Bioenergetics Assessment and ECM gene expression in Glaucoma Lamina Cribrosa cells: Role of Metformin

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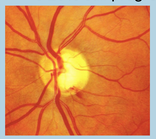
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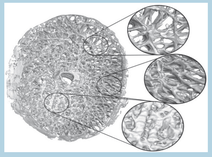
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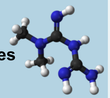
Primary Open Angle Glaucoma

- Leading cause of **irreversible blindness**
  - 70 million people worldwide
- **Lamina Cribrosa** (LC): connective tissue structure at Optic Nerve Head
  - Key site of Retinal Ganglion Cell axonal injury
- Molecular Hallmarks:
  - **Fibrosis**
  - **Mitochondrial Dysfunction**
- Current treatment – **lowering Intra-Ocular Pressure**
  - 20% still progress





Metformin



- Biguanide derivate – **Type 2 Diabetes**
- **Anti-Fibrotic**:
  - ↓ Pulmonary, Cardiac and Renal Fibrosis
- **Cohort** study: 150,016 patients
  - 25% reduced risk of POAG amongst diabetics taking metformin
  - Dose-dependent response
- **Rat Models:**
  - Eye drops of Metformin have been shown to prevent fibrosis after glaucoma filtration surgery

Aims

1. Assess Metformin's effect on **mitochondrial** function
2. Examine whether this correlated with an inhibition of **pro-fibrot**ic markers in human glaucomatous LC cells.

Methodology

Cell Culture

- Human Lamina Cribrosa cells – Normal (NLC) + Glaucoma (GLC)
  - Cells donated by Novartis Institutes for BioMedical Research (Cambridge, MA)



Mitochondrial Bioenergetic Assessment

Seahorse XFe96 Analyzer

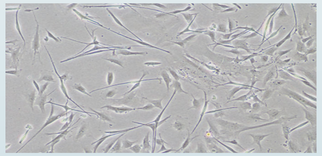
- GLC cells were treated with **Metformin** at different doses:
  - 10mM, 5mM, 2mM, 1mM, 0.5mM, 0.1mM and 0.05mM



ECM Gene expression

RT-PCR

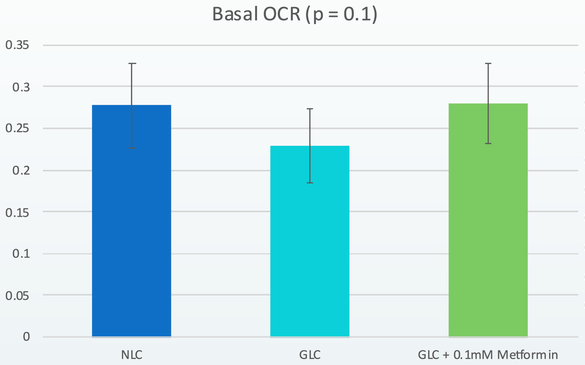
- GLC cells treated with **0.1mM** Metformin

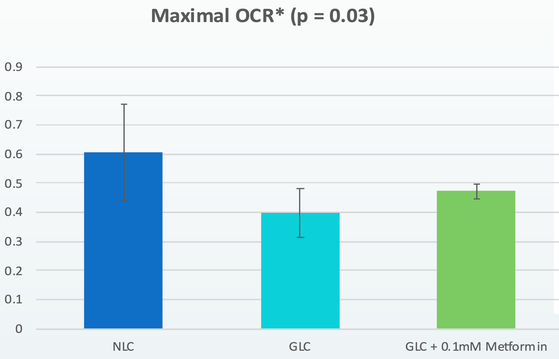


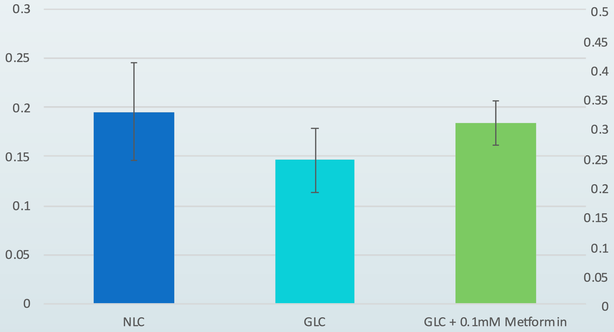
Results

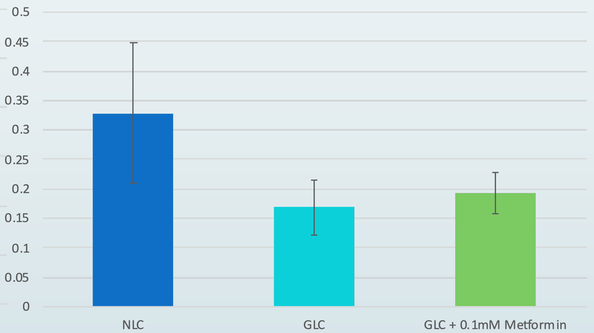
Mitochondrial Function

0.1mM Metformin



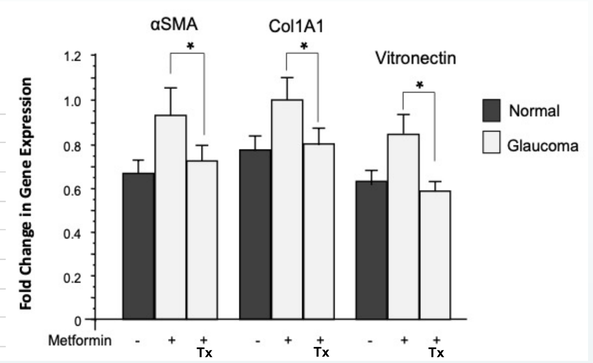






ECM Gene Expression

0.1mM Metformin



Conclusion

Improved Mitochondrial Respiratory Function

- **Dose-Dependent** effect
- *Inhibits permeability transition pore (**PTP**) -> Prevents cell death driven by oxidative stress*

Anti-Fibrotic

- ↓ **ECM production**
- ***AMPK** Activation -> TGF-β Inhibition*

Glaucoma – Multifactorial disease entity

- Unmet clinical need for a disease modifying agent
  - ➢ ***Is Metformin the future?***