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Title page

An open-label, non-randomized, Phase I study to evaluate the effect of copanlisib (a single intravenous dose of 60 mg) on the pharmacokinetics (PK) and pharmacodynamics (PD) of metformin (MATE2-K substrate) in healthy volunteers

Effect of copanlisib on metformin PK and PD

Bayer study drug	BAY 80-6946/Copanlisib		
Study purpose:	To evaluate the effect of copanlisib on the PK and PD of metformin		
Clinical study phase:	I	**Date:**	14 Dec 2018
Study No.:	19951	**Version:**	1.0
Author:	PPD		

This Statistical Analysis Plan is produced on a word-processing system and bears no signatures.

The approval of the Statistical Analysis Plan is documented in a separate Signature Document.

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Abbreviations

$\%A_{E,ur}(t_1\text{-}t_2)$	percentage of dose excreted into urine from t_1 to t_2
$\%AUC(t_{last}\text{-}\infty)$	percentage of AUC from the last data point > LLOQ to infinity
$A_{E,ur}$	Amount excreted into urine
$\%A_{E,ur}$	Percentage amount excreted into urine
$A_{E,ur}(0\text{-}24)$	Amount excreted into urine from 0 to 24 hours
$A_{E,ur}(t_1\text{-}t_2)$	Amount excreted into urine from t_1 to t_2
AE	adverse event
ANOVA	analysis of variance
AUC	area under the concentration vs. time curve from zero to infinity after single (first) dose
AUC(0-24)	AUC from time 0 to 24 hours
$AUC(0\text{-}t_{last})$	AUC from time 0 to the last data point > LLOQ
$AUC(0\text{-}t_{last})_u$	AUC from time 0 to the last data point > LLOQ of unbound drug
$AUC(t_{last}\text{-}\infty)$	AUC from the last data point to infinity
BAY no.	BAY number is the main identifier for compounds within the Bayer Pharmaceuticals Organization
CL/F	total body clearance of drug calculated after extravascular administration (e.g. apparent oral clearance)
CL_R	renal body clearance of drug
C_{max}	maximum observed drug concentration in measured matrix after single dose administration
C_{6h}	Concentration at 6 hours
C_{24h}	Concentration at 24 hours
CV	coefficient of variation
ECG	electrocardiogram
EOT	End of treatment
i.v.	intravenous
kg	kilogram
LLN	Lower limit of norm (upper limit of normal laboratory values)

LLOQ	lower limit of quantitation
MATE	Multidrug and toxin extrusion transporter
MedDRA	medical dictionary for regulatory activities
mg	milligram
min	minute
mL	milliliter
NCA	Non compartmental analysis
PD	pharmacodynamics
PK	pharmacokinetics
PKS	pharmacokinetic analysis set
PPS	Per-protocol set
SAE	serious adverse event
SAF	safety analysis set
SAP	Statistical Analysis Plan
SAS	Statistical Analysis System
$t_{1/2}$	half-life associated with the terminal slope
TEAE	treatment-emergent adverse event
t_{max}	time to reach C_{max} (in case of two identical C_{max} values, the first t_{max} will be used)
ULN	upper limit of norm (upper limit of normal laboratory values)
US(A)	United States (of America)
V_z/F	apparent volume of distribution during terminal phase after extravascular administration
WHO DD	World Health Organization Drug Dictionary

1. Introduction

Copanlisib (BAY 80-6946) was approved via accelerated approval in the US on 14 SEP 2017, as a monotherapy treatment in adult patients with relapsed follicular lymphoma who have received 2 prior systemic therapies. Copanlisib is being further developed for the treatment of advanced and refractory malignancies either as a single agent or in combination with other investigational agents. Copanlisib is the active ingredient (free base) of BAY 84-1236, the dihydrochloride salt, which for clinical use is formulated as an intravenous (i.v.) drug product solution.

This study will investigate the effect of copanlisib, a multidrug and toxin extrusion transporter (MATE)2-K inhibitor, on the pharmacokinetics (PK) and pharmcodynamics (PD) of metformin (substrate drug of MATE2-K) in healthy subjects.

This document is based on the

- Clinical Study Protocol BAY 80-6946 / 19951, Version 1.0, 15 May 2018[1].

This Statistical Analysis Plan (SAP) describes the final analysis of this study. No statistical interim analysis will be performed.

2. Study Objectives

The primary objective of this study is to

- Evaluate the effect of the co-administration of copanlisib on the PK of metformin

The secondary objectives of this study are to

- Assess the safety of copanlisib when administered to healthy subjects in combination with metformin
- Evaluate the PD effect of copanlisib on metformin in terms of plasma lactate levels

Exploratory objectives of this study are to

- Assess additional copanlisib and metformin PK parameters
- Assess additional PD parameters

3. Study Design

This study is a phase 1, non-randomized, single-center, open-label study to investigate the effect of copanlisib, a MATE2-K inhibitor, on the PK and PD of metformin in healthy subjects.

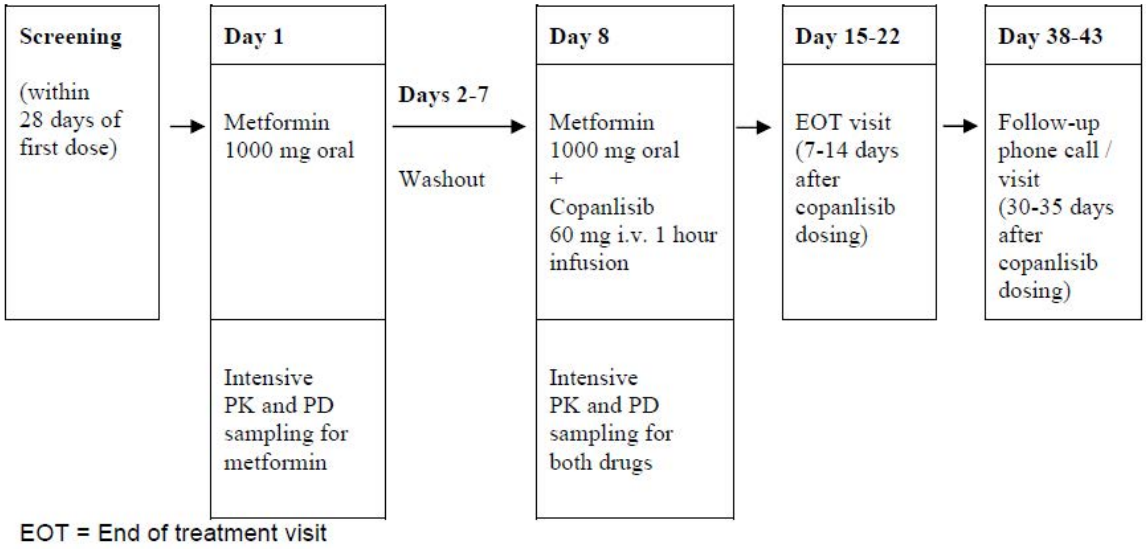
A total of approximately 15 subjects between the ages of 18 to 45 years will be enrolled, to achieve 12 evaluable subjects. All subjects will receive a single dose of metformin 1000 mg on Days 1 and 8 in a fasting state. Subjects will also receive a single i.v. dose of 60 mg copanlisib on Day 8 as part of the combination with metformin. After the first dose of metformin, there will be a 7-day wash-out before the second dose of metformin with copanlisib on Day 8, to assess the effect of copanlisib as a perpetrator on metformin.

Blood samples will be collected pre- and post-dose, and urine samples will be collected up to 24 hours for metformin; blood samples will be collected up to 24 hours for copanlisib. Additional blood samples will be collected up to 24 hours after each dose of metformin for the PD evaluations.

Subject will stay in-house at the study site from approximately 12 hours before until 24 hours after each dose.

An end of treatment (EOT) visit will be conducted 7 to 14 days after copanlisib dosing. Follow-up will be conducted 30 to 35 days after copanlisib dosing.

Figure 3—1: Flow chart of study design



EOT = End of treatment visit

The variables related to each study objective are as below:

Primary variables

The primary variables of this study, to be determined for metformin are:

- C_{max}, $AUC_{(0-24)}$, and AUC on Day 1 and Day 8

Secondary variable

The secondary variables of this study are :

- Frequency/severity of treatment emergent AEs
- PD parameter plasma lactate levels and maximum change from baseline on Day 1 and Day 8

Exploratory variable

The exploratory variables of this study are:

- Additional PK Parameters for Metformin on Day 1 and Day 8 – $\%A_{E,ur}$, $A_{E,ur}$, $\%A_{E,ur(t1-t2)}$, $A_{E,ur(t1-t2)}$ where t_1 and t_2 are the start and the stop times of the urine collection interval. t_{max}, V_Z/F, CL/F and $t_{1/2}$; and CLR (calculated as CLR = $A_{Eur(0-24)}/AUC_{(0-24)}$)
- Additional PK Parameters for Copanlisib on Day 8 – C_{max}, t_{max}, C_{6h}, C_{24h} and $AUC_{(0-24)}$
- Additional PD Parameters – Glucose, Insulin and C-Peptide on Day 1 and Day 8

4. General Statistical Considerations

4.1 General Principles

The statistical evaluation will be performed by using the software package SAS release 9.2 or higher (SAS Institute Inc., Cary, NC, USA).

All data will be listed and analyzed by descriptive statistical methods where appropriate. The number of measurements available, mean, standard deviation, minimum, median, and maximum will be presented for quantitative data. Whenever appropriate, summary statistics will be provided for the original data as well as for the change versus baseline. Graphical illustrations will be provided where appropriate. Frequency tables will be provided for qualitative data.

The analysis and reporting will be based on the Clinical Pharmacology Standards (CLIPS), version 2.0[2], and the Clinical Pharmacology Standard Listings_V1.0_final_09052018, version 1.0[3].

4.2 Handling of Dropouts

A subject who discontinues study participation prematurely for any reason is defined as a "dropout" if the subject has already been administered at least one dose of study medication.

Subjects who drop out or do not receive all doses of study drugs will be replaced to obtain a minimum of 12 subjects valid for PK of metformin.

4.3 Handling of Missing Data

Missing data will not be replaced. All available data will be used for statistical analysis.

All missing or partial data will be presented in the subject data listing as they are recorded.

4.4 Interim Analyses and Data Monitoring

No formal statistical interim analysis is planned.

4.5 Data Rules

Baseline is defined as the last observation prior to first administration of study medication (at Day -1/Day 1).

In case of more than one measurement at screening, the last observation of those will be used for descriptive statistics tables as well as for frequency tables.

If multiple measurements for a planned time point except screening are available, the first value (i.e. of the planned measurement) will be used for the calculation of descriptive statistics.

Measurements at unplanned visits, except for screening, will be listed but not summarized by descriptive statistics.

For laboratory data, values incorporated as '< x' into the database will be substituted by 1/2x, values incorporated as '> x' will be substituted by 'x' itself for calculation of summary statistics.

4.6 Blind Review

The results of the final data assessment will be documented in the final list of important deviations, validity findings and assignment to analysis sets. Any relevant changes to the statistical analysis prompted by the results of the review of study data will be documented in an amendment to this SAP.

Relevant changes to the approved SAP requested after Database Release will be described in the Clinical Study Report.

5. Analysis Sets

5.1 Assignment of analysis sets

Final decisions regarding the assignment of subjects 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 (see Section 4.6).

Safety Analysis Set (SAF)

All subjects who received at least one dose of the study medication will be included in the safety evaluation.

Safety evaluation will be done on the SAF. The PD data will also be analyzed using the Safety Analysis Set.

Pharmacokinetic Analysis Set (PKS)

All subjects with a valid PK profile for metformin will be included in the PKS for analyzing and evaluating the PK profile of metformin.

Per-protocol set (PPS)

All subjects with a valid PK profile for metformin both alone and in combination with copanlisib will be included in the PPS for analyzing and evaluating the effect of copanlisib on the PK profile of metformin.

In case, the number of subjects are identical in the PKS compared to the PPS, then the analysis will be done only on the PPS.

6. Statistical Methodology

6.1 Population Characteristics

6.1.1 Demographic and other baseline characteristics

Demographic and other baseline characteristics will be performed on the SAF.

Demographic data will be summarized using descriptive statistics (arithmetic mean, standard deviation, median, minimum and maximum for quantitative variables). Frequency tables for qualitative data will be provided.

6.1.2 Disposition

Disposition at the end of screening will be summarized for all enrolled subjects. Disposition at the end of treatment and for follow-up will be summarized for all subjects assigned to treatment.

6.1.3 Medical history

Medical history findings will be summarized using the most recent effective version of Medical Dictionary for Regulatory Activities (MedDRA, version 21.0 or higher) terms.

6.1.4 Prior and concomitant medication

The number of subjects that used prior medications (medications that began before the start of the study drug and are ongoing after administration of study drug) and/or concomitant medications (medications that began after the administration of study drug) will be analyzed separately by using frequency tables based on classified data. Concomitant and prior medication data will be classified according to the most recent effective WHO-DD version.

6.1.5 Other baseline characteristics

Data collected on smoking and alcohol consumption will be summarized and listed as appropriate.

6.2 Efficacy

Not applicable

6.3 Pharmacokinetics/pharmacodynamics

6.3.1 Pharmacokinetic Data

Non compartmental analysis (NCA)

The PK parameters will be calculated by the PK scientist/expert group and provided to the Statistical Analysis team for further analysis. Based on the concentration time data, the following calculated PK parameters will be provided:

Main PK Parameters: Metformin:

Day 1 and Day 8 : C_{max}, $AUC_{(0-24)}$ and AUC

Additional parameters: Metformin:

Day 1 and Day 8:

$\%A_E,ur$, A_E,ur, $\%A_E,ur(t_1-t_2)$, $A_E,ur(t_1-t_2)$ where t_1 and t_2 are the start and the stop times of the urine collection interval

t_{max}, V_Z/F, CL/F, $t_{1/2}$, and CLR (calculated as CLR = $A_{E,ur(0-24)}/AUC_{(0-24)}$)

Copanlisib:

Day 8 : C_{max}, t_{max}, C_{6h}, C_{24h} and $AUC_{(0-24)}$

The concentration–time data of all analytes will be tabulated by treatment condition. The following statistics will be calculated for each plasma sampling point: arithmetic mean, standard deviation and coefficient of variation (CV), geometric mean, geometric standard deviation (re-transformed standard deviation of the logarithms) and CV, minimum, median, maximum value and the number of measurements. Means at any time will only be calculated if at least 2/3 of the individual data were measured and were above the lower limit of quantification (LLOQ). For the calculation of the mean value a data point below LLOQ will be substituted by one half of this limit. In tables showing mean values, where values below LLOQ are included in the calculation of mean values, these means will be marked.

Individual and geometric mean concentration versus time profiles of all analytes (by using the actual sampling times for individual plots and the planned sampling times for mean plots) will be plotted on both linear and semi-logarithmic axes. The amount (%) of metformin excreted into urine will be graphically illustrated by treatment condition for each sampling interval as well as for the whole sampling period (bar-charts for the individual data and for the arithmetic means including standard deviation).

Pharmacokinetic characteristics (except t_{max}) will be summarized by the statistics mentioned above. t_{max} will be described utilizing minimum, maximum and median as well as frequency counts.

To investigate the primary objective of this study regarding the effect of copanlisib on metformin, an exploratory analysis of variance (ANOVA) with terms of treatment condition and subject will be performed on the natural logarithmic transformation of PK parameters (AUC, $AUC_{(0-24)}$ and C_{max}) of metformin. The estimate and 90% confidence interval of the Day 8 (with copanlisib) to Day 1 (without copanlisib) ratio of C_{max}, $AUC_{(0-24)}$ and AUC for metformin are derived by inverse transformation of the estimates and the 90% confidence interval of the least squares mean differences that are obtained from the model above. This analysis will be performed on the PPS.

A code, similar to one below, will be used for the above analysis:

```
proc glm data=tab.pk;
  class Day Subject;
  model l&var= Day Subject / solution;
  lsmeans Day  /pdiff cl alpha=0.1;
run;
```

6.3.2 Pharmacodynamic Data

Plasma lactate levels, glucose, insulin and c-peptide will be summarized by treatment conditions (Day 1 and Day 8) for the original data as well as for the difference from baseline. Maximum change from baseline on Day 1 and Day 8 and change in PD data between Day 1 and Day 8 will also be summarized.

6.4 Safety

6.4.1 Adverse Events (AEs)

Individual listings of AEs will be provided. The incidence of treatment-emergent AEs, worst severity per subject and drug-related AEs, respectively, will be summarized using MedDRA (version 21.0 or higher) terms, by treatment day and total. Listings of deaths, SAEs and AEs leading to discontinuation will be provided.

For presenting AEs by treatment day, following will be considered:

Day 1 : include all AEs starting/worsening post metformin administration on day 1 until pre-dose of metformin administration on day 8.

Day 8 : include all AEs starting post metformin administration on day 8.

AEs are considered to be treatment-emergent if they start or worsen after first administration of study medication up to 30 days after end of treatment with study medication.

AE of special interest will be tabulated and listed separately.

6.4.2 Safety examinations

Quantitative data (hematology, blood chemistry, vital signs, ECG) will be summarized. These summary statistics will be presented for the original data as well as for the difference to baseline, by treatment day. Frequency tables will be provided for qualitative data. Laboratory data outside the reference range will be listed and flagged with ‘L’ for low and ‘H’ for high. Additional tables with all abnormal values will be presented.

Graphical displays of individual data as well as mean values with standard deviation will be included.

7. Document history and changes in the planned statistical analysis

Not applicable

8. References

[1] Clinical Study Protocol BAY BAY 80-6946 / 19951, Version 1.0, 15 May 2018

[2] Clinical Pharmacology Standards (CLIPS), Version 2.0, Dated 09 May 2018

[3] Clinical Pharmacology Standard Listings_V1.0_final_09052018, Version 1.0, Dated 09 May 2018