lipophilicity of the linker that gives **13** and **98–104** good liver exposure precludes a further increase in overall lipophilicity through contributions from P3 substituents. Based on the exploration of the macrocyclic scaffold in the P2 isoindoline series, **13** was identified as the optimal compound in the series and was subsequently designated MK-7009.[42,43,100]

The synthesis of MK-7009 starts with 3-bromo-*o*-xylene (**105**), which was dibrominated with *N*-bromosuccinimide (NBS) and benzoyl peroxide (Scheme 7.4).[101] Displacement of the bromines with benzylamine with concomitant ring closure gave 2-benzyl-4-bromoisoindoline (**106**). Installation of the vinyl group and removal of the benzyl protecting group with 1-chloroethyl chloroformate[102] and methanol provided **107**. Standard carbamate-forming conditions with **108** and removal of the Boc protecting group gave key intermediate **109**. Compound **109** was deprotected and coupled to linker intermediate **110** to give the bis-olefin **111**. The Zhan 1b metathesis catalyst[88] was used to affect macrocyclization and the newly formed olefin could be hydrogenated, providing **112**. Ester hydrolysis and coupling to **113** gave MK-7009 (**13**). Subsequent optimization of synthetic routes led to improved approaches to MK-7009.[92,103]

In terms of off-target activity, MK-7009 demonstrated a large selectivity window against a broad panel of cathepsins, and also other proteases such as chymase and elastases (Table 7.17). Although MK-7009 does show a modest inhibitory activity against chymotrypsin, the selectivity ratio relative to NS3/4A potency is very large (8700-fold). The $> 10\,000$-fold selectivity observed in an array of 169 pharmacologically relevant ion channel, receptor and enzyme targets conducted at MDS Pharma (Panlabs) serves to demonstrate further the highly selective nature of MK-7009. In an assay to assess potential hERG activity, the ability of MK-7009 had $IC_{50} > 10\,\mu M$ in an MK-499 binding assay. In addition, an assessment of the ability of MK-7009 to inhibit p450 reversibly did not show significant inhibition of isoforms tested (3A4, 2D6 and 2C9), with $IC_{50} > 10\,\mu M$. This lack of significant activity observed against p450 isoforms examined is also clearly an attractive attribute for a drug targeting significant liver exposure.

The pharmacokinetic properties of MK-7009 were evaluated in multiple species (Table 7.18).[42] In rats, the compound showed a plasma clearance of $74\,mL\ min^{-1}\,kg^{-1}$ and a plasma half-life of $\sim 1\,h$. When dosed orally at $5\,mg\ kg^{-1}$, the plasma exposure was modest, with an AUC of $0.1\,\mu M\,h$. In contrast to the plasma exposure, the liver exposure of MK-7009 at 24 h after a $5\,mg\,kg^{-1}$ oral dose was significant ($0.4\,\mu M$).

When dosed to dogs, the plasma pharmacokinetics of MK-7009 were greatly improved (Table 7.18), with moderate clearance of $11\,mL\ min^{-1}\,kg^{-1}$ and a 1.2 h half-life after i.v. dosing and good plasma exposure ($AUC = 1.2\,\mu M\,h$) after a $5\,mg\,kg^{-1}$ oral dose. Dog liver biopsy studies showed that the liver concentrations of MK-7009 after the $5\,mg\ kg^{-1}$ oral dose were 34 and $0.5\,\mu M$ at the 2 and 24 h time points, respectively. Similarly to its behavior in rats, MK-7009 demonstrates effective partitioning into liver tissue and maintains high liver concentration, relative to potency, 24 h after oral dosing in dogs.