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Prediction of hemoglobin in blood donors using a latent class mixed-effects transition model

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Blood donors experience a temporary reduction in their hemoglobin (Hb) value after donation. At each visit, the Hb value is measured, and a too low Hb value leads to a deferral for donation. Because of the recovery process after each donation as well as state dependence and unobserved heterogeneity, longitudinal data of Hb values of blood donors provide unique statistical challenges. To estimate the shape and duration of the recovery process and to predict future Hb values, we employed three models for the Hb value: (i) a mixed-effects models; (ii) a latent-class mixed-effects model; and (iii) a latent-class mixed-effects transition model. In each model, a flexible function was used to model the recovery process after donation. The latent classes identify groups of donors with fast or slow recovery times and donors whose recovery time increases with the number of donations. The transition effect accounts for possible state dependence in the observed data. All models were estimated in a Bayesian way, using data of new entrant donors from the Donor InSight study. Informative priors were used for parameters of the recovery process that were not identified using the observed data, based on results from the clinical literature. The results show that the latent-class mixed-effects transition model fits the data best, which illustrates the importance of modeling state dependence, unobserved heterogeneity, and the recovery process after donation. The estimated recovery time is much longer than the current minimum interval between donations, suggesting that an increase of this interval may be warranted.

Keywords: latent class; mixed-effects model; transition model; blood donation; change point model

1. Introduction

Blood donation helps to save millions of lives each year and is an essential part of modern healthcare. Many blood donors come for donation on a regular basis. A blood donation implies a loss of erythrocytes and iron, resulting in a temporary decrease in hemoglobin (Hb) values. The minimum interval between two donations is internationally set at 8 weeks, but this interval seems to be too short for the body to completely recover the Hb value to its pre-donation value. Previous studies have shown that among donors with many visits, there is a decline in their Hb values at subsequent donations [1,2]. Low Hb values may potentially lead to anemia, which should be prevented. In the majority of blood banks, the Hb value is first measured to screen the potential donor for eligibility to give blood [3,4]. To protect potential donors from developing low Hb values, a Hb value of 8.4 mmol/l (135 g/l) and 7.8 mmol/l (125 g/l) for men and women, respectively, is widely accepted as the lower cut-off value of eligibility for donation [3,4].

Each year a considerable proportion of prospective blood donors are temporarily deferred from donation because of low Hb values [5,6]. Hb deferrals decrease the cost-effectiveness of blood supply, because (i) testing and deferring a donor are expensive; (ii) for every deferred donor, another donor needs to be invited to reach collection targets; and (iii) lapsing donors need to be replaced by new donors because deferred candidates rarely return for donation [7] .

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To limit the number of deferrals, it is important that blood banks are able to predict when donors have sufficiently recovered after a blood donation to be invited for a new donation. However, prediction of the Hb value for the subsequent visit of a blood donor is not straightforward. Longitudinal data of Hb values of blood donors have a high within-subject and between-subject variability due to unobserved individual heterogeneity and state dependence [8]. This requires a complex statistical model to capture both sources of variation for proper inference. Predictions of future Hb values of blood donors may be used to improve the invitation policy of potential blood donors.

Hence, for a model to be appropriate, two sources of variation among Hb values must be taken into account. There are various statistical techniques for analyzing longitudinal data. Two well-known approaches are (i) the mixed-effects model, which can capture unobserved individual heterogeneity; and (ii) the transition (auto-regressive) model, which can capture state dependence within an individual's observations. However, it may be that neither approach can adequately explain the correlation structure alone, because of the presence of both unobserved heterogeneity and state dependence. For these reasons, we may combine the mixed-effects model and the transition model by including the unobserved individual-specific effects and a lagged endogenous variable in a single regression model. Such a mixed-effects transition model is popular in econometrics for forecasting [9] but less commonly used in medical applications [10].

This paper aims to build an appropriate mixed-effects transition model for a longitudinal data set of Hb values collected from new entrant whole-blood donors from 2005 to 2012 in the Netherlands. We wish to predict not only the future Hb value but also the recovery time that is needed for Hb to return to its pre-donation value after a blood donation. This will help to improve the planning of the donors' visits to the blood bank. Predicting Hb value has been studied previously by Rikhtegaran *et al.* [11] and Nasserinejad *et al.* [12]. In the latter study, the authors applied mixed-effect models and transition models separately to predict the Hb value. In the former study, a mixed-effects transition model was applied on a subset of the longitudinal data, excluding all measurements after the candidate was deferred from donation for any reason. Recently, Nasserinejad *et al.* [13] used a latent class mixed-effects model to show how the trajectories of Hb values differ between donors.

The main contribution of this paper is the development of a realistic model for longitudinal Hb values. This model takes into account the unique aspects of these data, namely, (i) heterogeneity of the initial Hb value; (ii) state dependence of a donor's Hb values; (iii) varying time intervals between donations; (iv) the temporary reduction in Hb after blood donation; and (v) the fact that the recovery process may change with the number of donations and may differ between donors. To do so, the mixed-effects transition model is combined with a flexible non-linear specification for the recovery process, which enables us to estimate the shape and duration of the recovery, for which precise estimates are not available in the clinical literature. Latent classes are used to account for the heterogeneity of the recovery process between donors, and a time change point specification is used to determine how the recovery time changes with the number of donations. The observed data do not provide sufficient information to identify all model parameters in a frequentist setting. A further contribution of this paper is that it is shown how the model parameters can be estimated in a Bayesian way, using suitable prior information from the clinical literature. Finally, it is shown how to forecast a future Hb value based on the available history of donations in a fully Bayesian approach.

The remainder of the paper is organized as follows: Section 2 presents the motivating data set. Section 3 introduces the statistical models for the recovery process and the donation problem described earlier. Results are presented in Section 4. Section 5 deals with prediction in the latent class mixed-effects transition model. We discuss the findings of the study in Section 6.

2. Sanquin blood donor data set

The Donor InSight data set was collected by Sanquin Blood Supply in the Netherlands [14]. This data set is based on a self-administered questionnaire study aimed at gaining insight into characteristics and motivation of the Dutch donor population [14]. Our analysis comprises whole-blood donors who were registered as new donors from 1 January 2005 to 31 December 2012. Whole blood is a term used in transfusion medicine for a standard blood donation as opposed to plasma and platelet donation. To be

included in the study, the donors must have had at least one visit after the first donation. A total of 4461 donors (1552 male and 2909 female donors) fulfilled these criteria. The descriptive statistics of these donors are presented in Table I.

For blood collection, all measured data were entered into the blood-bank computer system (e)PROGESA (MAK-SYSTEM International Group, France). Prior to donation, Hb and other parameters undergo a check to determine whether the prospective donor is eligible. In the Netherlands, a newly registered donor is not allowed to donate blood at the first visit, the screening visit, which consists of a health check only. At every subsequent visit, donors who pass all eligibility checks may donate 500 ml of whole blood. Finally, guidelines impose a minimum interval of 56 days between donations, with a yearly maximum of 5 donations for men and 3 for women [15]. In Figure 1, profiles of the Hb value after the screening visit for a subset of male and female donors are displayed. The horizontal lines represent the eligibility thresholds for donation.

Several factors are known to be associated with the Hb value and hence may be used as predictors for Hb, i.e., sex [16], season [17], age [16], and body mass index (BMI) [16]. In this study, we take into account the effect of age at first visit and the season of the visit (a binary covariate, i.e., the cold season includes fall and winter and the warm season includes spring and summer). Because male and female donors have different Hb profiles, we analyze the data for men and women separately.

The Donor InSight study was approved by the Medical Ethical Committee Arnhem-Nijmegen in the Netherlands, and all participants gave their written informed consent.

Table I. Descriptive statistics of the Donor InSight data set.

	Male	Female
Age at 1st visit* (year)	35 (25–47)	30 (22–43)
Number of donations*	12 (6–19)	7 (4–13)
4–5th visit intervals* (day)	93 (77–133)	137 (121–189)
9–10th visit intervals* (day)	88 (88–124)	133 (118–175)
19–20th visit intervals* (day)	84 (70–109)	128 (113–147)
Deferral due to low Hb	5.29%	11.38%
Donors with at least one deferral due to low Hb	36.92%	54.40%

* Median and inter-quantile range.

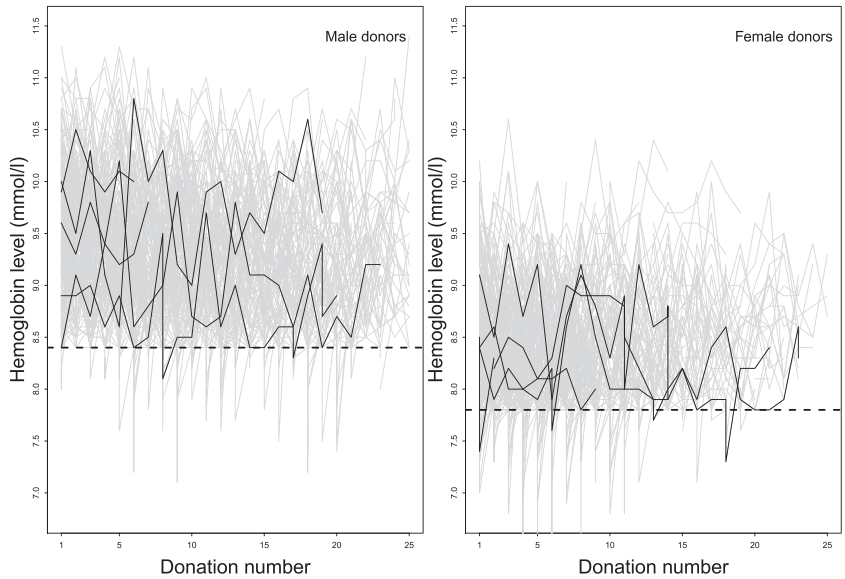

Figure 1. Hb profiles after the screening visit for a subset of male and female donors. Five random profiles are highlighted for both sexes. The bold dashed lines show the corresponding thresholds of eligibility for donation.

3. Statistical model for Hb values

In this section, we propose a mixed-effects model, a latent class mixed-effects model and a latent class mixed-effects transition model for the Donor InSight data. Formally, let Hb_{it} denote the Hb recorded for the ith individual ($i = 1, \ldots, N$) at T_i different times ($t = 1, \ldots, T_i$), together with a set of p strictly exogenous covariates $x_{it} = \{x_{i1t}, x_{i2t}, \ldots, x_{ipt}\}'$. Because the donation intervals are not equal, the data set is unbalanced (Table I). Furthermore, there is a reduction in Hb value after donation. Because the current Hb value is associated with the Hb value observed at the previous visit [11, 12] and the time since the previous donation [11, 18], we need to take into account the time interval the since previous donation and the reduction in Hb due to donation. All of these aspects must be incorporated into the statistical model for predicting Hb.

3.1. Hb recovery after blood donation

The Hb recovery process after blood donation is illustrated in Figure 2. In this figure, δ indicates the time that Hb reaches its minimum value after donation. After donation of 500 ml of blood, on average, a male donor loses 242 mg and a female donor 217 mg of iron [19]. This will cause Hb to decrease and to reach its nadir a few days after donation [20–22]. The Hb value will then gradually recover to its pre-donation value [20–22]. The recovery time needed for Hb_{it} to return to its pre-donation value is given by recovery time (RT).

Unfortunately, in this study, we have only the Hb value prior to donation at each visit, and there is no information on the Hb value between two invitations. The observed interdonation interval is at least 56 days in our data set. Therefore, we cannot accurately estimate the trajectory of Hb value during the first 56 days after donation. We incorporate into our model that the reduction of Hb after giving blood takes around 3 days [20, 22]. To estimate the amount of reduction after donation, we use the results of a recent randomized clinical trial on iron supplementation after blood donation. The donor characteristics and the donation policy in this trial were similar to the Dutch setting of the Donor InSight study; the amount of blood given per donation and the minimum interval between donations are the same for both data sets. The data of this trial showed that, 3 through 8 days after donation of 500 ml of whole blood, the amount of reduction in the Hb value had a mean (95% CI) of 0.68 (0.59, 0.77) and 0.96 (0.89, 1.03) mmol/l for male and female donors, respectively [22]. We use the following Hb recovery function (HRF):

$$HRF_{it} = \theta \left[\max \left(\frac{RT - (TSPD_{it} - \delta)}{RT}, 0 \right) \right]^{\psi}, \tag{1}$$

where HRF_{it} is the amount of Hb that needs to be recovered for person i at time t. δ and RT were defined earlier, and θ is the amount of Hb reduction. TSPD_{it} represents the time since the previous donation for the ith donor at time t. The parameter ψ indicates the shape of the recovery function of the Hb value after donation. Values of ψ smaller than one indicate a convex trajectory for Hb recovery, while values greater than one indicate a concave trajectory for Hb recovery (Figure 2). For $\psi = 1$, the Hb recovery is linear.

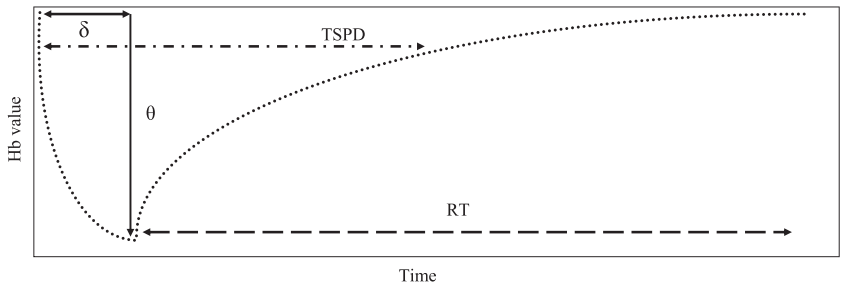

Figure 2. Hb recovery process after blood donation: δ indicates the time that Hb reaches its minimum value after donation, RT indicates the recovery time that is needed for Hb to return to its pre-donation value, and θ shows the amount of reduction in Hb after donation.

The recovery time may differ between donors because of diet, genetic factors, and other unobserved characteristics. In addition, some donors experience a reduction of their iron reserve after a few donations, which may cause the recovery time to increase with the number of donations. Therefore, the postulated function based on (1) can oversimplify the recovery process, because the RT may depend on the number of previous donations as well as the donor. However, a too flexible function (e.g., a different recovery time for each donor) is likely not to be estimable from the data. So, we use finite mixture modeling to capture the heterogeneity in recovery time by assuming that donors belong to two different latent classes. Namely, one class of donors is assumed to have a constant recovery time, and the other class is assumed to exhibit a nonconstant recovery time. The function could be

$$HRF_{it,g(i),\kappa} = \theta \left[\max \left(\frac{RT_{g(i),\kappa} - (TSPD_{it} - \delta)}{RT_{g(i),\kappa}}, 0 \right) \right]^{\psi}, \tag{2}$$

where $g(i)$ is the latent class membership of donor i, which has to be estimated from the data. It is assumed that donors who belong to (latent) class I ($g(i) = 1$) have a constant recovery time. For donors in class II, it is assumed that the recovery time changes after a certain number of donations (κ). This change point κ is assumed equal for all people in class II and must be estimated from the data.

3.2. Mixed-effects model

To accumulate the effects of the donation and recovery process, a non-linear model making use of (1) or (2) is needed. To also take into account the unobserved individual heterogeneity a random intercept is included. Therefore, a possible model with (1) as recovery function could be

$$\text{Model 1:} \quad Hb_{it} = \beta_0 + b_{i1} + \beta_1 Age_{i1} + \beta_2 Season_{it} - HRF_{it} + \varepsilon_{it}, \quad t = 1, ..., T_i, \tag{3}$$

where b_{i1} is the random intercept in the model to control heterogeneity. It is assumed that $\epsilon_{it} \overset{\text{iid}}{\sim} N(0, \sigma^2_{\epsilon})$ and $b_{i1} \overset{\text{iid}}{\sim} N(0, \sigma^2_{b1})$ and that b_{i1} and ϵ_{it} are mutually independent.

3.3. Latent class mixed-effects model

To take into account the heterogeneity in recovery time in Model 1, a possible model based on (2) as recovery function could be

$$\text{Model 2:} \quad Hb_{it} = \beta_0 + b_{i1} + \beta_1 Age_{i1} + \beta_2 Season_{it} - HRF_{it,g(i),\kappa} + \varepsilon_{it}, \quad t = 1, ..., T_i. \tag{4}$$

3.4. Latent class mixed-effects transition model

To take into account the state dependence among Hb values of the same individual, one can add a first lag of Hb as an autoregressive term to Model 2. Because the recovery process depends on the time since the last donation, we also use the Hb value at the last donation (instead of the last visit) as an autoregressive term. This model is given by

$$\text{Model 3:} \quad Hb_{it} = \beta_0 + b_{i1} + \beta_1 Age_{i1} + \beta_2 Season_{it} + \gamma Hb.pd_{it} - HRF_{it,g(i),\kappa} + \varepsilon_{it}, \quad t = 2, ..., T_i, \tag{5}$$

where b_{i1} controls the heterogeneity now partly explaining the intra-subject correlation; γ is the lagged impact of Hb at the previous donation ($Hb.pd_{it}$). Note that $Hb.pd_{it}$ is equal to $Hb_{i,t-1}$ if the last visit was a donation. For a stationary process, i.e., $|\gamma| < 1$, the correlation between two subsequent measurements can be expressed as [11]

$$\rho_{Hb_{it},Hb_{it-1}} = \gamma + \frac{1 - \gamma}{1 + (1 - \gamma)\sigma^2_{\epsilon} / \left[(1 + \gamma)\sigma^2_{b1}\right]}. \tag{6}$$

When the lag impact γ is negligible, this correlation reduces to the intra-class correlation (ICC) in the mixed-effects model. On the other hand, when there is no heterogeneity between individuals, i.e., $\sigma^2_{b1} \approx 0$, the correlation is equal to the lag impact only.

3.5. The initial conditions problem

One of the assumptions in classical mixed-effects models is that the covariates in the model are exogenous, i.e., the covariance between covariates and the random effects is 0. But this assumption is violated in mixed-effects transition models where one of the covariates is the lagged variable, which is endogenous. This issue relates to the initial conditions problem (ICP), which is well known in the econometric literature. The ICP occurs because the individual effects, b_{i1}, that capture the unobserved heterogeneity are correlated with the initial Hb values, i.e., $\text{cov}(Hb_{i1}, b_{i1}) \neq 0$ [23, 24]. Ignoring the ICP and thus the endogeneity of Hb_{i1} results in inconsistent estimates in the model [23, 24], i.e., an upward bias of the estimated state dependence and a downward bias in the estimated coefficients of explanatory variables [24].

A possible solution to the ICP problem is to incorporate the association of the initial value and the random effects jointly into the model for the subsequent Hb values. The model is assumed to be similar to the dynamic equations (Models 2 and 3) but without the lagged response variables [24]. Using this solution, the regression parameters as well as the residual variance are allowed to differ between the initial and the subsequent observations. The joint-modeling approach enables one to capture the correlation between the individual effects, b_{i1}, and the initial Hb values and provides reliable estimates for the regression parameters [24].

A possible model for the initial Hb values could be

$$\text{Model } 3_0: \quad Hb_{i1} = \beta_{00} + \beta_{01} Age_{i1} + \beta_{02} Season_{i1} + b_{i0}, \tag{7}$$

where $b_{i0} \sim N(0, \sigma^2_{b0})$. Furthermore, we allow for a correlation ρ_{01} between b_{i0} and b_{i1} in Model 3.

Because of the complexity of the models as well as the lack of information on the first 56 days of the recovery process, we opted for a Bayesian approach with Markov chain Monte Carlo (MCMC) sampling to estimate the parameters in these models.

3.6. Prior specification

Vague normal priors were specified for the β's in Model 1, Model 2, and Model 3_0, and for the β's and γ in Model 3, i.e., $N(0, 10^3)$. Because Hb is measured only at visits to the blood bank and the minimum interval between donations is 56 days, the current data provide little information on the parameters θ and δ. Therefore, we used informative priors for the amount of Hb reduction after donation, θ, and the time at which Hb value reaches its minimum value, δ. The parameters of the informative prior distributions are based on previous clinical studies [20]. For θ, we specified a normal prior with mean 0.68 and standard deviation 0.038 for male donors and a normal prior with mean 0.96 and standard deviation 0.045 for female donors, based on the results of a recent clinical trial [22]. For δ, a normal prior with mean 3 and variance equal to 0.1 was specified. For the RT, a positive uniform distribution with a wide range, i.e., $U(0, 10^4)$ was specified. For the parameter ψ, which indicates the shape of the recovery trend, an exponential of normal distribution, i.e., $N(0, 10)$ was specified. An $IG(\varepsilon, \varepsilon)$ prior with small value for ε, i.e., 10^{-3}, was specified for the variance of the residuals. An $Inv - Wishart(R, df)$ distribution was specified for the variance–covariance structure of the residuals b_{i0} in Model 3_0 and the random intercept b_{i1} in Model 3. We set the degrees of freedom, df, to 3 and the scale parameter matrix, R, to a diagonal matrix with small values, i.e., 10^{-3} [25]. For the class membership probability, a Dirichlet distribution with small values (i.e., 0.1) for the mixing distribution was specified [26]. We chose a discrete uniform distribution for κ with range from 1 to the maximum number of donations, i.e., 43 and 26 for male and female donors, respectively. Finally, prior sensitivity analyses were performed for the non-informative prior distributions.

3.7. Model fit and assessment

To be able to validate the model, we randomly divided the available donors into a training data set (2,231 donors) and a validation data set (2,230 donors). All statistical models were estimated using the training data set only. Parameter estimates were obtained using the JAGS [27] interface to R [28]; the JAGS syntax is shown in the Appendix. The first 10,000 iterations (i.e., burn-in iterations) of each chain were discarded. The posterior medians and 95% HPD credible intervals (CI) were calculated using the remaining iterations of each chain, using a thinning factor of 10. We checked the convergence by running two chains from dispersed initial values and examining standard Bayesian diagnostics, such as trace plots,

the Brooks-Gelman-Rubin statistics [29], and the Geweke diagnostic [30]. To check whether the number of MCMC iterations was sufficient to obtain accurate estimates, sampling was continued until the Monte Carlo errors were less than 5% of the posterior standard deviation of each parameter [25]. The label switching problem could not occur in Models 2 and 3, because one class in these models was restricted to have a stable recovery time.

To find the best fitting model for the data, we computed the deviance information criterion (DIC) [31]. The DIC was computed outside Jags using a self-written R program [28] based on the MCMC and the data used to fit the models. For this calculation, the expectations of the class-membership and time-change-point parameters were chosen based on the mode, and for the other parameters, this expectation was based on the median. Finally, to assess goodness of fit, we used an omnibus posterior predictive check (PPC) [32]. We generated Hb values given the parameters (Φ) from a random sample of draws from the posterior distribution and calculated the chi-square statistic

$$X^2 = \sum (Hb_{it} - \mathrm{E}(Hb_{it}|\Phi))^T \mathrm{Var}(Hb_{it}|\Phi)^{-1} (Hb_{it} - \mathrm{E}(Hb_{it}|\Phi))$$

for both replicated and observed Hb values at each iteration. Then we computed a Bayesian P-value, i.e., the probability that X^2 based on the replicated data from the model is more extreme than the X^2 based on the observed data. Small or large values of this P-value (e.g., < 0.05 or > 0.95) indicate a poor fit of the model to the data [32].

3.8. Simulation study

To evaluate the ability of the proposed model to estimate the true parameters, we performed a simulation study using 20 artificially generated data sets. In each data set, Hb values and covariates of 200 male donors were generated according to Model 3 and Model 3_0, using the posterior medians of the parameters estimated using the Sanquin data set. In the simulated data, donors were only allowed to donate if the Hb value was above the cut-off for eligibility. The distributions of age, season, and the number of visits per donor were simulated from the observed data for male donors. For each model parameter, the posterior median and the exceedance probability (i.e. the posterior probability that the estimated parameter is greater than the true value) were calculated for each generated data set.

4. Results

4.1. Donor InSight study results

The DICs for different models are presented in Table II. The results show that Model 3 is the best model for both sexes. For this model, the Bayesian P-value of the PPC is 0.48 and 0.43 for males and females, respectively, which indicates that the model assumptions appear to be satisfied. The parameter estimates based on Model 3 (and Model 3_0) for both sexes are presented in Table III.

Based on the highest posterior probability, donors can be assigned to latent classes. For Model 3 (and Model 3_0), 48.7% and 45.7% of the male and female donors, respectively, are assigned to the class with a non-constant recovery time (class II). In class II, the recovery time change point (κ) for male and female donors is at the 7th and 4th donation, respectively.

To contrast the two latent classes, sex-specific descriptive statistics including the average of the time since previous donation (day), age (year), percentage of deferral due to low Hb values, percentage of

Table II. The effective number of parameters (p_D), and the deviance information criterion (DIC) for different models for each sex.

	Male		Female	
Model	p_D	*DIC*	p_D	*DIC*
Model 1	691	16,189	1237	19,576
Model 2	809	15,360	1475	19,211
Model 3 (and Model 3_0)	557	14,230	1185	17,397

Table III. The posterior medians and 95% HPD CIs based on Model 3 and Model3_0 for male and female donors, separately.

	Male donors			Female donors		
Parameter	Estimate	95% CI		Estimate	95% CI	
β_0	7.87	7.65	8.09	6.96	6.78	7.15
β_1	−0.004	−0.006	−0.002	0.003	0.002	0.005
β_2	−0.082	−0.100	−0.064	−0.068	−0.084	−0.053
γ	0.19	0.17	0.21	0.17	0.15	0.19
θ	0.70	0.64	0.76	0.89	0.82	0.96
δ	2.98	2.39	3.60	2.98	2.32	3.58
ψ	2.18	1.49	3.61	3.61	2.26	4.04
κ	7	7	8	4	4	5
RT_{g_1}	100	69	145	54	20	129
$RT_{g_{21}}$	117	80	168	343	270	450
$RT_{g_{22}}$	419	293	663	503	394	665
σ_{b1}	0.34	0.31	0.35	0.29	0.28	0.31
σ	0.46	0.45	0.47	0.44	0.43	0.45
$\rho_{b_{i1},Hb_{i1}}$	0.67	0.64	0.69	0.61	0.58	0.63

Table IV. Sex-specific descriptive statistics for the two classes pertaining to Model 3 (and Model 3_0).

	Male		Female	
	Class I	Class II	Class I	Class II
Class size	54.3%	45.7%	51.3%	48.7%
Age at 1st visit* (year)	36 (27–46)	33 (25–45)	31 (22–46)	27 (21–41)
Number of donations*	11 (6–17)	12 (6–20)	8 (4–13)	6 (4–11)
Visit intervals* (day)	90 (73–124)	86 (72–115)	133 (119–175)	134 (119–180)
Deferral due to low Hb	0.9%	4.7%	4.5%	13.7%
Donors with at least one deferral due to low Hb	6%	31%	25%	57%

* Median and inter-quantile range.

Table V. Average class probabilities by latent classes for both sexes based on Model 3 (and Model 3_0).

	Mean of posterior probabilities			
	Male		Female	
Latent Class	Class I	Class II	Class I	Class II
Class I	0.70	0.30	0.72	0.28
Class II	0.28	0.72	0.29	0.71

donors with at least one deferral due to low Hb values, and number of donations are presented in Table IV. These results show that the two classes are dramatically different in their percentage of deferral and the percentage of donors with at least one deferral due to low Hb values.

To determine the class membership discrimination, we computed the mean posterior probability of class membership for donors and report the results in Table V. The mean posterior probability of the class to which a donor is assigned is approximately 70%.

The estimated recovery time (95% CI) in class I is 100 (69, 145) and 54 (20, 129) days for male and female donors, respectively. These values are 117 (80, 168) and 343 (270, 450) days for male and female donors, respectively, prior to the change point in class II and increase to 419 (293, 663) and

503 (394, 665) days after the change, much larger than the minimum (56 days) intra-donation interval. These results show a longer recovery time for female donors than for male donors in class II, though the uncertainty in the estimates is large.

The lag impact of Hb for male and female (0.19 and 0.17, respectively) is highly significant, which shows that there is strong evidence for the state dependence between successive Hb values. This can also be induced from the estimated correlation between two successive observations in Models 2 and 3. For Model 2, the correlation (95% CI) is estimated as 0.45 (0.42, 0.48) and 0.42 (0.39, 0.44) between two successive observations in male and female donors, respectively (not shown here). For Model 3, these correlations increase to 0.54 (0.51, 0.57) and 0.49 (0.46, 0.52) for male and female donors, respectively.

The posterior distributions for θ and δ are very close to the corresponding prior distributions. The prior and posterior means (95% CI) of θ for male donors were 0.68 (0.59, 0.77) and 0.70 (0.64, 0.76), respectively; these values for female donors were 0.96 (0.82, 1.03) and 0.89 (0.82, 0.96), respectively. The prior and posterior means (95% CI) of δ for male donors were 3.00 (2.38, 3.62) and 2.98 (2.39, 3.60), respectively; these values for female donors were 3.00 (2.38, 3.62) and 2.98 (2.32, 3.58), respectively. These results indicate that there is little information in the data to estimate these parameters, so that the posterior results are determined by the informative priors.

The shape parameter ψ is estimated greater than 1 in both sexes, which means that the estimated recovery process is a concave curve. That is, the Hb recovery at the beginning is fast and becomes slower over time. This corresponds to the function exhibited in Figure 2. Finally, the Pearson correlation of initial Hb values and random intercepts in the main model is 0.67 (0.64, 0.69) and 0.61 (0.58, 0.63) for male and female donors, respectively, which shows the importance of the ICP.

To compare the prediction accuracy between different models, used the results of the training data set to predict the Hb values in the validation data set. Figure 3 shows the mean square error (MSE) values during successive donations across these donors for different models. The graph illustrates the superiority of Model 3 compared with Model 2 and Model 1.

The prior sensitivity analyses showed that the posterior results are stable with respect to reasonable choices for the non-informative priors (e.g., a truncated normal distribution with mean 0 and standard deviation of 1000 for the recovery times yielded similar results). We also checked the sensitivity of the posterior results by using a less informative prior for θ (i.e., prior standard deviation increased by a factor 4) and δ (i.e., prior standard deviation increased by a factor 10). Because there is little information in the data about δ and θ, using less informative priors for these parameters, different results would have been obtained for parameter estimates of the recovery function. This might be treated as a weakness of the analysis, but this is actually the strength of a Bayesian analysis. Namely, it is the approach that allows to include such information in an elegant manner. Detailed results of these sensitivity analyses can be found in the web supplementary material.

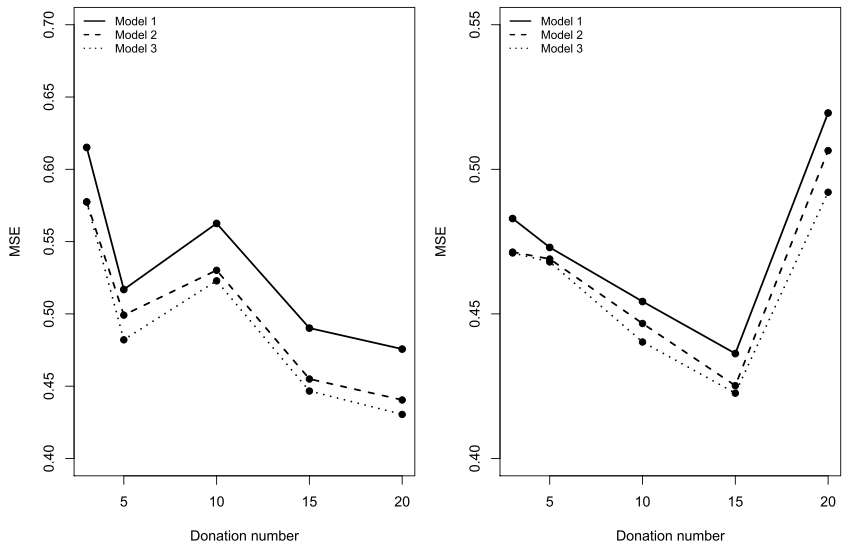

Figure 3. The mean square error (MSE) values for different models at successive donation numbers for male and female donors (male donors in the left panel, female donors in the right panel) in the validation data set.

Table VI. Results of Model 3 (and Model 3_0) for 20 artificially generated data sets of male donors.

Parameter	β_0	β_1	β_2	γ	θ	δ	ψ	RT_{g_1}	$RT_{g_{21}}$	$RT_{g_{22}}$	Class I size
True simulated value	7.87	−0.004	−0.082	0.19	0.70	2.98	2.18	100	117	419	54%
Estimated value	7.99	−0.004	−0.082	0.18	0.67	3.00	1.91	114	122	371	52%
Exceedance probability	0.65	0.51	0.47	0.38	0.25	0.52	0.38	0.46	0.50	0.32	

4.2. Simulation results

The results of Model 3 (and Model 3_0) for 20 artificially generated data sets of male donors are presented in Table VI. This table shows the true parameter values used to generate these data, the overall mean (over all 20 simulations) of the posterior medians of the parameters and the mean (over all simulations) of the corresponding exceedance probabilities. Extreme values for the exceedance probability (i.e., < 0.05 or > 0.95) indicate a significant difference. These simulation results indicate that the proposed model is able to estimate the true parameters without bias. The percentage of subjects correctly assigned to their true class was estimated to be 67% in this simulation study.

5. Predicting future Hb values

As mentioned in the introduction, the ultimate aim of this study is to improve the planning of the donors' visits to the blood bank. Predicting the Hb value after donation and forecasting the appropriate time for inviting again the prospective donor for the next donation may improve the planning of the donors' visits to the blood bank. Prediction of a future observation ($Hb_{i_{new_{t-1}}}$) is based upon the chosen model and the estimated parameters via the current data ($Data_{ref}$) together with any available observations ($Hb_{i_{new_{t-1}}}, ..., Hb_{i_{new_1}}$) for the prospective donor. The prediction consists of two stages. The first stage is to determine the class membership of the prospective donor. The class membership probability given all available information for a donor can be computed by applying Bayes' theorem:

$$P\left(g_i = k | Data_{ref}, Hb_{i_{new_{t-1}}}, ..., Hb_{i_{new_1}}\right) = \frac{P\left(Hb_{i_{new_{t-1}}}, ..., Hb_{i_{new_1}} | Data_{ref}, g_i = k\right) P(g_i = k)}{\sum_{k=1}^{2} P\left(Hb_{i_{new_{t-1}}}, ..., Hb_{i_{new_1}} | Data_{ref}, g_i = k\right) P(g_i = k)}, \quad (8)$$

where $P(g_i = k | Data_{ref}, Hb_{i_{new_{t-1}}}, ..., Hb_{i_{new_1}})$ is the marginal probability that donor i belongs to the kth class given the history of Hb values and the other covariates for that donor [33]. $P(Hb_{i_{new_{t-1}}}, ..., Hb_{i_{new_1}} | Data_{ref}, g_i = k)$ is the density of Hb values for this donor given that the donor belongs to class k. At this stage, we assign the donor to the class with the highest probability.

The second stage, which is predicting the random intercept for the donor $b_{i_{new}}$, also can be performed by applying Bayes' theorem:

$$P\left(b_{i_{new}} | Data_{ref}, Hb_{i_{new_{t-1}}}, ..., Hb_{i_{new_1}}, g_i\right) \propto P\left(Hb_{i_{new_{t-1}}}, ..., Hb_{i_{new_1}} | Data_{ref}, b_{i_{new}}, g_i\right) P\left(b_{i_{new}}\right). \quad (9)$$

Because the model is linear in the random effects, the posterior distribution for the random effects has the following closed-form expression [34]:

$$b_{i_{new}} | Data_{ref}, Hb_{i_{new_{t-1}}}, ..., Hb_{i_{new_1}}, g_i \sim N\left(\sigma_{b1}^2 Z_i^T V_i^{-1}\left(Hb_i - X_i\beta\right), \sigma_{b1}^2 Z_i^T K_i Z_i \sigma_{b1}^2\right), \quad (10)$$

where $K_i = V_i^{-1} - V_i^{-1} X_i \left(\sum_{i=1}^{N} X_i^T V_i^{-1} X_i\right)^{-1} X_i^T V_i^{-1}$, $V_i = \sigma_{b1}^2 1_{T_i} + \sigma^2 I_{T_i}$ and Z_i is a vector of 1s of length T_i. Finally, to compute the predicted Hb value, one can apply the equation for the corresponding model, with b_{i1} estimated as the mean of the posterior distribution in (10) and with the other parameters obtained from $Data_{ref}$. This is a dynamic prediction in the sense that it can be updated as soon as information from subsequent donations becomes available.

6. Conclusion

In this study, we have considered the prediction of a future Hb value for a potential blood donor given the previous observations and the estimation of the recovery time after a donation. The prediction of a donor's Hb value is complicated because of (i) heterogeneity of the initial Hb value; (ii) state dependence of a donor's Hb values; (iii) varying time intervals between donations; (iv) the temporary reduction in Hb after blood donation; and (v) the fact that the recovery process may change with the number of donations and may differ between donors. To account for these aspects of the data, we developed a mixed-effects model (Model 1), a latent class mixed-effects model (Model 2), and a latent class mixed-effects transition model (Model 3). The advantage of the mixed-effects transition model is that it simultaneously captures heterogeneity and state dependence. In all these models, the temporary reduction of Hb after donation was modeled using a flexible function. In the models with latent classes, the heterogeneity in the recovery process was controlled using latent classes and the dynamics of the recovery process using a change point model.

The latent class mixed-effects transition model was preferred over the simpler models according to the DIC and based on an evaluation of model fit. This finding shows that it is important to account for both unobserved individual heterogeneity and state dependence among the Hb values for an individual. The flexible function (2) enables us to estimate the recovery time, which is the time needed for Hb to return to its pre-donation value. In the model, the estimated pre-donation Hb value depends on the Hb value at the previous donation via the transition effect. If the value of the transition effect γ is small, the recovery time can be interpreted as approximately the time needed to return to the original Hb value, before the person started donating.

The model results show that the estimated recovery time is considerably longer than the mandatory interval between donations (i.e., 56 days). Also, our findings point to a concave Hb recovery process. That is, the recovery process is fastest at the beginning and becomes slower over time. The estimated recovery time should be seen as the ultimate recovery time, i.e., the time by which a donor's Hb value has fully recovered. Because of the concave shape of the recovery process, most of the recovery occurs before half of the recovery time has passed, which partially explains the long estimated recovery times in our data set. Furthermore, it should be noted that a recovery time that is longer than the average interval between donations is in line with the observed data, as there is a decline in the Hb trajectories with the number of donations.

Another interesting finding is that there is heterogeneity between donors in the recovery time, i.e., 54.3% and 51.3% of male and female donors have a constant recovery time during successive donations. The remaining donors have a longer recovery time and their recovery time increases after a number of donations. This increase in recovery time might be attributed to a reduction of the iron reserves in these donors. In a previous study, a faster Hb recovery was observed in donors with high pre-donation iron reserves [22], so these results require further investigation.

Our models also showed that male donors on average have a shorter recovery process than female donors. This finding is consistent with previous studies [18, 21]. The effects of age were estimated negative for male donors and positive for female donors, which is again consistent with previous studies [12, 15] and can be explained by the effects of menopause: women stop losing iron after menstruation [15]. The results also showed that the Hb value is lower on average in warm seasons than in cold seasons; see also [15, 17] for other evidence of this finding.

The model comparison (using DIC) hints that the latent class mixed-effects model with random intercepts is not sufficient to capture the entire within-subject correlation structure. An alternative approach would be to use a more complicated random-effects model, e.g., using a random slope of the time since the first visit or the number of donations. Although adding additional random effects to the model may improve the fit, the resulting parameters may be hard to interpret [10]. In our study, assuming that the time since the first donation or the number of donations affects the Hb values would imply that a part of the reduction in Hb value is permanent. This assumption would not be realistic from a clinical perspective; therefore, we did not include such random effects in the model. Also, adding random or fixed effects of time since the first donation or the number of donations to the model would have affected the estimated recovery time. Namely, the recovery time could no longer be interpreted as the time needed for Hb to return to its original value. Finally, yet another alternative model could be a random effects model using an autoregressive structure in the residuals instead of in the mean structure, which is not affected by previous covariates.

The transition model is not very often used in medical applications. One of the reasons is that the associated covariance matrix is more restrictive than for the mixed-effects model [10]. Furthermore, also transition models with random effects are not really popular in the medical area. The fact that the ICP

condition must be dealt with implies that standard estimation techniques cannot be applied. To handle the ICP in the latent class mixed-effect transition model, we used here a reduced-form equation for the initial period similar to the dynamic equation but excluding the lag effect from the model. We let the model take into account the correction between the unobserved individual effects, b_i, and the initial state Hb_{i1}.

Although we designed our model to be very flexible, it is only one out of many possible models. For instance, our model could be improved by incorporating more covariates that can affect Hb value such as physical activity [35], race [36], nutrition [37], BMI [38], and smoking status [39]. However, due to a lack of information, we could not incorporate them into our model. Class membership could be modeled to depend on some time-independent covariates such as genetic information of donors. This is the aim for a subsequent paper, once this information is available.

In conclusion, we developed a statistical modeling approach that allows to classify donors in two subgroups based on their Hb recovery time. This is of high practical importance because identification of the class for a donor could improve the planning of donors' visits to the blood banks and help to tailor donation intervals and prevent iron deficiency and donor deferrals.

Furthermore, our results support a donation interval longer than 56 days for both sexes, which has also been recommended in previous literature [11, 22, 40, 41]. The US Food and Drug Administration (FDA) is currently considering revising this interval to better protect donors [40]. For the candidates belonging to the group with non-constant recovery time, we suggest appropriate interventions (e.g., postponing the next invitation or using an iron-rich diet or taking over-the-counter vitamin supplements that contain iron or specific iron supplements) to prevent rejection at the next visit.

Appendix

JAGS syntax for running the Model 3 and Model 3_0 in JAGS.

```
model {
for(j in 1:n) {
       for(i in offset[j]:(offset[j+1]-1)){
              Hb[i]~dnorm(mu[i],tau) ### model for subsequent observations
              mu[i]<-b[j,1]+beta[1]+beta[2]*Age[j]+beta[3]*Season[i]+phi*Hbp[i]-
              theta*pow((max(RT[g[j],Time[i]]-(TSPD[i]-delta),0)/RT[g[j],Time[i]]),psi)
}
       g[j]~dcat(dsi[j,]) #Latent class indicator
       dsi[j, 1:2]~ddirch(alpha1[])
       Hb0[j]~dnorm(mu0[j], tau0) ### model for initial observations
       mu0[j]<-beta0[1]+b[j,2]+beta0[2]*Age[j]+beta0[3]*Season0[j]
}

for(i in 1:n){
       b[i,1:2]~dmnorm(mub,Omega[,])
}

for(i in 1:TT){
       RT[1,i]<-RT11
       RT[2,i]<-(1-step(i-kappa))*RT21+step(i-kappa)*RT22
}
### Priors
kappa ~ dcat(alpha2[])## Time change point in group II
for (k in 1:3) {
       beta[k]~dnorm(0,1.E-3)
       beta0[k]~dnorm(0,1.E-3)
}
RT11~dunif(0,1.E+4) ## Recovery time in group I
RT21~dunif(0,1.E+4) ## Recovery time before changing point in Group II
RT22~dunif(0,1.E+4) ## Recovery time after changing point in Group II
Omega[1:2,1:2]~dwish(R,3)
Sigma[1:2,1:2]<-inverse(Omega[,])
tau~dgamma(0.01,0.01)
tau0~dgamma(1.E+6, 1.E+3)
phi~dunif(0,1)
theta~dnorm(0.68,493) ## For male and theta~dnorm(0.96,703) for female
psi<-exp(lgpsi)
lgpsi~dnorm(0,10)
delta~dnorm(3,10)
}
```

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