

Using nonlinear mixed effects modeling to characterize the pharmacokinetics and pharmacodynamics of benazeprilat in dogs

J. Mochel ^[1,2,4], M. Fink ^[2], M. Peyrou ^[1], J. Giraudel ^[3] and M. Danhof ^[4]

[1] Novartis Centre de Recherche Santé Animale SA, Saint-Aubin, Switzerland
[2] Novartis Modeling & Simulation, Basel, Switzerland
[3] Novartis Animal Health Australasia Pty Ltd, Kemps Creek, Australia
[4] Leiden University Department of Pharmacology, 2300 RA Leiden, The Netherlands

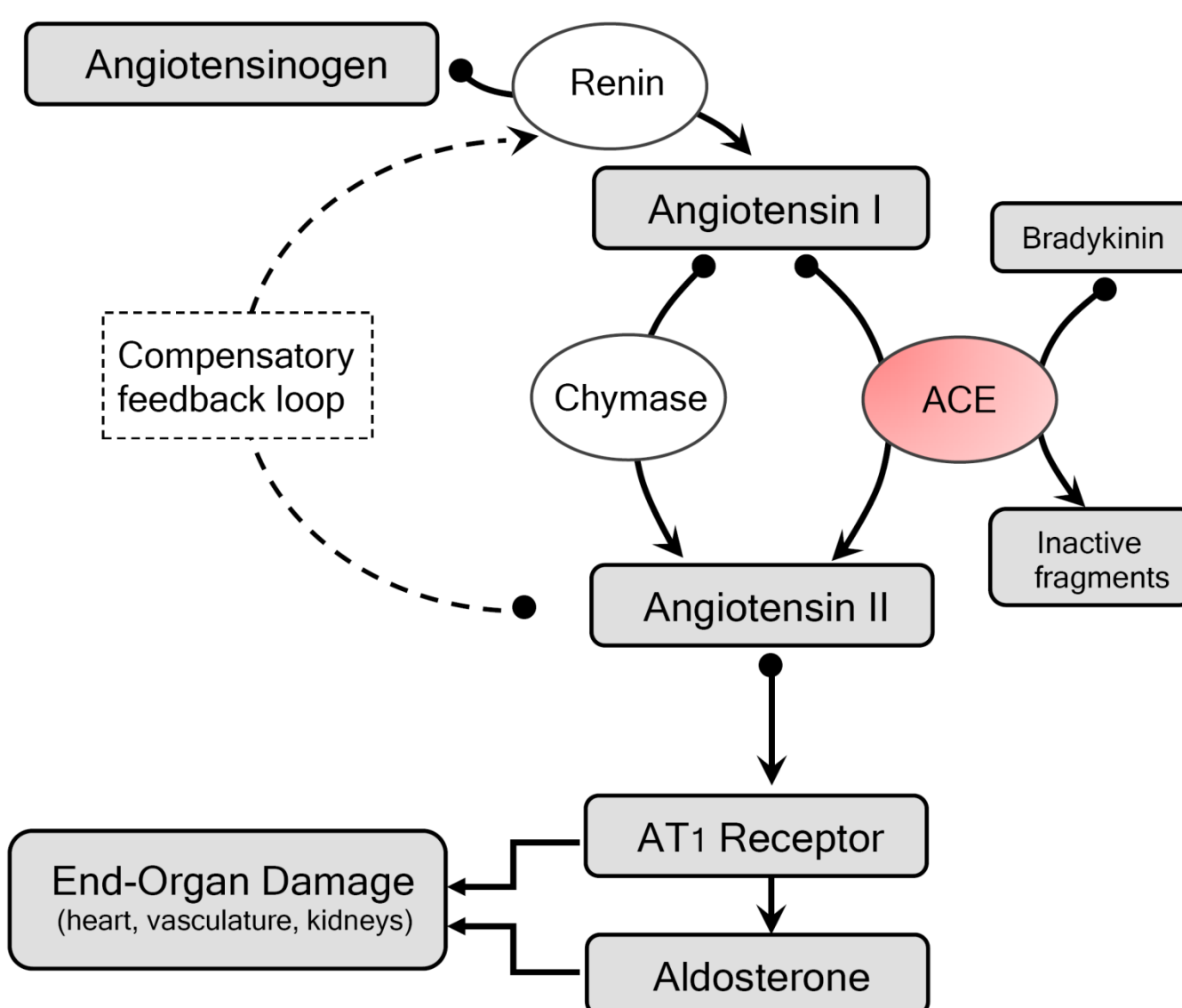
* Rationale & Objectives

- Long-term increases in angiotensin II and aldosterone lead to the progression of heart failure (HF) to its end stage
- ACE inhibitors are the foremost therapeutic option to offset activation of the Renin-Angiotensin-Aldosterone System (RAAS) ^[1] in canine HF
- Benazepril hydrochloride is a prodrug which is converted in vivo by esterases into its active metabolite, benazeprilat, a highly potent and selective inhibitor of ACE ^[2]
- Limited information on the effect of benazeprilat on circulating RAAS peptides is available in dogs
- The objective of this research was to determine the effect of benazeprilat on the renin-angiotensin cascade using nonlinear mixed effect modeling
- Chronobiology of RAAS biomarkers and determinants of their regulation were further investigated

* Results

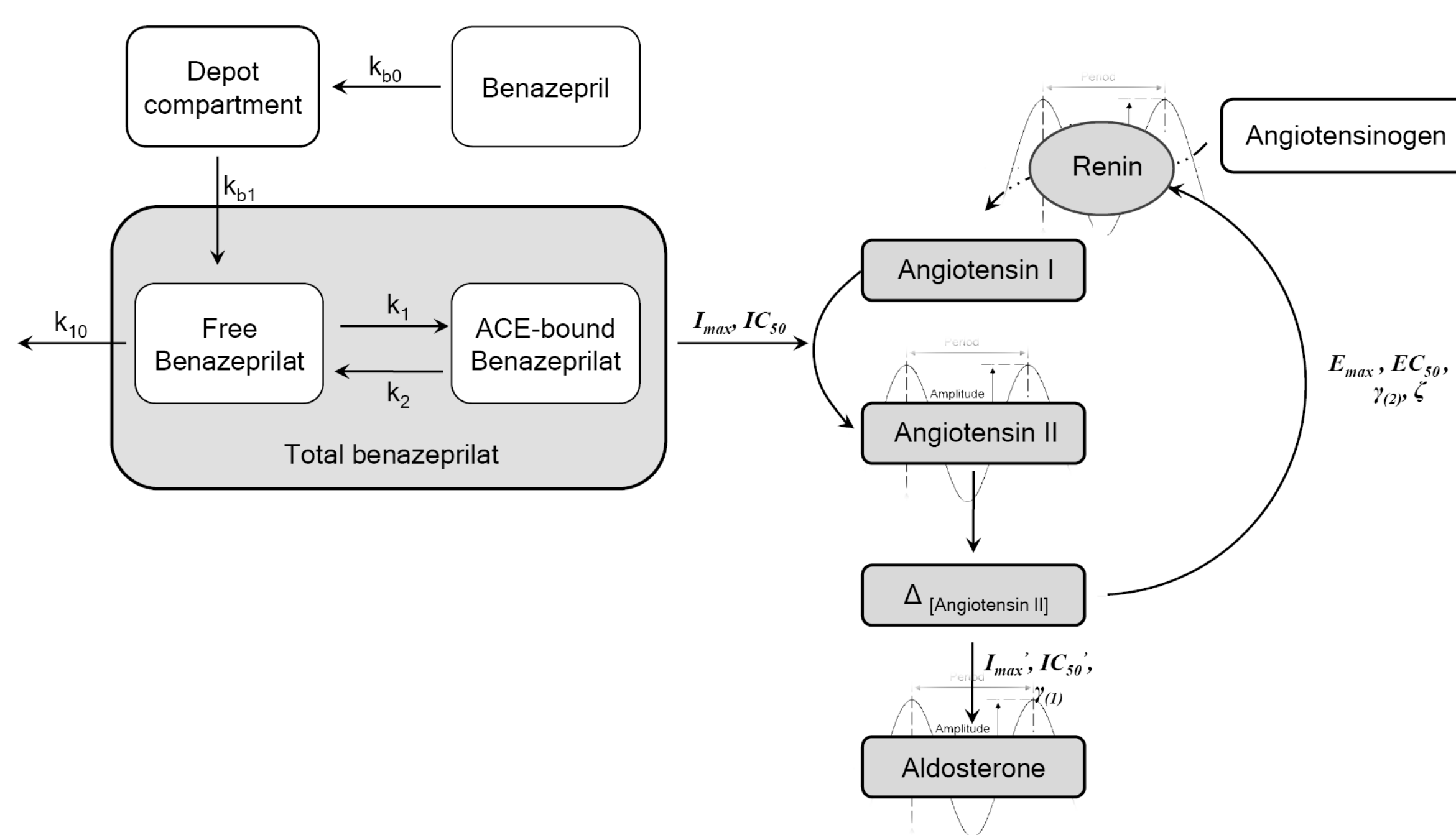
- The pharmacokinetic model consists of single compartment in which the ACE is either free or bound to benazeprilat, as reported by Toutain et al. (2000) ^[3]
- Cosine functions were identified to fit the periodic nature of RA, AII and ALD well, and served as baseline for the development of direct response models to assess the effect of benazepril on the RAAS
- Benazeprilat plasma concentrations served as a driving force for the prediction of AII levels
- RA and ALD concentrations were further derived from the predicted AII response during placebo and benazepril treatment
- Moving down from the “top” of the renin cascade, benazepril evoked a substantial increase in RA, while decreasing AII and ALD
- Bodyweight, gender and sodium intake were found to be significant covariates to explain part of the between-subject variability

Figure 1. Schematic view of the systemic renin-angiotensin cascade



Angiotensinogen is converted into angiotensin I by the activated form of renin. Angiotensin II is derived from AI by enzymatic cleavage catalyzed by ACE. AII stimulatory effect on aldosterone release is mediated by type 1 AII receptors (AT1R). Compensatory mechanisms contribute to the regulation of renin release. Long-term increases in AII and ALD lead to end-organ damage and progression of heart failure. ACE inhibitors, like benazepril, act by preventing the formation of AII and the degradation of bradykinin, which acts as a potent vasodilator

Figure 2. Integrated pharmacokinetic/pharmacodynamic model of benazeprilat disposition and effect on the canine renin-angiotensin cascade



k_{10} and $k_{10,1}$: 0 and 1st-order rate constants of absorption; k_1 and k_2 : 2nd and 1st-order rate constant of association/dissociation to ACE; k_{10} : 1st-order rate constant of elimination; IC_{50} : benazeprilat concentration leading to 50% of the total inhibition of angiotensin II; IC_{50}^* and EC_{50} : difference in angiotensin II (Δ) between placebo and benazepril required to achieve respectively 50% of the total inhibition of aldosterone, and 50% of activation of renin; γ and γ_{2D} : power terms representing the steepness of the concentration effect relationships; ζ : Richards asymmetry factor

* Methodology

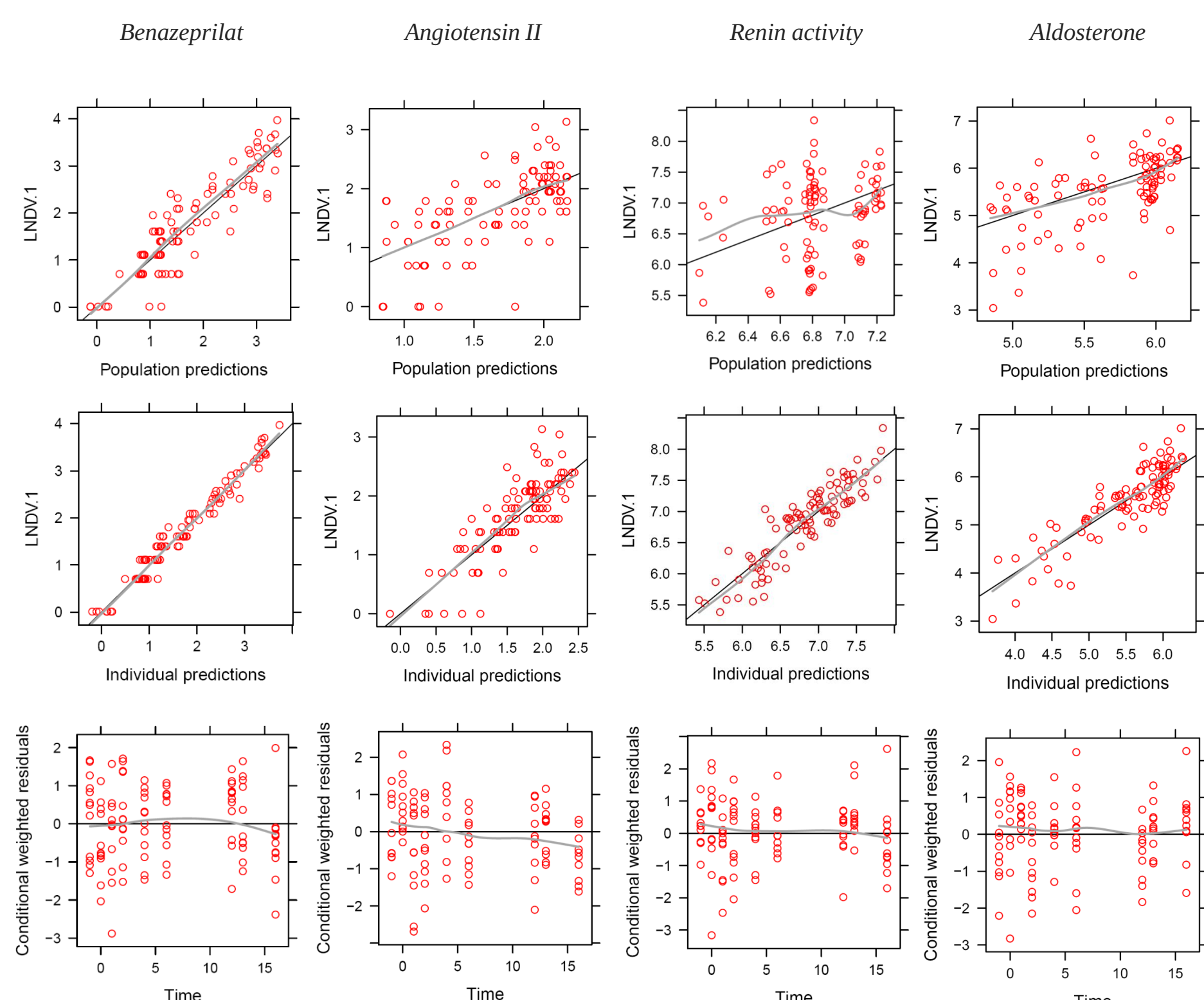
- Twelve healthy beagle dogs (sex ratio 1:1) were fed a low-sodium diet and dosed with placebo or benazepril tablets (5 mg PO, q24h) for 5 days using a crossover design
- Renin activity (RA) and angiotensin II levels (AII) were determined using immunoassay-based methods Benazeprilat and aldosterone concentration (ALD) were quantified in plasma by mass spectrometry
- Population PKPD models were developed using NONMEM version 7.2. Covariate search was performed using the stepwise covariate model building tool of PsN. Standard goodness-of-fit diagnostics, NPDEs, as well as posterior predictive checks and bootstraps were performed to assess the adequacy of selected models

Figure 3. Main parameter estimates of the final population model (point estimate, median obtained from 100 bootstrap runs, and %RSE)

Parameter [units]	NONMEM Point Estimate	Bootstrap Estimate (median)	%RSE
Fixed effect parameters			
k_{10} [hr ⁻¹]	2.03	1.96	39
$D1$ [hr]	1.10	1.21	16
k_1 [ng.hr ⁻¹]	0.00454	0.00368	58
ACE [ng]	24.6	32.8	20
k_{10} [hr ⁻¹]	0.598	0.665	16
$Base$ [pg.mL ⁻¹]	8.21	8.52	12
$Base^*$ [pg.mL ⁻¹]	349	300	37
AP [pg.mL ⁻¹]	-1.82	-1.62	11
IC_{50} [ng.mL ⁻¹]	16.2	17.1	30
I_{max}^*	0.80	0.50	40
E_{max}	4.87	5.76	23
Random effect parameters			
σ^2_{Base}	0.083	0.070	25
$\sigma^2_{Base^*}$	0.282	0.261	21
σ^2_{AP}	0.369	0.454	42
$\sigma^2_{Benazepril}$	0.20	0.19	10
σ^2_{Renin}	0.29	0.31	7

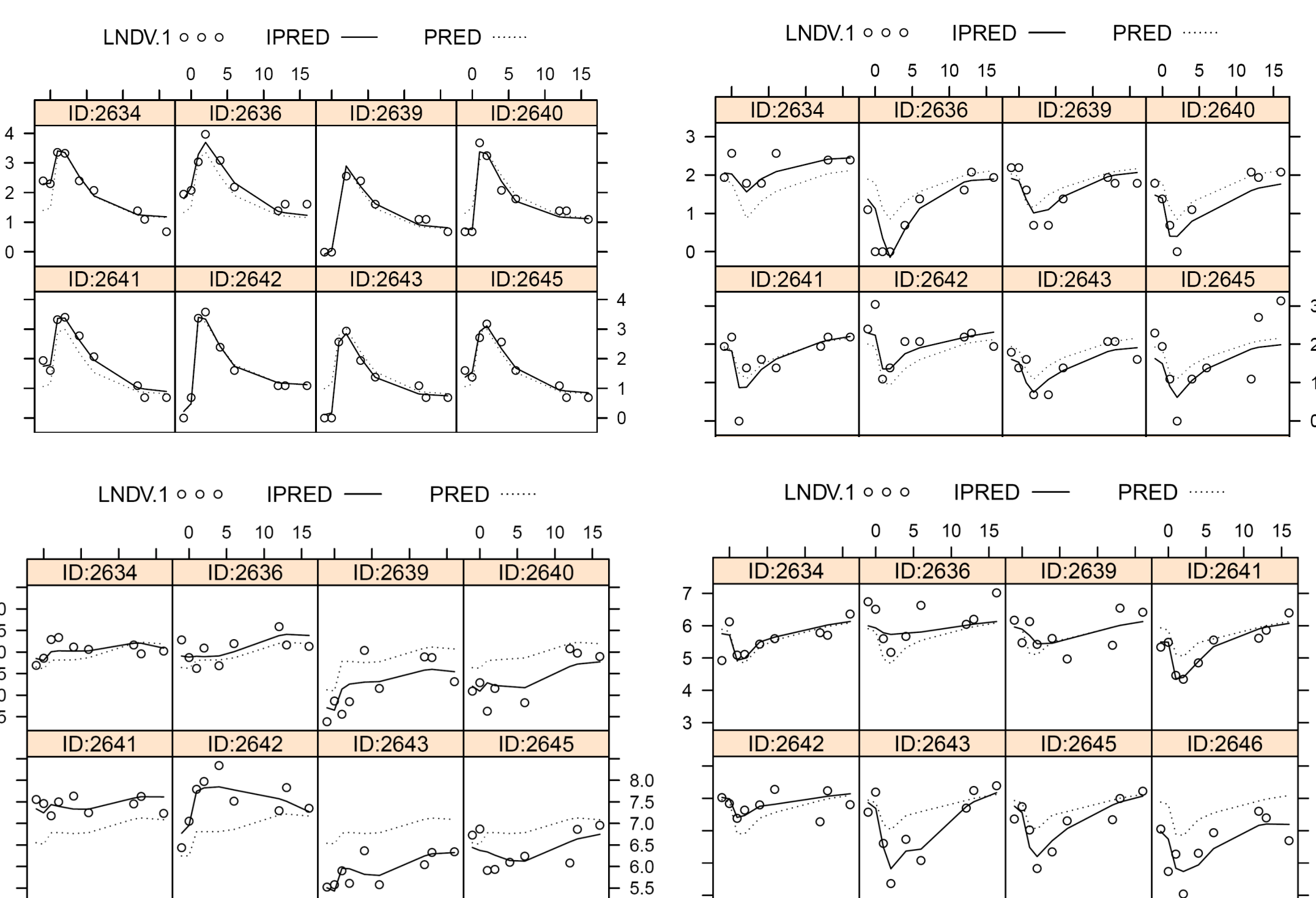
$D1$: estimated duration of the 0 order absorption process; ACE: total binding capacity to the angiotensin converting enzyme; Base: baseline angiotensin II; $Base^*$: baseline renin activity; AP: amplitude of angiotensin II circadian variations; I_{max}^* : maximum aldosterone inhibition; E_{max} : maximum stimulation of renin activity; σ^2 : variance terms; σ^2 : residual error terms; RSE: relative standard errors

Figure 4. Standard goodness-of-fit diagnostics for the final population pharmacokinetic/pharmacodynamic model



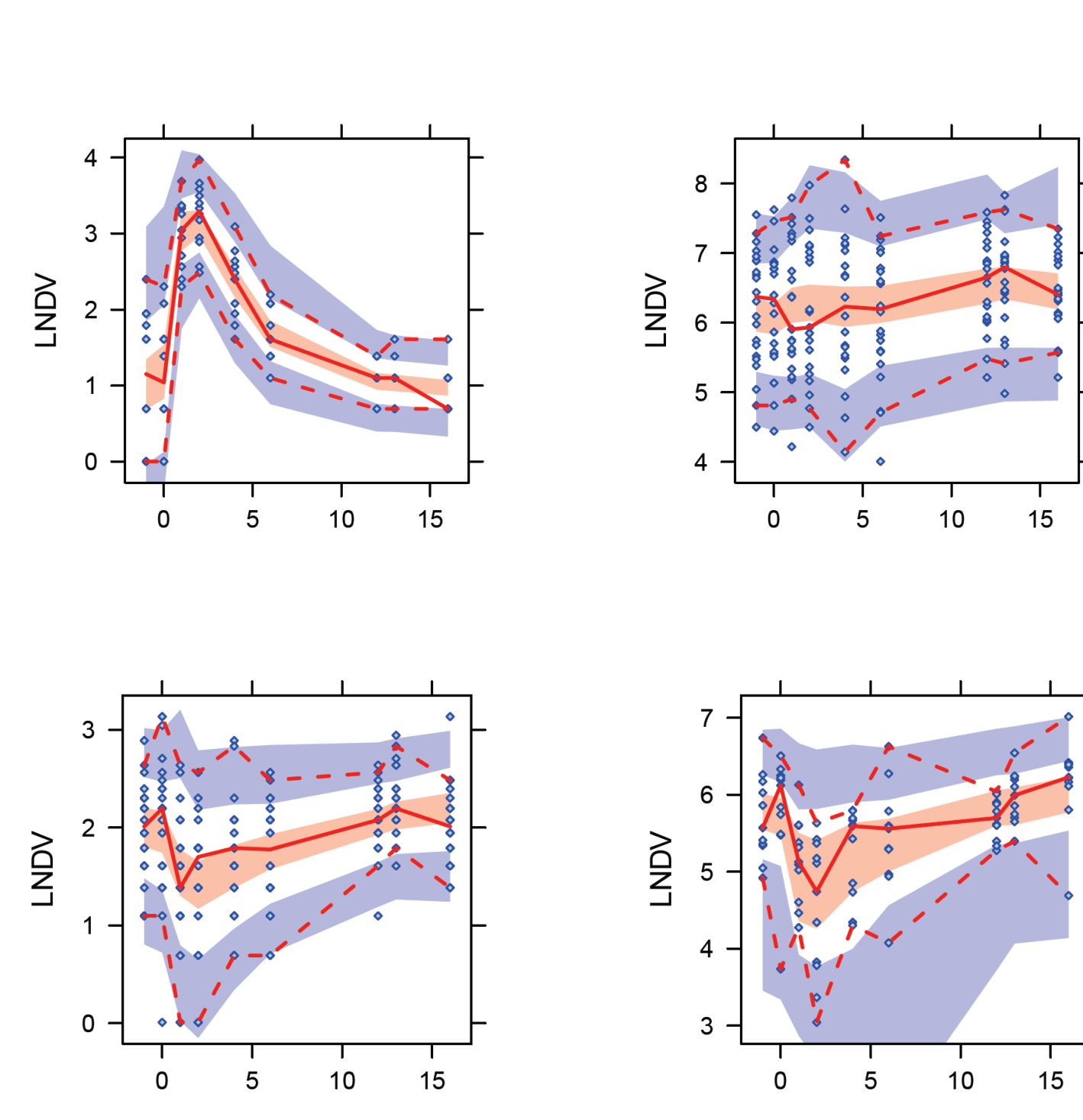
Top pane: population predictions vs. observations (log scale), middle pane: individual predictions vs. observations (log scale), bottom pane: conditional weighted residuals of population predictions vs. time after dose (hours). Solid line: identity line

Figure 5. Individual predictions (log scale) vs. time (hours) based on individual parameters obtained as Empirical Bayes Estimates



Scatter plot of observed (open circles) and predicted (continuous line) benazeprilat (top left pane), angiotensin II (top right pane), renin activity (bottom left pane), and aldosterone (bottom right pane) levels versus time after dose (hours). Dashed line: population predictions, ID: dog identification number. Out of clarity only a subset of individuals are represented herein

Figure 6. Visual predictive checks for the final population pharmacokinetic/pharmacodynamic model



Visual predictive checks for benazeprilat (top left pane), renin activity (top right pane), angiotensin II (bottom left pane), and aldosterone (bottom right pane) generated from 500 Monte Carlo simulations. Dots: observations, dashed red line: 5th and 95th percentile, red continuous line: model predicted median

Conclusions & Perspectives

- The model captured the dynamics of the renin cascade and aided our understanding of the physiology of this system in dogs
- Inclusion of baseline periodicity was necessary to describe benazepril-induced RAAS inhibition over time
- The model can be used to predict the effect and optimize the dose of a next ACE inhibitor
- The model underlines the effect of benazepril on AII and ALD which is assumed to drive the increased survival and improved quality of life in benazepril-treated dogs
- Profiling of RAAS peptides could support the determination of the severity of heart failure, complement clinical and echocardiographic findings, and be used for therapeutic drug monitoring purposes