concentration can be measured using either scintillation counting methods to detect $^{86}Rb^+$ or atomic absorption spectroscopy methods.[38]

The Rb^+ efflux assay and hERG binding assays are extensively used in the pharmaceutical industry, but ultimately they are both indirect methods of determining ion channel activity. As discussed in Chapter 4, electrophysiological patch clamp is the "gold standard" in determining the impact of candidate compounds on an ion channel. Experiments of this type can be conducted using an appropriately engineered cell line (hERG-expressing CHO or HEK293 cells), but this method is generally only used on advanced candidates. Electrophysiological patch clamp studies remain time and labor intensive processes that are not amenable to high throughput methods, and this significantly limits the number of compounds that can be screened for hERG activity using this method.

While hERG channel activity is an important aspect of cardiovascular safety, it is by no means the only issue that needs to be considered in determining cardiac safety risks associated with a candidate compound. Changes in factors such as blood pressure, heart rate, contractility, and ejection fraction must also be considered as alteration in any of these aspects of cardiovascular function can also lead to serious consequences. The impact of candidate compounds could be studied in a variety of animal models capable of providing direct insight into cardiovascular function, but it is not practical to study large numbers of compounds in this manner. The time and cost associated with these studies is simply too high to allow broad screening of candidate compounds in an *in vivo* setting. Fortunately, it is possible to identify compounds that may have an impact on cardiovascular function using *in vitro* screening systems. The cardiovascular system is exceedingly complex and there are a wide variety of enzymes, ion channels, GPCR, transporters, and other biomolecules that modulate its activity. *In vitro* screening methods designed to identify compounds that interact with these targets have been developed to identify useful drugs for the treatment of cardiovascular diseases such as hypertension and congestive heart failure. These same assays can be used as counterscreens to identify compounds that could induce cardiovascular side effects. SAR patterns can be determined in order to minimize the interaction of candidate compounds with biomolecules that control cardiovascular function. If a program is targeting a biomolecule that is closely related to a second biomolecule that is involved in cardiac function, then an assay focused on activity of the second biomolecule is often part of the screening paradigm used to identify compounds suitable for advancement. Consider, for instance, a program whose goal is the identification of 5-HT_7 antagonists for the treatment of irritable bowel disease (IBD).[39] The 5-HT_{2b} receptor shares a high degree of homology with 5-HT_7, and agonist activity at 5-HT_{2b} is associated with cardiovascular side effects.[40] As a result, the risk of designing a compound with activity at both of these receptors