8.3.2.1 Discovery of Filibuvir (PF-00868554)

The dihydropyrone **31** was discovered as a weak and reversible NS5B inhibitor ($IC_{50} = 0.9\,\mu M$) in a screening campaign at Agouron Pharmaceuticals (now Pfizer).[47b] The core of this molecule is reminiscent of tipranavir, an orally active HIV protease inhibitor.[48] Biochemical studies revealed that dihydropyrones behaved similarly to thumb pocket 1 inhibitors in that they did not compete with nucleotide substrates. A crystal structure of **31** bound to NS5B revealed a new, well-conserved binding site in the thumb domain, $\sim 30\,Å$ away from the active site. In addition to hydrophobic contacts, key protein–inhibitor interactions comprised direct hydrogen bonds between the enol–ketone oxygen of the inhibitor with the backbone amide NH of L476 in addition to a water-mediated H-bond to Y477 NH. Preliminary data suggested that dihydropyrones interfered with NS5B function through perturbation of protein dynamics, interference with RNA binding or disruption of enzyme oligomerization.[47b] Initial hit-to-lead activities guided by the available structural data allowed a 30-fold improvement of intrinsic potency, providing analogs such as **34** (Figure 8.9) with sub-micromolar potency in biochemical assays but that lacked significant antiviral potency in cell-based replicon assays.[49]

The large discrepancy between the nanomolar potencies achieved in enzymatic assays and the moderate activity in cell culture, and the poor rat PK properties (bioavailability) associated with sulfur-linked dihydropyrone

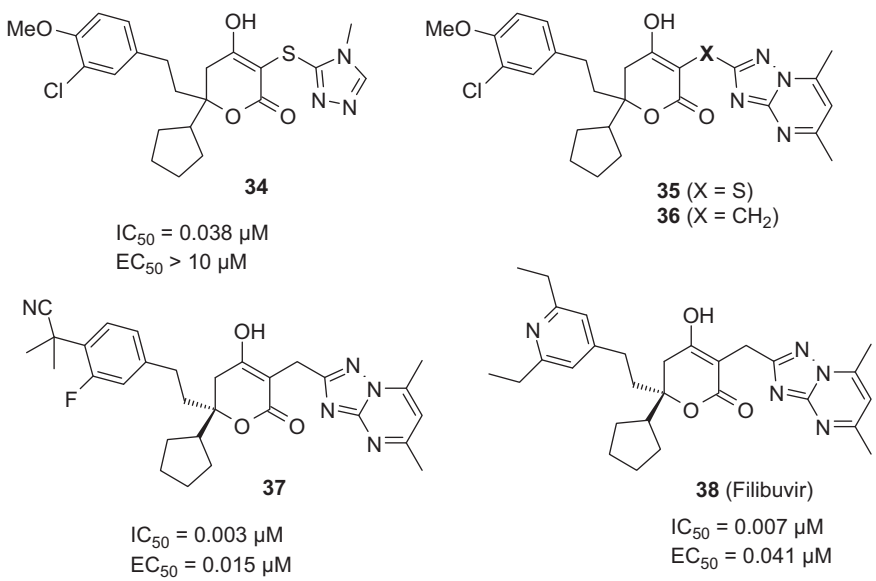

Figure 8.9 Evolution of dihydropyranone-based thumb pocket 2 inhibitors – discovery of filibuvir.