June 19, 2024
Medical Device Evaluation Division
Pharmaceutical Safety Bureau
Ministry of Health, Labour and Welfare

Report on the Deliberation Results

Classification	Human Cellular/Tissue-based Products 2. Human Somatic Stem Cell-processed Products
Non-proprietary Name	Vandefitemcel
Brand Name	Akuugo Suspension for Intracranial Implantation
Applicant	SanBio Company Limited
Date of Application	March 7, 2022 (Application for marketing approval)

Results of Deliberation

In the meeting held on June 19, 2024, the Committee on Regenerative Medicine Products and Biotechnology reached the following conclusion, and decided that this conclusion should be presented to the Pharmaceutical Affairs Council.

The product may be approved. The conditional and time-limited approval is applicable to the product. The approval conditions and the duration of approval are as follows. The product should be designated as a specified regenerative medical product.

The following approval conditions must be satisfied.

Approval Conditions

1. In view of the limited manufacturing experience with the product, the applicant is required to promptly collect information about the product's quality according to the plan, evaluate quality comparability between the product and the study product, and report the results. Based on these results, the applicant should file necessary partial change application. The product must not be shipped before the approval of the partial change application.
2. The applicant is required to ensure that the product is used at medical institutions fully prepared for emergencies and by physicians with adequate knowledge and experience in the diagnosis and treatment of traumatic brain injuries and stereotactic brain surgery techniques who are also fully knowledgeable about the clinical study results and adverse events, etc. associated with the product.
3. During the period after the conditional and time-limited approval until the re-application for marketing authorization, the applicant is required to conduct a post-marketing approval condition assessment covering all patients treated with the product.

4. During the period after the conditional and time-limited approval until the re-application for marketing authorization, the applicant is required to collect information on biological characteristics that reflect the mechanism of action of the product, and take necessary measures such as improving the quality control strategy.

Duration of Approval

7 years

Review Report

March 15, 2024
Pharmaceuticals and Medical Devices Agency

The following are the results of the review of the following regenerative medical product submitted for marketing approval conducted by the Pharmaceuticals and Medical Devices Agency (PMDA).

Brand Name	Akuugo Suspension for Intracranial Implantation
Classification	Human Cellular/Tissue-based Products 2. Human Somatic Stem Cell-processed Products
Non-proprietary Name	Vandefitemcel
Applicant	SanBio Company Limited
Date of Application	March 7, 2022

Shape, Structure, Active Ingredients, Quantities, or Definition

The product consists of cells derived from human (allogeneic) bone marrow mesenchymal stem cells, into which the human Notch-1 intracellular domain gene has been transfected using a pN-2 plasmid vector.

Application Classification	(1-1) New regenerative medical product
Items Warranting Special Mention	Orphan regenerative medical product (Orphan Regenerative Medical Product Designation No. 19 of 2020 [*R2 sai*]; PSEHB/MDED Notification No. 0623-4 dated June 23, 2020, by the Medical Device Evaluation Division, Pharmaceutical Safety and Environmental Health Bureau, Ministry of Health, Labour and Welfare) SAKIGAKE designation regenerative medical product (SAKIGAKE Regenerative Medical Product Designation No. 2 of 2018 [*30 sai*]; PSEHB/MDED Notification No. 0408-17 dated April 8, 2019, by the Medical Device Evaluation Division, Pharmaceutical Safety and Environmental Health Bureau, Ministry of Health, Labour and Welfare), Sakigake comprehensive assessment consultation conducted for regenerative medical products
Reviewing Office	Office of Cellular and Tissue-based Products

Results of Review

The decision on the approval of the product should be made based on additional evaluation results on the comparability between the study product and the product (see Attachment).

PMDA has concluded that when the comparability between the study product and the product is proven, the product will have a certain level of efficacy in improving chronic motor paralysis associated with traumatic brain injury, and that the product has acceptable safety in view of its benefits. Despite limited efficacy and safety information of the product, it is of significance to provide the product to clinical settings as one of the treatment options for improving chronic motor paralysis associated with traumatic brain injury.

Attachment

Review Report (1)

January 26, 2024

The following is an outline of the data submitted by the applicant and content of the review conducted by the Pharmaceuticals and Medical Devices Agency (PMDA).

Product Submitted for Approval

Brand Name	Akuugo Cell Suspension for Intracranial Implantation
Classification	Human Cellular/Tissue-based Products 2. Human Somatic Stem Cell-processed Products
Non-proprietary Name	Vandefitemcel
Applicant	SanBio Company Limited
Date of Application	March 7, 2022

Shape, Structure, Active Ingredients, Quantities, or Definition

The product consists of cells derived from human (allogeneic) bone marrow mesenchymal stem cells, into which the human Notch-1 intracellular domain gene has been transfected using a pN-2 plasmid vector.

Proposed Indication or Performance

Improvement of motor deficit after traumatic brain injury

Proposed Dosage and Administration or Method of Use

Usually, the cell suspension containing modified human (allogeneic) bone marrow-derived mesenchymal stem cells is prepared to contain 5×10^6 viable cells per dose (1.67×10^6 cells/100 μL), which is then transplanted to the surrounding area of the damaged tissue via stereotactic brain surgery.

1. Method of Use

See the manuals, etc. provided by the marketing authorization holder for detailed procedure.

1-1. Setup of invasive cranial fixation devices for neurosurgery

Before starting the surgical procedure using Akuugo, attach the guide and stop to the invasive cranial fixation device for neurosurgery. See the operating instructions for the detailed use of the accompanying invasive cranial fixation devices.

2-1. Preparation of cell suspension

(1) Thaw the cell suspension for intracranial transplantation (the primary component) that has been stored in the vapor phase of liquid nitrogen (≤–150°C). Add the dedicated preparation solution and stir gently.
(2) Centrifuge the obtained cell suspension. After removing the supernatant, add the dedicated preparation solution again to wash the suspension.

(3) Add the dedicated preparation solution and stir the suspension gently to repeat the procedure (2).
(4) After washing, count the viable cells and adjust the cell concentration using the dedicated preparation solution so as to obtain cell suspension at the concentration of 1.67×10^6 cells/100 μL for transplantation.

2-2. Preparation of administration cannula

(1) Secure the administration cannula to the microsyringe and cleanse both the microsyringe and the administration cannula with the dedicated preparation solution.
(2) Fill the microsyringe with the cell suspension from the plunger insertion side using the micropippeter.

2-3. Transplantation of Akuugo

(1) Using MRI images, etc., determine the site for 1 small opening on the skull as the starting point of 3 transplant routes that lead to the area surrounding the damage. Create a small opening on the skull as determined.
(2) Insert the stylet-equipped inserter through the small opening toward one of the transplant routes.
(3) Remove the stylet. Attach the cannula to the microsyringe filled with 100 μL of the cell suspension, and transplant it at 5 points 5 to 6mm apart, from the deepest point. The cell suspension of approximately 20 μL should be injected per point at approximately 10 μL/min.
(4) Take the same procedure for the remaining 2 transplant routes.

Table of Contents

List of Abbreviations

See Appendix.

1. Origin or History of Discovery, Use in Foreign Countries, and Other Information

1.1 Outline of the proposed product

Akuugo is a combination product consisting of a primary component and secondary components. The primary component is a human (allogeneic)-derived intracranial transplant cell suspension (SB623) containing mesenchymal stem cells (MSCs) that is produced from bone marrow aspirate of human healthy adults, cultured for isolation and proliferation, transfected with a plasmid vector encoding the intracellular domain of human Notch-1. The secondary components include a dedicated preparation solution used to prepare the transplant cell suspension and a dedicated delivery device set (comprising [1] microsyringe, [2] administration cannula, [3] inserter, [4] stylet, and [5] guide & stop). Akuugo is expected to promote the repair of neural cells through the proliferation and differentiation of endogenous neural stem cells, angiogenesis, and immunomodulation, etc. by secreting cytokines.

Akuugo was designated as an orphan regenerative medical product with the intended indication or performance for "improvement of sequelae in traumatic brain injury" dated June 23, 2020 (Orphan Regenerative Medical Product Designation No. 19 of 2020 [*R2 sai*], PSEHB/MDED Notification No. 0623-4). Akuugo is also designated as a regenerative medical product with the intended indication or performance for "improvement of motor deficit in moderate to severe traumatic brain injury" to be reviewed under the SAKIGAKE Designation system (SAKIGAKE Regenerative Medical Product Designation No. 2 of 2018 [*30-sai*]) dated April 8, 2019.

1.2 Development history etc.

Traumatic brain injury (TBI) is caused by physical impact to the head, such as in traffic accidents, and leads to motor deficit (e.g., difficulties in walking and coordination) and cognitive impairments (e.g., declines in communication and memory) (*Standard Neurosurgery* [in Japanese], 14th Edition. Igaku-Shoin; 2017.263-91, etc.). There is no curative treatment for TBI, while rehabilitation is provided to address sequelae. However, motor deficit stabilizes approximately 6 months after injury, and no further improvement is observed after ≥12 months post-injury (*Brain Inj.* 2015;29:1431-8, *J Rehabil Res Dev.* 2007;44:975-82, etc.). The 2020 patient survey by the Ministry of Health, Labour and Welfare reported that a total of 1,900 patients in Japan suffer from sequelae or complications from intracranial injuries.

The transplantation of bone marrow-derived MSCs into TBI model rats contributes to the recovery of neurological function after TBI (*Neurosurgery.* 2003;53:697-702). In the brain post-TBI, bone marrow-derived MSCs increases the production of humoral factors such as brain-derived neurotrophic factor (BDNF), and pharmacological actions mediated by these humoral factors may play an important role in the recovery process of neural tissue post-TBI (*J Neurosci Res.* 2002;69:687-91). SB623 is produced by expressing the intracellular domain of Notch-1, one of the transmembrane receptors in the Notch family, through genetic transfection. When Notch-expressing cells are in proximity to ligand-expressing cells, Notch-1, stimulated by the ligand, undergoes peptide cleavage, releasing its intracellular domain into the cytoplasm. This domain then moves into to the nucleus, thereby binds to RBP-J, etc. and activates transcription factors for multiple genes, leading to changes in the expression profile of secreted trophic factors, chemotactic factors, and extracellular matrix proteins (*Cell.* 2009;137:216-33). Thus, SB623 is expected to activate Notch signaling by expressing the intracellular domain of Notch-1, enhancing the

neurorestorative effects of MSCs on the damaged areas of the brain without the need for extracellular ligand stimulation.

A foreign multicenter, sham surgery-controlled, randomized, double-blind study (Study TBI-01) of Akuugo began in July 2016 targeting patients with chronic motor deficit from TBI.

In Japan, patient enrollment for Study TBI-01 started in [redacted], 20[redacted].

Recently, an application for marketing approval (application) of Akuugo has been submitted, based on the data from Study TBI-01 as the pivotal study.

As of December 2023, Akuugo has not been approved or marketed in any country or region.

Akuugo has been designated as a regenerative medical products under the SAKIGAKE Designation system, and PMDA expedited the review process. As a rule, SAKIGAKE-designated products are subjected to the "SAKIGAKE comprehensive assessment consultation" (quality, non-clinical, clinical, reliability, and Good Gene, Cellular, and Tissue-based Products Manufacturing Practice [GCTP]) to reduce the time taken for the processes from application to approval. Akuugo underwent these SAKIGAKE comprehensive assessment consultations, which concluded by pointing out some issues to be addressed before application. However, the following quality and GCTP-related issues still remained when the application for Akuugo was submitted.

- During the "SAKIGAKE comprehensive assessment consultation" (GCTP) (Reception Number: Saisen [redacted], Application Confirmation Document: PMDA/CPE Notification No. [redacted], dated [redacted], 20[redacted]), foreign matter contamination was identified in SB623, highlighting the need of a foreign matter control strategy to prevent contamination. However, the applicant filed the application for Akuugo without duly addressing the issue, and the foreign matter control strategy, involving changes in the manufacturing process including the change of the primary container, one of the cause of the contamination, was established after the application. The verification of the foreign matter control strategy at the actual manufacturing scale began in July 2022, approximately 3 months after the submission of application.

While the aforementioned actions contributed to the reduction of foreign matter contamination to a certain degree, the production yield continued to decrease significantly. As a result of process validation aiming to achieve a target minimum yield equivalent to that before application, including that achieved by the study product, the [redacted]th production post-application achieved a yield comparative to that before application [see Section 2.5]. Specifying this manufacturing process as commercial process, additional quality test results, which included the comparability between the product manufactured by the previously proposed process and the product manufactured by the commercial process, were submitted on November 28, 2023, approximately 1 year and 9 months after the application, causing a significant delay in the review schedule.

Considering it an unusual case, PMDA attributed it to the applicant's lack of awareness of critical matters that ensure the quality, safety, and efficacy of the product.

2. Quality and Outline of the Review Conducted by PMDA

Akuugo is a combination product consisting of the following components:

- Primary component: SB623, which is MSCs isolated and expanded through culture from the bone marrow fluid of healthy adults, transfected with a plasmid vector encoding the intracellular domain of human Notch-1.
- Secondary component 1: A dedicated preparation solution used for cell suspension preparation for transplantation
- Secondary component 2: A dedicated delivery device set (Figure 1) consisting of (1) a microsyringe made of stainless steel, polytetrafluoroethylene, fluororubber, and borosilicate glass; (2) an administration cannula, (3) an inserter, and (4) a stainless steel stylet; and (5) a guide & stop made of polyphenylsulfone resin.

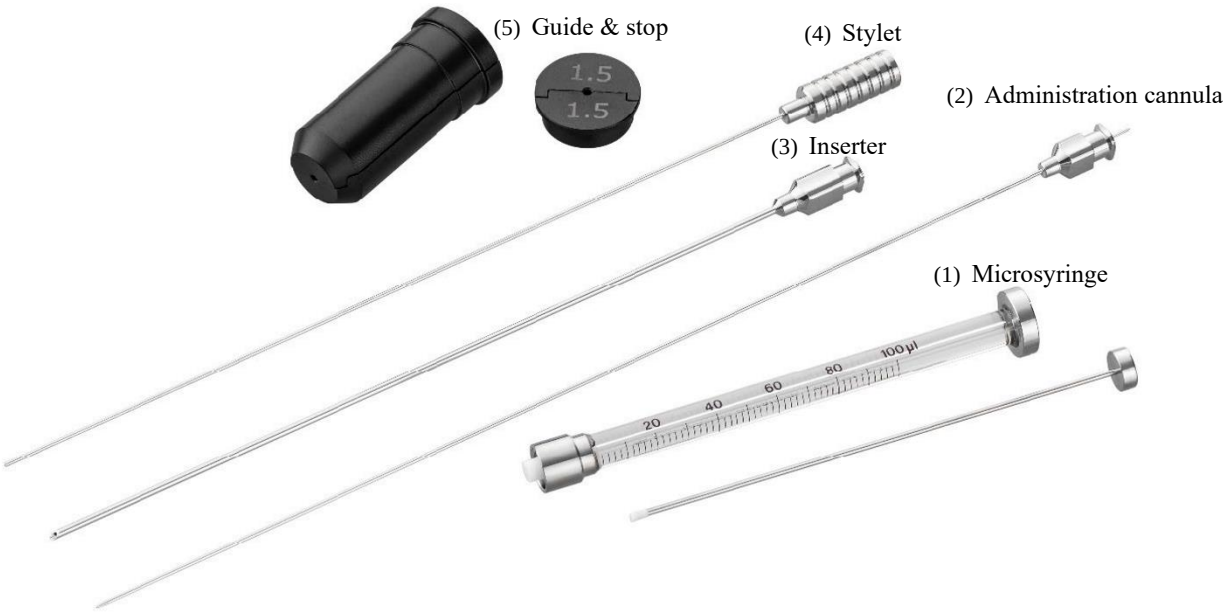


Figure 1. External view of the dedicated delivery device set

2.1 Manufacturing process

The primary component (SB623) is a cell suspension containing 12.5 × 10^6 live cells (1 mL) with a sub-component of cryopreservation solution containing 5% dimethyl sulfoxide ([redacted]). The suspension is filled in 2-mL [redacted] vials.

The manufacturing process includes [redacted], [redacted], [redacted], [redacted], [redacted], [redacted], [redacted], [redacted], [redacted], testing, packaging, and packaging/labeling/storage 1.

The critical steps are [redacted], [redacted], [redacted], [redacted], [redacted], [redacted], and [redacted].

The cells obtained by [redacted] are the critical intermediate, and are cryopreserved at temperatures ≤ [redacted]°C.

Secondary component 1 (dedicated preparation solution) is an electrolyte-containing solution filled in a [redacted] bag (100 mL).

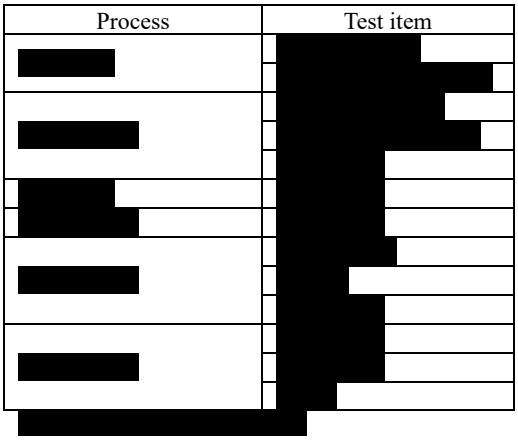
The manufacturing process involves packaging/labeling/storage 2.

Manufacturing processes of secondary component 2 (dedicated delivery device set) at the manufacturing site of Akuugo are packaging/labeling/storage 3.

2.1.1 In-process control tests

Table 1 shows the in-process control tests in the manufacturing process of the primary component.

Table 1. In-process control tests in the manufacture process of the primary component (SB623)



2.2 Safety evaluation of adventitious agents

The primary component of Akuugo contains the following human- and animal-derived materials. The secondary components do not contain any human- or animal-derived materials.

2.2.1 Bone marrow fluid

The bone marrow fluid used as raw material is collected from healthy adult male donors in the US and complies with the Standards for Biological Raw Materials (MHLW Public Notice No. 210, 2003). Donor eligibility is confirmed by medical history, lifestyle and behavioral risk questionnaire, as well as testing of the donor's blood sample for viruses such as human immunodeficiency virus (HIV)-1, HIV-2, hepatitis B virus (HBV), hepatitis C virus (HCV), human T-cell leukemia virus (HTLV)-1, HTLV-2, Treponema pallidum, cytomegalovirus (CMV), West Nile virus (WNV), Epstein-Barr virus (EBV), and parvovirus B19 (PVB19). Following donor qualification, bone marrow aspiration is performed within [redacted] days, and the collected bone marrow fluid is transported to the manufacturing site at [redacted]°C, ensuring that the initial [redacted] processing by [redacted] is completed within [redacted] hours of collection. A follow-up re-examination, considering the window period, is conducted 75 to 90 days after the initial blood draw for donor screening.

The bone marrow fluid contains heparin sodium injection as anticoagulant that has been approved in [redacted] for marketing.

2.2.2 Biological raw materials other than bone marrow fluid

The fetal bovine serum (FBS) (country of origin: [redacted], [redacted], and [redacted]) and porcine pancreas-derived trypsin used in the manufacturing process both comply with the Standards for Biological Raw Materials.

[redacted], one of the gene transfection reagents, contains bovine histone below ** w/v%. The bovine histone derives from healthy cows in the US, and is thymus-origin, the use of which as a raw material of pharmaceuticals, etc. has been banned by the Standards for Ruminant-derived Raw Materials (1) in Standards for Biological Raw Materials. The bovine spongiform encephalopathy (BSE) risk assessment and viral safety evaluation on [redacted] are provided in Section 2.R.1.

2.3 pN-2 Plasmid vector

The pN-2 plasmid vector is manufactured at [redacted], using a pCI-Neo mammalian expression plasmid vector. The Notch-1 intracellular domain [redacted] was amplified from [redacted] prepared from [redacted] extracted from [redacted], and inserted into the pCI-Neo plasmid vector to produce the pN-2 plasmid vector.

The control parameters for [redacted] include description, pH, [redacted], [redacted], sterility testing, bacterial endotoxin testing, DNA concentration, [redacted], [redacted], [redacted], [redacted], and [redacted]. The pN-2 plasmid vector is stored at [redacted]°C [redacted]°C and must be used within [redacted] months.

2.4 Critical intermediate

The quality and storage method of the cells to be used for gene transfection are highly likely to affect the quality of the final product. In this view, the cells obtained in [redacted] are determined as the critical intermediate. These cells obtained in [redacted] are suspended in [redacted] at a concentration of [redacted] cells/mL, filled at [redacted] mL per [redacted], and frozen at ≤[redacted]ºC. The control parameters for the critical intermediate are listed in Table 2. After culturing in [redacted], these cells are used for gene transfection.

Table 2. Specifications for the critical intermediate

Test item		Test method	Specification/ acceptance criteria
Identification	Cell surface marker analysis [redacted]	Flow cytometry	≥ [redacted]%
			≥ [redacted]%
			≥ [redacted]%
			≤ [redacted]%
			≤ [redacted]%
			≤ [redacted]%
Cell quality test*1	Viability	Automated cell counting	≥ [redacted]%
	Viable cell count		≥ [redacted] cells/mL
	[redacted]	[redacted]	≥ [redacted] vials
	[redacted]*2	[redacted]	≥ [redacted]
Safety tests	Sterility	Rapid sterility test (BacT/ALERT method)	No growth detected
	Test for mycoplasma*1	Culture and DNA staining methods (USP 63 and EP 2.6.7)	Not detected
	Virus-related tests*1	*In vitro* tests (indicator cells: MRC-5, Vero 76, HeLa)	Not detected
		qPCR method (HIV-1, HIV-2, HTLV-1, HTLV-2, HBV, HCV, CMV, EBV-1, EBV-2, PVB19, JCV)	Not detected
	Viral test of bone marrow fluid donor*3	Confirmation of the test results form	Not detected

*1 [redacted]
*2 Added after submission of application.
*3 In view of a window period, the second virus screening is performed using blood newly drawn from the donor 75 to 90 days after the initial blood collection for the first screening. The virus screening items are the same as those in the first screening [see Section 2.2.1].

The stability of the critical intermediate is described in Section 2.R.2.

2.5 Development of the manufacturing process (comparability)

Table 3 shows the main changes in the manufacturing process of the primary component (SB623) during development. The change from the proposed process to the commercial process occurred after the application, prompted by the development of the foreign matter control strategy to prevent contamination [see Section 1.2]. In conjunction with these manufacturing process changes, comparability assessment of the quality attributes was conducted.

Table 3. Main changes in manufacturing process

Manufacturing process change	Changes
Process A to B	[redacted] • Container and closure system
Process B to B’	[redacted]
Process B’ to proposed process	[redacted]
Proposed process to commercial process	[redacted] • Container and closure system

Table 4 shows the manufacturing processes for the primary component (SB623) used in non-clinical and clinical studies.

Table 4. Manufacturing process of the primary component (SB623) used in each study

Manufacturing process	Manufacturing site	Gene transfection reagent	Batch No*	Use
Process A	[redacted]	[redacted]	[redacted]	Study STR01 Pharmacological study Non-clinical safety study Stability study
Process B	[redacted]	[redacted]	[redacted]	Study STR02 Study TBI-01 Pharmacological study Stability study
Process B'	[redacted]	[redacted]	[redacted]	Study TBI-01 Stability study
Proposed process	[redacted]	[redacted]	[redacted]	Study of process parameters, etc. Stability study
Commercial process	[redacted]	[redacted]	[redacted]	Process verification

* [redacted]

The development of the commercial process after the application is detailed in Tables 5 and 6 below. Unless otherwise specified, "actions taken" were also taken as necessary in subsequent manufacturing operations.

Table 5. History of commercial process development after submission of application

Manufacturing No.	Critical intermediate	Evaluation results and responses
[redacted]	[redacted]	Evaluation results • [redacted]: The yield of the final product significantly decreased, and the [redacted] of [redacted] exceeded the upper limit, resulting in non-conformity. The likely reason for this non-conformity was the increase in [redacted] contained in [redacted] due to the reduced [redacted] during the cultivation process after the [redacted] process. The foreign matter control strategy included the change of the primary container (such as changing the [redacted] and the [redacted] of the cap), but [redacted], [redacted], or [redacted] was not implemented during production. • [redacted]: Although all foreign matter control strategies, including [redacted], were implemented during production, the reduction of foreign matter contamination could not be evaluated due to the low yield of the final product. Actions taken • Because the prolonged [redacted] and decreased yield of the final product showed [redacted], [redacted] was determined. Previously, while it was determined that [redacted], [redacted] was not determined. [redacted] was [redacted] in [redacted] the mean [redacted]. • Other actions: ➢ [redacted] was [redacted], resulting in [redacted] in the cultivation. [redacted] was incorrectly [redacted], so [redacted] was performed. ➢ During the [redacted] of [redacted], [redacted] caused the [redacted] to exceed [redacted], and [redacted] separating [redacted] and [redacted] became non-functional and [redacted]. Although [redacted] was attempted, [redacted] was not available. While [redacted], [redacted] was left [redacted] for [redacted] minutes. Considering this could have been due to different [redacted], [redacted] including [redacted] was conducted.
[redacted]	[redacted]	Evaluation results Due to the low [redacted], production was halted. Actions taken • [redacted]: After evaluating the cause of the production halt, it was concluded that the reduced [redacted] and the extended [redacted] were the primary factors. By changing [redacted] from [redacted] to [redacted], the [redacted] was shortened to [redacted] minutes. [redacted] was reverted to the original process submitted in the application, and [redacted]. • Other actions: ➢ Production was [redacted] by [redacted]. ➢ Thorough [redacted] of [redacted] and [redacted] was required before use.

[redacted]	[redacted]	Evaluation results Although [redacted] improved, the final product yield did not improve. Based on the foreign material control strategy review, foreign material contamination was detected in [redacted] out of [redacted] vials of the final product. Actions taken • [redacted] was changed to [redacted]. Multiple batches failed to secure sufficient cell count at the start of [redacted] after manufacturing by [redacted], [redacted] was increased to ensure a higher cell count at the start of [redacted]. • [redacted] with the increase in [redacted], the number of usable [redacted] was increased. • Selection of [redacted]: A study on the relationship between the quality of [redacted] and the final product yield suggested a correlation between [redacted] of [redacted] and the yield of the final product. Among [redacted], the batch number [redacted] ([redacted]), which was considered to be [redacted], was used for production. • Other actions: ➢ The optimal criteria for [redacted] were confirmed by the applicant and the manufacturer.
[redacted]	[redacted]	Evaluation results • There was no improvement in the final product yield. In addition, [redacted] following [redacted] was observed, and the [redacted] of [redacted] exceeded [redacted], resulting in non-conformity. • Upon the review of the foreign matter control strategy, foreign matter contamination was detected in [redacted] of [redacted] vials. Actions taken • Setting of [redacted]: Based on the evaluation of the cause of [redacted], it was determined from the production history of the final product using [redacted] that the final product yield tended to decrease with increased duration of [redacted]. Therefore, the [redacted] was used. • Other actions: ➢ [redacted] and [redacted] were increased to monitor the cultivation conditions. ➢ Parallel operations were implemented to shorten the overall process time.
[redacted]	[redacted]	Evaluation results • The final product yield improved with the production using [redacted]. • Foreign material contamination was observed in [redacted] of [redacted] vials of the final product. Actions taken Including [redacted], the manufacturing process was considered to have been optimized. In terms of the foreign matter control strategy, 3 batches ([redacted], [redacted], and [redacted]) showed the percentages of contaminated vials that were low enough for uninterrupted supply. Accordingly, the following changes were made in the proposed process to finalize the commercial production process: • In-process control tests: [redacted] and [redacted] were added (see Table 1). The specifications for the critical intermediate: [redacted] was added. Critical intermediate with [redacted] was to be used in production (see Table 2). • [redacted] method: [redacted] were changed to [redacted]. [redacted] until [redacted] was confirmed. • [redacted] was changed from [redacted] to [redacted]. • Expansion of [redacted]: [redacted] was increased. • Container and closure system: Modifications to the primary container (change of [redacted] and [redacted] of the cap, etc.).

Table 6. Production performance after the proposed process

	Item	Control values/ specification limits/ acceptance criteria	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
Specifications	Description	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
	pN-2 plasmid copy number	[redacted] (copy per haploid genome)	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
	Cell surfaces marker analysis [redacted]	≥[redacted]%	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
		≥[redacted]%						
		≥[redacted]%						
		≤[redacted]%						
		≤[redacted]%						
		≤[redacted]%						
	Viability	≥[redacted]%	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
	Viable cell count	≥[redacted] cells/vial	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
	[redacted]	≥[redacted]%	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
	Intracellular FGF-2	≥[redacted] molecules	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
	Sterility	No growth detected	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
	Bacterial endotoxin	≤[redacted] EU/mL	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
	Mycoplasma testing	Not detected	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]		[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]

*1 [redacted]
*2 [redacted]

The development of the manufacturing process and the evaluation of comparability are described in Section 2.R.2.

2.6 Evaluation of manufacturing process

2.6.1 Gene transduction and selective culture

In the manufacturing process of the primary component (SB623), gene-transfected cells, which are the target cells, are selected using [redacted].To confirm that only pN-2 plasmid-transfected cells are properly selected, [redacted] for each production and subjected to evaluations by [redacted] and [redacted]. [redacted] was evaluated in the study products used in Study TBI-01 ([redacted] and [redacted]), the proposed process formulation ([redacted]), and the commercial process formulation ([redacted]), which revealed [redacted] except for the [redacted] of the proposed process. The [redacted] of the proposed process became [redacted] at the end of [redacted], and no [redacted] was observed until the end of [redacted]. Since the remaining [redacted] showed no [redacted], it is expected to [redacted] the subsequent [redacted].

Based on the above, the majority of non-transfected cells perish, and [redacted].

2.6.2 Removal of process-related impurities

Impurity risk assessment was conducted on process-related impurities based on the expected maximum transplantation volume of impurities, calculating from the amount of the media used in the manufacturing process (Impurity A, Impurity B, and FBS), Impurity C, trypsin, pN-2 plasmid, Impurity

D, and Impurity E. The estimated exposure levels, which were calculated from measured or estimated residual amounts of each impurity in the cell suspension for transplantation, indicated low safety concerns in humans. Thus, no control items were specified for process-related impurities.

2.6.3 Implementation of verification

The quality attributes required for Akuugo include description, cell surface markers, pN-2 plasmid copy number, viable cell count, viability, [redacted], intracellular fibroblast growth factor (FGF)-2, and sterility, etc.

Because only one batch of the primary component (SB623) were produced by the commercial process, process variability factors have not yet been clearly identified. However, a quality control strategy has been established, taking into account the potential quality risks arising from variability in the quality attributes of the bone marrow fluid, which serves as the raw material, so that the desired quality attributes are achieved in every production. The quality control strategy involves verification including the following:

- In-process control tests (Table 1)
- Manufacturing process parameters (critical process parameter [CPP]/potential critical process parameter [pCPP])
- Specifications for critical intermediate (Table 2)
- Specifications for the primary component (SB623) (Table 9)
- Characterization (Table 7)

Table 7. Characterization tests conducted for verification

Test	Acceptance criteria
[redacted]	[redacted]
[redacted]	[redacted]

2.7 Characterization

The primary component (SB623) was subjected to characterization as shown in Table 8.

Table 8. Parameters for characterization of the primary component (SB623)

Characteristics	Parameters
Physicochemical	Cell surface marker analysis, pN-2 plasmid copy number, potency (intracellular FGF-2), process-related impurities, [redacted]
Biological	[redacted]

2.8 Control of product

Table 9 shows the specifications for the primary component (SB623).

Table 9. Specification for the primary component (SB623)

Test		Test method	Specification limits/acceptance criteria
Description		[redacted]	[redacted]
Identification	pN-2 plasmid copy number*	[redacted]	[redacted]
	Cell surface marker analysis	Flow cytometry	[redacted] ≥ [redacted]%
			[redacted] ≥ [redacted]%
			[redacted] ≥ [redacted]%
			[redacted] ≤ [redacted]%
			[redacted] ≤ [redacted]%
			[redacted] ≤ [redacted]%
Cell quality test	Viability	Automated cell counting	≥ [redacted]%
	Viable cell count		≥ [redacted] cells/vial
	[redacted]		≥ [redacted]%
Potency	Intracellular FGF-2	[redacted]	≥ [redacted] molecules
Safety test	Sterility	Sterility test (direct method [JP])	No detectable growth
	Bacterial endotoxin	Bacterial endotoxin test (chromogenic technique [JP])	≤ [redacted] EU/mL
	Mycoplasma test	Nucleic amplification test (General Information of JP)	Undetectable

* [redacted]

The proposed control items for secondary component 1 (dedicated preparation solution) include description, identification ([redacted]), osmotic ratio, pH, purity ([redacted]), bacterial endotoxin, extractable volume, foreign insoluble matter, insoluble particulate matters, sterility, and assay ([redacted]).

Table 10 shows performance and safety specifications for the secondary component 2 (dedicated delivery device set).

Table 10. Performance and safety specifications for the secondary component 2 (dedicated delivery device set)

	Secondary components	Test items
Performance Specification	Microsyringe	Shape dimensions, graduated capacity, airtightness, thermal shock, pressure resistance, extraction strength
	Administration cannula	Corrosion resistance, blade edge, fitting, size, leakage, extraction strength
	Inserter	Corrosion resistance, tip, size, junction strength between needle tube and needle hub
	Stylet	Corrosion resistance, size, extraction strength
	Guide & stop	Size
Safety specification	All	Biological safety,* sterility assurance (Sterility Assurance Level [SAL]: 10^{-6}), sterilization residues (JIS T 0993-7: 2012 "Temporary Contact Devices")

* Guide & stop is excluded as it does not come into contact with blood, body fluids, or tissues

2.9 Stability of product

Table 11 shows the outline of the stability study for the primary component (SB623) submitted with the application.

Table 11. Outline of the long-term stability testing for the primary component (SB623)

Number of batches	Manufacturing process	Storage condition	Study period	Storage form	Vial volume
3	Proposed process	In the vapor phase of liquid nitrogen (°C)	months (the stability study is planned to be continued up to months)	vial with cap	mL or mL

The long-term stability testing showed no significant changes in the quality attributes in any of the samples throughout the study period. The shelf life of months was proposed for the primary component (SB623) when stored in liquid nitrogen vapor (≤ °C) using vials.

Using the primary component produced by the proposed process, to be transplanted were evaluated. No significant changes were noted at room temperature for at least hours after preparation. Even when aspirating the cell suspension for transplantation using the microsyringe and letting it pass through the cannula attached to it at a predetermined rate, no significant changes were noted in . The same was true with cells that had been left at room temperature for 3 hours after preparation.

Based on the above, the cell suspension for transplantation should be used within 3 hours after preparation when stored at room temperature.

Table 12 is the summary of the long-term testing for secondary component 1 (dedicated preparation solution).

Table 12. The summary of the main stability studies for secondary component 1 (dedicated preparation solution)

Study	Number of batches	Storage condition	Study period	Storage form
Long-term testing	3	°C, % relative humidity	months	Bag with port Rubber cap: Cap (plug):
Accelerated testing	3	°C, % relative humidity	months	
Photostability testing	1	≥ lx•h, and integrated near ultraviolet energy of ≥ W•h/m^2		

In the long-term testing, no significant changes were observed in quality attributes in any of the samples throughout the study period. Similarly, in the accelerated testing and the photostability testing, no significant changes were observed in quality attributes in any of the samples during the study period. Based on these findings, the shelf life of 36 months was proposed for secondary component 1 (dedicated preparation solution) when stored at room temperature in bags with port, rubber stoppers, and caps.

The secondary component 2 (dedicated delivery device set) was subjected to a long-term testing for stability under conditions of °C and % relative humidity. No changes in quality were noted in the samples after 24 months. The shelf life of 24 months was proposed for secondary component 2 (dedicated delivery device set) when stored at room temperature.

2.R Outline of the review conducted by PMDA

Based on the submitted data and the following review results, PMDA concluded that the quality of the product, except for the matters related to Section 2.R.2 of the primary component (SB623), is appropriately controlled. The quality of secondary component 2 (dedicated delivery device set) was reviewed by Office of Medical Devices II, and it was confirmed that there were no specific issues identified.

2.R.1 BSE risk regarding bovine histones contained in [redacted]

The applicant's explanation about the evaluation of the BSE risk assessment of bovine histones contained in [redacted], a gene transfection reagent:

The histones in [redacted] are derived from bovine thymus, the use of which is prohibited as a pharmaceutical raw material under the Standards for Ruminant-derived Raw Materials (1) in Standards for Biological Raw Materials. The applicant is currently considering switching to a gene transfection reagent that complies with the Standards for Biological Raw Materials, and there are [redacted] chemically synthesized gene transfection reagents as candidates. This change, however, is difficult at this point, and there is no alternative but to use [redacted]. The theoretical risk of BSE was assessed based on the "BSE risk evaluation of drugs in view of the confirmed BSE cases in Canada" (Transmissible Spongiform Encephalopathy Investigation Committee, dated July 8, 2003) attached to "Handling of Risk Evaluation, etc. in Partial Change Applications for Pharmaceuticals, Medical Devices, etc. Using Bovine-derived Raw Materials (in Japanese)" (PFSB/ELD Notification No. 0801001 and PFSB/SD Notification No. 0801001, dated August 1, 2003). The applicant considers that the BSE risk is low and the use of [redacted] for the production of Akuugo is acceptable. Meanwhile, the patient will be informed of the use of ruminant-derived raw materials not conforming to Standards for Biological Raw Materials, and the product will be administered only with the patient's consent.

PMDA's view:

While the BSE risk associated with bovine histones contained in [redacted] cannot be completely ruled out, the risk is expected to decrease through the manufacturing process, etc. Given the current difficulty in switching to a gene transfection reagent conforming to the Standards for Biological Raw Materials, the use of non-conforming materials is considered inevitable for Akuugo production. At the same time, the use of raw materials conforming to the Standards for Biological Raw Materials is more appropriate. Considering that the risk should be eliminated by switching to a conforming gene transfection reagent, PMDA instructed the applicant to make this change as soon as possible.

2.R.2 Manufacturing process of the primary component (SB623)

The applicant's explanation about the manufacturing process of the primary component (SB623):

In response to PMDA's advice on a foreign matter control strategy for anti-contamination, the applicant identified the cause of contamination, planned an assessment to build a foreign matter control strategy, and filed the application, on the premise that evaluation data would be presented after the filing. However, at data acquisition, the yield of the final product significantly dropped, thus the applicant had to not only assess the foreign matter control strategy but also establish a manufacturing process. Tables 5 and 6 show the development history of the commercial process after the application.

The main changes between the proposed process and the commercial process are as follows:

- In-process control tests: [redacted], [redacted], etc., were added (see Table 1).
- Specifications for critical intermediates: [redacted] was added. A critical intermediate with [redacted] was used in the manufacturing process (see Table 2).
- [redacted]: Changed to [redacted], continuing [redacted] until [redacted] is confirmed.
- Changed from [redacted] to [redacted].
- [redacted]: Increased [redacted].
- Container and closure system: Changed the primary container (changes in [redacted] and [redacted] of the cap, etc.).

In view of significant decrease in cell yield in 5 consecutive batches manufactured after the application, PMDA asked the applicant to explain the comparability among the study product used in Study TBI-01, the formulation manufactured by the proposed process, and the formulation manufactured by the commercial process.

Comparability assessment, potency testing, and verification

The applicant's explanation about the comparability of the primary component (SB623) following the manufacturing process change:

Based on the results in Table 13, the commercial process formulation [redacted] is considered comparable to the study product used in Study TBI-01 ([redacted] manufactured using Process B and [redacted] manufactured using Process B'), as no differences were observed in (a) specification tests, (b) characterization tests, or (c) in-process control tests. Also, based on the results in Table 14, the commercial process formulation [redacted] is considered to be comparable to the proposed process formulation, because data on (a) specification tests, (b) characterization tests, or (c) in-process control tests in Table 14 fell within the range of that of the proposed process formulation.

Cell proliferation characteristics in each manufacturing process were assessed based on (d) the [redacted] of the critical intermediate and (e) [redacted] after thawing of the critical intermediate were evaluated. (d) The [redacted] of the critical intermediate was [redacted] in the manufacture of the commercial process formulation [redacted] and fell within the range of [redacted] in the manufacturing record of the study product and proposed manufacturing process, indicating the comparability. Furthermore, (e) [redacted] after thawing of the critical intermediate was [redacted] in the manufacture of the commercial process formulation [redacted] and fell within the range ([redacted]) of [redacted] in the manufacturing record of the study product and the proposed process formulation, excluding the [redacted] that did not meet the predetermined criteria of [redacted], thus considered comparable.

Table 13. Comparability assessment between the commercial process and the process used in the manufacture of the study product for Study TBI-01 (parameters after the thawing of the critical intermediate)

Test	Control values/ specification/acceptance criteria	Commercial process	Process B	Process B'
(a) Specifications				
Description				
pN-2 plasmid copy number	(copy per haploid genome)			
Cell surface marker analysis	≥ %			
	≥ %			
	≥ %			
	≤ %			
	≤ %			
	≤ %			
Viability	≥ %			
Viable cell count	≥ cells/vial			
	≥ %			
Intracellular FGF-2	≥ molecules			
Sterility	No detectable growth			
Bacterial endotoxin	≤ EU/mL			
Mycoplasma test	Not detected			
(b) Characterization test				
(c) In-process control test				
	≥ cells			
	≥ %			
	≥			
	≥			
	≤			
	≥ %			
	≥			
	≥			
(d)	of critical intermediate			
	≥ *2			
(e)	after thawing of the critical intermediate			

*1
*2

Table 14. Comparability assessment between the commercial process and the proposed process (parameters after the thawing of the critical intermediate)

Test	Control values/ specification limits/ acceptance criteria	Proposed process	Commercial process
(a) Specifications			
Description			
pN-2 plasmid copy number	(copies per haploid genome)		
Cell surface marker analysis	≥ %		
	≥ %		
	≥ %		
	≤ %		
	≤ %		
	≤ %		
Viability	≥ %		
Viable cell count	≥ cells/vial		
	≥ %		
Intracellular FGF-2	≥ molecules		
Sterility	No detectable growth		
Bacterial endotoxin	≤ EU/mL		
Mycoplasma test	Not detected		
(b) Characterization test			
(c) In-process control test			
	≥ cells		
	≥ %		
	≥		
	≥		
	≥		
	≤		
	≥ %		
	≥		
(d) of critical intermediate			
(e) after thawing of the critical intermediate			

*1
*2

The comparability assessment at the manufacturing process change involves critical testing, such as potency test that reflect the mechanism of action of SB623. Primary pharmacodynamics of the primary component (SB623) indicated its pharmacological effect induced by the release of intracellular FGF-2 into the extracellular space [see Section 3.1]. Accordingly, Intracellular FGF-2 was determined as a potency test reflecting one of the biological action mechanisms of SB623. Currently, intracellular FGF-

2 is the only established test method, but the exploration and development of potency tests other than intracellular FGF-2 have been underway. A total of 3 commercial process batches will be manufactured before the start of commercial product shipment following marketing authorization. These batches will be used for comparability assessment among the commercial process formulation, the proposed process formulation, and the study product used in Study TBI-01. The exploration will be continued to establish additional potency tests that reflect the action mechanisms of SB623, other than intracellular FGF-2, so as to be included in verification and specification test, as well as comparability assessment at manufacturing process change.

Shelf life of the primary component (SB623)

At present, stability data using the preserved commercial process formulation [redacted] have yet to be available. In the commercial process, changes in the primary container (changes of [redacted] and [redacted] of cap, etc.) were implemented after the application as an anti-foreign matter contamination measure. The [redacted] of the vial in the primary container remained the same, while there was a change in the cap [redacted] ([redacted]), and a [redacted] packing was added. These changes in the primary container are considered to have minimal impact on the stability of the primary component (SB623). Thus, the stability of the commercial process formulation can be evaluated based on the long-term testing results of the proposed process formulation. The assessment of the [redacted] of the primary container in the commercial process formulation is currently underway using samples of the [redacted] primary container stored in the vapor phase of liquid nitrogen. So far, the [redacted] of the [redacted]-month storage samples has been confirmed to meet the criteria. The assessment is scheduled to be continued for up to [redacted] months.

Based on the above, the shelf life is [redacted] months for the primary component (SB623) when stored in [redacted] vials in the vapor phase of liquid nitrogen (at ≤[redacted]°C).

Although only 1 batch of the commercial process formulation has been produced, the above results suggest that the optimization of the manufacturing process, including [redacted] of the critical intermediate, has been achieved. Before the start of commercial product shipment after marketing authorization, 3 additional batches of the commercial process formulation will be manufactured to verify the robustness of the process.

Shelf life of critical intermediate

The storage period for the critical intermediate used in the manufacture of the commercial process formulation, [redacted], was [redacted] months (Table 4). The storage period for the critical intermediate used in the production of 7 batches of the proposed process formulation was [redacted] months. Based on the fact that all batches met the specification tests (Table 9) for the primary component (SB623), it is considered possible to determine the shelf life of the critical intermediate as [redacted] months when stored in [redacted] vials with [redacted] caps under the vapor phase of liquid nitrogen (≤[redacted]°C). Additional data will be obtained to reconfirm the appropriateness of the shelf life of [redacted] months. Until the test results are available, the critical intermediate stored for up to [redacted] months will be used based on the storage period of the critical intermediate as in the manufacture of the commercial process formulation, [redacted].

PMDA's view:
Given that the comparability needs to be carefully assessed among the commercial process formulation, the proposed process formulation, and the study product used in Study TBI-01, further investigations, including additional quality attribute data, are required. It is inappropriate to conclude that intended product quality can permanently be obtained only based on the result with 1 batch of the commercial process formulation. The product's quality should be assured through verification in future production. PMDA instructed the applicant to conduct further investigations, including those on additional quality attribute data.

The shelf life of the primary component (SB623) is determined based on the results of the stability studies of the proposed process formulation, requiring the results of comparability between the commercial process formulation and the proposed process formulation.

Furthermore, PMDA asked the applicant to explain the duration and evaluation items of future stability studies, in view of the shelf life of the critical intermediate.

PMDA's final conclusion on the manufacturing process of the primary component (SB623) will be described in the Review Report (2).

3. Primary Pharmacodynamics or Performance and Outline of the Review Conducted by PMDA

The applicant submitted the results of the *in vitro* and *in vivo* studies as data relating to the primary pharmacodynamics or performance of Akuugo.

3.1 *In vitro* studies

Table 15 shows the main results of *in vitro* studies.

Table 15. Main *in vitro* primary pharmacodynamics of the efficacy of the primary component (SB623)

Summary of study	Main results	Document No. (study code No.)
SB623-induced neurogenesis	• The mRNA expression levels of factors likely involved in neurogenesis (*FGF1, FGF2, FGFR1, FGFR2, BMP2, BMP4, BMP6, HB-EGF, EGF,* and *HGF*) were evaluated on both SB623 and MSCs deriving from the same donor. The expression levels of *FGF-1* and *FGF-2* (involved in the differentiation of nestin-positive neural progenitor cells) and *BMP4* (involved in the differentiation of GFAP-positive astrocytes) were higher in SB623 than in MSC. In contrast, the expression levels of *FGFR1* and *FGFR2* were lower in SB623 as compared to MSC. • Rat cerebral cortical neurons were co-cultured with MSCs or SB623 on slide glasses coated with ECM derived from SB623 for immunostaining. Nestin (neural progenitor cell marker)-positive cells were more frequently observed in SB623 co-cultured samples as compared to MSC co-cultured samples or non-co-cultured samples after 5 days of co-culture. GFAP (astrocyte marker)-positive cells were observed in higher numbers in both MSC and SB623 co-cultured samples than in non-co-cultured samples. In SB623 co-cultured samples, CNP (oligodendrocyte marker) exhibited widespread cytoplasmic expression after 12 days of co-culture, while in MSC co-cultured samples, CNP expression was confined around the nucleus, and it was barely detectable in non-co-cultured samples. • Rat cerebral cortical neurons were co-cultured with MSCs, SB623, or CM derived from SB623 for 5 days. As compared to MSC co-cultured samples or non-co-cultured samples, SB623 co-cultured samples or samples supplemented with SB623-derived CM exhibited increased expression of *nestin*, *GFAP*, and *CNP* genes. The expression of *nestin*, which increased with the addition of SB623-derived CM, was reduced by FGF-1 or FGF-2 neutralizing antibody. The increased expression of *GFAP* gene induced by co-culture with SB623 was reduced by human noggin protein, an extracellular antagonist of BMP.	CTD 4.2.1.1-1 (PSP104)
Effect of SB623 on GABAergic neurons	• Rat cerebral cortical neurons were co-cultured with MSCs or SB623 on ornithine/fibronectin-coated slide glass. The number of VGAT-positive puncta (a marker of nerve terminals in GABAergic neurons) per 100 μm of neurites increased in SB623 co-cultures as compared to MSC co-cultures and non-co-cultures, while no difference was observed in the number of VGLUT-positive puncta (a marker of nerve terminals in glutamatergic neurons). • Rat cerebral cortical neurons were cultured for 7 days on PDL-coated plates in media containing 30% MSC-derived CM or SB623-derived CM prepared from the MSCs. GABA expression level was higher in the medium containing SB623-derived CM than in the medium containing MSC-derived CM.	CTD 4.2.1.1-2 (PSP105)
Effect of SB623-derived ECM on neurons	Rat cerebral cortical neurons were cultured on plates coated with either SB623-derived ECM or MSC-derived ECM. After 21 days of culture, the number of neurons on SB623-derived ECM was approximately twice that on MSC-derived ECM. Additionally, both SB623-derived and MSC-derived ECM-coated plates, showed the presence of MAP2-positive cells (a marker for neurons and neurites) and GFAP-positive cells (a marker for neurons and astrocytes) after 12 days of culture, and CNP-positive cells (a marker for oligodendrocytes) after 21 days of culture. In contrast, on the PDL-coated plates used as a negative control, only MAP2-positive cells were observed, with no GFAP-positive cells detected.	CTD 4.2.1.1-3 (PSP045)
Effect of SB623-derived CM on angiogenesis	• The concentrations of 10 cytokines (angiogenin, angiopoietin-2, EGF, FGF-2, HB-EGF, HGF, leptin, PDGF-BB, PlGF, and VEGF) present in CM derived from SB623 or MSCs were measured. A total of 9 cytokines, excluding EGF, were detected in both samples, with no difference in concentrations. VEGF was the cytokine detected at the highest concentration. • Tubular formation in HUVECs and the sprouting and branching of new blood vessels from rat aortic rings were enhanced by the addition of CM derived from SB623 or MSCs as compared to the negative control OptiMEM medium. This effect of SB623-derived CM was inhibited by the VEGF receptor-2 tyrosine kinase inhibitor SU5416.	CTD 4.2.1.1-4 (PSP106)

Effects of intracellular factors from SB623 on neurogenesis and angiogenesis	• The amount of FGF-2 in extracts and CM from SB623 and MSCs deriving from the same donor was measured. The amount of FGF-2 per unit cell count was highest in the SB623 extract. • Rat cerebral cortical neurons were cultured with the addition of MSC or SB623 extract. The addition of both MSC and SB623 extracts promoted the proliferation of rat cerebral cortical neurons and HUVECs, and the promoted proliferation of rat cerebral cortical neurons was inhibited by FGF-2 neutralizing antibody treatment. The cells proliferated by the addition of SB623 extracts were primarily nestin-positive neural progenitor cells.	
Investigation of the FGF-2 release mechanism in transplanted SB623	• Co-culture of PBMCs from different donors with SB623 or MSCs resulted in an increase in LDH activity, a cytoplasmic enzyme released in media through cell death, in a PBMC ratio-dependent manner. This was accompanied by an increase in FGF-2 release in the culture supernatant. • To model the brain microenvironment post-transplantation, SB623 or MSCs were seeded at a concentration of 3.5×10^5 cells/well in a 96-well plate and cultured under high-cell density/low-oxygen/low-nutrient conditions, sealed to prevent gas exchange, and intracellular FGF-2 concentration and FGF-2 release into the CM were evaluated. In both SB623 and MSCs, intracellular FGF-2 content decreased within 20 hours of seeding, while FGF-2 release into the CM remained stable from 4 hours to 2 days. The amount of FGF-2 released into the CM was higher with SB623 than with MSCs.	CTD 4.2.1.1-5 (PSP107)
Investigation of the effects of SB623 and neurotrophic factors secreted by SB623 on rat cerebral cortical neurons and hippocampal neurons under OGD conditions	• OGD-induced neuronal damage in rat cerebral cortical neurons and hippocampal slices was suppressed by co-culture with SB623 or MSCs, as well as by the addition of SB623-derived CM or MSC-derived CM. • The concentrations of 30 cytokines in SB623-derived CM and MSC-derived CM were measured. A total of 10 cytokines (BMP-4, DKK-1, FGF-7, HB-EGF, IL-6, IL-8, MCP-1, MMP-1, PDGF-AA, and VEGF) were secreted from the cells. DKK-1, IL-6, IL-8, and MCP-1 were found in higher concentrations in SB623-derived CM than in MSC-derived CM.	CTD 4.2.1.1-6 (PSP108)

3.2 *In vivo* studies

Table 16 shows the main results of *in vivo* studies. It should be noted that the study using the TBI animal model is provided as reference information. Because the development of Akuugo initially focused on stroke (cerebral infarction), studies in stroke animal models were submitted for evaluation.

Table 16. Main *in vivo* primary pharmacodynamic studies on primary component (SB623)

Summary of study	Main results	Document No. (study code No.)
Therapeutic effects of SB623 through scar tissue shrinkage in stroke model rats	At 28 days after tMCAo surgery, SB623 (1.8×10^5 cells) was transplanted, or vehicle was administered. Based on baselines including 2 days before tMCAo surgery, 7 days post-tMCAo, and 21 days post-tMCAo, sensorimotor function tests (Cylinder test and Paw Whisker test) were conducted at 35, 43, 57, and 71 days post-tMCAo. The average recovery index scores* improved in the SB623 transplant group as compared to the vehicle-administered group. The brain was extracted at 72 days post-tMCAo for histopathological examination to assess infarct volume, showing no difference in infarct volume between the SB623 transplant group and the vehicle-administered group.	CTD4.2.1.1-13 (PSP109)
Effects of SB623 on behavior, neurological impairment, and injured brain tissue in TBI model rats	A TBI model rat was created by removing the skull and applying an impact using a compressed-air-powered metal impactor. At 7 days post-TBI model creation, SB623 (3.0×10^5 cells) was transplanted, or vehicle was administered. Neurological and behavioral function tests (EBST, Bederson neurological test, and Rotarod test) were performed at 7 days, 1 month, 2 months, and 3 months post-TBI model creation. Brain tissues were extracted at 1 and 3 months post-TBI model creation for histopathological examination and immunostaining (including staining for rat Ki-67 [cell proliferation marker], rat Dcx [early neuron marker], and rat nestin). Neurological and behavioral function tests demonstrated improvements in neurological and behavioral function in the SB623 transplant group as compared to the vehicle-administered group at 1 month post-TBI model creation and beyond. Histopathological examination revealed reduced brain injury area in the SB623 transplant group as compared to the vehicle-administered group at both 1 and 3 months post-TBI model creation. Immunostaining showed a higher number of Ki-67 positive cells and nestin-positive cells in the injured cortex and SVZ in the SB623 transplant group as compared to the vehicle-administered group at 1 and 3 months post-TBI model creation. At 1 month post-TBI model creation, SB623 and Dcx-positive cells were present along the corpus callosum in the SB623 transplant group, whereas in the vehicle-administered group, Dcx-positive cells were scattered. At 3 months post-TBI model creation, Dcx-positive cells were predominantly located around the injury site in the vehicle-administered group, while in the SB623 transplant group, they were widely spread from the SVZ to the injured cortex.	CTD4.2.1.1-14 (reference data)

* The recovery index score was adjusted based on the results of sensorimotor function tests, which had stabilized by 21 days post-tMCAo surgery.

3.R Outline of the review conducted by PMDA

The applicant's explanation about the mechanism of action of Akuugo contributory to the treatment of TBI:

Based on the results of *in vitro* and *in vivo* studies shown in Sections 3.1 and 3.2, SB623 exhibits a variety of pharmacological properties, including neuroprotective effects via extracellular matrix (ECM) production (CTD 4.2.1.1-3), promotion of neuronal proliferation and maturation (CTD 4.2.1.1-1), and enhancement of angiogenesis (CTD 4.2.1.1-4).

The transplantation of bone marrow-derived MSCs in TBI model rats contributed to the restoration of neurological function post-TBI (*Neurosurgery.* 2003;53:697-702). In addition, in the brain with TBI, the production of humoral factors such as BDNF by bone marrow-derived MSC increases, and pharmacological effects mediated by these factors may play a significant role in the recovery process of neural tissues post-TBI (*J Neurosci Res.* 2002;69:687-91). Studies with supplemented SB623-conditioned medium (CM) in rat cerebral cortical neurons demonstrated that the increased expression of the neural progenitor cell marker, *nestin*, was suppressed by FGF-2 neutralizing antibody (CTD 4.2.1.1-1). This suggests that FGF-2 is one of the growth factors involved in neurogenesis facilitated by Akuugo. Additionally, SB623 cell apoptosis is suggested to induce the release of intracellular FGF-2

into the extracellular space (CTD 4.2.1.1-5). The release of FGF-2 in the process of cell death, due to cellular damage or metabolic stress post-transplantation, is thought to promote neuronal proliferation, and this is considered a mechanism of action of Akuugo. Comparative study of SB623 and MSC indicates that the level of extracellular release of FGF-2 by SB623 is comparable to MSC, but SB623 contributes to higher mRNA expression of *FGF-1*, *FGF-2*, and *BMP4*, as well as intracellular FGF-2 in cell extracts as compared to MSC (CTD 4.2.1.1-1 and CTD 4.2.1.1-5).

PMDA considers that, although the mechanism of action of Akuugo on TBI has yet to be fully elucidated in primary pharmacodynamics or performance studies, the applicant has had thorough discussions based on available data. PMDA instructed the applicant, in view of current limited information on the biological characteristics of SB623, to continue collecting relevant data for further clarification of the mechanism of SB623 related to its primary pharmacodynamics or performance.

4. Biological Disposition and Outline of the Review Conducted by PMDA

The applicant conducted studies on the biological disposition of the primary component (SB623) as shown in Table 17.

Table 17. Main *in vivo* studies on biological disposition of the primary component (SB623 cells)

Outline of study methods	Main results	Reference number (study code)
Study on brain distribution following intracerebral transplantation of SB623 in athymic nude rats	SB623 (2.5×10^5 cells per hemisphere; striatum: 1.5×10^5 cells, cerebral cortex: 1.0×10^5 cells) was transplanted into the striatum and cerebral cortex of male and female athymic nude rats (Crl:NIH-*Foxn1*^{rnu}). Evaluation of human nucleus-positive cells at 14, 28, 56, 90, and 120 days post-transplantation showed the presence of human nucleus-positive cells at the transplantation site in 1 of 3 each of male and female rates on Day 14. No human nucleus-positive cells were observed from Day 28 onwards.	CTD 4.2.3.1-1 (PSP036)
Study on brain distribution following intracerebral transplantation of SB623 in athymic nude rats	SB623 (2.5×10^5 cells per hemisphere; striatum: 1.5×10^5 cells, cerebral cortex: 1.0×10^5 cells) was transplanted into the striatum and cerebral cortex of male and female athymic nude rats (Crl:NIH-*Foxn1*^{rnu}). Evaluation of human mitochondria-positive cells at 26 and 52 weeks post-transplantation showed no positive cells in the brain sections evaluated.	CTD 4.2.3.1-2 (PSP001)
Study on brain distribution following intracerebral transplantation of rat analog*1 in athymic nude rats	The rat analog (2.5×10^5 cells per hemisphere; striatum: 1.5×10^5 cells, cerebral cortex: 1.0×10^5 cells) was transplanted into the striatum and cerebral cortex of male and female athymic nude rats (Crl:NIH-*Foxn1*^{rnu}). Evaluation of OX-27*2 positive cells in the brain, heart, kidney, liver, lung, and spleen at 1, 7, 28, 90, and 270 days post-transplantation showed that OX-27 positive cells at the transplantation site decreased from Days 1 to 90 and were no longer observed at Day 270. No OX-27 positive cells were observed in organs other than the brain.	CTD 4.2.3.1-3 (PSP047)
Study on the presence of SB623 at the transplantation site following intracerebral transplantation of SB623 in cynomolgus monkeys	SB623 (5.0×10^6 cells per hemisphere; cerebral cortex or subcortex: 1.25×10^6 cells at two sites) was transplanted into the cerebral cortex and subcortex of female cynomolgus monkeys. Evaluation of human nuclear matrix-positive cells in the brain and at the transplantation site at 1, 3, and 6 months post-transplantation showed no human nuclear matrix-positive cells at any evaluation time point.	CTD 4.2.3.1-4 (PSP004)
Study on the survival and migration potential of SB623 in athymic nude rats	SB623 (3.6×10^4 to 4.2×10^4 cells per hemisphere) was transplanted into the striatum of male athymic nude rats (*rnu/rnu*). Evaluation of human nucleus-positive cells in the brain at 5 and 48 hours, and 14 days post-transplantation suggested that approximately 10% of human nucleus-positive cells remained up to ≥48 hours post-transplantation and localized within an average area of 0.42 mm outside the transplant site. Human nuclear staining by anti-human nucleus antibody was observed at 48 hours post-transplantation but was no longer noted at 14 days post-transplantation.	CTD4.2.2.3-6 (PSP006) (reference data)
Study on brain distribution following intracerebral transplantation of SB623 in athymic nude rats	SB623 (2.5×10^5 cells per hemisphere; striatum: 1.5×10^5 cells, cerebral cortex: 1.0×10^5 cells) was transplanted into the striatum and cerebral cortex of male athymic nude rats (Crl:NIH-*Foxn1*^{rnu}). Evaluation of human nucleus- or mitochondria-positive cells around the transplantation site 16 days post-transplantation showed no human nucleus- or mitochondria-positive cells in any animal evaluated.	CTD 4.2.2.3-7 (PSP010) (reference data)
Study on *in vivo* distribution of SB623 in stroke model rats	SB623 (1.8×10^5 cells) was transplanted into the penumbra*3 of male tMCAo model rats. Evaluation of *Alu* sequences by RT-qPCR 2 weeks post-transplantation showed that *Alu* sequences were undetectable in all evaluated organs (spleen, heart, kidney, liver, lung, and testis).	CTD 4.2.2.3-8 (PSP034) (reference data)

*1 Prepared by expressing the intracellular domain of human Notch-1 in rat-derived MSC.
*2 Rat major histocompatibility complex (MHC) class 1A haplotype c, designated as an indicator for the rat analog.
*3 Reversible ischemic area induced by tMCAo.

4.R Outline of the review conducted by PMDA

The applicant's explanation:

Based on the following pharmacokinetics, SB623 transplanted into the brain (1) remains at the transplantation site for approximately 2 weeks and disappears by 4 weeks, and (2) migrates to only around the transplantation site, and are unlikely to migrate to other organs.

- Following the transplantation of SB623 into the striatum and cerebral cortex of both hemispheres in athymic nude rats, SB623 was detected at the transplantation site 14 days post-transplantation but undetectable at 28 days post-transplantation (CTD 4.2.3.1-1). Transplantation of SB623 into the cerebral cortex and subcortex of cynomolgus monkeys suggested that SB623 had disappeared within 30 days post-transplantation (CTD 4.2.3.1-4). The transplantation of a rat analog into the striatum

and cerebral cortex of both hemispheres in athymic nude rats showed the presence of the rat analog at the transplantation site even at 90 days post-transplantation. The results are likely attributable to the difference in properties between SB623 and the rat analog (CTD 4.2.3.1-3).

- Following transplantation of SB623 into the striatum of both hemispheres in athymic nude rats, SB623 was localized within an average area of 0.42 mm outside the transplant site 48 hours post-transplantation (CTD 4.2.2.3-6). When SB623 was transplanted into the penumbra of transient middle cerebral artery occlusion (tMCAo) model rats, SB623 was undetectable in all evaluated organs (spleen, heart, kidney, liver, lung, and testis) 2 weeks post-transplantation (CTD 4.2.2.3-8).

PMDA concluded that the applicant's explanation about the biological disposition of Akuugo is acceptable based on the submitted data.

5. Non-Clinical Safety and Outline of the Review Conducted by PMDA

The applicant submitted non-clinical safety data of SB623 including 4 studies on intracerebral transplantation SB623 or a rat analog in athymic nude rats or cynomolgus monkeys, and tumorigenicity studies (karyotype analysis and soft agar colony formation assay).

5.1 General toxicity (CTD4.2.3.1-1 to 4.2.3.1-4)

Studies on a single transplantation of SB623 or a rat analog into the striatum and cerebral cortex of athymic nude rats or cynomolgus monkeys that had received immunosuppressive agents. The histopathological examination of the brain in the 4 general toxicity studies yielded generally similar results, showing axonal and myelin degeneration, localized necrosis, neural network loss, gliosis (suggesting tissue repair in neural structures), fibrosis, etc. but no findings related to clinical symptoms (Table 18). Findings noted in the brain following transplantation of SB623 or the rat analog were also observed in the vehicle-treated group. The applicant explained that these findings were not from direct effect of SB623 or the rat analog but related to the transplantation technique.

Table 18. General toxicity study of single intracerebral transplantation

Animal species	Dose of test substance	Observation period	Main findings	Document number (Study ID)
Male and female athymic nude rats	SB623 5×10^5 cells/animal	14, 28, 56, 90, and 120 days	Deaths occurred in 1 of 6 animals in the 28-day observation group and 2 of 6 animals in the 90-day observation group. The deaths were caused by scab formation and deterioration of clinical symptoms associated with infection, and not considered related to SB623 transplantation. No findings related to SB623 transplantation were observed in clinical signs, body weight, or autopsy. Histopathological examination revealed necrosis at the transplantation site in the brain, loss of neural network, axonal and myelin degeneration, gliosis, and fibrosis.	CTD 4.2.3.1-1 (PSP036)
Male and female athymic nude rats	Vehicle, SB623 5×10^5 cells/animal	26 and 52 weeks	Deaths occurred in 5 of 25 animals in the vehicle-treated group and 1 of 21 animals in the SB623-transplanted group under 26-week observation, and in 5 of 34 animals in the vehicle-treated group and in 4 of 39 animals in the SB623-transplanted group under 52-week observation. Deaths were attributed to administration and transplantation techniques and not considered related to the test substance itself. No findings related to SB623 transplantation were observed in clinical signs, body weight, necropsy, or organ weight. Histopathological examination revealed necrosis, gliosis, and pigment-laden macrophages in the meninges at the transplantation site in the brain in both vehicle and SB623 groups.	CTD 4.2.3.1-2 (PSP001)
Male and female athymic nude rats	Vehicle, rat analog* 5×10^5 cells/animal	1, 7, 28, 90, and approximately 270 days	Deaths occurred in 6 of 50 animals in the vehicle-treated group and in 6 of 61 animals in the rat-analogue-transplanted group under observation for approximately 270 days. The deaths were due to scab formation and deterioration of clinical symptoms associated with infection, and not considered related to administration and transplantation of the test substance. No findings related to rat analog transplantation were observed in clinical signs, body weight, necropsy, or organ weight. Histopathological examination revealed necrosis, gliosis, pigment-laden macrophages, mineral deposits, etc. at the transplantation site in the brain in both vehicle and rat analog groups. In the rat analog-transplanted group, the survival of the rat analog was confirmed up to 90 days post-transplantation [see Section 4]; however, no evidence of cell proliferation was observed with immunostaining with anti-Ki-67 antibody.	CTD 4.2.3.1-3 (PSP047)
Female cynomolgus monkeys	Vehicle, SB623 5×10^6 cells/animal	1, 3, and 6 months	No deaths occurred. No findings related to SB623 transplantation were observed in clinical signs, body weight, clinical pathology, necropsy, or organ weight. Histopathological examination revealed necrosis, hematoma, meningitis, gliosis, fibrosis, etc., in the brain in both vehicle and SB623 groups.	CTD 4.2.3.1-4 (PSP004)

* The rat analog was manufactured by expressing the intracellular domain of human Notch-1 in rat-derived MSCs.

5.2 Other safety profiles

5.2.1 Tumorigenicity assessment

A cytogenetic analysis and soft agar colony formation assay of SB623 revealed no findings suggestive of tumorigenicity. Furthermore, no proliferative lesions were observed in the general toxicity studies with an observation period of up to approximately 9 months. Based on these findings, the applicant explains that the risk of tumorigenicity with SB623 is low.

5.2.1.1 Cytogenetic analysis (CTD4.2.3.4.3-1)

Chromosomal abnormalities and chromosomal number were analyzed in 100 cells from SB623. No chromosomal abnormalities were observed, and the numbers of cells displaying chromosome numbers of 43, 45, and 46 were 1, 3, and 96, respectively. An additional analysis of 1,000 SB623 cells identified no polyploid cells.

Karyotype analysis was conducted on 5 cells of SB623. In 1 karyotype, chromosomes 10, 19, and 21 respectively appeared as a single copy, with 3 unidentified chromosomes observed. No abnormalities were detected in the remaining 4 karyotypes. As there were no abnormalities identical among ≥2 cells, these findings are not indicative of clonal abnormalities (ISCN2020: International System for Human Cytogenetic Nomenclature [*Cytogenet Genome Res,* 2020;160:341-503]). The applicant explains that there is no concern about the genetic stability of Akuugo.

5.2.1.2 Soft agar colony formation assay (CTD4.2.3.4.3-2, reference data)

A soft agar colony formation assay was conducted using SB623, and no anchorage-independent cell growth was observed.

5.2.2 Safety evaluation of subcomponent, secondary component 1 (dedicated preparation solution), and process-related impurities

The applicant evaluated the safety of the subcomponent, secondary component 1 (dedicated preparation solution), and process-related impurities in Akuugo, as shown below, and considers that they have safety minimal concerns.

The subcomponent of Akuugo is [redacted], and a safety evaluation was conducted based on the results from a single intracerebral dose toxicity study in cynomolgus monkeys, using SB623 frozen in [redacted] and prepared following the same method as post-marketing use [see Section 5.1].

Each ingredient of secondary component 1 (sodium chloride, sodium gluconate, sodium acetate trihydrate, potassium chloride, magnesium chloride hexahydrate, and sodium hydroxide) was subjected to safety evaluation based on their historical use as pharmaceutical ingredients or the previous uses as excipients.

Impurities potentially remaining in the final product include medium components (Impurities A, B, and FBS), Impurity C, trypsin, pN-2 plasmid, Impurity D, and Impurity E. Safety evaluation was conducted based on the residual amounts of these impurities in Akuugo.

5.2.3 Biological safety evaluation of secondary component 2 (dedicated delivery device set)

The applicant considers that safety concerns of Akuugo's secondary component 2 (dedicated delivery device set) are minimal, based on the following biological safety outcome.

The microsyringe was subjected to a cytotoxicity test (colony formation assay using the extraction method) in accordance with the "Basic Principles of Biological Safety Evaluation Required for Application for Marketing Approval of Medical Devices (in Japanese)" (PFSB/ELD/OMDE No. 0301-

20, dated March 1, 2012) and ISO10993-1. Additionally, an alkaline elution test according to JIS T 3201:1979 and a fluoride elution test according to JIS T 3210:2011 were performed.

The testing for the administration cannula, inserter, and stylet was omitted because their materials and manufacturing processes were equivalent to those of previously approved devices.

Biological safety testing for guide & stop was omitted, as these parts do not come into contact with blood, body fluids, or tissues.

5.R Outline of the review conducted by PMDA

Based on the submitted data and the following reviews, PMDA concluded that there are no significant toxicological issues regarding the clinical use of Akuugo.

In the general toxicity studies, necrosis and other findings were noted in the brain, the intended clinical application site. These findings were mild and indicated potential reversibility, with no related findings in clinical signs. The safety of SB623 transplantation is considered acceptable. The localized brain safety in humans will be further discussed in Section "6.R.2 Safety."

The biological safety evaluation of secondary component 2 (dedicated delivery device set) was reviewed by the Office of Medical Devices II, confirming no significant issues.

6. Clinical Efficacy and Safety and Outline of the Review Conducted by PMDA

The applicant submitted efficacy and safety data from 1 global phase II study, as well as the reference data from 1 foreign Phase I/IIa study and 1 foreign Phase IIb study, as all listed in Table 19.

Table 19. List of clinical studies on efficacy and safety

Data category	Region	Study identifier	Phase	Study population	Dosage regimen	No. of patients enrolled	Main endpoints
Evaluation	Global (US, Ukraine, Japan)	TBI-01	II	Patients with chronic motor deficit from TBI	SB623 intracranial transplantation (1) 2.5 × 10^6 cells, (2) 5.0 × 10^6 cells, (3) 10.0 × 10^6 cells, or (4) sham surgery	63 ([1] 16, [2] 16, [3] 16, [4] 15)	Efficacy Safety
Reference	US	STR01	I/IIa	Patients with chronic cerebral infarction with hemiparesis	SB623 intracranial transplantation (1) 2.5 × 10^6 cells, (2) 5.0 × 10^6 cells, (3) 10.0 × 10^6 cells	18 ([1] 6, [2] 6, [3] 6)	Safety
	US	STR02	IIb	Patients with chronic motor deficit from cerebral infarction	SB623 intracranial transplantation (1) 2.5 × 10^6 cells, (2) 5.0 × 10^6 cells, or (4) sham surgery	163 ([1] 55, [2] 56, [4] 52)	Safety

6.1 Evaluation data

6.1.1 Global phase II study (CTD5.3.5.1, Study TBI-01, July 6, 2016 to March 5, 2019)

A multicenter, randomized, double-blind,[1)] sham-controlled study was conducted to investigate the efficacy and safety of Akuugo in patients with chronic motor deficit from TBI (target sample size, 52 subjects[2)]) at study sites in the US, Ukraine, and Japan.

Table 20 shows the main inclusion and exclusion criteria.

Table 20. Main inclusion/exclusion criteria

Inclusion criteria • Age 18-75 years • A history of TBI that can be confirmed by MRI or CT • At least 12 months post-TBI • Focal brain injury visible on MRI (regardless of the diffuse axonal injury) • Motor deficit due to focal brain injury visible on MRI • Glasgow Outcome Scale-Extended (GOS-E) score[3)] of 3-6 (moderate to severe disability) • Motricity Index[4)] score for upper extremity (UE Scale) of 10-81, at least 2 of 3 scores below 33 with one of these below 25 and at least 1 score greater than 0; or lower extremity (LE Scale) score of 10-78, at least 2 of 3 scores below 33 with one of these below 25 and at least 1 score greater than 0 • Able and willing to undergo head CT and MRI imaging • Agreement to use antiplatelet, anticoagulant, or non-steroidal anti-inflammatory drugs in accordance with the Anticoagulant guidelines • Willing to participate in study related exercises to the extent possible • Willing to discontinue herbal or alternative medications for 1 week before and 1 week after the head surgery • Able to undergo all neurological assessments scheduled • Ability to understand and sign the informed consent
Exclusion criteria • History or presence of major neurological disease other than TBI • Any seizures in 3 months prior to the study • Presence of contractures in any joint that would interfere with the interpretation of neurological assessments (e.g., contracture preventing the detection of any increase in motion range or task performance ability) • Other neurologic, neuromuscular, or orthopedic disease limiting motor function • Clinically significant finding unrelated to TBI on brain MRI • Uncontrolled major psychiatric illness (Center for Epidemiologic Studies Depression Scale-Revised [CESD-R] score of ≥16), including symptoms of depression

Patients deemed eligible were randomized in a 3:1 ratio into the SB623 group and the sham surgery group. Those in the SB623 group were further randomized in a 1:1:1 ratio into low-dose (2.5×10^6 cells), medium-dose (5.0×10^6 cells), and high-dose (10.0×10^6 cells) groups. The primary endpoint was assessed at Week 24 after Akuugo transplantation or sham surgery, with observation up to 48 weeks.

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1) In this study, the following personnel were unblinded:
Staff responsible for preparing Akuugo, unblinded clinical study coordinators, surgeons and surgical staff, designated unblinded study sponsor and contract research organization personnel, members of the Data Safety Monitoring Board (DSMB), and supporting statisticians and programmers for the DSMB.

2) Assuming an expected change from baseline in Fugl-Meyer Motor Scale (FMMS) score at Week 24 of 10.0 in the SB623 group and 3.0 in the sham surgery group, with a standard deviation (SD) of 7.25 in each dosing group, a total of 48 subjects (36 in the SB623 group, 12 in the sham surgery group) was calculated as necessary sample size to achieve a two-sided significance level of 5% and a power of 80% for comparison between the SB623 and sham surgery groups. Considering an 8% dropout rate, the target sample size for the entire study was 52 subjects.

3) The Glasgow Outcome Scale is a comprehensive indicator for assessing outcomes in terms of living status (Good recovery, Moderate disability, Severe disability, Vegetative state, and Dead in a 5-point scale). An extended version of this scale, known as the Glasgow Outcome Scale-Extended (GOS-E), evaluates neurological prognosis across 8 levels from 1 (Dead) to 8 (Good recovery-upper) by dividing Good recovery, Moderate disability, and Severe disability into upper and lower levels.

4) For muscle strength, the upper limb (UE Scale) is assessed in 3 areas: Pinch grip, elbow flexion, and shoulder abduction. The lower limb (LE Scale) is evaluated based on dorsiflexion, knee extension, and hip flexion, with each item scored out of 33, resulting in a 100-point scale for each limb.

Procedures performed in each group were as follows. Subjects were instructed to conduct an at-home exercise program[5] following Akuugo transplantation or sham surgery.

SB623 Group

Using stereotactic neurosurgical techniques, 3 insertion routes were established from a single cranial burr hole. In each insertion route, cells were transplanted at 5 points in various depths (20 μL per site).

Detailed transplantation method:

Under local anesthesia and sedation, a burr hole (1-1.5 cm) was drilled, the dura mater was opened, and the stylet-equipped inserter for fixation was inserted to a point just proximal to the damaged area. After removing the stylet, an administration cannula with attached microsyringe filled with the cell suspension was inserted in the inserter down to the deepest target point for the first transplantation. While the cannula is gradually withdrawn from the deepest target point, the cell suspension was transplanted in a total of 5 sites at 5-6 mm intervals. The cell suspension concentrations were 8.3×10^6 cells/mL for the low-dose group (2.5×10^6 cells), 17×10^6 cells/mL for the medium-dose group (5.0×10^6 cells), and 33×10^6 cells/mL for the high-dose group (10.0×10^6 cells), with 20 μL administered per point at approximately 10 μL/min. Subsequently, this procedure was repeated 2 times at the area surrounding the damage site, through 2 other different routes the start from the same burr hole. The target site was selected to be closest to the motor pathway, based the subject's own neuroanatomy, by neurosurgeons at respective study sites.

Sham Surgery Group

The sham procedure included stereotactic planning, burr hole creation in the outer cranial layer (without penetration of the inner cranial layer or dura mater) under local anesthesia and sedation. The sham surgery procedure was designed as closely as possible to the procedure performed in the SB623 group. Subjects were monitored in the operating room for the same duration as in the SB623 group.

A total of 63 subjects (17 Japanese subjects) were randomized. The surgical procedure was performed on 61 subjects (17 Japanese subjects) and they were included in the modified intention-to-treat (mITT) population, excluding 1 subject each in the 2.5×10^6 and 5.0×10^6 cells groups who had discontinued early at the physician's discretion.[6] The mITT population was used as the efficacy and safety analysis sets. The mITT population included 15 subjects (4 Japanese subjects) in the 2.5×10^6 cells group, 15 subjects (4 Japanese subjects) in the 5.0×10^6 cells group, 16 subjects (5 Japanese subjects) in the 10.0×10^6 cells group, and 15 subjects (4 Japanese subjects) in the sham surgery group. All subjects in the mITT population completed the 48-week observation.

Table 21 shows the characteristics of subjects.

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5) Subjects conducted a series of exercises every morning and afternoon, including gripping a cylinder, thumbs-up motions, standing and squatting, and walking.

6) Reasons for exclusion of the 2 subjects, respectively:
- The preoperative MRI revealed an extremely small volume in the target area that limited the surgical field for transplantation. Additionally, dense vascular structures and motor nerves near the target area posed a high safety risk.
- Preoperative MRI failed to clearly visualize the lesion on the side involved in motor deficit.

Table 21. Characteristics of subjects (mITT population)

	2.5×10^6 cells (n = 15)	5.0×10^6 cells (n = 15)	10.0×10^6 cells (n = 16)	SB623 (n = 46)	Sham surgery (n = 15)
Age (mean ± SD)	36.66 ± 13.57	31.22 ± 9.15	34.23 ± 11.46	34.04 ± 11.49	35.48 ± 12.96
Sex: Female (upper row), male (lower row) (Number of subjects [%])	4 (26.7)	3 (20.0)	5 (31.3)	12 (26.1)	6 (40.0)
	11 (73.3)	12 (80.0)	11 (68.8)	34 (73.9)	9 (60.0)
GOS-E Score (mean ± SD)	4.3 ± 1.1	4.2 ± 1.0	4.4 ± 1.0	4.3 ± 1.0	4.3 ± 1.0
Motricity Index (upper limb, mean ± SD)	60.7 ± 21.3	59.6 ± 21.2	58.0 ± 16.3	59.4 ± 19.3	58.7 ± 17.0
Motricity Index (lower limb, mean ± SD)	65.1 ± 13.6	54.5 ± 19.4	63.1 ± 13.9	60.9 ± 16.1	61.6 ± 15.6

Table 22 shows change from baseline in Fugl-Meyer Motor Scale (FMMS) score[7)] at Week 24 after transplantation of Akuugo or the sham surgery, the primary efficacy endpoint. Figure 2 shows the trend of change from baseline in FMMS scores. The primary comparison was between the pooled SB623 groups against the sham surgery group. A statistically significant difference was observed between the SB623 and sham surgery groups in the primary endpoint.

Table 22. Results of the primary endpoint (mITT population)

	2.5×10^6 cells (n = 15)	5.0×10^6 cells (n = 15)	10.0×10^6 cells (n = 16)	SB623 (n = 46)	Sham surgery (n = 15)
Baseline*1,*2	54.5 ± 18.1	51.3 ± 22.0	50.9 ± 18.7	52.2 ± 19.3	52.3 ± 15.1
Change from baseline in FMMS at Week 24*1,*2	6.0 ± 10.1	11.0 ± 8.4	8.1 ± 12.8	8.3 ± 10.6	2.3 ± 4.7
Difference from sham surgery group (95% CI)*1,*2	3.7 (–2.4, 9.7)	8.5 (3.4, 13.7)	5.7 (–1.3, 12.7)	6.0 (0.3, 11.8)	
P value*1,*2,*3				0.0401	

Mean ± SD

*1 For 1 subject whose baseline FMMS score of the left upper limb was measured in the right upper limb, all baseline FMMS component scores were missing. There was no specific instruction in the protocol on dealing with missing data in such cases. The baseline score of this subject was imputed from baseline data of all other subjects using a linear regression model.

*2 Mixed effect models for repeated measures (MMRM) assuming an unstructured covariance structure, with covariates including treatment group, time point, interaction of treatment group and time point, baseline FMMS score, interaction of baseline FMMS score and time point, screening GOS-E score, and interaction of screening GOS-E score and time point.

*3 Two-sided significance level of 5%.

7) A motor function index. Upper limb function is rated for 33 items, including reflex activity, synergistic movement of flexors and extensors, complex synergistic movements, movements without synergy, normal reflex activity, wrist, hand, coordination, and speed, on a 3-point scale from 0 (non-functional) to 2 (fully functional), (except for reflexes rated by 2 points; 0 or 2), with the highest score of 66. Lower limb function is rated for 17 items, including reflex activity, synergistic movement of flexors and extensors, complex synergistic movements, movements without synergy, normal reflex activity, coordination, and speed, also on 3-point scale, with the highest score of 34.

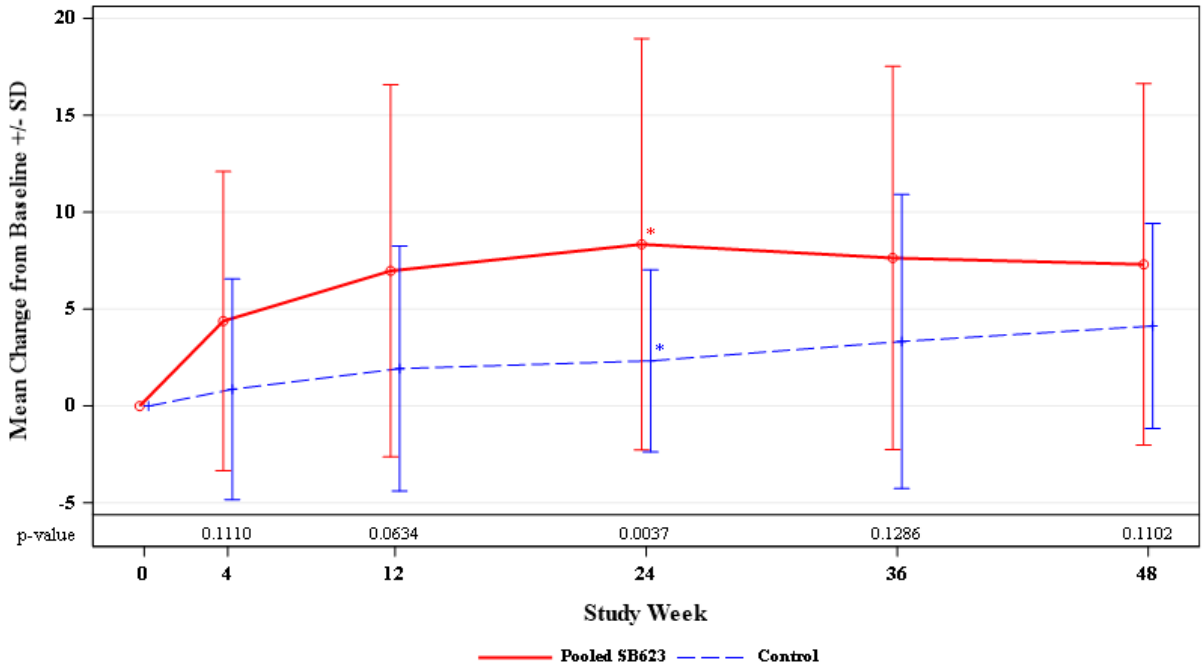

Figure 2. Change from baseline over time in FMMS score (Mean ± SD) (mITT population)

The change from baseline (mean ± standard deviation [SD]) in FMMS score at Week 24 in the Japanese population (13 subjects in the SB623 group, 4 subjects in the sham surgery group) was 8.2 ± 10.5 in the SB623 group and 2.3 ± 1.9 in the sham surgery group.

Tables 23 to 28 show the results of the main secondary endpoints.

Table 23. DRS score[8)] and change from baseline (mITT)

	2.5×10^6 cells (n = 15)		5.0×10^6 cells (n = 15)		10.0×10^6 cells (n = 16)		SB623 (n = 46)		Sham surgery (n = 15)	
		Change from baseline		Change from baseline		Change from baseline		Change from baseline		Change from baseline
Baseline	3.9 ± 2.1	-	6.2 ± 4.1*[1]	-	4.6 ± 2.4	-	4.8 ± 3.0*[2]	-	3.7 ± 2.0	-
Week 24	3.8 ± 2.6	−0.1 ± 1.2	4.6 ± 2.9	−1.4 ± 2.6*[1]	4.0 ± 1.8	−0.6 ± 2.2	4.1 ± 2.4	−0.7 ± 2.1*[2]	4.3 ± 2.6	0.6 ± 1.6
Week 48	3.9 ± 1.9	0.0 ± 1.4	4.7 ± 2.7	−1.4 ± 2.7*[1]	4.4 ± 2.1*[3]	−0.2 ± 2.0*[3]	4.3 ± 2.2*[2]	−0.5 ± 2.2*[4]	4.1 ± 2.7	0.4 ± 1.6

Mean ± SD; *[1] n = 14; *[2] n = 45; *[3] n = 15; *[4] n = 44

8) The rating for the degree of impairment using the following scale: Eye opening ability, 0 (spontaneous) to 3 (none) on a 4-point scale; communication ability, 0 (oriented) to 4 (none) on a 5-point scale; motor response from 0 (obeying) to 5 (none) on a 6-point scale; cognitive ability for feeding, toileting, and grooming, 0 (complete) to 3 (none) on a 4-point scale, respectively; level of functioning, 0 (completely independent) to 5 (totally dependent) on a 6-point scale; and employability, 0 (not restricted) to 3 (not employable) on a 4-point scale.

Table 24. ARAT[9] total score and change from baseline in subjects[10] with upper limb impairment (mITT)

	2.5×10^6 cells (n = 14)		5.0×10^6 cells (n = 13)		10.0×10^6 cells (n = 14)		SB623 (n = 41)		Sham surgery (n = 14)	
		Change from baseline		Change from baseline		Change from baseline		Change from baseline		Change from baseline
Baseline	21.0 ± 19.1	-	19.1 ± 20.8	-	17.1 ± 19.9	-	19.1 ± 19.5	-	20.1 ± 17.2	-
Week 24	24.0 ± 22.3	3.0 ± 6.7	23.3 ± 20.9	4.2 ± 5.2	16.9 ± 21.4	–0.3 ± 10.3	21.3 ± 21.3	2.3 ± 7.8	19.7 ± 19.0	–0.4 ± 11.5
Week 48	23.8 ± 22.8	2.8 ± 7.3	23.8 ± 21.5	4.7 ± 5.5	20.0 ± 23.0*1	1.9 ± 9.1*1	22.6 ± 21.9*2	3.1 ± 7.3*2	21.9 ± 17.4	1.9 ± 9.4

Mean ± SD; *1 n = 13; *2 n = 40

Table 25. Walking speed (time required for 10 m walk [s]) and change from baseline in subjects[11] with lower limb impairment (mITT)

	2.5×10^6 cells (n = 14)		5.0×10^6 cells (n = 13)		10.0×10^6 cells (n = 15)		SB623 (n = 42)		Sham surgery (n = 14)	
		Change from baseline		Change from baseline		Change from baseline		Change from baseline		Change from baseline
Baseline	44.5 ± 76.4	-	47.0 ± 79.9	-	58.4 ± 88.5*1	-	50.0 ± 80.0*2	-	56.5 ± 96.6	-
Week 24	41.4 ± 78.2	–3.1 ± 87.0	43.1 ± 79.5	–3.9 ± 6.7	72.5 ± 102.5	–2.2 ± 113.6*1	53.0 ± 87.2	–3.1 ± 81.7*2	54.1 ± 93.5	–2.4 ± 6.7
Week 48	26.7 ± 32.8*3	–20.8 ± 65.0*3	39.5 ± 79.6	–7.5 ± 11.3	60.6 ± 103.3*1	–20.0 ± 117.7*3	42.7 ± 77.7*4	–16.1 ± 76.1*5	56.9 ± 104.0	0.4 ± 15.7

Mean ± SD; *1 n = 14; *2 n = 41; *3 n = 13; *4 n = 40; *5 n = 39

Table 26. NeuroQOL upper limb function t-score[12] and change from baseline in subjects[10] with upper limb impairment (mITT)

	2.5×10^6 cells (n = 14)		5.0×10^6 cells (n = 13)		10.0×10^6 cells (n = 14)		SB623 (n = 41)		Sham surgery (n = 14)	
		Change from baseline		Change from baseline		Change from baseline		Change from baseline		Change from baseline
Baseline	35.46 ± 9.06	-	28.19 ± 11.25	-	33.57 ± 16.75	-	32.51 ± 12.86	-	32.15 ± 9.22	-
Week 24	41.11 ± 9.40	5.66 ± 8.06	33.46 ± 11.84	5.27 ± 8.90	32.80 ± 15.77	–0.77 ± 6.42	35.85 ± 12.90	3.34 ± 8.20	36.12 ± 7.71*1	2.48 ± 9.89*1
Week 48	38.95 ± 10.81	3.49 ± 8.46	36.04 ± 11.61	7.85 ± 8.96	34.42 ± 14.51*1	–0.68 ± 8.01*1	36.53 ± 12.20*2	3.55 ± 8.97*2	33.36 ± 10.09	1.21 ± 6.64

Mean ± SD; *1 n = 13; *2 n = 40

9) The rating for upper limb function across 19 items, 6 for grip, 4 for grasp, 6 for pinch, and 3 for gross movements, on a scale of 0 (no movement) to 3 (normal movement)

10) Patients with a Motricity Index UE Scale score of 10 to 81 at screening.

11) Patients with a Motricity Index LE Scale score of 10 to 78 at screening.

12) The rating for 8 daily living tasks involving fine motor upper limb function on a 5-point scale from 1 (unable) to 5 (no difficulty), with scores normalized to a mean of 50

Table 27. NeuroQOL lower limb function T-score[13] and change from baseline in subjects with lower limb impairment[11] (mITT)

	2.5×10^6 cells (n = 14)		5.0×10^6 cells (n = 13)		10.0×10^6 cells (n = 15)		SB623 (n = 42)		Sham surgery (n = 13)	
		Change from baseline		Change from baseline		Change from baseline		Change from baseline		Change from baseline
Baseline	43.41 ± 9.36	-	41.92 ± 13.08	-	39.29 ± 8.92	-	41.48 ± 10.40	-	44.25 ± 9.64	-
Week 24	46.47 ± 9.27	3.06 ± 6.58	43.87 ± 11.51	1.95 ± 6.82	42.67 ± 11.21	3.38 ± 7.10	44.31 ± 10.56	2.83 ± 6.70	44.59 ± 10.85*1	1.75 ± 7.05
Week 48	47.98 ± 9.18	4.57 ± 7.63	46.11 ± 12.47	4.19 ± 4.95	43.75 ± 9.70*1	5.12 ± 6.91*1	45.94 ± 10.38*2	4.64 ± 6.48*2	43.04 ± 11.65*1	0.62 ± 4.87

Mean ± SD; *1 n = 14; *2 n = 41

Table 28. Investigator/subinvestigator general assessment score[14] of subjective and objective changes from baseline (mITT)

	2.5×10^6 cells (n = 15)	5.0×10^6 cells (n = 15)	10.0×10^6 cells (n = 16)	SB623 (n = 46)	Sham surgery (n = 15)
Week 24	4.9 ± 1.4	5.3 ± 0.7	4.7 ± 1.3	4.9 ± 1.2	4.6 ± 0.8
Week 48	5.1 ± 1.5	5.6 ± 0.9	4.9 ± 1.1*1	5.2 ± 1.2*2	4.7 ± 1.0

Mean ± SD; *1 n = 15; *2 n = 45

Tables 29 and 30 show the upper limb subscale of FMMS (upper limb score [upper-extremity subscale of the Fugl-Meyer Motor Test (UE-FM) score]) and its change from baseline, respectively, in subjects with upper limb impairment.[10] Similarly, Tables 31 and 32 show the lower limb subscale of FMMS (lower limb score [lower-extremity subscale of the Fugl-Meyer Motor Test (LE-FM) score]) and its change from baseline, respectively, in subjects with lower limb impairment.[11]

Table 29. UE-FM scores in subjects with upper limb impairment (mITT)

	2.5×10^6 cells (n = 14)	5.0×10^6 cells (n = 13)	10.0×10^6 cells (n = 14)	SB623 (n = 41)	Sham surgery (n = 14)
Baseline	30.9 ± 13.0	26.9 ± 13.1*1	26.7 ± 14.6	28.2 ± 13.4*2	29.7 ± 11.9
Week 24	34.6 ± 15.7	36.3 ± 16.2	33.3 ± 16.2	34.7 ± 15.7	32.1 ± 12.7
Week 48	34.6 ± 15.5	36.1 ± 16.8	33.4 ± 16.6*3	34.7 ± 15.9*2	33.0 ± 13.5

Mean ± SD; *1 n = 12; *2 n = 40; *3 n = 13

Table 30. Change from baseline in UE-FM scores in subjects with upper limb impairment (mITT)

	SB623 (n = 41)	Sham surgery (n = 14)
Week 24	5.9 ± 8.0*1	2.4 ± 3.4
Week 48	5.6 ± 6.6*2	3.3 ± 3.2

Mean ± SD; *1 n = 40; *2 n = 39

Table 31. LE-FM scores in subjects with lower limb impairment (mITT)

	2.5×10^6 cells (n = 14)	5.0×10^6 cells (n = 13)	10.0×10^6 cells (n = 15)	SB623 (n = 42)	Sham surgery (n = 14)
Baseline	21.2 ± 4.6	18.3 ± 5.8	20.3 ± 4.5	20.0 ± 5.0	20.3 ± 6.8
Week 24	23.7 ± 6.0	21.0 ± 6.2	23.0 ± 5.9	22.6 ± 6.0	20.1 ± 7.0
Week 48	23.1 ± 5.9	20.8 ± 6.7	21.5 ± 6.4*1	21.8 ± 6.3*2	21.1 ± 6.5

Mean ± SD; *1 n = 14; *2 n = 41

13) The rating for 8 daily living tasks involving motor ability in lower limb function on a 5-point scale from 1 (unable) to 5 (no difficulty), with scores normalized to a mean of 50.

14) The rating for symptom improvement on a 7-point scale 1 (much worse) to 7 (much improved), as evaluated by physicians.

Table 32. Change from baseline in LE-FM score in subjects with lower limb impairment (mITT)

	SB623 (n = 42)	Sham surgery (n = 14)
Week 24	2.6 ± 3.4	–0.1 ± 2.7
Week 48	1.9 ± 3.4*	0.8 ± 3.6

Mean ± SD; * n = 41

The incidence of adverse events was 100% across all groups. The incidence of serious adverse events was 13.3% in the 2.5 × 10^6 cells group (2 of 15 subjects; haemorrhagic anaemia, femur fracture and delirium in 1 subject each); 6.7% in the 5.0 × 10^6 cells group (1 of 15 subjects; transient ischaemic attack); 12.5% in the 10.0 × 10^6 cells group (2 of 16 subjects; balance disorder, seizure, and delirium in 1 subject each); and 20.0% in the sham surgery group (3 of 15 subjects, wound infection, road traffic accident, and seizure in 1 subject each). No adverse events led to study discontinuation or resulted in death in any group. Table 33 shows adverse events occurring in ≥20% of subjects in any group.

Table 33. Adverse events observed in ≥20% of subjects in any group (safety analysis population)

	2.5 × 10^6 cells (n = 15)	5.0 × 10^6 cells (n = 15)	10.0 × 10^6 cells (n = 16)	Sham surgery (n = 15)
Headache	33.3% (5)	46.7% (7)	50.0% (8)	26.7% (4)
Wound complication	20.0% (3)	33.3% (5)	25.0% (4)	20.0% (3)
Nausea	20.0% (3)	20.0% (3)	18.8% (3)	6.7% (1)
Vomiting	13.3% (2)	13.3% (2)	25.0% (4)	6.7% (1)
Pyrexia	26.7% (4)	13.3% (2)	0	0
Dizziness	20.0% (3)	6.7% (1)	6.3% (1)	6.7% (1)
Constipation	0	6.7% (1)	6.3% (1)	20.0% (3)
Upper respiratory tract infection	0	20.0% (3)	6.3% (1)	0
Contusion	6.7% (1)	0	0	20.0% (3)
Incision site pain	20.0% (3)	0	6.3% (1)	6.7% (1)

Incidence (%) (Number of subjects with events)

The incidence of adverse events in the Japanese population was 100% across all groups, and there were no serious adverse events. The main adverse events are shown in Table 34.

Table 34. Adverse events observed in ≥2 subjects in any group (Japanese population)

	2.5 × 10^6 cells (n = 4)	5.0 × 10^6 cells (n = 4)	10.0 × 10^6 cells (n = 5)	Sham surgery (n = 4)
Wound complication	50.0% (2)	100.0% (4)	60.0% (3)	50.0% (2)
Influenza	0	0	40.0% (2)	50.0% (2)
Contusion	25.0% (1)	0	0	50.0% (2)
Headache	0	0	40.0% (2)	0
Pyrexia	75.0% (3)	25.0% (1)	0	0
Pruritus	0	0	40.0% (2)	25.0% (1)

Incidence (%) (Number of subjects with events)

6.2 Reference data (clinical studies with different target diseases)

6.2.1 Foreign phase I/IIa study (CTD5.3.5.4.1, Study STR01, August 22, 2011 to August 25, 2015)

A multicenter, open-label, dose-escalation study was conducted to assess the safety and efficacy of Akuugo in patients with chronic cerebral infarction with hemiparesis unresponsive to physical therapy or rehabilitation (target sample size, 18 subjects) at study sites in the US. The transplantation of Akuugo was performed via stereotactic neurosurgery, as in the Study TBI-01.

All 18 subjects enrolled were included in the safety analysis population, including 6 subjects each from the 2.5×10^6, 5.0×10^6, and 10.0×10^6 cells groups.

The incidence of adverse events[15] was 100% across all groups. The incidence of serious adverse events was 33.3% in the 2.5×10^6 cells group (2 of 6 subjects; carotid artery stenosis and seizure in 1 subject each), 50.0% in the 5.0×10^6 cells group (3 of 6 subjects; hypoaesthesia, transient ischaemic attack, dysphagia, and subdural haematoma in 1 subject each), and 33.3% in the 10.0×10^6 cells group (2 of 6 subjects; pneumonia, sepsis, and urinary tract infection in 1 subject each). No adverse events led to study discontinuation or resulted in death in any group. Table 35 shows adverse vents observed in ≥15% of the subjects in the SB623 group.

Table 35. AEs observed in ≥15% of the subjects in the SB623 group

	2.5×10^6 cells (n = 6)	5.0×10^6 cells (n = 6)	10.0×10^6 cells (n = 6)	SB623 (n = 18)
Headache	66.7% (4)	66.7% (4)	50.0% (3)	61.1% (11)
Nausea	0	50.0% (3)	50.0% (3)	33.3% (6)
Procedural headache	33.3% (2)	0	50.0% (3)	27.8% (5)
Muscle spasticity	33.3% (2)	16.7% (1)	16.7% (1)	22.2% (4)
Vomiting	0	33.3% (2)	33.3% (2)	22.2% (4)
Depression	0	33.3% (2)	33.3% (2)	22.2% (4)
Urinary tract infection	16.7% (1)	0	33.3% (2)	16.7% (3)
Constipation	0	33.3% (2)	16.7% (1)	16.7% (3)
Pain in extremity	33.3% (2)	16.7% (1)	0	16.7% (3)
Fatigue	0	16.7% (1)	33.3% (2)	16.7% (3)
Blood glucose increased	33.3% (2)	16.7% (1)	0	16.7% (3)
C-reactive protein increased	16.7% (1)	16.7% (1)	16.7% (1)	16.7% (3)

Incidence (%) (Number of subjects with events)

6.2.2 Foreign phase IIb study (CTD5.3.5.4.2, Study STR02, December 23, 2015 to December 10, 2018)

A multicenter, sham-controlled, randomized, double-blind study was conducted to assess the efficacy and safety of Akuugo in patients with chronic motor deficit from cerebral infarction (target sample size, 156 subjects) at study sites in the US. The transplantation of Akuugo was performed via stereotactic neurosurgery, as in Study TBI-01.

Patients deemed eligible were randomized in a 2:1 ratio to the SB623 and sham surgery groups. The SB623 group was further randomized in a 1:1 ratio into the 2.5×10^6 and 5.0×10^6 cells groups. A total of 163 subjects were randomized, and all randomized subjects were included in the safety analysis population, 55 subjects in the 2.5×10^6 cells group, 56 subjects in the 5.0×10^6 cells group, and 52 subjects in the sham surgery group.

The incidence of adverse events was 94.5% (52 of 55 subjects) in the 2.5×10^6 cells group, 96.4% (54 of 56) of subjects in the 5.0×10^6 cells group, and 98.1% (51 of 52 subjects) in the sham surgery group. The incidence of serious adverse events was 27.3% (15 of 55 subjects) in the 2.5×10^6 cells group (seizure in 4 subjects, subdural haematoma in 3 subjects, nausea and pneumonia in 2 subjects each, and haemorrhagic anemia, pericardial effusion, ventricular tachycardia, vertigo, abdominal pain, vomiting, chest pain, sepsis, wound infection, contusion, crush injury, humerus fracture, incision site complication,

[15] During the transplantation and follow-up periods (up to 2 years after the final Akuugo transplantation)

ligament sprain, upper limb fracture, wound dehiscence, positive anti-factor X antibody, muscle haemorrhage, basal ganglia haemorrhage, generalised tonic-clonic seizure, seizure-like symptoms, suicidal ideation, urinary retention, and hypotension in 1 subject each); 26.8% (15 of 56 subjects) in the 5.0×10^6 cells group (seizure in 4 subjects, pyrexia and subdural haemorrhage in 2 subjects each, and leukocytosis, atrial fibrillation, pancreatitis chronic, asthenia, pancreas infection, urinary tract infection, subdural haematoma, fall, hip fracture, pneumocephalus, hyponatremia, rhabdomyolysis, bladder cancer, syncope, renal failure in 1 subject each); and 21.2% (11 of 52 subjects) in the sham surgery group (seizure in 3 subjects, and bradycardia, pancreatitis, sepsis, bronchopneumonia, cellulitis, influenza, intervertebral discitis, staphylococcal osteomyelitis, basal cell carcinoma, encephalopathy, transient ischaemic attack, acute renal disorder, respiratory failure, and hypertension in 1 subject each). No adverse events leading to study discontinuation were observed in any group. The incidence of adverse events resulting in death was 1.8% (1 of 56 subjects) in the 5.0×10^6 cells group (bladder cancer in 1 subject), and a causal relationship to the study product or the surgical procedure was ruled out. Table 36 shows adverse events reported in ≥10% of subjects in the SB623 group.

Table 36. Adverse events reported in ≥10% of subjects in the SB623 group (safety analysis population)

	2.5×10^6 cells (n = 55)	5.0×10^6 cells (n = 56)	SB623 (n = 111)	Sham surgery (n = 52)
Headache	63.6% (35)	73.2% (41)	68.5% (76)	61.5% (32)
Nausea	25.5% (14)	21.4% (12)	23.4% (26)	5.8% (3)
Procedural headache	14.5% (8)	16.1% (9)	15.3% (17)	25.0% (13)
Vomiting	18.2% (10)	10.7% (6)	14.4% (16)	1.9% (1)
Procedural pain	20.0% (11)	8.9% (5)	14.4% (16)	11.5% (6)

Incidence (%) (Number of subjects with events)

6.R Outline of the review conducted by PMDA

PMDA considers that the comparability between the study product used in Study TBI-01 and the commercial process formulation cannot be determined from the data currently available [see Section 2.R.2]. However, the following assessments were conducted under the assumption that comparability exists between them.

6.R.1 Efficacy

In Study TBI-01, data from the pooled group of 3 dose levels (2.5×10^6, 5.0×10^6, and 10.0×10^6 cell groups; SB623 group) was compared against data from the sham surgery group.

6.R.1.1 Study design

6.R.1.1.1 Endpoint

The primary endpoint of Study TBI-01 was the FMMS score.

The applicant's explanation about FMMS score:

Fugl-Meyer scale, developed specifically for quantitative measurement of recovery from post-stroke hemiplegia (*Neurorehabil Neural Repair.* 2002;16:232-40), is the most widely recognized reference (*Stroke.* 2011;42:427-32). Fugl-Meyer scale is one of the tools to assess physical and mental functional impairment commonly used in clinical studies targeting motor dysfunction after neurological injury (*Circ Cardiovasc Qual Outcomes.* 2015;8(6 Suppl 3):S163-9). In particular, FMMS score, which

consists of the motor components of Fugl-Meyer scale, is considered reliable and valid (*Clin Rehabil.* 2005;19:404-11).

PMDA's view:
The use of FMMS score as the primary endpoint is understandable, as it is largely employed in Japan for motor paralysis evaluation. Yet, the score has yet to be recognized as an established efficacy index for the development of new treatments. Thus, the efficacy review of Akuugo was conducted comprehensively including the results of secondary endpoints as well.

6.R.1.1.2 Evaluation of the global study

Study TBI-01 was conducted as a global study. PMDA asked the applicant to explain the appropriateness of conducting a global study, from the viewpoint of any intrinsic and extrinsic ethnic factors that could affect the efficacy or safety evaluation of Akuugo.
Based on the following observations, the applicant explained that the evaluation of Akuugo in a global clinical study was appropriate:

- Akuugo is directly transplanted near the damage site of the brain, and is unlikely to show different pharmacodynamics in Japanese and non-Japanese patients.
- Although no established guidelines are available for the diagnosis or treatment of chronic TBI, there is a shared understanding that the degree of motor deficit stabilizes through the acute and recovery phases, i.e., approximately 3 weeks to 6 months post-injury, after which further recovery is no longer expected (*Arch Phys Med Rehabil.* 1998;79:488-93, *Brain Inj.* 2015;29:1431-8).
- Stereotactic brain surgery has been commonly performed across regions.

PMDA considered that the applicant's explanation was acceptable and that the evaluation of Akuugo based on Study TBI-01, a global study, be feasible.

6.R.1.1.3 Blinding

The rules set for safeguarding blinding in Study TBI-01 required any deviation cases to be shared with the sponsor and the contract research organization through a Protocol Deviation Tracker to prevent recurrence. After the study completion, the Protocol Deviation Tracker was rechecked for cases with suspected compromised blinding. Consequently, questionable blinding was found in 8 subjects (3 in the 2.5×10^6 cells group, 1 in the 5.0×10^6 cells group, and 4 in the 10.0×10^6 cells group).

PMDA considers that the assurance of blinding is essential for the efficacy evaluation of Akuugo, and the efficacy results of the Study TBI-01 should be interpreted carefully in light of the possibility of compromised blinding. Results of the subjects other than the 8 subjects with questionable blinding were also subjected to checking [see Section 6.R.1.2].

6.R.1.2 Results of efficacy evaluation

The applicant's explanation about the efficacy of Akuugo:
The results of FMMS score, the primary endpoint of study TBI-01, are of clinical significance in terms of the efficacy of Akuugo.

- At Week 24, a statistically significant difference was observed in the change from baseline in FMMS score between the SB623 group and the sham surgery group [see Section 6.1.1].
- Although there are no data indicating clinically significant change in FMMS score (total score of upper and lower limbs) in patients with chronic TBI, clinically significant change in FMMS score of upper limb (UE-FM scores) was 4.25 to 7.25 in patients with chronic stroke (*Phys Ther.* 2012;92:791-8). In Study TBI-01, the mean change from baseline in UE-FM score was 5.9 at Week 24 and 5.6 at Week 48 in the SB623 group, and 2.4 at Week 24 and 3.3 at Week 48 in the sham surgery group (Table 30).

The following secondary endpoint outcomes suggested a tendency of improvement in the SB623 group as compared to the sham surgery group.

- The change in Disability Rating Scale (DRS) score from baseline tended to decrease in the SB623 group at both Weeks 24 and 48. In particular, the 5.0×10^6 cells group showed a greater decrease than the low-dose and high-dose groups at all evaluation time points. In contrast, the sham surgery group showed a tendency of increase in DRS score from baseline at all evaluation points.
- In Action Research Arm Test (ARAT) total score, change from baseline tended to increase in the SB623 group at all evaluation points, and particularly the 5.0×10^6 cells group showed a greater increase than in the low-dose and high-dose groups at all evaluation time points. In the sham surgery group, ARAT total score decreased at Week 24 and increased at Week 48 but at a smaller degree than in the SB623 group.
- In the change from baseline in gait velocity (10-meter walk time), the time tended to shorten in the SB623 group at all evaluation points.
- In the change from baseline in Quality of Life in Neurological Disorders (NeuroQOL, upper and lower limbs) T-score, the SB623 group showed greater changes than in the sham surgery group for both limbs at all evaluation points.
- The general assessment score of subjective and objective changes from baseline tended to be higher in the SB623 group than in the sham surgery group at all evaluation points.

In those other than the 8 subjects (3 in the 2.5×10^6 cells group, 1 in the 5.0×10^6 cells group, 4 in the 10.0×10^6 cells group) with questionable blinding, the mean ± SD of the change from baseline in FMMS score, the primary endpoint, was 9.2 ± 10.8 in the SB623 group (38 subjects) and 2.4 ± 4.7 in the sham surgery group (15 subjects). Except for the following outcomes, the results of secondary endpoints had no impact on analysis results, demonstrating the efficacy of Akuugo even in the analysis excluding the subjects with questionable blinding.

- Change from baseline in gait velocity at Week 24
  Although, in the original analysis, gait velocity tended to increase in the SB623 group as compared to the sham surgery group, the results from the population excluding the 8 subjects tended to show decreased speed in the SB623 group. This result was attributed to the exclusion of the 8 subjects, among whom 1 subject had Week 24 change from baseline in gait velocity of –288, indicating the impact of substantial individual variability in this endpoint.
- Change from baseline in the NeuroQOL upper limb function T-score at Week 24
  The original analysis showed a dose-response tendency, which was not observed in the results excluding the 8 subjects.

PMDA's view:

The lack of sufficient information accountable for clinically significant change in FMMS scores in patients with chronic TBI precludes the conclusion on clear relationship between the primary endpoint results in Study TBI-01 and improvement in activities of daily living. However, based on the following outcomes, Akuugo is expected to have a certain level of efficacy in improving motor paralysis:

- In the change from baseline in FMMS score at Week 24, albeit possible bias due to handling of the analysis population[16] and missing data,[17] a statistically significant improvement was observed in the SB623 group as compared to the sham surgery group, and similarly, both UE-FM and LE-FM scores (Tables 29-32) also showed improvement tendencies.
- Secondary endpoints such as DRS score and ARAT total score suggested an improvement tendency in the SB623 group as compared to the sham surgery group.
- In the Japanese population, the change from baseline in FMMS score at Week 24 was greater in the SB623 group than in the sham surgery group, consistent with overall study results.
- The results excluding the subjects with questionable blinding showed a tendency toward improving efficacy in the SB623 group as compared to the sham surgery group, except the change from baseline in gait velocity Week 24, which was highly variable across individuals.

Meanwhile, the efficacy of Akuugo is subject to further investigation because of the current limited data, and given that the difference between the SB623 and sham surgery groups in Week 48 change from baseline was smaller at than Week 24.

6.R.2 Safety

The applicant conducted a categorized safety analysis of Akuugo, focusing on events recognized as significant risks associated with Akuugo treatment.

The applicant's explanation about causal relationships to Akuugo:

Nervous system disorders

In Study TBI-01, the incidence of all adverse events classified under "Nervous system disorders" (system organ class [SOC][18]) was 65.2% (30 of 46 subjects) in the SB623 group and 46.7% (7 of 15 subjects) in the sham surgery group. Postoperative adverse events observed in ≥10% of subjects in either the SB623 or sham surgery groups (preferred term [PT][18]) included headache (41.3% [19 of 46 subjects] in the SB623 group and 26.7% [4 of 15 subjects] in the sham surgery group) and dizziness (10.9% [5 of 46 subjects] in the SB623 group and 6.7% [1 of 15 subjects] in the sham group). Of 27 events of headache,[19] the most frequent, occurred in the SB623 group, 23 manifested from the day of surgery to 2 days post-surgery, and all recovered or improved. A causal relationship to the study product was ruled out for 22 of 27 events, while a causal relationship to surgical procedure could not be ruled out for 24 events. The higher incidence of adverse events in the SB623 group than in the sham surgery group could be attributable to different surgical procedures, although the influence of SB623 remained undeniable;

16) Two subjects assigned to the Akuugo transplant group discontinued early before transplantation at the discretion of the physician [see Footnote 6].
17) Handling of missing baseline FMMS score data [see Note 1 of Table 22].
18) Medical Dictionary for Regulatory Activities Japanese version (MedDRA) ver.18.0
19) Sum of "Headache" and "Procedural headache" in PT

the SB623 group involved the penetration of a burr hole into the subdural space, whereas in the sham group, only a superficial burr hole was made on the outer layer of the skull. A total of 4 serious adverse events occurred in 3 subjects in the SB623 group (2 seizures in 1 subject, balance disorder in 1 subject, and transient ischaemic attack in 1 subject), and in 1 subject in the sham group (1 event, seizure). All events resolved, except balance disorder unresolved. In the SB623 group, a causal relationship to the study product was ruled out for these events, while 1 each of seizure and balance disorder were assessed as causally related to the surgical procedure. A postoperative adverse event of cerebral or intracranial haemorrhage were noted in 2 subjects in the SB623 group and in 1 subject in the sham surgery group. A causal relationship to the study product was ruled out in both subjects of the SB623 group, while that to the surgical procedure was assessed as "related."

The incidence of "nervous system disorders" was higher in the SB623 group than in the sham group. However, in view of their seriousness and no causal relationship to the study product, the events will raise no particular safety concerns. Taking into account of a certain number of adverse events suggested to be causally related to surgical procedures, perioperative monitoring for these events will be requested via materials for healthcare professionals and patients, with caution against possible nervous system disorders.

Injury, poisoning and procedural complications

In Study TBI-01, the incidence of all adverse events classified under "Injury, poisoning and procedural complications" (SOC[18)]) was 65.2% (30 of 46 subjects) in the SB623 group and 60.0% (9 of 15 subjects) in the sham surgery group. Postoperative adverse events (PT[18)]) reported in ≥10% of subjects in either group were wound complication (26.1% [12 of 46 subjects] in the SB623 group, 20.0% [3 of 15 subjects] in the sham surgery group), procedural pain (4.3% [2 of 46 subjects] in the SB623 group, 13.3% [2 of 15 subjects] in the sham surgery group), and contusion (2.2% [1 of 46 subjects] in the SB623 group, 13.3% [2 of 15 subjects] in the sham surgery group). One serious adverse events occurred in 1 subject in the SB623 group (femur fracture [before transplant]) and 1 subject in the sham surgery group (traffic accident). Femur fracture resolved, and a causal relationship to both the study product or the surgical procedure was ruled out. A serious adverse event of head wound infection, under the "Infections and Infestations," was noted post-surgery in the sham surgery group.

A causal relationship to surgical procedures was suggested for a certain number of adverse events. Careful perioperative monitoring will be requested for these events via materials for healthcare professionals and patients, with caution against possible injuries, poisoning, and procedural complications.

Immune response

In Study TBI-01, the incidence of all adverse events suggestive of immune response (PT[18)] [pyrexia, pruritus, swelling face, fatigue, rhinorrhoea, oedema peripheral, nasal inflammation, dermatitis bullous, rash, and rosacea]) was 37.0% (17 of 46 of subjects) in the SB623 group and 33.3% (5 of 15 of subjects) in the sham surgery group. Events reported in ≥10% of subjects were pruritus (6.5% [3 of 46 subject] in the SB623 group, 13.3% [2 of 15 subjects] in the sham surgery group), and pyrexia (13.0% [6 of 46 subjects] in the SB623 group, 0% [0 of 15 subjects] in the sham group). No serious adverse events

suggestive of immune response occurred in either group. Although the incidence of pyrexia was higher in the SB623 group than in the sham surgery group, the occurrence of all adverse events suggestive of an immune response did not imply that immune response was triggered in the SB623 group. In Study TBI-01, 2 subjects exhibited an increase in donor-specific antibodies, and no clinically concerning adverse events were observed.

Neoplasm

No adverse events classified under the SOC[18)] of "Neoplasms benign, malignant and unspecified (incl cysts and polyps)" were observed in Studies TBI-01 and STR01. Study STR02 revealed 1 each of bladder cancer and colon adenoma in the SB623 group, and basal cell carcinoma in the sham surgery group. A causal relationship to both the study product or the surgical procedure was ruled out for these events. In view of non-clinical evaluations on tumorigenicity [see Section 5.2.1], the tumorigenic risk of SB623 is considered minimal.

Psychiatric disorders

In Study TBI-01, the incidence of adverse events classified under the SOC[18)] of "Psychiatric Disorders" was 17.4% (8 of 46 subjects) in the SB623 group and 13.3% (2 of 15 subjects) in the sham surgery group. No events were observed in ≥10% of subjects in both groups. Serious adverse events were observed in the SB623 group (delirium in 2 subjects) but not in the sham surgery group. Delirium in 2 subjects in the SB623 group resolved, and a causal relationship to the study product or surgical procedure was assessed as "related" in 1 subject.

Based on incidence and causality assessments, Akuugo is unlikely to raise significant safety concerns associated with psychiatric disorders. However, Akuugo is transplanted into the brain. In view of its risks of mood disorders and suicide risk in subjects with traumatic brain injury, the effect of Akuugo treatment on the central nervous system will be assessed in post-marketing clinical studies using the Columbia Suicide Severity Rating Scale (C-SSRS).

PMDA' view:

Adverse events noted in the SB623 group in Study TBI-01 did not reveal obviously problematic risks as compared to the sham surgery group, indicating acceptable safety of Akuugo at this point. Akuugo is intended for local intracranial administration via stereotactic surgery, which requires careful attention to adverse events observed in the clinical studies, including procedural complications such as intracranial hemorrhage. It is essential that Akuugo is used by physicians with adequate experience and knowledge in TBI treatment and stereotactic surgery who are also fully knowledgeable about Akuugo, and at well-equipped facilities with a system to address postoperative complications, etc. Because of the extremely limited number of subjects investigated in Study TBI-01, post-marketing data should be collected, and new findings should be offered to healthcare professionals appropriately.

6.R.3 Clinical positioning, indication or performance

At the time of application, the "Indication or Performance" of Akuugo was proposed as "Improvement of motor deficit after traumatic brain injury." The proposed "Precautions Concerning Indication or Performance" describes that "Akuugo should not be used in patients with acute TBI. Generally, motor

deficit stabilizes in 6 to 12 months post-injury, but the stabilization of motor deficit must be confirmed on a patient-by-patient basis. [Efficacy in patients with acute TBI has not been established.]"

PMDA's view:
Post-injury period and the severity of condition can significantly affect the efficacy and safety of Akuugo, and the clinical study data are extremely limited. Therefore, it is important that Akuugo's efficacy and safety be based on those demonstrated in subjects of Study TBI-01. Based on the discussions in Sections "6.R.1 Efficacy" and "6.R.2 Safety," and the outcomes described later, "Indication or Performance" and "Precautions Concerning Indication or Performance" for Akuugo should be described as follows:

Indication or Performance (Underline denotes additions and strikethrough denotes deletions.)
Improvement of chronic motor ~~deficit~~ paralysis associated with ~~after~~ traumatic brain injury

Precautions Concerning Indication or Performance (**Underline denotes additions and strikethrough denotes deletions.**)
~~Akuugo should not be used in patients with acute TBI. Generally, motor deficit stabilizes in 6 to 12 months post-injury, but the stabilization of motor deficit must be confirmed on a patient-by-patient basis. [Efficacy in patients with acute TBI has not been established.]~~

- Akuugo should be used in patients with moderate to severe motor deficit fixed for ≥6 months post-injury, with Glasgow Outcome Scale Extended (GOS-E) scores of 3 to 6.
- Akuugo should be used in patients with TBI having focal brain injury identifiable by MRI, etc. as responsible lesion for motor paralysis.
- Cellular proliferation may be promoted. The use of Akuugo should be carefully determined for patients who have brain tumors or a history of brain tumors, in light of its mechanism of action and tumor location, etc.
- The selection of eligible patients should be based on adequate knowledge from the "Clinical Studies" section, including the characteristics of the patients enrolled in the clinical studies, and a full understanding of the efficacy and safety of Akuugo.

6.R.3.1 Clinical positioning of Akuugo

The applicant's explanation:
The only therapeutic option for motor deficit associated with TBI is rehabilitation that is aimed at the preservation of functions for patients with chronic residual paralysis after acute or subacute treatments. There is no effective treatments intended for functional recovery.

Akuugo will be recognized as a novel treatment acting directly on the brain injury site to restore motor function in patients with chronic TBI beyond recovery.

PMDA's conclusion:
There are currently no effective treatments that promote functional recovery from chronic TBI with residual paralysis after acute or subacute interventions. Study TBI-01 demonstrated promising efficacy of Akuugo in improving motor paralysis with the acceptable safety profile. Thus, the provision of Akuugo as a new treatment is possible.

6.R.3.2 Target population, indication or performance of Akuugo

The applicant's explanation about the target population:

The inclusion criteria of Study TBI-01 specified participants' post-injury period as ≥12 months. This criterion was to ensure to select patients with stable motor deficit from central nervous system injury for the following reasons: Neurological recovery from TBI well stabilizes after 12 months; prolonged motor deficit can lead to the progression of contracture and decreased range of motion; and motor deficit can result from factors unrelated to central nervous system damage. However, "≥12 months post-injury" is merely a guideline for selecting patients with chronic impairment. In most patients, motor deficit stabilizes in approximately 6 months post-injury, the time period for the completion of recovery-phase inpatient rehabilitation. The effective target injury site of Akuugo will be the cortical motor area (precentral gyrus) or near the pyramidal tract, which is responsible for motor transmission. Due to the potential theoretical risk of tumor cell proliferation promoted by growth factors released by the transplanted cells, Akuugo should not be used in patients with brain tumors.

PMDA's view:

In light of the study population and primary efficacy endpoint in Study TBI-01, efficacy evaluation pertained only to the improvement of motor paralysis, and did not yield the outcomes pertaining to the improvement in overall motor deficit including involuntary movements. "Indication or Performance" should be, therefore, "improvement of chronic motor paralysis associated with traumatic brain injury."

The severity and post-injury period can have substantial impact on the efficacy and safety of Akuugo. Akuugo should be used in patients deemed eligible based on the criteria set in Study TBI-01. Eligible patients must have injury in the motor area or pyramidal tract with focal lesions identifiable by MRI, etc., which enables proper selection of the transplantation site. In patients with brain tumor, FGF-2 released from dying transplanted cells may promote tumor cell proliferation, as one of Akuugo's mechanism of action [see Section 3.R], and the theoretical risk of Akuugo to enhance tumor cell proliferation still remains. Even so, this theoretical risk does not support the contraindication of Akuugo in patients with current or prior brain tumors, unless the tumors are or were present near transplantation cells.

Based on the above, characteristics of patients participated in Study TBI-01 should be elaborated in the "Clinical Studies" section of the package insert, and the following advice should be given in Precautions Concerning Indication or Performance, and healthcare professionals should be provided with information to ensure the appropriate selection of patients.

- Akuugo should be used in patients with moderate to severe motor deficit fixed for ≥6 months post-injury, with Glasgow Outcome Scale Extended (GOS-E) scores of 3 to 6.
- Akuugo should be used in patients with TBI having focal brain injury identifiable by MRI, etc. as responsible lesion for motor paralysis.
- Cellular proliferation may be promoted. The use of Akuugo should be carefully determined for patients who have brain tumors or a history of brain tumors, in light of its mechanism of action and tumor location, etc.

- The selection of eligible patients should be based on adequate knowledge from the "Clinical Studies" section, including the characteristics of the patients enrolled in the clinical studies, and a full understanding of the efficacy and safety of Akuugo.

The descriptions will be finalized taking account of comments raised in the Expert Discussion.

6.R.4 Dosage and administration or method of use

The proposed Dosage and Administration or Method of Use was as follows:

Dosage and Administration or Method of Use

Usually, the cell suspension containing modified human (allogeneic) bone marrow-derived mesenchymal stem cell is prepared to contain 5×10^6 viable cells per dose (1.67×10^6 cells/100 μL), which is then transplanted to the surrounding area of the damaged tissue via stereotactic brain surgery.

1. Method of Use

See the manuals, etc. provided by the marketing authorization holder for detailed procedure.

1-1. Setup of invasive cranial fixation devices for neurosurgery

Before starting the surgical procedure using Akuugo, attach the guide and stop to the invasive cranial fixation device for neurosurgery. See the operating instructions for the detailed use of the accompanying invasive cranial fixation devices.

2-1. Preparation of cell suspension

(1) Thaw the cell suspension for intracranial transplantation (the primary component) that has been stored in the vapor phase of liquid nitrogen (≤–150°C). Add the dedicated preparation solution and stir gently.

(2) Centrifuge the obtained cell suspension. After removing the supernatant, add the dedicated preparation solution again to wash the suspension.

(3) Add the dedicated preparation solution and stir the suspension gently to repeat the procedure (2).

(4) After washing, count the viable cells and adjust the cell concentration using the dedicated preparation solution so as to obtain cell suspension at the concentration of 1.67×10^6 cells/100 μL for transplantation.

2-2. Preparation of administration cannula

(1) Secure the administration cannula to the microsyringe and cleanse both the microsyringe and the administration cannula with the dedicated preparation solution.

(2) Fill the microsyringe with the cell suspension from the plunger insertion side using the micropippeter.

2-3. Transplantation of Akuugo

(1) Using MRI images, etc., determine the site for 1 small opening on the skull as the starting point of 3 transplant routes that lead to the area surrounding the damage. Create a small opening on the skull as determined.

(2) Insert the stylet-equipped inserter through the small opening toward one of the transplant routes.

(3) Remove the stylet. Attach the cannula to the microsyringe filled with 100 μL of the cell suspension, and transplant it at 5 points 5 to 6 mm apart, from the deepest point. The cell suspension of approximately 20 μL should be injected per point at approximately 10 μL/min.

(4) Take the same procedure for the remaining 2 transplant routes.

On the basis of reviews in Sections "6.R.1 Efficacy," "6.R.2 Safety" and the following review, PMDA concluded that the "Dosage and Administration or Method of Use" for Akuugo should be modified as follows:

Dosage and Administration or Method of Use

(Underline denotes additions and strikethrough denotes deletions.)

Usually in adults, the cell suspension containing 5×10^6 viable (300 µL) of ~~modified~~ human (allogeneic) bone marrow-derived mesenchymal stem cells is ~~prepared to contain 5×10^6 viable cells per dose (1.67×10^6 cells/100µL), which is then~~ transplanted to the surrounding area of the damaged tissue via stereotactic brain surgery using a dedicated delivery device set. The cell suspension is injected through a single small opening on the skull that leads to 3 transplantation routes, reaching the area surrounding the injury. The dose of the cell suspension is 100 µL per route, 20 µL each of which is transplanted at 5 points 5 to 6 mm apart, from the deepest point. The infusion rate should be approximately 10 µL/min. The following steps should be taken prior to the transplantation.

~~1. Method of Use~~

~~See the manuals, etc. provided by the marketing authorization holder for detailed procedure.~~

~~1-1. Setup of invasive cranial fixation devices for neurosurgery~~

• Before starting the surgical procedure ~~using Akuugo,~~ attach the guide ~~and~~ & stop in the dedicated delivery device set and the stylet-equipped inserter to the invasive cranial fixation device for neurosurgery. ~~See the operating instructions for the detailed use of the accompanying invasive cranial fixation devices.~~

~~2-1. Preparation of cell suspension~~

• ~~(1)~~ Thaw the cell suspension for intracranial transplantation ~~(the primary component) that has been stored in the vapor phase of liquid nitrogen (≤ −150ºC). Add the dedicated preparation solution, and stir gently~~.

~~(2) Centrifuge the obtained cell suspension. After removing the supernatant, add the dedicated preparation solution again to wash the suspension~~.

~~(3) Add the dedicated preparation solution and stir the suspension gently to repeat the procedure (2).~~

~~(4)~~ After washing using the dedicated preparation solution, ~~count the viable cells and~~ adjust the cell concentration using the dedicated preparation solution so as to obtain cell suspension at the concentration of 1.67×10^6 cells/100 µL for transplantation.

~~2-2. Preparation of administration cannula~~

~~(1)~~ Cleanse ~~Secure~~ the administration cannula-fixed ~~to the~~ microsyringe in the dedicated delivery device set ~~and cleanse both the microsyringe and the administration cannula~~ using the dedicated preparation solution, and fill the microsyringe with the cell suspension.

~~(2) Fill the microsyringe with the cell suspension from the plunger insertion side using the micropippeter.~~

~~2-3. Transplantation of Akuugo~~

~~(1) Using MRI images, etc., determine the site for 1 small opening on the skull as the starting point of 3 transplant routes that leads to the area surrounding the damage. Create a small opening on the skull as determined.~~

~~(2) Insert a stylet-equipped inserter through the small opening toward one of the transplant routes.~~
~~(3) Remove the stylet. Attach the cannula to the microsyringe filled with 100 μL of the cell suspension, and transplant it at 5 points 5 to 6 mm apart, from the deepest point. The cell suspension of approximately 20 μL should be injected per point at approximately 10 μL/min.~~
~~(4) Take the same procedure for the remaining 2 transplant routes.~~

6.R.4.1 Cell count

The applicant's explanation regarding the rationale for setting the cell count:
In the change from baseline in FMMS score at Week 24, the primary efficacy endpoint of Study TBI-01, the mean change from baseline in FMMS score by dose group, was 6.0, 11.0, and 8.1 in the 2.5×10^6, 5.0×10^6, and 10.0×10^6 cell groups, respectively, showing the largest increase in the 5.0×10^6 cell group. After excluding 8 subjects with questionable blindness, the mean change from baseline in FMMS score by dose group was 7.2, 10.9, and 9.3, respectively, with the 5.0×10^6 cell group again showing the largest increase.

The incidence of postoperative headache[20] tended to increase with increased number of transplanted cells (26.7% in sham surgery group, 33.3% in 2.5×10^6 cell group, 40.0% in 5.0×10^6 cell group, 50.0% in 10.0×10^6 cell group). However, no other adverse events had a notably higher incidence in any specific dose group or an increased incidence with increasing dose.

Based on the above, the cell count of 5.0×10^6 was expected to offer the highest efficacy with a safety profile comparable to other doses. Accordingly, the number of transplanted cells for Akuugo was determined as 5.0×10^6.

PMDA's view:
Study TBI-01 was an exploratory study with a pooled Akuugo group consisting of 3 dose levels compared against the sham surgery group. Comparisons of each Akuugo dose group against the sham surgery group were not designed with adjusted multiplicity of hypothesis testing. In addition, the small number of subjects in respective Akuugo dose groups that was only approximately 15 each, and no clear reason for the decreased efficacy in the 10.0×10^6 cell group, higher dose, preclude a conclusion that the 5.0×10^6 cell dose has greater efficacy than the other dose levels. The applicant's observations on adverse events that the frequency of headaches was dose-dependent does not seem to be reproducible, in light of the incidence of postoperative headache[21] by dose group in the clinical studies on cerebral infarction (Study STR01, 100% [6 of 6 subjects] in the 2.5×10^6 cell group, 66.7% [4 of 6 subjects] in the 5.0×10^6 cell group, 83.3% [5 of 6 subjects] in the 10.0×10^6 cell group; Study STR02, 67.3% [37 of 55 subjects] in the 2.5×10^6 cell group, 73.2% [41 of 56 subjects] in the 5.0×10^6 cell group; and 71.2% [37 of 52 subjects] in the sham surgery group). Nevertheless, conducting further investigation on the efficacy and safety of Akuugo at the dose of 5.0×10^6 cells is considered acceptable in view of available information. These dose were, however, determined based on clinical study results in adults, which must be clearly highlighted.

[20] "Headache" and "Procedural headache" in PT
[21] "Headache" and "Procedural headache" in PT

6.R.4.2 Details of the transplantation method, including selection of the transplantation site

The rationale for the transplantation method explained by the applicant:
The transplantation of 20 μL per site 5 to 6 mm apart was determined based on the clinical study on cellular transplantation in patients with cerebral infarction (e.g., *Neurology.* 2000;55:565-9) that was underway at the time of planning of Study TBI-01.

The applicant's explanation about the selection of transplantation sites, based on the procedural manual used in Study TBI-01:

- The procedure targets an area near the motor cortex and pyramidal tract (corticobulbar and corticospinal tracts) responsible for motor nerve conduction.
- The transplantation site is specified by coordinate axes (X, Y, Z) using navigation software, based on previous MRI images overlaid with CT images taken at the facility or MRI images from the facility alone.
- The transplantation site is determined within subcortical perilesional tissue surrounding the brain injury area closest to motor nerve pathways, depending on the patient's neural structure.
- A total of 3 transplantation routes are determined to encircle the injury site, avoiding cerebral blood vessels, sulci, and ventricles.

PMDA's view:
Akuugo is expected to locally exert its effect [see Section 4.R] and has risks of procedural complications [see Section 6.R.2], and the method of transplantation including site determination is significantly influential to efficacy and safety. Akuugo should be used by physicians with adequate experience and knowledge in TBI treatment and stereotactic brain surgery. Furthermore, the transplantation method should be specified in "Dosage and Administration or Method of Use" based on the procedure in Study TBI-01, and the transplantation procedure, including prior selection of the transplantation site, should be explained in details through written materials.

6.R.5 Use in pediatric patients

The applicant's explanation about use in pediatric patients:
The subjects of Study TBI-01 were 18 years or older. There are no efficacy or safety data of patients aged <18 years and, preferably, pediatric use of Akuugo should be avoided. Having said that, the need of Akuugo will rise in patients aged <18 years as well. Stereotactic brain surgery has been employed in pediatric cases (*Eur J Pediatr Neurol.* 2017;21:168-75, *Neurosurgery.* 2011;68:738-43). In teenagers, the volume of the cerebral cortex reaches adult levels (*Nat Neurosci.* 1999;2:861-3). With these views, pediatric use of Akuugo is considered possible.

PMDA's view:
Considering the lack of study data demonstrating efficacy and safety in pediatric patients at present, Akuugo should be used in adults as a rule. However, there are limited treatment options for chronic motor paralysis from TBI, and the transplantation may be feasible in children who have reached adult body size. Information about actual pediatric cases of Akuugo transplantation should be collected in an appropriate manner.

The conclusion will be finalized taking account of the comments raised in the Expert Discussion.

6.R.6 Use in elderly patients

The applicant's explanation:

Study TBI-01 targeted patients aged 18 to 75 years. The mean age (mean ± SD [Min-Max]) of enrolled subjects was 34.40 ± 11.77 (18.5-67.5) years. Subjects aged ≥60 years were included in the SB623 group (2 subjects) and the sham-surgery group (1 subject). In these subjects, the change from baseline in FMMS score at Week 24 was 4 and 6 in the SB623 group and –7 in the sham-surgery group. Each of the 3 subjects aged ≥60 years experienced adverse events. Serious adverse events included delirium in 1 subject in the SB623 group and road traffic accident in 1 subject in the sham-surgery group. A causal relationship to Akuugo or the procedure was ruled out for delirium and the road traffic accident. Despite the small number of elderly subjects in Study TBI-01, FMMS score from baseline to Week 24 showed improvement without frequent serious adverse events, indicating no major concerns on efficacy or safety of Akuugo in elderly patients. The use of Akuugo is considered possible with careful monitoring of patient condition.

PMDA's view:

In Study TBI-01, the majority of subjects were relatively young, limiting experience in elderly subjects. Nevertheless, the study results showed promising efficacy in patients aged ≥60 years with acceptable safety, indicating the possibility of the use of Akuugo in elderly patients. However, particularly elderly patients can have ongoing non-TBI associated motor paralysis, thus the use of Akuugo should be determined with clear understanding of the advice given in "Precautions Concerning Indication or Performance" after identifying responsible lesions, etc. Information on the efficacy and safety in elderly patients should be collected in the post-marketing setting in an appropriate manner.

This conclusion will be finalized taking account of comments raised in the Expert Discussion.

7. Risk Analysis and Outline of the Review Conducted by PMDA

7.1 Post-marketing investigations

7.1.1 Post-marketing clinical study

The applicant's explanation about the plan for the post-marketing clinical study on Akuugo:

Study TBI-01 was designed as an exploratory study, in which only limited number of subjects received the cell transplantation at the proposed dose of 5.0×10^6. Therefore, post-marketing efficacy and safety of Akuugo needed to be assessed in a confirmatory manner. The post-marketing clinical study was planned as follows.

Table 37. Outline of the post-marketing clinical study (draft)

Objective	To confirm the efficacy of Akuugo in patients with chronic motor deficit from TBI
Study design	A multicenter, randomized, open-label, parallel-group study. Patients are allocated in a 2:1 ratio to either a group receiving Akuugo transplantation plus rehabilitation (Akuugo group) or a group receiving rehabilitation alone (control group).
Population	Adults with stable chronic motor deficit from TBI During a screening period, patients will receive rehabilitation. FMMS scores are measured every 4 weeks. Motor deficit is considered stable if the most recent 2 consecutive FMMS scores show fluctuations within ± 10%, and the patient proceeds to randomization.
Observation period	48 weeks after starting treatment and observation (Akuugo group, after transplantation) (with a separate ≥4-week screening period)
Dosage and administration and method of use	Akuugo group: As per the dosage and administration or usage method of Akuugo. After transplantation, patients undergo rehabilitation considered appropriate by the attending physician. Control group: No transplantation of Akuugo is performed. Patients are provided with rehabilitation that the attending physician consider appropriate.
Number of study sites	Surgery: 5-7 sites Rehabilitation: 10-21 sites
Primary efficacy endpoint	Change from baseline in FMMS score at Week 24 Third-party evaluators assess FMMS scores in a blinded manner.
Primary efficacy endpoint analysis	The primary analysis compares the change from baseline in FMMS score at Week 24 in the Akuugo group to that in the control groups. MMRM is employed assuming an unstructured covariance structure with allocated group, visit time, baseline FMMS score, and the interaction between allocated group and visit time as covariates.
Secondary efficacy endpoints	• Change from baseline in FMMS score at Week 48 • Change from baseline in UE-FM score and LE-FM score at Weeks 24 and 48 • Change from baseline in FIM motor score at Weeks 24 and 48 • Change from baseline in total FIM score at Weeks 24 and 48
Safety endpoints	• Death, adverse events leading to study discontinuation, serious adverse events, adverse events • Adverse events of special interest: Neuropsychiatric symptoms, immune response, infections, cerebral hemorrhage, cognitive dysfunction, suicide, and suicidal ideation
Target sample size	40 patients (30 in the Akuugo group, 10 in the control group) Rationale: In Study TBI-01, the mean ± SD of the change from baseline in FMMS score at Week 24 was 11.0 ± 8.4 in the group receiving 5.0 × 106 cells and 2.3 ± 4.7 in the sham surgery group. Assuming the reproducibility of these results, 32 patients need to be allocated to the Akuugo or control group in 3:1 for efficacy analysis, with a two-sided significance level of 5% and a power of 90% as per Study TBI-01. There were no dropouts after Akuugo transplantation in Study TBI-01, a double-blind clinical study. However, in this study, for being a post-marketing clinical study conducted in an open-label manner, the target sample size is 40, assuming Week-24 dropout rate as approximately 20%.

In the post-marketing clinical study, it is ethically difficult to perform only sham surgery in the control group as in Study TBI-01. Therefore, the study should be designed with a group of patients who will not undergo Akuugo transplantation but receive individually-tailored appropriate rehabilitation program.

The primary efficacy endpoint is the change from baseline in FMMS score at Week 24.

7.1.2 Use-results survey

The applicant planned a use-results survey as outlined in Table 38 to collect data from patients who are not included in the post-marketing clinical study. It should be noted that the FMMS score is not always used in clinical practice in the treatment of stable chronic motor deficit from TBI. Unlike in the clinical studies, there is a difficulty assessing FMMS score strictly in the use-results survey, and the FMMS score is, therefore, not included in efficacy assessment items.

Table 38. Outline of the use-results survey (draft)

Objective	To investigate the safety and efficacy of Akuugo in clinical use
Target patients	All patients receiving Akuugo transplant (excluding patients assessed in the post-marketing clinical study).
Observation period	48 weeks after Akuugo transplantation
Survey period	Survey period: 6 years from the market launch Registration period: 4 years from the market launch (Assuming a 7-year approval period)
Safety survey items	Occurrences of adverse events Key survey items: Neuropsychiatric symptoms, symptoms associated with immune response, infections, cerebral hemorrhage, cognitive impairment, and suicide and suicidal ideation
Efficacy survey items	FIM, Motricity Index, general assessment score of subjective and objective changes
Planned sample size	70 patients

7.R Outline of the review conducted by PMDA

PMDA's view:

Given the limited clinical experience with Akuugo, its efficacy and safety should be evaluated in the post- marketing setting. The applicant's plan for the post-marketing clinical study aiming at efficacy and safety assessments is considered appropriate.

In order to ensure the objectivity of post-marketing clinical study evaluations, the study should have been designed as randomized, double-blind, parallel-group study. At the same time, the applicant's concern, the difficulty in performing sham surgery in control patients in the post-marketing setting, is understandable. Thus, the proposed approach, i.e., FMMS score-based efficacy evaluation by blinded assessors, should be inevitably accepted to maintain the objectivity of evaluation. In light of potential impact of rehabilitation on efficacy, it is important that study sites prepare for the same rehabilitation program for both groups.

The proposed efficacy evaluation in the post-marketing clinical study is primarily based on the FMMS score as in Study TBI-01 for comparison with the control group. Akuugo treatment is aimed to improve motor paralysis. The proposed dose of Akuugo is subject to further investigation, as Study TBI-01 did not compare each dose group against the control. In these views, the efficacy evaluation in the post-marketing study has been designed appropriately.

The safety evaluation items are considered appropriate based on the review in Section "6.R.2 Safety."

The proposed observation period set for efficacy and safety items are considered appropriate based on the reviews in Sections "6.R.1 Efficacy" and "6.R.2 Safety."

Efficacy and safety information of Akuugo should be collected from patients who do not participate in the post-marketing clinical study. To this end, the target of the use-results survey, all patients receiving Akuugo (excluding patients evaluated in the post-marketing clinical study), is appropriate. As in the post-marketing clinical study, efficacy items of the survey should include FMMS score that is commonly used to evaluate efficacy against motor paralysis, which should be collected for evaluation. The safety items and observation period in the use-results survey are similar to those in the post-marketing clinical study, and are considered appropriate.

The details of the post-marketing clinical study and the use-results survey will be finalized taking account of comments on the efficacy and safety evaluation of Akuugo raised in the Expert Discussion.

8. Results of Compliance Assessment Concerning the New Regenerative Medical Product Application Data and Conclusion Reached by PMDA

8.1 PMDA's conclusion concerning the results of document-based GLP/GCP inspections and data integrity assessment

The new regenerative medical product application data were subjected to a document-based inspection and a data integrity assessment in accordance with the provisions of the Act on Securing Quality, Efficacy and Safety of Products Including Pharmaceuticals and Medical Devices. On the basis of the inspection and assessment, PMDA concluded that there were no obstacles to conducting its review based on the application documents submitted.

8.2 PMDA's conclusion concerning the results of the on-site GCP inspection

The new regenerative medical product application data (CTD5.3.5.1-1) were subjected to an on-site GCP inspection, in accordance with the provisions of the Act on Securing Quality, Efficacy and Safety of Products Including Pharmaceuticals and Medical Devices. On the basis of the inspection, PMDA concluded that the clinical studies were generally conducted in compliance with GCP, and that there were no obstacles to conducting its review based on the application documents submitted. PMDA pointed out the following error made by the sponsor, although it had no significant impact on the review of the overall clinical studies. PMDA notified the sponsor of the error.

Finding requiring corrective action

Sponsor

- SOPs pertaining to the preparation of the study protocol and the investigator's brochure were not ready at the start of the clinical study.

9. Overall Evaluation during Preparation of the Review Report (1)

PMDA's view:

The data submitted have not adequately demonstrated the comparability between the study product used in Study TBI-01 and the commercial process formulation [see Section 2.R.2]. However, when assuming the comparability between the two, Akuugo is expected to have a certain level of efficacy in improving chronic motor paralysis associated with TBI, and that Akuugo has acceptable safety in view of its benefits. Although current efficacy and safety information of Akuugo is limited, it is considered significant to provide Akuugo to clinical settings as a treatment option for the improvement of TBI-associated chronic motor paralysis.

When the comparability between the study product and the commercial process formulation is demonstrated in further investigations, and no particular efficacy or safety concerns of Akuugo are raised by the Expert Discussion, Akuugo may be approved with conditions and timelines based on Article 23-26 of the Act on Securing Quality, Efficacy, and Safety of Pharmaceuticals and Medical Devices, which will require continued efficacy assessment and safety data collection in a specified time period post-marketing. The timeline as stipulated in this article will be determined with consideration of the post-

marketing clinical study and use-result survey plans, (time required for marketing preparation, enrollment, patient observation, and preparation for application, etc.) as well as the outcomes of the Expert Discussion.

Review Report (2)

March 13, 2024

Product Submitted for Approval

Brand Name	Akuugo Suspension for Intracranial Implantation
Non-proprietary Name	Vandefitemcel
Applicant	SanBio Company Limited
Date of Application	March 7, 2022

List of Abbreviations
See Appendix.

1. Content of the Review

Comments made during the Expert Discussion and the subsequent review conducted by the Pharmaceuticals and Medical Devices Agency (PMDA) are summarized below. The expert advisors present during the Expert Discussion were nominated based on their declarations etc. concerning the product submitted for marketing approval, in accordance with the provisions of the Rules for Convening Expert Discussions etc. by Pharmaceuticals and Medical Devices Agency (PMDA Administrative Rule No. 8/2008 dated December 25, 2008).

The proposed brand name of the product has been changed from "Akuugo Cell Suspension for Intracranial Transplantation" to "Akuugo Suspension for Intracranial Implantation."

1.1 Efficacy

On the basis of review in Section "6.R.1 Efficacy" of the Review Report (1), PMDA concluded that Akuugo is expected to have a certain level of efficacy in improving motor paralysis.

Because of the extremely limited current information, the efficacy of Akuugo is subject to further evaluation in the post-marketing setting.

The above conclusions of PMDA were supported by the expert advisors at the Expert Discussion.

1.2 Safety

PMDA's conclusion:
On the basis of review in Section "6.R.2 Safety" of the Review Report (1), the safety of Akuugo is considered acceptable at this point because adverse events in the SB623 group of Study TBI-01 did not indicate any significant risk as compared to the sham surgery group. Akuugo is a product locally administered to the brain via stereotactic surgery. Potential procedure-associated complications, such as intracranial hemorrhage, and events observed in the clinical studies of Akuugo deserve thorough attention. PMDA concluded that Akuugo should be used by physicians with adequate knowledge and

experience in TBI treatment and stereotactic surgery who are also fully knowledgeable about Akuugo, and at well-equipped facilities with a system to address postoperative complications.

The above conclusions of PMDA were supported by the expert advisors at the Expert Discussion.

1.3 Clinical positioning and indication or performance

As a result of the review in Section "6.R.3 Clinical positioning, indication or performance" of the Review Report (1), PMDA concluded that the "Indication or Performance" and "Precautions Concerning Indication or Performance" sections should be described as follows, while the characteristics of patients in Study TBI-01 are detailed in the "Clinical Studies" section of the package insert.

Indication or Performance

Improvement of chronic motor paralysis associated with traumatic brain injury

Precautions Concerning Indication or Performance

- Akuugo should be used in patients with moderate to severe motor deficit fixed for ≥6 months post-injury, with Glasgow Outcome Scale Extended (GOS-E) scores of 3 to 6.
- Akuugo should be used in patients with TBI having focal brain injury identifiable by MRI, etc. as responsible lesion for motor paralysis.
- Cellular proliferation may be promoted. The use of Akuugo should be carefully determined for patients who have brain tumors or a history of such tumors, in light of its mechanism of action and the tumor location, etc.
- The selection of eligible patients should base on adequate knowledge from the "Clinical Studies" section, including the characteristics of the patients enrolled in the clinical studies, and a full understanding of the efficacy and safety of Akuugo.

The above conclusions of PMDA were supported by the expert advisors at the Expert Discussion.

PMDA requested the applicant to modify the "Indication or Performance" and "Precautions Concerning Indication or Performance" sections as above. The applicant appropriately responded, and PMDA accepted.

1.4 Dosage and administration or method of use

As a result of the review in Section "6.R.4 Dosage and administration or method of use" of the Review Report (1), PMDA has concluded that the "Dosage and Administration or Method of Use" section should be described as follows based on the Study TBI-01:

Dosage and Administration or Method of Use

Usually in adults, the cell suspension containing 5×10^6 viable cells (300 μL) of human (allogeneic) bone marrow-derived mesenchymal stem cells is transplanted to the surrounding area of the damaged tissue via stereotactic brain surgery using a dedicated delivery device set. The cell suspension is injected through a single small opening on the skull that leads to 3 transplantation routes, reaching the area

surrounding the injury. The dose of the cell suspension is 100 μL per route, 20 μL each of which is transplanted at 5 points 5 to 6 mm apart, from the deepest point. The infusion rate should be approximately 10 μL/min. The following steps should be taken prior to the transplantation.

- Before starting the surgical procedure, attach the guide & stop in the dedicated delivery device set and the stylet-equipped inserter to the invasive cranial fixation device for neurosurgery.
- Thaw the cell suspension for intracranial transplantation. After washing using the dedicated preparation solution, adjust the cell concentration using the dedicated preparation solution so as to obtain cell suspension at the concentration of 1.67×10^6 cells/100 μL for transplantation. Cleanse the administration cannula-fixed microsyringe in the dedicated delivery device set using the dedicated preparation solution, and fill the microsyringe with the cell suspension.

The above conclusions of PMDA were supported by the expert advisors at the Expert Discussion.

PMDA requested the applicant to modify the "Dosage and Administration or Method of Use" section as above. The applicant appropriately responded to the request, and PMDA accepted.

1.5 Plan for post-marketing approval condition evaluation (draft)

The applicant submitted draft plans for a post-marketing clinical study and a use-results survey to further evaluate post-marketing efficacy and safety of Akuugo.

Based on the review outlined in Section "7. Risk Analysis and Outline of the Review Conducted by PMDA," PMDA has concluded that it is appropriate to gather FMMS score, a common tool to assess efficacy against motor paralysis, for efficacy evaluation in the use-results survey, as in the post-marketing clinical studies.

PMDA's conclusions were supported by the expert advisors at the Expert Discussion. The expert advisors also raised the following views for further discussion.

- In view of the risk of brain tumors, etc., data on post-transplant long-term safety of Akuugo should be collected as much as possible.
- There is little justification for a 3:1 allocation ratio between the Akuugo group and the control group in the post-marketing clinical study aimed to confirm the efficacy of Akuugo.

Based on the results of the Expert Discussion, PMDA requested the applicant to review their post-marketing surveillance plan. In response, the applicant submitted the outline of the post-marketing clinical study (draft) and the outline of the use-results survey (draft) as in Tables 39 and 40, respectively, which include the following corrections. PMDA concluded that the post-marketing approval condition evaluation should be conducted based on these plans.

Main changes:

- The observation period of the use-results survey should be specified as "from the day of transplantation until the day of decision on the re-application, which is to be filed within the time frame of conditional and time-limited approval" to allow for the collection of long-term safety data. "Brain tumor" should be added to adverse events of special interest in the safety endpoints of the

post-marketing clinical study and safety survey items of the use-results survey. Patients undergoing Akuugo transplantation and assessment in the post-marketing clinical study will be enrolled in the use-results survey for further long-term safety data collection, after the completion or discontinuation of assessment in the post-marketing clinical study.

- The allocation ratio between the Akuugo group and the control group in the post-marketing clinical study is to be 2:1. With this change, the target sample size will be changed to 42 (28 in the Akuugo group, 14 in the control group).
- FMMS score is to be added in in the use-results survey efficacy specification.

Table 39. Outline of the post-marketing clinical study (draft)

Objective	To confirm the efficacy of Akuugo in patients with chronic motor deficit from TBI.
Study design	A multicenter, randomized, open-label, parallel-group study. Patients are allocated in a 2:1 ratio to either a group receiving Akuugo transplantation plus rehabilitation (Akuugo group) or a group receiving rehabilitation alone (control group).
Population	Adults with stable chronic motor deficit from TBI During a screening period, patients will receive rehabilitation. FMMS scores are measured every 4 weeks. Motor deficit is considered stable if the most recent 2 consecutive FMMS scores show fluctuations within ± 10%, and the patient will proceed to randomization.
Observation period	48 weeks after starting treatment and observation (Akuugo group, after transplantation) (with a separate ≥4-week screening period). Patients in the Akuugo group who have completed or withdrawn from the evaluation in the post-marketing clinical study are enrolled in the use-results survey for further observation.
Dosage and administration and method of use	Akuugo group: As per the dosage and administration or usage method of Akuugo. After transplantation, patients are provided with rehabilitation that the attending physician considers appropriate. Control group: No transplantation of Akuugo is performed. Patients are provided with rehabilitation that the attending physician considers appropriate
Number of study sites	Surgery: 5-7 sites Rehabilitation: 10-21 sites
Primary efficacy endpoint	Change from baseline in FMMS score at Week 24 Third-party evaluators assess FMMS scores in a blinded manner.
Primary efficacy endpoint analysis	The primary analysis compares the change from baseline in FMMS score at Week 24 in the Akuugo group to that in the control groups MMRM is employed assuming an unstructured covariance structure with allocated group, visit time, baseline FMMS score, and the interaction between group allocated and visit time as covariates.
Secondary efficacy endpoints	• Change from baseline in FMMS score at Week 48 • Change from baseline in UE-FM and LE-FM scores at Weeks 24 and 48 • Change from baseline in FIM motor score at Weeks 24 and 48 • Change from baseline in total FIM score at Weeks 24 and 48
Safety endpoints	• Death, adverse events leading to study discontinuation, serious adverse events, and adverse events • Adverse events of special interest: Neuropsychiatric symptoms, immune response, infections, cerebral hemorrhage, cognitive impairment, suicide, suicidal ideation, and brain tumor
Target sample size	42 patients (28 in the Akuugo group, 14 in the control group) Rationale: In Study TBI-01, the mean ± SD of the change from baseline in FMMS score at Week 24 was 11.0 ± 8.4 in the group receiving 5.0×10^6 cells and 2.3 ± 4.7 in the sham surgery group. Assuming a common SD as 6.8, 33 patients need to be allocated to the Akuugo or control group at 2:1 allocation ratio for efficacy analysis, with a two-sided significance level of 5% and a power of 90% based on the t-test. There were no dropouts after Akuugo transplantation in Study TBI-01, a double-blind clinical study. However, in this study, for being a post-marketing clinical study conducted in an open-label manner with a control group, the target sample size is 42, assuming Week 24 dropout rate as approximately 20%.

Table 40. Outline of the use-results survey (draft)

Objective	To investigate the safety and efficacy of Akuugo in clinical use.
Target patients	All patients receiving Akuugo transplant (patients who undergo the procedure and assessment in the post-marketing clinical study are enrolled after the completion or discontinuation of assessment in the post-marketing clinical study).
Observation period	From the day of transplantation until the day of decision on the approval of re-application, which is to be filed within the timeframe of conditional and time-limited approval*
Safety survey items	Incidence of adverse events Key survey items: Neuropsychiatric symptoms, symptoms associated with immune response, infections, cerebral hemorrhage, cognitive impairment, suicide and suicidal ideation, and brain tumor
Efficacy survey items	FMMS score, FIM, Motricity Index, and general assessment score of subjective and objective changes
Planned sample size	Approximately 25 cases per year

* Data will be aggregated for analysis after the cut-off at 18 months before the reapplication for approval and will be attached with the reapplication filed within the conditional time-limited approval period, which is assumed as 7 years.

1.6 Manufacturing process of the primary component (SB623)

Based on the review in the Section "2.R.2 Manufacturing process of the primary component (SB623)" in the Review Report (1), PMDA instructed the applicant to provide (a) the results of additional investigations on the comparability among the commercial process formulation, the proposed process formulation, and the study product used in Study TBI-01, and (b) the duration and evaluation items of future stability studies on critical intermediates.

The applicant's explanation:

(a) Results of additional investigations on the comparability among the commercial process formulation, the proposed process formulation, and the study product used in Study TBI-01

For the cell surface marker analysis in the specification test, in addition to the calculation of positive cell the percentage, the expression level (fluorescence intensity) of each marker was compared based on the fluorescence intensity of individual cells obtained from histograms. The expression level (fluorescence intensity) of each cell surface marker in the commercial process formulation was similar to that of the study product used in Study TBI-01, and was within the range of the proposed process formulation, suggesting equivalence with both the study product and the proposed process formulation (Table 41).

Table 41. Expression level of cell surface marker (fluorescence intensity)

Manufacturing process	Clinical study process		Proposed process							Commercial process
Batch No.	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]

Currently, no additional data are available from comparability assessments other than the above. For further studies, the development of a test system is underway to obtain data on [redacted], [redacted], [redacted], and [redacted].

Before starting the shipment of the commercial product post-approval, 3 batches of the commercial process formulation will be produced and assessed for the comparability among the commercial process formulation, the proposed process formulation, and the study product used in Study TBI-01 in term of the following:

- In-process control test items, specification test items, and characterization test items [see Tables 1, 7, and 9]
- Other comparability assessment items ([redacted], [redacted], [redacted], [redacted], and [redacted])

(b) Duration and evaluation items of future stability studies on critical intermediates

In the future stability studies on the critical intermediate, [redacted] will be tested for stability up to [redacted] months.

PMDA's view:

The quality attributes of 1 batch of the commercial process formulation did not show a significant deviation from 7 batches of the proposed process formulation and 2 batches of the study product. Because of limited information obtained after the compilation of the Review Report (1), the comparability between the study product used in Study TBI-01 and the commercial process formulation has yet to be confirmed until now. The applicant needs to collect data on the quality of Akuugo promptly through additional production of the commercial process formulation and assess the comparability with the study product.

The shelf life of the primary component (SB623), which will be assessed based on the stability study data of the proposed process formulation, needs to be determined based on the results of comparability between the commercial process formulation and the proposed process formulation.

The shelf life of critical intermediate needs to be determined based on the results of the stability study, the proposed duration and evaluation items of which are acceptable.

The mechanism of action of Akuugo against TBI has not been fully elucidated. Despite possible importance of pharmacological actions mediated by multiple humoral factors derived from bone marrow MSCs in the brain post-TBI, the potency test has been conducted only on intracellular FGF-2, which is currently understood as the biological properties showing the mechanism of action of Akuugo [see Sections 2.R.2 and 3.R]. The applicant should further collect information on biological properties of Akuugo to clarify its action mechanism against TBI and conduct potency tests on biological properties that reflect of Akuugo's action mechanisms, other than intracellular FGF-2.

1.7 Others

1.7.1 Designation of specified regenerative medical product

On the basis of "Principles for designation of biological products, specified biological products, and specified regenerative medical products" (PFSB/ELD Notification No. 1105-1 and No. 1105-2, dated

November 5, 2014), PMDA has concluded that Akuugo should be designated as a specified regenerative medical product, because it is a regenerative medical product that uses cells derived from homologous sources as raw material.

2. Overall Evaluation

Based on the above review, the decision on the approval of Akuugo be made taking into account the results of the additional comparability assessment between the study product and Akuugo. PMDA has concluded that once their comparability, which remains unclear at this point, is proven, the product will have a certain level of efficacy in improving chronic motor paralysis associated with TBI, and that the product has acceptable safety in view of its benefits. Albeit current limited efficacy and safety data, it is considered significant to provide Akuugo to the clinical setting as one of treatment options for chronic motor paralysis associated with TBI.

Appendix

List of Abbreviations

AE	adverse event
Akuugo	Akuugo Suspension for Intracranial Implantation
Application	application for marketing approval
ARAT	Action Research Arm Test
BDNF	brain derived neurotrophic factor
bFGF	basic fibroblast growth factor
BMP	bone morphogenetic protein
BSE	bovine spongiform encephalopathy
CD	cluster of differentiation
CI	confidence interval
CM	conditioned medium
CMV	cytomegalovirus
CNP	2',3'-cyclic nucleotide 3'-phosphodiesterase
CPP	critical process parameter
CQA	critical quality attribute
C-SSRS	Columbia Suicide Severity Rating Scale
Dcx	doublecortin
DKK-1	dickkopf
D-PBS	Dulbecco's phosphate-buffered saline
DRS	Disability Rating Scale
EBST	elevated body swing test
EBV	Epstein-Barr virus
ECM	extracellular matrix
EGF	epidermal growth factor
FBS	fetal bovine serum
FGF	fibroblast growth factor
FGFR	fibroblast growth factor receptor
FIM	Functional Independence Measure
FMMS	Fugl-Meyer Motor Scale
GABA	γ-aminobutyric acid
GCP	good clinical practice
GCTP	good gene, cellular, and tissue-based products manufacturing practice
GFAP	glial fibrillary acidic protein
GOS-E	Glasgow Outcome Scale-Extended
HB-EGF	heparin binding-epidermal growth factor
HBV	hepatitis B virus
HCV	hepatitis C virus
HeLa cell	Human cervical cancer cell
HGF	hepatocyte growth factor
HIV	human immunodeficiency virus
HTLV	human T-cell leukemia virus
HUVEC	human umbilical vein endothelial cell
IL	interleukin
JCV	JC virus
LDH	lactate dehydrogenase
LE-FM	lower-extremity subscale of the Fugl-Meyer Motor Test
[redacted]	[redacted]
MCP-1	monocyte chemoattractant protein
MedDRA	Medical Dictionary for Regulatory Activities Japanese version
MHC	major histocompatibility complex
MMP-1	matrix metalloproteinase 1

MMRM	mixed effect models for repeated measures
[redacted]	[redacted]
MRC-5 cell	Human fetal lung fibroblast cell
[redacted]	[redacted]
MSC	mesenchymal stem cell
NeuroQOL	Quality of Life in Neurological Disorders
NGF	nerve growth factor
OGD	oxygen-glucose deprivation
pCPP	potential critical process parameter
[redacted]	[redacted]
PDGF	platelet derived growth factor
PDL	poly-D-lysine
PMDA	Pharmaceuticals and Medical Devices Agency
PT	preferred term
PVB19	parvovirus B19
QOL	quality of life
qPCR	quantitative polymerase chain reaction
RT-qPCR	reverse transcriptase quantitative polymerase chain reaction
SMQ	standardised MedDRA queries
SOC	system organ class
SVZ	subventricular zone
TBI	traumatic brain injury
tMCAo	transient middle cerebral artery occlusion
UE-FM	upper-extremity subscale of the Fugl-Meyer Motor Test
VEGF	vascular endothelial growth factor
Vero cell	African green monkey kidney epithelial cell
VGAT	vesicular GABA transporter
VGLUT	vesicular glutamate transporter
WHO	World Health Organization
WNV	West Nile virus

Second Review Report

June 6, 2024
Pharmaceuticals and Medical Devices Agency

The following are the results of the review of the following regenerative medical product submitted for marketing approval conducted by the Pharmaceuticals and Medical Devices Agency (PMDA).

Brand Name	Akuugo Suspension for Intracranial Implantation
Classification	Human Cellular/Tissue-based Products 2. Human Somatic Stem Cell-processed Products
Non-proprietary Name	Vandefitemcel
Applicant	SanBio Company Limited
Date of Application	March 7, 2022

Shape, Structure, Active Ingredients, Quantities, or Definition

The product consists of cells derived from human (allogeneic) bone marrow mesenchymal stem cells, into which the human Notch-1 intracellular domain gene has been transfected using a pN-2 plasmid vector.

Application Classification	(1-1) New regenerative medical product
Items Warranting Special Mention	Orphan regenerative medical product (Orphan Regenerative Medical Product Designation No. 19 of 2020 [*R2 sai*]; PSEHB/MDED Notification No. 0623-4 dated June 23, 2020, by the Medical Device Evaluation Division, Pharmaceutical Safety and Environmental Health Bureau, Ministry of Health, Labour and Welfare) SAKIGAKE designation regenerative medical product (SAKIGAKE Regenerative Medical Product Designation No. 2 of 2018 [*30 sai*]; PSEHB/MDED Notification No. 0408-17 dated April 8, 2019, by the Medical Device Evaluation Division, Pharmaceutical Safety and Environmental Health Bureau, Ministry of Health, Labour and Welfare), Sakigake comprehensive assessment consultation conducted for regenerative medical products
Reviewing Office	Office of Cellular and Tissue-based Products

Results of Review

Based on the currently available information, it is difficult to reach a conclusion on the comparability between the study product and Akuugo (see Attachment).

As described in the review report dated March 15, 2024, when the comparability with the study product is proven, Akuugo will have a certain level of efficacy in improving chronic motor paralysis associated with traumatic brain injury, and the product has acceptable safety in view of its benefits. Albeit current limited efficacy and safety information, it is considered significant to provide Akuugo to the clinical setting as one of treatment options for chronic motor paralysis associated with TBI.

If based on the currently available data, Akuugo should be approved for the following indication or performance as well as dosage and administration or method of use, with the following approval conditions that specify conditions and time limit in accordance with Article 23-26 of the Act on Securing Quality, Efficacy, and Safety of Pharmaceuticals, Medical Devices, etc.

Indication or Performance

Improvement of chronic motor paralysis associated with traumatic brain injury

Dosage and Administration or Method of Use

Usually in adults, the cell suspension containing 5×10^6 viable cells (300 μL) of human (allogeneic) bone marrow-derived mesenchymal stem cells is transplanted to the surrounding area of the damaged tissue via stereotactic brain surgery using a dedicated delivery device set. The cell suspension is injected through a single small opening on the skull that leads to 3 transplantation routes, reaching the area surrounding the injury. The dose of the cell suspension is 100 μL per route, 20 μL each of which is transplanted at 5 points 5 to 6 mm apart, from the deepest point. The infusion rate should be approximately 10 μL/min. The following steps should be taken prior to the transplantation:

- Before starting the surgical procedure, attach the guide & stop in the dedicated delivery device set and the stylet-equipped inserter to the invasive cranial fixation device for neurosurgery.
- Thaw the cell suspension for intracranial transplantation. After washing using the dedicated preparation solution, adjust the cell concentration using the dedicated solution so as to obtain cell suspension at the concentration of 1.67×10^6 cells/100 μL for transplantation. Cleanse the administration cannula-fixed microsyringe in the dedicated delivery device set using the dedicated preparation solution, and fill the microsyringe with the cell suspension.

Approval Conditions

1. In view of the limited manufacturing experience with the product, the applicant is required to promptly collect information about the product's quality according to the plan, evaluate quality comparability between the study product and the product, and report the results. Based on these results, the applicant should file necessary partial change application. The product must not be shipped before the approval of the partial change application.
2. The applicant is required to ensure that the product is used at medical institutions fully prepared for emergencies and by physicians with adequate knowledge and experience in the diagnosis and

treatment of traumatic brain injuries and stereotactic brain surgery techniques who are also fully knowledgeable about the clinical study results and adverse events, etc. associated with the product.

3. During the period after the conditional and time-limited approval until the re-application for marketing authorization, the applicant is required to conduct a post-marketing approval condition assessment covering all patients treated with the product.
4. During the period after the conditional and time-limited approval until the re-application for marketing authorization, the applicant is required to collect information on biological characteristics that reflect the mechanism of action of the product, and take necessary measures such as improving the quality control strategy.

Review Report (3)

June 5, 2024

Product Submitted for Approval

Brand Name	Akuugo Suspension for Intracranial Implantation
Non-proprietary Name	Vandefitemcel
Applicant	SanBio Company Limited
Date of Application	March 7, 2022

List of Abbreviations
See Appendix.

1. Content of the Review

The Committee on Regenerative Medicine Products and Biotechnology had discussions on Akuugo on March 25, 2024, and concluded that the comparability assessment between the study product and Akuugo would require additional data. It also concluded that when the said comparability is proven, Akuugo will have a certain level of efficacy in improving chronic motor paralysis associated with TBI, and that the product has acceptable safety in view of its benefits. Albeit current limited efficacy and safety information, it is considered significant to provide Akuugo to the clinical setting as one of treatment options to improve chronic motor paralysis associated with TBI.

Recently, the applicant submitted additional data on the quality of the product, and PMDA conducted a review.

1.1 Manufacturing method of the primary component (SB623)

In response to the PMDA's conclusion requiring (1) the collection of quality-related information based on additionally manufactured commercial process formulation of Akuugo's and (2) the comparability assessment of the study product with the commercial process formulation, the applicant submitted results from the following additional assessment and a plan for post-approval comparability assessment.

(a) Additional comparability assessment between Akuugo and the study product used in Study TBI-01

Of the comparability assessment plan outlined in Section "1.6 Manufacturing process of the primary component (SB623)" of the Review Report (2), test systems were established for [redacted], [redacted], and [redacted]. One batch of the commercial process formulation manufactured ([redacted]) yielded the following results. All results were similar to those of the study product used in Study TBI-01, demonstrating the equivalence of quality between the commercial process formulation and the study product.

1) [redacted]

For Akuugo activity-related [redacted] and [redacted]), [redacted], [redacted] and [redacted]), and [redacted] ([redacted], [redacted], and [redacted]), the [redacted] of Akuugo against [redacted] was calculated. The results of the commercial process formulation were compared with those of the study product ([redacted], [redacted], and [redacted]) as shown in Figure 1.

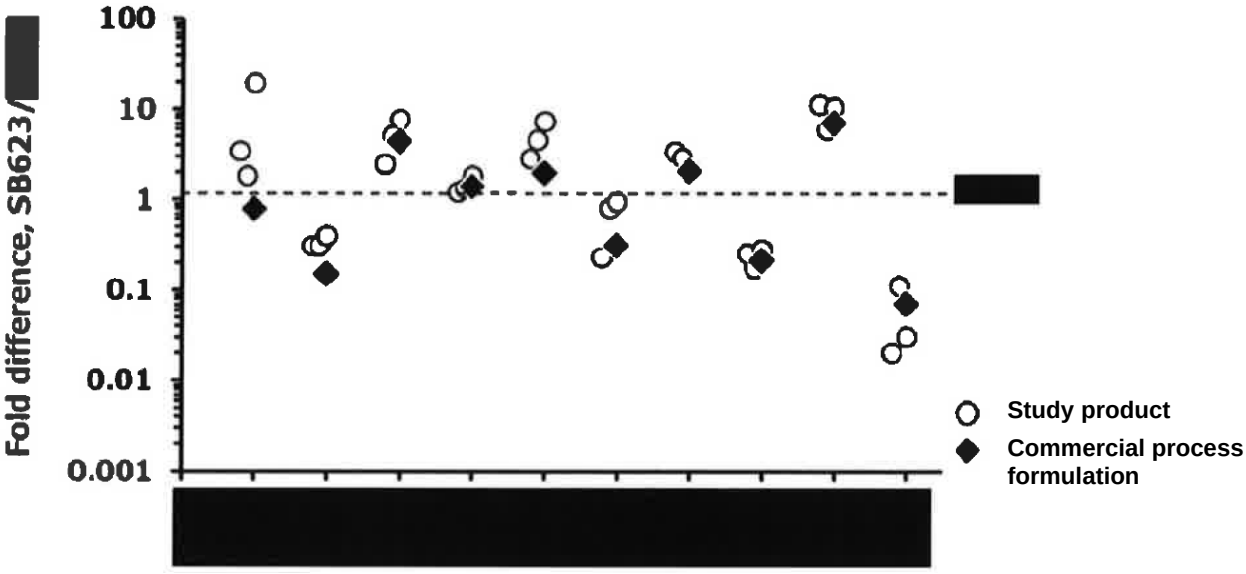

Figure 1. Results of [redacted]

2) [redacted]

To examine [redacted], cryopreserved SB623 was thawed for culture, and [redacted] was measured by [redacted]. Table 1 shows the results with the study product and the commercial process formulation.

Table 1. Results of [redacted]

	Batch No.	[redacted]
Study product	[redacted]	[redacted]
Commercial process formulation	[redacted]	[redacted]

3) Evaluation of [redacted]

Using [redacted], [redacted] SB623 and [redacted] were cultured [redacted], respectively, and [redacted] was evaluated by measuring [redacted] based on [redacted]. Figures 2 and 3 show the results of the study product and the commercial process formulation.

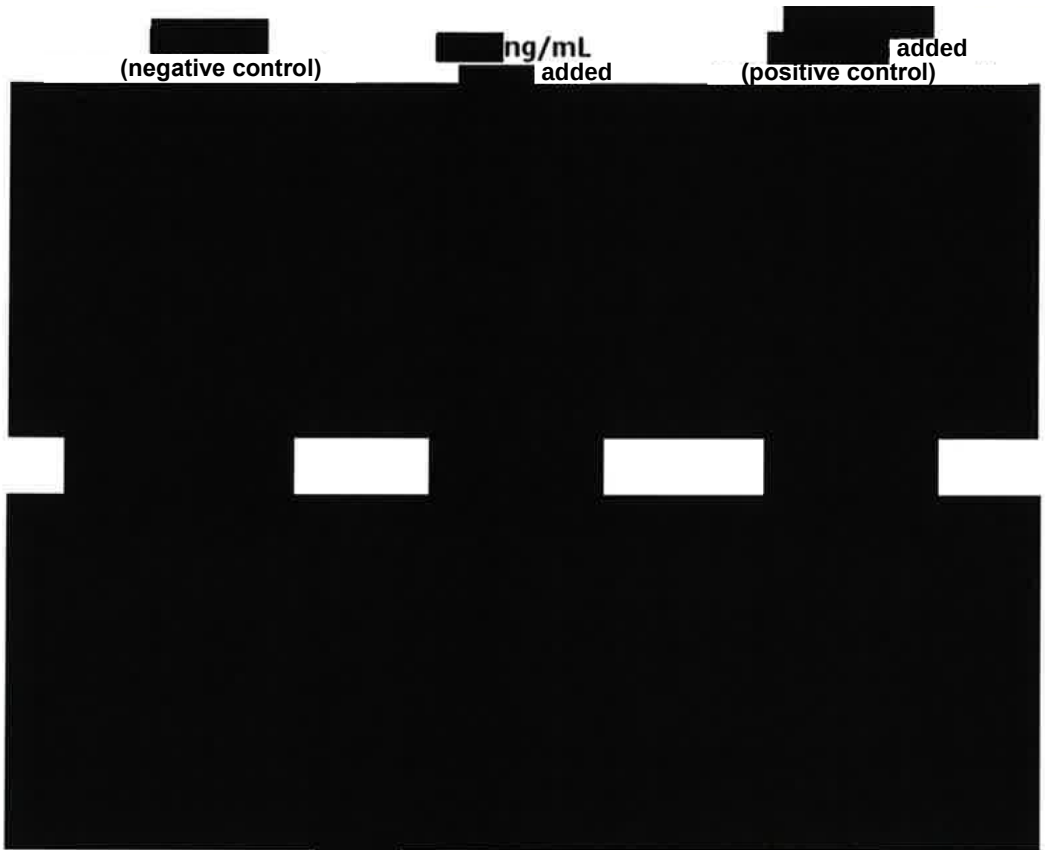

Figure 2. Microscopic photograph of

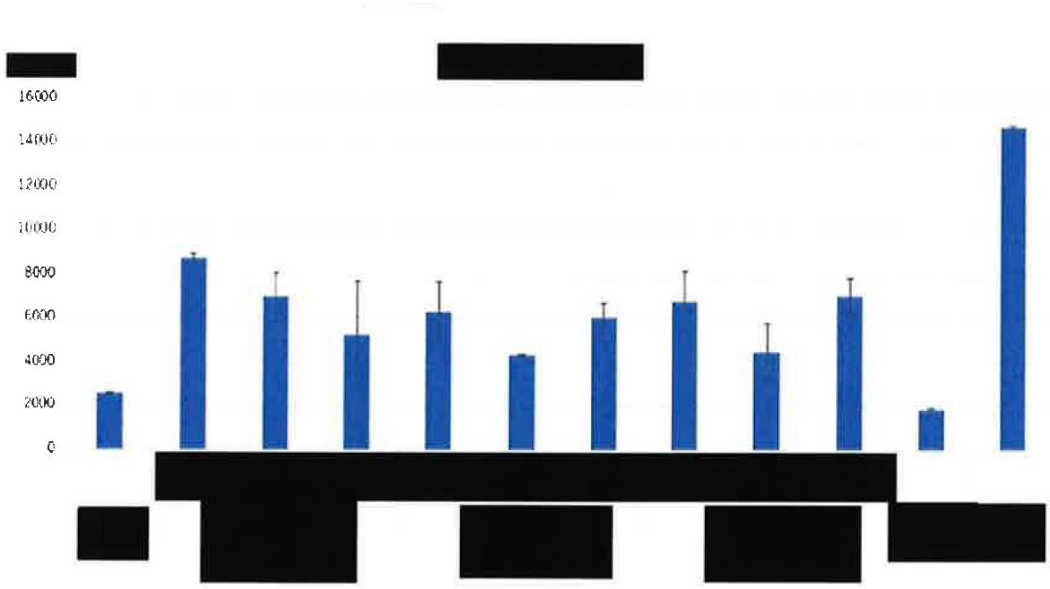

Figure 3.

(b) Plan for future comparability assessment between Akuugo and the study product

After the marketing approval for Akuugo, additional 2 batches of the commercial process formulation will be produced before the start of shipment. The 3 batches of the commercial process formulation are to be assessed for the comparability against the study product used in Study TBI-01 based on the test items listed in Table 2.

Table 2. Comparability test items to be evaluated after marketing authorization

Test items	Control level/specification limit/criteria
(a) Specification tests	
pN-2 plasmid copy number	[redacted] (copy per haploid genome)
Cell surface marker analysis [redacted]	≥ [redacted]%
[redacted]	≥ [redacted]%
[redacted]	≥ [redacted]%
[redacted]	≤ [redacted]%
[redacted]	≤ [redacted]%
[redacted]	≤ [redacted]%
Viability	≥ [redacted]%
Viable cell count	≥ [redacted] cells/vial
[redacted]	≥ [redacted]%
Intracellular FGF-2	≥ [redacted] molecules
Sterility	No observable growth
Bacterial endotoxin	≤ [redacted] EU/mL
Mycoplasma test	Not detected
(b) Characterization	
[redacted]	[redacted]
[redacted]	[redacted]
(c) In-process control tests	
[redacted]	
[redacted]	≥ [redacted]
[redacted]	
[redacted]	≥ [redacted]
[redacted]	
[redacted]	≥ [redacted]
[redacted]	
[redacted]	< [redacted]
[redacted]	≥ [redacted]%
[redacted]	≥ [redacted]
(d) Other test items	
[redacted]	
[redacted]	
[redacted]	
[redacted]	

PMDA's view:

The quality attributes of 1 batch of the commercial process formulation did not show any significant deviation from those observed in the study product, allowing for evaluation to a certain extent. Because of limited information obtained after the compilation of Review Report (2), the comparability between the study product in Study TBI-01 and the commercial process formulation has remains unconfirmed at this point. The applicant should promptly collect data on the quality of Akuugo through additional production of commercial process formulation and assess its comparability with the study product.

2. Overall Evaluation

As found in the above review, the additional production has not yielded sufficient data on the quality of Akuugo, and the comparability remains unconfirmed between the study product in Study TBI-01 and the commercial process formulation. As stated in the Review Report dated March 15, 2024, when the product is confirmed to be comparable with the study product in future evaluations involving 3 additional batches of the commercial process formulation and the study product in Study TBI-01, Akuugo will have a certain level of efficacy in improving chronic motor paralysis associated with TBI, and that the product has acceptable safety in view of its benefits. Albeit current limited efficacy and

safety information, it is considered significant to provide Akuugo to the clinical setting as one of treatment options for chronic motor paralysis associated with TBI.

If based on the currently available data, Akuugo should be approved after modifying the proposed indication or performance as well as dosage and administration or method of use as follows, on the premise of the provision of necessary cautions in the package insert and the dissemination of information on proper use in the post-marketing setting, and with the following approval conditions, which specify conditions and time limit in accordance with Article 23-26 of the Act on Securing Quality, Efficacy, and Safety of Pharmaceuticals, Medical Devices, etc. The time limit under the said article should be 7 years. The product should be designated as a specified regenerative medical product.

Indication or Performance

Improvement of chronic motor paralysis associated with traumatic brain injury

Dosage and Administration or Method of Use

Usually in adults, the cell suspension containing 5×10^6 viable cells (300 μL) of human (allogeneic) bone marrow-derived mesenchymal stem cells is transplanted to the surrounding area of the damaged tissue via stereotactic brain surgery using a dedicated delivery device set. The cell suspension is injected through a single small opening on the skull that leads to 3 transplantation routes, reaching the area surrounding the injury. The dose of the cell suspension is 100 μL per route, 20 μL each of which is transplanted at 5 points 5 to 6 mm apart, from the deepest point. The infusion rate should be approximately 10 μL/min. The following steps should be taken prior to the transplantation.

- Before starting the surgical procedure, attach the guide & stop in the dedicated delivery device set and the stylet-equipped inserter to the invasive cranial fixation device for neurosurgery.
- Thaw the cell suspension for intracranial transplantation. After washing using the dedicated preparation solution, adjust the cell concentration using the dedicated solution so as to obtain cell suspension at the concentration of 1.67×10^6 cells/100 μL for transplantation. Cleanse the administration cannula-fixed microsyringe in the dedicated delivery device set using the dedicated preparation solution, and fill the microsyringe with the cell suspension.

Approval Conditions

1. In view of the limited manufacturing experience with the product, the applicant is required to promptly collect information about the product's quality according to the plan, evaluate quality comparability between the product and the study product, and report the results. Based on these results, the applicant should file necessary partial change application. The product must not be shipped before the approval of the partial change application.
2. The applicant is required to ensure that the product is used at medical institutions fully prepared for emergencies and by physicians with adequate knowledge and experience in the diagnosis and treatment of traumatic brain injuries and stereotactic brain surgery techniques who are also fully knowledgeable about the clinical study results and adverse events, etc. associated with the product.
3. During the period after the conditional and time-limited approval until the re-application for marketing authorization, the applicant is required to conduct a post-marketing approval condition assessment covering all patients treated with the product.

4. During the period after the conditional and time-limited approval until the re-application for marketing authorization, the applicant is required to collect information on biological characteristics that reflect the mechanism of action of the product, and take necessary measures such as improving the quality control strategy.

Appendix

List of Abbreviations

Akuugo	Akuugo Suspension for Intracranial Implantation
Application	Application for marketing approval
[redacted]	[redacted]
CD	cluster of differentiation
[redacted]	[redacted]
FGF	fibroblast growth factor
[redacted]	[redacted]
MSC	mesenchymal stem cell
[redacted]	[redacted]
PMDA	Pharmaceuticals and Medical Devices Agency
TBI	traumatic brain injury
[redacted]	[redacted]