INTRODUCTION

- Post-traumatic Osteoarthritis (PTOA) comprises ~12% osteoarthritic cases\(^1\).
- Chondrocytes respond to joint injuries by releasing inflammatory mediators which aggravate PTOA\(^2\).
- Activation of Nod-like receptor family protein 3 (NLRP3) inflammasome through NF-κB signalling induces Interleukin-1β (IL-1β) secretion and mediates inflammatory cell death\(^3\).

**MCC950** is a selective NLRP3 inflammasome inhibitor and prevents IL-1β secretion\(^4\).

**Overall Goal:**
To develop a novel drug delivery platform releasing MCC950 to target NLRP3 inflammasome in order to reduce cartilage destruction and demonstrate therapeutic efficacy in PTOA.

**AIMS AND METHODS**

**Aim 1:** To obtain sustained release of MCC950 from PLGA microparticles

- PLGA and MCC950 dissolved in dichloromethane
- Spray-dried PLGA particles
- Release studies in PBS
- HPLC to quantify amount of MCC950 released

**Aim 2:** To investigate the potential of MCC950 released from PLGA microparticles to inhibit LPS-induced IL-1β secretion

- 100ng/mL LPS
- 3 hours
- MCC950 (stock/released from PLGA particles)
- 5mM ATP
- 40 minutes
- Inflamed macrophages
- 45 minutes
- IL-1β released

**Aim 3:** To explore the adverse effects of inflamed macrophages on chondrocytes

- Inflamed macrophages releasing IL-1β into cell media
- IL-1β released cell media added to healthy chondrocytes
- Polymerase Chain Reaction

RESULTS

**Spray-dried PLGA microspheres provided sustained release of MCC950**

- 1:1 ratio 3% PLGA 75:25 (molecular weight 4000-15000) + 75:25 (molecular weight 66,000-309,000) and 1mM MCC950
- 2.1 ratio 3% PLGA 50:50 (molecular weight 54,000-69,000) + 75:25 (molecular weight 66,000-309,000) and 1mM MCC950

**MCC950 inhibits LPS-induced IL-1β secretion in macrophages**

- Both 50µM MCC950 and MCC950 released from PLGA particles over 2 weeks inhibits pyroptosis significantly.
- Addition of MCC950 reduced cytotoxic effects on inflamed macrophages

**MCC950 reverses adverse effects of inflamed macrophages on healthy chondrocytes**

- IL-1β release from inflamed macrophages inhibited chondrocyte collagen formation and increased inflammatory protein level MMP-13. Both 1µM MCC950 and MCC950 released from PLGA particles reversed these effects.

DISCUSSION AND CONCLUSIONS

1. Spray-dried MCC950-PLGA microparticles allow for sustained release of MCC950 over 2 weeks.
2. MCC950 released from particles remain strongly bioactive and inhibit IL-1β release from inflamed macrophages.
3. MCC950 protected chondrocytes from the adverse effect of IL-1β released from inflamed macrophages.
4. MCC950 is potential drug delivery candidate for targeting IL-1β, which is believed to contribute to the pathogenesis of PTOA.

FUTURE WORK

- Optimisation of spray-dried PLGA particles for particle size and drug encapsulation
- Incorporate optimised PLGA particles into microparticles for joint delivery (Collaborator in Georgia Tech) to evaluate its safety and efficacy in the joint
- In-vivo study in PTOA rodent model

REFERENCES


ACKNOWLEDGMENTS

Development of a novel drug delivery platform to target NLRP3 inflammasome and its treatment in Post-traumatic Osteoarthritis

Hanitha Jacob\(^1,2,3\), James Early\(^1\), Sean McGrath\(^1\), Lauren Fagan\(^2,3\), Sarinj Fatatrah\(^1\), Amri Robertson\(^1\), Oran Kennedy\(^2,3\), Annie Curtis\(^2,3\), Cathal Kearney\(^1,2,3\)

\(^1\)Kearney Lab, Dept. of Anatomy and Regenerative Medicine, Royal College of Surgeons in Ireland (RCSI), Dublin, Ireland
\(^2\)Thermos Engineering Research Group TERRG, Dept. of Anatomy, and Regenerative Medicine, RCSI, Dublin, Ireland
\(^3\)Tissue Engineering Research Group (TERRG), Dept. of Anatomy, and Regenerative Medicine, RCSI, Dublin, Ireland

\(^1\)Kearney Lab, Royal College of Surgeons in Ireland (RCSI), Dublin, Ireland

\(^2\)Drug delivery and Advanced Materials Team, School of Pharmacy & Biomolecular Sciences, RCSI, Dublin, Ireland

\(^3\)Center Research in Medical devices (CURAM), RCSI and NUI, Galway, Ireland

\(^4\)School of Chemistry and Mater Science, The University of Queensland, Australia

\(^5\)Kearney Lab, Royal College of Surgeons in Ireland (RCSI), Dublin, Ireland