Annals of Anatomy 195 (2013) 475–483

Contents lists available at ScienceDirect

Annals of Anatomy

journal homepage: www.elsevier.de/aanat

Research article

Development of the oxytalan fiber system in the periodontal space of rat incisors

Kouji Inoue [a,*], Yaiko Hara [b], Noriyuki Kuroda [b], Tetsuji Sato [b]

[a] Research Center of Electron Microscopy, School of Dental Medicine, Tsurumi University, Yokohama 230-8501, Japan
[b] Department of Anatomy and Histocytology, School of Dental Medicine, Tsurumi University, Yokohama 230-8501, Japan

ARTICLE INFO

Article history:
Received 26 February 2013
Received in revised form 29 March 2013
Accepted 30 March 2013

Keywords:
Oxytalan fiber system
Incisor
Periodontal space
Enamel side
Cementum side
Development
Rat

SUMMARY

The present study clarifies developmental organization of the oxytalan fiber system in the periodontal space of both the enamel (labial) and cementum (lingual) sides of rat incisors. The number of oxytalan fibers per unit area (μm^2) was counted in rat incisors at stages of embryonic day 20 (E20) to postnatal day 35 (P35). Oxytalan fibers in the periodontal space of the enamel side were apt to decrease in number during the postnatal period, whereas their number remained almost unchanged on the cementum side during the developmental period. When the incisor emerged through the gum at P11, thinner oxytalan fibers distributed in the apical growing periodontium of the cementum side seemed to be fused with one another to become thicker fibers as has been reported for rat molars (Inoue et al., 2012). Thus, the oxytalan fiber system in the periodontal space represented significant differences in its distributional density between the enamel and cementum sides after E23. At the stage of P35, oxytalan fibers presented significantly denser distribution in all territories of the periodontal ligament of the cementum side versus the enamel side. The present findings claim that the oxytalan fiber system might bind the tooth to the periodontal ligament and provide equilibrium of vascular system and control of blood flow in the periodontal ligament of the cementum side, while it might exclusively regulate the high level of physiologically adapted vasculature in the periodontal space of the enamel side.

1. Introduction

In our previous study (Inoue et al., 2012), we demonstrated developmental topographic organizations of the oxytalan fiber system in the periodontal ligament of rat molars using specific stainings for oxytalan, elastic and collagen fibers, and electron-microscopic analyses. The elastic fiber system can be divided into three groups consisting of oxytalan, elaunin and elastic fibers, depending on the relative populations of microfibrils and elastin. Interestingly, only oxytalan fibers have been identified in the periodontal ligament, whereas all types of the elastic fiber system have been demonstrated in the gingiva (Chavrier et al., 1988; Sculean et al., 1999; Inoue et al., 2012). We have clearly confirmed the earliest oxytalan fibers in a bell-staged tooth germ of rat molars at embryonic day 18, using the specific staining for oxytalan fibers (Inoue et al., 2012). As development proceeds, the oxytalan fiber system composed of longer fibers is located in the apico-occlusal guidance together with vascular vessels. In addition, immunohistochemical and electron-microscopic analyses strongly suggest the adhesion of oxytalan fibers to vascular basement membranes. Our previous results claim that the oxytalan fiber system regulates periodontal ligament function through tensional variations registered on the walls of vascular structures.

In the rat, the periodontal ligament plays an active function when the incisor emerges through the gingiva during the process of tooth development (Berkovitz and Thomas, 1969; Berkovitz, 1971; Beertsen et al., 1974). Rat incisors are rootless teeth and continually increasing in size, and thus its periodontal ligament may differ markedly from molar periodontal ligament of rooted tooth. The connective tissue in the periodontal ligament of the cementum (lingual) side of the incisor fastens the tooth to the supporting alveolar bone. This connective tissue appears to be made up of two regions varied in histological structures: an avascular, cell-rich territory adjacent to the tooth, and a territory over the alveolar bone including the connective tissue located in the perivascular spaces (Beertsen et al., 1979). Although it has been suggested that the oxytalan fiber system in the incisor periodontal ligament probably remains stationary in the tooth eruption (Shore et al., 1985), no decisive data in favor of this sight have presented themselves from the literature. Furthermore, information concerning how the connective tissue on the enamel (labial) side of the incisor tooth

* Corresponding author at: Research Center of Electron Microscopy, School of Dental Medicine, Tsurumi University, 2-1-3 Tsurumi, Tsurumi-ku, Yokohama 230-8501, Japan. Tel.: +81 45 581 1001; fax: +81 45 573 9599.
E-mail address: inoue-k@tsurumi-u.ac.jp (K. Inoue).

0940-9602/$ – see front matter © 2013 Elsevier GmbH. All rights reserved.
http://dx.doi.org/10.1016/j.aanat.2013.03.012

functions and the specific organizations of oxytalan fibers during the development of the incisor periodontal ligament have not been obtained. In this investigation, we have examined the developing oxytalan fiber system on both the enamel and cementum sides of the rat incisor by means of light and electron microscopic analyses. Thus, the aim of this study was to compare the present results on the oxytalan fiber system in the incisor with those on it in the molar obtained in the previous study (Inoue et al., 2012), and to delineate its putative function in the periodontal ligament.

2. Materials and methods

In the present study, we explored the development of the oxytalan fiber system in the rat incisor from embryonic day 13 to postnatal day 60 (E13–P60) using light- and electron-microscopy.

2.1. Animals

Seventy-six male or female Wistar rats including fetuses were used for this study. Female rats were mated and the day of the first appearance of spermatozoa in vaginal smears was denoted embryonic day 1 (E1). The day of birth was designated postnatal day 0 (P0). At E13, E17, E18, E20, E23 ($n = 8$ each for E13–23), P1, P2, and P11 ($n = 6$ each for P1–11), the mandibles including dental germs were enucleated under deep narcosis with intra-peritoneal injection of a mixture of 0.15 mg/kg medetomidine, 2.0 mg/kg midazolam, and 2.5 mg/kg butorphanol. Narcotized postnatal rats at P14, P15, P35, and P60 ($n = 4$, 6, 6, 2 for P14–60, respectively) were perfusion-fixed. Postnatal animals in each age-group were divided into two, which included one half of the group being used for light microscopic studies and the other for electron microscopic observations. The use and treatment of the animals in the present study followed the Guideline for the Treatment of Experimental Animals at the

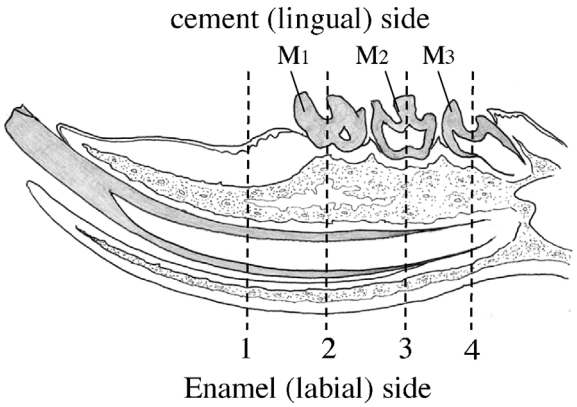

Fig. 1. Drawing showing sagittal sections in the lower jaw at postnatal day 35 (P35). The upper direction of the figure is the cementum (lingual) side, the lower is the enamel (labial) side, the left is the occlusal (incisal) side, and the right is the apical side. Broken lines (1–4) of the incisor give faces sectioned in labial-lingual directions shown in Fig. 8.

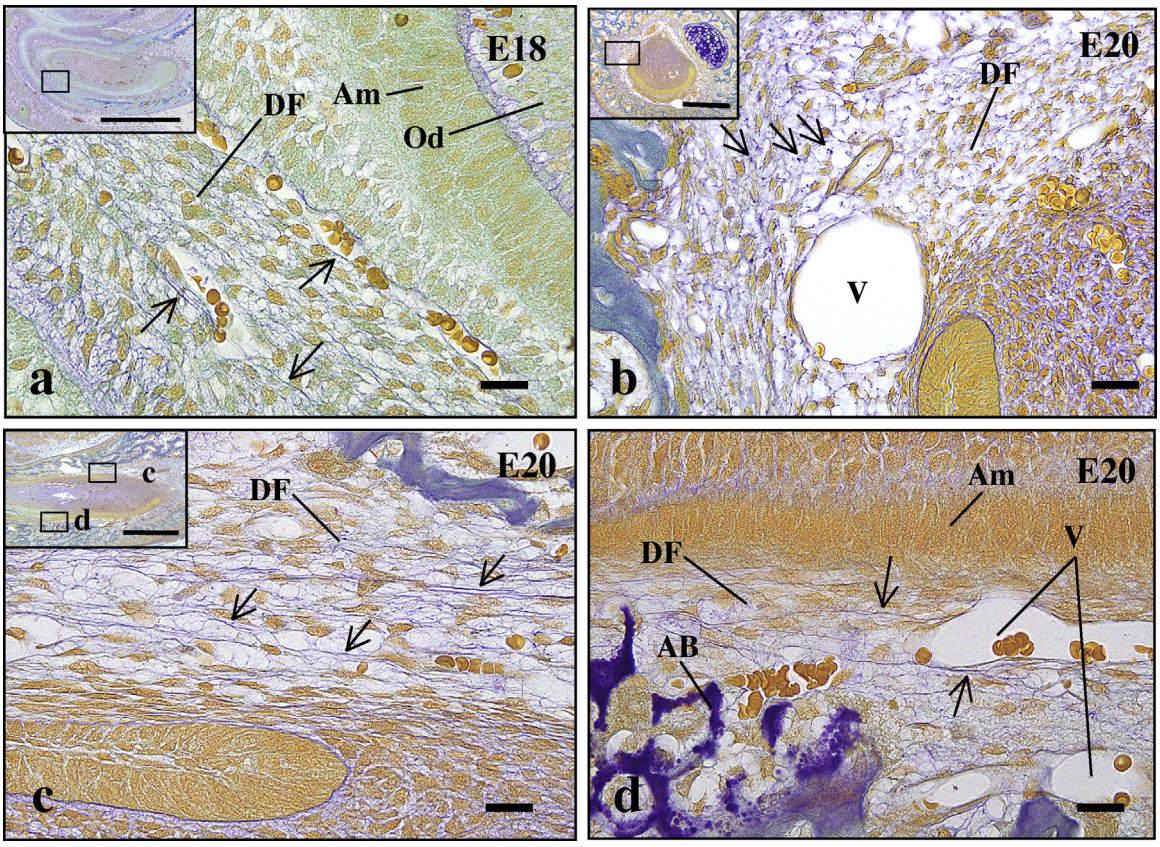

Fig. 2. Tooth germ of a bell-stage in a rat incisor. *Insets* in figures (a–c) represent lower magnifications of the tooth germ and figures (a–d) represent higher magnification of the respective regions marked by *black rectangles* in the *insets* of figures a–c. (a) In a bell-stage of the tooth germ at embryonic day 18 (E18), oxytalan fibers (*arrows*) are labeled in the dental follicle (*DF*) using oxytalan staining. (b–d) At E20, oxytalan fibers (*arrows*) show to be arranged in parallel to the tooth axis on the cementum side of a transverse section (b), and on the cementum (c) and enamel sides (d) of a lingual-labial directed section. The left sides in figures a and c present an incisal part, and the right sides an apical part (a formative part). *Am*: ameloblast; *DF*: dental follicle; *AB*: alveolar bone, *Od*: odontoblast, *V*: blood vessel. *Scale bars* = 25 μm for a–d of the larger magnifications: *Scale bars* = 500 μm for the *insets* in figures a–c.