ABSTRACT:

To determine the effect of age on exposure to the circulating major verapamil metabolites norverapamil, \(N\)-dealkylnorverapamil (D-617), and \(N\)-dealkylverapamil (D-620), plasma concentrations of verapamil and the three metabolites were determined during the last dose interval of a 14-day administration period of 240 mg of sustained release verapamil once daily in 11 older (aged 65–75 years) and 8 younger (20–28 years) healthy male volunteers. Area under the plasma concentration time curve (AUC) was greater for verapamil (mean ± S.D.) (2815 ± 733 older versus 1639 ± 466 ng/ml h\(^{-1}\) young; \(P < .0007\)) and norverapamil (2927 ± 655 versus 2143 ± 471 ng/ml h\(^{-1}\); \(P < .007\)); however, it was not significantly different for D-617 [2386 ± 772 versus 1894 ± 418 ng/ml h\(^{-1}\)]; not significantly different (NS) and \(N\)-dealkynorverapamil (897 ± 366 versus 757 ± 104 ng/ml h\(^{-1}\); NS) in older as compared with young subjects. These data indicate that impaired verapamil oral clearance previously described in older men does not result in decreased exposure to the formed major metabolites, rather there is increased exposure to norverapamil and the same or a trend toward greater exposure to D-617 as well. This suggests that in addition to the impaired clearance mechanisms for verapamil, which are thought to be primarily mediated by CYP3A, biotransformation processes distal to the formation of norverapamil and D-617 are impaired as well.

Verapamil disposition clearance has been described as decreased in older as compared with younger subjects and patients (Abernethy et al., 1986, 1993; Schwartz, 1990). This is apparently true after single doses of either i.v. or oral verapamil and during chronic i.v. infusion or oral administration of verapamil as would more commonly occur during therapy. Verapamil biotransformation in humans is predominantly mediated by CYP3A species, with gut wall CYP3A thought to contribute substantially to oral clearance (Kroemer et al., 1992, 1993; Fromm et al., 1996). Previous reports indicate that formation of D-617 (\(N\)-dealkylnorverapamil)\(^1\) may be in part mediated by CYP3A2 (Kroemer et al., 1992); however, the involvement of CYP2C8 has also been implicated in the formation of both D-617 and D-620 (\(N\)-dealkylnorverapamil) in human liver microsomal studies (Tracy et al., 1999). In addition, indirect studies have implicated hepatic flavin-containing monoxygenase in the \(N\)-oxygenation reaction that leads to formation of both D-617 and D-620 (Cashman, 1989). Therefore, it remains uncertain which drug metabolizing enzyme activities may be decreased in older individuals, and which result in the observed decreases in parent drug verapamil clearance. Although norverapamil has usually been reported as the formed metabolite that attains substantial plasma concentrations in humans after verapamil exposure (Abernethy et al., 1984), D-617 and D-620 have also been noted in patients and healthy volunteers in concentrations of the same magnitude as verapamil and norverapamil (Barbieri et al., 1985; Padrini et al., 1985; Piotrovskii et al., 1986).

Pharmacological activity of the metabolites has been studied in the dog (Neugebauer, 1978) and perfused rabbit heart (Johnson et al., 1991). Norverapamil may have about 20% of the potency of verapamil to block cardiac atrioventricular conduction and mediate coronary vasodilatation in these preparations, whereas D-617 may have limited effects on cardiac contractility and D-620 may have some effect on slowing atrioventricular conduction. Both are much less potent than parent drug verapamil and somewhat less potent than norverapamil for any of the measured pharmacodynamic effects.

This study is a further analysis of our previous report (Abernethy et al., 1993), which noted markedly decreased clearance of both verapamil enantiomers in older as compared with young healthy male volunteers (Table 1). Here we report the analysis of plasma concentrations of racemic verapamil, norverapamil, D-617, D-620, (2-[4-hydroxy-3-methoxyphenyl]-8-[3,4-dimethoxyphenyl]-6-methyl-2-isopropyl-6-azaoctanitrile) (PR-22), and ([3,4-dimethoxyphenyl]-acetic acid) (PR-25) and note that exposure to all detectable species is substantially greater (verapamil, norverapamil) or tending to be greater (D-617, D-620) in older individuals.

Materials and Methods

Samples were analyzed from 11 older (aged 65–75 years) and 8 younger (aged 20–28 years) healthy males who were on no medications and in good health. All were nonsmokers and of similar height (67–75 inches, older; 68–76
Subject characteristics

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Older N = 11</th>
<th>Younger N = 8</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>69 (65–75)</td>
<td>23 (20–26)</td>
</tr>
<tr>
<td>Weight (kilograms)</td>
<td>81 (69–100)</td>
<td>77 (61–91)</td>
</tr>
<tr>
<td>Body mass index (kilograms per square meter)</td>
<td>26.2 (22.6–30.5)</td>
<td>23.0 (20.5–25.7)</td>
</tr>
</tbody>
</table>

Species measured in this study were verapamil, norverapamil, D-617, D-620, PR-22, and PR-25. PR-22 and PR-25 were not detectable in any subject (<10 ng/ml at all time points).

**Results**

The human metabolic pathway of verapamil is shown in Fig. 1. The plasma concentration data reported here represent exposure to either
verapamil or the metabolite, and reflect both formation and clearance rate of these various intermediate metabolites. Plasma concentrations of verapamil, norverapamil, D-617, and D-620 at each time point are shown in Fig. 2(A–D). At the same dose, older subjects appear to have greater exposure to verapamil and norverapamil, and the same or somewhat greater exposure to D-617 and D-620 as compared with younger subjects. As previously reported, intersubject variability is large for both groups. To compare exposure to the various species within a subject age group, mean concentrations of each are demonstrated in Fig. 3 (A and B). Within each group, exposure to verapamil, norverapamil, and D-617 is similar, and to D-620 approximately one-third that of the other species. AUC comparisons reflected these observations, with exposure to verapamil and norverapamil greater in older subjects and exposure to D-617 and D-620 tending to be greater in older subjects, although this was not statistically significant (Table 2; Fig. 4). To further explore the relative changes in metabolite formation as reflected by steady-state plasma concentrations in older versus younger subjects, the metabolite AUC/verapamil AUC ratio was determined and compared between groups (Fig. 5). The ratio of norverapamil to verapamil AUC was decreased in older subjects, as was that of D-620 to verapamil AUC, whereas that of D-617 to verapamil AUC tended to be lower in older subjects, although not significantly so. There are many limitations on interpretation of such data; however, they suggest in general that the greater exposure to norverapamil and tendency to greater exposure to D-617 and D-620 in older subjects is due to decreased verapamil clearance and the attendant higher concentrations. Additionally, such findings support the possibility that enzyme activities that mediate clearance of norverapamil, D-620, and perhaps D-617 are decreased in older as compared with younger subjects.

**Discussion**

These data indicate that with chronic dosing of sustained release verapamil at a dose of 240 mg/day healthy older men have greater exposure to verapamil and norverapamil, tend to have increased exposure to D-617 and D-620 as compared with young men. The clinical importance of such an observation is unknown, but unlikely to be associated with the altered verapamil responses observed in older individuals (Abernethy et al., 1986, 1993; Schwartz, 1990) due to the limited pharmacological activity of these metabolites (Neugebauer, 1978; Johnson et al., 1991). In contrast, these findings do add insight regarding which enzymes of drug biotransformation have diminished activity with increasing age. CYP3A4 and CYP3A5 have been implicated in the formation of both
norverapamil and D-617 (Kroemer et al., 1992, 1993; Fromm et al., 1996; Tracy et al., 1999) and they have an important role in verapamil gut wall metabolism (Fromm et al., 1996). These findings are consistent with decreased clearance in older male subjects of other CYP3A substrates such as midazolam and triazolam (Greenblatt et al., 1983, 1984). Previous reports of CYP1A2 and CYP2C8 having a role in verapamil and metabolite biotransformation (Kroemer et al., 1992, 1993; Tracy et al., 1999) are difficult to place into the context of the present data as the in vivo contribution of these CYP enzymes is uncertain. Confounding these and other clinical pharmacokinetic findings that rather consistently note decreased CYP3A activity with age is the report of no age-related change in CYP3A activity as measured by erythromycin N-demethylation in resected human liver preparations (Hunt et al., 1992). The more recently described feature of verapamil disposition is the role of P-glycoprotein in its gut absorption and distribution (Doppenschmitt et al., 1999). There are no data

**Fig. 3. Pharmacokinetic curves which represent:**

A, mean verapamil and metabolite concentrations for elderly subjects over one dose interval; B, mean verapamil and metabolite concentrations for young subjects over one dose interval.
available that relate to age-related changes in expression or activity of P-glycoprotein; therefore, any potential contribution to the present data cannot be predicted.

In summary, these data indicate that exposure to not only verapamil, but also its metabolite norverapamil is increased during chronic oral verapamil administration to older subjects. In addition, exposure to D-617 tends to be increased with age and exposure to D-620 is similar in younger and older individuals when the same dose regimen of oral verapamil is administered to both groups, whereas PR-22 and

<table>
<thead>
<tr>
<th>Measured Species</th>
<th>Older Mean ± S.D. (ng/ml · h⁻¹)</th>
<th>Younger Mean ± S.D. (ng/ml · h⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Verapamil</td>
<td>2815 ± 733*</td>
<td>1639 ± 466</td>
</tr>
<tr>
<td>Norverapamil</td>
<td>2927 ± 655**</td>
<td>2143 ± 471</td>
</tr>
<tr>
<td>D-617</td>
<td>2386 ± 772</td>
<td>1894 ± 418</td>
</tr>
<tr>
<td>D-620</td>
<td>897 ± 366</td>
<td>757 ± 104</td>
</tr>
</tbody>
</table>

* P < .0007; ** P < .007.
PR-25 are not detectable in the plasma of older or younger subjects during steady-state oral verapamil treatment.

Acknowledgments. We thank Nektaria Markoglou for assistance in determination of verapamil and metabolite plasma concentrations.

References


