[redacted]

STATISTICAL ANALYSIS PLAN

INTERIM ANALYSIS

A Phase II, Randomized, Double-blind, Active Comparator, Parallel Group Study to Evaluate the Efficacy and Safety of K-285 Compared with Menthol Gel for the Treatment of Delayed Onset Muscle Soreness (DOMS) in the Lower Extremity

PROTOCOL NUMBER: K-285-201

Protocol Version 2.0 (28MAY2020)

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LIST OF ABBREVIATIONS

AE	Adverse event
API	active pharmaceutical ingredients
ANCOVA	analysis of covariance
AST	aspartate aminotransferase
BMI	body mass index
CFR	Code of Federal Regulations
CRF	case report/record form
CRO	Contract Research Organization
DOMS	delayed onset muscle soreness
ECG	electrocardiogram
eCRF	electronic Case Report/Record Form
e-diary	electronic diary device
eVAS	electronic visual analog scale
ET	early termination
FAS	full analysis set
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GI	gastrointestinal
ICF	informed consent form
ICH	International Council for Harmonisation
IEC	Independent Ethics Committee
IRB	Institutional Review Board
LOCF	last observation carried forward
LOE	lack of efficacy
OTC	Over the counter
PPS	per protocol set

1 PURPOSE OF THE ANALYSES

This statistical analysis plan (SAP) is based on protocol number K-285-201 from Kowa Research Institute Inc. This SAP will focus on analyses that will be performed for the unblinded interim analysis after a total of 120 subjects are enrolled and have completed the 24-hour assessment or are discontinued from the study. A final SAP will be created and signed off before the final database lock that will describe the full description of the analyses that will be performed for the final clinical study report (CSR).

The SAP contains detailed information to aid in the performance of the interim statistical analysis and is not a complete description of all analyses that will be performed at the end of the study. This interim SAP is being written with due consideration of the recommendations outlined in the most recent International Council on Harmonization (ICH) E9 Guideline entitled Guidance for Industry: Statistical Principles for Clinical Trials, the ICH E9 (R1) Addendum, the most recent ICH E3 Guideline and the Guidance for Industry: Structure and Content of Clinical Study, and the most recent FDA draft Guidance for Industry – Analgesic Indications: Developing Drug and Biological Products, dated February 2014.

This SAP describes the data that will be analyzed and the futility assessments that will be evaluated for the interim analysis. This SAP provides details of the specific statistical methods that will be used for that analysis.

2 PROTOCOL SUMMARY

2.1 Study Objectives

2.1.1 Primary Objective

To evaluate the efficacy of K-285 compared with menthol gel between 0 to 24 hours after the first gel application for the treatment of delayed onset muscle soreness (DOMS).

2.1.2 Secondary Objectives

Secondary objectives of this study are to determine:

- To evaluate the efficacy of K-285 compared with menthol gel after the first gel application for the treatment of DOMS
- To evaluate the safety and tolerability of K-285 compared with menthol gel after the first gel application for the treatment of DOMS

2.2 Overall Study Design and Plan

2.2.1 Study Design

This is a phase II, randomized, double-blind, active comparator, parallel group study to evaluate the efficacy and safety of K-285 compared with menthol gel for the treatment of DOMS in the lower extremity. The study will consist of screening, an exercise visit (DOMS induction), in-patient and out-patient periods up to 3 weeks, and a follow-up telephone call.

During the Screening Visit, subjects will provide informed consent and complete screening assessments including medical history, identification of the dominant leg, physical and skin examinations, blood and urine tests, vital signs measurements, electrocardiogram (ECG), and other screening procedures. Subjects who are able to participate in the study (Period 1 and 2) and who meet the pre-exercise inclusion and exclusion criteria will be enrolled into the study and return to the clinic on Day -1.

Eligibility will be confirmed, and enrolled subjects will perform a standardized repetitive quadriceps muscle exercise to create DOMS to both legs (Day -1). The exercise regimen will consist of a warm-up, determination of maximum tolerated weight, and timed and untimed workout phases. Within 24 to 30 hours after the exercise session (Day 1), subjects will rate the pain of both legs using a categorical pain scale and record pain intensity of both legs at rest and while standing using a 0-100 point electronic visual analog scale (eVAS) (a capture program will linearly convert the pixel touched to an integer between 0 and 100) to determine eligibility for randomization. The specific post-exercise [redacted] inclusion criterion will be masked to subjects. [redacted] If both legs meet post-exercise inclusion criteria, the study leg will be the dominant leg, based on hand dominance. Subjects not qualifying for randomization will be given appropriate standard of care.

A total of 240 subjects who meet post-exercise inclusion criteria will be randomly assigned in a 1:1 ratio to Treatment Arm A (K-285 gel) or Treatment Arm B (Menthol gel). [redacted]

[redacted]

In period 1 ([redacted] study gels will be applied to randomized subjects on a [redacted] on the study leg as follows:

- Treatment Arm A: Each subject will apply K-285 to the study leg
- Treatment Arm B: Each subject will apply menthol gel to the study leg

The first study gel application to the study leg will be applied by the subject with investigator (or designee) oversight to ensure proper application at the clinic on Day 1 within 24 to 30 hours after DOMS induction. Subsequent study gel applications at the clinic will be applied to the study leg by the subject with investigator (or designee) oversight to ensure proper application. On Day 2, photographic and residue evaluation of the application site will be performed prior to the [redacted] study gel application. Subjects will remain in the clinic until they are discharged following completion of study assessments on Day 2.

Pain intensity assessments will be performed for the study leg and non-study leg at rest and while standing using a 0-100 point eVAS on Day 1 at Baseline (0 hours) and at [redacted] after the first study gel application. Pain assessments scheduled between midnight and 6 am may be skipped if subjects are asleep but subjects may not skip more than two sequential assessments. A time window of +30 minutes will be allowed between qualifying pain assessments and first study gel application (0 hours).The following time windows will be allowed for each pain assessment (eVAS, Perceptible Pain Relief, and Meaningful Pain Relief) time point: [redacted]

Following discharge from the clinic on Day 2, subsequent study gel applications and pain intensity assessments will be performed by the subject on an out-patient basis (Day 2 through Day 8). Subjects will be instructed not to shower or bathe for at least 30 minutes before and at least 1 hour after gel application.

All pain intensity assessments including eVAS, Perceptible Pain Relief, and Meaningful pain relief will be recorded in e-diaries through Day 8 (Period 1), and all study gel applications and rescue medications will be recorded in the e-diary by the subject through Day 22 (Period 1 and 2). Perceptible Pain Relief and Meaningful Pain Relief will be recorded in the e-diary until achieved in Period 1. Due to unexpected problems that occurred after 20 subjects randomized, the e-diary vendor was switched from ERT to [redacted]. Newly enrolled subjects will utilize the [redacted] e-diary system. A telephone call will be conducted by study staff on Day 5 (±1 day) to assess safety. Subjects will return to the clinic for assessments on Day 8 (±1 day). For subjects who are assigned to receive study gel for 1 week, Day 8 (+1 day) will be an end of study (EOS) clinic visit and a follow-up telephone call to assess safety will be conducted by study staff on Day 15 (±1 day). Subjects who are assigned to receive study gel for 3 weeks will continue to apply study gel on an out-patient basis from Day 8 to Day 22 (Period 2) according to their assigned dosing regimen ([redacted]) (Table 1: Treatment Assignment). For subjects who are dosing in Period 2, telephone calls to assess safety will be conducted by study staff on Day 11 (±1 day) and Day 18 (±1 day), study gel applications and use of any rescue medications will be recorded in e-diaries by the subjects through Day 22, and an end of study (EOS) clinic visit will occur on Day 22 (+1 day). A follow-up telephone call to assess safety will be conducted by study staff on Day 29 (±1 day).

[redacted]

2.2.2 Study Population

The study population will be adult male and female subjects aged 18 to 35 years, inclusive, with a body mass index (BMI) between 18 and 32 kg/m2, inclusive.

2.2.3 Treatment Regimens

A description of each regimen is provided below:

- K-285:
- Menthol gel:

2.2.4 Treatment Group Assignments or Randomization

The study will be double-blind. The investigator, subjects, and sponsor personnel involved in the monitoring or conduct of the study will be blinded to the identity of the treatment from the time of randomization until the time of unblinding.

Subjects will be assigned to study treatment via appropriate randomization methods, in accordance with the randomization schedule generated by the allocation vendor), prior to the start of the study, using validated internal software. Once a

randomization number has been assigned to a subject, it must not be re-assigned to a different subject.

[redacted]) will prepare the randomization schedule before the study. Enrolled, eligible subjects will receive a kit containing study gels according to an allocation table that randomly assigns subjects by order of enrollment to receive study gel for [redacted]) in 1 of 2 treatment arms (A or B). Separate allocation tables will be prepared for 120 male and 120 female subjects. Male or female enrollment may exceed the target sample size (n=120 each gender) by ±10%. If the target sample size is exceeded in one gender, the next available kit number in the other gender will be assigned.

2.2.5 Sample Size Determination

The parameters are conservatively estimated based on the DOMS study with [redacted] gel. The between-group difference in the mean [redacted] (using NPRS: numerical pain rating scale from 0 to 10) is 11.07 with a SD of 25.62. To have statistical power of 90%, 113 subjects per group are required. Taking into account information loss caused by missing data, it is estimated that 120 subjects need to be treated with each study gel.

2.2.6 Interim Analysis for Futility Assessment

[redacted]. The interim analysis will assess the futility of the trial (conditional power <0.05) based on efficacy of K-285 to menthol gel using the FAS. That is, if the conditional power is less than 0.05 (i.e., < 5%), the study would have met the stopping rule/decision criteria to stop for futility. The conditional power will be calculated under the assumption that the effect size estimate at the interim analysis is true. See Section 11 of SAP for more details.

An independent team of biostatisticians and statistical programmers will perform the interim analysis within a folder that has restricted access to only the independent team members to maintain the blinding of the study.

3 GENERAL ANALYSIS AND REPORTING CONVENTIONS FOR INTERIM ANALYSES

To maintain the blinding of the study, all outputs for interim analysis will be presented in blinded format (e.g., data will be summarized overall (not by treatment group) and treatment group will not be displayed in data listings). Data captured from both eDiary vendors (ERT for the first 20 randomized patients and Trialogics for the remaining patients) will be included for analyses as appropriate.

All continuous study assessments will be summarized overall and by time point (as applicable) using the descriptive statistics n, mean, SD, median, and range (minimum, and maximum). All of the categorical study assessments will be summarized overall and by time point (as applicable) using frequency counts and rates of occurrence (%). Changes from baseline for continuous

outcomes will be presented as their corresponding continuous measures for post-baseline visits. All study data will be listed by subject, and time point (as applicable).

No preliminary rounding will be performed; rounding will only occur after the analysis. To round, consider the digit to the right of the last significant digit: if <5, then round down; if ≥5, then round up. Means and medians will be presented with one more decimal place than the precision of the data. Standard deviations will be presented with two more decimal places than the precision of the data. Percentages will be presented with one decimal place. A percentage of 100% will be reported as 100%. Minimums and maximums will be presented with the same precision as the original data.

All analyses will be performed using the SAS System® version 9.4 or higher. The domain (Study data tabulation Model [SDTM]) and analysis (Analysis Data Model [ADaM]) data sets will be taken as input to the SAS programs that generate the report-ready tables, figures and listings. The submission ready SDTM and ADaM datasets will be provided to the sponsor along with display deliveries.

The following conventions will be used throughout the study analysis:

- Time T0 is the time of first treatment (first gel application) with study drug.
- The date of first treatment with study drug is Day 1.
- Assessment visit times are defined by time of treatment administration.
- Baseline value is defined as the last valid measurement prior to the dosing of study treatment.
- The change from baseline will be calculated as post-baseline value minus baseline value. If either the baseline or post baseline value is missing, the change from baseline is set to missing as well.
- Pain intensity difference (PID) is to be calculated by subtracting the pain intensity at each time point from the pain intensity at time 0
  $\mathbf{PID_t = PI_t - PI_0}$
- The number of days in the study is computed as: [Date of study completion or withdrawal minus the date of first study drug administration] + 1.

4 STUDY ASSESSMENTS

4.1 Efficacy Evaluations

4.1.1 Pain Intensity

Prior to exercise to induce DOMS, pain will be assessed for both legs at rest and while standing using a 0-100 point paper VAS to exclude subjects with other pain.

Following exercise to induce DOMS, pain at rest and while standing will be assessed for both legs using a 0-100 point eVAS and a categorical pain scale to determine qualification for

randomization and to identify the study leg (baseline assessment). All pain assessments will be recorded in the e-diary.

After randomization, pain at rest and while standing will be assessed for the study leg and the non-study leg using a 0-100 point eVAS at all post-study gel application time points in Period 1 (see Table 2 of Protocol). When study gel application and pain assessment occur at the same time point, pain assessments will be performed before study gel application.

The subject will rest quietly for at least 5 minutes before assessing pain at rest for the study leg and non-study leg (the study leg will be assessed first). The subject will sit for at least 5 minutes, then stand up and rate the worst pain caused by standing (the act of moving from a sitting to standing position) for the study leg and non-study leg (the study leg will be assessed first).

4.1.2 Visual Analogue Scale

The pain VAS, a unidimensional measure of pain intensity, is a continuous scale from 0 to 100 comprised of a horizontal line anchored by 2 descriptors, one for each symptom extreme: “no pain” (score of 0) and “worst possible pain” (score of 100). A paper version of the pain VAS will be used for pre-exercise pain assessments (see Appendix 1 of Protocol).

The pain eVAS is electronically displayed and is completed by the subject for baseline and each post-randomization pain intensity assessment by touching the point on the eVAS that represents their pain intensity. (A capture program will linearly convert the pixel touched to an integer between 0 and 100). The integers of the eVAS will be referred to as points. The assessment date and time for the study leg and for the non-study leg is recorded in the e-diary (see Appendix 1 of Protocol).

4.1.3 Categorical Pain Scale

A 4-point categorical pain scale (none, mild, moderate, severe) at rest will be used to assess pain intensity following the exercise session. A categorical pain scale is provided in Appendix 2 of Protocol.

4.1.4 Rescue Medication Use

Use of any rescue medication for DOMS is strongly discouraged until 168 hours after the first study gel application, at which time over the counter (OTC) acetaminophen (one 500 mg acetaminophen tablet twice daily, not more than every 6 hours) as rescue medication is permitted if required. Subjects experiencing severe pain, or inability to move or walk due to pain, may be permitted to use rescue medication (one 500 mg OTC acetaminophen tablet twice daily, not more than every 6 hours) prior to 168 hours after the first study gel application. If subjects take rescue medication within the first 168 hours after the first study gel application, it will be recorded in the e-diary as rescue medication and entered into the eCRF as a concomitant medication, and will be programmatically categorized as a concomitant medication. Subjects will record their pain

intensity in the e-diary before taking the rescue medication. The number of tablets taken and the date and time will be recorded in the e-diary.

4.2 Safety Evaluations

No safety analyses will be performed for interim analysis.

5 SUBJECT SUMMARIES

5.1 Analysis Populations

Analysis populations are defined as follows:

- Full Analysis Set (FAS) will include all randomized subjects who have a baseline pain score and at least 1 study gel applied. Subjects will be analyzed according to the treatment they are randomized to.
- The per-protocol set (PPS) will include all randomized subjects who have a baseline pain score, at least 1 study gel applied, and no major protocol violations. Subjects will be analyzed by treatment group according to the actual treatment received, i.e., “as treated”.
- Safety set will include all subjects who have at least 1 study gel applied. Subjects will be analyzed by treatment group according to the actual treatment received, i.e., “as treated”.

Analysis populations will be determined before final data base lock and study unblinding.

The primary and secondary efficacy analyses will be performed using FAS population. Efficacy analyses will also be carried out using PPS populations for selected endpoints as appropriate. Baseline characteristics will be summarized using FAS population and safety endpoints will be summarized using Safety population.

5.2 Disposition of Subjects

The number of subjects screened, randomized, and treated will be summarized. The number of subjects discontinuing treatment/withdrawing from the study will be summarized along with the reasons for discontinuation/withdrawal. Subjects who are screen failures along with the inclusion/exclusion criteria violation will also be summarized.

A listing of randomized and subject disposition will also be produced.

6 DEMOGRAPHIC AND OTHER BASELINE CHARACTERISTICS

6.1 Demographics and Baseline Characteristics

Demographic variables include age, sex, race, and ethnicity. Baseline characteristics include height, weight (kg), and body mass index (BMI; kg/m2). Demographics and baseline characteristics will be presented in a by-subject listing and summarized overall and by treatment group using FAS population.

For quantitative data, means, medians, standard deviations, minimum and maximum values will be determined. For qualitative data, counts and percentages will be calculated.

7 STUDY DRUG EXPOSURE

Gel application information is captured on eDiary. Subjects who use eDiary from ERT are not allowed to enter prior missed gel application. However, for subjects who use eDiary from Trialogics, if subjects who miss the previous gel application entry (and only the previous one) will be allowed to enter the missed gel application when they login in prior to the next scheduled gel application diary. Since the actual gel application time for the missing gel application won't be captured at the next entry, the date/time for the missing gel application will be imputed with the previous scheduled gel application time.

8 Details of each gel application will be displayed in data listings. EFFICACY EVALUATION

8.1 Efficacy Endpoints

8.1.1 Primary Efficacy Endpoint

The primary efficacy endpoint for this study is the sum of PID between 0 (baseline) to 24 hours (SPID0-24) for the study leg after the first study gel application while standing using a 0-100 point eVAS.

8.2 Handling of Dropouts or Missing data

Missing VAS values caused by early withdrawal from the study due to events which are not preferable to subjects such as an adverse event (AE) and/or lack of efficacy (LOE) will be imputed by last observation carried forward (LOCF).

If missing VAS values caused by withdrawal other than AE and/or LOE will be imputed under the missing at random (MAR) assumption with the multiple imputation (MI) technique. The imputation will be carried out in SAS version 9.4 or later, using the full conditional specification method (FCS) with a regression model approach (Carpenter and Kenward, 2013). By using the MI technique, 100 datasets will be generated. After calculating the SPID, an analysis of covariance (ANCOVA) model will be applied for the primary endpoint with gender and treatment as fixed effects and baseline pain as a covariate in each dataset. Multiple results will be combined using Rubin's rule (Little and Rubin, 1987), and will be done in SAS using PROC MIANALYZE. A significance test of the treatment difference at the two-sided 0.05 level and corresponding 95% confidence intervals

will be calculated. However, the multiple imputation is used for subjects that have some baseline and post baseline data, i.e., if a subject is missing all pre and post data, no data will be imputed.

If there exist missing VAS values to be imputed, any such values will be imputed before calculating the SPID. Intermediate missing VAS values due to an omission of recording pain intensity or subject's sleep will not be imputed, therefore, the observed values before and after the missing measurement will be directly connected in order to calculate the SPID. If subjects need to take the rescue medication, they will record their pain intensity in the eDiary before taking the rescue medication. If the rescue medication is taken within 6 hours of a nominal pain intensity assessment time point, the pain intensity score that was recorded before taking the rescue medication will be carried forward to the next pain intensity assessment time point.

8.2.1 Assessment Time Windows

Data are generally reported according to the nominal time of clinic visits and assessments as specified in the protocol. Refer Section 2.2.1 of this SAP (study design) for time windows for different assessments.

8.3 Analysis Method for the Primary Efficacy Endpoint

The primary efficacy endpoint for this study is the sum of PID between [redacted]) for the study leg after the first study gel application while standing using a 0-100 point eVAS. The primary analysis of the primary endpoint will be based on the FAS.

[redacted]

The durations between nominal time points will be calculated using the actual times of the pain score measurement. If the actual time is missing the nominal planned time will be used.

The primary endpoint will be analyzed using an ANCOVA model, which will include treatment and gender as fixed effects and baseline eVAS score as a covariate. The least square (LS) mean, standard error (SE) and associated 95% confidence interval (CI) for each treatment group will be estimated. In addition, the LS mean difference between each treatment group, SE, p-value and the associated 95% CI will also be computed. Descriptive summaries (including mean, SD, median, minimum and maximum) will also be presented. Missing data imputation methodology is described in Section 8.2 in this SAP.

Distributional assumptions underlying the model used for analysis will be examined by obtaining a normal probability plot of the residuals and a plot of the residuals versus the fitted values (i.e.

checking the normality assumption and constant variance assumption of the model respectively) to gain confidence that the model assumptions are reasonable.

To explore the impact of collecting e-diary data from different vendors, a sensitivity analysis by including both baseline eVAS score and the e-diary vendor with and without the interaction term of treatment and the e-diary vendor in the model will be performed.

9 SAFETY EVALUATION

Not applicable for interim analysis.

10 PHARMACOKINETIC EVALUATION

Not applicable for this study.

11 INTERIM ANALYSES

[redacted]

[redacted]

12 REFERENCES

US Federal Register. (1998) International Council on Harmonization; Guidance for Industry: Statistical Principles for Clinical Trials. Department of Health and Human Services: Food and Drug Administration. Federal Register, Vol. 63, No. 179, September 16, 1998, page 49583.

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Chen, Y. H. J., D. L. DeMets, and K. K. G. Lan. 2004. Increasing the samples size when the unblinded interim result is promising. Statistics in Medicine

Jennison, C., and Turnbull, B.W. 2000. Group Sequential Methods with Applications to Clinical Trials. Chapman & Hall/CRF. New York.

13 APPENDICES

13.1 Example of SPID Calculation

Following provide an example of SPID using the trapezoidal rule described in Section 8.3 in this SAP:

[redacted]	PID	[redacted]
	0	
	-4	
	-14	
	-15	
	-13	
	-20	
	-13	
	-27	
	-21	
	-16	
	-16	
	-12	
	-21	
	-29	

13.2 Planned Tables, Figures and Listings for Interim Analysis

13.2.1 Tables

Table 14.1.1	All Subjects	Summary of Subject Disposition
Table 14.1.2.1	Full Analysis Set	Summary of Demographic and Baseline Characteristics

13.2.2 Listings

Listing 16.2.1	All Randomized Subjects	Randomization and Analysis Population Inclusion
Listing 16.2.2	All Randomized Subjects	Informed Consent and Re-Consent
Listing 16.2.3	All Randomized Subjects	Eligibility
Listing 16.2.3.1	Screen Failure Subjects	Eligibility
Listing 16.2.4	All Randomized Subjects	Subject Completion/Early Termination
Listing 16.2.5	All Subjects	Demographics and Baseline Characteristics
Listing 16.2.6	All Randomized Subjects	Study Drug Administration
Listing 16.2.7	All Subjects	Rescue Medications