February 26, 2020
Medical Device Evaluation Division
Pharmaceutical Safety and Environmental Health Bureau
Ministry of Health, Labour and Welfare

Report on the Deliberation Results

Classification	Gene therapy products, 2. Viral vector products
Non-proprietary name	Onasemnogene abeparvovec
Brand name	ZOLGENSMA Intravenous Infusion
Applicant	Novartis Pharma K.K.
Date of application	November 1, 2018 (marketing application)

Results of Deliberation

In its meeting held on February 26, 2020, the Committee on Regenerative Medicine Products and Biotechnology reached the following conclusion, and decided that this conclusion should be presented to the Pharmaceutical Affairs Department of the Pharmaceutical Affairs and Food Sanitation Council.

The product may be approved. The conditional and time-limited approval scheme is not applicable to this application. The re-examination period should be 10 years.

Approval Conditions

1. Because the number of Japanese patients participating in clinical trials is very limited, the applicant is required to conduct a post-marketing use-results survey covering all patients treated with the product, until data from a certain number of patients are collected, in order to identify the characteristics of patients using the product and collect data on the safety and efficacy of the product as early as possible, thereby taking necessary measures to ensure the proper use of the product. The applicant is also required to report the results of analysis of the long-term data from post-marketing surveillance etc. to the Ministry of Health, Labour and Welfare and the Pharmaceuticals and Medical Devices Agency, and take appropriate measures as needed.
2. The applicant is required to disseminate the proper use guide developed in cooperation with the relevant academic societies and take other necessary measures, so as to ensure that physicians with adequate knowledge of and experience in the treatment of spinal muscular atrophy fully understand the results from clinical trials of the product, adverse events reported, and other data and then use the product in accordance with the “INDICATION OR PERFORMANCE” and “DOSAGE AND ADMINISTRATION OR METHOD OF USE” statements at medical institutions prepared to treat spinal muscular atrophy.

3. The applicant is required to take necessary measures, for example, ensuring that relevant physicians are well informed of the Regulations on Type-1 Use approved under the Act on the Conservation and Sustainable Use of Biological Diversity through Regulations on the Use of Living Modified Organisms (Act No. 97 of 2003), so that the product is used in compliance with the approved Regulations on Type-1 Use.

April 8, 2020
Medical Device Evaluation Division
Pharmaceutical Safety and Environmental Health Bureau
Ministry of Health, Labour and Welfare

Report on the Deliberation Results (Supplement)

Classification	Gene therapy products, 2. Viral vector products
Non-proprietary name	Onasemnogene abeparvovec
Brand name	ZOLGENSMA Intravenous Infusion
Applicant	Novartis Pharma K.K.
Date of application	November 1, 2018 (marketing application)

In its meeting held on March 19, 2020, the Pharmaceutical Affairs Department of the Pharmaceutical Affairs and Food Sanitation Council advised as follows: "The Ministry of Health, Labour and Welfare (MHLW) is accountable to patients and the public for its decision on this application. The regulatory authority should therefore disclose in some way the basis for concluding that there are no quality, efficacy, or safety problems, in spite of deficiencies in the submitted data."

In light of this advice, the data manipulation issue involving the *in vivo* relative potency assay and the results of its assessment (only the outline of the issue is given in the review report) are documented in details.

1. An overview of the data manipulation case and the firm's actions

(1) How the problem came to light

- In March 2019, a whistleblower from AveXis alleged that the company's personnel had manipulated data from an assay at the San Diego facility. Novartis global headquarters (hereinafter referred to as the "Novartis headquarters") launched an investigation.
- At the end of June in 2019, the Novartis headquarters reported the results of its internal investigation to the United States Food and Drug Administration (US FDA). At the same time, Novartis Pharma K.K. (the applicant of the present application) also learned of this issue. In July 2019, the applicant reported the developments of the issue, to the Pharmaceuticals and Medical Devices Agency (PMDA).

(2) An outline of internal investigation

- Upon learning of the allegations from the whistleblower, the Novartis headquarters conducted an internal investigation[1] in cooperation with an external law firm. There was evidence of inappropriate manipulation[2] in the raw data from the *in vivo* relative potency assay (hereinafter referred to as "Assay A") among 3 potency assays as quality tests of the clinical product.
- Two senior executives responsible for the data manipulation were interviewed during the internal investigation, but could not offer a credible explanation for the issue. Based on the remaining records etc., the Novartis headquarters inferred that the senior executives had an incentive to make the data look better, for example, by reducing test variability. These two senior executives were later terminated.

(3) Impact assessment and actions implemented by the Novartis headquarters based on the results of internal investigation

- All lots of the clinical product released based on the results of Assay A were tested using other potency assays for reassessment of quality at release. The assay results for all clinical product lots were within the commercial product specification. The applicant concluded from the results that they had adequate quality.

2. The regulators' conclusion on the quality, efficacy, and safety of Zolgensma, with consideration of the data manipulation issue

Based on the following points, MHLW and PMDA concluded that there are no problems with the quality, efficacy, or safety of Zolgensma, in spite of the data manipulation.

(1) Assurance of quality of the clinical product, and confirmation of comparability of quality attributes between clinical product lots and commercial products

- Because the data manipulation compromised the integrity of the data from Assay A, PMDA decided to remove the results of Assay A from evaluation data.
- All lots of the clinical product released based on the results of Assay A were tested using other potency assays included in the commercial product specification[3] for reassessment of quality. Since the assay results were within the commercial product specification, it was concluded that their quality had been assured. Other potency assays included an *in vitro* cell-based potency assay, which was less variable compared with Assay A using mice, and considered to enable more appropriate evaluation.

---

[1] The primary components of internal investigation
· Interviews with key AveXis employees
· Analysis of the underlying data to determine if there were unexplained discrepancies in the data referenced in the allegations
· Review of relevant documents and electronic records
· An impact assessment to determine the impact of manipulated data in results from Assay A on lot release decisions, or clinical data that may have been generated through use of those lots, as well as the impact of data manipulation on subject safety and product efficacy or quality

[2] An overview of data manipulation
· A lack of real-time recording/maintenance of raw data
· Spreadsheets created to record mouse weights etc. were found to exist in different versions, and there was evidence of inappropriate revisions to data, etc.
· There was evidence of replacement of the original source documents (some data sheets to be used for data analysis). There was evidence that some data had been altered by a person other than the person who prepared the data sheets.

[3] Other potency assays in the commercial product specification do not include Assay A.

- Even when the results of Assay A were removed, the potency results obtained by reassessment were considered to demonstrate the comparability of quality attributes between clinical product lots and commercial products.
- After the revelation of the data manipulation issue, PMDA conducted an additional document-based compliance inspection of AveXis's San Diego testing site. The inspection cited an additional observation of failure to appropriately retain some source documents. Despite this observation, however, PMDA confirmed that there were no major problems with the integrity of the data from quality tests other than Assay A.

(2) Assessment of impact on efficacy and safety

- Assay A in which the data manipulation occurred is an animal potency assay, which is a quality test to qualitatively assess the potency of the clinical product. In this data manipulation case, the clinical trial data were not manipulated, but data were manipulated in the quality tests of the clinical product. This did not directly affect the assessment of clinical efficacy and safety of the product. No efficacy or safety problems have been reported in clinical trials using the clinical product lots released based on the results of Assay A.

Reference information: FDA's actions

- In July 2019, FDA initiated an inspection of AveXis's San Diego site.
- After its review of AveXis's responses to the inspectional observations and internal discussions, FDA completed its inspection review in February 2020, and finally classified the inspection as "Voluntary Action Indicated." FDA decided to take no further enforcement action.

3. Others

In its meeting held on February 26, 2020, the Committee on Regenerative Medicine Products and Biotechnology advised that handling of Japanese patients participating in a clinical trial, in response to the data manipulation, should be checked. The results of inquiries to the applicant are shown below.

- Three Japanese patients were enrolled in the clinical trial, of whom 2 received the product from the clinical product lot released based on the manipulated data.
- After the revelation of the data manipulation, the investigator explained to the 3 patients and their families about this issue, and they understood the situation and agreed to continue participation in the clinical trial.
- The details of this issue were reported to the Institutional Review Board (IRB). The IRB did not recommend termination of the clinical trial, etc. The trial was continued.

Review Report

February 7, 2020
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	ZOLGENSMA Intravenous Infusion
Classification	Gene therapy products, 2. Viral vector products
Non-proprietary name	Onasemnogene abeparvovec
Applicant	Novartis Pharma K.K.
Date of application	November 1, 2018

Shape, Structure, Active Ingredient, Quantities, or Definition

ZOLGENSMA is a non-replicating, recombinant adeno-associated virus (AAV) vector-based gene therapy. It is composed of a non-replicating, recombinant adeno-associated virus serotype 9 (AAV9) capsid shell containing the human survival motor neuron (SMN) gene under the control of a cytomegalovirus enhancer and a chicken-β-actin-hybrid promoter. One of the two AAV inverted terminal repeats (ITRs) from the AAV serotype 2 (AAV2) DNA has been modified to promote intramolecular annealing of the transgene, thus forming a double-stranded transgene ready for transcription.

Application Classification (1-1) New regenerative medical products

Items Warranting Special Mention

Orphan regenerative medical product (Orphan Regenerative Medical Product Designation No. 6 of 2018 [*30 sai*]; PSEHB/MDED Notification No. 1001-1 dated October 1, 2018, issued 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. 3 of 2017 [*29 sai*]; PSEHB/MDED Notification No. 0327-15 dated March 27, 2018, issued by the Medical Device Evaluation Division, Pharmaceutical Safety and Environmental Health Bureau, Ministry of Health, Labour and Welfare)

Reviewing Office Office of Cellular and Tissue-based Products

Results of Review

On the basis of the data submitted, PMDA has concluded that the product has efficacy in the treatment of patients with spinal muscular atrophy (including those with genetically diagnosed pre-symptomatic SMA), and that the product has acceptable safety in view of its benefits (see Attachment).

As a result of its review, PMDA has concluded that the product may be approved for the indication or performance and dosage and administration or method of use shown below, with the following conditions.

Indication or Performance

Treatment of patients with spinal muscular atrophy (including those with genetically diagnosed pre-symptomatic SMA), who had tested negative for anti-AAV9 antibodies.

Dosage and Administration or Method of Use

The usual dose of ZOLGENSMA for patients weighing ≥2.6 kg (<2 years of age) is 1.1×10^{14} vector genomes (vg)/kg. ZOLGENSMA should be administered as a single, one-time intravenous infusion over 60 minutes. Do not re-administer ZOLGENSMA.

The dose volume is determined based on patient body weight as per the table below.

Patient Weight Range (kg)	Dose Volume (mL)[Note)]
2.6 – 3.0	16.5
3.1 – 3.5	19.3
3.6 – 4.0	22.0
4.1 – 4.5	24.8
4.6 – 5.0	27.5
5.1 – 5.5	30.3
5.6 – 6.0	33.0
6.1 – 6.5	35.8
6.6 – 7.0	38.5
7.1 – 7.5	41.3
7.6 – 8.0	44.0
8.1 – 8.5	46.8
8.6 – 9.0	49.5
9.1 – 9.5	52.3
9.6 – 10.0	55.0
10.1 – 10.5	57.8
10.6 – 11.0	60.5
11.1 – 11.5	63.3
11.6 – 12.0	66.0
12.1 – 12.5	68.8
12.6 – 13.0	71.5
13.1 – 13.5	74.3

Note) Dose volume is calculated using the upper limit of the patient weight range.

The dose volume for patients <2 years of age weighing ≥13.6 kg is determined based on patient body weight.

Approval Conditions

1. Because the number of Japanese patients participating in clinical trials is very limited, the applicant is required to conduct a post-marketing use-results survey covering all patients treated with the product, until data from a certain number of patients are collected, in order to identify the characteristics of patients using the product and collect data on the safety and efficacy of the product as early as possible, thereby taking necessary measures to ensure the proper use of the product. The applicant is also required to report the results of analysis of the long-term data from post-marketing surveillance etc. to the Ministry of Health, Labour and Welfare and the Pharmaceuticals and Medical Devices Agency, and take appropriate measures as needed.
2. The applicant is required to disseminate the proper use guide developed in cooperation with the relevant academic societies and take other necessary measures, so as to ensure that physicians with adequate knowledge of and experience in the treatment of spinal muscular atrophy fully understand the results from clinical trials of the product, adverse events reported, and other data and then use the product in accordance with the “INDICATION OR PERFORMANCE” and “DOSAGE AND ADMINISTRATION OR METHOD OF USE” statements at medical institutions prepared to treat spinal muscular atrophy.
3. The applicant is required to take necessary measures, for example, ensuring that relevant physicians are fully aware of the Regulations on Type-1 Use approved under the Act on the Conservation and Sustainable Use of Biological Diversity through Regulations on the Use of Living Modified Organisms (Act No. 97 of 2003), so that the product is used in compliance with the approved Regulations on Type-1 Use.

Attachment

Review Report (1)

December 5, 2019

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	ZOLGENSMA Intravenous infusion
Classification	Gene therapy products, 2. Viral vector products
Non-proprietary name	Onasemnogene abeparvovec
Applicant	Novartis Pharma K.K.
Date of application	November 1, 2018

Shape, Structure, Active Ingredient, Quantities, or Definition

ZOLGENSMA is a non-replicating, recombinant adeno-associated virus (AVV) vector-based gene therapy. It is composed of a non-replicating, recombinant adeno-associated virus serotype 9 (AAV9) capsid shell containing the human survival motor neuron (SMN) gene under the control of the cytomegalovirus enhancer/chicken-β-actin-hybrid promoter. One of the two AAV inverted terminal repeats (ITRs) from the AAV serotype 2 (AAV2) DNA has been modified to promote intramolecular annealing of the transgene, thus forming a double-stranded transgene ready for transcription.

Proposed Indication or Performance

Infantile spinal muscular atrophy

Proposed Dosage and Administration or Method of Use

The usual dose of ZOLGENSMA for infants weighing between 2.6 and 8.5 kg is 1.1×10^{14} vector genomes (vg)/kg. ZOLGENSMA should be administered as a single, one-time intravenous infusion. The dose volume is determined based on patient body weight as per the table below.

Patient Weight Range (kg)	Dose Volume (mL)*
2.6 – 3.0	16.5
3.1 – 3.5	19.3
3.6 – 4.0	22.0
4.1 – 4.5	24.8
4.6 – 5.0	27.5
5.1 – 5.5	30.3
5.6 – 6.0	33.0
6.1 – 6.5	35.8
6.6 – 7.0	38.5
7.1 – 7.5	41.3
7.6 – 8.0	44.0
8.1 – 8.5	46.8

*: Dose volume is calculated using the upper limit of the patient weight range.

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

Zolgensma is a regenerative medical product, which is a non-replicating, recombinant AAV containing the human *SMN* gene, i.e., the gene responsible for spinal muscular atrophy (SMA). It consists of an AAV9 capsid packaging an expression construct with the modified inverted terminal repeat (ITR), the cytomegalovirus (CMV) enhancer/chicken-β-actin-hybrid promoter, the cDNA of the human *SMN* gene, and the ITR. When intravenously administered Zolgensma infects motor neurons or muscle cells of the patient, the expression construct packaged in Zolgensma persists as an episome in the nucleus of transduced cells, leading to long-term stable expression of the human *SMN* gene. Zolgensma is expected to improve the survival of SMA patients by replacing the missing or defective *SMN1* gene (the gene responsible for SMA) with a functional copy that makes SMN protein, thereby improving the survival of muscle cells and the neuromuscular function.

Zolgensma received an orphan regenerative medical product designation with the intended indication or performance of "spinal muscular atrophy (SMA)" on October 1, 2018 (Orphan Regenerative Medical Product Designation No. 6 of 2018 [*30 sai*]).

Zolgensma also received a SAKIGAKE designation as a regenerative medical product with the intended indication or performance of "spinal muscular atrophy" on March 27, 2018 (SAKIGAKE Designation No. 3 of 2017 [*29 sai*]).

1.2 Development history etc.

SMA is a lower motor neuron disease characterized by loss and degeneration of the anterior horn cells of the spinal cord, which lead to progressive proximal muscle weakness of the trunk and limbs and muscle atrophy. It is a genetic rare disease caused by bi-allelic mutations in the *SMN1* gene on chromosome 5q13 (a lack of a functional *SMN1* gene), resulting in diminished levels of SMN protein required mainly for the survival of spinal motor neurons. The SMN protein is ubiquitously expressed in all cells, and involved in RNA metabolism and splicing. Especially, protein expressed in spinal motor neurons is considered to maintain their survival and function and prevent neurodegeneration, etc. Although the *SMN2* gene serves as a back-up to the *SMN1* gene, a cytosine-to-thymine substitution occurs at position 840 in the *SMN2* gene compared with the sequence of the *SMN1* gene, resulting in an alternative splicing pattern. As a result, approximately 90% of the transcripts produced from the *SMN2* gene are missing exon 7 and translated into truncated SMN proteins, which are nonfunctional and rapidly degraded. Thus, only 10% to 15% of the SMN protein generated from *SMN2* is full-length, functional SMN protein.

Diagnosis of SMA is made by genetic testing and clinical findings. Genetic testing is used to detect deletion or mutation in the *SMN1* gene and the number of copies of the back-up *SMN2* gene (increased *SMN2* copy number is correlated with milder phenotypes).

SMA is categorized into 5 types based on age of onset and maximum motor function achieved: Type 0 (prenatal

onset SMA) and Types I, II, III, and IV (the disease with postnatal onset) (Table 1).

Table 1. SMA Classification

Type	Age at Symptom Onset	Maximum Motor Function	Life Expectancy	*SMN2* copy number
0	Fetal	Nil	Days - Weeks	1
I	0-6 months	Never sits	<2 years	1, 2*, 3
II	6-18 months	Never walks	20-40 years	2, 3*, 4
III	1.5-10 years	Walks, regression	Normal	3, 4*, 5
IV	≥35 years	Slow decline	Normal	4, 5

*: Predominant SMN2 copy number that defines the SMA Type

The clinical features and course of each type of the disease are described below.

- SMA Type I. This disease type is the most severe and common form of the disease, and ≥90% of patients die or require permanent ventilation by 20 months of age. Patients with SMA Type I are bedridden and unable to move about throughout their lifetime due to debilitation, hypotension, and growth failure.
- SMA Type II. The survival rate for patients with SMA Type II has been reported to be 68.5% at 25 years. However, most of the patients are unable to stand or walk without support, thus having difficulty in social life.
- SMA Type III. This disease type is further divided into 2 subtypes: IIIA (onset before the age of 3 years) and IIIB (onset at or after the age of 3 years). Both subtypes of patients are initially able to walk, but lose this ability as the disease progresses.
- SMA Type IV. Although patients with SMA Type IV have a normal life expectancy, clinical symptoms associated with disease progression persist throughout their lifetime. Patients have difficulty in social life once ambulation is lost.

Outside of Japan, AveXis initiated a phase I study of Zolgensma in patients with SMA Type I (Study CL-101) in May 2014, followed by a long-term follow-up study in patients participating in Study CL-101 (Study LT-001). AveXis also initiated a foreign phase I study in patients with SMA Type II or III (Study CL-102) in December 2017, a foreign phase III study in patients with SMA Type I or patients with a genetic diagnosis of SMA Type I (Study CL-303) in October 2017, and a global phase III study in patients with a genetic diagnosis of SMA Type I, II, or III (Study CL-304) in April 2018.

In the US, the biologics license application for Zolgensma was filed based mainly on the results of Study CL-101 in September 2018. Zolgensma was approved for the following indications and usage in May 2019. FDA granted this application Fast Track and Breakthrough Therapy designations in September 2013 and July 2016, respectively.

ZOLGENSMA (onasemnogene abeparvovec-xioi) is an adeno-associated virus vector-based gene therapy indicated for the treatment of pediatric patients less than 2 years of age with spinal muscular atrophy (SMA) with bi-allelic mutations in the *survival motor neuron 1* (*SMN1*) gene.

Limitation of Use

- The safety and effectiveness of repeat administration of ZOLGENSMA have not been evaluated.

- The use of ZOLGENSMA in patients with advanced SMA (e.g., complete paralysis of limbs, permanent ventilator-dependence) has not been evaluated.

A marketing authorization application for Zolgensma in the EU was filed based mainly on the results of Study CL-101 in October 2018 and is under review as of December 2019. In the EU, Zolgensma received PRIME (PRIority MEdicines) designation in January 2017. Zolgensma was granted an accelerated assessment status in July 2018 but lost it in July 2019.

In Japan, AveXis started patient enrollment in Study CL-304 in [redacted] 20[redacted], and the applicant has submitted a marketing application for Zolgensma based mainly on the results from Study CL-101.

Since Zolgensma received a SAKIGAKE designation as a regenerative medical product, PMDA took expedited action to shorten the review period. However, the review schedule was substantially delayed by the applicant's responses, which are described below.

(1) As a rule, SAKIGAKE-designated products undergo SAKIGAKE comprehensive evaluation consultation (quality; non-clinical; clinical; compliance; Good Gene, Cellular, and Tissue-based Products Manufacturing Practice [GCTP]) in order to shorten the time from submission to approval. The applicant applied for this evaluation consultation for Zolgensma. However, before the consultation was initiated, the applicant submitted a marketing application for Zolgensma. This precluded PMDA from identifying and sorting out key issues on the quality, efficacy, and safety of Zolgensma prior to regulatory submission.

(2) The information and data included in the application documents submitted were limited and difficult to interpret. PMDA instructed the applicant to revise the following documents immediately after starting the review of the application, but the applicant submitted all of the revised versions of the documents on December 25, 2018 (i.e., about 2 months after the date of regulatory submission).
   - Package insert (draft)
   - Indication or Performance, and Dosage and Administration or Method of Use with justifications
   - Precautions with justifications
   - Shape, Structure, Active Ingredient, Quantities, or Definition
   - Manufacturing process
   - Specifications
   - Clinical Overview
   - Clinical efficacy and safety
   - Summary of individual clinical trials
   - Post-marketing use-results survey plan and post-marketing clinical study plan
   - Information materials to be distributed to healthcare professionals and to the patient's family and relatives

(3) There were also many flaws in the responses to the inquiries from PMDA (especially, the responses to the inquiries about quality), resulting in repeated revision of the responses. This took a considerable amount of time to handle.

(4) PMDA sent inquiries about the key issues of the present application on the 22nd day after regulatory submission (November 22, 2018), but the applicant submitted the final responses about 10 months later (September 20, 2019).

(5) PMDA requested the inclusion of the *in vivo* potency assay in the specification in Japan since the assay had been included in the US product specification. However, it took several months for the applicant to do so.

(6) In July 2019, it was revealed that the data from an assay performed for release of the clinical product lots used in clinical studies (Studies CL-102, CL-302, CL-303, and CL-304) had been manipulated intentionally. At this time point, therefore, PMDA had to undertake an additional thorough review of quality control data and the responses to the inquiries for impact assessment. Amendments to the submitted data and the responses to the inquiries after the thorough review were completed on September 30, 2019. In the course of an additional compliance inspection associated with the data manipulation, further observations requiring amendments were found, and amendments to the data affecting the regulatory review were completed on November 22, 2019.

The above case was extremely unusual. PMDA considers that such numerous and extensive problems were not merely the matter of the adequacy of data, but were also attributed to the applicant's very poor awareness of the important matters to assure the quality, safety, and efficacy of the product for use in patients in Japan.

2. Manufacturing Process and Specifications and Outline of the Review Conducted by PMDA

Zolgensma is a recombinant AAV containing the human *SMN* gene. It has AAV9 capsid proteins and the modified ITR of AAV2. The expression construct delivered by Zolgensma contains the human *SMN* cDNA expression cassette flanked by AAV2 ITR sequences. The expression cassette comprises the CMV enhancer/chicken-β-actin hybrid promoter, simian virus 40 (SV40) intron, the human SMN cDNA, and the bovine growth hormone (BGH) polyadenylation (poly A) termination signal.

Zolgensma is unable to replicate due to the removal of the *rep* and *cap* genes from the wild-type AAV genome. The Rep2/Cap9 packaging component; the E2a, E4, and VA packaging component; and the viral vector component containing the SMN expression cassette, which are required for the production of Zolgensma, are divided into 3 plasmid vectors. Zolgensma has been designed not to generate a replication competent virus by homologous recombination.

2.1 Drug substance

2.1.1 Plasmid vectors

2.1.1.1 Generation and control of the cell substrate for the production of plasmid vectors

Three expression constructs were generated by inserting the SMN cDNA, the Rep2/Cap9 genes, and the E2a, E4, and VA1 RNA genes into respective cloning vectors. These expression constructs were transfected into *Escherichia coli* ([redacted] strain). The most suitable clone was selected and used to prepare a master cell bank (MCB) and a working cell bank (WCB) for the production of each plasmid vector.

The MCB for the production of the plasmid vector was characterized and controlled by host cell identity, host genotype ([redacted]), [redacted], viable cell count, the genetic stability of the plasmid, colony morphology, Gram staining, purity, restriction digest map, and sterility. All test results met the acceptance criteria. To control the MCB for the plasmid that encodes [redacted], plasmid sequencing was added.

The WCB for the production of the plasmid vector was characterized and controlled by host cell identity, [redacted], viable cell count, colony morphology, Gram staining, purity, restriction digest map, sterility, and plasmid sequence. All test results met the acceptance criteria.

2.1.1.2 Control of plasmid vectors

The controls of the plasmid vector consist of [redacted], endotoxin, and bioburden.

2.1.2 Generation and control of the cell substrate for the production of drug substance

Human embryonic kidney 293 (HEK293) cells are used to produce the drug substance. HEK293 cells (Catalog [redacted] Lot [redacted]) supplied from the American Type Culture Collection (ATCC) were used to prepare a MCB and a WCB for the production of the drug substance.

The MCB and WCB for the production of the drug substance were characterized and subjected to purity tests in accordance with the ICH Q5A (R1), Q5B, and Q5D guidelines. Tests for adventitious agents performed are shown in Table 2. The test results demonstrated genetic stability during production. No viral or non-viral adventitious agents were detected in any of the tests conducted.

The MCB and WCB for the production of the drug substance are stored at ≤−140°C. There is no plan for generating a new MCB, but a new WCB will be generated as necessary.

Table 2. Tests for adventitious agents for MCB and WCB for production of drug substance

In vivo virus tests (suckling and adult mice, guinea pigs, embryonated eggs)
In vitro virus tests (MRC-5 cells, Vero cells, HeLa cells)
In vitro tests for bovine viruses (9CFR; BVDV, PI3, BTV, BAV, IBRV, BPV, RSV, Reo-3, and RABV)
In vitro tests for porcine viruses (9CFR; PAV, PPV, RABV, Reo-3, TGEV, PHEV, and BVDV)
Electron microscopy
Reverse transcriptase activity assay
Test for adenovirus
Test for AAV (serotypes 1-13)
Tests for human viruses (HSV-1/2, PVB19, EBV, SV40, CMV, HHV-6, HHV-7, HHV-8, HBV, HIV-1, HIV-2, HTLV-1, HTLV-2, HCV, and HAV)
Sterility test
Test for mycoplasma

2.1.3 Manufacturing process

The manufacturing process for the drug substance consists of thawing of the WCB for the production of the drug substance, cell expansion, plasmid transfection, [redacted], [redacted], harvest, [redacted], [redacted], [redacted], [redacted], [redacted], [redacted], [redacted], [redacted], [redacted], filling, storage, and testing.

[redacted] have been defined as critical steps.

The commercial-scale drug substance manufacturing process has undergone validation.

2.1.4 Safety evaluation of adventitious agents

Raw materials of biological origin other than HEK293 cells used in the drug substance manufacturing process are shown in Table 3.

Bovine serum is derived from healthy bovine blood sourced from New Zealand, which is γ-irradiated to inactivate pathogenic agents and tested for bovine viruses in accordance with 9CFR.

Transferrin is derived from human plasma collected from donors who are qualified based on interview. The donors have been checked by serological testing (HBs antigen, anti-HCV antibody, anti-HIV-1 antibody, anti-HIV-2 antibody, and syphilis) and nucleic acid amplification testing of plasma or pooled plasma (hepatitis A virus [HAV], hepatitis B virus [HBV], hepatitis C virus [HCV], human immunodeficiency virus-1 [HIV-1], and parvovirus B19 [PVB19]), with negative results. In its manufacturing process, cold ethanol fractionation and heat treatment are used to inactivate pathogenic agents.

Trypsin is derived from healthy porcine pancreas sourced from the US, which is treated with low pH to inactivate pathogenic agents and tested for porcine viruses and porcine circovirus in accordance with 9CFR.

Fetal bovine serum (FBS) is derived from healthy bovine blood sourced from New Zealand or Australia, which is γ-irradiated to inactivate pathogenic agents and tested for bovine viruses in accordance with 9CFR.

Casamino acids are derived from healthy bovine milk sourced from Australia or New Zealand, which are treated with heat to inactivate pathogenic agents.

Table 3. Raw materials of biological origin etc. other than HEK293 cells

Raw material	Animal species	Specific part of animal used	Process
Bovine serum	Bovine	Blood	[redacted]
Transferrin	Human	Blood	[redacted]
Trypsin	Porcine	Pancreas	[redacted]
FBS	Bovine	Blood	[redacted]
Casamino acids	Bovine	Milk	[redacted]

Each intermediate before and after [redacted] step is tested for adventitious agents as shown in Table 4.

Table 4. Controls for adventitious agents in the manufacturing process

Test sample	Control item
[redacted] culture medium	*In vitro* virus tests (MRC-5 cells, Vero cells, HEK293 cells)
[redacted] culture medium	Sterility test, Test for mycoplasma, *In vitro* virus tests (MRC-5 cells, Vero cells, HEK293 cells)
Intermediate after the end of [redacted] step	Bioburden

Viral clearance studies of the purification process were performed with model viruses. The results demonstrated the robustness of the purification process (Table 5).

Table 5. Results of viral clearance studies

Process step	Virus reduction factor (log_{10})			
	XMuLV	PRV	HAV	MVM
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
Overall reduction factor	12.61	15.31	1.88	1.48

2.1.5 Manufacturing process development

The following are major changes made to the drug substance manufacturing process during development (Process A and Process B [the proposed commercial process]). The finished product produced from the drug substance manufactured by Process A was used in [redacted] study, and the finished product produced from the drug substance manufactured by Process B (the proposed commercial process) was used in non-clinical studies and [redacted] studies.

- Process A → Process B (the proposed commercial process): changes in the process parameters in [redacted], [redacted], [redacted], and [redacted] steps, changes in [redacted], changes in [redacted] used in [redacted] step and [redacted] used in [redacted] step, [redacted], [redacted], [redacted], addition of [redacted] and [redacted] after [redacted], change of [redacted] from [redacted] to [redacted] in [redacted], and deletion of [redacted]

For these process changes, comparability of quality attributes between pre-change and post-change finished products has been demonstrated. Since the change from Process A to Process B (the proposed commercial

process) for the drug substance and the change from Process C to Process D for the finished product occurred at the same time, the comparability study was performed with the pre-change and post-change finished products [see Section 2.2.3].

A quality by design (QbD) approach was used to develop the manufacturing process.

2.1.6 Characterization

2.1.6.1 Structure and properties

Table 6 shows characterization tests performed.

Table 6. Characterization attributes

Characterization of [redacted]	titer (genomic copy number), [redacted], [redacted], analysis of viral vector genome, plasmid DNA, host cell DNA, *in vitro* SMN protein expression in [redacted]
Characterization of finished product	viral vector genome sequence, titer (genomic copy number), [redacted], osmolarity, pH, [redacted], rcAAV, [redacted], plasmid DNA, host cell DNA, HCP, [redacted], microbial limits, endotoxin, sterility, infectious titer, *in vitro* SMN protein expression in [redacted], *in vivo* potency (prolongation of survival of SMNΔ7 mice)

2.1.6.2 Product-related substances/Product-related impurities

Based on the results of characterization tests presented in Section 2.1.6.1, [redacted] and [redacted] were considered product-related substances. [redacted], [redacted], and [redacted] were considered product-related impurities. All of the product-related impurities are adequately controlled by the drug substance and finished product specifications.

2.1.6.3 Process-related impurities

Replication competent AAV (rcAAV), Impurity A, plasmid DNA, host cell DNA, host cell protein (HCP), Impurity B, Impurity C, [redacted], [redacted], Impurity D, and [redacted] were considered process-related impurities. All of the process-related impurities have been demonstrated to be adequately removed by the manufacturing process. rcAAV, Impurity A, plasmid DNA, host cell DNA, HCP, Impurity B, and Impurity C are adequately controlled by the drug substance specification, and Impurity D is adequately controlled by the finished product specification.

2.1.7 Control of drug substance

The proposed specifications for the drug substance consist of appearance, identification ([redacted]PCR[redacted]), osmolality, pH, purity ([redacted], Impurity A, plasmid DNA, host cell DNA, HCP, Impurity B, Impurity C), microbial limits, rcAAV, and titer ([redacted]PCR).

2.2 Finished product

2.2.1 Description and composition of finished product and finished product development

The finished product is supplied in vials as a solution for infusion. Each vial (5.5 or 8.3 mL) contains 1.1×10^{14} or 1.7×10^{14} vg of the active substance and the following excipients: trometamol, magnesium chloride, sodium chloride, Poloxamer 188, hydrochloric acid, and water for injection.

2.2.2 Manufacturing process

The finished product is manufactured through a process consisting of thawing of the drug substance, [redacted], sterile filtration, filling, packaging/labeling, and storage/testing.

[redacted] and sterile filtration have been defined as critical steps.

The commercial-scale finished product manufacturing process has undergone validation.

2.2.3 Manufacturing process development

The following are major changes made to the finished product manufacturing process during development (Process C, Process D, and Process E [the proposed commercial process]). The finished product manufacturing processes used for non-clinical and clinical studies are shown in Table 7.

- Process C → Process D: changes in [redacted], addition of [redacted]
- Process D → Process E (the proposed commercial process): changes in [redacted]

Following these process changes, the comparability of quality attributes between pre-change and post-change finished products has been demonstrated. Because the test methods of all specification tests for the finished product manufactured by Process C were different from those for the finished product manufactured by Process D or Process E, PMDA asked the applicant to present the results of comparability assessment using the same test methods during the review. The submitted test results demonstrated the comparability between the finished product manufactured by Process C and the finished product manufactured by Process D or Process E.

A quality by design (QbD) approach was used to develop the manufacturing process.

Table 7. Finished product manufacturing processes used for non-clinical studies and clinical trials

Process	Non-clinical studies or clinical trials
Process C	[redacted]
Process D	[redacted]
Process E (the proposed commercial process)	Studies CL-302, CL-303, and CL-304

2.2.4 Control of finished product

The proposed specifications for the finished product consist of appearance, identification ([redacted]PCR[redacted]), osmolarity, pH, purity ([redacted]), endotoxin, foreign insoluble matter, insoluble particulate matter, sterility, Impurity D, *in vivo* potency (prolongation of survival of SMNΔ7 mice), *in vitro* potency (SMN protein expression), infectious titer, [redacted], and titer ([redacted]PCR). *In vivo* potency (prolongation of survival of SMNΔ7 mice) was added in the course of regulatory review.

2.3 QbD

A QbD approach was used to develop the drug substance and the finished product, and a quality control strategy was established based on the following studies:

- Identification of critical quality attributes (CQAs):
  To assess product-related substances/product-related impurities, process-related impurities, and formulation quality attributes, the following CQAs were identified based on the information obtained during the development of Zolgensma, the relevant knowledge, etc.
  - ➢ titer, [redacted], infectious titer, Impurity D, *in vivo* potency (prolongation of survival of SMNΔ7 mice), *in vitro* potency (SMN protein expression), [redacted], plasmid DNA, host cell DNA, [redacted]HCP[redacted], rcAAV, endotoxin, bioburden, [redacted], viral safety, mycoplasma, osmolarity, pH, appearance, insoluble particulate matter, foreign insoluble matter, sterility
- Process characterization:
  Process parameters were classified by risk assessment based on their impact on CQAs, and each process step was characterized.
- Development of method of control:
  Process knowledge including the above process characterization demonstrated that the quality attributes of Zolgensma were adequately controlled through the combination of process parameter controls, in-process controls, and the specifications [for the control of product-related substances/product-related impurities and process-related impurities, see Sections 2.1.6.2 and 2.1.6.3].

2.R Outline of the review conducted by PMDA

Based on the submitted data and the following considerations, PMDA has concluded that the quality of the drug substance and the finished product is adequately controlled.

2.R.1 Data manipulation involving specification test of finished product

It was revealed that the data from the *in vivo* relative potency assay[1)] performed for release of the clinical product lots used in Studies CL-102, CL-302, CL-303, and CL-304 had been manipulated intentionally. The problem came to light as described below.

(1) In March 2019, [redacted] of AveXis received the allegations of inappropriate handling (alteration) of the records from the manufacturing facility where the assay in question was conducted. In response to the report, an internal investigation was launched.

(2) The internal investigation, including interviews with the key employees and a thorough review of the records and other data, concluded that the data had been manipulated due to the combination of the following factors: a lack of adequate training of the staff at this manufacturing site, a lack of the staff's understanding of the GMP/GCTP standards and ALCOA principles,[2)] poor internal QA/QC systems, corporate culture, and other problems.

(3) In July 2019, the applicant reported the developments of the above issues and related information to PMDA.

The applicant's explanation about the impact of this quality data manipulation on the quality of the clinical product lots used in clinical studies, and the appropriateness of quality control of the product for use in patients:

Impact on the quality of clinical product lots used in clinical studies

- In clinical studies using the clinical product lots released based on the results of the *in vivo* relative potency assay in which quality data were manipulated, no efficacy or safety problems were reported, and the efficacy and safety data raised no major concerns. In Study CL-304, the clinical product lot in question was administered to 2 Japanese patients, but there were no problematic findings from the safety or efficacy standpoint.
- All lots of the clinical product released based on the results of the *in vivo* relative potency assay in which quality data were manipulated were tested using the *in vitro* potency assay (SMN protein expression) for reassessment of potency. The test results for all clinical product lots were within the release specification for the *in vitro* potency assay.
- For quality assessment of the finished product, including assessment of comparability of quality attributes between the clinical product lots and commercial products, the present application must exclude the results of the *in vivo* relative potency assay due to the manipulation of quality data, and the data from the *in vitro* potency assay (SMN protein expression) and the *in vivo* potency assay (prolongation of survival of SMNΔ7 mice), which are included in the finished product specification, are to be used for the assessment of the potency of the finished product.

[1)] A potency assay to support Zolgensma development. The assay is different from the *in vivo* potency assay (prolongation of survival of SMNΔ7 mice) included in the commercial product specification, in terms of test system.
[2)] Source data should be Attributable, Legible, Contemporaneous, Original, and Accurate.

Appropriateness of quality control of product for use in patients

- The *in vitro* potency assay (SMN protein expression) and the *in vivo* potency assay (prolongation of survival of SMNΔ7 mice) included in the specification for the clinical product used in the ongoing clinical studies and the commercial product, were developed separately from the *in vivo* relative potency assay in question. These assays are performed in accordance with the newly developed SOPs. As the quality of the product for clinical use is assured appropriately by these two assays, there is no problem.

PMDA decided to remove the results of the *in vivo* relative potency assay from evaluation data because the quality data manipulation compromises the integrity of the results of the *in vivo* relative potency assay. Meanwhile, PMDA concluded that appropriate quality control is performed by the *in vitro* potency assay (SMN protein expression) and the *in vivo* potency assay (prolongation of survival of SMNΔ7 mice) in the commercial product specification.

3. Stability and Outline of the Review Conducted by PMDA

3.1 Stability of drug substance

The primary stability study on the drug substance is outlined in Table 8.

Table 8. Overview of primary stability study on drug substance

Study	No. of batches*	Storage conditions	Testing period	Storage package
Long-term	6	≤−60°C	[redacted]	[redacted] bottle

*: The drug substance manufactured by the proposed commercial process was used in the study.

Under the long-term condition, a decrease in [redacted] was observed in [redacted] at the [redacted]-month time point and in [redacted] at the [redacted]-month time point. For other batches, there were no significant changes in quality attributes throughout the testing period.

Based on the above, a shelf-life of 6 months has been proposed for the drug substance when stored in [redacted] bottle at ≤−60°C.

3.2 Stability of finished product

The primary stability studies on the finished product are outlined in Table 9.

Table 9. Overview of primary stability studies on finished product

Study	No. of batches[*1]	Storage conditions	Testing period	Fill volume	Storage package
Long-term	2	≤−60°C	12 months[*2]	[redacted]mL	Chlorobutyl rubber stopper and cyclic PO vial
	1			[redacted]mL	
	1		6 months[*2]	8.3 mL	
	1[*3]			5.5 mL	
Accelerated	2	2°C-8°C	[redacted]	[redacted] mL	
	1			[redacted] mL	
	1			8.3 mL	
	1			5.5 mL	
Stress	2	20°C-25°C	[redacted]	[redacted] mL	
	1			[redacted]mL	
	1			8.3 mL	
	1			5.5 mL	
Photostability	1	An overall illumination of ≥1.2 million lux·h and an integrated near ultraviolet energy of ≥200 W·h/m^2, [redacted]		[redacted] mL	
	1	An overall illumination of ≥1.2 million lux·h and an integrated near ultraviolet energy of ≥200 W·h/m^2, 5°C		[redacted] mL	

*1: The finished product produced by the proposed commercial process from the drug substance manufactured by the proposed commercial process was used in the studies.
*2: The stability studies will be continued for up to 36 months.
*3: Additional 1 batch will be tested for stability.

Under the long-term condition, [redacted] and [redacted] tended to decrease at the [redacted]-month time point. There were no significant changes in other attributes tested throughout the testing period.

Under the accelerated condition, [redacted] tended to decrease in some batches at the [redacted]-month time point.

Under the stress condition, a decrease in [redacted] was observed at the [redacted]-month time point.

In the photostability testing, there were no significant changes in quality attributes.

Based on the above, a shelf-life of 12 months has been proposed for the finished product when stored in a cyclic polyolefin (PO) vial with a chlorobutyl rubber stopper at ≤−60°C.

3.R Outline of the review conducted by PMDA

Based on the submitted data, PMDA accepted the storage conditions and shelf-lives for the drug substance and the finished product.

4. Indication or Performance

4.1 *In vivo* studies

In vivo pharmacology studies of Zolgensma conducted are shown in Table 10.

Table 10. Summary of *in vivo* studies

Study Title[*1]	Principal findings
AAV9 transduction efficiency study in P1 neonatal mice (*Nat Biotechnol*. 2009; 27: 59-65)	Following intravenous administration of scAAV9.CB.GFP vector (4×10^{11} vg) that expressed GFP (green fluorescent protein), instead of SMN protein, to neonatal C57BL/6 mice (postnatal days 1-2), >56% of neurons were transduced. On the other hand, following intravenous administration of scAAV9.CB.GFP vector (4×10^{11}-4×10^{12} vg) to adult mice (postnatal day 70), predominant glial transduction was observed. The percentage of transduced neurons ranged from 5% to 10%.
Transduction efficiency study in neonatal and juvenile monkeys (*Molecular Therapy*. 2011; 19: 1971-80)	Cynomolgus monkeys (postnatal days 1, 30, 90) received intravenous injection of scAAV9.CB.GFP vector ($1\text{-}3\times10^{14}$ vg/kg). Predominant neuronal transduction (approximately 70%) was observed.
Efficacy study using intrathecal delivery in a piglet model of SMA (*Ann Neurol*. 2015; 77: 399-414)	A model for SMA in piglets was generated by transducing piglets on postnatal day 5 with a short hairpin RNA (shRNA) construct targeting pig SMN to reduce endogenous SMN protein expression. Piglets on postnatal day 6 (pre-symptomatic SMA) received intrathecal injection of scAAV9.CBA.SMN vector (8×10^{12} vg/kg) but did not develop SMA signs or severe hind limb weakness during the period prior to euthanasia (6-10 weeks post-injection). On the other hand, in piglets intrathecally treated with scAAV9.CBA.SMN vector ($2\text{-}3.8\times10^{13}$ vg/kg)[*3] on postnatal day 33 to 36 (symptomatic SMA), partial amelioration of the disease[*6] was observed by the time of euthanasia (6-10 weeks post-injection).

*1: The percent homology of the *SMN* mRNA sequence between humans and mice/pigs/cynomolgus monkeys was 84%, 89%, and 98%, respectively. The percent homology of the amino acid sequence of SMN protein between humans and mice/pigs/cynomolgus monkeys was 82%, 89%, and 97%, respectively.

*3: It has the same genome as the proposed product, but the comparability of quality attributes with those of the proposed product has not been demonstrated.

*6: Compound muscle action potential (CMAP) values were similar to values obtained in the control group (normal piglets) and the treated pre-symptomatic group. On postnatal day 54, motor unit number estimation (MUNE) in treated animals was not similar to that in the control animals (normal piglets), but was partially corrected with a value higher than in vehicle-injected animals.

5. Biodistribution of the Product and Outline of the Review Conducted by PMDA

5.1 Non-clinical biodistribution

5.1.1 Biodistribution

Following single intravenous administration of Zolgensma to neonatal mice, the distribution of Zolgensma in different organs was evaluated (Table 11).

Table 11. Single-dose biodistribution studies

Test system	Route of administration	Observation period	Dose (vg/kg)	Summary of findings	Analytical method	Submitted data (CTD)
Male and female neonatal mice[*1] (FVB/NJ)	IV	3, 7, 12 weeks	2.37×10^{14}	The brain, heart, liver, spleen, lumbar spinal cord, quadriceps muscle, and lung tissues were evaluated. Zolgensma genomic DNA (gDNA) was detected in all tissues evaluated at all time points. This showed the persistence of Zolgensma. Human *SMN* transgene expression was also detected in these tissues.	gDNA and mRNA samples were isolated from mouse tissues, and ddPCR method was used for Zolgensma vector copy number determination and human *SMN* transgene expression analysis. A primer set designed to specifically detect a 131 bp sequence of Zolgensma was used.	4.2.2.1-1
Male and female neonatal mice[*2] (FVB/NCrl)	IV	3, 6, 12 weeks	1.5×10^{14} 2.4×10^{14} 3.0×10^{14}	The brain, heart, liver, spleen, lumbar spinal cord, quadriceps muscle, lung, and gonads (testis and ovary) tissues were evaluated. Zolgensma gDNA was detected in all tissues evaluated at all time points. This showed the persistence of Zolgensma. Human *SMN* transgene expression was also detected in these tissues.		4.2.3.1-6
Male and female neonatal mice[*1] (FVB/NCrl)	IV	3, 6, 12, 24 weeks	6.7×10^{13} 3.3×10^{14}	The brain, gonads, heart, jejunum, kidney, liver, lung, inguinal lymph node, masseter muscle (injection site), pancreas, quadriceps muscle, spinal cord, and spleen tissues were evaluated. Zolgensma gDNA was detected in all tissues evaluated at all time points. This showed the persistence of Zolgensma. The lowest levels were detected in the gonads, and Zolgensma gDNA levels in the gonads declined over time. Human *SMN* transgene expression profile was evaluated in male and female mice following single intravenous administration of 3.3×10^{14} vg/kg. The gonads, spleen, kidney, jejunum, pancreas, lung, brain, heart, liver, spinal cord, and quadriceps muscle tissues were collected from the mice. Human *SMN* transgene expression was found in almost all male and female mice at almost all time points, while no human *SMN* transgene expression was detected in the gonads in either males or females across all time points.	gDNA and mRNA samples were isolated from mouse tissues, and Q-PCR method was used for Zolgensma vector copy number determination and human *SMN* transgene expression analysis. A primer set designed to specifically detect a 123 bp sequence of Zolgensma was used.	4.2.3.1-4[*3]

*1: Mice at postnatal day (PND) 1

*2: Mice at PND 0

*3: This was a non-GLP study and submitted as reference data. The infectious titer of the test product used was lower than that of Zolgensma and the comparability of quality attributes has not been assured. In addition, the analytical procedures for assay have not been validated.

5.1.2 Shedding

No study for non-clinical shedding of Zolgensma was conducted.

5.R Outline of the review conducted by PMDA

The applicant's explanation about the biodistribution of Zolgensma:

Following intravenous administration of Zolgensma to neonatal mice, genomic DNA (gDNA) of Zolgensma was detected in the brain and spinal cord. This indicated that Zolgensma was able to cross the blood brain barrier. Zolgensma gDNA and human *SMN* mRNA were uniformly detected in all tissues evaluated at all time points. Especially, higher gDNA and transgene mRNA levels were detected in the heart, liver, lung, brain, spinal cord, and quadriceps muscle, whereas the levels were relatively low in the spleen and gonads (the testis

and ovary). These findings suggested the systemic distribution and persistence of Zolgensma.

PMDA accepted the applicant's explanation.

The possibility that Zolgensma remains in the reproductive organs for a long period of time and its impact on germ cells are discussed in the section of Toxicity [see Section 6.R.4].

6. Non-clinical Safety and Outline of the Review Conducted by PMDA

The applicant submitted the results from single-dose toxicity studies as non-clinical safety studies. Since the target patient population of Zolgensma is neonates, neonatal mice were used in the toxicity studies.

6.1 Single-dose toxicity

Single intravenous dose toxicity studies were conducted in neonatal mice (Table 12). The principal findings related to Zolgensma were inflammatory changes in the heart and hepatocyte necrosis, and there were deaths associated with heart thrombi at $\geq 2.4\times10^{14}$ vg/kg. The inflammatory changes in the liver and heart were reversible. The applicant explained that these findings were of little toxicological significance because inflammatory changes in the lung were slight and observed only at scheduled sacrifice at Week 6.

Table 12. Single-dose toxicity studies

Test system	Route of administration	Observation period	Dose	Principal findings	Submitted data[*4] (CTD)
Male and female neonatal mice[*1] (FVB/NJ)	IV	3, 6, 12 weeks	0 (0.9% saline) 7.9×10^{13} vg/kg 2.37×10^{14} vg/kg 3.91×10^{14} vg/kg	Findings at 3.91×10^{14} vg/kg: Deaths associated with heart thrombi. Decreased activity; abdominal distension; dehydration; labored breathing; dark eyeball; increase in white blood cells; decreases in red blood cells and hemoglobin; increase in reticulocytes; decreases in total protein, albumin, and globulin; degeneration in the heart and liver; and heart thrombi. NOAEL in the study: 2.37×10^{14} vg/kg	4.2.3.1-5
Male and female neonatal mice[*2] (FVB/NCrl)	IV	3, 6, 12 weeks	0 (vehicle[*3]) 1.5×10^{14} vg/kg 2.4×10^{14} vg/kg 3.0×10^{14} vg/kg	Findings at $\geq1.5\times10^{14}$ vg/kg: Inflammation in the myocardium, myocardial fibrosis, edema, hepatocellular hypertrophy. Findings at $\geq2.4\times10^{14}$ vg/kg: Deaths associated with atrial thrombosis. Thrombosis, fibroplasia, myocardial degeneration/necrosis, and dilation in the atrium; perivascular inflammation and chronic inflammation in the lung; and increased sinusoidal macrophages, hepatocyte necrosis, and perinuclear vacuolation of hepatocytes in the liver Findings at 3.0×10^{14} vg/kg: Irregular or labored respiration, hypoactivity, hunched appearance, eye protrusion and discolored eyes, enlarged hearts and abnormal shape The maximum tolerated dose in the study: 1.5×10^{14} vg/kg	4.2.3.1-6
Cynomolgus monkeys	IT	2 weeks	3.0×10^{13} vg/animal	Inflammatory mononuclear cell infiltrates with neuronal necrosis/complete neuronal loss with mineralization in the dorsal root ganglia of the spinal cord (cervical, thoracic, lumbar, or sacral)	4.2.3.1-9[*5]

*1: Mice at PND 1
*2: Mice at PND 0
*3: 20 mM Tris, 1 mM $MgCl_2$, 200 mM NaCl, and 0.005% Poloxamer 188 solution was used as vehicle.
*4: Single-dose toxicity studies in mice or monkeys (CTD 4.2.3.1-1, 4.2.3.1-2, 4.2.3.1-3, 4.2.3.1-4, 4.2.3.1-7, 4.2.3.1-8) other than the studies presented in Table 12 were submitted as reference data because the consistency of quality between the test product used in these studies and Zolgensma has not been assured, etc.
*5: The study was a non-GLP study, and submitted as reference data.

6.2 Other safety evaluation

6.2.1 Potential chromosomal integration

The applicant explained that as with wild-type AAV, recombinant AAV vectors are considered to integrate into chromosome at low frequencies (*Clin Microbiol Rev*. 2008; 21: 583-93).

6.2.2 Tumorigenic and carcinogenic potential

The applicant's explanation:

The risk of tumorigenesis and carcinogenesis associated with the use of Zolgensma should be low, based on the following points:

- Generally, the AAV9 vector genome does not integrate into the host genome, and therefore there is no risk of insertional mutagenesis (Draft Guidance for Industry: Long Term Follow-Up After Administration of Human Gene Therapy Products, 2008).
- Tumor formation related to the overexpression of the SMN protein which is produced by Zolgensma has not been reported (*Curr Genomics.* 2018; 19: 339-55, *Neurobiol Aging*. 2014; 35: 906-15), and the association of the *SMN* gene with cancer has not been reported (the Catalogue of Somatic Mutations in Cancer [COSMIC v90, https://cancer.sanger.ac.uk/cosmic, accessed on September 5, 2019]).
- Pharmacology and toxicity studies of Zolgensma indicated no Zolgensma-related proliferative changes or tumor formation.

6.2.3 Reproductive and developmental toxicity

In view of the target patient population for Zolgensma, no reproductive and developmental toxicity studies were conducted.

6.2.4 Safety assessment of process-related impurities

The applicant's explanation about the safety of Impurity C, a process-related impurity:

Results of single-dose toxicity studies of Zolgensma (CTD 4.2.3.1-5 and 4.2.3.1-6) and other studies indicate that these are no safety concerns about Impurity C in the clinical use of Zolgensma.

6.2.5 Safety assessment of inactive ingredients of the product

The applicant's explanation about the safety of the inactive ingredients (excipients) of the product, i.e., buffer (Tris), sodium chloride, magnesium chloride, and Poloxamer 188:

Given the results of a single-dose toxicity study of Zolgensma (CTD 4.2.3.1-6) and taking into account that these excipients have been used in the approved pharmaceutical products in Japan, there are no safety concerns in the clinical use of Zolgensma.

6.R Outline of the review conducted by PMDA

Based on the following considerations on the submitted data, PMDA considers that the clinical use of Zolgensma is acceptable from a non-clinical safety perspective.

6.R.1 Effects on liver and heart

Hepatocyte necrosis in the liver, and thrombi, inflammation, and necrosis in the heart were observed in single-dose toxicity studies in mice. PMDA asked the applicant to explain their mechanism of development and safety in humans.

The applicant's explanation:

For the following reasons, the changes observed in single-dose toxicity studies in mice may occur also in humans. Precautionary statements about these risks will be included in the package insert.

- Hepatocyte necrosis in the liver, and inflammation, degeneration, and necrosis in the heart are considered related to the AAV9 capsid and/or SMN protein because Zolgensma gDNA was distributed in the liver and heart in a single-dose biodistribution study in neonatal mice (see CTD4.2.3.1-6, Section 5.1.1). However, these toxicological findings are considered related to the immune response to the AAV9 capsid of Zolgensma because no effects of SMN protein overexpression in humans or animals have been reported (*Curr Genomics*. 2018; 19: 339-55, *Neurobiol Aging*. 2014; 35: 906-15).
- The mechanism of development of heart thrombi is unknown, and its relevance to humans cannot be ruled out.

PMDA's view:

The applicant's explanation about the mechanism of development of the changes observed in single-dose toxicity studies in mice is merely an inference. The applicant should continue to collect information on safety in humans via post-marketing surveillance. Hepatocyte necrosis in the liver, and inflammation, degeneration, and necrosis in the heart are considered important risks that may occur during the clinical use of Zolgensma, and information on these risks should therefore be appropriately communicated to healthcare professionals in clinical practice, through the use of the package insert and other materials. Hepatotoxicity and cardiotoxicity are discussed also in the section of clinical safety [see Sections 8.R.4.1 and 8.R.4.2].

6.R.2 Effects on the dorsal root ganglia of the spinal cord

Findings reported in a single intrathecal dose toxicity study in cynomolgus monkeys (CTD 4.2.3.1-9) included inflammatory changes with neuronal necrosis in the dorsal root ganglia (DRGs) of the spinal cord. PMDA asked the applicant to explain the safety of intravenously administered Zolgensma in humans.

The applicant's response is as follows:

- Inflammatory changes in the DRG of the spinal cord were reported also in studies in which other AAV9 vectors were administered intravenously or intrathecally to monkeys and piglets (*Hum Gene Ther*. 2018; 29: 285-98, *Mol Ther Methods Clin Dev*. 2018; 10: 79-88). These findings are considered related to Zolgensma.
- Although the reversibility of the findings reported in a single intrathecal dose toxicity study in cynomolgus monkeys is unknown, the changes are inferred to have occurred due to accumulation of Zolgensma in the DRG surrounded by the cerebrospinal fluid, resulting from an increased Zolgensma concentration in the

cerebrospinal fluid following intrathecal administration of Zolgensma 6 mL into the monkey's cerebrospinal fluid <15 mL.

- Compared with intrathecal administration, intravenous administration of Zolgensma is inferred to result in lower concentrations of Zolgensma in the cerebrospinal fluid and thus less accumulation of Zolgensma in the DRG.

In addition, the applicant explained that though the safety of intravenously administered Zolgensma in humans is unknown, no adverse reactions related to these findings occurred in patients receiving intravenous Zolgensma in clinical trials of Zolgensma (data cutoff date of March 8, 2019).

PMDA's view:

Results of a single intravenous dose biodistribution study in mice (CTD 4.2.3.1-4) showed that Zolgensma was distributed in the spinal cord [see Section 5.1.1]. This suggests that intravenous administration of Zolgensma in humans potentially leads to the risk of inflammation in the DRG of the spinal cord. Thus, information on the findings reported in a single intrathecal dose toxicity study in cynomolgus monkeys should be communicated to healthcare professionals via the package insert and other materials.

6.R.3 Observation period of general toxicity studies

Given that long-term expression of the *SMN* gene delivered by a single dose of Zolgensma in humans is desired, PMDA asked the applicant to explain the appropriateness of the observation period of single-dose toxicity studies (CTD 4.2.3.1-5, 4.2.3.1-6) (up to 12 weeks).

The applicant explained that the observation period of general toxicity studies was appropriate, with the following justifications:

- The heart and liver findings observed in single-dose toxicity studies in mice (CTD 4.2.3.1-5, 4.2.3.1-6) were reversible at Week 12.
- Zolgensma is replication incompetent, and most of the DNA from the wild-type AAV has been replaced with the genes for the expression of the SMN protein. These factors are unlikely to lead to chronic toxicity associated with AAV.
- Syndrome related to the overexpression of the *SMN* gene in humans or animals has not been identified, nor are there any reports on effects of the overexpression of the *SMN* gene in specific tissues (*Curr Genomics*. 2018; 19: 339-55, *Neurobiol Aging*. 2014; 35: 906-15).

PMDA's view:

Since the *SMN* gene delivered by Zolgensma in humans may be expressed persistently, an observation period of >12 weeks should have been chosen. However, given that SMA is a serious disease and that no new toxicity findings were reported in general toxicity studies with a prolonged observation period (6 weeks → 12 weeks), the use of Zolgensma in patients is acceptable, taking also account of the currently available clinical study results. The applicant should collect information on the long-term safety of Zolgensma in humans via post-marketing surveillance.

6.R.4 Effects on germ cells

According to biodistribution evaluation in single intravenous dose toxicity studies in mice (CTD 4.2.3.1-5, 4.2.3.1-6), there was no trend towards decreasing levels of Zolgensma in the reproductive organs until Week 12. PMDA asked the applicant to explain how Zolgensma potentially affects germ cells if it remains persistently in the reproductive organs.

The applicant explained that the effects of Zolgensma on the reproductive organs in the target patient population are limited, with the following justifications:

- Given that Zolgensma is a non-integrating, non-replicating AAV vector product, the viral capsid or the transgene is very unlikely to remain in germ cells for ≥10 years, until patients treated with Zolgensma <1 year of age reach the reproductive age.
- In a single intravenous dose toxicity study in mice (CTD 4.2.3.1-6), Zolgensma gDNA levels in the reproductive organs were about 10- to 100-fold lower than those in the major tissues (the heart, CNS, muscle), and there were no toxicological changes in the ovary or testis at Week 12.
- Following intravenous injection of AAV9 vector in monkeys, AAV9-mediated transgene expression was detected in the Leydig cells of the testes only as a reproductive organ (*Molecular Therapy*. 2011; 19: 1971-80).

PMDA's view:

According to biodistribution evaluation of Zolgensma, there was no trend towards decreasing levels of Zolgensma gDNA in the reproductive organs until Week 12, and a clinical pharmacology study also showed that Zolgensma gDNA had not been cleared from blood at Month 12 [see Section 7.1.1]. Given these findings, Zolgensma remaining persistently in the reproductive organs potentially adversely affects germ cells. Thus, the package insert and other materials should provide information on this risk.

7. Clinical pharmacology and Outline of the Review Conducted by PMDA

7.1 Clinical pharmacology

7.1.1 Blood concentration after intravenous administration

The time course of blood Zolgensma gDNA levels was determined in 3 patients in Study CL-101 (Figure 6). Blood samples were collected at 7, 14, and 21 days and 1, 2, 3, 6, 9, and 12 months after Zolgensma infusion, and Zolgensma gDNA levels in blood were determined by droplet-digital polymerase chain reaction (ddPCR).

The blood Zolgensma gDNA levels declined rapidly to approximately 0.07% to 0.15% of the estimated blood level immediately after infusion based on body weight and blood volume[3)] by Day 7. At Month 6, the blood Zolgensma gDNA levels declined towards 1.0×10^5 vg/mL (lower limit of quantification [LLOQ], 2.0×10^4 vg/mL), and remained constant until Month 12, the end of observation period.

[3)] Assuming that boys 6.5 months of age with a median weight of 8.16 kg have a blood volume of 77 mL/kg, the blood level was estimated at 1.4×10^{12} vg/mL.

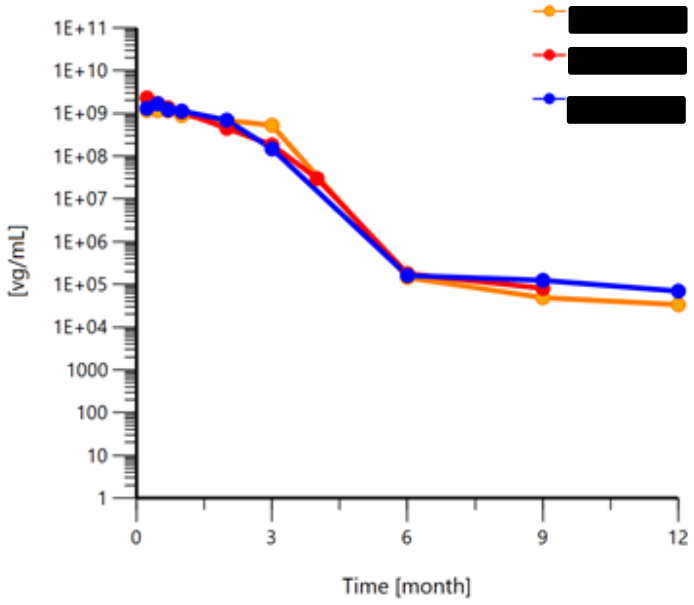

Figure 6. Time course of blood Zolgensma gDNA levels in Study CL-101

7.1.2 Shedding

The time courses of Zolgensma gDNA levels in stool, urine, and saliva after Zolgensma infusion were determined in 5 patients in Study CL-101. Samples of stool, urine, and saliva were collected at 1, 7, 14, 21, and 30 days after infusion, monthly through Month 12, and every 3 months thereafter, and Zolgensma gDNA levels were determined by ddPCR. In stool, urine, and saliva, Zolgensma gDNA declined to unquantifiable levels (LLOQ, 1.1×10^7 vg/g in stool; 1.1×10^6 vg/mL in urine and saliva) within 30 days after infusion, and remained constant at near undetectable levels (lower limit of detection, 1.1×10^6 vg/g in stool; 1.1×10^5 vg/mL in urine and saliva) from Day 60 until up to Month 18.

7.2 Antibody formation following administration of Zolgensma

In Study CL-101, anti-AAV9 antibody titers and anti-human SMN antibody titers were measured by an enzyme linked immunosorbent assay (ELISA) at 1, 2, and 3 weeks and 1, 2, 3, 6, 9, 12, 15, 18, and 24 months after Zolgensma infusion.

Although both anti-AAV9 and anti-SMN antibody titers were undetectable[4)] at baseline, all patients were positive for anti-AAV9 antibodies after Zolgensma infusion. On the other hand, all patients were negative for anti-SMN antibodies also after Zolgensma infusion.

[4)] A cutoff value of 1:50 was used. A cut-off value of 1:12.5 was used at screening for anti-SMN antibody titers.

7.R Outline of the review conducted by PMDA

The applicant's explanation about the biodistribution of Zolgensma:

Zolgensma was persistently detected in patient samples because the expression construct delivered by Zolgensma does not integrate into the chromosome of the infected cell and exists as an extrachromosomal episome in the cell for long periods of time.

PMDA accepted the applicant's explanation.

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

The applicant submitted efficacy and safety evaluation data, in the form of the results from 1 study presented in Table 13. The applicant also submitted the results from 4 studies presented in Table 13 as reference data. The studies other than Study CL-101 are ongoing.

Table 13. Listing of clinical studies to evaluate efficacy and safety

Data category	Geographical location	Study Identifier	Phase	Study population	No. of subjects enrolled	Dosing regimen*	Main endpoints
Evaluation	Foreign	CL-101	I	SMA Type I patients	Cohort 1 3 Cohort 2 12	Cohort 1 Zolgensma 6.7×10^{13} vg/kg Cohort 2 Zolgensma 2.0×10^{14} vg/kg	Safety Efficacy
Reference	Foreign	LT-001	Long-term follow-up	Patients who were enrolled in Study CL-101 and completed 24 months of follow-up	13	No additional Zolgensma infusion	Safety Efficacy
		CL-102	I	SMA Type II or III patients	Cohort 1 3 Cohort 2 25 Cohort 3 2	Cohort 1 Zolgensma 6.0×10^{13} vg/kg Cohort 2 Zolgensma 1.2×10^{14} vg/kg Cohort 3 Zolgensma 2.4×10^{14} vg/kg	Safety Efficacy
		CL-303	III	Patients with SMA Type I or genetically diagnosed SMA Type I	22	Zolgensma 1.1×10^{14} vg/kg	Efficacy Safety
	Global	CL-304	III	Patients with genetically diagnosed SMA Type I, II, or III	Cohort 1 8 Cohort 2 9 Cohort 3 1	Zolgensma 1.1×10^{14} vg/kg	Efficacy Safety

*: Zolgensma was administered intrathecally in Study CL-102 and intravenously in other studies.

The clinical studies are summarized below.

8.1 Evaluation data

8.1.1 Foreign phase I study (CTD5.3.5.1-1, Study CL-101 [May 2014 to December 2017])

An open-label, uncontrolled study was conducted at 1 site overseas to evaluate the safety and efficacy of Zolgensma in patients with SMA Type I defined by bi-allelic mutations (deletion or point mutation) in the *SMN1* gene, and 2 copies of the *SMN2* gene; the onset of clinical symptoms before age of 6 months; ≤6 months

of age (≤9 months of age[5)] for the first 9 subjects) at the time of infusion; and negative for elevated anti-AAV9 antibodies[6)] (target sample size, 12-15 subjects (3 in Cohort 1, 3-6 in Cohort 2A, 3 in Cohort 2B, 3 in Cohort 3[7)]) [for the key inclusion and exclusion criteria, see Table 14].

Table 14. Key inclusion and exclusion criteria

Inclusion criteria	Patients meeting all of the following criteria ● Bi-allelic mutations (deletion or point mutation) in the *SMN1* gene and 2 copies of the *SMN2* gene ● Absence of the c.859G>C substitution in *SMN2*[*] ● Onset of clinical symptoms at birth to 6 months of age, and 6 months of age or younger on the day of infusion ● Hypotonia by clinical evaluation with delay in motor skills, poor head control, round shoulder posture, and hypermobility of joints
Exclusion criteria	Patients meeting any of the following criteria ● Use of invasive ventilatory support (tracheotomy with positive pressure) or SpO_2 <95% at screening ● Presence of anti-AAV9 antibody ● Clinically significant abnormal laboratory values (GGT >3 × upper limit of normal (ULN), bilirubin ≥3.0 mg/dL, creatinine ≥1.8 mg/dL, hemoglobin <8 or >18 g/dL, or white blood cell count >20,000/mm^3)

*: Given that c.859G>C results in a milder phenotype (*Am J Hum Genet*. 2009; 85: 408-13) and that only 0.5% to 3.8% of SMA patients harbor c.859G>C (*J Med Genet.* 2010; 47: 640-2, etc.), these patients were excluded in order to enroll a homogeneous patient population in Study CL-101, wherever possible, and evaluate the efficacy and safety of Zolgensma.

Dosage regimen or method of use[8)] was as follows: The doses of Zolgensma were 6.7×10^{13} vg/kg for Cohort 1, 2.0×10^{14} vg/kg for Cohorts 2A and 2B, and 3.3×10^{14} vg/kg for Cohort 3, and Zolgensma was to be administered as a single, one-time intravenous infusion over approximately 60 minutes.

An overview of the study is shown in Figure 7. Subjects were assessed for safety for 24 months after Zolgensma infusion. Patients who completed 24 months of follow-up were to be enrolled in Study LT-001 [see Section 8.2.1] for 15-year, long-term followed-up.

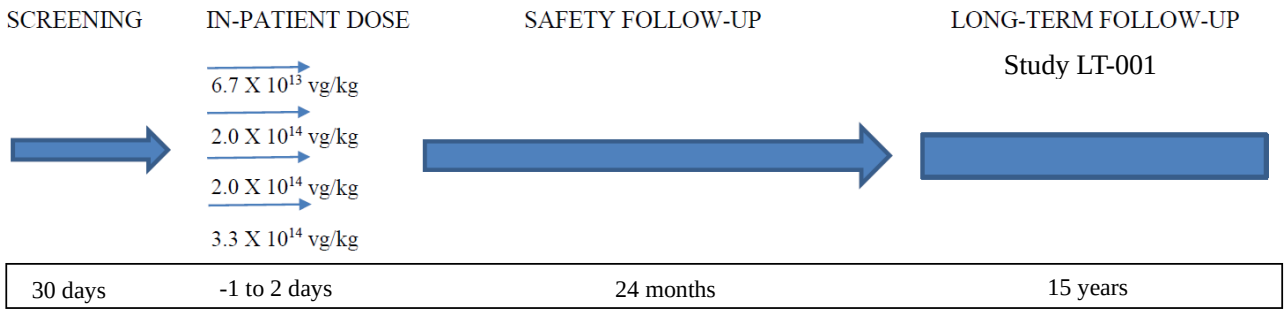

Figure 7. Overview of Study CL-101

[5)] Taking into account the age at onset of SMA Type I (<6 months of age) (the study population for CL-101) and the time to definitive diagnosis, the age for enrollment was "≤9 months of age" at the time of initiating the study. However, based on the following considerations on the first 9 patients enrolled in Study CL-101 (3 in Cohort 1, 6 in Cohort 2), the age for enrollment was changed to ≤6 months of age (Protocol Addendum 2 [as of [redacted], 20[redacted]]).
- Among the 9 patients, the youngest patient (1.9-month-old) showed highest efficacy (achieved the ability to walk without support).
- In Cohort 2 (6 patients), 5 patients aged ≤6 months showed higher efficacy than 1 patient aged 7.9 months.

[6)] Defined as anti-AAV9 antibody titer of ≤1:50. A potential patient with anti-AAV9 antibody titer >1:50 was eligible to participate in the study if anti-AAV9 antibody titer upon retesting was ≤1:50. Actually, 2 patients (Subject Numbers 12 and 13) were enrolled in Study CL-101 based on the results of retesting, and received Zolgensma.

[7)] When some data supporting the efficacy of Zolgensma were obtained in Cohort 2B, without improvement of ≥8 points in score on the Bayley Scales of Infant and Toddler Development, the study was to escalate to Cohort 3.

[8)] At the time of initiating the study, the doses of Zolgensma were 6.7×10^{13} vg/kg for Cohort 1 and 2.0×10^{14} vg/kg for Cohorts 2A and 2B, based on the titers measured by Q-PCR. Then, AveXis established a ddPCR based viral genome titer assay, and the titers for Cohorts 1 and 2 measured by the ddPCR method were 3.7×10^{13} and 1.1×10^{14} vg/kg, respectively. [redacted]PCR method will be used for determination of viral titers after marketing of Zolgensma.

After 3 subjects in Cohort 1 and 6 subjects in Cohort 2A received Zolgensma, whether to initiate enrollment of Cohort 3 was determined. It was decided not to escalate to Cohort 3, because (i) the data from Cohort 2A patients showed the promising efficacy of Zolgensma and (ii) an increased dose of Zolgensma may increase the risk of liver function abnormalities. Cohort 2B was expanded to include 6 patients (Protocol Version 13.0 [as of [redacted], 20[redacted]] and Protocol Addendum 2 [as of [redacted], 20[redacted]]). As the same dose of Zolgensma was used for Cohorts 2A and 2B, Cohorts 2A and 2B combined is hereinafter referred to as Cohort 2.

All of 15 subjects enrolled in the study[9)] (3 in Cohort 1, 12 in Cohort 2) received Zolgensma and included in the safety analysis set. The same population was used for the efficacy analysis.

Adverse events reported by ≥2 subjects are shown in Table 15. Those for which a causal relationship to Zolgensma could not be ruled out were transaminases increased (1 subject) in Cohort 1 and transaminases increased (3 subjects) and AST increased (1 subject) in Cohort 2.

Serious adverse events reported are shown in Table 16. Those for which a causal relationship to Zolgensma could not be ruled out were transaminases increased (1 subject) in Cohort 1 and transaminases increased (1 subject) in Cohort 2. No adverse events leading to death were reported.

Table 15. Adverse events reported by ≥2 subjects in Cohorts 1 and 2 combined (Safety analysis set)

	Cohort 1 (N = 3)		Cohort 2 (N = 12)		Total (N = 15)	
	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4
Any adverse event	3 (100)	3 (100)	12 (100)	10 (83.3)	15 (100)	13 (86.7)
Respiratory, thoracic and mediastinal disorders						
Nasal congestion	0	0	6 (50.0)	0	6 (40.0)	0
Cough	0	0	5 (41.7)	0	5 (33.3)	0
Atelectasis	0	0	4 (33.3)	3 (25.0)	4 (26.7)	3 (20.0)
Respiratory failure	1 (33.3)	1 (33.3)	3 (25.0)	2 (16.7)	4 (26.7)	3 (20.0)
Rhinorrhoea	0	0	3 (25.0)	0	3 (20.0)	0
Pneumonia aspiration	0	0	2 (16.7)	2 (16.7)	2 (13.3)	2 (13.3)
Respiratory distress	0	0	2 (16.7)	2 (16.7)	2 (13.3)	2 (13.3)
Wheezing	0	0	2 (16.7)	0	2 (13.3)	0
Infections and infestations						
Upper respiratory tract infection	1 (33.3)	0	10 (83.3)	2 (16.7)	11 (73.3)	2 (13.3)
Pneumonia	0	0	7 (58.3)	7 (58.3)	7 (46.7)	7 (46.7)
Gastroenteritis viral	0	0	5 (41.7)	1 (8.3)	5 (33.3)	1 (6.7)
Enterovirus infection	1 (33.3)	0	4 (33.3)	2 (16.7)	5 (33.3)	2 (13.3)
Rhinovirus infection	1 (33.3)	0	4 (33.3)	2 (16.7)	5 (33.3)	2 (13.3)
Parainfluenzae virus infection	1 (33.3)	1 (33.3)	3 (25.0)	2 (16.7)	4 (26.7)	3 (20.0)
Otitis media	2 (66.7)	0	2 (16.7)	0	4 (26.7)	0
Ear infection	1 (33.3)	0	2 (16.7)	0	3 (20.0)	0
Bronchiolitis	0	0	3 (25.0)	1 (8.3)	3 (20.0)	1 (6.7)
Viral upper respiratory inflammation	0	0	3 (25.0)	1 (8.3)	3 (20.0)	1 (6.7)
Pharyngitis streptococcal	1 (33.3)	0	2 (16.7)	0	3 (20.0)	0
Pneumonia respiratory	1 (33.3)	1 (33.3)	2 (16.7)	2 (16.7)	3 (20.0)	3 (20.0)

[9)] Although 16 patients were screened, 1 was not enrolled owing to the presence of anti-AAV9 antibody.

	Cohort 1 (N = 3)		Cohort 2 (N = 12)		Total (N = 15)	
	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4
syncytial viral						
Respiratory syncytial virus bronchiolitis	1 (33.3)	0	2 (16.7)	1 (8.3)	3 (20.0)	1 (6.7)
Adenovirus infection	0	0	2 (16.7)	1 (8.3)	2 (13.3)	1 (6.7)
Conjunctivitis	0	0	2 (16.7)	0	2 (13.3)	0
Urinary tract infection	0	0	2 (16.7)	0	2 (13.3)	0
Influenza	1 (33.3)	1 (33.3)	1 (8.3)	0	2 (13.3)	1 (6.7)
Gastrointestinal disorders						
Vomiting	0	0	8 (66.7)	1 (8.3)	8 (53.3)	1 (6.7)
Constipation	1 (33.3)	0	6 (50.0)	0	7 (46.7)	0
Gastrooesophageal reflux disease	1 (33.3)	0	5 (41.7)	0	6 (40.0)	0
Diarrhoea	0	0	3 (25.0)	1 (8.3)	3 (20.0)	1 (6.7)
Skin and subcutaneous tissue disorders						
Rash	0	0	5 (41.7)	0	5 (33.3)	0
Decubitus ulcer	0	0	2 (16.7)	1 (8.3)	2 (13.3)	1 (6.7)
Erythema	1 (33.3)	0	1 (8.3)	0	2 (13.3)	0
General disorders and administration site conditions						
Pyrexia	1 (33.3)	0	7 (58.3)	0	8 (53.3)	0
Injury, poisoning and procedural complications						
Fall	0	0	3 (25.0)	0	3 (20.0)	0
Investigations						
Transaminases increased	1 (33.3)	1 (33.3)	3 (25.0)	1 (8.3)	4 (26.7)	2 (13.3)
Human rhinovirus test positive	0	0	3 (25.0)	2 (16.7)	3 (20.0)	2 (13.3)
Enterovirus test positive	0	0	2 (16.7)	1 (8.3)	2 (13.3)	1 (6.7)
Metabolism and nutrition disorders						
Dehydration	0	0	2 (16.7)	1 (8.3)	2 (13.3)	1 (6.7)
Cardiac disorders						
Tachycardia	0	0	2 (16.7)	1 (8.3)	2 (13.3)	1 (6.7)

MedDRA ver.20.0, Severity grade based on CTCAE v4.03
n (%)

Table 16. Serious adverse events (Safety analysis set)

	Cohort 1 (N = 3)	Cohort 2 (N = 12)	Total (N = 15)
Any adverse event	3 (100)	10 (83.3)	13 (86.7)
Infections and infestations			
Pneumonia	0	7 (58.3)	7 (46.7)
Upper respiratory tract infection	0	3 (25.0)	3 (20.0)
Parainfluenzae virus infection	1 (33.3)	2 (16.7)	3 (20.0)
Pneumonia respiratory syncytial viral	1 (33.3)	2 (16.7)	3 (20.0)
Respiratory syncytial virus bronchiolitis	1 (33.3)	2 (16.7)	3 (20.0)
Adenovirus infection	0	2 (16.7)	2 (13.3)
Enterovirus infection	0	2 (16.7)	2 (13.3)
Rhinovirus infection	0	2 (16.7)	2 (13.3)
Gastroenteritis	0	1 (8.3)	1 (6.7)
Gastroenteritis viral	0	1 (8.3)	1 (6.7)
Lower respiratory tract infection	0	1 (8.3)	1 (6.7)
Pneumonia parainfluenzae viral	0	1 (8.3)	1 (6.7)
Pneumonia viral	0	1 (8.3)	1 (6.7)
Postoperative wound infection	0	1 (8.3)	1 (6.7)
Viral upper respiratory tract infection	0	1 (8.3)	1 (6.7)
Bronchitis	1 (33.3)	0	1 (6.7)
Influenza	1 (33.3)	0	1 (6.7)
Respiratory, thoracic and mediastinal disorders			
Pneumonia aspiration	0	2 (16.7)	2 (13.3)

	Cohort 1 (N = 3)	Cohort 2 (N = 12)	Total (N = 15)
Respiratory distress	0	2 (16.7)	2 (13.3)
Atelectasis	0	1 (8.3)	1 (6.7)
Respiratory failure	1 (33.3)	0	1 (6.7)
Investigations			
Human rhinovirus test positive	0	2 (16.7)	2 (13.3)
Transaminases increased	1 (33.3)	1 (8.3)	2 (13.3)
Enterovirus test positive	0	1 (8.3)	1 (6.7)
Norovirus test positive	0	1 (8.3)	1 (6.7)
Oxygen saturation decreased	0	1 (8.3)	1 (6.7)
Injury, poisoning and procedural complications			
Femur fracture	0	1 (8.3)	1 (6.7)
Post procedural haemorrhage	0	1 (8.3)	1 (6.7)
Cardiac disorders			
Tachycardia	0	1 (8.3)	1 (6.7)
Metabolism and nutrition disorders			
Dehydration	0	1 (8.3)	1 (6.7)

MedDRA ver.20.0
n (%)

Table 17 shows the efficacy endpoints. The primary efficacy endpoint was compared with a natural history cohort of SMA Type I (N = 23) derived from the US Pediatric Neuromuscular Clinical Research (PNCR) (*Neurology.* 2014; 83: 810-7).

Table 17. Efficacy endpoints

Primary efficacy endpoint	● Time from birth to either the need for permanent ventilatory support (defined as tracheostomy or ≥16 hours of respiratory assistance per day [including bi-level positive airway pressure (BiPAP)] continuously for ≥14 days in the absence of an acute reversible illness, excluding perioperative ventilation) or death
Secondary efficacy endpoints	● Achievement of motor milestones (head control, sitting, walking) (review by the study site and central review*) ● Change from baseline in Children's Hospital of Philadelphia Infant Test of Neuromuscular Disorders (CHOP-INTEND) score
Exploratory efficacy endpoints	● Patient function assessment with the CHOP-INTEND ● Bayley Scales of Infant and Toddler Development, Third Edition (the Bayley Scales of Infant and Toddler Development test), as a standardized, norm-referenced infant assessment ➢ The gross and fine motor portions as well as language and cognition portions of this test were administered in patients who achieved a score ≥60 of 64 on the CHOP-INTEND. ● Motor neuron function assessment by compound muscle action potential (CMAP) ● The proportion of patients not requiring non-oral nutrition (enteral feeding) prior to therapy who maintained the ability to thrive (defined by able to tolerate thin liquids, demonstrated through a formal swallowing test, not receiving nutrition through mechanical support [i.e., feeding tube], and maintaining weight >3rd percentile for age and gender) ● The proportion of patients who were independent of ventilatory support (defined as requiring no daily ventilator support/usage in the absence of acute reversible illness and excluding perioperative ventilation)

*: For objective and fair assessment of motor milestones, the protocol was amended to add the following during the study (Protocol Version 14.0 [as of [redacted], 20[redacted]]): The video recordings made by the medical staff, the patient's family, etc. at hospital, at home, etc. should be edited by an external contractor based on the advice of the study physical therapist, investigator, and sub-investigator at the study site, and reviewed by an independent, external reviewer (1 person) as central review. As a result, assessment at Month 24 only was to be subjected also to central review. In the event of a discrepancy in assessment between the study site and the independent external reviewer, motor milestone achievement was to be determined by the independent external reviewer after discussion with the study site.

The primary efficacy endpoint of "the time from birth to either the need for permanent ventilatory support or death" was evaluated. All of 15 subjects (100%)[10)] survived without permanent ventilation through 13.6 months of age and at the time of completing 24 months of follow-up (median age [range], 28.1 [25.3-32.4] months). In the natural history cohort of SMA Type I derived from the PNCR (*Neurology.* 2014; 83: 810-7) for a control comparison, the proportions of patients who achieved permanent ventilation-free survival at the age of 13.6 and 20 months were 25% and 8%, respectively.

Furthermore, the efficacy data other than "the time from birth to the need for permanent ventilatory support or death" are shown below.

Ventilator-use results and motor milestone achievement are shown in Table 18.

Table 18. Ventilator-use results and motor milestone achievement
(Efficacy analysis set, review by study site and central review [at Month 24 only])

Cohort		1			2											
Subject Number		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Sex		F	F	M	F	M	F	F	F	F	M	M	F	F	M	M
Age at symptom onset (months)		3	1	1	3	1	1	1	2	3	0	1	2	0	2	1
Age at infusion (months)		5.9	5.9	7.2	5.6	4.2	1.9	3.6	7.9	4.9	0.9	2.3	2.6	0.9	4.1	2.1
Independent of ventilatory support including non-invasive support	Baseline	–	–	–	✔	✔	✔	✔	–	✔	✔	✔	✔	✔	✔	–
	12 months	–	–	–	–	–	✔	–	–	–	✔	–	–	–	–	–
	18 months	–	–	–	–	–	✔	–	–	–	✔	–	–	–	–	–
	24 months	–	–	–	–	–	✔	–	–	–	✔	–	–	–	–	–
Hold head erect ≥3 seconds, unsupported	Baseline	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
	12 months	–	–	–	✔*	✔	✔	–	–	✔*	✔	✔	✔	–	✔	–
	18 months	–	–	–	✔*	✔	✔	✔*	–	✔*	✔	✔	✔	✔*	✔	✔*
	24 months	–	–	–	✔*	✔	✔	✔*	–	✔*	✔	✔	✔	✔*	✔	✔
	24 months (central review)	–	–	–	✔	✔	✔	✔	–	✔	✔	✔	✔	✔	✔	✔
Sits without support ≥5 seconds	Baseline	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
	12 months	–	–	–	–	–	✔	–	–	✔	✔	–	✔	–	–	–
	18 months	–	–	–	–	✔	✔	✔	–	✔	✔	–	✔	✔	✔	✔
	24 months	–	–	–	–	✔	✔	✔	–	✔	✔	–	✔	✔	✔	✔
	24 months (central review)	–	–	–	✔	✔	✔	✔	–	✔	✔	✔	✔	✔	✔	✔
Sits without support ≥30 seconds	Baseline	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
	12 months	–	–	–	–	–	✔	–	–	✔	✔	–	✔	–	–	–
	18 months	–	–	–	–	✔	✔	–	–	✔	✔	–	✔	–	✔	–
	24 months	–	–	–	–	✔	✔	–	–	✔	✔	–	✔	–	✔	–
	24 months (central review)	–	–	–	–	✔	✔	–	–	✔	✔	✔	✔	✔	✔	✔
Stands with assistance	Baseline	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
	12 months	–	–	–	–	–	✔	–	–	–	✔	–	–	–	–	–
	18 months	–	–	–	–	–	✔	–	–	–	✔	–	–	–	–	–

[10)] Although 1 subject in Cohort 1 transiently met the definition of permanent ventilatory support due to salivary hypersecretion when 22 months had passed since Zolgensma infusion, the subject required <16 hours of respiratory assistance per day at Month 24. Thus, the subject was considered not to meet the definition of permanent ventilatory support.

Cohort		1			2											
Subject Number		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Sex		F	F	M	F	M	F	F	F	F	M	M	F	F	M	M
Age at symptom onset (months)		3	1	1	3	1	1	1	2	3	0	1	2	0	2	1
Age at infusion (months)		5.9	5.9	7.2	5.6	4.2	1.9	3.6	7.9	4.9	0.9	2.3	2.6	0.9	4.1	2.1
	24 months	−	−	−	−	−	✔	−	−	−	✔	−	−	−	−	−
	24 months (central review)	−	−	−	−	−	✔	−	−	−	✔	−	−	−	−	−
Stands alone	Baseline	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
	12 months	−	−	−	−	−	✔	−	−	−	✔	−	−	−	−	−
	18 months	−	−	−	−	−	✔	−	−	−	✔	−	−	−	−	−
	24 months	−	−	−	−	−	✔	−	−	−	✔	−	−	−	−	−
	24 months (central review)	−	−	−	−	−	✔	−	−	−	✔	−	−	−	−	−
Walks with assistance	Baseline	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
	12 months	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
	18 months	−	−	−	−	−	✔	−	−	−	✔	−	−	−	−	−
	24 months	−	−	−	−	−	✔	−	−	−	✔	−	−	−	−	−
	24 months (central review)	−	−	−	−	−	✔	−	−	−	✔	−	−	−	−	−
Walks alone	Baseline	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
	12 months	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
	18 months	−	−	−	−	−	✔	−	−	−	✔	−	−	−	−	−
	24 months	−	−	−	−	−	✔	−	−	−	✔	−	−	−	−	−
	24 months (central review)	−	−	−	−	−	✔	−	−	−	✔	−	−	−	−	−

✔: Achieved, −: Not achieved

* Achievement of head control was determined based on achievement of milestones of rolling over or sitting.

The CHOP-INTEND[11)] scores (the maximum score is 64) are shown in Table 19. Although it has been reported that patients with SMA Type I never reach a CHOP-INTEND score of 40 (*Neurology*. 2014; 83: 810-7), 11 subjects (91.7%) in Cohort 2 reached a CHOP-INTEND score of ≥40. No one in Cohort 1 reached a score of ≥40.

11) A 16-item (spontaneous upper and lower extremity movements, head control, etc.) scale to assess motor function, in which higher scores indicate better motor function (total score ranges from 0 to 64)

Table 19. CHOP-INTEND scores (Efficacy analysis set)

Cohort		1			2											
Subject Number		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Sex		F	F	M	F	M	F	F	F	F	M	M	F	F	M	M
Age at symptom onset (months)		3	1	1	3	1	1	1	2	3	0	1	2	0	2	1
Age at infusion (months)		5.9	5.9	7.2	5.6	4.2	1.9	3.6	7.9	4.9	0.9	2.3	2.6	0.9	4.1	2.1
CHOP-INTEND total score*	Baseline	27	6	16	29	29	47	25	12	34	50	16	35	14	30	17
	12 months	31	8	－	51	50	64	－	－	55	－	54	60	46	52	50
	18 months	36	－	－	－	58	－	58	－	－	－	－	53	50	60	55
	24 months	－	－	－	－	58	－	54	－	57	－	－	－	54	56	54
	Last assessment	37	15	20	55	58	64	54	16	57	64	56	55	54	56	54

－: Not assessed

*: In the case of a missed assessment at a given visit, the assessment closest to the scheduled visit date is given for Months 12, 18, and 24, and a total score at the last time point where all scores were obtained among the data from Study CL-101 collected by the cutoff date (including unscheduled visits) is given for the last assessment.

The gross and fine motor, language, and cognition subtests of Bayley Scales of Infant and Toddler Development were administered in patients who achieved a score ≥60 of 64 on the CHOP-INTEND (4 patients [Subject Numbers 6, 10, 12, and 14]), because the CHOP-INTEND was unlikely to be able to assess their motor function precisely. The subtest results are shown below.

- The mean gross motor score (a maximum score of 72) (min.-max.) increased from 13.0 (5-17) at the initial assessment to 36.3 (17-54) at the last assessment.
- The mean fine motor score (a maximum score of 66) (min.-max.) increased from 18.8 (4-36) at the initial assessment to 40.3 (36-43) at the last assessment.
- The mean receptive language score (a maximum score of 49) (min.-max.) increased from 18.3 (14-24) at the initial assessment to 26.8 (22-32) at the last assessment. The mean expressive language score (a maximum score of 48) (min.-max.) increased from 19.8 (16-29) at the initial assessment to 29.8 (29-31) at the last assessment.
- The mean cognition score (a maximum score of 91) (min.-max.) increased from 44.3 (37-52) at the initial assessment to 62.5 (58-67) at the last assessment.

The proportion of patients who were independent of ventilatory support (defined as requiring no daily ventilator support/usage in the absence of acute reversible illness and excluding perioperative ventilation) at baseline was 10 of 15 patients (all in Cohort 2). Of the 10 patients, 7 remained free of ventilatory support at Month 24.

The proportion of patients not requiring enteral feeding prior to therapy who maintained the ability to thrive (defined as the ability to tolerate thin liquids, demonstrated through a formal swallowing test, not receiving nutrition through mechanical support [i.e., feeding tube], and maintaining weight >3rd percentile for age and gender) at baseline was 5 of 7 patients. Of the 7 patients, 5 maintained the ability to thrive at Month 24 (all in Cohort 2).

The CMAP results are shown in Table 20.

Table 20. CMAP results (Efficacy analysis set)

Cohort		1			2											
Subject Number		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Sex		F	F	M	F	M	F	F	F	F	M	M	F	F	M	M
Age at symptom onset (months)		3	1	1	3	1	1	1	2	3	0	1	2	0	2	1
Age at infusion (months)		5.9	5.9	7.2	5.6	4.2	1.9	3.6	7.9	4.9	0.9	2.3	2.6	0.9	4.1	2.1
Tibialis anterior muscle (mV)	Baseline	0.2	NR	0.3	1.0	0.6	1.2	0.4	0.1	0.3	4.1	0.5	0.5	0.6	0.1	0.3
	1 month	0.6	－	0.4	1.1	0.3	0.8	1.0	0.1	0.8	3.6	0.7	0.5	0.5	0.6	0.5
	3 months	1.1	<0.1	0.3	1.2	0.6	3.0	0.6	0.1	1.0	5.2	0.9	0.6	－	1.2	0.3
	6 months	1.2	0.1	0.2	1.7	0.9	3.7	1.5	－	－	－	1.8	0.7	0.7	1.0	1.1
	9 months	1.2	0.1	0.1	1.7	1.0	4.9	1.7	0.2	2.1	5.6	2.4	1.7	1.1	1.9	1.3
	12 months	0.5	0.1	0.2	2.3	1.1	4.6	1.2	－	1.5	8.4	2.7	1.3	－	1.1	1.5
	15 months	0.9	0.2	－	－	0.4	4.9	1.7	<0.1	0.9	6.4	－	0.9	2.2	2.3	1.6
	18 months	0.9	－	0.1	1.8	1.3	6.3	－	NR	1.3	8.7	－	2.3	1.2	3.1	1.8
	24 months*	0.8	0.2	0.1	2.5	1.1	5.2	3.1	<0.1	2.3	8.3	－	2.0	2.6	3.6	2.5
Abductor digiti minimi muscle (mV)	Baseline	0.3	NR	NR	0.2	0.3	2.5	0.5	0.1	0.2	3.4	0.3	0.5	0.5	0.3	0.1
	1 month	0.3	－	NR	0.2	0.2	2.9	0.6	0.1	0.2	4.1	0.2	0.7	0.2	0.2	0.1
	3 months	0.1	<0.1	0.1	0.3	0.2	3.3	0.4	0.1	0.2	5.3	0.2	0.7	－	0.3	0.2
	6 months	0.2	<0.1	<0.1	0.4	0.3	5	0.7	<0.1	－	－	0.6	0.6	0.2	0.5	0.2
	9 months	0.2	<0.1	<0.1	0.5	0.3	4.6	0.5	<0.1	0.4	5.9	0.6	0.9	0.4	0.5	0.2
	12 months	0.2	<0.1	<0.1	0.6	0.3	6.1	0.6	－	0.9	6.1	0.6	1.1	－	0.4	0.3
	15 months	0.2	<0.1	－	－	0.4	5.5	0.8	<0.1	0.9	5.8	－	1.9	0.5	0.4	0.3
	18 months	0.2	－	NR	0.7	0.5	4.4	－	<0.1	1.2	5.4	－	1.0	0.6	0.5	0.2
	24 months*	0.2	0.1	0.1	0.9	0.5	6.9	0.8	0.1	1.4	5.2	－	0.6	0.5	0.7	0.2

NR: no response, －: Not measured
*: If repeated measurements were obtained, the last CMAP reading is given.

8.2 Reference data

8.2.1 Long-term follow-up study (CTD5.3.5.2-4, Study LT-001 [ongoing since September 2017 (data cutoff date of March 8, 2019)])

A follow-up study involving patients who participated in Study CL-101 and completed 2 years of follow-up after Zolgensma infusion is ongoing at 1 site overseas to evaluate the long-term safety and efficacy of Zolgensma (up to 15 years after Zolgensma infusion).

Of 15 subjects who completed Study CL-101, 13 subjects (3 from Cohort 1 of Study CL-101, 10 from Cohort 2 of Study CL-101) were enrolled the follow-up study. The remaining 2 subjects did not participate in Study LT-001 due to a geographical reason or their parents' decision (both in Cohort 2 of Study CL-101). Safety and efficacy analyses were performed. During the study, 7 subjects started treatment with nusinersen.

For safety analysis, reports on serious adverse events and adverse events requiring particular attention[12] only were collected. Adverse events reported are shown in Table 21. All of the events were serious, but those events were all considered unrelated to Zolgensma. No deaths were reported. In this study, severity grades of adverse events were not clarified

Table 21. Adverse events reported (Safety analysis set)

	Cohort 1 of Study CL-101 (N = 3)	Cohort 2 of Study CL-101 (N = 10)	Total (N = 13)
	All Grades	All Grades	All Grades
Any adverse event	1 (33.3)	5 (50.0)	6 (46.2)
Infections and infestations			
Pneumonia	1 (33.3)	2 (20.0)	3 (23.1)
Bronchitis	0	1 (10.0)	1 (7.7)
Gastroenteritis	0	1 (10.0)	1 (7.7)
Respiratory, thoracic and mediastinal disorders			
Acute respiratory failure	0	2 (20.0)	2 (15.4)
Respiratory distress	1 (33.3)	1 (10.0)	2 (15.4)
Respiratory failure	1 (33.3)	0	1 (7.7)
Metabolism and nutrition disorders			
Dehydration	0	2 (20.0)	2 (15.4)
Hypoglycaemia	0	1 (10.0)	1 (7.7)
Cardiac disorders			
Cardiac arrest	1 (33.3)	0	1 (7.7)

MedDRA ver.20.0
n (%)

Efficacy analysis was performed. The motor milestones achieved by 9 subjects from Cohort 2 of Study CL-101 [see Table 18] were sustained. Moreover, 2 subjects from Cohort 2 (Subject Numbers 4 and 7) gained the new milestone of sitting without support for ≥30 seconds, and 2 subjects from Cohort 2 (Subject Numbers 11 and 14) gained the new milestone of standing with assistance.

8.2.2 Foreign phase I study (CTD5.3.5.2-3, Study CL-102 [ongoing since December 2017 (data cutoff date of March 8, 2019])

An open-label, uncontrolled study is ongoing at 11 sites overseas to evaluate the safety and efficacy of Zolgensma administered intrathecally in patients with SMA Type II or III defined by bi-allelic deletion of the *SMN1* gene, and 3 copies of the *SMN2* gene; the onset of clinical symptoms before age of 12 months; ≥6 months and <60 months of age at the time of infusion; and negative for anti-AAV9 antibodies (target sample size, 27 subjects) (for key inclusion and exclusion criteria, see Table 22).

[12] Since only patients who completed 2 years of follow-up after Zolgensma infusion were to be enrolled in Study LT-001, adverse events observed in Study LT-001 were unlikely to be causally related to Zolgensma. It was considered that there is no need for collecting all adverse events. Thus, it was decided to collect the following events (1) to (6) only as safety information.
(1) Gene therapy-related events, (2) liver function enzyme elevations, (3) new occurrences of a malignancy, (4) new occurrences or exacerbations of neurologic disorder, (5) new occurrences or exacerbations of rheumatism or autoimmune disorder, and (6) new occurrences of hematologic disorder

Table 22. Key inclusion and exclusion criteria

Inclusion criteria	Patients meeting all of the following criteria ● Bi-allelic deletion of the *SMN1* gene, and 3 copies of the *SMN2* gene ● Absence of the c.859G>C substitution in *SMN2* ● Onset of clinical symptoms before age of 12 months ● Demonstrated the ability to sit unassisted for ≥10 seconds but had never been able to stand or walk ● ≥6 months and <60 months of age at the time of infusion
Exclusion criteria	Patients meeting any of the following criteria ● Use of invasive ventilatory support (tracheotomy with positive pressure) or SpO_2 <95% at screening ● Medical necessity for a gastric feeding tube, where the majority of feedings were given by non-oral methods ● Presence of anti-AAV9 antibody ● Clinically significant abnormal laboratory values (GGT >3×ULN of the laboratory range, bilirubin ≥3.0 mg/dL, creatinine ≥1.0 mg/dL, hemoglobin <8 or >18 g/dL, or white blood cell count >20,000/mm^3)

Dosage regimen or method of use was as follows: The doses of Zolgensma were 6.0×10^{13} vg/body for Cohort 1, 1.2×10^{14} vg/body for Cohort 2, and 2.4×10^{14} vg/body for Cohort 3, and Zolgensma was to be administered as a single, one-time intrathecal infusion over 60 to 120 seconds.

Thirty subjects who were enrolled in the study (3 in Cohort 1, 25 in Cohort 2, 2 in Cohort 3) (the mean age at infusion [range], 25.1 [7.0-54.5] months) received Zolgensma. All the subjects were included in the safety analysis set. Efficacy was assessed separately for patients ≥6 and <24 months of age at infusion (3 in Cohort 1 and 13 in Cohort 2) and patients ≥24 and <60 months of age at infusion (12 in Cohort 2). In the present application, 2 subjects in Cohort 3 were not assessed for efficacy because sufficient time had not elapsed since Zolgensma infusion.

Adverse events reported by ≥2 subjects are shown in Table 23. A causal relationship between Zolgensma and following adverse events could not be ruled out: hypertension (3 subjects), AST increased (2 subjects), lymphadenopathy (2 subjects), pyrexia (2 subjects), and sinus tachycardia (1 subject) in Cohort 2. Serious adverse events reported by ≥1 subject are shown in Table 24. A causal relationship between Zolgensma and the following adverse events could not be ruled out: ALT increased and AST increased (1 subject each) in Cohort 2. No deaths were reported.

Table 23. Adverse events reported by ≥2 subjects in any cohort (Safety analysis set)

	Cohort 1 (N = 3)		Cohort 2 (N = 25)		Cohort 3 (N = 2)	
	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4
Any adverse event	3 (100)	1 (33.3)	25 (100)	5 (20.0)	2 (100)	0
General disorders and administration site conditions						
Pyrexia	3 (100)	0	12 (48.0)	0	0	0
Infections and infestations						
Upper respiratory tract infection	2 (66.7)	0	13 (52.0)	0	1 (50.0)	0
Otitis media	1 (33.3)	0	3 (12.0)	0	0	0
Pneumonia	1 (33.3)	0	2 (8.0)	1 (4.0)	0	0
Nasopharyngitis	0	0	4 (16.0)	0	0	0
Ear infection	0	0	2 (8.0)	1 (4.0)	0	0
Respiratory, thoracic and mediastinal disorders						
Nasal congestion	1 (33.3)	0	3 (12.0)	0	0	0
Cough	0	0	8 (32.0)	0	0	0
Rhinorrhoea	0	0	3 (12.0)	0	0	0
Upper respiratory tract congestion	0	0	3 (12.0)	0	0	0
Sleep apnoea syndrome	0	0	2 (8.0)	1 (4.0)	0	0
Skin and subcutaneous tissue disorders						
Rash	1 (33.3)	0	4 (16.0)	0	0	0
Dermatitis diaper	1 (33.3)	0	2 (8.0)	0	0	0
Erythema	0	0	2 (8.0)	0	0	0
Musculoskeletal and connective tissue disorders						
Scoliosis	0	0	3 (12.0)	0	0	0
Extremity contracture	0	0	2 (8.0)	0	0	0
Pain in extremity	0	0	2 (8.0)	0	0	0
Tendinous contracture	0	0	2 (8.0)	0	0	0
Gastrointestinal disorders						
Vomiting	0	0	9 (36.0)	0	1 (50.0)	0
Constipation	0	0	3 (12.0)	0	0	0
Investigations						
Blood ALP increased	1 (33.3)	1 (33.3)	2 (8.0)	2 (8.0)	0	0
AST increased	0	0	2 (8.0)	0	0	0
Cardiac disorders						
Sinus tachycardia	0	0	2 (8.0)	0	0	0
Vascular disorders						
Hypertension	0	0	3 (12.0)	0	0	0
Injury, poisoning and procedural complications						
Contusion	0	0	2 (8.0)	0	0	0
Skin abrasion	0	0	2 (8.0)	0	0	0
Blood and lymphatic system disorders						
Lymphadenopathy	0	0	3 (12.0)	0	0	0
Metabolism and nutrition disorders						
Weight gain poor	0	0	2 (8.0)	0	0	0

MedDRA ver.20.1, Severity grade based on CTCAE v4.03
n (%)

Table 24. Serious adverse events (Safety analysis set)

	Cohort 1 (N = 3)	Cohort 2 (N = 25)	Cohort 3 (N = 2)
Investigations			
Blood ALP increased	1 (33.3)	0	0
ALT increased	0	1 (4.0)	0
AST increased	0	1 (4.0)	0
Respiratory, thoracic and mediastinal disorders			
Respiratory failure	1 (33.3)	0	0
Infections and infestations			
Influenza	0	1 (4.0)	0
Pneumonia	0	1 (4.0)	0
Respiratory syncytial virus infection	0	1 (4.0)	0

MedDRA ver.20.1
n (%)

Table 25 shows the efficacy endpoints.

Table 25. Efficacy endpoints

Primary efficacy endpoint	[Patients ≥6 and <24 months of age at the time of infusion] ● The proportion of patients who achieved the ability to stand without support for ≥3 seconds [Patients ≥24 and <60 months of age at the time of infusion] ● Change from baseline in the Hammersmith Functional Motor Scale-Expanded (HFMSE) score
Secondary efficacy endpoint	The proportion of patients who achieved the ability to walk independently for ≥5 steps

The efficacy results as of the cutoff date of March 8, 2019 are shown in Table 26.

Table 26. Number of patients who achieved each motor milestone

Motor milestone	Patients ≥6 and <24 months of age				Patients ≥24 and <60 months of age	
	Cohort 1 (N = 3)		Cohort 2 (N = 13)		Cohort 2 (N = 12)	
	Baseline	After Zolgensma infusion	Baseline	After Zolgensma infusion	Baseline	After Zolgensma infusion
Stands without support ≥3 seconds	0	1	0	1	0	0
Holds head erect ≥3 seconds, unsupported	3	3	13	13	12	12
Rolls over	3	3	9	13	10	11
Sits without support ≥30 seconds	3	3	10	13	12	12
Stands with assistance ≥2 seconds	1	1	0	2	1	3
Crawls on hands and knees	0	2	1	2	3	3
Pulls to stand	0	1	0	1	1	1
Walks with assistance	1	1	0	1	0	1
Walks alone	0	0	0	1	0	0

8.2.3 Foreign phase III study (CTD5.3.5.2-1, Study CL-303 [ongoing since October 2017 (data cutoff date of March 8, 2019])

An open-label, uncontrolled study is ongoing at 16 sites overseas to evaluate the efficacy and safety of Zolgensma in patients with SMA Type I or genetically diagnosed SMA Type I, defined by bi-allelic mutations (deletion or point mutation) in the *SMN1* gene, and 1 or 2 copies of the *SMN2* gene; <6 months of age at the time of infusion; and negative for anti-AAV9 antibodies (target sample size, 15 subjects) (for key inclusion and exclusion criteria, see Table 27).

Table 27. Key inclusion and exclusion criteria

Inclusion criteria	Patients meeting all of the following criteria ● Bi-allelic mutations (deletion or point mutation) in the *SMN1* gene, and 1 or 2 copies of the *SMN2* gene (inclusive of c.859G>C) ● <6 months of age at the time of infusion ● Able to undergo swallowing evaluation test prior to infusion
Exclusion criteria	Patients meeting any of the following criteria ● SpO_2 <96% (for altitudes >1,000 m, SpO_2 <92%) while awake or asleep without any supplemental oxygen or respiratory support ● Tracheostomy ● Current use or requirement of non-invasive ventilatory support averaging ≥6 hours/day over the 7 days prior to the screening visit; or ≥6 hours/day on average during the screening period ● Requiring ventilatory support while awake over the 7 days prior to screening or at any point during the screening period ● Weight-for-age below the 3rd percentile based on WHO Child Growth Standards (*Acta paediatrica*. 2006; 95 :76-85) ● Presence of anti-AAV9 antibody ● Clinically significant abnormal laboratory values (GGT, AST, and ALT >3 × ULN of the laboratory range, bilirubin ≥3.0 mg/dL, creatinine ≥1.0 mg/dL, hemoglobin <8 or >18 g/dL, or white blood cell count >20,000/mm^3)

Dosage regimen or method of use was as follows: Zolgensma 1.1×10^{14} vg/kg was to be administered as a single, one-time intravenous infusion over approximately 30 to 60 minutes.

All of 22 subjects enrolled in the study (the mean age at infusion [range], 3.7 (0.5-5.9) months) received Zolgensma. All of them were included in the efficacy and safety analysis sets.

The following 2 co-primary endpoints were selected:

- The proportion of patients who achieved functional independent sitting for ≥30 seconds at 18 months of age.
- The proportion of patients who survived without permanent ventilatory support (defined by tracheostomy or ≥16 hours of respiratory assistance per day continuously for ≥14 days in the absence of an acute reversible illness, excluding perioperative ventilation) at 14 months of age.

The efficacy results as of the data cutoff date of March 8, 2019 are shown below.

- Eleven of 22 subjects were able to sit without support for ≥30 seconds.
- Twenty-one of 22 subjects were alive, free of permanent ventilatory support.

Safety analysis was performed. Adverse events reported by ≥3 subjects are shown in Table 28. A causal relationship between Zolgensma and the following events could not be ruled out: AST increased (6 subjects), ALT increased (5 subjects), blood CK-MB increased (1 subject), vomiting (1 subject), and diarrhoea (1 subject).

Serious adverse events reported by ≥2 subjects are shown in Table 29. The possibility of a causal relationship between Zolgensma and all those events could not be ruled out.

Table 28. Adverse events reported by ≥3 subjects (Safety analysis set)

	N = 22	
	All Grades	Grade 3 or 4
Any adverse event	22 (10.0)	10 (45.5)
Gastrointestinal disorders		
Vomiting	7 (31.8)	0
Constipation	6 (27.3)	0
Teething	5 (22.7)	0
Diarrhoea	4 (18.2)	0
Gastrooesophageal reflux disease	4 (18.2)	0
Infections and infestations		
Upper respiratory tract infection	10 (45.5)	1 (4.5)
Respiratory syncytial virus bronchiolitis	3 (13.6)	3 (13.6)
General disorders and administration site conditions		
Pyrexia	12 (54.5)	0
Investigations		
AST increased	6 (27.3)	2 (9.1)
ALT increased	5 (22.7)	1 (4.5)
Blood CK-MB increased	3 (13.6)	0
Respiratory, thoracic and mediastinal disorders		
Cough	6 (27.3)	0
Respiratory distress	3 (13.6)	0
Sleep apnoea syndrome	3 (13.6)	1 (4.5)
Skin and subcutaneous tissue disorders		
Rash	5 (22.7)	0
Musculoskeletal and connective tissue disorders		
Scoliosis	7 (31.8)	0

MedDRA ver.20.1, Severity grade based on CTCAE v4.03
n (%)

Table 29. Serious adverse events reported by ≥2 subjects (Safety analysis set)

	N = 22
Infections and infestations	
Bronchiolitis	2 (9.1)
Pneumonia	2 (9.1)
Respiratory syncytial virus bronchiolitis	2 (9.1)
Respiratory, thoracic and mediastinal disorders	
Respiratory distress	2 (9.1)

MedDRA ver.20.1
n (%)

One patient died (respiratory arrest [1 subject]). The patient was a 7-month-old girl. The patient had respiratory failure on Day 138 and once recovered, but died due to respiratory arrest, attributed to progression of SMA on Day 171. Its causal relationship to Zolgensma was denied.

8.2.4 Global phase III study (CTD5.3.5.2-2, Study CL-304 [ongoing since April 2018 (data cutoff date of March 8, 2019)])

An open-label, uncontrolled study is ongoing at 29 sites overseas and in Japan to evaluate the efficacy and safety of Zolgensma in pre-symptomatic patients with genetically diagnosed SMA Type I, II, or III[13)] defined

13) Since Zolgensma-related histopathological changes in the heart were observed in a single-dose toxicity study in neonatal mice (CTD 4.2.3.1-6), it was decided to suspend enrollment of pre-symptomatic SMA Type III patients with 4 copies of the *SMN2* gene, until Zolgensma safety information in humans is accumulated (Protocol Version 2.0 [as of [redacted], 20[redacted]]). On the other hand, pre-symptomatic SMA Type I or II patients with 2 or 3 copies of the *SMN2* gene are predicted to develop more severe disease than SMA Type III patients. Enrollment of this patient population was continued because the protocol was amended to include more rigorous cardiac safety monitoring.

by bi-allelic mutations (deletion or point mutation) in the *SMN1* gene, and 2, 3, or 4 copies of the *SMN2* gene; and ≤6 weeks of age at the time of infusion (target sample size, ≥44 subjects [Type I, ≥15; Type II, ≥12; and Type III, ≥17]) (for key inclusion and exclusion criteria, see Table 30).

Table 30. Key inclusion and exclusion criteria

Inclusion criteria	Patients meeting all of the following criteria ● Pre-symptomatic patients with SMA Type I with 2 copies of the *SMN2* gene, SMA Type II with 3 copies of the *SMN2* gene, or SMA Type III with 4 copies of the *SMN2* gene (inclusive of c.859G>C) ● ≤6 weeks of age at the time of infusion ● Ability to tolerate thin liquids as demonstrated through a formal swallowing test prior to infusion ● CMAP ≥2 mV at baseline
Exclusion criteria	Patients meeting any of the following criteria ● Body weight at screening <2 kg ● SpO_2 <96% (for altitudes >1,000 m, SpO_2 <92%) while awake or asleep without any supplemental oxygen or respiratory support ● Any clinical signs or symptoms at screening or immediately prior to dosing that are strongly suggestive of SMA (tongue fasciculations, hypotonia, areflexia, etc.) ● Tracheostomy or current prophylactic use or requirement of noninvasive ventilatory support at any time and for any duration prior to screening or during the screening period ● Weight-for-age below the 3rd percentile based on WHO Child Growth Standards (*Acta paediatrica*. 2006; 95 :76-85) ● Presence of anti-AAV9 antibody ● Clinically significant abnormal laboratory values

Dosage regimen or method of use was as follows: Zolgensma 1.1×10^{14} vg/kg was to be administered as a single, one-time intravenous infusion in all of Cohort 1 (2 copies of the *SMN2* gene), Cohort 2 (3 copies of the *SMN2* gene), and Cohort 3 (4 copies of the *SMN2* gene).

The efficacy results obtained from 8 subjects in Cohort 1 and 9 subjects in Cohort 2 as of the data cutoff date of March 8, 2019 (the range of follow-up, 1-8.7 months in Cohort 1 and 12 days to 4.7 months in Cohort 2) are shown below.

- All subjects enrolled in Cohorts 1 and 2 were alive, free of permanent ventilatory support.
- The motor milestone achievement was as follows: 4 of 8 subjects in Cohort 1 achieved the ability to sit without support for ≥30 seconds, and 3 of 8 subjects in Cohort 1 achieved the ability to sit without support for ≥10 seconds. In Cohort 2, 4 of 9 subjects achieved head control.

Adverse events reported by ≥2 subjects are shown in Table 31. A causal relationship between Zolgensma and the following adverse events was could not be ruled out: vomiting (2 subjects) in Cohort 1; and liver function test abnormal (2 subjects), gastrooesophageal reflux disease (2 subjects), and vomiting (1 subject) in Cohort 2.

Serious adverse events reported were croup infectious (1 subject) and hypercalcaemia (1 subject) in Cohort 1 and lethargy (1 subject) in Cohort 2, but all those events were considered unrelated to Zolgensma. No adverse events leading to death were reported.

Table 31. Adverse events reported by ≥2 subjects in any cohort (Safety analysis set)

	Cohort 1 (N = 8)		Cohort 2 (N = 9)		Cohort 3 (N = 1)	
	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4
Any adverse event	6 (75.0)	2 (25.0)	6 (66.7)	1 (11.1)	1 (100)	0
Gastrointestinal disorders						
Constipation	2 (25.0)	0	1 (11.1)	0	0	0
Vomiting	2 (25.0)	0	1 (11.1)	0	0	0
Gastrooesophageal reflux disease	0	0	2 (22.2)	0	0	0
Infections and infestations						
Upper respiratory tract infection	2 (25.0)	0	1 (11.1)	0	0	0
Investigations						
Liver function test abnormal	0	0	2 (22.2)	0	0	0

MedDRA ver.21.0, Severity grade based on CTCAE v4.03
n (%)

8.R Outline of the review conducted by PMDA

8.R.1 Use of foreign clinical study data and review strategy

PMDA asked the applicant to explain the appropriateness of evaluating the efficacy and safety of Zolgensma in Japanese patients based on the data from Study CL-101 in non-Japanese patients submitted as evaluation data and the data from Study LT-001 (a long-term follow-up study of CL-101) after examining intrinsic and extrinsic ethnic factors that may affect the evaluation of the efficacy and safety of Zolgensma.

The applicant's response:

The data from Study CL-101 in non-Japanese patients can be extrapolated to the Japanese population because there are no clear differences in the genetic cause, diagnosis and classification, clinical symptoms, or treatment of SMA between Japanese and non-Japanese populations. This is supported by the following evidence:

- There are no differences in variations in *SMN1* deletion (the genetic cause of SMA) between Japanese and non-Japanese populations: Homozygous deletions of *SMN1* (absence of exon 7 only, or absence of both exons 7 and 8) have been reported to account for approximately 95% of SMA cases both in and outside Japan (*Neuromuscul Disord*. 2018; 28: 103-15, *Brain Dev.* 2014; 36: 914-20).
- There are no differences in SMA diagnostic method between Japanese and non-Japanese populations: When SMA is suspected from clinical symptoms, clinical course, family history, etc., the diagnosis of SMA is made by genetic testing both in and outside Japan (*Neuromuscul Disord*. 2018; 28: 103-15, Clinical Practice Manual for Spinal Muscular Atrophy 2012, Editorial Committee of Clinical Practice Manual for SMA ed.).
- There are no differences in SMA classification between Japanese and non-Japanese populations: SMA is classified into Types I to IV on the basis of age of onset and maximum motor function achieved, and this practice is common in and outside Japan (*Arch Neurol*. 2011; 68: 979-84, *Brain Dev*. 2014; 36: 914-20).
- There are no differences in the major clinical symptoms of SMA: Clinically, both Japanese and non-Japanese patients with SMA present with hypotonia and muscle weakness (*Neuromuscul Disord*. 2018; 28: 103-15, Clinical Practice Manual for Spinal Muscular Atrophy 2012, Editorial Committee of Clinical Practice Manual for SMA ed.).

- There are no differences in treatment of SMA between Japanese and non-Japanese populations: Nusinersen is the only treatment option for SMA both in and outside Japan.
- A high transduction efficiency of the target tissue (CNS) is considered to be achieved by a single, one-time intravenous infusion of Zolgensma, and delivered Zolgensma expresses the SMN protein in the target cells. Thus, there should be no racial or ethnic differences in the onset of therapeutic effects or degradation of the SMN protein.

PMDA accepted the applicant's explanation, and decided to focus its review on Studies CL-101 and LT-001, which are the pivotal clinical studies to evaluate the efficacy and safety of Zolgensma among the clinical studies submitted. Since no Japanese patients were enrolled in Study CL-101 or LT-001, PMDA decided to also review the latest results from Study CL-304 that enrolled Japanese patients, in order to evaluate the efficacy and safety of Zolgensma in Japanese patients.

8.R.2 Clinical positioning of Zolgensma

The applicant's explanation about the clinical positioning of Zolgensma for the treatment of SMA patients:
SMA is caused by the deficiency of the SMN protein necessary for the survival of spinal motor neurons (*Neurol Clin*. 2015; 33: 831-46 etc.). The *SMN1* gene and the *SMN2* gene encode the SMN protein in humans. Since only approximately 10% to 15% of the protein product of *SMN2* is in the form of the full-length, functional SMN protein, patients with bi-allelic mutations in the *SMN1* gene develop SMA due to reduced levels of full-length functional SMN protein (*Arch Neurol.* 2011; 68: 979-84, *Neurol Clin*. 2015; 33: 831-46, etc.).

The available therapy for SMA is nusinersen. Nusinersen is considered to act by modulating the splicing of the *SMN2* gene and thus increasing the production of the SMN protein. On the other hand, Zolgensma is a gene therapy product containing a transgene encoding the SMN protein. The gene therapy is considered to have efficacy in the treatment of SMA by increasing the expression of the SMN protein through a different mechanism from nusinersen. Zolgensma will offer a new treatment option for SMA.

Treatment guidelines have yet to clearly show the positioning of Zolgensma or nusinersen in the treatment algorithm for SMA. However, taking account of different routes of administration (Zolgensma is administered intravenously, and nusinersen is administered intrathecally), presence of anti-AAV9 antibody (if positive, instead of Zolgensma, nusinersen is recommended), or differences in safety profile, the treating physician will choose the optimal therapy, according to the clinical condition of individual patients.

PMDA accepted the applicant's explanation.

8.R.3 Efficacy

The applicant's explanation:
Studies CL-101 and LT-001 support the promising efficacy of Zolgensma because the data from the studies have demonstrated the following:

- Data analyses for “the time from birth to either permanent ventilatory support or death” (the primary efficacy endpoint of Study CL-101) showed that Zolgensma is expected to increase event-free survival compared with the natural history of SMA Type I.
- Improvements in motor milestone achievement such as the CHOP-INTEND score were seen in most patients in Cohort 2 of Study CL-101, though no improvements are expected in the natural history cohort of SMA Type I.
- Motor milestones achieved during Study CL-101 were sustained even at >3 years after Zolgensma infusion, according to data from Study LT-001.
- In Study CL-101, 3 subjects in Cohort 1 and 1 subject in Cohort 2 (Subject Number 8) did not achieve motor milestones, but were alive without permanent ventilation at Month 24. Thus, the efficacy of Zolgensma has been demonstrated also in these 4 subjects.

PMDA requested the applicant to submit the latest results from Study LT-001.

The applicant’s response:
The results from Study LT-001 (13 subjects [3 from Cohort 1 and 10 from Cohort 2 of Study CL-101]) as of the latest data cutoff date (May 31, 2019) are shown below (the mean time since Zolgensma administration [min.-max.] was 4.1 years [3.5-5.1 years]). As of this data cutoff date, 7 of 13 subjects (3 of 3 from Cohort 1, 4 [Subject Numbers 5, 7, 12, and 13] of 10 from Cohort 2) had started treatment with nusinersen.

- Two (Subject Numbers 1 and 3) of 3 subjects from Cohort 1 and all of 10 subjects from Cohort 2 were alive and free of permanent ventilation. Among them, 5 (Subject Numbers 5, 6, 10, 12, and 14) of 10 subjects from Cohort 2 did not require ventilatory support of any kind.
- The motor milestones achieved during Study CL-101 had been maintained for all patients, and 2 patients (Subject Numbers 11 and 14) gained the new milestone of standing with assistance.

The applicant’s explanation about the efficacy of Zolgensma in Japanese patients:
As of November 7, 2019, 3 Japanese patients enrolled in Study CL-304 received Zolgensma. The details of information/data on the 3 patients, including the efficacy data (the data up to 9 months of age for the first and second patients, the data up to 55 days after Zolgensma infusion for the third patient) are described below. Given that, among others, all of the 3 patients achieved motor milestones (e.g., the first patient who achieved the ability to sit without support for 10 seconds), the results are expected to support the efficacy of Zolgensma also in Japanese patients.

- The first patient was a girl with 3 copies of the *SMN2* gene who received Zolgensma at the age of 21 days. The patient weighed 3.5 kg at the time of infusion, and did not present with clinical symptoms of SMA. The patient achieved the motor milestone of “head control without support for ≥3 seconds” on Day 27 and the motor milestones of “rolling over” and “sitting without support for ≥10 seconds” at the 9 months of age study visit. The fine motor subtest scores (a range from 0 to 66) of the Bayley Scales of Infant and Toddler Development were 3 at screening and 21 at 9 months of age, and the gross motor subtest scores (a range from 0 to 72) were 5 at screening and 26 at 9 months of age.

- The second patient was a girl with 2 copies of the *SMN2* gene who received Zolgensma at the age of 17 days. The patient weighed 3.2 kg at the time of infusion, and did not present with clinical symptoms of SMA. The patient achieved the motor milestone of "head control without support for ≥3 seconds" on Day 28 and the motor milestone of "rolling over" at the 9 months of age study visit. The CHOP-INTEND total scores[14)] (a range from 0 to 64) were 49 at screening and 62 at 9 months of age. The fine motor subtest scores of the Bayley Scales of Infant and Toddler Development were 3 at screening and 21 at 9 months of age, and the gross motor subtest scores were 5 at screening and 24 at 9 months of age.
- The third patient was a boy with 3 copies of the *SMN2* gene who received Zolgensma at the age of 15 days. The patient weighed 4.1 kg at the time of infusion, and did not present with clinical symptoms of SMA. The patient achieved the motor milestone of "head control without support for ≥3 seconds" on Day 28. The fine motor subtest scores of the Bayley Scales of Infant and Toddler Development were 3 at screening and 6 on Day 55, and the gross motor subtest scores were 4 at screening and 8 on Day 55.

PMDA's view:
In Study CL-101, the motor milestones achieved with Zolgensma exceed the natural history of SMA Type I. The achieved motor milestones have been maintained up to the latest data cutoff date. The results support a certain level of efficacy of Zolgensma.

The latest results from Japanese patients in Study CL-304 are expected to support the efficacy of Zolgensma also in Japanese patients.

8.R.4 Safety

The adverse events reported in all clinical studies submitted in the present application (Studies CL-101, LT-001, CL-102, CL-303, and CL-304) are presented in Sections 8.1.1 to 8.2.4.

There were 2 deaths in clinical studies. The detailed clinical courses are described in Sections 8.2.3 and 8.R.4.4.

An analysis was performed to evaluate the safety of Zolgensma in Japanese patients. The details of safety results in 3 Japanese patients enrolled in Study CL-304 as of November 7, 2019 (the data from the first and second patients who were followed up until 9 months of age, the data from the third patient who was followed up to 59 days after Zolgensma infusion) are shown below.

- The first patient (a girl with 3 copies of the *SMN2* gene who received Zolgensma at the age of 21 days) had 4 adverse events (conjunctivitis [2 episodes], genital rash [1], dacryostenosis acquired [1]), but all of those events were considered unrelated to Zolgensma.
- The second patient (a girl with 2 copies of the *SMN2* gene who received Zolgensma at the age of 17 days) had 5 adverse events (influenza, constipation, rhinitis, upper respiratory tract infection, and nasopharyngitis [1 episode each]), but all of those events were considered unrelated to Zolgensma.

[14)] Measured only for patients with 2 copies of the *SMN2* gene.

- The third patient (a body with 3 copies of the *SMN2* gene who received Zolgensma at the age of 15 days) had 2 adverse events (pharyngitis and miliaria [1 episode each], of which pharyngitis required hospitalization and was classified as a serious adverse event), but those events were considered unrelated to Zolgensma.

PMDA's view:
Although the currently available information on the safety of Zolgensma is very limited, particular attention should be paid to the risk of hepatotoxicity, cardiotoxicity, and thrombocytopenia, which were reported in clinical studies of Zolgensma and may run a serious course. Only 3 Japanese patients received Zolgensma and were not followed-up for a sufficient period of time. However, PMDA concluded that no events requiring particular attention have been observed in the Japanese patients at present.

In the following sections, PMDA focuses its review on serious adverse events etc. reported in clinical studies and in the overseas post-marketing experience.

8.R.4.1 Hepatotoxicity

The applicant provided explanation about hepatotoxicity associated with Zolgensma which highlighted the following: (a) the incidence of hepatotoxicity in clinical studies and in the overseas post-marketing experience, (b) the mechanism of development of hepatotoxicity and prednisolone treatment to mitigate hepatotoxicity, (c) the dosing regimen of prednisolone and the criteria for tapering/discontinuation, and (d) liver function monitoring.

(a) The incidence of hepatotoxicity in clinical studies and in the overseas post-marketing experience
Events coded to the MedDRA SMQ "liver related investigations, signs and symptoms" were tabulated as hepatotoxicity.

The incidences of hepatotoxicity in each of the clinical studies submitted in the present application (data cutoff date of March 8, 2019) are shown in Table 32 to Table 34.

Table 32. Incidence of hepatotoxicity (Study CL-101)

Preferred term*	n (%)			
	Cohort 2 of Study CL-101 N = 12		Cohorts 1 and 2 of Study CL-101 N = 15	
	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4
Hepatotoxicity	3 (25.0)	1 (8.3)	4 (26.7)	2 (13.3)
Transaminases increased	3 (25.0)	1 (8.3)	4 (26.7)	2 (13.3)
AST increased	1 (8.3)	0	1 (6.7)	0

*: MedDRA ver.20.0, Severity grade based on CTCAE v4.03

The possibility that Zolgensma may be causally related to any adverse events of hepatotoxicity observed in Study CL-101 could not be ruled out.

Serious adverse events reported were transaminases increased (2 subjects) (1 each in Cohorts 1 and 2).

The median time to onset of hepatotoxicity (range) and the median duration (range) were 27 (9-64) days and 78 (19-216) days, respectively.

Table 33. Incidence of hepatotoxicity (Studies CL-303 and CL-304)

Preferred term*	n (%)					
	CL-303 N = 22		Cohort 1 of Study CL-304 N = 8		Cohort 2 of Study CL-304 N = 9	
	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4
Hepatotoxicity	7 (31.8)	4 (18.2)	1 (12.5)	0	3 (33.3)	0
AST increased	6 (27.3)	2 (9.1)	0	0	0	0
ALT increased	5 (22.7)	1 (4.5)	0	0	0	0
GGT increased	2 (9.1)	0	0	0	0	0
Transaminases increased	2 (9.1)	2 (9.1)	1 (12.5)	0	0	0
Ammonia increased	1 (4.5)	0	0	0	0	0
Liver function test abnormal	0	0	0	0	2 (22.2)	0
Hepatic enzyme abnormal	0	0	0	0	1 (11.1)	0

*: MedDRA ver.20.1 for Study CL-303, MedDRA ver.21.0 for Study CL-304, Severity grade based on CTCAE v4.03

The possibility that Zolgensma may be causally related to any adverse events of hepatotoxicity observed in Studies CL-303 and CL-304 could not be ruled out.

Serious adverse events reported were AST increased, ALT increased, and transaminases increased (1 subject each) in Study CL-303. No serious adverse events were reported in Study CL-304.

The median time to onset of hepatotoxicity (range) and the median duration (range) in Study CL-303 were 11 (6-163) days and 9 (1-131) days, respectively. The median time to onset of hepatotoxicity (range) and the median duration (range) in Study CL-304 were 7 days and 7 days, respectively, in Cohort 1 (1 subject) and 26.5 (7-70) days and 32.5 (8-46) days, respectively, in Cohort 2.

Table 34. Incidence of hepatotoxicity (Study CL-102)

Preferred term*	n (%)			
	Cohort 1 of Study CL-102 N = 3		Cohort 2 of Study CL-102 N = 25	
	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4
Hepatotoxicity	1 (33.3)	1 (33.3)	4 (16.0)	3 (12.0)
Blood ALP increased	1 (33.3)	1 (33.3)	2 (8.0)	2 (8.0)
AST increased	0	0	2 (8.0)	0
ALT increased	0	0	1 (4.0)	1 (4.0)
Hepatomegaly	0	0	1 (4.0)	0

*: MedDRA ver.20.1, Severity grade based on CTCAE v4.03

The possibility of a causal relationship between Zolgensma and the following events could not be ruled out: AST increased (2 subjects), ALT increased (1 subject), and hepatomegaly (1 subject) in Cohort 2 of Study CL-102.

Serious adverse events reported were blood ALP increased (1 subject) in Cohort 1 and AST increased (1 subject) and ALT increased (1 subject) in Cohort 2.

The median time to onset of hepatotoxicity (range) and the median duration (range) were 182 days and 36 days, respectively, in Cohort 1 (1 subject) and 84 (16-150) days and 33 (13-131) days, respectively, in Cohort 2.

No hepatotoxicity was reported in Cohort 3 of Study CL-102.

In any of the above studies, no Hy's law cases (Guidance for industry. Drug-Induced Liver Injury: Premarketing Clinical Evaluation. U.S. Department of Health and Human Services, Food and Drug Administration. July 2009) were identified as of the data cutoff date of March 8, 2019.

In addition, the details of patients with serious hepatotoxicity in Study CL-302 (an ongoing foreign phase III study to evaluate the efficacy and safety of Zolgensma 1.1×10^{14} vg/kg administered intravenously in SMA Type I patients with *SMN1* bi-allelic mutations and 1 or 2 copies of *SMN2*) and the ongoing US Managed Access Program (MAP) (a program to make Zolgensma available to patients with serious or life-threatening SMA in countries where Zolgensma has not yet received regulatory approval) are shown in Table 35.

Table 35. Listing of patients with serious hepatotoxicity

Study Identifier	Age (months)	Sex	Preferred term*	Grade	Time to onset (days)	Duration (days)	Causality to Zolgensma	Outcome
CL-302	3	F	Hypertransaminasaemia	2	42	65	Yes	Resolved
	5	M	Transaminases increased	3	27	—	Yes	Unresolved
MAP	8	M	Acute hepatic failure	Unknown	51	—	Yes	Improved
	Unknown	Unknown	AST increased	Unknown	Unknown	Unknown	No	Unknown

*: MedDRA ver.21.1, Severity grade based on CTCAE v4.03

(b) Mechanism of development of hepatotoxicity and prednisolone treatment to mitigate hepatotoxicity

AAV vector administration in humans has been reported to induce capsid-specific cytotoxic T-cells (*Nat Rev Drug Discov*. 2019; 18: 358-78, *Journal of Clinical and Experimental Medicine*. 2018; 265: 344-50). Cytotoxic T-cells are considered to cause damage to AAV-infected hepatocytes, resulting in hepatotoxicity. Published literature has reported that suppression of cytotoxic T-cells with corticosteroids is effective in mitigating the immune response to AAV, including hepatotoxicity (*Hum Gene Ther*. 2016; 27: 947-61).

In the first-in-human study of Zolgensma, i.e., Study CL-101, the protocol did not initially stipulate the use of corticosteroid to mitigate hepatoxicity. However, the first patient (Subject Number 1) experienced a serious adverse event of transaminases increased on Day 27, received prednisolone (up to 2.25 mg/kg) from Day 27 to Day 75, and recovered on Day 90.

Based on the above, administration of prednisolone was considered effective in reducing Zolgensma-associated hepatotoxicity, and prednisolone was to be administered to prevent hepatotoxicity in the second and subsequent

patients in Study CL-101.

(c) The dosing regimen of prednisolone and the criteria for tapering/discontinuation

The dosing regimen of prednisolone and monitoring of liver function tests (Table 36) will be included in the IMPORTANT PRECAUTIONS section of the package insert for Zolgensma.

Table 36. Summary of the package insert (draft)
concerning prednisolone dosing regimen and monitoring of liver function tests

Dosing regimen of prednisolone	i) Administer prednisolone at 1 mg/kg/day 1 day prior to Zolgensma infusion, and then continue the same dose for 30 days after Zolgensma infusion. ii) If ALT and AST values are ≤2 × ULN and other liver function tests are normal at the end of the 30-day period of prednisolone treatment, taper the prednisolone dose over the next ≥4 weeks. If liver function abnormalities persist, continue prednisolone at 1 mg/kg/day until ALT and AST values are ≤2 × ULN and other liver function tests return to normal, and then taper the prednisolone dose over the next ≥4 weeks.
Monitoring of live function tests	Assess liver function (clinical symptoms, AST, ALT, total bilirubin, prothrombin time) prior to Zolgensma infusion. Continue to monitor liver function for 3 months after Zolgensma infusion (weekly for the first month, and then every other week for the second and third months), and continue prednisolone until AST and ALT values are ≤2 × ULN and other liver function test abnormalities return to normal.

(i) The dosing rationale for prednisolone (1 day prior to Zolgensma infusion and for 30 days after Zolgensma infusion)

Prednisolone was to be initiated with the following regimen in the second and subsequent patients in Study CL-101.

[Dosing regimen of prednisolone used in Study CL-101]

- Prednisolone should be administered at approximately 1 mg/kg/day for approximately 30 days, starting at 1 day prior to Zolgensma infusion.

Prednisolone was administered in accordance with the dosing regimen of prednisolone used in Study CL-101. As of the data cutoff date of June 29, 2018, of 25 patients who received the proposed intravenous dose of Zolgensma (not including Subject Number 1 in Study CL-101), 7 (28.0%) had hepatotoxicity for which a causal relationship to Zolgensma could not be ruled out. However, none of them presented with clinical symptoms or met Hy's law criteria.

Later, despite prophylactic administration of prednisolone in accordance with the dosing regimen of prednisolone used in Study CL-101, 1 case of acute hepatic failure (an 8-month-old boy listed in Table 35) was reported in the US MAP. Taking account of this case, the dosing regimen of prednisolone was changed in the ongoing phase III studies of Zolgensma (Studies CL-304 and CL-306[15)]) in order to more effectively dampen the immune response through a short course of strong immunosuppression during the early phase of treatment with Zolgensma. The modified dosing regimen of prednisolone is as follows:

15) A global study to evaluate the efficacy and safety of a single intravenous dose of Zolgensma 1.1×10^{14} vg/kg in patients with SMA Type I

[Dosing regimen of prednisolone used in Studies CL-304 and CL-306]

- Prednisolone should be administered at approximately 2 mg/kg/day at 1 day prior to Zolgensma infusion and for 2 days post-infusion (for a total of 3 days) and continued Zolgensma at approximately 1 mg/kg/day for 30 days starting on the third day post-infusion.

By [redacted] 20[redacted], 4 subjects (3 in Study CL-304, 1 in Study CL-306) received prednisolone in accordance with the dosing regimen of prednisolone used in Studies CL-304 and CL-306. Among these 4 subjects, 3 subjects with data available (in Study CL-304) experienced no significant infections, and their hepatotoxicity was manageable.

However, the recommended dosing regimen of prednisolone in the present application is the dosing regimen of prednisolone used in Study CL-101, for the following reasons:

- Many patients have received prednisolone in accordance with the dosing regimen of prednisolone used in Study CL-101, and hepatotoxicity is considered well manageable with this regimen.
- At present, there is limited clinical experience with the dosing regimen of prednisolone used in Studies CL-304 and CL-306.
- Three Japanese patients enrolled in Study CL-304 received prednisolone in accordance with the dosing regimen of prednisolone used in Study CL-101, and no hepatic dysfunction has been reported as of the data cutoff date of September 2, 2019.

(ii) The criteria for tapering prednisolone and the procedures for tapering/discontinuation

In Study CL-101, prednisolone was to be administered for 30 days after Zolgensma infusion, and was to be tapered and discontinued if AST and ALT values were below 120 IU/L.[16)] However, Studies CL-303 and CL-304 employed the following criteria/procedures: if AST and ALT values are ≤2×ULN, the dose of prednisolone are to be tapered over ≥4 weeks (at 0.5 mg/kg/day for the first 2 weeks and then at 0.25 mg/kg/day for another 2 weeks). The new prednisolone regimen was intended to reliably control Zolgensma-associated hepatotoxicity and avoid the long-term use of prednisolone for the treatment of relapsed or persistent hepatotoxicity wherever possible.

In Studies CL-303 and CL-304, 2 subjects had serious hepatotoxicity, which was managed in accordance with these criteria/procedures, and resolved. Information on the criteria/procedures for tapering prednisolone used in Studies CL-303 and CL-304 will be provided to healthcare professionals through the package insert and other materials, so that they can reliably control Zolgensma-associated hepatotoxicity in the post-marketing setting. Though not stipulated in the protocols of Studies CL-303 and CL-304, other liver function tests should be assessed before the start of tapering prednisolone. Thus, the package insert will advise physicians to taper prednisolone after ascertaining that other liver function test abnormalities also return to normal.

16) Although the protocol did not stipulate the use of prednisolone initially in Study CL-101, a cutoff of 120 IU/L was used for AST and ALT elevations under the Protocol Version 10 (as of [redacted], 20[redacted]).

(d) Liver function monitoring

An analysis of data from clinical studies of Zolgensma (as of the data cutoff date of March 8, 2019) showed that 16 subjects experienced hepatotoxicity which was possibly related to Zolgensma (3 in Study CL-101, 7 in Study CL-303, 4 in Study CL-304, 2 in Study CL-102), and that most of the events (12 of the 16 subjects) occurred within 30 days, and none beyond 90 days. For this reason, it is necessary to monitor liver function on a regular basis within 90 days after Zolgensma infusion.

PMDA's view:

Since serious hepatotoxicity has been reported in clinical studies of Zolgensma, a close attention should be paid to the risk of hepatotoxicity in patients to whom Zolgensma is administered. Thus, the applicant should provide information on the incidence of hepatotoxicity, administration of prednisolone to mitigate hepatotoxicity, and liver function monitoring to healthcare professionals, using the package insert and other materials.

The dosing regimen of prednisolone and the criteria for tapering prednisolone are very important information for the management of adverse events, such as Zolgensma-associated hepatotoxicity and infections associated with prednisolone. This information should be included in the PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE section, instead of the IMPORTANT PRECAUTIONS section, of the package insert [see Section 8.R.6]. Furthermore, information on the optimal dosing regimen of prednisolone should be collected also after marketing of Zolgensma. Any new information should be appropriately communicated to healthcare professionals in clinical practice as soon as possible.

8.R.4.2 Cardiotoxicity

The applicant provided explanation about cardiotoxicity associated with Zolgensma which highlighted the following: (a) the incidence of cardiotoxicity in clinical studies and in the overseas post-marketing experience, (b) the mechanism of development of cardiotoxicity, and (c) the need for monitoring creatine kinase, MB form (CK-MB) and cardiac troponin-I.

(a) The incidence of cardiotoxicity in clinical studies and in the overseas post-marketing experience

The incidences of cardiotoxicity in each of the clinical studies submitted in the present application (data cutoff date of March 8, 2019) are shown in Table 37 to Table 39.

Events coded to the MedDRA SOC "cardiac disorders" and MedDRA HLGTs "cardiac and vascular investigations (excl enzyme tests)," "enzyme investigations NEC," and "musculoskeletal and soft tissue investigations (excl enzyme tests)" were tabulated as cardiotoxicity.

Table 37. Incidence of cardiotoxicity (Study CL-101)

Preferred term*	n (%)	
	Cohort 2 of Study CL-101 N = 12	
	All Grades	Grade 3 or 4
Cardiotoxicity	3 (25.0)	1 (8.3)
Tachycardia	2 (16.7)	1 (8.3)
Bradycardia	1 (8.3)	0
Ventricular hypertrophy	1 (8.3)	0

*: MedDRA ver.20.0, Severity grade based on CTCAE v4.03

No cardiotoxicity was reported in Cohort 1 of Study CL-101. In Cohort 2 of Study CL-101, all events listed in Table 37 were considered unrelated to Zolgensma.

A serious adverse event of tachycardia occurred in 1 subject.

The median time to onset of cardiotoxicity (range) and the median duration (range) were 229.5 (29-661) days and 8.5 (2-169) days, respectively.

Table 38. Incidence of cardiotoxicity (Studies CL-303 and CL-304)

Preferred term*	n (%)					
	CL-303 N = 22		Cohort 1 of Study CL-304 N = 8		Cohort 2 of Study CL-304 N = 9	
	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4
Cardiotoxicity	3 (13.6)	0	1 (12.5)	0	1 (11.1)	0
Tachycardia	2 (9.1)	0	0	0	0	0
Cyanosis	1 (4.5)	0	0	0	0	0
Blood CK-MB increased	3 (13.6)	0	0	0	1 (11.1)	0
Blood pressure diastolic decreased	1 (4.5)	0	0	0	0	0
Blood pressure systolic increased	1 (4.5)	0	0	0	0	0
Cardiac murmur	1 (4.5)	0	0	0	0	0
Blood CPK increased	0	0	1 (12.5)	0	0	0
Troponin increased	0	0	0	0	1 (11.1)	0

*: MedDRA ver.20.1 for Study CL-303, MedDRA ver.21.0 for Study CL-304, Severity grade based on CTCAE v4.03

The events reported in Studies CL-303 and CL-304 are shown in Table 38. The possibility of a causal relationship between Zolgensma and the following events could not be ruled out: blood CK-MB increased (1 subject) and blood pressure diastolic decreased (1 subject) in Study CL-303, and blood CK increased (1 subject) in Cohort 1 and blood CK-MB increased (1 subject) and troponin increased (1 subject) in Cohort 2 in Study CL-304.

A serious adverse event of cyanosis occurred in 1 subject in Study CL-303. No serious adverse events were reported in Study CL-304.

The median time to onset of cardiotoxicity (range) and the median duration (range) in Study CL-303 were 174 (1-322) days and 64 (1-341) days, respectively. The median time to onset of cardiotoxicity (range) and the

median duration (range) in Study CL-304 were 14 days and 22 days, respectively, in Cohort 1 (1 subject) and 10.5 (8-13) days and 17 (9-25) days, respectively, in Cohort 2.

Table 39. Incidence of cardiotoxicity (Study CL-102)

Preferred term*	n (%)			
	Cohort 1 of Study CL-102 N = 3		Cohort 2 of Study CL-102 N = 25	
	All Grades	Grade 3 or 4	All Grades	Grade 3 or 4
Cardiotoxicity	1 (33.3)	0	4 (16.0)	0
Sinus tachycardia	0	0	2 (8.0)	0
Tachycardia	1 (33.3)	0	1 (4.0)	0
Mitral valve incompetence	0	0	1 (4.0)	0
Blood CK-MB increased	0	0	1 (4.0)	0
Blood pressure diastolic increased	0	0	1 (4.0)	0
Cardiac murmur	0	0	1 (4.0)	0
Electrocardiogram QT prolonged	0	0	1 (4.0)	0

*: MedDRA ver.20.1, Severity grade based on CTCAE v4.03

The events reported in Study CL-102 are listed in Table 39. The possibility of a causal relationship between Zolgensma and the following events could not be ruled out: sinus tachycardia, blood CK-MB increased, and cardiac murmur (1 subject each) in Cohort 2.

No serious adverse events were reported.

The median time to onset of cardiotoxicity (range) and the median duration (range) were 62 days and 5 days, respectively, in Cohort 1 (1 subject) and 38.5 (1-182) days and 20.5 (1-131) days, respectively, in Cohort 2.

The cases of serious cardiotoxicity reported in other clinical studies and the ongoing overseas MAP are presented in Table 40.

Table 40. Listing of patients with serious cardiotoxicity

Study Identifier	Age (months)	Sex	Preferred term*	Grade	Time to onset (days)	Duration (days)	Causality to Zolgensma	Outcome
MAP	7	F	Tachycardia	Unknown	0	—	Yes	Improved
	4	M	Tachycardia	Unknown	0	2	Yes	Resolved
	Unknown	Unknown	Blood CK increased	Unknown	Unknown	—	No	Unknown

*: MedDRA ver.21.1, Severity grade based on CTCAE v4.03

(b) Mechanism of development of cardiotoxicity

The definitive mechanism of development of cardiotoxicity is unknown. However, given that Zolgensma was distributed in the heart of mice and that changes such as inflammation and edema in the myocardium were observed in a single-dose toxicity study in neonatal mice [see Section 6.1], cardiotoxicity is possibly related to the immune response to AAV9 capsid and/or transgene expression.

(c) The need for monitoring CK-MB and cardiac troponin-I

In Study CL-101, CK-MB and cardiac troponin-I were measured as cardiac markers, and the results and related information are shown below.

- All 15 subjects had elevated CK-MB levels at baseline and during the study period, none of which were considered clinically significant by the investigator. Although there are no reports on CK-MB changes in SMA patients, CK-MB in skeletal muscles may change in patients with diseases affecting skeletal muscles such as SMA, and especially, elevated CK-MB levels were observed in children under 3 months of age (*Clin Biochem*. 1999; 32: 77-80). Thus, elevated CK-MB is unlikely to be related to Zolgensma.
- Of the 15 subjects, 8 (53.3%) had elevated cardiac troponin-I levels (the pre-defined criterion for potential clinical significance in the protocol: >0.05 µg/L). Of the 8 subjects, 2 had elevated cardiac troponin-I levels prior to administration of Zolgensma. None of the elevations in cardiac troponin-I were considered clinically significant by the investigator. According to a study in healthy neonates (N = 869), the upper reference limit for cardiac troponin-I has been reported to be 0.183 µg/L (*Clin Biochem*. 2004; 37: 1079-82). No patients had cardiac troponin-I of >0.183 µg/L in Study CL-101.

However, given the results from non-clinical studies etc., attention should be paid to the risk of cardiotoxicity, and monitoring of cardiac troponin-I for the first 3 months after Zolgensma infusion is needed for the following reasons:

- CK-MB is considered to change depending on the underlying disease (i.e., SMA) and is therefore unlikely to be suitable for monitoring for cardiotoxicity, whereas cardiac troponin-I is considered a specific marker for myocardial injury.
- In Study CL-101, cardiac troponin-I increased for the first 2 months after Zolgensma infusion and returned towards baseline at ≥3 months post-infusion.

PMDA's view:

Cardiotoxicity was observed in clinical trials of Zolgensma and other studies. Attention should therefore be paid to the risk of cardiotoxicity following Zolgensma infusion. Thus, the applicant should appropriately provide information on the incidence of cardiotoxicity, the need for monitoring cardiac troponin-I, etc., to healthcare professionals in clinical practice, using the package insert and other materials.

8.R.4.3 Thrombocytopenia

The applicant's explanation about thrombocytopenia associated with Zolgensma:

No thrombocytopenia was reported in Studies CL-101, LT-001, CL-304, and CL-102 and MAP.

In Study CL-303, thrombocytopenia occurred in 3 of 22 subjects (13.6%), which included thrombocytopenia in 2 subjects (9.1%) and platelet count decreased in 1 subject (4.5%). All those events were of Grade ≤2.

The median time to onset of thrombocytopenia (range) and the median duration (range) were 10 (7-63) days and 13 (8-42) days, respectively.

Events coded to the MedDRA SMQ “thrombocytopenia” were tabulated as thrombocytopenia.

PMDA’s view:
Because thrombocytopenia was reported in Study CL-303, attention should be paid to the risk of thrombocytopenia following Zolgensma infusion. Thus, the applicant should appropriately provide information on the incidence of thrombocytopenia etc. to healthcare professionals in clinical practice, using the package insert and other materials.

8.R.4.4 Leukoencephalopathy

In a foreign phase III study (Study CL-302), 1 subject had leukoencephalopathy, respiratory distress, transaminases increased, thrombocytopenia, and bronchiolitis, and died. The clinical course is described below.

The patient was a 5-month-old boy with SMA Type I. The patient had bronchiolitis 13 days after Zolgensma infusion, and then presented with respiratory distress twice (15-16 days and 17-53 days post-dose). Because tonic-clonic seizure occurred at 31 days post-dose, electroencephalography was performed, which revealed findings suggestive of encephalitis. An MRI scan of the brain at 32 days post-dose showed leukoencephalopathy, which was considered to be caused by electrolyte imbalance (plasma sodium concentration at 33 days post-dose, 157 mmol/L). Then, epilepsy-like symptoms persisted and leukoencephalopathy did not resolve. The patient died at 53 days post-dose.

An autopsy was performed, and the brain findings (an excerpt) are shown below. The possibility of a causal relationship of leukoencephalopathy to Zolgensma was ruled out.

- There were extensive lesions in the brain, and especially, the findings in the cerebral cortex were consistent with hypoxic/ischemic damage. Unusual features (e.g., cerebellar vacuolation and granulation pattern) were also observed.
- Patients with SMA may also have brain lesions, which are different from the pattern noted in this patient.
- There were no findings specific to CNS infection. Although the possibility of infection cannot be ruled out, the morphological pattern did not suggest the possibility of infection.
- The pattern noted in the brain of this patient was not typical of those caused by a poison or drug.

PMDA’s view:
Leukoencephalopathy occurred in 1 subject in Study CL-302. The patient had respiratory distress and was in a very serious state. Although the relationship between Zolgensma and the risk of leukoencephalopathy is unknown, the applicant should provide information on the risk of leukoencephalopathy after Zolgensma infusion to healthcare professionals through information materials and other materials, and should collect post-marketing information on the risk of leukoencephalopathy.

8.R.5 Indication or performance

The proposed indication or performance was infantile spinal muscular atrophy, and no statements were included in the PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE section.

After the submission of the marketing application for Zolgensma, the applicant explained that they intend to include the following statements in the PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE section.

(1) Zolgensma is indicated for symptomatic or pre-symptomatic patients.

(2) Do not administer Zolgensma to patients with ≥4 copies of the *SMN2* gene.

Moreover, after the submission of the present application in Japan, Zolgensma was approved in the US on May 24, 2019. The applicant requested that the proposed statements in the INDICATION OR PERFORMANCE and PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE sections be amended as described below.

Indication or Performance

Spinal muscular atrophy

Precautions Concerning Indication or Performance

(1) Administer Zolgensma to patients with confirmed bi-allelic deletion or mutation in the *SMN1* gene.
(2) Administer Zolgensma to patients <2 years of age.
(3) Zolgensma is indicated for symptomatic or pre-symptomatic patients.
(4) The efficacy and safety of Zolgensma in patients with advanced SMA (e.g., permanent ventilator-dependence) have not been established. Prior to the use of Zolgensma in this patient population, closely monitor the patient's condition and weigh the risks and benefits of the therapy.

Based on the considerations in Sections 8.R.5.1 and 8.R.5.2, PMDA concluded that the following statements should be included in the INDICATION OR PERFORMANCE and PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE sections.

Indication or Performance

Treatment of patients with spinal muscular atrophy who have tested negative for elevated anti-AAV9 antibodies.

Precautions Concerning Indication or Performance

(1) Administer Zolgensma to patients with confirmed bi-allelic deletion or mutation in the *SMN1* gene.
(2) Administer Zolgensma to patients <2 years of age.
(3) Zolgensma is indicated for symptomatic or pre-symptomatic patients.

(4) The efficacy and safety of Zolgensma in patients with advanced SMA (e.g., permanent ventilator-dependence) have not been established. Prior to the use of Zolgensma in this patient population, weigh the risks and benefits of the therapy carefully.
(5) Administer Zolgensma to patients who tested negative for elevated anti-AAV9 antibodies. The approved *in vitro* diagnostic should be used for testing.

8.R.5.1 Intended population for Zolgensma

The applicant's explanation about the rationale of selecting "infantile spinal muscular atrophy" for the proposed indication or performance:
The statements in the INDICATION OR PERFORMANCE and PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE sections for Zolgensma were proposed based on the patient population of Study CL-101 that demonstrated the clinical usefulness of Zolgensma.

However, for the following reasons, the applicant considered that (a) the indication or performance of Zolgensma should be "infantile spinal muscular atrophy," instead of the patient population of Study CL-101 ("symptomatic SMA Type I"), and Zolgensma should be indicated for symptomatic or pre-symptomatic patients; (b) Zolgensma should be indicated for patients with the c.859G>C substitution in the *SMN2* gene as well, and (c) Zolgensma should not be used in patients with ≥4 copies of the *SMN2* gene.

(a) The indication or performance of Zolgensma should be "infantile spinal muscular atrophy," instead of the patient population of Study CL-101 ("symptomatic SMA Type I"), and Zolgensma should be indicated for symptomatic or pre-symptomatic patients:
- Generally, a physician makes the diagnosis of SMA before the onset of clinical symptoms by genetic testing of the *SMN1* and *SMN2* genes early after birth of a child who is suspected of having SMA based on family history etc. On the other hand, SMA is classified into 5 types (Types 0-IV) on the basis of age of onset (Type I, about 6 months of age) and severity (SMA Type I patients never attain the ability to sit without support). Despite the genetic diagnosis of SMA, the patient needs to be followed-up untreated until about 6 months of age because they have a definitive diagnosis of SMA Type I after the onset of clinical symptoms. However, since the progression of irreversible neurodegeneration and motor neuron loss begins before the onset of clinical symptoms in SMA patients, treatment should be initiated as early as possible. If the indication or performance of Zolgensma is "spinal muscular atrophy Type I," Zolgensma cannot be administered to pre-symptomatic patients, who will lose the opportunity of benefiting from treatment with Zolgensma in a timely manner.
- Zolgensma is a product that expresses the SMN protein, which is reduced in SMA patients, and Zolgensma is expected to be effective in all types of SMA.
- The diagnosis of SMA should be made by physicians with expert knowledge and experience. There may be some patients whose SMA type cannot be determined due to delay in visiting a hospital with such a SMA specialist, resulting in an unknown age of onset. These patients may lose the opportunity of benefiting from treatment with Zolgensma.

- Nusinersen is approved for the same patient population as Zolgensma, and the indication for nusinersen was “infantile spinal muscular atrophy.” Thus, the same indication or performance should be proposed also for Zolgensma.

(b) Zolgensma should be indicated for patients with the c.859G>C substitution in the *SMN2* gene as well:
- Study CL-101 did not include patients with c.859G>C predicting milder disease because the study was intended to evaluate the safety and efficacy of Zolgensma in a homogeneous patient population. Although patients with c.859G>C were allowed to be enrolled in Studies CL-302, CL-303, and CL-304, no such patients have been enrolled as of March 8, 2019. Thus, the efficacy and safety of Zolgensma in patients with c.859G>C are unknown. However, Zolgensma is a product that expresses the SMN protein, which is reduced in SMA patients, and the efficacy of Zolgensma is promising, regardless of the presence or absence of c.859G>C.

(c) Zolgensma should not be used in patients with ≥4 copies of the *SMN2* gene:
- There is no clinical experience with Zolgensma in patients with ≥4 copies of the *SMN2* gene, nor are any evidence to support the efficacy and safety of Zolgensma in these patients. The applicant therefore considered that Zolgensma cannot be recommended.

Then, Zolgensma was approved for the following “INDICATIONS AND USAGE” in the US. The applicant made a suggestion that the INDICATION OR PERFORMANCE and PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE sections of the Japanese package insert should follow the US labeling.

“INDICATION AND USAGE” in the US:
ZOLGENSMA (onasemnogene abeparvovec-xioi) is an adeno-associated virus vector-based gene therapy indicated for the treatment of pediatric patients less than 2 years of age with spinal muscular atrophy (SMA) with bi-allelic mutations in the *survival motor neuron 1* (*SMN1*) gene.
Limitation of Use
- The safety and effectiveness of repeat administration of ZOLGENSMA have not been evaluated.
- The use of ZOLGENSMA in patients with advanced SMA (e.g., complete paralysis of limbs, permanent ventilator-dependence) has not been evaluated.

In response to the above suggestion by the applicant, PMDA asked the applicant to explain the following changes [(d) to (g)] made to the INDICATION OR PERFORMANCE and PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE sections of the proposed package insert:

(d) Zolgensma should be administered to patients <2 years of age. The indication or performance is SMA, instead of infantile SMA. [change from (a)]

(e) A precautionary statement about the use of Zolgensma in patients with ≥4 copies of the *SMN2* gene is omitted. [change from (c)]

(f) Zolgensma should be administered to patients with confirmed bi-allelic deletion or mutation in the *SMN1* gene. (addition of statement)

(g) The efficacy and safety of Zolgensma in patients with advanced SMA (e.g., permanent ventilator-dependence) have not been established. (addition of statement)

The applicant's response:

(d) Zolgensma should be administered to patients <2 years of age. The indication or performance is SMA, instead of infantile SMA:

The details of regulatory review in the US

In the US, the proposed indication was as follow: "Zolgensma is indicated for the treatment of pediatric patients with spinal muscular atrophy (SMA) Type 1 with or without disease onset."

During the review in the US, FDA made a suggestion to modify the indication as follows: "Zolgensma is indicated for the treatment of infants with spinal muscular atrophy (SMA) with confirmed biallelic mutations in the *survival motor neuron 1* (*SMN1*) gene."

FDA's suggestion was based on the following points:

- Given that clinical studies of Zolgensma in patients <6 months of age at the time of infusion demonstrated the clinically meaningful efficacy of Zolgensma (e.g., prolonged survival without permanent ventilation) and its manageable safety profile, the balance of risks and benefits of Zolgensma was considered favorable for SMA patients <2 years of age who have a high unmet medical need in the US.
- External experts commented that a historical SMA type is not appropriate as the indication.

On the other hand, FDA regulations define infants as persons 29 days to less than 2 years of age, and the indication suggested by FDA would exclude patients <29 days of age from treatment. However, since a positive outcome likely depends on early treatment in SMA patients, exclusion of patients <29 days of age from treatment with Zolgensma was considered inappropriate. Therefore, the indication for the US suggested by FDA was modified so that Zolgensma can be used in patients <29 days of age.

The reasons for considering that the target population defined by the indication in the US is applicable to Japan

For the following reasons etc., the use of Zolgensma in patients <2 years of age is recommended also in Japan:

- The mean age of diagnosis in Japanese patients with SMA Type I has been reported to be about 20 months (*BIO Clinica*. 2018; 33: 780-3 etc.). If Zolgensma is indicated for patients <2 years of age, most SMA Type I patients will benefit from treatment with Zolgensma.
- The percent of the total increment of neural growth at the age of 2 years has been reported to be approximately 50% (Patterns of human growth. 2nd ed. Cambridge University Press; 1999. 41), and normal neural growth after treatment with Zolgensma is reliably expected in patients aged <2 years.

- As of November 25, 2019, 139 patients with SMA ≥6 months of age received Zolgensma in the US post-marketing experience, the ongoing US MAP, etc. (92 in the post-marketing experience [21 patients ≥6 and <12 months of age; 22 patients ≥12 and <18 months of age; 48 patients ≥18 and <24 months of age; 1 patient ≥24 months of age], 35 in the MAPs ongoing in the US and other countries[17] [6.1-30.3 months of age], 12 in the Early Access Request Programs[18] [a majority of patients with unknown age]). It should be noted that these data are not from clinical studies, with limited efficacy and safety information. However, the data showed improvements in CHOP-INTEND (29 points at baseline → 41 points at 4 weeks after Zolgensma infusion), indicating the promising efficacy of Zolgensma. Safety data indicated that there were no adverse events requiring particular attention compared with adverse events reported in patients <6 months of age.

(e) A precautionary statement about the use of Zolgensma in patients with ≥4 copies of the *SMN2* gene is omitted:

The details of regulatory review in the US

For the following reasons, the use of Zolgensma was not restricted by *SMN2* copy number in the US.

- Based on its mechanism of action, the efficacy of Zolgensma is promising, regardless of *SMN2* copy number.
- External experts commended as follows:
  Treatment should be initiated before irreversible motor neuron loss, regardless of *SMN2* copy number. Taking also into account that it is difficult to uniformly define SMA types or disease severity only by *SMN2* copy number, patients eligible for the therapy should not be determined based on *SMN2* copy number.

The reasons for considering that the target population defined by the indication in the US is applicable to Japan

The findings suggestive of cardiotoxicity of Zolgensma were observed in non-clinical studies, and information on cardiotoxicity in humans was also limited. This resulted in the exclusion of SMA patients with ≥4 copies of the *SMN2* gene who have a milder disease from clinical studies. However, given that no clinically significant cardiotoxicity has been reported in clinical studies, the expected therapeutic benefits may outweigh the possible risks in some patients with ≥4 copies of the *SMN2* gene.

Moreover, given that the efficacy of Zolgensma is promising, regardless of *SMN2* copy number, there is no need for restricting its use by *SMN2* copy number in Japan.

(f) Zolgensma should be administered to patients with confirmed bi-allelic deletion or mutation in the *SMN1* gene:

Zolgensma is a gene replacement therapy, which makes up for the missing or nonworking *SMN1* in

17) Available in the US, South America, Canada, and the Asia Pacific regions where Zolgensma has not yet received regulatory approval, excluding Japan.
18) Available in Europe and the Middle East where Zolgensma has not yet received regulatory approval.

patients with *SMN1* deletion or mutation resulting in reduced levels of the SMN protein expression. There are no differences in the cause of SMA (bi-allelic deletion or mutation in the *SMN1* gene) between Japanese and non-Japanese populations. It is desirable also in Japan that Zolgensma be administered only to patients with confirmed bi-allelic deletion or mutation in the *SMN1* gene. Thus, this statement has been added.

(g) The efficacy and safety of Zolgensma in patients with advanced SMA (e.g., permanent ventilator-dependence) have not been established:

The details of regulatory review in the US

Since Zolgensma is indicated for patients <2 years of age in the US, the use of Zolgensma in patients with advanced SMA is likely to be within the scope of the indication. However, no patients with advanced SMA have been treated with Zolgensma in clinical studies, and adequate efficacy may not be obtained. Thus, "The use of ZOLGENSMA in patients with advanced SMA (e.g., complete paralysis of limbs, permanent ventilator-dependence) has not been evaluated." has been added as "Limitation of Use."

The reasons for considering that the target population defined by the indication in the US is applicable to Japan

Because the details of regulatory review in the US is relevant to Japan, the relevant precautionary statement should be included also in the Japanese package insert. Since the phrase "complete paralysis of limbs" in the US labeling does not appear in the package insert for nusinersen (the package insert for nusinersen states that the efficacy and safety of nusinersen in patients who required permanent ventilation have not been established.), the phrase "e.g., permanent ventilator-dependence" will be used also in the package insert for Zolgensma. The package insert will advise the following: "The efficacy and safety of Zolgensma in patients with advanced SMA (e.g., permanent ventilator-dependence) have not been established. Prior to the use of Zolgensma in these patients, closely monitor the patient's condition and weigh the risks and benefits the therapy."

Based on the above considerations presented in the above (b) and (d) to (g), the INDICATION OR PERFORMANCE and PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE sections of the package insert for Zolgensma have been proposed in accordance with the US labeling.

Furthermore, baseline anti-AAV9 antibody titers were measured by an ELISA in clinical studies, and patients positive for anti-AAV9 antibodies (those with anti-AAV9 antibody titer of >1:50) were not allowed to participate in the studies [see Section 8.1.1]. There is no clinical experience with Zolgensma in patients positive for anti-AAV9 antibodies. However, given the following points, the use of Zolgensma in patients positive for anti-AAV9 antibodies is considered acceptable if the physician considers that the potential therapeutic benefits outweigh the possible risks. Once Zolgensma is marketed, the anti-AAV9 antibody assay method used in clinical studies will be transferred to healthcare professionals so that anti-AAV9 antibody titers will be measured using this assay method to decide whether each patient is eligible for the use of Zolgensma.

- In Cohort 2 of Study CL-101, 2 subjects (Subject Numbers 12 and 13) had higher baseline antibody titers (1:50) than other subjects (≤1:25). These 2 subjects achieved motor milestones. This result and other data seemed to support the efficacy of Zolgensma. Safety data were also analyzed. Serious adverse events reported by these 2 subjects were gastroenteritis and pneumonia in Subject Number 12 and pneumonia aspiration in Subject Number 13, and all of these events were considered unrelated to Zolgensma. Thus, there is no clear impact of anti-AAV9 antibody titers on the efficacy and safety of Zolgensma. Considering the seriousness of SMA, the use of Zolgensma should be acceptable also in anti-AAV9 antibody-positive patients who have limited treatment option.

PMDA's view:
The applicant's explanations in the above subsections (b), (e), (f), and (g) are acceptable.

In the subsection (d), the scientific rationale for administering Zolgensma to patients <2 years of age is weak. However, selecting patients <2 years of age as the target population in Japan is acceptable, considering the following points:

- SMA is a congenital disease with a very poor survival prognosis, and there are limited existing therapies. In addition, given that the US post-marketing data etc. support the efficacy of Zolgensma in patients >6 months of age as well, offering the opportunity of benefiting from treatment with Zolgensma to patients older than the patient population of Study CL-101 (0.9-7.9 months of age) is clinically meaningful.
- However, Study CL-101 indicated that higher efficacy would be achieved by earlier treatment with Zolgensma. Given that the upper limit of age for enrollment was changed from 9 to 6 months during the study and that there are also safety concerns about Zolgensma, such as serious hepatotoxicity, a certain upper age limit is necessary for the therapy. The severity of symptoms varies depending of the SMA type and most patients with SMA Type I with the worst prognosis have a definitive diagnosis by the age of 2 years. Thus, these patients will have the opportunity of benefiting from treatment with Zolgensma if the patients <2 years of age are eligible for Zolgensma therapy.
- If Zolgensma is indicated only for the use in patients ≤6 months of age (the patient population of Study CL-101), the applicant will have to conduct a clinical study in Japanese patients aged ≥6 months and <2 years and file a partial change application for approval of the use of Zolgensma in Japanese patients <2 years of age. However, because of the very limited number of patients with SMA, it is extremely difficult to conduct a Japanese clinical study. Further, patients aged ≥6 months and <2 years will lose the opportunity of benefiting from treatment with Zolgensma.
- If the target population in Japan is harmonized with that in the US, a clinical study to evaluate the efficacy and safety of Zolgensma in patients ≥2 years of age will be conducted as a global study. This would contribute to earlier clinical development of Zolgensma for patients who are not eligible for the therapy so far.

The use of Zolgensma in patients positive for anti-AAV9 antibodies is not acceptable because (i) the applicant's discussion is not based on the results from a study in patients positive for anti-AAV9 antibodies, (ii) Zolgensma

is a non-replicating recombinant AAV having an AAV9 capsid, which may induce adverse events such as hypersensitivity and (iii) Zolgensma may not be effective in the treatment of this patient population. Thus, the INDICATION OR PERFORMANCE and PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE sections should clearly state that prior to the use of Zolgensma, anti-AAV9 antibody titers should be measured using the approved *in vitro* diagnostic and that Zolgensma should be used in patients tested negative for elevated anti-AAV9 antibodies.

Based on the above, the following statements should be included in the INDICATION OR PERFORMANCE and PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE sections of the package insert.

Indication or Performance

Treatment of patients with spinal muscular atrophy who have tested negative for anti-AAV9 antibodies

Precautions Concerning Indication or Performance

(1) Administer Zolgensma to patients with confirmed bi-allelic deletion or mutation in the *SMN1* gene.
(2) Administer Zolgensma to patients <2 years of age.
(3) Zolgensma is indicated for symptomatic or pre-symptomatic patients.
(4) The efficacy and safety of Zolgensma in patients with advanced SMA (e.g., permanent ventilator-dependence) have not been established. Prior to the use of Zolgensma in these patients, weigh the risks and benefits of the therapy carefully.
(5) Administer Zolgensma to patients who tested negative for anti-AAV9 antibodies. The approved *in vitro* diagnostic should be used for testing.

8.R.5.2 Use of nusinersen before or after Zolgensma infusion

PMDA asked the applicant to explain the safety of nusinersen in patients previously treated with Zolgensma and the safety of Zolgensma in patients previously treated with nusinersen.

The applicant's explanation:

Use of nusinersen in patients previously treated with Zolgensma

In Study LT-001, whether to start treatment with nusinersen after Zolgensma infusion was decided, in consultation with the investigator, upon request of the patient's family or caregiver. Actually, of 13 patients, 7 (3 [Subject Numbers 1, 2, and 3] from Cohort 1 of Study CL-101, 4 [Subject Numbers 5, 7, 12, and 13] from Cohort 2 of Study CL-101) started treatment with nusinersen.

None of the patients had worsening of clinical symptoms or lost motor milestones after start of treatment with nusinersen. As of the data cutoff date of March 8, 2019, of the 7 patients treated with nusinersen, 3 (1 [Subject Number 2] from Cohort 1 of Study CL-101, 2 [Subject Numbers 5 and 7] from Cohort 2 of Study CL-101) had serious adverse events (Table 41).

Table 41. Listing of patients previously treated with Zolgensma who had serious adverse events after administration of nusinersen (Study LT-001)

Subject Number	Age (months)	Sex	Time from Zolgensma infusion to start of treatment with nusinersen (days)	Complications	Serious adverse events reported	Time from start of treatment with nusinersen to onset of serious adverse event (days)	Grade	Causality to Zolgensma
2	3	F	1,358	Salivary hypersecretion, Bilevel positive airway pressure, Gastrostomy, Oesophagogastric fundoplasty	Respiratory failure	67	4	No
					Cardiac arrest	68	4	No
					Respiratory distress	118	3	No
					Pneumonia	123	3	No
					Respiratory distress	170	4	No
5	3	M	990	Constipation, Ear infection, Hypoglycaemia, Skin discolouration	Dehydration	146	3	No
					Hypoglycaemia	175	3	No
7	2	F	952	Constipation, Gastrooesophageal reflux disease, Hiatus hernia, Eczema, Livedo reticularis	Gastroenteritis	26	3	No
					Acute respiratory failure	398	3	No

*: MedDRA 20.1, Severity grade based on CTCAE v4.03

All serious adverse events were related to the patient's complications or SMA, and there was also no consistent trend in the time to onset. As of September 30, 2019, no new serious adverse events have been reported by these 7 patients.

As described above, there was no trend towards increased risk of adverse events after administration of nusinersen in patients previously treated with Zolgensma. Physicians should decide whether to administer nusinersen to patients previously treated with Zolgensma, according to the individual patient's condition.

Use of Zolgensma in patients previously treated with nusinersen

Based on safety information regarding the use of Zolgensma in patients previously treated with nusinersen in the US MAP and post-marketing experience, the cases were examined. As of September 30, 2019, 12 patients (6 patients received nusinersen, only before Zolgensma infusion, 6 patients received nusinersen before and after Zolgensma infusion) had serious adverse events.

The details of patients with serious adverse events after Zolgensma infusion are shown in Table 42 and Table 43.

Table 42. Listing of patients treated with nusinersen only before Zolgensma infusion who had serious adverse events after Zolgensma infusion (MAP and US post-marketing experience)

Age (months)	Sex	Time from last dose of nusinersen to Zolgensma infusion (days)	Complications	Serious adverse events reported	Time from Zolgensma infusion to onset of serious adverse event (days)	Grade	Causality to Zolgensma
Unknown	M	6	Reflux gastritis	Clostridium difficile colitis	50	Unknown	No
9	F	20	Respiratory failure, Dysphagia	Parainfluenzae virus infection	7	Unknown	Yes
				Thrombocytopenia	7	3	Yes
				AST increased	21	3	Yes
11	F	118	Dysphagia, Respiratory failure, Bilevel positive airway pressure	Influenza	46	3	No
11	M	125	Brachycephaly, Constipation	Vomiting	1	3	Yes
				Feeding intolerance	1	3	Yes
17	M	Unknown	Gastrooesophageal reflux disease, Constipation	Transaminases increased	30	Unknown	Yes
15	F	36	None	Vomiting	1	Unknown	Yes
				Transaminases increased	7	Unknown	Yes
				Thrombocytopenia	7	Unknown	Yes

*: MedDRA 22.0, Severity grade based on CTCAE v4.03 for MAP and CTCAE v5.0 for US post-marketing experience

Table 43. Listing of patients treated with nusinersen before and after Zolgensma infusion who had serious adverse events after Zolgensma infusion (MAP and US post-marketing experience)

Age (months)	Sex	Time from nusinersen dose immediately before Zolgensma infusion to Zolgensma infusion (days)	Time from Zolgensma infusion to nusinersen dose immediately after Zolgensma infusion (days)	Complications	Serious adverse events reported	Time from Zolgensma infusion to onset of serious adverse event (days)	Grade	Causality to Zolgensma
7	F	Unknown	Unknown	Failure to thrive, Weight gain poor	Respiratory syncytial virus infection	33	Unknown	Unknown
					Influenza	45	Unknown	Unknown
6	M	Unknown	Unknown	Hypotonia	Acute hepatic failure	51	Unknown	Yes
					Laryngospasm	52	Unknown	No
					Bradycardia	52	Unknown	No
5	M	3	28	None	Pneumonia viral	72	Unknown	No
					Respiratory distress	72	3	No
11	F	Unknown	91	Acute respiratory failure, Renal disorder, Hypertension, Drug tolerance, Developmental hip dysplasia, Osteoporosis	Cardiac arrest	16	4	No
					Cardiac arrest	51	4	No
19	F	Unknown	22	Dysphagia, Gastrooesophageal reflux disease	Liver function test increased	42	3	Yes
2 years	F	Unknown	Unknown	Hypoxia	Cerebral atrophy	75	Unknown	No

*: MedDRA 22.0, Severity grade based on CTCAE v4.03 for MAP and CTCAE v5.0 for US post-marketing experience

As described above, there was no trend towards increased risk of adverse events after administration of Zolgensma in patients previously treated with nusinersen. Physicians should decide whether to administer Zolgensma to these patients, according to the individual patient's condition.

PMDA's view:

At present, there is no information indicating that administration of nusinersen before or after Zolgensma infusion increases the safety risk of Zolgensma or affects its efficacy. However, the currently available information is limited, and it is necessary to continue to collect post-marketing information on the effects of nusinersen on Zolgensma.

8.R.6 Dosage and Administration or Method of Use

The following statements were included in the DOSAGE AND ADMINISTRATION OR METHOD OF USE and PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE sections of the proposed package insert.

Dosage and Administration or Method of Use

The usual dose of Zolgensma for infants weighing between 2.6 and 8.5 kg is 1.1×10^{14} vector genomes (vg)/kg.

Zolgensma should be administered as a single, one-time intravenous infusion.

The dose volume is determined based on patient body weight as per the table below.

Patient Weight Range (kg)	Dose Volume (mL)[Note)]
2.6-3.0	16.5
3.1-3.5	19.3
3.6-4.0	22.0
4.1-4.5	24.8
4.6-5.0	27.5
5.1-5.5	30.3
5.6-6.0	33.0
6.1-6.5	35.8
6.6-7.0	38.5
7.1-7.5	41.3
7.6-8.0	44.0
8.1-8.5	46.8

Note) Dose volume is calculated using the upper limit of the patient weight range.

Precautions Concerning Dosage and Administration or Method of Use

Follow the steps below for infusion:

(1) Place a primary catheter into a peripheral vein in the arm or leg. Insertion of a back-up catheter is recommended.
(2) Prime tubing with saline prior to Zolgensma infusion.
(3) Administer Zolgensma as a slow infusion over approximately 60 minutes. Do not infuse as an intravenous push or bolus.
(4) Flush line with saline following completion of infusion.

Later, Zolgensma was approved in the US. The applicant made a suggestion that the DOSAGE AND ADMINISTRATION OR METHOD OF USE and PRECAUTIONS CONCERNING DOSAGE AND

ADMINISTRATION OR METHOD OF USE sections of the Japanese package insert should follow the US labeling.

Dosage and Administration or Method of Use

The usual dose of Zolgensma for patients weighing ≥2.6 kg (<2 years of age) is 1.1×10^{14} vector genomes (vg)/kg. Zolgensma should be administered as a single, one-time intravenous infusion.

The dose volume is determined based on patient body weight as per the table below.

Patient Weight Range (kg)	Dose Volume (mL)[Note)]
2.6-3.0	16.5
3.1-3.5	19.3
3.6-4.0	22.0
4.1-4.5	24.8
4.6-5.0	27.5
5.1-5.5	30.3
5.6-6.0	33.0
6.1-6.5	35.8
6.6-7.0	38.5
7.1-7.5	41.3
7.6-8.0	44.0
8.1-8.5	46.8
8.6-9.0	49.5
9.1-9.5	52.3
9.6-10.0	55.0
10.1-10.5	57.8
10.6-11.0	60.5
11.1-11.5	63.3
11.6-12.0	66.0
12.1-12.5	68.8
12.6-13.0	71.5
13.1-13.5	74.3

Note) Dose volume is calculated using the upper limit of the patient weight range.

The dose volume calculated by patient body weight should be adjusted also for patients <2 years of age weighing ≥13.6 kg.

Precautions Concerning Dosage and Administration or Method of Use

(1) Follow the steps below for infusion:

1) Place a primary catheter into a peripheral vein in the arm or leg. Insertion of a back-up catheter is recommended.
2) Prime tubing with saline prior to Zolgensma infusion.
3) Administer Zolgensma as a slow infusion over approximately 60 minutes. Do not infuse as an intravenous push or bolus.
4) Flush line with saline following completion of infusion.

(2) The safety and efficacy of repeat administration of Zolgensma have not been established.

The applicant's explanation about the basis for the proposed dosage and administration or method of use:
Since the clinical efficacy of Zolgensma was demonstrated in Cohort 2 of Study CL-101, the dosing regimen of Zolgensma was proposed based on Cohort 2 of Study CL-101.

For the following reasons, a weight range from 2.6 to 8.5 kg was proposed for the target population.

- Patients enrolled in Study CL-101 weighed up to 8.4 kg, and there is no clinical experience with Zolgensma in patients weighing ≥8.5 kg. Thus, an upper limit of 8.5 kg was chosen.
- In clinical studies of Zolgensma, the minimum patient weight at baseline was 3.6 kg in Study CL-101, 4.7 kg in Study CL-302, 3.9 kg in Study CL-303, and 3.0 kg in Study CL-304. There is no clinical experience with Zolgensma in patients weighing <3.0 kg. Because a small quantity of Zolgensma is administered to low-body-weight pediatric patients, dosing in such patients would raise no major safety concern. However, considering that Zolgensma should not be used in patients weighing ≤2.5 kg because low birth weight is defined as weight less than 2500 g in the US (*Vaccine*. 2016; 34: 6047-56), a lower limit of 2.6 kg was chosen.

PMDA asked the applicant to explain the reason for omitting the upper limit of patient weight (8.5 kg) in response to the approval of Zolgensma in the US.

The applicant's response:
In the course of regulatory review in the US, external experts recommended that the decision to provide gene therapy to heavier or older patients should be left to the clinical judgment of the treating physician, in consultation with the patient's family. For this reason, the upper limit of body weight (8.5 kg) was omitted from "the Dosage and Administration" in the US labeling.

The age of patients eligible for Zolgensma therapy has been determined to be <2 years of age [see Section 8.R.5.1], and if patients weighing ≤8.5 kg at the time of infusion are eligible for Zolgensma therapy, only a limited number of patients may be treated with Zolgensma, for the following reasons:

- According to the Infant Physical Development Survey (MHLW, 2010), in healthy infants, 8.5 kg corresponds to the 97th percentile of body weight (8.72 kg) for 4- to 5-month old boys or the 50th percentile of body weight (8.50 kg) for 8- to 9-month-old boys, and the 97th percentile of body weight (8.67 kg) for 5- to 6-month-old girls or the 50th percentile of body weight (8.51 kg) for 11- to 12-month-old girls.
- Publication overseas has reported that SMA patients are 0.12 kg lighter than healthy infants at 3 months of age, 0.97 kg lighter at 6 months of age, 1.17 kg lighter at 12 months of age, 1.46 kg lighter at 18 months of age, and 2.27 kg lighter at 24 months of age (*Ann Neurol*. 2017; 82: 883-91).

In addition, 44 patients weighing ≥8.5 kg (the maximum weight of 13.5 kg) received Zolgensma in the US post-marketing experience by September 25, 2019 and 9 patients weighing ≥8.5 kg (the maximum weight of 13.1 kg) received Zolgensma in the ongoing US MAP by September 30, 2019. As of October 31, 2019, no new adverse events requiring safety measures have been reported by these patients.

Based on the above, the dose volumes for patients weighing ≤13.5 kg were tabulated, referring to the Infant Physical Development Survey (MHLW; 2010) etc., in order to cover the body weight of SMA patients <2 years of age (the target population for Zolgensma). However, the table was not intended to restrict the use of Zolgensma to patients weighing ≤13.5 kg, and it was added that "The dose volume calculated based on patient body weight should be administered to patients <2 years of age weighing ≥13.6 kg."

Furthermore, the statements in the PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE section were proposed based on the written procedures for clinical product management, which specified the method of administration of Zolgensma for Study CL-101. However, in the course of regulatory review in the US, the FDA instructed the sponsor in the US to include the following 2 statements in the US labeling. Thus, the relevant statements will be included in the Japanese package insert as well.

- Place a primary catheter into a peripheral vein in the arm or leg. Insertion of a back-up catheter is recommended.
- Do not infuse as an intravenous push or bolus.

PMDA's view:

The applicant's explanation is largely acceptable. However, the following statements contain important information, and the statements should therefore be included in the DOSAGE AND ADMINISTRATION OR METHOD OF USE section, instead of the PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE section.

- Administer Zolgensma as a slow infusion over 60 minutes.
- Do not re-administer Zolgensma.

Moreover, since the dosing regimen of prednisolone used to manage hepatotoxicity is important, this information needs to be included in the PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE section. Because the first precautionary statement ("Place a primary catheter into a peripheral vein in the arm or leg. Insertion of a back-up catheter is recommended.") merely explains routine procedures for intravenous administration, there is little need for expressly stating it in the PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE section.

Based on the above, the following statements should be included in the DOSAGE AND ADMINISTRATION OR METHOD OF USE and PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE sections of the package insert.

Dosage and Administration or Method of Use

The usual dose of Zolgensma for patients weighing ≥2.6 kg (<2 years of age) is 1.1×10^{14} vector genomes (vg)/kg. Zolgensma should be administered as a single, one-time intravenous infusion over 60 minutes. Do not re-administer Zolgensma.

The dose volume is determined based on patient body weight as per the table below.

Patient Weight Range (kg)	Dose Volume (mL)[Note)]
2.6-3.0	16.5
3.1-3.5	19.3
3.6-4.0	22.0
4.1-4.5	24.8
4.6-5.0	27.5
5.1-5.5	30.3
5.6-6.0	33.0
6.1-6.5	35.8
6.6-7.0	38.5
7.1-7.5	41.3
7.6-8.0	44.0
8.1-8.5	46.8
8.6-9.0	49.5
9.1-9.5	52.3
9.6-10.0	55.0
10.1-10.5	57.8
10.6-11.0	60.5
11.1-11.5	63.3
11.6-12.0	66.0
12.1-12.5	68.8
12.6-13.0	71.5
13.1-13.5	74.3

Note) Dose volume is calculated using the upper limit of the patient weight range.

The dose volume also for patients <2 years of age weighing ≥13.6 kg is determined based on patient body weight.

Precautions Concerning Dosage and Administration or Method of Use

(1) Follow the steps below for infusion:
   1) Prime tubing with saline prior to Zolgensma infusion.
   2) Do not infuse as an intravenous push or bolus.
   3) Flush line with saline following completion of infusion.

(2) Since hepatotoxicity may occur following administration of Zolgensma, administer prednisolone as per the table below.

Table. Dosing regimen of prednisolone

Prednisolone should be administered at 1 mg/kg/day 24 hours prior to Zolgensma infusion and then continued for 30 days after Zolgensma infusion. If AST and ALT values are ≤2 × ULN at the end of the 30-day period of prednisolone treatment, taper the prednisolone dose over the next ≥4 weeks (0.5 mg/kg/day for the first 2 weeks and 0.25 mg/kg/day for the subsequent 2 weeks) to discontinue prednisolone. If AST and ALT values are >2 × ULN at the end of the 30-day period of prednisolone treatment, continue prednisolone at 1 mg/kg/day until AST and ALT values are ≤2 × ULN and other liver function tests return to normal range, and then taper the prednisolone dose over the next ≥4 weeks (0.5 mg/kg/day for the first 2 weeks and 0.25 mg/kg/day for the subsequent 2 weeks) to discontinue prednisolone.

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

The applicant's explanation about the post-marketing surveillance plan for Zolgensma:

The applicant is planning all-case post-marketing surveillance, covering all SMA patients treated with Zolgensma, to assess the safety etc. of Zolgensma in clinical practice. This surveillance will be conducted as part of a global registry to collect long-term observational data from patients with SMA in Japan, the US, Europe, and other nations (16 countries plan to participate in the registry at present) (Title: RESTORE [a planned recruitment period of 5 years from 2018 through 2023]).

In the post-marketing surveillance, all adverse events will be collected for 12 months after Zolgensma infusion and during treatment with SMA drugs including nusinersen. Also during other periods, serious adverse events including deaths, adverse events of special interest associated with Zolgensma (liver injury, thrombocytopenia, cardiac adverse events), and all non-serious adverse reactions will be collected.

The planned sample size is not determined, and all patients treated with Zolgensma by June 2023 will be included in the survey. During the period between 2020 and 2023, approximately 68 patients with SMA are estimated to be enrolled in Japan.

Patients will be followed for 15 years because the long-term effects of Zolgensma should be assessed. However, an interim analysis will be performed in order to compile application data for re-examination of Zolgensma.

PMDA accepted the applicant's explanation.

10. Regulations on Type-1 Use of Living Modified Organisms under Article 4 of the Act on Conservation and Sustainable Use of Biological Diversity through Regulations on the Use of Living Modified Organisms ("the Cartagena Act")

The use of Zolgensma is classified as Type-1 Use of Living Modified Organisms under Article 4 of the Cartagena Act and the Regulations on Type-1 Use of Living Modified Organisms has been approved (Approval Number: 18-36V-0003).

11. Results of Compliance Assessment Concerning the Application Data and Conclusion Reached by PMDA

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

The inspection and assessment are currently ongoing, and their results and PMDA's conclusion will be reported in the Review Report (2).

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

The inspection is currently ongoing, and its results and PMDA's conclusion will be reported in the Review Report (2).

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

On the basis of the data submitted, PMDA has concluded that Zolgensma has a certain level of efficacy in the treatment of patients with spinal muscular atrophy of who tested negative for anti-AAV9 antibodies, and that Zolgensma has acceptable safety in view of its benefits. Thus, offering Zolgensma as a new treatment option for patients with SMA to clinical practice has its significance.

PMDA has concluded that Zolgensma may be approved for marketing if Zolgensma is not considered to have any particular problems based on comments from the Expert Discussion.

Review Report (2)

February 5, 2020

Product Submitted for Approval

Brand name	ZOLGENSMA Intravenous Infusion
Non-proprietary name	Onasemnogene abeparvovec
Applicant	Novartis Pharma K.K.
Date of application	November 1, 2018

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).

1.1 Efficacy

Based on the considerations in Section "8.R.3 Efficacy" in the Review Report (1), PMDA concluded that the efficacy of Zolgensma in the treatment of patients with SMA is supported by Cohort 2 of Study CL-101, Study LT-001 (a long-term follow-up study of CL-101), and Study CL-304 involving Japanese patients.

At the Expert Discussion, the expert advisors supported the above conclusion by PMDA.

1.2 Safety

PMDA's conclusion:

Based on the considerations in Section "8.R.4 Safety" in the Review Report (1), adverse events that require attention following administration of Zolgensma are hepatotoxicity, cardiotoxicity, and thrombocytopenia.

Among the above adverse events, hepatotoxicity requires particular attention. It is necessary to specifically advise the dosing regimen of prednisolone, which is used to mitigate hepatotoxicity, in the PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE section of the package insert [see Section 1.4 in the Review Report (2)].

At the Expert Discussion, the expert advisors supported the above conclusion by PMDA.

1.3 Indication or performance

Based on the considerations in Section "8.R.5 Indication or performance" in the Review Report (1), PMDA has concluded that the following statements should be included in the INDICATION OR PERFORMANCE and PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE sections of the package insert.

Indication or Performance

Treatment of patients with spinal muscular atrophy who have tested negative for anti-AAV9 antibodies.

Precautions Concerning Indication or Performance

(1) Administer Zolgensma to patients with confirmed bi-allelic deletion or mutation in the *SMN1* gene.
(2) Administer Zolgensma to patients <2 years of age.
(3) Zolgensma is indicated for symptomatic or pre-symptomatic patients.
(4) The efficacy and safety of Zolgensma in patients with advanced SMA (e.g., permanent ventilator-dependence) have not been established. Prior to the use of Zolgensma in these patients, weigh the risks and benefits of the therapy carefully.
(5) Administer Zolgensma to patients who have tested negative for elevated anti-AAV9 antibodies. The approved *in vitro* diagnostic should be used for testing.

At the Expert Discussion, the expert advisors supported PMDA's conclusion, and made the following comment:

- In Japan, the diagnosis of SMA needs to be made based on both genetic testing and the onset of clinical symptoms. If the "INDICATION OR PERFORMANCE" is spinal muscular atrophy, the physicians may misunderstand that only symptomatic patients with a definitive diagnosis of SMA may be treated with Zolgensma, even though the PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE section advises that Zolgensma is indicated for symptomatic or pre-symptomatic patients. Thus, the INDICATION OR PERFORMANCE section needs to clearly state that even pre-symptomatic patients without a definitive diagnosis of SMA may be treated with Zolgensma as long as genetic testing indicates that they are expected to develop SMA.

PMDA's view:

Taking account of the comment from the Expert Discussion, the following statements should be included in the INDICATION OR PERFORMANCE and PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE sections of the package insert.

Indication or Performance

Treatment of patients with spinal muscular atrophy (including those with genetically diagnosed pre-symptomatic SMA) who have negative for anti-AAV9 antibodies.

Precautions Concerning Indication or Performance

(1) Administer Zolgensma to patients with confirmed bi-allelic deletion or mutation in the *SMN1* gene.
(2) Administer Zolgensma to patients <2 years of age.
(3) The efficacy and safety of Zolgensma in patients with advanced SMA (e.g., permanent ventilator-dependence) have not been established. Prior to the use of Zolgensma in these patients, weigh the risks and benefits of the therapy carefully.
(4) Administer Zolgensma to patients who have tested negative for elevated anti-AAV9 antibodies. The approved *in vitro* diagnostic should be used for testing.

Based on the above, PMDA instructed the applicant to modify the INDICATION OR PERFORMANCE and PRECAUTIONS CONCERNING INDICATION OR PERFORMANCE sections of the proposed package insert accordingly. The applicant responded to it accordingly, and PMDA accepted the response.

1.4 Dosage and Administration or Method of Use

PMDA's conclusion:

Based on the considerations in Section "8.R.6 Dosage and administration or method of use" in the Review Report (1), the statements presented in said section of the Review Report (1) should be included in the DOSAGE AND ADMINISTRATION OR METHOD OF USE and PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE sections of the package insert.

At the Expert Discussion, the expert advisors supported the above conclusion by PMDA, and made the following comment:

- It is desirable that the route of administration of prednisolone used to manage hepatotoxicity should also be mentioned in the PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE section.

PMDA's view:

Based on the comment from the Expert Discussion and modifications to the text of the package insert (the thawing/preparation procedures are included in the PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE section, etc.), the following statements should be included in the DOSAGE AND ADMINISTRATION OR METHOD OF USE and PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE sections of the package insert.

Dosage and Administration or Method of Use

The usual dose of Zolgensma for patients weighing ≥2.6 kg (<2 years of age) is 1.1×10^{14} vector genomes (vg)/kg. Zolgensma should be administered as a single, one-time intravenous infusion over 60 minutes. Do not re-administer Zolgensma.

The dose volume is determined based on patient body weight as per the table below.

Patient Weight Range (kg)	Dose Volume (mL)[Note)]
2.6 – 3.0	16.5
3.1 – 3.5	19.3
3.6 – 4.0	22.0
4.1 – 4.5	24.8
4.6 – 5.0	27.5
5.1 – 5.5	30.3
5.6 – 6.0	33.0
6.1 – 6.5	35.8
6.6 – 7.0	38.5
7.1 – 7.5	41.3
7.6 – 8.0	44.0
8.1 – 8.5	46.8
8.6 – 9.0	49.5
9.1 – 9.5	52.3
9.6 – 10.0	55.0
10.1 – 10.5	57.8
10.6 – 11.0	60.5
11.1 – 11.5	63.3
11.6 – 12.0	66.0
12.1 – 12.5	68.8
12.6 – 13.0	71.5
13.1 – 13.5	74.3

Note) Dose volume is calculated using the upper limit of the patient weight range.

The dose volume for patients <2 years of age weighing ≥13.6 kg is determined based on patient body weight.

Precautions Concerning Dosage and Administration or Method of Use

(1) Follow the steps below for preparation and infusion of Zolgensma:

1) Zolgensma should be handled aseptically.
2) The frozen product will be thawed in approximately 16 hours if refrigerated at 2°C to 8°C, or in approximately 5.5 hours if placed at room temperature. Once thawed, the product should not be re-frozen.
3) Once thawed, the product should be stored at 2°C to 8°C.
4) Do not shake the thawed product.
5) To administer Zolgensma, draw the appropriate dose volume from vials into a syringe.
6) Visually inspect vials for particulate matter and discoloration prior to administration. Do not use vials if any particles or discoloration are present.
7) Infuse Zolgensma within 8 hours after the dose volume has been drawn into syringe. Discard the vector containing syringe if not infused within the 8-hour timeframe.
8) Prime tubing with saline prior to Zolgensma infusion.
9) Flush line with saline following completion of infusion.
10) Any unused product, vials, syringe, and waste material should be placed into a waste bag or container to be sealed and appropriately disposed of in accordance with local guidelines on handling of infectious waste.

(2) Since hepatic dysfunction may occur following administration of Zolgensma, administer prednisolone as per the table below.

Table. Dosing regimen of prednisolone [Note 1)]

Prednisolone should be administered at 1 mg/kg/day 24 hours prior to Zolgensma infusion and then continued for 30 days after Zolgensma infusion. If AST and ALT values are ≤2 × ULN at the end of the 30-day period of prednisolone treatment, taper the prednisolone dose over the next ≥4 weeks (0.5 mg/kg/day for the first 2 weeks and 0.25 mg/kg/day for the subsequent 2 weeks) to discontinue prednisolone. If AST and ALT values are >2 × ULN at the end of the 30-day period of prednisolone treatment, continue prednisolone at 1 mg/kg/day until AST and ALT values are ≤2 × ULN and other liver function tests return to normal range, and then taper the prednisolone dose over the next ≥4 weeks (0.5 mg/kg/day for the first 2 weeks and 0.25 mg/kg/day for the subsequent 2 weeks) to discontinue prednisolone. As a rule, oral prednisolone should be administered.

Note 1: If patients have an inadequate response to or are intolerant of prednisolone, administer other systemic corticosteroids equivalent to oral prednisolone 1 mg/kg/day.

Based on the above, PMDA instructed the applicant to include the above statements in the DOSAGE AND ADMINISTRATION OR METHOD OF USE and PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION OR METHOD OF USE sections. The applicant responded to the instruction appropriately, and PMDA accepted it.

1.5 Post-marketing surveillance plan (draft)

During the preparation of the Review Report (1), the applicant presented a plan for post-marketing surveillance, covering all patients treated with Zolgensma, to assess the long-term safety and efficacy of Zolgensma in routine clinical practice after marketing. This surveillance will be conducted as part of a global registry to collect long-term observational data from patients with SMA in Japan, the US, Europe, and other nations (16 countries plan to participate in the registry at present) (Title: RESTORE [a planned recruitment period of 5 years from 2018 through 2023 and at least 500 patients (400 patients treated with Zolgensma, 100 patients not treated with Zolgensma) are planned to be enrolled]).

PMDA's conclusion:

Based on the considerations presented in Section "9. Risk Analysis and Outline of the Review Conducted by PMDA" in the Review Report (1), the post-marketing surveillance as planned by the applicant is acceptable. In addition to the post-marketing surveillance, the ongoing global phase III studies (Studies CL-304 and CL-306[19)]) will be reclassified as post-marketing clinical studies after approval in Japan, and Study LT-002, a long-term follow-up safety and efficacy study of participants in these 2 studies will also be conducted.

At the Expert Discussion, the expert advisors supported the above conclusion by PMDA.

19) A study to evaluate the efficacy and safety of a single intravenous dose of Zolgensma 1.1×10^{14} vg/kg in patients with SMA Type 1 or genetically diagnosed SMA Type 1 with biallelic *SMN1* mutations (deletion or point mutations) and 1 or 2 copies of *SMN2*, <6 months of age at the time of infusion, and negative for anti-AAV9 antibodies

In view of the above discussion and the following modification presented by the applicant, PMDA has concluded that the applicant should conduct post-marketing surveillance shown in Table 44.

[Major modification]
Taking account of SMA experts' opinions etc., the number of patient enrollment has been re-estimated. Approximately 50 patients with SMA (42 patients receiving Zolgensma as an initial therapy for SMA, 8 patients switching from nusinersen to Zolgensma) have been estimated to be enrolled between 2020 and 2023 in Japan.

Table 44. Outline of post-marketing surveillance

Objective	To assess the long-term safety and efficacy of Zolgensma in patients with SMA.
Survey method	All-case surveillance (Recruitment until June 2023)
Population Planned sample size	SMA patients treated with Zolgensma 50 patients
Observation period	up to 15 years from enrollment (until June 2038)
Survey items	Patient clinical characteristics Date of diagnosis, age (months), SMA type, genetic information, *SMN2* copy number, family history, etc. Treatment Date of administration/dose of Zolgensma, date of administration/dose of prednisolone, nusinersen, and other SMA drugs Adverse events ● 12 months after Zolgensma infusion: all adverse events ● During treatment with SMA drugs such as nusinersen: all adverse events ● Other periods: serious adverse events, adverse events of special interest (liver injury, platelet disorder, cardiac adverse events), all non-serious adverse reactions Efficacy Motor milestone achievement, information on ventilatory support (tracheotomy or respiratory assistance), CHOP-INTEND, etc.
Analysis plan	Interim report at filing of a re-examination application Data collection will end in October 2038, and a final report will be completed in February 2039.

2. Results of Compliance Assessment Concerning the Application Data and Conclusion Reached by PMDA

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

The application data were subjected to a document-based compliance inspection and a data integrity assessment in accordance with the provisions of the Act on Securing Quality, Efficacy and Safety of Pharmaceuticals, Medical Devices, Regenerative and Cellular Therapy Products, Gene Therapy Products, and Cosmetics. After the initial inspection, the applicant reported that the data from the *in vivo* relative potency assay in CTD 3.2.P.5.4 had been manipulated intentionally. An additional inspection revealed that the raw data from the *in vivo* relative potency assay in CTD 3.2.P.5.4 were not accurate and were not maintained appropriately. Further, the raw data from the *in vivo* potency assay (prolongation of survival of SMNΔ7 mice) in CTD 3.2.P.5.6 were not maintained appropriately. Thus, PMDA concluded that it should conduct its review after taking measures, e.g., removing the relevant data from the application documents submitted.

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

The application data (CTD 5.3.5.1-1) were subjected to an on-site GCP inspection, in accordance with the

provisions of the Act on Securing Quality, Efficacy and Safety of Pharmaceuticals, Medical Devices, Regenerative and Cellular Therapy Products, Gene Therapy Products, and Cosmetics. The inspection showed that the clinical study as a whole was performed in compliance with GCP. PMDA concluded that there were no obstacles to conducting its review based on the application documents submitted. The inspection revealed the following findings at some of the study sites and at the sponsor, though the findings had no significant impact on the overall assessment of the study. The relevant medical institutions and the applicant (the sponsor) were notified of these findings requiring corrective action.

Findings requiring corrective action

Study sites

- Protocol deviations (non-compliance with the rules for reporting serious adverse events, non-compliance with the rules for assessment of Developmental Milestones)
- Failure to properly obtain patient's new consent for administration of Bayley Scales of Infant and Toddler Development and Gross Motor Skills Checklist

Sponsor

- The sponsor failed to prepare the investigator's brochure before the initiation of the clinical study.
- Failure to appropriately detect protocol deviations (non-compliance with the rules for assessment of Developmental Milestones) during monitoring visits.

3. Overall Evaluation

As a result of the above review, PMDA has concluded that the product may be approved for the indication or performance and dosage and administration or method of use shown below, with the following conditions, provided that necessary precautionary statements are included in the package insert and information on the proper use of the product is appropriately disseminated in the post-marketing setting. Since the product has been designated as an orphan regenerative medical product, the re-examination period is 10 years. The product needs not be classified as a designated regenerative medical product.

Indication or Performance

Treatment of patients with spinal muscular atrophy (including those with genetically diagnosed pre-symptomatic SMA) who have tested negative for anti-AAV9 antibodies

Dosage and Administration or Method of Use

The usual dose of ZOLGENSMA for patients weighing ≥2.6 kg (<2 years of age) is 1.1×10^{14} vector genomes (vg)/kg. ZOLGENSMA should be administered as a single, one-time intravenous infusion over 60 minutes. Do not re-administer ZOLGENSMA.

The dose volume is determined based on patient body weight as per the table below.

Patient Weight Range (kg)	Dose Volume (mL)[Note)]
2.6 – 3.0	16.5
3.1 – 3.5	19.3
3.6 – 4.0	22.0
4.1 – 4.5	24.8
4.6 – 5.0	27.5
5.1 – 5.5	30.3
5.6 – 6.0	33.0
6.1 – 6.5	35.8
6.6 – 7.0	38.5
7.1 – 7.5	41.3
7.6 – 8.0	44.0
8.1 – 8.5	46.8
8.6 – 9.0	49.5
9.1 – 9.5	52.3
9.6 – 10.0	55.0
10.1 – 10.5	57.8
10.6 – 11.0	60.5
11.1 – 11.5	63.3
11.6 – 12.0	66.0
12.1 – 12.5	68.8
12.6 – 13.0	71.5
13.1 – 13.5	74.3

Note) Dose volume is calculated using the upper limit of the patient weight range.

The dose volume for patients <2 years of age weighing ≥13.6 kg is determined based on patient body weight.

Approval Conditions

1. Because the number of Japanese patients participating in clinical trials is very limited, the applicant is required to conduct a post-marketing use-results survey covering all patients treated with the product, until data from a certain number of patients are collected, in order to identify the characteristics of patients using the product and collect data on the safety and efficacy of the product as early as possible, thereby taking necessary measures to ensure the proper use of the product. The applicant is also required to report the results of analysis of the long-term data from post-marketing surveillance etc. to the Ministry of Health, Labour and Welfare and the Pharmaceuticals and Medical Devices Agency, and take appropriate measures as needed.
2. The applicant is required to disseminate the proper use guide developed in cooperation with the relevant academic societies and take other necessary measures, so as to ensure that physicians with adequate knowledge of and experience in the treatment of spinal muscular atrophy fully understand the results from clinical trials of the product, adverse events reported, and other data and then use the product in accordance

with the “INDICATION OR PERFORMANCE” and “DOSAGE AND ADMINISTRATION OR METHOD OF USE” statements at medical institutions prepared to treat spinal muscular atrophy.

3. The applicant is required to take necessary measures, for example, ensuring that relevant physicians are well informed of the Regulations on Type-1 Use approved under the Act on the Conservation and Sustainable Use of Biological Diversity through Regulations on the Use of Living Modified Organisms (Act No. 97 of 2003), so that the product is used in compliance with the approved Regulations on Type-1 Use.

List of Abbreviations

AAV	Adeno-associated virus
ALP	Alkaline phosphatase
ALT	Alanine aminotransferase
application	marketing application
AST	Aspartate aminotransferase
ATCC	American Type Culture Collection
AveXis	AveXis, Inc
BAV	Bovine adenovirus
BGH	bovine growth hormone
BHV	Bovine herpesvirus
BPV	Bovine parvovirus
BTV	Bluetongue virus
BVDV	Bovine viral diarrhea virus
BSA	Bovine Serum Albumin
c.859G>C	homozygous substitution of guanine by cytosine at nucleotide 859
CB	Chicken Beta Actin
cDNA	Complementary Deoxyribonucleic Acid
[redacted]	[redacted]
CHOP-INTEND	Children's Hospital of Philadelphia Infant Test of Neuromuscular Disorders
CK	Creatine kinase
CK-MB	Creatine kinase, MB form
CMAP	Compound Muscle Action Potential
CMV	Cytomegalovirus
CQA	Critical Quality Attribute
CTCAE	Common Terminology Criteria for Adverse Events
ddPCR	droplet-digital polymerase chain reaction
DNA	Deoxyribonucleic Acid
EBV	Epstein-Barr virus
ELISA	Enzyme linked immunosorbent assay
EV	Enterovirus
FBS	Fetal Bovine Serum
FDA	Food and Drug Administration
gDNA	Genomic DNA
GCTP	Good Gene, Cellular, and Tissue-based Products Manufacturing Practice
GFP	Green Fluorescent Protein
GGT	gamma-glutamyltransferase
GLP	Good Laboratory Practice
GMP	Good Manufacturing Practice
HAV	Hepatitis A Virus
HBV	Hepatitis B Virus
HCP	Host Cell Protein
HCV	Hepatitis C Virus
[redacted]	[redacted]
HEK293 cells	Human embryonic kidney 293
HeLa cells	Human cervical cancer cells
HHV	Human herpes virus
HIV	Human Immunodeficiency Virus
HSV	Herpes simplex virus
HTLV	Human T-cell leukemia virus
IBRV	Infectious bovine rhinotracheitis virus

ITR	Inverted Terminal Repeat
MCB	Master Cell Bank
MedDRA	Medical Dictionary for Regulatory Activities Japanese version
MRC-5 cells	Human embryonic lung fibroblast cells
MRI	Magnetic Resonance Imaging
mRNA	Messenger RNA
MUNE	Motor Unit Number Estimation
MVM	Mouse minute virus
NPC	Neural Progenitor Cell
nusinersen	Nusinersen Sodium
PAV	Porcine adenovirus
PCR	Polymerase chain reaction
PETG	polyethylene terephthalate glycol-modified
PHEV	Porcine hemagglutinating encephalitis virus
PHV	Porcine herpesvirus
PI3	Parainfluenza-3
PMDA	Pharmaceuticals and Medical Devices Agency
PNCR	Pediatric Neuromuscular Clinical Research
PO	Polyolefin
PPV	Porcine parvovirus
PVB19	Parvovirus B19
PRV	Pseudorabies virus
QA	Quality Assurance
QbD	Quality by Design
QC	Quality Control
Q-PCR	Quantitative polymerase chain reaction
RABV	Rabies virus
rcAAV	Replication competent AAV
Reo	Reo virus
RNA	Ribonucleic acid
RSV	Respiratory syncytial virus
[redacted]	[redacted]
SMA	Spinal Muscular Atrophy
SMN	Survival Motor Neuron
SOP	Standard operating procedures
SpO_2	saturation of percutaneous oxygen
[redacted]	[redacted]
Study CL-101	Study AVXS-101-CL-101
Study CL-102	Study AVXS-101-CL-102
Study CL-302	Study AVXS-101-CL-302
Study CL-303	Study AVXS-101-CL-303
Study CL-304	Study AVXS-101-CL-304
Study CL-306	Study AVXS-101-CL-306
Study LT-001	Study AVXS-101-LT-001
Study LT-002	Study AVXS-101-LT-002
SV40	Simian virus 40
[redacted]	[redacted]
TGEV	Transmissible gastroenteritis virus
the Cartagena Act	the Act on the Conservation and Sustainable Use of Biological Diversity through Regulations on the Use of Living Modified Organisms

the product	Zolgensma
Vero cells	African green monkey kidney epithelial cells
vg	Vector genome
WCB	Working Cell Bank
XMuLV	Xenotropic murine leukemia virus-related virus