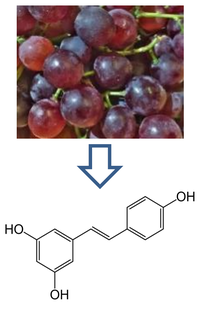
# Effects of Resveratrol on the Metabolic Activities of Chondrocytes

**Cynthia Wang[1], Yilu Zhou[2], Emily King[3], Yuechen Shi[4], Annie Porter[4], X. Lucas Lu[4]**

[1]MOT Charter High School, Middletown, DE. [2]University of Pennsylvania, Philadelphia, PA. [3]Rensselaer Polytechnic Institute, Troy, NY.
[4]Department of Mechanical Engineering, University of Delaware, Newark, DE.

## Introduction

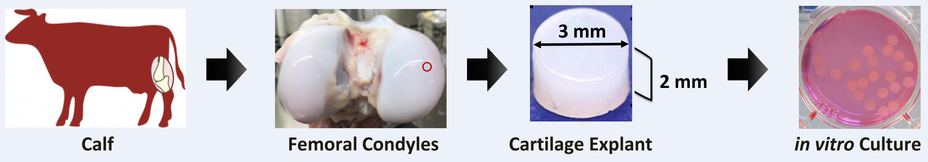
- Progression of osteoarthritis (OA) involves the loss of collagen and proteoglycan contents from cartilage extracellular matrix (ECM) [1].
- Resveratrol, a natural phenol found in the skin of grapes and various berries, has been shown to provide chondro-protective effects [2].
- Resveratrol can prevent OA initiation [3] and down-regulate catabolic gene expressions in chondrocytes [4].
- We recently found that resveratrol can directly bind to collagen fibrils and help collagen resist degradation caused by proteases [5].
- Chondro-protective mechanism of resveratrol is often attributed to its anti-oxidation and anti-inflammation functions. It is not clear how resveratrol regulates the metabolic activities of chondrocytes.




**Aim:** To evaluate the short- and long-term effects of resveratrol on the synthesis and degradation of cartilage ECM by *in situ* chondrocytes.

## Methods

### Cartilage Explant Harvest and *in vitro* Culture




### Labeling of Newly Synthesized ECM with Click Chemistry

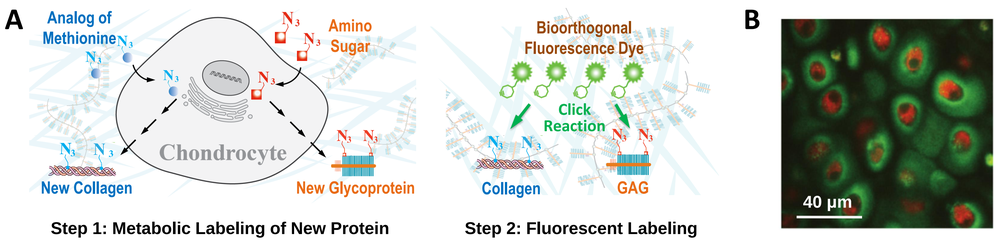
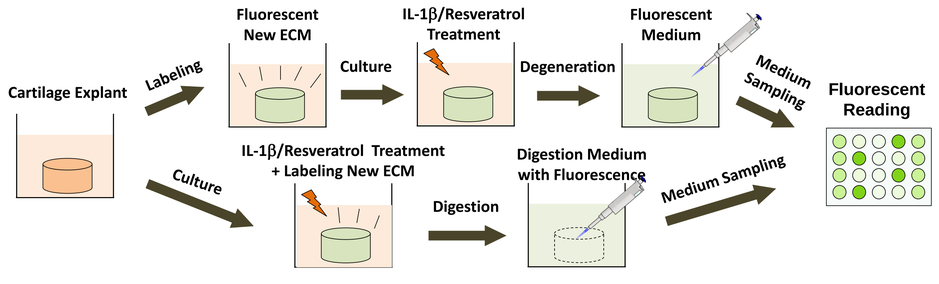



**Figure 1.** A new click chemistry-based technique was developed to quantify the newly synthesized proteoglycan and collagen by *in situ* chondrocytes. A) Modified amino acid or amino sugar with an azide functional group is fed to chondrocytes for collagen or glycoprotein synthesis, respectively. Fluorescent molecules are then conjugated to the azide groups on the new collagen or GAG. B) Newly synthesized GAG (green) surrounding *in situ* chondrocytes (red) in cartilage.

### Tracking the Loss of Collagen or GAG




### Tracking the Synthesis of Collagen or GAG

## Acknowledgements

This study was funded by the Osteo Science Foundation, DOD W81XWH-13-1-0148 and NIH P30GM103333-01. This study has no disclosure.

## References

[1] Mow, V. C. et al., 2005. *Basic Orthopaedic Biomechanics and Mechanobiology.* Lippincott Williams & Wilkins. [2] Csaki, C. et al., 2008. *Biochem Pharmacol.* 75.3: 677-687. [3] Li, W. et al., 2015. *J Orthop Res*. 33(7):1061-70. [4] Elmali, N. et al., 2005. *Inflamm Res*. 54(4):158-62. [5] Zhou *et al*., 2018. *Ann ORS Trans: 0466,* New Orleans, LA.

## Results

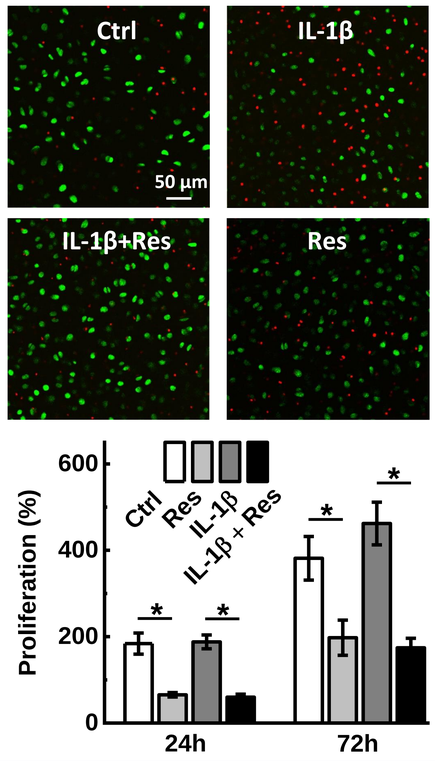



**Figure 2.** (Top) Resveratrol (100 μM) maintained the cell viability of *in situ* chondrocytes after 14-day culture. (Bottom) MTT assay showed that 100 μM resveratrol suppressed the primary chondrocyte proliferation at 24 h and 72 h. Results have been normalized to the value of control group at day 0 (n = 8; *: p<0.05).

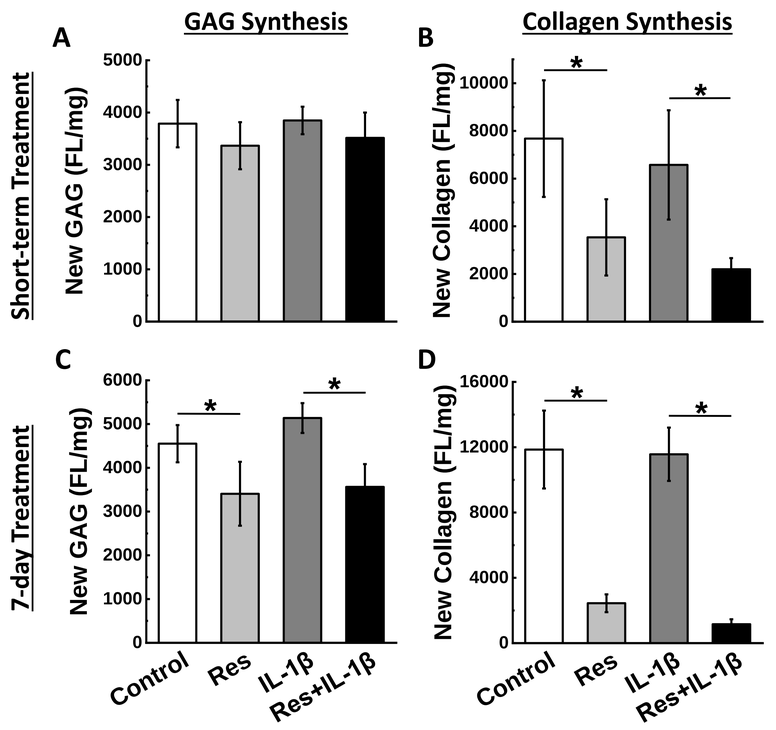



**Figure 3.** Effects of 100 μM resveratrol and 1 ng/ml IL-1β on the GAG and collagen synthesis by chondrocytes in cartilage explant. (A-B) GAG synthesis over 24 hours and collagen synthesis over 48 hours in four different culture conditions. Resveratrol did not affect the GAG synthesis, but significantly decreased the collagen synthesis regardless of IL-1β co-treatment (n=6). (C-D) After 7-day treatment, IL-1β had no effects on ECM synthesis. Resveratrol inhibited both GAG and collagen synthesis (n = 6; *: p<0.05).

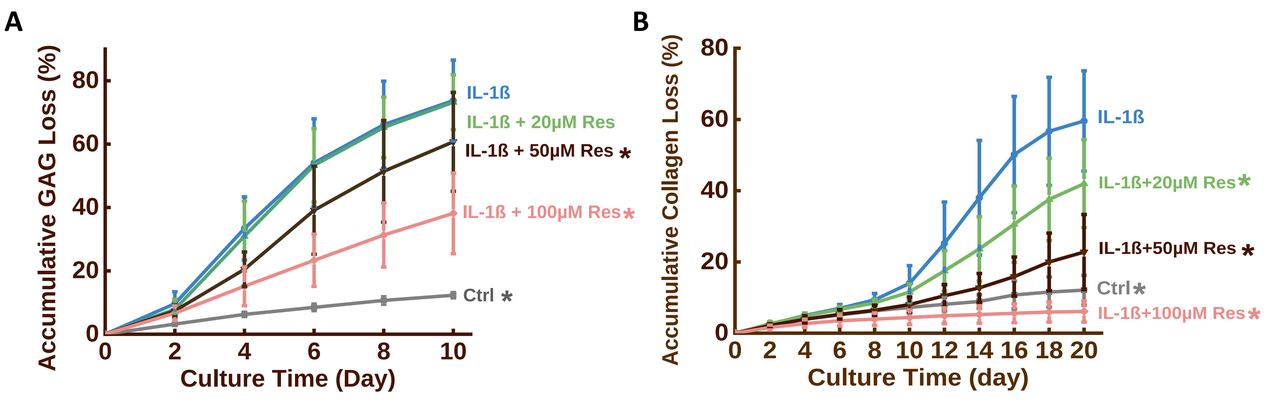



**Figure 4.** Resveratrol effectively inhibited the IL-1β-induced ECM loss with a dose-dependent manner. (A) Accumulative GAG loss from cartilage explant during 10-day culture. (B) Accumulative collagen loss from cartilage explants during 20-day culture. (n = 12; *: p<0.05, compared with IL-1β group)

## Conclusions

**Resveratrol regulates ECM synthesis by chondrocytes.**

- Short-term (2 days) treatment of resveratrol significantly reduces the synthesis of collagen but does not affect the synthesis rate of GAG in cartilage explants.
- Seven-day treatment of resveratrol significantly inhibited both GAG and collagen synthesis during the *in vitro* culture of cartilage explants.
- Resveratrol inhibits the proliferation of chondrocytes during *in vitro* culture.

**Resveratrol prevents inflammation-induced ECM degradation.**

- The reduced loss of GAG and collagen contents indicates that resveratrol can effectively protect cartilage from ECM degradation induced by pro-inflammatory cytokine.
- For chondrocytes under inflammatory attack, resveratrol can inhibit their catabolic activities, protect ECM integrity, and reduce the compensatory synthesis of collagen and GAG.