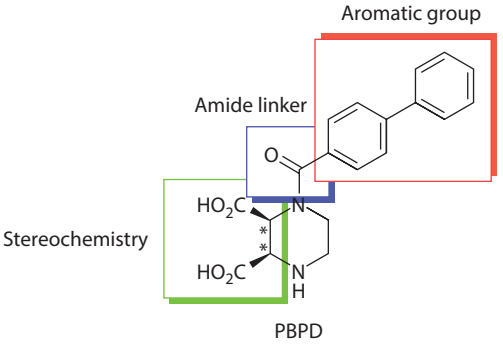

FIGURE 3.10 Structure–activity relationship study on PBPD derivatives to investigate structural features necessary for GluN2 subunit selectivity.

Although some of the compounds described earlier discriminated between the subunits more effectively than others, they still followed the same selectivity pattern that had been observed for the majority of "traditional" antagonists. Examining the existing pool of competitive antagonists, a series of NMDAR antagonists were found to go against this traditional selectivity pattern. These were derivatives of piperazine-2,3-dicarboxylate (PzDA) which are conformationally restricted derivatives of NMDA. Some of these PzDA derivatives showed selectivity for GluN2C/D *vs* GluN2A/B (Table 3.2). Most NMDAR competitive antagonists had 5 or 7 bonds between the acidic groups; unusually, the PzDA analogs had a short inter-acidic group chain length more commonly seen in agonists. It was hypothesized that the amino acid end of these compounds was binding to the same site as agonists such as NMDA and that the bulky aromatic ring was causing the antagonist activity. In particular, the biphenyl derivative PBPD (Figure 3.10) stood out as an interesting lead as it had highest affinity for GluN2D and lowest affinity for GluN2A (Table 3.2) which is the opposite of that observed for (*R*)-CPP. Therefore, a SAR study was carried out on a series of PBPD derivatives with variations to the biphenyl group to investigate the size and shape of the predicted hydrophobic pocket and investigate whether these changes would improve the GluN2D subunit selectivity. In addition to the aromatic group, the amide linking group and the stereochemistry were also examined for their effect on binding affinity and subunit selectivity (Figure 3.10). The affinity and selectivity of these compounds were evaluated at recombinant NMDARs using two-electrode voltage clamp electrophysiology. Each of the GluN2 subunits were individually co-expressed with GluN1a in *Xenopus* oocytes.

It was hypothesized that the aromatic ring was crucial for the antagonist activity and GluN2 subunit selectivity pattern observed in PBPD and that it occupied a hydrophobic pocket within the binding site. Without knowing the structure of the binding site, this could be attributed to a number factors relating to the size and shape of the pocket or the various interactions that aromatic groups can be involved in, such as hydrophobic interactions, cation–π interactions or hydrogen bonding interactions between the dense π system of the ring with strong hydrogen bond donors such as quaternary ammonium ions.

A number of PBPD analogs were synthesized, where the biphenyl group was systematically altered (Figure 3.11). Removing the second ring to leave a phenyl ring (UBP130) resulted in a loss of affinity across all the subunits. Replacing the first ring with an (*E*)-ethylene linker (UBP112) reduced affinity at GluN2B-D, but slightly increased affinity for GluN2A, showing a similar selectivity pattern to that of (*R*)-AP7 (Table 3.2). These alterations suggested that both rings were important in conferring GluN2D *vs* GluN2A selectivity.

As it appeared that the general size of the biphenyl group was favorable, its geometry was examined next. In a biphenyl ring system, the aromatic rings are twisted out of plane. To force a coplanar arrangement, the biphenyl group was substituted for a phenanthrene ring (PPDA) (Figure 3.11).