Influence of CYP2D6 Genetics on Opioid Kinetics, Metabolism and Response

Gerd Mikus* and Johanna Weiss

Department of Internal Medicine VI, Clinical Pharmacology and Pharmacoepidemiology, University of Heidelberg, Germany

Abstract: Pharmacogenetics does seem to play a key role in the use of so-called weak opioids. It has been shown for codeine, dihydrocodeine, oxycodone and hydrocodone, that their O-demethylation in the 3-position results in metabolites which have much stronger µ-receptor binding. These opioids may therefore exert their pharmacological actions predominantly through their O-demethylated metabolites. However, this metabolic step is under genetic control of the polymorphic cytochrome P450 2D6 isozyme (CYP2D6). Poor metabolisers of CYP2D6 (~10% of the Caucasian population) do not express this enzyme and hence can only form trace amounts of the O-demethylated metabolites of these four opioids. This might put these persons on risk of reduced or even abolished analgesic effects when given these weak opioids. From this point of view there are two major issues why weak opioids cannot wholeheartedly be recommended: large interindividual variability of the analgesic effect due to CYP2D6 polymorphism and 10% of patients with no benefit from these drugs. On the other hand it might be advantageous to use the O-demethylated metabolites morphine, oxymorphone and hydromorphone which are all strong opioids and have a smaller interindividual variability of the opioid effects. Instead of using weak opioids, small doses and controlled release formulations of strong opioids might be the future way to in analgesic therapy despite the fear of addiction and bureaucratic efforts involved with these compounds.

INTRODUCTION

Individual variation in drug response is a substantial clinical problem. The concentration at the target organ or receptor is the fundamental determinant of the drug effect. Normally this concentration is dependent on the dose of the drug (linear kinetics). Despite the same administered dose of a drug the concentration at the target organ often shows considerable differences which in consequence results in a wide variability of the drug effect. These differences can be caused by acquired or inherited variability of absorption, distribution, metabolism, and excretion (ADME) of a drug. Over the last decade the variability of drug metabolising enzymes was very much been focussed on because the large interindividual genetically determined differences in pharmacokinetics (multiplication of plasma concentrations and half-life) have been described.

VARIABILITY OF DRUG METABOLISING ENZYMES

At the level of drug metabolising enzymes molecular alterations such as gene deletion, gene duplication or single nucleotide polymorphisms (SNP) can occur which result in genetic variability in drug response. Numerous genetic polymorphisms in drug metabolising enzymes have been reported and the best characterised are those in the cytochrome P450 (CYP) superfamily. Although CYP3A4 is probably the most important isozyme of the cytochromes due to the large number of drugs being metabolised, CYP2D6 is much more interesting in terms of genetic variability.

CYP2D6 POLYMORPHISM

The *CYP2D6* gene is highly polymorphic with over 70 known alleles identified at the *CYP2D* locus on chromosome 22q13. At least 15 of these alleles encode nonfunctional gene products as a result of single nucleotide polymorphisms (SNPs), gene deletion, aberrant splicing or premature translation termination [Daly, 1996; Garte and Crosti, 1999; Meyer and Zanger, 1997]. Carriers of two nonfunctional alleles reveal a severely impaired metabolism of CYP2D6 substrates and are referred to as poor metabolisers (PMs). In contrast, individuals with at least one functional allele and thus normal CYP2D6 activity are called extensive metabolisers (EMs). Among Caucasians, 5-10% are PMs and further 10-15% show impaired yet residual activity of CYP2D6, the so called intermediate metabolisers (IMs). The genetic cause for the IMs is not totally understood, but over 60% in Caucasians seems to be attributed to the C-1496G polymorphism in the 5'-flanking region of the *CYP2D6* gene [Raimundo, 2000]. Aside from this polymorphism there are others linked to the IM phenotype, e.g. the CYP2D6*10 allele [Ramamoorthy, 2001], which has a prevalence of about 50% in Asians [Bradford, 2002; Ji, 2002; Johansson, 1994] and the CYP2D6*17 allele, which is frequent among Africans, but rare in Caucasians [Bradford, 2002; Masimirembwa, 1996; Wennerholm, 1999]. 1-5% of the Caucasian population has a duplication or multiduplication of the *CYP2D6* gene [Johansson, 1993; Lundqvist, 1999] leading to the phenotype of ultra rapid metabolisers (UMs).

*Address correspondence to this author at the Department of Internal Medicine VI, Clinical Pharmacology and Pharmacoepidemiology, University of Heidelberg, Im Neuenheimer Feld 410, D-69120 Heidelberg, Germany; Tel: + 49 6221 56 39197; Fax: + 49 6221 56 4642;
E-mail: gerd_mikus@med.uni-heidelberg.de

1570-1603/05 $50.00+.00 ©2005 Bentham Science Publishers Ltd.

Genotyping

Genotyping provides direct information of an individual's genetic information, is less invasive than phenotyping and is not influenced by concurrent drug administration, alterations in hormonal levels and disease states [Ensom, 2001]. It is, however, limited by the lack of functional significance of many of the specific genotypes.

In Caucasians CYP2D6*3, *4, *5, and *6 are responsible for over 93% of the PM phenotype, together with *7 and *8 even for approximately 99% [Stuven, 1996]. Therefore, genotyping is often restricted to these four to six alleles. Several polymerase chain reaction (PCR) based methods have been developed including allele specific PCR, PCR-restriction fragment length polymorphism (PCR-RFLP), multiplex PCR and LightCycler methods [Heim and Meyer, 1991; Hersberger, 2000; Muller, 2003; Stamer, 2002; Stuven, 1996]. UMs and IMs as well can be detected by PCR methods [Ji, 2002; Johansson, 1993; Lovlie, 1996; Muller, 2003; Skoda, 1988]or by sequencing [Raimundo, 2000].

Phenotyping

Phenotyping requires the intake of a probe drug, which metabolism is solely dependent on CYP2D6. The excretion of parent compound and/or metabolite in urine allows to calculate the metabolic ratio as a measure of the individual CYP2D6 activity. Several probe drugs have been used for phenotyping including sparteine, debrisoquine, and dextrometorphan [Streetman, 2000], as well as metoprolol [Lennard, 1982] and codeine [Yue, 1989]. Although phenotyping can lead to incorrect results due to coadministration of CYP2D6 inhibitors [LLerena, 1993; Madsen, 1995], to confounding effects of disease [Caporaso, 1992; Rost, 1995], or to other factors like hormonal levels [Ensom, 2001], it is the only possibility to evaluate the enzyme function and to detect variations in the enzyme activity due to posttranslational variations or defects in the overall process of drug metabolism [Linder, 1997].

OPIOIDS

Opioids are the most powerful analgesic drugs, showing wide variability in opioid response among patients. This can partly be attributed to differences in the pharmacogenetics of opioid receptors, G proteins and neuroplasticity due to pain stimulus which influence opioid responses.

Alkaloids of Papaver Somniferum L.

Since the isolation of morphine in 1803 by Sertürner more than 40 alkaloids have been isolated from *Papaver somniferum L.* and tested for their pharmacological activities [Lindner, 1985] thereby the most important ones are morphine, codeine, thebaine, papaverine and noscapine (Fig. (**1**)). Thebaine is largely used as a precursor for the synthesis of semisynthetic opioids. For 2003 the world total estimates of requirements are: codeine 376 000 kg, morphine 285 000 kg, thebaine 80 000 kg, hydrocodone 42 000 kg, dihydrocodeine and oxycodone 37 000 kg each [International Narcotics Control Board, 2003].

Codeine

Codeine was first isolated from *Papaver somniferum L.* in 1832 and has been used since then as an analgesic, antitussive and antidiarrhoeal agent [Eddy, 1968]. Meanwhile codeine is regarded as a prodrug at least for its analgesic effect [Gutstein and Akil, 2001], therefore an understanding of its metabolism is crucial to its mode of action.

Metabolism

Codeine is metabolised to three primary metabolites (Fig. (**2**)). The major metabolic step is a glucuronidation at the 6-position to yield codeine-6-glucuronide. The enzyme responsible for this is UGT2B7 [Coffman, 1997]. Two oxidative pathways of codeine metabolism are mediated by the cytochrome P450 enzyme system namely O-demethylation to morphine by CYP2D6 [Dayer, 1988; Mikus, 1991] and N-demethylation to norcodeine by

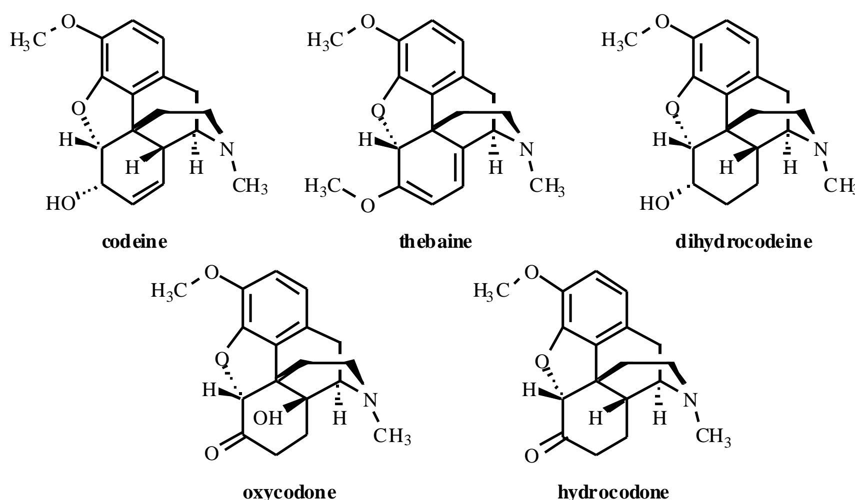

Fig. (1). Chemical structures of natural and semisynthetic opioids.

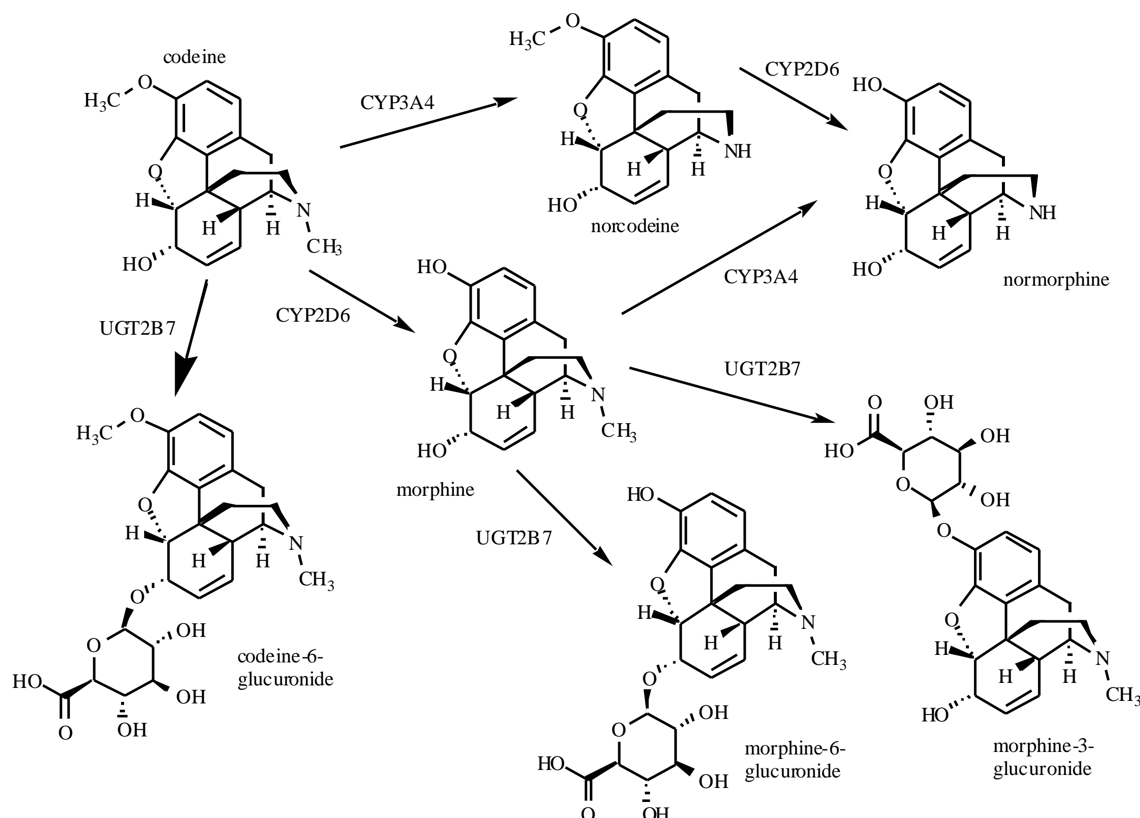

Fig. (2). Chemical structures and metabolic pathways of codeine and enzymes contributing to the metabolism.

CYP3A4 [Caraco, 1996b]. Codeine-6-glucuronide is not been further metabolised in contrast to morphine and norcodeine which can both be glucuronidated to morphine-3- and –6-glucuronide and norcodeine-6-glucuronide. Oxidative metabolism of morphine and norcodeine where also CYP2D6 and CYP3A4 (and CYP2C8) are involved results in a common metabolite normorphine [Projean, 2003; Xu, 1997], which then is further metabolised to the 3- and 6-glucuronide.

Pharmacokinetics

Due to the polymorphic codeine O-demethylation to form morphine the pharmacokinetics of codeine and its metabolites has been studied extensively during the past decades. Because of the small contribution of the O-demethylation to the overall metabolism of codeine the pharmacokinetics of codeine itself is independent of the CYP2D6 phenotype [Caraco, 1999; Chen, 1991b; Eckhardt, 1998; Mikus, 1997; Yue, 1991]. Terminal elimination half-life ranges between 2 and 4 h and the apparent oral clearance between 1000 and 2000 ml/min. Renal clearance of the parent drug is only a minor pathway of elimination (50 – 100 ml/min), metabolism to codeine-6-glucuronide accounts to ~1000 ml/min being the major elimination pathway of codeine. Partial metabolic clearances to norcodeine and morphine are similar in EM subjects (100 – 200 ml/min) [Ammon, 2002], whereas in PM subjects the partial metabolic clearance to morphine is reduced to 5-10% of the clearance in EM subjects [Chen, 1991b; Chen, 1991a; Mikus, 1997].

Pharmacodynamics

Codeine shows a very weak binding to μ-opioid receptors as well as weak opioid agonistic properties in the electrically stimulated guinea-pig ileum model [Kirkwood, 1995; Mignat, 1995]. A hydroxyl group seems to be important for strong agonistic effects of the opioids but other substitutes like a methoxyl group decrease μ-receptor binding and also reduce analgesic activity [Chen, 1991a]. The chemical group at position 6 in the opioid molecule has little effect on the binding affinity whereas a demethylation at position 17 results in compounds with reduced binding affinities (norcodeine, normorphine).

It has been shown that pharmacogenetics determines the effects of codeine. It is widely used analgesic, antitussive and antidiarrhoeal drug. There is increasing evidence, that the analgesic effect of codeine is mediated by its O-demethylated metabolite morphine [Caraco, 1996a; Eckhardt, 1998; Poulsen, 1996b] and that the glucuronidated metabolite morphine-6-glucuronide possesses even greater analgesic potency than morphine itself [Pasternak, 1987]. After codeine administration the antidiarrhoeal effect occurs only if morphine was formed from codeine [Mikus, 1997]. However, in terms of unwanted side effects they are observed independently of the CYP2D6 phenotype [Eckhardt, 1998]. Therefore, in CYP2D6 poor metabolisers

codeine is an ineffective analgesic drug which might elicit only unwanted side effects. Inhibition of CYP2D6 by other drugs will also result in diminished or even absent pharmacodynamic effects of codeine in extensive metabolisers.

Dihydrocodeine

Dihydrocodeine is a semisynthetic opioid which was described firstly in 1911. It is clinically used as a moderately potent opioid analgesic and antitussive drug. Until recently, only limited data on dihydrocodeine pharmacokinetics and metabolism were known. Dihydrocodeine is structurally similar to codeine (Fig. (**3**)), varying only by the reduction of the C7-C8 double bond of codeine to a single bond. It has therefore been proposed that pharmacokinetics and metabolism of dihydrocodeine may be very similar to codeine [Rowell, 1983].

Metabolism

Similar to codeine the primary metabolic steps of dihydrocodeine are N- and O-demethylation and glucuronidation at the 6-position (Fig. (**3**)). Both oxidative pathways have been studied in an animal model for the human CYP2D6 polymorphism, the female Dark-Agouti and female Sprague-Dawley rat [Kirkwood, 1996] as well as in human liver microsomes [Kirkwood, 1997]. CYP2D6 is the major enzyme mediating O-demethylation to dihydromorphine whereas CYP3A predominantly catalyses the nordihydrocodeine formation [Kirkwood, 1997]. It has also been suggested from *in vitro* experiments using human liver microsomes that the glucuronidation of dihydrocodeine is mediated by the UDP-glucuronosyltransferase isoform UGT2B7 with very little interindividual variation [Kirkwood, 1998]. *In vivo* about 30% of a given dose of dihydrocodeine is recovered in urine unchanged independent of the CYP2D6 phenotype [Fromm, 1995]. In PM subjects only 1.2% were recovered as dihydromorphine and its glucuronides whereas in EMs the O-demethylated metabolites accounted for 9%. The excretion of the N-demethylated metabolites was not significantly different (EM: 22.1% vs PM: 24.9 %) [Fromm, 1995]. Major metabolite of dihydrocodeine is the 6-conjugate accounting for 30% of an administered dose [Fromm, 1995]. Almost no data are yet available on the secondary metabolite nordihydromorphine and its glucuronides which might account for 1-2% [Hufschmid, 1995]. This metabolite can be formed via O-demethylation of nordihydrocodeine and via N-demethylation of dihydromorphine. It can be proposed that CYP2D6 is involved in the O-demethylation step and CYP3A in the N-demethylation thereby probably leading to an increase of nordihydrocodeine in PMs due to the lack of the subsequent O-demethylation. The observed small increase of nordihydrocodeine in PMs [Fromm, 1995] might support this assumption.

Pharmacokinetics

The pharmacokinetics of dihydrocodeine itself are independent of the CYP2D6 phenotype due to the small

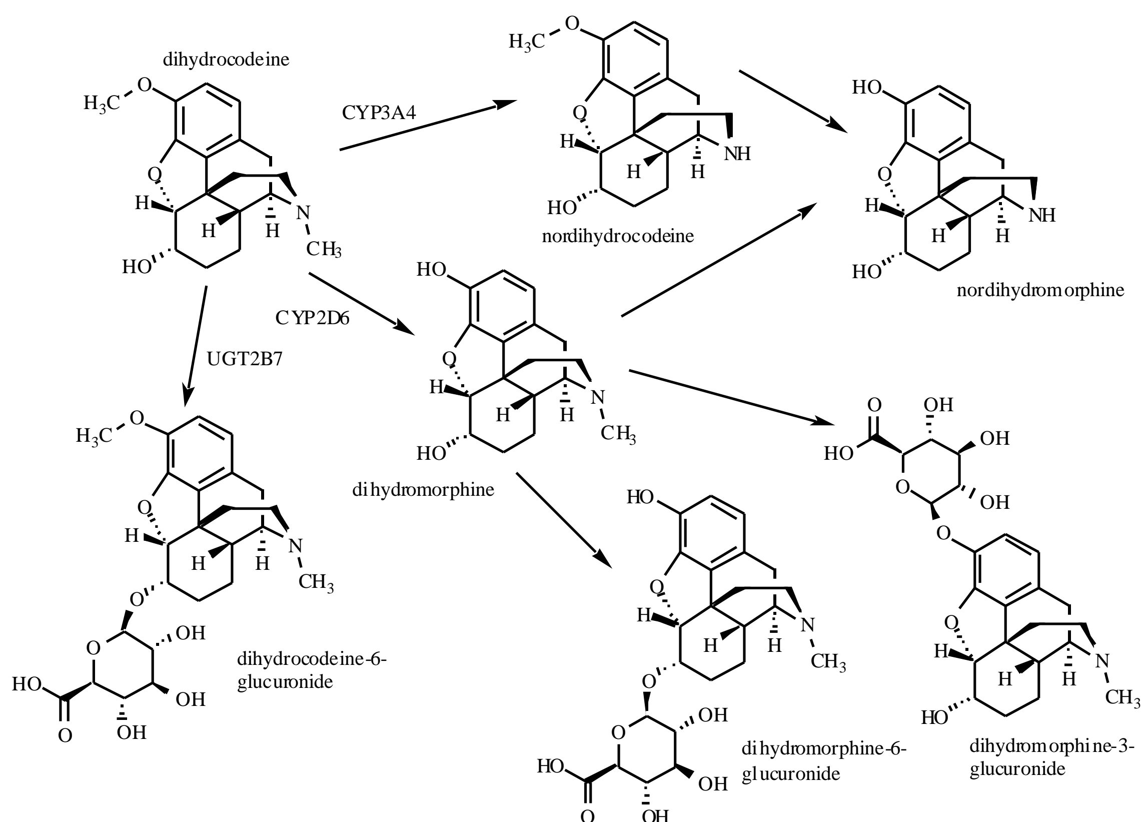

Fig. (3). Chemical structures and metabolic pathways of dihydrocodeine and enzymes contributing to the metabolism.

contribution of polymorphic O-demethylation to the overall metabolism. Maximum plasma concentrations (between 300 and 1000 nmol/L for a 60 mg single oral dose) are observed after 1 to 5 hours depending on the oral formulation used [Ammon, 1999; Fromm, 1995; Rowell, 1983]. Terminal elimination half-life ranges between 4 and 6 hours [Fromm, 1995; Rowell, 1983]. In the dose range up to 120 mg the pharmacokinetics of dihydrocodeine are linear [Ammon, 1999]. The absolute bioavailability of dihydrocodeine was determined to 20% [Rowell, 1983]. In relation to dihydrocodeine only very low plasma concentrations of the O-demethylated metabolite dihydromorphine were observed. The AUC ratio of dihydrocodeine and dihydromorphine is significantly related to the CYP2D6 metabolic ratio (phenotype) [Fromm, 1995]. The elimination half-life of this active metabolite is on average 9 hours [Fromm, 1995]. The formation of this metabolite is linear for a dose range up to 120 mg dihydrocodeine [Ammon, 1999]. The partial metabolic clearance of dihydrocodeine to its –6-glucuronide accounts for 50% of the apparent oral clearance [Ammon, 1999].

Pharmacodynamics

Binding studies to the µ-opioid receptor and studies of the effect on the electrically stimulated guinea-pig ileum revealed that dihydrocodeine and codeine have similar binding properties to the µ-receptor [Kirkwood, 1995; Mignat, 1995; Schmidt, 2002]. It has been suggested that dihydromorphine and also dihydromorphine-6-glucuronide are potent active metabolites of dihydrocodeine [Kirkwood, 1995; Mignat, 1995; Schmidt, 2002] whereas nordihydrocodeine and dihydrocodeine-6-glucuronide showed potencies similar to dihydrocodeine itself. Since the dihydrocodeine O-demethylation to dihydromorphine is under genetic control of CYP2D6 it was proposed that the CYP2D6 phenotype might influence the µ-receptor mediated effects after administration of dihydrocodeine. However, in contrast to codeine which is a prodrug, it was suggested that despite the essential role of CYP2D6 in formation of highly active metabolites the CYP2D6 phenotype might not have a major impact opioid receptor mediated effects [Schmidt, 2003; Webb, 2001]. So far this discrepancy has not been understood.

Oxycodone

Oxycodone is an analgesic agent which is recommended by the WHO for the management of mild to moderate cancer pain. It is a semisynthetic derivative of thebaine with similar chemical structure as the naturally occurring opioids (Fig. (**4**)). Oxycodone is increasingly used especially since controlled-release preparations became available [Davis, 2003]. However, a recent analysis of 16 studies showed no clinically significant advantage of controlled-release oxycodone over other long-acting opioids [Rischitelli and Karbowicz, 2002].

Metabolism

Oxycodone metabolism has not been extensively studied in humans. In analogy to codeine metabolism N- and O-demethylation of oxycodone have been described to occur *in vivo* (Fig. (**4**)) [Poyhia, 1991; Weinstein and Gaylord,

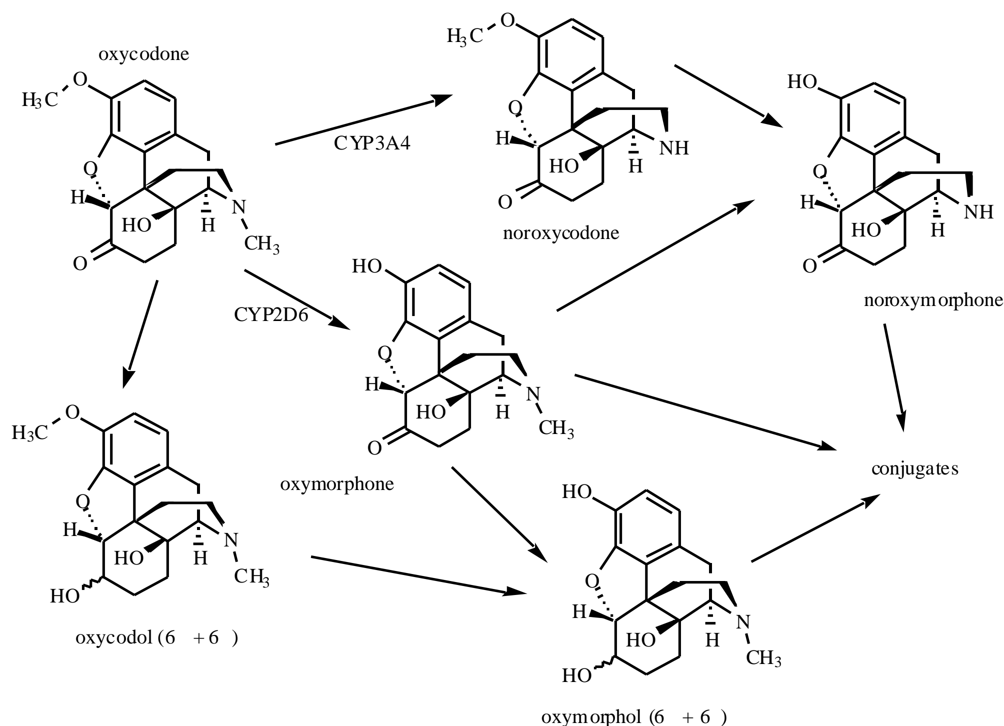

Fig. (4). Chemical structures and metabolic pathways of oxycodone and enzymes contributing to the metabolism.

1979]. The resulting metabolites are noroxycodone and oxymorphone. Both can further be metabolised to form noroxymorphone. Oxycodone and its metabolites can also be subject of conjugation although to our knowledge no detailed study on oxycodone metabolism has been carried out in man. Investigations in human liver microsomes revealed that the N-demethylated metabolite noroxycodone is formed to a substantially greater extent than the O-demethylated metabolite oxymorphone [Menelaou, 2003]. The oxycodone O-demethylation to oxymorphone is catalysed by the polymorphic CYP2D6 as microsomes from a poor metaboliser showed a 5-fold diminished formation of oxymorphone and a potent inhibition in microsomes from extensive metabolisers by quinidine was observed [Otton, 1993b]. As a further pathway of oxycodone metabolism a 6-keto reduction to form 6 - and 6 -oxymorphol has been described [Cone, 1983].

Pharmacokinetics

After intravenous administration of 0.07 mg/kg to 9 healthy male volunteers an average half-life of 3.7 h (range: 2.0 – 5.5 h) was observed [Poyhia, 1991]. Oxycodone clearance was 0.78 l/min (0.53 – 1.03 l/min) and V_{ss} 2.6 l/kg (2.2 – 3.0 l/kg) [Poyhia, 1991]. Noroxycodone was found only in low concentrations (C_{max}: 3 – 8 ng/ml) with an AUC ratio (nor/oxy) of 0.33. Oxymorphone concentrations were always below 0.5 ng/ml which was the limit of quantification of the analytical assay used [Poyhia, 1991].

After oral administration of 20 mg oxycodone controlled release to 10 healthy subjects maximum plasma concentrations of oxycodone (20.4 ± 5.4 ng/mL) were observed after 2.25 ± 1.1 h, mean AUC $_{0-24}$ was 228.8 ± 78.9 h ng/mL [Heiskanen, 1998]. In this study both metabolites noroxycodone and oxymorphone were determined in plasma. Noroxycodone was present in almost similar concentrations as the parent drug (C_{max}: 12.3 ± 2.9 ng/mL; AUC $_{0-24}$: 156.2 ± 38.5 h ng/mL) whereas oxymorphone concentrations were much lower (C_{max}: 0.34 ± 0.18 ng/mL; AUC $_{0-24}$: 1.5 ± 1.1 h ng/mL) [Heiskanen, 1998]. In a patient study it was confirmed that noroxycodone is the major metabolite while oxymorphone is only present in very low concentrations (median ratio oxycodone to noroxycodone 1:1.1; oxycodone to oxymorphone 35:1) [Heiskanen, 2000]. Interestingly in the volunteer study, pre-treatment with 200 mg quinidine, a potent inhibitor of CYP2D6, 3 h before and 100 mg quinidine 6 h after oxycodone administration did not change oxycodone pharmacokinetics but nearly abolished oxymorphone formation [Heiskanen, 1998], which was to be expected by *in vitro* metabolism data. Although noroxycodone formation is catalysed by CYP3A4 quinidine treatment increased noroxycodone plasma concentrations nearly 2-fold [Heiskanen, 1998]. In contrast to the authors of this study we suggest that this is due to inhibition of further metabolism to noroxymorphone mediated by CYP2D6 which is inhibited by quinidine treatment. Unfortunately, this cannot be proven since in this study no noroxymorphone was determined.

Only limited data have been published on the pharmacokinetics of oxycodone in patients with reduced liver and kidney function. The median elimination half-life of oxycodone was 13.9 hours (range, 4.6 to 24.4 hours) in 6 patients with cirrhosis before transplantation and noroxycodone was not measurable in plasma of most of the patients [Tallgren, 1997]. In 10 uremic patients elimination half-life of oxycodone was highly variable and prolonged compared to a control group. Moreover, elimination of the metabolite noroxycodone was even more impaired leading to elevated noroxycodone plasma concentrations [Kirvela, 1996]. No detailed recommendation can be made for dose adjustment in patients with reduced renal or liver function.

Pharmacodynamics

Radioreceptor assay indicate that oxycodone is a µ-receptor agonist, but its affinity is considerably lower compared to morphine [Chen, 1991a; Pert and Snyder, 1973]. However, data published on the µ-receptor binding of oxycodone metabolites indicate, that both noroxymorphone and oxymorphone are potent µ-receptor agonists [Chen, 1991a; Creese and Snyder, 1975]. It is thought that the analgesic effect of oxycodone is primarily due to the parent compound. Although oxymorphone has analgesic effects, it is only present in low concentrations and hence contributes to the analgesic effect little if at all.

Oxycodone is used clinically in the management of postoperative pain and chronic cancer pain [Kalso, 1991; Kalso and Vainio, 1990]. Equianalgesic dosing requires less oxycodone than the standard analgesic drug morphine [Kalso, 1991], which is in contrast to the µ-receptor binding data. Therefore, pharmacologically active metabolites of oxycodone may be important in oxycodone analgesia.

Hydrocodone

Hydrocodone is a semisynthetic congener of codeine which is used as an antitussive agent (Fig. (**5**)). The central antitussive effect of hydrocodone is more pronounced in relation to codeine and dihydrocodeine. In Germany the use of hydrocodone is restricted as it is a controlled substance like morphine. The O-demethylated metabolite hydromorphone is also marketed as a controlled substance and is used for the treatment of strongest pain.

Metabolism

Not many data exist on hydrocodone metabolism. In the 1970's the metabolic pathways of hydrocodone in man, rat, dog and other species have been published [Cone, 1978]. Like codeine and the other opioids O- and N-demethylation and the 6-keto-reduction are the primary metabolic steps (Fig. (**5**)) which are carried out enzymatically with the formation of norhydrocodone being the predominant step [Menelaou, 2003]. Recently, it was shown using human liver microsomes, that hydrocodone O-demethylation is mediated by CYP2D6 and hydrocodone N-demethylation by CYP3A4 [Hutchinson, 2004].

Pharmacokinetics

There is one study published on the pharmacokinetics of hydrocodone in relation to the CYP2D6 phenotype [Otton, 1993a]. No phenotypic differences in the pharmacokinetics of hydrocodone were observed, however total body

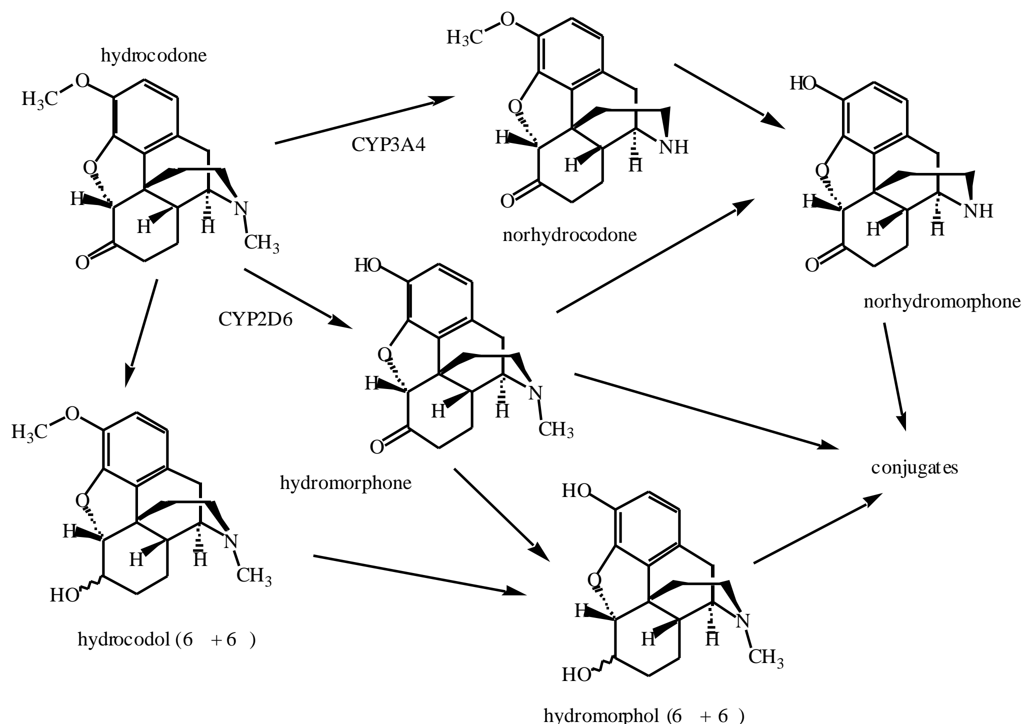

Fig. (5). Chemical structures and metabolic pathways of hydrocodone and enzymes contributing to the metabolism.

clearances was decreased by 40% in the 6 PM subjects. Because there are no AUC data given and the plasma concentration time profile of EM and PM subjects look very similar the clearance calculation cannot be verified. Terminal elimination half-life of hydrocodone varies between 3 and 9 hours and renal clearance is on average 60 ml/h/kg [Otton, 1993a]. The partial metabolic clearance to hydromorphone is 28 ml/h/kg in EMs and is reduced to 3 ml/h/kg in PMs. No further detailed pharmacokinetic data of hydrocodone are available to date.

Pharmacodynamics

Hydrocodone is an effective antitussive agent and analgesic agent for the treatment of moderate to moderately severe pain. The antitussive potency of hydrocodone is greater than that of codeine, however the drug appears to offer no advantage over codeine in the treatment of non-productive cough. Hydrocodone shows a μ-receptor binding affinity similar to methadone and its O-demethylated metabolite hydromorphone has a very strong binding [Chen, 1991a]. Clinical consequences of the CYP2D6 mediated hydrocodone O-demethylation to hydromorphone have not been studied yet.

Tramadol

Tramadol is a centrally acting analgesic drug with an opioid mechanism of action and additional effects on the noradrenergic and serotonergic neurotransmission. In contrast to pure opioid agonists tramadol causes only low incidences of respiratory depression, no tolerance and it shows a low dependency potential.

Metabolism

Tramadol has two chiral centers and the drug preparations used contain a 1:1 racemic mixture of (+)- and (-)-tramadol. It undergoes biotransformation mainly in the liver by two main metabolic pathways to form the O-demethylated (M1) and N-demethylated (M2) compounds [Lintz, 1981]. The enzymes responsibly for the primary metabolic steps have been identified *in vitro*: CYP2D6 (M1-formation), CYP2B6 and CYP3A4 (M2-formation) [Subrahmanyam, 2001]. The CYP2D6 dependency of M1-formation has also been demonstrated in panel studies with CYP2D6 EM and PM subjects [Abdel-Rahman, 2002; Poulsen, 1996a].

Pharmacokinetics

Stereoselective pharmacokinetics are observed after administration of racemic tramadol to humans [Liu, 2001; Poulsen, 1996a] and higher concentrations of both enantiomers are attained in CYP2D6 poor metaboliser subjects [Poulsen, 1996a]. In general, tramadol is well absorbed after oral administration with a moderate to high bioavailability (>60%) [Lintz, 1986]. Plasma protein binding is low (~20%) and volume of distribution is high indicating tissue affinity [Lintz, 1986]. The main route of elimination of tramadol is metabolism and subsequent renal excretion [Lee, 1993].

Pharmacodynamics

The (-)-enantiomers of tramadol and its M1 metabolite have a weaker affinity to the μ-opioid receptor than the (+)-enantiomers [Poulsen, 1996a; Raffa, 1993]. The affinity of

(+)-M1 is about 200 times greater than that of the parent compound suggesting the main analgesic activity after tramadol administration is mediated by the (+)-M1 metabolite which has been demonstrated in a clinical study using an experimental pain model [Poulsen, 1996a]. The therapeutic efficacy of tramadol in relation to the CYP2D6 genotype has not yet been studied.

CONCLUSION

The CYP2D6 polymorphism determines the formation of the O-demethylated metabolites of codeine, dihydrocodeine, oxycodone and hydrocodone. At least from *in vitro* data and μ-receptor affinities it is obvious that these O-demethylated metabolites are more potent opioids than the parent drugs. Because three of these metabolites (morphine, oxymorphone, hydromorphone) are available for drug therapy as parent drugs classified as strong opioids it is quite obvious that these compounds are able to elicit a strong opioid action by their own. If weak opioids like codeine, dihydrocodeine, oxycodone, and hydrocodone are administered to man the polymorphic CYP2D6 enzyme regulates the amount of potent O-demethylated metabolites. At least for codeine and morphine it is clear that codeine is a prodrug which elicits its pharmacological effects through the potent μ-receptor agonist morphine. Regarding the other opioids there are some uncertainties about the contribution of the parent drugs and their O-demethylated metabolites to the overall opioid effects. Due to the low metabolite concentration it is unlikely that they contribute much to the pharmacological effects. There are more studies required to elucidate the mechanisms behind the pharmacological effect of these weak opioids.

It has also been shown that CYP2D6 is involved in the metabolism of other μ-receptor agonists like tramadol and methadone.

REFERENCES

Abdel-Rahman, S. M.; Leeder, J. S.; Wilson, J. T.; Gaedigk, A.; Gotschall, R. R.; Medve, R.; Liao, S.; Spielberg, S. P. and Kearns, G. L. (2002) Concordance between tramadol and dextromethorphan parent/metabolite ratios: the influence of CYP2D6 and non-CYP2D6 pathways on biotransformation. *J. Clin. Pharmacol.* **42**, 24-29.

Ammon, S.; Hofmann, U.; Griese, E. U.; Gugeler, N. and Mikus, G. (1999) Pharmacokinetics of dihydrocodeine and its active metabolite after single and multiple oral dosing. *Br. J. Clin. Pharmacol.* **48**, 317-322.

Ammon, S.; Marx, C.; Behrens, C.; Hofmann, U.; Murdter, T.; Griese, E. U. and Mikus, G. (2002) Diclofenac does not interact with codeine metabolism *in vivo*: A study in healthy volunteers. *BMC Clin. Pharmacol.* **2**, 2.

Bradford, L. D. (2002) CYP2D6 allele frequency in European Caucasians, Asians, Africans and their descendants. *Pharmacogenomics* **3**, 229-243.

Caporaso, N. E.; Shields, P. G.; Landi, M. T.; Shaw, G. L.; Tucker, M. A.; Hoover, R.; Sugimura, H.; Weston, A. and Harris, C. C. (1992) The debrisoquine metabolic phenotype and DNA-based assays: implications of misclassification for the association of lung cancer and the debrisoquine metabolic phenotype. *Environ. Health Perspect.* **98**, 101-105.

Caraco, Y.; Sheller, J. and Wood, A. J. (1996a) Pharmacogenetic determination of the effects of codeine and prediction of drug interactions. *J. Pharmacol. Exp. Ther.* **278**, 1165-1174.

Caraco, Y.; Sheller, J. and Wood, A. J. (1999) Impact of ethnic origin and quinidine coadministration on codeine's disposition and pharmacodynamic effects. *J. Pharmacol. Exp. Ther.* **290**, 413-422.

Caraco, Y.; Tateishi, T.; Guengerich, F. P. and Wood, A. J. (1996b) Microsomal codeine N-demethylation: cosegregation with cytochrome P4503A4 activity. *Drug Metab. Dispos.* **24**, 761-764.

Chen, Z. R.; Irvine, R. J.; Somogyi, A. A. and Bochner, F. (1991a) Mu receptor binding of some commonly used opioids and their metabolites. *Life Sci.* **48**, 2165-2171.

Chen, Z. R.; Somogyi, A. A.; Reynolds, G. and Bochner, F. (1991b) Disposition and metabolism of codeine after single and chronic doses in one poor and seven extensive metabolisers. *Br. J. Clin. Pharmacol.* **31**, 381-390.

Coffman, B. L.; Rios, G. R.; King, C. D. and Tephly, T. R. (1997) Human UGT2B7 catalyzes morphine glucuronidation. *Drug Metab. Dispos.* **25**, 1-4.

Cone, E. J.; Darwin, W. D.; Buchwald, W. F. and Gorodetzky, C. W. (1983) Oxymorphone metabolism and urinary excretion in human, rat, guinea pig, rabbit, and dog. *Drug Metab. Dispos.* **11**, 446-450.

Cone, E. J.; Darwin, W. D.; Gorodetzky, C. W. and Tan, T. (1978) Comparative metabolism of hydrocodone in man, rat, guinea pig, rabbit, and dog. *Drug Metab. Dispos.* **6**, 488-493.

Creese, I. and Snyder, S. H. (1975) Receptor binding and pharmacological activity of opiates in the guinea-pig intestine. *J. Pharmacol. Exp. Ther.* **194**, 205-219.

Daly, A. K.; Brockmoller, J.; Broly, F.; Eichelbaum, M.; Evans, W. E.; Gonzalez, F. J.; Huang, J. D.; Idle, J. R.; Ingelman-Sundberg, M.; Ishizaki, T.; Jacqz-Aigrain, E.; Meyer, U. A.; Nebert, D. W.; Steen, V. M.; Wolf, C. R. and Zanger, U. M. (1996) Nomenclature for human CYP2D6 alleles. *Pharmacogenetics* **6**, 193-201.

Davis, M. P.; Varga, J.; Dickerson, D.; Walsh, D.; LeGrand, S. B. and Lagman, R. (2003) Normal-release and controlled-release oxycodone: pharmacokinetics, pharmacodynamics, and controversy. *Support Care Cancer* **11**, 84-92.

Dayer, P.; Desmeules, J.; Leemann, T. and Striberni, R. (1988) Bioactivation of the narcotic drug codeine in human liver is mediated by the polymorphic monooxygenase catalyzing debrisoquine 4-hydroxylation (cytochrome P-450 dbl/bufI). *Biochem. Biophys. Res. Commun.* **152**, 411-416.

Eckhardt, K.; Li, S.; Ammon, S.; Schanzle, G.; Mikus, G. and Eichelbaum, M. (1998) Same incidence of adverse drug events after codeine administration irrespective of the genetically determined differences in morphine formation. *Pain* **76**, 27-33.

Eddy, N. B.; Friebel, H.; Hahn, K. J. and Halbach, H. (1968) Codeine and its alternates for pain and cough relief. I. Codeine, exclusive of its antitussive action. *Bull. World Health Organ* **38**, 673-741.

Ensom, M. H.; Chang, T. K. and Patel, P. (2001) Pharmacogenetics: the therapeutic drug monitoring of the future? *Clin. Pharmacokinet.* **40**, 783-802.

Fromm, M. F.; Hofmann, U.; Griese, E. U. and Mikus, G. (1995) Dihydrocodeine: a new opioid substrate for the polymorphic CYP2D6 in humans. *Clin. Pharmacol. Ther.* **58**, 374-382.

Garte, S. and Crosti, F. (1999) A nomenclature system for metabolic gene polymorphisms. *IARC Sci. Publ.* 5-12.

Gutstein, H. B. and Akil, H. (2001) Opioid analgesics. In *The pharmacological basis of therapeutics;* Hardman, J. G.; Limbird, L. E.; Eds; McGraw-Hill: New York, pp. 569-619.

Heim, M. H. and Meyer, U. A. (1991) Genetic polymorphism of debrisoquine oxidation: restriction fragment analysis and allele-specific amplification of mutant alleles of CYP2D6. *Methods Enzymol.* **206**, 173-183.

Heiskanen, T.; Olkkola, K. T. and Kalso, E. (1998) Effects of blocking CYP2D6 on the pharmacokinetics and pharmacodynamics of oxycodone. *Clin. Pharmacol. Ther.* **64**, 603-611.

Heiskanen, T. E.; Ruismaki, P. M.; Seppala, T. A. and Kalso, E. A. (2000) Morphine or oxycodone in cancer pain? *Acta Oncol.* **39**, 941-947.

Hersberger, M.; Marti-Jaun, J.; Rentsch, K. and Hanseler, E. (2000) Rapid detection of the CYP2D6*3, CYP2D6*4, and CYP2D6*6 alleles by tetra-primer PCR and of the CYP2D6*5 allele by multiplex long PCR. *Clin. Chem.* **46**, 1072-1077.

Hufschmid, E.; Theurillat, R.; Martin, U. and Thormann, W. (1995) Exploration of the metabolism of dihydrocodeine via determination of its metabolites in human urine using micellar electrokinetic capillary chromatography. *J. Chromatogr. B Biomed. Appl.* **668**, 159-170.

Hutchinson, M. R.; Menelaou, A.; Foster, D. J.; Coller, J. K. and Somogyi, A. A. (2004) CYP2D6 and CYP3A4 involvement in the primary

oxidative metabolism of hydrocodone by human liver microsomes. *Br. J. Clin. Pharmacol.* **57**, 287-297.

International Narcotics Control Board (2003) Narcotic drugs - Estimated World Requirements for 2003 - Statistics for 2001. Technical Reports - E/INCB/2002/2; United Nations: New York.

Ji, L.; Pan, S.; Marti-Jaun, J.; Hanseler, E.; Rentsch, K. and Hersberger, M. (2002) Single-step assays to analyze CYP2D6 gene polymorphisms in Asians: allele frequencies and a novel *14B allele in mainland Chinese. *Clin. Chem.* **48**, 983-988.

Johansson, I.; Lundqvist, E.; Bertilsson, L.; Dahl, M. L.; Sjoqvist, F. and Ingelman-Sundberg, M. (1993) Inherited amplification of an active gene in the cytochrome P450 CYP2D locus as a cause of ultrarapid metabolism of debrisoquine. *Proc. Natl. Acad. Sci. USA* **90**, 11825-11829.

Johansson, I.; Oscarson, M.; Yue, Q. Y.; Bertilsson, L.; Sjoqvist, F. and Ingelman-Sundberg, M. (1994) Genetic analysis of the Chinese cytochrome P4502D locus: characterization of variant CYP2D6 genes present in subjects with diminished capacity for debrisoquine hydroxylation. *Mol. Pharmacol.* **46**, 452-459.

Kalso, E.; Poyhia, R.; Onnela, P.; Linko, K.; Tigerstedt, I. and Tammisto, T. (1991) Intravenous morphine and oxycodone for pain after abdominal surgery. *Acta Anaesthesiol. Scand.* **35**, 642-646.

Kalso, E. and Vainio, A. (1990) Morphine and oxycodone hydrochloride in the management of cancer pain. *Clin. Pharmacol. Ther.* **47**, 639-646.

Kirkwood, L. C.; Nation, R. L.; Reynolds, G. D.; Somogyi, A. and Sansom, L. N. (1996) Oxidative metabolism of dihydrocodeine in Dark-Agouti and Sprague-dawley rat liver microsomes. *Pharm. Sci.* **2**, 299-303.

Kirkwood, L. C.; Nation, R. L. and Somogyi, A. A. (1997) Characterization of the human cytochrome P450 enzymes involved in the metabolism of dihydrocodeine. *Br. J. Clin. Pharmacol.* **44**, 549-555.

Kirkwood, L. C.; Nation, R. L. and Somogyi, A. A. (1998) Glucuronidation of dihydrocodeine by human liver microsomes and the effect of inhibitors. *Clin. Exp. Pharmacol. Physiol.* **25**, 266-270.

Kirkwood, L. C.; Venning, M. G.; Nation, R. L.; Reynolds, G. D.; Somogyi, A. A. and Sansom, L. N. (1995) Comparative activity of dihydrocodeine and its metabolites in the electrically stimulated guinea-pig isolated ileum. *Pharm. Sci.* **1**, 573-575.

Kirvela, M.; Lindgren, L.; Seppala, T. and Olkkola, K. T. (1996) The pharmacokinetics of oxycodone in uremic patients undergoing renal transplantation. *J. Clin. Anesth.* **8**, 13-18.

Lee, C. R.; McTavish, D. and Sorkin, E. M. (1993) Tramadol. A preliminary review of its pharmacodynamic and pharmacokinetic properties, and therapeutic potential in acute and chronic pain states. *Drugs* **46**, 313-340.

Lennard, M. S.; Silas, J. H.; Freestone, S. and Trevethick, J. (1982) Defective metabolism of metoprolol in poor hydroxylators of debrisoquine. *Br. J. Clin. Pharmacol.* **14**, 301-303.

Linder, M. W.; Prough, R. A. and Valdes, R. Jr. (1997) Pharmacogenetics: a laboratory tool for optimizing therapeutic efficiency. *Clin. Chem.* **43**, 254-266.

Lindner, E. (1985) Structure activities and pharmacologicalproperties of the opium alkaloids. In *The chemistry and biology of isoquinoline alkaloids;* Phillipson, J. D.; Roberts, M. F.; Zenk, M. H.; Eds; Springer-Verlag: Berlin Heidelberg New York Tokyo, pp. 38-46.

Lintz, W.; Barth, H.; Osterloh, G. and Schmidt-Bothelt, E. (1986) Bioavailability of enteral tramadol formulations. 1st communication: capsules. *Arzneimittelforschung.* **36**, 1278-1283.

Lintz, W.; Erlacin, S.; Frankus, E. and Uragg, H. (1981) [Biotransformation of tramadol in man and animal (author's transl)]. *Arzneimittelforschung.* **31**, 1932-1943.

Liu, H. C.; Liu, T. J.; Yang, Y. Y. and Hou, Y. N. (2001) Pharmacokinetics of enantiomers of trans-tramadol and its active metabolite, trans-O-demethyltramadol, in human subjects. *Acta Pharmacol. Sin.* **22**, 91-96.

LLerena, A.; Herraiz, A. G.; Cobaleda, J.; Johansson, I. and Dahl, M. L. (1993) Debrisoquin and mephenytoin hydroxylation phenotypes and CYP2D6 genotype in patients treated with neuroleptic and antidepressant agents. *Clin. Pharmacol. Ther.* **54**, 606-611.

Lovlie, R.; Daly, A. K.; Molven, A.; Idle, J. R. and Steen, V. M. (1996) Ultrarapid metabolizers of debrisoquine: characterization and PCR-based detection of alleles with duplication of the CYP2D6 gene. *FEBS Lett.* **392**, 30-34.

Lundqvist, E.; Johansson, I. and Ingelman-Sundberg, M. (1999) Genetic mechanisms for duplication and multiduplication of the human CYP2D6 gene and methods for detection of duplicated CYP2D6 genes. *Gene* **226**, 327-338.

Madsen, H.; Nielsen, K. K. and Brosen, K. (1995) Imipramine metabolism in relation to the sparteine and mephenytoin oxidation polymorphisms--a population study. *Br. J. Clin. Pharmacol.* **39**, 433-439.

Masimirembwa, C.; Persson, I.; Bertilsson, L.; Hasler, J. and Ingelman-Sundberg, M. (1996) A novel mutant variant of the CYP2D6 gene (CYP2D6*17) common in a black African population: association with diminished debrisoquine hydroxylase activity. *Br. J. Clin. Pharmacol.* **42**, 713-719.

Menelaou, A.; Hutchinson, M. R.; Quinn, I.; Christensen, A. and Somogyi, A. A. (2003) Quantification of the O- and N-demethylated metabolites of hydrocodone and oxycodone in human liver microsomes using liquid chromatography with ultraviolet absorbance detection. *J. Chromatogr. B Analyt. Technol. Biomed. Life Sci.* **785**, 81-88.

Meyer, U. A. and Zanger, U. M. (1997) Molecular mechanisms of genetic polymorphisms of drug metabolism. *Annu. Rev. Pharmacol. Toxicol.* **37**, 269-296.

Mignat, C.; Wille, U. and Ziegler, A. (1995) Affinity profiles of morphine, codeine, dihydrocodeine and their glucuronides at opioid receptor subtypes. *Life Sci.* **56**, 793-799.

Mikus, G.; Somogyi, A. A.; Bochner, F. and Eichelbaum, M. (1991) Codeine O-demethylation: rat strain differences and the effects of inhibitors. *Biochem. Pharmacol.* **41**, 757-762.

Mikus, G.; Trausch, B.; Rodewald, C.; Hofmann, U.; Richter, K.; Gramatte, T. and Eichelbaum, M. (1997) Effect of codeine on gastrointestinal motility in relation to CYP2D6 phenotype. *Clin. Pharmacol. Ther.* **61**, 459-466.

Muller, B.; Zopf, K.; Bachofer, J. and Steimer, W. (2003) Optimized Strategy for Rapid Cytochrome P450 2D6 Genotyping by Real-Time Long PCR. *Clin. Chem.* **49**, 1624-1631.

Otton, S. V.; Schadel, M.; Cheung, S. W.; Kaplan, H. L.; Busto, U. E. and Sellers, E. M. (1993a) CYP2D6 phenotype determines the metabolic conversion of hydrocodone to hydromorphone. *Clin. Pharmacol. Ther.* **54**, 463-472.

Otton, S. V.; Wu, D.; Joffe, R. T.; Cheung, S. W. and Sellers, E. M. (1993b) Inhibition by fluoxetine of cytochrome P450 2D6 activity. *Clin. Pharmacol. Ther.* **53**, 401-409.

Pasternak, G. W.; Bodnar, R. J.; Clark, J. A. and Inturrisi, C. E. (1987) Morphine-6-glucuronide, a potent mu agonist. *Life Sci.* **41**, 2845-2849.

Pert, C. B. and Snyder, S. H. (1973) Opiate receptor: demonstration in nervous tissue. *Science* **179**, 1011-1014.

Poulsen, L.; Arendt-Nielsen, L.; Brosen, K. and Sindrup, S. H. (1996a) The hypoalgesic effect of tramadol in relation to CYP2D6. *Clin. Pharmacol. Ther.* **60**, 636-644.

Poulsen, L.; Brosen, K.; Arendt-Nielsen, L.; Gram, L. F.; Elbaek, K. and Sindrup, S. H. (1996b) Codeine and morphine in extensive and poor metabolizers of sparteine: pharmacokinetics, analgesic effect and side effects. *Eur. J. Clin. Pharmacol.* **51**, 289-295.

Poyhia, R.; Olkkola, K. T.; Seppala, T. and Kalso, E. (1991) The pharmacokinetics of oxycodone after intravenous injection in adults. *Br. J. Clin. Pharmacol.* **32**, 516-518.

Projean, D.; Baune, B.; Farinotti, R.; Flinois, J. P.; Beaune, P.; Taburet, A. M. and Ducharme, J. (2003) *In vitro* metabolism of chloroquine: identification of CYP2C8, CYP3A4, and CYP2D6 as the main isoforms catalyzing N-desethylchloroquine formation. *Drug Metab. Dispos.* **31**, 748-754.

Raffa, R. B.; Friderichs, E.; Reimann, W.; Shank, R. P.; Codd, E. E.; Vaught, J. L.; Jacoby, H. I. and Selve, N. (1993) Complementary and synergistic antinociceptive interaction between the enantiomers of tramadol. *J. Pharmacol. Exp. Ther.* **267**, 331-340.

Raimundo, S.; Fischer, J.; Eichelbaum, M.; Griese, E. U.; Schwab, M. and Zanger, U. M. (2000) Elucidation of the genetic basis of the common 'intermediate metabolizer' phenotype for drug oxidation by CYP2D6. *Pharmacogenetics* **10**, 577-581.

Ramamoorthy, Y.; Tyndale, R. F. and Sellers, E. M. (2001) Cytochrome P450 2D6.1 and cytochrome P450 2D6.10 differ in catalytic activity for multiple substrates. *Pharmacogenetics* **11**, 477-487.

Rischitelli, D. G. and Karbowicz, S. H. (2002) Safety and efficacy of controlled-release oxycodone: a systematic literature review. *Pharmacotherapy* **22**, 898-904.

Rost, K. L.; Brockmoller, J.; Esdorn, F. and Roots, I. (1995) Phenocopies of poor metabolizers of omeprazole caused by liver disease and drug treatment. *J. Hepatol.* **23**, 268-277.

Rowell, F. J.; Seymour, R. A. and Rawlins, M. D. (1983) Pharmacokinetics of intravenous and oral dihydrocodeine and its acid metabolites. *Eur. J. Clin. Pharmacol.* **25**, 419-424.

Schmidt, H.; Vormfelde, S.; Klinder, K.; Gundert-Remy, U.; Gleiter, C. H.; Skopp, G.; Aderjan, R. and Fuhr, U. (2002) Affinities of dihydrocodeine and its metabolites to opioid receptors. *Pharmacol. Toxicol.* **91**, 57-63.

Schmidt, H.; Vormfelde, S. V.; Walchner-Bonjean, M.; Klinder, K.; Freudenthaler, S.; Gleiter, C. H.; Gundert-Remy, U.; Skopp, G.; Aderjan, R. and Fuhr, U. (2003) The role of active metabolites in dihydrocodeine effects. *Int. J. Clin. Pharmacol. Ther.* **41**, 95-106.

Skoda, R. C.; Gonzalez, F. J.; Demierre, A. and Meyer, U. A. (1988) Two mutant alleles of the human cytochrome P-450db1 gene (P450C2D1) associated with genetically deficient metabolism of debrisoquine and other drugs. *Proc. Natl. Acad. Sci. USA* **85**, 5240-5243.

Stamer, U. M.; Bayerer, B.; Wolf, S.; Hoeft, A. and Stuber, F. (2002) Rapid and reliable method for cytochrome P450 2D6 genotyping. *Clin. Chem.* **48**, 1412-1417.

Streetman, D. S.; Bertino, J. S., Jr. and Nafziger, A. N. (2000) Phenotyping of drug-metabolizing enzymes in adults: a review of in-vivo cytochrome P450 phenotyping probes. *Pharmacogenetics* **10**, 187-216.

Stuven, T.; Griese, E. U.; Kroemer, H. K.; Eichelbaum, M. and Zanger, U. M. (1996) Rapid detection of CYP2D6 null alleles by long distance- and multiplex-polymerase chain reaction. *Pharmacogenetics* **6**, 417-421.

Subrahmanyam, V.; Renwick, A. B.; Walters, D. G.; Young, P. J.; Price, R. J.; Tonelli, A. P. and Lake, B. G. (2001) Identification of cytochrome P-450 isoforms responsible for cis-tramadol metabolism in human liver microsomes. *Drug Metab. Dispos.* **29**, 1146-1155.

Tallgren, M.; Olkkola, K. T.; Seppala, T.; Hockerstedt, K. and Lindgren, L. (1997) Pharmacokinetics and ventilatory effects of oxycodone before and after liver transplantation. *Clin. Pharmacol. Ther.* **61**, 655-661.

Webb, J. A.; Rostami-Hodjegan, A.; Abdul-Manap, R.; Hofmann, U.; Mikus, G. and Kamali, F. (2001) Contribution of dihydrocodeine and dihydromorphine to analgesia following dihydrocodeine administration in man: a PK-PD modelling analysis. *Br. J. Clin. Pharmacol.* **52**, 35-43.

Weinstein, S. H. and Gaylord, J. C. (1979) Determination of oxycodone in plasma and identification of a major metabolite. *J. Pharm. Sci.* **68**, 527-528.

Wennerholm, A.; Johansson, I.; Massele, A. Y.; Lande, M.; Alm, C.; Aden-Abdi, Y.; Dahl, M. L.; Ingelman-Sundberg, M.; Bertilsson, L. and Gustafsson, L. L. (1999) Decreased capacity for debrisoquine metabolism among black Tanzanians: analyses of the CYP2D6 genotype and phenotype. *Pharmacogenetics* **9**, 707-714.

Xu, B. Q.; Aasmundstad, T. A.; Christophersen, A. S.; Morland, J. and Bjorneboe, A. (1997) Evidence for CYP2D1-mediated primary and secondary O-dealkylation of ethylmorphine and codeine in rat liver microsomes. *Biochem. Pharmacol.* **53**, 603-609.

Yue, Q. Y.; Hasselstrom, J.; Svensson, J. O. and Sawe, J. (1991) Pharmacokinetics of codeine and its metabolites in Caucasian healthy volunteers: comparisons between extensive and poor hydroxylators of debrisoquine. *Br. J. Clin. Pharmacol.* **31**, 635-642.

Yue, Q. Y.; Svensson, J. O.; Alm, C.; Sjoqvist, F. and Sawe, J. (1989) Codeine O-demethylation co-segregates with polymorphic debrisoquine hydroxylation. *Br. J. Clin. Pharmacol.* **28**, 639-645.

Received: 17 November, 2003 Accepted: 19 October, 2004