A NEW PLAYER IN GLUCOSE TRANSPORT
Type 2 Diabetes

• Increases in Glucose Levels

http://www.deo.ucsf.edu/images/graphs/graph_target_glucose.gif
• Two pathways in which muscle regulates glucose transport
  • Insulin dependent
  • Insulin independent

AMP activated protein kinase
AKT and AMPK activation leading to glucose transport

AMPK

ATM’s role


Specific Aim 1: To determine whether the ATM activators, chloroquine and resveratrol, play a role in AMPK phosphorylation, AKT phosphorylation, and glucose transport in C2C12 myotubes.
Methods

• Western Blot

http://www.molecularstation.com/protein/western-blot/
Signaling Hypothesis in C2C12 myotubes

resveratrol

ATM

KU-55933

chloroquine

shRNA against ATM

P-AMPK
http://drugster.info/medic/term/short-hairpin-rna/
shRNA against ATM

A. ATM/Tubulin

B. CQ

P-AMPK/AMPK

C. CQ

P-AKT/THR308/AKT

GFP vs. KD Cell Type
Conclusion

Signaling Hypothesis in C2C12 myotubes

KU-55933

resveratrol

ATM

AMPK

AKT

chloroquine

shRNA against ATM
Mechanism?

• Mechanism of inhibition of bovine F$_1$-ATPase by resveratrol and related polyphenols
  • Jonathan R. Gledhill *, Martin G. Montgomery *, Andrew G. W. Leslie †, ‡, and John E. Walker *, ‡
Summary

- In C2C12 myotubes, chloroquine and resveratrol increase phosphorylation of AMPK, and chloroquine increases phosphorylation of AKT independent of ATM.
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  • Brian Downes
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  • Yuqi Wang
• Specific Aim 2: To determine whether insulin stimulated AKT phosphorylation leading to glucose transport is dependent on ATM in EDL and SOLEUS muscle.
Methods

- Isolated Muscle Incubations
- Western Blot
- Glucose Transport
Figure A: 
- Y-axis: soleus P-Akt S473/Akt, arbitrary units.
- X-axis: insulin, ATM genotype (+/+, +/+, +/-, +/-).
- Bar graphs showing the effect of insulin and ATM genotype on soleus P-Akt S473/Akt.
- Asterisks (*) indicate significant differences.

Figure B: 
- Y-axis: soleus P-Akt T308/Akt, arbitrary units.
- X-axis: insulin, ATM genotype (+/+, +/+, +/-, +/-).
- Bar graphs showing the effect of insulin and ATM genotype on soleus P-Akt T308/Akt.
- Asterisks (*) indicate significant differences.
**A**

<table>
<thead>
<tr>
<th>ATM genotype</th>
<th>soleus 2DG transport, nmol/mg protein/10 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>+/-</td>
<td>5</td>
</tr>
<tr>
<td>++</td>
<td>25</td>
</tr>
<tr>
<td>-/-</td>
<td>10</td>
</tr>
<tr>
<td>+/-</td>
<td>15</td>
</tr>
</tbody>
</table>

* Insulin effect marked with an asterisk.

**B**

- GLUT4/tubulin
- soleus GLUT4/tubulin, relative units
- ATM genotype
- +/-
- -/-
C

p-AS/AS160, arbitrary units

- - - +/+
- ++ +/+ - -
- +/- -/or - -

insulin
ATM genotype
Glucose Transport

Conclusion