Data Supplement 1: Methods

Definition of Dose Limiting Toxicity

Dose-limiting toxicities (DLTs) were defined as any of the following occurring during cycle 1 and were considered to be related to the study treatment:

Hematologic toxicities:

- Absolute neutrophil count (ANC) <500/mm^3 for ≥7 days
- Febrile neutropenia (single temperature of >38.3°C [101°F] or a sustained temperature of ≥38°C [100.4°F] for ≥1 hour) with ANC <500/mm^3
- Platelets <25,000/mm^3 for >7 days despite platelet transfusion or grade ≥3 thrombocytopenic bleeding

Non-hematologic grade 3 or 4 toxicity, except:

- Drug-related grade 3 diarrhea (but will be classified as DLT if ***not*** manageable by dose interruption within 3 days/adequate treatment)
- Drug-related grade 3 hand–foot skin reaction (HFSR, but will be classified as DLT if ***not*** manageable by dose interruption within 3 days and adequate treatment)
- Drug-related grade 3 aspartate aminotransferase (AST) or alanine aminotransferase (ALT) ≤8× the upper limit of normal (ULN) without an increase in bilirubin (but will be classified as DLT if ***not*** manageable [i.e., does not return to baseline] by dose interruption within 3 days). However, if the AST and/or ALT increase is >3× ULN and associated with a concurrent increase in bilirubin (>2× ULN), it will be considered a DLT

In addition, the following situations may be considered DLTs after discussion between the investigators and the study sponsor:

- Relative dose intensity of <80% for regorafenib and irinotecan and two infusions of vincristine in cycle 1 due to an adverse event considered to be study-drug related
- A delay of more than 14 days to start cycle 2, due to treatment-related toxicity

Exceptions included grade 3 diarrhea or HFSR not manageable by dose interruptions and adequate treatment, or sustained grade 3 ALT and/or AST elevations without concurrent increased bilirubin.

Pharmacokinetics Analysis

Pharmacokinetics (PK) of regorafenib and irinotecan were assessed in all patients. On average, 3.9 and 4.5 samples per patient were available for estimation of the individual PK of regorafenib and irinotecan, respectively.

Among patients receiving the concomitant regorafenib regimen (cohort 1a), those aged 2 to <18 years provided blood samples to measure regorafenib and irinotecan plasma concentrations on the following days and timepoints:

- Irinotecan PK were measured on days 3, 4, and 5 at 1 hour, 2-3, 6-8, 22-26 (day 4), and 46-50 (day 5) hours, following irinotecan infusion on day 3
- Regorafenib PK were measured on days 3, 4, and 5 at 2-3, 22-26 (day 4), and 46-50 (day 5) hours, following irinotecan infusion on day 3. Samples were collected on day 14 (pre-dose) and 2-4 hours post-dose

In patients aged 6 months to <2 years, PK of regorafenib only were measured pre-dose and 24 hours post-dose on day 14 of cycle 1 or day 14 of cycle 2 if samples could not be collected in cycle 1.

Among patients receiving the sequential regorafenib regimen (cohort 1b), those aged 2 to <18 years provided blood samples to measure regorafenib and irinotecan plasma concentrations on the following days and timepoints:

- Irinotecan PK were measured on days 3, 4, and 5 of cycle 2 at 1 hour, 23, 68, 2226(day 4), and 4650 (day 5) hours, following irinotecan infusion on day 3
- Regorafenib PK were measured at 24 hours post-dose on day 14 of cycle 1 and pre-dose, 24 and 58 hours post-dose on day 21 of cycle 1

In patients aged 6 months to <2 years, PK of regorafenib only were measured pre-dose and 24 or 58 hours post-dose on day 21 of cycle 1.

Regorafenib was determined in plasma after protein precipitation with acetonitrile/ammonium acetate buffer containing the internal standards followed by separation employing high pressure liquid chromatography and tandem mass spectrometric detection. The calibration range of the procedure was from 2.00 (lower limit of quantitation) to 2,000 µg/L (upper limit of quantitation).

A previously developed population PK model for adults was applied to estimate the individual PK based on available (sparse) PK samples. This model considers the effect of enterohepatic recirculation on the PK of regorafenib to estimate regorafenib exposure based on sparse sampling. This model describes regorafenib disposition by distribution between central and peripheral compartments with linear elimination from the central

compartment into a gallbladder compartment from which regorafenib can be re-absorbed into the central compartment after gallbladder emptying following meal intake (1). In the pediatric population PK model, the parameters for clearance and volume of distribution for adults were allometrically scaled by individual body weight (normalized by the median body weight of 70 kg in the adult population), with exponent of 0.75 and 1 for clearance and volume of distribution, respectively.

References

1. Keunecke A, Hoefman S, Drenth HJ, Zisowsky J, Cleton A, Ploeger BA. Population pharmacokinetics of regorafenib in solid tumours: Exposure in clinical practice considering enterohepatic circulation and food intake. *Br J Clin Pharmacol* **2020**;86:2362-76.