Vitamin D Impacts Nrf-2 Signaling and Antioxidant Pathways that Modulate Cellular Aging

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Research Question: Does Vitamin D increase Nrf-2 activity?

Methodology

- The nuclear factor (erythroid-derived 2)-like 2 protein (Nrf-2) is a potential therapeutic target against oxidative stress, protection against disease, and induction of cellular proliferation.
- It has been proposed that the active vitamin D hormone, 1,25-dihydroxyvitamin D (1,25D), is able to effectively promote "healthful aging". One possible mediator of this effect is through association of 1,25D/VDR with Nrf-2 signaling.

The Optimal Concentration of Nrf-2 Plasmid

The 1,25D target gene, CYP24a1, was measured as a positive control. In this study, 1,25D/DMSO treated cells showed the highest concentration of Nrf-2 while the lowest concentration of 1,25D revealed an increase in Nrf-2 activity.

Table 1. Summary of Expression of Nrf-2 Target Genes

<table>
<thead>
<tr>
<th>Treatment</th>
<th>CYP</th>
<th>GCLC</th>
<th>HMOX1</th>
</tr>
</thead>
<tbody>
<tr>
<td>1,25D/DMSO</td>
<td>1.6</td>
<td>3.7</td>
<td></td>
</tr>
<tr>
<td>10^-10M 1,25D/UA</td>
<td>0.93</td>
<td>0.86</td>
<td></td>
</tr>
<tr>
<td>5x10^-10M 1,25D/UA</td>
<td>0.59</td>
<td>0.74</td>
<td></td>
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</tbody>
</table>

Figure 2. Investigation of different concentrations of pcDNA3.1-Nrf2 to identify a dose-response relationship for the ARE-luciferase plasmid in the presence and absence of 1,25D.

Conclusions

- 1,25D has the ability to inhibit Nrf-2/ARE activity, however this effect is reversed by the addition of UA.
- 50 ng of 1,25D expression plasmid represented the most responsive ARE-luciferase reporter system.
- 1,25D-VDR has a DUAL role in Nrf-2/anti-oxidation. High concentrations of 1,25D inhibit Nrf-2 activity, lower concentrations boost Nrf-2 activity.
- Inhibition by high levels of 1,25D was observed in Nrf-2 target genes (GCLC and HMOX1) as measured by qPCR.
- Modulation of Nrf-2 activity by the vitamin D pathway and by Urolithin A may have a regulatory role in anti-oxidation and cellular aging pathways.

Future Directions

- We will continue to examine how 1,25D interacts with or affects Nrf-2 /pathway by performing the following:
  - A series of qPCRs utilizing Nrf-2 target genes to test the effects of different concentrations of 1,25D and urolithin A on expression of these anti-oxidation genes.
  - Continue to test 1,25D impact on the Nrf-2 pathway using the ARE-luciferase system.
  - Further test additional pathways in the presence of 1,25D and/or UA.
  - How and where 1,25D/VDR interacts in the Nrf-2 pathway, including potential association with the Maf protein.

Acknowledgments/References

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