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Title Page

Protocol Title: An open-label, non-randomized, multi-center, phase 4 pharmacokinetic study to evaluate the systemic exposure after bilateral intravitreal administration of high dose (8 mg) aflibercept in adults with diabetic macular edema or neovascular age-related macular degeneration

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Compound Number: BAY 865321, 8 mg aflibercept

Short Title: Study to evaluate exposure after 8 mg aflibercept bilateral therapy

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Table of Contents

Title Page ---- **1**

Table of Contents ---- **2**

Table of Tables ---- **3**

Table of Figures ---- **3**

Version History ---- **4**

List of Abbreviations ---- **6**

1. Introduction ---- **8**
1.1 Objectives, Endpoints, and Estimands ---- 8
1.2 Study Design ---- 11

2. Statistical Hypothesis ---- **14**
2.1 Multiplicity Adjustment ---- 14

3. Analysis Sets ---- **15**

4. Statistical Analyses ---- **16**
4.1 General Considerations ---- 16
4.2 Pharmacokinetics ---- 17
4.2.1 Primary Endpoint/Estimand Analysis ---- 17
4.2.2 Exploratory PK Endpoints/Estimands Analysis ---- 19
4.2.3 Other Exploratory Endpoints Analysis ---- 20
4.3 Efficacy Analysis ---- 21
4.4 Safety Analysis ---- 21
4.4.1 Extent of Exposure ---- 21
4.4.2 Adverse Events ---- 22
4.4.3 Additional Safety Assessments ---- 22
4.5 Analysis of Immunogenicity Data ---- 23
4.5.1 Other Variables and/or Parameters ---- 23
4.5.2 Subgroup Analyses ---- 24
4.6 Interim Analyses ---- 24
4.7 Changes to Protocol-planned Analyses ---- 24

5. Sample Size Determination ---- **26**

6. Supporting Documentation ---- **27**
6.1 Appendix 1: Participant Disposition ---- 27
6.2 Appendix 2: Demographic and Other Baseline Characteristics ---- 27
6.3 Appendix 3: Protocol Deviations ---- 28
6.4 Appendix 4: Medical History ---- 28
6.5 Appendix 5: Prior and Concomitant Medication and Procedures ---- 28
6.5.1 Prohibited Medications and Procedures ---- 28
6.6 Appendix 6: Schedule of Assessment for PK Samples ---- 29
6.7 Appendix 7: Missing/Incomplete Date Imputation ---- 35

6.8 Appendix 8: PTs for Selection of Safety Topics ---------- 38

7. References ---------- 40

Table of Tables

Table 3-1: Analysis Sets 15
Table 4-1: Strategies for handling ICE occurrence for PK endpoints concerning the maximum observed concentrations 18
Table 6-1: PK Sampling Schedule Regimen 1 30
Table 6-2: PK Sampling Schedule Regimen 2 32
Table 6-3: PK Sampling Schedule Regimen 3 33
Table 6-4: PTs for Hypertension 38

Table of Figures

Figure 1-1: Study Flow Diagram 13
Figure 1-2: Dosing Schedule 13

Version History

This Statistical Analysis Plan (SAP) for study 22578 is based on the Protocol Version 5.0, Amendment Number 3, dated 21 JAN 2025 and electronic Case Report Form (eCRF), Version 1.0 (29 AUG 2024).

SAP Version	Date	Change	Rationale
1.0	Date of last Signature	Not applicable	Original version
2.0	Date of last Signature	Section 1.1: Intercurrent event and strategy added for additional aflibercept injection	Describe additional ICEs occurred in the study.
		Section 3: Definition of mPKS updated, i.e. handling of PK samples outside of the sample window added.	Exclude samples taken at incorrect timepoints (i.e. invalid data).
		Handling of PK value outliers added.	Exclude samples which have an impact on the analysis of the PK data.
		Section 4 / Section 6: Removal of data listings, except of PD, SAE and procedures listing.	To remove analyses that are not necessary for the clinical study report.
		Section 4.1: Update of baseline definition for blood pressure.	Baseline definition in consistency with pivotal phase III studies in AMD and DME (PULSAR and PHOTON).
		Section 4.1: Update of mapping for EOS/EOT visits.	EOS/EOT visit mapping: Consistency with pivotal studies, to use data from similar timepoints in statistical analysis by visit (according to the visit schedule defined in the protocol).
		Section 4.2.1.1:	Exclude samples from participants who took an additional aflibercept

		Intercurrent event and strategy added for additional aflibercept injection. Section 4.2.2: References to estimand approach of section 4.2.1.1 added. Section 4.4.2: Removal of summaries for nasal mucosal events and intraocular inflammation. Section 4.4.3.1: Handling of vital signs taken outside of the time window added. Section 4.6: Removal of section "Other Analyses". Section 4.7: Details added for mPKS definition. Section 6.3: Reference to section 4.2.1.1 and 4.4.3.1 added. Section 6.5: Update of coding to be used for procedures. Section 6.8: Removal of tables 6.5, 6.6 and 6.7 (preferred terms for intraocular inflammation, nasal mucosal events, and intraocular pressure).	injection, which has an impact on the analysis of their PK data. Minor updates/correction. Exclude samples taken at incorrect timepoints (i.e. invalid data). Minor updates/correction.

List of Abbreviations

AE	Adverse event
ADA	Anti-drug antibodies
APTC	Anti-Platelet Trialists' Collaboration
ATC	Anatomical Therapeutic Chemical
ATE	Arterial thromboembolic event
BCVA	Best Corrected Visual Acuity
BLOQ	Below limit of quantification
CI-DME	Central involved diabetic macular edema
C_{max}	Maximum observed concentration
CRT	Central retinal thickness
CSR	Clinical Study Report
C_t	Concentration at time t
DBP	Diastolic blood pressure
DME	Diabetic macular edema
DR	Diabetic retinopathy
ETDRS	Early Treatment Diabetic Retinopathy Study
ECG	Electrocardiogram
eCRF	electronic Case Report Form
EDC	Electronic data capture
EOS	End of study
EOT	End-of-Treatment
FAS	Full Analysis Set
HD	High dose or 8 mg aflibercept
ICE	Intercurrent event
IOP	Intraocular pressure
IVT	Intravitreal
LLOQ	Lower limit of quantification
MedDRA	Medical Dictionary for Regulatory Activities
mPKS	Modified pharmacokinetic analysis set
NAb	Neutralizing antibodies
nAMD	Neovascular age-related macular degeneration
PCSV	Potentially clinically significant value
PK	Pharmacokinetic
PKS	Pharmacokinetic analysis set
PT	Preferred Term
SAE	Serious adverse event
SAF	Safety analysis set
SAP	Statistical Analysis Plan

SAS	Statistical analysis software
SBP	Systolic blood pressure
SD	Standard deviation
SOC	System Organ Class
TEAE	Treatment-emergent adverse event
t_{max}	Timepoint where maximum concentration was observed
VEGF	Vascular endothelial growth factor

1. Introduction

The SAP describes the final analysis of the study which is relevant for the Clinical Study Report (CSR). One additional SAP describes the safety analysis not relevant for the CSR. No statistical interim analysis will be performed.

There are no changes to the analyses described in the protocol.

Bilateral treatment with 8 mg aflibercept is expected to be employed in clinical practice after the launch of EYLEA 114.3 mg/mL. While pivotal studies allowed for concurrent dosing of 8 mg aflibercept in the study eye and 2 mg aflibercept or other anti-vascular endothelial growth factor (VEGFs) in the fellow eye, bilateral dosing of 8 mg aflibercept has yet to be investigated.

Despite available treatments for Neovascular age-related macular degeneration (nAMD) and Diabetic macular edema (DME), there remained an unmet medical need for the development of anti-VEGF therapies with the potential to improve important clinical outcomes while decreasing the treatment burden via a reduction in the required frequency of intravitreal injections. 8 mg aflibercept was developed to respond to this unmet medical need.

This Phase 4 pharmacokinetic (PK) study is planned to help better understand systemic exposure to free and adjusted bound aflibercept after bilateral dosing with 8 mg aflibercept.

This SAP details the statistical methodology and data presentations for the clinical study report. Table, figure and listing specifications are contained in a separate document.

The analyses described in this SAP are based upon the following study documents:

- Study Protocol Version 5.0, Amendment Number 3 (21 JAN 2025)
- electronic Case Report Form (eCRF), Version 1.0 (26 JUN 2024)

1.1 Objectives, Endpoints, and Estimands

The objectives and endpoints for the study are:

Objectives	Endpoints
Primary	
• To evaluate the systemic PK after bilateral 8 mg aflibercept treatment	Primary endpoint: • Maximum observed concentration (C_{max}) of free aflibercept Exploratory endpoints: • Maximum observed concentration (C_{max}) of adjusted bound aflibercept • Maximum observed concentration (C_{max}) of total aflibercept • Timepoint (t_{max}) where C_{max} of free aflibercept was observed • Timepoint (t_{max}) where C_{max} of adjusted bound aflibercept was observed

Objectives	Endpoints
	• In Regimen 2: Observed concentration (C_t) of free aflibercept at Weeks 4, 8, 20, 32, and 44
Exploratory	Exploratory endpoints
• To evaluate immunogenicity of bilateral 8 mg aflibercept treatment	• Incidence of treatment-emergent and treatment-boosted neutralizing antibodies (NAb) and anti-drug antibodies (ADA) titers
• To assess the safety of bilateral 8 mg aflibercept treatment	• Occurrence of ocular and non-ocular treatment-emergent AEs (TEAEs) and all serious adverse events (SAEs)
• To explore the relationship between dose, drug concentration and/or PK parameters and selected systemic response variables after bilateral 8 mg aflibercept treatment	• Maximum change from baseline of systolic/diastolic blood pressure and associated concentration of free and adjusted bound aflibercept
• To generate additional data for the effect of bilateral 8 mg aflibercept treatment on anatomical measures of response and on visual acuity	• Change from baseline in Best Corrected Visual Acuity (BCVA) at the end of the study[1] • Change from baseline in Central retinal thickness (CRT) at the end of the study[1]

Footnotes:

1. End of study is Week 52 for Regimen 1, Week 48 for Regimen 2 and Week 49 for Regimen 3

Primary Estimand

The primary estimand for the primary objective is described by the following attributes:

- Population: Adult patients with DME or nAMD with either active disease in one eye and inactive disease under maintenance treatment in fellow eye OR active disease in both eyes
- Treatment condition: Aflibercept 8 mg administered to both eyes in one of 3 treatment regimens depending on the patient's clinical presentation:
    - Treatment Regimen 1 (for participants with active disease in one eye and fellow eye with inactive disease under maintenance):
        - Active eye: Aflibercept 8 mg administered with 3 initial monthly initiation doses followed by extension of treatment interval to 12 weeks

        - Fellow eye: Aflibercept 8 mg with first dose administered on the same day as the active eye followed by extension of treatment interval to 12 weeks
    - Treatment Regimen 2 (for participants with active disease in both eyes at screening and bilateral simultaneous treatment):
        - Aflibercept 8 mg administered with 3 initial monthly initiation doses followed by extension of treatment interval to 12 weeks. Participants receive treatment in both eyes at the same day
    - Treatment Regimen 3 (for participants with active disease in both eyes at screening and bilateral staggered treatment):
        - First eye: Aflibercept 8 mg administered with 3 monthly initiation doses followed by extension of treatment interval to 12 weeks
        - Second eye: Aflibercept 8 mg administered with 3 monthly initiation doses followed by extension of treatment interval to 12 weeks. All doses to second eye administered one week after treatment administration to the first eye
- Variable: Maximum observed concentration (C_{max}) of free aflibercept
- Intercurrent events and strategies:
    - Premature treatment discontinuation prior to end of dense PK sampling – addressed by the hypothetical strategy
    - Premature treatment discontinuation after end of dense PK sampling – addressed by the while-on-treatment strategy
    - Additional aflibercept injection not planned as per protocol (e.g. any amount of 2mg or 8mg concentrations) – addressed by a hypothetical strategy
    - Use of prohibited medication – addressed by the treatment policy strategy
    - Missed study intervention – addressed by the treatment policy strategy
- Population-level summary: geometric mean C_{max} per treatment regimen

Rationale for estimand strategies: For patients that discontinue treatment prior to the end of the dense PK sampling period as defined in Table 4-1 (also see Section 6.6, and Table 6-1, Table 6-2, and Table 6-3), calculation of the maximum concentration of free aflibercept is not meaningful. For these events, a hypothetical strategy is applied whereby such patient is excluded from the analysis. This approach is sensible since it is not assumed that the PK profile of these patients would be systematically different from those who remained in the study, regardless of their reason for treatment discontinuation.

Regarding premature treatment discontinuation after end of the dense PK sampling period, only the on-treatment period is relevant for the estimation of C_{max} of free aflibercept. Hence the intercurrent event ‘premature treatment discontinuation’ is addressed using the while-on-treatment strategy.

Administration of any additional aflibercept injections not planned as per protocol will increase the concentration of free aflibercept and might distort the maximum to be estimated. For these events a hypothetical strategy is applied: If the additional aflibercept injection is administered prior to the end of the dense PK sampling period, such patient is excluded from the analysis. If the additional aflibercept injection was administered after the end of the dense PK sampling

period, data after the event will be excluded. This approach is sensible since it is not assumed that the PK profile of these patients would be systematically different from those who are treated according to protocol.

Intake of prohibited medication is not assumed to have an impact on the concentration of free aflibercept and is therefore addressed using the treatment policy strategy. Missing an injection may also be reflected in clinical practice and is thus addressed by the treatment policy strategy.

1.2 Study Design

This open-label, non-randomized, multi-center, Phase 4 PK study to evaluate the systemic exposure to free and adjusted bound aflibercept after bilateral intravitreal administration of high dose (HD) (8 mg) aflibercept in adults with DME or nAMD will be conducted in the European Union.

The study will evaluate up to 45 participants administered bilateral 8 mg aflibercept injections in 3 different treatment regimens. Upon enrollment participants with active central involved DME (CI-DME) or nAMD in at least one eye will be assigned by the study investigator based upon their clinical presentation to one of the 3 defined treatment regimens.

Two populations of patients with DME or nAMD will be eligible for participation in this PK study.

- The first population will be patients who present with one eye with active disease (as defined by inclusion criterion 2) with the fellow eye (without active disease) already under maintenance intravitreal (IVT) anti-VEGF treatment with an approved therapy. These patients will be assigned to Regimen 1.
- The second population will be patients with active disease in both eyes at the time of presentation. These patients will be assigned to Regimen 2 or Regimen 3.

In all treatment regimens the eyes with active disease will receive six 8 mg aflibercept injections per eye, while the fellow eye in Regimen 1 will receive only five 8 mg aflibercept injections during the up to 48-week study treatment period.

The sequence of 8 mg aflibercept IVT administrations to each eye will vary in each assigned treatment regimen as follows:

Treatment Regimen 1 – Initial treatment and maintenance schedule

- 15 participants presenting with one eye with active disease and the fellow eye (without active disease) already under maintenance treatment with an approved IVT anti-VEGF therapy will be assigned to Regimen 1
- In Regimen 1, the eye with active CI-DME or nAMD will receive a series of 3 monthly 8 mg aflibercept injections (Visit 2-Baseline, Visit 7-Week 4 and Visit 8-Week 8) followed by 3 maintenance doses given every 12 weeks after the last monthly injection (at Visit 10-Week 20, Visit 12-Week 32 and Visit 14-Week 44).
- The fellow eye without active disease will have received anti-VEGF treatment before the participant enters the study and within the study will be switched to 8 mg aflibercept. This eye will be treated with 5 maintenance 8 mg aflibercept doses administered every 12 weeks: at Visit 2-Baseline, Visit 9-Week 12, Visit 11-Week 24, Visit 13-Week 36 and Visit 15-Week 48.
- In this treatment regimen, both eyes will be treated on the same day at the baseline visit only.

Treatment Regimen 2 – Bilateral simultaneous initial treatment and maintenance schedule

- Up to 15 participants presenting with bilaterally active disease will be assigned to Regimen 2 and treated with six 8 mg aflibercept injections on the same day in each eye (3 initial monthly injections and then 3 injections every 12 weeks), refer Figure 1-1 for more details.
- Participants assigned to Regimen 2 will receive a series of 3 simultaneous bilateral monthly 8 mg aflibercept injections (Visit 2-Baseline, Visit 4-Week 4 and Visit 5-Week 8), followed by 3 bilateral maintenance injections given at 12-week intervals (Visit 11-Week 20, Visit 12-Week 32 and Visit 13-Week 44).
- Both eyes will be treated with HD aflibercept on the same day at each study treatment visit.

Treatment Regimen 3 – Bilateral staggered initial treatment and maintenance schedule

- Up to 15 participants presenting with bilaterally active disease may be assigned to Regimen 3.
- Participants in Regimen 3 will be treated with six bilateral 8 mg aflibercept injections (3 initial monthly injections and then 3 injections every 12 weeks) in each eye. administered 1 week apart, refer Figure 1-2 for more details.
- In Regimen 3 both eyes will receive a series of 3 bilateral monthly 8 mg aflibercept injections followed by 3 bilateral maintenance injections given at 12-week intervals.
    - The first eye will be treated with 3 monthly HD aflibercept at Visit 2-Baseline, Visit 5-Week 4, Visit 7-Week 8 followed by 3 maintenance injections given at 12-week intervals at Visit 14-Week 20, Visit 16-Week 32 and Visit 18-Week 44.
    - The second eye, to be dosed one week after the first eye, will receive 3 monthly 8 mg aflibercept injections on Visit 3-Week 1, Visit 6-Week 5, Visit 8-Week 9 followed by 3 maintenance injections given at 12-week intervals at Visit 15-Week 21, Visit 17-Week 33 and Visit 19-Week 45.

During the up to 48-week treatment period, participants assigned to each regimen will undergo a battery of ocular and safety assessments as well as immunogenicity and PK sampling corresponding to their assigned treatment regimen to evaluate the systemic exposure after bilateral 8 mg aflibercept treatment. The 8 mg aflibercept dosing, safety and efficacy assessments and PK sampling schedules will vary between the 3 regimens.

Participants assigned to Regimen 1 not completing treatments through Visit 8 will be replaced. However, it may not be possible to replace participants assigned to Regimen 2 or 3 prematurely discontinued from the study intervention or the study as participants with bilaterally active DME or nAMD are infrequently seen in routine clinical practice.

An overview of the study design and dosing schedule is presented in Figure 1-1 and Figure 1-2.

Figure 1-1: Study Flow Diagram

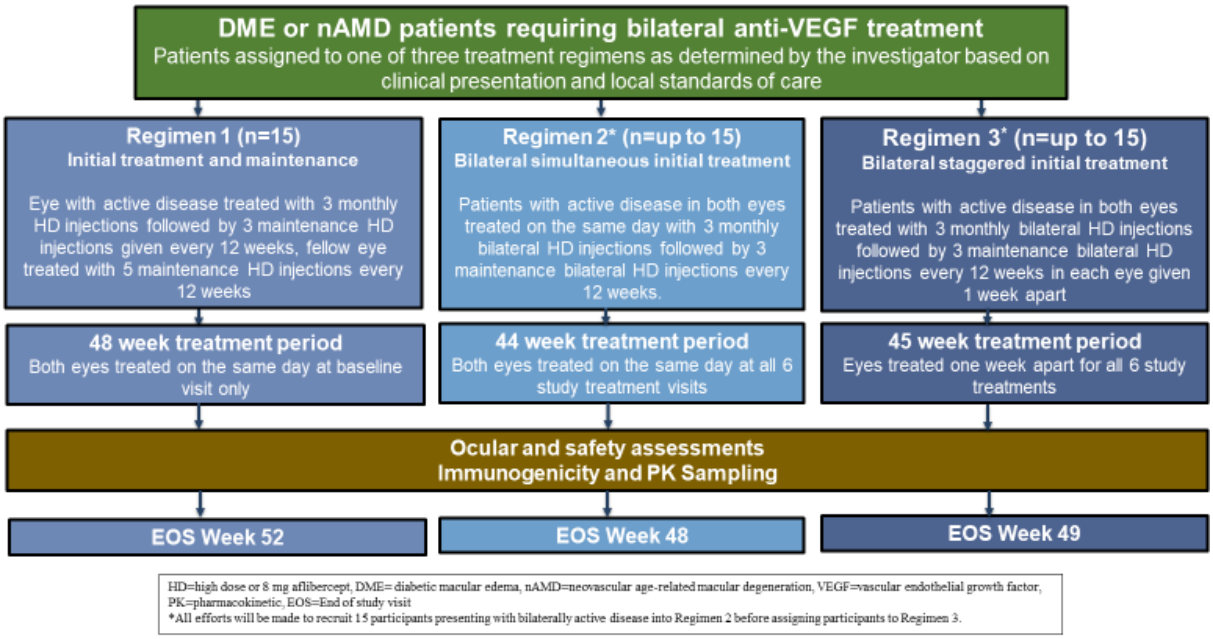


Figure 1-2: Dosing Schedule

a. REGIMEN 1 Dosing Schedule

Visit	2^	3 – 6+	7	8	9	10	11	12	13	14	15	16#
Week	0	0 – 2	4	8	12	20	24	32	36	44	48	52
1st eye~	X	-	X	X	-	X	-	X	-	X	-	-
2nd eye~	X	-	-	-	X	-	X	-	X	-	X	-

b. REGIMEN 2 Dosing Schedule (both eyes treated on the same day)*

Visit	2	3+	4	5	6 – 10+	11	12	13	14#
Week	0	0	4	8	8 – 12	20	32	44	48
1st eye"	X	-	X	X	-	X	X	X	-
2nd eye"	X	-	X	X	-	X	X	X	-

c. REGIMEN 3 Dosing Schedule (eyes treated one week apart)*

Visit	2	3	4+	5	6	7	8	9 – 13+	14	15	16	17	18	19	20#
Week	0	1	1	4	5	8	9	9 – 12	20	21	32	33	44	45	49
1st eye"	X	-	-	X	-	X	-	-	X	-	X	-	X	-	-
2nd eye"	-	X	-	-	X	-	X	-	-	X	-	X	-	X	-

Visit 1 is screening visit in all Regimens, ^Both eyes treated on same day at baseline visit only, +PK sampling visits only, #End of study visit
~In Regimen 1, the 1st eye will be the eye with active disease and the 2nd eye will be the fellow eye with non -active disease,
"In Regimens 2 and 3, the 1st eye will be the right eye and the 2 nd eye will be the left eye
*All efforts will be made to recruit 15 participants presenting with bilaterally active disease into Regimen 2 before assigning participants to Regimen 3.

2. Statistical Hypothesis

No statistical hypotheses are defined for this study.

2.1 Multiplicity Adjustment

Not applicable.

3. Analysis Sets

The following analysis sets are defined:

Table 3-1: Analysis Sets

Participant Analysis Set	**Description**
Full analysis set (FAS)	All participants who received at least one dose of study intervention in each eye.
Safety analysis set (SAF)	All participants who received at least one dose of study intervention in each eye.
Safety analysis set 2 (SAF2)	All participants who received any dose of study intervention (i.e. including participants who received study intervention in one eye only).
Pharmacokinetic analysis set (PKS)	All participants who received at least one dose of study intervention in each eye and who had at least one non-missing PK result after the first dose of study intervention.
Modified PKS (mPKS)	All participants in the PKS who additionally fulfilled: • for treatment Regimen 1: who did not discontinue and did not receive an additional aflibercept injection not planned as per protocol prior to Visit 5 and who had no missing or invalid PK results for Visits 2-5. • for treatment Regimen 2: who did not discontinue and did not receive an additional aflibercept injection not planned as per protocol prior to Visit 8 and who had no missing or invalid PK results for Visits 5-8. • for treatment Regimen 3: who did not discontinue and did not receive an additional aflibercept not planned as per protocol prior to Visit 11 and who had no missing or invalid PK results for Visits 8-11.
Immunogenicity analysis set 1 (ADA)	All participants who received at least one dose of study intervention in each eye and who had at least one non-missing result in the ADA assay after the first dose of study intervention.
Immunogenicity analysis set 2 (NAb)	All participants who received at least one dose of study intervention in each eye, were included in the ADA analysis set and tested negative at all ADA sampling times or tested positive at one or more post-dose ADA sampling times or had at least 1 non-missing post-dose NAb result (either imputed or analysis result).

The FAS is used to analyze endpoints related to efficacy, the SAF and SAF2 are used to analyze the endpoints related to safety, the PKS and mPKS are used to analyze endpoints related to PK, as appropriate, and ADA and NAb are used to analyze the endpoints related to immunogenicity.

Prior to database lock, all PK samples collected outside the sample window defined in the protocol will be assessed for validity of the sampling timepoint and potential exclusion from the statistical analysis. Not all violations of samples taken out of window will substantially influence the PK analysis and thus not all these violations might be excluded from the statistical analysis. The decision which of these violations have a substantial impact on the planned PK analysis and will be excluded from the statistical analysis will be taken case by case with the help of the Bayer PK expert and documented in a report prior to database lock.

Prior to database lock, all vital signs measurements taken outside the time window defined in the protocol section 1.3 will be assessed for validity of the measurement timepoint and potential exclusion from the statistical analysis (considering them as missing data).

Final decision regarding the assignment of participants to analysis sets will be made during the review of study data and documented in the final list of important deviations, validity findings and assignment to analysis sets.

An assessment of PK value outliers will be performed post database lock and might lead to exclusion of single PK values. This decision will be taken with the help of the Bayer PK Expert and documented accordingly.

4. Statistical Analyses

4.1 General Considerations

Imputation of missing data is not planned. Unless specified otherwise, all available data will be used for statistical analysis.

All planned statistical analysis will be exploratory. A confirmatory statistical analysis is not intended.

Baseline is defined as the last available assessment prior to administration of the first dose of study intervention.

For systolic and diastolic blood pressure the baseline value is defined as the average of all measurements prior to administration of the first dose of study intervention (for participants who failed the initial screening, measurements taken at the initial screening visit will not be included).

If there are any values below limit of quantification (BLOQ) they will be substituted by ½ of the lower limit of quantification (LLOQ) for the calculation of geometric statistics and 0 for arithmetic statistics. The rule of substitution of ½ of LLOQ also applies for the calculation of C_{max}. The geometric statistics will only be calculated if 2/3 of values are above LLOQ.

Summary of each intercurrent event will be provided by treatment regimen. Handling of each intercurrent event is described in detail in Section 4.2.1, Table 4-1.

Participants may discontinue prematurely from study intervention. At the same time or later participants may discontinue prematurely from study. At the time of premature discontinuation from the study/intervention, if possible, an early discontinuation or EOS visit should be conducted, as shown in the schedule of activities in protocol (Section 1.3, Table 1–1 for participants assigned to Regimen 1, Table 1–2 for participants assigned to Regimen 2 and Table 1–3 for participants assigned to Regimen 3) for data to be collected at the time of premature study discontinuation.

Any measurements taken at unscheduled visits will not be included in any summary tables in general. If more than one measurement of a variable is taken at an unscheduled visit, all measurements will be shown in listings (if applicable).

If EOS/EOT visit is performed within visit window of a regular study visit (as specified in the "Schedule of Activities" in the protocol), then re-mapping to regular study visit should be performed, unless a regular study visit was already performed for the respective visit window. Visit-based information recorded in the EOS/EOT visit folder which cannot be re-mapped to the regular study visit will be mapped to "EOT Visit" and handled like unscheduled assessments, described in the section above.

Rules on determining treatment-emergent adverse events and prior/concomitant medications in case of partial/missing dates are specified in Section 6.7.

The definition of the 1st eye and 2nd eye depends on the treatment regimen and is described in Figure 1-2. All summaries presented for each eye will summarize the 1st eye and 2nd eye separately.

The statistical evaluation will be performed by using the software SAS© (release 9.4 or higher; SAS Institute Inc., Cary, NC, USA). All variables will be analyzed by descriptive statistical methods. The number of data available and missing data, mean, standard deviation, minimum, quartiles, median, and maximum will be calculated for metric data. Frequency tables will be generated for categorical data.

In participant data listing data will be presented as they are recorded on the Case Report Form (CRF), i.e., partially missing data will appear as such.

4.2 Pharmacokinetics

4.2.1 Primary Endpoint/Estimand Analysis

4.2.1.1 Definition of Endpoint

The primary endpoint is the maximum observed concentration (C_{max}) of free aflibercept.

Maximum observed concentration (C_{max}) of free aflibercept for each participant will be derived by statistical programmer based on PK concentration data collected at below mentioned timepoints.

- Treatment regimen 1: Visit 2 (Day 1), Visit 3 (Day 2), Visit 4 (Day 4), Visit 5 (Day 8), Visit 6 (Day 15), Visit 7 (Day 29). More details on visit window can be found in Table 6-1 in Section 6.6.
- Treatment regimen 2: Visit 5 (Day 57), Visit 6 (Day 58), Visit 7 (Day 60), Visit 8 (Day 64), Visit 9 (Day 71), Visit 10 (Day 85). More details on visit window can be found in Table 6-2 in Section 6.6.
- Treatment regimen 3: Visit 8 (Day 64), Visit 9 (Day 65), Visit 10 (Day 67), Visit 11 (Day 71), Visit 12 (Day 78), Visit 13 (Day 92). More details on visit window can be found in Table 6-3 in Section 6.6.

As described in Table 4-1 PK concentrations after premature treatment discontinuation after the dense PK sampling period will not be used for the calculation of C_{max} of free aflibercept. The corresponding time points for each treatment regimen are described in Table 4-1. For this purpose, the time window from the last injection up to the next expected injection is considered as still on-treatment. Furthermore, participants with premature treatment discontinuation prior to the end of dense PK sampling period will be completely removed from the calculation of

C_{max}. Also, PK concentrations will be excluded from the calculation of C_{max} as described in Table 4-1, if additional aflibercept injections were administered that were not planned as per protocol.

Table 4-1: Strategies for handling ICE occurrence for PK endpoints concerning the maximum observed concentrations

Intercurrent Event	Primary Estimand Strategy	Primary Estimand Analysis
Premature treatment discontinuation prior to end of dense PK sampling	Hypothetical	Exclude all PK concentration of a participant from analysis if the participant is assigned to: • treatment regimen 1 and discontinues treatment prior to Visit 5 (Week 1; Day 8). • treatment regimen 2 and discontinues treatment prior to Visit 8 (Week 9; Day 64). • treatment regimen 3 and discontinues treatment prior to Visit 11 (Week 10; Day 71).
Premature treatment discontinuation after end of dense PK sampling	While-on-treatment	After premature treatment discontinuation, no further injections of study drug take place, but PK samples might still be taken. Therefore: If a participant discontinues treatment prematurely • at Visit 5 (Week 1; Day 8) or later (treatment regimen 1) • at Visit 8 (Week 9; Day 64) or later (treatment regimen 2) • at Visit 11 (Week 10; Day 71) or later (treatment regimen 3) then exclude PK concentrations obtained after the on-treatment time window. Valid PK concentrations obtained in the time window from the last injection up to the next expected injection is still considered to be on-treatment, i.e. data will be censored/excluded only after the date when the next injection would be expected (+/-7 days depending on regimen and visit).
Additional aflibercept injection not planned as per protocol (e.g. any amount of 2mg or 8mg concentrations)	Hypothetical	Exclude all PK concentration of a participant from analysis if the participant is assigned to: • treatment regimen 1 and received additional aflibercept injection prior to Visit 5 (Week 1; Day 8).

Intercurrent Event	Primary Estimand Strategy	Primary Estimand Analysis
prior to end of dense PK sampling		• treatment regimen 2 and received additional aflibercept injection prior to Visit 8 (Week 9; Day 64). • treatment regimen 3 and received additional aflibercept injection prior to Visit 11 (Week 10; Day 71).
Additional aflibercept injection not planned as per protocol (e.g. any amount of 2mg or 8mg concentrations) after end of dense PK sampling	Hypothetical	If a participant received an additional aflibercept injection • at Visit 5 (Week 1; Day 8) or later (treatment regimen 1) • at Visit 8 (Week 9; Day 64) or later (treatment regimen 2) • at Visit 11 (Week 10; Day 71) or later (treatment regimen 3) then exclude PK concentrations obtained after the additional injection.
Use of prohibited medication	Treatment policy	No observations of PK concentrations will be removed from analysis after start of prohibited medication.
Missed study intervention	Treatment policy	No observations of PK concentrations will be removed from analysis after a missed study intervention.

4.2.1.2 Main Analytical Approach

The primary endpoint will be summarized using descriptive statistics by treatment regimen using the mPKS. The descriptive statistical assessment will include the geometric mean and the geometric coefficient of variation. No formal statistical hypothesis test will be performed.

Figures will present individual and arithmetic mean free aflibercept concentrations over all sampling time points by treatment regimen.

4.2.1.3 Sensitivity Analysis

Not applicable.

4.2.1.4 Supplementary Analysis

Not applicable.

4.2.2 Exploratory PK Endpoints/Estimands Analysis

The below mentioned exploratory PK endpoints will be analyzed using the mPKS by considering the estimand approach as defined in Table 4-1: Strategies for handling ICE occurrence for PK endpoints concerning the maximum and similar to the primary endpoint as described in Section 4.2.1.2.

- Maximum observed concentration (C_{max}) of adjusted bound aflibercept.

- Maximum observed concentration (C_{max}) of total aflibercept.
- Timepoint (t_{max}) where C_{max} of free aflibercept was observed.
- Timepoint (t_{max}) where C_{max} of adjusted bound aflibercept was observed.

The following exploratory PK endpoint will be analyzed using the mPKS and the analysis will be similar to the primary endpoint as described in Section 4.2.1.2. However, it will not consider the estimand approach defined in Table 4-1 which is tailored to estimating maximum values instead of values by timepoint.

- In Regimen 2: Observed concentration (C_t) of free aflibercept at Weeks 4, 8, 20, 32 and 44.

Individual concentrations of adjusted bound aflibercept will be calculated as:

0.717 x individual concentrations of bound aflibercept.

Individual concentrations of total aflibercept will be calculated as:

the sum of individual concentrations of free and adjusted bound aflibercept.

The C_{max} of adjusted bound aflibercept and total aflibercept and t_{max} of adjusted bound aflibercept will be derived for each participant by statistical programmer based on data collected at below mentioned timepoints.

- Treatment regimen 1: Visit 2 (Day 1), Visit 3 (Day 2), Visit 4 (Day 4), Visit 5 (Day 8), Visit 6 (Day 15), Visit 7 (Day 29), Visit 8 (Day 57), Visit 9 (Day 85), Visit 10 (Day 141), Visit 11 (Day 169), Visit 14 (Day 309), Visit 15 (Day 337), and Visit 16 (Day 365). More details on visit window can be found in Table 6-1 in Section 6.6.
- Treatment regimen 2: Visit 2 (Day 1), Visit 3 (Day 4), Visit 4 (Day 29), Visit 5 (Day 57), Visit 6 (Day 58), Visit 7 (Day 60), Visit 8 (Day 64), Visit 9 (Day 71), Visit 10 (Day 85), Visit 11 (Day 141), Visit 12 (Day 225), Visit 13 (Day 309), and Visit 14 (Day 337). More details on visit window can be found in Table 6-2 in Section 6.6.
- Treatment regimen 3: Visit 2 (Day 1), Visit 3 (Day 8), Visit 4 (Day 11), Visit 5 (Day 29), Visit 6 (Day 36), Visit 7 (Day 57), Visit 8 (Day 64), Visit 9 (Day 65), Visit 10 (Day 67), Visit 11 (Day 71), Visit 12 (Day 78), Visit 13 (Day 92), Visit 14 (Day 141), Visit 15 (Day 148), Visit 18 (Day 309), Visit 19 (Day 316) and Visit 20 (Day 343). More details on visit window can be found in Table 6-3 in Section 6.6.

No summary statistics for the maximum observed concentration of total aflibercept will be provided.

Figures will present individual and geometric mean adjusted bound aflibercept concentrations by timepoint.

4.2.3 Other Exploratory Endpoints Analysis

Below graphical summaries will be provided to explore relationships between systolic and diastolic BP with drug concentration for the mPKS.

- Figures for relationship between the absolute change in systolic blood pressure (SBP) from baseline and the drug concentration of free and adjusted bound aflibercept over all timepoints for each regimen and over all dosing regimens.

- Figures for relationship between the absolute change in diastolic blood pressure (DBP) from baseline and the drug concentration of free and adjusted bound aflibercept over all timepoints for each regimen and over all dosing regimens.
- Figures for the maximum SBP change per participant and corresponding concentration of free and adjusted bound aflibercept over all timepoints for each regimen and over all dosing regimens.
- Figures for the maximum DBP change per participant and corresponding concentration of free and adjusted bound aflibercept over all timepoints for each regimen and over all dosing regimens.

The following figures will be presented using the Safety Analysis Set for each regimen (SAF):

- Spaghetti plot (time-proportional scaling of x-axis) for individual change from baseline for SBP and DBP over time per regimen.
- Arithmetic mean/standard deviation (SD) for change from baseline for SBP and DBP (time-proportional scaling of x-axis).
- Arithmetic mean/standard deviation (SD) for absolute SBP and DBP (time-proportional scaling of x-axis).

4.3 Efficacy Analysis

The following exploratory efficacy endpoints will be analyzed by indication (AMD and DME) by treatment regimen based on the FAS using descriptive statistics:

- Change from baseline in BCVA at the end of the study per eye.
- Change from baseline in CRT at the end of the study per eye.

End of the study corresponds to Week 52 for treatment Regimen 1, Week 48 for treatment Regimen 2 and Week 49 for Regimen 3.

Due to the limited sample size per indication per treatment regimen and due to potential differences in disease activity per geographical regions, the results must be interpreted with caution.

4.4 Safety Analysis

All safety variables will be summarized by treatment regimen based on the SAF. The SAF2 will be used only in the case of patients who received treatment only in one eye and for these patients only a SAE listing (if applicable) will be provided.

4.4.1 Extent of Exposure

Exposure and compliance parameters will be analyzed per treatment regimen using the SAF.

Treatment duration [weeks] for each participant is defined as the number of weeks from first day of study drug administration till the last day of the study drug administration divided by 7, i.e. [end date – start date + 1] / 7.

The average dose will be calculated as follows:

- The cumulative dose [mg] is the total amount of dose given to a participant over time.
- The average dose [mg/week] is the cumulative dose divided by treatment duration.

Compliance (%) for each patient will be calculated as [the cumulative dose received up to end of treatment divided by cumulative dose up to end of treatment planned as per protocol] * 100 for each participant.

4.4.2 Adverse Events

All AEs will be coded using the latest version of Medical Dictionary for Regulatory Activities (MedDRA). AEs will be summarized by the SOCs and PTs by treatment regimen based on the SAF. AEs within each SOC will be displayed in alphabetical order.

Pre-treatment AEs are defined as AEs that started after the participant has signed the informed consent, but prior to administration of first dose of the study intervention (Visit 2, date and time of the participant's first dose of study intervention).

TEAEs are defined as AEs that occurred on or after the administration of first dose of the study intervention to the last injection plus 30 days.

Post-treatment AEs are defined as AEs that started more than 30 days after the last injection of study drug in the study.

An overview of AEs will be provided along with the following summary tables by treatment regimen:

- TEAEs: Ocular (any eye), Non-ocular
- Serious TEAEs: Ocular (any eye), Non-ocular
- Drug related TEAEs: Ocular (any eye), Non-ocular
- AEs leading to discontinuation
- Deaths

A frequency table of adjudicated treatment-emergent Anti-Platelet Trialists' Collaboration (APTC) events terms, cross-tabulated with related MedDRA PT and SOC, will also be displayed by treatment arms. The adjudication of AE is described in the "APTC adjudication committee charter".

Additionally, a frequency table of TEAEs of **hypertension** terms, cross-tabulated with related MedDRA PT and SOC, will be displayed by treatment arm (see Section 6.8 for the definition of the term).

A listing for SAEs will be provided.

4.4.3 Additional Safety Assessments

Additional safety parameters will be analyzed per treatment regimen using the SAF.

4.4.3.1 Vital Signs

Descriptive summary tables will be provided by visit for absolute values along with change from baseline for the following vital signs parameters.

- Systolic and diastolic blood pressure (mmHg)
- Heart rate (beats/min)
- Body temperature (^{o}C)

Additionally, summaries will be provided for participants with at least one systolic/diastolic blood pressure treatment-emergent PCSV (by visit and at any visit) of

- ≤ 95 mmHg and decrease from baseline ≥ 20 mmHg, in systolic blood pressure.
- ≥160 mmHg and increase from baseline ≥ 20 mmHg, in systolic blood pressure.
- ≤ 45 mmHg and decrease from baseline ≥ 10 mmHg, in diastolic blood pressure.
- ≥110 mmHg and increase from baseline ≥ 10 mmHg, in diastolic blood pressure.

4.5 Analysis of Immunogenicity Data

The following exploratory immunogenicity endpoints will be analyzed using frequency tables based on NAb analysis set:

- Incidence of treatment-emergent NAb: defined as a positive result in the NAb assay post-dose, when baseline results are negative or missing

The following exploratory immunogenicity endpoints will be analyzed using frequency tables based on ADA analysis set:

- Incidence of pre-existing immunoreactivity: either a positive response in the ADA assay at baseline with all post first dose ADA results negative OR a positive response at baseline with all post first dose ADA responses less than 4-fold of baseline titer levels.
- Incidence of treatment-emergent ADA: defined as a positive result in the ADA assay post-dose, when baseline results are negative or missing.
- Incidence of treatment-boosted ADA: defined as a positive result in the ADA assay post-dose, that is greater than or equal to 4-fold over baseline titer levels, when baseline results are positive.
- Incidence of ADA Negative: negative response in the ADA assay at all time points, regardless of any missing samples.
- Incidence of missing ADA samples.

The number and percentages of patients who develop treatment-emergent or treatment-boosted ADA and neutralizing antibodies (NAb) status will be provided by treatment regimen and visit. The immunogenicity assessments will be summarized based on participants included in the ADA Analysis Set. The maximal ADA titers will be shown in categories for participants by visit and over the entire study:

- Low (titer < 1000)
- Moderate (1000 ≤ titer ≤ 10000)
- High (titer > 10000)

The influence of ADAs and NAbs on individual concentration-time profiles (free aflibercept) will be evaluated based on PK analysis set as follows:

Individual concentrations of free aflibercept in plasma by treatment-emergent and treatment-boosted aflibercept antibody status and treatment regimen.

4.5.1 Other Variables and/or Parameters

Not applicable.

4.5.2 Subgroup Analyses

Due to the limited number of participants in this study, no subgroup analyses will be performed.

4.6 Interim Analyses

No formal interim analysis is planned.

4.7 Changes to Protocol-planned Analyses

- mPKS:

CSP, section 9.2	SAP, section 3	Reason
All participants in the PKS who additionally fulfilled: • for treatment Regimen 1: who did not discontinue prior to Visit 5 and who had no missing PK results for Visits 2-5. • for treatment Regimen 2: who did not discontinue prior to Visit 8 and who had no missing PK results for Visits 5-8. • for treatment Regimen 3: who did not discontinue prior to Visit 11 and who had no missing PK results for Visits 8-11.	All participants in the PKS who additionally fulfilled: • for treatment Regimen 1: who did not discontinue **and did not receive an additional aflibercept injection not planned as per protocol** prior to Visit 5 and who had no missing **or invalid** PK results for Visits 2-5. • for treatment Regimen 2: who did not discontinue **and did not receive an additional aflibercept injection not planned as per protocol** prior to Visit 8 and who had no missing **or invalid** PK results for Visits 5-8. • for treatment Regimen 3: who did not discontinue **and did not receive an additional aflibercept injection not**	To align with the updated SAP version, i.e. removing participants from the analysis set, if additional aflibercept injections were given as this will influence the PK analysis. To clarify, that participants with invalid PK results as described in the SAP will be handled the same way as missing PK results.

	planned as per protocol prior to Visit 11 and who had no missing **or invalid** PK results for Visits 8-11.	

5. Sample Size Determination

No formal statistical sample size estimation has been performed for this study.

The selected number of participants to be included refers to the PK objective of the study. In previous studies of 8 mg aflibercept (CANDELA phase 2, PHOTON phase 2/3 and PULSAR phase 3) a dense PK cohort aimed to include between 8 and 15 participants per dose group. Based on the PK and population PK results generated from those studies including measures of variability, at least 8 participants were deemed sufficient to characterize systemic exposures for the most frequent scenarios of expected bilateral 8 mg aflibercept treatment to be implemented in routine clinical practice.

Up to 45 participants with DME or nAMD requiring anti-VEGF treatment in both eyes will be recruited and assigned to receive treatment with bilateral HD aflibercept according to one of three treatment regimens in the planned study. The sample size is expected to provide an adequate understanding of the systemic exposure resulting from bilateral 8 mg aflibercept treatment.

The study will remain open for recruitment until 15 participants are assigned to Regimen 1 and all efforts will be made to recruit 15 participants with bilaterally active disease to Regimen 2 before assigning participants to Regimen 3. No minimum number of participants is pre-defined for Regimen 3.

6. Supporting Documentation

6.1 Appendix 1: Participant Disposition

The number of participants enrolled in the study, assigned to study intervention and valid for the FAS, SAF, SAF2, PKS, mPKS, ADA and NAb sets will be summarized for by treatment regimen and overall and by study site using FAS.

The number of participants enrolled and assigned to study intervention will further be summarized by country/region and by treatment regimen and overall.

The number of sites will be summarized by country/region.

The number of participants with important protocol deviations and the number of screen failures will be summarized overall and by treatment regimen.

The number of participants per important deviation category and validity finding will be presented by treatment regimen.

A disposition summary for each study period (screening, treatment) will be presented summarizing the number of participants starting and completing the respective study period, the number of participants discontinuing it and the primary reason for discontinuation. This will be presented overall and for each treatment regimen. The number of participants completing the overall study will be provided by study regimen and overall.

6.2 Appendix 2: Demographic and Other Baseline Characteristics

Demographic and other baseline characteristics will be summarized based on FAS.

The following baseline demographic data will be summarized:

- Age (years; as entered in eCRF)
- Age categorized (<55 years, ≥ 55 to < 75 years, ≥ 75 years)
- Sex (Male vs Female)
- Race
- Height (cm)
- Weight (kg)
- Body Mass Index (BMI) (kg/m^2)
- BMI categorized (≤ 25 kg/m², 25 kg/m² $< BMI \leq 30$ kg/m², 30 kg/m² $< BMI \leq 35$ kg/m², $BMI > 35$ kg/m²)

The following additional baseline disease characteristics will be summarized:

- Systolic blood pressure (mmHg)
- Diastolic blood pressure (mmHg)
- Heart rate (bpm)
- Hypertension (yes, no)
- BCVA (ETDRS letters score)
- Intraocular pressure (IOP in mmHg)

- CRT

6.3 Appendix 3: Protocol Deviations

Prior to database lock, final decision regarding the assignment of participants to analysis sets as well as the exclusion of PK samples (as described in section 3) and vital signs measures (as described in section 3) will be made during the review of study data and documented in the final list of important deviations, validity findings and assignment to analysis sets.

After receipt of the final PK data, these will also be assessed for validity and implausible outliers might be excluded and documented in a report.

A summary table of the number and percentage of participants with any important protocol deviation by type of deviation will be provided using the SAF.

A by-participant listing of important protocol deviations will be provided.

6.4 Appendix 4: Medical History

Medical history will be coded using the latest version of MedDRA coding system and will be summarized by SOC and PT based on SAF.

6.5 Appendix 5: Prior and Concomitant Medication and Procedures

All medications will be coded using the latest version of the World Health Organization Drug Global reference available at time of database release for analysis and will be classified by Anatomical Therapeutic Chemical (ATC) categories. Procedures will be coded using the latest version of MedDRA coding system.

For prior and concomitant medications, the following definitions apply:

- Prior medication: Medication taken before start of the study intervention intake, (regardless of when it ended).
- Concomitant medication: Medication taken during treatment period, i.e., between first and last study intervention intake (regardless of when it started or ended).

If a prior or concomitant medication cannot be determined (due to missing or partially missing stop date), Please refer to Section 6.7 on how prior or concomitant medication will be determined.

Prior and Concomitant medications will be summarized separately by ATC classification level 4 and PT based on SAF.

By participant listings for procedures will be provided.

6.5.1 Prohibited Medications and Procedures

Ocular (both eyes):

Participants are not allowed to receive any standard or investigational treatment for DME, DR or nAMD other than their assigned study treatment with bilateral 8 mg aflibercept treatment, until they have completed the EOS/early discontinuation visit assessments. This includes medications administered locally (e.g., IVT, topical, juxta scleral, or periorbital routes) or systemically with the intent of treating DME, DR or nAMD.

Non-ocular (systemic):

Non-ocular (systemic) standard or investigational treatments for DME, DR or nAMD are not permitted.

6.6 Appendix 6: Schedule of Assessment for PK Samples

Study assessments and procedures are presented by regimen. The timing and intervals between visits, the dosing schedule of the intervention and the timing of the PK sampling varies in the 3 treatment regimens.

The dense PK sample periods are defined as follows:

- Regimen 1: Visit 2 to Visit 5 (Week 1; Day 8)
- Regimen 2: Visit 5 to Visit 8 (Week 9; Day 64)
- Regimen 3: Visit 8 to Visit 11 (Week 10; Day 71)

Table 6-1: PK Sampling Schedule Regimen 1

Initial Treatment and Maintenance Dosing Group		Visit	Week	Assessment Day	PK Sample	Dense PK Sampling Period	HR, BP[1] Measurement
1st Eye	2nd Eye						
		1	Screening	-21 to -1			
X	X	2	0	1	X[2]	X	X[1]
		3	During Week 0	2	X[3]	X	X[1]
		4	During Week 0	4	X[4]	X	X[1]
		5	1	8	X[5]	X	X[1]
		6	2	15	X[6]		X[1]
X		7	4	29	X[2,7]		X[1]
X		8	8	57	X[2]		X[1]
	X	9	12	85	X[2]		X[1]
X		10	20	141	X[2]		X[1]
	X	11	24	169	X[2]		X[1]
X		12	32	225			
	X	13	36	253			
X		14	44	309	X[2]		X[1]
	X	15	48	337	X[2]		X[1]
		16	52	365	X		X[1]

1. Heart rate (HR) and blood pressure (BP) measurements must be taken before the pharmacokinetic (PK) blood sample is collected and must be within 2 hours of the clock time of dosing on Day 1. BP will be obtained, using automated office blood pressure (AOBP) measurement (e.g Omron Model HEM 907XL or comparable). BP measurements displayed by the device will be recorded in the electronic data capture (EDC). Detailed instructions can be found in the study reference manual.

2. On dosing visits, PK sampling should be performed prior to the administration of study drug and within ±2 hours of the clock time of dosing on Day 1.

3. PK sample must be collected 24 ± 2 hours after dosing on Day 1/Visit 2.

4. PK sample should be collected 3 days after dosing on Day 1/Visit 2. If unavoidable, the visit can be shifted by ± 1 day, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 1.

5. PK sample should be collected 7 days after dosing on Day 1/Visit 2. If unavoidable, the visit can be shifted by ± 1 day, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 1.

6. PK sample should be collected 14 days after dosing on Day 1/Visit 2. If unavoidable, the visit can be shifted by ± 1 day, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 1.

7. PK sample should be collected 28 days after dosing on Day 1/Visit 2. If unavoidable, the visit can be shifted by ± 2 day, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 1.

Table 6-2: PK Sampling Schedule Regimen 2

Bilateral Simultaneous Initial Treatment Dosing Group		Visit	Week	Assessment Day	PK Sample	Dense PK Sampling Period	HR, BP[1] Measurement
1st Eye	2nd Eye						
		1	Screening	-20 to -1			
X	X	2	0	1	X[2]		X[1]
		3	During Week 0	4	X[3]		X[1]
X	X	4	4	29	X[2]		X[1]
X	X	5	8	57	X[2]	X	X[1]
		6	8 + 1 day	58	X[4]	X	X[1]
		7	8+ 3 days	60	X[5]	X	X[1]
		8	9	64	X[6]	X	X[1]
		9	10	71	X[7]		X[1]
		10	12	85	X[8]		X[1]
X	X	11	20	141	X[2]		X[1]
X	X	12	32	225	X[2]		X[1]
X	X	13	44	309	X[2]		X[1]
		14	48	337	X		X[1]

1. Heart rate (HR) and blood pressure (BP) measurements must be taken before the pharmacokinetic (PK) blood sample is collected and must be within 2 hours of the clock time of dosing on Day 1. BP will be obtained, using automated office blood pressure (AOBP) (e.g Omron Model HEM 907XL or comparable). BP measurements displayed by the device will be recorded in the electronic data capture (EDC). Detailed instructions can be found in the study reference manual.
2. On dosing visits, PK sampling should be performed prior to the administration of study drug and within ±2 hours of the clock time of dosing on Day 1.
3. PK sample should be collected 3 days after dosing on Day 1/Visit 2. If unavoidable, the visit can be shifted by ± 1 day, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 1.

4. PK sample must be collected 24 ± 2 hours after dosing on Day 57/Visit 5.
5. PK sample should be collected 3 days after dosing on Day 57/Visit 5. If unavoidable, the visit can be shifted by ± 1 day, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 57.
6. PK sample should be collected 7 days after dosing on Day 57/Visit 5. If unavoidable, the visit can be shifted by ± 1 day, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 57.
7. PK sample should be collected 14 days after dosing on Day 57/Visit 5. If unavoidable, the visit can be shifted by ± 1 day, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 57.
8. PK sample should be collected 28 days after dosing on Day 57/Visit 5. If unavoidable, the visit can be shifted by ± 2 days, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 57.

Table 6-3: PK Sampling Schedule Regimen 3

Bilateral Staggered Initial Treatment Dosing Group		**Visit**	**Week**	**Assessment Day**	**PK Sample**	**Dense PK Sampling Period**	**HR, BP[1] Measurement**
1st Eye	**2nd Eye**						
		1	Screening	-20 to -1			X[1]
X		2	0	1	X[2]		X[1]
	X	3	1	8	X[2]		X[1]
		4	During Week 1	11	X[3]		X[1]
X		5	4	29	X[2]		X[1]
	X	6	5	36	X[2]		X[1]
X		7	8	57	X[2]		X[1]
	X	8	9	64	X[2]	X	X[1]
		9	9 + 1 day	65	X[4]	X	X[1]
		10	9 + 3 days	67	X[5]	X	X[1]
		11	10	71	X[6]	X	X[1]
		12	11	78	X[7]		X[1]
		13	12	92	X[8]		X[1]

Bilateral Staggered Initial Treatment Dosing Group		Visit	Week	Assessment Day	PK Sample	Dense PK Sampling Period	HR, BP[1] Measurement
1st Eye	2nd Eye						
X		14	20	141	X[2]		X[1]
	X	15	21	148	X[2]		X[1]
X		16	32	225			
	X	17	33	232			
X		18	44	309	X[2]		X[1]
	X	19	45	316	X[2]		X[1]
		20	49	343	X		X[1]

1. Heart rate (HR) and blood pressure (BP) measurements must be taken before the pharmacokinetic (PK) blood sample is collected and must be within 2 hours of the clock time of dosing on Day 1. BP will be obtained, using automated office blood pressure (AOBP) (e.g. Omron Model HEM 907XL or comparable). BP measurements displayed by the device will be recorded in the electronic data capture (EDC). Detailed instructions can be found in the study reference manual.
2. On dosing visits, PK sampling should be performed prior to the administration of study drug and within ±2 hours of the clock time of dosing on Day 1.
3. PK sample should be collected 3 days after dosing on Day 8/Visit 3. If unavoidable, the visit can be shifted by ± 1 day, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 1.
4. PK sample must be collected 24 ± 2 hours after dosing on Day 64/Visit 8.
5. PK sample should be collected 3 days after dosing on Day 64/Visit 8. If unavoidable, the visit can be shifted by ± 1 day, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 64.
6. PK sample should be collected 7 days after dosing on Day 64/Visit 8. If unavoidable, the visit can be shifted by ± 1 day, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 64.
7. PK sample should be collected 14 days after dosing on Day 64/Visit 8. If unavoidable, the visit can be shifted by ± 1 day, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 64.
8. PK sample should be collected 28 days after dosing on Day 64/Visit8. If unavoidable, the visit can be shifted by ± 2 days, but the clock hour of PK sampling should be kept within ±2 hours of the clock hour of dosing on Day 64.

6.7 Appendix 7: Missing/Incomplete Date Imputation

Imputation of missing endpoint data is not planned. Unless specified otherwise, all available data will be used for statistical analysis.

The following rules will be applied where applicable to ensure that participants/data are not excluded from statistical analysis due to missing or incomplete data:

For Adverse Events (AEs) and prior/concomitant medications, complete start and stop dates must be recorded to determine if the AE occurred during the study intervention period (i.e. treatment-emergent) or the medication was taken during the study intervention period (i.e. concomitant).

AE Start Date	**AE End Date**	**Action**
known	known	Considered as a treatment-emergent adverse event (TEAE) if start date and time is on or after the date and time of the first IVT administration.

Medication Start Date	**Medication End Date**	**Action**
known	known	If start date is prior to the date for the first IVT administration, considered as prior; if start date is on or after the date for the first IVT administration, considered as concomitant.

In case of partial dates, the following rules will be applied:

For partial/missing start dates of medications and AEs, impute as follows to determine whether concomitant and treatment-emergent, respectively:

Start Date	End Date	Action
If only the month and year is available	End date is after the date of the first study intervention	If the month/year of the start date equals the month/year of first study intervention, then impute as the date of first study intervention. If the month/year of the start date is before the month/year of first study intervention, then impute as the last day of that month/year. If the month/year of the start date is after the month/year of first study intervention, then impute as the first day of that month/year.

If only the year is available	End date is after the date of the first study intervention	If the year of the start date equals the year of first study intervention, then impute as the date of first study intervention. If the year of the start date is before the year of first study intervention, then impute as the last day and month of that year. If the year of the start date is after the year of first study intervention, then impute as the first day and month of that year.
Missing	End date is after the date of the first study intervention	Impute as the date of first study intervention

For partial/missing end dates of medications, impute as follows to determine whether concomitant:

Start Date	End Date	Action
Start date is before the date of the last study intervention	If only the month and year is available	If the month/year of the end date equals the month/year of last study intervention, then impute as the date of last study intervention. If the month/year of the end date is before the month/year of last study intervention, then impute as the last day of that month/year. If the month/year of the end date is after the month/year of last study intervention, then impute as the first day of that month/year.
Start date is before the date of the last study intervention	If only the year is available	If the year of the end date equals the year of last study intervention, then impute as the date of last study intervention. If the year of the end date is before the year of last study intervention, then impute as the last day of that year. If the year of the end date is after the year of last study intervention, then impute as the first day of that year.

Start date is before the date of the last study intervention	Missing	impute as the date of last study intervention

Imputed dates will only be used to determine whether medications are concomitant and AEs are treatment-emergent. Original (partial/missing) entries will be retained in listings (if applicable).

6.8 Appendix 8: PTs for Selection of Safety Topics

Table 6-4: PTs for Hypertension

Preferred term (MedDRA version 26.0)
Accelerated hypertension
Blood pressure ambulatory increased
Blood pressure diastolic increased
Blood pressure inadequately controlled
Blood pressure increased
Blood pressure systolic increased
Diastolic hypertension
Endocrine hypertension
Essential hypertension
Hypertension
Hypertension neonatal
Hypertensive angiopathy
Hypertensive cardiomegaly
Hypertensive cardiomyopathy
Hypertensive cerebrovascular disease
Hypertensive crisis
Hypertensive emergency
Hypertensive encephalopathy
Hypertensive end-organ damage
Hypertensive heart disease
Hypertensive nephropathy
Hypertensive urgency
Labile hypertension
Malignant hypertension
Malignant hypertensive heart disease
Malignant renal hypertension
Maternal hypertension affecting foetus
Mean arterial pressure increased
Neurogenic hypertension
Orthostatic hypertension
Page kidney

Preferred term (MedDRA version 26.0)
Prehypertension
Renal hypertension
Renovascular hypertension
Retinopathy hypertensive
Supine hypertension
Systolic hypertension
White coat hypertension

7. References

Not applicable.