regulatory guidelines.[49] The potency of cobicistat as an inhibitor of human hepatic microsomal CYP3A was compared with that of ritonavir, using substrates that include terfenadine, midazolam, testosterone, atazanavir, telaprevir and elvitegravir. The inhibitory potency was either measured with marker activities for the enzymes or determined by monitoring substrate depletion. As shown in Table 13.4, the broad spectrum of inhibitory activity of cobicistat against human CYP3A was confirmed.

The data in Tables 13.3 and 13.4 indicate that cobicistat shares a similar spectrum of CYP3A substrate specificity to that of ritonavir. It retains the characteristic of mechanism-based inhibition of CYP3A and is equipotent to ritonavir for the substrates tested. Inhibition studies with the most important human CYP enzymes showed no significant inhibition at concentrations likely to be achieved clinically (Table 13.5). In addition, cobicistat is more selective than ritonavir, with much reduced inhibitory activity towards CYP2D6, CYP2C8 and CYP2C9.

As discussed in Session 13.3, ritonavir activates the pregnane X receptor (PXR) and induce metabolic proteins including CYP3A, CYP2B6, CYP2C9 and P-gp, further complicating the potential for drug–drug interactions.[29,30] It is therefore desirable to eliminate or reduce this drug interaction potential for new PK enhancers. Studies assessing the induction liability of cobicistat showed that neither cobicistat nor ritonavir showed significant stimulatory activity in the AhR-responsive assay. However, at 10 μM in the PXR assay, ritonavir

Table 13.4 Inhibitory potencies against human hepatic microsomal CYP3A-dependent activities.

	IC_{50} (nM)	
CYP3A activity	*Ritonavir*	*Cobicistat*
Midazolam 1′-hydroxylase[50]	107	154
Testosterone 6β-hydroxylase[51]	116	151
Terfenadine oxidase[52]	275	285
Elvitegravir oxidase	26	33
Atazanavir oxidation	40	44
Telaprevir oxidation	18	30

Table 13.5 Inhibitory potencies against activities catalyzed by major human hepatic microsomal cytochromes P450.

		Calculated IC_{50} (μM)	
Enzyme	*Activity*	*Ritonavir*	*Cobicistat*
CYP1A2	Phenacetin *O*-deethylase	>25	>25
CYP2B6	Bupropion 4-hydroxylase	2.9	2.8
CYP2C8	Paclitaxel-6α-hydroxylase	2.8	>25
CYP2C9	Tolbutamide 4-hydroxylase	4.4	>25
CYP2C19	*S*-Mephenytoin 4′-hydroxylase	>25	>25
CYP2D6	Dextromethorphan *O*-demethylase	2.8	9.2