An assessment of the preclinical development of long-acting biodegradable emtricitabine implants

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Introduction

- Long-acting (LA) antiretroviral therapy enables therapeutic plasma exposures to be maintained over extended time periods, minimising the impact of sub-optimal adherence on clinical response.
- Novel LA antiretrovirals are in preclinical development for HIV pre-exposure prophylaxis or treatment.
- The methods used for preclinical development of LA HIV antiretroviral therapies differs based on the intended delivery route, site of administration and dosing interval.
- LA formats are extremely difficult to develop and present major challenges in preclinical (*in vitro–in vivo* extrapolation) and clinical (species scaling) development.

Aims

- This study analysed the *in vitro* release and *in vivo* pharmacokinetic (PK) exposure profiles of emtricitabine (FTC) from novel biodegradable implants formed from polymers containing FTC within the polymer backbone.
- The overarching aim of this work was to develop an appropriate approach for extrapolating *in vitro* data to make *in vivo* predictions of pharmacokinetics (PK) for this technology.

In vitro release study:

- *Polymer manufacture*: FTC-derived implants were manufactured with varying properties, using previously published methodologies. [1]
- 25 polymer implants were screened.
- Each FTC-derived polymer implant was incubated at 37 ^{o}C/250 rpm in 1 mL of phosphate buffered saline (PBS) containing human liver microsomes for 14 days.
- At each timepoint a 250 µL sample was taken and replaced with fresh buffer/microsomes to maintain sink conditions.
- Each sample was quantified for FTC using a previously validated LC-MS/MS assay. [2]

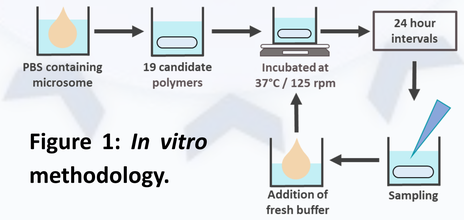

Figure 1: *In vitro* methodology.

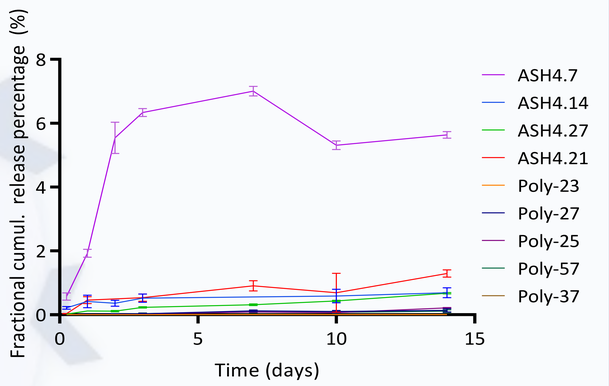

Figure 2: Fractional cumulative % release of emtricitabine *in vitro* from the nine implants selected for further study.

- Cumulative % release of FTC from the 9 candidates selected for further study ranged between 0.01-8% over 14 days.
- The candidates selected for *in vivo* testing were the best performers for each of the four chemistry types included in the study.

In vivo PK exposure study:

- 9 polymer candidates from the *in vitro* study were selected.
- 2 implants containing a mean total mass of 66.8 mg FTC (33.4 mg per implant) were implanted subcutaneously (SC) into the scapular region of male Wistar rats anaesthetised with 3% isoflurane, via a disposable implant syringe with a 12-gauge Lauer lock implant needle attached.[4.]
- All rats received a 0.05 mg/kg SC injection of buprenorphine post administration of the implant. [4.]
- A serial timecourse of plasma samples were taken via the tail vein 1-5 hours and 1-28 days post implantation. [4.]
- FTC concentrations in plasma were quantified using a validated LC-MS/MS assay. [2]

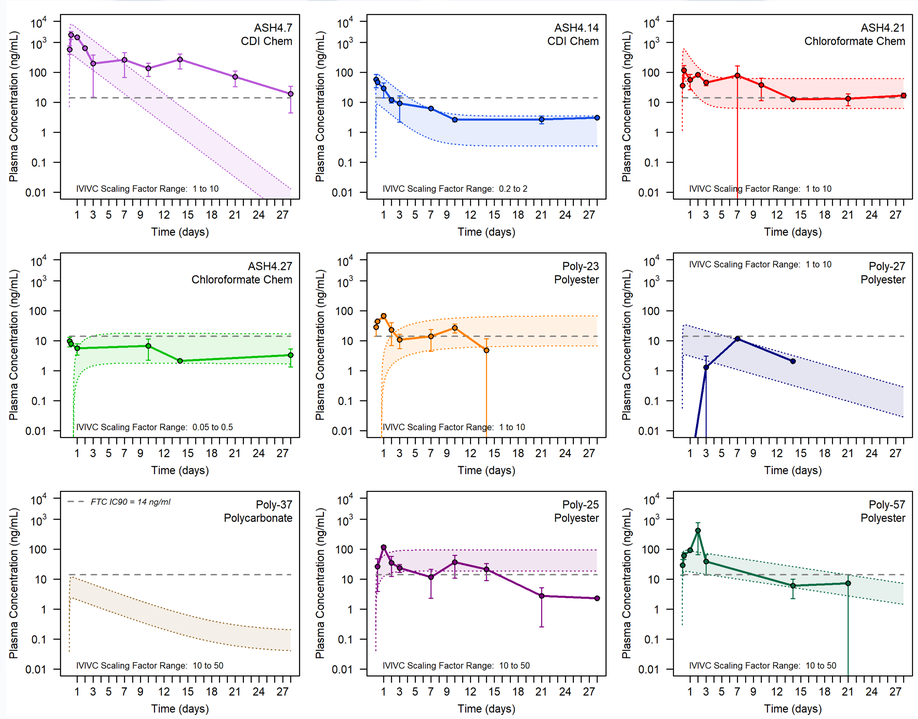

Figure 3: 28-day rat plasma PK exposure profiles of FTC following SC implantation of polymer implants

- Exposure profiles are overlaid with predictions from *in-vitro* release given by IVIVC convolution (see left).
- ASH4.7 FTC concentrations were above the non-protein adjusted FTC IC90 (14 ng/mL), for up to 28 days, Poly-23 FTC concentrations were above the IC90 for up to 10 days and ASH4.21 for up to 7 days.
- A consistent IVIVC scaling factor (or pattern in scaling factors) was not found across polymers tested , or for polymers grouped by chemistry type.

In vitro-in vivo correlation (IVIVC):

- Predictions of *in vivo* exposure were derived from *in vitro* release profiles using R (v4.0.3) by:
  - Fitting a biexponential mathematical model to the 14-day *in vitro* release profiles and extrapolating to 28 days
  - Multiplication of the extrapolated profiles by a scaling factor range and then convolution with a previously reported IV PK disposition profile for FTC [3.]
- *IVIVC* plots of release per day *in vitro* vs. FTC $AUC_{0\text{-tlast}}$, and C_{max} from each FTC implant in the *in vivo* study are also presented in **Figure 4**. *in vivo* FTC $AUC_{0\text{-tlast}}$ and C_{max} gave R^2 values of 0.9, and 0.84 vs. *in vitro* respectively.
- *In vitro* release rates correlated with *in vivo* exposure and C_{max}, which may facilitate selection of candidates with a favourable profile of high FTC exposure but moderate C_{max}.

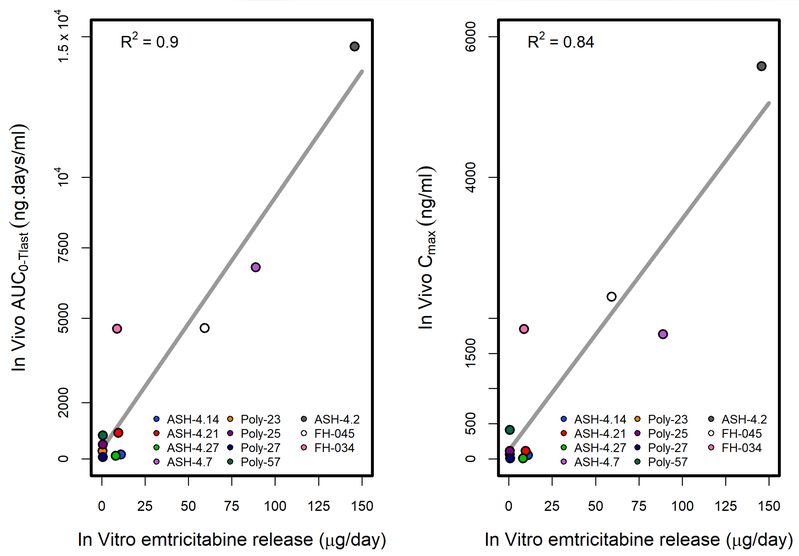

Figure 4: *IVIVC* of *in vivo* FTC $AUC_{0\text{-tlast}}$ or C_{max} vs. FTC release per day *in vitro*

Conclusions

- Prediction of *in vivo* exposure profiles by convolution of *in vitro* release with IV disposition of FTC did not reveal consistent, *a priori*, scaling factors suitable for robust *in vitro-in vivo* extrapolation.
- *In vitro-in vivo* correlation demonstrated that the *in vitro* methodology was able to predict adequately the ranked release rate and exposure of FTC *in vivo* in a rat model.
- A more refined *in vitro* experiment that better models the subcutaneous site, could enhance prediction of polymer *in vivo* release profiles and enable better extrapolation.
- These studies highlight the need for further development of *in vitro* methods that better model the *in vivo* performance of LA technologies.

References: [1] Shakil *et al.* 2022. [2] Curley *et al.* 2021. [3.] Nirogi *et al.* 2012. [4.] All animal work was conducted in accordance with the Animals (Scientific Procedures) Act 1986 (ASPA) implemented by the UK Home Office.

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