# Physiologically Based Pharmacokinetic (PBPK) Modeling of the Central Nervous System (CNS) Pharmacokinetics of Tucatinib in Patients with Breast Cancer Brain Metastasis

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## Background

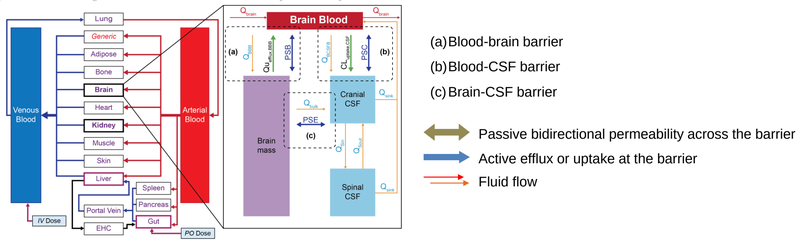
- Tucatinib (TUKYSA) is a HER2 selective tyrosine kinase inhibitor indicated in combination with trastuzumab and capecitabine for treatment of adult patients with advanced unresectable or metastatic HER2-positive breast cancer, including patients with brain metastases, who have received one or more prior anti-HER2-based regiments in the metastatic setting.
- First approved in USA on 17-Apr-2020.
- PBPK models integrate drug (substance) and system (physiology) information into a mathematical modeling framework. PBPK models have been used not only to describe the clinical observations, but also to predict the untested clinical outcomes using simulations.
- To quantitatively understand the CNS penetration of tucatinib and outcome of the HER2CLIMB trial demonstrating clinical efficacy of tucatinib in patients with breast cancer brain metastasis, a PBPK model for predicting the CNS PK of tucatinib in patients was developed and verified.

## Methods

- Tucatinib transcellular permeability and interaction with efflux transporters were determined using MDCKII cell lines.
- A whole-body PBPK model integrated with a 4-compartment permeability-limited brain model was developed and verified for predicting tucatinib concentration-time profiles in the plasma, cerebrospinal fluid (CSF), brain, and brain tumors.
- Target engagement ratio (TER), defined as the ratio of brain or tumor steady-state average concentration ($C_{ss,ave}$) of unbound drug to the $IC_{50}$ for inhibition of HER2 kinase, was used as a crude predictor of efficacy.

## PBPK Model

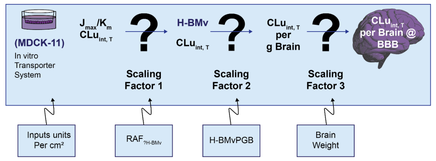
**Figure 1. Model structure of the whole-body PBPK model integrated with a 4-compartment permeability-limited brain model (4Brain)**




- 4Brain model has four compartments representing the brain blood, brain mass, cranial CSF, and spinal CSF[1]:
  1) Drug transport across the BBB is governed by bidirectional passive permeability (PSB) and ABCB1- and ABCG2-mediated active efflux clearance ($CL_{efflux,BBB}$)
  2) Drug transport across the blood-CSF barrier is controlled by bidirectional passive permeability (PSC) and ABCB1-mediated active influx clearance ($CL_{uptake,CSF}$)
  3) Drug transport between the brain mass and cranial CSF is governed by bidirectional passive permeability (PSE)
  4) Fluid balance is maintained by the circulation of CSF between spinal and cranial compartments and reabsorbed into the brain blood
  5) The cerebral blood flow rate ($Q_{Brain}$) links the 4Bain model to whole-body model
  6) Only unbound and unionized drug can passively pass through all barriers, while transporters act upon unbound drug (including both unionized and ionized species)
  7) All compartments are well-stirred with defined volumes
- Flow rates are described by the CSF secretion rate ($Q_{BCSFB}$), bulk flow rate from brain mass to cranial CSF ($Q_{bulk}$), CSF flow rate out of cranial and spinal compartments ($Q_{sink}$), CSF shuttle flow rate between cranial and spinal compartments ($Q_{Sin}$ and $Q_{Sout}$), and water transfer rate from the brain blood to brain mass ($Q_{BBB}$).

## In Vitro-In Vivo Extrapolation

**Figure 2. Mechanistic IVIVE (in vitro-in vivo extrapolation) scaling of transporter-mediated active clearance at human BBB. In vitro active efflux clearance was scaled to active efflux clearance in vivo, using experimentally determined amount of efflux transporters in cellular systems and human brain tissue**




$$CL_{efflux,vivo} = CL_{efflux,vitro} \times RAF \times BMvPGB \times BW$$

$$RAF = \frac{Transporter\ Protein\ Abundance\ at\ BBB}{Transporter\ Protein\ Abundance\ in\ vitro}$$

- $CL_{efflux,\ vitro}$ was scaled to the whole-brain in vivo efflux transporter-mediated clearance ($CL_{efflux,vivo}$; µL/min/mg) by multiplying a relative activity factor (RAF).
- Abundance in vitro or in vivo (at BBB) represents the ABCB1 (P-gp) and ABCG2 (BCRP) transporter protein expression levels in brain microvessels (pmol/mg microvessels) or in cellular models (pmol/mg cells), respectively.[2]
- BMvPGB is the milligrams of brain microvessels per gram brain.
- BW is the brain weight (gram).

## In Vitro Permeability and Efflux

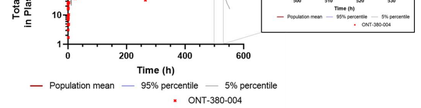
**Table 1. Apparent transcellular permeability and efflux ratios of tucatinib**
**Tucatinib showed high passive permeability (Papp, 12.6×10$^{-6}$ cm/s)**
**Tucatinib is a substrate for ABCB1 (net efflux ratio, 13.8) and ABCG2 (7.7)**

| | Without inhibitor | With ABCB1/ABCG2 inhibitor |
|---|---|---|
| **Parental MDCKII** | | |
| Apparent permeability ($P_{app,A-B}$) (×10$^{-6}$ cm/s)[a] | 7.45 ± 1.58 | 12.60 ± 0.57 |
| Efflux ratio | 2.50 ± 0.52 | 0.82 ± 0.11 |
| Net efflux ratio[b] | 3.11 ± 1.05 | NA |
| **MDCKII-ABCB1** | | |
| Apparent permeability ($P_{app,A-B}$) (×10$^{-6}$ cm/s)[a] | 2.47 ± 0.25 | 11.45 ± 2.76 |
| Efflux ratio | 9.48 ± 0.45 | 0.69 ± 0.04 |
| Net efflux ratio[b] | 13.77 ± 1.49 | NA |
| **MDCKII-ABCG2** | | |
| Apparent permeability $P_{app,A-B}$ (×10$^{-6}$ cm/s)[a] | 2.91 ± 0.81 | 11.65 ± 1.91 |
| Efflux ratio | 7.79 ± 1.41 | 1.01 ± 0.16 |
| Net efflux ratio[b] | 7.74 ± 0.20 | NA |

[a] Bidirectional permeability experiments were performed at pH 7.4 in both apical and basolateral chambers
[b] Net efflux ratio was the efflux ratio in the absence of an ABCB1 inhibitor (elacridar) or ABCG2 inhibitor (Ko145) divided by the efflux ratio in the presence of the inhibitor
Data are expressed as the mean ± standard deviation from 2 independent experiments (with triplicates in each experiment)

## Simulated Plasma Concentrations

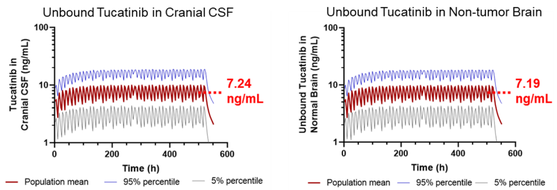
**Figure 3. PBPK model-simulated tucatinib plasma concentrations over time (red lines) agree well with observed tucatinib concentrations (red x) in patients with HER2-positive metastatic breast cancer**




- Simulations of 10 trials with 10 subjects in each were performed in the Simcyp virtual cancer patient population following 22-day tucatinib treatment at 300 mg BID.
- Observed clinical plasma concentrations of tucatinib were obtained from patients treated with tucatinib at 300 mg BID (ONT-380-004, NCT01983501).

## Simulated CSF and Normal Brain Concentrations

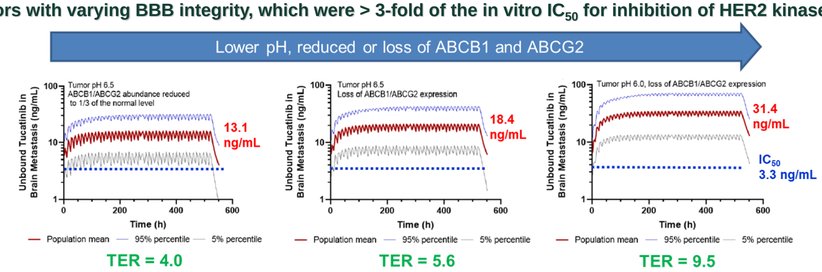
**Figure 4. PBPK model-simulated tucatinib concentrations in CSF and normal brain were similar**




- The model-predicted population mean steady-state average CSF concentration was 7.24 ng/mL and mean CSF-to-unbound plasma ratio (CSF $K_{p,uu}$) was 0.66.
- In the normal brain with an intact BBB (i.e., brain pH 7.12, ABCB1 abundance 3.38 pmol/mg, ABCG2 6.21 pmol/mg), the simulated population mean steady-state average brain concentration ($C_{ss,ave}$) of unbound tucatinib was 7.19 ng/mL and mean unbound brain-to-plasma ratio ($K_{p,uu}$) was 0.65.

## Simulated Breast Cancer Brain Metastasis Concentrations

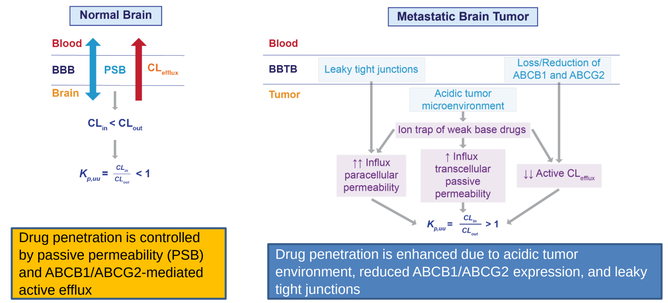
**Figure 5. PBPK model-simulated unbound tucatinib concentrations in breast cancer brain metastasis tumors with varying BBB integrity, which were > 3-fold of the in vitro $IC_{50}$ for inhibition of HER2 kinase**




$$\text{TER (Target Engagement Ratio)} = \frac{mean\ steady\text{-}state\ unbound\ concentration}{in\ vitro\ IC_{50}\ for\ HER2\ inhibition}$$

- Quantitative proteomics analyses of human normal brain (n = 30) and breast cancer brain metastasis (n = 30) specimens suggest that the median protein abundances of ABCB1 and ABCG2 are 3.38 and 6.21 pmol/mg in human normal brain microvessels, respectively[3]; and their protein levels are lower in breast cancer brain metastasis specimens (submitted).
- Electrode measurements of pH in human brain tumors are as low as 5.9 with a mean around 6.8.[4-6]
- To predict tucatinib concentrations in the human normal brain and brain metastasis tumors, different ABCB1/ABCG2 protein abundances at the BBB and varying brain pH were applied in the simulations following 22-day treatment at 300 mg BID.
- In brain tumors with pH 6.5 and ABCB1/ABCG2 abundance reduced to 1/3 of the normal level (*left*), pH 6.5 and loss of ABCB1 and ABCG2 expression (*middle*), and pH 6.0 and loss of ABCB1 and ABCG2 expression (*right*), the simulated population mean $C_{ss,ave}$ of unbound tucatinib was 13.1, 18.4, and 31.4 ng/mL, respectively.
- Target engagement ratio, defined as the ratio of the average steady-state unbound drug brain concentration to the *in vitro* $IC_{50}$ for HER2 inhibition, was used as a crude predictor of efficacy.[7]
- Tucatinib showed an $IC_{50}$ of 3.3 ng/mL in the in vitro HER2 kinase assay.[8] Therefore, following the standard dosing regimens, tucatinib achieved the HER2 target engagement ratios > 2 in the normal brain and brain metastases.

**Figure 6. Proposed mechanisms of CNS penetration of tucatinib: Drug penetration is enhanced due to acidic tumor environment, reduced expression of efflux transporters, and leaky tight junctions**




## Conclusions

- The PBPK modeling indicates that tucatinib achieves pharmacologically active concentrations for HER2 inhibition in brain tumor metastases with a disrupted BBB as well as in infiltrating tumor regions behind an intact BBB, which is key to effective systemic treatment of brain metastases.
- This study provides quantitative and mechanistic insights into the outcome of the HER2CLIMB trial[9] which demonstrated clinical efficacy of tucatinib in patients with breast cancer brain metastasis.

**References**

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