

ISOLATION AND CHARACTERIZATION OF A POTASSIUM CHANNEL INHIBITOR ISOLATED FROM *BUNODOSOMA CAPENSIS*

Wynand van Losenoord¹, Jason Krause¹, Shirley Parker Nance² and Carminita Frost¹

Nelson Mandela University, Republic of South Africa

Voltage gated potassium channels have recently become a subject of investigation as possible pharmaceutical targets. Research has linked the activity of potassium channels directly to both anti-inflammatory pathways and energy homeostasis. Sea anemones secrete a diverse array of bioactive compounds including potassium channel inhibitors. A novel potassium channel inhibitor was isolated and characterized from *Bunodosoma capensis* using a modified stimulation technique to induce the secretion of the neurotoxin rich mucus. The presence of a neurotoxin was confirmed using an *Artemia* shrimp swimming assay, through the induction of mild paralysis within the nauplii. The peptide component of the mucus was extracted using a desalting C18 cartridge. The potassium channel inhibitor was isolated using fast protein liquid chromatography and confirmed using the FluxOR potassium channel assay. Final purification of the peptide was performed by using a C18 reverse phase high performance liquid chromatography system and purity confirmed with a modified tricine SDS-PAGE system. N-terminal sequencing determined similarities in homology to potassium channel inhibitors isolated from *B. granulifera* and *Stichodactyla helianthus*. MCF-7 cells displayed higher sensitivity to inhibition by the isolated peptide in comparison to 3T3-L1 cells due to the presence of a Kv 1.3 channels. The disulfide bond containing peptide was determined to have a mass of ± 4.2 kDa. It can be concluded that the potassium channel inhibitor displays similarities to reported potassium channel inhibitors isolated from anemone species. The isolated inhibitor at 3.98 nM inhibited the potassium channels in MCF-7 cells to 70% as compared to 20 μ M for the controls (gliclazide and digoxin), indicating a significant (500 fold) increase in inhibition of the channels as compared to the controls and comparable to other potassium channel inhibitors isolated from anemones cited in literature.

s213203669@mandela.ac.za