Journal of Oral Biosciences 55 (2013) 85–90

Contents lists available at SciVerse ScienceDirect

Journal of Oral Biosciences

journal homepage: www.elsevier.com/locate/job

Review

Odontogenic ameloblast-associated protein (ODAM) and amelotin: Major players in hypermineralization of enamel and enameloid

Kazuhiko Kawasaki *

Department of Anthropology, Pennsylvania State University, 409 Carpenter Building, University Park, PA 16802, USA

ARTICLE INFO

Article history:
Received 22 December 2012
Received in revised form
21 January 2013
Accepted 2 February 2013
Available online 22 April 2013

Keywords:
Dental enamel
Amelogenesis
Basement membrane
Amelotin
Odontogenic ameloblast-associated protein (ODAM)

ABSTRACT

Odontogenic ameloblast-associated protein (ODAM) and amelotin (AMTN) both belong to the secretory calcium-binding phosphoprotein family, which is critical to biomineralization in vertebrates. In mammals, both *ODAM* and *AMTN* are expressed by ameloblasts in the maturation stage, when immature enamel grows into a hypermineralized inorganic tissue. At the onset of this stage, ameloblasts produce a specialized basal lamina (BL), over which both ODAM and AMTN are distributed. Enameloid is a different hypermineralized tissue that is found on the tooth surface of most ray-finned fish. Unlike amelogenesis, no such BL is produced during the maturation of enameloid. Nevertheless, *ODAM* is also found in ray-finned fish, and the expression of this gene has been detected in inner dental epithelial cells, which correspond to ameloblasts, after the enameloid is considerably mineralized. This specific gene expression suggests that ODAM is not a constituent of the BL but is still involved in the hypermineralization of enameloid. Both ODAM and AMTN are unusually rich in Pro and Gln, and they have 1 or 2 clusters of phospho-Ser residues. These characteristics suggest that ODAM and AMTN associate with weak interactions between relatively hydrophobic regions and further bind calcium phosphate via phospho-Ser clusters, similar to milk caseins that are evolutionary descendants of ODAM. Based on these considerations, I hypothesized that ODAM and AMTN generate and maintain the interface between unmineralized and hypermineralizing domains through weak protein–protein interactions and associations with calcium phosphate. This interface presumably facilitates hypermineralization, efficient removal of degraded proteins from the matrix, and the transfer of calcium phosphate to the matrix.

Contents

1. Introduction

The secretory calcium-binding phosphoprotein (SCPP) gene family is essential to biomineralization in vertebrates [1–3]. SCPP genes arose from a common ancestor by gene duplication [4], and they are classified into 2 types, either acidic or Pro and/or Gln (P/Q)-rich, based on the amino acid composition of the coded protein [5]. In tetrapods, acidic SCPP genes are involved in the mineralization of bone and/or dentin, whereas many P/Q-rich SCPP genes are expressed during amelogenesis. For example, P/Q-rich SCPPs include 3 principal enamel matrix proteins—amelogenin, ameloblastin, and enamelin. These matrix proteins are secreted by

* Tel.: +1 814 863 0032; fax: +1 814 863 1474.
E-mail address: kuk2@psu.edu

1349-0079/$ - see front matter © 2013 Japanese Association for Oral Biology. Published by Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.job.2013.02.001

ameloblasts and constitute the initial organic matrix. However, these proteins are subsequently processed by proteolytic digestion and eventually removed from the matrix [6]. The enamel, thus, matures into a hypermineralized inorganic tissue [7].

The enamel is commonly found in sarcopterygians (tetrapods, lungfish, and coelacanth), but it is not common in other vertebrates. In cartilaginous fish and most actinopterygians (ray-finned fish), the tooth surface is covered with enameloid instead of the enamel [8]. Unlike the enamel, the matrix of enameloid contains collagen and is secreted mainly by odontoblasts in cartilaginous fish and by both odontoblasts and inner dental epithelial cells (IDE cells, which correspond to ameloblasts) in ray-finned fish [5,9–12]. Nevertheless, enameloid grows into a hypermineralized tissue through maturation processes, similar to enamel. Interestingly, the odontogenic ameloblast-associated protein (ODAM, previously called APin) gene is expressed in the maturation stage of the enamel in mammals and enameloid in the zebrafish [12,13]. These findings suggest that *ODAM* is a major player in hypermineralization [3].

The initial study of *ODAM* identified only a 3′ region of this gene, which is expressed in the enamel organ but not in the dental pulp or other organs in the rat [14]. Subsequently, ODAM was found in amyloid obtained from patients with calcifying epithelial odontogenic tumors [15]. However, this protein turned out to be a C-terminal portion of the entire ODAM [16]. More recently, Moffat et al. described the entire *ODAM* gene in mammals [17]. In fact, prior to this report, we discovered this gene in fugu as a member of the SCPP gene family and named it *SCPP2* [18]. Subsequently, *SCPP2*/*ODAM* was identified in the frog and the zebrafish [12].

Before details of *ODAM* were reported, amelotin (*AMTN*) was identified as a gene that was expressed in maturation-stage ameloblasts but not in odontoblasts, dental pulp cells, or alveolar osteoblasts [19]. Independently, *AMTN* was characterized as a gene that was expressed in the enamel organ in the maturation stage of amelogenesis [17,20]. Furthermore, we reported *AMTN* as an uncharacterized SCPP gene, showing a close affinity to the ameloblastin gene (*AMBN*) and the enamelin gene (*ENAM*) [21].

In addition to *ODAM* and *AMTN*, SCPP-P/Q-rich 1 (SCPPPQ1) was cloned as a P/Q-rich SCPP gene from the jaw of a neonate mouse [12], and it was identified as a gene that was expressed in the enamel organ [17] in the maturation stage [22]. Whereas we detected *SCPPPQ1* in the lizard [23], the expression of this gene has not yet been confirmed in humans. No detailed analysis has been reported for this gene.

In addition to ameloblasts, both ODAM and AMTN have been immunolocalized to the junctional epithelium and various tumors. However, in this review, I will focus on the roles of these genes in the maturation of the enamel and/or enameloid. As described below, both ODAM and AMTN are distributed over the basal lamina (BL); hence, I begin by discussing the BL formed during amelogenesis.

2. Basal laminae formed during amelogenesis

The BL evolved early in the history of Eumetazoa, which includes jellyfish and the hydra among modern species [24]. These animals have laminin, type-IV collagen, perlecan (a core protein of heparan-sulfate proteoglycan; HSPG), and nidogen, the 4 major constituents of the BL in vertebrates [25]. At the epidermal–dermal junction of mammalian skin, laminins assemble with other molecules, including type-IV collagen and nidogen, and form an electron-dense layer, called the lamina densa, which is separated from the keratinocyte plasma membrane by an electron-lucent layer, the lamina lucida. Keratinocytes tightly attach to the BL through hemidesmosomes (HDs). The core component of the HD is integrin $\alpha_6\beta_4$, a heterodimer of transmembrane proteins, which binds laminin 332 in the BL [26]. The HD protects the tissue from distractive shear forces; and mutations in any of the 3 components of laminin 332, laminin $\alpha3$, $\beta3$, and $\gamma2$, have been associated with junctional epidermolysis bullosa, which is characterized by blister formation within the lamina lucida [27].

In the presecretory stage of amelogenesis, a BL separates the layer of preameloblasts from the dental pulp [28–31] (Fig. 1). This BL consists of type-IV collagen, laminin 332, nidogen, and HSPG [32–38], similar to the skin BL. Indeed, junctional epidermolysis bullosa is accompanied by enamel hypoplasia [39], and mice with a disrupted laminin $\alpha3$ gene show abnormalities in the differentiation of ameloblasts [40]. As the initial dentin matrix is laid down, preameloblasts extend cell processes through the BL to the predentin; shortly thereafter, the BL disintegrates before the predentin initiates mineralization [41] (Fig. 1). After a thin layer of enamel matrix accumulates on the mineralized dentin surface, the ameloblast extends the Tomes' process and secretes matrix proteins from 2 sites, the proximal and the distal portion of the Tomes' process [42], forming an interrod and a rod, respectively [43]. The rod–interrod structure (enamel prism) is found in the enamel of most mammals and *Uromastyx* among non-mammals [44]. Because the Tomes' process is not formed at the beginning and at the end of the secretory stage, the initial and final layers of the enamel are prismless [45,46].

After the full thickness of the immature enamel matrix is formed, ameloblasts undergo a major reorganization, preparing for the maturation of the enamel [29,30,47]. In this transition stage, ameloblasts reestablish the BL, which is sustained throughout the maturation stage [48,49] (Fig. 1). In the maturation stage, enamel matrix proteins are proteolytically degraded and removed from the matrix, which allows preexisting crystals to grow width and thickness [50]. The maturation-stage ameloblasts modulate the morphology between a highly invaginated, ruffle-ended apical surface and a smooth-ended surface [51]. The ruffle-ended ameloblasts show endocytotic activity and appear to pump calcium ions into the enamel through the plasma membrane Ca^{2+}-ATPase [52],

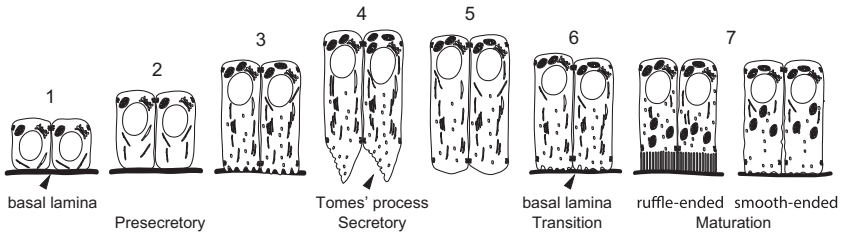

Fig. 1. Amelogenesis and the basal lamina (BL) in mammals. Presecretory ameloblasts differentiate from inner dental epithelial cells (1–3) and extend their cell processes through the BL (3). Shortly thereafter, the BL disintegrates. In the secretory stage, a thin layer of prismless enamel is initially formed, and then ameloblasts develop the Tomes' process and produce prismatic enamel (4). At the end of the secretory stage, the Tomes' process recesses and the prismless final enamel is formed (5). In the transition stage, ameloblasts reestablish a highly specialized BL (6). This BL is sustained throughout the maturation stage, while ameloblasts modulate the morphology between a ruffle-ended apical surface and a smooth-ended surface (7). See Hu et al. [29] for more detail.