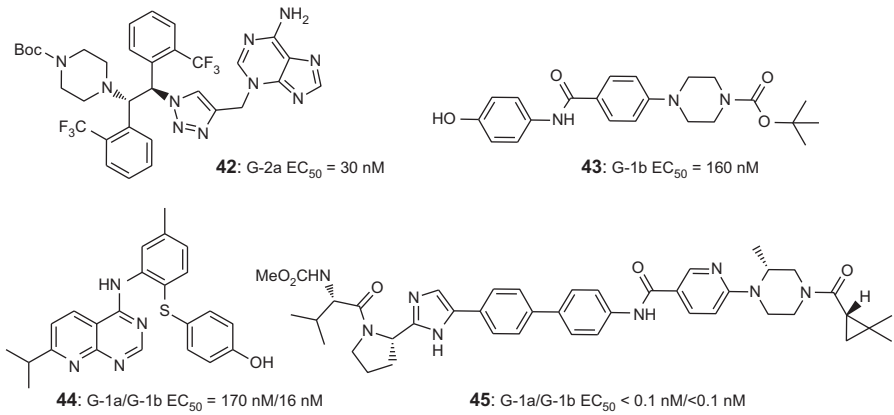

Figure 1.11 Miscellaneous chemotypes with NS5A resistance mutations.

1.5 Clinical Trials with HCV NS5A Replication Complex Inhibitors

Clinical validation that inhibitors of NS5A represented an effective approach to the control of HCV replication was obtained in Phase 1 studies with **1**.[1] A single ascending dose (SAD) study conducted in normal healthy volunteers with doses of **1** ranging from 1 to 200 mg established dose-proportional exposure of the drug, with plasma concentrations at 24 h post-dose in all subjects significantly exceeding that required to express antiviral activity in replicons. Administration of an oral solution of **1** to subjects chronically infected with G-1a and G-1b HCV at doses of 1, 10 and 100 mg produced a rapid and dose-related reduction of virus RNA levels. Viral RNA was reduced by an average of 1.8 $\log_{10}$ measured 24 h following a single 1 mg dose of **1**, whereas the 10 and 100 mg doses exerted more profound effects on viral load, with reductions of 3.2 $\log_{10}$ and 3.3 $\log_{10}$, respectively, at 24 h post-dose. The single 100 mg dose was associated with a maximum 3.6 $\log_{10}$ decline in viral RNA levels that was maintained for 144 h following drug administration, with one G-1b virus-infected subject in this cohort achieving RNA levels below the 25 IU mL^{-1} level of quantification at the 144 h time point, and the viral load for a second subject was measured at 35 IU mL^{-1}.

Virus sequencing, conducted prior to dosing and at 24 and 144 h post-dosing, revealed that when reduction in viremia was significant, HCV variants were detectable, with M28T, Q30H/R and L31M observed in G-1a-infected subjects and Y93H in the G-1b patients. These observations presumably reflect potent and effective restriction of wild-type virus replication by **1** that reveals virus-possessing resistant mutations as the thriving species. This scenario is anticipated based on the replication rate of the virus, estimated to be 10^{12} virions per day, combined with the low fidelity of the polymerase, the error