

# Inhibition of Cytochrome P450 2D6 Metabolism of Hydrocodone to Hydromorphone Does Not Importantly Affect Abuse Liability<sup>1</sup>

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Accepted for publication November 5, 1996

## ABSTRACT

Enzymatic conversion of hydrocodone to hydromorphone is catalyzed by cytochrome P450 2D6, which is inactive in about 7% of Caucasians [poor metabolizers (PMs)] and can be inhibited by quinidine pretreatment in the remainder [extensive metabolizers (EMs)]. If hydromorphone, having a substantially higher  $\mu$ -receptor affinity than hydrocodone, contributes importantly to the physiological and subjective effects of oral hydrocodone, then PMs should be less responsive to the same doses, and quinidine pretreatment should cause EMs to temporarily respond as PMs. Seventeen EMs and 8 PMs who previously responded positively to hydromorphone s.c. re-

ceived placebo and hydrocodone (10 mg, 15 mg and 22.5 mg p.o.) and were retested with their favorite dose after placebo or quinidine (100 mg) pretreatment; physiological and subjective measures were collected at base line and four times after drug administration, and urine was collected for 8 hr. EMs and PMs were equally responsive to oral hydrocodone, and quinidine had no consistent effect on their responses, even though quinidine abolished the pre-existing metabolic differences in hydromorphone production, as measured in urine. These data suggest only a small role of hydromorphone in eliciting abuse-related responses to oral hydrocodone.

The genetic polymorphism of the drug-metabolizing enzyme CYP2D6 results in phenotypic differences in the pharmacokinetics of many drugs (Eichelbaum and Gross, 1990). Approximately 7% of Caucasians have no CYP2D6 (PM phenotype) and therefore have a deficient ability to perform oxidative reactions normally catalyzed by this enzyme; CYP2D6 activity in the remainder of the population (EMs) is highly variable, ranging as much as 10000-fold among individuals (Bertilsson *et al.*, 1991). Quinidine is not metabolized by CYP2D6 but is a potent inhibitor of its activity. A single dose of quinidine sulfate (50–250 mg) temporarily inhibits CYP2D6 activity, thus converting EMs to apparent PMs in a process known as phenocopying (Inaba *et al.*, 1986; Leeman *et al.*, 1986; Speirs *et al.*, 1986).

One drug for which there is evidence of phenotypic differences in response is codeine, which is *O*-demethylated by CYP2D6 to form morphine (Yue *et al.*, 1991; Chen *et al.*, 1991b). In binding assays using rodent brain preparations,

the affinity of morphine for the  $\mu$ -opiate receptor is 2 to 3 orders of magnitude higher than that of codeine (Hennies *et al.*, 1988; Chen *et al.*, 1991a). PMs administered codeine may therefore receive less active drug than do EMs, and coadministration of other substrates/inhibitors of CYP2D6 may attenuate the effect of codeine in EMs. In a preliminary study, Desmeules *et al.* (1991) showed small but statistically significantly increased pain thresholds in EMs receiving codeine, whereas no significant analgesia was detectable in the same subjects coadministered quinidine or in the one PM studied.

Hydrocodone differs structurally from codeine in that the C6-position is occupied by a keto-group, and thus the drug does not undergo the extensive conjugation (>60%) that codeine undergoes (Chen *et al.*, 1991b). Like codeine, hydrocodone is *O*-demethylated by CYP2D6, but it forms hydromorphone instead of morphine. Hydromorphone, which binds with 10- to 33-fold greater affinity than hydrocodone to  $\mu$ -opiate receptors (Hennies *et al.*, 1988; Chen *et al.*, 1991a), is itself marketed as Dilaudid, a potent analgesic. When administered s.c., the analgesic potency of hydromorphone is 7 to 8 times greater than that of morphine (Jaffe and Martin,

Received for publication April 29, 1996.

<sup>1</sup>This work was supported in part by National Institute on Drug Abuse Grant DA06889.

**ABBREVIATIONS:** ARCI, Addiction Research Center Inventory; CYP, cytochrome P450; EM, extensive metabolizer; MBG, morphine-benzedrine group; MR, metabolic ratio; PM, poor metabolizer.

1990), and hydromorphone has been shown experimentally to be 7 times more potent than morphine in suppressing morphine abstinence (Himmelsbach, 1941).

The major metabolic pathways of hydrocodone are *O*-demethylation to form hydromorphone, *N*-demethylation to form nor-hydrocodone and C6-keto reduction to form approximately equal amounts of 6- $\alpha$ -hydrocol and 6- $\beta$ -hydrocol (Cone *et al.*, 1978; Cone and Darwin, 1978). Trace amounts of reduced hydromorphone are also excreted. Hydromorphone, like morphine, has a C3-phenolic site suitable for conjugation, but those authors measured only total hydromorphone.

Our own work has shown that CYP2D6 is responsible for the conversion of hydrocodone to hydromorphone (Otton *et al.*, 1993). The partial metabolic clearance to hydromorphone after 10 mg of oral hydrocodone bitartrate was 8 times faster in five EMs than in six PMs studied ( $28.1 \pm 10.3$  vs.  $3.4 \pm 2.4$  ml/hr/kg,  $P < .001$ ). Furthermore, pretreatment of the EMs with 100 mg of quinidine p.o. reduced their clearance to levels similar to those seen in PMs ( $5.0 \pm 3.6$  ml/hr/kg), and the maximum plasma concentration for hydromorphone was 5 times higher in EMs than in PMs or in EMs pretreated with quinidine. In that earlier, single-blind study, subjective and physiological measures were taken coincidentally with blood sampling over the first 2 hr after dosing. No statistically significant differences in physiological measures were observed but, over the first 1 hr after dosing, EMs reported more positive opiate effects and fewer unpleasant opiate effects, compared with PMs or their own quinidine-pretreated days. This investigation served as the pilot study for the larger, partially double-blind, placebo-controlled study described here.

## Methods

**Subjects.** The participants were 17 EMs and 8 PMs, as determined by the use of the *O*-demethylation MR for dextromethorphan, computed as urinary  $([\text{dextromethorphan}] + [3\text{-methoxymorphinan}]) / ([\text{dextromethorphan}] + [3\text{-hydroxymorphinan}])$ . The individual MRs can be read from the abscissa of figure 1. The 16 male and 9 female subjects (19–42 years of age) were recruited by word of mouth or replied to an advertisement in a weekly newspaper. CYP2D6 genotypes were subsequently determined by the use of leukocyte DNA after polymerase chain reaction analysis (Heim and Meyer, 1990) and were found to be consistent with the phenotypes; all EMs either were homozygous wild-type or had one allele with the B mutation, and all PMs were homozygous for the B mutation, except for one borderline PM (log MR =  $-0.03$ ), who was heterozygous wild-type/B.

Eligible subjects were required to reliably report opioid effects after a hydromorphone screening test (see below), and all signed informed consent (as approved by the Ethics Committee, University of Toronto and Addiction Research Foundation). Only subjects with normal medical and mental status examinations and laboratory screenings and without a history of drug abuse or dependence (DSM-III-R criteria) were admitted into the study. Of the 48 subjects assessed, 22 were excluded, primarily because of their inability to reliably report positive effects of hydromorphone but not placebo ( $n = 18$ ), subsequently discovered ineligibility for the study ( $n = 2$ ) or disinterest in further participation ( $n = 2$ ). One subject did not want to continue the study after the second testing session.

**Drugs and chemicals.** Hydromorphone HCl and quinidine sulfate tablets were obtained as generic products from The Drug Trading Co. Ltd. (Toronto, Canada), and hydrocodone bitartrate was purchased as Hycodan syrup from DuPont Merck Pharma. For injections of hydromorphone s.c., the placebo was sterile water; for hydrocodone p.o., it was artificial wild cherry-flavor syrup. The

200-mg quinidine tablets were halved and administered p.o. in gelatin capsules; the corresponding placebo was dextrose. All analytical standards, reagents and methods were the same as reported previously for dextromethorphan (Schadel *et al.*, 1995) and hydrocodone (Otton *et al.*, 1993).

**Hydromorphone screening day.** Prospective subjects participated in a hydromorphone screening day to ensure that they could reliably distinguish active drug from placebo and report positive subjective effects of hydromorphone HCl at 10 or 20  $\mu\text{g}/\text{kg}$  s.c. Subjects were told that the purpose of the procedure was to test their ability to differentiate active drug from placebo and to assess their mood and feelings about the drug. In this single-blind procedure, each subject received three injections; the third of these was hydromorphone, 10  $\mu\text{g}/\text{kg}$  s.c., with placebo injections 150 and 90 min earlier. A battery of tests similar to those of the double-blind study were conducted at 30 min after the first placebo, 30 and 60 min after the second placebo, and 30, 60 and 75 min after the active injection; having three predrug test cycles provided an opportunity to screen out subjects who responded positively to the second placebo dose. At the day's end, staff workers decided whether the subject was able to differentiate drug from placebo, preferring the hydromorphone, based on scores on nine scales associated with the detection of subjective drug effects. The six subjects who did not have drug effects at 10  $\mu\text{g}/\text{kg}$  were invited to participate in retesting at 20  $\mu\text{g}/\text{kg}$ . Additional details on the hydromorphone screening procedure have been provided elsewhere (U. Busto, H.L. Kaplan, S.V. Otton, M. Schadel, B. Gomez-Mancilla and E.M. Sellers, unpublished conference presentation, 1994).

**Study design.** The study consisted of six 1-day study sessions. On study days 1 and 2 the subject received either hydrocodone, 10 mg (expressed as the base), or placebo p.o., in a random order. On study days 3 and 4 subjects received 15 mg and 22.5 mg of hydrocodone p.o., respectively; an option to substitute lower doses, had these doses not been well tolerated (according to the study physician), was designed but never required. On study days 5 and 6 the subject's "favorite dose," the one preferred to the other two active doses in a majority of subjective liking measures, was again administered p.o. This dose was 10 mg for one subject, 15 mg for 10 subjects and 22.5 mg for 14 subjects. However, 8 hr before receiving this dose, the subjects received a placebo/quinidine sulfate (100 mg) capsule p.o., in a randomized double-blinded order.

**General procedures.** Subjects participated as outpatients at the Clinical Research and Treatment Institute, Addiction Research Foundation (Toronto, Canada), using a study room equipped with a bed and a computer. They were told that the purpose of the study was to determine how the drug affected their mood and behavior. Subjects were instructed to fast overnight before each study day. Typically, subjects were tested weekly, although testing sessions could be scheduled 3 to 21 days apart.

On subject arrival each day, a urine sample for drug screening was obtained before hydrocodone administration and was tested for benzodiazepines, barbiturates, cannabis, cocaine, amphetamines and opiates, and a Breathalyzer sample was measured. No subject required rescheduling or termination because of these test results. During the study days, subjects received a light standardized breakfast and lunch. Juice and water were available throughout the study day.

On each study day, a battery of objective and subjective tests was performed, twice at base line and at 30, 60, 105 and 180 min after hydrocodone administration. All measures were recorded directly into the computer (Kaplan, 1992, 1996). Subjects and experimenters responded to questionnaires using a light pen. Each test cycle required approximately 20 min to complete.

Each test cycle included both objective and subjective measures. The principal objective measure was pupil constriction, assessed using a custom-built system based on digitized video images (MacLean and Frecker, 1992). Heart rate, respiration rate, blood pressure and pulse oximetry were also measured, using a Hewlett

Packard 78354C automatic patient monitor. The principal subjective measure was the ARCI, with five scales from the 49-item short form (Martin *et al.*, 1971) plus seven derived by Cole *et al.* (1982), for a total of 77 questions. Other subjective measures were the Profile of Mood States, the Johanson and Uhlenhuth (1980) modification of a 72-adjective check list, and a collection of four- to seven-step rating scales, such as drug liking, anxiety and nausea, analyzed as individual items. In addition, the experimenters rated externally evident drug effects, such as motor ataxia and eye signs of intoxication, on additional four- or seven-step scales.

Urine was collected for 8 hr after drug administration and was analyzed for hydrocodone and hydromorphone according to the method previously reported (Otton *et al.*, 1993). The hydrocodone MR was computed as urinary [hydrocodone]/[hydromorphone].

**Statistical analyses.** All statistics were computed using the general linear models procedure of SAS, version 6.04, under MS-DOS (SAS Institute Inc., 1988). The statistical significance criterion was  $P \leq .05$ .

Data were prepared for analysis by imputation of missing data and extraction of post-drug peak scores. Missing or clearly unreasonable data (*e.g.*, because of technical malfunctions) were replaced with imputed values: within each drug condition (*e.g.*, repeat of the favorite dose without quinidine), data were fit to a general linear model with only subject and cycle as terms, and the predicted value from the model was used to replace any missing values. This imputation was performed using the pooled EM and PM subjects, after preliminary analyses showed that the two groups almost never differed. Data from 3 days on which the urine assays were inconsistent with the hydrocodone doses were eliminated from further analysis.

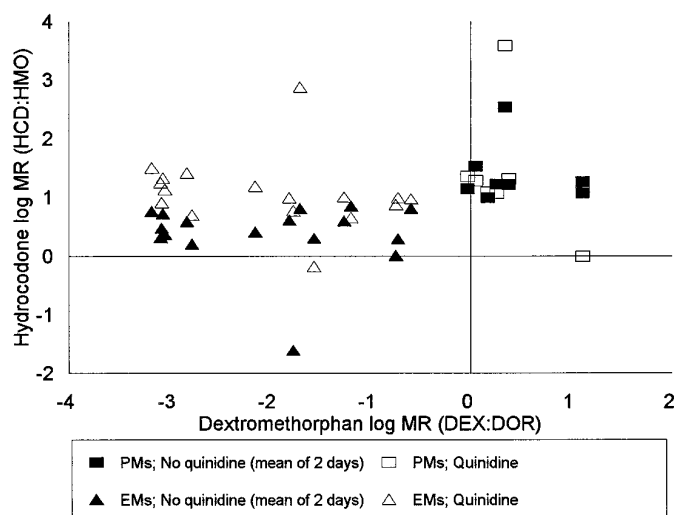
For each measure, the peak was computed in the direction of the typical opiate effect (*e.g.*, a minimum for pupil diameter but a maximum for the ARCI MBG scale). The mean of each day's two base-line cycles was subtracted from each cycle's score and from the peak, to yield a change from base line, and then the grand mean base-line score was added back to restore these base line-adjusted scores to the same scale as the original measurements.

The data for each post-drug cycle separately and for the peak were analyzed in models having terms for metabolic status (EM or PM) as a between-subjects factor, drug condition as a within-subjects factor and their interaction. This was done separately for two groupings of four drug conditions, *i.e.*, the four dose-ranging days of 0, 10, 15 and 22.5 mg, to test for a hydrocodone effect, and the 4 days of 0 mg, the favorite dose on original presentation, the repeat with quinidine and the repeat without quinidine, to test for quinidine modulation of that effect. If the CYP2D6 enzyme is responsible for the effects of hydrocodone in EMs, then quinidine should attenuate of the scores of EMs but not PMs, changing the scores of EMs in the direction opposite to the usual effect of hydrocodone; this will be detected as an interaction of metabolic status with quinidine/placebo.

## Results

**Relationship between dextromethorphan and hydrocodone MRs.** Figure 1 shows the MRs for both dextromethorphan and hydrocodone, the latter under both placebo and quinidine conditions. Considering only the placebo condition, the regression of hydrocodone MR on dextromethorphan MR across the pooled EM and PM groups was statistically significant (slope = 0.24,  $P = .0025$ ); however, within each of the EM and PM groups, there was no relationship between the dextromethorphan and hydrocodone MRs (EM, slope =  $-0.01$ ,  $P = .89$ ; PM, slope =  $-0.12$ ,  $P = .82$ ).

**Dose and time hydrocodone effects.** Orderly dose- and time-related responses to hydrocodone were observed in pupil constriction, an objective measure of opioid effects (fig. 2a), and in subjective measures such as the ARCI MBG scale

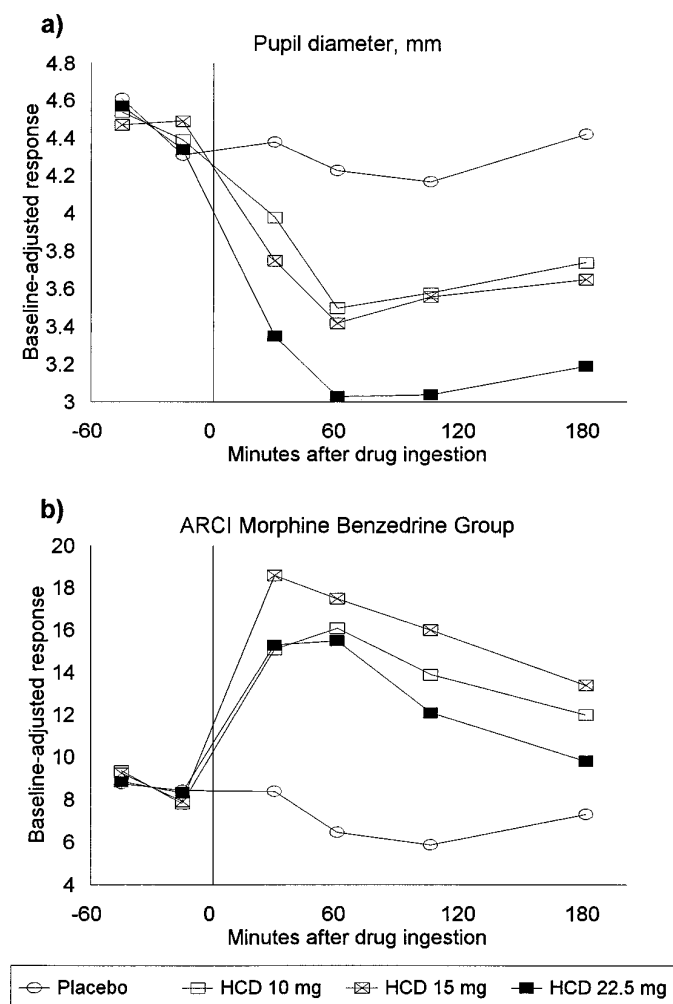


**Fig. 1.** Hydrocodone MR as a function of dextromethorphan MR and quinidine condition. The placebo points are the mean of the original and placebo repeat presentations of the favorite dose of hydrocodone. EM and PM hydrocodone log MRs differed under placebo but not quinidine pretreatment (interaction  $P = .02$ ).

(fig. 2b). For pupil constriction, all doses differed from placebo ( $P \leq .05$ ) at all four times tested and at the peak, and responses increased with dose. For the MBG scale, wide interindividual variations were observed, and the dose-response relationship was not monotonic; hydrocodone subjective effects differed from placebo except at 3 hr ( $P \leq .05$ ), but there was little difference in effect among doses, with the intermediate dose (15 mg) eliciting the largest effects. Hydrocodone also produced elevations in numerous other subjective effects measures, including measures of positive effects, such as the Cole/ARCI abuse potential scale, the Profile of Mood States elation scale and individual drug liking questions, and measures of negative effects, such as the Cole/ARCI sedation-motor, sedation-mental and unpleasantness-dysphoria scales. The observer-rated scales were generally consistent with the subjective scales. In the objective measures other than pupil diameter, hydrocodone produced transient elevations in systolic blood pressure and finger temperature. The results reported below are all based on the peak responses; analysis of individual time points did not reveal any patterns other than those evident in the peak responses.

**Effects of quinidine on the MR.** Quinidine (100 mg) produced inhibition of CYP2D6 activity, as shown in figure 1. For most EM subjects, the MR was increased after quinidine; for two, the MR was slightly decreased. Quinidine essentially abolished the EM/PM difference in the hydrocodone MR; the regression of hydrocodone MR on O-demethylation MR for quinidine days was not significant (slope = 0.01,  $P = .90$ ), and the interaction of EM/PM status with quinidine was significant ( $P = .02$ ), confirming the larger quinidine-induced changes for EMs than for PMs.

**Hydrocodone effects in EMs and PMs.** No significant differences were found between EMs and PMs in either objective or subjective effects of hydrocodone (figs. 3 and 4). Significant effects of the hydrocodone dose were found for daily minimum pupil diameter and daily maximum ARCI MBG scale results (dose main effect,  $P < .0001$ ), among other measures, but no phenotype differences were detected (EMs



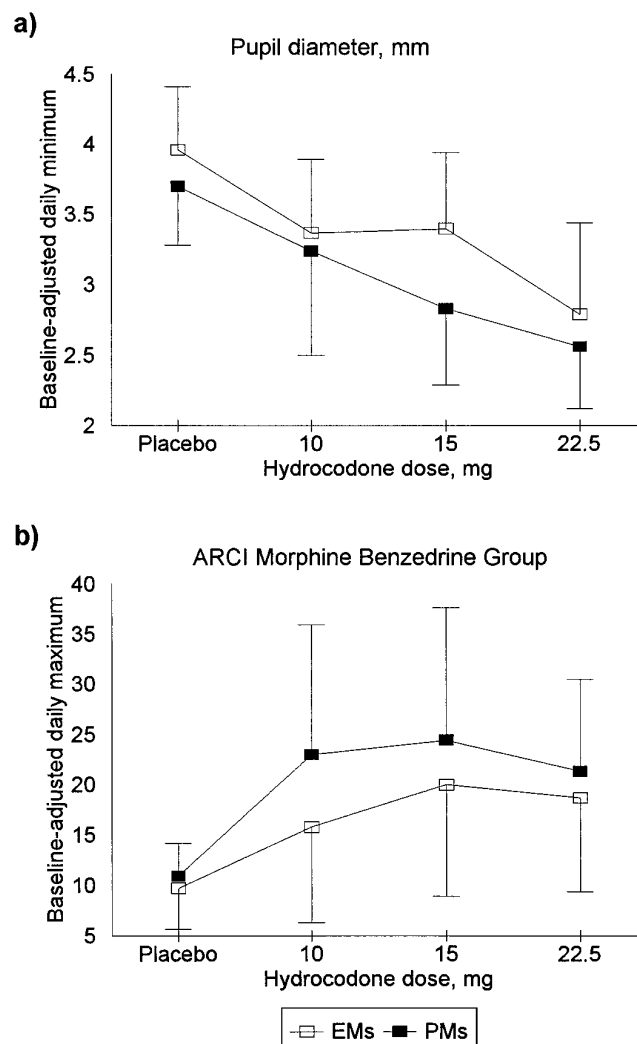
**Fig. 2.** Dose and time hydrocodone (HCD) effects on pupil diameter and the ARCI MBG scale responses, pooling the EM and PM data, which were not significantly different. All active dose responses differed from placebo ( $P \leq .05$ ) except for the ARCI MBG scale responses at 180 min.

*vs.* PMs main effects, not significant; interaction with hydrocodone dose, not significant.).

**Effects of hydrocodone with and without quinidine.** The peak pupil constrictions were almost identical after the favorite dose of hydrocodone on its original presentation and its re-presentations, with and without quinidine pretreatment (placebo *vs.* hydrocodone, main effect,  $P < .0001$ ; with *vs.* without quinidine, not significant); similar patterns were found for subjective measures such as the ARCI MBG scale (fig. 4). The proportion of measures on which any quinidine effect could be detected was smaller than the 5% expected by chance.

### Discussion

Hepatic conversion of hydrocodone to hydromorphone is of almost no consequence in determining the effects of hydrocodone. Active CYP2D6 is required for that metabolism, as measured by the urinary MR, and quinidine causes EMs to temporarily metabolize, excrete and behave as PMs. However, EMs with active enzyme, EMs with inhibited enzyme and PMs all respond virtually identically to hydrocodone with respect to pupil diameter and subjective measures. It is not because the measures are insensitive or the sample size

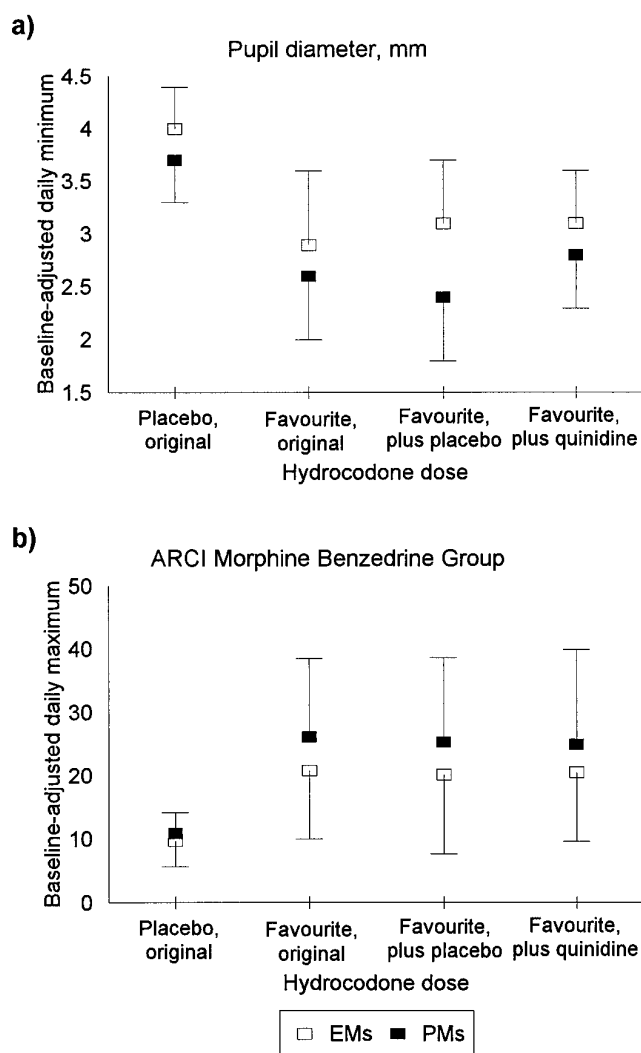


**Fig. 3.** Peak hydrocodone effects on pupil diameter and the ARCI MBG scale in EMs and PMs during the dose-ranging phase. In both panels, the hydrocodone effect was significant ( $P = .0001$ ), but neither the main effect of metabolic status nor its interaction with hydrocodone was significant. Error bars, 1 S.D. of the peak response.

is small that no difference was found; hydrocodone *vs.* placebo differences were easily detected, but not hydrocodone *vs.* hydrocodone differences which depend on the presence *vs.* absence of active CYP2D6 enzyme.

This lack of behavioral differences between EMs and PMs for hydrocodone effects, as found in the current experiment, is not unique. Other work done in rhesus monkeys and in Wistar rats also showed that the behavioral effects of hydrocodone were not modified by marked inhibition of the conversion of hydrocodone to hydromorphone (T.A. Berns, W.A. Corrigan, S.V. Otton and E.M. Sellers, unpublished poster, 1996; France *et al.*, 1996).

One reason for the lack of detectable difference is the limited range of hydrocodone MRs, due to genetic variability or pharmacological intervention. In a histogram of dextromethorphan *O*-demethylation MRs, EMs and PMs form two distinct modes, covering a 10,000-fold range. The hydrocodone MRs in this study formed a single mode with only a 15-fold range, with PMs and EMs concentrating at different ends of the distribution. There was no antimode, even though



**Fig. 4.** Peak effects of placebo and the favorite hydrocodone dose on pupil diameter and the ARCI MBG scale. In both panels, the placebo vs. pooled hydrocodone main effect was significant ( $P = .0001$ ), but the main effect of quinidine within the hydrocodone days was not. For pupil diameter, but not MBG scale results, there was a main effect of metabolic status ( $P = .024$ ); metabolic status did not interact with the hydrocodone or quinidine effects. Error bars, 1 S.D. of the peak response.

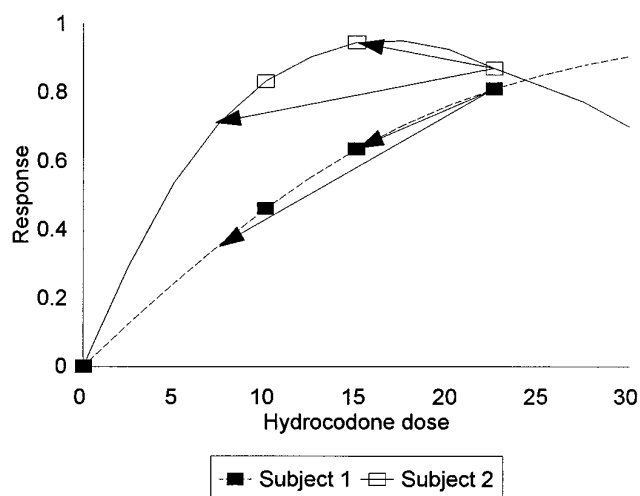
we guaranteed by recruitment that the distribution would include dextromethorphan PMs. As a substrate, dextromethorphan is unusual in its ability to separate such a population; for most CYP2D6 substrates, the range of kinetic responses is much more compressed (Yue *et al.*, 1991). Furthermore, an MR axis is not an arbitrary scale for discriminating among subpopulations; the numeric values are meaningful. As shown in figure 1, the log hydrocodone MR for EMs without quinidine is typically about +0.5, which means that the urinary concentration of hydrocodone is approximately 3 times that of hydromorphone. At best, only one-fourth of the parent drug is converted to hydromorphone. These results are consistent with those we reported earlier (Otton *et al.*, 1993), in which plasma concentrations of hydrocodone in EMs were approximately 4 times as large as those of hydromorphone. Cone *et al.* (1978) also reported that the amount of hydromorphone found even in EMs is relatively small, representing only 4.6% of the total clearance. Even

EMs may not produce enough hydromorphone to cause differences in the effects. Hydrocodone, although less potent than hydromorphone, clearly has agonist actions, and perhaps plasma hydrocodone concentrations are high enough to account for the observed pharmacological effects, with the low plasma concentrations of hydromorphone contributing only modestly to these effects.

Although the receptor binding of hydromorphone is greater than that of hydrocodone (Hennies *et al.*, 1988; Chen *et al.*, 1991a), morphinan precursors such as hydrocodone enter the brain faster and to a greater extent than do their *O*-demethylated metabolites (Wu *et al.*, 1995). Thus, the effective contribution of hydromorphone may be less than its plasma concentration suggests. In addition, brain CYPs appear to be regulated and expressed differently than hepatic CYPs (Joharchi *et al.*, 1995; Li *et al.*, 1996). Hydrocodone produces hydromorphone and four other metabolites in rat brain homogenates, which is qualitatively and quantitatively different from the pattern produced by rat liver microsomes. Conversion in the brain among hydrocodone metabolites of different activity could account for the lack of correspondence between plasma hydromorphone concentrations and observed responses.

If plasma hydromorphone were the critical determinant of hydrocodone responses, then one might expect quinidine inhibition of the metabolism to be equivalent to removing most of the original hydrocodone dose, returning responding nearly to placebo levels. However, given the relatively small actual importance of plasma hydromorphone, quinidine inhibition is equivalent to only a minor lowering of the effective hydrocodone dose, moving leftward only a small distance along the dose-response curve. In such circumstances, the overall slope of the dose-response curve (greater response for a greater dose) is less important than the individual local slope. At what we called their favorite doses, many subjects were already on a declining portion of the dose-response curve for some or all measures; for such measures, attenuating the effective hydrocodone dose could result in an augmentation, not a diminution, of their response, as shown in figure 5. Thus, the direction of response may be inconsistent across subjects, and this would not be detected by conventional analysis. In figures 2 and 3, the subjective response to hydrocodone at 15 mg is slightly greater than that to either 10 or 22.5 mg; this is consistent with the mean dose-response slope around 15 mg being nearly 0, possibly because it is negative in some subjects and positive in others.

When we chose each subject's favorite dose for retesting, we selected the one dose (of three possibilities) for which the overall positive response was greatest, to maximize the power of the placebo vs. drug retest. This optimization, however, almost eliminated the power of the quinidine effect test, by minimizing the local slope of the dose-response curve, which (given the small reduction in effective hydrocodone dose) is important for detecting quinidine effects. We can deduce that the local slope for the subjective response was most often flat or negative for subjects whose favorite dose was 10 or 15 mg, because they liked the higher dose less, and possibly also for many subjects whose favorite dose was 22.5 mg, because we never tested any higher doses. In the study by Otton *et al.* (1993), in contrast, all subjects were tested with 10 mg of hydrocodone. In the current study, the dose-response slope was less consistently positive at the favorite doses actually chosen than at 10 mg, which may



**Fig. 5.** Hypothetical dose-response curves for two subjects, showing the effects of two different treatments that reduce the effect of a 22.5-mg reference dose. The doses used in this study are indicated (squares). For each subject, one treatment reduces the effects to match those of a 15-mg dose, the other to match a 7.5-mg dose. For subject 1 both treatments decrease the response, but for subject 2 the treatment causing a smaller change in the effective dose actually increases the response.

account for the failure to replicate the earlier results for subjective measures.

This argument suggests a different strategy for selecting a dose for retesting in an enzyme inhibition study such as this one. Instead of selecting either the favorite dose or the maximum tolerable dose (another option we considered), it might be best to select the dose of the greatest positive slope in the dose-response curve. Such a strategy would be optimized for the detection of small changes in the effective dose, at the possible sacrifice of some power to detect differences from placebo.

Neither quinidine nor any other CYP2D6 inhibitor, used on a schedule that affects liver but not brain CYPs, is likely to be of any practical use in attenuating the effect of oral hydrocodone, in either experimental or clinical settings. This does not rule out the possibility of these inhibitors being of use in other situations where they can affect opiate metabolites that are more important determinants of the response. For example, we have conducted a study of codeine abuse liability and seen some quinidine modulation of the responses of EMs (K. Kathiramalainathan, H.L. Kaplan, U.E. Busto, M.K. Romach, R.F. Tyndale and E.M. Sellers, unpublished poster presentation, 1966).

Further studies to identify hydrocodone metabolites in brain regions are needed to identify the CYPs or other metabolizing enzymes and their relationship to liver microsomal enzymes. These studies may help to better explain the discrepancy between the great theoretical importance of plasma hydromorphone and its slight practical importance in oral hydrocodone effects. Finally, some of these studies should also be conducted with oral opiate abusers, using a large range of test doses appropriate to their drug tolerance.

#### References

- BERTILSSON, L., DAHL, M.-L., EKQVIST, B., JERLING, M. AND LIERENA, A.: Genetic regulation of the disposition of psychotropic drugs. *In* Current Practices and Future Developments in the Pharmacotherapy of Mental Disorders, ed. by H. Y. Meltzer and D. Nerozzi, pp. 73–80, Elsevier Scientific Publications, Amsterdam, 1991.
- CHEN, Z. R., IRVINE, R. J., SOMOGYI, A. A. AND BOCHNER, F.: *Mu* receptor binding

of some commonly used opioids and their metabolites. *Life Sci.* **48**: 2165–2171, 1991a.

- CHEN, Z. R., SOMOGYI, A. A., REYNOLDS, G. AND BOCHNER, F.: Disposition and metabolism of codeine after single and chronic doses in one poor and seven extensive metabolizers. *Br. J. Clin. Pharmacol.* **31**: 381–390, 1991b.
- COLE, J. O., ORZACK, M. H., BEAKE, B., BIRD, M. AND BAR-TAL, Y.: Assessment of the abuse liability of bupirone in recreational sedative users. *J. Clin. Psychiatry* **43**: 69–75, 1982.
- CONE, E. J. AND DARWIN, W. D.: Simultaneous determination of hydromorphone, hydrocodone and their 6- $\alpha$ - and 6- $\beta$ -hydroxy metabolites in urine using selected ion recording with methane chemical ionization. *Biomed. Mass Spectrom.* **5**: 291–295, 1978.
- CONE, E. J., DARWIN, W. D., GORODETZKY, C. W. AND TAN, T.: Comparative metabolism of hydrocodone in man, rat, guinea pig, rabbit and dog. *Drug Metab. Dispos.* **6**: 488–493, 1978.
- DESMEULES, J., GASCON, M. P., DAYER, P. AND MAGISTRIS, M.: Impact of environmental and genetic factors on codeine analgesia. *Eur. J. Clin. Pharmacol.* **41**: 23–26, 1991.
- EICHELBAUM, M. AND GROSS, A. S.: The genetic polymorphism of debrisoquine/sparteine metabolism: Clinical aspects. *Clin. Pharmacol. Ther.* **46**: 377–394, 1990.
- FRANCE, C. P., WEGERT, S., OTTON, S. V. AND SELLERS, E. M.: Inhibition of CYP2D6 does not alter the behavioral effects of hydrocodone in rhesus monkeys. *Natl. Inst. Drug Abuse Res. Monogr.* **162**: 103, 1996.
- HEIM, M. AND MEYER, U. A.: Genotyping of poor metabolizers of debrisoquine by allele-specific PCR amplification. *Lancet* **336**: 529–532, 1990.
- HENNIES, J. J., FRIDERICH, F. AND SCHNEIDER, J.: Receptor binding, analgesic and antitussive potency of tramadol and other selected opioids. *Arzneim. Forsch.* **38**: 877–880, 1988.
- HIMMELSBACH, C. K.: The effects of certain chemical changes on the addiction characterization of drugs of the morphine, codeine series. *J. Pharmacol. Exp. Ther.* **71**: 42–48, 1941.
- INABA, T., TYNDALE, R. F. AND MAHON, W. A.: Quinidine: Potent inhibition of sparteine and debrisoquine oxidation *in vivo*. *Br. J. Clin. Pharmacol.* **22**: 199–200, 1986.
- JAFFE, J. H. AND MARTIN, W. R.: Opioid analgesics and antagonists. *In* The Pharmacological Basis of Therapeutics, ed. by L. S. Goodman, A. Gilman, T. W. Rall, A. S. Nies and P. Taylor, pp. 485–521, Pergamon Press, New York, 1990.
- JOHANSON, C. E. AND UHLENHUTH, E. H.: Drug preference and mood in humans: Diazepam. *Psychopharmacology* **71**: 269–273, 1980.
- JOHARCHI, N., SELLERS, E. M., BERNIS, T. A., OTTON, S. V. AND BALSTER, R. L.: Inhibition of CYP2D1 enzyme activity alters hydrocodone metabolism but not drug discrimination in the rat (Abstract). *Natl. Inst. Drug Abuse Res. Monogr.* **153**: 188, 1995.
- KAPLAN, H. L.: Representation of on-line questionnaires in an editable, auditable database. *Behav. Res. Methods Instruments Comput.* **24**: 373–384, 1992.
- KAPLAN, H. L.: SMS: A software strategy for acute effects studies. *Natl. Inst. Drug Abuse Res. Monogr.* **162**: 188, 1996.
- LEEMAN, T., DAYER, P. AND MEYER, U. A.: Single-dose quinidine treatment inhibits metoprolol oxidation in extensive metabolizers. *Eur. J. Clin. Pharmacol.* **29**: 739–741, 1986.
- LI, N.-Y., CHEUNG, S. W., TYNDALE, R. F. AND SELLERS, E. M.: Evidence for conversion of hydrocodone to hydromorphone by rat brain (Abstract). *Natl. Inst. Drug Abuse Res. Monogr.* **162**: 254, 1996.
- MACLEAN, W. J. AND FRECKER, R. C.: Clinical pupillometry system running under MS-Windows (Abstract 2272–58). *In* Proceedings of the Investigative Ophthalmology and Visual Science Annual Meeting (Sarasota, FL), 1992.
- MARTIN, W. R., SLOAN, J. W., SAPIRO, J. D. AND JASINSKI, D. R.: Physiologic, subjective and behavioural effects of amphetamine, methamphetamine, ephedrine, phenmetrazine and methylphenidate in man. *Clin. Pharmacol. Ther.* **12**: 245–258, 1971.
- OTTON, S. V., SCHADEL, M., CHEUNG, S. W., KAPLAN, H. L., BUSTO, U. E. AND SELLERS, E. M.: CYP2D6 phenotype determines the metabolic conversion of hydrocodone to hydromorphone. *Clin. Pharmacol. Ther.* **54**: 463–472, 1993.
- SAS INSTITUTE INC.: SAS/STAT User's Guide, release 6.03 edition, SAS Institute Inc., Cary, NC, 1988.
- SCHADEL, M., WU, D., OTTON, S. V., KALOW, W. AND SELLERS, E. M.: Pharmacokinetics of dextromethorphan and metabolites in humans: Influence of the CYP2D6 phenotype and quinidine inhibition. *J. Clin. Psychopharmacol.* **15**: 263–269, 1995.
- SPEIRS, C. J., MURRAY, S., BOOBIS, A. R., SEDDON, C. E. AND DAVIES, D. S.: Quinidine and the identification of drugs whose elimination is impaired in subjects classified as poor metabolizers of debrisoquine. *Br. J. Clin. Pharmacol.* **22**: 739–743, 1986.
- WU, D. F., OTTON, S. V., KALOW, W. AND SELLERS, E. M.: Effects of route of administration on dextromethorphan pharmacokinetics and behavioral response in the rat. *J. Pharmacol. Exp. Ther.* **274**: 1431–1437, 1995.
- YUE, Q. Y., HASSELSTROM, J., SVENSSON, J. O. AND SAWE, J.: 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, 1991.

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