STATISTICAL ANALYSIS PLAN

STUDY TITLE:	**A CORNEAL ENDOTHELIAL CELLS SUBSTUDY IN ASSOCIATION WITH GR42691 STUDY (AVONELLE-X): A MULTICENTER, OPEN-LABEL EXTENSION STUDY TO EVALUATE THE LONG-TERM SAFETY AND TOLERABILITY OF FARICIMAB IN PATIENTS WITH NEOVASCULAR AGE-RELATED MACULAR DEGENERATION**
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PLAN PREPARED BY:	[redacted], PhD

STATISTICAL ANALYSIS PLAN APPROVAL

SPONSOR: F. Hoffmann-La Roche Ltd
LEGAL REGISTERED ADDRESS: Grenzacherstrasse 124, 4070 Basel, Switzerland

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STATISTICAL ANALYSIS PLAN VERSION HISTORY

This Statistical Analysis Plan (SAP) was developed based on Roche SAP model document eMD: 01 May 2024 (SAP eMD_v3.0) and 01 Aug 2024 (Ophtha SAP Template_v1.0)

SAP Version	Approval Date	Based on Protocol (Version, Approval Date)
1.0	see electronic date stamp on the last page of this document	Substudy Protocol V2, 02 Nov 2022

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LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviation or Term	Description
AE	adverse event
AESI	adverse event of special interest
APTC	Anti-Platelet Trialists' Collaboration
ATE	arterial thromboembolic event
BCVA	best-corrected visual acuity
CEC	corneal endothelial cells
CSR	Clinical Study Report
CST	central subfield thickness
CV	coefficient of variation
ECD	endothelial cell density
GCP	Good Clinical Practices
IOI	intraocular inflammation
HEX	hexagonal endothelial cells
MedDRA	Medical Dictionary for Regulatory Activities
nAMD	neovascular age-related macular degeneration
mITT	modified intent-to-treat
PMR	post-marketing requirement
PTI	personalized treatment interval
PT	preferred term
SAEs	serious adverse events
SAP	Statistical Analysis Plan
SMQs	standardized MedDRA queries
SOC	System Organ Class
USFDA	United States Food and Drug Administration

1. INTRODUCTION

This Statistical Analysis Plan (SAP) specifies the planned analyses for the corneal endothelium cell (CEC) substudy of the main Study GR42691.

The main Study GR42691 is a multicenter long-term extension study designed to evaluate the long-term safety and tolerability of intravitreal faricimab 6 mg administered at a personalized treatment interval (PTI) to patients with neovascular age-related macular degeneration (nAMD) who enrolled in and completed one of the Phase III studies, GR40306 and GR40844.

This substudy is designed to evaluate the impact on corneal endothelial cells in the study eyes of patients with nAMD receiving intravitreal faricimab (6 mg) to fulfill a United States Food and Drug Administration (USFDA) post-marketing requirement (PMR). Specifically, the U.S. FDA PMR is stated as “conduct a controlled trial to evaluate the corneal endothelial health of eyes treated with faricimab by monitoring the number/density of corneal endothelial cells using specular microscopy at baseline and over a period of at least one year in at least 100 patients receiving faricimab.”.

The corneal endothelium is a single layer of cells lining the posterior portion of the cornea that regulates the health and optical transparency of the cornea (Van den Bogerd et al. 2019). The structural and functional integrity of the corneal endothelium can be impacted by a variety of extrinsic and intrinsic factors, including genetics, race, age, corneal dystrophy, ocular trauma, intraocular surgery, ultraviolet radiation, and intraocular infection. Morphological changes in the corneal endothelium occur throughout life, including a significant decline in corneal endothelial cell density (ECD) and percentage of hexagonal cells with age (Bourne et al. 1997; Zavala et al. 2013).

Specular microscopy is an imaging modality that allows for direct assessment of the corneal ECD and morphology and is routinely used in the assessment of corneal endothelial cell health (Chaurasia and Vanathi 2021). Variables studied in the specular microscopy examination include ECD, percentage of hexagonal endothelial cells (HEX), and coefficient of variation (CV) of cell area and collectively allow the clinician to interpret the health status of the CECs (Chaurasia and Vanathi 2021). Human corneal endothelium consists of a monolayer of hexagonal cells situated in the posterior surface of the cornea (Schmedt et al. 2012) and healthy cornea has approximately 60% of HEX (Chaurasia and Vanathi 2021). The percentage of HEX decreases (pleomorphism) and the CV of cell area increases (polymegathism) with age and endothelial cell attrition due to various causes (Chaurasia and Vanathi 2021).

The purpose of this document is to provide details of the planned analyses. The analyses and endpoints specified in this document supersede the analysis plan described in the study protocol. Changes to the protocol-planned analyses are described in Section 4.8.

1.1 OBJECTIVES AND ENDPOINTS

This substudy will evaluate the impact of faricimab on corneal endothelial cells in patients with nAMD. Specific objectives and corresponding endpoints for this substudy are outlined below.

The primary objective for this substudy is to evaluate the impact of faricimab on corneal endothelial cell health on the basis of the following endpoints, as assessed by specular microscopy:

- **Primary endpoint:** Percent change in corneal endothelial cell density from baseline at Year 1 (defined as the earliest substudy visit closest to Week 52 occurring between Week 48 and Week 64) in the study eye as compared with the fellow eye
- **Secondary endpoint:** Percent change in corneal endothelial cell density from baseline at substudy midpoint (defined as the earliest substudy visit closest to Week 24 occurring between Week 20 and Week 28) in the study eye as compared with the fellow eye
- **Exploratory endpoints:**
    - Percent change in the coefficient of variation of corneal endothelial cell area (standard deviation of the cell area/mean cell area) from baseline at substudy midpoint and Year 1 in the study eye as compared with the fellow eye
    - Percent change of hexagonal endothelial cells from baseline at substudy midpoint and Year 1 in the study eye as compared with the fellow eye

1.2 STUDY DESIGN

The main Study GR42691 (AVONELLE-X) is a multicenter long-term extension study designed to evaluate the long-term safety and tolerability of intravitreal faricimab 6 mg administered at a PTI to patients with nAMD who enrolled in and completed one of the Phase III studies, GR40306 and GR40844.

This substudy is designed to evaluate the impact of intravitreal faricimab 6 mg administered at a PTI on the health of the corneal endothelial cells in patients with nAMD. Fellow eyes of enrolled patients will be used as a comparator for corneal endothelial cell analyses. Prior and/or current administration of faricimab in the fellow eye is one of the exclusion criteria for this substudy (refer to protocol Section 4.1.2). During the substudy, the following therapies are prohibited:

- faricimab
- brolucizumab (Beovu®)
- bevacizumab (Avastin®)
- Port Delivery System implantation (Susvimo™)

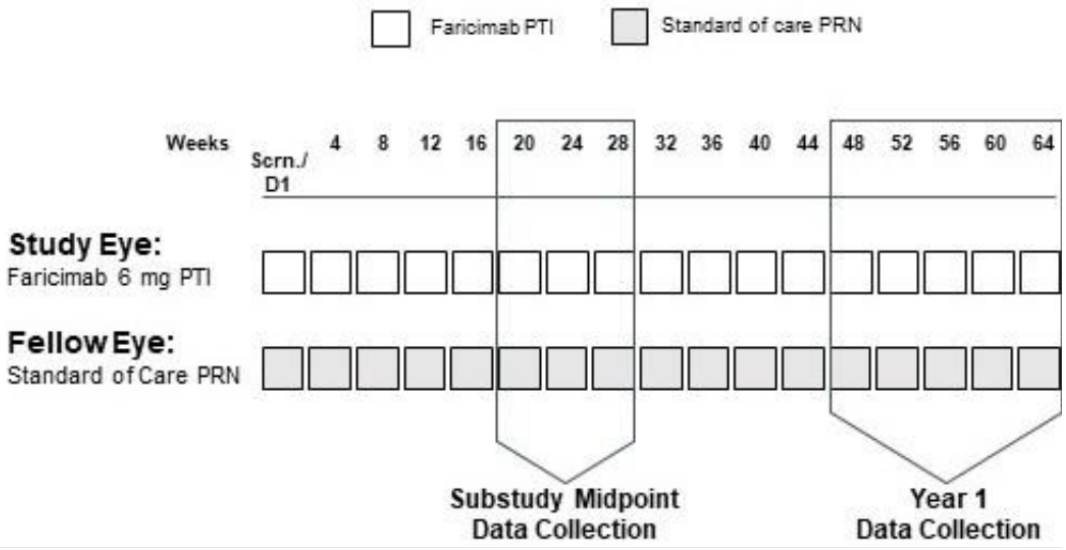
Patients in the main Study GR42691 will be eligible to screen for participation in this substudy. During the substudy screening visit, eligibility to participate will be determined

based on the inclusion and exclusion criteria for this substudy (refer to protocol Section 4.1). Study GR42691 patients are required to be willing to participate in the CEC substudy for at least 48 weeks and have at least their first CEC substudy visit while enrolled in the main Study GR42691.

Patients who do not meet the criteria for participation in this substudy (screen failure) may qualify for one re-screening opportunity (for a total of two screenings per patient) at the investigator's discretion. Patients are not required to re-sign the consent form if they are re-screened within 90 days after previously signing the consent form. The investigator will maintain a record of reasons for screen failure (refer to protocol Section 4.4.1).

Figure 1 presents an overview of the substudy design. A schedule of activities is provided in protocol Appendix 1.

Figure 1 Substudy Assessment Schema



D1 = Day 1; PRN = pro renata (as needed); PTI = personalized treatment interval; Scrn = screening visit.

Substudy midpoint data collection timepoint is defined as the earliest substudy visit closest to Week 24 occurring between Week 20 and Week 28. Year 1 data collection timepoint is defined as the earliest substudy visit closest to Week 52 occurring between Week 48 and Week 64.

When possible, patients will follow the scheduled assessments per the main protocol (see main Study GR42691 protocol Appendix 1). However, for some patients, an additional visit may need to be scheduled in between main protocol visits for the substudy midpoint visit. Substudy patients will undergo specular microscopy imaging in both eyes to assess number/density of corneal endothelial cells at screening, substudy

midpoint, and Year 1 prior to the application of any topical ophthalmic anesthetic, tonometry, or any other study treatment.

Completion of the substudy may occur prior to or after main study completion. If the patient withdraws from the substudy only, then the patient may remain in the main Study GR42691.

If the patient withdraws from the main Study GR42691, then the patient will also be withdrawn from this substudy.

If the patient discontinues treatment but remains in the main Study GR42691 for follow-up, then the patient will remain in this substudy. All patients who remain in this substudy will be strongly encouraged to complete specular microscopy imaging at substudy midpoint and Year 1.

The end of this substudy is defined as the date when the last patient, last visit occurs. The end of the substudy is expected to occur approximately 48 weeks after the last substudy patient is enrolled.

1.2.1 Treatment Assignment and Masking

There is no additional treatment compared to the main Study GR42691.

1.2.2 Independent Review Facility

A central reading center with expertise in specular microscopy assessment provided sites with the specular microscopy manual and training materials for specified substudy corneal images. Before any substudy images are obtained, site personnel, test images, systems, and software (where applicable) are certified and validated by the reading center as specified in the corneal reading center manual. All specular microscopy images are obtained by trained site personnel at the study sites, forwarded to the central reading center for independent analysis and/or storage, and later transferred to the Sponsor.

1.2.3 Data Monitoring

Not applicable

2. STATISTICAL HYPOTHESES AND SAMPLE SIZE DETERMINATION

2.1 STATISTICAL HYPOTHESES

No formal hypothesis testing is planned for this study.

The paired t-tests are planned for efficacy analyses for reference purposes and thus not considered formal. In general, the paired t-tests will compare the percent change from baseline between the study eye and fellow eye in each CEC parameter described in

Section 4.2, Section 4.3 and Section 4.4, respectively in the specified groups. The test will be 2-sided, with the null hypothesis of no difference in percent change from baseline between the study eye and fellow eye in each patient. P-values and 95% confidence interval of the difference from the paired t-tests will be provided.

2.2 SAMPLE SIZE DETERMINATION

No formal sample size calculations were performed for this substudy. A total of 117 patients were enrolled in this substudy from the main Study GR42691.

3. ANALYSIS SETS

Similar to main study analysis, patients from Good Clinical Practices (GCP) non-compliance affected site will not be included in the analyses due to possible impact of the GCP non-compliance. Details about the non-compliance can be found in AVONELLE-X main study SAP Section 3 and main study Clinical Study Report (CSR).

The analysis sets for the purposes of analyses are defined in Table 1.

Table 1 Analysis Sets

Analysis Set	Description
mITT set	All enrolled participants who received at least one injection of faricimab in the study eye during this substudy (with patients at GCP non-compliance affected site excluded from the analyses).
Safety-Evaluable set	All enrolled participants who received at least one injection of faricimab in the study eye during this substudy (with patients at GCP non-compliance affected site excluded from the analyses).

mITT = modified intent-to-treat

In general, analyses will include observations collected from substudy screening through to substudy completion/discontinuation, regardless of previous parent study treatment assignment.

Specific data points to be included for each analysis will be described in Section 4.

4. STATISTICAL ANALYSES

The analyses described in this section are based on patients enrolled in this CEC substudy. The analysis will be performed at the same time of the final analysis of the main study (refer to main SAP of GR42691).

4.1 GENERAL CONSIDERATIONS

Unless specified otherwise, efficacy analyses will be performed on the modified intent-to-treat (mITT) set. Safety analyses will be performed on safety-evaluable set.

For efficacy analyses specified in Section 4.2, Section 4.3 and Section 4.4, baseline will be defined as the screening/Day 1 CEC image taken date. Midpoint will be defined as

the earliest analysis substudy visit closest to Week 24 occurring between Week 20 and Week 28. Year 1 will be defined as the earliest substudy visit closest to Week 52 occurring between Week 48 and Week 68.

Descriptive summaries for continuous endpoints will include number of observations, arithmetic mean, standard deviation, 95% CI for mean, median, minimum, maximum, first quartile (Q1), and third quartile (Q3). Descriptive summaries for binary/categorical endpoints will include frequency and/or percentages.

Data (from both study eye and fellow eye) collected after the fellow eye use of the prohibited therapies (listed below) will be excluded from the efficacy analysis:

- faricimab
- brolucizumab (Beovu®)
- bevacizumab (Avastin®)
- Port Delivery System implantation (Susvimo™)

Missing data will not be imputed.

4.2 PRIMARY EFFICACY ENDPOINT ANALYSIS

4.2.1 Definition of Primary Endpoint/Estimand

The primary efficacy endpoint is the percent change from baseline in study eye ECD value at Year 1.

4.2.2 Main Analytical Approach for Primary Endpoint

Available corneal ECD values and percent change from baseline in ECD values will be summarized descriptively at baseline and at Year 1, for study and fellow eyes respectively.

In addition, the percent change in corneal ECD from baseline at Year 1 will be analyzed using the paired t-test comparing study and fellow eyes. P-value and 95% interval from the paired t-test will be provided.

Paired t-test will be performed among the subset of patients who had an interpretable baseline and Year 1 specular microscopic image in both eyes.

Paired t-test comparing study eyes and fellow eyes within each of the following subgroups will also be provided:

- Baseline age (<65, ≥ 65 to <75, and ≥ 75 years)

4.3 SECONDARY EFFICACY ENDPOINT ANALYSIS

The secondary efficacy endpoint is the percent change from baseline in study eye ECD value at midpoint. The secondary endpoint will be analyzed using the same analysis methods as the primary endpoint, as described in Section 4.2.

4.4 EXPLORATORY EFFICACY ENDPOINT ANALYSIS

Exploratory endpoints include:

1. Percent change in the coefficient of variation of corneal endothelial cell area (standard deviation of the cell area/mean cell area) from baseline at substudy midpoint and Year 1 in the study eye
2. Percent change of hexagonal endothelial cells from baseline at substudy midpoint and Year 1 in the study eye

Exploratory endpoints will be analyzed using the same analysis methods as the primary endpoint, as described in Section 4.2, with the exception that subgroup analyses will not be performed for exploratory endpoints.

4.5 SAFETY ANALYSES

Safety analyses will be performed using the safety evaluable population, all data points collected will be used. Missing data will not be imputed.

4.5.1 Extent of Exposure

Exposure to faricimab treatment (number of treatments and duration of treatment) in the study eye during the substudy will be summarized using the safety evaluable set. Faricimab treatment in the study eye after completion of AVONELLE-X will be included.

The exposure period will be calculated from the screening/Day 1 CEC image taken date to the last faricimab injection (including injections during AVONELLE-X or after the AVONELLE-X completion as concomitant medication) in the study eye or last dose hold, whichever is later.

4.5.2 Adverse Events

All Adverse Events (AEs) during the substudy are reported in the main study GR42691 database. If a patient's duration on the CEC substudy extends beyond the duration of the main study, safety data will continue to be collected per the same reporting mechanism as the main study period until the final CEC substudy visit. Only AEs occurring during the substudy will be analyzed, which is defined as any new adverse event reported or any worsening of an existing condition on or after the substudy screening in this CEC substudy.

All verbatim AEs terms will be coded using the latest version of Medical Dictionary for Regulatory Activities (MedDRA).

Non-ocular AEs will be tabulated by System Organ Class (SOC) and Preferred Term (PT). Ocular AEs in the study eye will be tabulated by PT.

Frequency tables, including patient incidence rates, will be provided for the adverse events listed below. Ocular AEs will be reported for study eyes. Selected summaries and listings of AE in the fellow eyes may be provided.

- Ocular AEs and serious adverse events (SAEs)
- Ocular AEs by severity
- Adverse events of special interest (AESI) defined as follows:
    - Cases of potential drug-induced liver injury that include elevated alanine aminotransferase or aspartate aminotransferase in combination with either an elevated bilirubin or clinical jaundice, as defined by Hy's Law (see Section 5.2.3 of the main study protocol)
    - Suspected transmission of an infectious agent by the study drug
    - Sight-threatening AEs (see Section 5.2.3 of the main study protocol for the definitions)
- Selected ocular AEs
    - Intraocular inflammation
    - Infectious endophthalmitis
    - Retinal vascular occlusion
- Treatment related ocular AEs and SAEs as determined by the investigator
- AEs leading to discontinuation from AVONELLE-X
- AEs leading to discontinuation of AVONELLE-X study treatment
- AEs leading to interruption of AVONELLE-X study treatment
- Non-ocular AEs and SAEs
- Non-ocular adverse events of arterial thromboembolic event (ATE) and cerebrovascular haemorrhagic adverse events
- Externally adjudicated APTC events
- Deaths

As there is no standardized MedDRA queries (SMQ) encompassing the medical concepts of intraocular inflammation (IOI), infectious endophthalmitis or retinal vascular occlusion, the definitions will be assessed based on the review of clinical database PTs or reported AEs from clinical database prior to data cut and final database lock, and with subsequent MedDRA version updates, to ensure no event terms are missed.

Listings of AEs, SAEs, AESIs, APTCs, ATEs, Intraocular Inflammation and deaths will be provided.

A listing of AEs, including ocular AEs in the study eye and non-ocular AEs, occurring to the patient at GCP noncompliance affected site (described in Section 3) will be provided.

4.5.3 Additional Safety Assessments

No additional safety assessments, laboratory data, vital signs or ocular assessment, were planned for this substudy. No summaries are planned.

4.6 OTHER ANALYSES

4.6.1 Summaries of Conduct of Study

Patient disposition including the number of participants enrolled and completing the entire substudy will be summarized in the mITT set. Reasons for premature substudy discontinuation will be tabulated. Listing of substudy discontinuations will be provided. Detailed, free text reasons for discontinuations due to physician decision, withdrawal by subject and 'other' reasons will be included in the listing.

The number of participants will be tabulated by country and site.

Eligibility criteria and other major protocol deviations will be tabulated in the mITT set, and a listing will be provided.

4.6.2 Summaries of Treatment Group Comparability

There is only one treatment group in the main study therefore, comparability across treatment groups is not applicable. Clinically important demographics will be summarized using the mITT set. The variables will be summarized using means, standard deviations, medians, and ranges for continuous variables, as well as using counts and proportions for categorical variables. The baseline summary will include:

- Age
- Sex
- Race
- Ethnicity
- Disease characteristics (BCVA, CST, etc)

Demographic data (e.g., sex and race or ethnicity) will originate from the main Phase III studies. Baseline age will be calculated using the date of baseline CEC image.

Baseline ocular disease characteristics (e.g., baseline BCVA, CST) will be assessed with values originating from the main study (GR42691).

Baseline ocular characteristics collected at the substudy screening visit will be summarized by eye (study eye versus control eye).

4.6.3 Pharmacokinetic Analyses

Not planned for this substudy.

4.6.4 Immunogenicity Analyses

Not planned for this substudy.

4.7 INTERIM ANALYSES

No interim analysis is planned for this substudy.

4.8 CHANGES TO PROTOCOL-PLANNED ANALYSES

Per-protocol analysis was removed from the analysis plan as the study intends to reflect the impact of treatment in a real-world setting, where participants may or may not adhere to a clinical study protocol when taking/receiving medication.

Adverse Event analyses, which were not specified in the protocol, are currently planned in this SAP document, in order to provide a comprehensive safety summary during the substudy, in light of the fact that a patient's duration on the CEC substudy could extend beyond the duration of the main study.

Analyses by the main Phase III study treatment group in this substudy will not be conducted. This is due to the fact that the substudy consists of a single treatment group (faricimab), which differs from the Phase III study that includes a comparative group.

The subgroup analysis based on the number of faricimab injections received during the substudy has been removed. This decision is made because participants enrolled in the substudy at different timepoints during the main study and received faricimab prior to the substudy enrollment. Hence, the injection numbers received during the substudy do not accurately represent the cumulative exposure to faricimab in the patients, rendering such analysis less clinically meaningful and potentially misleading.

Subgroup analyses of primary and secondary endpoints by age are considered sufficient to understand the effect of age on efficacy, hence no further subgroup analyses are planned for exploratory endpoints.

5. REFERENCES

Bourne WM, Nelson LR, Hodge DO. Central corneal endothelial cell changes over a ten-year period. Invest Ophthalmol Vis Sci 1997; 38: 779-82.

Chaurasia S, Vanathi M. Specular microscopy in clinical practice. Indian Journal of Ophthalmology 2021; 69: 517-2.

Schmedt T, Chen Y, Nguyen TT, et al. Telomerase immortalization of human corneal endothelial cells yields functional hexagonal monolayers. PLoS ONE 2012; 7:e514.

Van den Bogerd B, Zakaria N, Adam B, et al. Corneal endothelial cells over the past decade: are we missing the mark(er)?. Transl Vis Sci Technol 2019; 8:13.

Zavala J, López Jaime GR, Rodríguez Barrientos CA, et al. Corneal endothelium: developmental strategies for regeneration. Eye (Lond) 2013; 27:579.

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