enzymes (e.g., CYP1A2, monoamine oxidase, (MAO) etc.) may be assessed in order to further determine DDI risks.

Another pathway that can lead to drug–drug interaction is changes in expression of metabolic enzymes. There are many reported cases of compounds that induce increases in expression of CYP enzymes, an effect referred to as CYP induction. This can have a significant impact on the metabolic fate of candidate compounds. Increased CYP expression will lead to increased metabolism of susceptible compounds, decreasing their potential for efficacy, while at the same time increasing the rate of formation of metabolites. If any of the metabolites are associated with safety or toxicity issues, the increased concentration caused by CYP induction may lead to negative effects that would not have occurred in absence of CYP induction (e.g., drug–drug interaction). Identifying CYP inducers early in a drug discovery program can be accomplished using *in vitro* techniques similar to those used to identify compounds that inhibit CYP enzymes. Hepatocytes are exposed to candidate compounds for a sustained period to provide an opportunity for CYP induction and then assessed for their ability to metabolize substrates known to be specific to a single CYP isozyme (e.g., CYP3A4, CYP2D6, CYP2C9, etc.). If the rate of metabolism is increased relative to control hepatocytes (those not exposed to the candidate compound), then CYP induction is indicated. It is also possible quantify the amount of enzyme or mRNA present using biochemical means.[32]

CARDIOVASCULAR SAFETY AND TOXICOLOGY STUDIES

The manifestation of cardiovascular side effects, even in a very small portion of the patient population, can lead to significant marketing restrictions or even removal from the market. The painkiller Vioxx®, for example, had 2003 sales of $2.5 billion, but was removed from the market when it was determined that it increased the rate of cardiovascular events from 0.78 per 100 patients to 1.50 per 100 patients.[33] In order to minimize the risk to both the patient and sponsor company, the cardiovascular safety profile of candidate compounds is thoroughly examined prior to the initiation of clinical trials. It is, of course, cost prohibitive to assess large numbers of compounds in full animal models of cardiovascular safety. Fortunately, there are a number of assays available that can be used to identify and eliminate compounds with the potential to negatively impact cardiovascular function.

Assays designed to detect blockade of the hERG channel (also known as $K_v11.1$) are generally the first step in determining whether or not a candidate compound is at risk for cardiovascular side effects. This channel is an important component of the electrical system that allows the heart to beat in the rhythmic fashion required to move blood through the body. Specifically, the hERG channel conducts potassium ions out of cardiac myocytes (the "rapid"