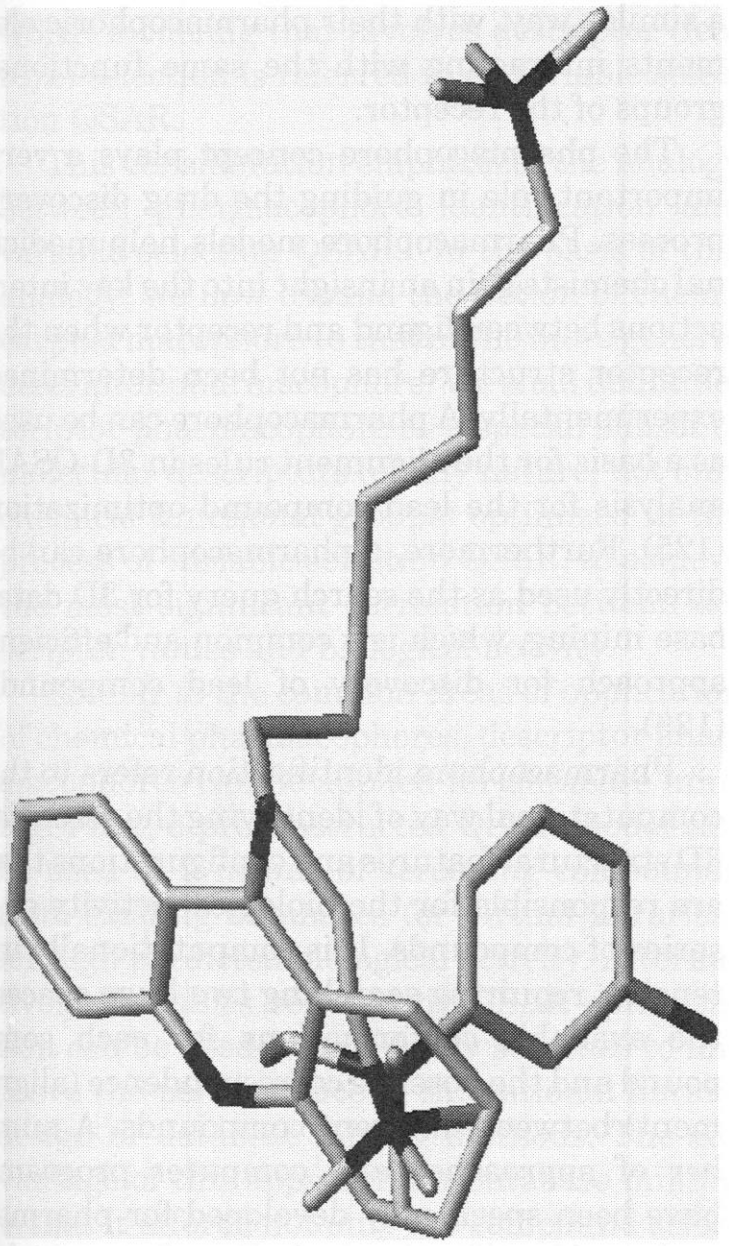

Figure 2.6. Superposition of three inhibitors of AChE in the active site of the enzyme based on crystallographic structures of enzyme-inhibitor complexes. Obviously, no common pharmacophore can be found for these molecules.

ment or even by the researcher's imagination on the basis of the ligand chemical structure alone. This consideration demonstrates the general difficulty of generating a unique and meaningful alignment in 3D-QSAR studies that leads to interpretable and predictive models.

The 3D alignment problem is the main source of ambiguity in obtaining and analyzing CoMFA results, especially in the case of structurally diverse compounds. However, it was also shown that, even if the structural alignment is fixed, the resulting q^2 value could also be sensitive to the orientation of rigidly aligned molecules on the user terminals (75), which can be explained as follows.

The grid orientation in CoMFA is fixed in the coordinate system of the computer; thus, every time when the orientation of the molecular aggregate is changed, the size of the grid may change but not its orientation. The orientation of the assembled molecules therefore affects the placement of probe atoms, which, in turn, influences the field sampling process. This leads to the variability of the q^2 values, mostly attributable to the reasons outlined earlier. The effect of variability of q^2 as a function of molecular aggregate orientation was more pronounced in the case of structurally diverse molecules (e.g., cephalotaxine esters and 5-HT_3 receptor ligands) than in the case of much less structurally diverse molecules (e.g., HIV protease inhibitors) (75). This effect may be attributed to the fact that the pattern of probe atom placement with respect to the aligned molecules changes more dramatically when one changes the orientation of more structurally diverse molecules than it does when the data set is composed of structurally similar molecules.

In the conventional CoMFA implementation, the steric and electrostatic fields, which theoretically form a continuum, are sampled on a fairly coarse grid. As a result, these fields are represented inadequately, and the results are not strictly reproducible. Intuitively, decreasing the grid spacing may increase the adequacy of sampling, as was suggested by Cramer et al. (120). Indeed, it was shown that decreasing the grid spacing from 2.0 to 1.0 Å minimized the fluctuation in the observed q^2 values (75). Most probably, the reason for this phenomenon is that the decrease in grid spacing increases the number of probe atoms, which in turn should raise the probability of placing the probe atoms in a region where the steric and electrostatic field changes can be best correlated with biological activity. However, as was noticed by Cramer et al. (120), the increase in the number of probe atoms also increases the noise in PLS analysis and leads to a less statistically significant q^2 (121).

An important feature of conventional CoMFA routine is that it assumes equal sampling and *a priori* equal importance of all lattice points for PLS analysis, whereas the final CoMFA result actually emphasizes the limited areas of three-dimensional space as important