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RESEARCH PAPER

Dopamine D2 Receptor Occupancy as a Predictor of Catalepsy in Rats: A Pharmacokinetic-Pharmacodynamic Modeling Approach

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ABSTRACT

Objectives Dopamine D_2 receptor occupancy (D_2RO) is the major determinant of efficacy and safety in schizophrenia drug therapy. Excessive D_2RO (>80%) is known to cause catalepsy (CAT) in rats and extrapyramidal side effects (EPS) in human. The objective of this study was to use pharmacokinetic and pharmacodynamic modeling tools to relate CAT with D_2RO in rats and to compare that with the relationship between D_2RO and EPS in humans.

Methods Severity of CAT was assessed in rats at hourly intervals over a period of 8 h after antipsychotic drug treatment. An indirect response model with and without Markov elements was used to explain the relationship of D_2RO and CAT.

Results Both models explained the CAT data well for olanzapine, paliperidone and risperidone. However, only the model with the Markov elements predicted the CAT severity well for clozapine and haloperidol. The relationship between CAT scores in rat and EPS scores in humans was implemented in a quantitative manner. Risk of EPS not exceeding 10% over placebo correlates with less than 86% D_2RO and less than 30% probability of CAT events in rats.

Conclusion A quantitative relationship between rat CAT and human EPS was elucidated and may be used in drug discovery to predict the risk of EPS in humans from D_2RO and CAT scores measured in rats.

KEY WORDS catalepsy · dopamine D_2 receptor antagonist · EPS · Markov model · schizophrenia

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INTRODUCTION

In schizophrenia drug therapy and research, dopamine D_2 receptor occupancy (D_2RO) is used as a biomarker for both efficacy and incidence of side effects (1). Several studies suggest that blockade of 65 to 80% of D_2 receptors is the basis for the antipsychotic efficacy of both the conventional neuroleptics and the novel antipsychotics (2–4). D_2RO higher than 80% increases the risk of adverse effects such as extrapyramidal symptoms (EPS) (5). In preclinical research, catalepsy (CAT) is used as a rodent model for evaluating EPS liability (6). CAT is a condition characterized by wax-like muscular rigidity, in which an abnormal body posture is maintained over an extended period of time. This effect is generally considered to be an animal model for the antipsychotic-induced EPS in human. In general, the procedure to assess CAT measures the time that an animal maintains an unusual position. It has been suggested that mechanisms involved in the mediation of CAT in rats and EPS in humans might indeed be similar (7). However, a translation of dose-response relationships for CAT and EPS thus far has not been established.

Recently, pharmacokinetic and pharmacodynamic (PKPD) models are increasingly being used to characterize and predict the time course of pharmacodynamic responses for both preclinical and clinical scenarios. These models use the principles of capacity limitation and turnover processes to describe the time course of pharmacological effects in a mechanistic manner (8). A key feature of these mechanistic models is their ability to differentiate between the system- and drug-specific parameters, what has proven to be useful in the prediction and extrapolation of treatment effects (9–11). These PKPD models were applied in the present study, in order to elucidate the D_2RO-CAT relationship in rats and to predict any side effects in humans. Modeling is not only useful to summarize and characterize data, but also for predicting answers to questions without performing new experiments (12). This "applied" modeling concept can be used to simulate and extrapolate treatment effects to different scenarios. This characteristic of PKPD modeling tools may be used to predict the relationship between D_2RO and side effects during early drug discovery phases.

Hence, the objective of this modeling study was to characterize a relationship between D_2RO and CAT in rats using PKPD modeling tools. We related the different scales of side effects in rats and human, in a quantitative manner. Furthermore, we aimed at a model structure that can predict CAT severity for other antipsychotics. These tools and approaches might be useful to predict side effects in human and thereby accelerate the drug development process.

METHODS

Data

This work was performed within the framework of the Dutch Top Institute Pharma project: Mechanism-based PKPD modeling (http://www.tipharma.com). This mechanism-based PKPD modeling platform involves leading pharmaceutical companies worldwide, and academic institutes from The Netherlands. The pharmaceutical companies who are the members of this mechanism-based PKPD modeling platform, namely, Janssen Research and Development–Belgium, Merck Sharp & Dohme—The Netherlands and Pfizer Worldwide Research and Development–USA, provided data on CAT scores after treatment with clozapine (CLZ), haloperidol (HAL), olanzapine (OLZ), paliperidone (PAL) and risperidone (RIS).

Catalepsy Experiments

CAT studies were performed using Wiga SPF rats at different dose levels for each drug (modified after Janssen *et al.*, 1965) (13). CAT was assessed in rats at hourly intervals over a period of 8 h after the administration of test compound or vehicle. All the test compounds were administrated subcutaneously. A control group (vehicle treatment) was included in all these experiments and the historical data from vehicle treatment was available and used for the following categorization. Each rat was scored based on the severity of the CAT as pronounced (score = 3), moderate (score = 2), slight (score = 1), and absent (score = 0). Evaluations of CAT were based on the sum of the scores from two independent observers, resulting in a score that ranged from 0 to 6. However, the criteria for drug-induced CAT were determined as absent for the scores $\leq$2, as mild for the scores between 3 and 5 (occurrence 0.1% of control animals) and severe for scores of 6 (not observed in controls). During this model development, CAT scores of absent, mild and severe were coded as 0, 1 and 2, respectively. Data obtained from different pharmaceutical companies were pooled, after confirming that the experimental procedure was similar between the different sources of the data. Details of these animal studies are presented in Table I.

Dopamine D_2 Receptor Occupancy Simulations

The animal study protocol for CAT experiments was aimed to measure the time course of CAT severity. Hence, it was not possible to measure D_2RO in the same animals and plasma drug concentrations were not available for these rats.

Previously, we developed hybrid physiology-based pharmacokinetic and pharmacodynamic (PBPKPD) models to describe the relationship between drug exposure in plasma and brain with D_2RO in rats for OLZ, PAL and RIS. We applied modeling approaches to simulate D_2RO for each dose level using previously developed PKPD models (14,15). These models were evaluated and showed good predictability of D_2RO at these dose levels. These models have been described elsewhere (14,15). Briefly, classical pharmacokinetic models based on a population approach were used to describe the plasma concentration time profiles. Distribution of drug to the brain was described based on passive and if applicable, active drug transport mechanisms. Subsequent binding to D_2 receptors in striatum was characterized using association and dissociation constants of these drugs to D_2 receptors. We used these previously developed PBPKPD models to simulate D_2RO for OLZ, PAL and RIS and used the simulated D_2RO to develop a model to explain the time course of CAT severity in rats. Since the individual PK data was not available, we used population values for the simulation.

In addition, we also had CAT severity information for CLZ and HAL in rats. However, there was not enough information available on plasma and brain drug concentrations and D_2RO to develop a PBPKPD model. We therefore simulated the time course of D_2RO based on a published empirical PKPD models (14–17). We refer to the original publications for more details on PKPD modeling (14–17). For CLZ, the pharmacokinetic parameters were obtained from literature

Table I Details of Animal Studies Used in this Analysis

Name of the antipsychotics	Dose as mg/kg (number of animals/dose group)	Number of rats used	Number of observations
Clozapine	0.63 (5), 1.25 (10), 2.5 (10), 5 (10), 10 (10), 20 (10), 40 (10), 80 (10), 160 (5), 320(5)	85	680
Haloperidol	0.16 (10), 0.31 (10), 0.63 (10), 1.25 (10), 2.5 (10), 5 (10), 10 (10)	70	560
Olanzapine	0.16 (8), 0.31 (6), 0.63 (10), 1.25 (15), 2.5 (10), 5 (15), 10 (10), 20 (15), 40 (10)	99	792
Paliperidone	0.08 (5), 0.16 (5), 0.31 (5), 0.63 (5), 1.25 (5), 2.50 (5), 5 (5), 10 (5), 20 (3), 40(3)	46	368
Risperidone	0.04 (5), 0.08 (5), 0.16 (10), 0.31 (10), 0.63 (10), 1.25 (10), 2.5 (10), 5 (10), 10 (10)	80	640
Control	This control group consists of all the above experiments and the historical data from vehicle treatment	445	3560

reference (15). For HAL, the pharmacokinetic parameters were obtained from a PK model developed by us. We used the simulated D_2RO of CLZ and HAL to predict the CAT scores using the PKPD model that was developed using the CAT scores from OLZ, PAL and RIS. These predicted CAT scores were compared with observations in order to judge the predictive performance of our D_2RO-CAT model.

Pharmacodynamic Model

Modeling the ordered categorical nature of CAT scores consisted of an indirect response (IDR) model combined with a logistic regression model to describe the relationship between D_2RO and the CAT scores. Further, we explored the requirement for a Markov element to properly account for the correlation between consecutive CAT scores.

Indirect Response Model

During exploratory data analysis, a time delay between D_2RO and severity of CAT scores (Fig. 1) was evident. Model building was therefore initiated by accounting for this time delay using biophase, transduction and IDR models. It was found that an IDR model gave better results than other models on the basis of objective function and model fit.

The IDR (18) model as implemented in this analysis, utilizes k_{in} as zero-order rate constant for the production of response and k_{out} as first-order rate constant for the loss of response.

$$d(R)/dt = K_{in} * \left(Q/\left(Q_{50} + Q\right)\right) - k_{out} * R$$

Q is the transformed form of D_2RO which was derived as RO / (100-RO), where RO refers to the D_2 receptor occupancy. This transformation allows Q to have a value from 0 to infinity for D_2RO from 0 to 100%. A similar transformation was applied to RO_{50}, as Q_{50} = (RO_{50}/ (100-RO_{50})). This model assumed that there was no CAT severity in the absence of D_2RO. The response variable, R, is an observed response which is a function of D_2RO. RO_{50} is the receptor occupancy at which the production of CAT response is 50% of the maximal K_{in}. RO_{50} was estimated by fitting this model to the D_2RO and CAT score data.

Severity of CAT is an ordered categorical variable that can take a value of 0 (absence of catalepsy), 1 (a mild catalepsy) or 2 (severe catalepsy). Hence, the probability of each severity was modeled with a logistic regression model (19,20). This model is intended to describe the relationship between D_2RO and severity of CAT in rats. Simulated D_2RO was used to sequentially fit this model to CAT score data. The mixed effects logistic regression model was implemented as explained in the following equation:

$$logit\left[P\left(CAT_{ij} \geq m | CAT_i, \eta_i\right)\right] = \sum_{k=1}^{m} \beta_k + R + \eta_i$$

where CAT_{ij} denotes the CAT severity score for the i^{th} individual at time t_j; $logit[P(CAT_{ij} \geq m | CAT_i, \eta_i)]$ denotes the logit function of the cumulative probability that the CAT severity score is $\geq m$ (m=1 or 2) for rat i at time t_j ; β_k specifies the baseline set of logit probabilities of the various degrees of CAT severity. For example, β_1 specifies the baseline set of probabilities for the CAT score to be ≥1. R is an observed response which is a function of D_2RO; and η_i is a random individual effect determining the individual sensitivity. In our study it was assumed to be 0, since due to the lack of individual plasma PK and D_2RO, it's value was expected to be inflated if estimated.

Indirect Response-Markov Model

The CAT severity scores were observed every hour during the animal experiments. Therefore, there may be a correlation between neighboring observations within a rat. This model estimates the cumulative probabilities of having a certain CAT score given the previous observation:

$$logit\left[P\left(CAT_{ij} \geq m \middle| CAT_i, CAT_{ij-1} = h, \eta_i\right)\right] = \sum_{k=1}^{m} \beta_{kh} + R + \eta_i$$

where CAT_{ij} denotes the CAT severity score for the i^{th} individual at t_j, and CAT_{ij-1} is the CAT severity for that rat at t_{j-1} (the previous CAT score), the β_{kh} specifies the baseline set of

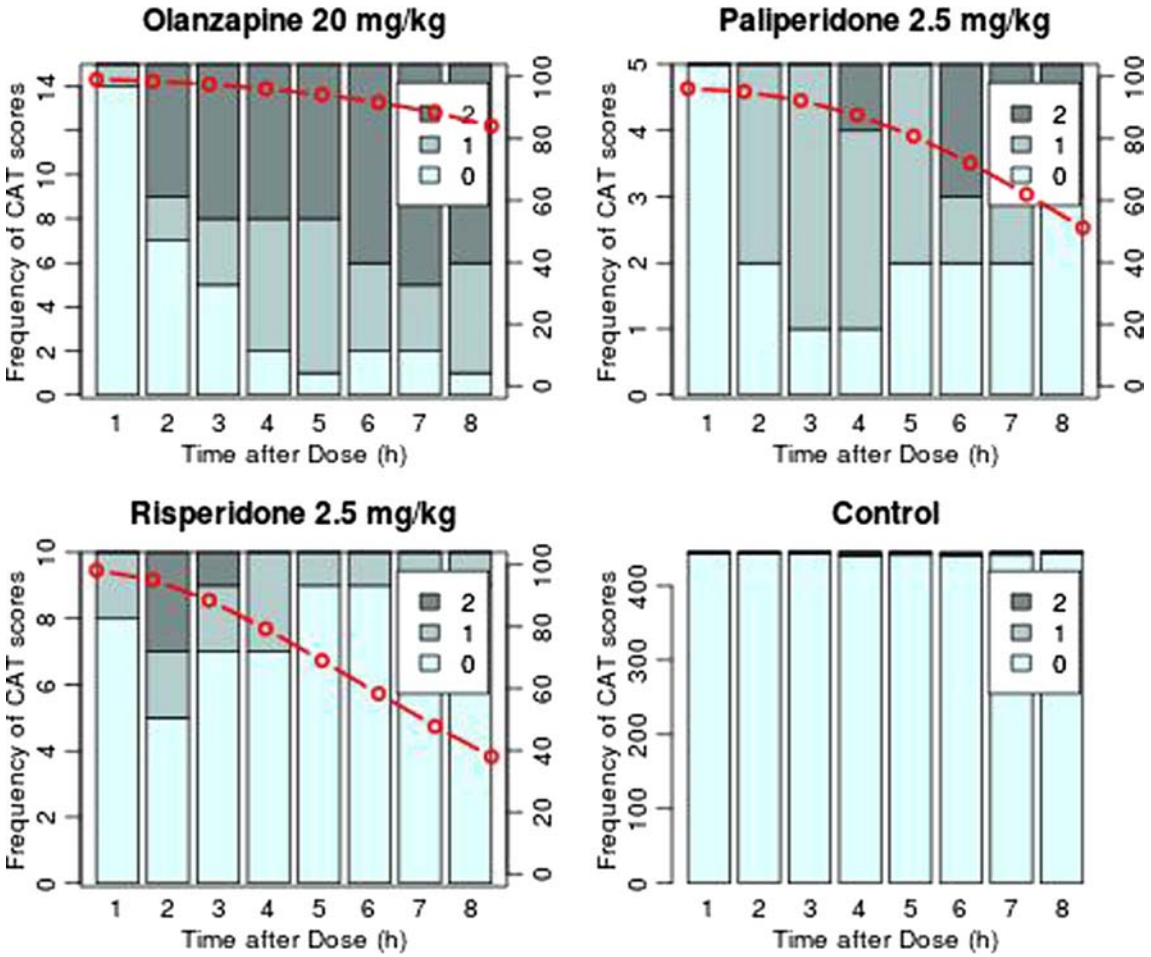

Fig. 1 Summary of catalepsy—time profiles illustrating the time delay between dopamine D_2 receptor occupancy (D_2RO) and its effect on catalepsy (CAT) scores for all drugs. The *bar chart* represents the time course of CAT scores (0, 1 and 2) and the *red line* indicates the D_2RO for the respective antipsychotics and control. Left-y axis depicts the frequency of CAT scores and right-y axis represents the percentage D_2RO.

logit probabilities of the various degrees of CAT severity, given the previous state of CAT (h). For example, β_{10} specifies the baseline set of probabilities for the CAT score to be ≥1, given the previous observation is 0. R is the outcome of the IDR model, which describes the effect of D_2RO on CAT response; and η_i is a random individual effect determining the individual sensitivity assumed in our model to be 0.

Model Building

Model fitting was performed using a population analysis approach as implemented in NONMEM (version VII level 2.0) (21). Diagnostic graphics, post processing of NONMEM output and data simulations were performed using R (version 2.10) (22).

During the model building, the goodness-of fit of different models to the data was evaluated using the change in objective function relative to the change in the number of parameters, assuming a chi-square distribution.

Model Evaluation

Standard visual predictive check (VPC) was performed to check the adequacy of the models. If the model provides an adequate description of the data, then the simulated data should mimic the important features of the observed data. To evaluate the integrity of the model, non-parametric 95% confidence intervals (CIs) were calculated by bootstrap methods. VPC and bootstrap were done as implemented in PsN (version 3.2.4) (23).

The performance of the IDR and IDR-Markov model for explaining the different transitions was evaluated by a predictive check based on the simulations obtained from these models. Transition refers to the change of CAT severity from one state to another state. One hundred datasets with identical design to the original dataset were used to simulate the distribution of the number of transitions using the parameters obtained from both the IDR and the IDR-Markov models and compared with the original number of transitions.

Table II Parameter Estimates for IDR Model

Parameters	Description	Population mean (SE)	95% CI (Lower … Upper)
β_1	Baseline probability (logit) for CAT ≥ 1	−4.87 (0.26)	−5.74 … −4.66
β_2	Baseline probability (logit) for CAT ≥ 2	−1.69 (0.12)	−1.98 … −1.52
K_{in} (h^{-1})	zero-order rate constant for the production of response	4.38 (0.24)	4.22 … 5.20
K_{out} (h^{-1})	first-order rate constant for the loss of response	0.248 (0.024)	0.206 … 0.297
RO_{50} (%)	Receptor occupancy at which the production of CAT response is 50% of the maximal production K_{in}	95.0 (0.9)	94.0 … 96.9

SE standard error as obtained from the COVARIANCE option of NONMEM, *CI* confidence interval estimated using likelihood profiling

Extrapolation to Other Drugs

The performance of the IDR and IDR-Markov models were evaluated by predicting CAT severity scores for two other antipsychotics, CLZ and HAL. This prediction was done as described below. Initially, D_2RO for CLZ and HAL was predicted using an empirical PKPD model (16,17). The IDR and IDR-Markov model parameters were used in this simulation study to predict CAT severity over time for CLZ and HAL. One hundred datasets with identical design to the original design were simulated using both models and the simulations were graphically compared with the observations.

EPS and Catalepsy

The secondary objective of this study was to relate the rat CAT scores with the human EPS scores at steady-state conditions. To simulate the CAT scores at steady-state conditions, the IDR-Markov model estimated parameters were used. Since the Markov model accounts for the correlation with previous observations, the probability of severity at the given time-point is conditioned on the previous observation, hence, $P_0 = P_0^{'} * (P_{00}) + P_1^{'} * (P_{10}) + P_2^{'} * (P_{20})$, where P_{00}, P_{10}, P_{20} are the probabilities of being at 0, when their previous observation was 0, 1 and 2 respectively. At steady-state conditions, $P_0^{'}$ is assumed to be equal to P_0. Similar equations can be written for P_1 and P_2. The probability of each severity was plotted against D_2RO.

Pilla Reddy *et al* (24) described the relationship between human D_2RO obtained from different antipsychotics and EPS using PKPD modeling with a Markov approach. This model was used to simulate the probability of EPS as absent, mild and moderate in steady-state conditions after 6 weeks of drug treatment. The relationship between D_2RO and probability of CAT in rats was related to the D_2RO-EPS relationship in humans using a polynomial equation.

RESULTS

Pharmacodynamic Model

Indirect response models with and without Markov elements were used to describe the drug effect on the severity of CAT in rats. The IDR with proportional odds model for ordered

Table III Parameter Estimates for IDR-Markov Model

Parameters	Description	Population mean (SE)	95% CI (Lower … Upper)
β_{10}	Baseline probability (logit) for CAT ≥ 1, when the previous CAT was 0	−5.21 (0.38)	−6.00 … −4.49
β_{20}	Baseline probability (logit) for CAT ≥ 2, when the previous CAT was 0	−1.43 (0.18)	−1.86 … −1.12
β_{11}	Baseline probability (logit) for CAT ≥ 1, when the previous CAT was 1	−0.827 (0.377)	−1.74 … −0.213
β_{21}	Baseline probability (logit) for CAT ≥ 2, when the previous CAT was 1	−3.46 (0.30)	−4.03 … −2.87
β_{12}	Baseline probability (logit) for CAT ≥ 1, when the previous CAT was 2	−0.054 (0.549)	−1.11 … 1.09
β_{22}	Baseline probability (logit) for CAT ≥ 2, when the previous CAT was 2	−2.52 (0.39)	−3.36 … −1.83
K_{in} (h^{-1})	zero-order rate constant for the production of response	6.84 (1.02)	4.94 … 8.85
K_{out} (h^{-1})	first-order rate constant for the loss of response	0.964 (0.200)	0.493 … 1.28
RO_{50} (%)	Receptor occupancy at which the production of CAT response is 50% of the maximal production K_{in}	92.3 (2.5)	89.1 … 97.4

SE standard error as obtained from the COVARIANCE option of NONMEM, *CI* confidence interval estimated using likelihood profiling

Table IV Objective Function Values and RO_{50} Estimates of IDR-Markov Model

Model Description	Objective function value	Estimated RO_{50} (%)
Common RO_{50} for OLZ, PAL and RIS	1044	95.0
Separate RO_{50} for OLZ, PAL and RIS	1030	OLZ – 96.0 PAL – 92.6 RIS – 95.2

RO_{50} is the receptor occupancy at which the production of CAT response is 50% of the maximal production K_{in}

OLZ Olanzapine, *PAL* Paliperidone, *RIS* Risperidone

categorical data provided a good fit to the olanzapine, paliperidone and risperidone data; K_{in}, k_{out}, RO_{50} and baseline probabilities were estimated with good precision (Tables II and III). We also estimated separate RO_{50} values for each drug for IDR-Markov, which turned out to be similar among OLZ, PAL and RIS. The objective function was significantly ($P<0.01$) decreased when compared to a model with common RO_{50} for all these drugs (Table IV).

Model Evaluation

For both IDR and IDR-Markov models, 95% CI obtained by bootstrapping was within acceptable limits for all the parameters, except for the baseline probability estimate β_{12} (Table III). VPC plots demonstrated that the model fits obtained by both IDR and IDR-Markov models were similar (Fig. 2). The time-dependent transition from one state of catalepsy to other state of catalepsy is depicted in Fig. 3. Both IDR and IDR-Markov models predicted the transition states adequately. However, the pattern of predicted transitions obtained using the model with Markov property is closer to observed transitions than those without Markov property (Fig. 3a and b). The Akaike information criterion (AIC) for model with Markov elements (1062) was lower than the AIC for model without Markov elements (1396).

Extrapolation to Other Drugs

For CLZ and HAL, the CAT score profiles could be predicted reasonably well, although some model misspecification is apparent, *e.g.* for category 2 of clozapine. For both drugs, the IDR-Markov model performed better than the IDR model in predicting the CAT severity score (Fig. 4).

CAT and EPS Relationship

The relationship between D_2RO and side effect scores in rat and human is depicted in Fig. 5. Excessive D_2RO (>80%) changed the probability towards mild and severe CAT in rats and EPS in humans. However, in humans the change in the severity of EPS was less pronounced in comparison to the changed severity of CAT in rats. The relationship between CAT and EPS scores is shown in Fig. 6. A polynomial function was used to empirically relate the CAT and EPS scores.

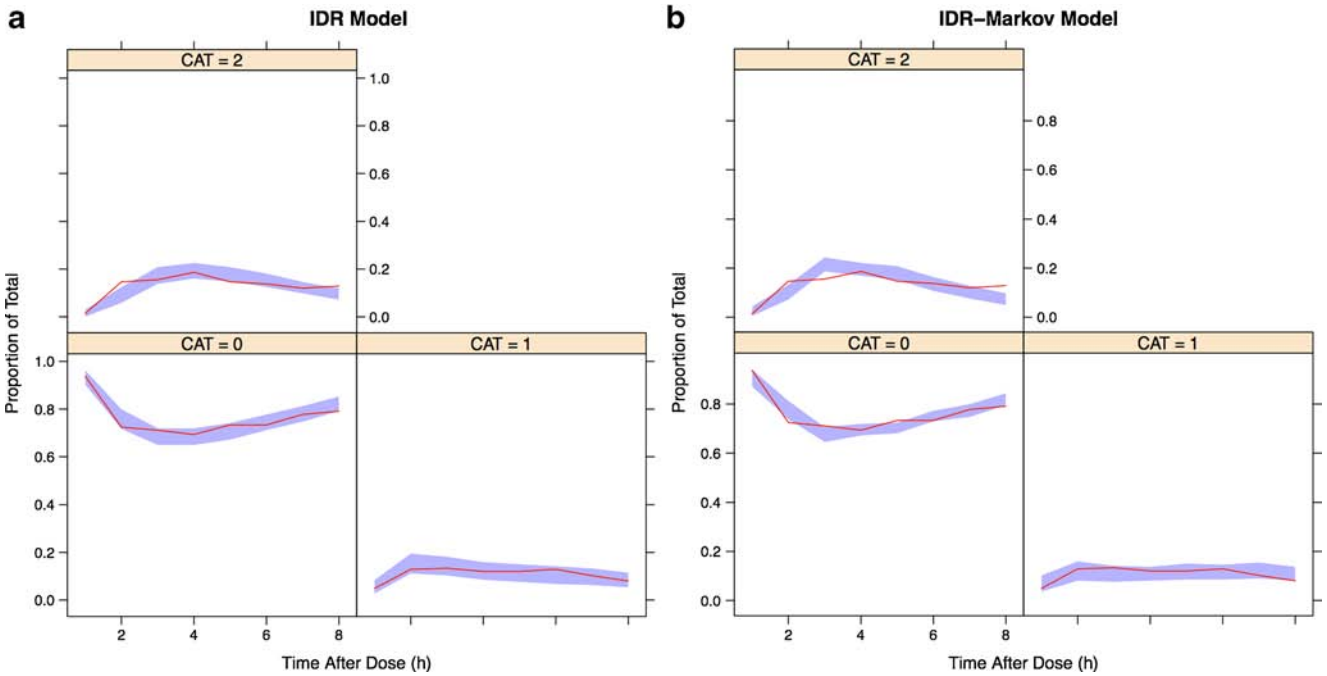

Fig. 2 Visual predictive check results for the adequacy of indirect response (IDR; (**a**)) and indirect response with Markov (IDR-Markov; (**b**)) model. *Shaded* area depicts the 90% prediction interval for the simulated probabilities and the *red solid line* represents the proportion from the original data.

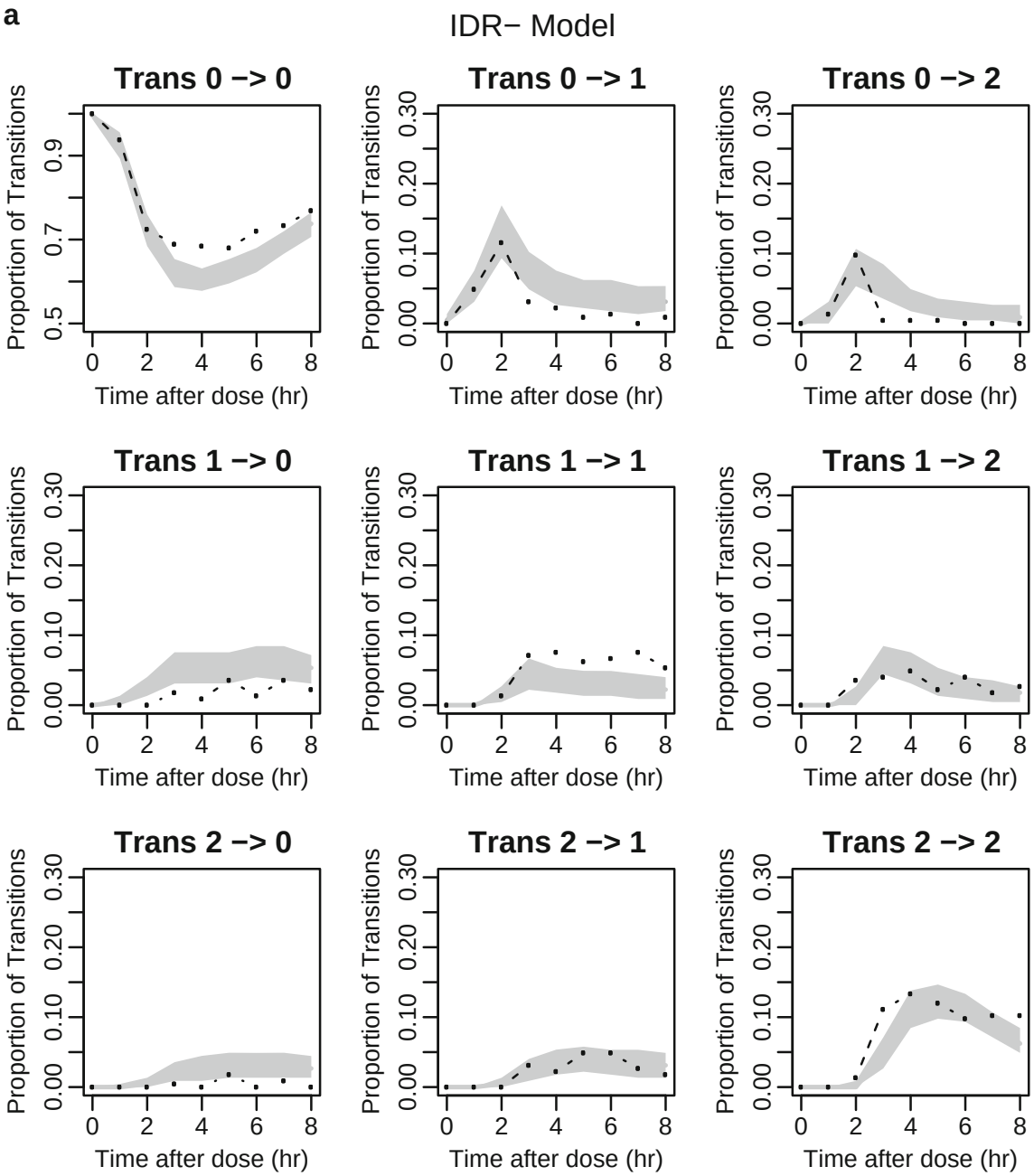

Fig. 3 Visual predictive check results for the distribution of transitions in catalepsy severity. *Black dotted lines* represent the observed proportion of transitions and *shaded areas* the 90% prediction interval from the indirect response (IDR; (**a**)) and indirect response with Markov (IDR-Markov; (**b**)) model simulations.

DISCUSSION

The objective of our PKPD analysis was to utilize the information on CAT in rats to describe the relationship between D_2RO and CAT and to extrapolate this relationship to other drugs to predict CAT scores. We also linked the relationship between D_2RO and CAT in rat to the relationship between D_2RO and EPS in human using PKPD modeling and simulation approaches. Several studies indicate that the dopaminergic pathway involved in the mediation of CAT in rats and EPS in humans could be similar (7). Moreover, it was reported that disappearance of CAT in rats is a reliable indicator of gradual dissociation of the antipsychotic agent from D_2 receptors (6). Hence, our approach to relate D_2RO and CAT severity is substantiated. Moreover, using the D_2RO-CAT relationship rather than drug exposure-CAT data provides a way to introduce a compound-independent variable, which may be used to extrapolate the D_2RO-CAT relationship to other drugs and also to relate this relationship between different species.

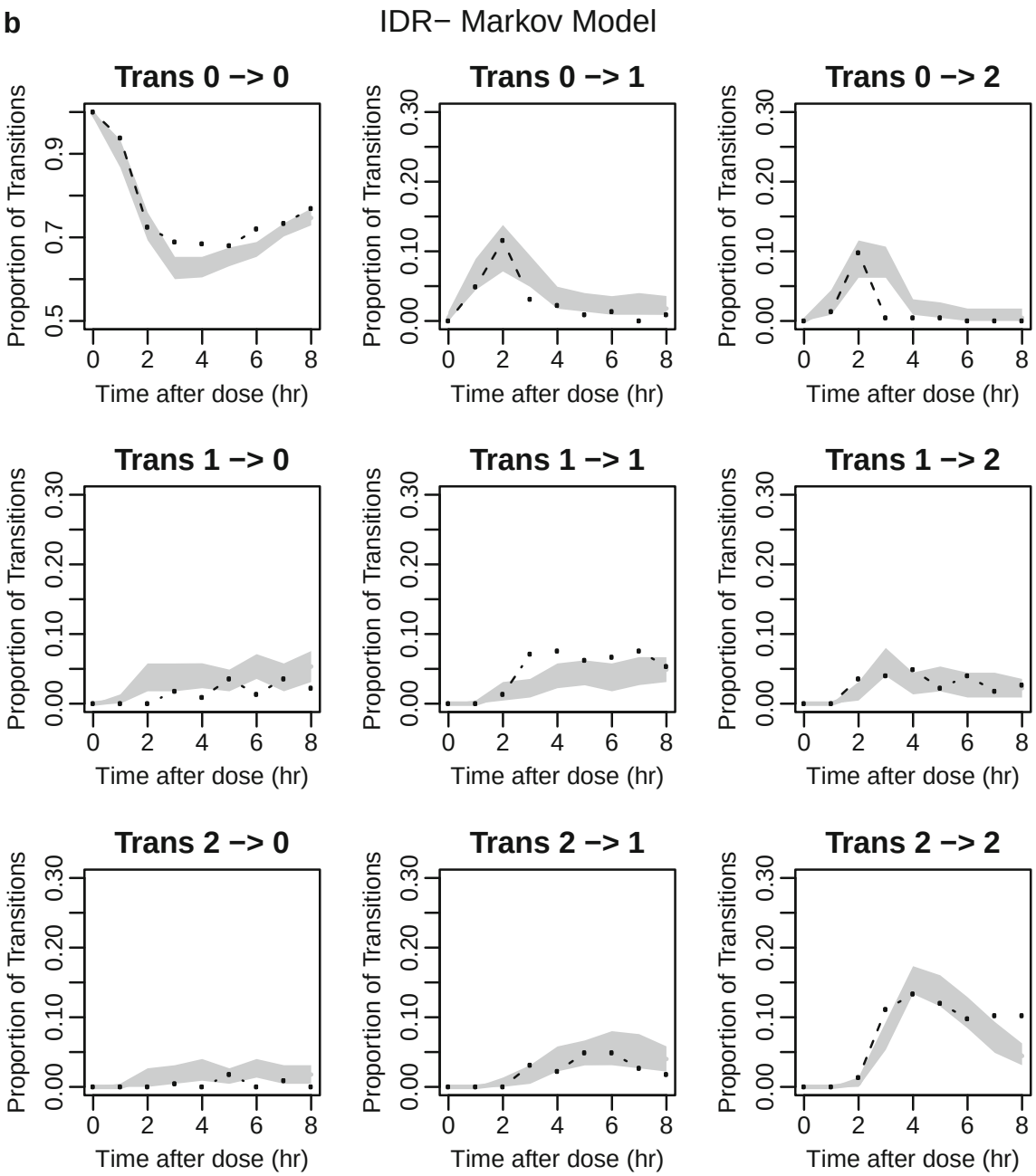

Fig. 3 (continued)

In general, proportional odds models, introduced in the field of PKPD modeling by Sheiner *et al* (20), have been proven to describe the ordered categorical data well. In our data analysis, the proportional odds model was used to explain the probability of the degree of CAT.

It is evident that D_2RO and CAT severity scores are not directly related with time (Fig. 1). So, it is appropriate to use an IDR model to explain the relationship between D_2RO and CAT scores rather than a direct response model (*e.g.*, Emax model). The semi-mechanistic nature of IDR models allows for distinguishing the drug-specific parameters and system-specific parameters (K_{in} and k_{out}). It is reasonable to assume that these system-specific parameters could be the same for different antipsychotic drugs. This characteristic of the IDR model is used as a base to extrapolate the D_2RO-CAT relationship to other drugs (discussed later). In our analysis we assumed a common RO_{50} to determine the receptor occupancy to produce a half-maximal effect. We also estimated separate RO_{50} for each drug and these separate RO50 estimates fall within the confidence interval (85.7 and 97.0) of the common RO50 estimate. This assumption (common RO_{50}) allows for considering RO_{50} as a system-specific parameter,

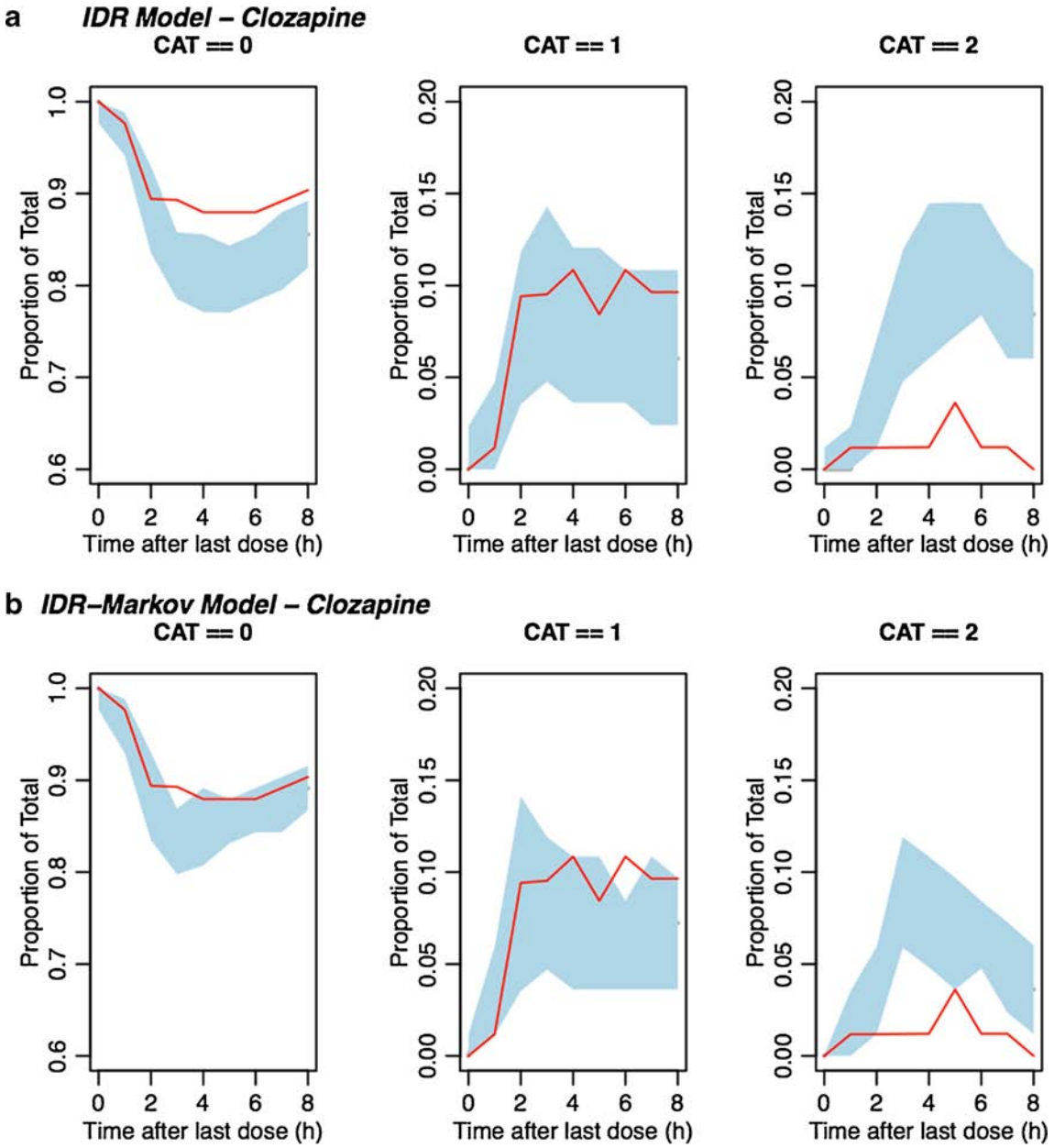

Fig. 4 Predicted and observed probabilities of catalepsy for clozapine and haloperidol. *Red lines* represent the observed proportion and shaded areas the 90% prediction interval from the indirect response (IDR) and indirect response with Markov (IDR-Markov) model simulations. (**a**) and (**b**) show the probabilities of catalepsy for clozapine using IDR and IDR-Markov models, respectively. (**c**) and (**d**) show the probabilities of catalepsy for haloperidol using IDR and IDR-Markov models, respectively.

which can be used for extrapolating the D_2RO-CAT relationship to other drugs.

It was expected that for frequently measured categorical type data, there might be a correlation between neighboring observations. The standard proportional odds model with IDR model may not be able to account for these correlations. These series of probable transitions between states can be described with Markov modeling (19,25). Zingmark *et al* (25) modeled spontaneously reported side-effects using a Markov approach which is a hybrid between the proportional odds model and the transition model. We adopted this approach in our modeling work in combination with the IDR model.

In other systems Markov models have been reported to describe frequently observed and correlated scores better than the models without Markov elements (19,25). The VPC plots representing the predicted probability at the three states of CAT were similar between these two models. It was also reported that Markov models predict the number of transitions much better than non-Markov models (19,25), as was confirmed in our analysis (Fig. 3a and b).

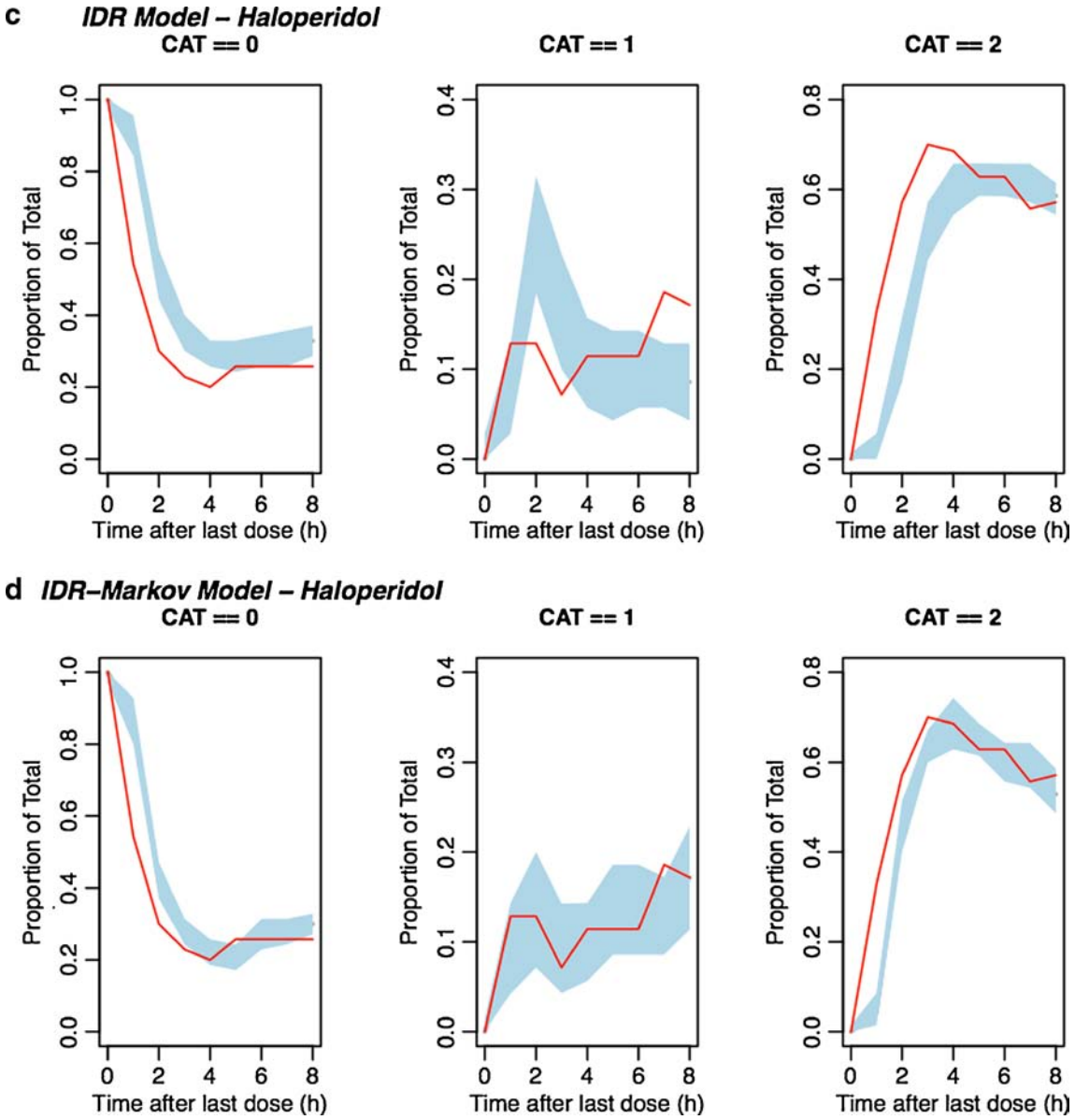

Fig. 4 (continued)

The precision of the predictions for clozapine (Fig. 4a) and haloperidol (Fig. 4b) was limited by the fact that the D_2RO was not observed for individual rats, but predicted based on previously developed models. In addition, for several practical reasons it is not possible to measure D_2RO and CAT severity score in the same animal at different time points. As an illustration, animals need to be scarified to excise the brain to measure D_2RO in brain tissue. Hence, obtaining time course of CAT severity score and D_2RO information in the same animal is experimentally not possible. This lack of information explains some of the inadequacy in simulating the time course of transitions on CAT score. For this same reason, inter-rat variability was assumed to be zero in these models. Besides, the objective of this modeling work was to relate D_2RO to CAT in preclinical levels and using this relationship to predict the probability of EPS in human for a typical D_2RO. Hence, including variability in individual D_2RO level was not needed. One of the baseline probabilities (β_{12}) was estimated with lower precision and wide confidence intervals were seen in the bootstrap analysis. It is common that Markov models are over-parameterized to handle the correlation between adjacent observations (25).

Dopamine D_2 receptor occupancy was simulated using previously developed PBPKPD models. These models are available only for OLZ, PAL and RIS. So, we used the CAT scores from OLZ, PAL and RIS to develop the IDR and IDR-Markov models. However, we intended to extend the D_2RO-CAT relationship to CLZ and HAL. Since we do not have PBPKPD models for CLZ and HAL, we utilized empirical PKPD models to simulate D_2RO for CLZ and

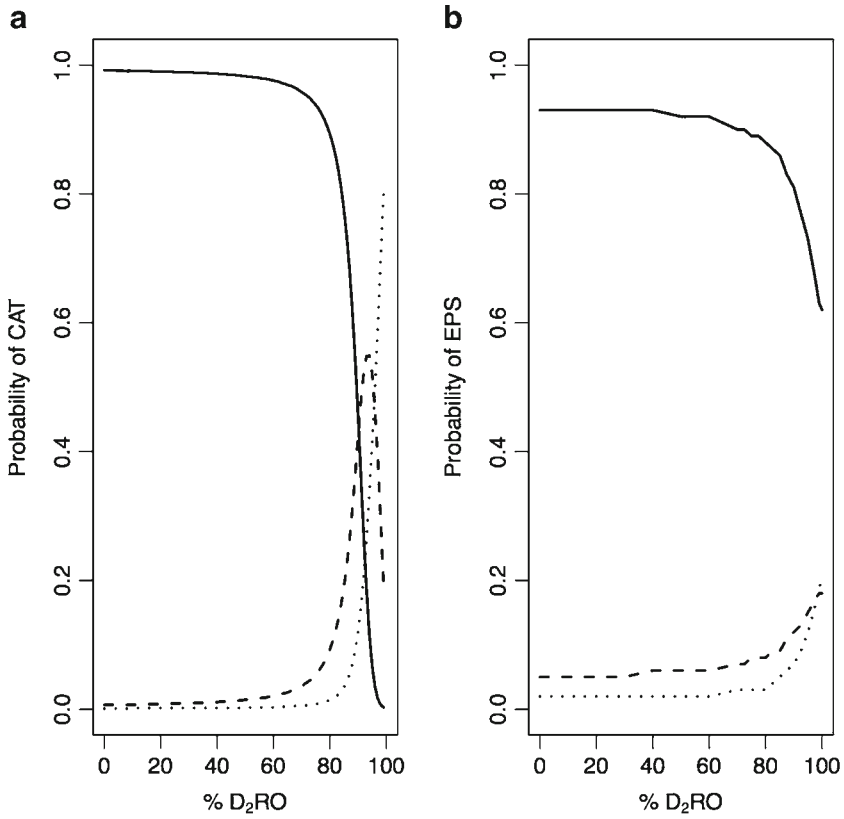

Fig. 5 Relationship between D_2 receptor occupancy and side effects. Panel (**a**) represents the probability of catalepsy (CAT) scores in rats at steady-state conditions. *Solid, dashed and dotted lines* depict the probabilities of CAT severity as absent, mild and severe, respectively. Panel (**b**) represents the probability of extrapyramidal symptom (EPS) scores in humans at steady-state conditions. *Solid, dashed and dotted lines* depict the probabilities of CAT severity as absent, mild and moderate to severe, respectively.

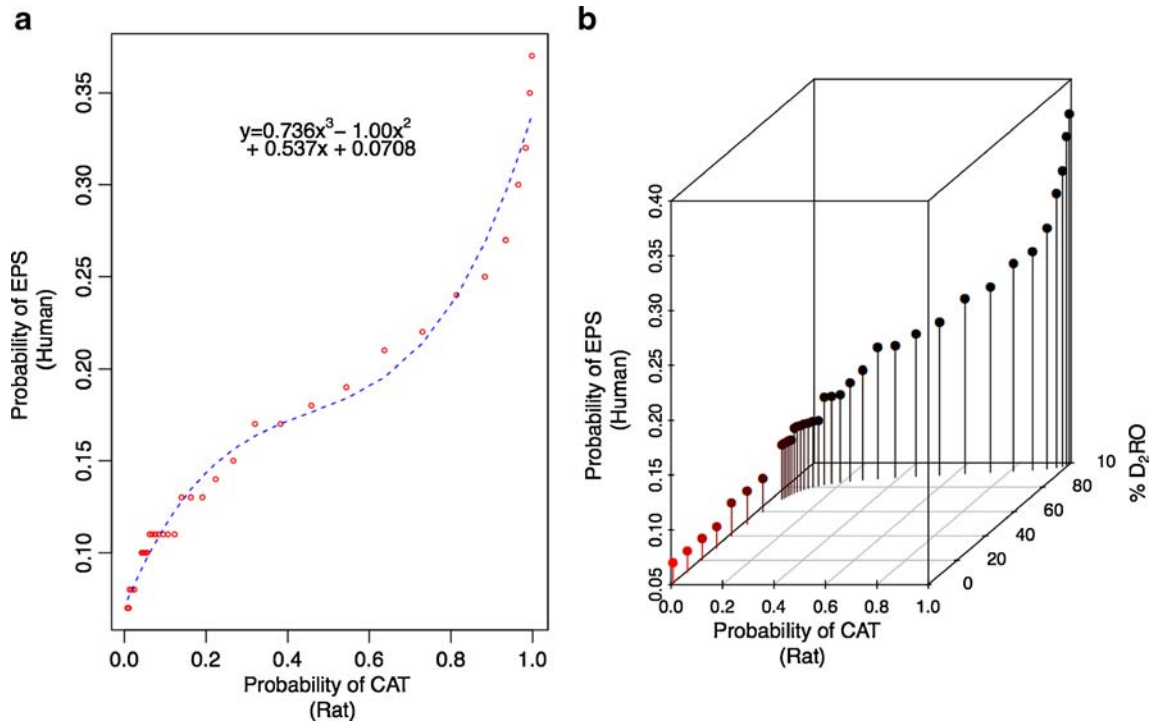

Fig. 6 Panel (**a**) represents the relationship between the probability of side effects in rats and humans. Probabilities of side effects were calculated based on D_2RO in rats and humans. The *red open circles* represent the calculated probability of side effects in rats and humans. The *blue dotted line* is obtained by fitting a polynomial model to the probability of side effects in rats (CAT) and humans (EPS). In panel (**b**), the *dots* with *dropped lines* depict the relationship between catalepsy in rats and the probability of extrapyramidal symptoms in humans for respective D_2 receptor occupancies.

HAL and to predict CAT severity in rats. Using both IDR and IDR-Markov model structure and parameter estimates, CAT scores were predicted for CLZ and HAL. IDR-Markov models performed slightly better than the IDR models in predicting CAT scores for CLZ and HAL. For CLZ and HAL, we needed to rely on literature information for receptor binding information (for example, EC_{50}) and pharmacokinetic information (for CLZ). We have not accounted for study specific covariates (strain of rats, route of administration of compounds) related change in pharmacokinetics and pharmacodynamics (*e.g.* typical and atypical antipsychotics, slow and fast dissociation from dopamine D_2 receptor) for CLZ and HAL. This may explain the inadequateness of the simulations to predict CAT score more precisely. Extending this external validation with other atypical antipsychotics is needed to substantiate the applicability of this model describing the relationship between D_2RO and catalepsy in rats. Nevertheless, it is clear from the simulations that Markov model is able to capture the trend and intensity of the CAT severity better than the model without Markov element. The unique feature of the Markov model to account for the correlation between adjacent observations may be responsible for its better predictive properties. Despite this moderate predictive performance of the IDR-Markov model, it should be noted that this approach has not been tested on antipsychotics, which have a different mechanisms of action than D_2 antagonism (*e.g.* aripiprazole).

In drug discovery phase, catalepsy model in rats are often used as a predictor for EPS in humans (7). However, there scores are not closely related in explaining the severity of side effects and this leaves out less scope for direct translation of CAT scores in rats to EPS scores in human. Hence, the relationship between CAT scores and EPS scores needs to be understood in a quantitative manner. Understanding the relationship of these scores to a biomarker (*i.e.* D_2RO) which is quantitated in both rats and human would provide room for translation of D_2RO and side effect relationship between species. To this end, our modeling work was not intended to predict EPS scores based on CAT scores in rats, but to understand the relationship between these scores and D_2RO in both species and descriptively derived the D_2RO-EPS relationship in human. Results of our modeling depict that severity and occurrence of side effects and their relation with D_2RO are not same between these species. The probability for having any EPS event is less than 40% in clinical studies, even for very high D_2RO, whereas the probability of any CAT event is close to 100% for very high D_2RO in rats. Interestingly in humans, the probability of having EPS for 0% D_2RO is approximately 5%, which shows the effect of placebo on EPS.

Utility of this model is to predict the certain probability of EPS in humans based on CAT scores and D_2RO in rats. Using this empirical modeling, a risk of EPS not exceeding 10% over placebo may be predicted for D_2RO up to 86% with less than 30% probability of CAT events in rats (Fig. 6). However, this relationship may be applied with caution considering several assumptions and limited number of antipsychotic compounds involved in the model development. Hence, these elucidations may be used in drug discovery to carefully consider the compound selection. A more mechanistic approach would be useful for a better understanding and better predictive power of the model. However, the observed relationships of rat CAT and human EPS with RO were quite different, as shown in Fig. 5. Therefore it is not surprising that we did not succeed in finding a mechanistic relationship.

Brain mechanisms involved in catalepsy in rats and EPS in humans may be expected to be similar and involve certain threshold of D_2 receptor occupancy (7). With this understanding, it may be speculated that these differences between species may be due to dissimilarities in susceptibility and tolerability of these effects between species. With this modeling effort, we achieved an empirical way to account for these differences by relating the chance of side-effects given a D_2RO level between species. Though the utility of this model is limited to only the investigated system, the concepts behind this modeling effort could be applied to other disease areas as well.

CONCLUSION

The relationship between D_2RO and CAT scores was elucidated for three drug (OLZ, RIS and PAL) using PKPD modeling tools. The IDR-Markov model predicted the severity of CAT for CLZ and HAL better than the IDR model. The outcome of our simulations directed towards a link between CAT as observed in rats and EPS as observed in humans in a quantitative manner.

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