Oxidative stress in vascular calcification: cause or consequence?

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Vascular calcification is one of current threats in chronic kidney disease. An increase in reactive oxygen species (ROS) occurs during vascular calcification. H2O2 is a causative factor of the trans-differentiation from vascular smooth muscle cells (VSMC) to osteoblast-like cells. Other lines of thought suggested that increased ROS is a consequence of vascular calcification process, highlighting the utility of ROS as markers of this process.

The cells used were both A7r5 rat cell line and primary cells from catalase over-expressing mice. To induce VSMC phenotypic changes, cells were cultured in DMEM / F12 + 0.1% BSA (Control) + Calcium and Phosphorus 2mM and 3mM (pro-calcifying) for 8 days. Mineral deposits were determined by Alizarin Red staining. Fluorescent probes were used to determine the presence of different ROS in VSMC by flow cytometry. Levels for catalase, MnSOD, Runx2, and markers of oxidative modification (nitrotyrosine) were determined by Western blot.

After 8 days with pro-calcifying treatment, increased deposition of calcium, expresion of Runx2 and MnSOD2 and H2O2 and O2 levels were observed. This resulted in increased nitrotirosine levels.

However, primary cultures of VSMCs overexpressing catalase showed prevention of both mineralization and ROS increase.

Increased oxidative stress in later stages of the development of vascular calcification suggests that higher H2O2, MnSOD and nitrotirosine might be a consequence of this process; however, the ability to prevent mineralization exhibited by the cells with higher levels of catalase, points at the increase in H2O2 levels as independent cause of vascular calcification.