A randomized, double-blind, placebo-controlled, multicenter phase IIa clinical study to evaluate safety and to explore efficacy of N-Rephasin® SAL200 in patients with persistent Staphylococcus aureus bacteremia

ClinicalTrials.gov ID: NCT03089697

Protocol

Study Title: 황색포도알균에 의한 지속성 균혈증 환자에서 **N-Rephasin® SAL200**의 안전성 평가 및 유효성을 탐색하기 위한 무작위배정, 이중 눈가림, 플라시보-대조, 다기관 전기 **2**상 임상시험

A randomized, double-blind, placebo-controlled, multicenter phase IIa clinical study to evaluate safety and to explore efficacy of N-Rephasin® SAL200 in patients with persistent *Staphylococcus aureus* bacteremia

Protocol No.	ITB-101
Version No.	2.1
Version Date	05/09/2019

iNtRON Biotechnology, Inc.

Signature

Sponsor of the Clinical Study Protocol [ITB-101 / ver 2.1]

iNtRON Biotechnology, Inc.
Seong Jun Yoon, Chief Executive Officer

Signature: _____ **Date:** _____

As I have read and understood all the information in this protocol, I hereby sign at the bottom.
In addition, I agree with the information specified in this protocol and agree to conduct the clinical study as specified in the protocol. I hereby confirm that I will conduct this clinical study in accordance with all relevant regulations applicable to the Korea Good Clinical Practice (KGCP) and ICH Good Clinical Practice (GCP). I will fulfill my major responsibilities as an investigator in accordance with the Declaration of Helsinki and the Institutional Review Board (IRB) regulations, and fulfill my responsibilities towards the sponsor and contract representative.

Investigator of the Clinical Study Protocol [ITB-101 / ver 2.1]

Name of the study site: _____
Principal investigator: _____

Signature: _____ **Date:** _____

▣ Definitions of Terms and Abbreviations

ADR	Adverse Drug Reaction
AE	Adverse Event
ALT	Alanine Transaminase
AST	Aspartate Transaminase
CPK	Creatine Phosphokinase
CRF	Case Report Form
ECG	Electrocardiogram
(K)GCP	(Korea) Good Clinical Practice
HED	Human Equivalent Dose
IBW	Ideal Body Weight
ICH	International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use
IRB	Institutional Review Board
LDH	Lactate Dehydrogenase
LLOQ	Lower Limit of Quantification
MRSD	Maximum Recommended Starting Dose
NAS	Network-Attached Storage
NCE	New Chemical Entity
NOAEL	No-Observed-Adverse-Effect Level
PK	Pharmacokinetic(s)
PT	Prothrombin time
RBC	Red Blood Cell
SBP	Systolic Blood Pressure
t_{max}	Time to reach the maximum concentration (C_{max}) after a single dose
WBC	White Blood Cell
MSSA	Methicillin-sensitive *Staphylococcus aureus*
MRSA	Methicillin-resistant *Staphylococcus aureus*

▣ Protocol Synopsis

Study title	황색포도알균에 의한 지속성 균혈증 환자에서 N-Rephasin® SAL200의 안전성 평가 및 유효성을 탐색하기 위한 무작위배정, 이중 눈가림, 플라시보-대조, 다기관 전기 2상 임상시험 A randomized, double-blind, placebo-controlled, multicenter phase IIa clinical study to evaluate safety and to explore efficacy of N-Rephasin® SAL200 in patients with persistent *Staphylococcus aureus* bacteremia
Sponsor	iNtRON Biotechnology, Inc.
Study drug	N-Rephasin® SAL200
Study sites	2 sites in total • Seoul National University Bundang Hospital (Professor Hong Bin Kim, Division of Infectious Diseases) • Seoul National University Hospital (Professor Wan Beom Park, Division of Infectious Diseases)
Clinical study objective	To evaluate the safety and explore the efficacy after a single intravenous administration of N-Rephasin® SAL200 (3 mg/kg) in addition to a conventional standard treatment for persistent *Staphylococcus aureus* bacteremia in *S. aureus* bacteremia patients whose *S. aureus* bacteremia lasts for 48 hours or longer despite antibiotic treatment to which *S. aureus* is susceptible.
Phase and design	Phase IIa, randomized, double-blind, placebo-controlled, multicenter clinical study
Target patients	Patients whose *S. aureus* bacteremia lasts even after 48 hours from the time when treatment using a susceptible antibiotic was started despite antibiotic treatment to which *S. aureus* is susceptible
Number of subjects	50 subjects in total: 25 subjects in each group (including a dropout rate of approximately 20%)

Arms	Sample size	Drug regimen
Control group	n=25	Conventional standard of care + placebo
Study group	n=25	Conventional standard of care + N-Rephasin® SAL200

Clinical study duration

36 months from the date of the approval for the clinical study plan by the IRB

Method of administration of the investigational product

Investigational product	Active ingredient	Method of administration
Study drug **N-Rephasin® SAL200**	SAL200 (SAL-1 18 mg/mL)	A single intravenous injection of the study drug in addition to a standard treatment
Placebo	Remaining formulation buffer excluding the active ingredient of the study drug	A single intravenous injection of the placebo in addition to a standard treatment

Subject eligibility

▶ **Inclusion criteria**

1) Those with MSSA/MRSA bacteremia who are confirmed to have more than a pair of Gram-positive bacteria in a blood culture performed at 48–96 hours after the start of antibiotic treatment to which *S. aureus* is susceptible
2) Males and females aged ≥ 19 years
3) Those who understand the information in the subject information sheet and received the informed consent form

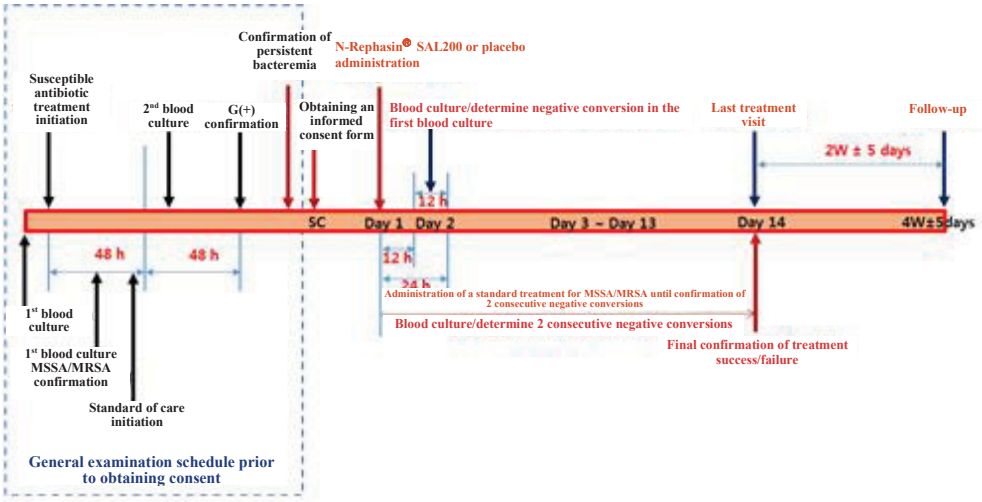
▶ **Exclusion criteria**

1) In the cases that an appropriate antibiotic has not been administered within 48 hours after the occurrence of bacteremia (the report time point of Department of Laboratory Medicine)
2) In the cases that the Gram-positive strain, identified in a blood culture performed at 48–96 hours after the start of antibiotic treatment to which *S. aureus* is susceptible, is not the same strain of *S. aureus* which was cultured when the definite diagnosis of *S. aureus* bacteremia was made
3) Those who have passed 48 hours after confirmation of persistent *S. aureus* bacteremia through a blood culture performed at 48–96 hours after the start of antibiotic treatment to which *S. aureus* is susceptible
4) Those who have symptoms of septic shock at the time of acquisition of the informed consent form
   - In the cases that systolic blood pressure is < 90 mmHg or blood pressure is lower than usual by ≥ 40 mmHg despite appropriate fluid treatment is given
   - In the cases that a hypertensor is required to be used to maintain systolic blood pressure at ≥ 90 mmHg
5) Those who are infected with mixed bacterial species

6) Those who are hypersensitive to N-Rephasin® SAL200 or have a clinically significant hypersensitivity to it or who have a past history thereof
7) Pregnant or breastfeeding women and women of child-bearing potential (those who have a possibility to become pregnant and do not agree to take appropriate contraceptive measures during the study period)
8) Those who participated in other clinical studies within 30 days prior to enrollment as subjects
9) Patients with any conditions that may interfere with study participation or accurate evaluation according to the investigator's judgment
10) Those who may die within 72 hours due to other serious complications (e.g., cerebral infarction) according to the investigator's judgment

Study method

Clinical study process



1) Patients whose *S. aureus* bacteremia lasts for 48 hours or longer even after the standard treatment for *S. aureus* bacteremia
2) Randomization according to the study site
3) The control group will receive a single intravenous injection of the placebo in addition to the standard treatment for persistent *S. aureus* bacteremia.
4) The study group will receive a single intravenous injection of N-Rephasin® SAL200 at 3 mg/kg in addition to the standard treatment for persistent *S. aureus* bacteremia.
5) A blood culture is performed 18 hours (± 6 hours) after administration of N-Rephasin® SAL200.
6) Blood cultures will continue to be performed every 24 hours (± 6 hours) or 48 hours (± 6 hours) after the time point of the previous blood culture performed until the results of the blood culture at 2 consecutive days (e.g., Day 2 and Day 3, or Day 8 and Day 10) are confirmed as negative conversions (treatment completion)
7) Side effects will be observed at the time point of the first blood culture after

	administration of N-Rephasin® SAL200 or the placebo and at the time point at intervals of 24 hours or 48 hours thereafter

Clinical evaluation	▶ **Primary endpoints** • Safety endpoints - Checking on adverse events - Laboratory tests - Anaphylaxis test, inflammatory cytokine test - Physical examination/vital signs ▶ **Secondary endpoints** • Efficacy Endpoint ① - Proportion of patients whose first blood culture performed after administration of the investigational product is negative • Efficacy Endpoint ② - Proportion of patients who die due to *S. aureus* bacteremia by Day 14 • Efficacy Endpoint ③ - Proportion of treatment failure of *S. aureus* bacteremia by Day 14 (if 2 consecutive negative conversions were not observed in blood cultures performed until Day 14)

Statistical analysis	▶ **Primary endpoints** • Safety analysis is performed in the safety set. A distribution table of the number of patients who experienced at least one side effect (incidence) and distribution tables of the relationship of the investigational product to the reported adverse events (distribution tables for severity and the relationship to the drug) will be presented for each group (study group, control group) to confirm the safety. The results of the laboratory tests, anaphylaxis test, inflammatory cytokine test, and vital signs at baseline and the last visit will be summarized as mean values and standard deviations, and the changes before and after the treatment within each group will be confirmed. Categorical data will be divided into normal and abnormal data and summarized as frequency and percentage, and the differences before and after treatment within each group will be confirmed. ▶ **Secondary endpoints** • Proportion of patients whose first blood culture performed after administration of the investigational product is negative - The descriptive statistics for the proportion of patients who are negative in the first blood culture after administration of the treatment (the rate of negative conversion) will be presented by each treatment group. Whether the rate of negative conversion in the study group is superior to that in the control group will be evaluated in a descriptive statistical manner. • Proportion of patients who die due to *S. aureus* bacteremia at Day 14 after the onset of bacteremia (according to the first confirmatory decision of bacteremia) - The descriptive statistics for the rate of mortality due to *S. aureus* bacteremia by Day14 will be presented and evaluated by treatment group. • Proportion of treatment failure of *S. aureus* bacteremia by Day 14 (if 2 consecutive negative conversions were not observed in blood cultures performed until Day 14) - The descriptive statistics for the treatment failure rate of *S. aureus* bacteremia by Day14 will be presented and evaluated.
Interim analysis	• Analysis time point: When 28 or more and 32 or less subjects have terminated participation in the clinical study • Purpose: Safety assessment (all adverse events occurred during the clinical study period) is the main purpose, and efficacy assessment (to present the proportion of each of the secondary endpoints and to compare between the treatment group) will be performed as well for an exploratory purpose.

▣ Clinical Study Schedule Table

Observation item		Screening[10]	Treatment				Follow-up[11]
		D-1	D1[12]	D2	D3~D13	D14	4W ± 5d
Written consent		√					
Checking on the inclusion/exclusion criteria		√					
Randomization[1]			√				
History of illness		√					
Data on demographic statistics		√					
Vital Signs[2]		√	√[2]	√	√	√	(√)
Physical examination[3]		√		√	√[3]	√	(√)
Laboratory tests[4, 13]	Hematology	√		√	√[4]	√	(√)
	Blood chemistry	√		√	√[4]	√	(√)
	Urinalysis	√		√	√[4]	√	(√)
Anaphylaxis test[5, 13]			√	√	√[5]		
Inflammatory cytokine test[5, 13]			√	√	√[5]		
Pregnancy test[6]		√					(√)
Skin reaction test		√					
Administration of the investigational product[7]			√				
Administration of drugs required for concomitant treatments[8]			√	√	√	√	
Blood culture/evaluation (blood collection)[9]				√	√[9]	√	
Checking on medication history/concomitant medications		√	√	√	√	√	√
Checking on concomitant treatments			√	√	√	√	√
Adverse events			√	√	√	√	√

1. Subjects who have passed screening will be assigned a subject number via randomization.
2. Vital signs will be measured once daily during the remaining treatment period except for Day 1 (including screening), and they will be measured 12 times at the following time points at Day 1:
   - Before administration of the investigational product (1 time)
   - 15 and 30 minutes after initiation of the investigational product administration (2 times)
   - Immediately after the end of the investigational product administration, 30 minutes, 1, 2, 3, 4, 8, 16, and 24 hours after the administration (9 times)
3. Physical examination will be performed at screening and Day 2, and it will be performed at an interval of "once in 2 days" after Day 2 (Days 4, 6, 8, 10, 12, and 14). It will be performed only until when the results of the blood culture/evaluation at 2 consecutive days are confirmed (treatment completion) as negative conversions.
4. Laboratory tests will be performed at screening and Day 2, and will be performed at Day 7 (± 48 hours) and Day 14 (± 48 hours) after Day 2.

Hematology	:	WBC, RBC, Hemoglobin, Platelets
Blood biochemistry	:	Total protein, Albumin, Total bilirubin, AST, ALT, ALP, Total cholesterol, Serum creatinine
Urinalysis	:	Protein, Glucose, Blood, Ketones

5. The anaphylaxis test and inflammatory cytokine test will be performed at Day 1 (before the investigational product administration and immediately after the end of the investigational product administration), Day 2, and Day 7 (± 48 hours).

Anaphylaxis test	:	C3a, C4a, Mast Cell Tryptase
Inflammatory cytokine test	:	IL-1b, IL-2, IL-6, TNF-α

6. A pregnancy test will be performed as a urine test or a serum hCG test only for women of childbearing potential.
7. The enrolled subjects will receive administration of the investigational product at Day 1.
8. Administration of drugs required for concomitant treatment will be among the drugs in "11.7.1 Drugs required for concomitant treatments" as determined by the investigator.
9. Collection of blood samples for blood cultures at Day 2 will be carried out to fulfill 18 hours (± 6 hours) from the administration of the investigational product, collection of blood samples for blood cultures at Days 3–8 will be carried out to fulfill 24 hours (± 6 hours) from collection of blood samples for blood cultures for the previous time point (once daily), and collection of blood samples for blood cultures at Days 8–14 will be carried out to fulfill 48 hours (± 6 hours) from collection of blood samples for previous blood cultures (once in 2 days). Collection of blood samples for blood cultures will be performed throughout the clinical study period until negative conversions (treatment completion) are confirmed at 2 consecutive days.
10. In the case that a subject has records of the relevant laboratory tests 7 days before screening, patient records may be used without performing additional laboratory tests at screening.
11. The tests for the relevant date will be performed when the subject visits for a follow-up at W4 ± 5 days, and concomitant medications, concomitant treatment, and adverse events will be checked via telephone monitoring. In the case of the subjects who can visit the hospital as outpatients, vital signs, physical examination, and laboratory tests will be performed, and concomitant medications, concomitant treatments, and adverse events will be checked as well.
12. Subjects who are suitable for the subject eligibility can have procedures for Day −1 and Day 1 simultaneously.
13. In the case that negative conversions (treatment completion) are confirmed at 2 consecutive days before Day 7, laboratory tests, anaphylaxis test and inflammatory cytokine test will be performed additionally.

▣ Clinical Study Process

Pre-enrollment	**All Subjects:** Written consent, screening subjects by the inclusion and exclusion criteria, and selection of subjects eligible for the clinical study based on the medical history data
⇩	
Enrollment	Randomization
⇩	
Study Group	Study drug treatment group (n=25) / Control drug treatment group (n=25)
⇩	
Treatment Period (D1~D14)	Clinical and adverse event observation (analysis of the efficacy and safety assessment items, discontinuation of the study drug administration)
⇩	
Follow-up Duration (4 week)	Clinical and adverse event observation (safety assessments and confirmation of occurrence of adverse events)
⇩	
Clinical Study Report	Evaluation and analysis of the final result of the clinical study/completion of the clinical study report

Table of Contents

1. Name and phase of the clinical study

A randomized, double-blind, placebo-controlled, multicenter phase IIa clinical study to evaluate safety and to explore efficacy of N-Rephasin® SAL200 in patients with persistent *Staphylococcus aureus* bacteremia

2. Name and address of the study site

Name of the study site	Address
Seoul National University Bundang Hospital	82, Gumi-ro 173beon-gil, Bundang-gu, Seongnam-si, Gyeonggi-do, 13620, Republic of Korea
Seoul National University Hospital	101, Daehak-ro, Jongno-gu, Seoul, 03080, Republic of Korea

3. Principal investigator of the clinical study

Name	Affiliation and title
Hong Bin Kim	Professor, Division of Infectious Diseases, Seoul National University Bundang Hospital
Wan Beom Park	Professor, Division of Infectious Diseases, Seoul National University Hospital

4. Independent Data Monitoring Committee (IDMC)

Name	Affiliation and title
Dong Gun Lee	Professor, Department of Infectious Disease, Seoul St. Mary's Hospital of the Catholic University of Korea
Chung Jong Kim	Professor, Department of Infectious Diseases, Ewha Womans University Seoul Hospital
Yong Seong Joo	Professor, Department of Statistics, Dongguk University

The IDMC will be established and operated to ensure the safety of the subjects and to implement recommendations for the progress, termination, and change of the clinical study. The IDMC will convene meetings at predefined time points and other requests to evaluate the safety data.

The IDMC will have a role to implement interim evaluation of the clinical study, and may evaluate the safety of the investigational product and monitor the overall conduct of the study as necessary. The IDMC will complete the IDMC charter and will confirm the committee member selection and the meeting schedule prior to opening a meeting for the predefined interim evaluation. The IDMC charter includes methods to secure supplementation of information, to guarantee appropriate communications, etc.

In addition, in the case that a serious adverse event such as death occurs, the IDMC may have access to the information of the group to which the relevant subject is assigned and evaluate the adverse event, if deemed necessary by the sponsor.

5. Name and address of the sponsor

5.1 Name and address of the sponsor

Company name	iNtRON Biotechnology, Inc.
Chief executive officer	Seong Jun Yoon
Address	137, Sagimakgol-ro, Jungwon-gu, Seongnam-si, Gyeonggi-do, 13202, Republic of Korea
Telephone number	+82-31-739-5352
Fax number	+82-31-736-7246

5.2 Name and address of the contract research organization

Company name	Symyoo Inc.
Address	2nd floor, 6, Hannam-daero 42-gil, Yongsan-gu, Seoul, 04417, Republic of Korea
Telephone number	+82-70-4335-5468
Fax number	+82-2-749-2050 +82-2-6442-4753 (eFax)

6. Objective and background of the clinical study

6.1 Background of the clinical study

Bacteremia is a bacterial infectious disease caused by an infection with active bacteria in circulating blood [4]. Bacteremia may not have clinical significance since it can also occur as a temporary harmless bacteremia as a result of dental treatment or other minor medical procedure, but in the case of serious bacteremia, it can progress to a more serious local or systemic infection if proper treatment is not given. Even though most bacteremia resolves, serious sequelae are common. Such bacteremia may cause pneumonia, pyogenic arthritis, osteomyelitis, meningitis, and cerebral edema, leading to death.

Staphylococcus aureus, which acts as a primary cause of bacteremia, is a bacterium that is most commonly isolated from clinical specimens among gram-positive micrococci, and it is known as a pathogenic organism that causes purulent disease, sepsis, encephalomeningitis, and food poisoning clinically, spreads from the nasal cavity or skin to other person, causes nosocomial infections such as surgical site infections and pneumonia [1][2]. In addition, it is being reported that the chances of getting an infection with a disease by direct contact between the physician and the patient through blood, saliva, or medical devices are quite high in dental care. Recently, nosocomial infections with *Staphylococcus aureus* has been steadily increasing, and infections with *Staphylococcus aureus* make treatment of patients more difficult, extend the duration of hospital stay, increase medical costs, and moreover, they can be fatal. Therefore, pathogenic infections with *Staphylococcus aureus* can be important problems [3][4][5]. Several antibacterial agents have been designed to treat infectious diseases caused by *Staphylococcus aureus*, but the frequent use of antibiotics has started to isolate bacteria strains resistant to methicillin as well as several other

antibacterial agents [6].

Methicillin-resistant *Staphylococcus aureus* (MRSA) was first isolated in the United States in 1968, and 20 to 25% of *Staphylococcus aureus* isolated from hospitalized patients hospitalized in early 1990s were identified as MRSA. The rate has been increasing continuously, and according to the National Nosocomial Infection Surveillance System (NNIS) in the United States, 50% or more of *Staphylococcus aureus* isolated from patients in intensive care units in 1999 and 59.5% or more in 2003 were identified as MRSA [7]. The frequency of MRSA isolation is increasing every year and is increasing globally in Europe and the United States as well as Singapore, Japan, Australia, the Republic of Korea, etc. recently, and the rate of increase is especially high in the Republic of Korea. The MRSA rate investigated in 12 universities and general hospitals in the nation in 2004 was 67%, and it was 86% in patients in intensive care units, which was very high [8]. According to preceding overseas studies, antibiotic resistance has been shown to cause prolonged hospitalization, expensive treatment replacement, increase in surgery frequency, increase in ICU admission, and so on [9].

Persistent bacteremia caused by methicillin-resistant *Staphylococcus aureus* (MRSA) and methicillin-susceptible *Staphylococcus aureus* (MSSA) occurs in between 6% and 38% of all *Staphylococcus aureus* bacteremia cases, and its clearance median time is between 7 to 9 days. Persistent bacteremia is defined as a condition that bacteremia persists for 3 to maximum of 7 days or more, despite appropriate active antibiotic treatment, and it is defined somewhat differently depending on the investigators. Its risk factors include sources of the infection (e.g., endocarditis or spinal osteomyelitis), pathogenic phenotypes (vancomycin heteroresistance), antibiotic treatment, maintenance or presence of prosthetic materials and its capability to remove infection clusters (e.g., surgical drainage). MRSA (clearance median time: 8–9 days) persists longer than MSSA (clearance median time: 3 days). Such a difference in the persistence between MRSA and MSSA appears to be due to pathogen-specific factors [38].

Accordingly, the development of a new antibiotic substance is urgent that can lead the public health and the pharmaceutical technology and even treat bacteria resistant to conventional antibiotics.

Based on the preceding basic study data mentioned above, iNtRON Biotechnology, Inc. found a part of the gene that is directly related to lytic activity from the genome of a bacteriophage, produced and isolated the enzyme protein belonging to endolysin from this using molecular biology technology and biotechnology, and named this as SAL-1. After formulating this, N-Rephasin® SAL200 has been developed.

Currently, antibiotics such as vancomycin or daptomycin are being used to treat MRSA bacteremia [1], but MRSA is often resistant to other antibiotics as well [2], and *Staphylococcus aureus* resistant to vancomycin or daptomycin has already appeared [3][4]. In consideration of these, a new antibacterial substance is needed that has a new mechanism, is safer, and has no concerns of developing resistance as well as potent antibacterial activity.

The study drug that has been developed according to such a demand and being used in this clinical study (N-Rephasin® SAL200) is an intravenous injection that was developed as a treatment for methicillin-resistant *Staphylococcus aureus* (MRSA) bacteremia and that contains the endolysin SAL-1, a lytic protein derived from the bacteriophage SAP-1 which is a microorganism having a specific antibacterial activity against *Staphylococcus aureus*, as an active ingredient. Unlike conventional antibiotics, SAL-1, the active ingredient of the study drug, has an antibacterial effect that destroys a specific structure of the peptidoglycan layer in the cell wall of

methicillin-resistant and methicillin-non-resistant *Staphylococcus aureus*, and therefore, it is effective in treatment of infectious diseases caused by *Staphylococcus aureus* and in treatment of infectious diseases caused by methicillin-resistant *Staphylococcus aureus*.

6.2 Rationale and objective of the clinical study

The study drug used in this clinical study (N-Rephasin® SAL200) is an intravenous injection that was developed as a treatment for methicillin-resistant *Staphylococcus aureus* (MRSA) bacteremia and that contains the endolysin SAL-1, a lytic protein derived from the bacteriophage SAP-1 which is a microorganism having a specific antibacterial activity against *Staphylococcus aureus*, as an active ingredient. Unlike conventional antibiotics, SAL-1, the active ingredient of the study drug, has an antibacterial effect that destroys a specific structure of the peptidoglycan layer in the cell wall of methicillin-resistant and methicillin-non-resistant *Staphylococcus aureus*, and therefore, it is effective in treatment of infectious diseases caused by *Staphylococcus aureus* and in treatment of infectious diseases caused by methicillin-resistant *Staphylococcus aureus*.

Currently, antibiotics such as vancomycin or daptomycin are being used to treat MRSA bacteremia [1], but MRSA is often resistant to other antibiotics as well [2], and *Staphylococcus aureus* resistant to vancomycin or daptomycin has already appeared [3][4]. In consideration of these, a new antibacterial substance is needed that has a new mechanism, is safer, and has no concerns of developing resistance as well as potent antibacterial activity.

Among bacteriophages that are microorganisms with antibacterial activity by specifically infecting only bacteria, only the bacteriophage that specifically affects *Staphylococcus aureus* (named as SAP-1) was isolated, and the genome sequence was obtained from the isolated bacteriophage. Using genetic engineering and biotechnology, an endolysin (named as SAL-1), which is an enzyme protein that is directly involved in the lysis of *Staphylococcus aureus*, was produced/purified, and this has been used as an active ingredient to formulate the study drug N-Rephasin® SAL200. Bacteriophages induce lysis of host bacteria in order to release offsprings proliferated in bacteria after infecting specific bacteria. In this process, two proteins called holin and endolysin are involved. Holins form micropores on the cell membrane, and endolysins are released through these micropores, which hydrolyze the bond in the peptidoglycan layer of the cell wall, causing the lysis of the host bacteria as a result [5]. In the natural world, endolysins are produced from the genetic information of bacteriophages and act inside of the bacteria; however, lysis of the bacteria can be induced even if endolysins are produced using a genetic recombination technology and treated outside of the cell [6]. Therefore, considering that the conventional antibiotic substances has a mechanism that inhibits cell wall synthesis, inhibits protein synthesis, destroys cell membranes, or inhibits DNA synthesis, this study drug has a novel mechanism of action that has never been applied so far, and therefore, it acts even against bacteria that have acquired resistance to conventional antibiotics regardless of their resistance [6].

The genus *Staphylococcus* includes many bacterial species, including *Staphylococcus aureus* and *Staphylococcus epidermidis*. Through the nonclinical efficacy studies on N-Rephasin® SAL200, it has been confirmed that the below 22 bacterial species in the genus *Staphylococcus* among the bacterial species belonging to the genus *Staphylococcus* are susceptible to N-Rephasin® SAL200.

1. *Staphylococcus aureus*	12. *Staphylococcus hemolyticus*
2. *Staphylococcus arlettae*	13. *Staphylococcus hominis*
3. *Staphylococcus auricularis*	14. *Staphylococcus intermedius*
4. *Staphylococcus carnosus*	15. *Staphylococcus kloosii*
5. *Staphylococcus carprae*	16. *Staphylococcus lentus*
6. *Staphylococcus chromogenes*	17. *Staphylococcus lugdunensis*
7. *Staphylococcus cohnii*	18. *Staphylococcus muscae*
8. *Staphylococcus delphini*	19. *Staphylococcus pasteuri*
9. *Staphylococcus epidermidis*	20. *Staphylococcus saprophyticus*
10. *Staphylococcus equorum*	21. *Staphylococcus warneri*
11. *Staphylococcus gallinarum*	22. *Staphylococcus xylosus*

As described above, it was confirmed that N-Rephasin® SAL200 has antibacterial activity against various species that belong to the genus *Staphylococcus*, but the Phase IIa clinical study is intended to be conducted on *Staphylococcus aureus* among the bacteria that belong to the genus *Staphylococcus* against which N-Rephasin® SAL200 displays antimicrobial activity.

Through preceding nonclinical efficacy studies, it has been confirmed that N-Rephasin® SAL200 has antibacterial activity against various *Staphylococcus aureus* strains, and N-Rephasin® SAL200 also has antibacterial activity against MSSA, MRSA, and bacteria resistant to other antibiotics (linezolid, vancomycin, daptomycin, oxacillin, etc.). These facts show that N-Rephasin® SAL200 is effective in treating bacteremia caused by *Staphylococcus aureus*.

6.2.1 Result of the nonclinical studies on the study drug (N-Rephasin® SAL200)

The peptidoglycan layer, which is the target site of the bacteriophage and its derived lytic protein endolysin, does not exist in animal or plant cells, and therefore, according to its mechanism, it does not affect cells in a target of treatment for infectious diseases, such as human cells, and only acts on bacteria specifically. As a result of the study on the cytotoxicity of SAL-1 against actual animal cells, it was found that there was no effect on the animal cells [Table 1].

[Table 1] Data related to the nonclinical efficacy studies on N-Rephasin® SAL200

No.	Title	Study site
1	Evaluation of the efficacy of lysin formulations against *Staphylococcus aureus*	Kyungpook National University School of Medicine Department of Microbiology
2	Efficacy study on N-Rephasin® SAL200 using mice (efficacy study on N-Rephasin® SAL200 against MRSA in mice)	Orient Genia Inc.
3	Basic antibacterial activity of N-Rephasin® SAL200	iNtRON Biotechnology, Inc. Biotechnology Laboratory
4	Investigation of the activity of N-Rephasin® SAL200 against *S. aureus* in various conditions	iNtRON Biotechnology, Inc. Biotechnology Laboratory

5	Efficacy study on N-Rephasin® SAL200 in serum (MIC/MBC)	iNtRON Biotechnology, Inc. Biotechnology Laboratory
6	Search for antibacterial activity	Culture Collection of Antimicrobial Resistance Microbes
7	Biofilm removal activity of N-Rephasin® SAL200	iNtRON Biotechnology, Inc. Biotechnology Laboratory
8	Study in animals treated with N-Rephasin® SAL200 after inducing infection with *Staphylococcus aureus*	iNtRON Biotechnology, Inc. Biotechnology Laboratory
9	Efficacy study on N-Rephasin® SAL200 against *Staphylococcus aureus* which has developed resistance to conventional antibiotics	BioScience LABORATORIES, INC. in the United States
10	Study on antibacterial activity of N-Rephasin® SAL200 against *Staphylococcus genus*	iNtRON Biotechnology, Inc. Biotechnology Laboratory

The results of the nonclinical studies on the study drug (N-Rephasin® SAL200) shows promise that the study drug will have great efficacy for treatment of MRSA bacteremia in human subjects.

6.2.2 Result of the Phase 1 clinical study on the study drug (N-Rephasin® SAL200)

The following results were found through the Phase 1 clinical study conducted to evaluate the safety and explore the characteristics of pharmacokinetics and pharmacodynamics after the continuous intravenous infusion of the study drug (N-Rephasin® SAL200) for 1 hour in healthy male adult volunteers. The continuous intravenous infusion of N-Rephasin® SAL200 for 1 hour has excellent tolerability within the dose range of 0.1–3 mg/kg, and it showed pharmacokinetic nonlinearity within the dose range of 0.1–10 mg/kg. In the *ex vivo* pharmacodynamic evaluation, methicillin-resistant *Staphylococcus aureus* was inhibited as the dose increased within the dose range of 0.1–10 mg/kg.

6.3 Clinical design

Randomized, double-blind, placebo-controlled, multicenter, Phase IIa clinical study

6.4 Objective of the clinical study

To conduct a Phase IIa clinical study to evaluate the safety and explore the efficacy after a single intravenous administration of N-Rephasin® SAL200 (3 mg/kg) in addition to a conventional standard treatment for persistent *S. aureus* bacteremia in *S. aureus* bacteremia patients whose *S. aureus* bacteremia lasts for 48 hours or longer despite antibiotic treatment to which *Staphylococcus aureus* is susceptible.

6.5 Data related to the nonclinical studies

The data of the nonclinical studies on the study drug (N-Rephasin® SAL200) are the data related to the efficacy study [Table 2.1], the data related to the safety pharmacology study [Table 2.2], the data related to the pharmacokinetic study (1) [Table 2.3], the data related to the pharmacokinetic study (2) [Table 2.4], and the data related to the toxicity studies [Tables 2.5–2.13] as shown below.

6.5.1 Data related to the efficacy study

[Table 2.1] Data related to the nonclinical efficacy studies on N-Rephasin® SAL200

No.	Title	Study site
1	Evaluation of the efficacy of lysin formulations against *Staphylococcus aureus*	Kyungpook National University School of Medicine Department of Microbiology
2	Efficacy study on N-Rephasin® SAL200 using mice (efficacy study on N-Rephasin® SAL200 against MRSA in mice)	Orient Genia Inc.
3	Basic antibacterial activity of N-Rephasin® SAL200	iNtRON Biotechnology, Inc. Biotechnology Laboratory
4	Investigation of the activity of N-Rephasin® SAL200 against *S. aureus* in various conditions	iNtRON Biotechnology, Inc. Biotechnology Laboratory
5	Efficacy study on N-Rephasin® SAL200 in serum (MIC/MBC)	iNtRON Biotechnology, Inc. Biotechnology Laboratory
6	Search for antibacterial activity	Culture Collection of Antimicrobial Resistance Microbes
7	Biofilm removal activity of N-Rephasin® SAL200	iNtRON Biotechnology, Inc. Biotechnology Laboratory
8	Study in animals treated with N-Rephasin® SAL200 after inducing infection with *Staphylococcus aureus*	iNtRON Biotechnology, Inc. Biotechnology Laboratory
9	Efficacy study on N-Rephasin® SAL200 against *Staphylococcus aureus* which has developed resistance to conventional antibiotics	BioScience LABORATORIES, INC. in the United States
10	Study on antibacterial activity of N-Rephasin® SAL200 against *Staphylococcus genus*	iNtRON Biotechnology, Inc. Biotechnology Laboratory

6.5.2 Data related to the safety pharmacology study

[Table 2.2] Summary of the data on the safety pharmacology study on N-Rephasin® SAL200

Test item	Detailed item	Animal species, strain, gender, and number of animals per group	Route of application	Dose administered	Test result	Compliance with GLP	Study #
Central nervous system	Modified Irwin (1964) method Temperature	32 each of male and female SPF Crl:CD SD rats	IV bolus	0, 0.5, 12.5, 25 (mg/kg)	As a result of measuring at 0.5, 1, 2, 6, and 24 hours, no changes in general behavior and temperature were observed in both male and female animals. Therefore, it was determined that there is no effect on the central nervous system.	O	G11044
Respiratory system	Tidal volume, minute respiratory volume, and respiration rate	32 male SPF Crl:CD SD rats	IV bolus	0, 0.5, 12.5, 25 (mg/kg)	As a result of measuring the tidal volume before the administration and at 0.5, 1, 2, 6, and 24 hours after the administration, no differences were observed at all time points for measurement when compared to the vehicle control group. There were significant increases in the respiratory rate at 24 hours in the 0.5 mg/kg treatment group, and significant increases in the minute respiratory volume were observed at 0 hours in the 0.5 mg/kg treatment group. Such increases were not determined to be an effect by the test substance.	O	G11045
Cardiovascular system	Blood pressure, heart rate, electrocardiogram, and general symptoms	4 male beagle dogs	IV bolus	0, 0.5, 12.5, 25 (mg/kg)	It was determined that the cardiovascular system was not affected by the dose up to 25 mg/kg when administered intravenously twice at an interval of a week in beagle dogs. When administered 3 times or more repeatedly at an interval of a week, changes were observed in the cardiovascular system that were determined to be due to immune responses.	O	G11046
	Whole-cell patch-clamp	CHO cells expressing hERG		0, 50, 200, 350, 500 (mg/mL)	IC50 > 500 μg/mL	O	G11047

6.5.3 Data related to the pharmacokinetic study

[Table 2.3] Summary of the data on the pharmacokinetic study (1)

<table>
<tr><td>Study title</td><td colspan="2">Single-intravenous-dose pharmacokinetic study on N-Rephasin® SAL200 using rats</td></tr>
<tr><td>Data suitability</td><td colspan="2">□ Data submitted upon approval (approved country) □ Specialized academic journal (SCI)
☑ Domestic and overseas institutions □ GLP</td></tr>
<tr><td>Study objective</td><td colspan="2">Conducted to confirm pharmacokinetics after intravenous administration of the test substance N-Rephasin® SAL200 in rats.</td></tr>
<tr><td rowspan="2">Study drug</td><td>Target drug</td><td>N-Rephasin® SAL200</td></tr>
<tr><td>Control drug</td><td>Formulation buffer-1</td></tr>
<tr><td rowspan="2">Details of the study</td><td>Test system</td><td>SPF [Crl:CD (SD)] rats</td></tr>
<tr><td>Study method</td><td>1) Composition of the study groups, volume of the solution administered, and dose
<table>
<tr><th>Group</th><th>Gender</th><th>Number of animals (animals)</th><th>Animal number</th><th>Volume of the solution administered (mL/kg)</th><th>Dose (mg/kg)</th></tr>
<tr><td rowspan="2">V.C.</td><td>M</td><td>4(3+1)</td><td>1-4</td><td rowspan="2">1.26</td><td rowspan="2">0</td></tr>
<tr><td>F</td><td>4(3+1)</td><td>45-48</td></tr>
<tr><td rowspan="2">T1</td><td>M</td><td>10(9+1)</td><td>5-14</td><td rowspan="2">0.02</td><td rowspan="2">0.4</td></tr>
<tr><td>F</td><td>10(9+1)</td><td>49-58</td></tr>
<tr><td rowspan="2">T2</td><td>M</td><td>10(9+1)</td><td>15-24</td><td rowspan="2">0.08</td><td rowspan="2">1.6</td></tr>
<tr><td>F</td><td>10(9+1)</td><td>59-68</td></tr>
<tr><td rowspan="2">T3</td><td>M</td><td>10(9+1)</td><td>25-34</td><td rowspan="2">0.31</td><td rowspan="2">6.25</td></tr>
<tr><td>F</td><td>10(9+1)</td><td>69-78</td></tr>
<tr><td rowspan="2">T4</td><td>M</td><td>10(9+1)</td><td>35-44</td><td rowspan="2">1.26</td><td rowspan="2">25</td></tr>
<tr><td>F</td><td>10(9+1)</td><td>79-88</td></tr>
</table>
2) Observation and test items
- Observation of general symptoms and dead animals: During the acclimation, administration and observation periods, the animals were observed once daily for occurrence of general symptoms and death. In the case that there were any abnormalities, the severity of symptoms was summarized using Parh/Tox System (v. 4.2.2) according to the types of symptoms, date of onset, and frequency.
- Weight measurement: It was performed once each at the time of group separation (acquisition) and on the day of the test substance administration.</td></tr>
</table>

Results (tabulated data, etc.)

Dose (mg/kg)	Gender	K_{el} (1/h)	$t_{1/2}$ (h)	C_{max} (μg/mL)	C_0 (μg/mL)	AUC_{last} (μg·h/mL)	AUC_{inf} (μg·h/mL)	CL (mL/h/kg)	MRT_{last} (h)
1.6	Male	8.138	0.09	0.85	1.25	0.21	0.22	7337.1	0.13
	Female	3.797	0.18	0.25	0.29	0.09	0.10	15566.5	0.19
6.25	Male	0.390	1.78	14.47	68.88	4.85	5.87	1064.1	0.26
	Female	4.197	0.17	15.24	49.09	4.15	4.18	1496.6	0.13
25	Male	0.645	1.07	87.41	175.57	30.10	30.80	811.7	0.45
	Female	0.397	1.75	19.52	20.89	15.02	17.06	1465.2	0.93

Conclusion and review comments

When the test substance N-Rephasin® SAL200 was administered intravenously to rats at a single dose of 0.4–25 mg/kg, the systemic exposure (AUC_{inf}) showed a dose-dependent increase. In addition, a pattern of bi-exponential decay was observed in the 25 mg/kg treatment group in which the completion of a concentration profile over time is easy. There was no apparent difference between male and female animals, but the systemic exposure was somewhat higher in male animals.

[Table 2.4] Summary of the data on the pharmacokinetic study (2)

<table>
<tr><td>Study title</td><td colspan="2">Single-intravenous-dose pharmacokinetic study on N-Rephasin® SAL200 using beagle dogs</td></tr>
<tr><td>Data suitability</td><td colspan="2">□ Data submitted upon approval (approved country) □ Specialized academic journal (SCI)
☑ Domestic and overseas institutions □ GLP</td></tr>
<tr><td>Study objective</td><td colspan="2">Conducted to confirm pharmacokinetics after intravenous administration of the test substance N-Rephasin® SAL200 in beagle dogs.</td></tr>
<tr><td rowspan="2">Study drug</td><td>Target drug</td><td>N-Rephasin® SAL200</td></tr>
<tr><td>Control drug</td><td>Formulation buffer-1</td></tr>
<tr><td rowspan="2">Details of the study</td><td>Test system</td><td>Canis familiaris (beagle dog)</td></tr>
<tr><td>Study method</td><td>1) Composition of the study groups, volume of the solution administered, and dose
<table>
<tr><th>Group</th><th>Gender</th><th>Number of animals (animals)</th><th>Animal number</th><th>Volume of the solution administered (mL/kg)</th><th>Dose (mg/kg)</th></tr>
<tr><td rowspan="2">V.C.</td><td>M</td><td>1</td><td>1</td><td rowspan="2">1.26</td><td rowspan="2">0</td></tr>
<tr><td>F</td><td>1</td><td>6</td></tr>
<tr><td rowspan="2">T1</td><td>M</td><td>1</td><td>2</td><td rowspan="2">0.02</td><td rowspan="2">0.4</td></tr>
<tr><td>F</td><td>1</td><td>7</td></tr>
<tr><td rowspan="2">T2</td><td>M</td><td>1</td><td>3</td><td rowspan="2">0.08</td><td rowspan="2">1.6</td></tr>
<tr><td>F</td><td>1</td><td>8</td></tr>
<tr><td rowspan="2">T3</td><td>M</td><td>1</td><td>4</td><td rowspan="2">0.31</td><td rowspan="2">6.25</td></tr>
<tr><td>F</td><td>1</td><td>9</td></tr>
<tr><td rowspan="2">T4</td><td>M</td><td>1</td><td>5</td><td rowspan="2">1.26</td><td rowspan="2">25</td></tr>
<tr><td>F</td><td>1</td><td>10</td></tr>
</table>
2) Observation and test items
- Observation of general symptoms and dead animals: During the acclimation, administration and observation periods, the animals were observed once daily for occurrence of general symptoms and death. In the case that there were any abnormalities, the severity of symptoms was summarized using Parh/Tox System (v. 4.2.2) according to the types of symptoms, date of onset, and frequency.
- Weight measurement: It was performed once each at the time of group separation (acquisition) and on the day of the test substance administration.</td></tr>
<tr><td>Results (tabulated data, etc.)</td><td colspan="2">
<table>
<tr><th>Dose (mg/kg)</th><th>Gender</th><th>K_{el} (1/h)</th><th>$t_{1/2}$ (h)</th><th>C_{max} (μg/mL)</th><th>C_0 (μg/mL)</th><th>AUC_{last} (μg·h/mL)</th><th>AUC_{inf} (μg·h/mL)</th><th>CL (mL/h/kg)</th><th>MRT_{last} (h)</th></tr>
<tr><td>6.25</td><td>Male</td><td>0.132</td><td>5.25</td><td>0.75</td><td>1.99</td><td>0.36</td><td>2.16</td><td>2894.4</td><td>0.37</td></tr>
<tr><td></td><td>Female</td><td>1.151</td><td>0.60</td><td>1.12</td><td>3.44</td><td>0.45</td><td>0.61</td><td>10234.1</td><td>0.31</td></tr>
<tr><td>25</td><td>Male</td><td>0.505</td><td>1.37</td><td>18.57</td><td>134.15</td><td>10.71</td><td>11.61</td><td>2152.7</td><td>0.73</td></tr>
<tr><td></td><td>Female</td><td>0.639</td><td>1.08</td><td>17.93</td><td>27.15</td><td>9.71</td><td>10.25</td><td>2440.1</td><td>0.79</td></tr>
</table>
</td></tr>
</table>

Conclusion and review comments	When a single dose of the test substance N-Rephasin® SAL200 was administered intravenously to beagle dogs, the systemic exposure (AUC_{last}) of N-Rephasin® SAL200 showed a dose-dependent increase, and there was no apparent difference between the male and female animals. Additionally, the elimination half-life of the test substance in the 25 mg/kg treatment group was 1.1–1.4 hours, and therefore, it is thought to be eliminated from the body within a relatively short time.

6.5.4 Data related to the toxicity studies

The data on the toxicity studies on the study drug (N-Rephasin® SAL200) are the summary of the data on the single-dose toxicity study in rats [Table 2.5], the summary of the data on the 2-week DRF toxicity study in rats [Table 2.6], the summary of the data on the 4-week repeat-dose toxicity study in rats [Table 2.7], the summary of the data on the 2-week DRF toxicity study in beagle dogs [Table 2.8], the summary of the data on the 2-week repeat-dose toxicity study in beagle dogs [Table 2.9], the summary of the data on other toxicity studies (1) [Table 2.10], the summary of the data on other toxicity studies (2) [Table 2.11], the summary of the data on the DES toxicity study in monkeys [Table 2.12], and the summary of the data on the 5-day repeat-dose toxicity study in monkeys [Table 2.13] as shown below.

[Table 2.5] Summary of the data on the single-dose toxicity study in rats

Study title	**Single-intravenous-dose toxicity study on N-Rephasin® SAL200 using rats**								
Compliance with GLP	☑ Yes □ No □ Performed before 01/01/2003 □ Documents submitted upon approval ☑ Domestic and overseas specialized institutions								
Test substance (substance name)	N-Rephasin® SAL200				Study No.		G11039		
Test animals	Species	□ Mice ☑ Rats □ Beagle dogs □ Monkeys □ Others (rabbits, guinea pigs)							
	Strain	Crl:CD(SD)							
Route of administration	□ Oral □ Subcutaneous ☑ Intravenous □ Intraperitoneal □ Intramuscular □ Others (percutaneous, inhalation)								
Administration period (observation period)	Single dose (observation for 15 days)								
Dosage form	☑ Solution □ Suspension □ colloid								
Dose (mL/kg)	5 each of male and female animals per group, 5.4 (vehicle control), 1.35, 2.7, and 5.4 mL/kg for each test group								
Details of the test	Test group	T1		T2		T3		Vehicle control	
	Concentration administered (mL/kg)	1.35 (25.11 mg/kg)		2.7 (50.22 mg/kg)		5.4 (100.44 mg/kg)		5.4 (0 mg/kg)	
	Test animals (M/F)	M	F	M	F	M	F	M	F
	Number of	5	5	5	5	5	5	5	5

	animals per group								
Reviewer's comments (test results)	(1) Dead animals No dead animals were observed in male and female animals in all study groups during the study period. (2) General symptoms No adverse events related to the test substance administration were observed in male and female animals in all study groups during the study period. (3) Weight changes No weight changes related to the test substance administration were observed in male and female animals in all study groups during the study period. (4) Necropsy findings No macroscopic abnormal findings related to the test substance administration were observed in all the male and female animals. As a result of the study, no mortality, general symptoms, weight changes, and macroscopic necropsy findings related to the test substance administration were observed in male and female animals in all study groups. In conclusion, the median lethal dose by a single intravenous administration of this test substance in rats was determined to exceed 100 mg/kg.								

[Table 2.6] Summary of the data on the 2-week DRF toxicity study in rats

Study title	**2-week repeat-intravenous-dose toxicity study and toxic dose range-finding study on N-Rephasin® SAL200 using rats**							
Compliance with GLP	☑ Yes □ No □ Performed before 01/01/2003 □ Documents submitted upon approval ☑ Domestic and overseas specialized institutions							
Test substance (substance name)	N-Rephasin® SAL200				Study No.		G11040	
Test animals	Species	□ Mice ☑ Rats □ Beagle dogs □ Monkeys □ Others (rabbits, guinea pigs)						
	Strain	Crl:CD(SD)						
Route of administration	□ Oral □ Subcutaneous ☑ Intravenous □ Intraperitoneal □ Intramuscular □ Others (percutaneous, inhalation)							
Administration period (observation period)	June 14, 2011–June 27, 2011 (study end date: July 5, 2011)							
Dosage form	☑ Solution □ Suspension □ colloid							
Dose (mL/kg)	In 5 each of male and female animals, 1.26 (vehicle control), 0.32, 0.63, and 1.26 mL/kg for each test group							
Details of the test	Test group	V. Control		T1		T2		T3	
	Concentration administered (mg/kg/day)	0.00		6.25		12.50		25.00	
	Test animals (M/F)	M	F	M	F	M	F	M	F
	Number of animals per group	5	5	5	5	5	5	5	5
Reviewer's	(1) Mortality								

comments (test results)	No deaths of animals occurred during the study period. (2) General symptoms No abnormal findings were observed for general symptoms due to the test substance. (3) Weight changes No weight changes caused by the test substance administration were observed. (4) Feed consumption No changes caused by the test substance administration were observed in all the test substance treatment groups. (5) Ophthalmic examination No abnormal findings caused by the test substance administration were observed. (6) Urinalysis For the urine volume, male animals showed significant increases or a trend of increase in the test substance treatment groups (6.25, 12.5 and 25 mg/kg/day) compared to the vehicle control group (approximately 1.2 times, 1.4 times, and 1.7 times, respectively, compared to the vehicle control group), but there was no difference in female animals. No changes caused by the test substance administration were observed in other tests. (7) Hematology Neutrophils decreased in male animals in the 6.25 and 25 mg/kg/day treatment groups (approximately 0.5 times and 0.6 times, respectively, compared to the vehicle control group), and female animals in the 6.25 and 12.5 mg/kg/day treatment groups (approximately 0.6 times and 0.7 times, respectively, compared to the vehicle control group) showed a trend of decrease, but dose-dependent trends were not observed. It was confirmed that eosinophils showed significant dose-dependent decreases in female animals in the 6.25, 12.5 and 25 mg/kg/day treatment groups (approximately 0.6 times, 0.5 times, and 0.4 times, respectively, compared to the vehicle control group). Monocytes decreased significantly in male animals in the 6.25, 12.5 and 25 mg/kg/day treatment groups (approximately 0.5 times, 0.6 times, and 0.5 times, respectively, compared to the vehicle control group), but dose-dependent trends were not observed. It was observed that basophils decreased significantly by approximately 0.7 times compared to the vehicle control group only in female animals in the 12.5 mg/kg/day treatment group. Lymphocytes increased significantly by approximately 1.1 times compared to the vehicle control group in male animals in all treatment groups, but dose-dependent trends were not observed. It was determined that the significant changes in the hematology item observed in this study were not related to the test substance as the changes were small, dose-dependent trends were not observed, or changes in related organs were not observed. (8) Blood biochemistry Total bilirubin increased significantly in male animals in the 6.25 and 12.5 mg/kg/day treatment groups (approximately 1.4 times and 1.4 times, respectively, compared to the vehicle control group), but dose-dependent trends were not observed. Gamma-glutamyltransferase increased significantly by approximately 4.3 times compared to the vehicle control group in female animals in the 12.5 mg/kg/day treatment group, but it was confirmed as an insignificant result since there were major differences among the animals. It was determined that the significant changes in the blood biochemistry item observed in this study were not related to the test substance as dose-dependent trends were not observed or changes in related organs were not observed. (9) Necropsy findings In male animals, 2 cases of enlarged spleen were observed in the 25 mg/kg/day treatment group, and 1 case of enlarged thymus was observed in the 6.25 mg/kg/day treatment group. In female animals, 1 and 3 cases of enlarged spleen were observed in the 12.5 and 25 mg/kg/day treatment groups, respectively, and 2, 2 and 4 cases of enlarged thymus were observed in the 6.25, 12.5 and 25 mg/kg/day treatment groups, respectively. Among the necropsy findings observed in this study, the incidence of increased spleen and

	thymus size in female animals were found to increase in correlation with the dose of the test substance. But, it was difficult to confirm the relationship to the test substance, as they were not severe and histological findings were not confirmed. (10) Organ weights In male animals, the absolute weights of seminal vesicles increased significantly in the 6.25 and 25 mg/kg/day treatment groups (approximately 1.3 times and 1.3 times, respectively, compared to the vehicle control group), the relative weights increased significantly by approximately 1.3 times, 1.3 times and 1.3 times in the 6.25, 12.5 and 25 mg/kg/day treatment groups, respectively, and the absolute weights of the pituitary gland increased 1.2 times in the 25 mg/kg/day group. In female animals, the absolute weights and relative weights of the spleen increased by 1.4 times, respectively, at 25 mg/kg/day. The organ weights that showed significant changes observed in this study had small changes, or their changes were not recognized as dose-dependent changes, and the histological findings need to be confirmed.
Consideration and conclusion	The no-observed-adverse-effect level of N-Rephasin® SAL200 induced by 2-week repeat-dose in rats was estimated to exceed 25 mg/kg/day.

[Table 2.7] Summary of the data on the 4-week repeat-dose toxicity study in rats

Study title	**4-week repeat-intravenous-dose toxicity and toxicokinetic study on N-Rephasin® SAL200 using rats (including a 2-week recovery period)**		
Compliance with GLP	☑ Yes □ No □ Performed before 01/01/2003 □ Documents submitted upon approval ☑ Domestic and overseas specialized institutions		
Test substance (substance name)	N-Rephasin® SAL200	Study No.	G11041
Test animals	Species	□ Mice ☑ Rats □ Beagle dogs □ Monkeys □ Others (rabbits, guinea pigs)	
	Strain	Crl:CD(SD)	
Route of administration	□ Oral □ Subcutaneous ☑ Intravenous □ Intraperitoneal □ Intramuscular □ Others (percutaneous, inhalation)		
Administration period (observation period)	Administration period/study end date: 1) Main group (♂): November 22, 2011–December 19, 2011/December 20, 2011 2) Main group (♀): November 22, 2011–December 20, 2011/December 21, 2011 3) Recovery group: November 22, 2011–December 19, 2011/January 3, 2012 (recovery period: 2 weeks)		
Dosage form	☑ Solution □ suspension □ colloid		
Dose (mL/kg)	1.5		

Details of the test

Toxicity group								
Test group	G1		G2		G3		G4	
Concentration administered (mg/kg/day)	0		4		10		25	
Test animals (M/F)	M	F	M	F	M	F	M	F
Number of animals per group	16	16	10	10	10	10	16	16

Toxicokinetics group								
Test group	G1		G2		G3		G4	
Concentration administered (mg/kg)	0		4		10		25	
Test animals (M/F)	M	F	M	F	M	F	M	F
Number of animals per group	4	4	14	14	14	14	14	14

Reviewer's comments (test results)

(1) Mortality:
No deaths of animals related to the test substance occurred during the study period.
(2) General symptoms
No changes in general symptoms related to the test substance were observed during the study period.
(3) Weight changes
No weight changes related to the test substance administration were observed.
(4) Feed consumption
No changes in feed consumption related to the test substance administration were observed.
(5) Ophthalmic examination
No adverse findings related to the test substance administration were observed.
(6) Urinalysis
As a result of the urinalysis in the main group and recovery group, no changes related to the test substance administration were observed.
(7) Hematology
No changes related to the test substance administration were observed. It was determined that some of the changes observed in the hematology performed in the main group and recovery group were not related to the test substance administration, as they were not dose-dependent, the ranges of changes were small, the values were within the normal ranges, or no associated tissue findings were observed.
(8) Blood biochemistry
As a result of the blood biochemistry in the main group and recovery group, no changes in blood biochemistry item results caused by the test substance administration were observed. It was determined that some of the statistically significant changes had no relationship to the test substance, as they were not dose-dependent, the ranges of changes were small, the values were within the normal ranges, or no associated tissue findings were observed. However, some changes in the blood biochemistry item were observed in some animals in the main group. Similar items had changes that were observed, although they were not dose-dependent. In addition, tissue findings and urinalysis findings that seem to be associated with these were observed, although they were not observed in all animals. It was determined that the changes observed in each animal were not due to the test substance,

as they were not dose-dependent changes and their patterns of occurrence were incidental. The items that changed are as shown below.

Total cholesterol (TCHO), phospholipid (PL) or triglyceride (TG) increased, and albumin (ALB) and alkaline phosphatase (ALP) decreased in Animal Nos. 21, 22 and 32 in male animals administered at doses of 4 and 10 mg/kg/day and Animal Nos. 69 and 97 in female animals administered at doses of 4 and 25 mg/kg/day in the main group. In the urinalysis results, urine protein (PRO, 3+) was detected in all the relevant animals. In addition, edematous findings were observed in the pancreas, skeletal muscles or hind limbs in some animals (Animal Nos. 22, 32 and 34) in necropsy findings, and edema was also confirmed in histopathological examination. In Animals Nos. 21, 32, 69 and 97, kidneys' organ weights were increased. In histopathological examination, tubular dilatation, hyaline droplets, and casts were commonly observed in the kidneys of Animal Nos. 32, 69, and 97. According to the comprehensive results above, decreased albumin in the blood and detection of increased urine protein indicate loss of protein through urine, and it was determined that this caused edema by changing the osmotic pressure in the body. Although significant kidney findings were not observed in all animals, it was determined that detection of increased urine protein observed in urinalysis was associated with histological changes observed in the kidneys.

(9) Necropsy findings

No changes related to the test substance were observed. It was determined that some of the necropsy findings observed during visual inspection in the main group and recovery group were incidental changes rather than changes caused by the test substance, as no dose-dependent changes were observed, and the frequency was low.

(10) Organ weights

No changes related to the test substance were observed. It was determined that some of the changes observed in the measurement of organ weights in the main group and recovery group were not related to the test substance administration, as they were not dose-dependent, the ranges of changes were small, the values were within the normal ranges, or no associated tissue findings were observed.

(11) Histopathological examination

Findings related to the test substance administration were observed in the femur/joint/bone marrow. Very mild to mild cases of increased hypertrophic zone in the middle physis of a tibia were observed in 9 out of 10 male animals administered at a dose of 25 mg/kg/day, and a very mild case was observed in 1 out of 10 female animals administered at a dose of 25 mg/kg/day. In the recovery group, very mild cases were observed in 2 out of 6 male animals and 0 out of 6 female animals administered at a dose of 25 mg/kg/day, respectively.

Besides, the findings observed during histopathological examination in the main group and recovery group were determined to be spontaneous findings or incidental findings observed in animals of similar age of the same species.

(12) Toxicokinetics

- Dose dependence: When the test substance N-Rephasin® SAL200 was administered intravenously to rats at doses of 10 and 25 mg/kg for 4 weeks, the systemic exposure (to be evaluated with AUC_{last}) at Day 1 increased with an increase in the dose administered in both male and female animals.
- Effect by repeated administration: The systemic exposure (AUC_{last}) of N-Rephasin® SAL200 after the 4-week repeat-dose increased or tended to increase with repeated administration.
- Differences depending on the gender: The differences between the male and female animals in the study substance treatment group tended to be higher in the male animals than the female animals, but the differences between the genders were not clear.

(13) Immunogenicity

- Before the administration (Day 1) and 2 weeks after initiation of the administration (Day

	14) All animals were determined to be negative or false positive. - 4 weeks after initiation of the administration (Day 28) In male animals, it was determined that 2 out of 3 cases at the dose of 4 mg/kg/day, 2 out of 3 cases at the dose of 10 mg/kg/day, and 2 out of 3 cases at the dose of 25 mg/kg/day were true positive, and all other cases were all negative or false positive. In female animals, it was determined that 1 out of 3 cases at the dose of 0 mg/kg/day, 3 out of 3 cases at the dose of 4 mg/kg/day, 3 out of 3 cases at the dose of 10 mg/kg/day, and 1 out of 3 cases at the dose of 25 mg/kg/day were true positive, and all other cases were all negative or false positive. - 6 weeks after initiation of the administration (Day 42) In male animals, it was determined that 3 out of 3 cases at the dose of 4 mg/kg/day, 3 out of 3 cases at the dose of 10 mg/kg/day, and 3 out of 3 cases at the dose of 25 mg/kg/day were true positive, and all other cases were all negative. In female animals, it was determined that 2 out of 3 cases at the dose of 0 mg/kg/day, 3 out of 3 cases at the dose of 4 mg/kg/day, 3 out of 3 cases at the dose of 10 mg/kg/day, and 2 out of 3 cases at the dose of 25 mg/kg/day were true positive, and all other cases were all negative or false positive.
Consideration and conclusion	As a result of histopathological examination, changes related to the test substance administration were observed in the femur/joint/bone marrow. Increased hypertrophic zone, middle physis, tibia was observed in male and female animals administered at a dose of 25 mg/kg/day, and it was characterized by the increased hypertrophic zone in the middle physis of a tibia. However, it was determined as a non-adverse finding, as there were no findings related to cell damage in the relevant area. In the recovery group, very mild cases were observed in male and female animals at a dose of 25 mg/kg/day, and the frequency and severity decreased compared to the main group. Therefore, it seems that the changes were reversible. As a result of analyzing toxicokinetics, when N-Rephasin® SAL200 was administered intravenously to rats at doses of 4, 10 and 25 mg/kg for 4 weeks repeatedly, the systemic exposure (AUC_{last}) of N-Rephasin® SAL200 in the 10 and 25 mg/kg treatment groups increased with an increase in the dose administered. The systemic exposure after the 4-week repeat-dose showed an increase or an increasing trend compared to that at Day 1, and no differences were observed between the male and female animals. As a result of the antibody analysis, the production of antibodies to N-Rephasin® SAL200 was observed in most samples of the test substance treatment group from 4 weeks after the administration, and a trend of an increase in the antibody titer was observed as groups went through the recovery period. In conclusion, as a result of the 4 weeks of repeated intravenous administration of the test substance N-Rephasin® SAL200 in SD rats at doses of 0, 4, 10 and 25 mg/kg/day, no adverse effects associated with the test substance were observed in mortality, observation of general symptoms, weight measurement, feed consumption, ophthalmic examination, urinalysis, hematology, blood biochemistry, necropsy findings, organ weight measurement, histopathological examination, toxicokinetic analysis, and immunogenicity analysis during the study period. Therefore, the no-observed-adverse-effect level (NOAEL) in this study was determined to be 25 mg/kg/day in male and female animals.

[Table 2.8] Summary of the data on the 2-week DRF toxicity study in beagle dogs

<table>
<tr><td>Study title</td><td colspan="10">2-week repeat-intravenous-dose toxic dose range-finding (DRF) study on N-Rephasin® SAL200 using beagle dogs</td></tr>
<tr><td>Compliance with GLP</td><td colspan="10">☑ Yes □ No
□ Performed before 01/01/2003 □ Documents submitted upon approval
☑ Domestic and overseas specialized institutions</td></tr>
<tr><td>Test substance (substance name)</td><td colspan="5">N-Rephasin® SAL200</td><td colspan="3">Study No.</td><td colspan="2">G11042</td></tr>
<tr><td rowspan="2">Test animals</td><td>Species</td><td colspan="9">□ Mice □ Rats ☑ Beagle dogs □ Monkeys □ Others (rabbits, guinea pigs)</td></tr>
<tr><td>Strain</td><td colspan="9">Canis familiaris (beagle dog)</td></tr>
<tr><td>Route of administration</td><td colspan="10">□ Oral □ Subcutaneous ☑ Intravenous □ Intraperitoneal □ Intramuscular
□ Others (percutaneous, inhalation)</td></tr>
<tr><td>Administration period (observation period)</td><td colspan="10">May 25, 2011–June 7, 2011 (study end date: September 23, 2011)</td></tr>
<tr><td>Dosage form</td><td colspan="10">☑ Solution □ Suspension □ colloid</td></tr>
<tr><td>Dose (mL/kg)</td><td colspan="10">In 1 each of male and female animals, 1.4 (vehicle control), 0.35, 0.7, and 1.4 mL/kg for each test group</td></tr>
<tr><td rowspan="4">Details of the test</td><td>Test group</td><td colspan="2">V. Control</td><td colspan="2">T1</td><td colspan="2">T2</td><td colspan="2">T3</td></tr>
<tr><td>Concentration administered (mg/kg/day)</td><td colspan="2">0.00</td><td colspan="2">6.25</td><td colspan="2">12.50</td><td colspan="2">25.00</td></tr>
<tr><td>Test animals (M/F)</td><td>M</td><td>F</td><td>M</td><td>F</td><td>M</td><td>F</td><td>M</td><td>F</td></tr>
<tr><td>Number of animals per group</td><td>1</td><td>1</td><td>1</td><td>1</td><td>1</td><td>1</td><td>1</td><td>1</td></tr>
<tr><td>Reviewer's comments (test results)</td><td colspan="10">(1) Mortality
No deaths related to the test substance administration occurred throughout the study.
(2) General symptoms
As a result of the observation of general symptoms, subdued behavior was observed at Days 14, 10, and 10–14 in male animals in the 6.25, 12.5 and 25 mg/kg/day treatment groups, respectively, and at Days 10–14 in female animals in the 6.25, 12.5 and 25 mg/kg/day treatment groups. In female animals, prone position was observed at Days 11–12, and 11 in the 6.25 and 12.5 mg/kg/day treatment groups, respectively, and decreased respiration rate was observed at Day 14 in the 12.5 and 5 mg/kg/day treatment groups. Also, in female animals, vomiting was observed at Day 8 in the 6.25 mg/kg/day treatment group, at Days 10 and 13 in the 12.5 mg/kg/day treatment group, and at Days 8–11 in the 25 mg/kg/day treatment group, and salivation was observed at Day 14 in the 6.25 mg/kg/day treatment group and at Days 12–14 in the 12.5 mg/kg/day treatment group. Eye coloration was observed at Days 13–14 in female animals in all test substance treatment groups, and skin coloration was observed at Day 13 in the 25 mg/kg/day treatment group.
(3) Weight changes
As a result of the weight measurement, no changes caused by the test substance administration were observed in all the test substance treatment groups.
(4) Feed consumption
As a result of the measurement of feed consumption, no changes caused by the test</td></tr>
</table>

	substance administration were observed. (5) Ophthalmic examination As a result of the ophthalmic examination, no changes caused by the test substance administration were observed. (6) Urinalysis As a result of the urinalysis, no changes caused by the test substance administration were observed. (7) Hematology As a result of the hematology, no adverse effects caused by the test substance administration were observed. In male animals, WBCs increased in the 6.25, 12.5 and 25 mg/kg/day treatment groups (1.66 times, 1.24 times and 1.91 times, respectively, compared to the vehicle control group), platelets decreased (0.70 times, 0.58 times and 0.66 times, respectively, compared to the vehicle control group), and basophils and large unstained cells (%) increased (1.53 times, 1.4 times and 1.6 times, respectively, and 2.25 times, 2 times and 3 times, respectively, compared to the vehicle control group). In female animals, platelets, reticulocytes and neutrophils decreased in the 6.25, 12.5 and 25 mg/kg/day treatment groups (0.67 times, 0.60 times and 0.40 times, respectively; 0.74 times, 0.63 times and 0.84 times, respectively; 0.56 times, 0.94 times and 0.71 times, respectively, compared to the vehicle control group), and monocytes and leukocytes (%) increased (1.32 times, 1.47 times and 1.61 times, respectively, and 2 times, 2 times and 3.8 times, respectively, compared to the vehicle control group). As a result of the hematology, it was determined that the changes in the hematology item observed were not related to the test substance, as the changes were small in general, or dose-dependent trends were not observed. (8) Blood biochemistry As a result of the blood biochemistry, no adverse effects caused by the test substance administration were observed. In male animals in the study group, blood urea nitrogen (BUN), total protein (TP), total bilirubin (TBIL), and creatine phosphokinase (CK) decreased in the 6.25, 12.5 and 25 mg/kg/day treatment groups (0.96 times, 0.79 times and 0.84 times, respectively; 0.86 times, 0.93 times and 0.76 times, respectively; 0.86 times, 0.58 times and 0.48 times, respectively; 0.74 times, 0.60 times and 0.46 times, respectively, compared to the vehicle control group). In female animals, BUN increased in the 6.25, 12.5 and 25 mg/kg/day treatment groups (1.33 times, 1.46 times and 1.11 times, respectively, compared to the vehicle control group). As a result of the blood biochemistry, it was determined that the changes in the blood biochemistry item observed were not related to the test substance, as the changes were small, or dose-dependent trends were not observed. (9) Necropsy findings As a result of the necropsy, no changes caused by the test substance administration were observed. In all treatment groups, including female animals in the vehicle control group, dark red discoloration at the site of administration was observed, but it was determined that this is related to the hemorrhage observed in the histopathological examination and is a response caused by repeated administration and irritation with a needle. Other findings were the changes that occur spontaneously or that are observed incidentally in beagle dogs, and it was determined that these changes were not related to the test substance administration. (10) Organ weights As a result of the measurement of organ weights, no changes caused by the test substance administration were observed. (11) Histopathological examination As a result of the histopathological examination, slight renal tubular degeneration/regeneration was observed in a male animal in the 25 mg/kg/day treatment

	group, but it is difficult to determine the relationship between the histopathological finding and the test substance administration, as each group in this study has only 1 animal and no finding was observed in the female animal in the same concentration group. Other findings observed in the histopathological examination were the changes that occur spontaneously or that are observed incidentally in beagle dogs, and it was determined that these changes were not related to the test substance administration. (12) Measurement of antibodies in blood As a result of the analysis of antibodies in serum, antibodies were produced at Day 15 in the 6.25 and 12.5 mg/kg/day treatment groups, and antibodies were not measured in the 25 mg/kg/day treatment group. In the 25 mg/kg/day treatment group, it is thought that antibodies were not measured due to the effect caused by drug interference and/or the removal from the blood by the formation of antigen–antibody complexes. [Test-Table. Analysis of samples on Day 15]
Consideration and conclusion	Through the 2-week repeated intravenous administration of N-Rephasin® SAL200 to beagle dog, various clinical symptoms presumed to be serum sickness were observed temporarily after the administration in all treatment groups administered at 6.25 mg/kg/day or higher, but these symptoms disappeared immediately. On the other hand, since the histopathological relationship to the test substance administration could not be confirmed, the no-observed-adverse-effect level (NOAEL) was determined to be 25 mg/kg/day.

Sex	Group	Dose (mg/kg/day)	Animal No.	Negative	Positive		Titer
					False	True	
Male	V.Control	0.00	1	0/1*	0/1	0/1	0
	T1	6.25	2	0/1	0/1	1/1	60
	T2	12.50	3	0/1	0/1	1/1	540
	T3	25.00	4	1/1	0/1	0/1	0
Female	V.Control	0.00	5	1/1	0/1	0/1	0
	T1	6.25	6	0/1	0/1	1/1	540
	T2	12.50	7	0/1	0/1	1/1	180
	T3	25.00	8	1/1	0/1	0/1	0

* Number of corresponding animals/number of animals tested

[Table 2.9] Summary of the data on the 2-week repeat-dose toxicity study in beagle dogs

<table>
<tr><td>Study title</td><td colspan="16">2-week repeat-intravenous-dose toxicity and toxicokinetic study on N-Rephasin® SAL200 using beagle dogs (including a 4-week recovery period)</td></tr>
<tr><td>Compliance with GLP</td><td colspan="16">☑ Yes □ No
□ Performed before 01/01/2003 □ Documents submitted upon approval
☑ Domestic and overseas specialized institutions</td></tr>
<tr><td>Test substance (substance name)</td><td colspan="8">N-Rephasin® SAL200</td><td colspan="4">Study No.</td><td colspan="4">G11043</td></tr>
<tr><td rowspan="2">Test animals</td><td>Species</td><td colspan="15">□ Mice □ Rats ☑ Beagle dogs □ Monkeys □ Others (rabbits, guinea pigs)</td></tr>
<tr><td>Strain</td><td colspan="15">Canis familiaris (beagle dog)</td></tr>
<tr><td>Route of administration</td><td colspan="16">□ Oral □ Subcutaneous ☑ Intravenous □ Intraperitoneal □ Intramuscular
□ Others (percutaneous, inhalation)</td></tr>
<tr><td>Administration period (observation period)</td><td colspan="16">Administration initiation date/necropsy date/study end date
- Male animals in the main group: November 10, 2011/November 24, 2011/April 27, 2012
- Female animals in the main group: November 10, 2011/November 25, 2011/April 27, 2012
- Recovery group: November 10, 2011/December 22, 2011/April 27, 2012</td></tr>
<tr><td>Dosage form</td><td colspan="16">☑ Solution □ Suspension □ colloid</td></tr>
<tr><td>Dose (mL/kg)</td><td colspan="16">1.5</td></tr>
<tr><td rowspan="4">Details of the test</td><td>Test group</td><td colspan="4">G1</td><td colspan="4">G2</td><td colspan="4">G3</td><td colspan="4">G4</td></tr>
<tr><td>Concentration administered (mg/kg)</td><td colspan="4">0</td><td colspan="4">2.5</td><td colspan="4">10</td><td colspan="4">25</td></tr>
<tr><td rowspan="2">Test animals (M/F)</td><td colspan="2">Main group</td><td colspan="2">Recovery group</td><td colspan="2">Main group</td><td colspan="2">Recovery group</td><td colspan="2">Main group</td><td colspan="2">Recovery group</td><td colspan="2">Main group</td><td colspan="2">Recovery group</td></tr>
<tr><td>M</td><td>F</td><td>M</td><td>F</td><td>M</td><td>F</td><td>M</td><td>F</td><td>M</td><td>F</td><td>M</td><td>F</td><td>M</td><td>F</td><td>M</td><td>F</td></tr>
<tr><td>Number of animals per group</td><td>3</td><td>3</td><td>2</td><td>2</td><td>3</td><td>3</td><td>-</td><td>-</td><td>3</td><td>3</td><td>-</td><td>-</td><td>3</td><td>3</td><td>2</td><td>2</td></tr>
<tr><td>Reviewer's comments (test results)</td><td colspan="16">(1) Mortality
No deaths of animals occurred.
(2) General symptoms
In male and female animals in the 2.5 (2 out of 3 animals, 2 out of 3 animals), 10 (3 out of 3 animals, 3 out of 3 animals) and 25 (5 out of 5 animals, 5 out of 5 animals) mg/kg treatment groups, vomiting was observed in each group continuously during the administration from 7 days after initiation of the administration.
Prone position was observed in male and female animals in the 2.5 (0 out of 3 animals, 2 out of 3 animals), 10 (0 out of 3 animals, 3 out of 3 animals) and 25 (3 out of 5 animals, 2 out of 5 animals) mg/kg treatment groups, and lateral recumbent position was observed in male and female animals in the 2.5 (0 out of 3 animals, 1 out of 3 animals), 10 (1 out of 3 animals, 2 out of 3 animals) and 25 (1 out of 5 animals, 3 out of 5 animals) mg/kg treatment groups. Subdued behavior was observed in male and female animals in the 2.5 (2 out of 3 animals, 3 out of 3 animals), 10 (1 out of 3 animals, 3 out of 3 animals) and 25 (5 out of 5 animals, 5 out of 5 animals) mg/kg treatment groups. These findings were observed during the administration period from approximately 9 or 10 days after initiation of the administration.
Irregular respiration was observed in male and female animals in the 2.5 (0 out of 3 animals, 1 out of 3 animals), 10 (0 out of 3 animals, 2 out of 3 animals) and 25 (2 out of 5 animals, 3 out of 5 animals) mg/kg treatment groups, and increased respiration rate was observed in the 10 (0 out of 3 animals, 1 out of 3 animals) and 25 (1 out of 5 animals, 0 out of 5 animals) mg/kg</td></tr>
</table>

	treatment groups. These findings were observed during the administration period from 10 days after initiation of the administration. The findings above that were observed during the administration period showed a trend of recovery during the observation of symptoms in the afternoon on the day of the administration after being observed immediately after the administration. No findings related to the test substance administration were observed during the recovery period. Decreased feed consumption, rubbing, and hypersalivation were observed as other findings during the administration period, but these findings occurred less frequently, and they were observed in the vehicle control group as well. Therefore, it was determined that they were not due to the test substance administration. (3) Weight changes No changes related to the test substance administration were observed in male and female animals in all treatment groups. (4) Feed consumption No changes related to the test substance administration were observed in male and female animals in all treatment groups. (5) Ophthalmic examination No changes related to the test substance administration were observed in male and female animals in all treatment groups. (6) Urinalysis No changes related to the test substance administration were observed in male and female animals in all treatment groups. (7) Hematology Decreased platelet (PLT) levels were observed in male and female animals in the main groups of the 2.5, 10 and 25 mg/kg treatment groups. But, the association with the test substance could not be determined, as the differences were small among the treatment groups, and no associated changes were observed in the histopathological examination. It was determined that other changes observed in the main groups and recovery groups were not related to the test substance administration, as they were not dose-dependent, the ranges of changes were small, and no associated changes were observed in the histopathological examination. (8) Blood coagulation test No changes related to the test substance administration were observed in male and female animals in all treatment groups. (9) Blood biochemistry No changes related to the test substance administration were observed in male and female animals in all treatment groups. (10) Electrocardiogram No changes related to the test substance administration were observed in male and female animals in all treatment groups. (11) Necropsy findings No changes related to the test substance administration were observed in male and female animals in all treatment groups. Dark red discoloration at the site of administration was observed in all treatment groups, including the main groups, recovery groups, and vehicle control group. It was determined that this is related to the congestion/hemorrhage observed in the histopathological examination and is a response caused by irritation from repeated administration with an injection needle or the anesthetization process at the time of necropsy. Other findings observed in the visual inspection were the changes that occur spontaneously or that are observed incidentally in beagle dogs, and it was determined that these changes were not related to the test substance administration.

(12) Organ weights
No changes related to the test substance administration were observed in male and female animals in all treatment groups.
The relative weight of kidneys increased (by approximately 1.2 times the control group) in male animals in the main group of the 25 mg/kg treatment group. However, it was determined to be an incidental change, as no associated changes were observed in the histopathological examination.
(13) Histopathological examination
No changes related to the test substance administration were observed in male and female animals in all treatment groups.
Mild to severe congestion (hemorrhage), very mild to mild vasculitis/perivasculitis, and very mild to moderate edema and inflammatory cell infiltration were observed at the site of administration in all treatment groups, including the main groups, recovery groups, and vehicle control group. It was determined that these changes were secondary responses caused by physical irritation from repeated administration with an injection needle or the process of anesthetization of the heart rather than direct effects of the test substance. Other findings related to the histopathological examination were the changes that occur spontaneously or that are observed incidentally in beagle dogs, and it was determined that these changes were not related to the test substance administration.
(14) Toxicokinetics
After the intravenous administration of the test substance N-Rephasin® SAL200 to beagle dogs at doses of 0, 2.5, 10 and 25 mg/kg/day once daily for 2 weeks, the concentration of N-Rephasin® SAL200 in serum was measured. The analysis result of the control group has not been tabulated, as all samples in the control group showed that the concentration of N-Rephasin® SAL200 was below the limit of quantification (1 μg/mL). Toxicokinetics analysis was performed only in the 10 and 25 mg/kg treatment groups in which biological samples are relatively easy to quantify.
After the intravenous administration of the test substance N-Rephasin® SAL200 at doses of 10 and 25 mg/kg, the systemic exposure (to be evaluated with AUC_{last}) to N-Rephasin® SAL200 increased 3.2 times in male animals and 4.1 times in female animals with an increase in the administered dose by 2.5 times at Day 1. The rate of increase was higher than the increase in the administered dose. In addition, the difference in the systemic exposure between the gender in the 25 mg/kg treatment group was 1.3 times higher in male animals compared to female animals at Day 1 and 1.8 times higher at Day 14, which were somewhat higher, but the difference between the gender was not clear.
The systemic exposure (AUC_{last}) to N-Rephasin® SAL200 after repeated administration for 2 weeks increased 1.9 times in male animals and 3.3 times in female animals in the 10 mg/kg treatment group at Day 14 compared to Day 1. In the 25 mg/kg treatment group in which the change in concentration over time was easy to check, it increased 1.9 times in male animals and 1.4 times in female animals, showing an increasing trend after repeated administration. This is thought to be due to the decrease in the total body clearance and the increase in MRT_{last}. On the other hand, referring to the antibody measurement test result, the systemic exposure to N-Rephasin® SAL200 has slightly increased despite the production of antibodies in the serum of some animals after repeated administration for 2 weeks. Therefore, it was determined that the production of antibodies did not significantly affect the toxicokinetic profile itself.
(15) Antibody analysis
Before the administration, all animals were determined to be negative or false positive.
It was determined that male and female animals in the 0 (1 out of 5 animals, 1 out of 5 animals), 2.5 (2 out of 3 animals, 3 out of 3 animals), 10 (1 out of 3 animals, 3 out of 3 animals) and 25 (1 out of 5 animals, 0 out of 5 animals) mg/kg treatment groups were each true positive

	at 2 weeks after the administration. It was determined that male and female animals in the 0 (1 out of 2 animals, 1 out of 2 animals) and 25 (1 out of 2 animals, 1 out of 2 animals) mg/kg treatment groups were each true positive at 2 weeks of the recovery. It was determined that male and female animals in the 0 (1 out of 2 animals, 2 out of 2 animals) and 25 (1 out of 2 animals, 1 out of 2 animals) mg/kg treatment groups were each true positive at 4 weeks of the recovery. Among these antibody measurement results, an increase in antibodies was observed in some animals in the vehicle treatment group, but the titer was lower than the N-Rephasin® SAL200 treatment groups generally, and there were not many cases. Therefore, it was determined that there was no effect on the immunogenicity evaluation in this study. In addition, according to the results of the serum C3 complement measurement assay performed additionally, the concentration of C3 in the N-Rephasin® SAL200 treatment group was lower than that in the vehicle control group.
Consideration and conclusion	Vomiting, prone position, lateral recumbent position, subdued behavior, irregular respiration, and increased respiration rate that were determined to be caused by the effect of the test substance administration were observed in male and female animals in all test substance treatment groups from 7 days after initiation of the administration and throughout the administration period. This was determined to be temporary symptoms occurred by activation of the complement system by the test substance–antibody complexes produced by administration of the test substance. Such a possibility was confirmed by the serum C3 complement measurement assay result. In this assay, the concentration of C3 in the N-Rephasin® SAL200 treatment groups was lower than that in the vehicle control group, and it was determined that this was due to the activation of complement by the immune complex formation. On the other hand, these findings were observed immediately after the administration, and they showed a trend of recovery immediately after the onset of symptoms and were not observed during the 4-week recovery period after the end of the administration. As a result of analyzing toxicokinetics, when N-Rephasin® SAL200 was administered intravenously to beagle dogs at doses of 2.5, 10 and 25 mg/kg for 2 weeks repeatedly, the systemic exposure (AUC_{last}) of N-Rephasin® SAL200 in the 10 and 25 mg/kg treatment groups, which were relatively easy to quantify, increased proportionally with an increase in the dose administered. The rate of increase was higher than the increase in the administered dose, and no differences were observed between the male and female animals. The systemic exposure of N-Rephasin® SAL200 after the 2-week repeat-dose showed a somewhat increasing trend when compared to that at Day 1. As a result of the antibody analysis, the production of antibodies to N-Rephasin® SAL200 was observed in most of the N-Rephasin® SAL200 treatment group from 2 weeks after the administration, and it was observed that higher doses administered resulted in lower titer values in both male and female animals. Especially in the treatment group in which the highest concentration (25 mg/kg) was administered, it was confirmed that several animals showed a pattern that there was almost no antibody production until 2 weeks after the administration, but it increased as the group went through the recovery period (at 4 weeks and 6 weeks). This might be the result of drug interference that interferes with the measurement of the antibody due to the remaining of the high concentration of N-Rephasin® SAL200 in blood by producing complexes with antibodies, and the drug interference limit in this assay identified in the GV11038 study was 150 ng/mL. Therefore, if N-Rephasin® SAL200 is present in serum at a concentration higher than 150 ng/mL, there is a possibility that antibody measurement may be interrupted. On the other hand, in the result of the toxicokinetic analysis, the concentration of N-Rephasin® SAL200 was observed lower than the lower limit of quantification (LLOQ) in all

	samples prior to the administration, and it was observed that the concentrations became lower than the lower limit of quantification in animal samples of all treatment groups within 1 hour after the administration. However, the lower limit of quantification of the relevant test method is 1 μg/mL, which is higher than the drug interference limit of the antibody measurement assay, and therefore, it was determined that the possibility of drug interference in the antibody measurement could not be ruled out. Also, it was determined that the antibody in the serum could be measured low due to the formation of immune complexes between the produced antibodies and excessive N-Rephasin® SAL200, resulting in deposition in the tissues or induction of clearance by complement. Such a possibility was confirmed by the serum C3 complement measurement assay result. In this assay result, the concentration of C3 in the N-Rephasin® SAL200 treatment groups was lower than that in the vehicle control group, and it was determined that this was resulted due to the activation of complement by the immune complex formation. Also, in the case of blood samples collected from the high-dose groups, it was determined that no antibody in the blood sample was detected, as the relevant dose group had more antigens compared to other dose groups, and this resulted in more effective removal of the antigen–antibody complexes by activation of the complement. From the above results, vomiting, prone position, lateral recumbent position, subdued behavior, irregular respiration, and increased respiration rate were observed as general symptoms immediately after the administration in male and female animals in the 2.5, 10 and 25 mg/kg treatment groups as the changes associated with the test substance administration. Therefore, the no-observed-adverse-effect level (NOAEL) of the test substance N-Rephasin® SAL200 in this study was determined to be < 2.5 mg/kg in both male and female animals.

[Table 2.10] Summary of the data on other toxicity studies (1)

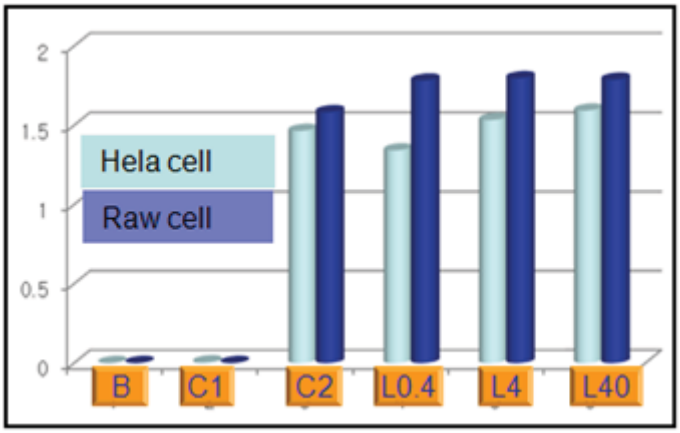
<table>
<tr><th>Study title</th><td colspan="3">In vivo safety test on N-Rephasin® SAL200 in mice</td></tr>
<tr><th>Data suitability</th><td colspan="3">□ Data submitted upon approval (approved country) □ Specialized academic journal (SCI)
☑ Domestic and overseas institutions □ GLP</td></tr>
<tr><th>Study objective</th><td colspan="3">To demonstrate the safety of N-Rephasin® SAL200 in animals even if an excessive dose compared to the effective dose is administered intravenously.</td></tr>
<tr><th rowspan="3">Study drug</th><td>Target drug</td><td>Substance name</td><td>N-Rephasin® SAL200</td></tr>
<tr><td rowspan="2">Control drug</td><td>Positive</td><td>Not applicable</td></tr>
<tr><td>Negative</td><td>Vehicle
[0.01 M L-histidine, pH 6.0, 5% (w/v) sorbitol, 10 mM $CaCl_2$, 0.1% (w/v) poloxamer 188]</td></tr>
<tr><th rowspan="2">Details of the study</th><td>Test system</td><td colspan="2">Mice (BALB/C, 6 weeks old female)</td></tr>
<tr><td>Study method</td><td colspan="2">Investigation of general symptoms and weights after single intravenous administration by applying various doses (15 mg/kg, 30 mg/kg and 50 mg/kg)</td></tr>
<tr><th>Evaluation indices</th><td colspan="3">Weights and general symptoms</td></tr>
<tr><th>Results (tabulated data, etc.)</th><td colspan="3">As a result of the observation, no abnormal signs in all respects, such as changes in shine of fur and behavior, were observed in the treatment groups compared to the control group. In addition, weight changes also showed no significant differences in the control group and treatment groups.

<Weight measurement results>
<table><tr><th>Target</th><th>Weight (unit: g)</th></tr></table></td></tr>
</table>

	Group	Animal	0 hr	24 hr	48 hr	72 hr	96 hr
	Control (Group 1)	1	20	19.5	19	19	19
		2	19	18	18	18	18
		3	19	18.5	18.5	18.5	18.5
		4	20	19	19.5	19.5	19.5
	Average		19.5	18.8	18.8	18.8	18.8
	15 mg/kg treatment group (Group 2)	1	20	19	19.5	19.5	19
		2	19	19	19	18.5	19
		3	19	18.5	18.5	18	18.5
		4	19	18	18	18	18
		5	20	18.5	19.5	19.5	19
	Average		19.4	18.6	18.9	18.9	18.7
	30 mg/kg treatment group (Group 3)	1	20	18.5	18.5	18.5	18.5
		2	20	19.5	19.5	19.5	19.5
		3	20	19.5	19.5	19.5	20
		4	19.5	18.5	19	19	18.5
		5	20	19	20	20	20
	Average		19.9	19	19.3	19.3	19.3
	50 mg/kg treatment group (Group 4)	1	19	19	19	19	19.5
		2	20	19	19.5	19.5	20
		3	19	18	18.5	18.5	18.5
		4	20	18.5	18.5	18.5	18
		5	20	19	18	18	17.5
	Average		19.6	18.7	18.7	18.7	18.7
Conclusion	It has been confirmed that N-Rephasin® SAL200 has no particular toxicity even after administration at dose of 50 mg/kg.						
Review comments	It has been determined that N-Rephasin® SAL200 is considerably safe.						

[Table 2.11] Summary of the data on other toxicity studies (2)

Study title	**Measurement of the cytotoxicity of N-Rephasin® SAL200 against animal cells**		
Data suitability	□ Data submitted upon approval (approved country) ☑ Domestic and overseas institutions	□ Specialized academic journal (SCI) □ GLP	
Study objective	To demonstrate that N-Rephasin® SAL200 is not cytotoxic, as it does not act on animal cells.		
Study drug	Target drug	Substance name	N-Rephasin® SAL200
	Control drug	Positive	Not applicable
		Negative	1× PBS
Details of the study	Test system	Animal cells (Hela cells and Raw cells)	
	Study method	Performing an MTT assay, which is the cell viability assay and proliferation assay	
Evaluation indices	Cytotoxicity (the quality of being toxic to cells) of N-Rephasin® SAL200 to animal cells		

Results (tabulated data, etc.)	When 1×10^4 animal cells were seeded in the well and proliferated for 72 hours, it was confirmed that the presence of N-Rephasin® SAL200 in the contents of the medium has no significant level of effect on cell growth.  ▪ C2: Negative control ▪ L0.4, L4, L40: N-Rephasin® SAL200 added
Conclusion	It was confirmed that N-Rephasin® SAL200 has no cytotoxicity to animal cells.
Review comments	It has been determined that N-Rephasin® SAL200 has no cytotoxicity to animal cells.

[Table 2.12] Summary of the data on the DES toxicity study in monkeys

<table>
<tr><td>Study title</td><td colspan="13">Single dose-escalation, intravenous-dose toxicity study on N-Rephasin® SAL200 using cynomolgus monkeys</td></tr>
<tr><td>Compliance with GLP</td><td colspan="13">□ Yes ☑ No
□ Performed before 01/01/2003 □ Documents submitted upon approval
☑ Domestic and overseas specialized institutions</td></tr>
<tr><td>Test substance (substance name)</td><td colspan="6">N-Rephasin® SAL200</td><td colspan="3">Study No.</td><td colspan="4">N13036</td></tr>
<tr><td>Test animals</td><td>Species</td><td colspan="12">□ Mice □ Rats □ Beagle dogs ☑ Monkeys □ Others (rabbits, guinea pigs)</td></tr>
<tr><td>Test animals</td><td>Strain</td><td colspan="12">Cynomolgus Monkey (Macaca fascicularis)</td></tr>
<tr><td>Route of administration</td><td colspan="13">□ Oral □ Subcutaneous ☑ Intravenous □ Intraperitoneal □ Intramuscular
□ Others (percutaneous, inhalation)</td></tr>
<tr><td>Administration period (observation period)</td><td colspan="13">August 28, 2013–September 2, 2013 (study end date: October 8, 2013)</td></tr>
<tr><td>Dosage form</td><td colspan="13">☑ Solution □ Suspension □ colloid</td></tr>
<tr><td>Dose (mL/kg)</td><td colspan="13">Administration in 1 each of male and female animals while increasing the doses over a total of 6 times (1, 5, 10, 20, 40 and 80 mg/kg) once daily</td></tr>
<tr><td rowspan="5">Details of the test</td><td>Test group</td><td colspan="12">G1</td></tr>
<tr><td>Concentration administered (mg/kg/day)</td><td>1</td><td>5</td><td>10</td><td>20</td><td>40</td><td>80</td><td>1</td><td>5</td><td>10</td><td>20</td><td>40</td><td>80</td></tr>
<tr><td>Day of administration (Day)</td><td>1</td><td>2</td><td>3</td><td>4</td><td>5</td><td>6</td><td>1</td><td>2</td><td>3</td><td>4</td><td>5</td><td>6</td></tr>
<tr><td>Test animals (M/F)</td><td colspan="6">M</td><td colspan="6">F</td></tr>
<tr><td>Number of animals</td><td colspan="6">1</td><td colspan="6">1</td></tr>
<tr><td>Reviewer's comments (test results)</td><td colspan="13">(1) Mortality
No deaths of animals occurred during the study period.
(2) General symptoms
No symptoms related to the test substance administration were observed. Vaginal discharge was observed in the female animal, but it was determined to be a temporary change that was not related to the test substance administration.
(3) Weight changes
No weight changes related to the test substance administration were observed.
(4) Feed consumption
No changes in feed consumption related to the test substance administration were observed.
(5) Hematology
No changes in the hematological indices related to the test substance administration were observed. The values that showed changes compared to the period before the administration were minor and within the normal range, and therefore it was determined that they had no relationship to the test substance administration or toxicological significance.
(6) Blood biochemistry
Increases in blood urea nitrogen (BUN, ×2.88 of baseline data) and creatinine (CREA, ×1.56 of baseline data) were observed in the male animal. Besides, the values that showed changes</td></tr>
</table>

	compared to the period before the administration were minor and within the normal range, and therefore it was determined that they had no relationship to the test substance administration or toxicological significance. **(7) Necropsy findings** No findings caused by the test substance administration were observed. **(8) Pharmacokinetic analysis** After the intravenous administration of the test substance N-Rephasin® SAL200 at doses of 10 mg/kg (Day 3) and 80 mg/kg (Day 6), the systemic exposure (to be evaluated with AUC_{last}) to N-Rephasin® SAL200 increased at a ratio of 1:9 in male animals and 1.8 in female animals with a proportional increase in the administered dose by at a ratio of 1.8 at Day 1. At the time of the first blood collection, the highest blood concentration was observed, and it decreased by multi-exponential decay. In addition, in terms of the systemic exposure, the differences between the male and female animals showed 1.3 and 1.1 times in female animals compared to those of the male animals, showing no gender differences in the male and female animals. Moreover, the elimination half-life ($t_{1/2}$) of the drug was 2.0–2.2 hours at 10 mg/kg and 2.7–5.0 hours at 80 mg/kg. The elimination half-life tended to increase with an increase in the dose administered, and the volume of distribution was approximately 0.28–0.52 L/kg. Therefore, it was expected that the tissue distribution of the test substance was not so large.
Consideration and conclusion	In this study, to investigate the toxicity of a single dose of the test substance N-Rephasin® SAL200, single dose was administered intravenously once daily in 1 each of male and female cynomolgus monkeys with single dose-escalating over a total of 6 times (1, 5, 10, 20, 40 and 80 mg/kg). The mortality rate, general symptoms, weight changes, feed consumption, hematology, blood biochemistry, and pharmacokinetic analysis were performed during the study period, and the animals were necropsied at 8 days after the first administration and visually inspected for findings. No deaths of animals occurred during the study period, and no changes in general symptoms, body weights, feed consumption, hematology values, and necropsy findings related to the test substance administration were observed. As a result of the blood biochemistry, increases in blood urea nitrogen (BUN) and creatinine (CREA) levels were observed in the male animal, but no other hematology or necropsy findings were accompanied, and the changes were observed only in the male animal, not in both male and female animals. Therefore, it was difficult to determine that they were the toxicity caused by the test substance. As a result of the pharmacokinetic analysis, when administered at doses of 10 and 80 mg/kg, the systemic exposure (to be evaluated with AUC_{last}) to the test substance N-Rephasin® SAL200 increased in proportional to the dose administered with an increase in the dose administered in male and female animals. No differences were observed between the male and female animals, the elimination half-life ($t_{1/2}$) of the drug was 2.0–2.2 hours at 10 mg/kg and 2.7–5.0 hours at 80 mg/kg. It tended to increase with an increase in the dose administered, and the volume of distribution was approximately 0.28–0.52 L/kg. Therefore, it was expected that the tissue distribution was not so large. From the results above, as a result of the single intravenous administration with an increase in the dose of the test substance N-Rephasin® SAL200, it was determined that the maximum tolerance dose (MTD) will exceed 80 mg/kg/day.

[Table 2.13] Summary of the data on the 5-day repeat-dose toxicity study in monkeys

<table>
<tr><td>Study title</td><td colspan="6">N-Rephasin® SAL200: 5-day repeat-intravenous-dose toxicity study using cynomolgus monkeys</td></tr>
<tr><td>Compliance with GLP</td><td colspan="6">☑ Yes □ No
□ Performed before 01/01/2003 □ Documents submitted upon approval
☑ Domestic and overseas specialized institutions</td></tr>
<tr><td>Test substance (substance name)</td><td colspan="3">N-Rephasin® SAL200</td><td>Study No.</td><td colspan="2">G13138</td></tr>
<tr><td rowspan="2">Test animals</td><td>Species</td><td colspan="5">□ Mice □ Rats □ Beagle dogs ☑ Monkeys □ Others (rabbits, guinea pigs)</td></tr>
<tr><td>Strain</td><td colspan="5">Cynomolgus Monkey (Macaca fascicularis)</td></tr>
<tr><td>Route of administration</td><td colspan="6">□ Oral □ Subcutaneous ☑ Intravenous □ Intraperitoneal □ Intramuscular
□ Others (percutaneous, inhalation)</td></tr>
<tr><td>Administration period (observation period)</td><td colspan="6">Male animals: February 6, 2014–February 11, 2014 (study end date: April 2, 2014)
Female animals: February 7, 2014–February 12, 2014 (study end date: April 2, 2014)</td></tr>
<tr><td>Dosage form</td><td colspan="6">☑ Solution □ Suspension □ colloid</td></tr>
<tr><td>Dose (mL/kg)</td><td colspan="6">Administration in 3 each of male and female animals in each group twice daily for 5 days at doses of 0 (vehicle control group), 10, 20 and 40 mg/kg/day</td></tr>
<tr><td rowspan="5">Details of the test</td><td rowspan="2">Study group</td><td rowspan="2">Gender</td><td rowspan="2">Number of animals (animals)</td><td rowspan="2">Animal number</td><td colspan="2">Volume of the solution administered (mL/kg/day)</td><td rowspan="2">Dose (mg/kg/day)</td></tr>
<tr><td>1st</td><td>2nd</td></tr>
<tr><td>G1</td><td>Male
Female</td><td>3
3</td><td>1 - 3
13 - 15</td><td>0.98</td><td>0.98</td><td>0</td></tr>
<tr><td>G2</td><td>Male
Female</td><td>3
3</td><td>4 - 6
16 - 18</td><td>0.245</td><td>0.245</td><td>10</td></tr>
<tr><td>G3</td><td>Male
Female</td><td>3
3</td><td>7 - 9
19 - 21</td><td>0.49</td><td>0.49</td><td>20</td></tr>
<tr><td>G4</td><td>Male
Female</td><td>3
3</td><td>10 - 12
22 - 24</td><td>0.98</td><td>0.98</td><td>40</td></tr>
<tr><td>Reviewer's comments (test results)</td><td colspan="6">(1) Mortality
No deaths of animals occurred during the study period.
(2) Observation of general symptoms
No general symptoms related to the test substance were observed. Besides, other general symptoms observed, including loss of fur, scratch wounds, vaginal discharge, soft feces, swelling, scars, skin coloration, and loss of tail, occurred less frequently and only occurred in specific animals. Therefore, they were determined to be spontaneous or incidental changes.
(3) Weight
No significant weight changes related to the test substance were observed.
(4) Feed consumption
No changes in feed consumption related to the test substance were observed.
(5) Ophthalmological examination
No adverse findings related to the test substance were observed.
(6) Temperature measurement
No temperature changes related to the test substance were observed. Other observed changes occurred less frequently and only occurred in specific animals. Therefore, they were</td></tr>
</table>

	determined to be spontaneous or incidental changes. **(7) Electrocardiogram** No changes related to the test substance were observed. **(8) Hematology** No changes related to the test substance were observed. It was determined that the changes observed in the hematology were not related to the test substance administration, as they were not dose-dependent, the ranges of changes were small, or the values were within the normal ranges. **(9) Blood biochemistry** No changes related to the test substance were observed. **(10) Urinalysis** No changes related to the test substance were observed. **(11) Macroscopic observation** No changes related to the test substance were observed. Other observed changes were determined to be the changes caused by intravenous administration (site of administration) or the spontaneous or incidental changes. **(12) Measurement of organ weights** No changes related to the test substance were observed. The increases and decreases observed as a result of the measurement of organ weights were determined to have no relationship to the dose or to be incidental changes not related to the test substance administration. **(13) Toxicokinetics** Dose dependence: When the test substance N-Rephasin® SAL200 was administered intravenously to monkeys for 5 days at doses of 10–40 mg/kg, the systemic exposure (to be evaluated with AUC_{last}) of N-Rephasin® SAL200 showed a dose-dependent increase. N-Rephasin® SAL200 showed the maximum blood concentration (C_{max}) of the test substance mostly at the first blood collection time (0.083–0.2 hr) after the intravenous administration, and then it decreased rapidly. Effect by repeated administration: The systemic exposure of N-Rephasin® SAL200 after the repeated administration at an interval of 12 hours for 5 days (AUC_{last}, Day 5, 2nd dosing) showed no difference compared to that at Day 1 (1st dosing). This pattern was also observed for the trough level. Differences depending on the gender: No differences were observed between the male and female animals in the study substance treatment group.
Consideration and conclusion	To investigate the toxicity of the test substance N-Rephasin® SAL200 after repeated intravenous administration for 5 days, the test substance was administered intravenously in 3 each of male and female cynomolgus monkeys in each group twice daily for 5 days repeatedly at doses of 0 (vehicle control group), 10, 20 and 40 mg/kg/day. During the study period, the mortality, observation of general symptoms, weight changes, and feed consumption were observed, and the temperature measurement, ophthalmic examination, electrocardiogram, urinalysis, hematology, blood biochemistry, necropsy findings, and organ weight measurement did not show any changes related to the test substance. According to the toxicokinetic results, the systemic exposure increased with an increase in the doses administered in male and female animals, and the difference in the systemic exposure and the accumulation in the body were not observed after the repeated administration for 5 days. In addition, there were no differences in the exposure according to the gender of the animals.

6.6 Data related to the clinical study

The study drug (N-Rephasin® SAL200) is an injection that contains 18 mg/mL of SAL-1 as the active ingredient. The study drug is a new drug and has no clinical use experience, but its safety was secured and its efficacy was demonstrated through preclinical studies. As a result of the Phase 1 clinical study conducted based on these demonstrated the safety and tolerability of the study drug [Table 3].

6.6.1 Data related to the Phase 1 clinical study

[Table 3] Synopsis of the Phase 1 clinical study

Study title	A Randomized, Double-Blind, Placebo-Controlled, Single-Dosing, Dose-Escalating Study to Evaluate the Safety, Pharmacokinetics, and Pharmacodynamics of N-Rephasin® SAL200 after Continuous Intravenous Infusion in Healthy Volunteers
Study drug	N-Rephasin® SAL200
Study site and principal investigator	Department of Clinical Pharmacology and Therapeutics, Seoul National University College of Medicine and Department of Clinical Pharmacology and Therapeutics, Seoul National University Hospital In Jin Jang, MD, PhD
Clinical study duration	November 27, 2013 Date of Last Completion: February 7, 2014
Study objective	To evaluate the safety and explore the characteristics of pharmacokinetics and pharmacodynamics after continuous intravenous infusion of N-Rephasin® SAL200 for 1 hour in healthy male volunteers
Study method	This study was conducted as a randomized, double-blind, placebo-controlled, single-dosing, dose-escalating clinical study to evaluate the safety and explore the characteristics of pharmacokinetics and pharmacodynamics after continuous intravenous infusion of N-Rephasin® SAL200 for 1 hour by dose group in healthy volunteers. As a preliminary study, 4 subjects were randomized to the 0.1 mg/kg dose group (3 subjects for study drug and 1 subject for the placebo) to conduct the clinical study. Then, 8 subjects in the 0.3, 1, 3 and 10 mg/kg dose groups were randomized (6 subjects for study drug and 2 subjects for the placebo) to conduct the clinical study. After completion of the administration in each dose group, the safety was evaluated to determine whether to proceed to the next dose group. The subjects deemed eligible for this clinical study were selected by performing screening tests, such as history taking, physical examination, and clinical laboratory tests, etc. within 4 weeks (Day −28 to Day −2) from a day before the clinical study initiation (Day −1) only in volunteers. Then, the subjects visited the research unit of the Seoul National University Hospital Clinical Trials Center by 1 p.m. on the day before the clinical study initiation (Day −1), and they had a skin test. After the skin test, the subjects who did not have specific reactions were admitted to the research unit of the Seoul National University Hospital Clinical Trials Center. Subjects who passed the skin test were assigned a subject number. The subjects fasted from 10 p.m. on the day of admission (Day −1), except for drinking water. The study drug or placebo was infused intravenously in all subjects, depending on the relevant treatment group, at around 9 a.m. on the next day of the admission, and the clinical study was conducted according to the clinical study schedule. After a certain period of time, post-study visit tests were performed in the subjects who completed the entire clinical study.

Number of subjects

- 57 subjects screened
- 36 subjects enrolled, 36 subjects received the administration, 34 subjects completed

Dose group (mg/kg)	Planned (N)	Enrolled (N)	Withdrawn (N)
0.1	4	4	1
0.3	8	8	0
1	8	8	0
3	8	8	1
10	8	8	0

Subject eligibility

1. Inclusion criteria

1) Healthy male adults aged 20 to 45 years at screening
2) Those whose weigh is ≥ 50 kg and < 90 kg and within the range of ±20% of the ideal body weight
   ☞ Ideal body weight (kg) = (Height [cm] − 100) × 0.9
3) Those who fully understood this clinical study after the detailed explanation, decided to voluntarily participate, and gave a written consent to comply with precautions

2. Exclusion criteria

1) Those who have clinically significant liver, kidney, nervous system, endocrine system, respiratory system, hemato-oncology, cardiovascular system, or mental diseases or past history
2) Those who have had an infectious disease or suspected clinical findings within 30 days prior to the date of the investigational product administration
3) Those who are hypersensitive to drugs containing N-Rephasin® SAL200 or other drugs (aspirin, antibiotics, etc.) or have a clinically significant hypersensitivity to them or who have a past history thereof
4) Those who have received administration of drugs containing N-Rephasin® SAL200
5) Those who are antibody-positive to N-Rephasin® SAL200
6) Those who have SBP < 90 mmHg or DBP < 50 mmHg, or SBP > 150 mmHg or DBP > 100 mmHg in vital signs when measured after taking a 3-minute rest in a sitting position
7) Those who have a past history of drug abuse or who are positive to urine drug screening
8) Those who have taken any prescription drugs, oriental medicines or herbal medicines within 14 days prior to the date of the investigational product administration, or have taken over-the-counter (OTC) drugs or vitamin supplements within 7 days (However, if other conditions are appropriate upon judgment of the investigator, the subject may be selected as a subject.)
9) Those who have received administration of other investigational products within 2 months prior to the date of the investigational product administration
10) Those who have donated whole blood within 2 months or apheresis blood within 1 month prior to the date of the investigational product administration, or received blood transfusion within 1 month prior to the date of the initial administration
11) Those who smoke cigarettes or who have been found to be nicotine metabolite-positive in urinalysis
12) Those who continuously drink alcohol (exceeding 21 units/week; 1 unit = 10 g of

<table>
<tr><td></td><td>pure alcohol) or who cannot abstain from drinking alcohol and smoking cigarettes during hospitalization
13) Those who are judged ineligible to participate in the clinical study by the investigator due to other reasons, including the results of clinical laboratory tests
14) Those who do not agree to practice contraception using appropriate methods for 60 days after the date of the administration, or those who do not consent to voluntarily report the female partner's pregnancy to the sub-investigator until 90 days after the date of the administration</td></tr>
<tr><td>Method of administration of the investigational product</td><td>
<table>
<tr><th>Dose group (mg/kg)</th><th>Subject (N)</th><th>Treatment</th><th>Volume</th></tr>
<tr><td>0.1</td><td>3
1</td><td>Study drug, Placebo</td><td>0.0056 ml/kg
0.0056 ml/kg</td></tr>
<tr><td>0.3</td><td>6
2</td><td>Study drug, Placebo</td><td>0.017 ml/kg
0.017 ml/kg</td></tr>
<tr><td>1</td><td>6
2</td><td>Study drug, Placebo</td><td>0.056 ml/kg
0.056 ml/kg</td></tr>
<tr><td>3</td><td>6
2</td><td>Study drug, Placebo</td><td>0.167 ml/kg
0.167 ml/kg</td></tr>
<tr><td>10</td><td>6
2</td><td>Study drug, Placebo</td><td>0.556 ml/kg
0.556 ml/kg</td></tr>
</table>
At around 9 a.m. on Day 1, each of the subject received intravenous infusion of the investigational product corresponding to the treatment group for 1 hour continuously on an empty stomach. Drinking water was not allowed 1 hour before and after the administration, and the subjects kept fasting for 4 hours after starting the administration.</td></tr>
<tr><td>Clinical evaluation</td><td><u>Safety evaluation</u>
- Monitoring of adverse events, such as subject and objective symptoms
- Physical examination, vital signs, clinical laboratory tests, 12-lead ECG

<u>Pharmacokinetic evaluation</u>
Blood was collected to evaluate the pharmacokinetics of N-Rephasin® SAL200 after intravenous administration of the study drug.
- Blood sampling (15 times): Pre-dose (0), 0.25, 0.5, 0.75, 1, 1.5, 2, 3, 4, 5, 6, 8, 10, 12, and 24 hours post-dose
- Endpoints: C_{max}, AUC_{last}, AUC_{inf}, t_{max}, $t_{1/2}$, CL/F</td></tr>
<tr><td>Statistical analysis</td><td><u>Data presentation/Descriptive statistics</u>
- All demographic information, safety and pharmacokinetic data were summarized using descriptive statistics according to appropriate factors, such as dose group and treatment group.

<u>Safety and tolerability data</u>
- About the occurrence of adverse events, the number of cases occurred, number of subjects experiencing the cases, severity, seriousness, and causality relationship to the study drug were classified using the system organ classes and preferred terms of the MedDRA, and they were analyzed by dose group and treatment group in a descriptive statistical manner.
- Results of tests, such as vital signs, electrocardiogram, and clinical laboratory tests were reviewed collectively.</td></tr>
</table>

	Pharmacokinetic data - For pharmacokinetic parameters, the values were obtained for each subject, and their mean, standard deviation, etc. were provided in a descriptive statistical manner. - The main pharmacokinetic parameters AUC_{last}, AUC_{inf}, and C_{max} were calculated using a noncompartmental analysis method, and the values corrected by dose administered were also analyzed.
Results	**Study subjects** A total of 57 volunteers visited the Seoul National University Hospital Clinical Trials Center for screening, and 21 of them failed as ineligible subjects. A total of 36 subjects received the study drug or the placebo, and 2 of the subjects who received the administration withdrew their consent. Accordingly, a total of 34 subjects completed the clinical study. Subjects who received the investigational product were included in the safety evaluation set, and 27 subjects who received the administration of N-Rephasin® SAL200 and completed blood collection for pharmacokinetics were included in the set for pharmacokinetic evaluation. **Safety** A total of 42 adverse events were reported in 12 out of 36 subjects in total who received either the study drug or the placebo during this study, and 3 of these cases occurred in 1 subject who received the placebo. No adverse events were reported in the 0.1 mg/kg dose group. A total of 11 adverse events were reported in the 0.3–3 mg/kg dose groups, and 6 of these were related to the drug. The severity of the reported adverse events were evaluated as “mild” for 5 cases and “moderate” for 1 case, and all of them recovered without sequelae and disappeared without any medical intervention except for 1 case in 1 subject (R207) who was positioned in a Trendelenburg position due to syncope that occurred before the administration. It was determined that hyperbilirubinemia (Subject R208 in the 0.3 mg/kg dose group), which was evaluated as a “moderate” case, was not a clinically significant result considering that the relevant level was high at screening, it decreased without any medical intervention afterward, and no related adverse events were reported or specific findings found during a physical examination. 28 adverse events occurred in 6 subjects who received the study drug in the 10 mg/kg dose group. Of these, 25 cases were found to be related to the drug. Of the adverse events that were found to be related to the drug, 23 cases were reported as mild and 2 cases were reported as moderate. A total of 5 subjects received the administration of a drug (acetaminophen) for treatment of adverse events, and all of them recovered without sequelae. On the other hand, no subjects discontinued the study due to serious adverse events and adverse events. Two subjects (subject numbers: R502, R503) who were evaluated as “moderate” felt discomfort due to rigors that occurred after the administration, and its severity was enough to interfere with other activities of daily living (using computers, reading books, etc.). All the relevant subjects received the administration of acetaminophen for treatment of adverse events as determined by the investigator, all of them recovered without sequelae, and they completed the clinical study. Based on the results above, it is evaluated that N-Rephasin® SAL200 is safe and well tolerated from 0.1 mg/kg to 3 mg/kg when infused intravenously for 1 hour in healthy male volunteers. **Pharmacokinetics** When N-Rephasin® SAL200 was infused intravenously for 1 hour at doses of 0.1, 0.3,

1, 3 and 10 mg/kg, the maximum blood concentration was reached at 25 minutes to 1 hour, and then it was eliminated rapidly afterwards until 4 hours. C_{max} and AUC_{last}, which reflect the highest blood concentration and the exposure in the body of N-Rephasin® SAL200, showed a pattern of a nonlinear increase pharmacokinetically in the dose groups ranging from 0.1 mg/kg to 10 mg/kg. The mean effective half-life of the drug, reflecting the mean residual time, was 0.04–1.23 (after baseline correction: 0.04–0.38), and it tended to increase with an increase in the dose.

Pharmacodynamics

For the pharmacodynamic evaluation of this study, clear zone formation was compared after dripping each of the calibration sample prepared with N-Rephasin® SAL200 at concentrations of 0–1.0 μg/mL and the serum samples obtained from subjects on the medium on which methicillin-resistant *Staphylococcus aureus* was smeared. In the 0.1 mg/kg and 0.3 mg/kg dose groups, no subjects showed results similar to the bacteria suppression effect observed in the calibration sample prepared with N-Rephasin® SAL200 at 0.05 μg/mL. In the 1 mg/kg and 3 mg/kg dose groups, the bacteria suppression effects were similar in the serum sample obtained 1 hour after the administration and the calibration samples prepared with N-Rephasin® SAL200 at concentrations of 0.1, 0.2 and 0.4 μg/mL. In the 10 mg/kg dose group, the bacteria suppression effect of the serum samples obtained at 1 and 1.5 hours after the administration in all subjects was similar to the bacteria suppression effect of the calibration samples prepared with N-Rephasin® SAL200 at concentrations of 0.1–0.6 μg/mL. In 3 subjects among them, the results corresponding to the bacteria suppression effect of the calibration samples prepared with N-Rephasin® SAL200 at concentrations of 0.05–0.1 μg/mL were confirmed in the serum samples obtained 2 hours after the administration. According to the correlation between the serum N-Rephasin® SAL200 concentration of the entire subjects who received the study drug and the N-Rephasin® SAL200 concentration of the calibration sample, the calibration sample prepared with N-Rephasin® SAL200 at a concentration of 0.1 μg/mL or higher has the similar effect to the result of bacteria suppression when the blood concentration of N-Rephasin® SAL200 is approximately 9.054 (9.015 after baseline correction) μg/mL or higher, and the serum measured 1 hour after the administration showed the same bacteria suppression effect as above in all subjects who received administration of N-Rephasin® SAL200 at the 1 mg/kg dose or higher.

Conclusion

The continuous intravenous infusion of N-Rephasin® SAL200 for 1 hour has excellent tolerability within the dose range of 0.1–3 mg/kg, and it showed pharmacokinetic nonlinearity within the dose range of 0.1–10 mg/kg. In the *ex vivo* pharmacodynamic evaluation, methicillin-resistant *Staphylococcus aureus* was inhibited as the dose increased within the dose range of 0.1–10 mg/kg.

7. Code name of the investigational product, etc. or generic name of the active ingredient, ingredients and their quantities, formulation, etc.

7.1 Investigational product

7.1.1 Study drug

Product name	**N-Rephasin® SAL200**	**Remark**
Content of the active ingredient	100% SAL200 (18 mg/mL as SAL-1)	
Formulation and appearance	Injection filled with a colorless clear liquid in a colorless transparent vial	
Dosage and dosing regimen	A single intravenous injection of N-Rephasin® SAL200 in addition to a standard treatment	
Storage method	Store in a freezer (−70 ± 5°C)	
Shelf life (effective period)	18 months from the date of manufacture	
Manufacturer	BINEX Co., Ltd.	Entire manufacturing process contracted

7.1.2 Placebo

Product name	**INT200 – Placebo (Saline)**	**Remark**
Content of the active ingredient	Remaining formulation buffer excluding the active ingredient of the study drug	
Formulation and appearance	Injection that has the same formulation and appearance as the study drug	
Dosage and dosing regimen	A single intravenous injection of the placebo in addition to a standard treatment	
Storage method	Store in a freezer (−70 ± 5°C)	
Shelf life (effective period)	18 months from the date of manufacture	
Manufacturer	BINEX Co., Ltd.	Entire manufacturing process contracted

7.2 Storage method and shelf life

Store a sealed container in a freezer (−70 ± 5°C); 18 months from the date of manufacture

7.3 Packaging and labeling of the investigational product

7.3.1 Packaging

Each investigator will receive a sufficient quantity of the below investigational products as necessary from iNtRON Biotechnology, Inc., the sponsor of this study.

Category	Product name	Packaging unit	Ingredients and contents
Study group	N-Rephasin® SAL200	1 mL/vial	100% SAL200 (18 mg/mL as SAL-1)
Control group	INT200 – Placebo (Saline)	1 mL/vial	Remaining formulation buffer excluding the active ingredient of the study drug

7.3.2 Labeling

The labels provided by the study site will be used for the study drug and placebo, and if they are not provided, the sponsor will provide the study drug and placebo with the label with each of below information entered. The relevant visit cycle will be entered separately to distinguish the baseline and treatment visit cycles.

[For Clinical Trial Use Only]	Identification number assigned to the investigational product
[Protocol number]	
[Code name of the product] or [generic name of the active ingredient]	
[Lot number]	
[Shelf life]	
[Storage method]	
Name and address of the Investigational New Drug application holder	
DO NOT USE FOR OTHER PURPOSES THAN CLINICAL TRIALS	

7.3.3 Management and records of the investigational product

The investigational products (hereinafter referred to as the “clinical drug”) will be manufactured by iNtRON Biotechnology, Inc. and supplied to the clinical study site (hereinafter referred to as the “study site”), and they will be managed by the clinical trial pharmacist designated by the head of the study site. Whenever the clinical drugs are delivered, iNtRON Biotechnology, Inc. will deliver the quality assurance certificate for the clinical drugs to the clinical trial pharmacist at each time, and the clinical trial pharmacist must confirm the receipt and quantity of the clinical drugs provided by the sponsor and sign in writing.

The clinical trial pharmacist should properly store and manage the study drugs and the control drugs according to the KGCP regulations and the protocol and should communicate with the investigator closely to mutually confirm the accountability of all clinical drugs used in the clinical study.

The clinical drugs must be dispensed according to a prescription signed by the attending physician participating

as an investigator in this study, and the initial of the subject, subject number, date of dispensation, quantity, etc. should be recorded in the "investigational product accountability log."

In addition, the clinical trial pharmacist should accurately record the quantity of the drugs supplied by iNtRON Biotechnology, Inc., the quantity of dispensation, and the quantity of remaining drugs, and all drugs not used in the clinical study at the end of the study should be returned to the sponsor.

The clinical research associate should periodically check the quantity of the clinical drugs stored by the clinical trial pharmacist in order to confirm the history of use of all clinical drugs. If the clinical study is completed or is discontinued in the middle, issues related to the collection or disposal of all clinical drugs shall be handled based on the sponsor's request.

In any case, the investigator should not supply the clinical drugs and related products to other investigators or study sites or use them for any purpose other than that specified in the protocol, unless approved by the sponsor. The clinical drug provided to the subjects will be covered by iNtRON Biotechnology, Inc.

8. Target disease

Blood infections (bacteremia) caused by methicillin-susceptible and -resistant *Staphylococcus aureus*

9. Criteria for the inclusion and exclusion of subjects, target number of subjects and its rationale

This clinical study will be conducted in patients with MSSA/MRSA bacteremia caused by *Staphylococcus aureus* who meet the inclusion criteria and do not fall under the exclusion criteria.

9.1 Inclusion criteria

This clinical study will select those who meet the following conditions as the subjects:

1) Those with MSSA/MRSA bacteremia who are confirmed to have more than two pairs of Gram-positive bacteria in a blood culture performed at 48–96 hours after the start of antibiotic treatment to which *S. aureus* is susceptible
2) Males and females aged ≥ 19 years
3) Those who understand the information in the subject information sheet and received the informed consent form

9.2 Exclusion criteria

Patients who fall under any of the following conditions cannot participate in this clinical study:

1) In the cases that an appropriate antibiotic has not been administered within 48 hours after the occurrence of bacteremia (the report time point of Department of Laboratory Medicine)
2) In the cases that the Gram-positive strain, identified in a blood culture performed at 48–96 hours after the start of antibiotic treatment to which *S. aureus* is susceptible, is not the same strain of *S. aureus* which was cultured when the definite diagnosis of *S. aureus* bacteremia was made
3) Those who have passed 48 hours after confirmation of persistent *S. aureus* bacteremia through a blood culture performed at 48–96 hours after the start of antibiotic treatment to which *S. aureus* is susceptible

4) Those who have symptoms of septic shock at the time of acquisition of the informed consent form
   - In the cases that systolic blood pressure is < 90 mmHg or blood pressure is lower than usual by ≥ 40 mmHg despite appropriate fluid treatment is given
   - In the cases that a hypertensor is required to be used to maintain systolic blood pressure at ≥ 90 mmHg
5) Those who are infected with mixed bacterial species
6) Those who are hypersensitive to N-Rephasin® SAL200 or have a clinically significant hypersensitivity to it or who have a past history thereof
7) Pregnant or breastfeeding women and women of child-bearing potential (those who have a possibility to become pregnant and do not agree to take appropriate contraceptive measures during the study period)
8) Those who participated in other clinical studies within 30 days prior to enrollment as subjects
9) Patients with any conditions that may interfere with study participation or accurate evaluation according to the investigator's judgment
10) Those who may die within 72 hours due to other serious complications (e.g., cerebral infarction) according to the investigator's judgment

9.3 Target number of subjects and its rationale

This is a Phase IIa clinical study to evaluate the clinical efficacy and safety of N-Rephasin® SAL200 in addition to a conventional standard treatment in patients with persistent bacteremia caused by *Staphylococcus aureus*.

The primary endpoints are the safety endpoints to be evaluated by presenting a distribution table of the number of subjects who experienced at least one side effect (incidence) and distribution tables of the relationship of the investigational product to the reported adverse events (distribution tables for severity and the relationship to the drug) for each group (study group, control group). Also, the purpose of this study is to obtain baseline data on the study drug and control drug.

The primary evaluation will use descriptive statistics for the study drug and control drug, and no hypothesis testing will be performed. The number of subjects required was 20 subjects respectively in the study group and control group considering their representativeness, and it has been decided to enroll 25 subjects in each group considering a dropout rate of 20%.

Arms	**Drug regimen**	Number of subjects for safety evaluation	Number of subjects including a dropout rate (approximately 20%)
Control group	Standard treatment for MSSA/MRSA + placebo	20	25
Study group	Standard treatment for MSSA/MRSA + N-Rephasin® SAL200	20	25

In 50 subjects in total, the clinical study will be divided into 2 steps—Step 1 (28 or more and 32 or less subjects) and Step 2 (approximately 22 subjects).

Upon completion of Step 1 of this clinical study at the discretion of the sponsor, the IDMC will perform an interim evaluation of the safety and efficacy, and the study will proceed to Step 2, if the investigational product is determined to be safe.

Number of subjects in each step

Step 1	Interim evaluation	Step 2
28–32 subjects		Approximately 22 subjects

10. Duration of the clinical study

The duration of this study will be 36 months from the date of approval for the clinical study by the IRB.

However, in the case of a situation that may affect the progress of the clinical study, such as a difficulty in selecting subjects, the duration may be changed.

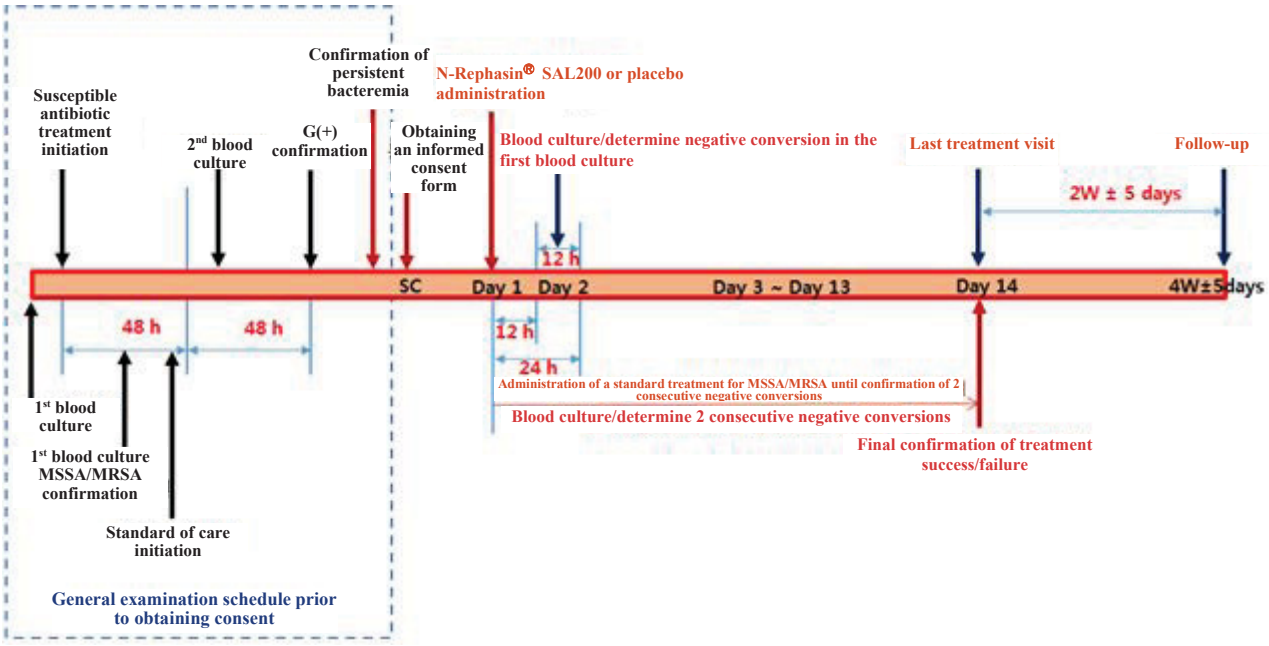
11. Methods of the clinical study

11.1 Design of the clinical study

This is a multi-center, randomized, double-blind, placebo-controlled, parallel-group, Phase IIa clinical study to explore the efficacy and confirm the safety through the confirmation of microbiological eradication of pathogens by performing blood cultures in each group after 18 hours (± 6 hours) from the time of administration of the study drug or control drug (placebo) after a single intravenous administration of N-Rephasin® SAL200 and the control drug (placebo) in addition to treatment with a conventional standard treatment in patients with persistent MSSA/MRSA bacteremia despite treatment with a standard treatment. The subjects will be randomized using a random code program of the statistical SAS program to design the study to have approximately the same number of subjects in the two groups.

To maintain the blinding status, neither the investigator nor the subject will know to which group each subject is assigned while conducting the study. Unblinding will not happen unless a medical emergency occurs, and the confidentiality of the assigned groups will be maintained. In the case of unblinding due to an adverse event, etc., it must be reported to the IRB and informed to the sponsor.

11.2 Process of the clinical study



<Clinical study schedule>

1) Patients whose *S. aureus* bacteremia lasts for 48 hours or longer even after the standard treatment for *S. aureus* bacteremia
2) Randomization according to the study site
3) The control group will receive a single intravenous injection of the placebo in addition to the standard treatment for persistent *S. aureus* bacteremia.
4) The study group will receive a single intravenous injection of N-Rephasin® SAL200 at 3 mg/kg in addition to the

standard treatment for persistent *S. aureus* bacteremia.

5) A blood culture is performed 18 hours (± 6 hours) after administration of N-Rephasin® SAL200.
6) Blood cultures will continue to be performed every 24 hours (± 6 hours) or 48 hours (± 6 hours) after the time point of the previous blood culture performed until the results of the blood culture at 2 consecutive days (e.g., Day 2 and Day 3, or Day 8 and Day 10) are confirmed as negative conversions (treatment completion) in blood cultures
7) Side effects will be observed at the time point of the first blood culture after administration of N-Rephasin® SAL200 or the placebo and at the time point at intervals of 24 hours or 48 hours thereafter

11.3 Randomization of subjects

A randomization method will be used to guarantee the scientific validity of the clinical study and to ensure that the investigator's personal opinion is not involved in assigning subjects to each treatment group.
Once the subjects who satisfy the subject inclusion criteria are selected, the investigator will assign the randomization numbers to the subjects sequentially in the order of randomization.

For the randomization, a stratified block randomization method, which has been stratified by study site, will be used.
Randomization codes will be generated using the PROC PLAN procedure of SAS (version 9.2).
The enrolled subjects will be assigned to the study group and control group at a ratio of 1:1 according to the randomization codes.

The randomization number will consist of 1 letter of a study site code and 2 digits assigned in the order of randomization.

R			

Serial No.

The investigator will use this number to prescribe the investigational product, and the clinical trial pharmacist will dispense the investigational product with the number that matches the prescribed assignment number to the subject. The subject numbers and randomization numbers of the subjects dropped out from the study will remain continuously. In the case of dropping out, the relevant numbers will also drop out. A new subject must be assigned a new subject number and randomization number.

11.4 Dosage and method of administration

Subjects will be determined whether they are eligible for this clinical study at the screening visit. Eligible subjects will be assigned randomization numbers, and the clinical study will be conducted according to the schedule of clinical study plan. In this step, the drugs used in the following clinical study will be administered:

Investigational product	Active ingredient	Method of administration
Study drug N-Rephasin® SAL200	SAL-1 18 mg/mL	A single intravenous injection of N-Rephasin® SAL200 in addition to a standard treatment
Placebo	Remaining formulation buffer excluding the active ingredient of the study drug	A single intravenous injection of the placebo in addition to a standard treatment

During the treatment period, the standard treatment for MSSA/MRSA bacteremia will be given, and at the same time, a single dose of the study drug (or placebo) will be administered intravenously to the subject. The placebo and study drug are in the same kind of containers that cannot be distinguished visually. Each case packaged by serial number will be each labeled separately, and the clinical trial pharmacist will deliver them to the investigator according to the prescription issued separately. The placebo or study drug will be administered directly to the subject.

In 50 subjects in total, the clinical study will be divided into 2 steps—Step 1 (28 or more and 32 or less subjects) and Step 2 (approximately 22 subjects).

Upon completion of the clinical study in subjects in Step 1, the IDMC will perform an interim evaluation of the safety and efficacy, and the study will proceed to Step 2, if the investigational product is determined to be safe.

Step	Number of subjects	Dose and method of administration
1	Approximately 28 subjects (28 or more and 32 or less subjects)	① Calculate the dose in consideration of the body weight in order to administer the investigational product at a dose of 3 mg/kg. : Weight of the subject (kg) × 3 mg/kg = Dose of the investigational product (mg) ② Dilute the dose calculated in ① in normal saline, and prepare the injection to have 110 mL as the volume of the entire solution. ③ Administer the injection solution prepared in ② to the subject intravenously at an infusion rate of 155 mL/hr.
The IDMC will perform an interim evaluation of the safety and efficacy in subjects in Step 1 and issue a report. The safety will be confirmed in the interim evaluation report, and the study will proceed to Step 2		
2	Approximately 22 subjects	The study will proceed using the same method of administration as in Step 1.
Total Subjects	50 subjects	25 subjects in the study group : 25 subjects in the control group

11.5 Administration period

The study drug or placebo will be administered to all randomized subjects once at Day 1 during the clinical study period depending on the group to which the subjects are assigned. Along with this, treatment using a standard treatment will be performed concomitantly until the blood culture results are confirmed as negative conversions (treatment completion) at 2 consecutive days.

11.6 Blinding and unblinding

This clinical study will maintain the blinding of the investigator and the subject from the information on the type of drug assigned to each subject. By maintaining double-blinding of the investigator and subject, the investigator's and subject's bias in the evaluation of the therapeutic effect and adverse events, etc. can be prevented.

When unblinding is deemed necessary due to such a reason as an emergency situation threatening the subject's safety, the investigator is required to notify this immediately to the sponsor or the clinical research associate of a contract research organization outsourced by the sponsor. The clinical research associate is required to contact the relevant study staff member of the sponsor immediately. After receiving the relevant information, the relevant study staff member of the sponsor will discuss with the investigator to decide whether or not to unblind and will document the decision made. After discussing with the sponsor, the investigator will obtain the investigational product information of the relevant subject, check and print the email for notification of unblinding, and keep it in the investigator file. In the case that the sponsor cannot be reached immediately, the investigator will open the emergency unblinding envelope kept in the file, obtain the investigational product information of the relevant subject, write the date and time of opening on the emergency unblinding envelope, sign, and keep it in the investigator file. In this case, the investigator will notify the sponsor of the unblinding of double-blinding as soon as possible and document the reason for unblinding without having a discussion with the sponsor.

If the investigator becomes aware of the subject's code during the course of the study, the investigator will endeavor to eliminate bias during the efficacy and safety evaluation.

In the case that a serious adverse event such as death occurs, regardless of the unblinding status of the investigator, the IDMC may have access to the information of the group to which the relevant subject is assigned and evaluate the adverse event, if deemed necessary by the sponsor.

After the end of the study, problems will be resolved through discrepancies (queries) of all data. Once the database is confirmed to be complete and accurate, the data will be locked, and the randomization code information will be disclosed. Subsequent changes to the database can only be made with the written consent of the sponsor and the database manager.

11.7 Concomitant medication administration criteria

11.7.1 Drugs required for concomitant treatments

This is a comparative clinical study between the monotherapy group in which the standard treatment for persistent bacteremia caused by *Staphylococcus aureus* was administered and the concomitant therapy group in which the study drug N-Rephasin® SAL200 was administered in addition to the standard treatment, and it will be conducted using the concomitant treatment with the treatments shown below, which are the standard treatments for persistent bacteremia caused by *Staphylococcus aureus*.

Standard treatment type	Active ingredient	Method of administration
1. MSSA	**nafcilin**	1-2 g IV q4-6h
	cefazolin	1-2 g IV q8h
	cefepime	1-2 g IV q8-12h
	ceftriaxone	1-2 g IV q12-24h
	cefotaxime	1-2 g IV q6-8h
	vancomycin	1 g IV q12h (to be adjusted based on the renal function)
	teicoplanin	400 mg (6 mg/kg) IV q12h × 3, then 400 mg (6 mg/kg) IV q24h
	linezolid	600 mg IV or PO q12h
	rifampin	450-600 mg PO qd
2. MRSA	**vancomycin**	1 g IV q12h (to be adjusted based on the renal function)
	teicoplanin	400 mg (6 mg/kg) IV q12h × 3, then 400 mg (6 mg/kg) IV q24h
	linezolid	600 mg IV or PO q12h
	rifampin	450-600 mg PO qd

11.7.2 Medications and treatments allowed to be administered and performed concomitantly

During this clinical study period, drugs and treatments will not be prohibited separately except for the items in Section "11.7.3" below. In addition, drugs that the subject has been taking before participating in this clinical study can be continued and their dosage and dosing regimen can be changed even after participating in the clinical study, and new drugs and treatments started during the participation period will not be prohibited separately. However, the types and increases/decreases in doses of the drugs and treatments should be documented accurately.

11.7.3 Medications and treatments prohibited from being administered and performed concomitantly

The below drugs and treatments will be prohibited during this clinical study period.

- Investigational products other than for this clinical study
- In the case that the investigator determines that other drugs and treatments may significantly affect the result of the clinical study

11.8 Discontinuation of the administration

Subjects may withdraw their consent at any time without a justifiable reason. In addition, the investigator may discontinue the administration, if the subject's clinical condition is deemed difficult to proceed with the clinical study. Similarly, if the sponsor or relevant government agencies find situations that require discontinuation of the clinical study, they may require early termination. Reasons for discontinuation of administration include the following:

1) Withdrawal of consent by the subject
2) In the case that the investigator determines that it is difficult to proceed with the clinical study due to any other reasons (such as serious adverse events)

3) Medications and treatments prohibited from being administered and performed concomitantly
4) Deviation from the inclusion/exclusion criteria (in the case that the blood culture obtained after the investigational product administration is infected with other bacterial organisms, the subject will not be dropped out due to the criterion "Those who are infected with mixed bacterial species.")
5) In the case that the subject does not follow the specified treatment schedule

In the case of discontinuation of the subject's drug administration due to the above reasons, the reason for discontinuation and the date of implementation should be recorded in the case report form, medical record, etc. of the subject.

12. Observation items, clinical test items, and observation test methods

12.1 Observation items

Once the eligibility of the subject based on the inclusion and exclusion criteria is confirmed through screening tests, the investigational product will be supplied to conduct the clinical study. The planned visits and the plan for tests and assessments required at each visit are as shown in [Table 4] Clinical study schedule table.

The subject will participate in the clinical study for a total of 15 days (screening window period: ±1 day) except for the follow-up (W4 ± 5 days). In the case of a subject who complete the study early, the subject will participate in the clinical study from the day of screening until the day of early completion (screening window period: ± 1 day). In the case of the follow-up (W4 ± 5 days), telephone monitoring will be performed mandatorily, and if the subject is able to make a visit, the subject will visit the hospital as an outpatient to have the test items performed.

Visits other than the treatment visits will be documented in the unscheduled visit CRF, and the purpose of visit and the results processed will be documented. Data obtained from these additional visits will be recorded in source data as well as in the case report form. If this is reported to the investigator, it should be recorded in the section for the occurrence of adverse events that must always be recorded, or in the case of discontinuation of this study, it should be recorded in the relevant section in the case report form.

In the case of early termination of the study, the subject will be asked to make the final visit, if possible, to have the procedures for the follow-up (W4 ± 5 days) performed and complete the relevant pages in the case report form that include an end-of-study form and a confirmation statement form. The primary reason for dropping out (adverse event, noncompliance with the protocol, or other reasons) should be specified in the case report form. The end-of-study form should be completed even if the subject is not able to make the final visit.

12.1.1 Demographic information

The demographic information of subjects, such as the name, gender, and age, will be investigated.

12.1.2 Medical history and medication history

- Medical history: Medical/surgical history, comorbidities, drug hypersensitivity, and history of surgery (within the last 1 year)
- Medication history: The drugs that have been taken since 1 month prior to screening will be investigated.

12.1.3 Vital signs

- Vital signs: Blood pressure, pulse, and temperature

12.1.4 Physical examination

- Physical examination: This test will use history taking, inspection, palpation, etc. to examine the general conditions, skin/mucous membrane, eyes, otorhinolaryngological system, cardiovascular system, respiratory system, gastrointestinal system, genitourinary system, nervous system, musculoskeletal system, lymphatic system, obstetric and gynecological system, and other body organs. Physical examination will be performed at screening and Day 2, and it will be performed at an interval of once in 2 days after Day 2—Days 4, 6, 8, 10, 12, and 14. It will be performed until negative conversions are confirmed (treatment completion) in blood culture/evaluation at 2 days consecutively.

12.1.5 Laboratory tests and pregnancy test

- Laboratory tests: Hematology, blood biochemistry, urinalysis

  Laboratory tests will be performed at screening and Day 2, and will be performed at Day 7 (± 48 hours) and Day 14 (± 48 hours) after Day 2.

Hematology	:	WBC, RBC, Hemoglobin, Platelets
Blood chemistry	:	Total protein, Albumin, Total bilirubin, AST, ALT, ALP, Total cholesterol, Serum creatinine
Urinalysis	:	Protein, Glucose, Blood, Ketones

* In the case that negative conversions (treatment completion) are confirmed at 2 consecutive days before Day 7, the treatment schedule (Day 1 to Day 14) will be completed after performing these test items.

- Pregnancy test: A pregnancy test (serum/urine HCG) will be included in the case of women of childbearing potential.

12.1.6 Anaphylaxis test and inflammatory cytokine test

The tests will be outsourced to an external laboratory in order to examine the anaphylaxis and inflammatory cytokine aspects of the investigational product, and blood samples for these tests will be collected and stored (when collecting blood samples for anaphylaxis testing, use a blood collection tube that contains Futhan [FUT-175], which is a protease inhibitor). The anaphylaxis test and inflammatory cytokine test will be performed at Day 1 (before the investigational product administration and immediately after the end of the administration), Day 2, and Day 7 (± 48 hours).

- Anaphylaxis test: C3a, C4a, mast cell tryptase
- Inflammatory cytokine test: IL1b, IL-2, IL-6, TNF-α

* In the case that negative conversions (treatment completion) are confirmed at 2 consecutive days before Day 7, the treatment schedule (Day 1 to Day 14) will be completed after performing these test items.

12.1.7 Skin test

The study drug (N-Rephasin® SAL200) will be diluted in normal saline at a concentration of approximately 0.05% (v/v) in order to make a test solution. Using this solution, a skin test will be performed on one arm.

Subjects with an unusual response will drop out from the clinical study, and only those who are negative according to the acceptance criteria may be enrolled in this clinical study.

1) Injection site: Inside the forearm
2) Method of injection: Place a 26G needle with bevel up, and puncture at an angle of 10 to 15 degrees, but only insert the bevel of the needle under the skin. Administer the minimum amount (100 μL at a concentration of approximately 0.05% [v/v]) of the diluted solution intradermally to test skin sensitization. Observe skin redness, etc. 15 minutes after the injection, and evaluate based on the acceptance criteria.
3) Acceptance criteria

(a) If a wheal of at least 3 mm diameter appears along with redness: Positive
(b) If there is a wheal of at least 3 mm diameter but no redness: Undetermined and re-test required
(c) If the diameter of a wheal is 3 mm or smaller regardless of redness: Negative
(d) Retest if the result is equivocal

12.1.8 Blood culture/evaluation (blood collection)

- Blood cultures: Collection of blood samples for blood cultures will be carried out based on the following schedule: the collection at Day 2 will be carried out to fulfill 18 hours (± 6 hours) from the administration of the investigational product, the collection at Days 3–8 will be carried out to fulfill 24 hours (± 6 hours) from collection of blood samples for blood cultures for the previous time point (once daily), and the collection at Days 8–14 will be carried out to fulfill 48 hours (± 6 hours) from collection of blood samples for previous blood cultures (once in 2 days). Collection of blood samples for blood cultures will be performed throughout the clinical study period until negative conversions (treatment completion) are confirmed at 2 consecutive days.

  Two sets of blood samples for blood cultures will be collected (in principle, there should be an interval of at least 30 minutes between 2 sets of blood samples, but the time difference is not required, if blood samples are collected from different parts).

  1) Select a blood vessel at the site of blood collection, wrap the part with a rubber band, and unwrap after checking the vein. Wrap the rubber band at 10 cm above the site of collection, and do not wrap it too tight and longer than 2 minutes.
  2) Wipe the area around the skin at the site of blood collection thoroughly with a 70% alcohol swab.
  3) Apply 2% povidone iodine to the area that was wiped with an alcohol swab.
  4) After 2 minutes, draw 10 mL of blood without touching the disinfected area.
  5) After removing the injection needle, press the area with an alcohol swab.
  6) Add 5 mL aseptically to an aerobic culture bottle for culturing.
  7) Add 5 mL aseptically to a 50-mL anaerobic culture bottle for culturing.
  8) Record the subject's initials, enrollment number, date, time, and site of collection on the culture bottle.

- Evaluation of blood cultures:
  1) If the organism is growing during the culture period, an automated device will sound an alarm, and at this time, the growth of the organism can be checked in the laboratory. The laboratory will confirm that the bacterium growing is a gram-negative organism or a gram-positive organism, and it will be reported in the electronic medical record and informed to the medical staff. Thus, the culture status in the automated device will be checked frequently.
  2) The usual blood culture period is 2 to 3 days.

12.2 Clinical study schedule

[Table 4] Clinical study schedule table

Observation item		Screening[10]	Treatment				Follow-up[11]
		D-1	D1[12]	D2	D3~D13	D14	4W ± 5d
Written consent		√					
Checking on the inclusion/exclusion criteria		√					
Randomization[1]			√				
History of illness		√					
Data on demographic statistics		√					
Vital Signs[2]		√	√[2]	√	√	√	(√)
Physical examination[3]		√		√	√[3]	√	(√)
Laboratory tests[4, 13]	Hematology	√		√	√[4]	√	(√)
	Blood chemistry	√		√	√[4]	√	(√)
	Urinalysis	√		√	√[4]	√	(√)
Anaphylaxis test[5, 13]			√	√	√[5]		
Inflammatory cytokine test[5, 13]			√	√	√[5]		
Pregnancy test[6]		√					(√)
Skin reaction test		√					
Administration of the investigational product[7]			√				
Administration of drugs required for concomitant treatments[8]			√	√	√	√	
Blood culture/evaluation (blood collection)[9]				√	√[9]	√	
Checking on medication history/concomitant medications		√	√	√	√	√	√
Checking on concomitant treatments			√	√	√	√	√
Adverse events			√	√	√	√	√

1. Subjects who have passed screening will be assigned a subject number via randomization.
2. Vital signs will be measured once daily during the remaining treatment period except for Day 1 (including screening), and they will be measured 12 times at the following time points at Day 1:
   - Before administration of the investigational product (1 time)
   - 15 and 30 minutes after initiation of the investigational product administration (2 times)
   - Immediately after the end of the investigational product administration, 30 minutes, 1, 2, 3, 4, 8, 16, and 24 hours after the administration (9 times)
3. Physical examination will be performed at screening and Day 2, and it will be performed at an interval of "once in 2 days" after Day 2 (Days 4, 6, 8, 10, 12, and 14). It will be performed only until when the results of the blood culture/evaluation at 2 consecutive days are confirmed (treatment completion) as negative conversions.

4. Laboratory tests will be performed at screening and Day 2, and will be performed at Day 7 (± 48 hours) and Day 14 (± 48 hours) after Day 2.
   - Hematology : WBC, RBC, Hemoglobin, Platelets
   - Blood biochemistry : Total protein, Albumin, Total bilirubin, AST, ALT, ALP, Total cholesterol, Serum creatinine
   - Urinalysis : Protein, Glucose, Blood, Ketones
5. The anaphylaxis test and inflammatory cytokine test will be performed at Day 1 (before the investigational product administration and immediately after the end of the investigational product administration), Day 2, and Day 7 (± 48 hours).
   - Anaphylaxis test: C3a, C4a, Mast Cell Tryptase
   - Inflammatory cytokine test: IL-1b, IL-2, IL-6, TNF-α
6. A pregnancy test will be performed as a urine test or a serum hCG test only for women of childbearing potential.
7. The enrolled subjects will receive administration of the investigational product at Day 1.
8. Administration of drugs required for concomitant treatment will be among the drugs in "11.7.1 Drugs required for concomitant treatments" as determined by the investigator.
9. Collection of blood samples for blood cultures at Day 2 will be carried out to fulfill 18 hours (± 6 hours) from the administration of the investigational product, collection of blood samples for blood cultures at Days 3–8 will be carried out to fulfill 24 hours (± 6 hours) from collection of blood samples for blood cultures for the previous time point (once daily), and collection of blood samples for blood cultures at Days 8–14 will be carried out to fulfill 48 hours (± 6 hours) from collection of blood samples for previous blood cultures (once in 2 days). Collection of blood samples for blood cultures will be performed throughout the clinical study period until negative conversions (treatment completion) are confirmed at 2 consecutive days.
10. In the case that a subject has records of the relevant laboratory tests 7 days before screening, patient records may be used without performing additional laboratory tests at screening.
11. The tests for the relevant date will be performed when the subject visits for a follow-up at W4 ± 5 days, and concomitant medications, concomitant treatment, and adverse events will be checked via telephone monitoring. In the case of the subjects who can visit the hospital as outpatients, vital signs, physical examination, and laboratory tests will be performed, and concomitant medications, concomitant treatments, and adverse events will be checked as well.
12. Subjects who are suitable for the subject eligibility can have procedures for Day −1 and Day 1 simultaneously.
13. In the case that negative conversions (treatment completion) are confirmed at 2 consecutive days before Day 7, laboratory tests, anaphylaxis test and inflammatory cytokine test will be performed additionally.

12.3 Visit schedule

The test procedures based on the clinical study schedule are as shown below, and during this participation period, subjects will continue the administration of conventional drugs for the "standard treatment" used to treat bacteremia.

12.3.1 Screening (Day −1)

- After providing a detailed explanation of this clinical study prior to participation of the patient, obtain a written consent, and assign the screening no. in the order of enrollment.
- Investigate the demographic data and medical history (for the medical history, obtain the items within 1 year prior to participation in the clinical study).
- Examine vital signs (blood pressure, pulse, and temperature).
- Perform a physical examination.
- Medication history: Investigate the drugs taken by the subject within 1 month of obtaining the informed consent form (all drugs including oral contraceptives).
- Obtain blood samples for laboratory testing and urine or blood samples for urinalysis and pregnancy testing, and investigate the observation items (perform a pregnancy test in female patients who may be pregnant or who have been postmenopausal for less than a year, and enroll only those who are negative as subjects).

Hematology	:	WBC, RBC, Hemoglobin, Platelets
Blood chemistry	:	Total protein, Albumin, Total bilirubin, AST, ALT, ALP, Total cholesterol, Serum creatinine
Urinalysis	:	Protein, Glucose, Blood, Ketones

- Perform a skin test to check the sensitivity to the study drug (N-Rephasin® SAL200).
- Check the inclusion/exclusion criteria for eligibility, and perform procedures in the next visit schedule in only the eligible subjects.
- To use for randomization, identify the type of infecting organism (MRSA, MSSA) and the route of infection in the eligible subjects.
  - ➔ The procedures for the day of the screening visit (Day −1) and Day 1 can be performed together, and the clinical study can be initiated on the same day as determined by the principal investigator.

12.3.2 Administration of the investigational product (Day 1)

The subject will receive the investigational product and will proceed with the study in the following order:

- Assign the randomization numbers to the eligible subjects based on the inclusion/exclusion criteria at screening.
- Examine vital signs (blood pressure, pulse, and temperature) at the time intervals stated below. (12 times in total)
  - Before administration of the investigational product (1 time),
  - 15 and 30 minutes after initiation of the investigational product administration (2 times),
  - Immediately after the end of the investigational product administration (1 time),
  - 30 minutes, 1, 2, 3, 4, 8, 16, and 24 hours after the end of the investigational product administration (8 times)

- Before administration of the investigational product and immediately after the end of the investigational product administration, collect and store blood samples for anaphylaxis testing and inflammatory cytokine testing (when collecting blood samples for anaphylaxis testing, use a blood collection tube that contains Futhan [FUT-175], which is a protease inhibitor).

  Anaphylaxis test: C3a, C4a, mast cell tryptase
  Inflammatory cytokine test: IL-1b, IL-2, IL-6, TNF-α

- Administer the investigational product (N-Rephasin® SAL200 or placebo) by intravenous drip infusion over approximately 60 minutes.
- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.
  * Adverse events will be collected from the time of initiation of the investigational product administration until the follow-up visit, and concomitant medications and concomitant treatments will be collected from within 1 month prior to obtaining the informed consent form until the follow-up visit (W4 ± 5 days).

12.3.3 Follow-up observation period (Day 2 to Day 14)

▶ Day 2

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment and have follow-up observation until Day 14 or when the results of the blood culture at 2 consecutive days (e.g., Day 2 and Day 3, or Day 8 and Day 10) are confirmed as negative conversions (treatment completion), and the subjects will proceed with in the following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- Perform a physical examination.
- Obtain blood and urine samples for laboratory testing, and investigate the observation items.

Hematology	:	WBC, RBC, Hemoglobin, Platelets
Blood chemistry	:	Total protein, Albumin, Total bilirubin, AST, ALT, ALP, Total cholesterol, Serum creatinine
Urinalysis	:	Protein, Glucose, Blood, Ketones

- Collect and store blood samples for anaphylaxis testing and inflammatory cytokine testing (when collecting blood samples for anaphylaxis testing, use a blood collection tube that contains Futhan [FUT-175], which is a protease inhibitor).

  Anaphylaxis test: C3a, C4a, Mast Cell Tryptase
  Inflammatory cytokine test: IL-1b, IL-2, IL-6, TNF-α

- At the time that 18 hours (± 6 hours) from the administration of the investigational product is satisfied, examine for the persistence of bacteremia by performing blood cultures using 2 sets of blood samples (10 mL for each sample) collected from the subject.
- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.

▶ **Day 3**

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment and have follow-up observation until Day 14 or when the results of the blood culture at 2 consecutive days are confirmed as negative conversions (treatment completion), and the subjects will proceed with the study in the following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- At the time that 24 hours (± 6 hours) from the "time of blood collection at Day 2" is satisfied, examine for the persistence of bacteremia by performing blood cultures using 2 sets of blood samples (10 mL for each sample) collected from the subject.
- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.

▶ **Day 4**

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment and have follow-up observation until Day 14 or when the results of the blood culture at 2 consecutive days are confirmed as negative conversions (treatment completion), and the subjects will proceed with the study in the following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- Perform a physical examination.
- At the time that 24 hours (± 6 hours) from the "time of blood collection at Day 3" is satisfied, examine for the persistence of bacteremia by performing blood cultures using 2 sets of blood samples (10 mL for each sample) collected from the subject.
- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.

▶ **Day 5**

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment and have follow-up observation until Day 14 or when the results of the blood culture at 2 consecutive days are confirmed as negative conversions (treatment completion), and the subjects will proceed with the study in the following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- At the time that 24 hours (± 6 hours) from the "time of blood collection at Day 4" is satisfied, examine for the persistence of bacteremia by performing blood cultures using 2 sets of blood samples (10 mL for each sample) collected from the subject.
- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.

▶ **Day 6**

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment and have follow-up observation until Day 14 or when the results of the blood culture at 2 consecutive days are confirmed as negative conversions (treatment completion), and the subjects will proceed with the study in the

following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- Perform a physical examination.
- At the time that 24 hours (± 6 hours) from the "time of blood collection at Day 5" is satisfied, examine for the persistence of bacteremia by performing blood cultures using 2 sets of blood samples (10 mL for each sample) collected from the subject.
- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.

▶ **Day 7**

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment and have follow-up observation until Day 14 or when the results of the blood culture at 2 consecutive days are confirmed as negative conversions (treatment completion), and the subjects will proceed with the study in the following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- At the time that 24 hours (± 6 hours) from the "time of blood collection at Day 6" is satisfied, examine for the persistence of bacteremia by performing blood cultures using 2 sets of blood samples (10 mL for each sample) collected from the subject.
- Obtain blood and urine samples for laboratory testing, and investigate the observation items.

Hematology:	WBC, RBC, hemoglobin, platelets
Blood chemistry:	Total Protein, Albumin, Total Bilirubin, AST, ALT, ALP, Total Cholesterol, Serum Creatinine
Urinalysis:	Protein, Glucose, Blood, Ketone

- Collect and store blood samples for anaphylaxis testing and inflammatory cytokine testing (when collecting blood samples for anaphylaxis testing, use a blood collection tube that contains Futhan [FUT-175], which is a protease inhibitor).

Anaphylaxis test:	C3a, C4a, mast cell tryptase
Inflammatory cytokine test:	IL-1b, IL-2, IL-6, TNF-α

- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.

▶ **Day 8**

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment and have follow-up observation until Day 14 or when the results of the blood culture at 2 consecutive days are confirmed as negative conversions (treatment completion), and the subjects will proceed with the study in the following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- At the time that 24 hours (± 6 hours) from the "time of blood collection at Day 7" is satisfied, examine for the persistence of bacteremia by performing blood cultures using 2 sets of blood samples (10 mL for each sample) collected from the subject.
- Perform a physical examination.
- Perform the required concomitant treatments.

- Investigate the adverse events, concomitant medications, and concomitant treatments.

▶ **Day 9**

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment, and the subjects will proceed with the study in the following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.

▶ **Day 10**

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment and have follow-up observation until Day 14 or when the results of the blood culture at 2 consecutive days are confirmed as negative conversions (treatment completion), and the subjects will proceed with the study in the following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- Perform a physical examination.
- At the time that 48 hours (± 6 hours) from the "time of blood collection at Day 8" is satisfied, examine for the persistence of bacteremia by performing blood cultures using 2 sets of blood samples (10 mL for each sample) collected from the subject.
- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.

▶ **Day 11**

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment, and the subjects will proceed with the study in the following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.

▶ **Day 12**

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment and have follow-up observation until Day 14 or when the results of the blood culture at 2 consecutive days are confirmed as negative conversions (treatment completion), and the subjects will proceed with the study in the following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- Perform a physical examination.
- At the time that 48 hours (± 6 hours) from the "time of blood collection at Day 10" is satisfied, examine for the persistence of bacteremia by performing blood cultures using 2 sets of blood samples (10 mL for each sample) collected from the subject.
- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.

▶ **Day 13**

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment, and the subjects will proceed with the study in the following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.

▶ **Day 14 (time to determine treatment success/failure)**

The subjects will receive treatment for bacteremia with standard treatments until the completion of treatment, and the subjects will proceed with the study in the following order:

- Examine vital signs (blood pressure, pulse, and temperature).
- Perform a physical examination.
- At the time that 48 hours (± 6 hours) from the "time of blood collection at Day 12" is satisfied, examine for the persistence of bacteremia by performing blood cultures using 2 sets of blood samples (10 mL for each sample) collected from the subject. At Day 14, collect the last blood sample for a blood culture, and if negative conversion rates are not confirmed in the blood culture/evaluation at Day 12 and Day 14, consider it as a failure to treat *S. aureus* bacteremia.
- Obtain blood samples for laboratory testing and urine samples for urinalysis, and investigate the observation items.

Hematology	:	WBC, RBC, Hemoglobin, Platelets
Blood chemistry	:	Total protein, Albumin, Total bilirubin, AST, ALT, ALP, Total cholesterol, Serum creatinine
Urinalysis	:	Protein, Glucose, Blood, Ketones

- Perform the required concomitant treatments.
- Investigate the adverse events, concomitant medications, and concomitant treatments.
- Evaluate the clinical outcome of the treatment (clinical success or failure).

12.3.4 Follow-up (W4 ± 5 days)

If the subject is unable to make a visit as an outpatient for this follow-up, adverse events and concomitant medications will be checked via telephone monitoring. If the subject is able to make a visit, measurement of vital signs, physical examination, and laboratory tests can be performed if necessary through an outpatient visit. In the case of subjects who are unable to make an outpatient visit, telephone monitoring will be carried out, and at this time, the reason for not being able to make a visit will be documented (even in the case of death or inability to make a visit due to other diseases, the relevant subjects will not be dropped out from this clinical study).

The following tests will be performed via telephone monitoring:

- Investigate the adverse events, concomitant medications, and concomitant treatments.

In the case of subjects who are able to make an outpatient visit, laboratory tests, vital sign measurement, and physical examination will be carried out if necessary in the following order:

- Investigate the adverse events, concomitant medications, and concomitant treatments.
- Obtain blood samples for laboratory testing and urine samples for urinalysis, and investigate the observation items.

Hematology	:	WBC, RBC, Hemoglobin, Platelets
Blood chemistry	:	Total protein, Albumin, Total bilirubin, AST, ALT, ALP, Total cholesterol, Serum creatinine
Urinalysis	:	Protein, Glucose, Blood, Ketones

- Examine vital signs (blood pressure, pulse, and temperature).
- Perform a physical examination.

Subjects who drop out at any time during the study will make the termination visit, and the reason for dropping out will be documented.

12.4 Unscheduled visit and hospitalization

In the case that the subject is visiting or hospitalized on a day that is not the scheduled date, adverse events related to the study, changes in the concomitant medications, dropout status, outcome of the clinical measurement, cases requiring concomitant treatment, etc. should be recorded. The clinical study schedule should not be changed due to unscheduled visits. At this time, visits scheduled before the initiation of the clinical study are not unscheduled visits.

13. Expected side effects and precautions for use

(1) Adverse events found in the results of the nonclinical studies

The study drug used in this clinical study is a new drug of a new class, and the absence of drugs of the same or similar class makes it difficult to infer side effects. However, as a result of the nonclinical studies and the Phase 1 clinical study, the adverse events shown below have been reported.

① Rodents

a. Single-dose study

- According to the result of a single-dose toxicity study in SD rats, no abnormal findings related to the study drug, such as death, general abnormal symptoms, weight changes, and abnormal necropsy findings, were observed in all observed items.

b. 2-week repeated dose selection study

- Urine volume increased, neutrophil decreased, eosinophil decreased, monocyte decreased, basophil decreased, lymphocyte increased, TBIL increased, GGT increased, spleen size increased, thymus size increased, seminal vesicle weight increased, pituitary gland weight increased

c. 4-week repeat-dose study

- TCHO increased, PL increased, TG increased, ALB decreased, ALP decreased, proteinuria; edematous findings in the pancreas, skeletal muscles or hind limbs; edema according to histopathological examination; kidney weight increased, renal tubule dilated, hyaline droplets, cast, and increased hypertrophic zone, middle physis, tibia

② Beagle dogs

a. 2-week repeated dose selection study

- Subdued behavior, prone position, respiration rate decreased, vomiting, hypersalivation, ocular pigmentation, skin pigmentation, WBC increased, platelet decreased, basophil increased, LUC increased, reticulocyte decreased, neutrophil decreased, monocyte increased, leukocyte increased, BUN decreased, TP decreased, TBIL decreased, CK decreased, dark red discoloration at the site of administration, mild renal tubular degeneration/regeneration

b. 4-week repeat-dose study

- Vomiting, prone position, lateral recumbent position, subdued behavior, irregular breathing, respiration rate increased, feed consumption decreased, rubbing, hypersalivation, platelet decreased, dark red discoloration at the site of administration, relative weight of the kidney increased, mild to severe congestion/hemorrhage, very mild to mild vasculitis/perivasculitis, very mild to moderate edema and inflammatory cell infiltration

③ Monkeys

a. DES study

- BUN level increased, creatinine level increased

(2) Adverse events found in the results of the Phase 1 clinical study

A total of 42 adverse events were reported in 12 out of 36 subjects who received the investigational product during the Phase 1 clinical study. All adverse events recovered without sequelae, serious adverse events did not occur, and none of the subjects were discontinued from the study due to adverse events.

a. 0.3 mg/kg dose group

- Total bilirubin increased

b. 10 mg/kg dose group

- Rigors, fatigue, headache, myalgia, tenderness, nausea, dyspepsia

14. Discontinuation and dropout, and progress and early termination of the clinical study

14.1 Discontinuation and dropout criteria for individual subjects

Subjects have the right to drop out from the clinical study at any time and for any reason without prejudice to subsequent treatment. The investigator also has the authority to drop out subjects from the clinical study for the benefit of subjects. Unnecessary dropouts should be avoided, as an excessive dropout rate can result in the results of the clinical study being unable to be interpreted.

If the clinical study is discontinued prior to termination of the clinical study, the reason for discontinuation should be recorded in the clinical study termination report in the case report form, and the case report form should be completed.

In all cases, the reason for dropout and the date of dropout should be recorded in the CRF and the subject's medical records. It should be followed up whether the reason for dropout is due to adverse events, and if so, it should be reported in accordance with the processes described in Section 16.7 "Documentation of adverse events and serious adverse events." For example, if a subject withdraws his or her consent to participate in the study due to recurrence of a disease during treatment, "recurrence of the disease" should be recorded in the case report form as a reason for dropping out. All subjects who received the clinical drug but did not complete the study in accordance with the

protocol should have all tests scheduled for the final visit completed as much as possible. The investigator should make best efforts to contact a subject who is lost to follow-up. Attempts to contact such a subject should be recorded in the subject's medical records (e.g., the date and time of a call, a receipt for sending a registered mail).

The subject's participation in the clinical study may be discontinued for the following reasons:

1) If it is determined that the situation observed while conducting the clinical study is unreasonable to continue the clinical study, the principal investigator should request discontinuation of the clinical study to the Institutional Review Board and is able to discontinue the clinical study based on the decision made by the Institutional Review Board.
2) If the sponsor wants to discontinue the clinical study due to reasons such as the safety of the investigational product, the sponsor can request discontinuation of the clinical study to the Institutional Review Board and discontinue the clinical study based on the decision made by the Institutional Review Board.

The subject may drop out from participation in the clinical study for the following reasons:

1) In the case that the subject (or the authorized representative) withdraws the consent for participation in the clinical study
2) In the case that surgery or other drugs that may affect the evaluation of the safety and efficacy have been carried out or used concomitantly
3) In the case that the investigator determines that serious adverse events may occur and may cause disadvantages to the subject's health status
4) In the case that the efficacy evaluation items cannot be observed continuously due to the absence of the subject
5) In the case that the subject dies for reasons unrelated to the clinical study
6) In the case that the sub-investigator determines that there are other problems in conducting the clinical study
7) In the case that the subject discontinues contraception due to pregnancy or with an intention to become pregnant
8) In the case that the subject has an exclusion criterion occurred during the clinical study period, the relevant subject may be excluded from the clinical study (in the case that the blood culture obtained after the investigational product administration is infected with other organisms, the subject will not be dropped out due to the criterion "Those who are infected with mixed bacterial species").

14.2 Entry from Step 1 to Step 2

Proceeding to Step 2 will be determined considering the evaluation results of the IDMC. The IDMC will establish the charter and the standard operating procedures separately for its operations and will determine the safety and efficacy observed in Step 1 in accordance with the standard operating procedures established.

An interim analysis to evaluate whether to enter Step 2 will be performed by the IDMC when the number of subjects who have terminated the clinical study participation has reached 28 or more and 32 or less, and the safety will be evaluated to determine whether to enter Step 2.

14.3 Early termination of the clinical study

The clinical study may be terminated early for the reasons stated below. The reasons stated below are the cases when new information regarding the benefit/risk ratio for subjects of the investigational product obtained has a negative direction and include the following:

1) In the case that conducting the clinical study further after the interim analysis is considered meaningless
2) In the case that new and important adverse drug reactions have occurred or the incidence and severity of past adverse drug reactions are unexpectedly significant
3) Other safety reasons that negatively affect the benefit/risk ratio of the clinical study
4) In the case that the sponsor determines that continuation of this clinical study is not justifiable from medical and ethical perspectives
5) In the case that it is determined that the subject enrollment rate at the relevant study site will cause a significant setback of the subject enrollment schedule required for this clinical study
6) In the case that the sale or supply of investigational product is discontinued

If this clinical study is terminated early, a government regulatory agency or the Institutional Review Board will be notified in accordance with applicable regulations.

The clinical study may be discontinued or suspended if requested by the government regulatory agency.

14.4 Replacement of subjects who drop out

Subjects who are terminated early from this clinical study will not be replaced.

15. Effectiveness evaluation criteria, and methods of evaluation and interpretation

15.1 Primary endpoints

▶ Safety endpoints

- Checking on adverse events
- Laboratory tests
- Anaphylaxis test, inflammatory cytokine test
- Physical examination/vital signs

15.2 Secondary endpoints

▶ Efficacy Endpoint ①

- Proportion of patients whose first blood culture performed after administration of the investigational product is negative

▶ Efficacy Endpoint ②

- Proportion of patients who die due to *S. aureus* bacteremia by Day 14

▶ Efficacy Endpoint ③

- Proportion of treatment failure of *S. aureus* bacteremia by Day 14 (if 2 consecutive negative conversions were not observed in blood cultures performed until Day 14)

15.3 Statistical analysis methods

15.3.1 Demographic baseline data

The background and demographic data of the subjects included in this clinical study will be evaluated by treatment group, and the mean, standard deviation, minimum value, maximum value, etc. will be obtained and presented for continuous variables, and the frequency and percentage will be presented for categorical variables.

15.3.2 Primary endpoint analysis method

Safety analysis is performed in the safety set. A distribution table of the number of subjects who experienced at least one side effect (incidence) and distribution tables of the relationship of the investigational product to the reported adverse events (distribution tables for severity and the relationship to the drug) will be presented for each group (study group, control group) to confirm the safety.

The results of the laboratory tests, anaphylaxis test, inflammatory cytokine test, and vital signs at baseline and the last visit will be summarized as mean values and standard deviations, and the changes before and after the treatment within each group will be confirmed. Categorical data will be divided into normal and abnormal data and summarized as frequency and percentage, and the differences before and after treatment within each group will be confirmed.

15.3.3 Secondary endpoint analysis methods

15.3.3.1 Efficacy Endpoint ①

- Proportion of patients whose first blood culture performed after administration of the investigational product is negative

  The descriptive statistics for the proportion of patients who are negative in the first blood culture after administration of the treatment (the rate of negative conversion) will be presented by treatment group. Whether the rate of negative conversion in the study group is superior to that in the control group will be evaluated in a descriptive statistical manner.

15.3.3.2 Efficacy Endpoint ②

- Proportion of patients who die due to *S. aureus* bacteremia at Day 14 after the onset of bacteremia (according to the first confirmatory decision of bacteremia). The descriptive statistics for the rate of mortality due to *S. aureus* bacteremia by Day14 will be presented and evaluated by treatment group.

15.3.3.3 Efficacy Endpoint ③

- Proportion of treatment failure of *S. aureus* bacteremia by Day 14 (if 2 consecutive negative conversions were not observed in blood cultures performed until Day 14)

  The descriptive statistics for the treatment failure rate of *S. aureus* bacteremia by Day14 will be presented and evaluated.

15.4 Statistical analysis sets

The data obtained from the subjects in this clinical trial will be analyzed largely in three methods: safety set, FAS (full analysis set), and PP (per protocol) set.

15.4.1 Safety set

The safety set is intended for subjects who participated in this clinical study and used the study drug/control drug at least once.

15.4.2 FAS (Full Analysis Set)

The FAS is intended for subjects whose bacteremia (blood culture) test results have been confirmed at least once after using the study drug/control drug at least once.

15.4.3 PP (Per Protocol)

The PP set is intended for subjects who have completed this clinical study in accordance with the protocol among those who are included in the FAS. Among them, those who take medications prohibited from being administered concomitantly, those who receive treatments prohibited from being performed concomitantly, and those who do not comply with the clinical study schedule except for the F/U visit (however, the noncompliance due to death will be included in the PP set) will be excluded.

[Principle of result interpretation]

For data for efficacy evaluation, the FAS analysis will be the main analysis, and the PP analysis will be performed separately to evaluate whether there are differences compared to the PP analysis results.

Data for safety evaluation will be used to evaluate the safety of this study drug/control drug in the safety set.

15.5 Issues related to the statistical analysis and method to handle missing data

SAS Ver 9.2 will be used as an analysis program. If missing values are generated, observed cases will be analyzed.

15.6 Interim analysis

An interim analysis will be performed when the number of subjects who have terminated the clinical study participation has reached 28 or more (but to be performed at or before reaching 32). The main evaluation of the interim analysis is for safety purposes, and the efficacy evaluation will also be performed for exploratory purposes. The interim analysis and evaluation will be performed by the IDMC.

The safety will be evaluated by presenting the frequency and pattern (relationship to the investigational product, severity, seriousness, etc.) for all adverse events that occurred during the clinical study period for each treatment group. The efficacy evaluation will provide proportions for each of the secondary endpoints listed in Section 15.2 and compare them between the treatment groups.

16. Evaluation criteria for safety including side effects, method of evaluation, and reporting method

16.1 Safety evaluation method

① The safety evaluation items will be evaluated according to the clinical study schedule and "15.3.2 Primary endpoint analysis method."

In all subjects, the medical history prior to administration of the investigational product, concomitant medications, laboratory tests, and measurement of vital signs will be performed.

In addition, the sub-investigator will record all outliers, which are considered clinically significant, that meet the criteria in "16.2 Safety evaluation criteria," as adverse events or serious adverse events.

16.2 Safety evaluation criteria

16.2.1 Definition of an adverse event (AE)

An adverse event is any undesired and unintended sign (e.g., abnormality in a laboratory test) or symptom, syndrome, or disease that occurs or worsens in a patient or subject receiving the investigational product during the observation period of the clinical study. This does not necessarily have to have a causality relationship to the drug. In other words, a case that has no causality relationship to the investigational product or the clinical study is included in the use of the term "adverse event."

Clinically significant abnormal results in diagnostic procedures including abnormal findings of laboratory test results (e.g., those requiring unplanned diagnostic procedures/treatments or discontinuation of participation in the clinical study) will be considered as adverse events. Therefore, adverse events can be diseases caused by the use of a drug regardless of any undesirable and unexpected signs (which may be clinically significant abnormal laboratory test results), symptoms or relationship to the drug.

Worsening of signs or symptoms of the disease being treated is usually measured as a safety endpoint. However, if the result is applicable to the definition of a "serious adverse event," it should be recorded according to the procedure (see Section 16.7 "Rules for reporting of serious adverse events").

The surgery processes themselves will not be treated as adverse events. They are treatment options for conditions requiring surgery. In the case that events requiring surgery occur or are detected during the clinical study period, they are adverse events. If the planned surgical procedures allowed in the protocol and the situation(s) that

led to take these measures were known prior to initiation of the clinical study, these are not adverse events. In the latter case, they should be reported as medical histories.

▶ Cases that are included in adverse events
- Disease newly occurred
- Worsening of existing diseases
- Effects caused by the clinical study drugs (including the control drug [placebo]): In the case that preexisting reactions or conditions worsen or their frequencies increase
- Effects caused by the clinical study procedures
- Conditions that are diagnosed or detected after administration of the clinical drug (including the control drug [placebo]) even if it is assumed that they were present before initiation of the study
- In the case that diseases or symptoms observed at the baseline worsen after initiation of the study
- Complex effects of 2 or more of the factors above

▶ Cases that are not included in adverse events
- Temporary pain at the site of blood collection and injection
- Medical or surgical procedures (e.g., surgery, endoscopy, tooth extraction, blood transfusion)
- In the case that an existing disease or a condition that was present or detected at the beginning of the study does not worsen
- Situations where unexpected medical phenomena do not occur (e.g., hospitalization due to cosmetic surgery, hospitalization for personal convenience, etc.)
- Diseases or conditions that have been studied, conditions that the subject's condition is not more severe than expected, or signs or symptoms not related to the diseases
- No signs or symptoms resulting from an overdose of the clinical drug or concomitant medications

16.2.2 Definition of an adverse drug reaction

An adverse drug reaction (ADR) refers to any harmful and unintended reaction that occurred at a certain dose of the investigational product where a causality relationship to the investigational product cannot be denied.

16.2.3 Serious adverse event/adverse drug reaction

A serious adverse event/adverse drug reaction (serious AE/ADR) refers to an adverse event or an adverse drug reaction that occurred at a certain dose of the investigational product that falls under any one of the following:

▶ Cases that are included in serious adverse events
- In the case that results in death or is life-threatening
- In the case that requires hospitalization or prolongation of hospitalization
- In the case that results in permanent or serious disability and dysfunction
- In the case that results in congenital anomaly or birth defect

▶ Cases that are not included in serious adverse events
- Visiting emergency department and staying for within 24 hours
- Admission to a hospital for a day
- Prolongation of hospitalization period or transfer to another hospital for treatment of underlying diseases that have existed before participation in the study (obtaining of the informed consent form)
- Transfer to a rehabilitation hospital or a nursing hospital for conservative management

In the case of a situation where there is a significant impact on the subject's well-being and health status besides those listed above, the study doctor and relevant experts will make a medical judgement whether or not to consider it as a serious adverse event and take appropriate measures accordingly.

16.2.4 Unexpected adverse drug reaction

An unexpected adverse drug reaction refers to a reaction that shows a difference in the characteristics or degree of harm of the adverse drug reaction in view of the available information on the drug such as an investigator's brochure or a package insert of the drug.

16.2.5 Severity of adverse events

The severity of adverse events will be determined based on the below.

1) Mild: Symptoms or signs are perceivable, but they can be tolerated without discomfort, and no treatment is needed to relieve symptoms, if they do not affect behaviors or functions.
2) Moderate: Symptoms are uncomfortable enough to interfere with activities of daily living, they affect behaviors, and treatment may be needed to relieve symptoms.
3) Severe: Symptoms cause discomfort that incapacitates from working or performing activities of daily living, and treatment is needed to relieve symptoms, if discomfort causes discontinuation of a suspect drug.

16.2.6 Causality relationship of adverse events

The investigator must also systematically evaluate the causality relationship between the investigational product and adverse events according to the criteria listed below.

The following items should be considered to evaluate the relationship between the drug and adverse events:

1) Temporal continuity after the drug administration: An adverse drug reaction must occur after the investigational product administration. The temporal interval between the investigational product administration and the onset of an adverse drug reaction should be evaluated according to the clinical circumstances of the event.
2) Recovery after drug dechallenge, and recurrence after drug rechallenge: Subject's response after drug dechallenge and subject's response after drug rechallenge (Drug rechallenge will be considered in the aspect of the usual clinical stage of a problematic incident.)
3) Underlying, comorbid, and intercurrent diseases: Reports of each adverse event will be evaluated based on the characteristics and stages of the disease being treated and other disease that a subject has.
4) Concomitant medications and treatments: Other medications or treatments that a subject is receiving should be evaluated, as they may be related to the problematic event.
5) Trend of known responses to drugs of this class: Clinical/preclinical
6) Pharmacological and pharmacokinetic characteristics of the drug: Pharmacokinetic characteristics (absorption, distribution, metabolism, and excretion) of the investigational product should be considered along with the pharmacodynamic aspects of the subject.

The evaluation of the relationship between the investigational product administration and adverse events (not related, unlikely related, possibly related, probably related, certainly related) will be clinically determined based on all information that can be obtained at the time of completing the case report form. Of these, possibly related,

probably related, and certainly related will be evaluated as adverse drug reactions.

[Table 5] Causality Relationship Assessment

Causality Term	Assessment Criteria
Certainly related	When the temporal order of the investigational product administration and the adverse event onset is plausible When the adverse event is unexplained by other possible causes (such as diseases or other drugs) In the case that discontinuation or dose reduction of the investigational product shows a plausible result of the relationship between the event and the drug (the adverse event disappears or weakens) When an adverse event occurred is known to be definite in the pharmacological and symptomatologic aspects of the drugs in the same class as the investigational product ("gray baby syndrome" after administration of chloramphenicol, anaphylaxis immediately after rechallenge of the previously administered drug, etc.) When the event reappears when rechallenged (if performed)
Probably related	When the temporal order of the investigational product administration and the onset of the adverse event or laboratory abnormality is reasonable When the adverse event is unlikely to be caused by other possible causes (such as diseases or other drugs) When discontinuation or dose reduction of the investigational product shows a clinically valid result (the adverse event disappears or weakens) A result of rechallenge is not mandatory.
Possibly related	When the temporal order of the investigational product administration and the onset of the adverse event or laboratory abnormality is reasonable When the adverse event is believed to be caused by the investigational product as much as by other possible causes (such as diseases or other drugs) When there is no information or information is not apparent upon discontinuation of the investigational product administration
Unlikely related	When the temporal relationship of the investigational product administration and the onset of the adverse event or laboratory abnormality is improbable (but not impossible) When the adverse event is more plausibly explained by other causes (such as diseases or other drugs) than administration of the investigational product
Not related	When the subject has not received the investigational product When the temporal order of the investigational product administration and the onset of the adverse event or laboratory abnormality is impossible When there is a definitive cause (e.g., bleeding at the operative site) that can explain the reason Unreasonable (e.g., when there is no evidence for the loss of a sense of direction due to the investigational product although the patient was hit by a motorcycle, occurrence of cancer a couple days after the administration of the investigational product)

16.2.7 Exacerbation of underlying diseases or health condition that is already present

The expected change or expected exacerbation of the underlying disease or preexisting condition will not be recorded as an AE unless it meets at least one of the following criteria:

- When the exacerbated disease becomes an SAE
- When the investigational product is discontinued, or the dose is decreased or increased
- When the investigator determines that the disease exacerbated from baseline unexpectedly

16.3 Observation period

For the purpose of this clinical study, the duration of adverse event observation will be between the administration of the investigational product and the last visit at 4 weeks ± 5 days.

After the end of the observation period, if the investigator identifies a serious adverse event in the subject and considers that it is an adverse event likely to be related to the previous clinical study treatment or related to the clinical study procedures, the investigator should contact the sponsor to check how the adverse event should be documented and reported.

16.4 Safety evaluation set

It will be carried out only in the subjects who received the investigational product at least once.

16.5 Method, frequency, and time to detect adverse events and serious adverse events

The initiation of the study is the period after screening, and the end of the reporting period means the follow-up visit at W4 ± 5 days after the end of treatment of persistent bacteremia. The investigator should record each adverse event promptly and completely in the case report form, and the investigator should record adverse events, even if they are determined to be unrelated to the study drug treatment by the investigator. If possible, the investigator should try to diagnose based on the symptoms and signs that appear.

16.6 Documentation of adverse events and serious adverse events

All adverse events occurring during the study period will be recorded according to the following:

(1) All adverse events (serious or non-serious, or alert terms) must be recorded in the “Adverse Events” page of the case report form.

(2) If an adverse event is a serious adverse event (see Section 16.2.3), the investigator should complete the “Serious Adverse Event Report” form as well as the “Adverse Events” section of the CRF at the time when the serious adverse event is confirmed. This form should be sent to the clinical research associate, and then, this form will be sent to the pharmacovigilance (PV) staff member of the sponsor by the clinical research associate.

(3) In other cases where expedited reporting is required without an adverse event, the investigator does not need to complete the “Serious Adverse Event Report” form and the “Adverse Events” page of the case report form.

(4) Adverse events should be described using diagnostic terminologies as much as possible. If a definitive diagnosis is made, each signs and symptoms are not required to be recorded unless atypical or extreme symptoms have occurred. Each of atypical or extreme symptoms must be reported as separate adverse events. If a definitive diagnosis cannot be made, signs and symptoms should be recorded separately.

16.7 Rules for reporting of serious adverse events

16.7.1 Expedited reporting of serious adverse events

- In the case of serious adverse events, if the investigator determines that the event corresponds to a "serious adverse event" defined in the protocol, it should be reported promptly to the designated pharmacovigilance staff members designated by the sponsor and to the IRB as specified below. The reported information should be recorded identically in the serious adverse event report and the case report form.

- Submission of serious adverse event reports

The initial notification of all serious adverse events that newly occur during the clinical study period should be submitted immediately after recognizing the occurrence of serious adverse events. The investigator should complete a serious adverse event report within 24 hours of recognition and send via email or fax to the pharmacovigilance staff members. The contact information of the pharmacovigilance staff members is as follows:

▶ Sponsor: iNtRON Biotechnology, Inc.

Person in charge	Eun Ah Park
E - mail	euna0917@intron.co.kr
Address	137, Sagimakgol-ro, Jungwon-gu, Seongnam-si, Gyeonggi-do, Republic of Korea
Postal code	13202
Telephone number	+82-31-739-5032
Fax number	+82-31-736-7246

▶ Contract research organization: Symyoo Inc.

Person in charge	Clinical research associate
Address	2nd floor, 6, Hannam-daero 42-gil, Yongsan-gu, Seoul, Republic of Korea
Postal code	04417
Telephone number	+82-70-4335-5468
Fax number	+82-2-749-2050 +82-2-6442-4753 (eFax)

- In the case that iNtRON Biotechnology, Inc. receives a report of a suspected unexpected serious adverse reactions from the investigator, they should promptly report to the Ministry of Food and Drug Safety (MFDS) within the following deadline:

① Any suspected unexpected serious adverse reactions that cause death or that are life threatening should be reported within 7 days from the day when the sponsor received a report or recognized. If additional information

needs to be reported, additional reports should be submitted including the detailed information on the relevant adverse drug reaction within 15 days from the day when the sponsor received a report or recognized the relevant adverse drug reaction for the first time.

② All other suspected unexpected serious adverse reactions should be reported within 15 days from the day when the sponsor received a report or recognized suspected unexpected serious adverse reactions.

16.7.2 Obligations related to serious adverse events

☞ Reporting of serious adverse events

The obligations of each staff member related to “serious adverse events” that occur during the clinical study period are as follows:

(1) Obligations of the principal investigator

In the case that any serious adverse event occurs during the clinical study, the principal investigator should report it to the sponsor immediately and provide further reports with detailed information later. In the case that a death case has been reported, the principal investigator should provide additional information, such as an autopsy report (only if an autopsy was performed) and a death certificate, to the sponsor and the Institutional Review Board. In addition, the principal investigator should report serious adverse events occurred during the clinical study at the affiliated site or at other sites to the affiliated Institutional Review Board in accordance with the requirements for reporting serious adverse events, which are specified by each study site.

(2) Obligations of the sub-investigators

In the case that a serious adverse event occurs during the clinical study, the sub-investigator should report it to the principal investigator and the sponsor immediately and provide additional reports with detailed information later.

(3) Obligations of the Institutional Review Board

The Institutional Review Board should inform the principal investigator of the necessary measures in the case of occurrence of suspected unexpected serious adverse reactions or new information that may adversely affect the safety of the subject or the conduct of the clinical study.

(4) Obligations of the sponsor

The sponsor should report all suspected unexpected serious adverse reactions reported from the investigator to other related investigators and the Ministry of Food and Drug Safety within “15 days” from the day when the sponsor received a report or recognized suspected unexpected serious adverse reactions. However, in the case that they cause death or are life threatening, the sponsor should report within “7 days” from the day when the sponsor received a report or recognized suspected unexpected serious adverse reactions. If additional information needs to be reported, additional reports should be submitted within “15 days” from the day when the sponsor received a report or recognized the detailed information. When the sponsor is submitting an adverse drug reaction report, the sponsor will attach and submit the information received from the investigator.

In relation to the above report, the sponsor should periodically report additional safety information until the end of the relevant adverse event (disappearance of the relevant adverse event or impossible to follow up). The investigator should actively cooperate in providing data and information on the reports.

In accordance with the ICH GCP regulations, the sponsor should notify the investigator of “matters that may

negatively affect the subject's participation in the clinical study or that may result in a change in the approval of the Institutional Review Board for continuation of the clinical study," and especially should inform the investigator of suspected unexpected serious adverse reactions (SUSARs).

(5) Immediate reporting to the sponsor by the investigator

Serious adverse events and adverse events that require expedited reporting to the pharmacovigilance staff member should be recorded in the "Serious Adverse Event Report" form. This form must be reported to the sponsor and contract research organization within 24 hours. The "Serious Adverse Event Report" form and recording instructions will be provided in the investigator study file.

The initial report should be prepared as complete as possible, including the detailed information on intercurrent diseases and (serious) adverse events and the evaluation of causality relationship to the investigational product.

Information that is not obtained at the time of completing the initial report (e.g., the adverse event end date or the laboratory test result obtained after the initial report) should be recorded in the "Serious Adverse Event Report" form at the follow-up.

If a non-serious adverse event becomes a serious adverse event, it should be recorded in the "Serious Adverse Event Report" form and should be reported to the sponsor immediately.

16.8 Follow-up on adverse events

While the subject is participating in the clinical study, the investigator should ensure that proper medical treatment is given for all adverse events, including clinically significant laboratory test results related to the study. If medical treatment is needed for adverse events perceived by the investigator, the subject should be informed.

The follow-up information should only include new (updated and/or additional) information that reflects the situation at the time of the investigator's signature. The follow-up period for adverse events in this clinical study is to be 1 month after the subject's last visit. In the case of a chronic disease or death of a subject due to other adverse events, etc., these cases may be terminated with the result as "recovering" or "not recovered," and therefore, the follow-up does not require the result to be in the "recovered" category.

16.9 Emergency

In the case of a deviation from the protocol due to an emergency, the investigator should call and inform the sponsor, iNtRON Biotechnology, Inc, of the deviation as soon as possible, and determine whether the relevant subject should continue or discontinue to be in the clinical study. The details of the protocol deviation and its reason should be recorded in detail in the case report form or the protocol deviation (violation) form.

17. Informed consent form

The investigator should obtain a written informed consent form [Attachment 1] after providing sufficient information in accordance with the Declaration of Helsinki and KGCP before enrolling the patient in the clinical study.

The Institutional Review Board (IRB) should review the informed consent form and subject information sheet to be used by the investigator. It is the responsibility of the principal investigator to obtain the informed consent form from the patient or his/her legally authorized representative before initiating any act or treatment that is not the patient's usual care.

The informed consent form and subject information sheet should include all information required by the ICH, KGCP and applicable regulations. In addition, ethical principles based on the KGCP and Declaration of Helsinki should be followed. The informed consent form should also include the statement "iNtRON Biotechnology, Inc., the sponsor, and the Ministry of Food and Drug Safety will directly access the subject's medical records." Prior to initiation of the study, the investigator should obtain approval from the Institutional Review Board (IRB) for the written informed consent form and other information to be provided to the subject.

In the case that the clinical study includes subjects recruited after obtaining consent only from their authorized representative (e.g., minor, patient with severe dementia) or without obtaining consent (e.g., emergency), the investigator should explain the clinical study to the subject in person to the extent that the subject can understand it, and if possible, the investigator should have the subject sign and date the informed consent form as soon as possible.

If it is impossible for the subject to provide informed consent, the consent from the subject's authorized representative should be obtained. In the case that it is impossible for the subject to provide informed consent and authorized representative doesn't exist, the method of subject recruitment should be specified in the protocol and/or elsewhere. The investigator should obtain a written approval from the Institutional Review Board to protect the rights, safety and well-being of such a subject and to comply with applicable regulations in relation to the subject's participation. The subject or the subject's authorized representative should be informed about the clinical study as soon as possible.

The investigator should provide a copy of the informed consent form and a copy of the written subject information sheet, which explains the clinical study using terms that can be easily understood rather than using professional jargon, to the subject or the authorized representative. After giving the subject or the subject's authorized representative time to ask questions about the details of the clinical study, the investigator should have the subject or the subject's authorized representative sign and date the informed consent form in person, and if necessary, the person who discussed together when obtaining the informed consent form should also sign and date in person. The subject or the authorized representative should receive a copy of the informed consent form signed prior to participation in the study and a copy of the other written subject information sheet provided to the subject.

If the subject or the authorized representative is an illiterate, an impartial witness should be present during the course of obtaining consent.

After the subject or the authorized representative provides consent verbally and signs, if possible, the impartial witness should sign and date the informed consent form in person to testify that the information was accurate and the subject or the authorized representative understood and provided consent based on their free will.

The informed consent form and any other information provided to the subject or the authorized representative should be changed and provided whenever important information related to the subject's consent is newly obtained, and the Institutional Review Board's opinion should be obtained prior to use. The investigator or a person designated by the investigator should fully explain all the valid aspects of the clinical study and new information related to the subject's willingness to continue participating in the clinical study to the subject or the subject's authorized representative. Mutual exchange of opinions such as this should be recorded.

While the subject is participating in the clinical study, the newly updated informed consent form and written subject information sheet should be provided for the subject.

18. Subject indemnification policy

In the case that the subject experiences adverse events that have causality relationship to the study drug used in this clinical study despite the investigator has been conducting the clinical study in compliance with all laws and regulations and strictly in accordance with the protocol and various related literature, recommendations and suggestions provided by iNtRON Biotechnology, Inc, iNtRON Biotechnology, Inc. will implement and reimburse the reasonable treatment cost in accordance with the indemnification policy for victims [Attachment 2] submitted to the IRB of the study site. See the subject indemnification policy document.

19. Examination and treatment criteria for subjects after the clinical study

The test fees and the investigational product for the schedule planned for this study will be paid by iNtRON Biotechnology, Inc. only for the subjects who participate during the clinical study participation period. The examination and treatment of the subjects whose clinical study participation has been discontinued or terminated will be based on the general examination and treatment principles of the infectious diseases field. In other words, different types of treatments, procedures, and drug therapies will be performed depending on the subject's condition and the doctor's decision.

20. Measures for protection of the subject safety

This clinical study will be conducted scientifically and ethically in accordance with the Korea Good Clinical Practice (KGCP) and laws related to clinical studies. In addition, this clinical study will be conducted not only to respect the dignity and interests of human being in accordance with the Declaration of Helsinki but also to prevent disadvantages to the subject as well.

The Institutional Review Board of the study site will evaluate/approve this protocol according to the Korea Good Clinical Practice and will evaluate regularly to check whether this clinical study is being conducted in accordance with the protocol.

Before enrolling the subject in the clinical study, the investigator will check the health condition of each subject to confirm that the subject can participate in the study, and if there was an adverse event in the previous cycle prior to the administration for the next cycle, the investigator will check whether the adverse event has recovered and continue the study. In addition, the investigator will fully understand the investigational product and will put the best effort to ensure the safety of the subject.

In the case that the subject experiences adverse events due to the clinical study, the subject will receive appropriate medical treatment until recovery, and iNtRON Biotechnology, Inc. will indemnify against the injuries caused by the investigational product in accordance with the "indemnification policy for victims [Attachment 2]."

20.1 Study site

The head of the study site should fully equip the laboratory and professional personnel required for the conduct of the relevant clinical study for each step and should prepare perfectly to conduct the relevant clinical study appropriately.

20.2 Principal investigator

The principal investigator should fully understand the adverse events, precautions, etc. that are stated in this protocol in advance and report to the Institutional Review Board and the sponsor immediately in case of serious adverse events, etc. during the clinical study.

20.3 Sub-investigators

They should understand the expected adverse events, precautions for use, etc. that are stated in this protocol in advance and report to the principal investigator and the sponsor immediately in case of serious adverse events, etc. while conducting the clinical study.

20.4 Sponsor

The sponsor will designate a medical monitor to promote the safety of subjects participating in this clinical study. The medical monitor will conduct the review while blinded and will perform the following activities:

- Reviewing SAEs/SUSARs, and if necessary, performing separate evaluation in combination with the investigator's evaluation
- Attending the IDMC meeting if necessary (to attend only where blinding can be maintained)
- Reviewing the safety section of the clinical study report

21. Other matters required to conduct the clinical study safely and scientifically

21.1 Approval of the Institutional Review Board (IRB)

Prior to initiation of this clinical study, the investigator should obtain a written approval from the Institutional Review Board for the protocol, informed consent form, data and process related to the subject recruitment (e.g., advertisement), and written subject information sheet to be provided to the subject. The investigator should provide the investigator's brochure, a copy of the overview of the drug applied for approval (for marketed products, the package insert), the information to be provided to the subject, the most recent information, etc. to the Institutional Review Board. The investigator should provide the clinical study report, the most recent information, and other information (e.g., safety update, amendments, notification letters) to the Institutional Review Board according to applicable regulations or in-hospital procedures. The investigator should also submit the approval

letter obtained from the Institutional Review Board to iNtRON Biotechnology, Inc. before the clinical drug is supplied. If an audit is conducted by the study site or the Institutional Review Board in relation to this clinical study, the investigator should inform this to iNtRON Biotechnology, Inc. immediately. In this case, the investigator should inform iNtRON Biotechnology, Inc. regarding the result after the audit.

21.2 Approval of the protocol and the protocol amendment

Prior to initiation of the clinical study, the protocol, subject information sheet, informed consent form, and other relevant documents will be submitted to the IRB along with an application for a review (cover letter or a form) that contains the list of submitted documents, date of submitted documents issued, institution from which approval needs to be obtained (or region or district under jurisdiction). If applicable, these documents may be submitted to the regulatory authorities, as required by the legal requirements in the relevant country.

The sponsor may supply the clinical drug to the investigator to initiate the study only after obtaining all documents required ethically and legally. These documents should also include a list of IRB members whose occupations and qualifications are indicated. If the IRB does not disclose a list of members, a request should be made to issue a document confirming that the members will be composed based on the KGCP. The IRB approval letter should include the records of the clinical study title, clinical study number, study site (region if needed), and amendment number and other documents reviewed, if applicable. The approval letter should include the documentation of the date when the approval was determined, and it should include the official signature of a member. All ethical and legal requirements should be met before the first subject is enrolled in the clinical study.

Amendments to the protocol and administrative changes should be reported to the IRB and, if applicable, to the regulatory authorities, as required by the legal requirements in the relevant country. The protocol amendment should be evaluated to determine if official approval is required and to determine whether the informed consent form should be revised.

The investigator should keep all the records of information exchange with the IRB, and if applicable, the investigator should keep the records of information exchange documented between the coordinating investigator and the IRB. This is also applied to the information exchange between the investigator (or the coordinating investigator, if applicable) and the regulatory authorities.

21.3 Compliance with the protocol and amendments to the protocol

This clinical study will be conducted in accordance with the approved protocol. All amendments to the protocol should be discussed with the sponsor, iNtRON Biotechnology, Inc., and the protocol amendment should be completed by the sponsor (or the contract research organization). The investigator should not apply any of these amendments to the protocol before they are reviewed and approved by the Institutional Review Board, except for the cases to prevent harm to the subjects immediately. Serious protocol deviations should be recorded in the case report form.

If any of these modifications or amendments to the protocol are applied before obtaining approval from the Institutional Review Board in order to prevent harm to the subject immediately, these modifications or amendments should be submitted to the Institutional Review Board (for review and approval in the future), the

sponsor, the contract research organization, and the Ministry of Food and Drug Safety (if required by applicable regulations) as soon as possible. Then, the chairperson or the secretary of the Institutional Review Board should send a signed approval document to the sponsor.

If the protocol amendments are minor, it is sufficient for the investigator to just notify the Institutional Review Board. However, if the fundamental study design is being changed or if the possibility of risks to the subject is increasing,

① the informed consent form should be revised and submitted to the Institutional Review Board to be reviewed and approved,

② consent should be obtained again from the subjects already recruited using a newly changed informed consent form if such a change affects the subjects, and

③ the new informed consent form should be used to obtain consent from newly recruited subjects.

21.4 Protection of subjects

The sponsor, contract research organization, and investigator should thoroughly monitor the subject's progression of disease for the safety of the subject, and if the risks to the subject safety are expected, the relevant subject should be excluded from the clinical study and appropriate measures should be taken.

This study guarantees the confidentiality of all subjects, and records and evaluates using the subject number (ID) assigned during the study process. The subjects will be informed of this confidentiality guarantee for subjects. The principal investigator will keep the completed informed consent form. By signing this agreement, the investigator has agreed to obtain the informed consent form from the subject participating in the study.

21.5 Monitoring

The purposes of monitoring are as follows:

1) To protect the rights and welfare of subjects
2) To determine whether the reported clinical study-related data are accurate, complete, and verifiable when compared with the source documents
3) To confirm whether the clinical study is conducted in accordance with the protocol approved by the Minister of Food and Drug Safety and the Institutional Review Board as well as the KGCP

Therefore, the clinical research associate will regularly visit the study site during the study progression in order to confirm whether the principal investigator is complying with the approved protocol, whether source documents and other clinical study-related records are maintained to be accurate, complete and up to date, and whether all adverse events have been properly reported within the deadlines specified in the KGCP and the protocol. Also, the clinical research associate will visit the study site for purposes to check the storage (preservation) condition and quantity of the investigational product, and to confirm the accuracy, completeness, and consistency of case report forms, source documents and other clinical study-related documents (including electronic documents) by checking whether the clinical study data required by the protocol are being recorded in the case report forms accurately and whether the information in the case report forms is consistent with the source documents.

The clinical research associate acts as the key communicator between the sponsor and the principal investigator.

When the clinical research associate visits the study site or contacts the study staff member via telephone, fax or email, the relevant facts and information should be reported to the sponsor in writing. The monitoring report should include the following:

1) Date and location where monitoring is carried out, name of the clinical research associate, investigator (or the name of a person who came in contact)
2) Descriptions of a summary of what the clinical research associate has identified and clinically significant findings or events, deviations from the protocol, etc. or problems, and action taken or to be taken to maintain compliance with the protocol

The clinical research associate should report data related to the safety of the subjects that has been identified through the monitoring process to the sub-investigator (investigator) who can make medical judgement, and the sub-investigator (investigator) will finally determine the following recommendations resulting from the data safety monitoring:

- Recommendation whether to continue or discontinue the study
- Recommendation for the recruitment/selection/preservation and management of subjects, the improvement of protocol compliance, and the data management and quality control process in order to ensure the integrity of the study
- Recommendation of ways to reduce the risks of adverse events, etc. by evaluating the information on risks that exceed the benefits related to the investigational product, the adverse events, or the effect that is below the expectation
- Review of the completeness of data, etc. by receiving reports of monitoring results related to protocol deviations, dropout, etc. when such cases occur

The principal investigator should agree and cooperate to allow the clinical research associate or the delegate of this task to have access to the location of the investigational products stored and the clinical study-related documents (including electronic documents), and the clinical research associate can review all CRFs and written consents.

21.6 Case report forms

Source documents include all documents (including electronic documents), data, and records that contain source data such as hospital records, medical records, subject's records, memos, pathological test results, subject's diaries, assessment checklists, drug dispense records, data recorded on automated test equipment, test certificates and their official copies, microfiches, microfilms, radiological examination data, magnetic tapes, and pathological laboratory record data. Source data include any information contained in the original or an official copy of the original in which the relevant clinical findings, observations, and other actions required to reproduce or evaluate the clinical study are recorded.

Therefore, all information collected and recorded in the subject's case report form (CRF) should be prepared based on the source documents and source data and should be consistent with this information. A description should be attached to any information that does not match the source documents.

The investigator should ensure that the data contained in the case report form or any other report are accurate, complete, legible, and timely. In the case of changing or correcting information in the documented or electronic case report form, the records of any changes or corrections should be stored.

The clinical research associate will visit the study site at appropriate intervals to perform the duties to compare the case report forms and source documents. For this, the investigator should agree and cooperate to allow the clinical research associate to access source documents. Once the data to be recorded are obtained, the case report form should be completed as soon as possible.

The form of the case report form is the web-CRF, and the extent of authorization to use will be set and an ID will be assigned based on the scope of work of the investigator, CRA, CRC, etc. Only the clinical study staff members will have access to the web-CRF using the ID and PW, and the principal investigator should maintain confidentiality of the records that can identify the subject in order to ensure that subject's personal life and confidentiality is guaranteed in accordance with applicable regulations.

Other clinical study-related data (such as various source data and correspondence) should be properly signed and dated in accordance with the general Korea Good Clinical Practice (KGCP).

After the clinical study is terminated and data are transferred from the investigator to iNtRON Biotechnology, Inc., the relevant data will go through a verification process. If necessary, iNtRON Biotechnology, Inc. may request the clinical study staff member to verify or modify the data.

21.7 Preservation of records

The principal investigator will keep all documents related to the study, including the reports submitted to the Ministry of Food and Drug Safety, the Institutional Review Board, and iNtRON Biotechnology, Inc. Also, according to Article 30 Paragraph 2 of the Regulation on Safety of Pharmaceuticals, etc., the investigator should keep the protocol and the records and data related to the conduct of the clinical study (including electronic documents) for 3 years from the date of licensure for manufacturing, selling, importing products.

In the case that the investigator is unable to continue storing the documents due to appointment to another department or retirement, the study-related documents should be transferred to a mutually agreed designee (e.g., another investigator who participated in the clinical study, the Institutional Review Board), and in such a case, the sponsor should be notified in writing.

The investigator should contact the sponsor of the clinical study-related documents in order to obtain a written approval letter from the sponsor prior to the destruction. If the sponsor determines that the preservation of the data is no longer necessary, the sponsor will inform the principal investigator in writing.

The investigator should obtain a written approval letter from the sponsor prior to the destruction of the documents. The investigator should keep the records of use of the study drug, copies of case report forms (or electronic files), source data, etc. related to the clinical study for the period specified in the relevant regulations (KGCP basis: to be kept for 3 years from the date of licensure or the date of deletion of the conditions of permission) and the period specified by the sponsor, whichever is longer. The essential documents of the clinical study include the following:

① All informed consent forms signed
② Subject identification code list*, screening log (if applicable), enrollment log

③ Records of any contact between the investigator and the IEC/IRB
④ Composition of the IRB
⑤ Records of any contact between the investigator and the sponsor (or the contract research organization)
⑥ Recorded list of the investigators and staff members delegated to perform important clinical study tasks and their roles and signatures
⑦ CRFs of all subjects and copies of documents related to the changes
⑧ Investigational product accountability log
⑨ Records of the storage of body fluids or tissue samples (if applicable)
⑩ All other source documents (subject's medical records, hospital records, laboratory test records, etc.)
⑪ All documents corresponding to Section 8 of the ICH E6 Guideline for KGCP (essential documents for the conduct of clinical studies)

In general, these documents will be kept by the investigator. If the investigator cannot perform these obligations, the sponsor must be notified to perform an alternative, and this should be documented in detail.

21.8 Maintaining data and subject records

21.8.1 Maintaining confidentiality of data

The investigator should treat all information provided by iNtRON Biotechnology, Inc. in relation to this clinical study as confidential. In addition, in the case of disclosing this information to the Institutional Review Board or similar professional committees or related organizations and to people working at such organizations, the relevant information must be provided after they fully understand about maintaining the confidentiality of the information.

21.8.2 Maintaining subject records

The investigator should treat all personal information related to the subjects who are involved in this clinical study as confidential. However, if the Ministry of Food and Drug Safety or equivalent health authorities request that the study documents related to the subjects be reviewed or copied to verify the records in the case report forms, the investigator should accept this.

21.9 Termination of the clinical study

Upon completion of the clinical study, the relevant clinical study must be terminated at the study site. The sponsor or the investigator may discontinue the clinical study at any time, but early termination of the study should be done after having a sufficient consultation. In accordance with the regulations in the relevant country, it may be necessary to report to the IEC/IRB and regulatory authorities, at the end of the clinical study period. Clinical study-related supplies should be collected, destroyed, or stored as requested by the sponsor.

21.10 Reporting of early termination or discontinuation of the clinical study

The sponsor, the investigator, or the relevant regulatory agencies may discontinue the entire or part of the study

at any time, but the compliance process should be consulted.

The clinical study can be discontinued unexpectedly in the event of a safety issue while conducting the clinical study. The study can also be discontinued even when the overall risk–benefit evaluation is not favorable for continuing the study.

If the study is terminated early or discontinued, the investigator should notify the subjects promptly to allow them to have proper treatment and follow-up care. Furthermore, the investigator or the sponsor should immediately notify the IRB of the detailed information in writing. The regulatory agencies should be informed according to the relevant regulations of each country.

If the risk–benefit evaluation changes after the study is discontinued, it should be provided to the IRB in the case that the new evaluation can affect the follow up plan on the subjects who participated in the study. In such a case, the measures required to protect the subjects should be described.

21.11 Deviation from the protocol

No protocol deviation should occur. If a deviation occurs, the principal investigator should notify the clinical research associate and review and discuss the deviation. All deviations should be documented, and the reason, date, action taken, impact on the subject and the study should be documented. These documents should be kept in the investigator's study file and the sponsor's study file.

21.12 Essential documents

Before initiating the study, in other words, before obtaining consent from the first subject, the investigator should provide the following documents to iNtRON Biotechnology, Inc.:

① Curriculum vitae of the principal investigator and sub-investigators (signed and dated currently and/or supported by notarization documents)
② Signed and dated agreement for the final protocol
③ Signed and dated agreement for the actual protocol amendment (if applicable)
④ Approval/favorable comments from the IRB that clearly define the documents reviewed (protocol, actual protocol amendment, subject information sheet/informed consent form, other written information provided to subjects, and subject recruitment procedures)
⑤ Copies of the subject information sheet, informed consent form, other written information, and subject recruitment advertisement that are approved by the IRB (if applicable)
⑥ IRB members/composition list
⑦ Financial agreement(s)
⑧ Signed IRB receipt
⑨ Approval letter from the regulatory agencies and/or notification if required

22. Use of the study result

By signing this protocol, the principal investigator and the investigator agree to use the results of this study for purposes such as registration, presentation, and provision of information for medical and pharmaceutical experts. If necessary, the name, address, and qualifications of the investigator may be notified to the government authorities. The result of this clinical study will be presented in academic journals or academic conferences, and the sponsor has the right to review the information to be presented prior to presentation or submission of the clinical study result.

All information related to the study drug and the unpublished information provided by the sponsor, such as indications, formulations, and preparation methods of the study drug and other academic data related to the study drug are confidential and solely owned by the sponsor. The investigator will use the relevant information only for the purpose of conducting this study. Unless a prior written approval is obtained from the sponsor, the investigator will agree that it will not be used for any other purposes.

The original CRFs completed will be solely owned by the sponsor. By signing the protocol, the investigator will agree to the sponsor's use of the study results for the domestic and overseas application for approval of drugs, presentation and use as information for medical and pharmaceutical professionals. The name, address, and qualifications of the investigator will be reported to the regulatory authorities.

The information obtained during this clinical study at the discretion of iNtRON Biotechnology, Inc. may be provided to other investigators who are conducting other clinical studies using this clinical drug.

Although it is the opposite from the information described in the protocol, iNtRON Biotechnology, Inc. will agree with the investigator's right to publish the study results. All data and results obtained in this clinical study are owned by iNtRON Biotechnology, Inc., and the publications should state the participation of the principal investigator, staff members who assisted him/her, and other investigators who participated in this clinical study. Any scientific paper, presentation, communication, or other study-related information described in this protocol should be submitted to the chief executive officer of iNtRON Biotechnology, Inc. to review the information before submitting it for publication or presentation. The reviewed information will be provided within 4 weeks after the application of the manuscript. The guidelines for the copyright aims to follow the guidelines of the ICMJE (International Committee of Medical Journal Editors). In addition, when there is a request for a patent application or measures for the sponsor to firmly establish its ownership, the investigator will postpone the presentation for an additional 90 days.

Upon termination of the study, one or more manuscripts for joint publication will be prepared by the investigator and iNtRON Biotechnology, Inc. collaboratively. The copyrighted investigator will be requested to initiate and approve the publication. The completion of the clinical study report (CSR) is the responsibility of Symyoo Inc. and iNtRON Biotechnology, Inc. One principal investigator responsible for reviewing the CSR and signing the investigator's signature section will be designated by iNtRON Biotechnology, Inc.

If there is a disagreement about the publication, the publication should reflect the opinions of both the investigator and iNtRON Biotechnology, Inc. fairly and sufficiently.

23. Responsibilities

Symyoo Inc. and iNtRON Biotechnology, Inc. are responsible for completion of the protocol, case report form, supply of the clinical drug and other study materials, monitoring, data management, statistical analysis, procedures of iNtRON Biotechnology, Inc., consent form for internal uses, and the clinical study report (CSR) specified in the recent

KGCP guidelines.

The investigator is responsible for conducting the clinical study. If any responsibilities are delegated, the investigator should have a list of properly qualified persons to whom the investigator has delegated specific important responsibilities related to the study.

The investigator will take all technical and organized security measures needed to prevent accidental or undesirable damage, loss, or deterioration of data. The investigator will prevent unauthorized access to any data or other data processing that is not permitted by applicable laws.

In the case that there is a request from iNtRON Biotechnology, Inc., the investigator will provide iNtRON Biotechnology, Inc. with the necessary information which can confirm that such technical and organized security measures are being implemented.

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Attachments

[Attachment 1] Subject Information Sheet and Informed Consent Form

[Attachment 2] Indemnification Policy for Victims

[Attachment 3] Data and Safety Monitoring Plan