Differential effects of fingolimod and other transient receptor potential melastatin 7 (TRPM7) channel modulators on calcium paradox-induced myocardial injury in rat hearts

Matthew AMONI
- **Calcium** is drives cardiac cellular function.

- Abnormalities of calcium (Ca\(^{2+}\)) homeostasis underlie the pathophysiology of several cardiovascular conditions.

- **Calcium homeostasis** is critical for signalling.

- Non-selective cation channels such as transient receptor potential (TRP) channels, specifically TRPM7 are implicated in abnormal Ca\(^{2+}\) homeostasis (Gwanyanya et al., 2004)

- However, the contribution of such non-selective channels to cardiac injury at organ level is unknown.

- We present preliminary work interrogating background contributors to Ca\(^{2+}\) homeostasis.
INTRODUCTION

- **Calcium Paradox:** Ca$^{2+}$-mediated myocardial injury due to Ca$^{2+}$ paradox (CP) has long been established experimentally in hearts temporarily perfused with Ca$^{2+}$-deficient solutions (Zimmerman and Hulsmann, 1966).

- Clinically, varying degrees of the CP phenomenon may occur peri-operatively.
- Experimentally, cardiac tissue storage solutions and isolation of cardiomyocytes for various cellular studies.

Novel specific exogenous pharmacological blockers to evaluate TRPM7:
fingolimod (FTY720) (Qin et al., 2013) and
nordihydroguaiaretic acid (NDGA) (Chen et al., 2010)

Otto Loewi: A drug is a substance that when injected into an animal, produces A PAPER.
AIM

We therefore investigated the effects of TRPM7 channel inhibitors:

- fingolimod (FTY720)
- nordihydroguaiaretic acid (NDGA),
- as well as Mg\(^2+\) pre-treatment

… on Ca\(^{2+}\) Paradox-induced myocardial injury (CP).
METHODS

The presence of cardiac TRPM7 proteins was evaluated by immunoblotting n=5-6.

Langendorff-perfused Wistar rat hearts:
intraventricular balloon – Hemodynamics;
triphenyltetrazolium chloride stain - infarcts size.

FTY720 (1 µmol/L), NDGA (10 µmol/L), or vehicle infused prior to a CP protocol consisting of 3-min Ca²⁺ depletion, followed by 30-min Ca²⁺ repletion n=6-8.

MgSO₄ (270 mg/kg, intraperitoneally) or saline daily for 7 days were also subjected to CP n=6-9.

STATISTICS

GraphPad Prism 6 software package – ANOVA & t-test.
Data are presented as mean ± SEM; P < 0.05 = significant.
RESULTS

The expression of TRPM7 channels in cardiac ventricular tissue:

A

(KDa)

250

TRPM7

β-actin

37

Control  Mg treated

B

Fold expression of TRPM7 (normalised to beta-actin)

Control  Mg treated

n.s.
RESULTS

Effects of NDGA and FTY720 on CP-induced hemodynamic changes:

Although both FTY720 and NDGA minimized the CP-induced elevation of left ventricular end-diastolic pressure, only FTY720 improved LV developed pressure (p=0.029). Pretreatment with Mg2+ affected CP-induced infarct size.
**RESULTS**

Effects of NDGA and FTY720 on Ca2+ paradox (CP) induced infarcts:

FTY720, but not NDGA, decreased CP-induced infarct size from 64.6 ± 5.3% to 39.0 ± 6.8% (p=0.001; n=6).
RESULTS

Effects of Mg2+ pretreatment on CP-induced hemodynamic changes and infarcts:

A

B

C

Infarct size (% AAR)

Control   Mg  CP   Mg+CP
CONCLUSION

TRPM7 protein presence in cardiac ventricular tissue.

Among the TRPM7 channel modulators tested, FTY720, but not NDGA or pre treatment with Mg2+, reversed CP-induced myocardial damage and dysfunction (suggesting that the action of FTY720 seems to be unrelated to the capacity of the drugs to inhibit TRPM7 channels)

FTY720 may have a potential therapeutic role during cardiac perfusion, but future studies need to investigate the mechanisms underlying the cardioprotective effect of FTY720 in CP.
ACKNOWLEDGEMENTS

Dr Fatma Altrag
Dr Roisin Kelly-Laubscher
Dr Asfree Gwanyanya

Dept. of Human Biology:
Cardiac- Vascular and Regenerative Medicine Laboratory

NRF National Research Foundation