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Commentary

Target-mediated drug disposition and dynamics

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Abbreviations:
ACE, angiotensin-converting enzyme
ACEI, ACE inhibitor
AUC, area under the plasma concentration–time curve
CL, total systemic drug clearance
EGF, epidermal growth factor
EPO, erythropoietin
G-CSF, granulocyte colony-stimulating factor
GP IIB/IIIa, glycoprotein αIIbβ3 platelet-surface receptor
IFN-β, interferon-beta
LIF, leukemia inhibitory factor
mAb, monoclonal antibody
PD, pharmacodynamics
PK, pharmacokinetics
TMDD, target-mediated drug disposition
t-PA, tissue plasminogen activator
TPO, thrombopoietin
VEGF, vascular endothelial growth factor
V_{ss}, steady-state volume of distribution

ABSTRACT

Nonlinear pharmacokinetics and pharmacodynamics may result from several capacity-limited processes and often represent complicating factors in characterizing the pharmacological properties of drugs. Target-mediated drug disposition (TMDD) corresponds to a special case wherein a significant proportion of a drug (relative to dose) is bound with high affinity to a pharmacological target, such that this interaction is reflected in the pharmacokinetic properties of the drug. Dose-dependent effects on apparent pharmacokinetic parameters may manifest, including the steady-state volume of distribution and total systemic clearance. Although a few small molecular weight compounds have been identified to exhibit TMDD, the incidence of TMDD is likely to increase particularly among emerging biotechnology pharmaceuticals. The goal of this commentary is to describe the basic tenets of TMDD and discuss several mathematical modeling approaches for characterizing this phenomenon. Whereas traditional pharmacokinetic/pharmacodynamic models assume that the amount of the drug–target complex is negligible relative to the total amount of drug in the body, integrated mechanism-based models of TMDD incorporate the binding and stoichiometry of drug–target binding. These models may be utilized to infer the time-course of inaccessible system variables, such as the in vivo density of the drug–target complex, and provide a suitable platform for ascertaining the apparent pharmacodynamic implications of TMDD.

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1. Introduction

Nonlinear conditions in drug disposition and pharmacological effects are encountered frequently, and dose-dependent pharmacokinetic (PK) and pharmacodynamic (PD) processes complicate the characterization of drug concentration-effect and -toxicity relationships [1], often with significant clinical implications [2]. The fundamental principle that governs this behavior is that of capacity-limitation, where limited densities of enzymes or other proteins result in saturable processes and disproportionate changes in net drug exposure or responses with increasing dose. Levy introduced the term target-mediated drug disposition (TMDD) in reference to the observation that for some drugs, the capacity-limited substance responsible for their complex nonlinear pharmacokinetics was in fact the pharmacological target of the compound [3]. Whereas plasma concentrations of most drugs greatly exceed receptor or target concentrations, agents exhibiting TMDD are bound with high affinity and to a significant degree (relative to dose), such that this interaction influences the temporal profile of plasma drug concentrations. Although originally posed to describe the effects of extensive drug–target binding in tissues, TMDD has received considerable interest owing in part to its role in saturable clearance mechanisms for specific peptide and protein pharmaceuticals (e.g., receptor-mediated endocytosis) [4,5]. The utilization of lower doses of these potent compounds, coupled with the means for detecting relatively low drug concentrations in biological fluids offered by advanced analytical methods, further increases the probability of observing this phenomenon.

The pharmacokinetic characteristics imparted by TMDD are now well recognized; however, systematic approaches for understanding the pharmacodynamic implications of TMDD are still in early development. For some compounds, there is an apparent disconnect between the time-course of target-occupancy and the pharmacological response [3]. Pertinent aspects of the in vivo pharmacological properties of drugs derive from the integration of PK/PD systems [6], and progress in comprehending the significance of TMDD will most likely be made through an iterative consideration of experimental data and mechanism-based modeling. This paradigm should yield opportunities for the rational design of new analogues and/or delivery systems for drugs with TMDD properties, as well as clinical dosing regimens that optimize pharmacotherapy across populations and for individual patients.

2. General pharmacological expectations

The pharmacokinetic consequences of TMDD may be subtle or pronounced, but in either case, the effects are important and it is convenient to categorize compounds based on whether or not binding to the pharmacological target significantly contributes to the elimination of the drug. Classic examples of small molecular weight compounds that demonstrate TMDD characteristics, but for which saturable elimination mechanisms are not implicated, include various angiotensin-converting enzyme (ACE) inhibitors (ACEI) [7], imirestat (an aldose reductase inhibitor) [8], and warfarin [9]. Strong experimental evidence suggests that the targeted enzymes of imirestat and warfarin are responsible for their extensive and concentration-dependent tissue binding [9–12], and mathematical modeling has been used to infer TMDD for ACEI [13]. Plasma concentration–time profiles consistent with TMDD also have been shown for bosentan [14], a mixed endothelin-receptor antagonist, and selegiline and desmethylselegiline [15], both monoamine oxidase type B inhibitors; however, definitive experiments confirming TMDD have yet to be reported.

In agreement with the definition of nonlinear systems, dose-normalized concentration–time profiles of drugs exhibiting TMDD do not superimpose, and the most notable and common effects of saturable target binding include dose-dependent changes in the apparent steady-state volume of distribution (V_{ss} decreases to a limiting value as dose increases) and a long terminal elimination phase, particularly when the drug assay is sensitive enough to measure relatively low concentrations. For first doses, the pharmacological target rapidly acquires an initial portion of a dose (commensurate with the capacity of the target and affinity of the drug), prior to general non-specific distribution, thus conferring a dose-dependent V_{ss}, an initial lag phase for relatively low constant rate infusions, and steeper distribution phases for comparatively low doses following rapid administration [3]. As an example, simulated plasma warfarin pharmacokinetic profiles following single oral doses in humans are shown in Fig. 1. The more rapid distribution phase of the lowest dose can be observed (inset plot), along with prolonged parallel terminal elimination phases for all dose levels, which reflect the temporal aspects of warfarin binding to Vitamin K 2,3-epoxide reductase. Interestingly, as originally postulated by Till et al. [7], the terminal phases of drugs with TMDD properties tend to converge to similar concentration values such that the back-extrapolated intercept is constant or dose-independent (with the exception of extremely low doses).

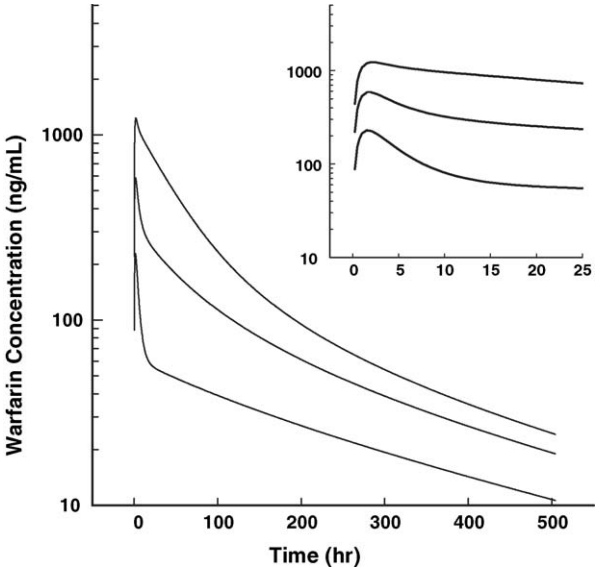

Fig. 1 – Racemic warfarin plasma concentrations after oral doses of 2, 5, and 10 mg in man. Profiles are simulated using a target-mediated pharmacokinetic model and parameter values reported by Levy et al. [34].

Binding to a pharmacological target may represent a major elimination pathway also, and it is well recognized that receptor-mediated endocytosis controls uptake and clearance disposition processes for many peptide and protein drugs [16]. For such systems, the limited capacity of the pharmacological receptors may result in saturable, dose-dependent clearance, which would be further susceptible to modulation based on receptor up- or down-regulation. The PK/PD properties of recombinant human granulocyte colony-stimulating factor (G-CSF) are a good example, as Terashi et al. have shown a significant correlation between G-CSF clearance and the percentage of G-CSF receptor-positive neutrophils in cancer patients [17]. Following chemotherapy, the pharmacokinetics of G-CSF (5 μg/kg IV) was evaluated in five cancer patients at neutropenia (<1000/μL) and neutrophilia (>5000/μL). The administration of G-CSF was associated with an increase in the number of circulating neutrophils, as well as the number of G-CSF receptors per cell (up-regulation), suggesting a mechanism by which G-CSF modulates its own clearance (increasing with increased receptor density). Another classical example is the specific and saturable internalization of erythropoietin (EPO) in erythroid progenitor cells [18]. Chapel et al. demonstrated that the bone marrow, a major site for erythropoietic cells, represents a significant EPO elimination pathway [19]. Bone marrow ablation in sheep, achieved via busulfan or 5-fluoruracil chemotherapy, was associated with a decrease in EPO clearance, thus revealing the in vivo significance of target-mediated uptake and elimination of this drug.

In a similar manner, the pharmacokinetics of murine monoclonal antibodies (mAb) directed against the human CD3 antigen, a component of the T cell receptor/CD3 complex on T lymphocytes, also is influenced by its pharmacological effect [20]. Meijer et al. found that non-mitogenic anti-CD3 mAb accumulated in the serum of renal transplant patients (n = 18) receiving 5 mg/day (IV bolus) for 10 days, and the pharmacokinetics could not be characterized with traditional linear compartmental models. Anti-CD3 mAb administration produced a rapid decrease in circulating T lymphocytes, which gradually returned toward baseline levels over 10 days; however, the density of the antigen was significantly down-regulated for the duration of treatment. Serum drug concentrations were inversely correlated with free antigen expression, demonstrating the target-mediated pharmacokinetic properties of this mAb. Complex nonlinear clearance mechanisms, as typified by G-CSF and anti-CD3 mAb, may accompany dose-dependent distribution characteristics for drugs showing TMDD and also suggest the need to assess the PK/PD properties of these drugs concurrently.

3. PK/PD systems analysis

Noncompartmental analysis of plasma drug concentration–time data represents a useful starting point in characterizing the pharmacokinetic properties of drugs, and curve-fitting single-dose data is a common method for resolving the slopes, heights, area, and moment (SHAM) properties of such curves that are used to calculate primary pharmacokinetic parameters [21,22]. This approach may be applied to initially identify the dose-dependent properties of TMDD. Natalizumab is a humanized mAb directed against the α4β1 integrin, a major adhesion molecule involved in lymphocyte migration, and is approved for the treatment of multiple sclerosis [23]. Sheremata et al. conducted a safety and pharmacokinetic study of natalizumab in multiple sclerosis patients [24], and selected pharmacokinetic parameters obtained from their noncompartmental analysis are listed in Table 1. Intravenous doses ranged from 0.03 to 3.0 mg/kg and both CL and V_{ss} decrease with increasing dose levels, seemingly approaching limiting values. Interestingly, the pharmacokinetic profiles reveal a steeper distribution phase for lower doses, a prolonged terminal elimination phase, and estimates of V_{ss} approach plasma volume as might be expected for relatively large macromolecules. These properties suggest that natalizumab may exhibit TMDD; however, further studies are needed to test this hypothesis. In addition, although noncompartmental analysis may be used to ascertain system linearity, the calculation methods assume in fact that the system is linear and time-invariant. Thus, the numerical values of the dose-dependent V_{ss} may be an artifact and complex nonlinear systems require structural modeling for describing their kinetic properties [25].

Table 1 – Noncompartmental analysis of natalizumab pharmacokinetics in humans

Measurement	Dose, mg/kg (n = 3 or 6)				
	0.03	0.1	0.3	1.0	3.0
AUC (μg h/mL)	0.3	13.5	217	1660	9900
CL (mL/h/kg)	86.9	7.8	1.5	0.6	0.3
V_{ss} (mL/kg)	187	255	74.9	90.7	67.1
$T_{1/2}$ (h)	1.6	24.1	36.9	92.4	108

Adapted from Sheremata et al. [24].

Important methodological issues involved in PK/PD modeling, such as structural model development and selection, choice of minimization algorithm, variance or residual error model specification, objective model-fitting criteria, and model validation and interpretation, are beyond the scope of this commentary [26]. However, one must be cognizant of the fact that all models are approximations that attempt to mimic the salient properties of the system under investigation. The subsequent mathematical models and examples described in this section have been shown to be useful for characterizing the complex nonlinear properties of TMDD systems. Interested readers should consult the original papers for a more complete discussion of these concepts and the implementation of specific PK/PD models.

The classical equation for characterizing nonlinear kinetics is the Michaelis-Menten equation [27]:

$$v = \frac{V_{max}C}{K_m + C} \quad (1)$$

where v traditionally represents the velocity of a disposition process, such as metabolism or transport, V_{max} the capacity constant, K_m represents a sensitivity or affinity term, and C is the substrate or ligand concentration. The nature of this equation is fundamental to quantitative pharmacology and various forms of the equation are used to describe a range of capacity-limited systems including: drug metabolism and

transport, protein and receptor binding, and direct reversible pharmacological effects [28]. For protein and receptor binding, the analogous Michaelis-Menten equation is defined as:

$$RC = \frac{R_{tot}C}{K_D + C} \quad (2)$$

where RC represents the concentration of bound drug or the drug–receptor complex, R_{tot} the capacity term, which is defined by the product of the total protein or receptor concentration and the number of binding sites per target molecule, and K_D is the equilibrium dissociation constant. Thus, as free drug concentrations approach high values (i.e., $C \gg K_D$), the bound concentration approaches a limiting value ($RC \rightarrow R_{tot}$), and at relatively low concentrations, $RC \rightarrow R_{tot}C/K_D$.

Based on the equilibrium condition described by Eq. (2), several investigators have evaluated the effects of saturable plasma protein and tissue binding on pharmacokinetic profiles [29–31]. Computer simulations of such nonlinear binding models reveal that log plasma concentration–time profiles may be linear, convex, or concave depending on the extent of drug distribution and the efficiency of elimination [29,30]. In addition, certain scenarios are associated with time-dependent pharmacokinetic parameters, such as the apparent volume of distribution approaching a limiting value with increasing time. Wagner introduced a series of equilibrium nonlinear binding models, which represent various cases of an overall general theory [31]. The general scheme resembles a two-compartment model with drug input and elimination occurring to and from one of the compartments. Drug may bind to one or more ''tissue'' types in either compartment, each characterized by separate capacity and affinity parameters, and only free drug is available for distribution and elimination. Consider the case where drug binds to one specific tissue, accessible from the central compartment, and no saturable binding occurs in the peripheral compartment. The rate of change of drug concentration in the central compartment (C_p) following an IV bolus dose for this specific model is:

$$\frac{dC_p}{dt} = \frac{k_{21}C_t - (k_{12} + k_{el})C_p}{1 + (R_{tot}K_D/(K_D + C_p)^2)} \quad (3)$$

where the first-order rate constants k_{12} and k_{21} govern distribution to and from the peripheral compartment (C_t), and k_{el} is a first-order elimination rate constant. A combination of simulations and analytical solutions were used to derive an expression for the initial plasma concentration (C_0) and slope of the terminal elimination phase (λ_z), and to show that bound tissue concentrations decrease in parallel with plasma concentrations.

Modifications to these classical models of plasma protein and tissue binding have been used to describe the kinetics of several drugs exhibiting TMDD. For example, the pharmacokinetics of several ACEI have been well characterized with the nonlinear model derived by Lees et al. [13]. Although utilizing an alternative derivation, the structure of the final model resembles the one-compartment model of Wagner that includes access to two different tissue types [31]. In this case, the two tissue types reflect the same target, namely circulating and tissue bound ACE. Whereas the maximum binding capacity may be divided between the two sites, the identical affinity (or dissociation constant) parameter may be used to describe the thermodynamics of target binding at each site. In an analogous manner, the model for imirestat pharmacokinetics in humans consists of saturable tissue binding in a peripheral compartment and linear binding in the central compartment, with linear drug distribution and elimination operating only on free drug [8]. Using an alternative parameterization, Snoeck et al. coupled the target-binding process with rate constants to describe the nonlinear pharmacokinetics of draflazine in humans [32]. Similar to the ACEI model, distinct capacity terms were included to reflect specific binding to red blood and endothelial cells, but a single K_D parameter was used to define drug affinity for the common nucleoside transporters. These traditional concepts have been implemented also in physiologically based pharmacokinetic models of drugs, such as methotrexate [33] and warfarin [34]. For these models, the tissue-serum or plasma partition coefficients (K_P) are concentration dependent. As an example, the K_P values of S-warfarin for non-adipose tissues in rats were described by the equation [34]:

$$K_p = \frac{R_{tot}}{(K_D/ff) + C} + P \quad (4)$$

where ff is the drug free fraction and the P term represents a proportionality constant when specific binding is relatively low or negligible. Interestingly, the rank-order of tissue binding capacities (R_{tot}) correlates with that of Vitamin K 2,3-epoxide reductase activity [3], supporting the concept of target-mediated warfarin kinetics.

Receptor-mediated clearance represents an additional capacity-limited system that reflects the concept of TMDD and also has been described using classical Michaelis-Menten kinetics. The significance of the receptor-mediated clearance mechanism for human anti-CD11 mAb has been investigated with in vitro and in vivo systems [35], and an open two-compartment pharmacokinetic model, with parallel nonlinear and linear elimination, well captured the dose-dependent kinetics of this drug in chimpanzees and humans [36,37]. This approach corresponds to the classical technique of defining a concentration-dependent clearance term according to Eq. (1). However, such models do not account for nonlinear distribution processes and assume apparent compartment volumes to be independent of concentration or dose. A combination of Michaelis-Menten functions to describe drug elimination and distribution to a peripheral compartment may be applied to simultaneously characterize nonlinear clearance and volume terms [38]. On the other hand, one might hypothesize that both phenomena manifest from interactions with a single pharmacological target and a more mechanistic model could be derived. Furthermore, classical nonlinear binding or transport models do not provide a means for describing the turnover or up- or down-regulation of the capacity term in a mechanistic manner.

A general mechanistic pharmacokinetic model of TMDD has been developed for suitable plasma concentration–time profiles [39], and most contemporary applications are reflected in the schemes shown in Fig. 2. For case A, the pharmacological target is directly accessible from the central compartment, whereas in case B, distribution to a peripheral compartment is required. This distinction may represent a

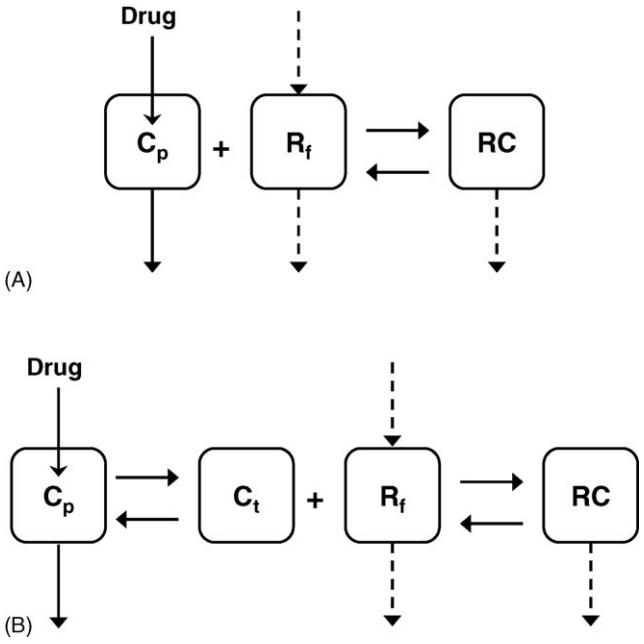

Fig. 2 – Schematic of general pharmacokinetic models of target-mediated drug disposition including a directly accessible pharmacological target-pool (A) or prior distribution to a peripheral compartment (B). Optional rates of "receptor" (R_f) turnover processes and elimination of the drug–target complex (RC) are shown with dashed arrows. Drug binding to the pharmacological target may be characterized with binding micro-constants or assuming equilibrium conditions as described in the text.

modeling artifact where the target in case A might be localized in a tissue; however, achieving concentrations at the biophase may not be rate-limiting as illustrated by case B. For both cases, optional rate constants describing the production and elimination of free receptor (or target, R_f) and the uptake or elimination of the drug–target complex are shown (dashed arrows). An additional compartment with first-order rate constants of distribution could be linked with the central compartment to account for non-specific drug distribution or binding as well. Assuming total receptor concentration is time-invariant, the free receptor concentration may be defined as $R_{tot} - RC$, and the rate of change of drug concentration in the central compartment for the simple one-compartment case after rapid IV administration is:

$$\frac{dC_p}{dt} = -k_{el}C_p - k_{on}(R_{tot} - RC)C_p + k_{off}RC \quad (5)$$

where k_{on} is the second-order association rate constant, and k_{off} is the first-order dissociation rate constant (N.B., concentration variables are often best modeled in molar units). Hence, at relatively high drug concentrations, $RC \rightarrow R_{tot}$ and $R_f \rightarrow 0$, thereby saturating the formation rate of the drug–target complex. This modeling approach depicts pharmacological target binding as the key process controlling the complex nonlinear profiles and is based on physiological [40] and cellular kinetic [41,42] models of peptide disposition. Computer simulations show that under such conditions (case A) the apparent volume of distribution decreases with increasing dose, approaching a limiting value [39]. In addition, if the target-complex undergoes significant internalization or metabolism, then this system exhibits a dose-dependent clearance, again decreasing with increasing dose and approaching a limiting value. Importantly, simulated pharmacokinetic profiles are in accordance with expected properties of TMDD, including: poly-exponential kinetics, steeper distribution phases at relatively lower doses, prolonged terminal phases, convergence of concentrations to similar values at later times (again with the exception of extremely low doses), and bound concentrations decrease in parallel with that of the central compartment. Applied variations of the general model were shown to capture the time-course of drug concentrations for imirestat, bosentan, and interferon-beta (IFN-β) 1a in humans [39].

The versatile and mechanistic nature of the general TMDD model provides a framework for the systematic evaluation of complex nonlinear PK/PD processes and has been used to characterize the pharmacokinetics of several drugs in animals and humans. A clinical study by King et al. [43] evaluated the pharmacokinetics of warfarin given as single oral doses of 2, 5, and 10 mg (racemic warfarin). These data were re-analyzed using a simple TMDD model (Fig. 2A), in which Eq. (5) was modified to include drug absorption characterized by a linear first-order rate constant [34]. The mean pharmacokinetic data were well described and the pharmacological properties of the system were qualitatively similar to pre-clinical data. Eppler et al. developed a target-mediated model to describe the pharmacokinetics of vascular endothelial growth factor (VEGF) following a 4-h IV infusion (17 or 50 ng/kg/min) in patients with coronary artery disease [44]. The final model reflects an open two-compartment model with zero-order input and first-order elimination to and from the central compartment, and included a saturable, irreversible target-binding component with first-order elimination of the bound species. Analogous to Eq. (5), the rate of change of plasma VEGF concentrations was defined as:

$$\frac{dC_p}{dt} = \frac{K_0}{V_c} - (k_{el} + k_{12})C_p - k_{on}(R_{tot} - RC)C_p + k_{21}C_t \quad (6)$$

in which K_0 is the zero-order infusion rate and V_c is the volume of the central compartment. The authors mention that a successful retrospective analysis of other VEGF pharmacokinetic data further validated the model, suggesting it is robust and may be used to simulate additional dosing regimens to optimize safety and efficacy to be evaluated in future trials.

Computer simulations and analytical expressions of R_{tot} at steady-state suggest that assuming a time-invariant R_{tot} is appropriate under certain conditions and supports the definition of free target concentration, as shown in Eqs. (5) and (6), as a parsimonious first step in defining a TMDD model [39,40]. However, an equation describing the kinetics of free target or receptor may be added if such information is known, if required for improving model fitting, or if functional adaptation processes result in significant changes in target or receptor turnover (for either single or multiple-dosing).

Such TMDD models have been developed for the pharmacokinetics of thrombopoietin (TPO) [45] and leukemia inhibitory factor (LIF) [46]. The turnover of free receptor might be specified as:

$$\frac{dR_f}{dt} = k_{syn} - k_{on}C_pR_f + k_{off}RC - k_{deg}R_f \quad (7)$$

where k_{syn} represents a zero-order production rate constant and k_{deg} is a linear first-order degradation rate constant. Assuming that no drug is present in the system at time zero, the initial condition of Eq. (7) is defined as $R_f(0) = R_{totss}$, which is estimated during model fitting, and if stationarity is assumed also, then $k_{syn} = k_{deg}R_{totss}$. Thus, the inclusion of Eq. (7) and the kinetics of free receptor or target concentrations only increases the number of system parameters by one (k_{deg}). The final model of TPO pharmacokinetics in humans also contained a positive feedback loop, whereby the bound drug–receptor complex served to stimulate the production of free receptors in accordance with its pharmacodynamic effect (TPO stimulates the production of platelets and thus the number of receptors available for binding) [45]. The pharmacokinetic study of LIF was conducted in sheep and included a large range of IV dose levels (12.5–750 μg/kg), as well as three doses administered subcutaneously (SC). Owing to the contribution of lymphatic transport of proteins after SC injection [47], a delayed absorption compartment was incorporated into the final TMDD model of LIF kinetics, similar to the approach developed by Radwanski et al. [48]. These studies serve to underscore the flexibility of the general TMDD model and several techniques for incorporating various system complexities.

One limitation of the general TMDD model is that the target-binding rate constants (k_{on} and k_{off}) are often not readily identifiable from typical in vivo pharmacokinetic data. As a result, constraints on model parameters are frequently utilized, where binding constants may be fixed according to in vitro estimates, or one of the constants may be estimated, such as k_{on}, and the other may be calculated from the relationship $k_{off} = k_{on}K_D$, with K_D fixed to a known value. Of course, if such a problem occurs and the system can be well described using a traditional equilibrium modeling approach, then implementing the kinetic processes of the general model may not be necessary. For example, Kemme et al. developed a three-compartment circulatory model, with specific saturable binding in the intermediate compartment, to characterize the pharmacokinetics of tissue plasminogen activator (t-PA) in healthy male volunteers [49]. This model essentially incorporates a distribution process prior to making drug available for interactions with free receptors (Fig. 2B). Interestingly, model simulated t-PA concentrations, evaluating a range of R_{tot} and K_D values, suggest that the model is relatively insensitive to changes in K_D as compared to the significant volume of distribution effects elicited by changes in R_{tot}. In order to allow for the turnover processes of receptor or target kinetics, which are not provided for in traditional nonlinear protein binding models, a quasi-equilibrium solution to the general TMDD model has been derived [50]. Instead of ordinary differential equations describing the time-course of free drug and receptor or target concentrations, the system is defined by differentials for total drug concentration ($C_{tot} = C_p + RC$) and R_{tot}, and the measurable free drug concentrations are calculated according to the explicit equation:

$$C_p = 0.5\left[(C_{tot} - R_{tot} - K_D) + \sqrt{(C_{tot} - R_{tot} - K_D)^2 + 4K_DC_{tot}}\right] \quad (8)$$

This new modeling approach was applied to the LIF pharmacokinetic data following IV administration, which were previously analyzed using the general TMDD model [46]. The equilibrium model resulted in essentially identical predicted profiles as compared with the general model, and parameter estimates were in relatively good agreement with previous values. Care must be taken to ensure that assumptions of equilibrium conditions are valid and additional metrics have been proposed for quick verification [50].

Mathematical models of TMDD present a means for exploring the pharmacodynamic implications of substantial binding to pharmacological targets. Although a general modeling approach for characterizing the pharmacokinetics of TMDD has been described, the plethora of mechanisms by which drugs elicit their pharmacological effects precludes the development of a single model for defining corresponding PK/PD relationships. The hypothesis that the time-course of target-occupancy directly correlates with the kinetics of pharmacological response is intuitive. However, the situation can be complicated for drugs such as warfarin, where anticoagulation can return toward baseline conditions in the presence of significant binding with the target enzyme [51]. Careful consideration must be made for mechanisms of action, and appropriate pharmacodynamic models should be based on the pharmacological properties of drugs and the relevant rate-limiting steps in the biology of the system [52,53].

Presently, it is convenient to distinguish between those compounds that act through antagonistic mechanisms from those that function as direct agonists. For drugs that exhibit TMDD and also inhibit or block enzymes or receptors, it appears that their pharmacodynamics are often correlated with free drug concentrations in plasma, directly or indirectly, as opposed to the time-course of target-occupancy. The ex vivo measurements of target enzyme activities following the administration of several ACEI and warfarin have been described by a simple inhibitory sigmoidal E_{max} model [51,54]:

$$E = E_0\left(1 - \frac{I_{max}C_p}{IC_{50} + C_p}\right) \quad (9)$$

where E represents the observed effect, E_0 the baseline effect or $E(0)$, I_{max} the maximal inhibitory effect, and IC_{50} is the plasma drug concentration producing 50% of E_{max}. This same pharmacodynamic model was linked with one of Wagner's nonlinear binding models (Eq. (3)) to characterize the PK/PD properties of abciximab simultaneously in patients undergoing coronary angioplasty [55]. Abciximab is a mAb Fab fragment directed against the glycoprotein (GP) IIb/IIIa platelet surface receptor, which mediates aggregation by binding to adhesive molecules, such as fibrinogen, after activation by endogenous agonists. The antiplatelet effects of abciximab are desired clinically to prevent ischemic complications during coronary angioplasty and atherectomy procedures. Simulations of the final PK/PD model for abciximab reasonably predicted ex vivo measurements of receptor-occupancy, and more importantly, suggest that patients with higher receptor

concentrations (R_{tot}) might exhibit response profiles reflecting reduced efficacy, which may be supported by recent clinical observations [56].

In contrast to enzyme antagonists that display TMDD properties, the temporal aspects of receptor-occupancy typically drive the effects of direct receptor agonists. Eppler et al. observed a decrease in mean arterial pressure following a 4-h infusion of VEGF, and although a pharmacodynamic model was not applied, simulated concentration–time profiles of irreversibly bound VEGF showed an apparent relationship with the time-course of this hemodynamic effect [44]. Mechanistic pharmacodynamic models that utilize the time-course of the bound drug–receptor complex as a forcing function have been linked to TMDD models to characterize simultaneously the PK/PD properties of digoxin in an isolated perfused rat heart experiment [57] and IFN-β 1a in cynomolgus monkeys [58] and humans [59]. The digoxin model included two distinct receptor populations, and the time-course of positive inotropic effect (E) was assumed to be directly proportional to receptor occupancy and defined by an explicit equation, $E = e_1RC_1 + e_2RC_2$, where e_1 and e_2 are estimated sensitivity parameters. For IFN-β 1a, plasma neopterin concentrations were measured after IV and SC drug administration, and resulting profiles of this biomarker were biphasic, with an initial onset delay, a rise to maximal values, and a gradual return toward baseline conditions (Fig. 3). The final pharmacodynamic model for neopterin dynamics following single-dose administration of IFN-β 1a is driven by the bound drug–receptor complex and incorporates a single transit compartment coupled to a precursor-dependent indirect response model [60]. This model has a mechanistic basis and reflects the major processes by which IFN-β 1a is thought to elicit neopterin induction. From a systems analysis perspective, model simulations were used to evaluate inaccessible variables and the apparent greater efficacy following SC dosing could be explained by the temporal profiles of the transit compartment relative to an estimated EC_{50} parameter [58]. Two submodels of tolerance, including receptor down-regulation and negative feedback inhibition by neopterin, were required to describe the slight drug accumulation and less than expected neopterin concentrations following multiple-dosing [58], which are directly or indirectly supported by in vitro and clinical observations.

The PK/PD model of IFN-β 1a depicts capacity-limited receptor binding as the central feature connecting complex nonlinear drug disposition with pharmacodynamic signal transduction processes. Such systems based on principles of TMDD may necessitate the simultaneous modeling of kinetic and dynamic data, and the step-wise model development and evaluation approach for IFN-β 1a may serve as a useful paradigm for characterizing respective PK/PD properties. Complex models of this type are likely to become more common as evidenced by the recent receptor-mediated PK/PD models for macromolecules targeting CD11a [37] and CD11b [61] cell surface receptors. The anti-CD11b PK/PD model constructed by Jonsson et al. successfully combines three mechanistic submodels of neutrophil maturation and proliferation, CD11b receptor up-regulation, and three drug elimination pathways including (1) linear first-order elimination, (2) capacity-limited receptor-mediated elimination, and (3) saturable non-receptor-mediated clearance. Importantly, their mechanism-based model is implemented using a population mixed-effects modeling approach and explains the apparent pharmacodynamic differences between patients and healthy volunteers, related to neutrophil dynamics and total number of CD11b receptors per neutrophil, which could not be obtained with traditional empirical models. Finally, the potential exists for linking receptor-mediated biomarkers with clinical outcomes. Ng et al. also utilized population modeling for assessing efalizumab PK/PD in psoriasis patients and included an efficacy model of disease severity, which correlates the rate of psoriasis skin production with the amount of free surface CD11a T cell receptors [37].

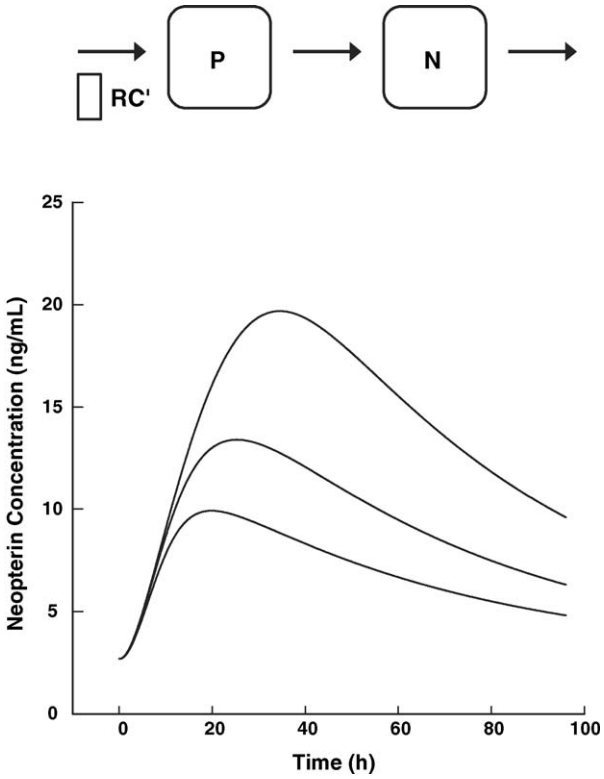

Fig. 3 – Neopterin plasma concentrations following IV doses of interferon-β 1a (1, 3, and 10 million IU/kg) in monkeys. Profiles are simulated using the integrated pharmacokinetic/pharmacodynamic model and parameter values reported by Mager et al. [58]. The pharmacodynamic component of the model (top) reflects a precursor-dependent indirect response model [60], where internalized drug–receptor complex (RC′) stimulates the production of a neopterin precursor (*P*), which gets converted to neopterin (*N*).

4. Future considerations

Appreciation for TMDD properties and the pharmacodynamic implications of such systems will indubitably increase following the acquisition of much needed experimental data coupled with the continued development and refinement of mechanism-based PK/PD models of this phenomenon. Major

goals continue to include the identification of drug and system specific parameters that control exposure-response relationships, as well as patient specific characteristics or covariates that account for inter-subject pharmacodynamic variability. One field that is likely to significantly contribute to our understanding of TMDD systems is in vivo molecular imaging. As noted by Levy et al., the general model of TMDD [39] (as well as other plasma concentration-based TMDD models) requires a suitable range of drug doses and concentrations, and direct detailed information regarding tissue and cellular pharmacokinetics are only obtainable from animal studies and physiologically based modeling approaches [34]. However, advanced in vivo imaging methods, such as positron emission tomography (PET) scanning, allow for the noninvasive imaging of the disposition and pharmacodynamics of targeted drugs [62–64]. The mathematical modeling of dynamic PET data has been used to quantify benzodiazepine receptor density and apparent in vivo receptor binding constants [65]. Such details may be used eventually to connect traditionally measured plasma drug concentrations with tissue kinetics and pharmacodynamic measurements of drugs exhibiting TMDD in humans. In addition, TMDD principles may be critical for interpreting data from human microdosing experiments, where a sub-pharmacological dose is administered and plasma drug concentrations are measured using accelerator mass spectrometry to obtain pharmacokinetic properties earlier in the drug development process [66]. Lappin and Garner note that, "... if high-affinity binding compartments exist then plasma kinetics at microdoses and pharmacological doses may differ substantially" [66].

Lastly, target-mediated PK/PD models may be of particular importance in characterizing the properties of advanced targeted drug delivery systems. For protein drugs with TMDD properties, drug delivery systems that alter protein–protein interactions, such as drug conjugation with polyethylene glycol [67] or specific peptide residue mutations [68], may substantially affect both drug disposition and pharmacological properties. A pegylated formulation of IFN-β 1a, for example, demonstrated enhanced drug exposure in monkeys; however, this was countered with a relatively large EC_{50} value (as compared with plasma drug concentrations) resulting in net pharmacological effects similar to those of unmodified drug [69]. Interferon-β also has been administered along with a soluble type I interferon receptor, resulting in enhanced exposure and antitumor activity in xenografted SCID mice [70]. Mathematical modeling of such delivery systems may provide key insights into appropriate dosing regimens (considering both amount/capacity and dosing frequency), optimal binding affinity, and whether an increase in net exposure and/or receptor-mediated effects may be anticipated from natural or artificially designed drug-binding ligands. These concepts and TMDD models may be extended directly to targeted drug delivery through specific receptor systems, such as the transferrin [71] and folate [72] receptor-mediated endocytosis pathways.

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