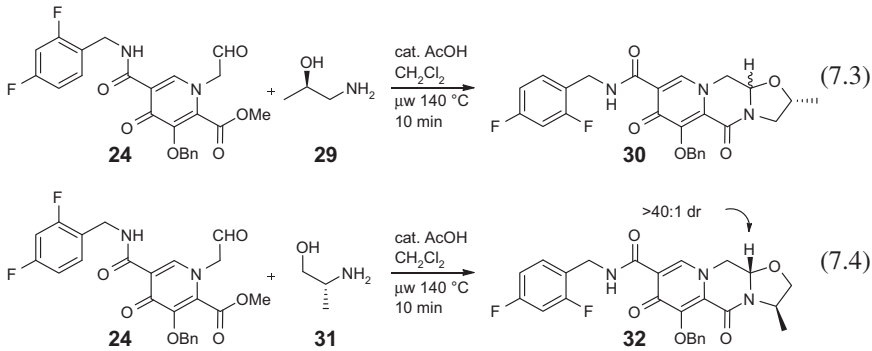
slow to cyclize and the product distribution is driven by the ring-closing step energetics, with the *trans* product having a $\sim$2.24 kcal mol^{-1} lower energy.



6.3.4 Choosing the Optimal Candidate

We were pleased to observe that the methyl isomers (**33–37**, **3**) all had high potency as good as or better than that of the unsubstituted original enantiomerically pure analogs of **19**. A detailed examination of the SAR is not included herein, but suffice it to say that the methyl analogs appeared to be the optimal molecules to deliver stereocontrol while preserving or improving the necessary virological and PK properties (Table 6.4). Further complexity, lipophilicity, polarity or size of the substituent did not appear to bring any advantages to justify the added challenges of accessing the necessary amino alcohol and increase in molecular weight.

A key parameter in our triage of compounds was the coverage of the protein-adjusted IC_{50} at 24 h post-dosing from a standard 5 mg kg^{-1} oral dose ($C_{24\,h}/^{pHIV}PAIC_{50}$). The formulation studies ultimately do become important, but for screening of analogs we typically began with either a solution or suspension dose and eventually moved to a fully crystalline drug in capsules as a measure of our worst-case bioavailability. The combination of the PK profile along with early virology data places compounds **3** and **4** in an area with great potential for more detailed virological assessment.[92]

6.3.5 Tricyclic Carbamoylpyridones Deliver 'Next-generation' Virological Profiles

Compounds **3** and **4** were examined in MT-4 cell and peripheral blood mononuclear cell (PBMC) HIV multiround replication assays (Table 6.5).[93,94] A more robust protein shift value was also determined *via* titrating various concentrations of human serum into the MT4 cell assay system.[35] From these data, a better approximation of the protein-adjusted clinical target was able to be determined. The overall fold shift values were somewhat higher than the