

Mathematical modelling of the molecular mechanisms of interaction of tenofovir with emtricitabine against HIV

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Background and Objective

The two nucleoside reverse transcriptase inhibitors (NRTI) Tenofovir disoproxil fumarate (TDF) and emtricitabine (FTC) are used in combination for treatment of HIV-1 infection and in pre-exposure prophylaxis. Administered as prodrugs, they are taken up by HIV infected target cells, undergo intracellular phosphorylation and compete with natural deoxynucleoside triphosphates (dNTP) for incorporation into nascent viral DNA during reverse transcription. Once incorporated, they halt reverse transcription.

In-vitro studies have proposed that TDF and FTC act synergistically within an HIV infected cell. However, it is unclear whether-, and which *direct* drug-drug interactions mediate the apparent synergy.

The goal of this work was to refine a mechanistic model for the molecular mechanism of action (MMOA) of nucleoside analogues in order to analyse whether putative direct interactions may account for the *in-vitro* observed synergistic effects.

MMOA model refinement

Two extended models were generated and used to assess - for realistic parameter ranges - if the proposed hypotheses can explain the observed levels of synergy between the drugs.

DEC-modified MMOA; FTC-TP may induce a dead-end complex after binding to a TFV-DP terminated primer [3](Figure 2B). This binding blocks access to the incorporated TFV-DP unless FTC-TP unbinds, which alters the excision of TFV-DP from the terminated primer (Figure 2A, in green).

dNTP MMOA; NRTIs may alter the pools of endogenous dNTPs [1], e.g. by binding to the respective intracellular kinases and blocking them. This increases $r_{term}(i+1)$ and decreases $r_{pol}(i+1)$ (Figure 2A, in blue).

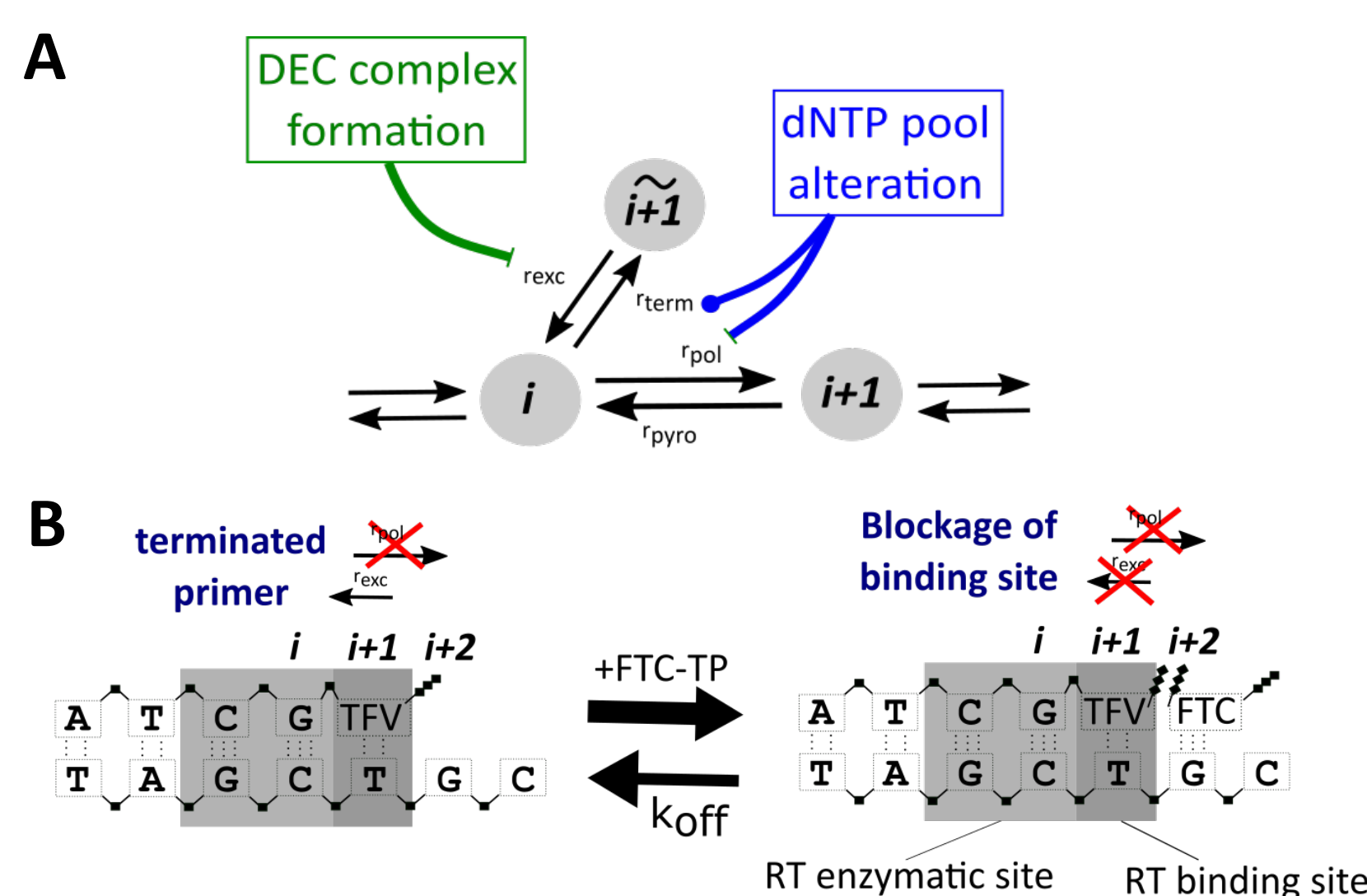


Figure 2: Modified molecular mechanisms of action of NRTIs: direct drug interactions (A) and mechanisms of dead-end complex formation (B)

References

- [1] Chen, X., et al., Analysis of the Endogenous Deoxynucleoside Triphosphate Pool in HIV-Positive and -Negative Individuals Receiving Tenofovir-Emtricitabine. *Antimicrob Agents Chemother*, 2016. 60(9): p. 5387-92.
- [2] Duwal, S. and M. von Kleist, Top-down and bottom-up modeling in system pharmacology to understand clinical efficacy: An example with NRTIs of HIV-1. *Eur J Pharm Sci*, 2016. 94: p. 72-83.
- [3] Feng, J.Y., et al., The triple combination of tenofovir, emtricitabine and efavirenz shows synergistic anti-HIV-1 activity in vitro: a mechanism of action study. *Retrovirology*, 2009. 6: p. 44.

The molecular mechanism of action (MMOA) model

The previously defined model [2] explicitly considers reverse transcriptase (RT)-mediated polymerization of nascent viral DNA and the effect of activated NRTI-triphosphates.

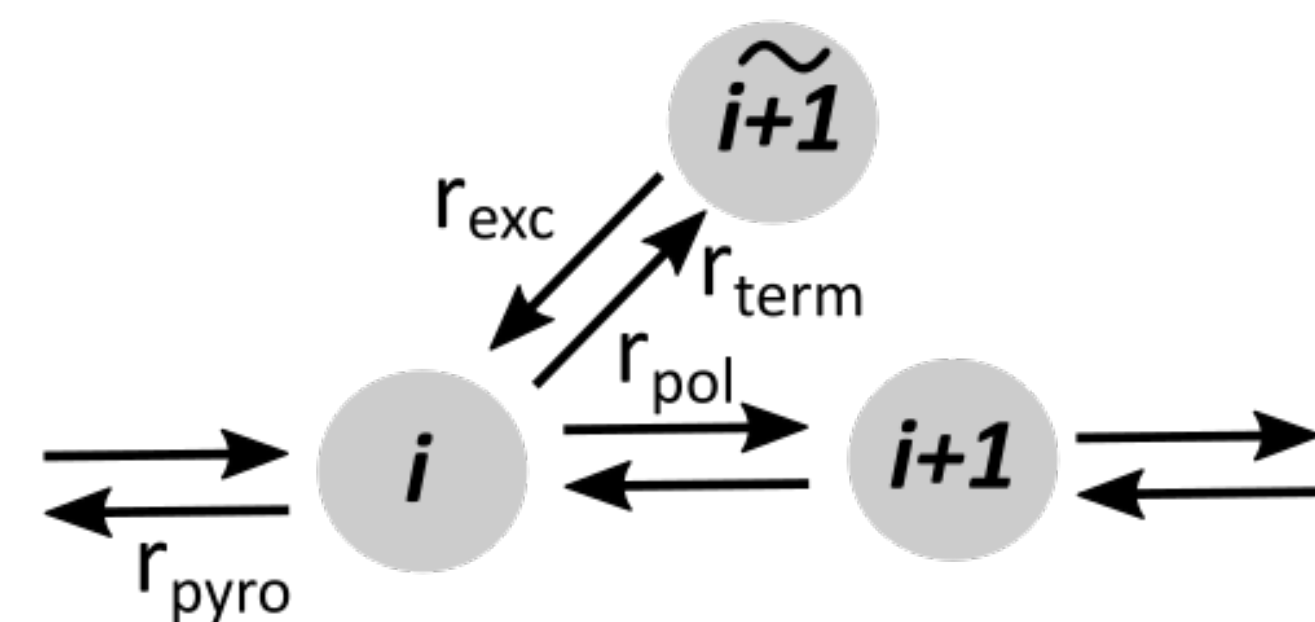


Figure 1: Model of the molecular mechanisms of action of NRTIs.

As shown in Figure 1, a primer sequence of length i may either be prolonged by the polymerase reaction $r_{pol}(i+1)$ or may be terminated by the incorporation of an NRTI-TP $r_{term}(i+1)$, depending on the nucleotide to be incorporated at position $(i+1)$. An incorporated NRTI may be excised with rate $r_{exc}(i+1)$. Also, the primer of length $(i+1)$ may be shortened by the pyrophosphorolysis reaction $r_{pyro}(i)$. It takes *in-vitro* measurable microkinetic parameters as input and computes the inhibition of reverse transcription (ε) by NRTIs. From there one can derive the inhibition of cell infection (η).

Modelling combination effects using the MMOA model

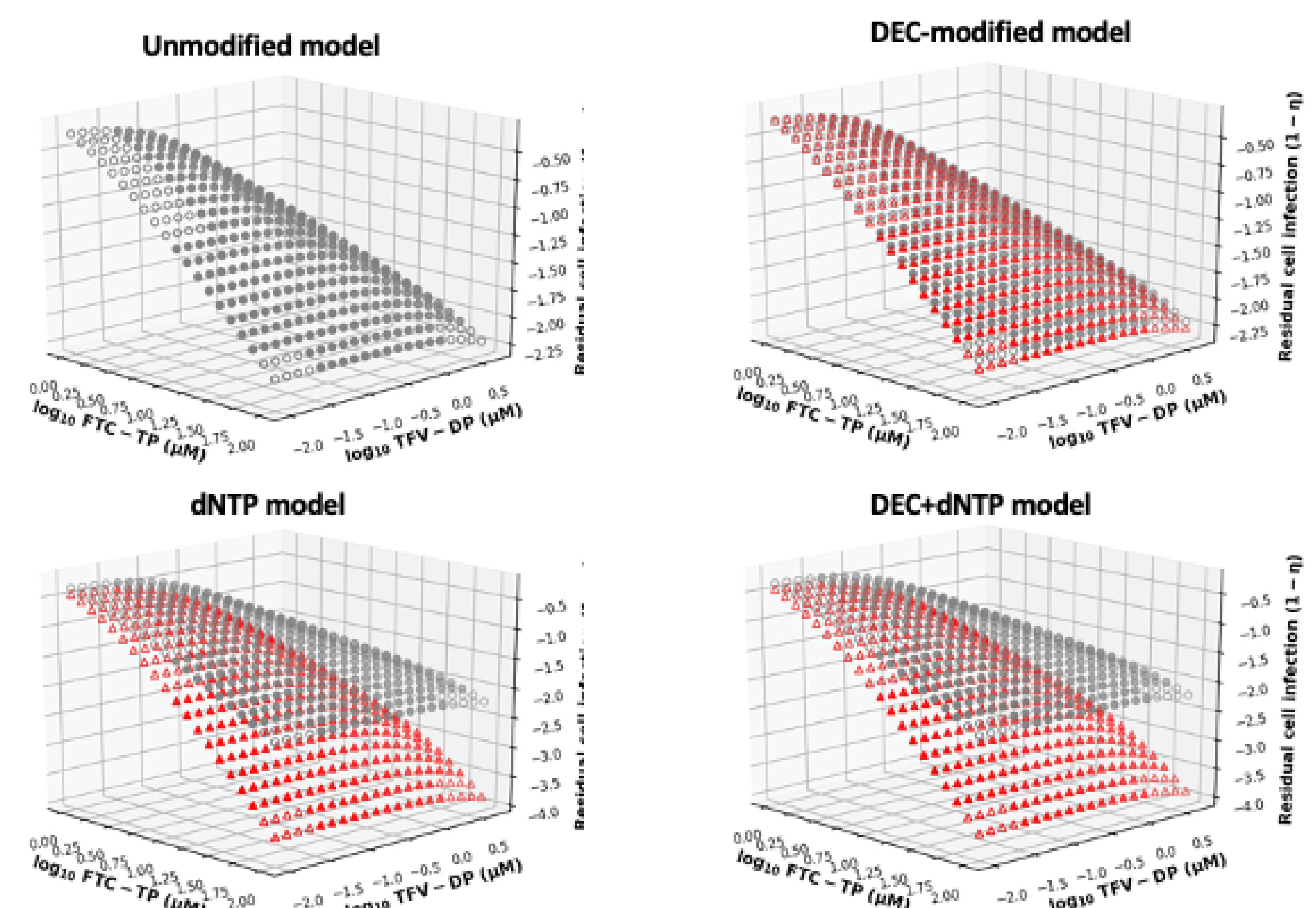


Figure 3: Drug combination surface plot. The x- and y-axis depict the concentration of FTC-TP and TDF-DP respectively. The solid dots mark clinically observed concentration ranges after once-daily oral administration of truvada (300mg TDF + 200mg FTC). The z-axis depicts the residual cell infection ($1-\eta$) computed from the MMOA model. Red dots indicated the modified models.

Analysis using the combination index

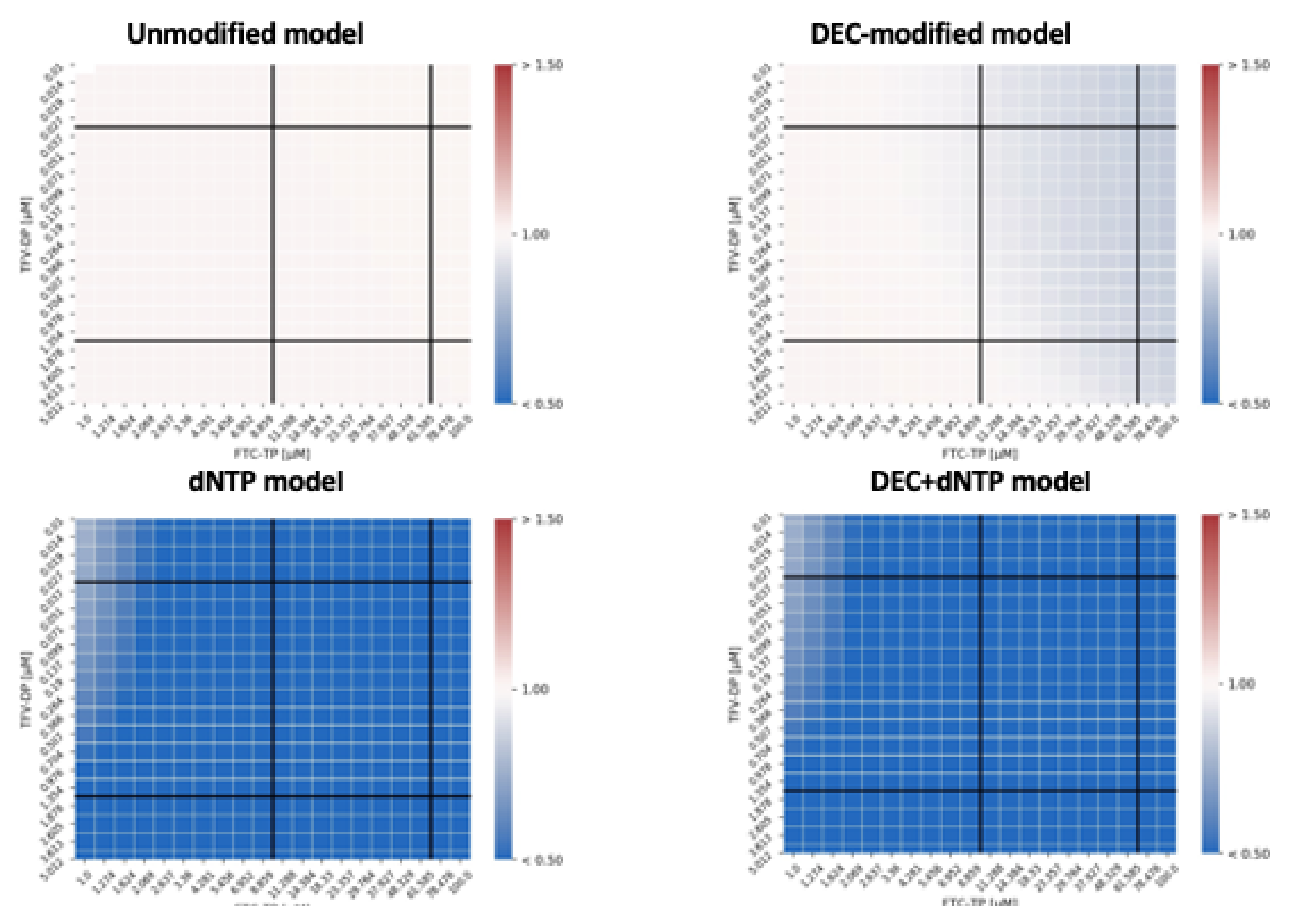


Figure 4: TFV-DP and FTC-TP interactions heatmaps (Combination Index). The concentrations that produced an effect classified as synergistic are shown in purple. The panels report results for the unmodified model and the other modifications applied.

Conclusion

Our predictions with the MMOA model, as well as available experimental data indicate that the direct interaction of FTC-TP and TFV-DP is mainly mediated by a depletion of dNTP pools. Interactions at the level of DEC formation play a minor role for this drug combination at physiologically meaningful drug concentrations. Concerning its applications, the refined model output (η) can be used as a drug efficacy parameter. It can be integrated in other routines, providing a link between the pharmacokinetic and pharmacodynamic of combination therapy.