SHORT COMMUNICATION

NANOTECHNOLOGY IN BIOMINERALIZATION: PROTEOLIPOSOMES AS MVs BIOMIMETICS

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During endochondral bone formation, chondrocytes and osteoblasts are responsible for the synthesis and mineralization of the extracellular matrix through a carefully orchestrated process, believed to initiate within membrane-invested matrix vesicles (MVs) at the sites of initiation of hydroxyapatite (HA) deposition and end with bone mineral propagation onto the collagenous scaffold. Inorganic Pyrophosphate (PPi), derived both from ENPP1-catalyzed production from extracellular nucleoside triphosphates and by cellular export via the ANK transporter, inhibits matrix mineralization. This inhibition is released through the action of TNAP, which hydrolyzes PPi, thus simultaneously removing the inhibitor and providing additional Pi for mineral formation. Ca2+ enters MVs via an annexin channels and phosphate enters via a type III Na+-dependent phosphate transporter (PiT), and possibly others, to form apatite within MVs. Acidic phospholipids and other MV components are thought to nucleate these intravesicular nanocrystals. Subsequently, the intravesicular mineral grows beyond the confines of MVs onto a collagenous matrix aided by a number of promoters and inhibitors of calcification. HA crystals are still present in TNAP-deficient MVs, and it has been shown that the soluble MV phosphatase PHOSPHO1 is involved in intravesicular mineralization. As we strive to understand the physiological interplay between TNAP, PHOSPHO1, NPP1, and other important MV-associated enzymes and channeling proteins in the initiation of biomineralization, we must keep in mind the microenvironment in which these proteins function, which can have a profound effect on their biological properties, since phospholipids play an important role in the initiation of the biomineralization process. The ability of synthetic or natural vesicles to mimic the organizational structure and function of biomembranes makes these structures an advantageous and convenient experimental model to help us advance our understanding of MV-mediated calcification. The proteoliposome system provides a means of reconstituting lipid vesicles that will function like MVs. Proteoliposomes can be manufactured using different methods and with controlled lipid and protein composition, electrolytes and sizes, representing a convenient experimental model to mimic the organizational structure and function of natural biomembranes and to reproduce some essential features of the biomineralization process. Such an MV biomimetic proteoliposome system would be useful for many important translational applications. The enzymatic defects associated with disease-causing mutations in the TNAP molecule, such as those found in hypophosphatasia, could be further elucidated in a membrane vesicle that better mimics their in vivo biological environment. This proteoliposome system can be used for the screening of small molecule

compounds able to modulate (inhibit or activate) the activity of MV enzymes for potential therapeutic uses. Once built and characterized, the proteoliposomes can be added to fixed amounts of MVs, wild-type or deficient in specific enzymes, as a way of modulating their in vitro calcification properties. Those experiments would validate the use of these nanovesicles in promoting or delaying calcification, and such an artificial nanovesicular system could also potentially prove useful for the repair/treatment of craniofacial and other skeletal defects and to facilitate the mineralization of titanium-based tooth implants. In addition, the property of liposome to carrying in aqueous or lipid bilayers could be used as a system for delivery of drug or dyes with many applications.

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