A Randomized, Double-Blind, Placebo-Controlled, Multiple-Dosing, Dose-Escalating Phase 1b Study to Evaluate the Safety, Pharmacokinetics, Pharmacodynamics and Immunogenicity of N-Rephasin® SAL200 after Continuous Intravenous Infusion in Healthy Volunteers

ClinicalTrials.gov ID: NCT03446053

# Protocol

*Title:* 건강한 남성자원자를 대상으로 N-Rephasin® SAL200 정맥 내 지속주입 시 안전성, 약동학, 약력학, 면역원성을 평가하기 위한 무작위배정, 양측눈가림, 위약대조, 단회 및 반복 투여, 단계적 증량 제 1b상 임상시험

*Title:* A Randomized, Double-Blind, Placebo-Controlled, Multiple-Dosing, Dose-Escalating Phase 1b Study to Evaluate the Safety, Pharmacokinetics, Pharmacodynamics and Immunogenicity of N-Rephasin® SAL200 after Continuous Intravenous Infusion in Healthy Volunteers

- Sponsor: iNtRON Biotechnology, Inc.
- Protocol No.: ITB-101_1b
- Protocol Version: 2.0

**05/ 17/ 2019**

**CONFIDENTIAL**

## iNtRON Biotechnology, Inc.

# 【Schedule of Clinical Study Plan】

## 1. Single-dose group

| Observation and Test Items | Schedule of Clinical Study | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Screening Period | Admission Period | | | Outpatient Period | | | | Post-study Visit |
| Visit Number | SCV | V1 | | | V2 | V3 | V4 | V5 | V6 |
| | Screening | Hospitalization | | | FU1 | FU2 | FU3 | FU4 | PSV |
| | D-28~D-2 | D-1 | D1 | D2 | D8±D1 | D15±D1 | D22±D1 | D29±D1 | D50±D2 |
| Written consent | ✘ | | | | | | | | |
| Demographic information/medical history | ✘ | | | | | | | | |
| Inclusion/exclusion criteria | ✘ | ✘ | | | | | | | |
| Randomization | | ✘ | | | | | | | |
| Admission/discharge[1] | | ✘ | | ✘ | | | | | |
| Administration of the investigational product[2] | | | ✘ | | | | | | |
| Vital signs[3] | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Electrocardiogram[4] | ✘ | | ✘ | ✘ | | ✘ | | | ✘ |
| Clinical laboratory tests[5] | ✘ | | ✘ | ✘ | ✘ | ✘ | | | ✘ |
| Breath alcohol test | ✘ | | | | | | | | |
| Physical examination[6] | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Blood collection for pharmacokinetics[7] | | | ✘ | ✘ | | | | | |
| Urine collection for pharmacokinetics[8] | | | ✘ | ✘ | | | | | |
| Blood collection for pharmacodynamics[9] | | | ✘ | | | | | | |
| Immunogenicity assessment[10] | | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Allergenicity test[11] | ✘ | ✘ | | | ✘ | ✘ | | ✘ | |
| Checking on adverse events | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Checking on medication history/concomitant medications | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Checking on whether medical treatment has been given | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |

Note

1. The subject will be admitted at 1 p.m. of Day −1 and will be discharged after completing the entire schedule of Day 2.
2. All schedules will be carried out based on the date and time of the first administration of the IP. [Date and time of the first IP administration - Day 1: 0 h]
3. Vital signs (sitting blood pressure/pulse rate/temperature) will be measured at SCV, V1, V2, V3, V4, V5, and V6 and will be measured in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day-1 • Day 1 : pre-dose, 1h, 4h, 8h, 12h, 13h. • Day 2 : 0h.
4. Electrocardiogram will be obtained at SCV, V1, V3, and V6 and will be measured in accordance with the detailed schedule below at V1 (hospitalization period).

- Day 1 : pre-dose, 0h~2h(continuous), 4h. • Day 2 : 0h.

5. Clinical laboratory tests will be performed at SCV, V1 (D1: pre-dose; D2: 0 h), V2, V3, and V6.
6. Physical examination will be performed at SCV, V1 (D−1; D1: pre-dose, 1.5 h; D2: 0 h), V2, V3, V4, V5, and V6.
7. Blood collection for pharmacokinetics will be performed in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : pre-dose, 0.25h, 0.5h, 0.75h, 1h, 1.5h, 2h, 3h, 4h, 5h, 6h, 8h, 10h, 12h.
   - Day 2 : 0h.
8. Urine collection for pharmacokinetics will be performed in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : 0~4h, 4~12h, 12~24h.
9. Blood collection for pharmacodynamics will be performed in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : pre-dose, 1h, 1.5h, 2h.
10. An immunogenicity assessment will be performed at V1 (D1: pre-dose; D2: 0 h), V2, V3, V4, V5, and V6.
11. An allergenicity test (Standard allergy skin test) will be performed at SCV, V1 (D−1), V2, V3, and V5.

## 2. Repeat-dose group

| Observation and Test Items | Schedule of Clinical Study | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Screening Period | Admission Period | | | | | | Outpatient Period | | | | Post-study Visit |
| Visit Number | SCV | V1 | | | | | | V2 | V3 | V4 | V5 | V6 |
| | Screening | Hospitalization | | | | | | FU1 | FU2 | FU3 | FU4 | PSV |
| | D-28D~D-2 | D-1 | D1 | D2 | D3 | D4 | D5 | D8± D1 | D15± D1 | D22± D1 | D29± D1 | D50±D2 |
| Written consent | ✘ | | | | | | | | | | | |
| Demographic information/medical history | ✘ | | | | | | | | | | | |
| Inclusion/exclusion criteria | ✘ | ✘ | | | | | | | | | | |
| Randomization | | ✘ | | | | | | | | | | |
| Admission/discharge[1] | | ✘ | | | | | ✘ | | | | | |
| Administration of the investigational product[2] | | | ✘ | ✘ | ✘ | ✘ | | | | | | |
| Vital signs[3] | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Electrocardiogram[4] | ✘ | | ✘ | ✘ | ✘ | ✘ | ✘ | | ✘ | | | ✘ |
| Clinical laboratory tests[5] | ✘ | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | | | ✘ |
| Breath alcohol test | ✘ | | | | | | | | | | | |
| Physical examination[6] | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Blood collection for pharmacokinetics[7] | | | ✘ | ✘ | ✘ | ✘ | ✘ | | | | | |
| Urine collection for pharmacokinetics[8] | | | ✘ | ✘ | | ✘ | ✘ | | | | | |
| Blood collection for pharmacodynamics[9] | | | ✘ | | | ✘ | | | | | | |
| Immunogenicity assessment[10] | | | ✘ | | | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Allergenicity test[11] | ✘ | ✘ | | | | | | ✘ | ✘ | | ✘ | |
| Anaphylatoxin test[12] | | | ✘ | | | ✘ | | | | | | |

| | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Checking on adverse events | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Checking on medication history/concomitant medications | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Checking on whether medical treatment has been given | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |

Note

1. The subject will be admitted at 1 p.m. of Day −1 and will be discharged after completing the entire schedule of Day 5.
2. All schedules will be carried out based on the date and time of the first administration of the IP. [Date and time of the first IP administration - Day: 0 h]
3. Vital signs (sitting blood pressure/pulse rate/temperature) will be measured at SCV, V1, V2, V3, V4, V5, and V6 and will be measured in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day-1
   - Day 1 / Day 4 : pre-dose, 1h, 4h, 8h, 12h, 13h.
   - Day 2 / Day 3 : pre-dose, 1h, 12h, 13h.
   - Day 5 : 0h.
4. Electrocardiogram will be obtained at SCV, V1, V3, and V6 and will be measured in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : pre-dose, 4h.
   - Day 2 / Day 3 / Day 4 : 0h, 4h.
   - Day 5 : 0h.
   - Day 1 / Day 4 : 0~2h(continuous).
5. Clinical laboratory tests will be performed at SCV, V1 (D1: pre-dose; D2, D3, D4, D5: 0 h), V2, V3, and V6.
6. Physical examination will be performed at SCV, V1 (D−1; D1: pre-dose, 1.5 h; D2, D3, D4: 0 h, 1.5 h; 5D: 0 h), V2, V3, V4, V5, and V6.
7. Blood collection for pharmacokinetics will be performed in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : pre-dose, 0.25h, 0.5h, 0.75h, 1h, 1.5h, 2h, 3h, 4h, 5h, 6h, 8h, 10h, 12h.
   - Day 2 / Day3 : 0h, 1h, 12h.
   - Day 4 : 0h, 0.25h, 0.5h, 0.75h, 1h, 1.5h, 2h, 3h, 4h, 5h, 6h, 8h, 10h, 12h.
   - Day 5 : 0h.
8. Urine collection for pharmacokinetics will be performed in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 / Day 4 : 0~4h, 4~12h, 12~24h.
9. Blood collection for pharmacodynamics will be performed in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : pre-dose, 1h, 1.5h, 2h.
   - Day 4 : 0h, 1h, 1.5h, 2h.
10. An immunogenicity assessment will be performed at V1 (D1: pre-dose; D5: 0 h), V2, V3, V4, V5, and V6.
11. An allergenicity test (Standard allergy skin test) will be performed at SCV, V1 (D−1), V2, V3, and V5.
12. An anaphylatoxin test will be performed on C3a, C4a, mast cell tryptase, IL-1b, IL-2, IL-6, and TNF-alpha in accordance with the detailed schedule below at V1 (hospitalization period).
    - Day 1 : pre-dose, 1h
    - Day 4 : 0h, 1h

## 【Definitions of Abbreviations or Terms】

| Abbreviation or Term | Definition |
|---|---|
| **Active** | Study group |
| **ADR** | **A**dverse **D**rug **R**eaction |
| **AE** | **A**dverse **E**vent |
| **Serious AE・ADR** | Serious Adverse Event / Adverse Drug Reaction |
| **$Ae_{t,ss}$** | Cumulative amount of a drug excreted in the urine during a dosing interval (t) at a steady state |
| **ALT** | **A**lanine **T**ransaminase |
| **ANOVA** | Analysis of Variance |
| **anti-HCV Ab** | anti-**H**epatitis **C** **V**irus Antibody |
| **anti-HIV Ab** | anti-**H**uman **I**mmunodeficiency **V**irus Antibody |
| **AST** | Aspartate Transaminase |
| **$AUC_{t,ss}$** | Area under the serum drug concentration–time curve within a dosing interval(t) at a steady state |
| **BP** | Blood Pressure |
| **BUN** | Blood Urea Nitrogen |
| **CI** | Confidence Interval |
| **$C_{max,ss}$** | Maximum concentration of a drug in serum after the repeated administration at a steady state |
| **$C_{min,ss}$** | Through concentration of a drug in serum at a steady state<br>Concentration immediately before the administration at a steady state (through concentration at a steady state) |
| **CL/F** | Apparent Clearance |
| **$CL_R$** | Renal Clearance |
| **CPK** | Creatine Phosphokinase |

| Abbreviation or Term | Definition |
|---|---|
| **CRF** | Case Report Form |
| **CRO** | **C**ontract **R**esearch **O**rganization |
| **DBP** | **D**iastolic **B**lood **P**ressure |
| **ECG** | Electrocardiogram |
| **$E_{max}$** | Peak concentration of glucose/insulin after glucose administration |
| **GCP** | **G**ood **C**linical **P**ractice |
| **γ-GT** | gamma-**G**lutamyl **Tr**anspeptidase |
| **HBsAg** | Hepatitis B Virus surface Antigen |
| **HR** | **H**eart **R**ate |
| **IRB** | **I**nstitutional **R**eview **B**oard |
| **LDH** | **L**actate **D**e**h**ydrogenase |
| **LLOQ** | Lower Limit of Quantification |
| **MR** | Metabolic ratio; ratio of the metabolite to the parent drug<br>MR= $AUC_{t,ss}$ of metabolite /$AUC_{t,ss}$ of parent drug |
| **PR** | **P**ulse **R**ate |
| **RBC** | **R**ed **B**lood **C**ell |
| **SBP** | **S**ystolic **B**lood **P**ressure |
| **SOP** | **S**tandard **O**perating **P**rocedure |
| **$t_{1/2}$** | Terminal half-life |
| **$T_{max,ss}$** | Time of maximum concentration at steady state |
| **WBC** | **W**hite **B**lood **C**ell |

# 【 Table of Contents 】

## Appendix

# 1. Title and phase of the clinical study, protocol identification number, establishment/revision history, etc.

## 1.1 Title of the clinical study

건강한 남성자원자를 대상으로 N-Rephasin® SAL200 정맥 내 지속주입 시 안전성, 약동학, 약력학, 면역원성을 평가하기 위한 무작위배정, 양측눈가림, 위약대조, 단회 및 반복 투여, 단계적 증량 제 1b상 임상시험

A Randomized, Double-Blind, Placebo-Controlled, Multiple-Dosing, Dose-Escalating Phase 1b Study to Evaluate the Safety, Pharmacokinetics, Pharmacodynamics and Immunogenicity of N-Rephasin® SAL200 after Continuous Intravenous Infusion in Healthy Volunteers

## 1.2 Phase of the clinical study

Phase 1b clinical study

## 1.3 Protocol identification number

ITB-101_1b

## 1.4 Protocol establishment/revision history

| Amendment No. | Protocol | | Reason for amendment | Remark |
|---|---|---|---|---|
| | version no. | Date | | |
| 0 | 1.0 | 07/07/2017 | Established | |
| 1 | 1.1 | 12/20/2017 | Addition of missing items, correction of typographical errors, and revision of the window period for the items during hospitalization | |
| 2 | 1.2 | 02/08/2018 | Revision of statements, and correction of typographical errors | |
| 3 | 1.3 | 06/28/2018 | Revision of the clinical study duration | |
| 4 | 1.4 | 07/24/2018 | Assignment of an unblinded CRA, and addition of a statement for unblinding clinical trial pharmacists | |
| 5 | 2.0 | 05/17/2019 | Change in the criteria for dose escalation, clarification of the early termination criteria, change of the CRO and change in the guidelines for safety reporting, and revision of statements | |

## 2. Protocol Synopsis

<table>
<tr><td>Clinical study title</td><td>건강한 남성자원자를 대상으로 N-Rephasin® SAL200 정맥 내 지속주입 시 안전성, 약동학, 약력학, 면역원성을 평가하기 위한 무작위배정, 양측눈가림, 위약대조, 단회 및 반복 투여, 단계적 증량 제 1b상 임상시험<br>[ A Randomized, Double-Blind, Placebo-Controlled, Multiple-Dosing, Dose-Escalating Phase 1b Study to Evaluate the Safety, Pharmacokinetics, Pharmacodynamics and Immunogenicity of N-Rephasin® SAL200 after Continuous Intravenous Infusion in Healthy Volunteers]</td></tr>
<tr><td>Sponsor</td><td>iNtRON Biotechnology, Inc.</td></tr>
<tr><td>Study site</td><td>Seoul National University Hospital (Professor In Jin Jang, Department of Clinical Pharmacology and Therapeutics)</td></tr>
<tr><td>Study objective</td><td>To evaluate the safety and explore the characteristics of pharmacokinetics, pharmacodynamics, and immunogenicity after single and repeated continuous intravenous infusion of N-Rephasin® SAL200 for 1 hour in healthy male volunteers.</td></tr>
<tr><td>Clinical study duration</td><td>36 months from the date of the initial approval by the Ministry of Food and Drug Safety</td></tr>
<tr><td>Target disease</td><td>Healthy male adults</td></tr>
<tr><td>Target number of subjects</td><td>• 40 subjects in total (8 subjects per group - 6 in the study group, 2 in the control group)
<table>
<tr><th>Arms</th><th>Number of Subjects</th><th>Dose</th></tr>
<tr><td>Group 1</td><td>8</td><td>6.0 mg/kg, Single dose</td></tr>
<tr><td>Group 2</td><td>8</td><td>3.0 mg/kg /day (1.5 mg/kg, b.i.d.)</td></tr>
<tr><td>Group 3</td><td>8</td><td>6.0 mg/kg /day (3.0 mg/kg, b.i.d.)</td></tr>
<tr><td>Group 4</td><td>8</td><td>9.0 mg/kg /day (4.5 mg/kg, b.i.d.)</td></tr>
<tr><td>Group 5</td><td>8</td><td>12.0 mg/kg /day (6.0 mg/kg, b.i.d.)</td></tr>
</table></td></tr>
<tr><td>Investigational product</td><td>• Study drug
<table>
<tr><td>Product name</td><td>N-Rephasin® SAL200</td></tr>
<tr><td>Content of the active ingredient</td><td>100% SAL200 (18 mg/mL as SAL-1)</td></tr>
<tr><td>Storage method</td><td>Store in a freezer (−70 ± 5°C)</td></tr>
<tr><td>Shelf life</td><td>18 months from the date of manufacture</td></tr>
</table>
• Control drug
<table>
<tr><td>Product name</td><td>INT200 –Placebo (Saline)</td></tr>
<tr><td>Content of the active ingredient</td><td>Remaining formulation buffer excluding the active ingredient of the study drug</td></tr>
<tr><td>Storage method</td><td>Store in a freezer. (-70 ± 5°C)</td></tr>
<tr><td>Shelf life</td><td>18 months from the date of manufacture</td></tr>
</table></td></tr>
</table>

| | |
|---|---|
| **Clinical study phase and design** | Phase 1b<br>Randomized, double-blind, placebo-controlled, single-center, single- and repeat-dose, gradual dose-escalation clinical study |
| **Subject eligibility** | **1. Inclusion criteria**<br>1) Healthy male adults aged 20 to 45 years at screening<br>2) Those whose weigh is ≥ 50 kg and < 90 kg and body mass index (BMI) is ≥ 18.0 and < 27.0<br>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<br><br>**2. Exclusion criteria**<br>1) Those who have clinically significant liver, kidney, nervous system, endocrine system, respiratory system, hemato-oncology, cardiovascular system, or mental diseases or past history<br>2) Those who have had an infectious disease or suspected clinical findings within 30 days prior to the date of the investigational product administration<br>3) Those who have hypersensitivity reactions to drugs containing N-Rephasin® SAL200 or other drugs (aspirin, antibiotics, etc.) or a history of such reactions<br>4) Those who have received administration of drugs containing N-Rephasin® SAL200<br>5) Those who are antibody-positive to N-Rephasin® SAL200<br>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<br>7) Those who have a past history of drug abuse or who are positive to urine drug screening<br>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.)<br>9) Those who have received administration of other investigational products within 3 months prior to the date of the investigational product administration<br>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<br>11) Those who smoke cigarettes or who have been found to be nicotine metabolite-positive in urinalysis<br>12) Those who continuously drink alcohol (exceeding 21 units/week; 1 unit = 10 g of pure alcohol) or who cannot abstain from drinking alcohol and smoking cigarettes during hospitalization<br>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<br>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 |

**Method and duration of administration of the investigational product**

**[Single-dose group]**
- 6 mg/kg, single intravenous dose

**[Repeat-dose group]**
- 1.5 mg/kg, 6 intravenous doses in total (3 mg/kg/day)
- 3.0 mg/kg, 6 intravenous doses in total (6 mg/kg/day)
- 4.5 mg/kg, 6 intravenous doses in total (9 mg/kg/day)
- 6.0 mg/kg, 6 intravenous doses in total (12 mg/kg/day)

| Repeat-dose schedule | Day 1 | Day 2 | Day 3 | Day 4 |
|---|---|---|---|---|
| At around 9 a.m. | ● | ● | ● | ● |
| At around 9 p.m. | | ● | ● | |

**Clinical study method (continued on the next page)**

**- Study method**

**[Single-dose group]**

| Screening | Admission (administration of the study drug) | Outpatient visits (blood collection for immunogenicity) | | | | Outpatient (PSV) |
|---|---|---|---|---|---|---|
| Screening visit | Visit 1 | Visit 2 | Visit 3 | Visit 4 | Visit 5 | Visit 6 |
| D -28 ~ D -2 | D -1 ~ D 2 | D 8 ±D1 | D 15 ±D1 | D 22 ±D1 | D 29 ±D1 | D 50 ±D2 |

**[Repeat-dose group]**

| Screening | Admission (administration of the study drug) | Outpatient visits (blood collection for immunogenicity) | | | | Outpatient (PSV) |
|---|---|---|---|---|---|---|
| Screening visit | Visit 1 | Visit 2 | Visit 3 | Visit 4 | Visit 5 | Visit 6 |
| D -28 ~ D -2 | D -1 ~ D 5 | D 8 ±D1 | D 15 ±D1 | D 22 ±D1 | D 29 ±D1 | D 50 ±D2 |

For 8 subjects in the first group, single-dose (6 mg/kg) group and then 3, 6, 9 and 12 mg/kg/day dose groups (repeat-dose group), 8 subjects in each group will be 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 will be evaluated to determine whether to proceed to the next dose group.
The subjects deemed eligible for this clinical study will be selected by performing screening tests, such as history taking, physical examination, clinical laboratory tests, and allergenicity testing, only in volunteers within 4 weeks (Day −28 to Day −2) from a day before the clinical study initiation (Day −1). Then, the subjects will visit the research unit of the Seoul National University Hospital Clinical Trials Center in the afternoon of a day before the clinical study initiation (Day −1), and they will have an allergenicity test.

| | |
|---|---|
| **Clinical study method (continued from the previous page)** | Subjects who do not have specific reactions will be admitted, and subject numbers will be assigned. The subjects will fast from 10 p.m. on the day of admission (Day −1), except for drinking water. The study drug or placebo will be infused intravenously in all subjects, depending on the relevant dose group, at around 9 a.m. on the next day of the admission, and the clinical study will be conducted according to the clinical study schedule.<br>After a certain period of time, post-study visit tests will be performed in all subjects who received the administration at least once, and the tests for immunogenicity assessment will be performed until about 50 days after the administration.<br><br>**2. Concomitant medications and treatments**<br>1) Medications allowed to be administered concomitantly<br>• Drugs can be administered as determined by the investigator, as needed in the cases such as when treating adverse events of the subjects.<br><br>2) Medications and treatments prohibited from being administered and performed concomitantly<br>• All medications other than those allowed to be administered concomitantly are prohibited.<br>However, if there is a drug that was administered arbitrarily without the investigator's judgment, the investigator will determine whether it may affect the safety and pharmacokinetic evaluation of this clinical study, and the subject will drop out if it is expected that the drug will have an effect.<br>• The following are prohibited as well:<br>Ingestion of foods containing caffeine (coffee, black tea, green tea, etc.), Korean distilled spirit, carbonated beverages, coffee milk, energy drinks, foods containing grapefruit, etc., drinking alcohol, or smoking cigarettes |
| **Discontinuation and dropout criteria (continued on the next page)** | **1. Discontinuation criteria**<br>1) If the principal investigator determines that the situation observed while conducting the clinical study is unreasonable to continue the clinical study, discontinuation of the clinical study can be requested to the IRB. The clinical study can be discontinued based on the decision made by the IRB, and this should be reported to the IRB.<br><br>2) The sponsor can discontinue the clinical study due to the safety of the investigational product, identification of an unfavorable risk–benefit ratio, supply of the investigational product, or other administrative or strategic reasons, and the principal investigator must report this to the IRB.<br><br>3) After the investigator evaluate the pattern of occurrence of adverse events, they will be evaluated based on the following criteria to determine whether the entire clinical study should be discontinued:<br>→ If moderate or worse adverse drug reactions that show a causality relationship to the study drug occur in 50% or more in one dose group or if severe or worse adverse events occur in 2 or more subjects, the subject's clinical study being conducted at the relevant time point should be discontinued, and the medical expert, sponsor, and principal investigator should discuss and decide whether to continue the study afterward. |

| | |
|---|---|
| **Discontinuation and dropout criteria (continued from the previous page)** | 4) The IRB or pharmaceutical regulatory agencies can discontinue the clinical study when the safety or the unfavorable risk–benefit ratio of the investigational product is identified<br><br>**2. Dropout criteria**<br>1) In the case that specific reactions occurred in an allergenicity test performed during the run-in period (day of admission, Day −1)<br>2) In the case that a subject requests discontinuation of the study drug administration during the clinical study or withdraws the consent to participation in the study<br>3) In the case that serious adverse events/adverse drug reactions occur<br>4) In the case that a subject has arbitrarily taken any of drugs that are expected to affect the safety and pharmacokinetics of the study drug<br>5) Those who ingested foods containing caffeine (coffee, black tea, green tea, etc.), Korean distilled spirit, carbonated beverages, coffee milk, energy drinks, foods containing grapefruit, etc., drank alcohol or smoked cigarettes during the hospitalization period<br>6) In the case that the investigator believes that the study needs to be discontinued for the subject<br>7) In the case that a subject does not visit as scheduled without previous notice and is unreachable<br>8) In the case that a serious protocol deviation is newly discovered during the clinical study<br>9) In the case that a disease determined to be significant by investigator develops |
| **Endpoints** | **1. Safety endpoints**<br>1) Safety-related tests<br>① Electrocardiogram (12-lead ECG, continuous ECG)<br>② Clinical laboratory tests<br>③ Vital signs<br>④ Adverse events<br>⑤ Physical examination<br>⑥ Allergenicity test<br>⑦ Anaphylatoxin test<br><br>**2. Pharmacokinetic endpoints**<br>1) Single-dose: $AUC_{last}$, $AUC_{inf}$, $C_{max}$, $T_{max}$, $T_{1/2}$, CL/F<br>2) Repeat-dose: $AUC_{last}$, $AUC_{inf}$, $AUC_{t.ss}$, $C_{max.ss}$, $C_{min.ss}$, $T_{max.ss}$, $CL_{ss}$, R(accumulation ratio). PTF(Peak Trough Fluctuation)<br><br>**3. Pharmacodynamic endpoint**<br>1) ex vivo antibacterial activity (using serum)<br><br>**4. Immunogenicity endpoint**<br>1) Analysis of concentration of anti-drug antibody (using serum) |

| | |
|---|---|
| **Statistical analysis methods** | **※ Description of results/Descriptive statistical analysis**<br>All demographic information, pharmacokinetic, pharmacodynamic, and safety/tolerability data will be summarized using descriptive statistics according to appropriate factors, such as dose group and collection time point.<br><br>**1. Assessment of the safety and tolerability**<br>- The patterns of occurrence of adverse events and the results of tests such as vital signs, electrocardiogram, clinical laboratory tests, and allergenicity test may be reviewed comprehensively, and test items judged to be clinically significant by the investigator may be compared statistically for each dose group as needed.<br><br>**2. Pharmacokinetic assessment**<br>- Pharmacokinetic parameters will be calculated using a non-compartmental method, and they will be described using descriptive statistics for each dose group.<br>- The pharmacokinetic parameters, such as $AUC_{last}$, $AUC_{inf}$, and $C_{max,ss}$, will be corrected by doses to confirm the linearity or proportionality according to the change in doses.<br><br>**3. Pharmacodynamic assessment**<br>- Pharmacodynamic parameters will be analyzed for the relationship between the concentration and the time passed after the administration in order to explore the concentration-dependency and time-dependency.<br><br>**4. Immunogenicity assessment**<br>- For immunogenicity assessment, the antibody titer for each subject will be obtained, and then the difference by the time of measurement for each dose group will be described using descriptive statistics such as differences in the mean, standard deviation, etc. at the relevant time point from the baseline values. |

# 3. Introduction

## 3.1 Background

### 3.1.1 Indication

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 more serious local or systemic infection if proper treatment is not given. Even if most of bacteremia resolves, serious sequelae are common. Such bacteremia may lead to pneumonia, pyogenic arthritis, osteomyelitis, meningitis, or cerebral edema, leading to death.

*Staphylococcus aureus*, which acts as a primary cause of bacteremia, is a bacteria 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 antimicrobial 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 antimicrobial 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 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 [10].

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.

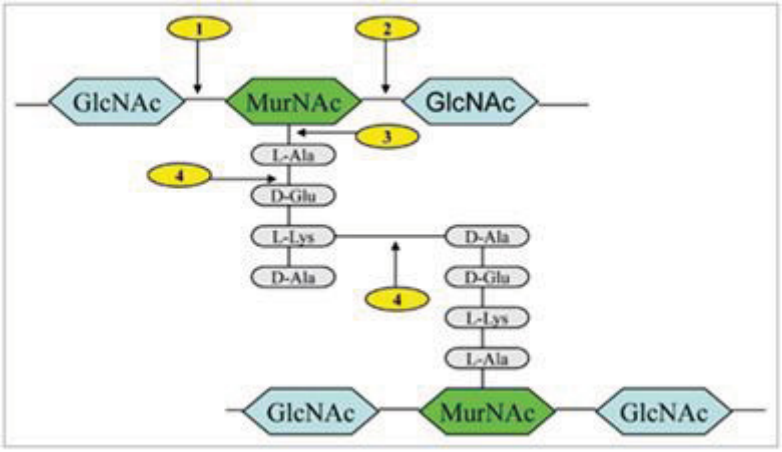
### 3.1.2 Current treatments and their limitations

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.

## 3.2 Rationale

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 that contains lysin 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.

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 (Jane D et al, 2006). The frequency of MRSA isolation is increasing every year, and as an alternative to this, a need for a substance that can replace conventional antibiotics is becoming prominent. A bacteriophage (or phage) is a bacterial virus that specifically penetrates into bacteria, and in the case of a lytic phage, it uses a specific endolysin (or lysin) to induce bacterial lysis when released from bacteria.



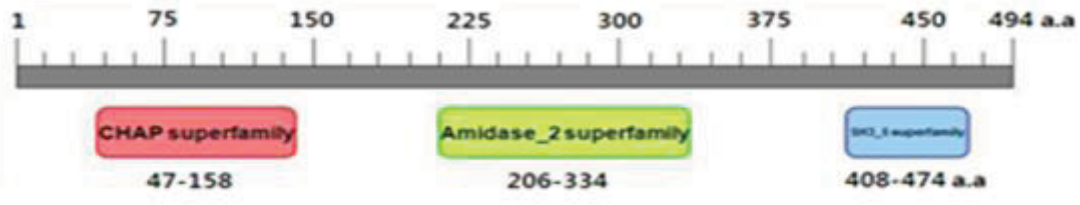

[Figure 1] Peptidoglycan structure of typical gram-positive bacteria and lysin cleavage sites

(1) N-acetylmuramidases (2) N-acetyl-β-D-glucosaminidases

(3) N-acetylmuramoyl-L-alanine amidases (4) L-alanoyl-D-glutamate endopeptidases

SAP-1 is a novel bacteriophage that belongs to the genus T4-like phage of the *Myoviridae* Family that can specifically kill *Staphylococcus aureus*, and its treatment effect against *Staphylococcus aureus* was confirmed in animal studies.

SAL-1 is a substance obtained by producing and isolating enzyme proteins that belongs to lysins among the bacterial genes of SAP-1, and it is composed of an N-terminal catalytic domain and a C-terminal binding domain. The N-terminal catalytic domain has activity that can cleave peptidoglycan of 1 to 4 bacteria [Figure 1]. The C-terminal binding domain functions to bind to a substrate in the cell wall of the target bacteria. It has been demonstrated that lysins originally produced inside hosts have the same lytic effect as those processed internally even when they were processed externally (Mark Fenton et al, 2010).




[Figure 2] Structure of the lysin SAL-1 (active ingredient)

SAL-1, the active ingredient of this study drug, is a protein with a molecular weight of 54.6 kDa consisting of 494 amino acids, and it has 3 major domains as shown in [Figure 2] constitutively. These 3 domains are divided into 2 catalytic

domains (CHAP domain and Amidase domain) and 1 cell wall binding protein (cell wall binding domain; SH3_5 domain), and they have lytic activity only against *Staphylococcus aureus* due to the substrate specificity of these domains. Therefore, SAL-1 has the advantage of having very high specificity against *Staphylococcus aureus* compared to conventional antibiotics.
In addition, the peptidoglycan layer, which is an active target of SAL-1, is an essential structure for bacterial survival (Loessner MJ, 2005. Fischetti VA, 2008). SAL-1 is an enzyme protein essential for the release of progeny by bacteriophages, and it is very difficult for *Staphylococcus aureus* to develop resistance to N-Rephasin® SAL200. In fact, no resistance to lysins was seen in experiments that observed the induction of resistance in bacteria after a considerable number of passages (Loeffler JM et al, 2001). Thus, the study drug N-Rephasin® SAL200 has an important advantage that it does not induce tolerance or resistance of pathogens while the range of use of conventional antibiotics gradually narrows as they are facing an increase in resistance. Therefore, the study drug is expected to have a potential to be a new antibiotic substance with a longer life cycle compared to conventional antibiotics.
In terms of its effectiveness, the study drug N-Rephasin® SAL200 showed highly significant antimicrobial activity in various in vitro and in vivo studies conducted previously. In particular, considering the effect of killing 99.9999% or more of bacteria in an in vitro efficacy study conducted on MRSA in various stages of growth and in an in vivo efficacy study on MRSA in high concentrations that is not found in human infections easily, it can be judged that its effectiveness is very high.
In addition, the bacteria suppression effect was observed in the first-in-human single-dose study conducted in 2013 as well. 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.

## 3.3 Benefit and risk assessment

For this study drug (N-Rephasin® SAL200), the pharmacological effect of the drug was confirmed in a preclinical study, and the safety, pharmacokinetic, and pharmacodynamic effects of the single dose was confirmed in the Phase 1 study. It is intended to confirm the safety, characteristics of pharmacokinetics, pharmacodynamics, and immunogenicity through repeat doses and dose escalation in subjects participating in this Phase 1b clinical study.
In this clinical study, the safety of a dose (6 mg/kg, single dose) that was not tested in the Phase 1 clinical study previously will be confirmed by administering a single dose, and the gradual dose escalation will be carried out from 3 mg/kg/day, which is the dose that the safety and efficacy have been confirmed in the Phase 1 clinical study conducted previously, through 6 mg/kg/day, which is the dose that the safety has been confirmed through the previous single dose administration, and finally up to 12 mg/kg/day (6 mg/kg b.i.d.). This clinical study will identify the dosage and dosing regimen of the dose that have not been administered previously. In order to ensure safety, subjects will be administered only if there is no hypersensitivity reaction to the investigational product (study drug and control drug) according to the allergenicity assessment, and the safety will be evaluated after completion of the administration in each dose group in order to determine whether to proceed to the next dose group.

Currently, antibiotics such as vancomycin or daptomycin are being used to treat MRSA bacteremia, but MRSA is often resistant to other antibiotics as well, and *Staphylococcus aureus* resistant to vancomycin or daptomycin has already appeared. 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.
Considering that the conventional antibiotic substances have a mechanism that inhibits cell wall synthesis, inhibits protein synthesis, destroys cell membranes or inhibits DNA synthesis, SAL-1, the active ingredient of the study drug (N-Rephasin® SAL200), has an antimicrobial effect that destroys a specific structure of the peptidoglycan layer in the cell wall of methicillin-resistant and methicillin-susceptible *Staphylococcus aureus*, and therefore, it acts against bacteria that have acquired resistance to conventional antibiotics regardless of their resistance.
Thus, it has an important advantage that it does not induce tolerance or resistance of pathogens while the range of use of conventional antibiotics gradually narrows as they are facing an increase in resistance, the study drug is effective for

the treatment of infectious diseases caused by *Staphylococcus aureus* and for the treatment of methicillin-resistant *Staphylococcus aureus* as a new antibiotic substance with a longer life cycle than conventional antibiotics.

This clinical study is conducted in healthy subjects, and there is no benefit to the treatment of subjects. But, the information obtained from the result of this clinical study may provide a beneficial outcome for future clinical studies to be conducted in patients with MSSA/MRSA bacteremia and their treatment.

Therefore, this study may be considered to have more potential benefits than risks.

## 3.4 Rationale for the dose selection and evaluation of the safety upon escalation

Based on the results of the repeat-dose study in primates and single-dose study in human subjects that were already conducted with the same drug, the dose setting in this clinical study was established with two aspects—safety and pharmacodynamics.

The single-dose study conducted in human subjects in 2013 was carried out with the treatment groups receiving doses of 0.1 mg/kg, 0.3 mg/kg, 1 mg/kg, 3 mg/kg, and 10 mg/kg, and no adverse event was 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 it was determined that the results were not clinically significant. However, 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 with 23 mild and 2 moderate cases. A total of 5 subjects received administration of drugs to treat adverse events, and all of them recovered without sequelae. In order to more accurately identify the maximum dose that can cause adverse events, it was determined desirable to test the 6 mg/kg single-dose group between the 3 mg/kg dose group and the 10 mg/kg dose group. Therefore, this clinical study was set to proceed with the repeat-dose group study after conducting the study with the 6 mg/kg single-dose group and completing the safety evaluation.

In addition, in the pharmacodynamic evaluation of a single-dose study, it was confirmed that the bacteria suppression effect was observed in the 1 mg/kg and 3 mg/kg dose groups for up to 1 hour after the administration, and the effect lasted for up to 1, 1.5 and 2 hours in the 10 mg/kg dose group.

The repeat-dose study conducted in primates in 2014 was carried out in 10 mg/kg/day, 20 mg/kg/day, and 40 mg/kg/day dose groups, and 3 male and 3 female animals in each group received the investigational product twice daily for 5 days. No adverse events related to the investigational product were observed in both male and female animals in the above study.

Exposure to the administration of the drug in the repeat-dose study in primates and single-dose study in human subjects can be considered as follows: The maximum dose in the repeat-dose study in primates was 40 mg/kg/day, which is 13.6 mg/kg/day when converted into the HED (multiply factor = 0.34). The $AUC_{last\ (0–12\ h)}$ measured on the first day of the administration was 66.2 h*mcg/mL in male animals and 81.6 h*mcg/mL in female animals, and the measured value observed at 5 days after completion of the administration was 49.1 h*mcg/mL in male animals and 35.6 h*mcg/mL in female animals. The maximum dose in the single-dose study conducted in human subjects was 10 mg/kg, showing no significant difference in the dose compared to that in primates, and the $AUC_{last\ (0–24\ h)}$ observed was 61.83 h*mcg/mL. When the AUC was compared in both studies, the result in the primates was similar to that in human subjects at Day 1, and the degree of exposure to primates reached 80% of the exposure to human subjects at Day 5, showing no significant difference in terms of exposure.

Considering the above information in general, the repeat-dose groups in this study to be conducted in human subjects were set as 3 mg/kg/day, 6 mg/kg/day, 9 mg/kg/day, and 12 mg/kg/day dose groups, and we would like to evaluate the safety and the duration of the bacteria suppression effect after repeated administration of the relevant doses.

In general, in the case of administering a new substance for the first time in human subjects, the safety of the investigational product is evaluated in the middle of the clinical study, and whether to continue the clinical study is determined. This clinical study is to administer the drug repeatedly in human subjects for the first time, and it has been designed to escalate the dose sequentially. Therefore, after the administration is completed in each dose group, the safety evaluation is required before proceeding to the next dose level. The medical experts will evaluate the study results, such as safety, comprehensively based on the tests and adverse events until up to 7 days after the administration in all subjects in each dose group, and the principal investigator and the sponsor will discuss the decision on the dose escalation based on the results. The test results at the post-study visit will be referenced afterward.

# 4. Objective of the clinical study

Since it is expected that the study drug N-Rephasin® SAL200 has considerable efficacy against MSSA/MRSA bacteremia in the human body, we would like to evaluate the safety and explore the characteristics of pharmacokinetics, pharmacodynamics, and immunogenicity after single and repeated continuous intravenous infusion of N-Rephasin® SAL200 for 1 hour in healthy male volunteers.

# 5. Study population

## 5.1 Target number of subjects

- Total number of subjects planned: 40 subjects

| | Daily administered amount (mg/kg/day) | Amount administered per dose (mg/kg/dose) | Study group (subjects) | Control group (subjects) |
|---|---|---|---|---|
| Single dose | 6.0 | 6.0 | 6 | 2 |
| Repeat dose | 3.0 | 1.5 | 6 | 2 |
| | 6.0 | 3.0 | 6 | 2 |
| | 9.0 | 4.5 | 6 | 2 |
| | 12.0 | 6.0 | 6 | 2 |

By setting 8 subjects (6 subjects in the study group, 2 subjects in the placebo group) for each dose level, a total of 40 subjects will be recruited.

## 5.2 Inclusion criteria

1. Healthy male adults aged 20 to 45 years at screening
2. Those whose weigh is ≥ 50 kg and < 90 kg and body mass index (BMI) is ≥ 18.0 and < 27.0
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

## 5.3 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 have hypersensitivity reactions to drugs containing N-Rephasin® SAL200 or other drugs (aspirin, antibiotics, etc.) or a history of such reactions
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 3 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 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

## 5.4 Dropout criteria

Subjects have the right to drop out from the clinical study at any time and for any reason without prejudice to subsequent treatment, and the investigator also has the authority to drop subjects out from the clinical study for the benefit of patients. In the case of dropping out, the reason for the dropout should be checked to ensure the safety of the subject, and the necessary procedures, such as observation/tests, should be performed.

**Subjects may drop out from the clinical study for the following reasons:**
1. In the case that specific reactions occurred in an allergenicity test performed during the run-in period (day of admission, Day −1)
2. In the case that a subject requests discontinuation of the study drug administration during the clinical study or withdraws the consent to participation in the study
3. In the case that serious adverse events/adverse drug reactions occur
4. In the case that a subject has arbitrarily taken any of drugs that are expected to affect the safety and pharmacokinetics of the study drug
5. Those who ingested foods containing caffeine (coffee, black tea, green tea, etc.), Korean distilled spirit, carbonated beverages, coffee milk, energy drinks, foods containing grapefruit, etc., drank alcohol or smoked cigarettes during the hospitalization period
6. In the case that the investigator believes that the study needs to be discontinued for the subject
7. In the case that a subject does not visit as scheduled without previous notice and is unreachable
8. In the case that a serious protocol deviation is newly discovered during the clinical study
9. In the case that a disease determined to be significant by investigator develops

**In the case of dropping out due to the above reasons, the case will be handled as follows:**
1. In the case of a dropout, the reason for the dropout shall be confirmed, and the test items described in the "Dropout" section in "10.2 Visit schedule" shall be performed.
2. If it is due to an adverse event, it should be reported according to the procedures described in Section 10.4.
3. Subjects who dropped out during the screening period will be considered screening failures and will be recorded as screening failures in the CRF, and they will not be further followed up.
4. In the case that a subject drops out, the reason for the dropout shall be recorded in the "Case Conclusion" in the CRF.
5. In the case of a subject who failed to comply with the protocol (failure to F/U, etc.), the case can be closed after a discussion between the sponsor and the investigator, and the subject will not be further followed up.

# 6. Clinical study design

## 6.1 Study duration

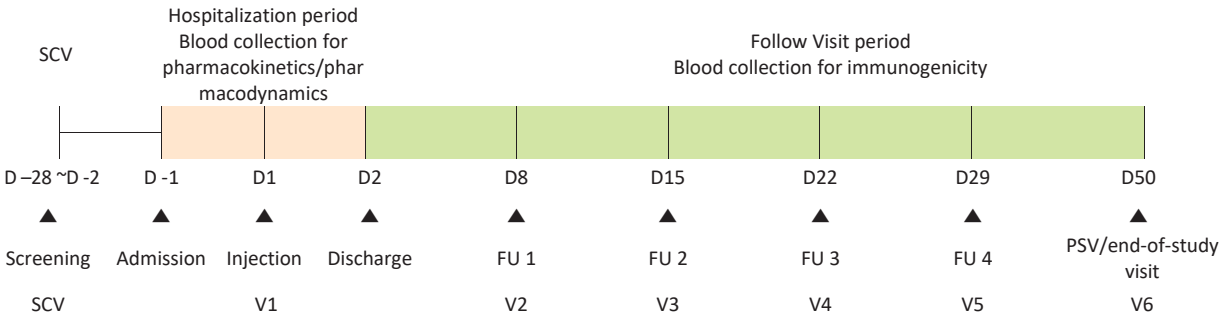
The duration of this study will be 36 months from the date of the initial approval by the Ministry of Food and Drug Safety. However, in the case of a situation that may affect the progress of the clinical study, such as a difficulty in selecting subjects, the duration may be changed.

- Expected subject recruitment duration: Approximately 9 months
- Clinical study conduct and observation duration: Approximately 2 to 3 months/subject

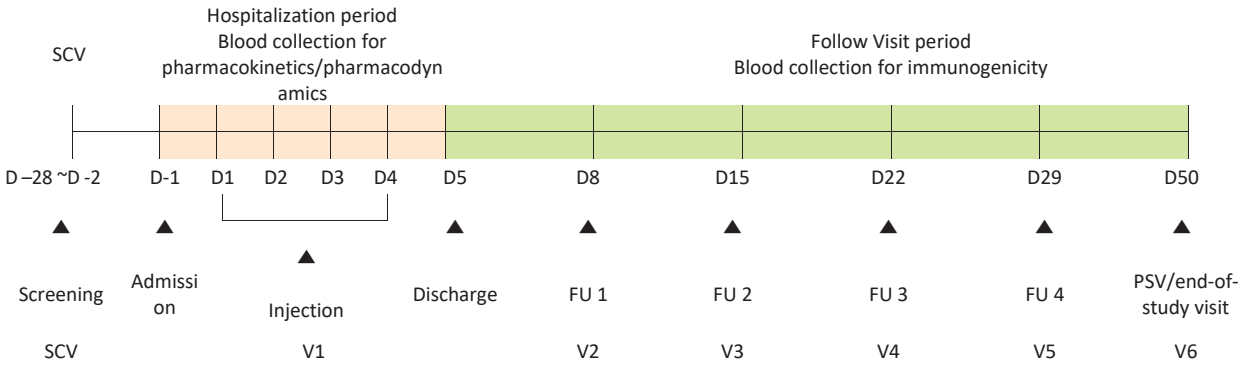
## 6.2 Study flow chart

This randomized, double-blind, placebo-controlled, single- and repeat-dose, gradual dose-escalation clinical study was conducted to explore the safety, characteristics of pharmacokinetics and pharmacodynamics, immunogenicity and allergenicity after single and repeated administration of the study drug "N-Rephasin® SAL200" only in healthy male volunteers who have been fully informed of the clinical study and have signed the informed consent form voluntarily. For this study, the eligibility of subjects was evaluated during the screening period, and those who are eligible to participate were randomized at a ratio of 3:1 to be divided into the study group to receive administration of "SAL200" and the control group (placebo) to receive administration of the "formulation buffer excluding the active ingredient of the study drug." After administering the investigational product once (single dose) or 6 times (repeat dose), the safety, pharmacokinetics, pharmacodynamics, and immunogenicity of "N-Rephasin® SAL200" were evaluated by following up for approximately 50 days.

The overall flow of the clinical study will proceed as shown in the following flow chart:




<Figure 1. Single-dose study flow>




<Figure 2. Repeat-dose study flow>

## 6.3 Study group and control group

This clinical study will be conducted with the following 2 groups: study group and control group.

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

## 6.4 Subject assignment method

### 6.4.1 Randomization

The screening numbers will be assigned in the order of the informed consent form signed by visiting the site to participate in this clinical study. Once it is confirmed in the screening that the subject meets all subject inclusion/exclusion criteria and is eligible for participation in the clinical study, the investigator will assign the subject to either the study group or the control group according to the method to operate randomization. The randomization table will be created using the program SAS Version 9.2 in order to randomize the subjects into each group at a ratio of 3:1.

### 6.4.2 Method to operate randomization

The created randomization table will be applied the IWRS (Interactive Web Response System) operated by the CRO (GDFI Braincell Laboratory), a third-party institution, and will be used to assign subjects. If subjects are found to be eligible through the screening tests until a day before the clinical study initiation (Day −1), the study site will comply with the following process in the order of passing the screening (in the order of the time of the consent provided in the case that subjects passed on the same day).

1. After entering the IWRS website into the web browser, access using the ID assigned by the IWRS administrator in accordance with the role and authority of each investigator.
   - IWRS site: http://iwrs2.gdfi.co.kr

2. Click “Subject management” in the menu tab and press the “Enrollment” button to enter the basic information of subjects (e.g., screening no.).
   - Screening no. assignment: These numbers are assigned in a form of serial numbers in the order of the informed consent form signed to participate in this study. One subject will have 1 screening no., and this number has a certain rule. The number already assigned cannot be applied to another subject.
     e.g., S001, S002, S003...; “S” stands for screening, and the following digits are in the order of the visit made. The number starts from S001, and it is assigned in the form of serial numbers without being divided according to each dose level.

3. Once you verify the entered information again and click the “Save” button, a randomization number will be assigned and the information will be saved. The format of the randomization numbers has a certain rule as shown below.
   - Randomization number format:
     Starting from R101, the numbers such as R102, R103... R108 are assigned in a form of serial numbers.
     In here, “R” in “R108” is an abbreviation of “randomization,” and the first digit “1” after “R” represents the first dose group, the second digit represents whether the subject has been replaced (e.g., 0: original subject; 1: the first replaced subject), and the third digit “8” represents the eighth subject who participated in the relevant dose level.

If a subject is replaced after a randomization number is assigned,
the second digit of the subject number, which represents whether the subject has been replaced, of the replacing subject will be changed as stated in the "Randomization number format" section above, and the investigational product to be administered to the replacing subject will receive the drug scheduled for the subject replaced after a randomization number has already been assigned.
For example, if the subject "R207" in the second dose group is replaced after being assigned according to the randomization table for "R201" to "R208," the number "R217" will be assigned to the replacing subject, and the drug assigned to "R207" (study drug or control drug) will be administered to the subject "R217."

4. The site will record this randomization number in the CRF, and the assigned randomization no. will be used as the subject identification code to identify the subject until the end of the clinical study.
   Once a subject number is assigned, it cannot be assigned to another subject, and all subjects will receive only one subject number.

## 6.5 Blinding and unblinding processes

### 6.5.1 Blinding

The double-blinding status of whether you have been assigned to receive the study drug or the control drug will be maintained—in other words, it will not be disclosed to the subject, investigators, sponsor, and other related personnel—until it is confirmed that the clinical study database is locked and the statistical analysis plan is fixed. However, the unblinded CRA and the clinical trial pharmacists will be unblinded to carry out the drug accountability.

### 6.5.2 Double-blinding method and its maintenance

To prevent subjective prejudice from the investigators and the subjects affecting the study results, this clinical study will be double-blinded. In order to conduct the clinical study double-blinded, the sponsor will manufacture and supply the control drug with the same appearance as the study drug. During the clinical study period, the study drug and control drug that were supplied with numbers pre-assigned based on the randomization code will be provided to maintain double-blinding. Unless the randomization number is disclosed, the investigator as well as the subject will not be able to distinguish the group to which they were assigned.
However, in the case of an emergency such as a serious adverse event during the study, the double-blinding code can be disclosed by "6.5.3 Emergency unblinding process" only if unblinding is required for proper clinical treatment or welfare of the subject, but the result of such disclosure does not mean unblinding of the entire clinical study.

### 6.5.3 Emergency unblinding process

In this double-blind study, the investigator or study physician may obtain information about the investigational product assigned to the subject by using the following method only in an emergency required for proper clinical management and well-being of the subject. Only the relevant subject's code should be disclosed when unblinded.

1. Request unblinding to the investigator who has the authority to unblind among the investigators who have been given the ID for the IWRS.
2. The investigator who has the authority to unblind shall login on the IWRS website.
   - IWRS site: http://iwrs2.gdfi.co.kr
3. Go to the "Subject management" section, double-click the relevant subject in the list, and click the "Unblinding request" button at the top to enter the reason for unblinding.
4. Request and receive unblinding approval from the sponsor, and identify the group to which the subject was assigned.

If the investigator determines that the case is urgent and needs to be unblinded immediately, the case can be handled by clicking "Emergency unblinding" next to the "Unblinding request" button in Step 3 above.

Before requesting unblinding, the investigator should try their best to discuss alternatives with the sponsor and the sub-investigator of iNtRON Biotechnology, Inc. (or the sub-investigator of Symyoo Inc., a CRO). Once unblinded, the investigator should inform the sponsor immediately and record the date of and reason for unblinding in writing. After unblinding, the investigator or the subject who does not remain blinded may discontinue the study. However, if the subject needs to continue the clinical study for ethical reasons, etc., the investigator may discuss with the sponsor and continue the treatment.

## 7. Clinical study termination and early termination criteria

When the last subject of the target number of subjects in this protocol completes the last visit schedule (including the early termination), the clinical study will be terminated. If the clinical study is terminated, the principal investigator should report the completion of the clinical study to the IRB with a summary of the clinical study result attached.

Also, the clinical study may be discontinued early for the following reasons during the course of the clinical study:

1. If the principal investigator determines that the situation observed while conducting the clinical study is unreasonable to continue the clinical study, the principal investigator can discontinue the clinical study, and this should be reported to the IRB.
2. The sponsor can discontinue the clinical study due to the safety of the investigational product, identification of an unfavorable risk–benefit ratio, supply of the investigational product, or other administrative or strategic reasons, and the principal investigator must report this to the IRB.
3. After the investigator evaluate the pattern of occurrence of adverse events, they will be evaluated based on the following criteria to determine whether the entire clinical study should be discontinued:
   → If moderate or worse adverse drug reactions that show a causality relationship to the study drug occur in 50% or more in one dose group or if severe or worse adverse events occur in 2 or more subjects, the subject's clinical study being conducted at the relevant time point should be discontinued, the medical expert, sponsor, and principal investigator should discuss and decide whether to continue the study afterward.
4. The IRB or pharmaceutical regulatory agencies can discontinue the clinical study when the safety or the unfavorable risk–benefit ratio of the investigational product is identified

In addition, subjects may drop out from the clinical study for the following reasons during the course of the clinical study:

1. In the case that specific reactions occurred in an allergenicity test performed during the run-in period (day of admission, Day −1) or before administration of the investigational product at Day 1
2. In the case that a subject requests discontinuation of the study drug administration during the clinical study or withdraws the consent to participation in the study
3. In the case that serious adverse events/adverse drug reactions occur
4. In the case that a subject has arbitrarily taken any of drugs that are expected to affect the safety and pharmacokinetics of the study drug
5. Those who ingested foods containing caffeine (coffee, black tea, green tea, etc.), Korean distilled spirit, carbonated beverages, coffee milk, energy drinks, foods containing grapefruit, etc., drank alcohol or smoked cigarettes during the hospitalization period
6. In the case that the investigator believes that the study needs to be discontinued for the subject
7. In the case that a subject does not visit as scheduled without previous notice and is unreachable
8. In the case that a serious protocol deviation is newly discovered during the clinical study
9. In the case that a disease determined to be significant by investigator develops

In the case that the clinical study has been terminated early or discontinued due to the situations above, the case should be handled as follows:

1. In the case that discontinuation occurs, the reason for discontinuation should be confirmed. If it is due to an adverse event, it should be reported in accordance with the procedures described in Section 10.4.
2. In the case that the principal investigator judges that it is difficult to continue the clinical study, the principal investigator should have a preliminary discussion with the sponsor and submit a letter of explanation of detailed reasons for early termination and discontinuation.
3. In the case that the IRB determines early termination/discontinuation of the clinical study, the head of the study site should notify the Minister of Food and Drug Safety immediately and submit a letter of explanation of detailed reasons for early termination.
4. In the case that the sponsor terminates the clinical study early or discontinues the clinical study, the sponsor should report the relevant decision and the reason for such a decision to the principal investigator and the Minister of Food and Drug Safety and notify the principal investigators of other study sites of the relevant decision and the reason for such a decision in writing.
5. In the case that the clinical study is terminated early, the principal investigator should inform the subject immediately and ensure that appropriate action is taken and follow-up is carried out.

# 8. Information of the investigational product and its management

## 8.1 Composition of the investigational product

### 8.1.1 Study drug

| | |
|---|---|
| Product name | N-Rephasin® SAL200 |
| Active ingredient | SAL200 1 mL (18 mg/mL as SAL-1) |
| Appearance and dosage form | Injection filled with a colorless clear liquid in a colorless transparent vial |
| Storage method and shelf life | Store a sealed container in a freezer at −70 ± 5°C; 18 months from the date of manufacture |
| Manufacturer | BINEX Co., Ltd. (entire process contracted) |

### 8.1.2 Control drug (placebo)

| | |
|---|---|
| Product name | INT200-Placebo (Saline) |
| Active ingredient | Remaining formulation buffer excluding the active ingredient of the study drug |
| Appearance and dosage form | Injection filled with a colorless clear liquid in a colorless transparent vial |
| Storage method and shelf life | Store a sealed container in a freezer at −70 ± 5°C; 18 months from the date of manufacture |
| Manufacturer | BINEX Co., Ltd. (entire process contracted) |

## 8.2 Labeling and packaging of the investigational product

### 8.2.1 Labeling

In accordance with Article 69 Paragraph 6 of the Regulation on Safety of Pharmaceuticals, etc., the sponsor will attach a label with the following information on the container or packaging of the investigational product and provide it to the IP manager of the study site.

① "For Clinical Trial Use Only" indication
② Code name of the product or generic name of the active ingredient
③ Lot number and shelf life (effective date) or re-test date
④ Storage method
⑤ Name and address of the Investigational New Drug application holder
⑥ "DO NOT USE FOR OTHER PURPOSES THAN CLINICAL TRIALS" indication

### 8.2.2 Packaging
Packaging unit: 1 mL (1 vial)

## 8.3 Route of administration
Administer intravenously.

## 8.4 Method of administration
Calculate the dose of the investigational product for the subject, and continue the intravenous infusion of the solution diluted in saline for 1 hour.

① Calculate the total amount administered(mL) taking into account the concentration per dose for each dose group and the subject's weight.

※ Total amount administered (mL) = Dose of the investigational product (mg) ÷ 18 mg/mL (study drug concentration)
↳Weight of the subject (kg) × **Concentration per dose by dose group* (mg/kg)**

| | Single dose | Repeat dose | | | |
|---|---|---|---|---|---|
| Dose group | 6.0 mg/kg | 3.0 mg/kg | 6.0 mg/kg | 9.0 mg/kg | 12.0 mg/kg |
| **Concentration per dose*** | **6.0 mg/kg** | **1.5 mg/kg** | **3.0 mg/kg** | **4.5 mg/kg** | **6.0 mg/kg** |

② Dilute the total amount administered (mL) calculated in ① in normal saline to have 110 mL as the volume of the entire injection solution.
③ Administer the injection solution prepared in ② to the subject intravenously.

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

## 8.6 Accountability
The investigational products will be manufactured by BINEX Co., Ltd., a company where manufacturing is contracted by the sponsor iNtRON Biotechnology, Inc., and supplied to the study site, and they will be managed by the clinical trial pharmacist designated by the head of the study site. Whenever the investigational products are delivered, BINEX Co., Ltd. will deliver the quality assurance certificate for the investigational products, and the clinical trial pharmacist must confirm the receipt and quantity of the investigational products provided by the sponsor and sign in writing.
The sponsor should prepare guidelines on how to receive, handle, and store the investigational products and provide the guidelines to the investigators and managers, and the investigational products should be stored under the storage conditions defined by the sponsor.
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 contact the unblinded CRA to mutually confirm the accountability of all investigational products used in the clinical study.

The investigational products will be dispensed only when prescribed by the principal investigator or co-investigators, and the randomization number of the subject, the date and quantity of dispensation, etc. will be recorded in the investigational product accountability log. In addition, the clinical trial pharmacist should accurately record the quantity of the investigational product supplied, the quantity of dispensation, and the quantity of remaining drugs, and all drugs not used at the end of the clinical study should be returned to the sponsor.

The investigator, who has been delegated as an investigational product manager, is responsible for managing and keeping records until the investigational product provided by the sponsor is received and administered. When the quantity of the drugs administered to the subject or in the case the drugs needs to be returned to the sponsor, the quantity of the relevant drugs should be checked and documented, and all investigational products should be stored in a secured space and managed to be handled only by those who are authorized to access the investigational products.
In any case, the investigator delegated to be the investigational product manager should not supply the investigational products 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.

### 8.7 Management, collection, and destruction

After administration of the investigational products, destruction, etc. of the investigational products supplied to the study site should be carried out by the investigational product manager in accordance with the hospital regulations, and the relevant information should be documented and stored. After the end of the clinical study, the unopened investigational products will be collected by the sponsor.

### 8.8 Management and destruction of the collected tissues

The collected blood and urine samples should be stored and tested according to the storage method of each assessment item in the protocol. The storage and destruction of the remaining samples after the measurement will be carried out in accordance with the items related to the storage of samples in the “SOP for samples” of the Department of Clinical Pharmacology and Therapeutics, Seoul National University Hospital, and the decision of whether to destroy them will be made after a discussion between the sponsor and the principal investigator.

## 9. Method of the clinical study and administration plan, etc.

### 9.1 Administration and treatment schedule

Subjects will receive either the study drug or control drug assigned through randomization.

- Study drug: N-Rephasin® SAL200
- Control drug: INT200-Placebo (Saline)

This clinical study will conduct a gradual dose escalation, repeat-dose study after administration of a single dose at 6 mg/kg.
Except for the schedule of the duration of hospitalization, the single-dose and repeat-dose groups will have the same clinical study schedule for the screening, outpatient period, and post study visit.

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| **IP administration schedule for the single-dose group** | **Screening** | **Hospitalization** | | | **Outpatient period** | | | | **Post study visit** |
| | Screening Visit (D-28~D-2) | Day -1 | **Day 1** | Day 2 | D8 ± D1 | D15 ± D1 | D22 ± D1 | D29 ± D1 | 50±D2 |
| | Wash-out | admission | **Single-dose administration and blood collection for pharmacodynamics** | discharge | Blood collection for allergenicity (D8, D15, D29) and blood collection for immunogenicity | | | | Blood collection for immunogenicity |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| **IP administration schedule for the repeat-dose group** | **Screening** | **Hospitalization** | | | **Outpatient period** | | | | **Post study visit** |
| | Screening Visit (D-28~D-2) | Day -1 | **Day 1 ~4** | Day 5 | D8 ± D1 | D15 ± D1 | D22 ± D1 | D29 ± D1 | 50±D2 |
| | Wash-out | admission | **Repeat-dose administration (BID, 6 doses in total) and blood collection for pharmacodynamics** | discharge | Blood collection for allergenicity (D8, D15, D29) and blood collection for immunogenicity | | | | Blood collection for immunogenicity |

## 9.2 Medications allowed to be administered concomitantly

1. In principle, no drugs other than the investigational products specified in the protocol should be administered during the study period; however, drugs may be administered at the discretion of the investigator if they are necessary for treatment of adverse events, etc.

## 9.3 Medications and treatments prohibited from being administered and performed concomitantly

1. All medications other than those allowed to be administered concomitantly are prohibited.

   However, if there is a drug that was administered arbitrarily without the investigator's judgment, the investigator will determine whether it may affect the safety and pharmacokinetic evaluation of this clinical study, and the subject will drop out if it is expected that the drug will have an effect.
2. The following are prohibited as well:
   Ingestion of foods containing caffeine (coffee, black tea, green tea, etc.), Korean distilled spirit, carbonated beverages, coffee milk, energy drinks, foods containing grapefruit, etc., drinking alcohol and smoking cigarettes

All drugs administered and reasons for administration must be documented in the case report form.

### 9.3 Treatment compliance

Since the study drug and the control drug are administered only by the investigator after being admitted to the study site, it will be managed based on the investigational product administration status.

## 10. Clinical study process and assessments

### 10.1 Study schedule table

During the clinical study period, subjects participating in the clinical study will have 1 inpatient visit and approximately 5 outpatient visits to the study site after being confirmed eligible at screening.
The subjects deemed eligible for this clinical study will be selected by performing screening tests, such as history taking, physical examination, clinical laboratory tests, and allergenicity testing, in volunteers who provided a written consent to participate in this clinical study within 4 weeks (Day −28 to Day −2) from a day before the clinical study initiation (Day −1). Then, the subjects will visit the research unit of the Seoul National University Hospital Clinical Trials Center in the afternoon of a day before the clinical study initiation (Day −1), and they will have an allergenicity test. Subjects who do not have specific reactions will be admitted, and subject numbers will be assigned through randomization. The subjects will fast from 10 p.m. on the day of admission (Day −1), except for drinking water.

In each dose group, the study drug or placebo will be infused intravenously in all subjects, depending on the relevant dose group by the randomization table generated in advance, at around 9 a.m. on the next day of the admission, and the clinical study will be conducted according to the clinical study schedule.
After completion of the administration in each dose group, the safety will be evaluated to determine whether to proceed to the next dose group.

The schedule for planned visits are as shown in Table 1 below.

[Table 1. Clinical study schedule for the single-dose group]

| Observation and Test Items | Schedule of Clinical Study | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Screening Period | Admission Period | | | Outpatient Period | | | | Post-study Visit |
| Visit Number | SCV | V1 | | | V2 | V3 | V4 | V5 | V6 |
| | Screening | Hospitalization | | | FU1 | FU2 | FU3 | FU4 | PSV |
| | D-28~D-2 | D-1 | D1 | D2 | D8±D1 | D15±D1 | D22±D1 | D29±D1 | D50±D2 |
| Written consent | ✘ | | | | | | | | |
| Demographic information/medical history | ✘ | | | | | | | | |
| Inclusion/exclusion criteria | ✘ | ✘ | | | | | | | |
| Randomization | | ✘ | | | | | | | |
| Admission/discharge[1] | | ✘ | | ✘ | | | | | |
| Administration of the investigational product[2] | | | ✘ | | | | | | |
| Vital signs[3] | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Electrocardiogram[4] | ✘ | | ✘ | ✘ | | ✘ | | | ✘ |
| Clinical laboratory tests[5] | ✘ | | ✘ | ✘ | ✘ | ✘ | | | ✘ |
| Breath alcohol test | ✘ | | | | | | | | |
| Physical examination[6] | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Blood collection for pharmacokinetics[7] | | | ✘ | ✘ | | | | | |
| Urine collection for pharmacokinetics[8] | | | ✘ | ✘ | | | | | |
| Blood collection for pharmacodynamics[9] | | | ✘ | | | | | | |
| Immunogenicity assessment[10] | | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Allergenicity test[11] | ✘ | ✘ | | | ✘ | ✘ | | ✘ | |
| Checking on adverse events | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Checking on medication history/concomitant medications | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Checking on whether medical treatment has been given | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |

Note

1. The subject will be admitted at 1 p.m. of Day −1 and will be discharged after completing the entire schedule of Day 2.
2. All schedules will be carried out based on the date and time of the first administration of the IP. [Date and time of the first IP administration - Day 1: 0 h]
3. Vital signs (sitting blood pressure/pulse rate/temperature) will be measured at SCV, V1, V2, V3, V4, V5, and V6 and will be measured in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day-1 Day 1 : pre-dose, 1h, 4h, 8h, 12h, 13h. • Day 2 : 0h.
4. Electrocardiogram will be obtained at SCV, V1, V3, and V6 and will be measured in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : pre-dose, 0h~2h(continuous), 4h. • Day 2 : 0h.

5. Clinical laboratory tests will be performed at SCV, V1 (D1: pre-dose; D2: 0 h), V2, V3, and V6.
6. Physical examination will be performed at SCV, V1 (D−1; D1: pre-dose, 1.5 h; D2: 0 h), V2, V3, V4, V5, and V6.
7. Blood collection for pharmacokinetics will be performed in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : pre-dose, 0.25h, 0.5h, 0.75h, 1h, 1.5h, 2h, 3h, 4h, 5h, 6h, 8h, 10h, 12h.
   - Day 2 : 0h.
8. Urine collection for pharmacokinetics will be performed in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : 0~4h, 4~12h, 12~24h.
9. Blood collection for pharmacodynamics will be performed in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : pre-dose, 1h, 1.5h, 2h.
10. An immunogenicity assessment will be performed at V1 (D1: pre-dose; D2: 0 h), V2, V3, V4, V5, and V6.
11. An allergenicity test (Standard allergy skin test) will be performed at SCV, V1 (D−1), V2, V3, and V5.

[Table 2. Clinical study schedule for the repeat-dose group]

| Observation and Test Items | Schedule of Clinical Study | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Screening Period | Admission Period | | | | | | Outpatient Period | | | | Post-study Visit |
| Visit Number | SCV | V1 | | | | | | V2 | V3 | V4 | V5 | V6 |
| | Screening | Hospitalization | | | | | | FU1 | FU2 | FU3 | FU4 | PSV |
| | D-28 ~D-2 | D-1 | D1 | D2 | D3 | D4 | D5 | D8± D1 | D15± D1 | D22± D1 | D29± D1 | D50±D2S |
| Written consent | ✘ | | | | | | | | | | | |
| Demographic information/medical history | ✘ | | | | | | | | | | | |
| Inclusion/exclusion criteria | ✘ | ✘ | | | | | | | | | | |
| Randomization | | ✘ | | | | | | | | | | |
| Admission/discharge[1] | | ✘ | | | | | ✘ | | | | | |
| Administration of the investigational product[2] | | | ✘ | ✘ | ✘ | ✘ | | | | | | |
| Vital signs[3] | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Electrocardiogram[4] | ✘ | | ✘ | ✘ | ✘ | ✘ | ✘ | | ✘ | | | ✘ |
| Clinical laboratory tests[5] | ✘ | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | | | ✘ |
| Breath alcohol test | ✘ | | | | | | | | | | | |
| Physical examination[6] | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Blood collection for pharmacokinetics[7] | | | ✘ | ✘ | ✘ | ✘ | ✘ | | | | | |
| Urine collection for pharmacokinetics[8] | | | ✘ | ✘ | | ✘ | ✘ | | | | | |
| Blood collection for pharmacodynamics[9] | | | ✘ | | | ✘ | | | | | | |
| Immunogenicity assessment[10] | | | ✘ | | | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Allergenicity test[11] | ✘ | ✘ | | | | | | ✘ | ✘ | | ✘ | |
| Anaphylatoxin test[12] | | | ✘ | | | ✘ | | | | | | |

| | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Checking on adverse events | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Checking on medication history/concomitant medications | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |
| Checking on whether medical treatment has been given | | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ | ✘ |

Note

1. The subject will be admitted at 1 p.m. of Day −1 and will be discharged after completing the entire schedule of Day 5.
2. All schedules will be carried out based on the date and time of the first administration of the IP. [Date and time of the first IP administration - Day: 0 h]
3. Vital signs (sitting blood pressure/pulse rate/temperature) will be measured at SCV, V1, V2, V3, V4, V5, and V6 and will be measured in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day-1
   - Day 1 / Day 4 : pre-dose, 1h, 4h, 8h, 12h, 13h.
   - Day 2 / Day 3 : pre-dose, 1h, 12h, 13h.
   - Day 5 : 0h.
4. Electrocardiogram will be obtained at SCV, V1, V3, and V6 and will be measured in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : pre-dose, 4h.
   - Day 2 / Day 3 / Day 4 : 0h, 4h.
   - Day 5 : 0h.
   - Day 1 / Day 4 : 0~2h(continuous).
5. Clinical laboratory tests will be performed at SCV, V1 (D1: pre-dose; D2, D3, D4, D5: 0 h), V2, V3, and V6.
6. Physical examination will be performed at SCV, V1 (D−1; D1: pre-dose, 1.5 h; D2, D3, D4: 0 h, 1.5 h; 5D: 0 h), V2, V3, V4, V5, and V6.
7. Blood collection for pharmacokinetics will be performed in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : pre-dose, 0.25h, 0.5h, 0.75h, 1h, 1.5h, 2h, 3h, 4h, 5h, 6h, 8h, 10h, 12h.
   - Day 2 / Day3 : 0h, 1h, 12h.
   - Day 4 : 0h, 0.25h, 0.5h, 0.75h, 1h, 1.5h, 2h, 3h, 4h, 5h, 6h, 8h, 10h, 12h.
   - Day 5 : 0h.
8. Urine collection for pharmacokinetics will be performed in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 / Day 4 : 0~4h, 4~12h, 12~24h.
9. Blood collection for pharmacodynamics will be performed in accordance with the detailed schedule below at V1 (hospitalization period).
   - Day 1 : pre-dose, 1h, 1.5h, 2h. ・Day 4 : 0h, 1h, 1.5h, 2h.
10. An immunogenicity assessment will be performed at V1 (D1: pre-dose; D5: 0 h), V2, V3, V4, V5, and V6.
11. An allergenicity test (Standard allergy skin test) will be performed at SCV, V1 (D−1), V2, V3, and V5.
12. An anaphylatoxin test will be performed on C3a, C4a, mast cell tryptase, IL-1b, IL-2, IL-6, and TNF-alpha in accordance with the detailed schedule below at V1 (hospitalization period).
    - Day 1 : pre-dose, 1h ・Day 4 : 0h, 1h

## 10.2 Visit schedule

The study procedures for each clinical study schedule planned are as follows:

■ **Screening Visit [SCV: D-28 ~ D-2]**

The screening tests to review the eligibility of the subjects who voluntarily provided a written consent to participate in the clinical study will be performed within 4 weeks from the expected day of the initial administration (D−28 to D−2), and any subjects who have clinically significant abnormalities in the following tests shall be excluded:

1) Demographic information and history taking

: Check demographic information, such as gender and age, of the subject, and examine the past medical history, recent medical history, and medication history. In addition, measure the height and weight to check the weigh is ≥ 50 kg and < 90 kg and the body mass index (BMI) is ≥ 18.0 and < 27.0.

☞ BMI (kg/m$^2$) = Weight (kg) / {Height (m)}$^2$

Round the height (cm) to the nearest whole number, and round the weight (kg) to the nearest tenth for the

record.

2) Vital signs and physical examination
: Measure the blood pressure and pulse rate after maintaining a sitting position for 3 minutes or longer without any sudden posture change. Measure the tympanic membrane temperature, and perform the physical examination.

3) Clinical laboratory tests
- Hematology
  : WBC with differential count (segmented neutrophil, lymphocyte, monocyte, eosinophil, basophil), RBC, hemoglobin, hematocrit, platelets
- Blood coagulation tests: PT(INR)/aPTT
- Blood chemistry
  : Calcium, phosphorus, glucose, BUN, uric acid, cholesterol, total protein, albumin, total bilirubin, alkaline phosphatase, AST, ALT, γ-GT, CPK, LDH, creatinine, sodium, potassium, chloride, triglyceride, amylase, lipase, ESR, hs-CRP
- Urinalysis
  : Color, pH, specific gravity, albumin, bilirubin, glucose, urobilinogen, ketone, nitrite, occult blood, leukocyte, microscopy
- Urine drug screening (to be performed only at screening)
  : Amphetamines, barbiturates, benzodiazepines, cannabinoids, cocaine, cotinine, opiates
- Serology (to be performed only at screening): HBsAg, anti-HCV Ab, anti-HIV Ab, RPR

4) Breath alcohol test (to be performed only at screening)

5) Allergenicity test
: After taking a certain amount of the study drug and placebo from 1 of each vial, dilute the active ingredient to 0.05% (v/v) with normal saline, and perform a standard allergy skin test (skin reaction test) on both arms against each of the clinical drugs (right arm: placebo; left arm: study drug) using the solution at a concentration of 0.009 mg/mL. Subjects who show specific reactions will be screened out.

a. Injection site: Inside the forearm
b. 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 mL of the solution at a concentration of approximately 0.009 mg/mL) 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.
c. Acceptance criteria
   ① If a wheal of at least 3 mm diameter appears along with redness: Positive
   ② If there is a wheal of at least 3 mm diameter but no redness: Undetermined and re-test required
   ③ If the diameter of a wheal is 3 mm or smaller regardless of redness: Negative
   ④ Retest if the result is equivocal

6) Electrocardiogram (12-lead ECG)
: In addition to the basic records, record the ventricular rate (beats/min), PR interval (msec), QRS (msec), and QT/QTc (msec) with automatic analysis & recording.

7) Check the results of 1) to 5) during the screening tests, and inform the subjects, who meet the inclusion/exclusion criteria based on the relevant test results, of the inpatient visit.

■ **Hospitalization schedule for the single-dose group [V1: D−1 to D2 (injection)]**

All subjects will be admitted to Clinical Trials Center by 1 p.m. at D−1. At this time, the subjects will have the following procedures:

**- Run-in period [D−1, day of admission]**

1) Vital signs and physical examination
2) Allergenicity test
: After the admission, perform the allergenicity test again to confirm the final inclusion/exclusion criteria.

Subjects with specific reactions will drop out and be replaced by other backup subjects.

3) Randomization number assignment
: Assign only to the eligible subjects who meet the inclusion/exclusion criteria.
4) Adverse event monitoring
5) Checking on concomitant medications
6) Checking on medical treatment given

**- Administration of the investigational product and various tests [D1 to D2]**

The time points for all of the items below are based on 0 h of D1 (basis) as the time of administration of the investigational product.

1) Administration of the investigational product
① D1: At around 9 a.m. (0 h)
2) Vital signs (sitting blood pressure/pulse rate/temperature)
Measure the blood pressure, pulse rate, and temperature after maintaining a sitting position for 3 minutes or longer without any sudden posture change.
① D1 : pre-dose, 1, 4, 8, 12, 13 h. ② D2 : 0 h.
3) Electrocardiogram (12-lead ECG)
① D1 : pre-dose, 4 h. ② D2 : 0 h.
4) Electrocardiogram (continuous ECG monitoring)
① D1 : 0 ~ 2 h.
5) Clinical laboratory tests (hematology, blood coagulation tests, blood chemistry, urinalysis)
① D1 : pre-dose. ② D2 : 0 h.
6) Physical examination
① D1 : pre-dose, 1.5 h. ② D2 : 0 h.
7) Blood collection for pharmacokinetics
① D1 : pre-dose, 0.25 h, 0.5 h, 0.75 h, 1 h, 1.5 h, 2 h, 3 h, 4 h, 5 h, 6 h, 8 h, 10 h, 12 h.
② D2 : 0h.
8) Urine collection for pharmacokinetics
① D1 : 0~4 h, 4~12 h, 12~24 h.
9) Blood collection for pharmacodynamics (ex vivo antibacterial activity)
① D1 : pre-dose, 1 h, 1.5 h, 2 h.
10) Immunogenicity assessment (analysis of concentration of anti-drug antibody) - See <Appendix 4>.
① D1 : pre-dose. ② D2 : 0 h.
11) Adverse event monitoring, checking on concomitant medications and medical treatment
: Check adverse events and concomitant medications regularly through the investigator's history taking, etc. in addition to voluntary reports by subjects. For adverse events, record the detailed information on the symptoms, time of appearance, duration, severity of adverse events, and causality relationship. For concomitant medications, record the detailed information including the name of drug (product name or substance name), route of administration, daily dose, duration of administration, and reason for administration. For medical treatment, record the detailed information including the name of treatment, indication, site of treatment, and duration of treatment.
12) Meal
: Subjects will fast from 10:00 p.m. on the day before the administration (D−1), except for drinking water, and the drug will be administered at D1 while fasting. Subjects will fast for 4 hours after the administration, and lunch and dinner will be provided afterwards.
13) Discharge
: Subjects will be discharged after completing the entire schedule for until D2.

■ **Hospitalization schedule for the repeat-dose group [V1: D−1 to D5 (injection)]**

All subjects will be admitted to Clinical Trials Center by 1 p.m. at D−1. At this time, the subjects will have the following procedures:

**- Run-in period [D−1, day of admission]**

1) Vital signs and physical examination
2) Allergenicity test
   : After the admission, perform the allergenicity test again to confirm the final inclusion/exclusion criteria. Subjects with specific reactions will drop out and be replaced by other backup subjects.
3) Randomization number assignment
   : Assign only to the eligible subjects who meet the inclusion/exclusion criteria.
4) Adverse event monitoring
5) Checking on concomitant medications
6) Checking on medical treatment given

**- Administration of the investigational product and various tests [D1 to D5]**

The time points for all of the items below are based on 0 h of D1 (basis) as the time of administration of the investigational product.

1) Administration of the investigational product
   ① D1: At around 9 a.m. (0 h)
   ② D2: At around 9 a.m. and at around 9 p.m.
   ③ D3: At around 9 a.m. and at around 9 p.m.
   ④ D4: At around 9 a.m.
2) Vital signs (sitting blood pressure/pulse rate/temperature)
   : Measure the blood pressure, pulse rate, and temperature after maintaining a sitting position for 3 minutes or longer without any sudden posture change.
   ① D1 : pre-dose, 1 h, 4 h, 8 h, 12 h, 13 h.
   ② D2 : pre-dose, 1 h, 12 h, 13 h.
   ③ D3 : pre-dose, 1 h, 12 h, 13 h.
   ④ D4 : pre-dose, 1 h, 4 h, 8 h, 12 h, 13 h.
   ⑤ D5 : 0 h.
3) Electrocardiograms (12-lead ECG)
   ① D1 : pre-dose, 4 h.
   ② D2 : 0 h, 4 h.
   ③ D3 : 0 h, 4 h.
   ④ D4 : 0 h, 4 h.
   ⑤ D5 : 0 h.
4) Electrocardiogram (continuous ECG monitoring)
   ① D1 : 0 ~ 2 h.
   ② D4 : 0 ~ 2 h.
5) Clinical laboratory tests (hematology, blood coagulation tests, blood chemistry, urinalysis)
   ① D1 : pre-dose.
   ② D2/D3/D4/D5: 0 h.
6) Physical examination
   ① D1 : pre-dose, 1.5 h.
   ② D2/D3/D4 : 0, 1.5 h.
   ③ D5 : 0 h.
7) Blood collection for pharmacokinetics
   ① D1: pre-dose, 0.25 h, 0.5 h, 0.75 h, 1 h, 1.5 h, 2 h, 3 h, 4 h, 5 h, 6 h, 8 h, 10 h, 12 h.
   ② D2 : 0 h, 1 h, 12 h.
   ③ D3 : 0 h, 1 h, 12 h.
   ④ D4 : 0 h, 0.25 h, 0.5 h, 0.75 h, 1 h, 1.5 h, 2 h, 3 h, 4 h, 5 h, 6 h, 8 h, 10 h, 12 h.
   ⑤ D5 : 0 h.
8) Urine collection for pharmacokinetics
   ① D1 : 0~4 h, 4~12 h, 12~24 h.
   ② D4 : 0~4 h, 4~12 h, 12~24 h.
9) Blood collection for pharmacodynamics (ex vivo antibacterial activity)

① D1 : pre-dose, 1 h, 1.5 h, 2 h.
② D4 : 0 h, 1 h, 1.5 h, 2 h.

10) Immunogenicity assessment (analysis of concentration of anti-drug antibody)
① D1 : pre-dose.
② D5 : 0 h.

11) Anaphylatoxin test
: C3a, C4a, mast cell tryptase, IL-1b, IL-2, IL-6, TNF-alpha
① D1 : pre-dose, 1 h.
② D4 : 0 h, 1 h.

12) Adverse event monitoring, checking on concomitant medications and medical treatment
: Check adverse events and concomitant medications regularly through the investigator's history taking, etc. in addition to voluntary reports by subjects. For adverse events, record the detailed information on the symptoms, time of appearance, duration, severity of adverse events, and causality relationship. For concomitant medications, record the detailed information including the name of drug (product name or substance name), route of administration, daily dose, duration of administration, and reason for administration. For medical treatment, record the detailed information including the name of treatment, indication, site of treatment, and duration of treatment.

13) Meal
① D1, D4: : Subjects will fast from 10:00 p.m. on the day before the administration, except for drinking water, and the drug will be administered at D1 while fasting. Subjects will fast for 4 hours after the administration, and lunch and dinner will be provided afterwards.
② D2, D3: Breakfast, lunch, and dinner will be provided.

14) Discharge
: Subjects will be discharged after completing the entire schedule for until D5.

■ **Outpatient visits [D8±D1(V2), D15±D1(V3), D22±D1(V4), D29±D1(V5)]**

For clinical laboratory tests, physical examination, immunogenicity assessment, and allergenicity test to evaluate the safety, subjects will visit Clinical Trials Center at D8, D15, D22, and D29. At this time, all subjects will have the following procedures:

**- FU 1 [V2: D8±D1]**

1) Vital signs (sitting blood pressure/pulse rate/temperature)
2) Clinical laboratory tests (hematology, blood coagulation tests, blood chemistry, urinalysis)
3) Physical examination
4) Immunogenicity assessment
5) Allergenicity test
6) Adverse event monitoring
7) Checking on concomitant medications
8) Checking on medical treatment given

**- FU 2 [V3: D15±D1]**

1) Vital signs (sitting blood pressure/pulse rate/temperature)
2) Physical examination
3) 12-lead ECG (electrocardiogram)
4) Clinical laboratory tests (hematology, blood coagulation tests, blood chemistry, urinalysis)
5) Immunogenicity assessment
6) Allergenicity test
7) Adverse event monitoring
8) Checking on concomitant medications
9) Checking on medical treatment given

**- FU 3 [V4: D22±D1]**

1) Vital signs (sitting blood pressure/pulse rate/temperature)
2) Physical examination
3) Immunogenicity assessment
4) Adverse event monitoring
5) Checking on concomitant medications
6) Checking on medical treatment given

**- FU 4 [V5: D29±D1]**

1) Vital signs (sitting blood pressure/pulse rate/temperature)
2) Physical examination
3) Immunogenicity assessment
4) Allergenicity test
5) Adverse event monitoring
6) Checking on concomitant medications
7) Checking on medical treatment given

■ **Post-study visit [V6: D50±D2]**

Considering the subject's convenience, perform the following observation and tests for the subject between D48 and D52:

1) Vital signs (sitting blood pressure/pulse rate/temperature)
2) Physical examination
3) 12-lead ECG (electrocardiogram)
4) Clinical laboratory tests (hematology, blood coagulation tests, blood chemistry, urinalysis)
5) Immunogenicity assessment
6) Adverse event monitoring
7) Checking on concomitant medications
8) Checking on medical treatment given

■ **Unscheduled Visit**

If a subject visits the site other than the scheduled visit based on the schedule above, such a visit will be managed as an unscheduled visit, and this visit should not change the planned study schedule.
Basically, the items shown below are performed at this visit, and other safety test items may be performed at the discretion of the investigator.

1) Vital signs (sitting blood pressure/pulse rate/temperature)
2) Physical examination
3) 12-lead ECG (electrocardiogram)
4) Clinical laboratory tests (hematology, blood coagulation tests, blood chemistry, urinalysis)
5) Adverse event monitoring
6) Checking on concomitant medications
7) Checking on medical treatment given

■ **Dropout**

If a subject drops out during the outpatient visit period (D8 ± 1 d to D29 ± 1 d), the subject will be requested to visit Clinical Trials Center within 1 week if possible to perform observation and tests for the post-study visit.
If a subject who receive the administration at least once drops out during the hospitalization period (single-dose group: D−1 to D2; repeat-dose group: D−1 to D5), the scheduled dose will be discontinued, and the subject will be discharged after completing the vital signs, electrocardiogram, clinical laboratory tests, physical examination, and monitoring of adverse events scheduled by the next morning if possible. After discharge, the subject will be requested to visit Clinical Trials Center within 1 week if possible to perform observation and tests for the post-study visit.

■ **Window period for each observation item**

Each window period is only applicable to the inpatient visit.

| Observation item | Window | |
|---|---|---|
| Vital signs | Before the administration (pre-dose, 0 h) | Within 60 minutes of the scheduled time |
| | After the administration | ±30 minutes of the scheduled time |
| Electrocardiogram | Before the administration (pre-dose, 0 h) | Within 60 minutes of the scheduled time |
| | After the administration | ±30 minutes of the scheduled time |
| Clinical laboratory tests | Before the administration (pre-dose, 0 h) | Within 60 minutes of the scheduled time |
| | After the administration | ±30 minutes of the scheduled time |
| Blood collection for pharmacokinetics | Before the administration (pre-dose, 0 h) | Within 30 minutes of the scheduled time |
| | Up to 4 h after the administration | ±5 minutes of the scheduled time |
| | 4 h after the administration | ±15 minutes of the scheduled time |
| Urine collection for pharmacokinetics | Before the administration (pre-dose, 0 h) | Within 60 minutes of the scheduled time |
| | After the administration | ±30 minutes of the scheduled time |
| Blood collection for pharmacodynamics | ±10 minutes of the scheduled time | |
| Immunogenicity assessment | Within 60 minutes of the scheduled time | |
| Anaphylatoxin test | ±10 minutes of the scheduled time | |

## 10.3 Pharmacokinetic, pharmacodynamic, immunogenicity, and safety endpoints and evaluation

### 10.3.1 Basic information items

1. Demographic information: Check the subject's gender, age, height, weight, BMI, smoking history, drinking history, etc.
2. Medical history: Medical and surgical history and history of surgery (within the last 1 month)
3. Medication history/concomitant medications: For medication history, check the history for 2 weeks (14 days) prior to the day of the screening visit.
4. Vital signs: Sitting blood pressure, pulse, and temperature
5. Physical examination
6. Clinical laboratory tests
   1) Hematology: WBC with differential count (segmented neutrophil, lymphocyte, monocyte, eosinophil, basophil), RBC, hemoglobin, hematocrit, platelets
   2) Blood coagulation tests: PT (INR)/aPTT
   3) Blood chemistry: Calcium, phosphorus, glucose, BUN, uric acid, cholesterol, total protein, albumin, total bilirubin, alkaline phosphatase, AST, ALT, γ-GT, CPK, LDH, creatinine, sodium, potassium, chloride, triglyceride,

amylase, lipase, ESR, hs-CRP
   4) Urinalysis: Color, pH, specific gravity, albumin, bilirubin, glucose, urobilinogen, ketone, nitrite, occult blood, leukocyte, microscopy
   5) Urine drug screening: amphetamins, barbiturates, benzodiazepines, cannabinoids, cotinine, cocaine, opiates (to be performed only at screening)
   6) Serology: HBsAg, anti-HCV Ab, anti-HIV Ab, RPR (to be performed only at screening)
7. Breath alcohol test (to be performed only at screening)
8. Allergenicity test
9. Electrocardiogram (12-lead ECG): In addition to the basic records, record the ventricular rate (beats/min), PR interval (msec), QRS (msec), and QT/QTc (msec) with automatic analysis & recording.

## 10.3.2 Blood and urine collection for pharmacokinetic evaluation and analysis

**1. Samples and substances to be analyzed**

The concentration of N-Rephasin® SAL200 in serum and urine will be analyzed in accordance with the dose group.

**2. Method of blood collection, pre-treatment of isolated serum and storage method**

- Blood collection will be performed according to the clinical study schedule table.
- Blood will be collected via venipuncture or a saline-locked angiocatheter inserted into the brachial vein. In the case of collecting blood using a saline-locked angiocatheter, approximately 1 mL of blood will be drawn and discarded, and approximately 6 mL of blood will be collected. Then, 1 mL of saline will be injected into the catheter to prevent coagulation of the blood.
- The collected blood will be immediately placed in an SST tube, shaken gently, and stored at room temperature. After about 30 minutes, the serum will be separated by centrifugation at a minimum of 2700 rpm for approximately 10 minutes using a centrifuge maintained at 4°C or less.
- The serum separated by centrifugation will be transferred to 4 Eppendorf tubes to have them contain at least 0.5 mL, respectively.
- The samples prepared as above will be transferred into a −70°C freezer and stored until analysis.

**3. Method of urine collection, pre-treatment of urine and storage method**

- The single-dose group will empty their bladder at D1 and the repeat-dose group will empty their bladder at D1 and D4 immediately before the administration, and they will collect urine continuously for 0–4, 4–12, and 12–24 hours after the administration. At the end of the urine collection period, all subjects should empty their bladder for urine collection.
- The urine collected during each urine collection period should be refrigerated immediately until the end of the urine collection period. When the urine collection period ends at 24 hours, each of the collected urine samples will be weighed in an appropriate way, and the results will be recorded in the CRF.
- After weighing, the samples will be shaken properly to make them homogeneous. Then, one 5 mL urine sample will be taken and transferred to 5 Eppendorf tubes to have them contain at least 1 mL, respectively.
- The samples prepared as above will be transferred into a −70ºC freezer immediately and stored until analysis.

**4. Pharmacokinetic analysis method**

See <Appendix 4>.

**5. Pharmacokinetic endpoints**

Using the measured value of the concentration of N-Rephasin® SAL200 in blood, the following pharmacokinetic variables will be calculated for N-Rephasin® SAL200. The actual blood collection time of each subject will be used for the blood collection time during the pharmacokinetic analysis. If the measured concentration is lower than the lower limit of quantification (LLOQ), the actual blood collection has not been performed (not applicable), or the blood sample is missing (missing data), "< LLOQ," "N/A" or "MD" will be entered in the blood drug concentration data. A blood concentration–time pattern of the study drug will be shown as a graph in a linear or log/linear shape for each subject, and the mean blood concentration–time curve by dose group will be shown in the same way. The obtained data will be used to calculate the following pharmacokinetic variables through a noncompartmental method by using Phoenix™ WinNonlin® (Pharsight, CA, USA):

| | |
|---|---|
| $AUC_{last}$ | Area under the blood concentration–time curve to the point of the time of the last blood collection that can be measured |
| $AUC_{inf}$ | Area under the blood concentration–time curve calculated by extrapolating to infinity after a single dose |
| $C_{max}$ | Maximum blood concentration after a single dose |
| $T_{max}$ | Time to reach the maximum blood concentration after a single dose |
| $T_{1/2}$ | Elimination half-life |
| CL/F | Apparent clearance |
| $AUC_{t,ss}$ | Area under the serum drug concentration–time curve within a dosing interval (t) at a steady state |
| $C_{max,ss}$ | Maximum concentration of a drug in serum after the repeated administration at a steady state |
| $C_{min,ss}$ | Concentration immediately before the administration at a steady state |
| $T_{max,ss}$ | Time to reach the maximum blood concentration at a steady state |
| $CL_{ss}$ | Clearance at a steady state |
| R | Cumulative index |
| PTF | peak trough fluctuation |
| $f_e$ | fraction excreted unchanged |
| $Ae_{t,ss}$ | Cumulative amount of a drug excreted in the urine during a dosing interval (t) at a steady state |
| $CL_R$ | Renal clearance |
| $CL_{R,ss}$ | Renal clearance at a steady state, $CL_{R,ss} = Ae_{t,ss} / AUC_{t,ss}$ |

**6. Storage and destruction of samples for pharmacokinetic evaluation**

The blood samples for pharmacokinetic evaluation should be stored and tested according to the relevant SOP. The storage and destruction of the remaining serum samples after the measurement will be carried out in accordance with the items related to the storage of samples in the “SOP for samples” of the Department of Clinical Pharmacology and Therapeutics, Seoul National University Hospital, and the decision of whether to destroy them will be made after a discussion between the sponsor and the principal investigator.

## 10.3.3 Blood collection for pharmacodynamic evaluation and analysis method

**1. Method of blood collection, pre-treatment of isolated serum and storage method**

- Blood collection will be performed according to the clinical study schedule table depending on the relevant study drug.
- Blood will be collected via venipuncture or a saline-locked angiocatheter inserted into the brachial vein.
  In the case of collecting blood using a saline-locked angiocatheter, approximately 1 mL of blood will be drawn and discarded, and approximately 6 mL of blood will be collected. Then, 1 mL of saline will be injected into the catheter

to prevent coagulation of the blood.

- The collected blood will be immediately placed in an SST tube, shaken gently, and stored at room temperature. After about 30 minutes, the serum will be separated by centrifugation at a minimum of 2700 rpm for approximately 10 minutes using a centrifuge maintained at approximately 4°C or less.
- The serum separated by centrifugation will be transferred to 4 Eppendorf tubes to have them contain the same amount in each tube.
  The samples prepared as above will be transferred into a −70ºC freezer and stored until analysis.

**2. Pharmacodynamic analysis method**
See <Appendix 4>.

## 10.3.4 Blood collection for immunogenicity evaluation and analysis method

**1. Method of blood collection, pre-treatment of isolated serum and storage method**

- Blood collection will be performed according to the clinical study schedule table depending on the relevant study drug.
- Blood will be collected via venipuncture or a saline-locked angiocatheter inserted into the brachial vein. In the case of collecting blood using a saline-locked angiocatheter, approximately 1 mL of blood will be drawn and discarded, and approximately 6 mL of blood will be collected. Then, 1 mL of saline will be injected into the catheter to prevent coagulation of the blood.
- The collected blood will be immediately placed in an SST tube, shaken gently, and stored at room temperature. After about 30 minutes, the serum will be separated by centrifugation at a minimum of 2700 rpm for approximately 10 minutes using a centrifuge maintained at approximately 4°C or less.
- The serum separated by centrifugation will be transferred to 4 Eppendorf tubes to have them contain at least 0.5 mL, respectively.

The samples prepared as above will be transferred into a −70ºC freezer and stored until analysis.

**2. Immunogenicity analysis method**
See <Appendix 4>.

## 10.3.5 Safety endpoints

1) Adverse events
2) Vital signs, electrocardiogram (12-lead ECG), clinical laboratory tests, allergenicity test, anaphylatoxin, etc.

The patterns of occurrence of adverse events and the results of tests such as vital signs, electrocardiogram (12-lead ECG), clinical laboratory tests, allergenicity test, and anaphylatoxin may be reviewed comprehensively in subjects who received administration of the investigational product at least once.
After administration of the investigational product, the occurrence, severity, and frequency of adverse drug reactions that are already known will be evaluated. Besides, if unexpected adverse events, especially serious adverse drug reactions occur, their causality relationship, frequency, severity, etc. will be evaluated. The descriptive statistics will be provided for test items judged to be clinically significant by the investigator for each treatment group as needed, and they can be compared by applying a nonparametric method as needed.

## 10.4 Adverse event reporting, etc.

The safety evaluation items will be evaluated according to the schedule in the protocol and “10.3.5 Safety endpoints.”
The investigator will record all outliers, which are considered clinically significant and meet the criteria in “10.4.1 Evaluation criteria,” as adverse events or serious adverse events.

## 10.4.1 Evaluation criteria

### 10.4.1.1 Adverse event (AE)

An adverse event refers to any harmful and unintended sign (including an abnormal laboratory test result), symptom,

or disease that occurred in a subject who was administered an investigational product, and it does not necessarily have to have a causality relationship to the investigational product.

### 10.4.1.2 Adverse drug reaction (ADR)

An adverse drug reaction 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.

### 10.4.1.3 Serious adverse event/adverse drug reaction (Serious AE/ADR)

It 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:

1) In the case that results in death or is life-threatening
2) In the case that requires hospitalization or prolongation of hospitalization
3) In the case that results in permanent or serious disability and dysfunction
4) In the case that results in congenital anomaly or birth defect
5) Besides the cases in 1) through 4) above, occurrence of other medically significant incidents such as drug dependence, drug abuse, or blood dyscrasia

However, hospitalization for the following is not considered as a serious adverse event:

- Hospitalization or prolongation of hospitalization for diagnosis or elective surgery for an existing disease
- Hospitalization or prolongation of hospitalization required to measure the effectiveness of the clinical study
- Hospitalization or prolongation of hospitalization for scheduled treatment for the indicated disease in the clinical study
- In the case of visiting an emergency department, the duration of the hospital visit is less than 24 hours

### 10.4.1.4 Unexpected adverse drug reaction (Unexpected ADR)

It 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.

► To determine whether it is an unexpected adverse drug reaction

Only for adverse events that are considered to be related to the study drug, the investigator and iNtRON Biotechnology, Inc. will determine whether adverse events have been expected or unexpected.

- Expected: For investigational products, an adverse event that matches the nature, severity, frequency and characteristics of the adverse event described in the current investigator's brochure or protocol shall be defined as "Expected."
- Unexpected: Adverse events that are not within the range of "Expected"

### 10.4.1.5 Severity of adverse events

The severity of adverse events will be classified based on the following criteria in consideration of the maximal intensity.

**<Severity of adverse events>**

| Grade | | Description |
|---|---|---|
| 1 | Mild | The degree that the subject is rarely aware of the event and the event does not affect (functional) activities of daily living. Most cases do not require treatment. |
| 2 | Moderate | The degree that the subject may feel discomfort and the event affects (functional) activities of daily living. The degree that the subject may continue to participate in the study, but this severity may require treatment. |

| | | |
|---|---|---|
| 3 | Severe | The degree that subject feels very uncomfortable, (functional) activities of daily living are impossible, and the subject cannot continue to participate in the study. The degree that requires treatment or hospitalization. |

### 10.4.1.6 Causality relationship of adverse events

For the causality relationship of adverse events, a medical decision should be made to determine the relationship taking into account all relevant factors, such as the type of adverse event, relation in respect to time, discontinuation or rechallenge of treatment, and concomitant medications and disturbance factors including medical history related to intercurrent diseases.

The result of a causality relationship determined and all information related to adverse events should be recorded in the case report form.

**<Causality relationship of adverse events>**

| Causality relationship | | Rationale for the decision |
|---|---|---|
| 1 | Certain | • When the sequential relation between the drug administration and the onset of an adverse event is reasonable<br>• When an adverse event cannot be explained by other drugs, chemical substances, or accompanying diseases<br>• When a clinically reasonable reaction is observed after discontinuation of the drug<br>• When rechallenged (to be done only if possible), the result is decisive pharmacologically or phenomenologically |
| 2 | Probable/likely | • When the relation in respect to time between the drug administration and the onset of an adverse event is proper<br>• When an adverse event does not appear to have been caused by other drugs, chemical substances, or accompanying diseases<br>• When a clinically proper reaction is observed after discontinuation of the drug<br>• When there is no information about rechallenge |
| 3 | Possible | • When the relation in respect to time between the drug administration and the onset of an adverse event is proper<br>• When an adverse event can be explained to have been caused by other drugs, chemical substances, or accompanying diseases<br>• When the information on discontinuation of the administration is insufficient or unclear |
| 4 | Unlikely | • When it is a temporary case which does not seem to be a causality relationship between the drug administration and the onset of an adverse event<br>• When it can also be a reasonable explanation that an adverse event has been caused by other drugs, chemical substances, or latent diseases |
| 5 | Conditional/unclassified | • When it requires more data for appropriate evaluation or its additional data are being reviewed |
| 6 | Unassessable/unclassifiable | • When it cannot be judged because the information is insufficient or contradictory, and the information cannot be supplemented or verified |

If a serious adverse event occurs while it is still blinded, the investigator should determine the causality relationship to all potential investigational product.

### 10.4.1.7 Action against adverse events

The action against adverse events will be classified as shown below and recorded in the case report form.

| | |
|---|---|
| 1. | No action taken |
| 2. | Study drug temporarily interrupted<br>(Temporary discontinuation of administration of the investigational product) |
| 3. | Study drug permanently discontinued due to this adverse event<br>(Discontinuation of administration of the investigational product) |
| 4. | Concomitant medication taken<br>(Concomitant administration of a therapeutic drug) |
| 5. | Non-drug therapy given |
| 6. | Hospitalization/prolonged hospitalization |

### 10.4.1.8 Exacerbation of underlying diseases or health condition that already exists

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 additional treatment is needed; in other words, when a concomitant medication is added or changed
- When the investigator determines that the disease exacerbated from baseline unexpectedly

### 10.4.2 Reporting of adverse events

The investigator should collect adverse events that occurred during the clinical study period (from the time of signing the informed consent form to the observation period) and report to the sponsor using CRF records, SAE forms, etc.

For each adverse event, the investigator will determine its relationship to the investigational product, record the date of onset, end date, grade, need for treatment, outcome, seriousness, etc., and report it within the relevant regulations and requirements/deadlines of the IRB.

- 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 | +82-70-4335-5469 |
| Fax number | +82-2-749-2050<br>+82-2-6442-4753 (eFax) |

In addition, the investigator should report unexpected serious adverse reactions to the IRB and the sponsor, and the sponsor should report all serious and unexpected adverse events to the investigator, IRB (applicable if the principal investigator has not reported the case to the IRB or the reported information needs to be changed), and Minister of Food and Drug Safety by the deadlines according to the following categories:

- In the case of death or life-threatening cases: Within 7 days from the day when the sponsor received a report or recognized the relevant fact. In this case, the sponsor should report further details of adverse drug reactions within 8 days from the date of initial report.
- For all other unexpected serious adverse reactions: Within 15 days from the day when the sponsor received a report or recognized

If there is additional information about the adverse drug reaction reported according to the process above, the investigator should report it until the relevant adverse drug reaction ends (meaning that the relevant adverse drug reaction disappeared or is lost of a follow-up). In addition, in the case of reporting a death case, the principal investigator should submit additional information, such as an autopsy note (only if autopsy was performed) and a death certificate, to the sponsor and the IRB.

# 11. Data analysis and statistical considerations

## 11.1 Analysis set

All subjects who were randomized, who received the drug administration, and who has evaluable pharmacokinetic results will be classified and analyzed. Demographic information will be analyzed for all subjects who have been randomized (intention-to-treat), and the safety assessment will be performed for subjects who have received the investigational product at least once. The pharmacokinetics, pharmacodynamics, and immunogenicity will be evaluated in those who completed all study procedures. However, if a subject drops out after receiving the drug administration, the test results of the relevant subject may be reviewed on the items that can be evaluated in the end up until the time of discontinuation/dropout.

## 11.2 Acceptance criteria

The results of the safety and pharmacokinetic assessments will be statistically analyzed with a level of significance of 0.05 using SAS® (SAS Institute, Cary, NC, USA) or SPSS®.

## 11.3 Statistical analysis methods

### 11.3.1 General principles of result analysis

The number of subjects to be observed, mean, and standard deviation will be provided for continuous variables, and the frequency and percentage will be obtained for categorical variables. All p-values will be presented to have 4 decimal places, and those numbers with digits to the right of the decimal point, such as mean and standard deviation, will be presented to have 2 decimal places.

### 11.3.2 Method to handle missing data

In the case of missing data of subjects who participated in the clinical study, replacement of excluded data will not be applied. Values below the lower limit of quantification will be treated as 0.

### 11.3.3 Analysis of demographic baseline data

Descriptive statistical analysis, including the mean and standard deviation, will be performed on demographic information such as the age, height, weight, and BMI. If needed, statistical analysis for each dose group can be performed in order to examine validity of the randomization and the subsequent analysis.

### 11.3.4 Analysis of pharmacokinetic endpoints

For pharmacokinetic parameters, the values will be obtained for each subject, and their mean, standard deviation, etc. will be provided for each dose group in a descriptive statistical manner. The pharmacokinetic linearity evaluation will be analyzed through the comparison between the dose groups of $C_{max,ss}$/dose, $AUC_{\tau}$/dose and the regression method for the doses of $AUC_{\tau}$, $C_{max,ss}$. Besides, $CL_{ss}$, $t_{1/2}$, and $f_e$ will be evaluated by using a nonparametric method comparing the difference among the dose groups.

### 11.3.5 Analysis of pharmacodynamic endpoints

For pharmacodynamic evaluation, the antimicrobial activity in serum samples will be analyzed by comparing the clear zone formed after incubating the calibration samples prepared with N-Rephasin® SAL200 at concentrations of 0-1.0 μg/mL and the serum samples obtained from the subjects dripped on a medium on which methicillin-resistant *Staphylococcus aureus* (isolated from dairy cows with mastitis, provided by iNtRON Biotechnology, Inc.) is smeared. Their mean, standard deviation, etc. will be provided for each dose group in a descriptive statistical manner.

### 11.3.6 Analysis of immunogenicity endpoints

For immunogenicity evaluation, the antibody titer of each subject will be obtained, and then the effects of the dose and systemic exposure of N-Rephasin® SAL200 on these will be analyzed. For their mean, standard deviation, etc., the difference of the time of measurement for each dose group will be provided using descriptive statistics such as difference of the relevant time point from the baseline value in order to validate the statistical significance of the dose and systemic exposure in relation to the antibody titer.

### 11.3.7 Analysis of safety endpoints

The patterns of occurrence of adverse events and the results of tests such as vital signs, electrocardiogram (12-lead ECG), clinical laboratory tests, and allergenicity test may be reviewed comprehensively in all subjects who received administration of the investigational product at least once. After administration of the investigational product, the occurrence, severity, and frequency of adverse drug reactions that are already known will be evaluated. Besides, if unexpected adverse events, especially serious adverse drug reactions occur, their causality relationship, frequency, severity, etc. will be evaluated. The descriptive statistics will be provided for test items judged to be clinically significant by the investigator for each treatment group as needed, and they can be compared by applying a nonparametric method as needed.

### 11.4 Time of analysis

This clinical study will be statistically analyzed after all scheduled subjects have had the clinical study period (the last visit of the last subject), confirmation of all data is completed, and the data are locked.

### 11.5 Rationale for the number of subjects

This clinical study is a typical Phase 1 clinical study to evaluate the safety and explore the characteristics of pharmacokinetics, pharmacodynamics and immunogenicity after repeated continuous intravenous infusion of N-Rephasin® SAL200 in healthy volunteers. Since the objective of this clinical study has explorative and descriptive nature, it is quite different from a typical study for a statistical hypothesis test in its nature. Therefore, it is advisable to conduct the study with the least number of subjects required empirically rather than establishing based on a certain formula.

By setting 8 subjects (6 subjects to receive the study drug, 2 subjects to receive the placebo) for each dose level, a total of 40 subjects will be required.

## 12. Data management

### 12.1 Record, collection, and access

The investigator will collect data of each subject who participated in this study by completing the case report form (CRF), which is designed to record all data needed to conduct the clinical study including observation results, properly and accurately based on source documents, and only the investigator, CRC, etc. delegated as staff members of this study can complete data. The investigator should ensure that the data included in the case report form or any other report are accurate, complete, legible, and timely.
In this clinical study, data will be collected using the web-CRF, and the CRO (GDFI Braincell Laboratory) will set the extent of authorized use (write, edit, read, etc.) according to the degree of authorization assigned based on the investigator's (investigator, CRA, CRC, etc.) scope of work and assign an ID to have access to this web-CRF.

### 12.1.1 Case report forms

Source documents include all documents (including electronic documents), data, and records that contain source documents such as hospital records, medical records, subject's records, memos, pathological test results, subject's diaries, assessment checklists, drug dispensing records from the pharmacy, 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.

### 12.1.2 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 IRB as well as the KGCP

Therefore, the clinical research associate will regularly visit the study site during the study 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 also acts as the key communicator between the sponsor and the principal investigator.
The clinical research associate should visit the study site or contact the study staff member via telephone, fax or email and report the relevant information and details 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, action taken or to be taken to maintain compliance with the protocol

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. If the CRF has been completed differently from the source document, a query should be issued to the investigator requesting confirmation. If it is found that the relevant information requires a correction, the investigator should rewrite the information along with the reason for correction.

### 12.1.3 Data safety monitoring plan

The principal investigator should manage the documents related to this clinical study to prevent disclosure in order to protect the information of the subjects. To guarantee the completeness of the study data, the principal investigator should periodically check whether the storage location of source documents has been changed and whether the documents have been accessed by others who are not involved. In addition, the safety of the subjects should be protected by reviewing whether the clinical study is being conducted in accordance with the study objective in the protocol (appropriateness of subject recruitment, safety and efficacy assessments in compliance with the protocol, and whether adverse events are reported/recorded properly when they occur), and the completeness of the data should be guaranteed by completing case report forms based on source documents.
Moreover, the clinical research associate should report data related to the safety of the subjects that has been identified through this monitoring process to the investigator who can make medical judgment, and the investigator will finally determine the following recommendations resulted 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

## 12.2 Protection and storage of clinical study-related data

The principal investigator will store 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 1 Subparagraph 12 of the Regulation on Safety of Pharmaceuticals, etc., the investigator should store the protocol and various data related to the conduct of the clinical study (including electronic documents) for 3 years from the date of the MFDS approval.
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 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.

# 13. Ethical considerations and administrative process

## 13.1 Good Clinical Practice and regulations on consenting process, etc.

The procedures prescribed in this protocol have been designed to comply with the Korea Good Clinical Practice (KGCP) and the fundamental philosophy of the Declaration of Helsinki when the sponsor and the investigator are conducting, evaluating, and recording the results of this study. Therefore, the investigator and the sponsor should conduct the clinical study in compliance with the protocol, which has been agreed in writing and approved by the IRB and the Minister of Food and Drug Safety. The principal investigator should not conduct the clinical study differently from the protocol until receiving a prior agreement with the sponsor and approval for amendments by the IRB and the Minister of Food and Drug Safety, and if there is anything that has been performed differently from the approved protocol, the principal investigator or the sub-investigator should record the relevant information and its reason.

In addition, prior to initiating the clinical study, the informed consent form, subject information sheet, and other documented information to be provided to subjects should be approved by the IRB, and it should be carried out in accordance with the consenting process shown below.

### 13.1.1 Consenting process

The investigator delegated by the principal investigator or the sub-investigator should inform the subject of the informed consent form and subject information sheet approved by the IRB and other information about all aspects of the clinical study prior to enrolling the patient in the clinical study, and should obtain a written consent from the subject. If a subject cannot provide consent, this should be informed to a subject's authorized representative (a parent, spouse, or guardian of the subject who can make decisions on the subject's participation in the clinical study on behalf of the subject).

The principal investigator or the sub-investigator should not coerce or have undue influence in the participation of subjects at any time, and the informed consent form used and the information related to the clinical study (including verbal or written information) should not restrict the rights of the subject or the subject's authorized representative or contain any information that implies such a restriction, and should not exempt the investigator, study site, sponsor or sponsor's representative from responsibilities or contain any information that implies such exemption.
In addition, terminologies easy for the subject, the subject's authorized representative, or impartial witness to understand should be used.
Before obtaining consent from the subject, the principal investigator or a physician designated by the principal investigator should provide the subject or the subject's authorized representative ample time and opportunity to inquire about details of the clinical study and to decide whether or not to participate in the relevant clinical study. All questions about the clinical study should be answered to the satisfaction of the subject or the subject's authorized representative.

If the subject or the subject's authorized representative cannot read the informed consent form, subject information sheet, or other documented information, the impartial witness should attend the entire process of obtaining the consent. In such a case, the principal investigator or a staff member delegated by the principal investigator should read and explain the informed consent form, subject information sheet, or other documented information to the subject or the subject's authorized representative. The subject or the subject's authorized representative should verbally consent to the participation in the clinical study, and if possible, sign and date the informed consent form in person. The impartial witness should sign and date the informed consent form in person, and confirm the following before signing the informed consent form:

- Whether the informed consent form, subject information sheet, and other documented information have been accurately explained to the subject or the subject's authorized representative
- Whether the subject or the subject's authorized representative has understood the relevant information
- Whether the process of obtaining the consent was carried out based on the free will of the subject or the subject's authorized representative

Prior to participation in the clinical study, the subject or the subject's authorized representative and the principal investigator or the sub-investigator delegated by the principal investigator should sign and date the informed consent form in person. A copy of the informed consent form and other documented information provided to the subject should be given to the subject or the subject's authorized representative prior to participation in the clinical study, and in the case that the informed consent form changes during the clinical study, a copy of the changed informed consent form should be given to the subject or the subject's authorized representative. Also, in the case that new information related to the clinical study obtained may affect the subject's consent, the informed consent form, subject information sheet, and other documented information should be revised accordingly and approved by the IRB. Then, the principal investigator should inform the subject or the subject's authorized representative in a timely manner and record the subject notified, the date and time of notification, and the information notified.

## 13.2 Ethical compliances

In order to protect the rights of the subject and ensure that the clinical study is conducted scientifically and ethically, this clinical study will be conducted to comply with the Korea Good Clinical Practice (KGCP) and regulations related to clinical studies, to respect the dignity of human being and interests in accordance with the Declaration of Helsinki, and to prevent disadvantages to the subject as well.

## 13.3 Measures for protection of the subject safety

For this clinical study, the Institutional Review Board (IRB) will evaluate/approve this protocol according to the Korea Good Clinical Practice (KGCP) and will evaluate regularly to check whether this clinical study is being conducted in accordance with the protocol.

The sub-investigators and participating investigators will thoroughly analyze and fully understand the study plan, and the principal investigator will take precautionary action for the onset of unexpected side effects, etc., such as taking sufficient action, reporting as necessary, and providing sufficient training for the investigators. The clinical study will be conducted in accordance with the KGCP standards.

In the case that the subject experiences adverse events due to the clinical study despite the investigator conducting the clinical study in compliance with all applicable laws and regulations and strictly in accordance with the protocol and various related literature, recommendations, and suggestions provided by iNtRON Biotechnology, Inc., the subject will receive appropriate medical treatment until recovery, and iNtRON Biotechnology, Inc. will indemnify against the damage caused by the investigational product in accordance with the “indemnification policy for victims.”

In addition, when enrolling vulnerable subjects, the information below and the regulations of each study site should be followed.

## 13.4 Information about the examination of 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 subject who participate during the clinical study participation period.

The examination and treatment of the subject whose clinical study participation has been discontinued or terminated will be based on the general examination and treatment principles of the study site. In other words, different types of treatments, procedures, and drug therapies will be performed, depending on the patient’s condition and the physician’s decision.

## 13.5 Publication of results

After analyzing the clinical study data, the sponsor will complete a clinical study report that includes the data of the subjects who participated in the clinical study, the statistical analysis results, etc. Then, if required by applicable regulations, the sponsor will confirm the principal investigator’s signature for approval of the clinical study report, and will provide the principal investigator with reasonable access to statistical tables, figures, and related reports. The principal investigator may share the result with the subject appropriately upon completion of the clinical study report.

By signing this protocol, the principal investigator and the investigator agree to use the results of this study for purposes of registration, presentation, and provision of information for medical and pharmaceutical experts.

The result of this clinical study can be submitted to the Ministry of Food and Drug Safety or presented in academic journals, academic conferences, etc. to obtain the pharmaceutical approval, and the sponsor has the right to review the information to be presented prior to presentation or submission of the clinical study result.

## 13.6 Confidentiality of patient records

For protection of personal information of the subject as well as the data, the subject will be managed using the subject identification code. Through this, the personally identifiable information will not be disclosed throughout the clinical study participation and will not be included in the study result in the future (such as publishing the study result and disclosing the study result). In addition, the principal investigator should be responsible for careful supervision to ensure that personal information collected before coding is not disclosed and should prevent disclosure of information on identity of the subject to others except for those who are allowed to have direct access to the data.

## 13.7 Quality control and reliability assurance

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 compliance with the protocol approved by the Ministry of Food and Drug Safety and the IRB of the study site.

The clinical research associate of this clinical study will train the investigators to analyze and fully understand the study plan. If a deviation of the protocol is found at the time of monitoring despite prior measures, such as sufficient training, have been taken, the clinical research associate will document the relevant information and train the investigators again to ensure that the clinical study is conducted in accordance with the KGCP standards.

In addition, the investigator's access to the web-CRF will be restricted depending on the tasks delegated in order to ensure the reliability and accuracy, and the person registered as the clinical research associate of the clinical study will visit the study site and perform monitoring regularly in order to check whether the investigators are complying with the most recent protocol, whether the CRFs have been completed accurately based on the source documents, whether the issued queries have been solved and corrections have been made, the investigational product management status, data storage (study files), serious adverse event reporting process, etc. As a result, quality control of the clinical study data will be carried out at all stages of data processing. In order to guarantee the reliability of the laboratory test results, it will be confirmed through the relevant certificate of the laboratory at the site whether the items (areas) to be performed in this study are included, and a copy will be stored.

# 14. Information on those who would like to conduct the clinical study, and name and title of the principal investigator

## 14.1 Sponsor and contact information

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

## 14.2 Contract research organization (CRO)

| | |
|---|---|
| Company name | Symyoo Inc. |
| Address | 2nd floor, 6, Hannam-daero 42-gil, Yongsan-gu, Seoul, 04417, Republic of Korea |
| Telephone number | +82-70-4335-5469 |
| Fax number | +82-2-749-2050<br>+82-2-6442-4753 (eFax) |

## 14.3 Name and title of the principal investigator

| Name of the study site | Classification | Name | Title |
|---|---|---|---|
| Seoul National University Hospital | Principal investigator | In Jin Jang | Professor, Department of Clinical Pharmacology and Therapeutics |

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

## 15.1 Approval of the Institutional Review Board (IRB)

This clinical study should be conducted ethically and scientifically and should be approved by the Institutional Review Board prior to initiation of the clinical study in accordance with the KGCP.

## 15.2 Protocol amendment and deviation

### 15.2.1 Protocol amendment

In the case that the study procedures becomes more extensive, the risk increases, there are changes in the inclusion criteria, or the protocol is amended due to additional safety information after obtaining approval for the protocol from

the Minister of Food and Drug Safety and the Institutional Review Board, the protocol should be approved again by the Minister of Food and Drug Safety and the Institutional Review Board. When revisions are made to the protocol, the date of revisions, reasons for revisions, and details of revisions should be recorded and stored.

### 15.2.2 Protocol deviation

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

## 16. Literature references

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4. Etiene J, Brun Y, Fleurette J. *Staphylococcus lugdunensis* endocarditis. J Clin Pathol. 1989;42:892-893
5. Frency J, Brun Y, Bes M, Meugnier H, Grimont F, Grimon PAD, Newi C, Fleurette J. *Staphylococcus lugdunensis* sp. and *Staphylococcus schleiferi* sp. novel two species from human clinical specimens. Int Syst Bacteriol. 1998;38:168-172
6. Seung-Ho Han et al., Monitoring of Methicillin-Resistant *Staphylococcus aureus* in Nasal Swabs Obtained fron Dental Clinic Healthcare Providers and Medical Environments Nurses. Int J Oral Biology. 2010;35:7-12
7. Jane D. Siegel, MD et al., the Healthcare Infection Control Practices Advisory Committee, Management of Multidrug-Resistant Organisms In Healthcare Settings, 2006
8. Lee HM, Yong DG, Lee KW et al. Antimicrobial Resistance of Clinically Important Bacteria Isolated from 12 Hospitals in Korea in 2004. Korean Journal of Clinical Microbiology 2005; 8 (1): 66–73
9. Eun Ja Park et al., Analysis of Economic Outcome of Methicillin-Resistant *Staphylococcus aureus*(MRSA) Bacteremia Using Retrospective Case-Control Study, Kor. J. Clin. Pharm 2007;17:59-64
10. Van Hal et al., Predictors of Mortality in *Staphylococcus aureus* Bacteremia, Clinical Microbiology Reviews 2012; 25(2):362-386

# [Attachment 1] Signature Section

## 1.1 Signature Section - Sponsor

## Signature Sheet

**Clinical study title: A Randomized, Double-Blind, Placebo-Controlled, Multiple-Dosing, Dose-Escalating Phase 1b Study to Evaluate the Safety, Pharmacokinetics, Pharmacodynamics and Immunogenicity of N-Rephasin® SAL200 after Continuous Intravenous Infusion in Healthy Volunteers**

**Protocol No. : ITB-101_1b**

**Version No. / Version date : 2.0 / May 17, 2019**

We have reviewed and hereby approve the information specified in this protocol, and we agree to conduct the clinical study as specified in the protocol.

We hereby confirm that we will conduct this clinical study in accordance with all relevant regulations applicable to the Korea Good Clinical Practice (KGCP, ICH GCP), and we will fulfill the responsibilities as a sponsor in accordance with the fundamental philosophy of the Declaration of Helsinki and in accordance with ethical and scientific principles.

**Sponsor**

| *Title / Name* | *Date* | *Signature* |
|---|---|---|

## 1.2 Signature Section - Principal Investigator

# Signature Sheet

**Clinical study title: A Randomized, Double-Blind, Placebo-Controlled, Multiple-Dosing, Dose-Escalating Phase 1b Study to Evaluate the Safety, Pharmacokinetics, Pharmacodynamics and Immunogenicity of N-Rephasin® SAL200 after Continuous Intravenous Infusion in Healthy Volunteers**

**Protocol No. : ITB-101_1b**
**Version No. / Version date : 2.0 / May 17, 2019**

I have read and understood all the information in this protocol and agree to conduct the clinical study as specified in the protocol.

In addition, I hereby confirm that I will conduct this clinical study in accordance with all relevant regulations applicable to the Korea Good Clinical Practice (KGCP, ICH GCP), and I will conduct the study in compliance with the major responsibilities as an investigator in accordance with the fundamental philosophy of the Declaration of Helsinki and in accordance with ethical and scientific principles.

**Study site: Seoul National University Hospital**

**Principal investigator**

| *Title / Name* | *Date* | *Signature* |
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# [Attachment 2] Expected Adverse Events and Precautions for Use

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 conducted in rodents and beagle dogs and the single dose study conducted in human subjects, the adverse events shown below have been reported.

≫ Rodents

a. Single dose

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

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

≫ 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

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

≫ Human subjects (single dose)

A total of 28 adverse events occurred in all 6 subjects who received administration of the study drug in the dose group in which N-Rephasin® SAL200 was administered at 10 mg/kg, and 25 cases of these adverse events were determined to be related to N-Rephasin® SAL200. Among the adverse events reported in the relevant dose group, rigors was the highest in frequency (16%), and fatigue, headache, and myalgia were reported in many cases. The intensity of rigors occurred after the administration in 2 subjects (subject numbers: R502, R503) was enough for the subjects to feel discomfort and to interfere with other activities of daily living (using computers, reading books, etc.), and the severity was evaluated as moderate. Other adverse events were evaluated as mild. The active ingredient of N-Rephasin® SAL200 is a lysin protein, and in general, intravenous administration of protein formulations can cause adverse events such as headache, rigors, fever, and myalgia. No clinically significant findings were observed in clinical laboratory tests or electrocardiogram in relation to adverse events, and based on the time of onset of adverse events, they are considered to be due to the protein substance of N-Rephasin® SAL200.