EFFECT OF ANTIBIOTICS-MEDIATED MICROBIOTA DEPLETION ON TACROLIMUS PHARMACOKINETICS IN MICE

Degraeve AL[1,4], Stillemans G[1,2], Moudio S[1], El-Hamdaoui H[1], Delongie K-A[3], Haufroid V[2,3], Bindels LB[4], Elens L[1]

[1]Department of Integrated PharmacoMetrics, PharmacoGenomics and PharmacoKinetics, LDRI, UCLouvain, Belgium;
[2] Louvain Centre for Toxicology and Applied Pharmacology, IREC, UCLouvain, Belgium;
[3] Department of Clinical Chemistry, Cliniques Universitaires Saint-Luc, Belgium; [4] Metabolism and Nutrition Research Group, LDRI, UCLouvain, Belgium.

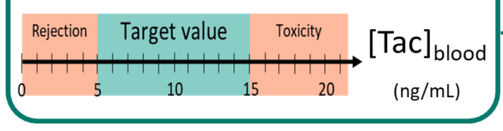
IATDMCT Virtual 2020 Congress
September 14-16

Context

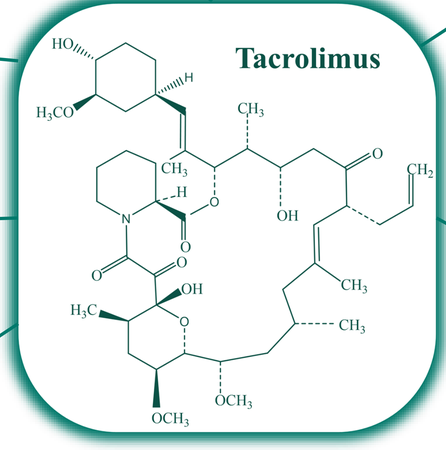
Immunosuppressive agent: prophylaxis of organ rejection after transplantation

Low bioavailability

Narrow therapeutic index



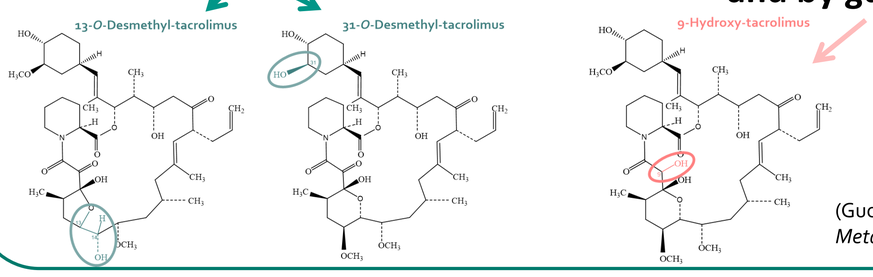
ABCB1 transport
Biliary excretion



Large PK variability inter- & intra-patients → **Low correlation : TAC dose ~[TAC]blood** → **Regular post-treatment monitoring**

?

Intensive hepatic metabolism (CYP3A) ... and by gut bacteria



(Guo et al., *Drug Metab Dispos.*, 2018)

Objectives

Regarding **tacrolimus (TAC) pharmacokinetics (PK) variability**, we hypothesized that beside an already known genetic component, there could be an **implication of gut bacteria**, especially to explain **intra-patient variation**. Using mice (male C57Bl6), we investigated the effect of antibiotics-mediated (ATB) microbiota depletion on tacrolimus PK.

Method

❖ TAC dose

In order to define the best dose for further experiments, TAC was administered by oral gavage in mice (n = 10) with different escalating doses (from 0,1 to 10 mg/kg of body weight [bw]). At steady-state, blood concentrations were determined at Toh and T2h using UHPLC-MS/MS.

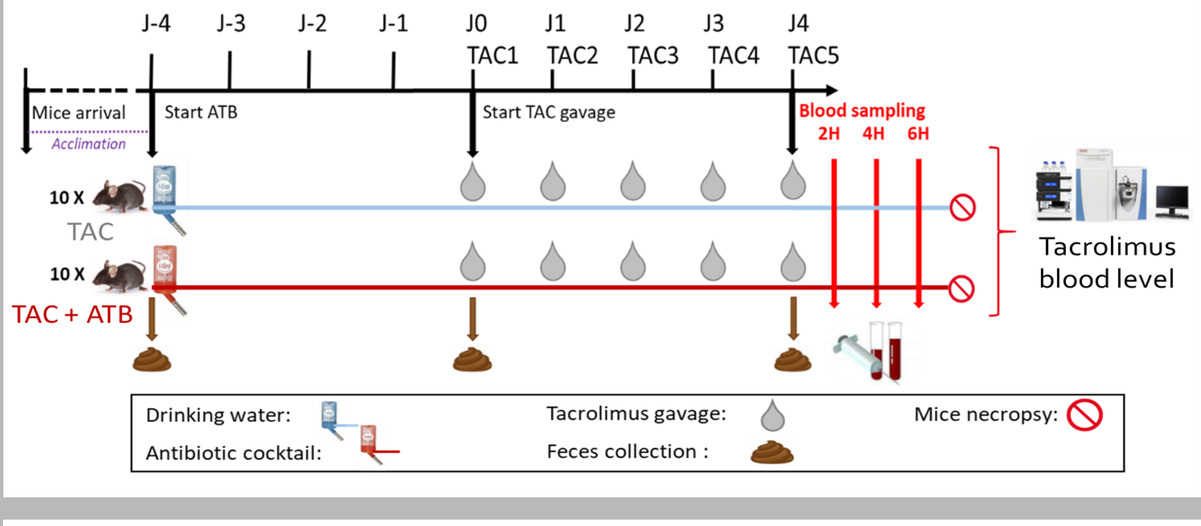
❖ Blood sampling strategy

A kinetic curve covering 6 times points was performed at steady-state, using the selected TAC dose [3mg/kg bw] (n = 24 mice, n = 72 PK data) to identify the best sampling strategy.

❖ Impact of gut microbiota depletion on TAC PK

Mice were treated with TAC ± a non-resorbable ATB cocktail that ensures gut microbial depletion. Bacterial load in faeces was measured by qPCR of the 16S rRNA gene. Blood levels were determined at T2h, T4h and T6h and compared between groups (n = 10 mice/group). All generated PK data were used to develop a TAC popPK model (n = 54 mice, n = 172 PK data) to estimate PK parameters. TAC AUC were compared between groups.
The experimental scheme is depicted in figure 1.

Figure 1.



Results

❖ TAC dose and blood sampling strategy selection

Figure 2A.

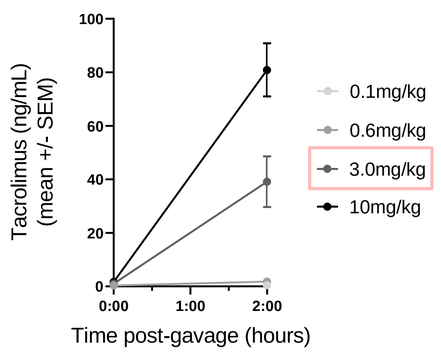
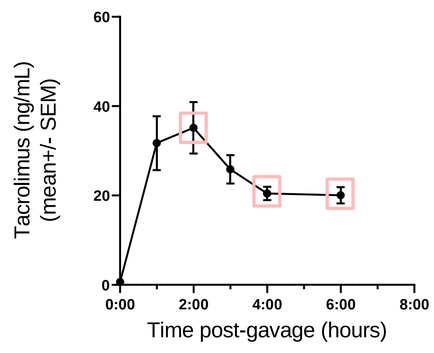


Figure 2B.



✓ Among the doses tested, the best experimental scheme was 3mg/kg bw by oral gavage (Figure 2A). Based on the kinetic profile, we continue with a blood sampling at 2, 4 and 6h post-gavage (Figure 2B).

❖ Gut microbiota depletion and TAC PK model

Figure 3.

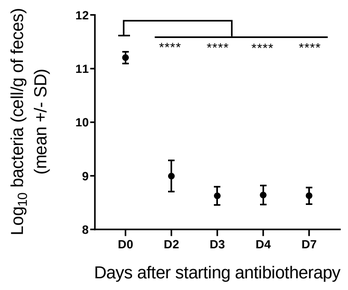


Table 1: Primary and derived PK parameters obtained with all generated data (n=172)

TAC PK model		Median	IC95%
Primary PK parameters	Cl/F	0,326 l/h	[0,290-0,362]
	V	2,1 l	[1,747-2,453]
	K_a	1,22 h^{-1}	[0,57-1,87]
Derived PK parameters	AUC_{0-24h}	220,1 ng.h/ml	-
	T ½	3,69 h	-

✓ The non-resorbable ATB cocktail induced a 2,6 log_{10} reduction in bacterial load after 4 days (Figure 3, One-way ANOVA, Dunnett's post-hoc test, **** p< 0,0001).

✓ A one-compartment popPK model was generated, PK parameters were computed (Table 1).

❖ Impact of the gut microbiota on TAC PK

Figure 4A.

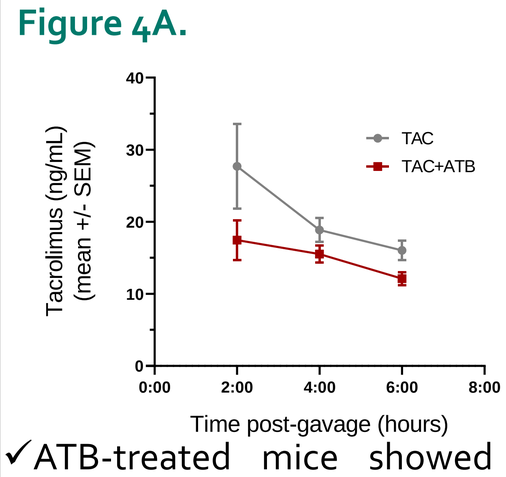
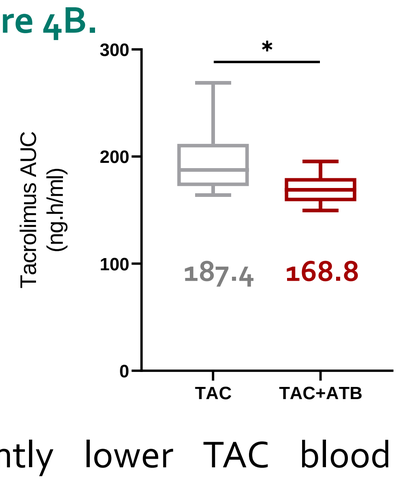


Figure 4B.



✓ ATB-treated mice showed significantly lower TAC blood whole exposure when compared to mice treated with TAC solely (Figures 4A and 4B, Wilcoxon-Mann-Whitney test, p = 0.023).

Conclusion

Our results demonstrate that **effective ATB-mediated microbial depletion decreases TAC exposure**. This observation suggests that the **gut microbiota influences TAC PK.**
If confirmed in humans, this information can be of importance for explaining inter- but also intra-individual variability in TAC exposure.
Further studies are required to understand the mechanisms by which the gut microbiota impacts TAC PK.

This work was supported by the Fonds de la Recherche Scientifique (F.R.S.-FNRS, Belgium) and the Fonds Spéciaux de la Recherche (FSR, UCLouvain).
DEGRAEVE Alexandra (alexandra.degraeve@uclouvain.be) is a FNRS fellow.