Pharmac. Ther. Vol. 48, pp. 71–94, 1990
Printed in Great Britain.

0163-7258/90 $0.00 + 0.50

Associate Editor: M. J. BRODIE

ENZYME INDUCTION AND INHIBITION

M. BARRY* and J. FEELY

Department of Pharmacology and Therapeutics, Trinity College Medical School, St James's Hospital, Dublin 8, Ireland

Abstract—The rate and extent of drug metabolism significantly influences drug effect. Enzyme induction by increasing the metabolism of drugs may result in important drug interactions. Other implications of enzyme induction include alterations in the metabolism of endogenous substrates, vitamins and activity of extrahepatic enzyme systems. Similarly a wide range of drugs may produce clinically significant drug interactions following enzyme inhibition. Assessment of enzyme induction and inhibition in man involves diverse methods including the use of model drugs.

CONTENTS

1. INTRODUCTION

The pharmacological effect of a drug results from its interaction with its receptor and is therefore critically dependent on the drug concentration available at the receptor site (Ariens, 1979). While several factors including absorption, distribution, protein binding and elimination may influence drug concentration at the receptor site, by and large the rate of drug elimination is a principal determinant for many drugs. Thus factors influencing drug metabolism may be expected to significantly alter drug effect.

*Present address: Department of Pharmacology and Therapeutics, University of Liverpool, New Medical Building, Ashton Street, P.O. Box 147, Liverpool L69 3BX, U.K.

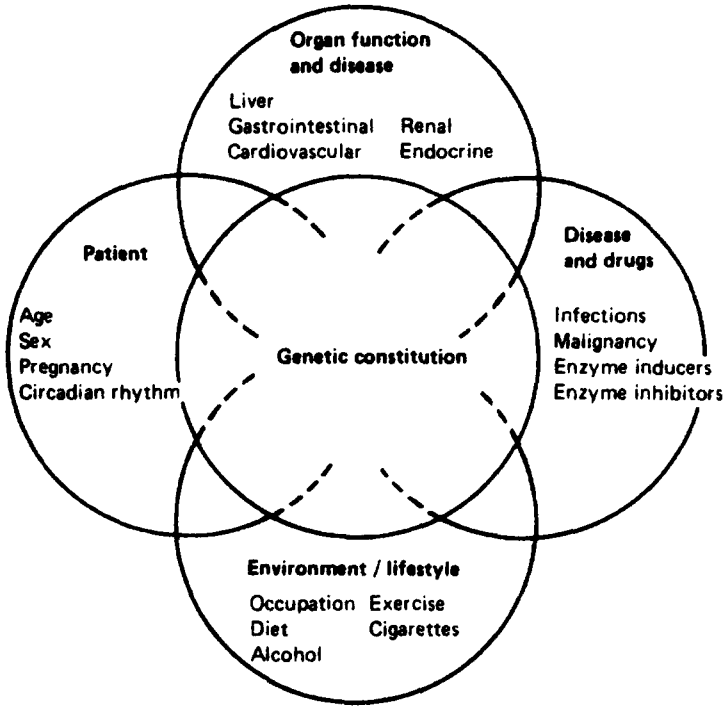

FIG. 1. Overlapping variables that influence rate and extent of drug metabolism in liver.

A large number of drugs, toxins and endogenous substances are lipid soluble and would be expected to accumulate in the body disrupting cellular activity if elimination did not take place. A fundamental role of drug metabolism is the conversion of these lipophilic compounds into more water soluble metabolites which are readily excreted thus limiting drug action and potential toxicity (Woolf and Jordan, 1987).

It is generally accepted that the liver is the principal site of drug metabolizing activity. However the contribution of extra hepatic sites, e.g. kidney, to drug metabolism is becoming increasingly recognized (Vainio and Hietanen, 1980). The enzymes concerned with drug metabolism are located primarily in the hepatic endoplasmic reticulum (Mannering, 1981). The conversion of lipophilic compounds to more polar metabolites proceeds via two metabolic conversions. The first conversion or phase one reactions include oxidation, reduction and hydroxylation reactions (Woolf and Jordan, 1987). The metabolic intermediates produced may then undergo phase two or conjugation reactions. This involves the conjugation of the metabolite with glucuronic, sulfuric and mercapturic acids and amino acids (Caldwell, 1982). The most influencial enzymes involved in the biotransformations are the hepatic microsomal P-450 mixed function oxidases. They catalyze phase one reactions with the insertion of molecular oxygen into drug molecules via the cytochrome P-450 enzyme–substrate complex. This enables transformations such as *N*, *S* and *O* dealkylation, aliphatic and aromatic hydroxylations to be carried out.

The enzymes catalyzing these reactions are versatile and nonspecific while the rate of activity may be under strong genetic influences. Numerous external and physiological factors may affect drug metabolizing enzyme systems including age, sex, dietary constituents, smoking and drugs (Fig. 1). The metabolizing activity of the enzymes may be increased following administration of certain xenobiotics. When this results from the selective increase in the concentration of enzyme the term enzyme induction is used to describe the process (Goldberg, 1980). Likewise the activity of the hepatic drug metabolizing enzymes may be decreased following drug administration. This usually involves a direct effect on the enzyme rather than a change in enzyme biosynthesis and is termed enzyme inhibition.

The consequences of enzyme induction and inhibition will be determined by the relative activity of the parent drug and the formed metabolite. Usually the metabolite will be less active than the original compound and therefore enzyme induction will result in a reduction in the pharmacological effect due to the increased drug metabolism. Enzyme inhibition would result in an increased concentration of parent drug at the receptor site and hence an increase in drug action. Also the metabolite formed during the biotransformation may be chemically reactive, e.g. cyclophosphamide to aldophosphamide, prednisone to prednisolone and hence enzyme induction may result in increased toxicity due to the increased production of the toxic intermediate (Bolt *et al.*, 1975).

One of the major factors leading to the realization of the significance of enzyme induction and inhibition in man is the common practice of polypharmacy. The resultant drug interactions observed have produced many relevant examples of the processes of induction and inhibition. In this clinical setting interactions involving drugs with a narrow therapeutic index, e.g. oral anticoagulants, antiepileptics etc. will be of greater importance as moderate changes in drug

metabolism could result in significant changes in pharmacological activity.

The effects of enzyme induction and enzyme inhibition are reversible on withdrawal of the inducing or inhibiting compound although the rate of onset and offset of these processes may differ (MacDonald and Robinson, 1968).

2. ENZYME INDUCTION, UNDERLYING MECHANISM

In recent years the mono-oxygenase system has been solubilized and various components purified. The system includes NADPH cytochrome P-450 reductase, phospholipid and a number of cytochrome P-450 isoenzymes (Goldstein, 1984) which are present in the uninduced state and possess overlapping specificities. Exposure to a variety of foreign compounds may preferentially increase the hepatic content of specific forms of cytochrome P-450. Therefore the process of enzyme induction involves the adaptive increase in the content of specific enzyme in response to the enzyme inducing agent. At least four distinct classes of enzyme inducing agents are known to exist (Adesnik *et al.*, 1981). They induce distinct subsets of P-450 isozymes, a fact that suggests the isozymes are under separate regulatory control. The classes of enzyme inducing agents include:

(i) phenobarbitone and a wide variety of structurally unrelated compounds;
(ii) polycyclic aromatic hydrocarbons such as 3-methylcholanthrene;
(iii) steroids including pregnenalone 16 α carbonitrile;
(iv) ethanol.

The hypolipidemic agent clofibrate may represent a fifth class of enzyme inducing agent. The proposed mechanism of enzyme induction involves the deactivation of the active repressor, a product of the regulatory gene component. The absence of the repressor at the operator gene facilitates the binding of the particular RNA polymerase to the promotor gene. This in turn facilitates the expression of the structural gene and the production of mRNA resulting in increased protein (enzyme) synthesis (Hardwick *et al.*, 1983).

Complementary DNA, RNA hybridization studies indicate that phenobarbitone induces nucleotide mRNA which codes for the translation of the phenobarbitone induced P-450 *in vitro*. An increase in mRNA by thirty-fold was observed with phenobarbitone increasing the transcription of the drug induced *P-450* gene (Wang *et al.*, 1983). Despite the fact that phenobarbitone appears to regulate cytochrome P-450 by increasing transcription of mRNA, for phenobarbitone induced cytochrome P-450s there is to date no evidence for interaction of the drug with a specific receptor. In contrast there is good evidence that 3-methylcholanthrene and other polycyclic aromatic hydrocarbons interact with a specific cytoplasmic receptor (Ah receptor) which may enter the nucleus. The drug receptor complex then interacts with the regulatory gene of the specific cytochrome P-450 isoenzyme resulting in increased synthesis of the isoenzyme (Jones *et al.*, 1986).

Proliferation of smooth endoplasmic reticulum may be regarded as a morphological expression of enzyme induction. An increase in the smooth endoplasmic reticulum content of animal and human hepatocytes has been observed following treatment with the enzyme inducing agent rifampicin (Jezequel *et al.*, 1971).

The number of compounds known to stimulate the activity of drug metabolizing enzymes is large and includes compounds of most pharmacologically active groups. The apparent nonspecificity of inducers suggests some common characteristics. In fact many enzyme inducing agents are lipid soluble at physiological pH and possess relatively long elimination half-lives. The ability to bind to cytochrome P-450 is another common characteristic. The ability of some water soluble barbiturate derivatives to stimulate drug metabolizing enzymes represents an exception to the general rule. The importance of achieving high hepatic concentrations of the inducing agent is also appreciated (Gelehrter, 1972).

The ability of enzyme incuding agents to increase the rate of phase one reactions is recognized. However administration of these agents may also increase the activity of phase two enzymes, such as glucuronyl transferase as demonstrated by the ability of oral contraceptive steroids to increase glycine and glucuronide conjugation pathways (Miners *et al.*, 1986).

The time course of induction will vary with different inducing agents. Increased transcription of P-450 mRNA has been detected in the rat nucleus as early as 1 hr after administration of phenobarbitone. The maximum induction with phenobarbitone may be apparent in animals after 48–72 hr (Eriksson, 1973). However induction following 3 methylcholanthrene may reach a maximum at 24 hr. Less potent inducers of hepatic drug metabolism may take a much longer time course of induction, e.g. antipyrine. The time course of induction is a reflection of the enzyme turnover for the particular inducing agent. In man a turnover rate ranging from 1 to 6 days was found resulting from the interaction between carbamazepine and clonazepam (Lai *et al.*, 1978). This may also reflect the considerable intersubject variation in enzyme induction. A number of studies suggest that subjects who are initially slow metabolizers of drugs have a greater potential for induction than subjects with initial high rates of metabolism (Vesell and Page, 1969). However it is now recognized that intersubject variance in steady state drug concentrations may be as wide following induction as before induction (Branch and Shand, 1979).

From the many experiments in animals and man it is apparent that enzyme induction is a dose-dependent phenomenon (Breckenridge *et al.*, 1973). Induction is easily evoked under appropriate conditions in animal experiments with numerous compounds in a high dose range. Because the high concentrations required for induction are rarely achieved without toxic side effects in man, pharmacological drug interactions from induction are only seen with a small number of compounds. These include the antiepileptic agents e.g. phenobarbital, phenytoin and carbamazepine and the antituberculous agent

TABLE 1. *Some Enzyme Inducers*

Barbiturates	Marijuana smoke
Carbamazepine	Meprobamate
Cigarette smoke	Phenytoin
Dichloralphenazone	Primidone (phenobarbitone)
Ethanol (chronic)	Rifampicin
Glutethimide	Sulfinpyrazone
Griseofulvin	

rifampicin (McInnes and Brodie, 1988). Given this limitation it is possible in man to compare drugs as enzyme inducers by the construction of dose–response curves. The effect of phenobarbitone on warfarin and antipyrine metabolism has been found to be dose dependent as is the effect of rifampicin on antipyrine and cortisol hydroxylation. In clinical practice most inducing agents administered in therapeutic doses will produce maximum effect within fourteen days. It is also recognized that many potent inducing agents may actually inhibit drug metabolism during the first few hours following administration prior to the onset of induction although this may not be detectable in man. Although numerous chemical and pharmacological compounds are capable of enzyme induction (Table 1), of greatest interest are those that produce clinically apparent drug interactions resulting from the induction process. Important examples of these drugs are discussed later. Numerous methods have been described for the detection of enzyme induction and inhibition. The methods are common to both and summarized here.

3. ASSESSMENT OF ENZYME INDUCTION AND INHIBITION IN MAN

The assessment of enzyme induction in man may be conducted by both *in vitro* and *in vivo* studies. Direct measurement of enzyme activities, e.g. 7 ethoxy phenacetin *O* de-ethylase from liver biopsy samples before and after administration of the inducing/inhibiting compound is an obvious method. However the sample may not be representative of the liver as a whole and there is not always a strong correlation with *in vivo* assessment. Furthermore ethical and practical considerations restrict the use of this method (Sesardic *et al.*, 1988). Therefore more indirect methods are required. Changes in the pharmacokinetics of a marker drug like antipyrine or changes in metabolite formation of the drug under study are commonly used to assess drug metabolizing activity (Vesell, 1979). In assessment of enzyme induction an alternative approach is to monitor changes in the disposition of an endogenous substance such as plasma glutamyl transpeptidase and bilirubin or urinary excretion of D-glucaric acid and 6 β hydroxycortisol (Hildebrandt *et al.*, 1975; Heirwegh and Blanckaert, 1981; Hunter *et al.*, 1974; Park, 1981). The advantage of this approach is the exclusion of possible effect of marker drugs on the metabolizing enzymes. It is the changes in the endogenous compounds rather than their absolute levels which are important. While the specificity and sensitivity of this approach may be limited in certain circumstances it may prove most informative.

3.1. MODEL DRUG SUBSTRATES

For a drug to be a useful substrate for assessing hepatic drug metabolizing enzyme activity, its clearance from plasma or the rate of appearance of a metabolite should be directly related to the activity of the enzyme(s) responsible for its metabolism. Other desirable properties required for marker drugs include simple, e.g. one or two compartmental, pharmocokinetics, with elimination entirely dependent on hepatic metabolism but metabolism independent of liver blood flow or plasma protein binding. All routes of metabolism and enzymes involved should be known. For drugs administered orally the absorption should be rapid and complete otherwise the substance is best given parenterally (Wilkinson and Shand, 1975).

3.2. ANTIPYRINE

Antipyrine has proved the most popular marker drug for assessing drug metabolism in man. It fulfils the above requirements for the ideal marker drug with almost complete hepatic metabolism, less than 10% plasma protein binding and less than 5% excreted unchanged in the urine. Its single compartment kinetics together with its relative ease of measurement in either plasma or saliva by gas liquid chromatography, radioimmunoassay or high performance liquid chromatography also render it a suitable marker drug (Stevenson, 1977). Vesell has shown a high degree of reproducibility of antipyrine disposition in individuals studied on up to 8 different occasions, enabling subjects to act as their own controls (Vesell, 1979). This is suited to the study of the effect of various environmental conditions on drug metabolism. Antipyrine clearance which relates the concentration of the drug to the rate of elimination should be used as it is a better indicator of drug elimination than antipyrine half life as assessed from the plasma drug concentration decay curve (Branch and Shand, 1979). Antipyrine clearance has been shown to increase and half life decrease following administration of a wide range of inducing agents including pesticides and drugs. Similarly enzyme inhibiting agents increase antipyrine elimination half life (Tables 2 and 3).

TABLE 2. *Substances which Decrease Antipyrine Half Life in Man*

Drug	Reference
Phenobarbitone	Vesell and Page, 1969
Organochlorine compounds	Kolmodin *et al.*, 1969
Ethanol	Vesell *et al.*, 1971b
Spironolactone	Huffman *et al.*, 1973
Tricyclic antidepressants	O'Malley *et al.*, 1973a
Chlorimipramine	O'Malley *et al.*, 1973b
Phenytoin	Petruch *et al.*, 1974
Quinine	Berlin *et al.*, 1975
Testosterone	Johnsen *et al.*, 1976
Rifampicin	Miguet *et al.*, 1977
Ascorbic acid	Houston, 1977
Glutethimide	Jackson *et al.*, 1978
Antipyrine	Ohnhaus and Park, 1979
Carbamazepine	Moreland *et al.*, 1981

TABLE 3. *Substances which Increase Antipyrine Half Life in Man*

Drug	Reference
Allopurinol	Vesell *et al.*, 1970
Levodopa	Vesell *et al.*, 1971a
Norethynodrel and Mestranol	Carter *et al.*, 1974
Disulfiram	Vesell *et al.*, 1975
Aminopyrine	Vesell *et al.*, 1976
Delta 9 tetrahydrocannabinol	Benowitz and Jones, 1977
Cimetidine	Serlin *et al.*, 1979
Propranolol	Bax *et al.*, 1981
Metoprolol	Bax *et al.*, 1983

While antipyrine clearance has been widely used to document environmental and drug induced change in hepatic drug metabolizing oxidative activity, changes in clearance are not usually predictable of the extent of alteration in the clearance of other drugs under the same influence. Another disadvantage of antipyrine as a model drug is its ability to act as a mild enzyme inducer; it therefore may stimulate its own metabolism and the metabolism of other drugs such as warfarin and diazepam (Bax *et al.*, 1981; Ohnhaus and Park, 1979; Breckenridge *et al.*, 1971). This effect is however only apparent after pretreatment for at least 7 days (Ohnhaus *et al.*, 1979b).

Measurement of the kinetics of antipyrine metabolites has also been used for assessing the activity of drug metabolizing enzymes. Several studies have indicated that the three major metabolites of antipyrine representing three separate enzymatic processes are produced by different forms of cytochrome P-450 in man (Danhof *et al.*, 1982). Drugs and disease states have been shown to exhibit either a nonselective or selective effect on antipyrine metabolism. Barbiturates, sulfinpyrazine and hyperthyroidism have been shown to nonselectively increase the rate of antipyrine oxidation (Saenger *et al.*, 1976). Selective induction by rifampicin results in increased formation of the norantipyrine metabolite. On the other hand phenytoin and carbamazepine selectively induce the formation of 4 hydroxy antipyrine and 3 hydroxymethyl antipyrine. Testicular cancer seemed to be associated with more pronounced induction of 3 hydroxymethyl antipyrine (Schellens and Breimer, 1987). The measurement of the activities of the separate metabolite pathways may be more reliable for correlation studies than the overall rate of metabolism. It has been found that the rate of formation of 4 hydroxy antipyrine was very strongly correlated with theophylline clearance (Teunissen *et al.*, 1985).

The lack of predictive value of antipyrine kinetics in estimating how the liver will handle other drugs has prompted the study of other potential marker drugs. Other drugs used as model substrates include diazepam, hexobarbitone, phenylbutazone, phenacetin, quinine, propranolol, tolbutamide and warfarin. The latter three have the added attraction of allowing one to follow concomitant changes in dynamics (heart rate, blood sugar, PTT) in addition to kinetics but all again have limited predictive value.

3.3. BREATH TEST ANALYSIS

Assessment of drug metabolism by breath analysis affords an alternative method of assessing possible enzyme inducers and inhibitors in man. The substrate most commonly used is aminopyrine although caffeine and diazepam have been employed also. The substrate is labeled with a ^{14}C which undergoes hepatic demethylation and the resultant radiolabeled carbon dioxide is excreted in the breath. In the aminopyrine breath test the percentage of administered radiolabeled ^{14}C aminopyrine excreted as $^{14}CO_2$ 2 hr after the oral dose (Hepner and Vesell, 1974) is used as the index of hepatic demethylation. Studies in animals demonstrate a correlation between exhaled $^{14}CO_2$ following administration of ^{14}C aminopyrine and the mass of hepatic microsomal mixed function monoxygenase system (Lauterburg and Bircher, 1976). The aminopyrine breath test has also been shown to be a useful test for enzyme induction in patients taking anticonvulsant drugs indicating that it is sensitive to changes in cytochrome P-450 (Hepner *et al.*, 1977). *N* demethylation is the major biotransformation for aminopyrine in man. In recent studies the aminopyrine breath test has been used to demonstrate the inhibitory effect of amiodarone and cotrimoxazole on hepatic drug metabolism (Barry *et al.*, 1986; Duenas-Laita *et al.*, 1986).

3.4. ENDOGENOUS SUBSTRATES

Noninvasive methods for the assessment of drug metabolizing capacity of the liver do not require the administration of a test drug such as antipyrine. Changes in the disposition of endogenous compounds or altered activity of a readily accessible enzyme may provide a useful noninvasive method of assessing enzyme induction in man. A variety of substances have been proposed.

3.4.1. *γ Glutamyltransferase*

This enzyme is found in many tissues such as kidney, liver, pancreas and testes (Orulowski, 1963). It catalyzes the transfer of γ glutamylpeptides (γ GT) from glutathione to other peptides or amino acids. Initially thought to be primarily located in the endoplasmic reticulum highest concentrations are found in plasma membranes of cells. The function of this enzyme is not related to drug metabolism. Nevertheless raised plasma concentrations have been found in patients receiving enzyme inducing agents such as anticonvulsants and hypnotics (Hildebrandt *et al.*, 1975; Table 4).

Following these observations the plasma level of the enzyme was proposed as an indicator of enzyme induction. However, drugs not regarded as enzyme inducers also resulted in increased γ GT levels, e.g. benzodiazepines and imipramine. Furthermore the recognized enzyme inducer rifampicin while increasing antipyrine clearance and urinary 6 β hydroxycortisol (other indices of enzyme induction) did not consistently result in significant increases in γ GT levels. Studies of phenobarbitone induction in rabbits showed an increase in γ GT concentrations in parallel with hepatic cytochrome P-450 content but only after

TABLE 4. *Substances which Increase Plasma γ GT Concentration in Man*

Drug	Reference
Anticonvulsants	Rosalki *et al.*, 1971
Ethanol	Rosalki and Rau, 1972
Nitrazepam	Whitfield *et al.*, 1973
Phenobarbitone	Hildebrandt *et al.*, 1975
Antipyrine	Ohnhaus and Park, 1979
Rifampicin	Back *et al.*, 1979
Imipramine	Tazi *et al.*, 1980b

eighteen days of treatment (Tazi *et al.*, 1980a). Parallel *in vitro* studies showed the longer phenobarbitone was administered the more readily γ GT was solubilized from hepatic plasma membranes. The plasma γ GT concentration is therefore dependent on at least two factors: (i) induction of hepatic γ GT and (ii) disturbances of hepatic plasma membranes. As these two functions are not necessarily associated the assessment of enzyme induction by plasma levels of γ GT must be regarded as unreliable. The effect of concomitant disease processes on γ GT levels must also be considered as raised levels are commonly found in patients with liver damage, high alcohol intake and pancreatitis (Kryszewski *et al.*, 1973).

3.4.2. *Urinary* D *Glucaric Acid*

D glucaric acid, first identified in mammalian urine in 1963, is an end product of carbohydrate metabolism via the glucuronic acid pathway which is a metabolic route, branching from glycogenolysis (Fig. 2) (Marsh, 1963).

Since the original report by Aarts that treatment with phenobarbitone or antipyrine caused an increase in urinary D glucaric acid, many workers have advocated the use of this parameter in assessing enzyme inducing agents (Aarts, 1965). Urinary D glucaric acid production reflects phenobarbitone type induction mainly, and by virtue of a parallel effect on enzyme activities of the glucaric acid pathway and the cytochrome P-450 system glucaric acid provides an indirect parameter of changes in cytochrome P-450 activity. Many drugs known to be enzyme inducers in man, stimulate D glucaric acid excretion in urine (Table 5).

Experimental data from animal studies found a significant correlation between hepatic cytochrome

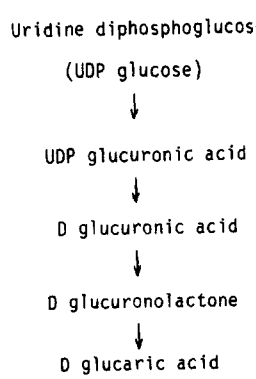

FIG. 2. Formation of D glucaric acid.

TABLE 5. *Some Substances which Increase Urinary Excretion of* D *Glucaric Acid in Man*

Drug	Reference
Aminopryrine	Aarts, 1965
Phenylbutazone	Aarts, 1965
Barbiturates	Hunter *et al.*, 1971a
DDT	Hunter *et al.*, 1971b
Spironolactone	Huffman *et al.*, 1973
Antipyrine	Davis *et al.*, 1974
Phenobarbitone	Lantham *et al.*, 1975
Carbamazepine	Lai *et al.*, 1978
Rifampicin	Ohnhaus and Park, 1979

P-450 content and urinary D glucaric acid following phenobarbitone treatment (Hunter *et al.*, 1973). Similarly in man phenobarbitone has been shown to increase D glucaric acid by seven-fold (Roots *et al.*, 1977). Increases in D-glucaric acid excretion have been shown to correlate with the clearance of the marker drug antipyrine though not with γ GT levels. No relation has been found between excretion of D glucaric acid and basal levels of metabolism. The dose-dependent effect of enzyme inducers is further suggested by the dose dependency in production of D glucaric acid with little or no change in the production following low dose phenobarbital (30 mg) and a seven-fold increase with higher doses of phenobarbitone (300–500 mg/day) (Roots *et al.*, 1977). However low dose phenobarbitone 30 mg does produce a significant increase in drug oxidation rates (Perucca *et al.*, 1981; Price *et al.*, 1986). Therefore induction of hepatic drug metabolizing enzymes may be dissociated from increases in D glucaric acid excretion. This lack of sensitivity renders D glucaric acid excreted less useful in detecting enzyme induction. The mechanism whereby enzyme induction results in increased D glucaric acid production is not entirely clear. It may possibly be due to a feedback mechanism involving NADPH production in the pentose phosphate pathway, NADPH being required for the reduction of cytochrome P-450. Alternatively D glucaric acid may be an index of phase two drug metabolism. As many mammals convert D glucuronic acid to ascorbic acid it is only in animals incapable of this conversion, e.g. primates and guinea pigs, that increases in D glucaric acid may be used as an index of increased enzyme activity. Despite the reservations D glucaric acid has proved a useful tool for the detection of enzyme induction.

3.4.3. *Urinary 6 β Hydroxycortisol*

6 β OH cortisol is a minor metabolite (2–6%) of cortisol formed primarily in the endoplasmic reticulum of hepatocytes by the mixed function oxidases and is excreted unconjugated in the urine (Frantz *et al.*, 1961). As cortisol hydroxylation is directly dependent upon the activity of the hepatic mixed function oxidase system the excretion of 6 β OH cortisol may be expected to reflect changes in hepatic drug metabolizing enzyme activity (Ohnhaus and Park, 1979). In fact changes in urinary 6 β OH cortisol excretion have been found to be a useful and sensitive index of microsomal enzyme induction in man (Park, 1981). A wide range of drugs and environmental chemicals

TABLE 6. *Substances Increasing the Urinary Excretion of 6β Hydroxycortisol in Man*

Drug	Reference
op DDD	Bledsoe *et al.*, 1964
Phenobarbitone	Burnstein and Klaiber, 1965
Phenylbutazone	Kuntzman *et al.*, 1966
DDT	Poland *et al.*, 1970
Pentobarbitone	Berman and Green, 1971
Spironolactone	Huffman *et al.*, 1973
Antipyrine	Davis *et al.*, 1974
Carbamazepine	Roots *et al.*, 1979
Rifampicin	Davis *et al.*, 1974

have been found to increase 6 β OH cortisol excretion (Table 6).

The extent of 6 β OH cortisol production depends on the daily cortisol formation whose variation may be compensated for by referring 6 β OH cortisol values to the excretion rates of 17 hydroxy corticosteroids. Hence many workers now use the 6 β OH cortisol/17 hydroxycorticosteroid ratio in the assessment of enzyme inducing agents. Recent studies have been greatly facilitated by the development of sensitive and specific radioimmunoassay and high performance liquid chromatography assays for 6 β OH cortisol (Roots *et al.*, 1979). The increase in 6 β OH cortisol excretion produced by enzyme induction has been shown to reflect an increase in 6 β OH cortisol production without a change in cortisol production (Burnstein *et al.*, 1967). A selectivity in the form(s) of cytochrome P-450 catalyzing cortisol 6 β hydroxylation is suggested from animal studies demonstrating cortisol 6 β hydroxylase activity, a function of cytochrome P-450 rather than cytochrome P-448 mixed function oxidase activity. This is supported by the observation that cigarette smoking does not influence 6 β OH cortisol excretion. Also poor metabolizers of spartine and debrisoquine in man have normal 6 β OH cortisol excretion rates (Park *et al.*, 1982).

Measurement of 6 β OH cortisol is particularly useful for monitoring the time course of enzyme induction. In comparative induction studies antipyrine clearance and 6 β OH cortisol, both direct measurements of hepatic mono-oxygenase activity, showed a significant correlation but no such correlation existed for plasma γ GT or D glucaric acid excretion (Ohnhaus and Park, 1979).

Unfortunately urinary 6 β OH cortisol cannot be used to predict interindividual differences in the 'basal' rate of drug metabolism in man as studies have demonstrated a lack of correlation between antipyrine clearance and 6 β OH cortisol excretion in healthy volunteers. This may be due to the many factors known to influence both 6 β OH cortisol production and antipyrine clearance including sex, thyroid function, liver disease, hypertension and adrenal status. It is also probable that the adrenal production of 6 β OH cortisol shows considerable interindividual variation. While measurements of 6 β OH cortisol excretion cannot be used to predict 'basal' drug metabolism rates in man they may be employed serially as a simple noninvasive method for detecting enzyme induction and as such are probably a better index of microsomal enzyme induction than either plasma γ GT or urinary D glucaric acid excretion (Park, 1981). Enzyme inhibitors including cimetidine and isoniazid may reduce 6 β hydroxy cortisol excretion but the changes are small and cannot be used to accurately monitor enzyme inhibition in man (Brodie *et al.*, 1981).

3.4.4. *Plasma Bilirubin*

Bilirubin is taken up by hepatocytes where it is conjugated with glucuronic acid and excreted into bile. The estimation of plasma bilirubin has been studied in relation to drug metabolism. Patients administered phenobarbitone, rifampicin and phenytoin were found to have significantly lower serum unconjugated bilirubin than control subjects. A significant correlation between plasma unconjugated bilirubin and antipyrine half life was found (Scott *et al.*, 1979). However plasma bilirubin has been considered an unreliable index of enzyme induction because plasma concentrations are affected by diet and numerous diseases and show diurnal variation (Hunter and Chasseaud, 1976).

Finally it must be stressed that no endogenous compound or probe drug can yet replace the measurement of the actual drug with which the patient is being treated. However, they do provide useful tools for better understanding the various aspects underlying enzyme induction and inhibition in man and may be useful in screening tests.

4. ENZYME INDUCERS

4.1. RIFAMPICIN

One of the most potent enzyme inducing agents known to man is the antituberculous agent rifampicin. Since its introduction to clinical practice there have been many reports of associated drug interactions largely as a consequence of its inducing properties. Rifampicin has been shown to increase the smooth endoplasmic reticulum of human and guinea-pig hepatocytes (Galehrter, 1972). The increase was noted after two days and reached a maximum by five days. The cytochrome P-450 content of human liver has also been shown to increase following rifampicin (Bolt *et al.*, 1975). Furthermore in common with other agents autoinduction is a property of rifampicin and the serum concentration of rifampicin falls on repeated administration to human volunteer subjects (Acocella *et al.*, 1978). The corresponding increase in the rate of formation of the major metabolite deacetylrifampicin supported the concept of induction as a cause of the fall in rifampicin levels. Rifampicin produces maximum changes in the activity of the hepatic drug metabolizing enzymes within 9–12 days (Park and Breckenridge, 1981). Many studies have demonstrated induction by rifampicin to be dose dependent (Ohnhaus *et al.*, 1987).

The induction of drug metabolizing enzymes following rifampicin treatment was of the order of 60–70% as assessed by the increase in antipyrine clearance in healthy human volunteers. Interestingly the induction of antipyrine clearance in patients with pulmonary tuberculosis following rifampicin was of the order of 30% (Teunissen *et al.*, 1984) possibly due

to concomitant therapy. The oxidative routes of antipyrine metabolism are differentially induced by rifampicin lending further support to the concept of multiple forms of cytochrome P-450, the different routes of antipyrine metabolism being catalyzed by different forms of cytochrome P-450 (Saenger *et al.*, 1976). In all studies the clearance of norantipyrine is increased to the greatest extent. Furthermore subjects with lower initial values of antipyrine clearance tended to be more inducible than those with higher clearance (Teunissen *et al.*, 1984). The observation that patients on long term anticoagulant treatment required an increase in their daily dose during concomitant rifampicin treatment was one of the first reported interactions of rifampicin. Further studies in healthy volunteers demonstrated a reduction in the prothrombin time and a corresponding decrease in the plasma half life of warfarin following rifampicin treatment (O'Reilly, 1974). Another clinically important interaction with rifampicin involves the concomitant administration of the oral contraceptive pill. Menstrual disturbances and unplanned pregnancies have been reported with this combination. The increased metabolism of both components of oral contraceptives i.e. ethynylestradiol and norethisterone is thought to be the underlying mechanism (Back *et al.*, 1979). The relative importance of the enhanced metabolism of estrogen and progestagen is unknown. The range of metabolic pathways induced by rifampicin is apparent from the wide range of exogenous and endogenous compounds whose metabolism is increased following rifampicin treatment (Table 7).

Rifapentine, the cyclopentyl derivative of rifampicin, is also capable of enzyme induction (Durand *et al.*, 1986).

4.2. PHENOBARBITONE

The enzyme inducing properties of phenobarbitone have been widely studied in both animals and man. In animal studies phenobarbital was found to increase the smooth endoplasmic reticulum of liver cells with resultant hypertrophy and increase in liver cell mass (Schulte-Hermann, 1974). In human studies administration of phenobarbitone resulted in increased smooth endoplasmic reticulum and cytochrome P-450 content. The activity of NADPH cytochrome P-450 reductase enzyme was also found to be increased. In addition an increase in liver size

TABLE 7. *Drug Interactions with Rifampicin*

Drug	Reference
Rifampicin	Acocella *et al.*, 1971
Warfarin	O'Reilly, 1974
Digitoxin	Peters *et al.*, 1974
Cortisol	Edwards *et al.*, 1974
Hexobarbitone	Zilly *et al.*, 1975
Dapsone	Gelber *et al.*, 1975
Ethynylestradiol	Bolt *et al.*, 1975
Norethisterone	Back *et al.*, 1979
Oral hypoglycemics	Kenny and Strates, 1981
Antipyrine	Bennett *et al.*, 1982
Metoprolol	Bennett *et al.*, 1982
Rifapentine	Durand *et al.*, 1986

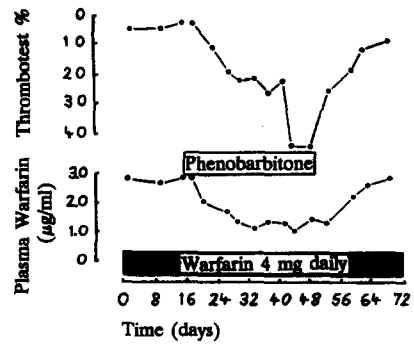

FIG. 3. Change in steady-state plasma warfarin concentration and thrombotest produced by phenobarbitone (120 mg/day for 30 days).

was seen. Unlike other enzyme inducing agents phenobarbitone was also found to increase liver blood flow (Pirttiaho *et al.*, 1982). The dose-dependent degree of enzyme induction in patients receiving phenobarbitone and other antiepileptic agents is well recognized (Perucca *et al.*, 1984).

Like rifampicin, phenobarbitone can stimulate a wide range of metabolic routes and one of the earlier reported interactions with phenobarbitone also involved oral anticoagulants. Breckenridge and Orme (1971) found that concurrent administration of phenobarbitone and warfarin resulted in changes in steady state plasma warfarin concentrations and anticoagulant effect after six days, the maximum effect recorded between fourteen and twenty one days (Fig. 3). Withdrawal of phenobarbitone in these patients may, in the absence of an appropriate reduction in warfarin dosage, result in fatal hemorrhage as drug metabolism reverts to normal resulting in higher warfarin concentrations (MacDonald and Robinson, 1968).

The hazards of polypharmacy in the treatment of epileptic patients is well recognized and this clinical setting provides another example of a drug interaction involving phenobarbitone, in this case with phenytoin whose metabolism is increased (Kutt *et al.*, 1969). Recent studies suggest that phenytoin inhibits phenobarbitone metabolism in epileptic patients (Kutt, 1984). However the interaction is not now thought to be of great clinical significance. Similar to rifampicin, pregnancies in women taking phenobarbitone with oral contraceptives have been described. Again the relative contribution of the induction of estrogen or progestogen components is unknown. Examples of drug interactions with phenobarbitone are shown in Table 8.

Enzyme induction by other barbiturates including amylobarbitone, barbitone, butobarbital, quinalbarbital and vinbarbital has been demonstrated (Anthintz *et al.*, 1968). A variation in the potency of induction of these agents is recognized and an inverse relationship between enzyme induction and plasma half life has been found.

4.3. PHENYTOIN

The anticonvulsant phenytoin has been found to induce the metabolism of other exogenous and

TABLE 8. *Drug Interactions with Phenobarbitone*

Drug	Reference
Phenytoin	Cucinell *et al.*, 1965
Bishydroxycoumarin	Cucinell *et al.*, 1965
Cortisol	Burnstein and Klaiber, 1965
Desmethylimipramine	Hammer and Sjoqvist, 1967
Testosterone	Southern *et al.*, 1969
Antipyrine	Vesell and Page, 1969
Chlorpromazine	Forrest *et al.*, 1970
Quinine	Saggers *et al.*, 1970
Diazepam	Viala *et al.*, 1971
Glyceryltrinitrate	Bogaert *et al.*, 1971
Warfarin	Breckenridge and Orme, 1971
Digitoxin	Soloman and Abrams, 1972
Oral contraceptives	Hempel *et al.*, 1973
Doxycycline	Neuvonen and Pentilla, 1974
Aminopyrine	Shaw *et al.*, 1985

endogenous compounds. Antipyrine clearance may be increased by up to 90% in some subjects following phenytoin dosing (Shaw *et al.*, 1985). Phenytoin has also been found to increase the hepatic cytochrome P-450 content in epileptic patients, the cytochrome P-450 content correlated linearly with antipyrine clearance (Pirttiaho *et al.*, 1978). Phenytoin appears to be a less potent inducer in man compared with phenobarbitone or rifampicin at doses used in clinical practice. Phenytoin blood levels remain stable for relatively long periods suggesting an absence of auto-induction (Kutt, 1971). Study of the metabolism of phenytoin is complicated by its dose related saturation kinetics.

Phenytoin induces the metabolism of exogenous compounds including anticoagulants, theophylline, digitoxin and carbamazepine (Hansen *et al.*, 1971; Marquis *et al.*, 1982; Patsalos *et al.*, 1988). The induction of endogenous compound metabolism including corticosteroids and thyroxine may also result from phenytoin treatment (Jubiz *et al.*, 1970; Larsen *et al.*, 1970).

4.4. CARBAMAZEPINE

Carbamazepine is another example of an anticonvulsant drug with enzyme inducing properties. The clearance of the model drug antipyrine was increased by 60% in healthy volunteers following carbamazepine treatment (Shaw *et al.*, 1985). Similarly in epileptic patients carbamazepine administration resulted in higher antipyrine clearance and increased urinary excretion of D glucaric acid. The enzyme induction is dose dependent and the relative potency for antipyrine clearance was 0.84 compared with phenobarbitone (Perucca *et al.*, 1984). It has been demonstrated that carbamazepine induces its own metabolism during maintenance therapy and concomitant treatment with phenobarbitone or phenytoin further induces its metabolism (Eichelbaum *et al.*, 1985). In contrast carbamazepine inhibits the metabolism of concomitantly administered phenytoin (Patsalos *et al.*, 1988). Further evidence for the enzyme inducing properties of carbamazepine is derived from studies with warfarin, doxycycline and clonazepam whose half lives were all decreased following treatment (Hansen *et al.*, 1971; Penttila *et al.*, 1974; Lai *et al.*, 1978).

Other drugs with enzyme inducing properties include the hypnotic glutethimide and the anti-inflammatory agent phenylbutazone (Jackson *et al.*, 1978; Lewis *et al.*, 1974).

Some evidence of enzyme inducing properties for meprobamate, spironolactone and diazepam have been produced but the clinical significance of these is undetermined.

4.5. CIGARETTE SMOKING

Cigarette smoking increases the metabolism of a number of drugs and chemicals in man (Jusko, 1978). The lower plasma concentrations of phenacetin in smokers was the first indication of the inductive properties of smoking in man (Pantuck *et al.*, 1974). Hepatic drug metabolism as assessed by antipyrine clearance may be increased by 30% in heavy smokers (Vestal *et al.*, 1975). Polycyclic aromatic hydrocarbons (PAH) are a prominent component of cigarette smoke and are known inducers of hepatic xenobiotic metabolism in experimental animals. It is thought that these compounds are responsible for the enzyme induction resulting from smoking in man. However differences in aryl hydrocarbon hydroxylase activity, which is induced by PAH, between smokers and nonsmokers are nonexistent (Boobis *et al.*, 1980). However the high affinity components of the *O* deethylation of both phenacetin and 7 ethoxyresorufin are two to three times greater in liver samples from smokers than in those from nonsmokers (Kahn *et al.*, 1985; Pelkonen *et al.*, 1986). Recent studies suggest that the form of cytochrome P-450 catalyzing the *O* deethylation of phenacetin in man is orthologous to rat form d, and that this is inducible by hydrocarbons in cigarette smoke (Sesardic *et al.*, 1988).

Studies of *in vivo* drug biotransformations have demonstrated increased metabolism of antipyrine, imipramine, nicotine, pentazocine and theophylline. The metabolism of diazepam, nortryptilline, pethidine, phenytoin and warfarin was unchanged (Park and Breckenridge, 1981).

Smoking habit of individuals must be considered when studying drug metabolism in humans. It is known that plasma propranolol steady state concentrations are lower in smokers than in nonsmokers and in younger compared to elderly patients (Feely *et al.*, 1981a). The latter effect may however be due to an age related change in drug metabolism influenced by smoking, the effect being greatest in the younger age group.

Smoking may indeed influence therapeutic intervention as demonstrated by the recent U.K. MRC trial on the effect of treatment in mild hypertension. Reduction in blood pressure following treatment with propranolol was less marked in patients who smoked cigarettes (MRC Working Party, 1985).

4.6. ETHANOL

Chronic alcoholism is a major cause of liver disease leading to abnormal drug metabolism. However, ethanol itself and independently of liver disease also influences the biotransformation of other drugs. The nature of this effect is largely determined by the

duration of ethanol exposure. Generally, acute ethanol administration depresses the rate of drug metabolism (Rubin *et al.*, 1970). This inhibitory effect on hepatic drug metabolizing enzyme activity may involve several possible mechanisms. Ethanol normally metabolized by the cytoplasmic alcohol dehydrogenase may also be metabolized by an inducible microsomal enzyme. Ethanol may compete for the same microsomal system required to metabolize other drugs. Ethanol may also inhibit metabolism by inhibiting the activity of NADPH cytochrome P-450 reductase, the rate limiting step in the oxidative biotransformation of drugs. The disturbance of the lipid bilayer membrane may also interfere with metabolizing enzymes.

Acute ethanol intake has been shown to result in higher plasma levels of many benzodiazepines including diazepam, chlordiazepoxide and clobazam. Acute ethanol administration prolonged the elimination half life of meprobamate, phenobarbitone, tolbutamide and antipyrine (Rubin *et al.*, 1970).

All the above studies focused on phase one biotransformations. However acute ethanol intake may also inhibit phase two reactions. The glucuronidation of disulfiram may be decreased and the glucuronidation of *p*-nitrophenol by microsomes is impaired by ethanol (Rubin *et al.*, 1970).

Whereas acute ethanol intake depresses metabolism, chronic ethanol intake enhances the rate of drug metabolism in man. Chronic ethanol intake has been associated with increased binding of ethanol to cytochrome P-450. The repeated exposure produces an increase in microsomal mass and a greater activity of the monooxygenase system, thus enhancing the elimination of drugs (Videla *et al.*, 1973).

Chronic intake has been found to decrease the elimination half life of meprobamate and phenobarbitone. The elimination of tolbutamide was increased by 50% in chronic alcoholic patients (Kater *et al.*, 1969). Other drugs whose elimination is increased during chronic alcohol intake include phenytoin and antipyrine. The problems associated with anticoagulation in chronic alcoholics may be due in part to alterations in the elimination of warfarin, usually increased in these patients, hence the difficulty in maintaining constant levels of warfarin.

Phase two reactions may also be affected by chronic ethanol intake with increased levels of glucuronyltransferase. As is seen with other inducers of drug metabolism, e.g. phenobarbitone, withdrawal of the inducing agent may in itself precipitate a drug interaction. In patients with alcoholic withdrawal the elimination of chlormethiazole also decreased.

Aside from the direct influence of ethanol on drug metabolism an indirect mechanism may possibly come into play involving ethanol induced release of corticosterone which may secondarily influence the rate of drug metabolism.

4.7. DIET AND NUTRITION

Concomitant food intake may influence the bioavailability of medications. The bioavailability of the antihypertensive agents propranolol, metoprolol and hydrochlorothiazide may be increased (Melander, 1978). In the case of propranolol a transient inhibition of presystemic primary conjugation may be the underlying mechanism (Liedholm and Melander, 1986). Food may also influence the bioavailability of drugs by altering absorption (Melander, 1978).

The degradation of drugs by phase one and two reactions is catalyzed by microsomal enzymes and the formation of metabolites requires the participation of a wide range of substances provided by nutrition. The ability of the nutritional state to influence drug metabolism and the activity of hepatic monooxygenase enzymes is increasingly appreciated (Anderson, 1988). A low protein diet in animals produces diminished activities of NADPH dependent enzymes (Campbell and Hayes, 1974). In children with protein-calorie malnutrition the elimination of chloramphenicol, antipyrine and sulfadiazine is impaired (Eriksson *et al.*, 1984). In adults a high protein low carbohydrate diet enhances the metabolism of antipyrine, theophylline and phenacetin (Kappas *et al.*, 1978b). A diet rich in carbohydrates causes a decrease in microsomal mixed function oxidase activities. The third macronutrient, fat, does not seem to affect the metabolism of drugs, e.g. theophylline or antipyrine (Anderson *et al.*, 1979). Preparation of food may also play a role in drug metabolism. The ingestion of charcoal broiled beef for four days was found to significantly decrease the plasma levels of phenacetin in healthy volunteers (Kappas *et al.*, 1978a). Whether the process of enzyme induction would fully explain the observations outlined remains to be seen. However animal studies suggest that dietary protein augments hepatic microsomal cytochrome P-450 content, liver weight and mitotic indices (Campbell and Hayes, 1974).

Since a number of endogenous substrates are metabolized by hepatic monooxygenases it is not surprising that induced alterations of enzyme activity will alter the metabolism of these substrates. Such endogenous substrates include bilirubin, vitamins, hormones and lipids. The alteration in metabolism of these substrates results in a variety of implications of enzyme induction.

5. IMPLICATIONS OF ENZYME INDUCTION

The ability of enzyme inducers to stimulate phase two biotransformation has provided a therapeutic application of phenobarbitone in conditions such as Crigler–Najjar syndrome type II and Gilbert's syndrome (Black and Sherlock, 1970). In these patients with unconjugated hyperbilirubinemia there is a reduction in the activity of glucuronyl transferase enzyme. The resultant jaundice is ameliorated by treatment with phenobarbitone which increases the clearance of bilirubin by increasing the activity of glucuronyl transferase enzyme. Phenobarbitone may, however, produce a number of additional changes such as increased liver blood flow, increased transport of bilirubin into cells and an increase in bile flow. The relative contribution of enzyme induction is not defined (Black *et al.*, 1974).

It is well established that long term treatment of epilepsy with known enzyme inducers, e.g. phenobarbitone and phenytoin, may lead to abnormalities

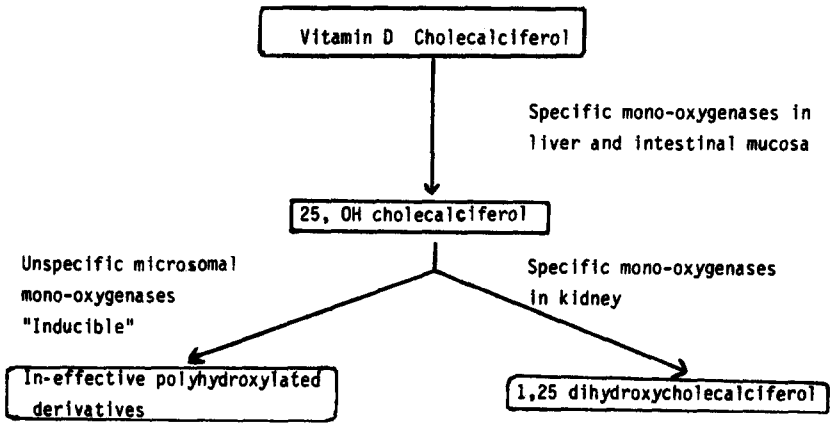

FIG. 4. Vitamin D metabolism, the effect of inducing agents.

in plasma calcium metabolism and resultant osteomalacia (Cinti *et al.*, 1978). The interference by enzyme inducers on Vitamin D metabolism is thought to be the underlying mechanism. Vitamin D, essential for normal calcium metabolism, is hydroxylated by hepatic microsomal enzymes to 25 hydroxycholecalciferol. This in turn is converted by the kidney to 1.25 dihydroxycholecalciferol, the active form of the vitamin. With enzyme induction there is a diversion in the hepatic metabolism of vitamin D to bioligically inactive metabolites (Hahn *et al.*, 1972) (Fig. 4). Since the level of 25 hydroxycholecalciferol regulates the formation of the active Vitamin D an increased breakdown can easily be compensated by an enhanced conversion rate. Therefore a sufficient intake of Vitamin D can prevent any deficiency. This may explain why Vitamin D deficiency develops in only a small percentage of patients receiving antiepileptics. The ability of enzyme inducing agents to affect Vitamin D metabolism has been used in the clinical setting of Vitamin D poisoning, where agents like phenobarbitone and glutethimide have been used to decrease Vitamin D concentrations (Lukaskiewicz *et al.*, 1987; Iqbal and Taylor, 1982).

A complication of long term anticonvulsant treatment is anemia thought to be due to folic acid deficiency. The underlying mechanism is unclear but impairment of folate absorption with associated inhibition of intestinal conjugation leading to polyglutamate malabsorption has been suggested (Hoffbrand and Necheles, 1968). Increased requirement of folate as a consequence of enzyme induction has also been suggested as an underlying cause (Labadarios *et al.*, 1978).

Increased cortisol metabolism occurs following treatment with phenobarbitone, phenytoin and other enzyme inducers (Werk *et al.*, 1964). An increased excretion of 6 β hydroxycortisol in urine has been found. Thus, results of dexamethasone suppression tests in patients receiving enzyme inducers may be unreliable (Werk *et al.*, 1969). The underlying mechanism is thought to be enhanced hepatic conjugation and biliary excretion of the steroid. A significant interaction between enzyme inducing agents and corticosteroids has been suggested in the following clinical settings: increased corticosteroid dosage requirements in the treatment of Addisons disease (Maisey *et al.*, 1974), the nonresponsiveness in the nephrotic syndrome (Hendrikse *et al.*, 1979) and the requirement of increased corticosteroid doses in the treatment of tuberculous pericarditis in patients on concurrent rifampicin therapy (Van Marle *et al.*, 1979).

Enzyme induction is known to interfere with peripheral thyroid metabolism. The decrease in protein bound plasma iodine in animals and man following phenytoin treatment is well recognized (Mendoze *et al.*, 1966). Further studies with chronic phenytoin treatment demonstrated an increased turnover of thyroxine (T_4) in rats (Oppenheimer *et al.*, 1968). A 60% increase in the peripheral conversion of T_4 to the more active form of thyroid hormone, triiodothyramine (T_3) following diphenylhydantoin administration in humans has been found. A decrease in thyroxine and the biologically inactive reverse T_3 of approximately 14% together with a 25% increase in T_3 has been found following administration of the enzyme inducer rifampicin (Ohnhaus *et al.*, 1981). The clinical relevance of these findings with respect to overall thyroid function remains to be established.

Experimental studies indicate a link between the hepatic microsomal carbohydrate and drug metabolizing systems (Stengard *et al.*, 1984). Phenobarbitone has been found to enhance insulin mediated glucose metabolism in healthy nondiabetic subjects (Lahtela *et al.*, 1984). Furthermore, addition of hepatic microsomal enzyme inducing compounds to the therapy of NIDDM patients improves the glycemic control (Sotaniemi *et al.*, 1983).

Glucose 6 phosphatase, a multicomponent enzyme system, and glycogen synthetase, are both microsomal glucose metabolizing enzymes (Wordlie, 1979). Therefore drugs having an effect on the mixed function oxidase system may nonspecifically influence other enzymes located in the endoplasmic reticulum of hepatocytes. Utilizing the euglycemic clamp technique Lahtela *et al.* studied the effect of phenobarbitone on insulin mediated glucose metabolism in noninsulin dependent diabetics on sulfonylurea treatment. Addition of phenobarbitone to the sulfonylurea therapy resulted in a reduction in fasting glucose and immunoreactive insulin levels. A significant increase in glucose disposal rate and metabolic clearance rate of glucose resulted. The authors

demonstrated a corresponding increased activity of the mixed function oxidase system as antipyrine clearance and insulin mediated glucose metabolism both enhanced by 30% following phenobarbital treatment (Lahtela *et al.*, 1985). The activation of postreceptor events in hepatocytes through microsomal enzyme inducing agents may therefore offer a new approach to the improvement of insulin sensitivity in noninsulin-dependent diabetes mellitus. Conversely, inhibitors of the mixed function oxidase system, e.g. cimetidine, may decrease the metabolic clearance rate of glucose and therefore impair glucose tolerance (Lahtela *et al.*, 1986).

The beneficial effects of enzyme inducing compounds may also extend to lipid metabolism. Drugs such as phenobarbitone, phenytoin, carbamazepine and alcohol, which induce the microsomal enzyme system, may also influence serum lipid and apolipoprotein concentrations. They result in increased concentrations of high density lipoproteins (HDL) which are cardioprotective and a reduction in low density lipoproteins (LDL) which are atherogenic with a resultant increase in the HDL:LDL ratio (Luoma, 1987).

Serum HDL concentrations have been shown to be directly proportional to cytochrome P-450 content of human liver (Luoma *et al.*, 1985) and also directly related to plasma antipyrine clearance (Luoma *et al.*, 1984). Therefore enzyme inducing drugs by lowering the concentration of atherogenic lipids may favorably influence coronary heart disease. Indeed mortality from ischemic heart disease is approx 30% lower in epileptic subjects treated with anticonvulsants when compared with age and sex matched members of the general population (Muuronen *et al.*, 1985). Not all enzyme inducing drugs influence lipid metabolism as rifampicin and antipyrine failed to affect serum lipoprotein profile (Ohnhaus *et al.*, 1979a).

The administration of enzyme inducing drugs may also be associated with adverse effects and they may exacerbate certain pathological states, e.g. acute intermittent porphyria. This disorder is inherited as autosomal dominant and is characterized by excessive urinary excretion of 5 aminolevulinic acid and porphobilinogen. The patients develop disorders of the nervous system such as psychosis and abdominal pain with tachycardia and raised blood pressure. The underlying mechanism is thought to be an increase in the activity of hepatic 5 aminolevulinic acid synthetase, the rate limiting enzyme in the biosynthetic chain. Acute attacks may be precipitated by enzyme inducing drugs like phenobarbitone (Cohn and Roth, 1983).

The increased activity of the microsomal monooxygenase system following enzyme inducing agents may have toxicological implications. It is becoming increasingly recognized that the formation of chemically reactive metabolites may produce a range of toxic effects. The reactive intermediate may react covalently with cellular macromolecules disrupting cellular activity. The toxicity produced will depend on a number of factors, e.g. the proportion of drug converted to the reactive intermediate, the proportion of metabolites bound covelently, the proportion of reactive metabolite binding to certain macromolecules within the cell and the resultant irreversible damage produced. The production of reactive metabolite may be offset by enzyme activity deactivating the metabolite. The situation may be exemplified by the hepatotoxicity of the reactive intermediate to paracetamol with the deactivating reduced glutathione. A reduction of the reduced glutathione results in unopposed toxic intermediate with resultant hepatotoxicity (Mitchell *et al.*, 1974). Animal studies have demonstrated the potential toxicity of inducing agents in the enhancement of toxic intermediate production (Garner and McLean, 1969) and have also demonstrated that inhibition of oxidative metabolism modifies the toxicity of compounds like carbon tetrachloride, isoniazid and paracetamol that require activation by hepatic microsomal enzymes before producing cellular toxicity (Homan *et al.*, 1985; Lauterburg *et al.*, 1983; Mitchell *et al.*, 1981). Further studies are required to clarify the situation in humans (Lauterburg *et al.*, 1983).

A number of phase one biotransformations are thought to increase the carcinogenicity of certain compounds. Metabolic products from aromatic carbon oxidation with the resultant production of epoxide intermediates may play a role in the carcinogenicity of polycyclic aromatic hydrocarbons, e.g. benzopyrine. Hydroxylation of amines adjacent to aromatic ring structures is well described and the resultant *N* oxidation products correlate with the appearance of bladder carcinoma (Vaught and King, 1984). Enzyme inducing agents through the promotion of intermediate formation may enhance the carcinogenicity of many compounds.

6. INDUCTION OF EXTRAHEPATIC DRUG METABOLIZING ENZYMES

There exists a marked variability in the ability of enzyme inducers to stimulate metabolism in various extrahepatic organs, this ability varying further with the different species (Jenner and Testa, 1976). The failure of inducers to increase oxidative drug metabolism in the rat extrahepatic tissues including kidney, brain, intestines and adrenals has been demonstrated. Phenobarbitone was shown to be without effect on aminopyrine demethylase, aniline hydroxylase, coumarin 3-hydroxylase and hexobarbital oxidase in these tissues. However, administration of inducing agents to the rabbit resulted in increased cytochrome P-450 levels and drug metabolizing ability at extrahepatic sites, e.g. kidney. The rate of response of extrahepatic tissues to inducing agents has been found to vary markedly suggesting qualitative as well as quantitative differences between the monooxygenases in various tissues. Recent animal studies suggest that extrahepatic metabolism may afford a protective mechanism as increases in cytochrome P-450 and NADPH cytochrome C reductase activities have been found in extrahepatic sites, e.g. kidney in animals, with hepatic dysfunction secondary to pharmacological and surgical interventions (Barry *et al.*, 1987). Induction of enzyme activities in tissues such as gut, skin and lymphocytes have been demonstrated in man. Induction of extrahepatic metabolism is a fruitful area for future research.

7. ENZYME INHIBITION, UNDERLYING MECHANISM

Inhibition of the metabolism of drugs which are biotransformed in the liver often leads to serious adverse effects because of accumulation of drugs to toxic concentrations. Interactions involving inhibitors of drug metabolism are probably of greater clinical significance than those involving enzyme induction (Kato *et al.*, 1964). The onset of inhibition is usually rapid following a single administration of the inhibitory compound. The increased plasma level of a drug following inhibition of its metabolism reaches a new steady state often approximately five half lives later. Therefore potentiation of the pharmacological effect usually occurs quickly if the drug has a short half life (e.g. tolbutamide $t_{1/2} = 4$–10 hr).

The prediction of drug interactions following enzyme inhibition is difficult as many different xenobiotics may inhibit metabolism. However, the most clinically relevant interactions will involve drugs with a narrow therapeutic ratio (e.g. warfarin, phenytoin, theophylline) (McInnes and Brodie, 1988). Drug metabolizing enzyme systems may be inhibited in numerous ways either indirectly by alterations of physiological parameters including hormonal levels and nutritional status and by inhibitors of the synthesis of microsomal components, or more commonly by directly arising from interaction at the monooxygenase site. Enzyme destruction may be implicated with some drugs (Muakkassah and Yang, 1981).

That the hepatic monooxygenases exhibit low substrate specificity is well recognized and on this basis substrate competition for the active sites may be expected to explain the mechanism of enzyme inhibition. This has been demonstrated for a large number of drugs, e.g. hexobarbitone, chlorpromazine, phenylbutazone and acetanilide which are competitive inhibitors of *N*-demethylation of ethylmorphine (Rubin *et al.*, 1964). Competition for the same substrate binding site is probably the most prevalent mechanism of inhibition of drug metabolism in man. However, not all substrates have the ability to inhibit the metabolism of an alternate substrate. The agent SKF 525 A while exhibiting marked inhibitory action on the metabolism of a large number of substrates fails to influence the *N* dealkylation of *N* methylamine or the sulfoxidation of chlorpromazine (Gillette and Kamm, 1960). The mechanism of alternate substrate inhibition requires that the inhibition must be competitive and the inhibitor must also serve as substrate. The efficacy of the inhibitor will depend on the magnitude of the inhibitory constant (K_I) of the inhibitor relative to the Michaelis constant (K_m) of the drug whose metabolism is inhibited. The lower the K_I value the higher is the inhibitory activity. The inhibitor should not have a V_{max} greatly exceeding that of the substrate if its presence at the metabolic site is to be maintained (Anders and Mannering, 1966). The inhibitory capacity increases as V_{max} for metabolism of the inhibitor decreases. That the inhibitor should be metabolized itself is not a prerequisite, as exemplified by the potent inhibitor 2.4 dichloro 6 phenyl-phenoxyethylamine (D.P.E.A.) which is a relatively poor substrate for the monooxygenase system. While competitive inhibition may be the most important mechanism in relation to xenobiotic metabolism it is not the only one as many cases of noncompetitive and mixed kinetics have been reported. Inhibition of hexobarbitone metabolism by chloramphenicol or the inhibition of diphenylhydantoin metabolism by isoniazid have been shown to be noncompetitive (Kutt *et al.*, 1968). The type of inhibition obtained appears to be influenced by such factors as the nature of the substrate, the concentration of the inhibitor, more than one enzyme system metabolizing the substrate and metabolism of the inhibitor during the course of the reaction (Sasame and Gillette, 1970). The demonstration of an interaction between the inhibitor and a component of the monooxygenase system supports the mechanism of an alternate substrate mechanism. Cytochrome P-450 is capable of combining with exogenous compounds to produce difference spectra of two basic types, i.e. type I and type II. The results of spectral studies provide further information with regard to the interaction of the inhibitor with the monooxygenase system. The production of a type II binding spectrum by cimetidine supports the suggestion that cimetidine interacts with the heme iron of cytochrome P-450 both with its imidazole and cyano coordinating groups (Reilly *et al.*, 1983). The interaction of inhibitors with cytochrome P-450 reductase is further evidence of the interaction of inhibitors with a component of the drug metabolizing enzyme system (Sasame and Gillette, 1970).

The route(s) of drug metabolism is an important parameter when looking at the clinical consequence of inhibition of drug metabolism. If a drug is metabolized by a single route only then inhibition of this route may be expected to produce a clinically significant interaction, e.g. tolbutamide (Thomas and Ikeda, 1966). However, most drugs are metabolized by more than one route and inhibition of a single pathway would be less likely to produce a significant interaction. A pharmacokinetic expression for the effect of inhibition on the ratio (R) of the new half life of a drug in the presence of inhibitor to the normal half life or for the steady state concentration in the absence and presence of inhibitor was developed by Rowland (1975). Determination of R requires knowledge of the fraction of the dose eliminated by the particular metabolic pathway in the absence of inhibitor (fm), the amount of inhibitor (I) and the inhibitor constant (K_I).

$$\frac{R = t_{1/2}\,\text{inhibited}}{t_{1/2}\,\text{normal}} = \frac{C_{ss}\,\text{inhibited}}{C_{ss}\,\text{normal}}$$

$$= \frac{1}{fm/(1 + I/K_2)} + (1 - fm).$$

If all the drug is eliminated by the inhibited route $fm = 1$ then the ratio will change dramatically with increasing inhibitor concentration. If less than 50% is eliminated by the inhibited route then it is unlikely that a clinically significant interaction will result, unless the therapeutic index of the drug is very narrow (Rowland, 1975).

Examples of enzyme inhibitors are found in a variety of drug groups including chemotherapeutic, anti-inflammatory, cardiovascular, and neuroleptic agents (Table 9).

TABLE 9. *Some Enzyme Inhibitors*

Allopurinol	Metronidazole
Amiodarone	Miconazole
Azapropazone	Nortriptyline
Chloramphenicol	Oral contraceptives
Chlorpromazine	Oxyphenbutazone
Cimetidine	Perphenazine
Danazol	Phenylbutazone
Dextropropoxyphene (propoxyphene)	Primaquine
	Propranolol
Diltiazem	Quinidine
Disulfiram	Sodium valproate
Ethanol (acute)	Sulfinpyrazone
Erythromycin	Sulfonamides
Imipramine	Thioridazine
Isoniazid	Trimethoprim
Ketoconazole	Verapamil
Metoprolol	

8. ENZYME INHIBITORS

8.1. CIMETIDINE AND OTHER ULCER HEALING DRUGS

The histamine H_2 antagonist cimetidine, widely used in the treatment of peptic ulcer, is a recognized enzyme inhibitor (Serlin *et al.*, 1979). As cimetidine is relatively nontoxic and is one of the most widely used drugs in clinical practice today, it is a useful agent for the further study of factors affecting the response to inhibition of drug metabolism in man. Cimetidine has been shown in man to impair the oxidative metabolism of a wide range of drugs including antipyrine, chlordiazepoxide, diazepam, propranolol and warfarin. The broad specificity of enzyme inhibition which was previously accepted is now being re-examined in the light of recent evidence suggesting selectivity of inhibition of cytochrome P-450 drug metabolism by cimetidine (Puurunen *et al.*, 1980). Cimetidine has been found to inhibit the 3 and 7 demethylation of theophylline with no inhibition of the 8 oxidation pathway. The absence of an inhibitory effect by cimetidine on the clearance of tolbutamide, ibuprofen, mexilitine and metronidazole has been demonstrated (Dey *et al.*, 1983). Steroid hydroxylation and epoxidation of carbamazepine are also spared of inhibition by cimetidine (Peden *et al.*, 1984; Levine *et al.*, 1984). The explanation may be forthcoming from recent studies showing cimetidine binding differentially to various forms of cytochrome P-450 enzyme which are known to exist (Boobis and Davies, 1984). The indications are therefore that cimetidine cannot be considered a universal inhibitor of phase one metabolism. However, cimetidine does provide useful information on the inhibition of drug metabolism in man.

That the inhibition of metabolism produced by cimetidine is dose dependent was suggested from the *in vitro* study of benzo[a]pyrine hydroxylation by human liver microsomes (Puurunen *et al.*, 1980). A concentration related effect on propranolol metabolism in man was also shown (Feely *et al.*, 1981b). The dose-dependent inhibitory effect of cimetidine on antipyrine clearance in healthy volunteers has also been shown (Fig. 5). The half life of antipyrine increased with increasing cimetidine concentration, the greater reduction in antipyrine clearance was associated with the higher cimetidine concentrations.

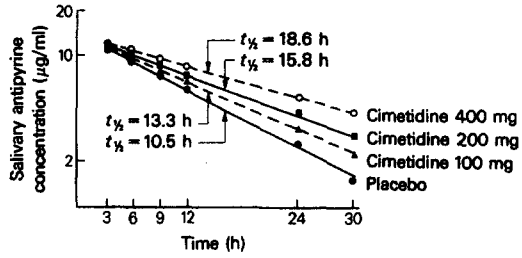

FIG. 5. Antipyrine concentrations and elimination half-life ($t_{1/2}$) during treatment with either placebo, cimetidine 100 mg, 200 mg or 400 mg four times daily.

The degree of enzyme inhibition may be related to the initial oxidizing capacity of the liver enzymes. An increased inhibitory effect by cimetidine on theophylline clearance in rats who had been pretreated with enzyme inducing agents has been found (Grygiel *et al.*, 1981). The effect of cimetidine was also more marked on the clearance of theophylline in smokers who had higher initial clearance rates, presumably due to enzyme induction related to smoking (Miners *et al.*, 1981). The percentage reduction in antipyrine clearance has been shown to be greater in subjects pretreated with the enzyme induced Rifampicin than in the control uninduced state (Fig. 6).

Studies suggest that the effect of cimetidine on oxidative metabolism is independent of the duration of pretreatment. The effect of pretreatment begun just 1 hr before administration of the test drug antipyrine was similar to that of 24 hr pretreatment and that reported for chronic cimetidine pretreatment (Somogyi and Gugler, 1982).

The responsiveness of hepatic enzymes to inducing agents may be reduced in the elderly (Wood *et al.*, 1979). However, the same may not be the case for enzyme inhibitors as elderly subjects are as sensitive to the inhibitory effects of cimetidine on oxidative metabolism as young subjects (Fig. 7). It has been shown in animals and in man that the effects of cimetidine on oxidative metabolism dissipate rapidly following cessation of therapy (Vestal *et al.*, 1983). Tolerance does not occur during chronic therapy (Patwardhan *et al.*, 1981). Thus as with enzyme induction the process of inhibition appears readily reversible but at a faster rate. This may also lead to

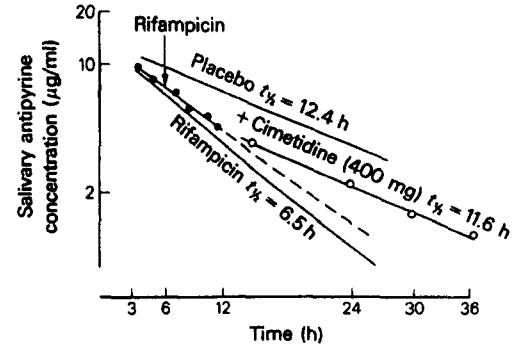

FIG. 6. Elimination half-life of antipyrine as control (placebo) during treatment with rifampicin (day 15; $t_{1/2}$ = 6.5 hr) and day 21 when cimetidine (400 mg four times daily) was commenced 12 hr following antipyrine.

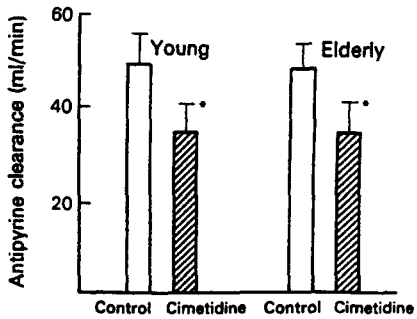

FIG. 7. Comparison of the reduction in antipyrine clearance produced by cimetidine (800 mg/day) in young and elderly subjects (mean ± SE mean, $n = 6$, $*P < 0.05$).

a pharmacological interaction with concurrently administered drugs. Cessation of cimetidine would result in decreased drug levels following removal of the inhibitory effect. Clinically important drug interactions resulting from enzyme inhibition have been described for cimetidine (Table 10).

The inhibitory effect of cimetidine is not shared by other H_2 antagonists such as famotidine and nitazidine. Ranitidine may have minor effects but these are not associated with clinically important interactions (Smith and Kendall, 1988). Another antiulcer drug omeprazole, a substituted benzimidazole compound, has also been found to inhibit the activity of drug metabolizing enzyme systems. *In vitro* studies using human liver microsomes demonstrated the inhibitory effect of omeprazole on both the high and low affinity components of 7 ethoxycoumarin deethylase (Jensen and Gugler, 1986). Omeprazole has been found to increase the half life and decrease the clearance of diazepam by 130% and 54% respectively. A clinically significant interaction may result from the administration of omeprazole to patients on phenytoin as omerazole may decrease phenytoin clearance by 15% (Gugler and Jensen, 1985).

8.2. Cardiovascular Drugs Including Calcium Antagonists and β-blockers

A number of cardiovascular drugs are known to be inhibitors of oxidative drug metabolism. These include β-blockers, calcium antagonists and the antiarrhythmics amiodarone and quinidine. The elimination of the calcium antagonist verapamil has been shown to be more rapid after the first dose than after chronic administration (Schwartz *et al.*, 1982). The mean serum concentrations of verapamil may be twice that predicted, during long term administration. Long term administration of verapamil has been found to be associated with prolongation of its half-life (64%) (Shand *et al.*, 1981). This apparent self inhibition of its metabolism is supported by further studies on antipyrine clearance which is reduced by 66% in patients on long term verapamil treatment (Rumiantsev *et al.*, 1986). The ability of verapamil to inhibit oxidative metabolism is likely to be of clinical consequence as exemplified by the recent reported interaction with the antiepileptic agent carbamazepine (CBZ) (MacPhee *et al.*, 1986). Administration of verapamil to patients with refractory partial epilepsy treated with carbamazepine resulted in symptoms of CBZ neurotoxicity within a few days. A mean rise of 46% in total, and 33% in free plasma CBZ concentrations in five out of six patients studied and a simultaneous fall of 36% in the ratio of the principal metabolite CBZ-10, 11-epoxide to CBZ was observed. Verapamil was found to increase the area under the CBZ concentration–time curve by 42% in one patient. The consequence of withdrawal of an enzyme inhibiting agent was demonstrated by the reduction of CBZ concentration from 12 mg/l to 7 mg/l following cessation of verapamil treatment. This was of clinical significance as seizure breakthrough was seen in the patient. As carbamazepine induces its own metabolism an inhibitory interaction is likely to produce a greater than expected increase in carbamazepine concentration. Verapamil also inhibits prazosin metabolism resulting in a greater hypotensive effect of the drug (Pasanisi *et al.*, 1984). Inhibition of theophylline metabolism resulting in toxic side effects has also been described (Burnakis *et al.*, 1983). A recent case report suggests that verapamil may inhibit metabolism of cyclosporin (Robson *et al.*, 1988).

Diltiazem is another calcium antagonist that inhibits oxidative drug metabolism. Diltiazem reduced antipyrine clearance by approximately 25% (Carrum *et al.*, 1986) and may also increase concentrations of cyclosporin (Grimo *et al.*, 1986). The calcium antagonist nifedipine does not produce significant enzyme inhibition in man (Bauer *et al.*, 1986).

Some β-blocking agents also demonstrate the ability to inhibit oxidative drug metabolism (Bax *et al.*, 1981). Greenblatt *et al.* (1978) showed that propranolol inhibits the metabolism of antipyrine. Subsequently both propranolol and metoprolol were found to inhibit antipyrine and theophylline metabolism (Bax *et al.*, 1983). Propranolol may increase plasma chlorpromazine concentrations accounting for the additive antipsychotic effects seen when these drugs are coadministered (Peet *et al.*, 1980). β-blocking drugs also inhibit lignocaine metabolism in rat liver microsomes and propranolol pretreatment in rats impaired its own metabolism and this may in part explain the unexpected accumulation of the drug with chronic dosing (Wood *et al.*, 1978).

The antiarrhythmic agent quinidine has been shown to inhibit metoprolol metabolism (Leeman *et al.*, 1986). It is a potent inhibitor of both sparteine

TABLE 10. *Some Important Interactions with Cimetidine*

Warfarin	Anticonvulsants
Theophylline	—Phenytoin
Antiarrhythmics—Lignocaine	Carbamazepine
Procainamide	Valproic Acid
β-blockers—Propranolol	Benzodiazepines
Metoprolol	Chlordiazepoxide
Labetolol	Clobazam
	Desmethyldiazepam
Calcium Antagonists—Nifedipine	Diazepam
Verapamil	Midazolam
Diltiazem	Nitrazepam
	Triazolam

From Smith and Kendall, 1988. Reprinted with permission of the authors and the copyright holder, ADIS Press, Ltd, Auckland.

and debrisoquine oxidation in healthy volunteers, thus raising the possibility that quinidine may interfere with phenotyping and convert an extensive metabolizer (Inaba *et al.*, 1986).

Amiodarone is another antiarrhythmic agent that may produce clinically important drug interactions. Potentiation of warfarin anticoagulation (Hamer *et al.*, 1982) and increased serum concentrations of concomitant quinidine or procainamide treatment has been found, enzyme inhibition by amiodarone was the proposed underlying mechanism (Saal *et al.*, 1984). Animal studies have demonstrated that amiodarone is a potent inhibitor of oxidative drug metabolism in the rat and markedly reduces cytochrome P-450 concentration (Duenas-Laita *et al.*, 1987).

8.3. CHEMOTHERAPEUTIC AGENTS

A number of chemotherapeutic agents have been found to inhibit drug metabolism. Pharmacokinetic interactions involving the macrolide antibiotics erythromycin and triacetyloleandomycin have been described. Both drugs are known to affect the cytochrome P-450 drug metabolizing system and *in vitro* studies suggest that a metabolite of these compounds forms a stable complex with the heme portion of reduced cytochrome P-450 thus producing enzyme inhibition (Pessayre *et al.*, 1981). Of the two agents triacetyloleandomycin appears to be the more potent inhibitor of microsomal drug metabolism. It has been found to significantly decrease the metabolism of methylprednisolone, theophylline (Weinberger *et al.*, 1977) and carbamazepine (Wroblewski *et al.*, 1986), the inhibition of methylprednisolone metabolism being dose dependent and time related (Szefler *et al.*, 1982). The ability of triacetyloleandomycin to cause ergotism in patients receiving ergot alkaloids and cholestatic jaundice in patients receiving oral contraceptives may also be related to the inhibitory effect on drug metabolism (Matthews and Havill, 1979). Erythromycin appears to be a weaker inhibitor of drug metabolism and in some individuals the metabolism of methylprednisolone, theophylline, carbamazepine and warfarin may be affected (Ludden, 1985).

Inhibition by erythromycin of the conversion of carbamazepine to its 10, 11 epoxide metabolite results in a significant clinical interaction with patients developing carbamazepine toxicity (Barzaghi *et al.*, 1987). Recent studies suggest the possibility of an interaction between erythromycin and cyclosporin with a resultant increase in cyclosporin concentration (Freeman *et al.*, 1987). The mean change in drug clearance is a reduction of the order of 25% in patients administered the macrolide antibiotics mentioned above.

Hypoglycemic coma has resulted from the concomitant administration of chloramphenicol and sulfaphenazole to noninsulin-dependent diabetic patients treated with the oral hypoglycemic agent tolbutamide presumably due to enzyme inhibition (Hansen and Christensen, 1977). Tolbutamide excretion is almost completely dependent on a single route of metabolism that is particularly sensitive to inhibition. Sulfamethazole, another sulfonamide, has similar though less pronounced inhibitory effects on the metabolism of other drugs including phenytoin, tolbutamide and warfarin. The trimethoprim sulfamethoxazole complex (Septrin) produces a stereoselective inhibition of warfarin, decreasing the clearance of the more potent *S* isomer of warfarin by approx 20%, increasing anticoagulant effect by 65% (O'Reilly, 1980). The heterocyclic nitro derivative metronidazole also inhibits the metabolism of *S*-warfarin increasing the anticoagulant effect by 40% (O'Reilly, 1976).

In vitro studies suggest isoniazid to be a noncompetitive inhibitor of drug metabolism. Isoniazid impaired the metabolism of phenytoin in patients found to be slow inactivators of isoniazid (Miller *et al.*, 1979). This interaction is of clinical importance as one study of twenty-two hospitalized medical patients who received phenytoin and isoniazid for at least five days reported a 27% incidence of central nervous system toxicity due to phenytoin. The incidence of phenytoin toxicity without isoniazid has been estimated to be approximately 3%. Isoniazid has also been found to inhibit the metabolism of carbamazepine (Wright *et al.*, 1982).

The new synthetic quinolone antibiotics have been found to reduce the hepatic clearance of some coadministered drugs (Edwards *et al.*, 1988). Enoxacin appears to be the most potent inhibitor consistently decreasing theophylline clearance by 50% or more (Wijands *et al.*, 1985). Ciprofloxacin and pefloxacin reduce theophylline clearance to a smaller extent, approximately 20–30% (Wijands *et al.*, 1986). Enoxacin decreases the clearance of *R*-warfarin and antipyrine with no effect on chlorpropamide, glibenclamide or phenytoin (Edwards *et al.*, 1988). Norfloxacin, ofloxacin and nalidixic acid appear to have minimal effects on theophylline clearance.

8.4. ANTI-INFLAMMATORY AND ANALGESIC DRUGS

Phenylbutazone is an anti-inflammatory agent capable of enzyme inhibition. The interaction between phenylbutazone and warfarin was one of the earliest examples of inhibition of metabolism and the complexity of the interaction was of interest (Lewis *et al.*, 1974). Coadministration of phenylbutazone produced an augmented anticoagulant effect without altering the apparent plasma concentration of warfarin. Study of the individual isomers revealed a reduction in clearance of the *S* isomer but an accelerated clearance of the *R* isomer. As the *S* isomer is five times more potent than the *R* isomer inhibition of its metabolism provides one mechanism for the increased anticoagulant effect.

Sulfinpyrazine, a derivative of phenylbutazone, is also capable of inhibition of drug metabolism in man. Clinically important interactions are potentiation of the effects of oral anticoagulants, theophylline and tolbutamide as a result of enzyme inhibition (Miners *et al.*, 1982a,b).

Dextropropoxyphene is similar structurally to methadone but it is a weaker analgesic and not classified as a narcotic despite having several properties in common with that group. Dextrapropoxyphene may be involved in clinically significant interactions as a result of its ability to inhibit drug

metabolism. It inhibits the metabolism of anticonvulsants phenytoin, phenobarbitone and carbamazepine (Kutt, 1971; Dam *et al.*, 1977), drug concentrations may be increased by approximately 70% and central nervous system adverse effects become apparent (Hansen *et al.*, 1980). Concurrent administration with the oral anticoagulant warfarin may result in raised warfarin levels and hemorrhage (Jones, 1976). Recent studies suggest that dextropropoxyphene may also inhibit theophylline metabolism (Robson *et al.*, 1987).

8.5. Neuroleptics

The phenothiazine derivative chlorpromazine has been shown to inhibit competitive drug metabolism *in vitro*. Chlorpromazine also inhibits the metabolism of propranolol in man, the clearance of propranolol being reduced by 30%. As a result of this inhibition an 80% increase in steady state propranolol concentrations was found (Vestal *et al.*, 1979).

Thioridazine, another phenothiazone drug, has been reported to produce phenytoin toxicity when the drugs are administered concomitantly (Vincent, 1980). Recent studies suggest that the hydroxylation of debrisoquine is decreased in patients receiving therapeutic doses of thioridazine and levomepromazine (Syvalahti *et al.*, 1986). High plasma concentrations of imipramine and desipramine have been observed in patients simultaneously treated with oral imipramine and intramuscular fluphenazine (Nelson and Tatlow, 1980). Tricyclic antidepressants may also inhibit the metabolism of neuroleptics if the oxidation is catalyzed by the same form of cytochrome P-450. Warfarin metabolism is also inhibited by tricyclic antidepressants (Barber and Douglas, 1980).

8.6. Oral Contraceptives

Oral contraceptives have been shown to influence the kinetics of concurrently administered drugs. Drugs eliminated by oxidative pathways such as antipyrine and diazepam have decreased metabolic clearance and prolonged elimination half life in oral contraceptive users (Carter *et al.*, 1974). Ethynylestradiol administered for five days decreased the cytochrome P-450 content of rat livers and was associated with a decreased turnover of the P-450 (Mackinnon *et al.*, 1977). Oral contraceptives may also decrease the metabolism of corticosteroids (Hunter *et al.*, 1973). In contrast some drugs metabolized by phase II conjugation reactions show increased clearance, e.g. paracetamol (Legler and Benet, 1986).

The clinical significance of these interactions is not entirely clear but the widespread use of oral contraceptives suggests caution in patients receiving the combination of potent drugs and oral contraceptives.

8.7. Valproic Acid

Valproic acid is a relatively recent addition to the treatment of epilepsy. However its ability to produce clinically significant drug interactions following enzyme inhibition is already apparent (Levy and Koch, 1982). Elevated blood levels of phenobarbitone producing increased sedation are seen when valproic acid is given to patients treated with phenobarbitone. A 32% increase in phenobarbitone half life with an associated 28% decrease in clearance follows valproic acid administration (Bruni *et al.*, 1980). Valproic acid also inhibits the elimination of ethosuxamide and carbamazepine (MacPhee *et al.*, 1988).

8.8. Disulfiram

Inhibition of the enzyme aldehyde dehydrogenase resulting in accumulation of acetaldehyde is the underlying mechanism of action of this drug used in the treatment of chronic alcoholism. It does, however, possess the ability to inhibit the monooxygenase enzymes system and impairs the metabolism of antipyrine, warfarin and phenytoin (Vesell *et al.*, 1975). The clearance of chlordiazepoxide and diazepam are reduced by 50% and 40% respectively when given with disulfiram. The drug does not inhibit phase two reactions.

8.9. Allopurinol

Allopurinol is another example of a drug capable of inhibiting both monooxygenase enzymes and enzymes not involved in drug metabolism. Inhibition of xanthine oxidase decreasing the production of uric acid is the underlying basis of its use in treatment of gout. As xanthine oxidase is also involved in the metabolism of mercaptopurine it is not surprising that an interaction between these two drugs has been reported. Concurrent use of these agents results in increased mercaptopurine activity and toxicity (Elien *et al.*, 1963). Allopurinol has also been found to inhibit the metabolism of antipyrine, bishydroxycoumarin and theophylline (Vesell *et al.*, 1970; Manfredi and Vesell, 1981). Recently we described a significant interaction between allopurinol and warfarin. Using the ^{14}C aminopyrine breath test as an index of drug metabolism an increase in the mean ^{14}C-aminopyrine half life by 44% following allopurinol treatment was noted (Barry and Feely, 1988). It is possible that the effect of allopurinol on oxidative metabolism is greater than hitherto appreciated.

8.10. Monoamine Oxidase Inhibitors

Drugs may also alter the pharmacological action of other drugs by inhibiting nonmicrosomal pathways of metabolism. The use of monoamine oxidase inhibitors, e.g. tranylcypromine, render patients sensitive to subsequent treatment with sympathomimetic agents which are metabolized by the mitochondrial enzyme. Patients treated with these inhibitors have developed hypertensive reactions following ingestion of cheese, other foods rich in tyramine, amphetamines or direct and indirect sympathomimetic agents (Tollefson, 1983).

The importance of genetic factors in determining susceptibility of an individual to drug interactions involving enzyme inhibition is exemplified by the phenytoin–isoniazid interaction mentioned above. All cases occur in slow metabolizers of isoniazid. Therefore many factors, both genetic and environmental, influence the rate of drug elimination and

hence drug response in man. The ability of drugs to modify the activity of drug metabolizing enzyme systems may result in increased or decreased activity of concurrently administered drugs. Clinically significant interactions are more likely following enzyme inhibition and when drugs with a narrow therapeutic index are used.

9. CONCLUSION

Too many of the interactions noted above as a consequence of induction or inhibition were at the expense of patient toxicity. Assessment of the potential for such effects using animal, *in vitro* and human screening studies should be mandatory in the assessment of new drugs. It is clear that no single test has a high predictive value. Nonetheless it is possible by combining *in vitro* and *in vivo* studies both in animal and man to identify drugs at particular risk. Close monitoring, including frequent measurement of drug levels in susceptible patients receiving drugs with a low therapeutic ratio should be included in this assessment.

REFERENCES

AARTS, E. M. (1965) Evidence for the function of D glucaric acid as an indicator for drug induced enhanced metabolism through the glucuronic pathway in man. *Biochem. Pharmac.* **11:** 359–363.

ACOCELLA, G., PANGANI, V., NORCHETTI, M., BARONI, G. C. and NICOLIS, F. B. (1971) Kinetic studies on rifampicin I. Serum concentration analysis in subjects treated with different oral doses over a period of two weeks. *Chemotherapy* **16:** 356–370.

ACOCELLA, G., NATTIUSSI, R. and SEGRE, G. (1978) Multicompartmental analysis of serum urine and bile concentrations of rifampicin and desacetyl rifampicin in subjects treated for one week. *Pharmac. Res. Commun.* **10:** 271–288.

ADESNIK, M., BAR-NUN, S., MASCHIO, F., ZUNICH, M., LIPPMAN, A. and BAIRD, E. (1981) Mechanism of induction of cytochrome P450 by Phenobarbital. *J. biol. Chem.* **256:** 10340–10345.

ANDERS, M. W. and MANNERING, G. J. (1966) Inhibition of drug metabolism I. Kinetics of the inhibition of the *N*-demethylation of ethylmorphine by 2 diethylaminoethyl 2.2 diphenylvalerate HCl (SKF 525 A) and related compounds. *Molec. Pharmac.* **2:** 319–327.

ANDERSON, K. E. (1988) Influences of diet and nutrition on clinical pharmacokinetics. *Clin. Pharmacokinet.* **14:** 325–246.

ANDERSON, K. E., CONNEY, A. H. and KAPPAS, A. (1979) Nutrition and oxidative drug metabolism in man, relative influence of dietary lipids, carbohydrate and protein. *Clin. Pharmac. Ther.* **26:** 493–501.

ANTHINTZ, M. A., TOLENTINO, M. and KODAI, M. F. (1968) Effect of butabarbital an orally administered anticoagulant. *Curr. Ther. Res.* **10:** 70–73.

ARIENS, E. J. (1979) Receptors: from fiction to fact. *Trends Pharmac. Sci.* **1:** 11–15.

BACK, D. J., BRECKENRIDGE, A. M., CRAWFORD, F. E., MCIVER, M., ORME, M., PARK, B. K., ROWE, P. H. and SMITH, E. (1979) The effect of rifampicin on norethisterane pharmacokinetics. *Eur. J. Clin. Pharmac.* **15:** 193–197.

BARBER, H. E. and DOUGLAS, A. S. (1980) Drug interactions involving anticoagulant drugs. In: *Clinically Important Adverse Drug Interactions. Cardiovascular and Respiratory Disease Therapy*, Vol. 1, pp. 141–152, PETRIE, J. (ed.) Elsevier, Amsterdam.

BARRY, M. and FEELY, J. (1988) Inhibition of drug metabolism by allopurinol. *Br. J. clin. Pharmac.* **26:** 649P–650P.

BARRY, M., DUENAS-LAITA, A., MAC MATHUNA, P. and FEELY, J. (1986) Amiodarone inhibition of hepatic oxidative drug metabolism. An acute and chronic study. *Br. J. clin. Pharmac.* **87:** 144.

BARRY, M., DUENAS LAITA, A., MAC MATHUNA, P. and FEELY, J. (1987) Increase in renal cytochrome P-450 and NADPH cytochrome c reductase activity following drug inhibition of hepatic monooxygenase activity. *Biochem. Pharmac.* **36:** 768–769.

BARZAGHI, N., GATTI, G., CREMA, F., MONTELEONE, M., AMIONE, C., LEONE, L. and PERUCCA, E. (1987) Inhibition by erythromycin of the conversion of carbamazepine to its active 10, 11-epoxide metabolite. *Br. J. clin. Pharmac.* **24:** 836–838.

BAUER, L. A., STENWALL, M., HORN, J. R., DAVIS, R., OPHEIM, K. and GREENE, L. (1986) Changes in antipyrine and indocyanine green kinetics during nifedipine, verapamil and diltiazem therapy. *Clin. Pharmac. Ther.* **40:** 239–242.

BAX, N. D. S., LENNARD, M. S. and TUCKER, G. T. (1981) Inhibition of antipyrine metabolism by β adrenoceptor antagonists. *Br. J. clin. Pharmac.* **12:** 779–784.

BAX, N. D. S., LENNARD, M. S., AL ASADY, S., DEACON, C. S., TUCKER, G. T. and WOODS, H. F. (1983) Inhibition of drug metabolism by β-adrenoceptor antagonists. *Drugs* **25** (Suppl. 2): 121–126.

BENNETT, P. N., JOHN, V. A. and WHITMARSH, U. B. (1982) Effect of rifampicin on metoprolol and antipyrine kinetics. *Br. J. clin. Pharmac.* **13:** 387–391.

BENOWITZ, N. L. and JONES, R. T. (1977) Effects of delta 9 tetrahydrocannabinol on drug disposition and metabolism. *Clin. Pharmac. Ther.* **22:** 259–268.

BERLIN, C. M., STTUDEMAN, J. M. and VESELL, E. S. (1975) Quinine induced alterations in drug disposition. *Clin. Pharmac. Ther.* **18:** 670–679.

BERMAN, M. L. and GREEN, O. C. (1971) Acute stimulation of cortisol metabolism by phenobarbital in man. *Anesthesiology* **34:** 365–369.

BLACK, M. and SHERLOCK, S. (1970) Treatment of Gilberts Syndrome with phenobarbitone. *Lancet* **1:** 1359–1362.

BLACK, M., FEVERY, J., PARKER, D., JACOBSON, J., BILLING, B. H. and CARSON, E. R. (1974) Effect of phenobarbitone on plasma [^{14}C] bilirubin hyperbilirubinaemia. *Clin. Sci. molec. Med.* **46:** 1–17.

BLEDSOE, T., ISLAND, D. P., NEY, R. L. and LIDDLE, G. W. (1964) An effect of op DDD on the extrarenal metabolism of cortisol in man. *J. clin. Endocr. Metab.* **24:** 1303–1311.

BOGAERT, M. G., ROSSELL, M. T. and BELPAIRE, F. M. (1971) Metabolism of nitroglycerine in man, influence of phenobarbital. *Archs int. Pharmacodyn. Thér.* **192:** 198–199.

BOLT, H. M., KAPPAS, H. and BOLT, M. (1975) Effect of rifampicin treatment on metabolism of oestradiol and 17α ethinyloestradiol by human liver microsomes. *Eur. J. clin. Pharmac.* **8** (5): 301–307.

BOOBIS, A. R. and DAVIES, D. S. (1984) Human cytochrome P-450. *Xenobiotica* **14:** 151–185.

BOOBIS, A. R., BRODIE, M. J., KAHN, G. C., FLETCHER, D. R., SAUNDERS, J. H. and DAVIES, D. S. (1980) Monooxygenase activity in human liver microsomal fractions of needle biopsy specimens. *Br. J. clin. Pharmac.* **9:** 11–19.

BRANCH, R. A. and SHAND, D. G. (1979) A re-evaluation of intersubject variation in enzyme induction in man. *Clin. Pharmacokinet.* **4:** 104–110.

BRECKENRIDGE, A. M. and ORME, M. L'E. (1971) Clinical implications of enzyme induction. *Ann. N.Y. Acad. Sci.* **179:** 421–431.

BRECKENRIDGE, A. M., ORME, M., THERGEIRSSON, S. S., DAVIES, D. S. and BROOKS, R. K. (1971) Drug interactions with warfarin: studies with dichloralphenazone chloral hydrate and phenazine (antipyrine). *Clin. Sci.* **40:** 351–364.

BRECKENRIDGE, A., ORME, M., DAVIES, L., THORGEIRSSON, S. S. and DAVIES, D. S. (1973) Dose dependent enzyme induction. *Clin. Pharmac. Ther.* **14:** 514–520.

BRODIE, M. J., BOOBIS, A. R., HILLYARD, C. J., ABEYASKEKERA, G., MACINTYRE and PARK, B. K. (1981) Effect of isoniazid on Vitamin D metabolism and hepatic monooxygenase activity. *Clin. Pharmac. Ther.* **30:** 363–367.

BRUNI, J., WILDER, B. J., PERCHALSKI, R. J., HAMMOND, E. J. and VILLAREAL, H. J. (1980) Valproic acid and plasma levels of phenobarbital. *Neurology* **30:** 94–97.

BURNAKIS, T. G., SELDEN, M. and CZAPLICKI, A. D. (1983) Increased serum theophylline concentrations secondary to oral verapamil. *Clin. Pharmac.* **2:** 458–461.

BURNSTEIN, S. and KLAIBER, E. L. (1965) Phenobarbital induced increase in 6 beta hydroxycortisol excretion, due to its significance in urine. *J. clin. Endocr. Metab.* **25:** 293–296.

BURNSTEIN, S., KIMBALL, H. L., KLAIBER, E. L. and FUT, M. (1967) Metabolism of 2 α and 6 β hydroxycortisol in man, determination of production rates of 6 β hydroxycortisol with and without phenobarbital administration. *J. clin. Endocr.* **27:** 491–499.

CALDWELL, J. (1982) Conjugation reactions in foreign compound metabolism. Definition, consequences and species variations. *Drug Metab. Rev.* **5:** 745–777.

CAMPBELL, T. C. and HAYES, T. R. (1974) Role of nutrition in the drug metabolizing enzyme system. *Pharmac. Rev.* **26:** 171–197.

CARRUM, G., EGAN, J. M. and ABERNETHY, D. R. (1986) Diltiazem treatment impairs hepatic drug oxidation. Studies of antipyrine. *Clin. Pharmac. Ther.* **40:** 140–143.

CARTER, D. E., GOLDMAN, J. M., BRESSLER, R., HUXTABLE, R. J., CHRISTIAN, C. D. and HEINE, M. W. (1974) Effect of oral contraceptives on drug metabolism. *Clin. Pharmac. Ther.* **15:** 22–31.

CINTI, D. L., GLORIEUX, F. H., DEVLIN, E. E., GOLUB, E. E. and BRENNER, F. (1978) 25 hydroxycholecalciferol high affinity substrate for hepatic cytochrome P450. *Adv. exp. Med. Biol.* **881:** 441–453.

COHN, R. M. and ROTH, K. S. (eds) (1983) Porphyria. In: *Metabolic Disease*, pp. 369–378, W. B. Saunders Co., Philadelphia.

CUCINELL, S. A., CONNEY, A. H., SANSUR, M. and BURNS, J. J. (1965) Drug interactions in man. Lowering effect of phenobarbital on plasma levels of bishdydroxycoumarin (Docoumaral) and diphenylhydantoin (Dilantin). *Clin. Pharmac. Ther.* **6:** 420–429.

DAM, M., KRISTENSEN, C. B., HANSEN, B. S. and CHRISTIANSEN, J. (1977) Interaction between carbamazepine and propoxyphene in man. *Acta neurol. scand.* **56:** 603–607.

DANHOF, M., VERBEEK, R. M. A., VAN BOXTEL, C. J., BOEIJINGA, J. K. and BREIMER, D. D. (1982) Differential effects of enzyme induction on antipyrine metabolite formation. *Br. J. clin. Pharmac.* **13:** 379–386.

DAVIS, M., SIMMONS, C. J., DORDONI, B. and WILLIAMS, R. (1974) Urinary D glucaric acid excretion and antipyrine kinetics during enzyme induction. *Br. J. clin. Pharmac.* **1:** 253–258.

DEY, N. G., CASTLEDEN, C. M., WARD, J., CORNHILL, J. and MCBURNEY, A. (1983) The effect of cimetidine on tolbutamide kinetics. *Br. J. clin. Pharmac.* **16:** 438–440.

DUENAS-LAITA, A., BARRY, M., GANUZA-BIDARTE, M., MAC MATHUNA, P. and FEELY, J. (1986) Effect of co-trimoxazole on hepatic drug metabolism. *Ir. J. med. Sci.* **155**(9): 330.

DUENAS-LAITA, A., BARRY, M., MAC MATHUNA, P. and FEELY, J. (1987) Effects of chronic treatment with amiodarone on hepatic demethylation and cytochrome P 450. *J. Pharm. Pharmac.* **18:** 757–759.

DURAND, D. V., HAMPDEN, C., BOOBIS, A. R., PARK, B. K. and DAVIES, D. S. (1986) Induction of mixed function oxidase activity in man by rifampicin (MDL 473) a long acting rifamycin derivative. *Br. J. clin. Pharmac.* **21:** 1–7.

EDWARDS, D. J., BOWLES, S. K., SVENSSON, C. K. and RYBAK, M. J. (1988) Inhibition of drug metabolism by quinolone antibiotics. *Clin. Pharmacokinet.* **15:** 194–204.

EDWARDS, O. M., COURTENAY-EVANS, R., GALLEY, J., HUNTER, J. and TAIT, A. J. (1974) Changes in cortisol metabolism following rifampicin therapy. *Lancet* **2:** 549–551.

EICHELBAUM, M., TOMSON, T., TYBRING, G. and BERTILSSON, L. (1985) Carbamazepine metabolism in man, Induction and pharmacogenetic aspects. *Clin. Pharmacokinet.* **10:** 80–90.

ELIEN, G. B., CALLAHAN, S., NATHAN, H., BIEBER, S. and RUNDLES, R. W. (1963) Potentiation by inhibition of drug degradation: 6 substitute purines and xanthine oxidase. *Biochem. Pharmac.* **12:** 85–93.

ERIKSSON, L. C. (1973) Studies of the biogenesis of endoplasmic reticulum in the liver cell. *Acta path. microbiol. scand.* (A) (Suppl. 239): 1–72.

ERIKSSON, M., PAALZOW BOLME, P. and MARIUM, T. W. (1984) Chloramphenicol pharmacokinetics in Ethiopian children of differing nutritional status. *Eur. J. clin. Pharmac.* **24:** 819–823.

FEELY, J., CROOKS, J. and STEVENSON, I. H. (1981a) The influence of age, smoking and hyperthyroidism on plasma propranolol steady state concentration. *Br. J. clin. Pharmac.* **12:** 73–78.

FEELY, J., WILKINSON, G. R. and WOOD, A. J. J. (1981b) Reduction of liver blood flow and propranolol metabolism by cimetidine. *New Engl. J. Med.* **304:** 692–695.

FORREST, F. M., FORREST, L. M. and SERRA, M. T. (1970) Modification of chlorpromazine metabolism by some other drugs frequently administered to psychiatric patients. *Biol. Psych.* **2:** 53–58.

FRANTZ, A. G., RUTZ, F. H. and JAILER, J. W. (1961) 6 β hydroxycortisol and other polar corticosteroids, measurement and significance in human urine. *J. clin. endocr. Metab.* **21:** 1290–1303.

FREEMAN, D. J., MARTELL, R., CORRUTHERS, S. G., HEINRICHS, D., KEOWN, P. A. and STILLER, C. R. (1987) Cyclosporin–erythromycin interaction in normal subjects. *Br. J. clin. Pharmac.* **23:** 776–778.

GARNER, R. C. and MCLEAN, A. E. M. (1969) Increased susceptibility of carbon tetrachloride poisoning in the rat after pretreatment with oral phenobarbitone. *Biochem. Pharmac.* **18:** 645–650.

GELBER, R. H., GOOI, H. C. and REES, R. J. W. (1975) The effect of rifampicin on dapsone metabolism. *Proc. Western Pharmac. Soc.* **18:** 330–334.

GELEHRTER, T. D. (1972) Enzyme Induction. *New Engl. J. Med.* **287:** 900–903.

GILLETTE, J. R. and KAMM, J. J. (1960) The enzymatic formation of sulfoxides, the oxidation of chlorpromazine and 4,4-diaminodiphenylsulphide by guinea pig liver microsomes. *J. Pharmac. exp. Ther.* **129:** 262–267.

GOLDBERG, D. M. (1980) Hepatic Protein synthesis in health and disease, with special reference to microsomal enzyme induction. *Clin. Biochem.* **13:** 216–226.

GOLDSTEIN, J. A. (1984) Mechanism of induction of hepatic drug metabolizing enzymes. Recent advances. *Trends Pharmac. Sci.* **5**(7): 290–292.

GREENBLATT, D. J., FRANKE, K. and HOFFMAN, D. H. (1978) Impairment of antipyrine clearance in humans by propranolol. *Circulation* **57:** 1161–1164.

GRIMO, J. M., SABATE, I., CASTELAO, A. M. and ALSINA, J. (1986) Influence of diltiazem on cyclosporin clearance. *Lancet* **i:** 1387.

GRYGIEL, J. J., DREW, J. O., MINERS, J., RENVELL, J., WILLOUGHBY, J. O. and BIRKETT, D. J. (1981) The effect of cimetidine on theophylline clearance in the rat. *Clin. exp. Physiol. Pharmac.* **8:** 632.

GUGLER, R. and JENSEN, J. C. (1985) Omeprazole inhibits oxidative drug metabolism. *Gastroenterology* **89:** 1235–1241.

HAHN, T. J., BENDIN, B. A., SCHARP, C. R. and HADDAD, J. G. (1972) Effect of chronic anticonvulsant therapy on serum 25 hydroxycholecalciferol levels in adults. *New Engl. J. Med.* **287:** 900–903.

HAMER, A., PETER, T., MANDEL, W. J., SCHEINMAN, M. M. and WEISS, D. (1982) The potentiation of warfarin anticoagulation by amiodarone. *Circulation* **65:** 1025–1029.

HAMMER, W. and SJOQVIST, F. (1967) Plasma levels of monomethylated tricyclic antidepressants during treatment with imipramine like compounds. *Life Sci.* **6:** 1895–1903.

HANSEN, B. S., DAM, M., BRANDT, J., HVIDBERG, E. F. and ANGELO, H. (1986) Influence of dextropropoxyphene on steady state serum levels and protein binding of three antiepileptic drugs. *Acta neurol. scand.* **61:** 357–367.

HANSEN, J. M. and CHRISTENSEN, L. J. (1977) Drug interactions with oral sulphonylurea hypoglycaemic drugs. *Drugs* **13:** 24–34.

HANSEN, J. M., SIERBOEK-NIELSEN, K., KRISTENSEN, M., SKOUSTED, L. and CHRISTENSEN, L. K. (1971) Effect of diphenylhydantoin on the metabolism of dicoumarol in man. *Acta med. scand.* **189:** 15–19.

HARDWICK, J. P., GONZALES, F. J. and KASPER, C. B. (1983) Cloning of DNA complimentary to cytochrome P450 induced by pregnenolone 16 α carbonitrile. *J. biol. Chem.* **258:** 10182–10186.

HEIRWEGH, P. H. and BLANKAERT, N. (1981) Analysis of bilirubin conjugates. In: *Methods in Enzymology*, Vol. 77, pp. 391–398, JAKOBY, W. B. (ed.) Academic Press, London.

HEMPEL, E., BOHM, W., CAROL, W. and KLINGER, G. (1973) Medikamen tose Enzymunduktion und harmancle kontrazeptian. *Zentrablatt fur Gynakologie* **95:** 1451–1457.

HENDRIKSE, W., MCKIERNAN, J., PICKUP, M. and LOWE, J. (1979) Rifampicin induced non responsiveness to corticosteroid treatment with nephrotic syndrome. *Br. med. J.* **1:** 306.

HEPNER, G. W. and VESELL, E. S. (1974) Assessment of aminopyrine metabolism in man by breath analysis after oral administration of ^{14}C-aminopyrine. *New Engl. J. Med.* **291:** 1384–1388.

HEPNER, G. W., VESELL, E. S., LIPTON, A., HARVEY, H. A., WILKINSON, G. R. and SCHENKER, S. (1977) Disposition of aminopyrine, antipyrine, diazepam and indocyanine green in patients with liver disease or on anticonvulsant drug therapy: diazepam breath test and correlations in drug elimination. *J. Lab. clin. Med.* **90:** 440–456.

HILDEBRANDT, A. G., ROOTS, I., SPECK, M., SAALFRANK, K. and KEWITZ, H. (1975) Evaluation of *in vivo* parameters of drug metabolizing enzyme activity in man after administration of clemastine, phenobarbital or placebo. *Eur. J. clin. Pharmac.* **8:** 327–336.

HOFFBRAND, A. V. and NECHELES, T. F. (1968) Mechanism of folate deficiency in patients receiving phenytoin. *Lancet* **2:** 528–530.

HOMAN, J., ROTTER, S., SCHNEIDER, S., ROTGER, P., KRUTZ, F., KROHER, R., KAMENISCH, W., PAUL, F. and MATTHES, J. K. (1985) Influence of cimetidine on CCl_4 induced liver injury and survival in rats. *Biochem. Pharmac.* **34:** 415–416.

HOUSTON, J. B. (1977) Effect of vitamin C supplement on antipyrine disposition in man. *Br. J. clin. Pharmac.* **4:** 236–239.

HUFFMAN, D. H., SHOEMAN, D. W., PENTIKAINEN, P. and AZARNOFF, D. L. (1973) The effect of spironolactone on antipyrine metabolism in man. *Pharmacology* **10:** 338–344.

HUNTER, J. and CHASSEAUD, L. F. (1976) Clinical aspects of microsomal enzyme induction. In: *Progress in Drug Metabolism I*, pp. 129–192, BRIDGES, J. W. and CHASSEAUD, L. F. (eds) Wiley, London.

HUNTER, J., MAXWELL, J. D., CARELLA, M., STEWART, D. A. and WILLIAMS, R. (1971a) Urinary D glucaric acid excretion as a test for enzyme induction in man. *Lancet* **i:** 572–575.

HUNTER, J., MAXWELL, J. D., STEWART, D. A., WILLIAMS, R., ROBINSON, J. and RICHARDSON, A. (1971b) Environmental exposure to organochlorine pesticides and hepatic enzyme induction in man. *Gut* **12:** 761.

HUNTER, J., MAXWELL, J. D., STEWART, D. A. and WILLIAMS, R. (1973) Urinary D glucaric acid excretion and total liver content of cytochrome P 450 in guinea pigs. Relationship during enzyme induction and following inhibition of protein synthesis. *Biochem. Pharmac.* **22:** 743–747.

HUNTER, J., BURNHAM, W. R., CHASSEAUD, L. F. and DOWN, W. (1974) Changes in D glucaric acid excretion in relationship to alterations in the rate of antipyrine metabolism. *Biochem. Pharmac.* **23:** 2480–2483.

INABA, T., TYNDALE, R. E. and MAHON, W. A. (1986) Quinidine potent inhibition of sparteine and debrisoquine oxidation *in vivo*. *Br. J. clin. Pharmac.* **22:** 199–200.

IQBAL, S. J. and TAYLOR, W. H. (1982) Treatment of Vitamin D2 poisoning by induction of hepatic enzymes. *Br. med. J.* **285:** 541–542.

JACKSON, L., HAMEIDA, M. and ROBERTS, C. J. C. (1978) The features of hepatic enzyme induction with glutethimide. *Br. J. clin. Pharmac.* **6:** 525–528.

JENNER, P. and TESTA, B. (1976) Induction of extrahepatic drug metabolising enzyme systems. In: *Drug Metabolism: Chemical and Biochemical Aspects*, p. 445, SWARBRICK, J. (ed.) Marcel Dekker, New York.

JENSEN, J. C. and GUGLER, R. (1986) Inhibition of human liver cytochrome P 450 by omeprazole. *Br. J. clin. Pharmac.* **21:** 328–330.

JEZEQUEL, A. M., ORLANDI, F. and TENCANI, L. T. (1971) Changes of the smooth endoplasmic reticulum induced by rifampicin in human and guinea pig hepatocytes. *Gut* **12:** 984–987.

JOHNSEN, S. G., KAMPMANN, J. P., BENNET, E. P. and JORGENSEN, F. S. (1976) Enzyme induction by oral testosterone. *Clin. Pharmac. Ther.* **20:** 233–237.

JONES, P. B. C., DURRIN, L. K., FISHER, J. M. and WHITLOCK, J. P. (1986) Control of gene expression by 2,3,7,8 tetrachlorodibenzo-*p* dioxin. *J. biol. Chem.* **261:** 6647–6653.

JONES, R. V. (1976) Warfarin and distalgesic interaction. *Br. med. J.* **1:** 460.

JUBIZ, W., MEIKLE, A. W., LEVINSON, R. A., MIZUTANI, S., WEST, C. D. and TYLER, F. H. (1970) Effect of diphenylhydantoin on the metabolism of dexamethasone. *New Engl. J. Med.* **283:** 11–24.

JUSKO, W. J. (1978) Role of tobacco smoking in pharmocokinetics. *J. Pharmcokinet. Biopharm.* **6:** 7–39.

KAHN, G. C., BOOBIS, A. R., BRODIE, M. J., TOVERUD, E. L., MURRAY, S. and DAVIES, D. S. (1985) Phenacetin O deethylase: an activity of a cytochrome P450 showing genetic linkage with that catalysing the 4 hydroxylation of debrisoquine. *Br. J. clin. Pharmac.* **20:** 67–76.

KAPPAS, A., ALVARES, A. P., ANDERSON, K. E., PANTUCK, E. J. and PANTUCK, C. B. (1978a) Effect of charcoal broiled beef on antipyrine and theophylline metabolism. *Clin. Pharmac. Ther.* **23:** 445–450.

KAPPAS, A., ANDERSON, K. E., CONNEY, A. H. and ALVERES, A. P. (1978b) Influence of dietary protein and carbohydrate on antipyrine and theophylline metabolism. *Clin. Pharmac. Ther.* **23:** 445–450.

KATER, R. M. H., TOBON, F. and IBER, F. L. (1969) Increased rate of tolbutamide metabolism in alcoholic patients. *JAMA.* **207:** 363–365.

KATO, R., CHIESARA, E. and VASSANELLI, P. (1964) Further studies on the inhibition and stimulation of microsomal drug metabolizing enzymes of rat liver by various compounds. *Biochem. Pharmac.* **13:** 69–83.

KENNY, M. T. and STRATES, B. (1981) Metabolism and pharmacokinetics of the antibiotic rifampicin. *Drug Metab. Rev.* **12:** 159–218.

KOLMODIN, B., AZARNOFF, D. L. and SJOQVIST, F. (1969) Effect of environmental factors on drug metabolism: Decreased plasma half life of antipyrine in workers exposed to hydrocarbon insecticides. *Clin. Pharmac. Ther.* **10:** 638–642.

KRYSZEWSKI, A. J., NEALE, G., WHITFIELD, J. B. and MOSS, D. W. (1973) Enzyme changes in experimental biliary obstruction. *Clinica chim. Acta* **47:** 175–182.

KUNTZMAN, R., JACOBSON, M. and CONNEY, A. H. (1966) Effect of phenylbutazone on cortisol metabolism in man. *Pharmacologist* **8:** 195.

KUTT, H. (1971) Biochemical and genetic factors regulating dilantin metabolism in man. *Ann. N.Y. Acad. Sci.* **179:** 705–722.

KUTT, H. (1984) Interactions between anticonvulsants and other commonly prescribed drugs. *Epilepsia* **25** (Suppl. 2): S118–S131.

KUTT, H., VEREBELY, K. and MCDOWELL, F. (1968) Inhibition of diphenylhydantoin metabolism in rats and in rat liver microsomes by anti tuberculous drugs. *Neurology* **18:** 706–710.

KUTT, H., HAYNES, J., VEREBELY, K. and MCDOWELL, F. (1969) The effect of phenobarbital on plasma diphenylhydantoin level and metabolism in man and in rat liver microsomes. *Neurology* **19:** 611–616.

LABADARIOS, D., DICKERSON, J. W. T., PARKE, D. V., LUCAS, E. G. and OBUWA, G. H. (1978) The effects of chronic drug administration on hepatic enzyme induction and alcohol in man. *Br. J. clin. Pharmac.* **5:** 167–174.

LAHTELA, J. T., SARKKA, P. and SOTANIEMI, E. A. (1984) Phenobarbital treatment enhances insulin mediated glucose metabolism in man. *Res. Commun. Chem. Path. Pharmac.* **44:** 215–216.

LAHTELA, J. T., ARRANTO, A. J. and SOTANIEMI, E. A. (1985) Enzyme inducers improve insulin sensitivity in non insulin dependent diabetic subects. *Diabetes* **34:** 911–916.

LAHTELA, J. T., GACHALYI, B., EKSYMA, S., HAMALAINEN, A. and SOTANIEMI, E. A. (1986) The effect of liver microsomal enzyme inducing and inhibiting drugs on insulin mediated glucose metabolism in man. *Br. J. clin. Pharmac.* **21:** 19–26.

LAI, A. A., LEVY, R. H. and CUTLER, R. E. (1978) Time course of interction between carbamazepine and clonazepam in normal man. *Clin. Pharmac. Ther.* **24:** 316–323.

LANTHAM, A. N., TURNER, P., FRANKLIN, C. and MACLAY, W. (1975) Phenobarbitone induced urinary excretion of D glucaric acid and 6 β hydroxycortisol in man. *Can. J. Physiol. Pharmac.* **54:** 778–782.

LARSEN, P. R., ATKINSON, A. J., WELLMAN, H. N. and GOLDSMITH, R. E. (1970) The effect of diphenylhydantoin on thyroxine metabolism in man. *J. clin. Invest.* **49:** 1266–1279.

LAUTERBURG, B. H. and BIRCHER, J. (1976) Expiratory measurement of maximal aminopyrine demethylation *in vivo*: effect of phenobarbital, partial hepatectomy, portacaval shunt and bile duct ligation in the rat. *J. Pharmac. exp. Ther.* **196:** 501–509.

LAUTERBURG, B. H., TODD, E. L. and MITCHELL, J. R. (1983) Cimetidine protects against isoniazid hepatotoxicity in rats but is not effective in man. *Hepatology* **3:** 880.

LEEMAN, T., DAYER, P. and MEYER, U. A. (1986) Single dose quinidine treatment inhibits metoprolol oxidation in extensive metabolizers. *Eur. J. clin. Pharmac.* **31:** 313–318.

LEGLER, U. F. and BENET, L. Z. (1986) Marked alterations in dose dependent prednisolone kinetics in women taking oral contraceptives. *Clin. Pharmac. Ther.* **39:** 425–429.

LEVINE, M., JONES, M. W. and SHEPPARD, J. (1984) Effect of cimetidine on steady state concentrations of carbamazepine and phenytoin. *Clin. Pharmac. Ther.* **35:** 256.

LEVY, R. H. and KOCH, K. M. (1982) Drug interactions with valproic acid. *Drugs* **24:** 543–556.

LEWIS, R. J., TRAGER, W. F., CHAN, K. K., BRECKENRIDGE, A., ORME, M. L., ROWLAND, M. and SCHARY, W. (1974) Warfarin. Stereochemical aspects of its metabolism and the interaction with phenylbutazone. *J. clin. Invest.* **53:** 1607–1617.

LIEDHOLM, H. and MELANDER, A. (1986) Concomitant food intake can increase the bioavailability of propranolol by transient inhibition of its presystemic primary conjugation. *Clin. Pharmac. Ther.* **40:** 29–36.

LUDDEN, T. M. (1985) Pharmacokinetic interactions of the macrolide antibiotics. *Clin. Pharmacokinet.* **10:** 63–79.

LUKASKIEWICZ, J., PROSZYNSKA, K., LORENC, R. and LUDWIRZAK, H. (1987) Hepatic microsomal enzyme induction, treatment of Vitamin D poisoning in a 7 month old baby. *Br. med. J.* **295:** 1173.

LUOMA, P. V. (1987) Enzyme induction, lipids and apolipoproteins. In: *Enzyme Induction in Man*, pp. 231–242, SOTANIEMI, E. A. and PELKONEN, R. O. (eds) Taylor & Francis, London.

LUOMA, P. V., SOTANIEMI, E. A. and PELKONEN, R. O. (1984) Alcohol HDL cholesterol and liver microsomal induction. *Lancet* **1:** 47.

LUOMA, P. V., SOTANIEMI, E. A., PELKONEN, R. O. and PIRTHIAHO, H. I. (1985) Serum low density lipoprotein and high density lipoprotein cholesterol and liver size in subjects on drugs inducing hepatic microsomal enzymes. *Eur. J. clin. Pharmac.* **28:** 615–618.

MACDONALD, M. G. and ROBINSON, D. S. (1968) Clinical observations of possible barbiturate interference with anticoagulants. *JAMA* **204:** 97–100.

MACKINNON, M., SUTHERLAND, E. and SIMON, F. E. (1977) Effects of ethinyl estradiol on hepatic microsomal proteins and the turnover of cytochrome P 450. *J. Lab. clin. Med.* **90:** 1096–1106.

MACPHEE, G. J. A., THOMPSON, G. G., MCINNES, G. T. and BRODIE, M. J. (1986) Verapamil potentiates carbamazepine neurotoxicity. A clinically important inhibitory interaction. *Lancet* **i:** 700–703.

MACPHEE, G. J. A., MITCHELL, J. R., WISEMAN, L., MCLELLAN, A. R., PARK, B. K., MCINNES, G. T. and BRODIE, M. J. (1988) Effect of sodium valproate on carbamazepine disposition and psychomotor profile in man. *Br. J. clin. Pharmac.* **25:** 59–66.

MAISEY, D. N., BROWN, R. C. and DAY, J. L. (1974) Rifampicin and cortisone replacement therapy. *Lancet* **2:** 896–897.

MANFREDI, R. L. and VESELL, E. S. (1981) Inhibition of theophylline metabolism by long term allopurinol administration. *Clin. Pharmac. Ther.* **29:** 224–229.

MANNERING, G. (1981) Hepatic cytochrome P450-linked drug metabolizing systems. In: *Concepts in Drug Metabolism*, Part B, pp. 53–166, JENNER, P. and TESTA, B. (eds) Marcel Dekker, New York.

MARQUIS, J. F., CARRUTHERS, S. G., SPENCE, J. D., BROWNSTONE, Y. S. and TOOGOOD, J. H. (1982) Phenytoin–theophylline interaction. *New Engl. J. Med.* **307:** 1189–1190.

MARSH, C. A. (1963) Metabolism of D glucuronolactone in mammalian systems. Identification of D glucaric acid as a normal constituent of urine. *Biochem. J.* **86:** 77–86.

MATTHEWS, N. T. and HAVILL, J. H. (1979) Ergotism with therapeutic doses of ergotamine tartrate. *N.Z. Med. J.* **89:** 476–477.

MCINNES, G. T. and BRODIE, M. J. (1988) Drug Interactions that matter. A critical reappraisal. *Drugs* **36:** 83–110.

MELANDER, A. (1978) Influence of food on the bioavailability of drugs. *Clin. Pharmacokinet.* **3:** 337–351.

MENDOZA, D. M., FLOCK, E. V., OWEN, C. A. and PARIS, J. (1966) Effect of 55 diphenylhydantoin on the metabolism of L thyroxine ^{131}I in the rat. *Endocrinology* **79:** 106–108.

MIGUET, J. P., MAVIER, P., SOUSSY, C. J. and DHUMEAUX, D. (1977) Induction of hepatic microsomal enzymes after brief administration of rifampicin in man. *Gastroenterology* **72:** 924–926.

MILLER, R. R., PORTER, J. and GREENBLATT, D. J. (1979) Clinical importance of the interaction of phenytoin and isoniazid, a report from the Boston Collaborative Drug Surveillance Program. *Chest* **75:** 356–358.

MINERS, J. O., GRYGIEL, J. J., DREW, R. and BIRKETT, D. J. (1981) Interaction between cimetidine and theophylline in smokers and non smokers. *Clin. exp. Physiol. Pharmac.* **8:** 633–634.

MINERS, J. O., FOENANDER, T., WANWIMOLEUK, S., GALLUS, A. S. and BIRKETT, D. J. (1982a) The effect of sulphinpyrazone on oxidative drug metabolism in man, inhibition of tolbutamide elimination. *Eur. J. clin. Pharmac.* **22:** 321–326.

MINERS, J. O., FOENANDER, T., WANWIMOLEUK, S., GALLUS, A. S. and BIRKETT, D. J. (1982b) Interaction of sulphinpyrazone with warfarin. *Eur. J. clin. Pharmac.* **22:** 327–331.

MINERS, J. O., ROBSON, R. A. and BIRKETT, D. J. (1986) Induction of glucuronidation and glycine conjugation in oral contraceptive steroid users. III World Conference on Clinical Pharmacology and Therapeutics, Stockholm, Abstr. (I), p. 207.

MITCHELL, J. R., THORGEIRSSON, S. S., POTTER, W. Z., JALLOW, D. J. and KEISER, H. (1974) Acetaminophen-induced hepatic injury: protective role of glutathione in man and rationale for therapy. *Clin. Pharmac. Ther.* **16:** 676–782.

MITCHELL, M. C., SCHENKER, S., AVANT, G. R. and SPEEG, K. V. (1981) Cimetidine protects against acetaminophen hepatotoxicity in rats. *Gastroenterology* **81:** 1052–1060.

MORELAND, T. A., PARK, B. K. and RYLANCE, G. W. (1981) Drug metabolizing enzyme induction in children. *Br. J. clin. Pharmac.* **11:** 420P.

MEDICAL RESEARCH COUNCIL WORKING PARTY (1985) MRC trial of treatment of mild hypertension: Principal results. *Br. med. J.* **291:** 97–104.

MUAKKASSAH, S. F. and YANG, W. C. T. (1981) Mechanism of the inhibitory action of phenalzone on microsomal drug metabolism. *J. Pharmac. exp. Ther.* **219:** 147–155.

MUURONEN, A., KASTE, M., WIKKILA, E. A. and TOLPPANEN, E. M. (1985) Mortality from ischaemic heart disease among patients using anticonvulsive drugs: a case control study. *Br. med. J.* **291:** 1481–1483.

NELSON, J. and TATLOW, P. (1980) Neuroleptic effect on desipramine steady state plasma concentrations. *Am. J. Psychiat.* **137:** 1232–1234.

NEUVONEN, P. J. and PENTILLA, O. (1974) Interaction between doxycycline and barbiturates. *Br. med. J.* **1:** 535–536.

OHNHAUS, E. E. and PARK, B. K. (1979) Measurement of urinary 6β hydroxycortisol excretion as an *in vivo* parameter in the clinical assessment of the microsomal enzyme inducing capacity of antipyrine, phenobarbitone and rifampicin. *Eur. J. clin. Pharmac.* **15:** 139–145.

ONHNHAUS, E. E., KIRCKHOF, B. and PEHEIM, E. (1979a) Effect of enzyme induction on plasma lipids using antipyrine phenobarbital and rifampicin. *Clin. Pharmac. Ther.* **25:** 591–517.

OHNHAUS, E. E., PARK, B. K., COLUMBO, R. P. and HEIZMANN, P. (1979b) The effect of enzyme induction on diazepam metabolism in man. *Br. J. clin. Pharmac.* **8:** 557–564.

OHNHAUS, E. E., BURGI, H., BURGER, A. and STUDER, H. (1981) The effect of antipyrine, phenobarbital and rifampicin on thyroid hormone metabolism in man. *Eur. J. clin. Invest.* **11:** 381–387.

OHNHAUS, E. E., BROCKMEYER, N. and DYLEWICZ HABICHT, H. (1987) The effect of antipyrine and rifampicin on the metabolism of diazepam. *Clin. Pharmac. Ther.* **42:** 148–156.

O'MALLEY, K., BROWNING, M., STEVENSON, I. H. and TURNBULL, M. J. (1973a) Stimulation of drug metabolism in man by tricyclic antidepressants. *Eur. J. Clin. Pharmac.* **6:** 102–106.

O'MALLEY, K., SAWYER, P. R. and STEVENSON, I. H. (1973b) Effects of tricyclic antidepressants on drug metabolism. *Br. J. Pharmac.* **44:** 372–373P.

OPPENHEIMER, J. H., BERNSTEIN, G. and SURKS, M. I. (1968) Increased thyroxine turnover and thyroid function after stimulation of hepatocellular binding of thyroxine by phenobarbital. *J. clin. Invest.* **47:** 1399–1406.

O'REILLY, R. A. (1974) Interactions of sodium warfarin and rifampicin studies in man. *Ann. int. Med.* **81:** 337–340.

O'REILLY, R. A. (1976) The stereoselective interaction of warfarin and metronidazole in man. *New Engl. J. Med.* **295:** 345–357.

O'REILLY, R. A. (1980) Stereoselective interaction of trimethoprim sulphamethoxazole with the separated enantiomorphs of racemic warfarin in men. *New Engl. J. Med.* **302:** 33–35.

ORULOWSKI, M. (1963) The role of gamma glutamyl transpeptidase in the internal disease clinic. *Archivm Immunoliae Ther. Exp.* **11:** 1–61.

PANTUCK, E. J., HOIAO, K. C., MAGGIO, A., WAKAMURA, K., KUNTZMAN, R. and CONNEY, A. H. (1974) Effect of smoking on phenacetin metabolism. *Clin. Pharmac. Ther.* **15:** 9–16.

PARK, B. K. (1981) Assessment of urinary 6 β hydroxycortisol as an *in vivo* index of mixed function oxygenase activity. *Br. J. clin. Pharmac.* **12:** 97–102.

PARK, B. K. and BRECKENRIDGE, A. M. (1981) Clinical implications of enzyme induction and enzyme inhibition. *Clin. Pharmacokinet.* **6:** 1–24.

PARK, B. K., EICHELBAUM, M. and OHNHAUS, E. E. (1982) 6 β hydroxycortisol excretion in relation to polymorphic *N* oxidation of sparteine. *Br. J. clin. Pharmac.* **13:** 737–740.

PASANISI, F., ELLIOTT, H. L., MEREDITH, P. A., MCSHARRY, D. R. and REID, J. L. (1984) Combined alpha adrenoceptor antagonism and calcium channel blockade in normal subjects. *Clin. Pharmac. Ther.* **36:** 716–723.

PATSALOS, P. N., DUNCAN, J. S. and SHORVON, S. D. (1988) Effect of the removal of individual antiepileptic drugs on antipyrine kinetics in patients taking polytherapy. *Br. J. clin. Pharmac.* **26:** 253–259.

PATWARDHAN, R., JOHNSON, R., SINCLAIR, A., SCHENKER, S. and SPEEG, K. V. (1981) Lack of tolerance and rapid recovery of cimetidine inhibited chlordiazepoxide (Librium) elimination in man. *Gastroenterology* **81:** 547–551.

PEDEN, N. R., REWHORN, I., CHAMPION, M. C., MUSSANI, R. and OOI, T. C. (1984) Cortisol and dexamethasone elimination during treatment with cimetidine. *Br. J. clin. Pharmac.* **18**(1): 101–103.

PEET, M., MIDDLEMISS, D. N. and YATES, R. A. (1980) Pharmacokinetic interaction between propranolol and chlorpromazine in schizophrenic patients. *Lancet* **2:** 978.

PELKONEN, O., PASANEN, M., KUHA, H., GACHALYI, B., KAIRALUOMA, M., SOTANEMI, E. A., PARK, S. S., FRIEDMAN, K. F. and GELBOIN, H. V. (1986) The effect of cigarette smoking on 7 ethoxyresorufin *O* deethylase and other monooxygenase activities in human liver. *Br. J. clin. Pharmac.* **22:** 125–130.

PENTTILA, O., NEUVONEN, P. J., AHO, K. and LEHTAVAARA, R. (1974) Interaction between doxycycline and antiepileptic drugs. *Br. med. J.* **2:** 470–472.

PERUCCA, E., RUPRAH, M., RICHENS, A., PARK, B. K., BETTERBRIDGE, D. J. and HEDGES, A. (1981) Effect of low dose phenobarbitone on five indirect indices of hepatic microsomal enzyme induction and plasma lipoproteins in normal subjects. *Br. J. clin. Pharmac.* **12:** 592–596.

PERUCCA, E., HEDGES, A., MAKKI, K. A., RIPRAH, M., WILSON, J. F. and RICHENS, A. (1984) A comparative study of the relative enzyme inducing properties of anticonvulsant drugs in epileptic patients. *Br. J. clin. Pharmac.* **18:** 401–410.

PESSAYRE, D., DESCATOIRE, V., KANSTANTINOVA-MITCHEVA, M., WANDSCHEER, J. C., COBERT, B., LEVEL, R., BENHAMOU, P. J., JAOUEN, M. and MANSUY, D. (1981) Self-induction by triacetyloleandomycin of its own transformation into a metabolite forming a stable 456 nonabsorbing complex with cytochrome P 450. *Biochem. Pharmac.* **30:** 553–538.

PETERS, U., HAUSEMAN, T. U. and CROSSE-BROCKHOFT, E. (1974) Einfluss van Tuberkulostatcka auf die Pharmakokinetikdes Digitoxins. *Deutsche med. Wschr.* **99:** 2381–2386.

PETRUCH, F., SCHUPPEL, R. V. A. and STEINHILBER, G. (1974) Effect of diphenylhydantoin on hepatic drug hydroxylation. *Eur. J. clin. Pharmac.* **7:** 281–285.

PIRTTIAHO, H. I., SOTANIEMI, E. A., AHOKAS, J. T. and PITKANEN, U. (1978) Liver size and indices of drug metabolism in epileptics. *Br. J. clin. Pharmac.* **6:** 273–278.

PIRTTIAHO, H. I., SOTANIEMI, E. A., PELKONEN, R. O. and PITKANEN, U. (1982) Hepatic blood flow and drug metabolism in patients on enzyme inducing anticonvulsants. *Eur. J. clin. Pharmac.* **22:** 441–445.

POLAND, A., SMITH, M. D., KUTNZMAN, R., JACOBSON, M. and CONNEY, A. H. (1970) Effect of intensive occupational exposure to DDT or phenylbutazone and cortisol metabolism in human subjects. *Clin. Pharmac. Ther.* **ii:** 724–729.

PRICE, D. E., MEHTA, A., PARK, B. K., HAY, A. and FEELY, M. P. (1986) The effect of low dose phenobarbitone on three indices of hepatic microsomal enzyme induction. *Br. J. clin. Pharmac.* **22:** 744–747.

PUURUNEN, J., SOTANIEMI, E. and PELKONEN, O. (1980) Effect of cimetidine on microsomal drug metabolism in man. *Eur. J. clin. Pharmac.* **18:** 185–187.

REILLY, P. E., CARRINGTON, L. E. and WINZOR, D. J. (1983) The interaction of cimetidine with rat liver microsomes. *Biochem. Pharmac.* **32:** 831–835.

ROBSON, R. A., MINERS, J. O., WHITEHEAD, A. G. and BIRKETT, D. G. (1987) Specificity of the inhibitory effect of dextropropoxyphene on oxidative drug metabolism in man: effects on theophylline and tolbutamide disposition. *Br. J. clin. Pharmac.* **23:** 772–775.

ROBSON, R. A., FRAENKEL, M., BARRATT, L. J. and BIRKETT, D. J. (1988) Cyclosporin–verapamil interaction. *Br. J. clin. Pharmac.* **25:** 402–403.

ROOTS, I., LEY, B. and HINDEBRANDT, A. G. (1977) *In vivo* parameters of drug metabolism—differences in specificity towards inducing agents. In: *Microsomes and Drug Oxidations*, pp. 581–588, ULLRICH, V., ROOTS, I., HILDEBRANDT, A. G., ESTABROOKE, R. W. and CONNEY, A. H. (eds) Pergamon Press, Oxford.

ROOTS, I., HOLBE, L., HOVERMANN, W., NIGAM, S., HEINEMEYER, G. and HILDEBRANDT, A. G. (1979) Quantitative determination by HPLC of urinary 6 β hydroxycortisol an indicator of enzyme induction by rifampicin and antieptileptic drugs. *Eur. J. clin. Pharmac.* **16:** 63–71.

ROSALKI, S. B. and RAU, D. (1972) Serum gamma glutamyl transpeptidase activity in alcoholism. *Clinica chim. Acta* **39:** 41–47.

ROSALKI, S. B., TARLOW, D. and RAU, D. (1971) Plasma gamma glutamyl transpeptidase elevation in patients receiving inducing drugs. *Lancet* **ii:** 376–377.

ROWLAND, M. (1975) Kinetics of drug–drug interactions. In: *Pharmacology and Pharmacokinetics*, pp. 321–386, TEORELL, T., DEDRICK, R. L. and CANDLIFFE, P. G. (eds) Plenum Press, London.

RUBIN, A., TEPHLY, T. R. and MANNERING, G. J. (1964) Kinetics of drug metabolism by hepatic microsomes. *Biochem. Pharmac.* **13:** 1007–1016.

RUBIN, E., GANG, H., MISRA, P. S. and LIEBER, C. S. (1970) Inhibition of drug metabolism by acute ethanol intoxication. *Am. J. Med.* **49:** 801–806.

RUMIANTSEV, D. O., PIOTROVSKII, V. R., RIABOKON, O. S., SLASTNIKOVA, I. D., KOKURINA, E. V. and METELITSA, V. I. (1966) The effect of oral verapamil therapy on antipyrine clearance. *Br. J. clin. Pharmac.* **22:** 606–609.

SAAL, A. K., WERNER, J. A., GREENE, H. L., SEARS, G. K. and GRAHAM, E. L. (1984) Effect of Amiodarone on serum quinidine and procainamide levels. *Am. J. Cardiol.* **53:** 1264–1267.

SAENGER, P., RIFKIND, A. B. and NEW, M. I. (1976) Changes in drug metabolism in children with thyroid disorders. *J. clin. Endocr. Metab.* **42:** 155–159.

SAGGERS, V. H., HARIRATNAJOTHI, N. and MCCLEAN, A. E. M. (1970) The effect of diet and phenobarbitone on Quinine metabolism in the rat and in man. *Biochem. Pharmac.* **19:** 499–503.

SASAME, H. A. and GILETTE, J. R. (1970) Studies on the inhibitory effects of various substances on drug metabolism by liver microsomes. The effect of nicotinamide in altering the apparent mechanism of inhibition. *Biochem. Pharmac.* **19:** 1025–1042.

SCHELLENS, J. H. M. and BREIMER, D. D. (1987) Antipyrine metabolite formation in man. Influence of induction and application as a tool in cytochrome P-450 characterization. In: *Enzyme Induction in Man*, pp. 109–123, SOTANIEMI, E. A. and PELKONEN, R. O. (eds) Taylor and Francis, London.

SCHULTE-HERMANN, R. (1974) Induction of liver growth by xenobiotic compounds and other stimuli. *CRC Crit. Rev. Toxic.* **3:** 97–158.

SCHWARTZ, J. B., KEEFE, D. L., KURSTEN, E., KATES, R. E. and HARRISON, D. C. (1982) Prolongation of verapamil elimination kinetics during chronic oral administration. *Am. Heart J.* **104:** 198–203.

SCOTT, A. K., JEFFERS, T. A., PETRIE, J. C. and GILBERT, J. C. (1979) Serum bilirubin and hepatic enzyme induction. *Br. med. J.* **2:** 310.

SERLIN, M., SIBEON, R. G., MOSSMAN, S., BRECKENRIDGE, A. M., WILLIAMS, J. R. B., ATWOOD, J. L. and WILLOUGHBY, J. M. T. (1979) Cimetidine: interaction with oral anticoagulants in man. *Lancet* **ii:** 317–319.

SESARDIC, D., BOOBIS, A. R., EDWARDS, R. J. and DAVIES, D. S. (1988) A form of cytochrome P450 in man, orthologous to form d in the rat, catalyses the *O* deethylation of phenacetin and is inducible by cigarette smoking. *Br. J. clin. Pharmac.* **26:** 363–372.

SHAND, D. G., HAMMILL, S. C., AANONSEN, L. and PRITCHETT, E. L. C. (1981) Reduced verapamil clearance during long term oral administration. *Clin. Pharmac. Ther.* **30:** 701–703.

SHAW, P. N., HOUSTON, J. B., ROWLAND, M. and HOPKINSK THIERCELIN, J. F. (1985) Antipyrine metabolite kinetics in healthy human volunteers during multiple dosing of phenytoin and carbamazepine. *Br. J. clin. Pharmac.* **20:** 611–618.

SMITH, S. R. and KENDALL, M. J. (1988) Ranitidine versus cimetidine. A comparison of their potential to cause clinically important drug interactions. *Clin. Pharmacokinet.* **15:** 44–56.

SOLOMAN, H. M. and ABRAMS, W. B. (1972) Interaction between digitoxin and other drugs in man. *Am. Heart J.* **83:** 277–280.

SOMOGYI, A. and GUGLER, R. (1972) Drug interactions with cimetidine. *Clin. Pharmacokinet.* **7:** 23–41.

SOTANIEMI, E. A., ARRANTO, A. J., SUTINEN, S., STENGARD, J. H. and SUTINEN, S. (1983) Treatment of non-insulin dependent diabetes mellitus with enzyme inducers. *Clin. Pharmac. Ther.* **33:** 826–835.

SOUTHERN, A. L., GORDON, G. G., TOCHIMATO, S., KRIKUN, E., KREIGER, D., JACKSON, M. and KUNTZMAN, R. (1969) Effect of *N*-phenylbarbital (Phetharbital) on the

metabolism of testosterone and cortisol in man. *J. clin. Endocr.* **29:** 251–256.

STENGARD, J. H., SAURNI, H. U. and SOTANIEMI, E. A. (1984) Effects of medroxyprogesterone acetate on hepatic glucose metabolism and microsomal enzyme activity in rats with normal and altered liver. *Pharmacology* **28:** 34–41.

STEVENSON, I. H. (1977) Factors influencing antipyrine elimination. *Br. J. Pharmac.* **4:** 261–265.

SYVALAHTI, E. K. G., LINDBERG, R., KALLIO, J. and DE VOCHT, M. (1986) Inhibitory effects of neuroleptics on debrisoquine oxidation in man. *Br. J. clin. Pharmac.* **22:** 89–92.

SZEFLER, S. J., BRENNER, M., JUSKO, W. J., SPECTOR, S. L., FLESHER, K. A. and ELLIS, E. F. (1982) Dose and time related effect of troleandomycin on methylprednisolone elimination. *Clin. Pharmac. Ther.* **32:** 166–171.

TAZI, A., GALTEAU, M. and SIEST, G. (1986a) γ-Glutamyltransferase of rabbit liver: kinetic study of phenobarbital induction and *in vitro* solubilization by bile salts. *Toxic. appl. Pharmac.* **55:** 1–7.

TAZI, A., GALEAU, M. and SIEST, G. (1980b) Effect of imipramine on hepatic gamma glutamyl transferase in female rats. Interactions with contraceptives. *Biochem. Pharmac.* **29:** 2874–2876.

TEUNISSEN, M. W. E., BAKKER, W., MEERBURG, VAN DE, TORREN, J. E. and BREIMER, D. D. (1984) Influence of rifampicin treatment on antipyrine clearance and metabolite formation in patients with tuberculosis. *Br. J. clin. Pharmac.* **18:** 701–706.

TEUNISSEN, M. W. E., DE LEEDE, L. G. J., BOEIJINGA, J. K. and BREIMER, D. D. (1985) Correlation between antipyrine metabolite formation and theophylline metabolism in humans after simultaneous single dose administration and at steady state. *J. Pharmac. exp. Ther.* **233:** 770–775.

THOMAS, R. C. and IKEDA, C. J. (1966) The metabolic fate of tolbutamide in man and in the rat. *J. med. Chem.* **9:** 507–510.

TOLLEFSON, G. D. (1983) Monoamine oxidase inhibitors: a review. *J. clin. Psych.* **44:** 280–286.

VAINIO, H. and HIETANEN, E. (1980) Role of extrahepatic metabolism. In: *Concepts in Drug Metabolism*, Part A, pp. 251–284, JENNER, P. and TESTA, B. (eds) Marcel Dekker, New York.

VAN MARLE, W., WOODS, K. L. and BEELEY, L. (1979) Concurrent steroid and rifampicin therapy. *Br. med. J.* **1:** 1020(c).

VAUGHT, J. B. and KING, C. M. (1984) Carcinogenesis in bladder cancer. In: *Bladder Carcinoma*, pp. 32–45, SMITH, P. H. and PROUT, G. R. (eds) Butterworths & Co., London.

VESELL, E. S. (1979) The antipyrine test in clinical pharmacology: Conceptions and misconceptions. *Clin. Pharmac. Ther.* **26:** 275–286.

VESELL, E. S. and PAGE, J. G. (1969) Genetic control of the phenobarbital induced shortening of plasma antipyrine half-lives in man. *J. clin. Invest.* **48:** 2202–2209.

VESELL, E. S., PASSANANTI, G. T. and GREENE, F. E. (1970) Inhibition of drug metabolism in man by allopurinol and nortriptyline. *New Engl. J. Med.* **283:** 1484–1488.

VESELL, E. S., NA, L., PASSANANTI, G. T. and CHASE, T. N. (1971a) Inhibition of drug metabolism by levodopa in combination with a dopa decarboxylase inhibitor. *Lancet* **ii:** 370.

VESELL, E. S., PAGE, J. G. and PASSANANTI, G. T. (1971b) Genetic and environmental factors affecting ethanol metabolism. *Clin. Pharmac. Ther.* **12:** 192–201.

VESELL, E. S., PASSANATI, G. T. and GLENWRIGHT, P. A. (1975) Anomalous results of studies on drug interaction in man. III. Disulfiram and antipyrine. *Pharmacology* **13:** 481–491.

VESELL, E. S., PASSANANTI, G. T. and HEPNER, G. W. (1976) Interaction between antipyrine and aminopyrine. *Clin. Pharmac. Ther.* **20:** 661–669.

VESTAL, R. E., NORRIS, A. H., TOBIN, J. D., COHEN, B. H., SCHOCK, N. W. and ANDRES, R. (1975) Antipyrine metabolism in man. Influence of age alcohol caffeine and smoking. *Clin. Pharmac. Ther.* **18:** 425–432.

VESTAL, R. E., KORNHAUSER, D. M., HOLLIFIELD, J. W. and SHAND, D. G. (1979) Inhibition of propranolol metabolism by chlorpromazine. *Clin. Pharmac. Ther.* **25:** 19–24.

VESTAL, R. E., THUMMEL, K. C. and MUSSER, B. (1983) Cimetidine inhibits theophylline clearance in patients with chronic obstructive pulmonary disease. A study using stable isotope methodology during multiple oral dose administration. *Br. J. clin. Pharmac.* **15:** 411–418.

VIALA, A., CANO, J. P., DRANET, C., TASSINARAI, C. A. and ROGER, J. (1971) Blood levels of diazepam (Valium) and *N* desmethyl diazepam in the epileptic child. A preliminary report. *Psychiat. Neurol. Neurosci.* **74:** 153–158.

VIDELA, L., BERNSTEIN, J. and ISRAEL. Y. (1973) Metabolic alterations produced in the liver by chronic ethanol administration. *Biochem. J.* **134:** 507–514.

VINCENT, F. M. (1980) Phenothiazine induced phenytoin intoxication. *Ann. intern. Med.* **93:** 56–57.

WANG, P. P., BEAUNE, P., KAMINSKY, L. S., DANNAN, G. A., KADLUBAR, F. F., LAMEY, D. and GUENGERICH, F. P. (1983) Purification and characterization of six cytochrome P450 isozymes from human liver microsomes. *Biochemistry* **22:** 5375–5383.

WEINBURGER, M., HUDGEL, D., SPECTOR, S. and CHIDSEY, C. (1977) Inhibition of theophylline clearance by troleandomycin. *J. All. clin. Immun.* **59:** 228–231.

WERK, E. E., MCGEE, J. and SHOLITON, L. J. (1964) Effect of diphenylhydantoin on cortisol metabolism in man. *J. clin. Invest.* **43:** 1824–1835.

WERK, E. E., CHOI, Y., SHOLITON, L. J., OLINGER, C. and HAQUE, N. (1969) Interference in the effect of dexamethasone by diphenylhydantoin. *New Engl. J. Med.* **281:** 42–34.

WHITFIELD, J. B., MOSS, D. W., NEALE, G., ORME, M. and BRECKENRIDGE, A. (1973) Changes in plasma by glutamyl transpeptidase activity associated with alterations in drug metabolism in man. *Br. med. J.* **1:** 316–318.

WIJANDS, W. J. A., VREE, T. B. and VAN HERWAARDEN, C. L. A. (1985) Enoxacin decreases the clearance of theophylline in man. *Br. J. clin. Pharmac.* **20:** 583–588.

WIJANDS, W. J. A., VREE, T. B. and VAN HERWAARDEN, C. L. A. (1986) The influence of quinolone derivatives on theophylline clearance. *Br. J. clin. Pharmac.* **22:** 677–683.

WILKINSON, G. R. and SHAND, D. G. (1975) A physiological approach to hepatic drug clearance. *Clin. exp. Ther.* **18:** 377–390.

WOOD, A. J. J., CARR, K., VESTAL, R. E., BELCHER, S., WILKINSON, G. R. and SHAND, D. G. (1978) Direct measurement of propranolol bioavailability during accumulation to steady state. *Br. J. clin. Pharmac.* **6:** 345–350.

WOOD, A. J. J., VESTAL, R. E., WILKINSON, G. R., BRANCH, R. A. and SHAND, D. G. (1979) Effect of aging and cigarette smoking on antipyrine clearance and indocyanine green elimination. *Clin. Pharmac. Ther.* **26:** 16–20.

WOOLF, T. F. and JORDAN, R. A. (1987) Basic Concepts in Drug Metabolism: Part I. *J. clin. Pharmac.* **27:** 15–17.

WORDLIE, R. C. (1979) Multifunctional glucose 6 phosphatase. Cellular biology. *Life Sci.* **24:** 2397–2404.

WRIGHT, J. M., STOKES, E. F. and SWEENEY, V. P. (1982) Isoniazid induced carbamazepine toxicity and vice versa a double drug interaction. *New Engl. J. Med.* **307:** 1325–1327.

WROBLEWSKI, B. A., SINGER, W. D. and WHYTE, J. (1986) Carbamazepine–erythromycin interaction: case studies and clinical significance. *JAMA* **255:** 1165–1167.

ZILLEY, W., BREIMER, D. D. and RICHTER, E. (1975) Induction of drug metabolism in man after rifampicin treatment measured by increased hexobarbital and tolbutamide clearance. *Eur. J. clin. Pharmac.* **9:** 219–227.