Store-operated Ca^{2+} entry and TRPC expression; possible roles in control of heart rate

Y-K. Ju and D.G. Allen, School of Medical Sciences, University of Sydney F13, NSW 2006, Australia.

Store-operated Ca^{2+} channels (SOCCs) were first identified in non-excitable cells by the observation that depletion of Ca^{2+} stores caused increased influx of extracellular Ca^{2+} (Putney, 1986). Recent studies have suggested that SOCCs might be related to the transient receptor potential (TRPC) gene family (Vazquez *et al.,* 2004). In a previous study, we found that activation of the $P2Y_1$ purinergic receptor by ATP results in modulation of pacemaker firing due to receptor-coupled phospholipase C (PLC) activation and depletion of sarcoplasmic reticulum (SR) Ca^{2+} stores (Ju *et al.,* 2003). Since activation of SOCCs also involves PLC, we speculated that SOCCs might also be present in pacemaker tissue (Ju & Allen, 2007).

To study SOCC in pacemaker tissue, we first developed a method for recording intracellular Ca^{2+} signals from mouse sinoatrial nodes (SAN) in which the structural integrity and activity of the node is preserved. Store-operated Ca^{2+} entry was investigated in isolated mouse sinoatrial nodes (SAN) dissected from right atria and loaded with Ca^{2+} indicators. Incubation of the SAN in Ca^{2+}-free solution caused a substantial decrease in resting intracellular Ca^{2+} ($[Ca^{2+}]_i$) and stopped pacemaker activity. Reintroduction of Ca^{2+} in the presence of cyclopiazonic acid, a selective sarcoplasmic reticulum Ca^{2+}-ATPase inhibitor, led to sustained elevation of $[Ca^{2+}]_i$; a characteristic of store-operated Ca^{2+} channel (SOCC) activity (Ju *et al.,* 2007). Transcripts for all TRPC isoforms, except TRPC5 have been detected in SAN preparation. Immunohistochemistry studies also revealed the localizations of TRPC1, 3, 4, and 6 proteins in both the central and peripheral SAN (Ju *et al.,* 2007).

The mechanism of cardiac pacemaking involves voltage-dependent pacemaker current; in addition there is growing evidence that intracellular sarcoplasmic reticulum (SR) Ca^{2+} release plays an important role. The SOCC antagonist, SKF-96365 (10 μmol/L) that inhibited Ca^{2+} influx reduced the spontaneous pacemaker rate and stopped pacemaker firing in the present of CPA (Ju *et al.,* 2007). These newer findings suggest that Ca^{2+} entry and inward current triggered by store depletion might contribute to the pacemaker current and may play a role in control of heart rate.

Ju YK, Huang WB, Jiang L, Barden JA & Allen DG. (2003) *Journal of Physiology,* **552:** 777-87.

Ju YK & Allen DG. (2007) *Heart, Lung and Circulation,* Sep 4; PMID: 17822952

Ju YK, Chu Y, Chaulet H, Lai D, Gervasio OL, Graham RM, Cannell MB & Allen DG (2007) *Circulation Research,* **100:** 1605-14.

Putney JW. (1986) *Cell Calcium*, **7:** 1-12.

Vazquez G, Wedel BJ, Aziz O, Trebak M, Putney JW Jr. (2004) *Biochimica et Biophysica Acta,* **1742:** 21-36.

Acknowledgements: This work was supported by NH&MRC.