NOVEL TOOLS TO CHARACTERIZE HERG CHANNELS

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The K^+ channel encoded by the human ether-á-go-go related gene (HERG) is one of many ion channels that are crucial for normal action potential repolarization in cardiac myocytes. HERG encodes the pore-forming subunit of the rapid component of the delayed rectifier K^+ channel, $I_{K(Vr)}$. Mutations in HERG result in a prolongation of the QT-interval on the electrocardiogram and give rise to Long QT Syndrome, a disorder that may cause syncope and sudden death resulting from episodic ventricular arrhythmias and ventricular fibrillation.
Thus, HERG K^+channels are of considerable pharmaceutical interest as possible therapeutic targets for anti-arrhythmic agents. Moreover, a large number of various pharmaceutical agents exerted severe cardiac side effects by simply blocking HERG channels.
Toxins isolated from a variety of venoms are tools for probing the physiological function and structure of ion channels. It has previously been demonstrated that BeKm, a toxin isolated from the venom of the Central Asian scorpion *Buthus eupeus*, is a novel, powerful, and specific blocker of the HERG K^+ channel. BeKm is a 36-amino acid peptide containing 6 cysteines and 6 positively charged residues and, on the basis of analysis of its amino acid sequence, shares sequence homology with other K^+ channels blocking toxins derived from scorpion venom.

A synthetic gene encoding BeKm was designed, synthesized and expressed as a cleavable fusion protein in *Escherichia Coli*. Thereafter, a BeKm-analogue which carries a single free sulfhydryl group in the 'back' of the molecule, most opposite from the toxin's interaction surface with the HERG channel will be constructed. This sulfhydryl group can be used for derivatization with fluorescent sulfhydryl-reactive dyes.
This BeKm analogue can now allow 'real time' imaging of channels, overcoming several limitations known to exist for radioligands or sequence specific antibodies. Additionally they will enable quantification of ligand binding on the cellular level or at the subcellular level.