Part IV

Pharmacological and Clinical Consequences of Drug-Protein Binding

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DRUG DISPLACEMENT INTERACTIONS: A CASE STUDY OF THE PHENYLBUTAZONE-WARFARIN INTERACTION

Edward M. Sellers

INTRODUCTION

The mechanisms, frequency, and clinical importance of drug interactions have generally not been carefully studied. Interactions allegedly based on displacement of highly protein-bound drugs from plasma proteins continue to be particularly misunderstood. The evidence supporting the role of such interaction is particularly incomplete (Sellers, 1979; Koch-Weser and Sellers 1976). On theoretical grounds, simple displacement interactions are not likely to be common, easy to detect, or serious. In those interactions where displacement has been most convincingly shown, and a clinically important consequence ensues, an additional interaction mechanism has always been present.

In this chapter, I shall review the theoretical basis of drug protein displacement interactions, examine the warfarin-phenylbutazone interaction in relationship to theory, and propose research tactics needed to document displacement effects in drug interaction studies.

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THEORETICAL BASIS OF INTERACTIONS AT STEADY STATE

Small Hepatic Extraction Ratio Drugs

Many drugs undergo extensive biotransformation in the liver, exhibit a low blood clearance, and therefore have a small hepatic extraction ratio (Wilkinson and Shand 1975). The principles that pertain to hepatic drug clearance are applicable to dispositional drug interactions involving the liver. The mean steady-state total concentration in blood of a chronically administered drug (Ct_{ss}) that undergoes only hepatic elimination is:

$$Ct_{ss} = \frac{F \cdot Dose}{Cl_h \tau} \qquad \text{Eqn 1}$$

where Cl_h, τ, and $F \cdot Dose$ are the hepatic clearance, dosing interval, and the dose absorbed, respectively. Hepatic clearance relates to hepatic blood flow (Q_h) and the hepatic extraction ratio (E):

$$Cl_h = Q_h E \qquad \text{Eqn 2}$$

Rowland, Benet, and Graham (1973) have defined the hepatic extraction ratio:

$$E = \frac{Cl_{int}}{Q_h + Cl_{int}} \qquad \text{Eqn 3}$$

where Cl_{int} is the total intrinsic clearance describing the maximal ability of the liver to remove an agent from the blood in the absence of any hepatic blood flow restrictions. If only the free (unbound) drug in the blood is available for extraction, then:

$$E = \frac{\alpha Cl_{int'}}{Q_h + \alpha Cl_{int'}} \qquad \text{Eqn 4}$$

where α is the fraction of free drug in the blood and $Cl_{int'}$ is the intrinsic hepatic clearance of free drug. Combining eqns. (2) and (4) and substituting into eqn. (1) results in eqn. (5).

$$Ct_{ss} = \frac{F \cdot Dose(Q_h + \alpha Cl_{int'})}{\tau Q_h \alpha Cl_{int'}} \qquad \text{Eqn 5}$$

Since the free concentration of a drug in the blood is the product of the free fraction (α) (expressed as percent unbound) and the total concentration (Ct_{ss}), the mean steady state free concentration (Cf_{ss}) is defined:

$$Cf_{ss} = \frac{F \cdot Dose(Q_h + \alpha Cl_{int'})}{\tau Q_h Cl_{int'}} \qquad \text{Eqn 6}$$

Provided processes embodied in the intrinsic hepatic clearance term do not become saturated, eqns. (5) and (6) predict a direct relationship between the effective dose absorbed (F • Dose) and the mean steady-state concentrations in blood. Therefore, chronic concomitant administration of any agent which decreases the effective dose of an oral anticoagulant absorbed will decrease both the mean free and total steady-state anticoagulant blood levels.

The consequences of changes in hepatic blood flow (Q_h), intrinsic clearance of the free drug ($Cl_{int'}$), or the free fraction (α) are not immediately apparent from eqns. (5) and (6). The effect of changes in these parameters can be illustrated with warfarin. Schary, Lewis, and Rowland (1975) report a long-term multiple dose study investigating the warfarin-phenylbutazone interaction in a single normal subject. In the absence of phenylbutazone, chronic dosing with warfarin (10 mg per day) resulted in mean steady-state free and total plasma warfarin concentrations of 0.02 and 4.10 mg per liter, respectively. Assuming the partition coefficient of warfarin between plasma and erythrocytes in whole blood is very high, the hematocrit is 0.5, and the hepatic blood flow is 1.5 L per min: the hepatic blood clearance (Cl_h), extraction ratio (E), total intrinsic clearance (Cl_{int}), and intrinsic clearance of the free drug ($Cl_{int'}$) are approximately 3.0 ml per min, 0.002, 3 ml per min, and 600 ml per min, respectively. Since a $Cl_{int'}$ is small compared to Q_h, eqns. (5) and (6) may be simplified to eqns. (7) and (8):

$$Ct_{ss} \simeq \frac{F \cdot Dose}{\tau \alpha Cl_{int'}} \qquad \text{Eqn 7}$$

$$Cf_{ss} \simeq \frac{F \cdot Dose}{\tau Cl_{int'}} \qquad \text{Eqn 8}$$

These equations indicate that:

1. The change in plasma protein binding results in a change in Ct_{ss} but not Cf_{ss}. The chronic concomitant use of an agent that dis-

places drug from plasma proteins (increase in α or decrease in the apparent association constant) would not change the mean steady-state free level, but would lower the mean steady-state total concentration in the blood. A variety of theoretical, animal, and other clinical studies provide evidence consistent with the usual lack of changes in free drug concentration at steady state (Wardell 1974). The most important determinant of the intensity of a drug's clinical action does not depend on the extent of protein binding. This is to be expected, since α and C_f are determined by two different and independent physiologic processes; α depends on the physicochemical interaction of the drug with protein (reflected in the association constant Ka = k1/k2), whereas C_f depends on the balance between the rate at which the drug enters the body and the rate of elimination of unbound drug by biotransformation or excretion (Greenblatt, Sellers, and Koch-Weser 1982).

2. Steady state blood drug concentrations are sensitive to changes in $Cl_{int'}$. Thus, any increase or decrease in this parameter as in hepatic microsomal enzyme induction, or metabolic inhibition, respectively, results in proportional inverse changes in Cf_{ss} and Ct_{ss}.

3. Steady-state blood drug concentrations are insensitive to hepatic blood flow changes. Therefore, concomitant therapy of a low clearance drug—e.g., warfarin with agents altering cardiac output, or hepatic blood flow will not appreciably change Ct_{ss} or Cf_{ss}.

Combined changes in α and $Cl_{int'}$ will also result in predictable changes. For example, a metabolic inhibitor of warfarin (i.e., decrease in $Cl_{int'}$) which also displaces warfarin from plasma proteins (i.e., increase in α) would be expected to increase Cf_{ss}. The net alteration of Ct_{ss} would depend upon the relative magnitude of changes of α and $Cl_{int'}$.

As long as Ct_{ss} is directly and linearly proportionate to Cf_{ss}, as is true at low drug to protein concentrations, measurement of α and total steady-state drug concentration may, in fact, be sufficient to appreciate the probable mechanism of an interaction and reason for change in drug effect—e.g., anticoagulant control. For example:

1. An increase in drug effect coupled with "normal" or depressed total drug concentration indicates inhibition of drug biotransformation and plasma protein displacement (Koch-Weser and Sellers 1976).

2. An increase in drug effect coupled with elevated total drug concentration indicates primarily metabolic inhibition.

3. A decrease in drug effect with a lower total drug concentration indicates enzyme induction with or without plasma protein displacement. A measurement of α would be necessary to clarify this.

4. An increase in drug effect and total drug concentration indicates metabolic inhibition.

These examples assume that the interaction is due to changes in disposition of the drug other than a change in the effective dose absorbed. Full investigation of drug interactions with drugs should always include studies of pharmacodynamic interaction—e.g., with anticoagulants this would include vitamin K disposition, alterations in turnover of the K-dependent clotting factors and effects on other components of the hemostatic processes.

Perfusion Limited ("High Extraction Drugs")

Theoretical model-specific calculations imply that drugs that are removed from the circulation by high capacity or affinity uptake mechanisms in kidney or liver will have decreased clearance when there is a decrease in total drug concentration caused by displacement (i.e., clearance depends on total drug concentration) (Wilkinson and Shand 1975). Thus, displacement of very high extraction ($\alpha > 0.9$) drugs could increase their total and free concentrations (Rowland, Benet, and Graham 1973). In practice, extraction ratios are probably not high enough to see this extreme. The failure of total concentration to fall to an extent equal to the increase in α may be a clue that such kinetic factors are present. Propranolol and other high extraction drugs and renal tubular secretion of highly bound penicillins could behave in this way. These theoretical possibilities have not been confirmed in vivo in humans, nor their biological importance established.

ACUTE DISPLACEMENT INTERACTIONS

In contrast to the considerations that apply to chronic dosing with displacers and low extraction drugs and "steady state" kinetics, the immediate consequences when a displacing drug is added to therapy can theoretically be the appearance of toxicity, or an otherwise altered response (Gillette and Pang 1977; Koch-Weser and Sellers 1976). Displacement of even a small amount of a highly plasma-bound drug causes a large relative increase in the free fraction of the drug in the plasma compartment (Figure 19-1). However, the displaced drug does not remain confined in the circulation but distributes throughout the body (Figure 19-1). Rapid displacement of one drug by another is analogous to injecting a pulse of free drug into the plasma compartment and having it distributed in the body. The rate

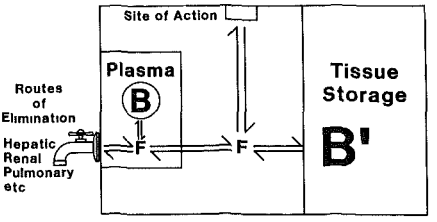

FIGURE 19-1
Protein drug displacement interactions. B = bound; F = free; total B + F.

of distribution of the "pulse" of drug will be determined primarily by tissue blood flow, the physicochemical properties of the drug, and the affinity and capacity of the sites of tissue localization. Tissue redistribution may be rapid; however, for highly tissue-localized or lipid soluble drugs complete distribution may take many hours. Because of its high capacity, tissue distribution is the single most important factor buffering wide fluctuations in free concentration.

After redistribution is complete, any increase in free drug concentration in serum and extracellular fluid depends on the apparent volume of distribution for the free drug. If the free drug distribution space is large, the increase in free drug concentration after distribution will be small and pharmacologically unimportant.

Other less important processes also act to buffer acute changes in free concentration after a drug's partial displacement from albumin (Figure 19-1). An increase in the concentration of unbound drug in the serum makes more drug available for glomerular filtration or hepatic biotransformation. For drugs restrictively eliminated by the liver, this displacement results in a greater elimination of the free drug (viz., first order and Michaelis-Mentin kinetics) which could be reflected in an initial shortening of the serum half-life of total drug. Since redistribution is occurring concurrently, early shortening of half-life should not be ascribed to increased biotransformation.

These processes continue until the free drug concentration in the serum has returned to almost the same concentration present before the displacement. The net post redistribution concentration will be: (amount of drug displaced - free drug volume of distribution). The amount displaced is: (α predisplacement - α postdisplacement) × plasma volume (3.5L) × total plasma concentration prior to displacement. At this new steady state, the total drug serum concentration is lower than before displacement, and the free drug serum concentration is a higher fraction of the total (Koch-Weser and Sellers 1976). Therefore, clearance of free drug will in most cases be virtu-

ally the same as before displacement (viz., dose/area under free drug concentration), but clearance calculated on the basis of total drug will be increased.

Finally, withdrawal of a displacing drug should theoretically cause an initial decrease in free concentration and pharmacologic action of the displaced drug. However, this has never been documented. Certain strict conditions would need to be met for this to be of biological importance (see next section).

PREDICTING IMPORTANT DISPLACEMENT INTERACTIONS

Table 19.1 summarizes features of drug protein interactions that could result in clinically important interactions. These are fairly stringent criteria and no putative protein binding drugs involved in drug interactions completely meet the criteria.

Displaced drug must be highly bound and have a small volume of free drug distribution (or redistribute slowly into its free volume of distribution). The drug's effects should be immediately detectable; hence, such drugs will usually have their pharmacologic action in tissues with high perfusion ratios. The displacing drug must achieve high drug to protein concentrations (e.g., > 1:10), to sufficiently modify the apparent association constant of the displaced drug (Koch-Weser and Sellers 1976). One practical implication of these criteria is that new, more selective and potent drugs (which will exert their effects at low concentrations) will have an even lower "risk" of clinically important drug displacement interactions.

TABLE 19.1
Necessary Conditions for Displacement Interactions to Be Important

Displaced Drug	Displacer
$\alpha < 0.05$	$\alpha < 0.05$
high correlation between concentration and response	competitive binding to same site on protein
rapid onset therapeutic or toxic effect	drug:protein concentration high
small volume of distribution	rapid increase in drug concentration
narrow therapeutic index	

Note:
α = free fraction.

Because the exact conditions are rarely met, acute displacement interactions between two drugs are difficult to detect. For example, if displacing drugs are absorbed or administered at rates only slightly faster than redistribution, the risk of serious interaction will be less. This risk is affected by the extent of cumulation in the body of the interacting drugs (which is, in turn, a function of their half-lives and dosage schedules), and by the sequence of their administration (Gillette and Pang 1977; Koch-Weser and Sellers 1976). If metabolites of one or both drugs also compete for binding sites, the situation becomes even more complicated. Since fatty acids and other drugs can decrease or increase drug binding, drug affinity can change continuously influenced by fatty acid changes due to meals, fasting, release of catechols (Romach et al. 1985).

Tissues represent a potentially major source of displaced drug since at steady-state the majority of drug in the body is outside the plasma compartment (e.g., diazepam 95 percent, warfarin 72 percent, phenylbutazone 66 percent). However, the importance of this is unclear since drugs "dissolved" in fat are not displaceable and "tissue binding" represents the sum of a multiplicity of discrete tissues each with a unique affinity and often high capacity for specific drugs. These factors suggest the tissue reservoir is rarely a source of displaced drug. The multiplicity of binding and localization and high capacity of sites for drug outside the plasma compartment suggest that the necessary conditions of selectivity and specificity for displacement from tissues may rarely be met. If displacement from tissues occurs, similar acute and chronic kinetic changes and buffering concentration changes will apply as described for displacement from plasma proteins.

One area which has not been studied in detail concerns the importance of interstitial albumin. Unfortunately, the importance of such albumin (30-60 percent of the total body albumin) for drug distribution is not known. Perhaps the interstitial space should be regarded as an extension of the plasma compartment, in which case the same general consideration outlined for the plasma compartment earlier applies.

INTERACTION OF WARFARIN AND PHENYLBUTAZONE

The administration of phenylbutazone to a patient already being treated with warfarin can lead to serious hemorrhagic complications. In 1956, Sigg et al. reported a decrease in the disappearance rate of ethylbiscoumacetate in one of their patients when phenylbutazone was given. They attributed this to delayed renal excretion of the coumarin dug. Weiner et al. (1965) reported that oxyphenbutazone slows

the disappearance of bishydroxycoumarin from plasma. On the basis of these results, Weiner (1964), Burns (1965), and Cucinell et al. (1965) suggested that enhancement of the anticoagulant effect might result from inhibition by the pyrazolone compounds of the coumarin biotransformation. Although he offered no experimental evidence, Brodie (1964) stated that phenylbutazone speeds up the metabolism of the coumarin anticoagulants. He also suggested that phenylbutazone might displace warfarin from plasma proteins, and therefore make more free warfarin available at its site of action in the liver (Brodie 1965). Since then, the interaction of the anti-inflammatory drug phenylbutazone and warfarin has become the most widely publicized and documented interaction of the oral anticoagulants (Table 19.2).

When concomitantly administered with warfarin, phenylbutazone causes a profound potentiation of hypoprothrombinaemic response, which can be life-threatening (Aggeler et al. 1967) (Table 19.2). The explanation commonly offered for the interaction is that phenylbutazone displaces warfarin from albumin, thereby increasing the unbound (and pharmacologically active) concentration of warfarin. If displacement were the basis of the interaction on theoretical grounds, the interaction should be transient (Koch-Weser and Sellers 1976). However, potentiation persists for as long as the drugs are co-administered, and other drugs which displace warfarin from albumin do not cause a similar potentiation of anticoagulation (Sellers and Koch-Weser 1970).

In fact, the situation is quite complex. The clinically available form of this drug is the racemate. In man, S(-)-warfarin is five times more potent, and is eliminated more rapidly than the R(+) isomer, and is more highly bound to plasma albumin than the R(+) warfarin enantiometer (Lewis et al. 1974; Sellers and Koch-Weser 1975). There also appear to be quantitative and qualitative differences in the metabolism of the two isomers (Hewick and McEwen 1973 and Table 19.3). When co-administered with phenylbutazone, the elimination half-life of S(-)-warfarin is increased four-fold (and clearance reduced), while the elimination half-life of the R(+)-warfarin is reduced (and clearance increased). This reduced elimination of the S(-)-warfarin explains, at least in part, the potentiating effect of phenylbutazone. Thus, phenylbutazone acts stereoselectively, inhibiting the metabolism of the S-isomer, but inducing the metabolism of the R-isomer (Lewis et al. 1974; O'Reilly et al. 1980). During the interaction, α increases 34 percent (1.09 percent to 1.46 percent) and total warfarin concentration is decreased by 50 percent (O'Reilly and Goulart 1981; Schary, Lewis, and Rowland 1975). These interpretations were based on measurements in plasma, following either the independent administration of warfarin isomers, or the administration of a stable labeled pseudoracemate and determining the

TABLE 19.2
Drug Interactions with Phenylbutazone and Warfarin

Warfarin Form	Warfarin Dose	Phenylbutazone Dose	Subjects No.	Subjects N	Observations	Reference
RS	S (1.5 mg/kg)	M (200 mg tid or bid) × 11–19 d	3 1	N P	RS $t_{\frac{1}{2}}$ ↓ 39%; RS conc ↓ 45%; PT ↑ 50%	Aggeler et al. 1967
RS	various	M (2–300 mg/d) × 5–7 d	5	P	Potentiation	Eisen 1965
RS	S (1.5 mg/kg)	M (200 mg/d) × 8 days	6	N	PT response ↑ 2×; RS total conc ↓ 30%; RS $t_{\frac{1}{2}}$ ↓ 46%	O'Reilly et al. 1968
RS	C (stabilized)	M (300 mg/d) × 1–2 wks	10	P	↑ mean prothrombin time (mean ↑ 38%)	Udall 1970
RS	S (91.5 mg/kg), control and day 4	M (200 mg, tid) × at least 8 d	6	N	↓ $t_{\frac{1}{2}}$ (mean 38%); pharmacologic receptor (69% ↓ in m in R_{syn} vs C_p)	O'Reilly and Levy 1970
RS	S (control and on day 10)	M (100 mg, tid) × 10 d)	4	N	↑ prothrombin time = clearance of racemic warfarin nor $t_{\frac{1}{2}}$ (half life); ↓ plasma 7-hydroxy-warfarin; ↑ plasma warfarin alcohol 2	Lewis et al. 1974
RS	M (10 mg/d) × 1 mth	M (100 mg, tid) × 10 d, beginning 12th d	1	N	↑ steady-state unbound warfarin (65%); ↓ steady-state total plasma warfarin (52%); ↓ plasma conc. and urinary excretion of 7-hydroxy-warfarin	Schary et al. 1975
RS	S (1.5 mg/kg)	M (300 mg/d)	8	N	↑ 34%	O'Reilly et al. 1981
S(–) R(+)	S (1.5 mg/kg) control and day 4	M (100 mg tid)	3	N	peak α ↑; S(–) free clearance ↓ four-fold; R+ clearance =	O'Reilly et al. 1980
S(–)	S (1.5 mg/kg) control and day 4	M (100 mg tid)	3	N	$t_{\frac{1}{2}}$ S(–) ↑ 84% $t_{\frac{1}{2}}$ R(+) ↓ 32%	Banfield et al. 1983

Notes:
The following key clarifies abbreviations: R(+) and S(–) warfarin represent the two racemates while RS warfarin is the racemic mixture; the anticoagulant or interaction agent may have been given as a single (S) dose, in a multiple dose (M) fashion; subjects may have been normal non-anticoagulated subjects (N), patients on anticoagulants (P).

The observations may have been an increase (↑), decrease (↓), or no change (=) in a parameter such as half-life ($t_{\frac{1}{2}}$), fraction of anticoagulant free (α), etc.; d = day; wk = week.

TABLE 19.3
Pharmacokinetics of Racemic Warfarin and Phenylbutazone

	Warfarin	Phenylbutazone
Absorption	F ≃ 1.0 T max 3-9h	F ≃ 0.8-1.0 T max 1.8-7.0h
Distribution	α = 0.01; S < R Vd = 8-27% TBW (12.3L) P: Vd area = 10.2L (30%)	α = 0.01 Vd free est = 1230L (in plasma) Oxyphenbutazone (O) α = 0.05
Mean Elimination Half Life (h)	R = 44 S = 30	72 oxyphenbutazone: 75 γ-hydroxy metabolite: 24
Plasma Concentrations (μg/ml)	1.0-4.0	80 oxyphenbutazone: 80
Biotransformation	to inactive hydroxylated forms and alcohol	to two active metabolites

Sources:
Madsen and Ellis 1981; O'Reilly et al. 1980; Kelly and O'Malley 1979; Levy et al. 1974; Wingard et al. 1978; Sioufi et al. 1980; Yasuda et al. 1982.

isomers by mass spectrometry. Finally, in rats phenylbutazone has a direct anticoagulant effect (Kelly and Bell 1981).

A review of the pharmacokinetic features of warfarin and phenylbutazone indicates that they meet only some of the criteria for a displacement interaction of potential importance (Table 19.3). Both are highly bound at the same site on albumin (Müller et al. 1984). However, free phenylbutazone and warfarin volumes of distribution are not small. Phenylbutazone is given in doses that result in drug protein ratios of approximately 1:3. Since oxyphenbutazone is highly bound it also will contribute to displacement. In this context, whether pyrazolones displace warfarin from tissue sites has not been studied extensively. Displacement of warfarin by phenylbutazone does occur in rats (G. Levy, personal communication). A decrease in Vd free would augment the importance of displacement.

An interesting and possibly frequent interaction which does meet most of the criteria for important interactions (Table 19.1) is

illustrated by recent studies of fluctuations in total and free fraction of diazepam concentrations. In these studies, young male volunteers were given diazepam 10 mg i.v. and multiple blood samples were taken over the next three days under controlled conditions. During the drug elimination phase, total diazepam and desmethyldiazepam levels fluctuated by 80 percent. Increases in free drug fraction accompanied each fall in total diazepam concentrations. These fluctuations are caused by the rapid increases and decreases in fatty acids due to meals and fasting which occur more rapidly than redistribution can be completed. Hence fluctuations in diazepam-free concentration occur and may be associated with fluctuations in acute clinical effect. Possibly such fluctuation in endogenous modulations of binding could account for the "unusual" increases and decrease in concentrations that are detected (Wong and Sellers 1979; Romach et al. 1985).

EXPERIMENTAL TACTICS

The experimental tactics required to demonstrate that the effect of a drug interaction is due to drug displacement from plasma proteins are clear.

In vitro or ex vivo studies can be used to demonstrate a displacement potential. Such studies must avoid all the usual pitfalls of low protein concentration, high drug concentration; high fatty acid content of protein, poor control of pH; inappropriate buffer. In human ex vivo studies of total concentration, free fraction and free concentration are needed. Kinetic calculations of clearance should be done using free drug concentrations and specific organ excretion. The rates of production of drug metabolite need to be determined. Interaction studies should include estimates of system bioavailability, tissue binding, and an assessment of changes in biotransformation. Increased drug effect should be shown to be correlated with free drug concentration.

In these respects the elucidation of the multifactorial, stereospecific warfarin-phenylbutazone interaction is instructive. The investigation of this interaction has extended over a 29-year period. Most drug interactions have not received so much attention. Most interaction studies were done some years ago without full awareness of the complexity of the problem. Inevitable deficiencies in the experimental design of these studies necessitate a reassessment of the clinical importance of all interactions ascribed to displacement.

In general, simple displacement interactions are clinically unimportant. However, the combination of decreased free drug clearance and plasma protein displacement will often result in a serious interaction.

REFERENCES

Aggeler, P. M., O'Reilly, R. A., Leong, L., and Kowitz, P. E. 1967. Potentiation of anticoagulant effect of warfarin by phenylbutazone. N Engl J Med 276: 496-501.

Banfield, C., O'Reilly, R. A., Chan, E., and Rowland, M. 1983. Phenylbutazone-warfarin interaction in man: Further stereochemical and metabolic considerations. Br J Clin Pharmacol 16: 669-675.

Brodie, B. B. 1965. Displacement of one drug by another from carrier or receptor sites. Proc Roy Soc Med 58: 946-955.

____. 1964. "Distribution and fate of drugs: Therapeutic implications." In: Absorption and Distribution of Drugs, pp. 199-251. Baltimore: Williams and Wilkins.

Burns, J. J. 1965. Discussion of panel discussion: Drug interaction in relation to acute poisoning. Proc Roy Soc Med 58: 995-998.

Cucinell, S. A., Conney, A. H., Sansur, M. S., and Burns, J. J. 1965. Drug interactions in man. I. Lowering effect of phenobarbital on plasma levels of bishydroxycoumarin (Dicumarol) and diphenylhydantoin (Dilantin). Clin Pharmacol Ther 6: 420-429.

Eisen, M. J. 1965. Combined effect of sodium warfarin and phenylbutazone. JAMA 189: 64-65.

Gillette, J. R., and Pang, K. S. 1977. Theoretic aspects of pharmacokinetic drug interactions. Clin Pharmacol Ther 22: 623-639.

Greenblatt, D. J., Sellers, E. M., Koch-Weser, J. 1982. Importance of protein binding for the interpretation of serum of plasma drug concentrations. J Clin Pharmacol 22: 259-263.

Hewick, S. D., and McEwen, J. 1973. Plasma half-lives, plasma metabolites and anticoagulant efficacies of the enantiomers of warfarin in man. J Pharm Pharmacol 25: 458-465.

Kelly, J. G., and O'Malley, K. 1979. Clinical pharmacokinetics of oral anticoagulants. Clin Pharmacokin 4: 1-15.

Kelly, L. J., and Bell, R. G. 1981. Mechanism for potentiation of warfarin by phenylbutazone. Biochem Pharmacol 30: 2443-2449.

Koch-Weser, J., and Sellers, E. M. 1976. Drug therapy: Binding of drugs to serum albumin. N Engl J Med 294: 311-316 (Part I); 526-531 (Part II).

____. 1971. Drug interactions with coumarin anticoagulants. N Engl J Med 285: 487-498 (Part I); 547-548 (Part II).

Levy, G., O'Reilly, R. A., and Wingard Jr., J. B. 1974. Relationship between the kinetics of the anticoagulant effects of racemic warfarin and its individual enantiomers in man. Res Comm Chem Path Pharmacol 1: 359-365.

Lewis, R. J., Trager, W. F., Chan, K. K., Breckenridge, A., Orme, M., Roland, M., and Schary, W. 1974. Warfarin: stereochemical aspects of its metabolism and the interactions with phenylbutazone. J Clin Invest 53: 1607-1617.

Madsen, B. W., and Ellis, G. M. 1981. Cooperative interaction of warfarin and phenylbutazone with human serum albumin. Biochem Pharmacol 30: 1169-1173.

Müller, W. E., Fehske, K. J., and Schläfer, S. A. C. 1985. Structure of binding sites on albumin. In: Drug-Protein Binding. New York: Praeger.

O'Reilly, R. A., and Aggeler, P. M. 1968. Phenylbutazone potentiation of anticoagulant effect: Fluorometric assay of warfarin. Proc Soc Exp Biol Med 128: 1080-1081.

O'Reilly, R. A., and Goulart, D. A. 1981. Comparative interaction of sulfinpyrazone and phenylbutazone with racemic warfarin. Alteration in vivo of free fraction of plasma warfarin. J Pharmacol Exp Ther 219: 691-694.

O'Reilly, R. A., and Levy, G. 1970. Pharmacokinetic analysis of potentiating effect of phenylbutazone on anticoagulant action of warfarin in man. J Pharm Sci 59: 1258-1261.

O'Reilly, R. A., Trager, W. F., Motley, C. H., and Howald, W. M. 1980. Stereoselective interaction of phenylbutazone with warfarin pseudoracemates in man. J Clin Invest 65: 746-753.

Romach, M. K., Sellers, E. M., Khouw, V., Naranjo, C. A. Interactions increasing drug protein binding. American Society for Clinical Pharmacology and Therapeutics, March 1985.

Rowland, M., Benet, L. Z., and Graham, G. G. 1973. Clearance concepts in pharmacokinetics. J Pharmaco Biopharm 1: 123-136.

Schary, W. L., Lewis, R., and Rowland, M. 1975. Warfarin-phenylbutazone interaction in man: A long term multiple dose study. Res Comm Chem Pathol Pharmacol 10: 663-672.

Sellers, E. M. 1979. Plasma protein displacement interactions are rarely of clinical significance. Pharmacol 18: 225-227.

Sellers, E. M., and Koch-Weser, J. 1975. Interaction of warfarin stereoisomers with human albumin. Pharmacol Res Comm 7: 331-336.

____. 1970. Potentiation of warfarin-induced hypoprothrombinemia by chloral hydrate. N Engl J Med 283: 827-831.

Sigg, A., Pestalozzi, H., Clauss, A., et al. 1956. Verstarkung der antikoagulantienwirkung durch butazolidin. Schweiz Med Wochenschr 86: 1194-1195.

Sioufi, A., Colussi, D., Caudal, F., Schoeller, J. P., and Massias, P. 1980. Pharmacokinetics of phenylbutazone in health subjects after oral administration of single and multiple doses. J Pharm Sci 69: 1413-1416.

Udall, J. A. 1970. Drug interference with warfarin therapy. Clin Med 77: 20-55.

Wardell, W. M. 1974. "Redistributional drug interactions. A critical examination of putative clinical examples." In: Morselli, Garattini, and Cohen, Drug Interactions, pp. 123-134. New York: Raven Press.

Weiner, M. 1964. The significance of the physiologic disposition of drugs in anticoagulant therapy. Seminars Hemat 1: 345-374.

Wilkinson, G. R., and Shand, D. G. 1975. A physiological approach to hepatic drug clearance. Clin Pharmacol Ther 18: 377-390.

Wingard Jr., J. B., O'Reilly, R. A., and Levy, G. 1978. Pharmacokinetics of warfarin enantiomers: A search for intrasubject correlations. Clin Pharmacol Ther 23: 212-217.

Wong, G. B., and Sellers, E. M. 1979. Intravascular factors affecting diazepam binding to human serum albumin. Biochem Pharmacol 28: 3265-3270.

Yasuda, Y., Shindo, T., Mitani, N., Ishida, N., Oono, F., and Kageyama, T. 1982. Comparison of the absorption, excretion and metabolism of suxibuzone and phenylbutazone in humans. J Pharm Sci 71: 565-572.