Review

Intrinsically disordered proteins and biomineralization

Adele L. Boskey and Eduardo Villarreal-Ramirez

Musculoskeletal Integrity Program, *Hospital for Special Surgery, New York, NY 10021, USA*

Correspondence to Adele L. Boskey: *Hospital for Special Surgery, 535 E 70th Street, New York, NY 10021, USA. Boskeya@HSS.edu.*
http://dx.doi.org/10.1016/j.matbio.2016.01.007
Edited by Marian Young

Abstract

In vertebrates and invertebrates, biomineralization is controlled by the cell and the proteins they produce. A large number of these proteins are intrinsically disordered, gaining some secondary structure when they interact with their binding partners. These partners include the component ions of the mineral being deposited, the crystals themselves, the template on which the initial crystals form, and other intrinsically disordered proteins and peptides. This review speculates why intrinsically disordered proteins are so important for biomineralization, providing illustrations from the SIBLING (small integrin binding N-glycosylated) proteins and their peptides. It is concluded that the flexible structure, and the ability of the intrinsically disordered proteins to bind to a multitude of surfaces is crucial, but details on the precise-interactions, energetics and kinetics of binding remain to be determined.

Introduction

Composition of mineralized tissues

Biomineralization occurs in a variety of organisms, from unicellular bacteria containing gold deposits [1] to diatoms [2] having silicon based mineral deposits, shells with calcium carbonate phases (aragonite, calcite and occasionally vaterite as well as amorphous calcium carbonate) [3], to hierarchical structures in vertebrate bones and teeth containing the calcium phosphate, hydroxyapatite (HA) [4]. Common features of these mineralized structures are (i) formation on an organic matrix, produced by the cells of that organism. The organic matrix within each organism or within their tissues, extra- or intracellular, plays a role in the mineralization process. This is well-known due to the observation that the removal or modification of the matrix, or any given component within the matrix, alters its properties and thus the manner in which the mineral is formed (Table 1). (ii) An additional feature of the matrix proteins regulating mineralization is that a large percentage of them, greater than that within the general protein data base [5], are intrinsically disordered proteins (IDPs). In this review, we suggest the reasons IDPs[†] are essential for the biomineralization process and provide examples to illustrate its importance.

Overview of the process of mineralization

Mineral deposition is a physical–chemical process that, in living organisms, is regulated by cells and by the intra- and extra-cellular matrices they produce. In purely chemical terms, a mineral will not precipitate unless its solubility is exceeded, either by having an increase in the concentration of its component ions,

[†] Abbreviation; BSP — bone sialoprotein; CAP — carbonate hydroxyapatite; CD — circular dichroism; DD — dentinal dysplasia; DGI — dentinogenesis imperfecta; DMP1 — dentin matrix protein 1; DPP — phosphophoryn; DSP — dentin sialophosphoprotein; DSPP — dentin sialophosphoprotein gene; HA — hydroxyapatite; IDP — intrinsically disordered protein; KO — knockout; MEPE — matrix extracellular phosphoglycoprotein; OI — osteogenesis imperfecta; OPN — osteopontin; SIBLING — small ligand binding N-glycoslated.

Table 1. Examples of alterations in matrix composition that affect the mineralization process.

Species	Type of mineral	Treatment of matrix	Change in mineral
Magnetic bacteria [7]	Magnetite (Fe_3O_4)	Deletion of operon	Irregular crystals
Avian egg shell [8]	Calcite	Incubation in dermatan sulfate	Delays formation
Abalone shell [9]	Aragonite/calcite	Perlucin variants	Modulate rate of precipitation
Diatoms [10]	Silicate	Composite of silk and silaffin	Promotes mineralization
Mice [11]	Hydroxyapatite	Knockout or overexpression of ameloblastin	Decreased enamel formation
Mice [12]	Hydroxyapatite	Knockout of phospho-1	Smaller crystals
Vertebrate [13]	Hydroxyapatite	Dephosphorylation of osteopontin	No inhibition of growth
Vertebrate [14]	Hydroxyapatite	Dephosphorylation of all phosphoproteins	Decreases extent of mineralization
Zebra-fish [15]	Hydroxyapatite	Mutation in BMP1 (enzyme)	Defective mineralization

a change in temperature or pressure, or the addition of a surface that lowers the activation energy for the initial mineral formation (Fig. 1). Precursor phases such as pre-nucleation clusters [6] and amorphous calcium phosphate can form in solution, reducing the activation energy barrier. Within the organism, the cells and vascular system provide the necessary increases in the concentration of component ions for initial crystal formation (critical nucleus). Intra- or extra-cellular matrices may also provide surfaces that facilitate this initial mineral deposition. Inhibitors of mineralization protect the matrix or extracellular matrix from being mineralized, when mineral is neither needed nor wanted, and are modified or removed by enzymes produced by the cell. This entire process is highly complex, even in the simplest organism, yet common themes exist; the elevation of local concentrations of precipitating ions and the regulation of the process by the extracellular matrix.

Protein regulation of mineralization

According to classic nucleation theory, deposition of crystals requires the formation of nuclei which support the growth and proliferation of mineral crystals. Nucleation can take place, de novo, when solution ionic concentrations are increased and ions collide with sufficient energy to form a critical nucleus. This critical nucleus has the same structure, only a few unit cells in size, as does the final crystal. Once a critical nucleus is formed, less energy is required for the addition of ions to the nucleus, thus allowing for growth or branching which facilitates the spread of the crystals. Pre-nucleation clusters or foreign surfaces can resemble the critical nucleus or the aggregates of these pre-nucleation clusters might themselves transform into crystals which will then grow, facilitating mineral deposition. It is believed that in the process of biomineralization, the matrix has a structure resembling that of the

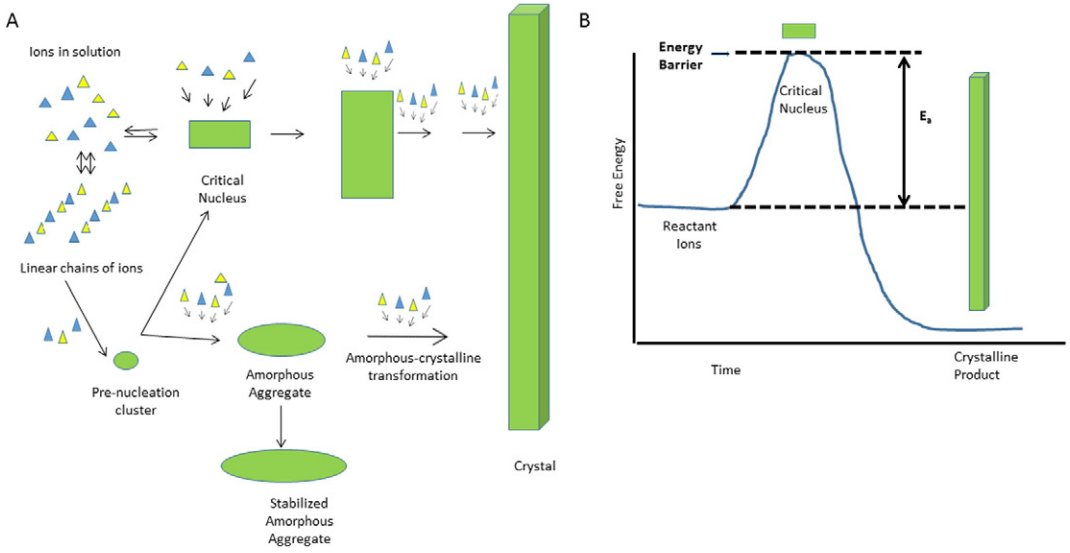

Fig. 1. Physical chemistry of mineralization. A) Schematic illustrating how ions in solution can either form linear chains of ions, critical nucleus (the classical concept of nucleation) or unstructured (amorphous) clusters. The critical nucleus continues to accumulate ions and develops into a crystal. The clusters may also associate to form a "critical nucleus". Proteins can interact with any of these forms to facilitate or inhibit the next step. B) The major expenditure of energy is in the formation of the critical nucleus as illustrated by this plot of activation energy vs. time.