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ORIGINAL ARTICLE

Dissociated Agonist of Glucocorticoid Receptor or Prednisone for Active Rheumatoid Arthritis: Effects on P1NP and Osteocalcin Pharmacodynamics

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Fosdagrocorat (PF-04171327), a dissociated agonist of the glucocorticoid receptor, has potent anti-inflammatory activity in patients with rheumatoid arthritis with reduced adverse effects on bone health. To identify fosdagrocorat doses with bone formation marker changes similar to prednisone 5 mg, we characterized treatment-related changes in amino-terminal propeptide of type I collagen (P1NP) and osteocalcin (OC) with fosdagrocorat (1, 5, 10, or 15 mg) and prednisone (5 or 10 mg) in a phase II randomized trial ($N = 323$). The time course of markers utilized a mixed-effects longitudinal kinetic-pharmacodynamic model. Median predicted changes from baseline at week 8 with fosdagrocorat 5, 10, and 15 mg were −18, −22, and −22% (P1NP), and −7, −13, and −17% (OC), respectively. Changes with prednisone 5 and 10 mg were −15% and −18% (P1NP) and −10% and −17% (OC). The probability of fosdagrocorat doses up to 15 mg being noninferior to prednisone 5 mg for P1NP and OC changes was >90%.

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Study Highlights

WHAT IS THE CURRENT KNOWLEDGE ON THE TOPIC?

☑ P1NP and OC are well-established bone formation markers and are good indicators of bone health. The time course of these bone markers may provide valuable input into predicting clinical response.

WHAT QUESTION DID THIS STUDY ADDRESS?

☑ As part of model-based drug development for fosdagrocorat, longitudinal P1NP and OC data were modeled to provide information about optimal doses of fosdagrocorat with reduced effects on bone formation.

WHAT THIS STUDY ADDS TO OUR KNOWLEDGE

☑ This study characterizes the time course of bone markers with a K-PD model in patients with RA receiving fosdagrocorat, prednisone, or placebo, on a background of methotrexate, and provides a case example of how a modeling and simulation approach can be applied to quantitative decision making.

HOW MIGHT THIS CHANGE DRUG DISCOVERY, DEVELOPMENT, AND/OR THERAPEUTICS?

☑ These findings might facilitate the application of K-PD modeling to longitudinal dose-response data. Use of probability of success estimates may help decision making.

Glucocorticoids (GCs) are well-established drugs used to treat a variety of inflammatory and autoimmune diseases. Side effects of GCs include bone loss/osteoporosis/osteopenia, hyperglycemia/diabetes/insulin resistance, adrenal suppression, weight gain, hypertension, cataracts, mood, and sleep disturbances.[1,2] GCs decrease bone formation by inhibiting osteoblast differentiation, reducing bone matrix synthesis in osteoblasts, and increasing bone resorption by activating osteoclasts.[3] The expression of osteocalcin (OC), an important component of bone matrix protein synthesized mainly in osteoblasts, is repressed by GCs in animals and human patients.[4,5] Most of the metabolic side effects considered mediated by the GC receptor (GR) are via transactivation, whereas most of the desired anti-inflammatory effects are via transrepression. Therefore, a significant focus has been on the separation of these two modes of GR activity.[6]

Fosdagrocorat (PF-04171327), a phosphate ester prodrug form of PF-00251802, is a dissociated agonist of the GR (DAGR),[7] and is being developed to retain the anti-inflammatory efficacy of GCs while reducing unwanted effects. A dissociated agonist is a selective GR agonist that has a strong transrepressive action, but reduced transactivation, and therefore produces a "dissociated" action by retaining the potent anti-inflammatory action, but reduces the damaging effects to bone, skin, and muscle.[8] It is investigated as a treatment for reducing signs and symptoms of rheumatoid arthritis (RA), inhibiting the progression of joint destruction, and improving physical function in patients with active RA. In preclinical *in vitro* and *in vivo* assays using prednisolone as a positive control, PF-00251802 showed robust anti-inflammatory activity and reduced effects on markers of bone and glucose metabolism at exposures associated with anti-inflammatory effects. The presence of such dissociation in the clinical setting was expected to represent a therapeutic improvement relative to the standard GC therapy.

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A phase II trial, assessing the safety and efficacy of fosdagrocorat in patients with RA, was conducted with various doses of fosdagrocorat and prednisone.[9] Results demonstrated that both fosdagrocorat 10 and 15 mg q.d. produced efficacy superior to placebo and comparable to prednisone 10 mg q.d.[9] As part of the model-based drug development for fosdagrocorat, modeling and simulation with the collected bone effect and efficacy data were conducted to quantitate the relationships and inform dose selection for subsequent confirmatory clinical trials.[7,10] Optimal fosdagrocorat doses were investigated, with efficacy comparable to, or greater than, prednisone, while providing acceptable changes in bone formation markers.

For efficacy, a longitudinal dose-response analysis on the Disease Activity Score showed that fosdagrocorat doses of $\geq$9 mg q.d. had an effect comparable to, or greater than, prednisone 10 mg q.d.[7] From a safety perspective, to determine the upper end of the optimal dose, changes in the following two bone formation markers were examined: (1) serum amino-terminal propeptide of type I collagen (P1NP), a specific biomarker for bone collagen synthesis; and (2) OC, a noncollagenous matrix protein in bone produced by osteoblasts.[11]

The two objectives of the present analysis were to: (1) characterize the concentration-time course of serum P1NP and OC after administration of fosdagrocorat, prednisone, or placebo in patients with RA on background methotrexate using a kinetic-pharmacodynamic (K-PD) model; and (2) conduct stochastic simulation to evaluate the probability of fosdagrocorat being noninferior to prednisone with regard to changes in bone formation biomarkers. Application of the K-PD approach in clinical trials has been reported to sufficiently describe the time course of PD data in the absence of drug concentration measurements.[12–15] A bone biomarker analysis in urine (bone resorption marker urine C-terminal telopeptide in osteoporosis treatment with bisphosphonate) used a similar approach to our biomarker analysis, with a K-PD model.[16] They evaluated drug regimens in ongoing clinical development with both K-PD and pharmacokinetic (PK)-PD models and concluded that the K-PD model adequately described the biomarker response. In our investigation, while PK data of fosdagrocorat were available, the model was under development. Additionally, PK data of prednisone were not available. As a comparison of the effects of fosdagrocorat and prednisone doses on the bone biomarker changes was required, and in order to provide timely dose information for ongoing clinical development, we investigated the longitudinal dose-response relationships using the K-PD model instead of PK-PD models.

MATERIALS AND METHODS

Clinical trial and assay methods

This analysis used data from a phase II global, randomized, double-blind, parallel-group trial in 323 patients with active RA on a background of methotrexate (clinicaltrials.gov NCT01393639).[17] The trial was conducted in accordance with Good Clinical Practice principles and the Helsinki Declaration, and informed consent was obtained from all patients in compliance with Good Clinical Practice and related requirements. Prospective approval of the trial protocol was obtained from the Institutional Review Board/ Independent Ethics Committee.

Each patient received either fosdagrocorat 1, 5, 10, or 15 mg, prednisone 5 or 10 mg, or placebo q.d. for 8 weeks followed by a 4-week taper (to evaluate the hypothalamic–pituitary–adrenal axis recovery) during which each dose was reduced to fosdagrocorat 1 mg, prednisone 5 mg, or placebo every other day (weeks 9 and 10) and every 3 days (weeks 11 and 12), in a blinded fashion. During the trial period, blood samples were taken from each subject at weeks 0, 2, 4, 6, 8, 10, and 12 prior to dosing and week 13 (**Supplementary Figure S1**).

Serum samples were analyzed for P1NP and OC concentrations using electro-chemiluminescent immunoassay methods at Synarc Laboratory, Newark, CA. The assay information is summarized in **Supplementary Table S1**. P1NP is regarded as the most critical biomarker for bone formation[11] and was the principal focus in the current study.

Kinetic-pharmacodynamic model

To describe the time course of the biomarkers, a mixed-effects longitudinal K-PD model was applied to the final analysis data. In the structural model (**Supplementary Figure S2)**, it is assumed that the drug (fosdagrocorat or prednisone) is administered to a virtual effect compartment and that the biomarker (P1NP or OC) is synthesized in, and eliminated from, another virtual response compartment. Changes over time (t) for the drug amount (A) in the effect compartment, and the biomarker response (R) in the response compartment are expressed as follows:

$$\frac{dA}{dt} = Input - KDE \cdot A$$

$$\frac{dR}{dt} = Ks \cdot u(t) - Kd \cdot R$$

$$R(t=0) = BL = Ks/Kd$$

It is assumed that multiple administration of the drug q.d. is input to the effect compartment with a zero-order rate *Input* (mg/week). For example, *Input* for fosdagrocorat 1 mg q.d. is assumed as 7 mg/week. A is eliminated with a first-order rate constant (*KDE*). The response (R) is assumed to be produced with a zero-order synthesis rate (*Ks*) and eliminated with a first-order degradation rate (*Kd*) and u(t) is defined as follows:

$$u(t) = \left(1 - \frac{Imax \cdot IR^{\gamma}}{EDK50^{\gamma} + IR^{\gamma}}\right)$$

The *BL* represents the biomarker concentration at time zero. The rate of the drug infusion to the response compartment ($IR = KDE \cdot A$) is assumed to indirectly inhibit synthesis of the biomarker in the compartment and is described by a sigmoidal maximum effect (E_{max}) model.[18]

Imax indicates the maximum fractional inhibition of the synthesis rate and *EDK50* represents *IR* that produces 50% of *Imax*. The γ indicates the Hill coefficient. A linear increase over time for P1NP or OC was incorporated to describe the underlying change in the biomarker response

as observed in the placebo group. Random-effects parameters for interindividual and intra-individual variability were incorporated into the model. Exploratory graphical analysis suggested a linear increase of P1NP and OC over time in placebo treatment (methotrexate only). The response was assumed to be independent of the response driven by GCs and DAGR and was incorporated as an additive linear effect outside the differential equations.

Accordingly, an observed serum P1NP or OC concentration for the *i*th individual at the *j*th time point, $Y_{(ij)}$, was modeled as follows:

$$\log Y_{(ij)} = \log F_{(ij)} + \varepsilon_{(ij)}$$
$$F_{(ij)} = R_{(ij)} + SLP_{(i)} \cdot t_{(ij)}$$

$F_{(ij)}$ represents a model-predicted serum P1NP or OC concentration. The $R_{(ij)}$ is a predicted response described earlier. A slope parameter, $SLP_{(i)}$, represents a coefficient for a linear change over time $t_{(ij)}$ to describe the natural history of P1NP or OC change. The $\varepsilon_{(ij)}$ indicates an independent random-effect parameter to explain the intra-individual variability, which is assumed to distribute normally with a mean zero and a variance σ^2. To incorporate the interindividual variability into the model, parameters for the *i*th individual are divided into fixed-effect and random-effect parameters. For example, *EDK50* for the *i*th individual, $EDK50_{(i)}$, is modeled as follows:

$$EDK50_{(i)} = \theta_{EDK50} \cdot \exp(\eta_{EDK50(i)})$$

where θ_{EDK50} indicates the fixed-effect parameter and $\eta_{EDK50(i)}$ the random-effect parameter (assumed to distribute normally with a mean zero and a variance ω^2_{EDK50}). For *SLP* only, the parameter for the *i*th individual is described with an additive error model as follows:

$$SLP_{(i)} = \theta_{SLP} + \eta_{SLP(i)}$$

where θ_{SLP} and $\eta_{SLP(i)}$ (a normal distribution with mean zero and variance ω^2_{SLP}) represent the fixed-effect and random-effect parameters.

One of our main objectives was to conduct stochastic simulations for decision making rather than to explore potential covariates. However, a limited number of covariates were available for investigation, and, therefore, a limited covariate investigation was performed. Covariates investigated were based on several basic demographic variables of interest, which may possibly influence future study design. Covariate effects were screened by visually investigating relationships between individual empirical Bayesian estimates (EBEs) for each model parameter and the following baseline covariates: treatment, sex, race, age, body weight, and body mass index.

Model evaluation

The model was evaluated by diagnostic plots,[19] nonparametric bootstrapping with 1,000 datasets, and visual predictive checks (VPCs). Details of these methods are included in the **Supplementary Information** (Model evaluation method). η-shrinkage and ε-shrinkage were also calculated.[20]

Simulations

To compare the biomarker response between fosdagrocorat and prednisone, 1,000 trials identical to the original phase II trial were simulated using the developed model with random effects and considering the parameter uncertainty (stochastic simulations).

In each trial, P1NP or OC median percent change from baseline (%CFB) at week 8 (following administration of fosdagrocorat or prednisone) and the median %CFB for each fosdagrocorat dose difference from that for prednisone 5 or 10 mg were calculated. Then, probability that the difference was not less than −20% was calculated. The noninferiority criteria of −20% was based on input from key opinion leaders and is the lower limit of equivalence criteria typically used in bioequivalence studies. Details of the simulation method are included in the **Supplementary Information** (Simulation method).

Software

Time-course data of the P1NP and OC concentrations were analyzed using the nonlinear mixed-effects modeling methodology, as implemented by NONMEM version 7.2/7.3 (ICON Development Solutions, Ellicott City, MA).[21] Analyses with the final model were conducted by NONMEM version 7.3. A statistical package R version 3.0.2/3.1.2 was used for performing the exploratory graphical analysis, summarizing the analysis, and assisting the model development and simulations.[22] Perl-speaks-NONMEM version 3.5.4/4.2.0 and Xpose version 4.4.1/4.5.3 were used for model evaluations.[19,23–25]

RESULTS

Patient characteristics and observed data

In the clinical trial, 323 patients were randomized in a blinded fashion to one of the fosdagrocorat, prednisone, or placebo treatment groups. Because baseline values for P1NP and OC concentrations were unavailable in 2 patients, 321 patients were included in this analysis. The majority of the patients were female (80%); patient characteristics were overall similar across the treatment groups (**Table 1**).

In total, 4,837 serum samples were available for the population K-PD modeling. Following administration of fosdagrocorat and prednisone, observed P1NP and OC concentrations decreased over time until approximately week 8 (**Figure 1**). Beyond week 8 (in the taper period), concentrations generally returned to the baseline levels. Following administration of placebo (methotrexate only), observed serum P1NP and OC concentrations underwent a small linear increase; estimates (95% confidence interval (CI)) for the linear background P1NP and OC increase were 0.330 (−0.199, 0.779) ng/mL per week and 0.134 (−0.0855, 0.354) ng/mL per week, respectively. Although the 95% CIs estimated using the placebo-only data were relatively wide, it might be considered due to the limited number of subjects. Therefore, it was considered meaningful to keep the placebo response with the interindividual variability in the next modeling steps. The linear

Table 1 Patient characteristics at baseline

	Fosdagrocorat				Prednisone			
Treatment	**1 mg**	**5 mg**	**10 mg**	**15 mg**	**5 mg**	**10 mg**	**Placebo**	**All**
No. of patients (male/female)	45 (12/33)	47 (9/38)	45 (11/34)	47 (11/36)	44 (6/38)	46 (5/41)	47 (10/37)	321 (64/257)
Race (white/black/Asian/other)	(38/2/5/0)	(41/1/3/2)	(43/1/1/0)	(41/2/3/1)	(39/0/3/2)	(38/1/5/2)	(41/0/4/2)	(281/7/24/9)
Median age, years [min, max]	54 [18, 78]	56 [29, 77]	56 [25, 80]	56 [27, 72]	52 [20, 78]	59 [34, 75]	56 [23, 79]	56 [18, 80]
Median height, cm [min, max]	163 [137, 190]	162 [137, 175]	163 [136, 185]	164 [139, 185]	163 [145, 185]	162 [135, 185]	162 [148, 181]	163 [135, 190]
Median body weight, kg [min, max]	66.4 [36.6, 144]	71.0 [40.0, 115]	70.0 [38.2, 128]	75.0 [46.8, 133]	73.2 [45.8, 103]	72.2 [40.5, 107]	70.0 [45.0, 110]	71.0 [36.6, 144]
Median body mass index, kg/m^2 [min, max]	25.9 [17.5, 51.0]	29.0 [18.3, 48.5]	26.9 [15.9, 40.0]	28.2 [17.5, 47.8]	27.1 [19.3, 37.7]	27.5 [20.2, 44.0]	27.0 [17.6, 42.0]	27.1 [15.9, 51.0]
Median P1NP, ng/mL [min, max]	57.2 [14.2, 163]	46.2 [14.4, 146]	44.2 [14.0, 153]	46.2 [10.9, 134]	47.8 [19.8, 184]	49.2 [18.4, 90.0]	52.2 [15.2, 145]	48.5 [10.9, 184]
Median OC, ng/mL [min, max]	26.6 [6.05, 85.9]	22.4 [5.86, 55.7]	21.1 [4.80, 66.5]	20.1 [5.70, 47.0]	23.3 [10.8, 71.7]	21.2 [9.39, 46.4]	25.3 [4.85, 81.1]	22.4 [4.80, 85.9]

OC, osteocalcin; P1NP, amino-terminal propeptide of type I collagen.

increase also appeared in the active treatment groups in the taper period (**Figure 1**).

Observed dose-response plots at week 8 suggested dose-response relationships for both P1NP and OC (**Figure 1**). The fosdagrocorat dose-response profile for P1NP appeared to reach a plateau at around 10–15 mg (fosdagrocorat), whereas the corresponding dose-response profile for OC appeared not to reach a plateau with fosdagrocorat 15 mg.

Pharmacodynamic analyses

To fit the P1NP and OC concentration-time courses without the drug PK data, several longitudinal dose-response models were tested. Empirical models, such as E_{max}, with exponential time-course functions were tested, but had difficulty in describing slight changes with large interindividual variability. In contrast, the K-PD model, which describes longitudinal dose-response profiles using virtual spaces for drug and response in the absence of PK data,[18] successfully described the P1NP and OC time course with precise parameter estimates. Therefore, the developed model was considered adequate to describe the biomarker time course semimechanistically, assuming that fosdagrocorat and prednisone indirectly inhibit synthesis of the biomarkers. The assumption was supported by a report showing that prednisone decreased P1NP and OC in a dose-dependent and time-dependent manner.[26]

Because serum P1NP and OC concentrations were correlated ($\rho = 0.73$) and the time-course profiles were similar, modeling was initially performed for P1NP and then the same model was used to fit OC observations. Following exploratory analyses, it was considered that the mixed-effects K-PD model was the most appropriate to describe the P1NP data.

For *SLP*, $\eta_{\mathrm{SLP}(i)}$ was highly and positively correlated with fosdagrocorat and prednisone doses. Observed time course for P1NP and OC also seemed to increase when the dose increased. This dose-dependent and time-dependent increase was assumed to be a rebound effect of the drug on positive bone-homeostatic feedback. To incorporate the increase, a time-dependent and dose-dependent function for the rebound effect was incorporated into the synthesis rate of the biomarker as follows:

$$u(t) = \left(1 + \frac{Dose \cdot RBmax \cdot t}{T50 + t}\right) \cdot \left(1 - \frac{Imax \cdot IR^{\gamma}}{EDK50^{\gamma} + IR^{\gamma}}\right)$$

where *Dose* and *t* represent fosdagrocorat or prednisone dose and time after the dose. *RBmax* and *T50* indicate the maximum rebound effect and time to achieve 50% of *RBmax*, respectively. The time-dependent and dose-dependent effect was assumed common for the same dose of fosdagrocorat and prednisone due to difficulty in estimating the parameters separately with data from only two doses of prednisone (i.e., *Imax* and *EDK50* were estimated for fosdagrocorat and prednisone, respectively, whereas *RBmax* and *T50* were assumed to be the same for both compounds). When γ was estimated, the estimate was not significantly different from 1 and the model led to estimation instability (γ was 0.920 when the estimation step was terminated). Additionally, stochastic simulations to compare the biomarker response between

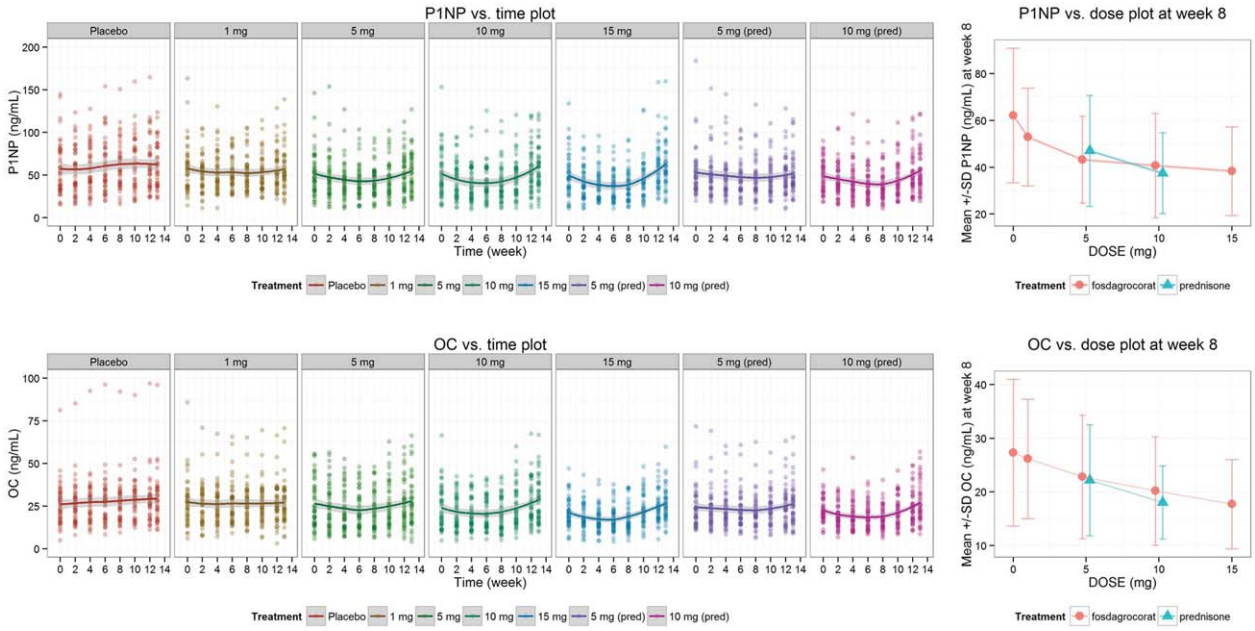

Figure 1 Left panels: observed serum amino-terminal propeptide of type I collagen (P1NP) (top) and osteocalcin (OC) (lower) concentrations vs. time plots for fosdagrocorat 1, 5, 10, and 15 mg, prednisone 5 and 10 mg, and placebo (methotrexate only). Right panels: observed dose-response plots for mean (±SD) at week 8. Each line represents a smooth local regression with the 95% confidence intervals calculated from locally weighted scatterplot smoothing (LOESS) function of R software. X mg on each label represents fosdagrocorat dose and Y mg (pred) indicates prednisone dose. In the dose-response plots in the right panels, fosdagrocorat dose at 0 mg represents placebo.

fosdagrocorat and prednisone in which γ was fixed at 1, 1.5, 2, and 3 showed that, as γ values were increased, the prediction profiles seemed lower than the observed profile. Consequently γ was fixed to 1.

Estimation of interindividual variability parameters for *KDE*, *EDK50*, *SLP*, and *BL* were supported by the data as models converged with reasonable estimates of the parameters. Because EBEs for *KDE*, *EDK50*, and *BL* showed a slight correlation, covariance structure for the parameters was incorporated (Δ objective function value $= -15.115$). There was no clear trend observed between EBEs and individual covariate values for age, body weight, body mass index, sex, treatment, and race (**Supplementary Figure S3**).

For OC, the developed model using P1NP data was used. Because estimation of the fixed-effect parameter for drug elimination (*KDE*) was unstable (high relative SE %), the value was fixed to the estimate in the P1NP analysis. Because *Imax* was estimated to be close to 1, the parameter was fixed to 1. To reduce estimation instability, individual random effect parameters for *KDE*, *EDK50*, and *BL* were assumed to be independent. Except for these changes, similar results were observed and no other modifications were made to the model. **Table 2** shows parameter estimates (% relative SE) of the developed K-PD model for P1NP and OC. In general, the parameters were precisely estimated. For P1NP, *EDK50* of fosdagrocorat (40.1 mg/week) was smaller than that of prednisone (45.9 mg/week), whereas, for OC, *EDK50* fosdagrocorat (148 mg/week) was higher than that of prednisone (122 mg/week). For P1NP (OC), *Ks* was increased by the rebound effect up to 1.05, 1.24, 1.48, and 1.72 (1.03, 1.14, 1.28, and 1.41) times the *Ks* values without the rebound effect at doses of 1, 5, 10, and 15 mg, respectively. The *T50* was estimated to be 1.13 and 2.24 weeks for P1NP and OC, respectively. As expected, relatively large interindividual variability was observed ranging from 46.6% to 95.0% (considering η_{KDE}, η_{EDK50}, and η_{BL}) for P1NP, and from 25.5% to 123% for OC. The interindividual variability for *SLP* was 0.928 and 0.338 ng/mL per week for P1NP and OC, which was also larger compared to the fixed effect parameter estimate. Meanwhile, intraindividual variability was small and estimated to be 15.2% (P1NP) and 14.1% (OC).

Model evaluations

The developed K-PD model was evaluated using diagnostic plots, nonparametric bootstrapping, and a VPC. The prediction-based plots indicated a central tendency to the identity line and no major bias (**Supplementary Figure S4**). The residual-based plots did not show any systematic trend with regard to population predicted concentrations (PRED) or time after the first dose (WEEK). The observed and predicted serum concentrations vs. time plots for each individual are provided in **Supplementary Figure S5**.

For the nonparametric bootstrapping, the success rates of the convergence for P1NP and OC were 81% and 86%, respectively. The median values for parameter estimates from the bootstrap were comparable with the final parameter estimates (**Table 2**). For the success runs, 95% CIs were estimated with the 2.5th and 97.5th percentiles. The bootstrap 95% CIs were generally similar to 95% CIs from

Table 2 Parameter estimates of the model for trough P1NP and OC concentrations

Parameter	Estimate (RSE%)		[95% CI]	Bootstrap median [95% CI]	
P1NP					
$KDE_{Fosdagrocorat}$ (/week)	0.597	(17.8)	[0.388, 0.806]	0.615	[0.288, 1.21]
$KDE_{Prednisone}$ [/week]	0.535	(28.9)	[0.232, 0.839]	0.551	[0.219, 1.36]
Kd [/week]	0.609	(17.5)	[0.400, 0.818]	0.579	[0.356, 1.47]
$Imax_{Fosdagrocorat}$	0.751	(4.85)	[0.679, 0.822]	0.766	[0.669, 0.913]
$Imax_{Prednisone}$	0.754	(8.89)	[0.622, 0.885]	0.772	[0.674, 0.943]
$EDK50_{Fosdagrocorat}$ [mg/week]	40.1	(17.8)	[26.1, 54.0]	40.2	[26.8, 59.5]
$EDK50_{Prednisone}$ [mg/week]	45.9	(30.2)	[18.7, 73.0]	46.3	[28.0, 74.9]
RBmax [/mg]	0.0479	(17.6)	[0.0313, 0.0644]	0.0494	[0.0296, 0.0858]
T50 [week]	1.13	(42.6)	[0.185, 2.07]	1.17	[0.290, 3.71]
BL [ng/mL]	47.0	(2.83)	[44.4, 49.6]	47.1	[44.6, 49.7]
SLP [ng/mL per week]	0.162	(67.3)	[−0.0516, 0.375]	0.173	[0.0162, 0.407]
IIV %CV[a] [η_{KDE}]	95.0	(25.1)	[67.7, 116]	98.3	[64.9, 159]
IIV %CV[a] [η_{EDK50}]	65.5	(32.7)	[39.3, 83.9]	63.9	[30.3, 99.6]
IIV %CV[a] [η_{BL}]	46.6	(8.51)	[42.6, 50.4]	46.6	[42.6, 50.8]
IIV SD[a] [η_{SLP}]	0.928	(11.5)	[0.817, 1.03]	0.909	[0.705, 1.09]
ρ[b] [$\eta_{KDE,}$ η_{EDK50}]	−0.312	(61.5)	[−0.687, 0.0640]	−0.253	[−0.743, 0.345]
ρ[b] [$\eta_{BL,}$ η_{KDE}]	−0.316	(36.1)	[−0.540, −0.0921]	−0.323	[−0.569, −0.0444]
ρ[b] [$\eta_{BL,}$ η_{EDK50}]	−0.410	(25.5)	[−0.614, −0.205]	−0.411	[−0.708, −0.101]
Residual variability %CV [ε]	15.2	(0.962)	[14.9, 15.5]	15.1	[14.0, 16.5]
OC					
$KDE_{Fosdagrocorat}$ [/week]	0.597	FIX		0.597	FIX
$KDE_{Prednisone}$ [/week]	0.535	FIX		0.535	FIX
Kd [/week]	0.939	(24.6)	[0.486–1.39]	0.910	[0.603–1.97]
$Imax_{Fosdagrocorat}$	1	FIX		1	FIX
$Imax_{Prednisone}$	1	FIX		1	FIX
$EDK50_{Fosdagrocorat}$ [mg/week]	148	(6.68)	[128–167]	147	[123–177]
$EDK50_{Prednisone}$ [mg/week]	122	(11.3)	[95.2–149]	122	[104–146]
RBmax [/mg]	0.0276	(21.5)	[0.0160, 0.0393]	0.0282	[0.0178, 0.0391]
T50 [week]	2.24	(54.8)	[−0.165, 4.64]	2.09	[0.783–5.73]
BL [ng/mL]	22.2	(2.58)	[21.1–23.4]	22.2	[21.1–23.4]
SLP [ng/mL per week]	0.0675	(53.6)	[−0.00342, 0.138]	0.0670	[−0.0150, 0.155]
IIV %CV[a] [η_{KDE}]	123	(22.7)	[92.1–148]	127	[81.7–185]
IIV %CV[a] [η_{EDK50}]	25.5	(56.3)	[0[c]–37.0]	24.6	[11.3–37.6]
IIV %CV[a] [η_{BL}]	43.6	(7.80)	[40.1–46.8]	43.4	[39.8–47.5]
IIV SD[a] [η_{SLP}]	0.338	(10.1)	[0.303, 0.370]	0.333	[0.253, 0.408]
Residual variability %CV [ε]	14.1	(0.887)	[13.9–14.3]	14.1	[12.9–15.2]

Model parameters γ described in the Methods section were fixed to 1.
BL, biomarker concentration at time zero (baseline); CI, confidence interval; CV, coefficient of variation; *EDK50*, drug infusion to the response compartment that leads to 50% of I_{max}; FIX, parameters were not estimated and were fixed; I_{max}, maximum inhibition of the synthesis rate; IIV, interindividual variability; *Kd*, first−order degradation rate; *KDE*, first−order elimination rate; NA, not applicable; OC, osteocalcin; P1NP, amino-terminal propeptide of type I collagen; *RBmax*, maximum rebound effect; RSE, relative SE; SLP, slope parameter; T50, time to 50% of maximum rebound effect.
[a]%CV = $\sqrt{(\omega^2)}\cdot 100$ or SD = $\sqrt{(\omega^2)}$; RSE% for ω^2. [b]$\rho[\eta_1, \eta_2]$ = covariance for η_1 and $\eta_2/(\sqrt{(\omega_1{}^2)}\cdot\sqrt{(\omega_2{}^2)})$. [c]Not calculated because the value was negative.

the variance-covariance matrix of the final parameter estimates. Overall, the model parameters derived from the bootstrapping were estimated with good precision.

A VPC for P1NP and OC was performed, and plots of observed and model-predicted concentration CFB vs. time for each treatment group were generated (**Figure 2**). Because the observed CFB at the 10th, 50th, and 90th percentile points were generally within the predicted 95% CIs, the K-PD model was considered to have adequately reproduced the actual P1NP and OC time profile.

The η-shrinkage and ε-shrinkage were calculated to evaluate model adequacy. There was a moderate degree of shrinkage in interindividual variability (η-shrinkage) for *KDE*, *EDK50*, and *SLP*: 28%, 34%, and 21% for P1NP, and 41%, 62%, and 27% for OC, respectively. The η-shrinkages for *BL* of P1NP and OC were low (3% and 2%, respectively). Degree of ε-shrinkage for the intra-individual variability was also low: 15% (P1NP) and 14% (OC).

Simulation results

Table 3 summarizes observed and simulated P1NP and OC %CFB at week 8 after administration of fosdagrocorat, prednisone, or placebo. These simulated median %CFB suggest that fosdagrocorat 5 and 10 mg (%CFB −18% and

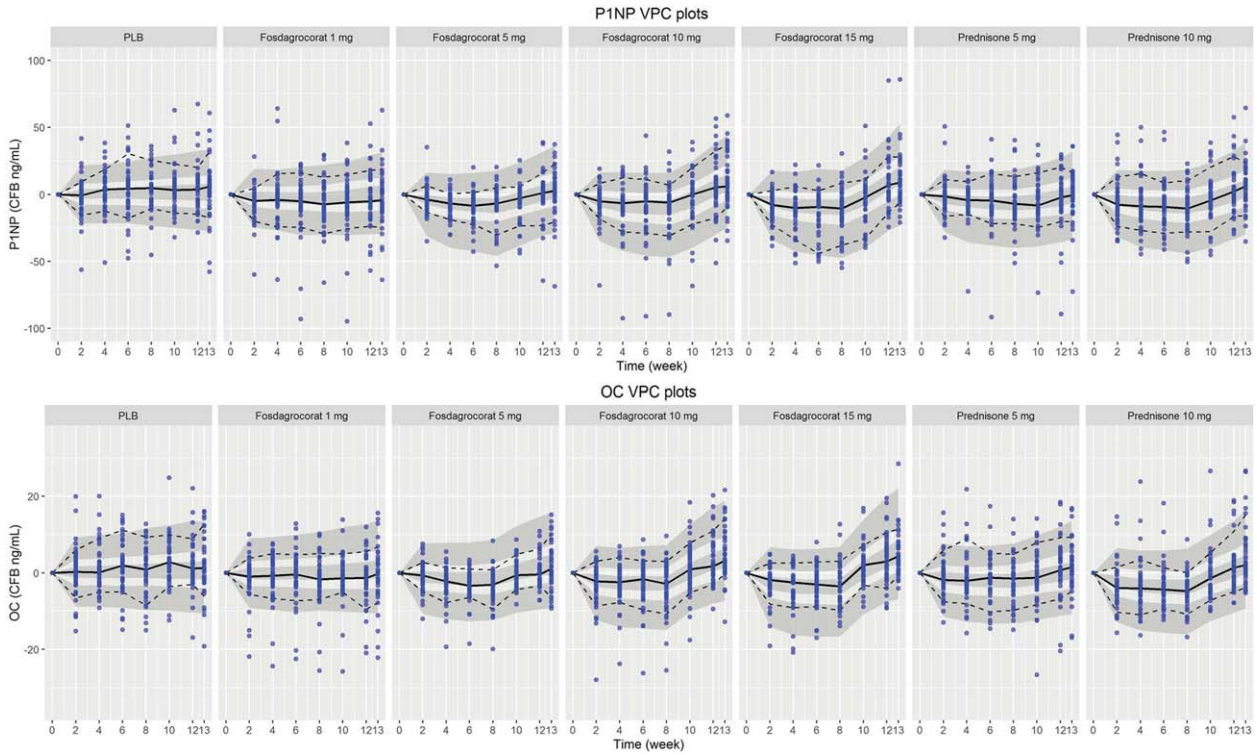

Figure 2 Visual predictive check (VPC) plots for change from baseline in concentration of amino-terminal propeptide of type I collagen (P1NP) and osteocalcin (OC), by treatment group. For each panel, the black line (black dashed lines) represents observed median (10th and 90th percentiles) P1NP and OC concentrations (change from baseline [CFB]) time course. The gray areas represent model-predicted 95% confidence intervals (CIs) of 10th, 50th, and 90th percentiles of the CFB time course. The closed circles indicate observed individual P1NP and OC CFB. PLB, placebo.

−22%) and prednisone 5 and 10 mg (%CFB −15% and −18%) would have a similar effect on P1NP change, where the difference of %CFB in fosdagrocorat from that in prednisone was <20% (18% and 19% at 5 and 10 mg, respectively). Meanwhile, fosdagrocorat doses would have less effect on OC (%CFB −7% and −13%) than the prednisone doses (%CFB −10% and −17%), where the difference was over −20% (−31% and −25% at 5 and 10 mg, respectively).

To investigate the effects of fosdagrocorat on biomarker changes relative to prednisone, the difference in median %CFB at week 8 for fosdagrocorat vs. prednisone was calculated. Upper panels in **Figure 3** show the %CFB difference from prednisone 5 mg vs. fosdagrocorat doses for P1NP and OC, respectively. Lower panels of **Figure 3** show the probability of fosdagrocorat being noninferior to prednisone 5 mg. There was a >90% probability that fosdagrocorat 10 mg and 15 mg were noninferior (prespecified noninferiority margin of 20%) to prednisone 5 mg for P1NP and OC changes. In addition, there was a >90% probability that fosdagrocorat doses were noninferior to prednisone 10 mg for both P1NP and OC changes (**Supplementary**

Table 3 Summary of observed and simulated P1NP and OC percentage change from baseline at week 8 following administration of fosdagrocorat, prednisone, and placebo, q.d.

	P1NP		OC	
	Observed mean, % [95% CI]	**Simulated median, % [95% CI]**	**Observed mean, % [95% CI]**	**Simulated median, % [95% CI]**
Fosdagrocorat 1 mg q.d.	−4.8 [−13.5, 3.8]	−5.7 [−15.8, 5.5]	−2.1 [−8.7, 4.4]	0.1 [−8.2, 9.5]
Fosdagrocorat 5 mg q.d.	−12.4 [−19.2, −5.6]	−18.2 [−28.4, −6.6]	−11.6 [−17.3, −5.9]	−6.7 [−15.3, 3.0]
Fosdagrocorat 10 mg q.d.	−16.1 [−24.3, −7.9]	−21.7 [−33.3, −9.0]	−11.2 [−18.7, −3.6]	−12.6 [−21.4, −3.0]
Fosdagrocorat 15 mg q.d.	−16.0 [−27.0, −5.0]	−21.6 [−33.6, −8.0]	−13.6 [−21.2, −6.0]	−16.8 [−26.5, −6.1]
Prednisone 5 mg q.d.	−5.2 [−16.7, 6.4]	−15.4 [−27.6, −2.7]	−5.7 [−14.6, 3.2]	−9.7 [−18.3, 0.1]
Prednisone 10 mg q.d.	−19.6 [−28.2, −10.9]	−18.3 [−30.3, −4.8]	−19.4 [−25.5, −13.4]	−16.9 [−25.8, −6.1]
Placebo q.d.	14.0 [3.8, 24.2]	2.5 [−7.7, 14.1]	7.6 [−3.4, 18.6]	1.8 [−7.5, 12.7]

For simulation, median [95% CI] indicates 50th percentile point [2.5th, 97.5th percentile points] for median percentage change from baseline of 1,000 clinical trials. Observed mean [95% CI] indicates mean ± 1.96·SE.

CI, confidence interval; OC, osteocalcin; P1NP, amino-terminal propeptide of type I collagen.

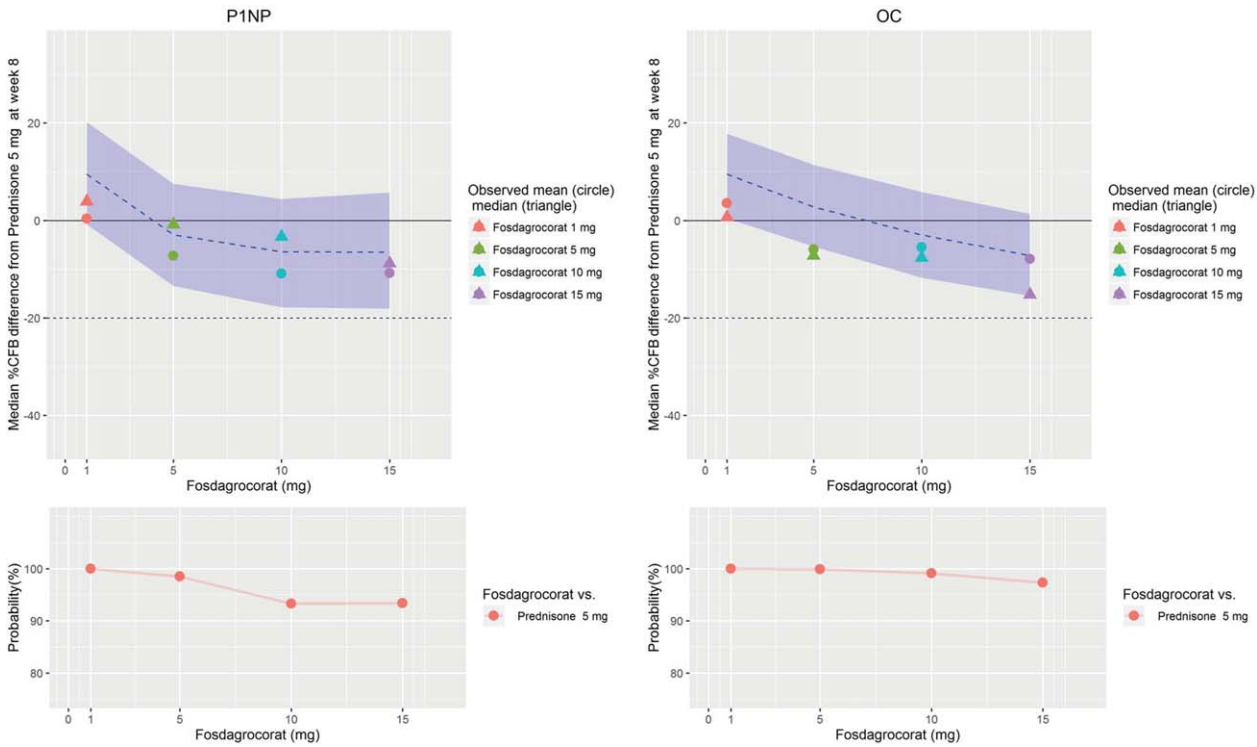

Figure 3 Simulation results for comparing percentage CFB in amino-terminal propeptide of type I collagen (P1NP) and osteocalcin (OC) with fosdagrocorat and prednisone 5 mg. Upper panels: the blue dashed line (blue shaded area) indicates model-predicted median (10th to 90th percentiles) of P1NP or OC percentage change from baseline (%CFB) for fosdagrocorat difference from the median %CFB for prednisone 5 mg at week 8. Closed circles and triangles represent observed mean and median of the difference, respectively. Lower panels: probability that fosdagrocorat is noninferior to prednisone 5 mg with a margin of 20%.

Figure S6). Although the model seemed to relatively underpredict the data for %CFB difference in OC with fosdagrocorat and prednisone 5 mg, the model predicted %CFB difference was considered to generally capture the observed %CFB difference. Simulations for P1NP or OC %CFB for fosdagrocorat difference from that for prednisone 5 or 10 mg are summarized in **Supplementary Table S2**.

DISCUSSION

Serum P1NP and OC concentration-time profiles following administration of fosdagrocorat, prednisone, or placebo, in patients with RA on a background of methotrexate, were adequately described by the mixed-effects longitudinal K-PD model with good precision. Based on the simulations, with a noninferiority criteria predefined as P1NP or OC change for fosdagrocorat no worse than that of prednisone with 20% margin, all doses of fosdagrocorat met the noninferiority criteria to prednisone 5 mg with a >90% probability for P1NP and OC. Together with efficacy results,[7,9] it is suggested that fosdagrocorat 10 and 15 mg would have efficacy comparable to, or greater than, prednisone 10 mg q.d., whereas the changes in bone formation markers of these doses of fosdagrocorat would be noninferior to prednisone 5 mg q.d., based on each bone biomarker change (probability of the noninferiority >90% for the fosdagrocorat doses).

For the developed model, moderate degrees of η-shrinkage for *KDE*, *EDK50*, and *SLP* were observed. When shrinkage is present (>20–30%), it is recommended that model diagnostics are not based on EBE but instead on simulation-based diagnostics.[20] In this analysis, the VPC showed adequacy of the model. Together with the other evaluations, such as diagnostic plots and nonparametric bootstrap results, it is unlikely that the observed moderate shrinkage resulted in biased model selection.

The observed P1NP and OC concentrations in the placebo group (on methotrexate) linearly increased over time, and increases in the simulation are shown in **Table 3**. These findings suggest a slight but significant positive effect of a mixture of methotrexate, placebo, and disease progression on the bone biomarker changes over a long-term period. In a clinical trial of patients with RA receiving tocilizumab or placebo with methotrexate, median P1NP, and OC, CFB in the placebo group seemed to increase at 24 weeks, although it was reported as nonsignificant.[27] In another study, urinary excretion levels of N-telopeptide of type I collagen (bone resorption marker) significantly decreased in patients with RA treated with methotrexate.[28]

In this analysis, the parameter (*SLP*) was estimated using both placebo-treated and drug-treated arms data simultaneously. With the placebo group data only, the *SLP*

estimate (95% CI) was 0.330 ng/mL per week (−0.119, 0.779) for P1NP, compared with 0.162 ng/mL per week (−0.0516, 0.375) in the simultaneous estimation. Although a slight difference between the *SLP* estimates was observed, the variability was large and the impact was <1.4 ng/mL for 8 weeks. Considering the baseline P1NP value (47.0 ng/mL), the difference was considered slight. When *SLP* was fixed to the estimate using placebo group data or estimated with the other parameters using both placebo-treated and drug-treated arms data, *RBmax* and *T50* showed a slight difference (11% and 17%, respectively), and changes for other parameters were ≤5%. Because the study design was not a crossover but a parallel study design, the parameter estimation may have some limitations with the possibility that they may be biased to some extent. However, as the potential extent of the difference was considered slight, the *SLP* estimate in this analysis was considered acceptable.

Regarding the linear biomarker increase observed with placebo, it was initially assumed that the response was the natural history of the disease or sample variability occurring with disease progression, which was common for fosdagrocorat, prednisone, and placebo treatment groups. However, distribution of the EBEs ($\eta_{SLP(i)}$) was different between the placebo and the active drug groups. Additionally, the EBEs were proportional to fosdagrocorat and prednisone doses. To explain the presence of the proportional increase as a rebound effect on positive bone-homeostatic feedback, several models were investigated, such as models that incorporated functions allowing *Ks* to increase when *R* (P1NP or OC) decreases (linearly with a coefficient *RB* for 1/*R* or *BL-R* or nonlinearly with a maximum effect *RBmax* and *RB50* for *1/R* to achieve 50% of *RBmax*, which were investigated with reference to feedback control models).[29] However, such models were unstable without improvement in objective function value. Meanwhile, as we developed this analysis, the empirical time-dependent and dose-dependent function for the rebound effect was successfully incorporated into the model. It should be noted that the common rebound effect for fosdagrocorat and prednisone at the same dose is considered a relatively strong assumption. Although better predictive mechanistic models were not obtained, the empirical model was considered adequate to achieve our objectives. To explain the dose-proportional increase observed in P1NP and OC, further investigation of the system would be required and is beyond the scope of this work. A limitation of this model is that the effect of concomitant medication was not considered.

Jacqmin *et al*.[18] described relationships between ED_{50} and *EDK50* ($ED_{50} = EDK50/KDE$). In the present study, multiple dosing of fosdagrocorat and prednisone q.d. was assumed to be a zero-order rate *Input* (mg/week). Under this assumption, when the drug amount in the effect compartment reaches steady state (i.e., $dA/dt = 0$ hence, $Input = IR$), the *EDK50* at steady state is considered the dosing rate *Input* (mg/week) leading to 50% inhibition of synthesizing the response (defined as $ED_{50,ss}$ hereafter). Considering five half-lives of the drug (ln2/*KDE*), the drug amount at week 8 is considered at steady state in the typical population. The $ED_{50,ss}$ for P1NP and OC correspond to fosdagrocorat 40.1 mg/week (fosdagrocorat 5.7 mg q.d.) and 148 mg/week (fosdagrocorat 21 mg q.d.), respectively (**Table 2**). It indicates that fosdagrocorat 15 mg q.d. exceeds the $ED_{50,ss}$ for P1NP, whereas all the fosdagrocorat doses do not reach the $ED_{50,ss}$ for OC. These results are consistent with the fact that the observed dose-response profile at week 8 for P1NP seemed to reach a plateau by fosdagrocorat 15 mg q.d., whereas the dose-response profile for OC did not reach a plateau up to fosdagrocorat 15 mg q.d. (**Figure 1**). No plateau observed in OC dose-response profile might lead to large uncertainty of the estimate for $ED_{50,ss}$ for OC. However, uncertainty of *EDK50* estimate for OC was considered acceptable based on the SE of the estimate and CIs in the bootstrapping. Therefore, although apparent, plateau was not observed in the dose-response profile for OC, the $ED_{50,ss}$ estimate derived from the *EDK50* would be informative for OC as well as P1NP.

Although the K-PD model is not considered a substitute to the PK-PD approach,[14] as reported in previous studies, we demonstrated the usefulness of the K-PD model and provided quantitative information about dose selection for late-stage clinical trials.

In conclusion, the K-PD model adequately describes the P1NP and OC time course following administration of fosdagrocorat, prednisone, or placebo and was demonstrated to be a useful quantitative tool for simulations to find optimal doses of fosdagrocorat with a reduced effect on bone formation.

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Conflict of Interest. M.C.P., J.H.-H., R.R., and B.K.T. are employees and shareholders of Pfizer Inc. S.S. and A.S. are employees and shareholders of Pfizer Japan Inc. D.J.C. and D.M. were employees and shareholders of Pfizer Inc at the time of the analysis.

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