The influence of plasma protein binding on the pharmacokinetics and pharmacodynamics of S(-)-Propranolol

Tamara van Steeg[a], Elke Krekels[a], Jan Freijer[b], Fiona Macintyre[c], Elizabeth C.M. de Lange[a] and Meindert Danhof[a,b]

[a] LACDR, Division of Pharmacology, Leiden University, P.O. Box 9502, 2300 RA Leiden, The Netherlands
[b] LAP&P Consultants BV, Archimedesweg 31, 2333 CM Leiden, The Netherlands
[c] Pfizer, Clinical Pharmacokinetics, Sandwich Laboratories, Sandwich Kent CT13 9NJ, United Kingdom

INTRODUCTION

Protein binding can have a major impact on a drug's pharmacokinetics (PK) and pharmacodynamics (PD). At present the theoretical basis of the influence of plasma protein binding on PK is well-established[1]. Elimination of a drug from the body is often described as restrictive or non-restrictive depending on the clearance properties of the compound. The "free drug hypothesis" states that the pharmacological activity of a drug (pharmacodynamics) is determined by the unbound drug concentration *in vivo*. This implies that plasma protein binding is always restrictive for the pharmacodynamics of a drug. There are however indications that for certain drugs the pharmacodynamics are non-restrictive (the total rather than the free drug is responsible for the effect)[2]. Therefore the role of plasma-protein binding on pharmacodynamics should be established in a systematic manner.

OBJECTIVE

It is hypothesised that plasma protein binding is non-restrictive for pharmacodynamics if the affinity for the receptor is higher than the affinity for the plasma protein. To investigate this hypothesis simulations were performed using a mechanism-based PD model describing receptor-occupancy as a function of both affinity for plasma protein and receptor.

Under the assumption that plasma protein binding is restrictive for the pharmacodynamics of a drug, the concentration-effect relationship will shift if plasma protein binding is altered. Therefore the objective of the presented research is to determine the influence of alterations in plasma protein binding on the concentration-effect relationship of S(-)-Propranolol both *in silico*, using the developed PD model, and corroborate the findings *in vivo*.

IN SILICO

Methods

In order to determine the influence of the affinity of the drug for both protein and receptor on in vivo drug effects, simulations were performed using Berkeley Madonna (Version 8.0.1, Macey & Oster, California, USA).

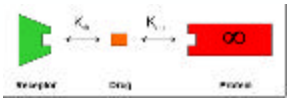


Figure 1 *Schematic representation of drug binding to receptor in presence of plasma protein*

Receptor occupancy was described as a function of both the equilibrium dissociation constant for receptor binding (Kdr) and protein binding (Kdp). Binding of the drug to protein was assumed to be non-saturable and meaningful values for Kdr and Kdp were obtained from literature. A schematic representation of the model is presented in figure 1.

Equations

The theoretical relationship between the free fraction (φ) and the affinity constant for protein binding (K_{dp}) in absence of receptor can be described by the following equation;

$$j = \frac{1}{\frac{Kdp}{[P]}+1}$$

The free drug ([A]) in presence of receptor can be described by the following equation:

$$[A] = \frac{[D]-[Rt]-Kdr-CKdr+\sqrt{([D]-[Rt]-Kdr-CKdr)^2+4Kdr(1+C)[D]}}{2(1+C)}$$

Where [D] is the total drug concentration, [Rt] is the total receptor concentration, K_{dr} is the equilibrium dissociation constant for receptor binding and C=[P]/K_{dp} is the binding constant for protein binding.

Consequently receptor occupancy can be described using the following equation:

$$Occupancy = \frac{[D]-[A]-C.[A]}{[Rt]}*100 = \frac{[RA]}{[Rt]}*100$$

Results

The theoretical relationship between the affinity constant for protein binding an the free fraction is presented in figure 2a. In figure 2b the influence of different K_{dp} values on receptor-occupancy is shown.

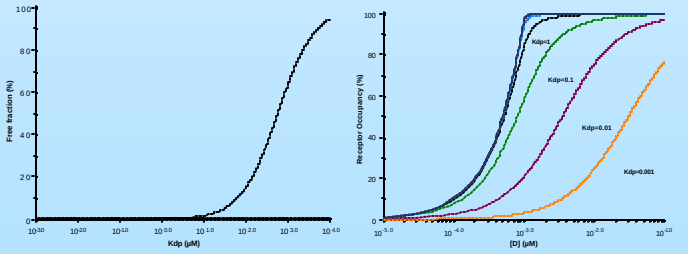


Figure 2 *A. Simulation of the relationship between the affinity constant for protein binding (K_{dp}) and the free fraction B. Shift in concentration-occupancy curve with different values for Kdp (µM)*

IN VIVO

Methods

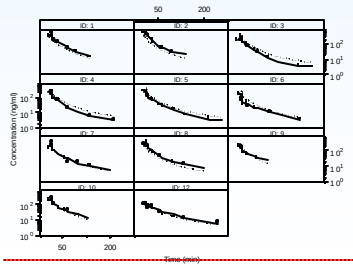
Male WKY rats, instrumented with four cannulas, received an IV-infusion (jugular vein) of S(-)-Propranolol (1mg/kg, 15 min) under isoprenaline-induced tachycardia (5 µg/kg/h). The other cannulas were implanted in the right and left femoral artery for both bloodsampling and heart rate measurements.

Previously results showed a ten tot fifteen fold increase in plasma a1-acid glyoprotein (AGP) levels in rats at two days post cannulation, with a return to baseline within seven days post cannulation. Therefore *In vivo* PK-PD experiments were performed on day two and seven post surgery.

An in-house available S-plus (Insightful corp., Seattle, USA) interface to NONMEM (Version V, level 1.1, NONMEM project group, university of California, San Francisco, USA) was used for data-analysis.

Preliminary Results

All individual pharmacokinetic profiles are described with a 3-compartment model (Figure 3). Inter individual variability was observed for both CL and V2. Both the IIV and the residual error variability were described using a proportional error model.



Parameter	Value (CV%)
V1(ml)	134 (15)
CL (ml/min)	30.3 (13)
V2 Day 2(ml)	569 (14)
V2 Day 7 (ml)	936 (19)
Q2 (ml/min)	68.4 (11)
V3(ml)	1230 (57)
Q3(ml/min)	8.91 (13)
IIV CL	0.1080 (34)
COV CL-V2	0.1320 (33)
IIV V2	0.1850 (39)
Residual error	0.0335 (19)

Figure 3 *Pharmacokinetics profiles S(-)-propranolol, increased plasma protein binding (ID 1-5) normal protein binding (ID 6-12)*

Data-analysis showed that a significant correlation between AGP, one of the major binding proteins, and V2 exists (-0.576, p<0.01). To fully characterise the relationship between V2 and AGP more data are required. The pharmacodynamic data-analysis is still in progress

DISCUSSION

• Usually in literature protein binding is reported in percentage bound instead of a K_{dp}. Therefore figure 2a shows the theoretical relationship between Kdp and the free fraction. For a drug with a high receptor affinity (K_{dr}=1e-6 µM) the simulations show that the influence of protein binding is limited if K_{dp}>0.1 µM. Theoretically a drug with a Kdp of 0.1 µM has a protein binding of approximately 99%. However in future literature data should be used to confirm the relationship between K_{dp} and the free fraction.

• The obtained concentration-effect relationship for S(-)-Propranolol will be used to evaluate whether or not the mechanism-based PD model predicts the influence of plasma protein binding on pharmacodynamics. Heart rate under isoprenaline-induced tachycardia is used as a biomarker for receptor occupancy. Figure 4 shows the predicted shift in the target-occupancy curve for S(-)-Propranolol.

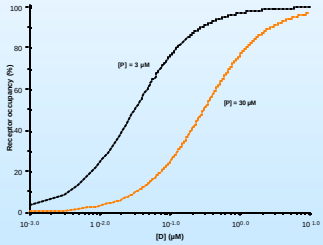


Figure 4 *Predicted shift in the target occupancy curve of S(-)-Propranolol due to a ten-fold change in protein binding.*

CONCLUSIONS

• Simulations show that the role of plasma protein binding in *in vivo* pharmacodynamics can be identified using a mechanism-based PD model.

• For drugs with a high affinity for the receptor protein binding can be non-restrictive for pharmacodynamics even though protein binding might be up to 99% (Kdp=0.1 µM).

REFERENCES

1. Rowland & Tozer, (1985), Clinical pharmacokinetics – concepts and applications, 3rd edition
2. Van der Graaf *et al*, (1997), J. Pharmacol. Exp. Ther 283: 809-816