized, double-blinded, placebo-controlled comparative study.

Study Procedures

Patients will be randomly assigned to G-CSF and placebo groups. A central registration system will be used for dynamic randomization into the investigational treatment group (G-CSF) and control group (placebo) based on age at registration (16–64 years of age, or 65–84 years of age) and severity of paralysis (AIS B or C) at 48 h after injury. Each subject will be randomly allocated to either the investigational treatment group (G-CSF) or control group (placebo) in a 1 to 1 ratio. The subject registration center uses a program based on an appropriate computer algorithm to allocate patients into groups.

The G-CSF group will be administered 400 μg/m^2/d × 5d of G-CSF in normal saline via intravenous infusion for 5 consecutive days. The placebo group will be similarly administered a placebo. Allocation will be concealed between blinded evaluators of efficacy/safety and those for laboratory data, as G-CSF markedly increases white blood cell counts that can reveal patient treatment.

Our primary endpoint is changes in ASIA motor scores from baseline to 3 months calculated as follows: 3-month ASIA motor score change = 3-month ASIA motor score – pretreatment ASIA motor score.

Secondary endpoints are as follows. [1] Change in ASIA motor scores at 6 months and 12 months after G-CSF administration compared with pretreatment. [2] Changes in sensory

paralysis over time: change in ASIA sensory scores at 3 months, 6 months, and 12 months after G-CSF administration compared with pretreatment. [3] Severity of functional compromise because of paralysis: AIS before administration and at 3, 6, and 12 months after administration of G-CSF. [4] Percentage of responders: percentage of patients whose AIS improved by 1 grade or more at 3, 6, and 12 months after administration compared with before administration of G-CSF. [5] Neurological level of injury (NLI): percentage of patients whose NLI decreased by 1 grade of more at 3, 6, or 12 months after administration of G-CSF compared with pretreatment. [6] SCIM: change in ASIA motor scores at 3, 6, and 12 months after G-CSF administration compared with pretreatment [7] EQ-5D: measured EQ-5D efficacy scores at 3, 6, and 12 months after G-CSF.

Each group will include 44 patients (88 patients in total). Our protocol was approved by the Pharmaceuticals and Medical Device Agency and this trial will be funded by the Center for Clinical Trials, Japan Medical Association.

Inclusion

Inclusion criteria are as follows. [1] Patients with cervical SCI (severity of AIS B/C) within 48 h of injury. [2] Patients reassessed for neurological status at 48 h after injury, and those whose palsy is AIS B/C will be enrolled. [3] Patients with neurological level of injury between C4 and C7. [4] Patients with age of 16 to 85 years. [5] Patients who agree to participate in the current trial and from whom informed consent was obtained orally and in writing. [6] Patients who can be followed up for 12 months after SCI.

Exclusion

Exclusion criteria are as follows. [1] Patients with neurological recovery to AIS D at neurological reassessment 48 h after SCI, because only AIS B/C patients will be included to standardize the severity of paresis in order to stratify the patients at the initiation of drug administration. [2] Allergy to G-CSF. [3] Hematological malignancy, [4] within 6 months after invasive coronary intervention, [5] splenomegaly, or [6] pregnancy, [7] neurological disorders that can affect neurological evaluation in the present trial, [8] fracture of

extremities that can affect the neurological evaluation. Exclusion criteria 2 - 6 are set for safety and criteria 1, 7 and 8 are set to maintain homogeneity of the patient population enrolled.

Concealment

Patients will be administered drug or placebo and be evaluated in a double-blinded manner. An evaluator blinded to the treatment will take charge of patient evaluation including clinical findings and paresis, without laboratory data because G-CSF induces an apparent increase of white blood cell count that makes it easy to distinguish G-CSF and placebo treatment. Therefore, a non-blinded evaluator for laboratory data will be assigned to evaluate laboratory data alone. From a safety point-of view, the dose of G-CSF will be modulated according to excessive increase of white blood cell count as shown in Table 2. Therefore, a non-blinded evaluator will instruct the pharmacy to modulate the dose of G-CSF based on white blood cell count.

Sample Size Calculation

The target sample size for this randomized trial is 88. This number is based on the results of previous clinical trials[16]. The estimated group difference (± standard deviation) of change in ASIA motor scores from baseline to 3 months is 13.9 (± 21.9). A sample size of 44 patients in each group will provide 80% power to detect a difference of the change in ASIA motor scores between the G-CSF and the placebo treatments, using a mixed-effects models for repeated measures (MMRM) at a two-sided 5% level of significance. A common correlation of 0.25 at each time point is assumed. A dropout rate of 10% is allowed. Thus, the total sample size of 88 patients is required for the trial.

Statistical analyses

The analyses of the primary and secondary end points will be performed in a full analysis set, which includes all patients who: (1) took at least one course of treatment during the study; (2) do not present any serious violation of the study protocol; and (3) have

data collected after commencement of treatment. For the baseline characteristics, summary statistics will comprise frequencies and proportions for categorical variables, and means and SDs for continuous variables. The patient characteristics will be compared using a chi-square test for categorical variables, and a *t* test or Wilcoxon rank sum test for continuous variables.

For the primary analysis, aimed at comparing treatment effects, a change in ASIA motor score from baseline to 3 months and its 95% confidence interval (CI) will be estimated using the MMRM. To test for significant association of the primary endpoint, a mixed effects model for repeated measures with an unstructured covariance matrix will be applied to adjust for age (<65 years or ≥65 years) and AIS at 48 h after the injury (B or C).

For the secondary analysis, the change in ASIA motor score will be compared using a Student *t* test and the 95% CI will be estimated. The same method will be applied to change in sensory score, SCIM, and EQ-5D. A chi-squared test will be applied to the frequencies of the responder in AIS and of the improvement in NLI. The frequency of AIS will be summarized. The frequency of AEs will be compared using a Fisher exact test.

All comparisons are planned and all p values will be two sided. $p < 0.05$ will be considered significant. All statistical analyses will be performed using SAS software (version 9.4; SAS Institute, Cary, NC, USA). The statistical analysis plan will be developed by the principal investigator and the biostatistician before completion of patient recruitment and fixing of data.

Data Monitoring Committee

The data monitoring committee consists of three clinical trial specialists, including a biostatistician, who are independent from the current study. The committee will meet at least 2 times per year and all the data obtained by the current trial will be checked by the committee.

Adverse Events

As for safety evaluation, adverse events will be collected as follows. “Adverse events”

refers to any untoward symptom or disease or signs of such (including clinical laboratory data abnormalities) in a clinical investigation subject after informed consent and does not necessarily have a causal relationship with the investigational product (G-CSF).

Increases in white blood counts will be considered an adverse event only when the count exceeds 50,000/μL from the perspective of a pharmacological effect of G-CSF, and any values below this will not be handled as an adverse event.

All adverse events will code terminology used by the investigators according to the ICH International Medical Dictionary for Regulatory Activities Japanese version (MedDRA/J).

Ethics and Dissemination

Ethics

The study was approved by the Institutional Review Board (IRB) of each institutions involved to the present trial and will be conducted according to the principles of the World Medical Association (WMA) Declaration of Helsinki—Ethical Principles for Medical Research Involving Human Subjects with the amendments made in Seoul, South Korea, October 2008, with a Note of Clarification on Paragraph 29 added by the WMA General Assembly, Washington 2002; Note of Clarification on Paragraph 30 added by the WMA General Assembly, Tokyo 2004, and in accordance with the Japanese Medical Research Involving Human Subjects Act (WMO) and other guidelines, regulations and Acts.

Public Disclosure and Publication Policy

Results of the clinical study will be submitted to the head of the respective clinical study site as a report after conclusion of the clinical study by the sponsor-investigator (includes study coordinating investigator). Even if the results are not favorable despite conducting the clinical study properly, the data will be published as a paper. Other sponsor-investigators (including the clinical study coordinating investigator), if they plan to publicize the data from this study in a specialized academic society conference or other external site, must first obtain permission from the other principal investigators and investigational product provider. In publicizing the results, the confidentiality of the subjects will be maintained and proofread in advance by the other sponsor-investigators

(includes coordinating investigator) and investigational product provider.

Discussion

The current trial is a confirmative trial to elucidate the therapeutic efficacy of G-CSF for SCI. If the current trial can successfully show significant improvement of motor paralysis of SCI by G-CSF, we will move forward to drug approval application to the Ministry for Health and Labor, Japan. The entire protocol of the current trial was approved beforehand for the initiation of the current trial by the Japanese Pharmaceutical and Medical Device Agency (PMDA). The PMDA will also permit a drug approval application if significant efficacy of G-CSF for SCI is proven.

The current trial is an important milestone for SCI clinics and research to explore G-CSF for SCI.

Trial Status

The present trial is now on-going.

Trial Sites

Nineteen major hospitals in Japan constituting the G-SPIRIT study group as follows: Tohoku University Hospital, Miyagi; Niigata University, Niigata; Dokkyo University Hospital, Tochigi; Tsukuba University Hospital, Ibaraki; Tsukuba Medical Center, Ibaraki; Chiba University Hospital, Chiba; Funabashi Municipal Medical Center, Chiba; Kimitsu Chuo Hospital, Chiba; Chiba Rosai Hospital, Chiba; Tokai University Hospital, Kanagawa; Hamamatsu University Hospital, Shizuoka; Gifu University Hospital, Gifu; Chubu Rosai Hospital, Aichi; Mie University Hospital, Mie; Kanazawa Medical Collage, Ishikawa; Kobe Red Cross Hospital, Hyogo; Hiroshima University Hospital, Hiroshima; Yamaguchi University Hospital, Yamaguchi; and Nagasaki Rosai Hospital, Nagasaki.

Funding Statement

This trial is supported by Center for Clinical trials, Japan Medical Association, Japan.

Competing Interests

No authors have any competing interests to declare.

Contributorship Statement

MK, TF and MY contributed to planning and conduct of the present trial and to reporting the present protocol paper. HH, TS, YF, MH and ST contributed to conception and design of the present trial protocol and to reporting the present protocol paper. YI, JS, MK, SO, YM, TA, KW, TH, MO, HS, TM, IT, NK, MK. YO, TS, MY, MF, KY, DS, HT, DT, SI, HM, HU, FA, YS, IA, YT, MM, JS, YS, SI, HS, KT, YI, FN, MH, TO, FH, TF, KK, MW, HK, YM, YY, DT, TH, SK, GY, SO, TB, HA, KA, EK, HI, TS, AS, YI, TK, SO, NT, KN, NK, SK, HB, TO, HK, TY, IK, KY, TM, TM, IM, HK, TA, KH, TE, TS, MM, KF, SN, CI, TT, TK, HS, NN and MF contributed to conducting the present trial and to acquisition of data.

References

1. Oyinbo CA. Secondary injury mechanisms in traumatic spinal cord injury: a nugget of multiply cascade. ***Acta Neurobiol Exp*** 2011;71:281-299.
2. Karsy M, Hawryluk G. Pharmacologic Management of Acute Spinal Cord Injury. ***Neurosurg Clin N Am*** 2017;28:49-62. doi: 10.1016/j.nec.2016.07.002.
3. Bracken MB, Shepard MJ, Collins WF, et al. A randomized controlled trial of methylprednisolone or naloxone in the treatment of acute spinal cord injury: results of the second national acute spinal cord injury study. ***N Engl J Med*** 1990;322: 1405-1411.
4. Matsumoto T, Tamaki T, Kawakami M, et al. Early complications of high-dose metyl-prednisolone sodium succinate treatment in the follow-up of acute cervical spinal cord injury. ***Spine*** 2001;26: 426-430.
5. Hurlbert RJ, Hadley, Walters BC, et al. Pharmacological Therapy for Acute Spinal cord Injury. ***Neurosurgery*** 2013;72: 93-105.
6. Nicola NA, Metcalf D, Matsumoto M, Johnson GR. Purification of a factor inducing

differentiation in murine myelomonocytic leukemia cells. Identification as granulocyte colony-stimulating factor. ***J Biol Chem*** 1983;258: 9017-9023.

7. Roberts AW: G-CSF: a key regulator of neutrophil production, but that's no all! ***Growth Factors*** 2005;23: 33–41.
8. Kawada H, Takizawa S, Takanashi T, et al. Administration of hematopoietic cytokines in the subacute phase after cerebral infarction is effective for functional recovery facilitating proliferation of intrinsic neural stem/progenitor cells and transition of bone marrow-derived neuronal cells. ***Circulation*** 2006;113: 701-710.
9. Schneider A, Kruger C, Steigleder T, et al. The hematopoietic factor G-CSF is a neuronal ligand that counteracts programmed cell death and drives neurogenesis. ***J Clin Invest*** 2005;115: 2083-2098.
10. Shyu WC, Lin SZ, Lee CC, Liu DD, Li H. Granulocyte colony-stimulating factor for acute ischemic stroke: a randomized controlled trial. ***CMAJ*** 2006;174: 927-933.
11. Koda M, Nishio Y, Kamada T, et al. Granulocyte colony-stimulating factor （G-CSF） mobilizes bone marrow-derived cells into injured spinal cord and promotes functional recovery after compression-induced spinal cord injury in mice. ***Brain Res*** 2007;1149: 223-231.
12. Nishio Y, Koda M, Kamada T, et al. Granulocyte colony-stimulating factor （G-CSF） attenuates neuronal death and promotes functional recovery after spinal cord injury in mice. ***J Neuropathol Exp Neurol*** 2007;66: 724-731.
13. Kadota R, Koda M, Kawabe J, et al. Granulocyte colony-stimulating factor (G-CSF) protects oligodendrocyte and promotes hindlimb functional recovery after spinal cord injury in rats. ***PLoS One*** 2012;7:e50391.
14. Kawabe J, Koda M, Hashimoto M, et al. Granulocyte colony-stimulating factor （G-CSF） exerts neuroprotective effects via promoting angiogenesis after spinal cord injury in rats. ***J Neurosurg Spine*** 2011;15: 414-421.
15. Takahashi H, Yamazaki M, Okawa A, et al. Neuroprotective therapy using granulocyte colony-stimulating factor for acute spinal cord injury: a phase I/IIa clinical trial. ***Eur Spine J*** 2012;21:2580-2587.

16. Inada T, Takahashi H, Yamazaki M, et al. A multicenter prospective non-randomized controlled clinical trial to prove neurotherapeutic effects of granulocyte colony-stimulating factor （G-CSF） for acute spinal cord injury: Analyses of follow-up cases after at least one year. ***Spine*** 2014;39:213-219.

CONSORT 2010 checklist of information to include when reporting a randomised trial*

Section/Topic	Item No	Checklist item	Reported on page No
Title and abstract			
	1a	Identification as a randomised trial in the title	1
	1b	Structured summary of trial design, methods, results, and conclusions (for specific guidance see CONSORT for abstracts)	3
Introduction			
Background and objectives	2a	Scientific background and explanation of rationale	4-5
	2b	Specific objectives or hypotheses	6
Methods			
Trial design	3a	Description of trial design (such as parallel, factorial) including allocation ratio	6
	3b	Important changes to methods after trial commencement (such as eligibility criteria), with reasons	N/A
Participants	4a	Eligibility criteria for participants	7
	4b	Settings and locations where the data were collected	11
Interventions	5	The interventions for each group with sufficient details to allow replication, including how and when they were actually administered	6-7
Outcomes	6a	Completely defined pre-specified primary and secondary outcome measures, including how and when they were assessed	6-7
	6b	Any changes to trial outcomes after the trial commenced, with reasons	N/A
Sample size	7a	How sample size was determined	8
	7b	When applicable, explanation of any interim analyses and stopping guidelines	N/A
Randomisation:			
Sequence generation	8a	Method used to generate the random allocation sequence	6
	8b	Type of randomisation; details of any restriction (such as blocking and block size)	6
Allocation concealment mechanism	9	Mechanism used to implement the random allocation sequence (such as sequentially numbered containers), describing any steps taken to conceal the sequence until interventions were assigned	8
Implementation	10	Who generated the random allocation sequence, who enrolled participants, and who assigned participants to interventions	6
Blinding	11a	If done, who was blinded after assignment to interventions (for example, participants, care providers, those	8

		assessing outcomes) and how	
	11b	If relevant, description of the similarity of interventions	N/A
Statistical methods	12a	Statistical methods used to compare groups for primary and secondary outcomes	8-9
	12b	Methods for additional analyses, such as subgroup analyses and adjusted analyses	N/A
Results			
Participant flow (a diagram is strongly recommended)	13a	For each group, the numbers of participants who were randomly assigned, received intended treatment, and were analysed for the primary outcome	N/A
	13b	For each group, losses and exclusions after randomisation, together with reasons	N/A
Recruitment	14a	Dates defining the periods of recruitment and follow-up	N/A
	14b	Why the trial ended or was stopped	N/A
Baseline data	15	A table showing baseline demographic and clinical characteristics for each group	N/A
Numbers analysed	16	For each group, number of participants (denominator) included in each analysis and whether the analysis was by original assigned groups	N/A
Outcomes and estimation	17a	For each primary and secondary outcome, results for each group, and the estimated effect size and its precision (such as 95% confidence interval)	N/A
	17b	For binary outcomes, presentation of both absolute and relative effect sizes is recommended	N/A
Ancillary analyses	18	Results of any other analyses performed, including subgroup analyses and adjusted analyses, distinguishing pre-specified from exploratory	N/A
Harms	19	All important harms or unintended effects in each group (for specific guidance see CONSORT for harms)	N/A
Discussion			
Limitations	20	Trial limitations, addressing sources of potential bias, imprecision, and, if relevant, multiplicity of analyses	N/A
Generalisability	21	Generalisability (external validity, applicability) of the trial findings	N/A
Interpretation	22	Interpretation consistent with results, balancing benefits and harms, and considering other relevant evidence	N/A
Other information			
Registration	23	Registration number and name of trial registry	3
Protocol	24	Where the full trial protocol can be accessed, if available	3
Funding	25	Sources of funding and other support (such as supply of drugs), role of funders	11

*We strongly recommend reading this statement in conjunction with the CONSORT 2010 Explanation and Elaboration for important clarifications on all the items. If relevant, we also recommend reading CONSORT extensions for cluster randomised trials, non-inferiority and equivalence trials, non-pharmacological treatments, herbal interventions, and pragmatic trials. Additional extensions are forthcoming: for those and for up to date references relevant to this checklist, see www.consort-statement.org.

BMJ Open

Study Protocol for the G-SPIRIT trial: A Randomized, Placebo-Controlled, Double-blinded Phase 3 Trial of Granulocyte Colony Stimulating Factor-Mediated Neuroprotection for Acute Spinal Cord Injury

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Primary Subject Heading:	Neurology
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Study Protocol for the G-SPIRIT trial: A Randomized, Placebo-Controlled, Double-blinded Phase 3 Trial of Granulocyte Colony Stimulating Factor-Mediated Neuroprotection for Acute Spinal Cord Injury

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Abstract

Introduction: Granulocyte Colony Stimulating Factor (G-CSF) is generally used for neutropenia. Previous experimental studies revealed that G-CSF promoted neurological recovery after spinal cord injury (SCI). Next we moved to early phase of clinical trials. In a phase 1/2a trial, no adverse events were observed. Next, we conducted a non-randomized, non-blinded, comparative trial, which suggested the efficacy of G-CSF for promoting neurological recovery. Based on those results, we are now performing a phase 3 trial.
Methods and Analysis: The objective of this study is to evaluate the efficacy of G-CSF for acute SCI. The study design is a prospective, multicenter, randomized, double-blinded, placebo-controlled comparative study. The current trial includes cervical SCI (severity of American Spinal Injury Association (ASIA) Impairment Scale B/C) within 48 hours after injury. Patients are randomly assigned to G-CSF and placebo groups. The G-CSF group is administered 400 μg/m^2/d×5d of G-CSF in normal saline via intravenous infusion for 5 consecutive days. The placebo group is similarly administered a placebo. Our primary endpoint is changes in ASIA motor scores from baseline to 3 months. Each group includes 44 patients (88 total patients).This trial is funded by the Center for Clinical Trials, Japan Medical Association.
Ethics and Dissemination: The study was approved by the Institutional Review Board of each institution and will be conducted according to the principles of the World Medical Association Declaration of Helsinki and in accordance with the Japanese Medical Research Involving Human Subjects Act and other guidelines, regulations and Acts. Results of the clinical study will be submitted to the head of the respective clinical study site as a report after conclusion of the clinical study by the sponsor-investigator. Even if the results are not favorable despite conducting the clinical study properly, the data will be published as a paper.
Registration: UMIN000018752
https://upload.umin.ac.jp/cgi-open-bin/ctr/ctr.cgi?function=brows&action=brows&type=summary&recptno=R000021694&language=J

Strengths and Limitations of This Study

Strengths

- ✓ Novel drug therapy for acute spinal cord injury is much-needed.
- ✓ Randomized, placebo-controlled, double-blinded design can eliminate bias.

Limitations

- ✓ Acute spinal cord injury patients are difficult to recruit to the trial.
- ✓ Patient's neurological status in acute phase is unstable, possibly resulting in dispersion of the patient's background.
- ✓ Concealment must be performed very strictly because G-CSF apparently increases

white blood cell count.

Introduction

Spinal cord injury (SCI) is a devastating injury by which the patient can suffer from long-lasting severe sequelae including palsy of extremities, sensory disturbance, bowel/bladder/sexual dysfunction, and neuropathic pain. Conceptually, SCI is divided into two chronological phases: a primary and a secondary phase. Primary injury is mechanical damage to spinal cord tissue itself caused by fracture and/or dislocation or compression. Secondary injury is triggered by the primary injury and is a biological reaction of the spinal cord, which includes ischemia, hemorrhage, excitotoxicity, hyperpermeability, and inflammation[1]. Because secondary injury can be the main target of treatment, extensive laboratory and clinical investigation of neuroprotection is needed to manage secondary injury[2].

To date, the only approved neuroprotective therapy for SCI is massive methylprednisolone sodium succinate (MPSS) therapy based on the NASCIS 2 study[3]. However, recent reports revealed that MPSS shows only a modest effect for SCI. In addition, several reports have described adverse events induced by MPSS for SCI including infections (pneumonia, urinary tract infection) and gastrointestinal disorders (gastric ulcer, etc.)[4]. Therefore, the use of MPSS for SCI has become controversial[5]. Accordingly, a new therapeutic drug for SCI is desirable.

Granulocyte colony-stimulating factor (G-CSF, generic name: filgrastim) is a growth factor that affects the hematopoietic system, promoting differentiation, proliferation, and survival of granulocytes[6]. Clinically, in Japan, G-CSF is administered to patients with leukopenia, and to peripheral stem cell transplantation donors, G-CSF is administered to mobilize hematopoietic stem cells into the peripheral blood[7]. In the central nervous system, G-CSF has properties to mobilize the movement of bone marrow cells into the brain[8] and spine, and in a stroke model, has shown neuroprotective properties[9]. In other countries, clinical studies of the effects of G-CSF in cerebral infarction have been reported[10].

To prove the hypothesis that G-CSF has neuroprotective properties against SCI, G-CSF was administered to rat and mouse animal models of spinal cord injury, and hind limb

function improved significantly after administration of G-CSF. Further investigations into the mechanism of action of G-CSF in SCI were conducted. Data obtained to date identify the following properties of G-CSF: [1] mobilization of bone marrow-derived stem cells causing their biointegration at the site of SCI[11], [2] direct inhibition of nerve cell death[12], [3] protection of the myelin sheath by inhibiting oligodendrocyte cell death[13], [4] inhibition of inflammatory cytokine expression (TNF-α, IL-1β)[13], and [5] promotion of neovascularization[14]. These properties suggest that G-CSF has a neuroprotective effect in acute SCI.

Based on these properties, a phase 1/2a clinical study was conducted where the main objective was to confirm the safety and feasibility of G-CSF for treatment of patients with acute SCI[15]. This study was an open-label, dose-titrating study with no control group. As the initial step, 5 patients were given 5 μg/kg/day of G-CSF for 5 consecutive days by i.v. infusion, and as the second step, 11 patients were given 10 μg/kg/day of G-CSF for 5 consecutive days by i.v. infusion. No serious adverse events were noted, and the safety of G-CSF administration in patients with acute SCI was confirmed[15].

As a next step, to validate the efficacy of G-CSF in neuroprotective treatment, a multicenter, prospective, nonrandomized, nonblinded, comparative control study (phase 2b clinical trial) was conducted[16]. Based on the results of the previous phase 1/2a clinical trial, the dosage and duration of treatment with G-CSF in this study was 10 μg/kg/day for 5 consecutive days. Patients with acute cervical SCI (within 48 h of injury) were registered in the clinical trial and allocated to either the G-CSF group (G-CSF 10 μg/kg/day × 5 days i.v. infusion) or control group (no G-CSF administration) at each treatment facility. A total of 19 patients in the G-CSF group and 26 patients in the control group were observed for 3 months or longer. American Spinal Injury Association (ASIA) motor score (motor: 0 to 100 points) was compared between the groups. ASIA motor scores were 26.1 ± 18.9 in the G-CSF group and 12.2 ± 14.7 in the control group showing a significant improvement of motor paralysis in the G-CSF group ($p < 0.01$). In addition, in cases that could be followed for 1 year or longer, a significant improvement of ASIA motor score was observed in the G-CSF group[16].

Based on results of these preclinical and early phase clinical trials, we are now conducting a phase 3 trial.

Specific Objective

The objective of this study is to evaluate the efficacy of G-CSF for improving motor paralysis in acute SCI.

Methods and Analysis

Design of the study

The study design of the current trial is a prospective, multicenter, randomized, double-blinded, placebo-controlled comparative study.

Study Procedures

The study outline is shown in Figure. Patients will be randomly assigned to G-CSF and placebo groups. A central registration system will be used for dynamic randomization into the investigational treatment group (G-CSF) and control group (placebo) based on age at registration (16–64 years of age, or 65–84 years of age) and severity of paralysis (AIS B or C) at 48 h after injury. Initially, screened patients with severity AIS B/C will be tentatively enrolled. Initial screening of the patients include clinical laboratory test, imaging studies including X-ray, magnetic resonance imaging and computed tomography, and neurological/functional evaluations.

Neurological re-assessment will be performed 48±4 hours after SCI and the patients who will recover to severity AIS D will be excluded. The patients with severity AIS B/C at neurological re-assessment 48±4 hours after SCI will be enrolled and randomly allocated to either the investigational treatment group (G-CSF) or control group (placebo) in a 1 to 1 ratio. The subject registration center uses a program based on an appropriate computer algorithm to allocate patients into groups. The first dose of investigative drugs will be administered to the patients after re-assessment of neurological status and enrollment 48±4 hours after SCI.

The G-CSF group will be administered 400 μg/m^2/d × 5d of G-CSF in normal saline via intravenous infusion for 5 consecutive days. The placebo group will be similarly administered a placebo. The first dose of investigative drugs will be administered to the patients 48±4 hours after SCI. The dosing schedule will be once a day in every morning (9-10 AM) for consecutive 5 days even in case that the first dosing was performed at night because of restriction on practices. Investigational drugs including both G-CSF and placebo will be stored in refrigerator which will be kept between 1- 6□ and has temperature-logger in pharmacies in each participating institutes. The investigational drugs is packaged in ample with label only printed as serial numbers, ten ample is packed in one box with label only printed serial numbers. Web-based allocation system will show the serial number of investigational drug which must be used to respective patients to ensure blinding.

We decided dosage of drug based on our previous non-randomized early phase clinical studies. On phase 1/2a clinical study, of which study design was open-label, dose-titrating study with no control group, SCI patients who were administered 10 μg/kg/day G-CSF for 5 days showed marked elevation of white blood cell number (reached nearly 50000/μl, of which WBC number might cause adverse effects of G-CSF) during G-CSF administration[15]. Therefore we decided to withdraw additional titration. In addition, next phase of clinical study, of which design was multicenter, prospective, nonrandomized, nonblinded, comparative control study, showed suggestive efficacy of G-CSF (10 μg/kg/d for 5 days) for acute SCI[16]. Based on those results, we finally decided the dosage of G-CSF as 10 μg/kg/d × 5d for 5 days. In the current clinical trial, the dosage of G-CSF is written as 400 μg/m^2/d (= 10 μg/kg/d) according to the Japanese Pharmaceutical and Medical Device Agency (PMDA)'s instruction for consolidation with product labeling.

We decided to 48hours after SCI as the therapeutic time window because our previous multicenter, prospective, nonrandomized, nonblinded, comparative control study, which recruited SCI patients within 48 hours after injury, showed that there was no significant difference in neurological outcome between the patient administered G-CSF very early after the injury and 48 hours after injury[16].

Allocation will be concealed between blinded evaluators of efficacy/safety and those

for laboratory data, as G-CSF markedly increases white blood cell counts that can reveal patient treatment.

Our primary endpoint is changes in ASIA motor scores (international standards for neurological classification of spinal cord injury (ISNSCI) suppl. Figure 1) from baseline to 3 months calculated as follows: 3-month ASIA motor score change = 3-month ASIA motor score – pretreatment ASIA motor score. To maintain consistency of neurological assessment among the each evaluators, attending lecture and e-learning (in website of International Spinal Cord Society: http://www.elearnsci.org/) of ASIA/ISNSCI scoring system is mandatory for every investigators participating to the present trial.

Secondary endpoints are as follows. [1] Change in ASIA motor scores at 6 months and 12 months after G-CSF administration compared with pretreatment. [2] Changes in sensory paralysis over time: change in ASIA sensory scores at 3 months, 6 months, and 12 months after G-CSF administration compared with pretreatment. [3] Severity of functional compromise because of paralysis: AIS before administration and at 3, 6, and 12 months after administration of G-CSF. [4] Percentage of responders: percentage of patients whose AIS improved by 1 grade or more at 3, 6, and 12 months after administration compared with before administration of G-CSF. [5] Neurological level of injury (NLI): percentage of patients whose NLI decreased by 1 grade of more at 3, 6, or 12 months after administration of G-CSF compared with pretreatment. [6] SCIM: change in SCIM scores at 3, 6, and 12 months after G-CSF administration compared with pretreatment [7] EQ-5D: measured EQ-5D efficacy scores at 3, 6, and 12 months after G-CSF.

To strengthen the analysis, more strict blindness of assessment by the evaluator for outcome measures is ideal. Therefore every prior score/measurement should be blinded and/or not the same person should assess every midpoint motor/sensory functional measurement. However, PMDA instructed us to assess one patients' functional evaluation by one evaluator for consistent assessment in respective patients. PMDA considers the consistency of assessment in respective patients is more important.

The precise explanation of abovementioned outcome measures is as followings (suppl. Figures). ASIA motor score is determined by examining a key muscle function within each

of 10 myotomes on each side of the body on manual muscle testing (MMT) in the supine position. There is a range of score from 0 to 25 for each extremity, totaling 0 to 50 for the upper limbs and 0 to 50 for the lower limbs, resulting in total of ASIA motor score ranges from 0 to 100. This score can reflect the degree of motor impairment associated with the SCI (suppl. Figure 1). ASIA sensory scores is completed through the testing of a key point in each of the 28 dermatomes (from C2 to S4-5) on the right and left sides of the body that can be readily located in relation to bony anatomical landmarks. At each of these key points, two aspects of sensation are examined: light touch and pin prick. Appreciation of light touch and pin prick sensation at each of the key points is separately scored on a three-point scale, with comparison to the sensation on the patients' cheek as a normal frame of reference: 0 = absent, 1= altered, 2 = normal or intact. As a result, ASIA sensory scores range from 0 to 112 (suppl. Figure 1). AIS designation is used in grading the degree of impairment: A = Complete. No sensory or motor function is preserved in the sacral segments S4-S5. B = Sensory incomplete. Sensory but not motor function is preserved below the neurological level. C = Motor incomplete. Motor function is preserved below the neurological level, and more than half of key muscle functions below the single neurological level of injury have a muscle grade less than 3 (Grades 0–2). D = Motor incomplete. Motor function is preserved below the neurological level, and at least half (half or more) of key muscle functions below the neurological level have a muscle grade >3. E = Normal. If sensation and motor function as tested are graded as normal in all segments, and the patient had prior deficits, then the AIS grade is E (suppl. Figure 1, footnote). NLI refers to the most caudal segment of the cord with intact sensation and antigravity muscle function strength, provided that there is normal (intact) sensory and motor function rostrally (suppl. Figure 1, footnote). SCIM is disability scale developed specifically for patients with spinal cord lesions in order to make the functional assessments of patients with paraplegia or tetraplegia more sensitive to changes. The SCIM includes the following areas of function: self-care (subscore (0-20), respiration and sphincter management (0-40) and mobility (0-40). Each area is scored according to its proportional weight in these patients' general activity. The final score ranges from 0 to 100 (suppl. Figure 2-4). EQ-5D

essentially consists of 2 pages (suppl. Figure 5, 6): the EQ-5D descriptive system and the EQ visual analogue scale (EQ VAS). The descriptive system comprises five dimensions: mobility, self-care, usual activities, pain/discomfort and anxiety/depression. Each dimension has 5 levels: no problems, slight problems, moderate problems, severe problems and extreme problems. The patient is asked to indicate his/her health state by ticking the box next to the most appropriate statement in each of the five dimensions. This decision results in a 1-digit number that expresses the level selected for that dimension. The digits for the five dimensions can be combined into a 5-digit number that describes the patient's health state and can be converted to efficacy score with calibration scale (ranges -0.111 to 1.000). The EQ VAS records the patient's self-rated health on a vertical visual analogue scale, where the endpoints are labelled 'The best health you can imagine' and 'The worst health you can imagine'. The VAS can be used as a quantitative measure of health outcome that reflect the patient's own judgement.

Each group will include 44 patients (88 patients in total). Our protocol was approved by PMDA and this trial will be funded by the Center for Clinical Trials, Japan Medical Association.

Inclusion

Inclusion criteria are as follows. [1] Patients with cervical SCI (severity of AIS B/C) within 48 h of injury. [2] Patients reassessed for neurological status at 48 h after injury, and those whose palsy is AIS B/C will be enrolled. [3] Patients with neurological level of injury between C4 and C7. [4] Patients with age of 16 to 85 years. [5] Patients who agree to participate in the current trial and from whom informed consent was obtained orally and in writing. [6] Patients who can be followed up for 12 months after SCI.

Exclusion

Exclusion criteria are as follows. [1] Patients with neurological recovery to AIS D at neurological reassessment 48 h after SCI, because only AIS B/C patients will be included to

standardize the severity of paresis in order to stratify the patients at the initiation of drug administration. [2] Allergy to G-CSF. [3] Hematological malignancy, [4] within 6 months after invasive coronary intervention, [5] splenomegaly, or [6] pregnancy, [7] consciousness impairment, [8] neurological disorders that can affect neurological evaluation in the present trial, [9] fracture of extremities that can affect the neurological evaluation and [10] massive dose administration of MPSS. Exclusion criteria 2 - 6 are set for safety, criteria 1, 7 - 9 are set to maintain homogeneity of the patient population enrolled, criteria 7-9 are set for maintenance of accuracy of functional assessment, criteria 7 is set to obtain patients' own informed consent upon participation to the trial and criteria 10 is set to omit the possible interference of MPSS on outcome assessment.

Concealment

Patients will be administered drug or placebo and be evaluated in a double-blinded manner. An evaluator blinded to the treatment will take charge of patient evaluation including clinical findings and paresis, without laboratory data because G-CSF induces an apparent increase of white blood cell count that makes it easy to distinguish G-CSF and placebo treatment. Therefore, a non-blinded evaluator for laboratory data will be assigned to evaluate laboratory data alone. From a safety point-of view, the dose of G-CSF will be modulated according to excessive increase of white blood cell count. Therefore, a non-blinded evaluator will instruct the pharmacy to modulate the dose of G-CSF based on white blood cell count.

Sample Size Calculation

The target sample size for this randomized trial is 88. This number is based on the results of previous clinical trials[16]. The estimated group difference (± standard deviation) of change in ASIA motor scores from baseline to 3 months is 13.9 (± 21.9). A sample size of 44 patients in each group will provide 80% power to detect a difference of the change in ASIA motor scores between the G-CSF and the placebo treatments, using a mixed-effects models for repeated measures (MMRM) at a two-sided 5% level of significance. A

common correlation of 0.25 at each time point is assumed. A dropout rate of 10% is allowed. Thus, the total sample size of 88 patients is required for the trial.

Statistical analyses

The analyses of the primary and secondary end points will be performed as intention-to-treat analyses in a full analysis set, which includes all patients who: (1) took at least one course of treatment during the study; (2) do not present any serious violation of the study protocol; and (3) have data collected after commencement of treatment. For the baseline characteristics, summary statistics will comprise frequencies and proportions for categorical variables, and means and SDs for continuous variables. The patient characteristics will be compared using a chi-square test for categorical variables, and a *t* test or Wilcoxon rank sum test for continuous variables. Missing data including loss to follow-up and missing measurement will be supplemented with MMRM.

For the primary analysis, aimed at comparing treatment effects, a change in ASIA motor score from baseline to 3 months and its 95% confidence interval (CI) will be estimated using the MMRM. To test for significant association of the primary endpoint, a mixed effects model for repeated measures with an unstructured covariance matrix will be applied to adjust for age (<65 years or ≥65 years) and AIS at 48 h after the injury (B or C).

For the secondary analysis, the change in ASIA motor score will be compared using a Student *t* test and the 95% CI will be estimated. The same method will be applied to change in sensory score, SCIM, and EQ-5D. A chi-squared test will be applied to the frequencies of the responder in AIS and of the improvement in NLI. The frequency of AIS will be summarized. The frequency of AEs will be compared using a Fisher exact test.

All comparisons are planned and all *p* values will be two sided. $p < 0.05$ will be considered significant. All statistical analyses will be performed using SAS software (version 9.4; SAS Institute, Cary, NC, USA). The statistical analysis plan will be developed by the principal investigator and the biostatistician before completion of patient recruitment and fixing of data.

Data Monitoring Committee

The data monitoring committee consists of three clinical trial specialists, including a biostatistician, who are independent from the current study. The committee will meet at least 2 times per year and all the data obtained by the current trial will be checked by the committee.

Adverse Events

As for safety evaluation, adverse events will be collected as follows. “Adverse events” refers to any untoward symptom or disease or signs of such (including clinical laboratory data abnormalities) in a clinical investigation subject after informed consent and does not necessarily have a causal relationship with the investigational product (G-CSF).

Increases in white blood counts will be considered an adverse event only when the count exceeds 50,000/μL from the perspective of a pharmacological effect of G-CSF, and any values below this will not be handled as an adverse event. Anaphylaxis and adult respiratory distress syndrome are the most representative G-CSF-related severe adverse events to be payed full attention.

All adverse events will code terminology used by the investigators according to the ICH International Medical Dictionary for Regulatory Activities Japanese version (MedDRA/J).

Ethics and Dissemination

Ethics

The study was approved by the Institutional Review Board (IRB) of each institutions involved to the present trial and will be conducted according to the principles of the World Medical Association (WMA) Declaration of Helsinki—Ethical Principles for Medical Research Involving Human Subjects with the amendments made in Seoul, South Korea, October 2008, with a Note of Clarification on Paragraph 29 added by the WMA General Assembly, Washington 2002; Note of Clarification on Paragraph 30 added by the WMA General Assembly, Tokyo 2004, and in accordance with the Japanese Medical Research Involving Human Subjects Act (WMO) and other guidelines, regulations and Acts.

Patient informed consent

The principal investigator will try to prepare the informed consent form and other explanatory materials in as simple language as possible in order to obtain informed consent from the patient and the patient's legal representative. In case that trial participants cannot sign the informed consent form due to upper extremity palsy caused by SCI, allograph by patients' representative will be allowed.

Public Disclosure and Publication Policy

Results of the clinical study will be submitted to the head of the respective clinical study site as a report after conclusion of the clinical study by the sponsor-investigator (includes study coordinating investigator). Even if the results are not favorable despite conducting the clinical study properly, the data will be published as a paper. Other sponsor-investigators (including the clinical study coordinating investigator), if they plan to publicize the data from this study in a specialized academic society conference or other external site, must first obtain permission from the other principal investigators and investigational product provider. In publicizing the results, the confidentiality of the subjects will be maintained and proofread in advance by the other sponsor-investigators (includes coordinating investigator) and investigational product provider.

Discussion

The current trial is a confirmative trial to elucidate the therapeutic efficacy of G-CSF for SCI. If the current trial can successfully show significant improvement of motor paralysis of SCI by G-CSF, we will move forward to drug approval application to the Ministry for Health and Labor, Japan. The entire protocol of the current trial was approved beforehand for the initiation of the current trial by the Japanese Pharmaceutical and Medical Device Agency (PMDA). The PMDA will also permit a drug approval application if significant efficacy of G-CSF for SCI is proven.

The current trial is an important milestone for SCI clinics and research to explore

G-CSF for SCI.

Trial Status

The present trial is now on-going.

Trial Sites

Nineteen major hospitals in Japan constituting the G-SPIRIT study group as follows: Tohoku University Hospital, Miyagi; Niigata University, Niigata; Dokkyo University Hospital, Tochigi; Tsukuba University Hospital, Ibaraki; Tsukuba Medical Center, Ibaraki; Chiba University Hospital, Chiba; Funabashi Municipal Medical Center, Chiba; Kimitsu Chuo Hospital, Chiba; Chiba Rosai Hospital, Chiba; Tokai University Hospital, Kanagawa; Hamamatsu University Hospital, Shizuoka; Gifu University Hospital, Gifu; Chubu Rosai Hospital, Aichi; Mie University Hospital, Mie; Kanazawa Medical Collage, Ishikawa; Kobe Red Cross Hospital, Hyogo; Hiroshima University Hospital, Hiroshima; Yamaguchi University Hospital, Yamaguchi; and Nagasaki Rosai Hospital, Nagasaki.

Funding Statement

This trial is supported by Center for Clinical trials, Japan Medical Association, Japan.

Competing Interests

No authors have any competing interests to declare.

Contributorship Statement

MK, TF and MY contributed to planning and conduct of the present trial and to reporting the present protocol paper. HH, TS, YF, MH and ST contributed to conception and design of the present trial protocol and to reporting the present protocol paper. YI, JS, MK, SO, YM, TA, KW, TH, MO, HS, TM, IT, NK, MK. YO, TS, MY, MF, KY, DS, HT, DT, SI, HM, HU, FA, YS, IA, YT, MM, JS, YS, SI, HS, KT, YI, FN, MH, TO, FH, TF, KK, MW, HK, YM, YY, DT, TH, SK, GY, SO, TB, HA, KA, EK, HI, TS, AS, YI, TK, SO, NT, KN, NK, SK, HB, TO, HK, TY, IK, KY, TM, TM, IM, HK, TA, KH, TE, TS, MM, KF, SN, CI, TT, TK, HS, NN and MF contributed to conducting the present trial and to acquisition of

data.

References

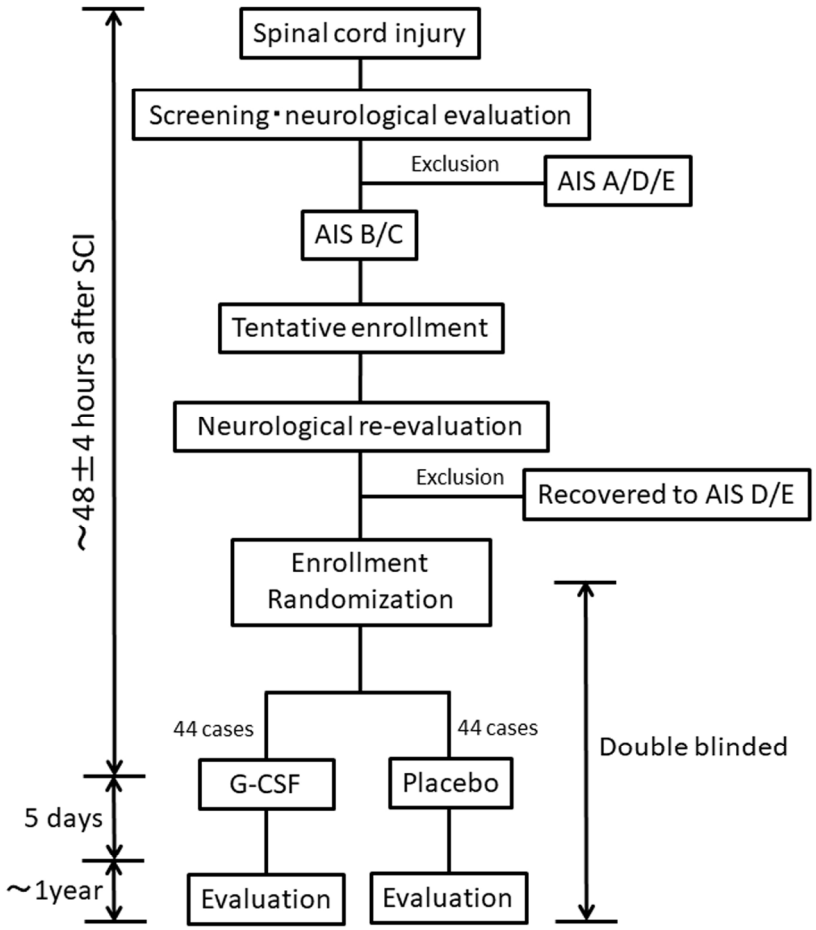
1. Oyinbo CA. Secondary injury mechanisms in traumatic spinal cord injury: a nugget of multiply cascade. ***Acta Neurobiol Exp*** 2011;71:281-299.
2. Karsy M, Hawryluk G. Pharmacologic Management of Acute Spinal Cord Injury. ***Neurosurg Clin N Am*** 2017;28:49-62. doi: 10.1016/j.nec.2016.07.002.
3. Bracken MB, Shepard MJ, Collins WF, et al. A randomized controlled trial of methylprednisolone or naloxone in the treatment of acute spinal cord injury: results of the second national acute spinal cord injury study. ***N Engl J Med*** 1990;322: 1405-1411.
4. Matsumoto T, Tamaki T, Kawakami M, et al. Early complications of high-dose metyl-prednisolone sodium succinate treatment in the follow-up of acute cervical spinal cord injury. ***Spine*** 2001;26: 426-430.
5. Hurlbert RJ, Hadley, Walters BC, et al. Pharmacological Therapy for Acute Spinal cord Injury. ***Neurosurgery*** 2013;72: 93-105.
6. Nicola NA, Metcalf D, Matsumoto M, Johnson GR. Purification of a factor inducing differentiation in murine myelomonocytic leukemia cells. Identification as granulocyte colony-stimulating factor. ***J Biol Chem*** 1983;258: 9017-9023.
7. Roberts AW: G-CSF: a key regulator of neutrophil production, but that's no all! ***Growth Factors*** 2005;23: 33–41.
8. Kawada H, Takizawa S, Takanashi T, et al. Administration of hematopoietic cytokines in the subacute phase after cerebral infarction is effective for functional recovery facilitating proliferation of intrinsic neural stem/progenitor cells and transition of bone marrow-derived neuronal cells. ***Circulation*** 2006;113: 701-710.
9. Schneider A, Kruger C, Steigleder T, et al. The hematopoietic factor G-CSF is a neuronal ligand that counteracts programmed cell death and drives neurogenesis. ***J Clin Invest*** 2005;115: 2083-2098.
10. Shyu WC, Lin SZ, Lee CC, Liu DD, Li H. Granulocyte colony-stimulating factor

for acute ischemic stroke: a randomized controlled trial. ***CMAJ*** 2006;174: 927-933.

11. Koda M, Nishio Y, Kamada T, et al. Granulocyte colony-stimulating factor（G-CSF）mobilizes bone marrow-derived cells into injured spinal cord and promotes functional recovery after compression-induced spinal cord injury in mice. ***Brain Res*** 2007;1149: 223-231.
12. Nishio Y, Koda M, Kamada T, et al. Granulocyte colony-stimulating factor（G-CSF）attenuates neuronal death and promotes functional recovery after spinal cord injury in mice. ***J Neuropathol Exp Neurol*** 2007;66: 724-731.
13. Kadota R, Koda M, Kawabe J, et al. Granulocyte colony-stimulating factor (G-CSF) protects oligodendrocyte and promotes hindlimb functional recovery after spinal cord injury in rats. ***PLoS One*** 2012;7:e50391.
14. Kawabe J, Koda M, Hashimoto M, et al. Granulocyte colony-stimulating factor（G-CSF）exerts neuroprotective effects via promoting angiogenesis after spinal cord injury in rats. ***J Neurosurg Spine*** 2011;15: 414-421.
15. Takahashi H, Yamazaki M, Okawa A, et al. Neuroprotective therapy using granulocyte colony-stimulating factor for acute spinal cord injury: a phase I/IIa clinical trial. ***Eur Spine J*** 2012;21:2580-2587.
16. Inada T, Takahashi H, Yamazaki M, et al. A multicenter prospective non-randomized controlled clinical trial to prove neurotherapeutic effects of granulocyte colony-stimulating factor（G-CSF）for acute spinal cord injury: Analyses of follow-up cases after at least one year. ***Spine*** 2014;39:213-219.

Figure legend

Schematic diagram showing trial timeline.



Schematic diagram showing trial timeline.

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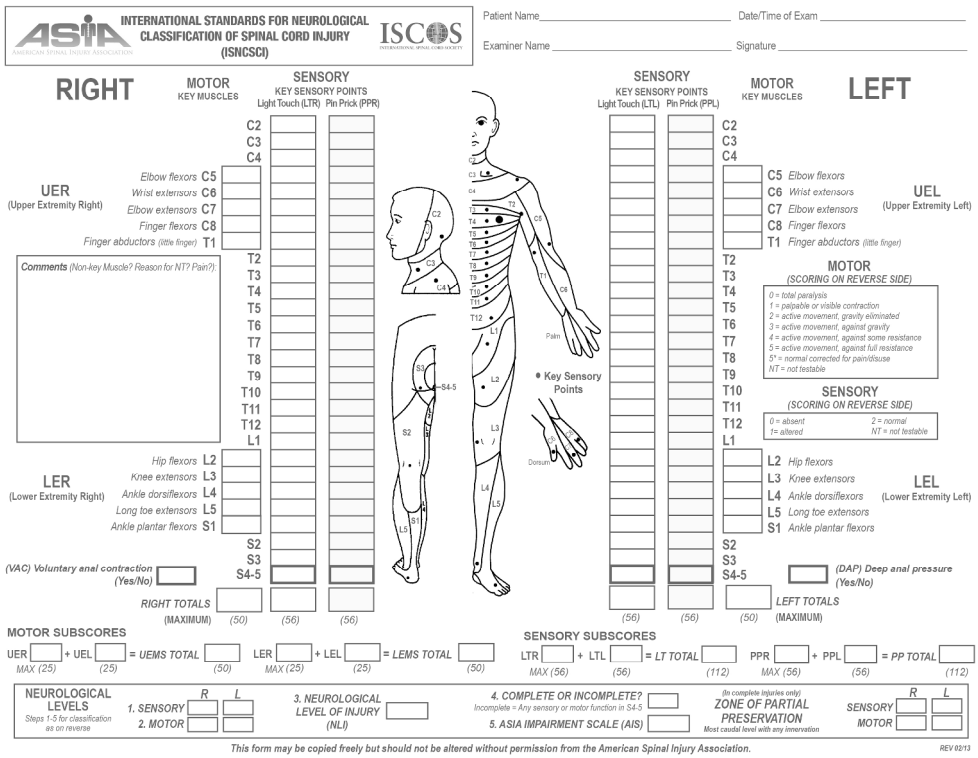

274x206mm (300 x 300 DPI)

セルフケア

(1) 食事 (切る, 容器を開ける, 飲み物を注ぐ, 食べ物を口に運ぶ, 飲み物の入ったコップをもつ)
- 0. 静脈栄養または胃瘻が必要である. あるいは経口摂取において全介助を要する.
- 1. 食べたり飲んだりすること, または補助器具の装着に部分介助を要する.
- 2. 食べることは自立, 補助器具を必要とするか, または食べ物を切ったり注いだりすること, 容器の開閉にのみ解介助を要する.
- 3. 食べることも飲むことも自立, 介助や補助器具を必要としない.

(2) 入浴 (頭と体を石鹸でこすって洗い, 乾かす, 蛇口の開け閉め) A —上半身, B —下半身

A. 0. 全介助を要する.
- 1. 部分介助を要する.
- 2. 補助器具, または特定の環境 (例えば, 手すりや椅子) が整っていれば, 自立して体を洗う.
- 3. 自立して体を洗う. (通常, 健常者なら用いることのない) 補助器具や特定の環境設定は不要

B. 0. 全介助を要する.
- 1. 部分介助を要する.
- 2. 補助器具, または特定の環境が整っていれば, 自立して体を洗う.
- 3. 自立して体を洗う. 補助器具や特定の環境設定は不要

(3) 更衣 (衣服, 靴, 常用している装具の着脱) A —上半身, B —下半身

A. 0. 全介助を要する.
- 1. ボタン, ファスナー, ひものない衣服で部分介助を要する.
- 2. ボタン, ファスナー, ひものない衣服であれば自立, 補助器具と特定の環境設定, またはそのどちらかが必要.
- 3. ボタン, ファスナー, ひものない衣服であれば自立, 補助器具も特定の環境設定も不要. ボタン, ファスナー, ひもの扱いにだけ介助, 補助器具, または特定の環境設定が必要.
- 4. (衣類の種類を問わず) 自立して着替える. 補助器具も特定の環境設定も不要.

B. 0. 全介助を要する.
- 1. ボタン, ファスナー, ひものない衣服で部分介助を要する.
- 2. ボタン, ファスナー, ひものない衣服であれば自立, 補助器具と特定の環境設定, またはそのどちらかが必要.
- 3. ボタン, ファスナー, ひものない衣服であれば自立, 補助器具も特定の環境設定も不要. ボタン, ファスナー, ひもの扱いにだけ介助, 補助器具, または特定の環境設定が必要.
- 4. (衣類の種類を問わず) 自立して着替える. 補助器具も特定の環境設定も不要.

(4) 整容 (手洗い, 洗顔, 歯磨き, 整髪, 髭剃り, 化粧)
- 0. 全介助を要する.
- 1. 部分介助を要する.
- 2. 補助器具があれば自立して整容動作をおこなう.
- 3. 補助器具を用いずに自立して整容動作をおこなう.

小計 (0–20)

呼吸と排泄管理

(5) 呼吸
- 0. 気管チューブと持続的補助換気または間欠的補助換気が必要.
- 2. 気管チューブを装用して自己呼吸, 酸素を必要としたり, 咳嗽時または気管チューブの管理に多大な介助を必要とする.
- 4. 気管チューブを装用して自己呼吸, 咳嗽時または気管チューブの管理に少ししか介助を必要としない.
- 6. 気管チューブなしで自己呼吸, 酸素, 咳嗽時の多大な介助, マスク (例 : 終末呼気陽圧 PEEP), または間欠的補助換気 (BiPAP) を必要とする.
- 8. 気管チューブなしで自己呼吸, 介助または咳嗽刺激を少ししか必要としない.
- 10. 介助も器具もなしで自己呼吸.

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(6) 排尿管理
0. 留置カテーテル
3. 残尿量＞100cc，不定期導尿または介助による間欠的導尿．
6. 残尿量＞100cc または間欠的自己導尿，集尿器[2] をあてがう際の介助は必要．
9. 間欠的自己導尿，集尿器を使用する．器具をあてがう際の介助は不要．
11. 間欠的自己導尿，導尿と導尿の間には失禁なし，集尿器は使用しない．
13. 残尿量＜100cc，集尿器のみ必要，集尿器の取扱いには介助不要．
15. 残尿量＜100cc，失禁なし，集尿器を使用しない．

(7) 排便管理
0. 排便が不規則またはごく低頻度（3 日に 1 回未満）．
5. 規則的ながら，（座薬を挿入するなどに）介助を要する，失敗はまれ（月 2 回未満）．
8. 規則的な排便で，介助を要しない，失敗はまれ（月 2 回未満）．
10. 規則的な排便で，介助を要しない，失敗はなし．

(8) トイレの使用（会陰部の清潔，使用前後での衣服の扱い，ナプキンまたはおむつの使用）
0. 全介助を要する．
1. 部分介助を要する．自分でお尻を拭けない．
2. 部分介助を要する．自立してお尻を拭ける．
4. 自立してトイレを使用するも，補助器具または特別な環境（例：手すり）が整っている必要あり．
5. 自立してトイレを使用．補助器具も特別な環境が整っている必要もない．

小計（0–40）

移動 (室内とトイレ)

(9) ベッド上での姿勢変換と褥瘡予防動作
0. ベッド上で上半身の向きを変えること，下半身の向きを変えること，起き上がること，および車椅子上でのプッシュアップのすべての動作に介助が必要，補助器具の要否は問わないが，電動器具は用いない．
2. 介助なくできる動作が 1 つある[3]．
4. 介助なくできる動作が 2 つまたは 3 つある．
6. ベッド上動作と除圧動作はすべて自立しておこなう．

(10) 移乗：ベッド―車椅子（車いすのブレーキ操作，フットレストの跳ね上げ，アームレストの脱着，乗り移り，足の持ち上げ）
0. 全介助を要する．
1. 部分介助，監視，および補助器具（例：スライディングボード）のすべて，もしくは，そのいずれかが必要．
2. 自立（または車椅子を必要としない）．

(11) 移乗：車椅子―トイレ，浴槽（トイレ用車椅子[4] を使用している場合は，それへの／からの乗り移り，通常の車椅子を使用している場合は，車椅子のブレーキ操作，フットレストの跳ね上げ，アームレストの脱着，乗り移り，足の持ち上げ）
0. 全介助を要する．
1. 部分介助，監視，および補助器具（例：手すり）のすべて，もしくは，そのいずれかが必要．
2. 自立（または車椅子は必要としない）．

移動 (屋内と屋外，平らな所で)

(12) 屋内の移動
0. 全介助を要する．
1. 電動車椅子を必要とするか，または手動車椅子を操作するのに部分介助を要する．
2. 手動車椅子で自立して移動する．
3. 歩行時に監視を必要とする（器具の要否は問わない）．
4. 歩行器または松葉杖で歩行（大振り・小振り歩行）．
5. 松葉杖または T 字杖 2 本で歩行（交互歩行）．
6. T 字杖 1 本で歩行．
7. 下肢装具のみを必要とする．
8. 歩行補助異なしで歩行．

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(13) まとまった距離の移動 (10〜100m)
0. 全介助を要する.
1. 電動車椅子を必要とするか, または手動車椅子を操作するのに部分介助を要する.
2. 手動車椅子で自立して移動する.
3. 歩行時に監視を必要とする(器具の要否は問わない).
4. 歩行器または松葉杖で歩行(大振り・小振り歩行).
5. 松葉杖または T 字杖 2 本で歩行(交互歩行).
6. T 字杖 1 本で歩行.
7. 下肢装具のみを必要とする.
8. 歩行補助具なしで歩行.

(14) 屋外の移動 (100m 以上)
0. 全介助を要する.
1. 電動車椅子を使用するか, または手動車椅子を操作するのに部分介助を要する.
2. 手動車椅子を自立して移動する.
3. 歩行時に監視を必要とする(器具の要否は問わない).
4. 歩行器または松葉杖で歩行(大振り・小振り歩行).
5. 松葉杖または T 字杖 2 本で歩行(交互歩行).
6. T 字杖 1 本で歩行.
7. 下肢装具のみを必要とする.
8. 歩行補助具なしで歩行.

(15) 階段昇降
0. 階段の上り下り不能.
1. 人に支えられるか, またはその監視下で少なくとも 3 段は上り下りできる.
2. 手すりにつかまったり, 松葉杖や T 字杖を用いて少なくとも 3 段は上り下りできる.
3. 支えも監視もなくで少なくとも 3 段は上り下りできる.

(16) 移乗:車椅子一車 (車に近づく, 車椅子のブレーキ操作, アームレストおよびフットレストの取り外し, 車への／からの乗り移り, 車椅子の積み降ろし)
0. 全介助を要する.
1. 部分介助, 監視, および補助器具のすべて, もしくはいずれかを必要とする.
2. 自立して移乗する, 補助器具を必要としない(または車椅子を必要としない).

(17) 移乗:床一車椅子
0. 介助を要する.
1. 補助器具の有無にかかわりなく, 自立して移乗する(または車椅子を必要としない).

小計(0-40)

SCIM 合計スコア(0-100)

訳者注:
1. 夜間の BiPAP を使用している場合も 6 点とする
2. 原著では external drainage instrument. 体外からあてるさまざま芯様尿器集尿器のことを指す. 例えば, コンドーム型集尿器 , しびんなどが含まれる. 尿道や膀胱に挿入するカテーテルは含まれない
3. ベッド柵やひもなどを使ってもよい
4. 原著では toilet wheelchair. 便器や浴槽に移乗する ことなく, 乗車したまま洋式便器が使用でき, シャワー浴にも使える車椅子のこと. 介助型と自走型がある. わが国ではトイレ兼用シャワーキャリー(介助型)が用いられることがある.

SCIM (Version III): 日本語版: 熊川博之・黒川真希子・出田良輔・里宇明元訳

190x254mm (300 x 300 DPI)

以下のそれぞれの項目の一つの四角に（このように☑）印をつけて、あなた自身の今日の健康状態を最も良く表している記述を示して下さい。

移動の程度

歩き回るのに問題はない ❑

歩き回るのにいくらか問題がある ❑

ベッド（床）に寝たきりである ❑

身の回りの管理

身の回りの管理に問題はない ❑

洗面や着替えを自分でするのにいくらか問題がある ❑

洗面や着替えを自分でできない ❑

ふだんの活動 *（例：仕事、勉強、家事、家族･余暇活動）*

ふだんの活動を行うのに問題はない ❑

ふだんの活動を行うのにいくらか問題がある ❑

ふだんの活動を行うことができない ❑

痛み／不快感

痛みや不快感はない ❑

中程度の痛みや不快感がある ❑

ひどい痛みや不快感がある ❑

不安／ふさぎ込み

不安でもふさぎ込んでもいない ❑

中程度に不安あるいはふさぎ込んでいる ❑

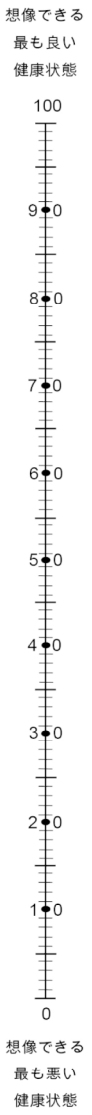
ひどく不安あるいはふさぎ込んでいる ❑

190x254mm (300 x 300 DPI)

健康状態がどのくらい良いか悪いかを表わしてもらうために、（温度計に似たような）目盛を描きました。目盛には、あなたの想像できる最も良い状態として 100、あなたの想像できる最も悪い状態として 0 が付けられています。

あなたの今日の健康状態がどのくらい良いか悪いかを、あなたの考えでこの目盛上に示して下さい。下の「あなたの今日の健康状態」と書かれた四角から、あなたの現在の健康状態の良し悪しを示す目盛上の点まで、線を引いて下さい。

あなたの
今日の
健康状態



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CONSORT 2010 checklist of information to include when reporting a randomised trial*

Section/Topic	Item No	Checklist item	Reported on page No
Title and abstract			
	1a	Identification as a randomised trial in the title	1
	1b	Structured summary of trial design, methods, results, and conclusions (for specific guidance see CONSORT for abstracts)	3
Introduction			
Background and objectives	2a	Scientific background and explanation of rationale	4-5
	2b	Specific objectives or hypotheses	6
Methods			
Trial design	3a	Description of trial design (such as parallel, factorial) including allocation ratio	6
	3b	Important changes to methods after trial commencement (such as eligibility criteria), with reasons	N/A
Participants	4a	Eligibility criteria for participants	7
	4b	Settings and locations where the data were collected	11
Interventions	5	The interventions for each group with sufficient details to allow replication, including how and when they were actually administered	6-7
Outcomes	6a	Completely defined pre-specified primary and secondary outcome measures, including how and when they were assessed	6-7
	6b	Any changes to trial outcomes after the trial commenced, with reasons	N/A
Sample size	7a	How sample size was determined	8
	7b	When applicable, explanation of any interim analyses and stopping guidelines	N/A
Randomisation:			
Sequence generation	8a	Method used to generate the random allocation sequence	6
	8b	Type of randomisation; details of any restriction (such as blocking and block size)	6
Allocation concealment mechanism	9	Mechanism used to implement the random allocation sequence (such as sequentially numbered containers), describing any steps taken to conceal the sequence until interventions were assigned	8
Implementation	10	Who generated the random allocation sequence, who enrolled participants, and who assigned participants to interventions	6
Blinding	11a	If done, who was blinded after assignment to interventions (for example, participants, care providers, those	8

		assessing outcomes) and how	
	11b	If relevant, description of the similarity of interventions	N/A
Statistical methods	12a	Statistical methods used to compare groups for primary and secondary outcomes	8-9
	12b	Methods for additional analyses, such as subgroup analyses and adjusted analyses	N/A
Results			
Participant flow (a diagram is strongly recommended)	13a	For each group, the numbers of participants who were randomly assigned, received intended treatment, and were analysed for the primary outcome	N/A
	13b	For each group, losses and exclusions after randomisation, together with reasons	N/A
Recruitment	14a	Dates defining the periods of recruitment and follow-up	N/A
	14b	Why the trial ended or was stopped	N/A
Baseline data	15	A table showing baseline demographic and clinical characteristics for each group	N/A
Numbers analysed	16	For each group, number of participants (denominator) included in each analysis and whether the analysis was by original assigned groups	N/A
Outcomes and estimation	17a	For each primary and secondary outcome, results for each group, and the estimated effect size and its precision (such as 95% confidence interval)	N/A
	17b	For binary outcomes, presentation of both absolute and relative effect sizes is recommended	N/A
Ancillary analyses	18	Results of any other analyses performed, including subgroup analyses and adjusted analyses, distinguishing pre-specified from exploratory	N/A
Harms	19	All important harms or unintended effects in each group (for specific guidance see CONSORT for harms)	N/A
Discussion			
Limitations	20	Trial limitations, addressing sources of potential bias, imprecision, and, if relevant, multiplicity of analyses	N/A
Generalisability	21	Generalisability (external validity, applicability) of the trial findings	N/A
Interpretation	22	Interpretation consistent with results, balancing benefits and harms, and considering other relevant evidence	N/A
Other information			
Registration	23	Registration number and name of trial registry	3
Protocol	24	Where the full trial protocol can be accessed, if available	3
Funding	25	Sources of funding and other support (such as supply of drugs), role of funders	11

*We strongly recommend reading this statement in conjunction with the CONSORT 2010 Explanation and Elaboration for important clarifications on all the items. If relevant, we also recommend reading CONSORT extensions for cluster randomised trials, non-inferiority and equivalence trials, non-pharmacological treatments, herbal interventions, and pragmatic trials. Additional extensions are forthcoming: for those and for up to date references relevant to this checklist, see www.consort-statement.org.

BMJ Open

Study Protocol for the G-SPIRIT trial: A Randomized, Placebo-Controlled, Double-blinded Phase 3 Trial of Granulocyte Colony Stimulating Factor-Mediated Neuroprotection for Acute Spinal Cord Injury

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Study Protocol for the G-SPIRIT trial: A Randomized, Placebo-Controlled, Double-blinded Phase 3 Trial of Granulocyte Colony Stimulating Factor-Mediated Neuroprotection for Acute Spinal Cord Injury

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Abstract

Introduction: Granulocyte Colony Stimulating Factor (G-CSF) is generally used for neutropenia. Previous experimental studies revealed that G-CSF promoted neurological recovery after spinal cord injury (SCI). Next we moved to early phase of clinical trials. In a phase 1/2a trial, no adverse events were observed. Next, we conducted a non-randomized, non-blinded, comparative trial, which suggested the efficacy of G-CSF for promoting neurological recovery. Based on those results, we are now performing a phase 3 trial.
Methods and Analysis: The objective of this study is to evaluate the efficacy of G-CSF for acute SCI. The study design is a prospective, multicenter, randomized, double-blinded, placebo-controlled comparative study. The current trial includes cervical SCI (severity of American Spinal Injury Association (ASIA) Impairment Scale B/C) within 48 hours after injury. Patients are randomly assigned to G-CSF and placebo groups. The G-CSF group is administered 400 μg/m^2/d×5d of G-CSF in normal saline via intravenous infusion for 5 consecutive days. The placebo group is similarly administered a placebo. Our primary endpoint is changes in ASIA motor scores from baseline to 3 months. Each group includes 44 patients (88 total patients).This trial is funded by the Center for Clinical Trials, Japan Medical Association.
Ethics and Dissemination: The study was approved by the Institutional Review Board of each institution and will be conducted according to the principles of the World Medical Association Declaration of Helsinki and in accordance with the Japanese Medical Research Involving Human Subjects Act and other guidelines, regulations and Acts. Results of the clinical study will be submitted to the head of the respective clinical study site as a report after conclusion of the clinical study by the sponsor-investigator. Even if the results are not favorable despite conducting the clinical study properly, the data will be published as a paper.
Registration: UMIN000018752
https://upload.umin.ac.jp/cgi-open-bin/ctr/ctr.cgi?function=brows&action=brows&type=summary&recptno=R000021694&language=J

Strengths and Limitations of This Study

Strengths

- ✓ Novel drug therapy for acute spinal cord injury is much-needed.
- ✓ Randomized, placebo-controlled, double-blinded design can eliminate bias.

Limitations

- ✓ Acute spinal cord injury patients are difficult to recruit to the trial.
- ✓ Patient's neurological status in acute phase is unstable, possibly resulting in dispersion of the patient's background.
- ✓ Concealment must be performed very strictly because G-CSF apparently increases

white blood cell count.

Introduction

Spinal cord injury (SCI) is a devastating injury by which the patient can suffer from long-lasting severe sequelae including palsy of extremities, sensory disturbance, bowel/bladder/sexual dysfunction, and neuropathic pain. Conceptually, SCI is divided into two chronological phases: a primary and a secondary phase. Primary injury is mechanical damage to spinal cord tissue itself caused by fracture and/or dislocation or compression. Secondary injury is triggered by the primary injury and is a biological reaction of the spinal cord, which includes ischemia, hemorrhage, excitotoxicity, hyperpermeability, and inflammation[1]. Because secondary injury can be the main target of treatment, extensive laboratory and clinical investigation of neuroprotection is needed to manage secondary injury[2].

To date, the only approved neuroprotective therapy for SCI is massive methylprednisolone sodium succinate (MPSS) therapy based on the NASCIS 2 study[3]. However, recent reports revealed that MPSS shows only a modest effect for SCI. In addition, several reports have described adverse events induced by MPSS for SCI including infections (pneumonia, urinary tract infection) and gastrointestinal disorders (gastric ulcer, etc.)[4]. Therefore, the use of MPSS for SCI has become controversial[5]. Accordingly, a new therapeutic drug for SCI is desirable.

Granulocyte colony-stimulating factor (G-CSF, generic name: filgrastim) is a growth factor that affects the hematopoietic system, promoting differentiation, proliferation, and survival of granulocytes[6]. Clinically, in Japan, G-CSF is administered to patients with leukopenia, and to peripheral stem cell transplantation donors, G-CSF is administered to mobilize hematopoietic stem cells into the peripheral blood[7]. In the central nervous system, G-CSF has properties to mobilize the movement of bone marrow cells into the brain[8] and spine, and in a stroke model, has shown neuroprotective properties[9]. In other countries, clinical studies of the effects of G-CSF in cerebral infarction have been reported[10].

To prove the hypothesis that G-CSF has neuroprotective properties against SCI, G-CSF was administered to rat and mouse animal models of spinal cord injury, and hind limb

function improved significantly after administration of G-CSF. Further investigations into the mechanism of action of G-CSF in SCI were conducted. Data obtained to date identify the following properties of G-CSF: [1] mobilization of bone marrow-derived stem cells causing their biointegration at the site of SCI[11], [2] direct inhibition of nerve cell death[12], [3] protection of the myelin sheath by inhibiting oligodendrocyte cell death[13], [4] inhibition of inflammatory cytokine expression (TNF-α, IL-1β)[13], and [5] promotion of neovascularization[14]. These properties suggest that G-CSF has a neuroprotective effect in acute SCI.

Based on these properties, a phase 1/2a clinical study was conducted where the main objective was to confirm the safety and feasibility of G-CSF for treatment of patients with acute SCI[15]. This study was an open-label, dose-titrating study with no control group. As the initial step, 5 patients were given 5 μg/kg/day of G-CSF for 5 consecutive days by i.v. infusion, and as the second step, 11 patients were given 10 μg/kg/day of G-CSF for 5 consecutive days by i.v. infusion. No serious adverse events were noted, and the safety of G-CSF administration in patients with acute SCI was confirmed[15].

As a next step, to validate the efficacy of G-CSF in neuroprotective treatment, a multicenter, prospective, nonrandomized, nonblinded, comparative control study (phase 2b clinical trial) was conducted[16]. Based on the results of the previous phase 1/2a clinical trial, the dosage and duration of treatment with G-CSF in this study was 10 μg/kg/day for 5 consecutive days. Patients with acute cervical SCI (within 48 h of injury) were registered in the clinical trial and allocated to either the G-CSF group (G-CSF 10 μg/kg/day × 5 days i.v. infusion) or control group (no G-CSF administration) at each treatment facility. A total of 19 patients in the G-CSF group and 26 patients in the control group were observed for 3 months or longer. American Spinal Injury Association (ASIA) motor score (motor: 0 to 100 points) was compared between the groups. ASIA motor scores were 26.1 ± 18.9 in the G-CSF group and 12.2 ± 14.7 in the control group showing a significant improvement of motor paralysis in the G-CSF group ($p < 0.01$). In addition, in cases that could be followed for 1 year or longer, a significant improvement of ASIA motor score was observed in the G-CSF group[16].

Based on results of these preclinical and early phase clinical trials, we are now conducting a phase 3 trial.

Specific Objective

The objective of this study is to evaluate the efficacy of G-CSF for improving motor paralysis in acute SCI.

Methods and Analysis

Design of the study

The study design of the current trial is a prospective, multicenter, randomized, double-blinded, placebo-controlled comparative study.

Study Procedures

The study outline is shown in Figure. Patients will be randomly assigned to G-CSF and placebo groups. A central registration system will be used for dynamic randomization into the investigational treatment group (G-CSF) and control group (placebo) based on age at registration (16–64 years of age, or 65–84 years of age) and severity of paralysis (AIS B or C) at 48 h after injury. With or without the surgical stabilization and/or decompression was not included in stratification. Initially, screened patients with severity AIS B/C will be tentatively enrolled. Initial screening of the patients include clinical laboratory test, imaging studies including X-ray, magnetic resonance imaging and computed tomography, and neurological/functional evaluations.

Neurological re-assessment will be performed 48±4 hours after SCI and the patients who will recover to severity AIS D will be excluded. The patients with severity AIS B/C at neurological re-assessment 48±4 hours after SCI will be enrolled and randomly allocated to either the investigational treatment group (G-CSF) or control group (placebo) in a 1 to 1 ratio. The subject registration center uses a program based on an appropriate computer algorithm to allocate patients into groups. The first dose of investigative drugs will be administered to the patients after re-assessment of neurological status and enrollment 48±4

hours after SCI.

The G-CSF group will be administered 400 μg/m^2/d × 5d of G-CSF in normal saline via intravenous infusion for 5 consecutive days. The placebo group will be similarly administered a placebo. The first dose of investigative drugs will be administered to the patients 48±4 hours after SCI. The dosing schedule will be once a day in every morning (9-10 AM) for consecutive 5 days even in case that the first dosing was performed at night because of restriction on practices. Investigational drugs including both G-CSF and placebo will be stored in refrigerator which will be kept between 1- 6□ and has temperature-logger in pharmacies in each participating institutes. The investigational drugs is packaged in ample with label only printed as serial numbers, ten ample is packed in one box with label only printed serial numbers. Web-based allocation system will show the serial number of investigational drug which must be used to respective patients to ensure blinding.

We decided dosage of drug based on our previous non-randomized early phase clinical studies. On phase 1/2a clinical study, of which study design was open-label, dose-titrating study with no control group, SCI patients who were administered 10 μg/kg/day G-CSF for 5 days showed marked elevation of white blood cell number (reached nearly 50000/μl, of which WBC number might cause adverse effects of G-CSF) during G-CSF administration[15]. Therefore we decided to withdraw additional titration. In addition, next phase of clinical study, of which design was multicenter, prospective, nonrandomized, nonblinded, comparative control study, showed suggestive efficacy of G-CSF (10 μg/kg/d for 5 days) for acute SCI[16]. Based on those results, we finally decided the dosage of G-CSF as 10 μg/kg/d × 5d for 5 days. In the current clinical trial, the dosage of G-CSF is written as 400 μg/m^2/d (= 10 μg/kg/d) according to the Japanese Pharmaceutical and Medical Device Agency (PMDA)'s instruction for consolidation with product labeling.

We decided to 48hours after SCI as the therapeutic time window because our previous multicenter, prospective, nonrandomized, nonblinded, comparative control study, which recruited SCI patients within 48 hours after injury, showed that there was no significant difference in neurological outcome between the patient administered G-CSF very early after the injury and 48 hours after injury[16].

Allocation will be concealed between blinded evaluators of efficacy/safety and those for laboratory data, as G-CSF markedly increases white blood cell counts that can reveal patient treatment.

Our primary endpoint is changes in ASIA motor scores (international standards for neurological classification of spinal cord injury (ISNSCI) suppl. Figure 1) from baseline to 3 months calculated as follows: 3-month ASIA motor score change = 3-month ASIA motor score – pretreatment ASIA motor score. To maintain consistency of neurological assessment among the each evaluators, attending lecture and e-learning (in website of International Spinal Cord Society: http://www.elearnsci.org/) of ASIA/ISNSCI scoring system is mandatory for every investigators participating to the present trial.

Secondary endpoints are as follows. [1] Change in ASIA motor scores at 6 months and 12 months after G-CSF administration compared with pretreatment. [2] Changes in sensory paralysis over time: change in ASIA sensory scores at 3 months, 6 months, and 12 months after G-CSF administration compared with pretreatment. [3] Severity of functional compromise because of paralysis: AIS before administration and at 3, 6, and 12 months after administration of G-CSF. [4] Percentage of responders: percentage of patients whose AIS improved by 1 grade or more at 3, 6, and 12 months after administration compared with before administration of G-CSF. [5] Neurological level of injury (NLI): percentage of patients whose NLI decreased by 1 grade of more at 3, 6, or 12 months after administration of G-CSF compared with pretreatment. [6] SCIM: change in SCIM scores at 3, 6, and 12 months after G-CSF administration compared with pretreatment [7] EQ-5D: measured EQ-5D efficacy scores at 3, 6, and 12 months after G-CSF.

To strengthen the analysis, more strict blindness of assessment by the evaluator for outcome measures is ideal. Therefore every prior score/measurement should be blinded and/or not the same person should assess every midpoint motor/sensory functional measurement. However, PMDA instructed us to assess one patients' functional evaluation by one evaluator for consistent assessment in respective patients. PMDA considers the consistency of assessment in respective patients is more important.

The precise explanation of abovementioned outcome measures is as followings (suppl.

Figures). ASIA motor score is determined by examining a key muscle function within each of 10 myotomes on each side of the body on manual muscle testing (MMT) in the supine position. There is a range of score from 0 to 25 for each extremity, totaling 0 to 50 for the upper limbs and 0 to 50 for the lower limbs, resulting in total of ASIA motor score ranges from 0 to 100. This score can reflect the degree of motor impairment associated with the SCI (suppl. Figure 1). ASIA sensory scores is completed through the testing of a key point in each of the 28 dermatomes (from C2 to S4-5) on the right and left sides of the body that can be readily located in relation to bony anatomical landmarks. At each of these key points, two aspects of sensation are examined: light touch and pin prick. Appreciation of light touch and pin prick sensation at each of the key points is separately scored on a three-point scale, with comparison to the sensation on the patients' cheek as a normal frame of reference: 0 = absent, 1= altered, 2 = normal or intact. As a result, ASIA sensory scores range from 0 to 112 (suppl. Figure 1). AIS designation is used in grading the degree of impairment: A = Complete. No sensory or motor function is preserved in the sacral segments S4-S5. B = Sensory incomplete. Sensory but not motor function is preserved below the neurological level. C = Motor incomplete. Motor function is preserved below the neurological level, and more than half of key muscle functions below the single neurological level of injury have a muscle grade less than 3 (Grades 0–2). D = Motor incomplete. Motor function is preserved below the neurological level, and at least half (half or more) of key muscle functions below the neurological level have a muscle grade >3. E = Normal. If sensation and motor function as tested are graded as normal in all segments, and the patient had prior deficits, then the AIS grade is E (suppl. Figure 1, footnote). NLI refers to the most caudal segment of the cord with intact sensation and antigravity muscle function strength, provided that there is normal (intact) sensory and motor function rostrally (suppl. Figure 1, footnote). SCIM is disability scale developed specifically for patients with spinal cord lesions in order to make the functional assessments of patients with paraplegia or tetraplegia more sensitive to changes. The SCIM includes the following areas of function: self-care (subscore (0-20), respiration and sphincter management (0-40) and mobility (0-40). Each area is scored according to its proportional weight in these patients'

general activity. The final score ranges from 0 to 100 (suppl. Figure 2-4). EQ-5D essentially consists of 2 pages (suppl. Figure 5, 6): the EQ-5D descriptive system and the EQ visual analogue scale (EQ VAS). The descriptive system comprises five dimensions: mobility, self-care, usual activities, pain/discomfort and anxiety/depression. Each dimension has 5 levels: no problems, slight problems, moderate problems, severe problems and extreme problems. The patient is asked to indicate his/her health state by ticking the box next to the most appropriate statement in each of the five dimensions. This decision results in a 1-digit number that expresses the level selected for that dimension. The digits for the five dimensions can be combined into a 5-digit number that describes the patient's health state and can be converted to efficacy score with calibration scale (ranges -0.111 to 1.000). The EQ VAS records the patient's self-rated health on a vertical visual analogue scale, where the endpoints are labelled 'The best health you can imagine' and 'The worst health you can imagine'. The VAS can be used as a quantitative measure of health outcome that reflect the patient's own judgement.

Each group will include 44 patients (88 patients in total). Our protocol was approved by PMDA and this trial will be funded by the Center for Clinical Trials, Japan Medical Association.

Inclusion

Inclusion criteria are as follows. [1] Patients with cervical SCI (severity of AIS B/C) within 48 h of injury. [2] Patients reassessed for neurological status at 48 h after injury, and those whose palsy is AIS B/C will be enrolled. [3] Patients with neurological level of injury between C4 and C7. [4] Patients with age of 16 to 85 years. [5] Patients who agree to participate in the current trial and from whom informed consent was obtained orally and in writing. [6] Patients who can be followed up for 12 months after SCI.

Exclusion

Exclusion criteria are as follows. [1] Patients with neurological recovery to AIS D at

neurological reassessment 48 h after SCI, because only AIS B/C patients will be included to standardize the severity of paresis in order to stratify the patients at the initiation of drug administration. [2] Allergy to G-CSF. [3] Hematological malignancy, [4] within 6 months after invasive coronary intervention, [5] splenomegaly, or [6] pregnancy, [7] consciousness impairment, [8] neurological disorders that can affect neurological evaluation in the present trial, [9] fracture of extremities that can affect the neurological evaluation and [10] massive dose administration of MPSS. Exclusion criteria 2 - 6 are set for safety, criteria 1, 7 - 9 are set to maintain homogeneity of the patient population enrolled, criteria 7-9 are set for maintenance of accuracy of functional assessment, criteria 7 is set to obtain patients' own informed consent upon participation to the trial and criteria 10 is set to omit the possible interference of MPSS on outcome assessment.

Concealment

Patients will be administered drug or placebo and be evaluated in a double-blinded manner. An evaluator blinded to the treatment will take charge of patient evaluation including clinical findings and paresis, without laboratory data because G-CSF induces an apparent increase of white blood cell count that makes it easy to distinguish G-CSF and placebo treatment. Therefore, a non-blinded evaluator for laboratory data will be assigned to evaluate laboratory data alone. From a safety point-of view, the dose of G-CSF will be modulated according to excessive increase of white blood cell count. Therefore, a non-blinded evaluator will instruct the pharmacy to modulate the dose of G-CSF based on white blood cell count.

Sample Size Calculation

The target sample size for this randomized trial is 88. This number is based on the results of previous clinical trials[16]. The estimated group difference (± standard deviation) of change in ASIA motor scores from baseline to 3 months is 13.9 (± 21.9). A sample size of 44 patients in each group will provide 80% power to detect a difference of the change in ASIA motor scores between the G-CSF and the placebo treatments, using a mixed-effects

models for repeated measures (MMRM) at a two-sided 5% level of significance. A common correlation of 0.25 at each time point is assumed. A dropout rate of 10% is allowed. Thus, the total sample size of 88 patients is required for the trial.

Statistical analyses

The analyses of the primary and secondary end points will be performed as intention-to-treat analyses in a full analysis set, which includes all patients who: (1) took at least one course of treatment during the study; (2) do not present any serious violation of the study protocol; and (3) have data collected after commencement of treatment. For the baseline characteristics, summary statistics will comprise frequencies and proportions for categorical variables, and means and SDs for continuous variables. The patient characteristics will be compared using a chi-square test for categorical variables, and a *t* test or Wilcoxon rank sum test for continuous variables. Missing data including loss to follow-up and missing measurement will be supplemented with MMRM.

For the primary analysis, aimed at comparing treatment effects, a change in ASIA motor score from baseline to 3 months and its 95% confidence interval (CI) will be estimated using the MMRM. To test for significant association of the primary endpoint, a mixed effects model for repeated measures with an unstructured covariance matrix will be applied to adjust for age (<65 years or ≥65 years) and AIS at 48 h after the injury (B or C).

For the secondary analysis, the change in ASIA motor score will be compared using a Student *t* test and the 95% CI will be estimated. The same method will be applied to change in sensory score, SCIM, and EQ-5D. A chi-squared test will be applied to the frequencies of the responder in AIS and of the improvement in NLI. The frequency of AIS will be summarized. The frequency of AEs will be compared using a Fisher exact test.

All comparisons are planned and all *p* values will be two sided. $p < 0.05$ will be considered significant. All statistical analyses will be performed using SAS software (version 9.4; SAS Institute, Cary, NC, USA). The statistical analysis plan will be developed by the principal investigator and the biostatistician before completion of patient recruitment and fixing of data.

Data Monitoring Committee

The data monitoring committee consists of three clinical trial specialists, including a biostatistician, who are independent from the current study. The committee will meet at least 2 times per year and all the data obtained by the current trial will be checked by the committee.

Adverse Events

As for safety evaluation, adverse events will be collected as follows. “Adverse events” refers to any untoward symptom or disease or signs of such (including clinical laboratory data abnormalities) in a clinical investigation subject after informed consent and does not necessarily have a causal relationship with the investigational product (G-CSF).

Increases in white blood counts will be considered an adverse event only when the count exceeds 50,000/μL from the perspective of a pharmacological effect of G-CSF, and any values below this will not be handled as an adverse event. Anaphylaxis and adult respiratory distress syndrome are the most representative G-CSF-related severe adverse events to be payed full attention.

All adverse events will code terminology used by the investigators according to the ICH International Medical Dictionary for Regulatory Activities Japanese version (MedDRA/J).

Ethics and Dissemination

Ethics

The study was approved by the Institutional Review Board (IRB) of each institutions involved to the present trial and will be conducted according to the principles of the World Medical Association (WMA) Declaration of Helsinki—Ethical Principles for Medical Research Involving Human Subjects with the amendments made in Seoul, South Korea, October 2008, with a Note of Clarification on Paragraph 29 added by the WMA General Assembly, Washington 2002; Note of Clarification on Paragraph 30 added by the WMA General Assembly, Tokyo 2004, and in accordance with the Japanese Medical Research

Involving Human Subjects Act (WMO) and other guidelines, regulations and Acts.

Patient informed consent

The principal investigator will try to prepare the informed consent form and other explanatory materials in as simple language as possible in order to obtain informed consent from the patient and the patient's legal representative. In case that trial participants cannot sign the informed consent form due to upper extremity palsy caused by SCI, allograph by patients' representative will be allowed.

Public Disclosure and Publication Policy

Results of the clinical study will be submitted to the head of the respective clinical study site as a report after conclusion of the clinical study by the sponsor-investigator (includes study coordinating investigator). Even if the results are not favorable despite conducting the clinical study properly, the data will be published as a paper. Other sponsor-investigators (including the clinical study coordinating investigator), if they plan to publicize the data from this study in a specialized academic society conference or other external site, must first obtain permission from the other principal investigators and investigational product provider. In publicizing the results, the confidentiality of the subjects will be maintained and proofread in advance by the other sponsor-investigators (includes coordinating investigator) and investigational product provider.

Discussion

The current trial is a confirmative trial to elucidate the therapeutic efficacy of G-CSF for SCI. If the current trial can successfully show significant improvement of motor paralysis of SCI by G-CSF, we will move forward to drug approval application to the Ministry for Health and Labor, Japan. The entire protocol of the current trial was approved beforehand for the initiation of the current trial by the Japanese Pharmaceutical and Medical Device Agency (PMDA). The PMDA will also permit a drug approval application if significant efficacy of G-CSF for SCI is proven.

The current trial is an important milestone for SCI clinics and research to explore G-CSF for SCI.

Trial Status

The present trial is now on-going.

Trial Sites

Nineteen major hospitals in Japan constituting the G-SPIRIT study group as follows: Tohoku University Hospital, Miyagi; Niigata University, Niigata; Dokkyo University Hospital, Tochigi; Tsukuba University Hospital, Ibaraki; Tsukuba Medical Center, Ibaraki; Chiba University Hospital, Chiba; Funabashi Municipal Medical Center, Chiba; Kimitsu Chuo Hospital, Chiba; Chiba Rosai Hospital, Chiba; Tokai University Hospital, Kanagawa; Hamamatsu University Hospital, Shizuoka; Gifu University Hospital, Gifu; Chubu Rosai Hospital, Aichi; Mie University Hospital, Mie; Kanazawa Medical Collage, Ishikawa; Kobe Red Cross Hospital, Hyogo; Hiroshima University Hospital, Hiroshima; Yamaguchi University Hospital, Yamaguchi; and Nagasaki Rosai Hospital, Nagasaki.

Funding Statement

This trial is supported by Center for Clinical trials, Japan Medical Association, Japan.

Competing Interests

No authors have any competing interests to declare.

Contributorship Statement

MK, TF and MY contributed to planning and conduct of the present trial and to reporting the present protocol paper. HH, TS, YF, MH and ST contributed to conception and design of the present trial protocol and to reporting the present protocol paper. YI, JS, MK, SO, YM, TA, KW, TH, MO, HS, TM, IT, NK, MK. YO, TS, MY, MF, KY, DS, HT, DT, SI, HM, HU, FA, YS, IA, YT, MM, JS, YS, SI, HS, KT, YI, FN, MH, TO, FH, TF, KK, MW, HK, YM, YY, DT, TH, SK, GY, SO, TB, HA, KA, EK, HI, TS, AS, YI, TK, SO, NT, KN, NK, SK, HB, TO, HK, TY, IK, KY, TM, TM, IM, HK, TA, KH, TE, TS, MM, KF, SN, CI,

TT, TK, HS, NN and MF contributed to conducting the present trial and to acquisition of data.

References

1. Oyinbo CA. Secondary injury mechanisms in traumatic spinal cord injury: a nugget of multiply cascade. ***Acta Neurobiol Exp*** 2011;71:281-299.
2. Karsy M, Hawryluk G. Pharmacologic Management of Acute Spinal Cord Injury. ***Neurosurg Clin N Am*** 2017;28:49-62. doi: 10.1016/j.nec.2016.07.002.
3. Bracken MB, Shepard MJ, Collins WF, et al. A randomized controlled trial of methylprednisolone or naloxone in the treatment of acute spinal cord injury: results of the second national acute spinal cord injury study. ***N Engl J Med*** 1990;322: 1405-1411.
4. Matsumoto T, Tamaki T, Kawakami M, et al. Early complications of high-dose metyl-prednisolone sodium succinate treatment in the follow-up of acute cervical spinal cord injury. ***Spine*** 2001;26: 426-430.
5. Hurlbert RJ, Hadley, Walters BC, et al. Pharmacological Therapy for Acute Spinal cord Injury. ***Neurosurgery*** 2013;72: 93-105.
6. Nicola NA, Metcalf D, Matsumoto M, Johnson GR. Purification of a factor inducing differentiation in murine myelomonocytic leukemia cells. Identification as granulocyte colony-stimulating factor. ***J Biol Chem*** 1983;258: 9017-9023.
7. Roberts AW: G-CSF: a key regulator of neutrophil production, but that's no all! ***Growth Factors*** 2005;23: 33–41.
8. Kawada H, Takizawa S, Takanashi T, et al. Administration of hematopoietic cytokines in the subacute phase after cerebral infarction is effective for functional recovery facilitating proliferation of intrinsic neural stem/progenitor cells and transition of bone marrow-derived neuronal cells. ***Circulation*** 2006;113: 701-710.
9. Schneider A, Kruger C, Steigleder T, et al. The hematopoietic factor G-CSF is a neuronal ligand that counteracts programmed cell death and drives neurogenesis. ***J Clin Invest*** 2005;115: 2083-2098.

10. Shyu WC, Lin SZ, Lee CC, Liu DD, Li H. Granulocyte colony-stimulating factor for acute ischemic stroke: a randomized controlled trial. ***CMAJ*** 2006;174: 927-933.

11. Koda M, Nishio Y, Kamada T, et al. Granulocyte colony-stimulating factor（G-CSF）mobilizes bone marrow-derived cells into injured spinal cord and promotes functional recovery after compression-induced spinal cord injury in mice. ***Brain Res*** 2007;1149: 223-231.

12. Nishio Y, Koda M, Kamada T, et al. Granulocyte colony-stimulating factor（G-CSF）attenuates neuronal death and promotes functional recovery after spinal cord injury in mice. ***J Neuropathol Exp Neurol*** 2007;66: 724-731.

13. Kadota R, Koda M, Kawabe J, et al. Granulocyte colony-stimulating factor (G-CSF) protects oligodendrocyte and promotes hindlimb functional recovery after spinal cord injury in rats. ***PLoS One*** 2012;7:e50391.

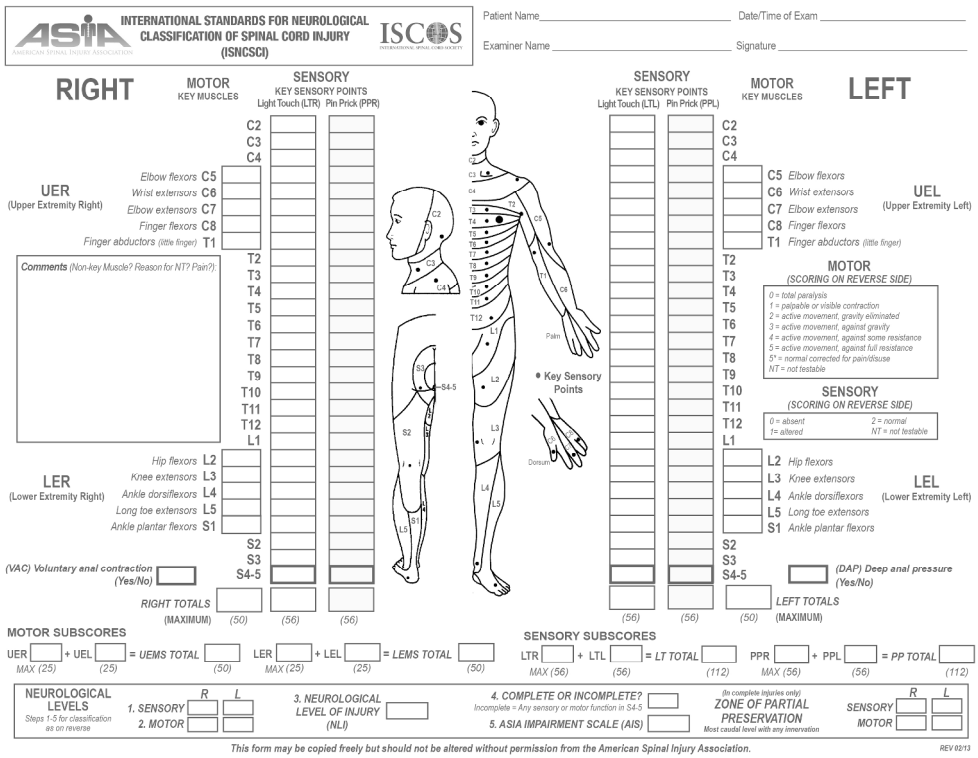
14. Kawabe J, Koda M, Hashimoto M, et al. Granulocyte colony-stimulating factor（G-CSF）exerts neuroprotective effects via promoting angiogenesis after spinal cord injury in rats. ***J Neurosurg Spine*** 2011;15: 414-421.

15. Takahashi H, Yamazaki M, Okawa A, et al. Neuroprotective therapy using granulocyte colony-stimulating factor for acute spinal cord injury: a phase I/IIa clinical trial. ***Eur Spine J*** 2012;21:2580-2587.

16. Inada T, Takahashi H, Yamazaki M, et al. A multicenter prospective non-randomized controlled clinical trial to prove neurotherapeutic effects of granulocyte colony-stimulating factor（G-CSF）for acute spinal cord injury: Analyses of follow-up cases after at least one year. ***Spine*** 2014;39:213-219.

Figure legend

Schematic diagram showing trial timeline.



274x206mm (300 x 300 DPI)

セルフケア

(1) 食事 (切る, 容器を開ける, 飲み物を注ぐ, 食べ物を口に運ぶ, 飲み物の入ったコップをもつ)
- 0. 静脈栄養または胃瘻が必要である. あるいは経口摂取において全介助を要する.
- 1. 食べたり飲んだりすること, または補助器具の装着に部分介助を要する.
- 2. 食べることは自立, 補助器具を必要とするか, または食べ物を切ったり注いだりすること, 容器の開閉にのみ解介助を要する.
- 3. 食べることも飲むことも自立, 介助や補助器具を必要としない.

(2) 入浴 (頭と体を石鹸でこすって洗い, 乾かす, 蛇口の開け閉め) A —上半身, B —下半身

A. 0. 全介助を要する.
- 1. 部分介助を要する.
- 2. 補助器具, または特定の環境 (例えば, 手すりや椅子) が整っていれば, 自立して体を洗う.
- 3. 自立して体を洗う. (通常, 健常者なら用いることのない) 補助器具や特定の環境設定は不要

B. 0. 全介助を要する.
- 1. 部分介助を要する.
- 2. 補助器具, または特定の環境が整っていれば, 自立して体を洗う.
- 3. 自立して体を洗う. 補助器具や特定の環境設定は不要

(3) 更衣 (衣服, 靴, 常用している装具の着脱) A —上半身, B —下半身

A. 0. 全介助を要する.
- 1. ボタン, ファスナー, ひものない衣服で部分介助を要する.
- 2. ボタン, ファスナー, ひものない衣服であれば自立, 補助器具と特定の環境設定, またはそのどちらかが必要.
- 3. ボタン, ファスナー, ひものない衣服であれば自立, 補助器具も特定の環境設定も不要. ボタン, ファスナー, ひもの扱いにだけ介助, 補助器具, または特定の環境設定が必要.
- 4. (衣類の種類を問わず) 自立して着替える. 補助器具も特定の環境設定も不要.

B. 0. 全介助を要する.
- 1. ボタン, ファスナー, ひものない衣服で部分介助を要する.
- 2. ボタン, ファスナー, ひものない衣服であれば自立, 補助器具と特定の環境設定, またはそのどちらかが必要.
- 3. ボタン, ファスナー, ひものない衣服であれば自立, 補助器具も特定の環境設定も不要. ボタン, ファスナー, ひもの扱いにだけ介助, 補助器具, または特定の環境設定が必要.
- 4. (衣類の種類を問わず) 自立して着替える. 補助器具も特定の環境設定も不要.

(4) 整容 (手洗い, 洗顔, 歯磨き, 整髪, 髭剃り, 化粧)
- 0. 全介助を要する.
- 1. 部分介助を要する.
- 2. 補助器具があれば自立して整容動作をおこなう.
- 3. 補助器具を用いずに自立して整容動作をおこなう.

小計 (0–20)

呼吸と排泄管理

(5) 呼吸
- 0. 気管チューブと持続的補助換気または間欠的補助換気が必要.
- 2. 気管チューブを装用して自己呼吸, 酸素を必要としたり, 咳嗽時または気管チューブの管理に多大な介助を必要とする.
- 4. 気管チューブを装用して自己呼吸, 咳嗽時または気管チューブの管理に少ししか介助を必要としない.
- 6. 気管チューブなしで自己呼吸, 酸素, 咳嗽時の多大な介助, マスク (例 : 終末呼気陽圧 PEEP), または間欠的補助換気 (BiPAP) を必要とする.
- 8. 気管チューブなしで自己呼吸, 介助または咳嗽刺激を少ししか必要としない.
- 10. 介助も器具もなしで自己呼吸.

190x254mm (300 x 300 DPI)

(6) 排尿管理
0. 留置カテーテル
3. 残尿量＞100cc，不定期導原または介助による間欠的導尿．
6. 残尿量＞100cc または間欠的自己導尿，集尿器[2] をあてがう際の介助は必要．
9. 間欠的自己導尿，集尿器を使用する．器具をあてがう際の介助は不要．
11. 間欠的自己導尿，導尿と導尿の間には失禁なし，集尿器は使用しない．
13. 残尿量＜100cc，集尿器のみ必要，集尿器の取扱いには介助不要．
15. 残尿量＜100cc，失禁なし，集尿器を使用しない．

(7) 排便管理
0. 排便が不規則またはごく低頻度（3 日に 1 回未満）．
5. 規則的ながら，（座薬を挿入するなどに）介助を要する，失敗はまれ（月 2 回未満）．
8. 規則的な排便で，介助を要しない，失敗はまれ（月 2 回未満）．
10. 規則的な排便で，介助を要しない，失敗はなし．

(8) トイレの使用（会陰部の清潔，使用前後での衣服の扱い，ナプキンまたはおむつの使用）
0. 全介助を要する．
1. 部分介助を要する．自分でお尻を拭けない．
2. 部分介助を要する．自立してお尻を拭ける．
4. 自立してトイレを使用するも，補助器具または特別な環境（例：手すり）が整っている必要あり．
5. 自立してトイレを使用．補助器具も特別な環境が整っている必要もない．

小計（0–40）

移動 (室内とトイレ)

(9) ベッド上での姿勢変換と褥瘡予防動作
0. ベッド上で上半身の向きを変えること，下半身の向きを変えること，起き上がること，および車椅子上でのプッシュアップのすべての動作に介助が必要，補助器具の要否は問わないが，電動器具は用いない．
2. 介助なくできる動作が 1 つある[3]．
4. 介助なくできる動作が 2 つまたは 3 つある．
6. ベッド上動作と除圧動作はすべて自立しておこなう．

(10) 移乗：ベッド―車椅子（車いすのブレーキ操作，フットレストの跳ね上げ，アームレストの脱着，乗り移り，足の持ち上げ）
0. 全介助を要する．
1. 部分介助，監視，および補助器具（例：スライディングボード）のすべて，もしくは，そのいずれかが必要．
2. 自立（または車椅子を必要としない）．

(11) 移乗：車椅子―トイレ，浴槽（トイレ用車椅子[4] を使用している場合は，それへの／からの乗り移り，通常の車椅子を使用している場合は，車椅子のブレーキ操作，フットレストの跳ね上げ，アームレストの脱着，乗り移り，足の持ち上げ）
0. 全介助を要する．
1. 部分介助，監視，および補助器具（例：手すり）のすべて，もしくは，そのいずれかが必要．
2. 自立（または車椅子は必要としない）．

移動 (屋内と屋外，平らな所で)

(12) 屋内の移動
0. 全介助を要する．
1. 電動車碕子を必要とするか，または手動車椅子を操作するのに部分介助を要する．
2. 手動車椅子で自立して移動する．
3. 歩行時に監視を必要とする（器具の要否は問わない）．
4. 歩行器または松葉杖で歩行（大振り・小振り歩行）．
5. 松葉杖または T 字杖 2 本で歩行（交互歩行）．
6. T 字杖 1 本で歩行．
7. 下肢装具のみを必要とする．
8. 歩行補助異なしで歩行．

190x254mm (300 x 300 DPI)

(13) まとまった距離の移動 (10〜100m)
0. 全介助を要する.
1. 電動車椅子を必要とするか, または手動車椅子を操作するのに部分介助を要する.
2. 手動車椅子で自立して移動する.
3. 歩行時に監視を必要とする(器具の要否は問わない).
4. 歩行器または松葉杖で歩行(大振り・小振り歩行).
5. 松葉杖または T 字杖 2 本で歩行(交互歩行).
6. T 字杖 1 本で歩行.
7. 下肢装具のみを必要とする.
8. 歩行補助具なしで歩行.

(14) 屋外の移動 (100m 以上)
0. 全介助を要する.
1. 電動車椅子を使用するか, または手動車椅子を操作するのに部分介助を要する.
2. 手動車椅子を自立して移動する.
3. 歩行時に監視を必要とする(器具の要否は問わない).
4. 歩行器または松葉杖で歩行(大振り・小振り歩行).
5. 松葉杖または T 字杖 2 本で歩行(交互歩行).
6. T 字杖 1 本で歩行.
7. 下肢装具のみを必要とする.
8. 歩行補助具なしで歩行.

(15) 階段昇降
0. 階段の上り下り不能.
1. 人に支えられるか, またはその監視下で少なくとも 3 段は上り下りできる.
2. 手すりにつかまったり, 松葉杖や T 字杖を用いて少なくとも 3 段は上り下りできる.
3. 支えも監視もなくで少なくとも 3 段は上り下りできる.

(16) 移乗:車椅子一車 (車に近づく, 車椅子のブレーキ操作, アームレストおよびフットレストの取り外し, 車への／からの乗り移り, 車椅子の積み降ろし)
0. 全介助を要する.
1. 部分介助, 監視, および補助器具のすべて, もしくはいずれかを必要とする.
2. 自立して移乗する, 補助器具を必要としない(または車椅子を必要としない).

(17) 移乗:床一車椅子
0. 介助を要する.
1. 補助器具の有無にかかわりなく, 自立して移乗する(または車椅子を必要としない).

小計(0-40)

SCIM 合計スコア(0-100)

訳者注:
1. 夜間のみ BiPAP を使用している場合も 6 点とする
2. 原著では external drainage instrument. 体外からあてるさまざま芯様尿器集尿器のことを指す. 例えば, コンドーム型集尿器 , しびんなどが含まれる. 尿道や膀胱に挿入するカテーテルは含まれない
3. ベッド柵やひもなどを使ってもよい
4. 原著では toilet wheelchair. 便器や浴槽に移乗する ことなく, 乗車したまま洋式便器が使用でき, シャワー浴にも使える車椅子のこと. 介助型と自走型がある. わが国ではトイレ兼用シャワーキャリー(介助型)が用いられることがある.

SCIM (Version III): 日本語版. 岡川惠之・黒川真希子・出田良輔・里宇明元訳
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190x254mm (300 x 300 DPI)

以下のそれぞれの項目の一つの四角に（このように☑）印をつけて、あなた自身の今日の健康状態を最も良く表している記述を示して下さい。

移動の程度

歩き回るのに問題はない ❑
歩き回るのにいくらか問題がある ❑
ベッド（床）に寝たきりである ❑

身の回りの管理

身の回りの管理に問題はない ❑
洗面や着替えを自分でするのにいくらか問題がある ❑
洗面や着替えを自分でできない ❑

ふだんの活動 *（例：仕事、勉強、家事、家族･余暇活動）*

ふだんの活動を行うのに問題はない ❑
ふだんの活動を行うのにいくらか問題がある ❑
ふだんの活動を行うことができない ❑

痛み／不快感

痛みや不快感はない ❑
中程度の痛みや不快感がある ❑
ひどい痛みや不快感がある ❑

不安／ふさぎ込み

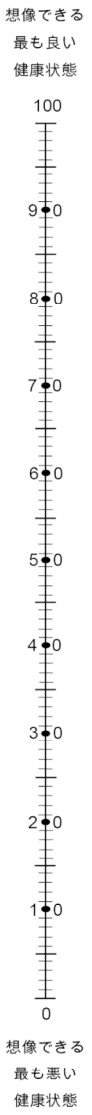
不安でもふさぎ込んでもいない ❑
中程度に不安あるいはふさぎ込んでいる ❑
ひどく不安あるいはふさぎ込んでいる ❑

190x254mm (300 x 300 DPI)

健康状態がどのくらい良いか悪いかを表わしてもらうために、（温度計に似たような）目盛を描きました。目盛には、あなたの想像できる最も良い状態として 100、あなたの想像できる最も悪い状態として 0 が付けられています。

あなたの今日の健康状態がどのくらい良いか悪いかを、あなたの考えでこの目盛上に示して下さい。下の「あなたの今日の健康状態」と書かれた四角から、あなたの現在の健康状態の良し悪しを示す目盛上の点まで、線を引いて下さい。

あなたの
今日の
健康状態



190x254mm (300 x 300 DPI)

CONSORT 2010 checklist of information to include when reporting a randomised trial*

Section/Topic	Item No	Checklist item	Reported on page No
Title and abstract			
	1a	Identification as a randomised trial in the title	1
	1b	Structured summary of trial design, methods, results, and conclusions (for specific guidance see CONSORT for abstracts)	3
Introduction			
Background and objectives	2a	Scientific background and explanation of rationale	4-5
	2b	Specific objectives or hypotheses	6
Methods			
Trial design	3a	Description of trial design (such as parallel, factorial) including allocation ratio	6
	3b	Important changes to methods after trial commencement (such as eligibility criteria), with reasons	N/A
Participants	4a	Eligibility criteria for participants	7
	4b	Settings and locations where the data were collected	11
Interventions	5	The interventions for each group with sufficient details to allow replication, including how and when they were actually administered	6-7
Outcomes	6a	Completely defined pre-specified primary and secondary outcome measures, including how and when they were assessed	6-7
	6b	Any changes to trial outcomes after the trial commenced, with reasons	N/A
Sample size	7a	How sample size was determined	8
	7b	When applicable, explanation of any interim analyses and stopping guidelines	N/A
Randomisation:			
Sequence generation	8a	Method used to generate the random allocation sequence	6
	8b	Type of randomisation; details of any restriction (such as blocking and block size)	6
Allocation concealment mechanism	9	Mechanism used to implement the random allocation sequence (such as sequentially numbered containers), describing any steps taken to conceal the sequence until interventions were assigned	8
Implementation	10	Who generated the random allocation sequence, who enrolled participants, and who assigned participants to interventions	6
Blinding	11a	If done, who was blinded after assignment to interventions (for example, participants, care providers, those	8

		assessing outcomes) and how	
	11b	If relevant, description of the similarity of interventions	N/A
Statistical methods	12a	Statistical methods used to compare groups for primary and secondary outcomes	8-9
	12b	Methods for additional analyses, such as subgroup analyses and adjusted analyses	N/A
Results			
Participant flow (a diagram is strongly recommended)	13a	For each group, the numbers of participants who were randomly assigned, received intended treatment, and were analysed for the primary outcome	N/A
	13b	For each group, losses and exclusions after randomisation, together with reasons	N/A
Recruitment	14a	Dates defining the periods of recruitment and follow-up	N/A
	14b	Why the trial ended or was stopped	N/A
Baseline data	15	A table showing baseline demographic and clinical characteristics for each group	N/A
Numbers analysed	16	For each group, number of participants (denominator) included in each analysis and whether the analysis was by original assigned groups	N/A
Outcomes and estimation	17a	For each primary and secondary outcome, results for each group, and the estimated effect size and its precision (such as 95% confidence interval)	N/A
	17b	For binary outcomes, presentation of both absolute and relative effect sizes is recommended	N/A
Ancillary analyses	18	Results of any other analyses performed, including subgroup analyses and adjusted analyses, distinguishing pre-specified from exploratory	N/A
Harms	19	All important harms or unintended effects in each group (for specific guidance see CONSORT for harms)	N/A
Discussion			
Limitations	20	Trial limitations, addressing sources of potential bias, imprecision, and, if relevant, multiplicity of analyses	N/A
Generalisability	21	Generalisability (external validity, applicability) of the trial findings	N/A
Interpretation	22	Interpretation consistent with results, balancing benefits and harms, and considering other relevant evidence	N/A
Other information			
Registration	23	Registration number and name of trial registry	3
Protocol	24	Where the full trial protocol can be accessed, if available	3
Funding	25	Sources of funding and other support (such as supply of drugs), role of funders	11

*We strongly recommend reading this statement in conjunction with the CONSORT 2010 Explanation and Elaboration for important clarifications on all the items. If relevant, we also recommend reading CONSORT extensions for cluster randomised trials, non-inferiority and equivalence trials, non-pharmacological treatments, herbal interventions, and pragmatic trials. Additional extensions are forthcoming: for those and for up to date references relevant to this checklist, see www.consort-statement.org.

BMJ Open

Study Protocol for the G-SPIRIT trial: A Randomized, Placebo-Controlled, Double-blinded Phase 3 Trial of Granulocyte Colony Stimulating Factor-Mediated Neuroprotection for Acute Spinal Cord Injury

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Primary Subject Heading:	Neurology
Secondary Subject Heading:	Medical management
Keywords:	Neurological injury < NEUROLOGY, Spine < ORTHOPAEDIC & TRAUMA SURGERY, Clinical trials < THERAPEUTICS

Study Protocol for the G-SPIRIT trial: A Randomized, Placebo-Controlled, Double-blinded Phase 3 Trial of Granulocyte Colony Stimulating Factor-Mediated Neuroprotection for Acute Spinal Cord Injury

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Word Count: 2516

Abstract

Introduction: Granulocyte Colony Stimulating Factor (G-CSF) is generally used for neutropenia. Previous experimental studies revealed that G-CSF promoted neurological recovery after spinal cord injury (SCI). Next we moved to early phase of clinical trials. In a phase 1/2a trial, no adverse events were observed. Next, we conducted a non-randomized, non-blinded, comparative trial, which suggested the efficacy of G-CSF for promoting neurological recovery. Based on those results, we are now performing a phase 3 trial.
Methods and Analysis: The objective of this study is to evaluate the efficacy of G-CSF for acute SCI. The study design is a prospective, multicenter, randomized, double-blinded, placebo-controlled comparative study. The current trial includes cervical SCI (severity of American Spinal Injury Association (ASIA) Impairment Scale B/C) within 48 hours after injury. Patients are randomly assigned to G-CSF and placebo groups. The G-CSF group is administered 400 μg/m^2/d×5d of G-CSF in normal saline via intravenous infusion for 5 consecutive days. The placebo group is similarly administered a placebo. Our primary endpoint is changes in ASIA motor scores from baseline to 3 months. Each group includes 44 patients (88 total patients).This trial is funded by the Center for Clinical Trials, Japan Medical Association.
Ethics and Dissemination: The study was approved by the Institutional Review Board of each institution and will be conducted according to the principles of the World Medical Association Declaration of Helsinki and in accordance with the Japanese Medical Research Involving Human Subjects Act and other guidelines, regulations and Acts. Results of the clinical study will be submitted to the head of the respective clinical study site as a report after conclusion of the clinical study by the sponsor-investigator. Even if the results are not favorable despite conducting the clinical study properly, the data will be published as a paper.
Registration: UMIN000018752
https://upload.umin.ac.jp/cgi-open-bin/ctr/ctr.cgi?function=brows&action=brows&type=summary&recptno=R000021694&language=J

Protocol version: Ver. 5.0 (2016/10/20)

Strengths and Limitations of This Study

Strengths

- ✓ Novel drug therapy for acute spinal cord injury is much-needed.
- ✓ Randomized, placebo-controlled, double-blinded design can eliminate bias.

Limitations

- ✓ Acute spinal cord injury patients are difficult to recruit to the trial.
- ✓ Patient's neurological status in acute phase is unstable, possibly resulting in dispersion of the patient's background.

- ✓ Concealment must be performed very strictly because G-CSF apparently increases white blood cell count.

Introduction

Spinal cord injury (SCI) is a devastating injury by which the patient can suffer from long-lasting severe sequelae including palsy of extremities, sensory disturbance, bowel/bladder/sexual dysfunction, and neuropathic pain. Conceptually, SCI is divided into two chronological phases: a primary and a secondary phase. Primary injury is mechanical damage to spinal cord tissue itself caused by fracture and/or dislocation or compression. Secondary injury is triggered by the primary injury and is a biological reaction of the spinal cord, which includes ischemia, hemorrhage, excitotoxicity, hyperpermeability, and inflammation[1]. Because secondary injury can be the main target of treatment, extensive laboratory and clinical investigation of neuroprotection is needed to manage secondary injury[2].

To date, the only approved neuroprotective therapy for SCI is massive methylprednisolone sodium succinate (MPSS) therapy based on the NASCIS 2 study[3]. However, recent reports revealed that MPSS shows only a modest effect for SCI. In addition, several reports have described adverse events induced by MPSS for SCI including infections (pneumonia, urinary tract infection) and gastrointestinal disorders (gastric ulcer, etc.)[4]. Therefore, the use of MPSS for SCI has become controversial[5]. Accordingly, a new therapeutic drug for SCI is desirable.

Granulocyte colony-stimulating factor (G-CSF, generic name: filgrastim) is a growth factor that affects the hematopoietic system, promoting differentiation, proliferation, and survival of granulocytes[6]. Clinically, in Japan, G-CSF is administered to patients with leukopenia, and to peripheral stem cell transplantation donors, G-CSF is administered to mobilize hematopoietic stem cells into the peripheral blood[7]. In the central nervous system, G-CSF has properties to mobilize the movement of bone marrow cells into the brain[8] and spine, and in a stroke model, has shown neuroprotective properties[9]. In other countries, clinical studies of the effects of G-CSF in cerebral infarction have been reported[10].

To prove the hypothesis that G-CSF has neuroprotective properties against SCI, G-CSF

was administered to rat and mouse animal models of spinal cord injury, and hind limb function improved significantly after administration of G-CSF. Further investigations into the mechanism of action of G-CSF in SCI were conducted. Data obtained to date identify the following properties of G-CSF: [1] mobilization of bone marrow-derived stem cells causing their biointegration at the site of SCI[11], [2] direct inhibition of nerve cell death[12], [3] protection of the myelin sheath by inhibiting oligodendrocyte cell death[13], [4] inhibition of inflammatory cytokine expression (TNF-α, IL-1β)[13], and [5] promotion of neovascularization[14]. These properties suggest that G-CSF has a neuroprotective effect in acute SCI.

Based on these properties, a phase 1/2a clinical study was conducted where the main objective was to confirm the safety and feasibility of G-CSF for treatment of patients with acute SCI[15]. This study was an open-label, dose-titrating study with no control group. As the initial step, 5 patients were given 5 μg/kg/day of G-CSF for 5 consecutive days by i.v. infusion, and as the second step, 11 patients were given 10 μg/kg/day of G-CSF for 5 consecutive days by i.v. infusion. No serious adverse events were noted, and the safety of G-CSF administration in patients with acute SCI was confirmed[15].

As a next step, to validate the efficacy of G-CSF in neuroprotective treatment, a multicenter, prospective, nonrandomized, nonblinded, comparative control study (phase 2b clinical trial) was conducted[16]. Based on the results of the previous phase 1/2a clinical trial, the dosage and duration of treatment with G-CSF in this study was 10 μg/kg/day for 5 consecutive days. Patients with acute cervical SCI (within 48 h of injury) were registered in the clinical trial and allocated to either the G-CSF group (G-CSF 10 μg/kg/day × 5 days i.v. infusion) or control group (no G-CSF administration) at each treatment facility. A total of 19 patients in the G-CSF group and 26 patients in the control group were observed for 3 months or longer. American Spinal Injury Association (ASIA) motor score (motor: 0 to 100 points) was compared between the groups. ASIA motor scores were 26.1 ± 18.9 in the G-CSF group and 12.2 ± 14.7 in the control group showing a significant improvement of motor paralysis in the G-CSF group ($p < 0.01$). In addition, in cases that could be followed for 1 year or longer, a significant improvement of ASIA motor score was observed in the

G-CSF group[16].

Based on results of these preclinical and early phase clinical trials, we are now conducting a phase 3 trial.

Specific Objective

The objective of this study is to evaluate the efficacy of G-CSF for improving motor paralysis in acute SCI.

Methods and Analysis

Design of the study

The study design of the current trial is a prospective, multicenter, randomized, double-blinded, placebo-controlled comparative study.

Study Procedures

The study outline is shown in Figure. Patients will be randomly assigned to G-CSF and placebo groups. A central registration system will be used for dynamic randomization into the investigational treatment group (G-CSF) and control group (placebo) based on age at registration (16–64 years of age, or 65–84 years of age) and severity of paralysis (AIS B or C) at 48 h after injury. With or without the surgical stabilization and/or decompression was not included in stratification. Initially, screened patients with severity AIS B/C will be tentatively enrolled. Initial screening of the patients include clinical laboratory test, imaging studies including X-ray, magnetic resonance imaging and computed tomography, and neurological/functional evaluations.

Neurological re-assessment will be performed 48±4 hours after SCI and the patients who will recover to severity AIS D will be excluded. The patients with severity AIS B/C at neurological re-assessment 48±4 hours after SCI will be enrolled and randomly allocated to either the investigational treatment group (G-CSF) or control group (placebo) in a 1 to 1 ratio. The subject registration center uses a program based on an appropriate computer algorithm to allocate patients into groups. The first dose of investigative drugs will be

administered to the patients after re-assessment of neurological status and enrollment 48±4 hours after SCI.

The G-CSF group will be administered 400 μg/m^2/d × 5d of G-CSF in normal saline via intravenous infusion for 5 consecutive days. The placebo group will be similarly administered a placebo. The first dose of investigative drugs will be administered to the patients 48±4 hours after SCI. The dosing schedule will be once a day in every morning (9-10 AM) for consecutive 5 days even in case that the first dosing was performed at night because of restriction on practices. Investigational drugs including both G-CSF and placebo will be stored in refrigerator which will be kept between 1- 6□ and has temperature-logger in pharmacies in each participating institutes. The investigational drugs is packaged in ample with label only printed as serial numbers, ten ample is packed in one box with label only printed serial numbers. Web-based allocation system will show the serial number of investigational drug which must be used to respective patients to ensure blinding.

We decided dosage of drug based on our previous non-randomized early phase clinical studies. On phase 1/2a clinical study, of which study design was open-label, dose-titrating study with no control group, SCI patients who were administered 10μg/kg/day G-CSF for 5 days showed marked elevation of white blood cell number (reached nearly 50000/μl, of which WBC number might cause adverse effects of G-CSF) during G-CSF administration[15]. Therefore we decided to withdraw additional titration. In addition, next phase of clinical study, of which design was multicenter, prospective, nonrandomized, nonblinded, comparative control study, showed suggestive efficacy of G-CSF (10 μg/kg/d for 5 days) for acute SCI[16]. Based on those results, we finally decided the dosage of G-CSF as 10 μg/kg/d × 5d for 5 days. In the current clinical trial, the dosage of G-CSF is written as 400 μg/m^2/d (= 10 μg/kg/d) according to the Japanese Pharmaceutical and Medical Device Agency (PMDA)'s instruction for consolidation with product labeling.

We decided to 48hours after SCI as the therapeutic time window because our previous multicenter, prospective, nonrandomized, nonblinded, comparative control study, which recruited SCI patients within 48 hours after injury, showed that there was no significant difference in neurological outcome between the patient administered G-CSF very early

after the injury and 48 hours after injury[16].

Allocation will be concealed between blinded evaluators of efficacy/safety and those for laboratory data, as G-CSF markedly increases white blood cell counts that can reveal patient treatment.

Our primary endpoint is changes in ASIA motor scores (international standards for neurological classification of spinal cord injury (ISNSCI)[17] suppl. Figure 1) from baseline to 3 months calculated as follows: 3-month ASIA motor score change = 3-month ASIA motor score – pretreatment ASIA motor score. To maintain consistency of neurological assessment among the each evaluators, attending lecture and e-learning (in website of International Spinal Cord Society: http://www.elearnsci.org/) of ASIA/ISNSCI scoring system is mandatory for every investigators participating to the present trial.

Secondary endpoints are as follows. [1] Change in ASIA motor scores[17] at 6 months and 12 months after G-CSF administration compared with pretreatment. [2] Changes in sensory paralysis over time: change in ASIA sensory scores at 3 months, 6 months, and 12 months after G-CSF administration compared with pretreatment. [3] Severity of functional compromise because of paralysis: AIS before administration and at 3, 6, and 12 months after administration of G-CSF. [4] Percentage of responders: percentage of patients whose AIS improved by 1 grade or more at 3, 6, and 12 months after administration compared with before administration of G-CSF. [5] Neurological level of injury (NLI): percentage of patients whose NLI decreased by 1 grade of more at 3, 6, or 12 months after administration of G-CSF compared with pretreatment. [6] SCIM[18]: change in SCIM scores at 3, 6, and 12 months after G-CSF administration compared with pretreatment [7] EQ-5D[19]: measured EQ-5D efficacy scores at 3, 6, and 12 months after G-CSF.

To strengthen the analysis, more strict blindness of assessment by the evaluator for outcome measures is ideal. Therefore every prior score/measurement should be blinded and/or not the same person should assess every midpoint motor/sensory functional measurement. However, PMDA instructed us to assess one patients' functional evaluation by one evaluator for consistent assessment in respective patients. PMDA considers the consistency of assessment in respective patients is more important.

The precise explanation of abovementioned outcome measures is as followings (suppl. Figures). ASIA motor score is determined by examining a function of 10 key muscles with manual muscle testing (MMT) in the supine position[17]. The score ranges from 0 to 25 for each extremity (MMT 0-5 in 5 muscles in each extremity), totaling 0 to 50 for the upper limbs and 0 to 50 for the lower limbs, resulting in total of ASIA motor score ranges from 0 to 100. ASIA motor score shows the degree of motor impairment induced by SCI (suppl. Figure 1)[17]. ASIA sensory scores is determined by sensory testing of a key point in each of the 28 dermatomes (from C2 to S4-5) on the each sides of the body. Two aspects of sensation are examined, light touch and pin prick, at all the key points. Light touch and pin prick sensation is separately scored on a three-point scale, 0 = absent, 1= altered, 2 = normal or intact. As a result, ASIA sensory scores range from 0 to 112 (suppl. Figure 1) in light touch and pin prick respectively[17]. AIS is used for gross grading of impairment: A = Complete. No sensory or motor function is preserved in the sacral segments S4-S5. B = Sensory incomplete. Sensory but not motor function is preserved below the neurological level. C = Motor incomplete. Motor function is preserved below the neurological level, and more than half of key muscle functions below the single neurological level of injury have a muscle grade less than 3 (Grades 0–2). D = Motor incomplete. Motor function is preserved below the neurological level, and at least half (half or more) of key muscle functions below the neurological level have a muscle grade >3. E = Normal[17]. If sensation and motor function as tested are graded as normal in all segments, and the patient had prior deficits, then the AIS grade is E (suppl. Figure 1, footnote)[17]. NLI refers to the most caudal segment of the cord with intact sensation and antigravity muscle function strength, provided that there is normal (intact) sensory and motor function rostrally (suppl. Figure 1, footnote)[17]. SCIM is disability scale developed specifically for patients with spinal cord lesions in order to make the functional assessments of patients with paraplegia or tetraplegia more sensitive to changes[18]. The SCIM includes the following areas of function: self-care (subscore (0-20), respiration and sphincter management (0-40) and mobility (0-40). Each area is scored according to its proportional weight in these patients' general activity[18]. The final score ranges from 0 to 100 (suppl.

Figure 2-4). EQ-5D essentially consists of 2 pages[19] (suppl. Figure 5, 6): the EQ-5D descriptive system and the EQ visual analogue scale (EQ VAS). The descriptive system comprises five dimensions: mobility, self-care, usual activities, pain/discomfort and anxiety/depression. Each dimension has 5 levels: no problems, slight problems, moderate problems, severe problems and extreme problems. The patient is asked to indicate his/her health state by ticking the box next to the most appropriate statement in each of the five dimensions[19]. This decision results in a 1-digit number that expresses the level selected for that dimension. The digits for the five dimensions can be combined into a 5-digit number that describes the patient's health state and can be converted to efficacy score with calibration scale (ranges -0.111 to 1.000)[19]. The EQ VAS records the patient's self-rated health on a vertical visual analogue scale, where the endpoints are labelled 'The best health you can imagine' and 'The worst health you can imagine'. The VAS can be used as a quantitative measure of health outcome that reflect the patient's own judgement[19].

Each group will include 44 patients (88 patients in total). Our protocol was approved by PMDA and this trial will be funded by the Center for Clinical Trials, Japan Medical Association.

Inclusion

Inclusion criteria are as follows. [1] Patients with cervical SCI (severity of AIS B/C) within 48 h of injury. [2] Patients reassessed for neurological status at 48 h after injury, and those whose palsy is AIS B/C will be enrolled. [3] Patients with neurological level of injury between C4 and C7. [4] Patients with age of 16 to 85 years. [5] Patients who agree to participate in the current trial and from whom informed consent was obtained orally and in writing. [6] Patients who can be followed up for 12 months after SCI.

Exclusion

Exclusion criteria are as follows. [1] Patients with neurological recovery to AIS D at neurological reassessment 48 h after SCI, because only AIS B/C patients will be included to

standardize the severity of paresis in order to stratify the patients at the initiation of drug administration. [2] Allergy to G-CSF. [3] Hematological malignancy, [4] within 6 months after invasive coronary intervention, [5] splenomegaly, or [6] pregnancy, [7] consciousness impairment, [8] neurological disorders that can affect neurological evaluation in the present trial, [9] fracture of extremities that can affect the neurological evaluation and [10] massive dose administration of MPSS. Exclusion criteria 2 - 6 are set for safety, criteria 1, 7 - 9 are set to maintain homogeneity of the patient population enrolled, criteria 7-9 are set for maintenance of accuracy of functional assessment, criteria 7 is set to obtain patients' own informed consent upon participation to the trial and criteria 10 is set to omit the possible interference of MPSS on outcome assessment.

Concealment

Patients will be administered drug or placebo and be evaluated in a double-blinded manner. An evaluator blinded to the treatment will take charge of patient evaluation including clinical findings and paresis, without laboratory data because G-CSF induces an apparent increase of white blood cell count that makes it easy to distinguish G-CSF and placebo treatment. Therefore, a non-blinded evaluator for laboratory data will be assigned to evaluate laboratory data alone. From a safety point-of view, the dose of G-CSF will be modulated according to excessive increase of white blood cell count. Therefore, a non-blinded evaluator will instruct the pharmacy to modulate the dose of G-CSF based on white blood cell count.

Sample Size Calculation

The target sample size for this randomized trial is 88. This number is based on the results of previous clinical trials[16]. The estimated group difference (± standard deviation) of change in ASIA motor scores from baseline to 3 months is 13.9 (± 21.9). A sample size of 44 patients in each group will provide 80% power to detect a difference of the change in ASIA motor scores between the G-CSF and the placebo treatments, using a mixed-effects models for repeated measures (MMRM) at a two-sided 5% level of significance. A

common correlation of 0.25 at each time point is assumed. A dropout rate of 10% is allowed. Thus, the total sample size of 88 patients is required for the trial.

Statistical analyses

The analyses of the primary and secondary end points will be performed as intention-to-treat analyses in a full analysis set, which includes all patients who: (1) took at least one course of treatment during the study; (2) do not present any serious violation of the study protocol; and (3) have data collected after commencement of treatment. For the baseline characteristics, summary statistics will comprise frequencies and proportions for categorical variables, and means and SDs for continuous variables. The patient characteristics will be compared using a chi-square test for categorical variables, and a *t* test or Wilcoxon rank sum test for continuous variables. Missing data including loss to follow-up and missing measurement will be supplemented with MMRM.

For the primary analysis, aimed at comparing treatment effects, a change in ASIA motor score from baseline to 3 months and its 95% confidence interval (CI) will be estimated using the MMRM. To test for significant association of the primary endpoint, a mixed effects model for repeated measures with an unstructured covariance matrix will be applied to adjust for age (<65 years or ≥65 years) and AIS at 48 h after the injury (B or C).

For the secondary analysis, the change in ASIA motor score will be compared using a Student *t* test and the 95% CI will be estimated. The same method will be applied to change in sensory score, SCIM, and EQ-5D. A chi-squared test will be applied to the frequencies of the responder in AIS and of the improvement in NLI. The frequency of AIS will be summarized. The frequency of AEs will be compared using a Fisher exact test.

All comparisons are planned and all *p* values will be two sided. $p < 0.05$ will be considered significant. All statistical analyses will be performed using SAS software (version 9.4; SAS Institute, Cary, NC, USA). The statistical analysis plan will be developed by the principal investigator and the biostatistician before completion of patient recruitment and fixing of data.

Data Monitoring Committee

The data monitoring committee consists of three clinical trial specialists, including a biostatistician, who are independent from the current study. The committee will meet at least 2 times per year and all the data obtained by the current trial will be checked by the committee[20].

Adverse Events

As for safety evaluation, adverse events will be collected as follows. "Adverse events" refers to any untoward symptom or disease or signs of such (including clinical laboratory data abnormalities) in a clinical investigation subject after informed consent and does not necessarily have a causal relationship with the investigational product (G-CSF).

Increases in white blood counts will be considered an adverse event only when the count exceeds 50,000/μL from the perspective of a pharmacological effect of G-CSF, and any values below this will not be handled as an adverse event. Anaphylaxis and adult respiratory distress syndrome are the most representative G-CSF-related severe adverse events to be payed full attention.

All adverse events will code terminology used by the investigators according to the ICH International Medical Dictionary for Regulatory Activities Japanese version (MedDRA/J).

Ethics and Dissemination

Ethics

The study was approved by the Institutional Review Board (IRB) of each institutions involved to the present trial and will be conducted according to the principles of the World Medical Association (WMA) Declaration of Helsinki—Ethical Principles for Medical Research Involving Human Subjects with the amendments made in Seoul, South Korea, October 2008, with a Note of Clarification on Paragraph 29 added by the WMA General Assembly, Washington 2002; Note of Clarification on Paragraph 30 added by the WMA General Assembly, Tokyo 2004, and in accordance with the Japanese Medical Research Involving Human Subjects Act (WMO) and other guidelines, regulations and Acts.

Patient informed consent

The principal investigator will try to prepare the informed consent form and other explanatory materials in as simple language as possible in order to obtain informed consent from the patient and the patient's legal representative. In case that trial participants cannot sign the informed consent form due to upper extremity palsy caused by SCI, allograph by patients' representative will be allowed.

Public Disclosure and Publication Policy

Results of the clinical study will be submitted to the head of the respective clinical study site as a report after conclusion of the clinical study by the sponsor-investigator (includes study coordinating investigator). Even if the results are not favorable despite conducting the clinical study properly, the data will be published as a paper. Other sponsor-investigators (including the clinical study coordinating investigator), if they plan to publicize the data from this study in a specialized academic society conference or other external site, must first obtain permission from the other principal investigators and investigational product provider. In publicizing the results, the confidentiality of the subjects will be maintained and proofread in advance by the other sponsor-investigators (includes coordinating investigator) and investigational product provider.

Discussion

The current trial is a confirmative trial to elucidate the therapeutic efficacy of G-CSF for SCI. If the current trial can successfully show significant improvement of motor paralysis of SCI by G-CSF, we will move forward to drug approval application to the Ministry for Health and Labor, Japan. The entire protocol of the current trial was approved beforehand for the initiation of the current trial by the Japanese Pharmaceutical and Medical Device Agency (PMDA). The PMDA will also permit a drug approval application if significant efficacy of G-CSF for SCI is proven.

The current trial is an important milestone for SCI clinics and research to explore

G-CSF for SCI.

Trial Status

The present trial is now on-going.

Trial Sites

Nineteen major hospitals in Japan constituting the G-SPIRIT study group as follows: Tohoku University Hospital, Miyagi; Niigata University, Niigata; Dokkyo University Hospital, Tochigi; Tsukuba University Hospital, Ibaraki; Tsukuba Medical Center, Ibaraki; Chiba University Hospital, Chiba; Funabashi Municipal Medical Center, Chiba; Kimitsu Chuo Hospital, Chiba; Chiba Rosai Hospital, Chiba; Tokai University Hospital, Kanagawa; Hamamatsu University Hospital, Shizuoka; Gifu University Hospital, Gifu; Chubu Rosai Hospital, Aichi; Mie University Hospital, Mie; Kanazawa Medical Collage, Ishikawa; Kobe Red Cross Hospital, Hyogo; Hiroshima University Hospital, Hiroshima; Yamaguchi University Hospital, Yamaguchi; and Nagasaki Rosai Hospital, Nagasaki.

Funding Statement

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Competing Interests

No authors have any competing interests to declare.

Contributorship Statement

Koda M, Hanaoka H, Sato T, Fujii Y, Hanawa M, Takahashi S, Furuya T, Ijima Y, Saito J, Kitamura M, Ohtori S, Matsumoto Y, Abe T, Watanabe K, Hirano T, Ohashi M, Shoji H, Mizouchi T, Takahashi I, Kawahara N, Kawaguchi M, Orita Y, Sasamoto T, Yoshioka M, Fujii M, Yonezawa K, Soma D, Taneichi H, Takeuchi D, Inami S, Moridaira H, Ueda H, Asano F, Shibao Y, Aita I, Takeuchi Y, Mimura M, Shimbo J, Someya Y, Ikenoue S, Sameda H, Takase K, Ikeda Y, Nakajima F, Hashimoto M, Ozawa T, Hasue F, Fujiyoshi T, Kamiya K, Watanabe M, Katoh H, Matsuyama Y, Yamamoto Y, Togawa D, Hasegawa T,

Kobayashi S, Yoshida G, Oe S, Banno T, Arima H, Akeda K, Kawamoto E, Imai H, Sakakibara T, Sudo A, Ito Y, Kikuchi T, Osaki S, Tanaka N, Nakanishi K, Kamei1 N, Kotaka S, Baba H, Okudaira T, Konishi H, Yamaguchi T, Ito K, Katayama Y, Matsumoto T , Matsumoto T, Idota M, Kanno H, Aizawa T, Hashimoto K, Eto T, Sugaya T, Matsuda M, Fushimi K, Nozawa S, Iwai C, Taguchi T, Kanchiku T, Suzuki H, Nishida N, Funaba M and Yamazaki M substantially contributed to the conception or design of the present trial protocol, to the acquisition and analyses of data for the present manuscript, to drafting the manuscript and revising the present manuscript critically for important intellectual content and final approval of the version of the present manuscript to be published to agreement to be accountable for all aspects of the present manuscript in ensuring that questions related to the accuracy or integrity of any part of the present manuscript are appropriately investigated and resolved, consequently fulfill the ICMJE criteria for authorship.

References

1. Oyinbo CA. Secondary injury mechanisms in traumatic spinal cord injury: a nugget of multiply cascade. ***Acta Neurobiol Exp*** 2011;71:281-299.
2. Karsy M, Hawryluk G. Pharmacologic Management of Acute Spinal Cord Injury. ***Neurosurg Clin N Am*** 2017;28:49-62. doi: 10.1016/j.nec.2016.07.002.
3. Bracken MB, Shepard MJ, Collins WF, et al. A randomized controlled trial of methylprednisolone or naloxone in the treatment of acute spinal cord injury: results of the second national acute spinal cord injury study. ***N Engl J Med*** 1990;322: 1405-1411.
4. Matsumoto T, Tamaki T, Kawakami M, et al. Early complications of high-dose metyl-prednisolone sodium succinate treatment in the follow-up of acute cervical spinal cord injury. ***Spine*** 2001;26: 426-430.
5. Hurlbert RJ, Hadley, Walters BC, et al. Pharmacological Therapy for Acute Spinal cord Injury. ***Neurosurgery*** 2013;72: 93-105.
6. Nicola NA, Metcalf D, Matsumoto M, Johnson GR. Purification of a factor inducing differentiation in murine myelomonocytic leukemia cells. Identification as

granulocyte colony-stimulating factor. ***J Biol Chem*** 1983;258: 9017-9023.

7. Roberts AW: G-CSF: a key regulator of neutrophil production, but that's no all! ***Growth Factors*** 2005;23: 33–41.
8. Kawada H, Takizawa S, Takanashi T, et al. Administration of hematopoietic cytokines in the subacute phase after cerebral infarction is effective for functional recovery facilitating proliferation of intrinsic neural stem/progenitor cells and transition of bone marrow-derived neuronal cells. ***Circulation*** 2006;113: 701-710.
9. Schneider A, Kruger C, Steigleder T, et al. The hematopoietic factor G-CSF is a neuronal ligand that counteracts programmed cell death and drives neurogenesis. ***J Clin Invest*** 2005;115: 2083-2098.
10. Shyu WC, Lin SZ, Lee CC, Liu DD, Li H. Granulocyte colony-stimulating factor for acute ischemic stroke: a randomized controlled trial. ***CMAJ*** 2006;174: 927-933.
11. Koda M, Nishio Y, Kamada T, et al. Granulocyte colony-stimulating factor （G-CSF） mobilizes bone marrow-derived cells into injured spinal cord and promotes functional recovery after compression-induced spinal cord injury in mice. ***Brain Res*** 2007;1149: 223-231.
12. Nishio Y, Koda M, Kamada T, et al. Granulocyte colony-stimulating factor （G-CSF） attenuates neuronal death and promotes functional recovery after spinal cord injury in mice. ***J Neuropathol Exp Neurol*** 2007;66: 724-731.
13. Kadota R, Koda M, Kawabe J, et al. Granulocyte colony-stimulating factor (G-CSF) protects oligodendrocyte and promotes hindlimb functional recovery after spinal cord injury in rats. ***PLoS One*** 2012;7:e50391.
14. Kawabe J, Koda M, Hashimoto M, et al. Granulocyte colony-stimulating factor （G-CSF） exerts neuroprotective effects via promoting angiogenesis after spinal cord injury in rats. ***J Neurosurg Spine*** 2011;15: 414-421.
15. Takahashi H, Yamazaki M, Okawa A, et al. Neuroprotective therapy using granulocyte colony-stimulating factor for acute spinal cord injury: a phase I/IIa clinical trial. ***Eur Spine J*** 2012;21:2580-2587.
16. Inada T, Takahashi H, Yamazaki M, et al. A multicenter prospective

non-randomized controlled clinical trial to prove neurotherapeutic effects of granulocyte colony-stimulating factor（G-CSF）for acute spinal cord injury: Analyses of follow-up cases after at least one year. ***Spine*** 2014;39:213-219.

Figure legends

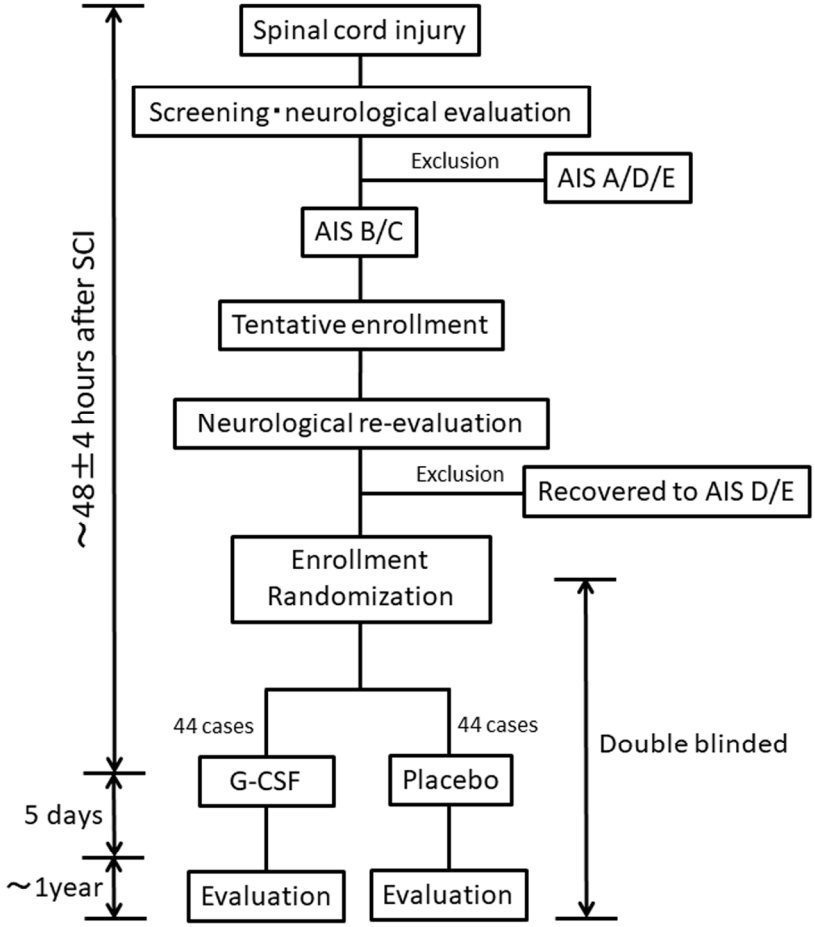
Figure: Schematic diagram showing trial timeline.

Cervical SCI with severity of AIS B/C within 48 hours after injury is recruited. Patients are randomly assigned to G-CSF and placebo groups. The G-CSF group is administered G-CSF in normal saline via intravenous infusion for 5 consecutive days. The placebo group is similarly administered a placebo.

Suppl. Figure 1: ASIA score chart.

Suppl. Figure 2-4: Spinal cord independence measure chart (Japanese version) for spinal cord injury-specific ADL evaluation.

Suppl. Figure 5, 6: Euro-QOL 5-dimension chart (Japanese version) for QOL evaluation.



Schematic diagram showing trial timeline.

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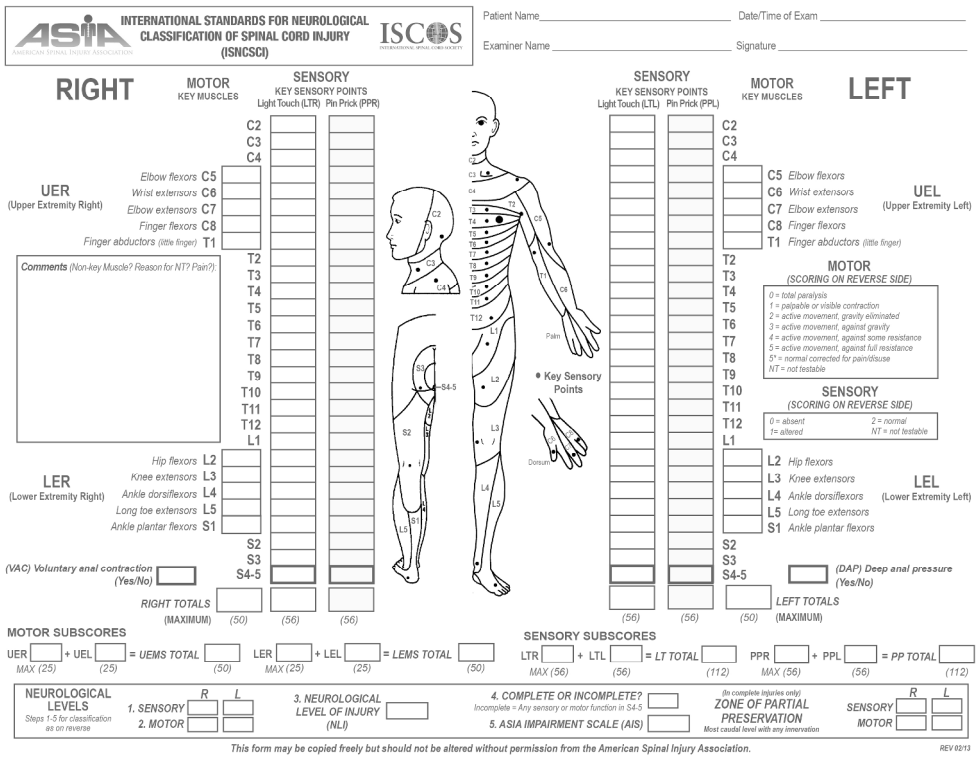

274x206mm (300 x 300 DPI)

セルフケア

(1) 食事 (切る, 容器を開ける, 飲み物を注ぐ, 食べ物を口に運ぶ, 飲み物の入ったコップをもつ)
- 0. 静脈栄養または胃瘻が必要である. あるいは経口摂取において全介助を要する.
- 1. 食べたり飲んだりすること, または補助器具の装着に部分介助を要する.
- 2. 食べることは自立, 補助器具を必要とするか, または食べ物を切ったり注いだりすること, 容器の開閉にのみ解介助を要する.
- 3. 食べることも飲むことも自立, 介助や補助器具を必要としない.

(2) 入浴 (頭と体を石鹸でこすって洗い, 乾かす, 蛇口の開け閉め) A —上半身, B —下半身

A. 0. 全介助を要する.
- 1. 部分介助を要する.
- 2. 補助器具, または特定の環境 (例えば, 手すりや椅子) が整っていれば, 自立して体を洗う.
- 3. 自立して体を洗う. (通常, 健常者なら用いることのない) 補助器具や特定の環境設定は不要

B. 0. 全介助を要する.
- 1. 部分介助を要する.
- 2. 補助器具, または特定の環境が整っていれば, 自立して体を洗う.
- 3. 自立して体を洗う. 補助器具や特定の環境設定は不要

(3) 更衣 (衣服, 靴, 常用している装具の着脱) A —上半身, B —下半身

A. 0. 全介助を要する.
- 1. ボタン, ファスナー, ひものない衣服で部分介助を要する.
- 2. ボタン, ファスナー, ひものない衣服であれば自立, 補助器具と特定の環境設定, またはそのどちらかが必要.
- 3. ボタン, ファスナー, ひものない衣服であれば自立, 補助器具も特定の環境設定も不要. ボタン, ファスナー, ひもの扱いにだけ介助, 補助器具, または特定の環境設定が必要.
- 4. (衣類の種類を問わず) 自立して着替える. 補助器具も特定の環境設定も不要.

B. 0. 全介助を要する.
- 1. ボタン, ファスナー, ひものない衣服で部分介助を要する.
- 2. ボタン, ファスナー, ひものない衣服であれば自立, 補助器具と特定の環境設定, またはそのどちらかが必要.
- 3. ボタン, ファスナー, ひものない衣服であれば自立, 補助器具も特定の環境設定も不要. ボタン, ファスナー, ひもの扱いにだけ介助, 補助器具, または特定の環境設定が必要.
- 4. (衣類の種類を問わず) 自立して着替える. 補助器具も特定の環境設定も不要.

(4) 整容 (手洗い, 洗顔, 歯磨き, 整髪, 髭剃り, 化粧)
- 0. 全介助を要する.
- 1. 部分介助を要する.
- 2. 補助器具があれば自立して整容動作をおこなう.
- 3. 補助器具を用いずに自立して整容動作をおこなう.

小計 (0–20)

呼吸と排泄管理

(5) 呼吸
- 0. 気管チューブと持続的補助換気または間欠的補助換気が必要.
- 2. 気管チューブを装用して自己呼吸, 酸素を必要としたり, 咳嗽時または気管チューブの管理に多大な介助を必要とする.
- 4. 気管チューブを装用して自己呼吸, 咳嗽時または気管チューブの管理に少ししか介助を必要としない.
- 6. 気管チューブなしで自己呼吸, 酸素, 咳嗽時の多大な介助, マスク (例 : 終末呼気陽圧 PEEP), または間欠的補助換気 (BiPAP) を必要とする.
- 8. 気管チューブなしで自己呼吸, 介助または咳嗽刺激を少ししか必要としない.
- 10. 介助も器具もなしで自己呼吸.

190x254mm (300 x 300 DPI)

(6) 排尿管理
0. 留置カテーテル
3. 残尿量＞100cc，不定期導原または介助による間欠的導尿．
6. 残尿量＞100cc または間欠的自己導尿，集尿器[2] をあてがう際の介助は必要．
9. 間欠的自己導尿，集尿器を使用する．器具をあてがう際の介助は不要．
11. 間欠的自己導尿，導尿と導尿の間には失禁なし，集尿器は使用しない．
13. 残尿量＜100cc，集尿器のみ必要，集尿器の取扱いには介助不要．
15. 残尿量＜100cc，失禁なし，集尿器を使用しない．

(7) 排便管理
0. 排便が不規則またはごく低頻度（3 日に 1 回未満）．
5. 規則的ながら，（座薬を挿入するなどに）介助を要する，失敗はまれ（月 2 回未満）．
8. 規則的な排便で，介助を要しない，失敗はまれ（月 2 回未満）．
10. 規則的な排便で，介助を要しない，失敗はなし．

(8) トイレの使用（会陰部の清潔，使用前後での衣服の扱い，ナプキンまたはおむつの使用）
0. 全介助を要する．
1. 部分介助を要する．自分でお尻を拭けない．
2. 部分介助を要する．自立してお尻を拭ける．
4. 自立してトイレを使用するも，補助器具または特別な環境（例：手すり）が整っている必要あり．
5. 自立してトイレを使用．補助器具も特別な環境が整っている必要もない．

小計（0–40）

移動（室内とトイレ）

(9) ベッド上での姿勢変換と褥瘡予防動作
0. ベッド上で上半身の向きを変えること，下半身の向きを変えること，起き上がること，および車椅子上でのプッシュアップのすべての動作に介助が必要，補助器具の要否は問わないが，電動器具は用いない．
2. 介助なくできる動作が 1 つある[3]．
4. 介助なくできる動作が 2 つまたは 3 つある．
6. ベッド上動作と除圧動作はすべて自立しておこなう．

(10) 移乗：ベッド―車椅子（車いすのブレーキ操作，フットレストの跳ね上げ，アームレストの脱着，乗り移り，足の持ち上げ）
0. 全介助を要する．
1. 部分介助，監視，および補助器具（例：スライディングボード）のすべて，もしくは，そのいずれかが必要．
2. 自立（または車椅子を必要としない）．

(11) 移乗：車椅子―トイレ，浴槽（トイレ用車椅子[4] を使用している場合は，それへの／からの乗り移り，通常の車椅子を使用している場合は，車椅子のブレーキ操作，フットレストの跳ね上げ，アームレストの脱着，乗り移り，足の持ち上げ）
0. 全介助を要する．
1. 部分介助，監視，および補助器具（例：手すり）のすべて，もしくは，そのいずれかが必要．
2. 自立（または車椅子は必要としない）．

移動（屋内と屋外，平らな所で）

(12) 屋内の移動
0. 全介助を要する．
1. 電動車碕子を必要とするか，または手動車椅子を操作するのに部分介助を要する．
2. 手動車椅子で自立して移動する．
3. 歩行時に監視を必要とする（器具の要否は問わない）．
4. 歩行器または松葉杖で歩行（大振り・小振り歩行）．
5. 松葉杖または T 字杖 2 本で歩行（交互歩行）．
6. T 字杖 1 本で歩行．
7. 下肢装具のみを必要とする．
8. 歩行補助異なしで歩行．

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(13) まとまった距離の移動 (10〜100m)
0. 全介助を要する.
1. 電動車椅子を必要とするか, または手動車椅子を操作するのに部分介助を要する.
2. 手動車椅子で自立して移動する.
3. 歩行時に監視を必要とする(器具の要否は問わない).
4. 歩行器または松葉杖で歩行(大振り・小振り歩行).
5. 松葉杖または T 字杖 2 本で歩行(交互歩行).
6. T 字杖 1 本で歩行.
7. 下肢装具のみを必要とする.
8. 歩行補助具なしで歩行.

(14) 屋外の移動 (100m 以上)
0. 全介助を要する.
1. 電動車椅子を使用するか, または手動車椅子を操作するのに部分介助を要する.
2. 手動車椅子を自立して移動する.
3. 歩行時に監視を必要とする(器具の要否は問わない).
4. 歩行器または松葉杖で歩行(大振り・小振り歩行).
5. 松葉杖または T 字杖 2 本で歩行(交互歩行).
6. T 字杖 1 本で歩行.
7. 下肢装具のみを必要とする.
8. 歩行補助具なしで歩行.

(15) 階段昇降
0. 階段の上り下り不能.
1. 人に支えられるか, またはその監視下で少なくとも 3 段は上り下りできる.
2. 手すりにつかまったり, 松葉杖や T 字杖を用いて少なくとも 3 段は上り下りできる.
3. 支えも監視もなくで少なくとも 3 段は上り下りできる.

(16) 移乗:車椅子一車 (車に近づく, 車椅子のブレーキ操作, アームレストおよびフットレストの取り外し, 車への／からの乗り移り, 車椅子の積み降ろし)
0. 全介助を要する.
1. 部分介助, 監視, および補助器具のすべて, もしくはいずれかを必要とする.
2. 自立して移乗する, 補助器具を必要としない(または車椅子を必要としない).

(17) 移乗:床一車椅子
0. 介助を要する.
1. 補助器具の有無にかかわりなく, 自立して移乗する(または車椅子を必要としない).

小計(0-40)

SCIM 合計スコア(0-100)

訳者注:
1. 夜間の BiPAP を使用している場合も 6 点とする
2. 原著では external drainage instrument. 体外からあてるさまざま芯様尿器集尿器のことを指す. 例えば, コンドーム型集尿器 , しびんなどが含まれる. 尿道や膀胱に挿入するカテーテルは含まれない
3. ベッド柵やひもなどを使ってもよい
4. 原著では toilet wheelchair. 便器や浴槽に移乗する ことなく, 乗車したまま洋式便器が使用でき, シャワー浴にも使える車椅子のこと. 介助型と自走型がある. わが国ではトイレ兼用シャワーキャリー(介助型)が用いられることがある.

SCIM (Version III): 日本語版. 岡川博之・黒川真希子・出田良輔・里宇明元訳
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190x254mm (300 x 300 DPI)

以下のそれぞれの項目の一つの四角に（このように☑）印をつけて、あなた自身の今日の健康状態を最も良く表している記述を示して下さい。

移動の程度

歩き回るのに問題はない ❑
歩き回るのにいくらか問題がある ❑
ベッド（床）に寝たきりである ❑

身の回りの管理

身の回りの管理に問題はない ❑
洗面や着替えを自分でするのにいくらか問題がある ❑
洗面や着替えを自分でできない ❑

ふだんの活動 *（例：仕事、勉強、家事、家族･余暇活動）*

ふだんの活動を行うのに問題はない ❑
ふだんの活動を行うのにいくらか問題がある ❑
ふだんの活動を行うことができない ❑

痛み／不快感

痛みや不快感はない ❑
中程度の痛みや不快感がある ❑
ひどい痛みや不快感がある ❑

不安／ふさぎ込み

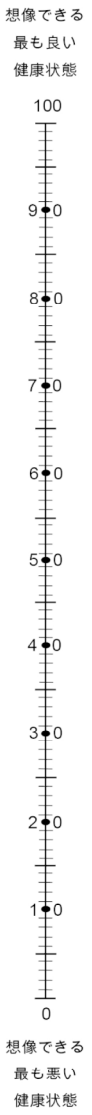
不安でもふさぎ込んでもいない ❑
中程度に不安あるいはふさぎ込んでいる ❑
ひどく不安あるいはふさぎ込んでいる ❑

190x254mm (300 x 300 DPI)

健康状態がどのくらい良いか悪いかを表わしてもらうために、（温度計に似たような）目盛を描きました。目盛には、あなたの想像できる最も良い状態として 100、あなたの想像できる最も悪い状態として 0 が付けられています。

あなたの今日の健康状態がどのくらい良いか悪いかを、あなたの考えでこの目盛上に示して下さい。下の「あなたの今日の健康状態」と書かれた四角から、あなたの現在の健康状態の良し悪しを示す目盛上の点まで、線を引いて下さい。

あなたの
今日の
健康状態



190x254mm (300 x 300 DPI)

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	3
	2b	All items from the World Health Organization Trial Registration Data Set	3
Protocol version	3	Date and version identifier	3
Funding	4	Sources and types of financial, material, and other support	15
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	1-2
	5b	Name and contact information for the trial sponsor	15
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	15
	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	13

Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	4-6
	6b	Explanation for choice of comparators	6
Objectives	7	Specific objectives or hypotheses	6
Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)	6-8
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	15
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	11
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	6-8
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	13
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	7
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	11
Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	8-10
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	Figure

Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	11-12
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	15
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	6
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	6
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	6-7
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	7, 11
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	11
Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	8-10
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	12-13

Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	13
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	12-13
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	12-13
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	12-13
Methods: Monitoring			
Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	13
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	N/A
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	13
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	Not in he protocol
Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	13-14
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	Patient consent form

Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	13
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	N/A
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	14
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	15
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	14
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	Patient consent form
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	14
	31b	Authorship eligibility guidelines and any intended use of professional writers	14-15
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	N/A
Appendices			
Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	Patient consent form
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	N/A

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.