That EE itself is a weak inhibitor (nM (Belle et al., 2002). Moreover, Jurima et al. (1985) have reported and peak plasma concentrations (total EE) range between 0.6 and 0.7
crease CYP2C19 activity. For example, Palovaara et al. (2003) eval-
ations containing EE decrease CYP2C19 activity. The observations of
the plasma concentration vs. time curve of omeprazole (38%) and the plasma concentration vs. time curve ratio (48%). LNG alone had no affect on the PK and metabolism of omeprazole. In addition, neither formulation in-
hibited CYP3A4-catalyzed omeprazole sulfone formation. Because the hydroxylation of omeprazole is widely accepted as an index of CYP2C19 activity (Chang et al., 1995; Lasker et al., 1998; Abel et al., 2000; Kita et al., 2001), the authors concluded that OC prepara-
tions decrease CYP2C19 activity. The observations of Palovaara et al. (2003) confirm the findings of Laine et al. (2000), who also reported an increase (~100%) in the omeprazole to 5-hy-
droxy omeprazole ratio in plasma. In the same study, the ratio of (S)-mephénytoin to (R)-mephénytoin in urine (S/R ratio) increased from 0.11 to 0.28, and the effect was similar to that observed with subjects genotyped CYP2C19*/1/2 (versus CYP2C19*/1/*). A comparable change in the S/R ratio has been reported by Hagg et al. (2001) and Tamminga et al. (1999).

EE-containing OC formulations have also been shown to affect the PK and metabolism of diazepam, propranolol, proguanil, and selegi-
line (Abernethy et al., 1982; Walle et al., 1996; Laine et al., 1999; McGreedy et al., 2003). All four drugs are reported to be CYP2C19 substrates. However, the effect of EE on their PK cannot be ascribed to CYP2C19 alone, because other cytochromes P450 are involved in metabolism (Jung et al., 1997; Yang et al., 1999; McGinnity et al., 2000; Hidestrand et al., 2001). The same cannot be said for the urinary mephénytoin S/R ratio and the omeprazole to 5-hydroxy omeprazole ratio in plasma. Both have served as a useful index of CYP2C19 activity and have been validated with genotyped subjects (Rodrigues and Rushmore, 2002).

Although the results of these various clinical drug interaction studies are compelling, it cannot be assumed that EE is a clinically relevant inhibitor of CYP2C19. The dose of EE is low (30–50 μg), and peak plasma concentrations (total EE) range between 0.6 and 0.7 nM (Belle et al., 2002). Moreover, Jurima et al. (1985) have reported that EE itself is a weak inhibitor (K \text{I} \approx 100 \mu M) of human liver microsomal mephénytoin (racemate) hydroxylase activity, whereas Laine et al. (2003) observed inhibition (70%) of omeprazole 5-hydroxy-
drolyase only at a high concentration of EE (0.1 mM). We have also determined that EE is a relatively weak, reversible inhibitor (IC_{50} = 19 \mu M) of CYP2C19 (4’-hydroxylation of (S)-mephénytoin) in human liver microsomes. The IC_{50} was determined at the K_{m} of (S)-mephénytoin (80 \mu M; substrate concentration/K_{m} \approx 1.0) and (R)-N-3-benzyl-phenobarbital (IC_{50} = 0.3 \mu M) served as a positive con-
trol. Therefore, these data imply that the concentration of inhibitor/K_{I} for EE is very low.

EE has been shown to be a mechanism-based inhibitor (K \text{I} \approx 18 \mu M; k_{\text{inact}} = 0.04 \text{ min}^{-1}) of CYP3A4 in vitro (Lin et al., 2002). In our hands, up to 70% inhibition of testosterone 6β-hydroxylase activity was observed when EE (50 μM) was preincubated (30 min) with NADPH-
fortified human liver microsomes. In contrast, preincubation of EE (0.1 to 50 μM) with human liver microsomes resulted in no inhibition of (S)-mephénytoin 4’-hydroxylase activity. Ticlopidine (10 μM), on the other hand, behaved as a preincubation time-dependent inhibitor (50% inhibition) and also served as a positive control (Tateishi et al., 1999; Ko et al., 2000; Ha-Duong et al., 2001). Based on our preliminary data, therefore, the k_{\text{inact}}/K_{I} ratio of EE for CYP2C19 is probably lower than that for CYP3A4 (0.002 min^{-1} · μM^{-1}). It is worth noting that despite overt mechanism-based inhibition of CYP3A4 in vitro, EE has a modest effect on the PK and metabolism of midazo-
lam, a sensitive CYP3A4 probe drug (Palovaara et al., 2000; Belle et al., 2002; Lin et al., 2002). The in vitro study described herein was expanded to include a number of EE metabolites (e.g., EE 3-O-sulfate, EE 3-O-glucuronide, and 2-methoxy EE), and none were shown to be inhibitors of human liver microsomal (S)-mephénytoin 4’-hydro-
xylase activity.

At first glance, it is difficult to conclude that the effect of OCs on CYP2C19 activity is due to inhibition (reversible or mechanism-
based) of the enzyme by EE. Further studies are needed to elucidate the mechanism of interaction involving omeprazole and mephénytoin. Toward this end, it will be important to evaluate carefully the estrogenic and progestogenic components of OC formulations and their metabolites, as reversible and mechanism-based inhibitors of CYP2C19 in vitro. In turn, in vitro-in vivo correlations can be at-
tempted. Such studies are important because it has been estimated that up to 70 million women worldwide take OC formulations (Belle et al., 2002), and relatively little mechanistic information is available concerning the effects of these formulations on cytochromes P450.

Department of Drug Metabolism, A. DAVID RODRIGUEZ A. DAVID RODRIGUEZ
Merck Research Laboratories, PING LU
West Point, PA

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Hidestrand M, Oscarson M, Salonen JS, Nyman L, Pelkonen O, Turpeinen M, and Ingelman-

\[ a \] Abbreviations used are: OC, oral contraceptive; EE, 17α-ethinyl estradiol; LNG, levonorgestrel; PK, pharmacokinetics; S/R ratio, ratio of (S)-mephénytoin to (R)-mephénytoin in urine.

\[ b \] Present address: Drug Metabolism and Pharmacokinetics, Bristol-Myers Squibb, Pharmaceutical Research Institute, Princeton, NJ.
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