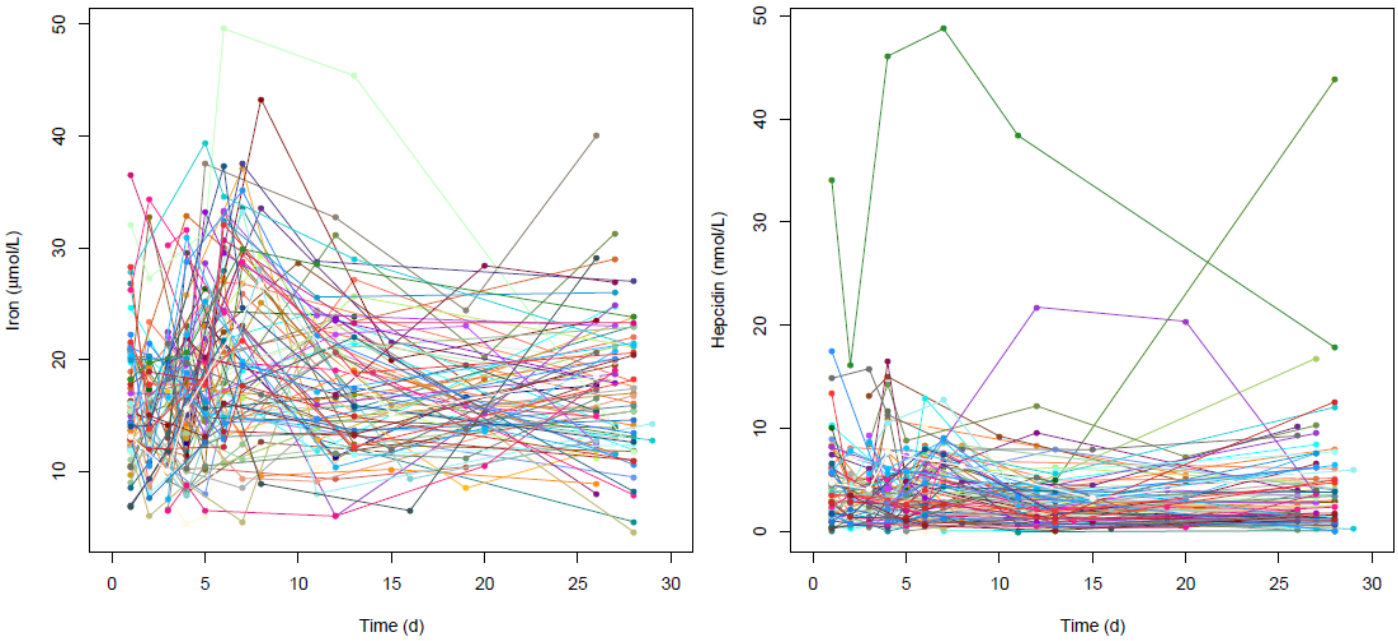
Joint model of iron and hepcidin during the menstrual cycle in healthy women

SUPPLEMENTARY DATA

Raw iron and hepcidin data

Supplementary Figure S1 represents the individual profiles of iron and hepcidin collected in the 90 subjects participating in the Hepmen study.



Supplementary Figure S1: Individual profiles for iron (left) and hepcidin (right) collected during one menstrual cycle in 90 healthy volunteers

SUPPLEMENTARY RESULTS

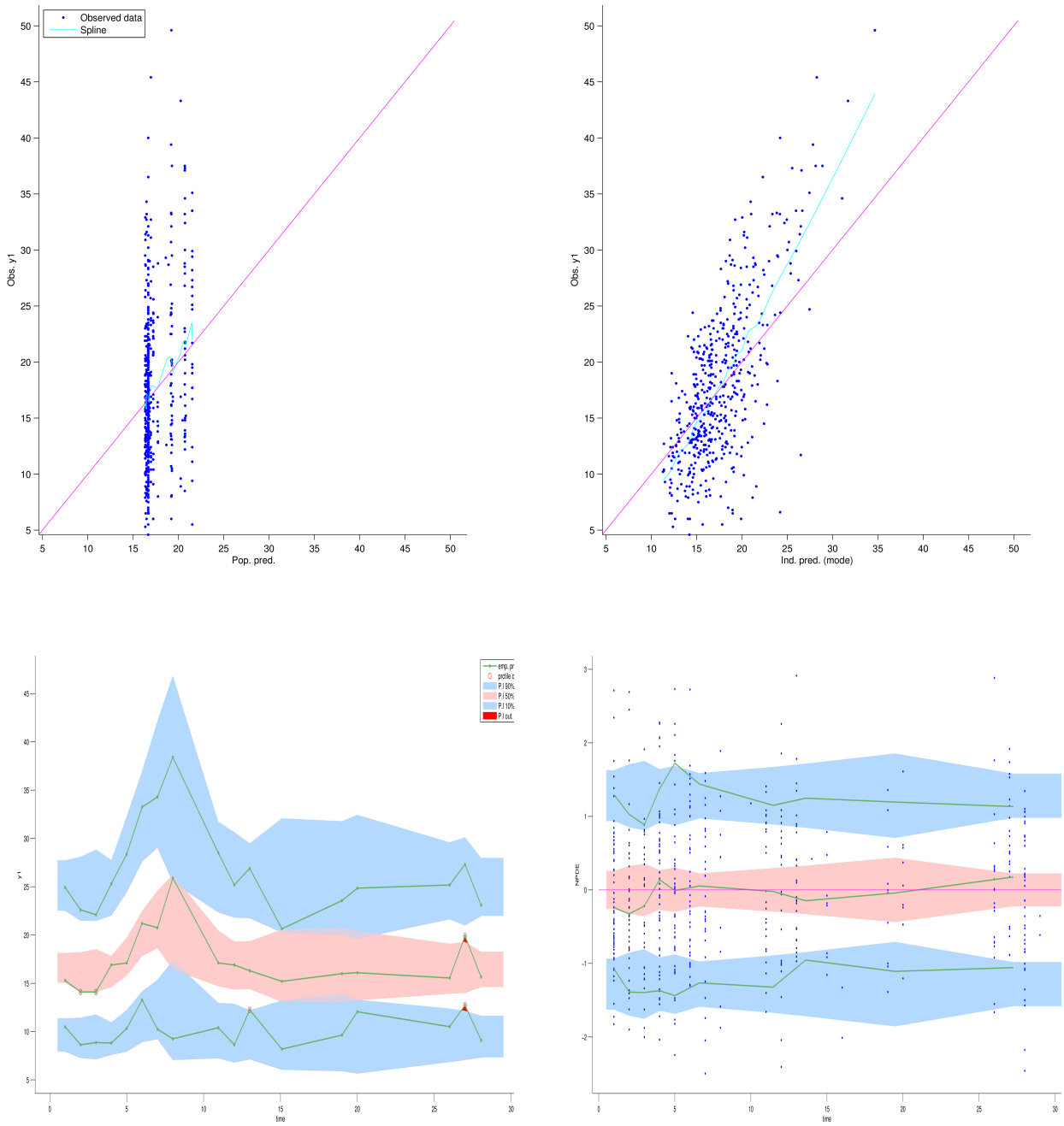
Separate modelling of iron and hepcidin

Table S1 shows the main models tested to describe the evolution of iron and hepcidin separately. A combined error model was used in all the runs to describe residual error variability.

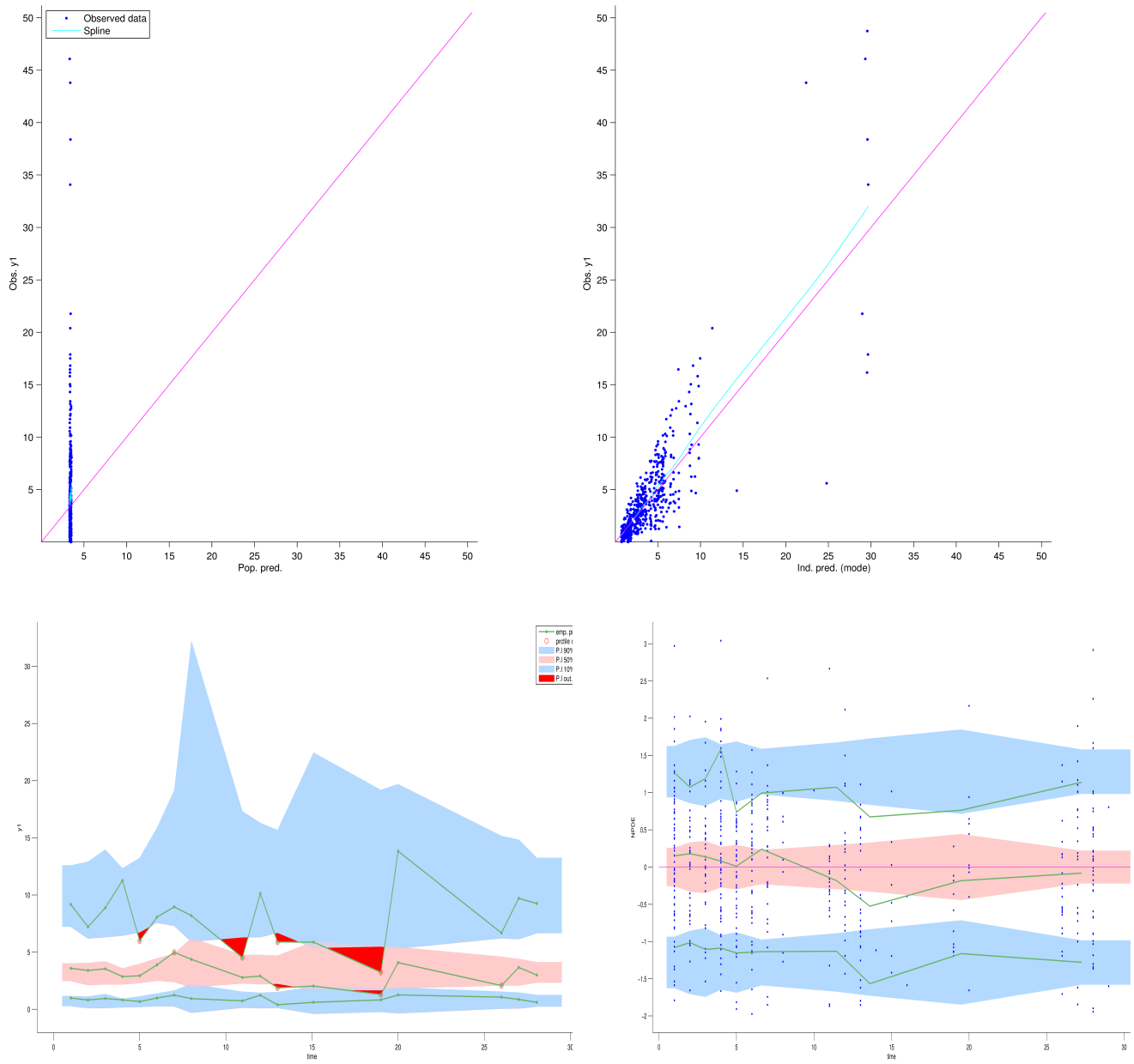
Nb	Description	BIC (IS)
	Modelling iron alone	
I1	Turnover model with iron loss during dloss	3400.5
I2	Turnover model with iron loss with dloss equal to length of menses	3394.0
I3	Push-pull model	3371.7
I4	Tolerance modelled through a precursor compartment	3402.7
I5	Empirical rebound model starting after the end of menses	3354.7
I6	Empirical rebound model starting at Tbegin, for a duration of drel (estimated)	3366.6
I7	Empirical rebound model with Tbegin=2, drel estimated	3358.7
I8	Mixture of models with and without rebound – between subjects	3364.4
I9	Mixture of models with and without rebound – within subjects	3361.7
	Modelling hepcidin alone	
H1	Turnover model with iron loss during dloss	2381.8
H2	Turnover model with iron loss with dloss equal to length of menses	2395.6
H3	Push-pull model	2393.3
H4	Tolerance modelled through a precursor compartment	2389.1
H5	Empirical rebound model starting after the end of menses	2377.5
H6	Empirical rebound model starting at Tbegin, for a duration of drel (estimated)	2382.4
H7	Empirical rebound model with Tbegin=2, drel estimated	2377.4
H8	Cyclic change in ksynH	2311.1
H9	Mixture of models with and without rebound – between subjects	2378.0
H10	Mixture of models with and without rebound – within subjects	2388.4

Supplementary Table S1: Models tested in the separate analyses of iron and hepcidin, with the corresponding Bayesian Information Criterion (BIC). The log-likelihood was computed by Importance Sampling (IS) for all models tested. The BIC for the different models can be compared only within the same variable (iron or hepcidin).

Figure S2 shows diagnostic plots for iron and Figure S3 shows diagnostic plots for hepcidin in the final models obtained in the separate analyses.



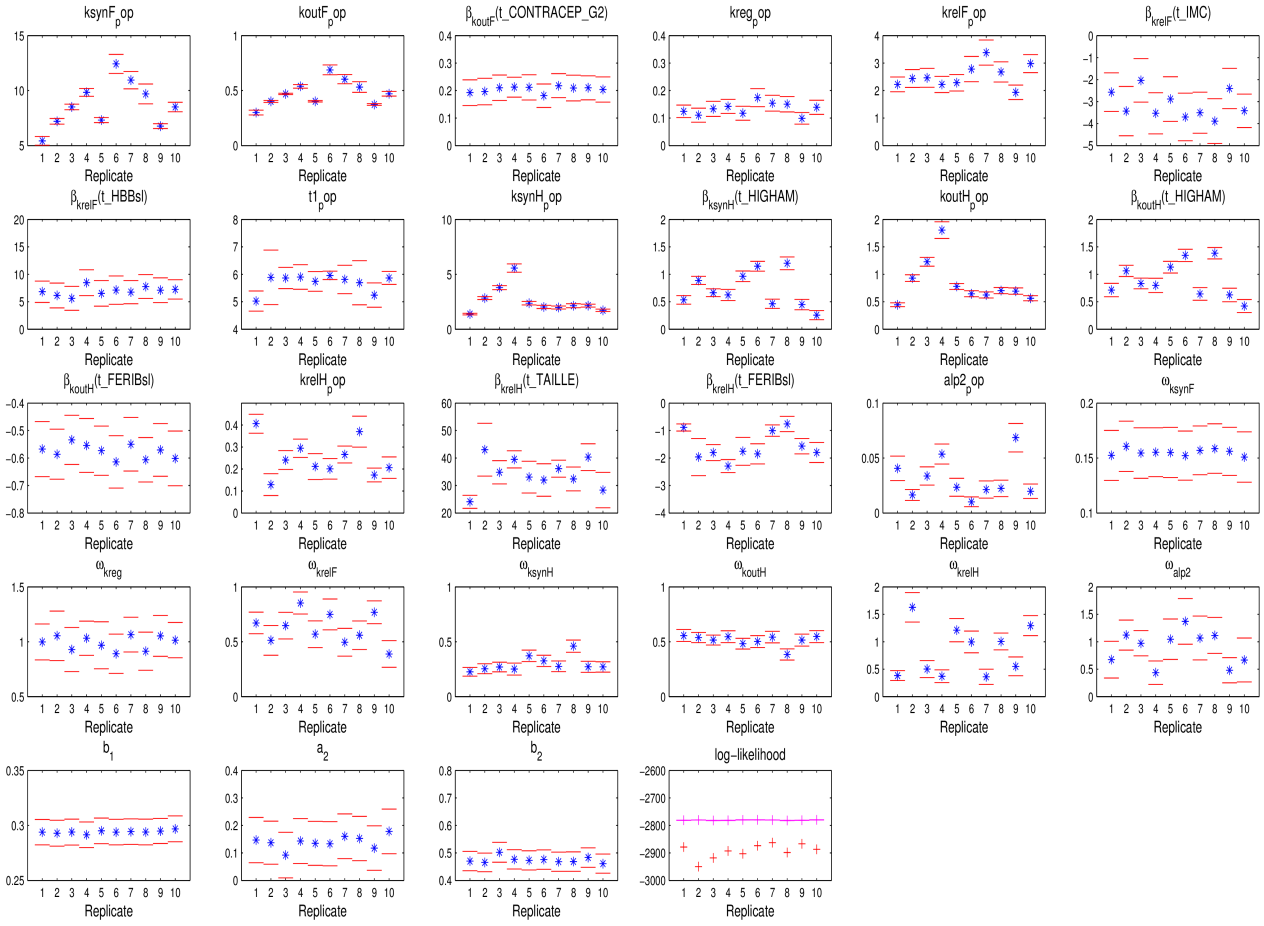
Supplementary Figure S2: Diagnostic plots for the analysis of iron alone. (a) Population predictions versus observations (b) Individual predictions versus observations (c) VPC (d) npde versus time.



Supplementary Figure S3: Diagnostic plots for the analysis of hepcidin alone. (a) Population predictions versus observations (b) Individual predictions versus observations (c) VPC (d) npde versus time.

Joint model of iron and hepcidin – additional information

Figure S4 shows the plots produced by the run assessment feature in Monolix. For each parameter, the ten estimates and their confidence intervals obtained in the ten runs, starting from different random seeds and initial conditions, are shown. Except for the estimate of the fixed effect for Higham score on ksynH, which was close to 0 in the third run, all fixed and variance parameters were significant in all runs.



Supplementary Figure S4: Run assessment plots for the final model. Each plot represents the estimates of one parameter and its associated confidence interval over 10 runs initiated with a different seed for the random number generator and different initial estimates for all parameters.

Supplementary table S2 shows the results of the run assessment procedure run in Monolix. From run to run, the release and elimination of iron from the blood stream tend to vary in the same direction, maintaining the baseline value similar across runs, and the same occurs for hepcidin, explaining some of the variation of these two parameters when different seeds are used. The covariate effects remained significant across all replicate runs. The table also shows the parameter estimates from the sensitivity analysis performed without the 3 subjects with high or abnormal values of hepcidin. Most of the parameter estimates remained very close to those obtained with the full dataset, and within the range of estimates obtained in the run assessment, showing that the estimates are reasonably robust. The residual variability of hepcidin decreased slightly when removing the outliers, from 0.47 to 0.45.

	Main run (N=90)		**Sensitivity (N=87)**		**Run assessment (n=10)**	
Parameter	**Estimate (*RSE*)**	**Variability (*RSE*)**	**Estimate (*RSE*)**	**Variability (*RSE*)**	**Estimate**	**Variability**
ksynI (µmol.d. L^{-1})	7.57 (8)	15 (*15*)	6.46 (8)	16 (14)	5.42-12.40	15-16
koutI (d^{-1})	0.42 (8)	-	0.36 (8)	-	0.30-0.69	-
No contraception	0.20 (*23*)	-	0.20 (25)	-	0.18-0.22	-
kloss (d^{-1})	0.14 (19)	95 (*19*)	0.10 (22)	110 (19)	0.10-0.17	89-106
krelI (µmol.d.L^{-1})	2.55 (*13*)	55 (*22*)	2.38 (14)	53 (24)	1.94-3.38	39-85
β_{BMI}	-3.31 (*31*)	-	-4.22 (24)	-	-3.89 - -2.03	-
$\beta_{Haemoglobin}$	6.93 (31)	-	5.07 (48)	-	5.61-8.47	-
drel (d)	5.74 (5)	-	5.50 (6)	-	5.02-5.96	-
ksynH (nmol.d.L^{-1})	2.48 (5)	24 (*19*)	1.85 (7)	28 (18)	1.38-5.56	22-46
β_{Higham}	0.66 (*11*)	-	0.20 (46)	-	0.26-1.20	-
koutH (d^{-1})	0.82 (7)	57 (9)	0.63 (8)	42 (12)	0.46-1.81	38-57
β_{Higham}	0.83 (*14*)	-	0.46 (24)	-	0.42-1.38	-
$\beta_{Ferritin}$	-0.60 (16)	-	-0.46 (19)	-	-0.61 - -0.53	-
krelH (nmol.d.L^{-1})	0.28 (21)	103 (*17*)	0.22 (24)	129 (14)	0.13-0.41	36-163
β_{Height}	32.70 (*16*)	-	30.70 (23)	-	24.1-43.0	-
$\beta_{Ferritin}$	-1.95 (18)	-	-1.10 (25)	-	-2.29 - -0.76	-
α (-)	0.03 (*31*)	87 (38)	0.03 (25)	66 (37)	0.01-0.07	44-137
b_{Iron}	0.29 (4)		0.29 (4)		0.29-0.30	
a_{Hep}	0.17 (*50*)		0.19 (46)		0.09-0.18	
b_{Hep}	0.47 (*8*)		0.45 (*8*)		0.46-0.50	

RSE, relative standard error, in %. The interindividual variability for each parameter is expressed in %.

Supplementary Table S2: Parameter estimates obtained in the final model with (main run) and without (sensitivity) the three subjects with high values of hepcidin. The last columns give the ranges obtained in the 10 run assessments on the full dataset.