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Does resveratrol act independently of SIRT1 to affect genes relevant to ageing?

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Some effects of the polyphenol resveratrol mimic responses to dietary restriction. Dietary resveratrol may thus have the potential to prolong lifespan and reduce the risk of age related diseases, such as cardiovascular disease, diabetes and cancer. Actions of the histone deacetylase Sirt1 are one of several pathways believed to play a role in responses to dietary restriction. Controversy remains over whether or not resveratrol is an activator of SIRT1 and thus over if SIRT1 is the conduit for the beneficial actions of resveratrol.

Previous work in human intestinal Caco-2 cells, based on DNA microarray analysis, indicated that resveratrol and SIRT1 affect distinct sets of genes, although genes affected by dietary restriction were significantly over-represented in both groups. The aim was to investigate if resveratrol and SIRT1 act independently to affect the expression of different genes with functions relevant to ageing.

Clusterin and *WWP1* were selected from the group of genes previously found to be affected by resveratrol and dietary restriction. *Clusterin* has been detected at high levels in Alzheimer's disease and modulates plaque solubility. *WWP1* is a ubiquitin ligase that increased lifespan when overexpressed in *C. elegans* and when mutated led to abolition of the longevity response to dietary restriction. *LAMP2*, a key player in chaperone-mediated autophagy, which modulates cellular ageing, was selected from the group of genes that were affected by SIRT1 and dietary restriction. RT-qPCR was used to measure mRNA levels of all three genes and of *SIRT1*in human intestinal Caco-2 cells after treatment with resveratrol or reduction of SIRT1 expression using siRNA.

Results are shown in Figure 1. SIRT1 knockdown increased *Clusterin* and *WWP1* mRNA and decreased *LAMP2* mRNA. Resveratrol decreased *WWP1* mRNA and increased SIRT1 mRNA. Resveratrol also increased transcription from a *SIRT1* promoter-reporter construct.

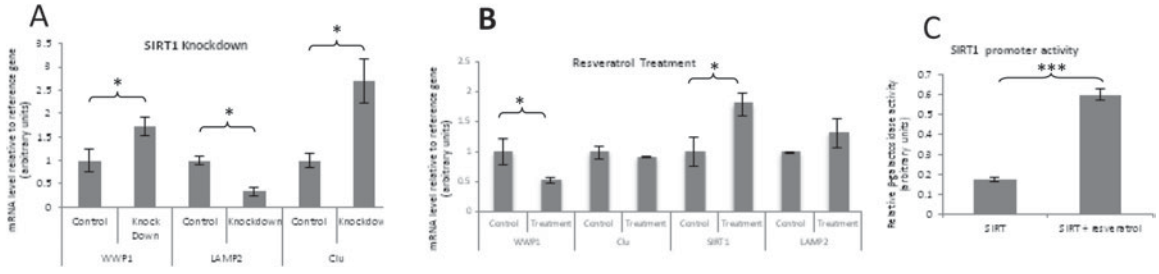

Fig. 1. A. mRNA levels of *WWP1, Clusterin (Clu)* and *LAMP2* relative to GAPDH in response to SIRT1 knockdown in Caco-2 cells **B.** mRNA levels of *WWP1, Clusterin, LAMP2* and *SIRT1* relative to GAPDH in response to treatment of Caco-2 cells over 24 h with 10 μM resveratrol. **C.** *SIRT1* promoter-reporter assay to investigate the effect of resveratrol on the transcription of the *SIRT1* gene. Data are mean, SEM ($n = 3$). $^{*}P<0.05$; $^{***}P<0.001$ by Student's *t*-test.

The observed response of the *WWP1* gene to SIRT1 knockdown and to resveratrol is consistent with resveratrol acting via SIRT1. The observed effects of resveratrol on SIRT1 expression indicate that resveratrol can affect the expression of genes through increasing *SIRT1* transcription. Thus effects of resveratrol on gene expression are not all independent of actions on SIRT1.