Pulp Regenerative Therapy Using Autologous Dental Pulp Stem Cells in a Mature Tooth with Apical Periodontitis: A Case Report

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ABSTRACT

The utility and feasibility of pulp regenerative therapy with autologous dental pulp stem cells (DPSCs) in mature teeth with irreversible pulpitis were clinically demonstrated. On the other hand, there is no evidence of the utility of DPSCs in mature teeth with apical periodontitis. The aim of this case report was to describe the potential utility of regenerative cell therapy in mature teeth with apical periodontitis. A 44-year-old man was referred for pulp regeneration due to a periapical lesion in his maxillary first premolar. Root canal disinfection was performed by irrigation and intracanal medication by nanobubbles with levofloxacin and amphotericin B in addition to conventional irrigation. Autologous DPSCs isolated from an extracted third molar were transplanted into the root canal after residual bacteria and fungi were below the detection level by polymerase chain reaction assay using universal genes to amplify specific regions within bacterial 16S ribosomal DNA and fungal ribosomal DNA (ITS1), respectively. There were no adverse events or systemic toxicity assessed for clinical evaluations during the 79-week-follow-up period and laboratory evaluations after 4 weeks. The affected tooth was responsive to the electric pulp test. Cone-beam computed tomographic imaging revealed a reduced lesion size, remission of the periapical tissue, and mineralized tissue formation in the apical part of the canal after 79 weeks. The signal intensity on magnetic resonance imaging of the regenerated tissue in the affected tooth was comparable to that of the normal pulp in the adjacent teeth after 24 weeks. This case report demonstrated the potential use of DPSCs for pulp regenerative therapy in mature teeth with apical periodontitis. *(J Endod 2024;50:189–195.)*

KEY WORDS

Apical periodontitis; case report; cone-beam computed tomographic imaging; dental pulp stem cells; nanobubbles; pulp regeneration

SIGNIFICANCE

The pulp regenerative therapy harnessing autologous dental pulp stem cells with granulocyte colony-stimulating factor after complete disinfection may be a favorable treatment option in permanent mature teeth with apical periodontitis.

Cell-free regenerative endodontic therapy (RET) has been used in treating necrotic immature teeth with apical periodontitis to allow dentinal wall thickening and continuous root development with apical closure as well as the resolution of clinical signs and symptoms and healing of periapical tissue[1]. This therapy also has been used in mature teeth with apical periodontitis, demonstrating a viable alternative treatment instead of conventional treatment[2,3]. However, the regenerated tissue is not true pulp tissue but rather ectopic tissues such as the periodontal ligament, cementum, and bone[4]. Cell-based RET, called pulp regenerative cell therapy, in mature teeth potentially regenerate a true pulp-dentin complex accompanied by reinnervation, revascularization, normal homeostatic, and immunomodulatory functions to re-establish tooth vitality and function[5]. A clinical study and case reports have demonstrated the utility of dental pulp stem cells (DPSCs) for irreversible pulpitis of mature teeth analyzed by electric pulp testing (EPT), dental radiography, and magnetic resonance imaging (MRI)[6-8]. Pulp regenerative cell therapy has the advantages of preventing infection and tooth fracture in mature teeth, especially with apical periodontitis, due to the recovery of various biological functions of vital pulp, including mechanical strengthening by lateral dentin formation with apical closure, metabolic processes of the dentin-pulp complex, and defense mechanisms against bacterial invasion into dentinal tubules and periapical tissues by outward flow of dentin fluid and immune actions[3].

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Regenerative therapy using autologous[9] and allogeneic stem cells from deciduous teeth[10] was performed in human traumatized immature teeth with apical periodontitis. However, in mature teeth with apical periodontitis, allogeneic mesenchymal stem cells (MSCs) either from the umbilical cord[11] or bone marrow[12] together with platelet-rich fibrin/platelet-rich plasma were used. Clinical success was demonstrated in the 2 reports assessed by the survival of a tooth, the absence of clinical symptoms, and radiographic evidence of reduction of the apical lesion size at more than a 1-year follow-up. In addition, pulp regeneration was suggested by the increased rate of the affected teeth with a positive pulp response on cold testing and EPT. However, histologic data are not included in these reports because of institutional review board restrictions and not demonstrated in the related animal experimental reports to confirm pulp regeneration. Furthermore, there is no clinical report on pulp regenerative therapy using DPSCs in mature teeth with apical periodontitis. Our previous histologic studies using canine models of mature teeth with apical periodontitis demonstrated that pulp tissues could be regenerated only if cell transplantation was performed in completely disinfected root canals[13]. Therefore, in this case report, we examined the potential utility of autologous DPSCs for pulp regenerative therapy in a mature tooth with apical periodontitis.

Case Report

Our provisional plan of pulp regenerative cell therapy as a clinical treatment was approved by the category II Certified Committee for Regenerative Medicine and further officially accepted by the Japanese Ministry of Health, Labour and Welfare (#PB5210021, #PB5220025) based on the Act of Securement and Safety of Regenerative Medicine.

Patient

A 44-year-old man was referred to our dental clinic for pulp regenerative cell therapy in a maxillary right first premolar. Clinical examination revealed caries on the medial and distal surfaces under metal inlay restoration (Fig. 1*Aa–Ac*). The tooth showed no clinical symptoms and a negative response to EPT (Vitality Scanner/Pulp Tester; Yoshida, Tokyo, Japan). Periodontal probing affirmed normal attachment and normal physiological mobility. An apical lesion with a periapical radiolucency was observed by periapical radiographic (Fig. 1*Ba*) and cone-beam computed tomographic (CBCT) examinations (Fig. 1*Bb–Bd*), and its total volume analyzed by OsiriX (Bernex, Switzerland) medical imaging software was 17.6 mm^3. The diagnosis for the tooth was asymptomatic apical periodontitis. The medical history, including no history of allergies to antimicrobial and local anesthetic agents, was reviewed in order to determine whether pulp regenerative therapy was indicated. All findings and the risks and benefits of pulp regenerative therapy compared with conventional artificial root canal filling were explained, and the patient opted for the former. After obtaining informed consent from the patient, the presence of any serious diseases and infectious diseases was further assessed by blood and urine tests and serologic tests for common infectious agents including bacteria, fungi, mycoplasma, and viruses (hepatitis B surface antigen, hepatitis C antibody, human immunodeficiency virus antigen, antibody, and human T-cell leukemia virus type 1).

Preparation for Autologous DPSC Transplantation

The patient's maxillary right third molar was deemed to be nonfunctional without opposing dentition, and soon after extraction the tooth was transported to the cell processing center facility (Aeras Bio Inc, Kobe, Japan) at 4°C. Enzymatically isolated and cultured DPSCs in a stable hypoxic condition formed colonies in 3 days and were further expanded to be cryopreserved at the 4th passage of culture as previously reported[14]. The quality and safety inspections confirmed that the patient's DPSCs passed our standards of quality control—positive for stem cell marker expression of CD29 and CD105 (99.8% and 99.3%, respectively) and negative for CD31 (0.21%), viability of 98.6%, doubling time of 27.6 hours, the absence of pathogenic microorganisms (bacteria [aerobe and anaerobe], fungus, and mycoplasmas), and endotoxin <1.0 endotoxin unit/mL.

Root Canal Treatment

The inlay and all the caries were removed (Fig. 1*Ac* and *Ad*), and a bulkhead was formed (Fig. 1*Ae*). Access to the root canal system was performed using a surgical operating microscope (Flexion; CJ-Optik GmbH & Co KG, Aßlar, Hessen, Germany). The gutta-percha and sealer were removed using the Peeso Reamer #3 (Dentsply Maillefer, Tulsa, OK), an ultrasound diamond chip (EMS Japan KK, Tokyo, Japan), and further with gutta-percha remover (YDM Corp, Tokyo, Japan) and K-files #30–#60 (MANI, Tokyo, Japan) without using any solvent. The K-file #10 (MANI, Tokyo, Japan) was used to determine the canal length using an electronic apex locator (Root ZX; Morita, Tokyo, Japan) and establish patency. The buccal and palatal canals were joined together to be 1 canal up to the apex (Fig. 1*Af*). The canal was shaped to the apex, which is defined as Root ZX digital display 0.5 on a digital scale, with K-files from #45–#70 (MANI, Tokyo, Japan). Irrigation was performed with 6% sodium hypochlorite (NaOCl) for less than 2 minutes and 3% EDTA (SmearClean; Nippon Shika Yakuhin Co Ltd, Simonoseki, Japan). The canals were further irrigated with nanobubbles (Aeras Bio Inc) for 2 minutes followed by nanobubbles with 0.015% levofloxacin (1.5% Cravit Ophthalmic Solution; Santen Pharmaceutical Co, Ltd, Osaka, Japan). Then, the canals were dried, and paper points moistened with nanobubbles containing 0.015% levofloxacin were placed into the canals as an intracanal medicament. The cavity was temporarily filled with water-setting hydraulic cement (Caviton; GC, Tokyo, Japan) and composite resin (Clearfil DC Core Automix ONE; Kuraray Noritake, Tokyo, Japan) with an adhesive procedure (Clearfil Mega Bond 2; Kuraray Noritake, Tokyo, Japan).

At the second treatment, 2 weeks later, the patient returned free of symptoms, and the intracanal paper points did show hemorrhagic exudate. To ascertain the presence of aseptic conditions in the canal, bacterial anaerobic culture and polymerase chain reaction (PCR) analyses were performed before irrigation and intracanal medication with the same procedures as the previous time except no EDTA irrigation. The sterilized paper point was inserted for 1 minute into the canal filled with nanobubbles and was cultured in PLADIA liquid anaerobic culture media (Showa Yakuhin Kako Co, Ltd, Tokyo, Japan) for 5 days under an anaerobic condition (Fig. 1*Cc*). Furthermore, the PCR samples were harvested from the root canal 2 times. In brief, after keeping nanobubbles in the canal for 2 minutes, a paper point was inserted for 2 minutes to be absorbed with microbiome and was suspended in a liquid culture medium. This process was repeated. The 2 DNA samples were isolated from each microbial suspension according to the manufacturer's protocol (Takara Bio Inc, Kusatsu, Japan). The PCR assay for bacteria was performed using universal primers (Sequencing Primer 10F and Sequencing Primer 800R; Takara Bio Inc, Kusatsu, Japan) to amplify a specific region (approximately 0.8 kilobases [kb]) within bacterial 16S ribosomal DNA (rDNA) (Bacterial 16S rDNA PCR Kit Fast [800], Takara Bio Inc). The PCR assay for fungi was performed using universal primers (Sequencing Primer ITS1 F

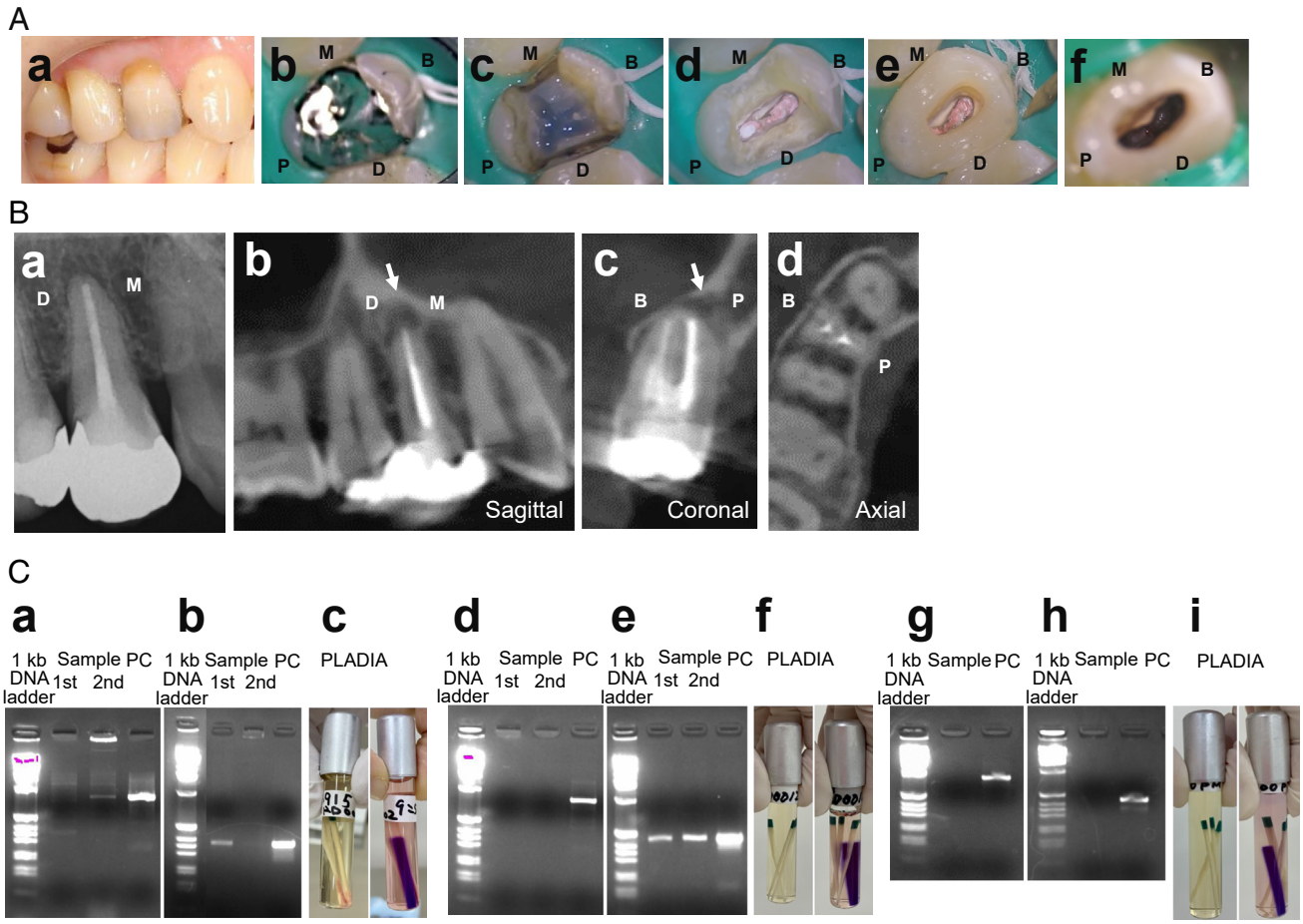

FIGURE 1 – The root canal treatment of the maxillary right first molar. *(Aa)* An image of the preoperative buccal view with discoloration, (*Ab*) mirror images of occlusal views of the preoperation with metal inlay restoration, (*Ac*) after removal of the inlay, (*Ad*) after removal of caries, (*Ae*) after forming the bulkhead, and (*Af*) after access cavity preparation. (*Ba*) A preoperative radiograph showing caries under the inlay restoration; CBCT images showing an extensive periapical lesion (*arrow*) in (*Bb*) the sagittal view and (*Bc*) the coronal view and (*Bd*) 2 orifices of the root canals in the axial view. Bacteriologic analyses for the examination of the aseptic condition in the canals (*Ca–Cc*) at the second root canal treatment, (*Cd–Cf*) at the third treatment, and (*Cg–Ci*) at the fourth treatment. PCR assays for (*Ca*, *Cd*, and *Cg*) bacteria and (*Cb*, *Ce*, and *Ch*) fungi and (*Cc*, *Cf*, and *Ci*) anaerobic bacterial culture after 5 days using PLADIA including resazurin assay. B, buccal; D, distal; M, medial; P, palatal; PC, positive control.

and Sequencing Primer ITS1 R; Takara Bio Inc, Kusatsu, Japan) to amplify a specific region (approximately 0.45 kb) within rDNA ITS1 region (Fungal rDNA [ITS1] PCR Kit Fast, Takara Bio Inc). The PCR assays showed positive results both for bacteria and fungi, although bacterial culture was negative (Fig. 1*Ca–Cc*).

At the third treatment, 2 weeks later, after harvesting microbiomes, irrigation and intracanal medication were further performed. The intracanal paper points did show a little exudate. Additional samples were obtained from the canal per the same protocols in the previous treatment. These samples were negative for bacteria but were positive for fungi (Fig. 1*Cd* and *Ce*). After harvesting microbiomes, the canals were further treated by nanobubbles containing 0.015% levofloxacin with the addition of 2.5 μg/mL amphotericin B (Fungizone; Clinigen K.K., Tokyo, Japan) as irrigant and intracanal medicament.

At the fourth treatment, 1 week later, because both the bacterial culture and PCR assays for bacteria and fungi showed negative results in the canal (Fig. 1*Cg–Ci*), informed consent to receive pulp regenerative therapy was provided by the patient again.

Autologous DPSC Transplantation

One month later, the DPSCs of 2 cryotubes at 1×10^6 cells/each tube was transported under −70°C from the cell processing center facility (Aeras Bio Inc) to the clean bench of the operating room in the clinic. The DPSCs were suspended in 200 μL clinical-grade atelocollagen (1% Koken atelocollagen implant; Koken, Tokyo, Japan) containing 1.5 μg granulocyte colony-stimulating factor (Neutrogin; Chugai Pharmaceutical Co Ltd, Tokyo, Japan). After irrigation with 6% NaOCl and nanobubbles containing 0.015% levofloxacin and further irrigation with 3% EDTA for 3 minutes and thoroughly with saline, the canal was dried well with sterilized paper points. Then, autologous transplantation was performed as previously described[8], and the cavity was sealed with Biodentine (Septodont, Lancaster, PA) and composite resin (Clearfil DC Core Automix) with a bonding agent (Clearfil Mega Bond).

Follow-up

The patient was followed up at 1, 4, 12, 24, 48, and 79 weeks after cell transplantation. There were no adverse events or toxicity related to cell transplantation assessed for clinical evaluations during the 79-week follow-up period and laboratory evaluations after 4 weeks. The pulp sensibility test using an electric pulp tester (VITALITY SCANNER;

Yoshida Dental Trade Distribution Co Ltd, Tokyo, Japan) demonstrated a positive response after 1 week, suggesting functional reinnervation. The response was continued during the follow-up duration of 48 weeks, although the response was very weak at 48 weeks. We could not examine the response to EPT at 79 weeks because of interference of the conduction of electricity by a hard jacket crown cemented with resin. Periapical radiographs showed a gradual decrease of lesion size during the 79 weeks of follow-up (Fig. 2*Aa–Ae*) and restitution of the lamina dura and a slightly widened periodontal ligament after 79 weeks (Fig. 2*Ae*). CBCT imaging showed a gradual decrease in the apical lesion size after 24 weeks, 48 weeks, and 79 weeks of follow-up (Fig. 2*Ba–Bf*), and only a very small periapical radiolucency was observed in the sagittal view (Fig. 2*Be*) and not in the coronal view (Fig. 2*Bf*) after 79 weeks. The mineralized tissue formation was observed in the apical part of the canal with apical closure after 48 weeks and 79 weeks (Fig. 2*Bc–Bf*). The volumes of the apical lesion analyzed by OsiriX medical imaging software were 8.60 mm^3 just after transplantation, 3.30 mm^3 after 24 weeks, 2.20 mm^3 after 48 weeks, and 0.01 mm^3 after 79 weeks, demonstrating a 61.6%, 74.4%, and 99.9% decrease at 24 weeks, 48 weeks, and 79 weeks, respectively, compared with the background (day 0). The volume of the regenerative pulp in the affected tooth was 25.7% reduced from 34.2 mm^3 at day 0 to 25.4 mm^3 after 79 weeks (Fig. 2*Ca* and *Cb*), although the volume of the normal pulp of the maxillary right canine tooth was identical after 79 weeks to that at day 0 (8.65 mm^3) (Fig. 2*Cc* and *Cd*), indicating lateral dentinlike mineralized tissue formation in the affected tooth. Furthermore, MRI was performed using a 1.5 Tesla scanner (SIGNA Explorer; GE Healthcare Japan, Tokyo, Japan) with the HNS Head Coil (GE Healthcare Japan) after 24 weeks with the use of T2 (repetition time = 5305 milliseconds, echo time = 90.432 milliseconds, and echo train length = 16) and short tau inversion recovery (repetition time = 4577 milliseconds, echo time = 87.768 milliseconds, and echo train length = 14), with slice spacing of 0.0977 mm, slice gap of 0.0977 mm, and slice thickness of 1 mm. The high signal intensity of the regenerated tissue in the affected tooth was comparable to that of the normal pulp in the adjacent maxillary right second premolar and canine tooth (Fig. 2*Da* and *Db*). These results suggest complete pulp regeneration in the affected mature tooth with apical periodontitis as previously demonstrated in the mature teeth with irreversible pulpitis [6,8].

DISCUSSION

This case report evaluates the use of human autologous DPSCs for pulp regenerative therapy in a mature tooth with an apical lesion. The recent clinical trial and case reports on cell-based RET for apical periodontitis have been performed using allogeneic MSCs derived from the umbilical cord[11,15] or bone marrow[12] in plasma-derived biomaterials, such as platelet-poor plasma and platelet-rich plasma. Previous animal studies in dogs demonstrated that DPSCs have higher potential for pulp regeneration, angiogenesis, and reinnervation compared with other tissue origin, although other tissue-derived MSCs can be alternative cell sources for pulp/dentin regeneration[16]. In this report, we presented clinical evidence of pulp regeneration and remission of periapical tissue also for apical periodontitis by autologous transplantation of DPSCs.

The mature teeth with irreversible pulpitis were successfully treated by transplantation of autologous DPSCs with atelocollagen and granulocyte colony-stimulating factor[8]. However, unlike treating irreversible pulpitis, apical periodontitis with an apical lesion is more challenging because microbial infection is in an inaccessible location of the root canal system and the apical lesions could be more of an impediment to disinfection. An animal study demonstrated that the presence of residual bacteria is a major drawback to the success of pulp regenerative therapy[17]. According to the American Association of Endodontists protocol of RET[18], the concentration of 1.5% NaOCl is recommended for disinfection of root canal systems followed by the application of an intracanal medication based on calcium hydroxide or triple antibiotic paste. However, several studies have indicated that failed RET may be caused by insufficiency of the disinfection levels recommended by the protocol[19]. Thus, it is proposed that careful reevaluation of the disinfection protocol of RET should be necessary to achieve a proper balance between disinfection of the root canal systems and survival of the resident stem cells and preservation of a variety of growth/differentiation factors in the dentinal wall, leading to a long-term successful outcome. The concentrations of NaOCl (5.25%–6%) demonstrated significantly greater antibacterial efficacy compared with mild concentrations (2%–2.5%) and lower concentrations (1%–1.5%)[20]. Some studies further demonstrated that high concentrations of NaOCl (5.25%–6%) are necessary to eliminate biofilms and completely inhibit the biofilm bacterial growth including *Enterococcus faecalis* and the multispecies type from the oral cavity[20]. The tissue-dissolving ability of NaOCl could be influenced by not only concentration but also duration. It has been reported that the killing of bacteria in the *E. faecalis* biofilms was faster during the first 3 minutes[21], and no further biofilm removal occurred after 5 minutes[22]. We used 6% NaOCl as an irrigant for less than 2 minutes using a 27-G cleaning needle in view of its cytotoxicity and related complications after accidental overflow beyond the root apex. It was followed by irrigation with nanobubbles to further remove the smear layer and biofilms[13]. Moreover, levofloxacin has a broad spectrum to have bactericidal effects on a variety of microorganisms responsible for apical periodontitis. Our recent canine experimental study on pulp regenerative cell therapy in apical periodontitis models demonstrated that true pulp-dentin complexes are regenerated after complete disinfection by nanobubbles with levofloxacin in addition to irrigation with 6% NaOCl and with nanobubbles alone[23]. Thus, in the current report, nanobubbles with levofloxacin were further used as irrigant and intracanal medicament for enhanced antimicrobial efficacy within the root canal systems.

The clinical efficacy of cell-based RET with allogeneic umbilical cord MSCs has been demonstrated in mature teeth with apical lesions by an increase of the positive response to EPT (from 17%–50% of the affected teeth) and cold testing (from 6%–56%) 12 months after transplantation and the remission of apical lesions[11]. Furthermore, the pulp vitality test by laser Doppler flowmetry demonstrated an increase of the perfusion unit percentage from 60.6%–78.1% between baseline and 12 months. Although Brizuela et al[11] could not demonstrate any histologic data, they suggested that these results were indicative of the formation of a vascularized tissue with a normal physiological response, probably regeneration of vital pulplike tissue, reported by histologic examination in allogeneic DPSC transplantation in dogs[24]. In the present case, the absence of any adverse events assessed by clinical evaluations within 79 weeks and any toxic response by laboratory evaluations at 4 weeks and highly reduced periapical radiolucency by CBCT imaging at 79 weeks indicate the safety of DPSC transplantation in mature teeth with apical periodontitis. The positive responses to EPT indicated functional reinnervation. The periapical radiographic and CBCT findings of restitution of the lamina dura and the 99.9% decrease in the lesion volume after 79 weeks indicated remission of the periapical tissue. The decrease of the pulp tissue, 25.7% in volume after 79 weeks, due to lateral dentinlike mineralized tissue

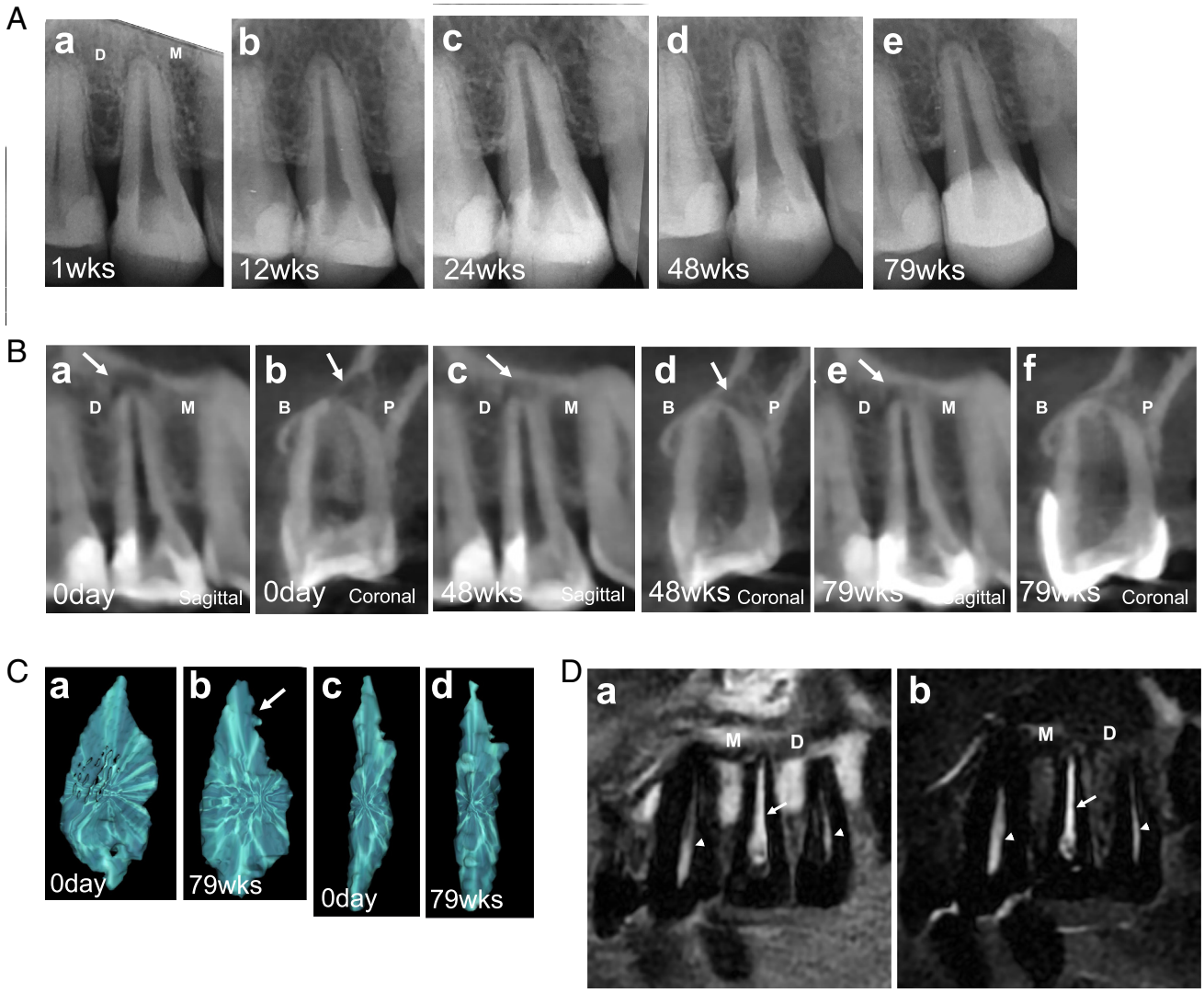

FIGURE 2 – Postoperative evaluation. (*A*) The 79-week follow-up periapical radiograph showing a minimal apical lesion and restitution of the lamina dura after 79 weeks. (*B*) The 79-week follow-up CBCT images showing the gradually reduced size of the apical lesion (*arrows*), apical bone regeneration, and apical remodeling. (*Ba*, *Bc*, and *Be*) The sagittal view. (*Bb*, *Bd*, and *Bf*) The coronal view. (*Ba* and *Bb*) Immediately after cell transplantation (day 0), (*Bc* and *Bd*) after 48 weeks, and (*Be* and *Bf*) after 79 weeks. (*Be*) Note a very small periapical radiolucency in the sagittal view at 79 weeks. (*C*) Analysis of 3-dimensional CBCT images of the root canals using the OsiriX program demonstrates a decrease in low-density areas (*Cb*) after 79 weeks compared with (*Ca*) day 0. (*Ca* and *Cb*) The cell-transplanted tooth (maxillary right first molar) and (*Cc* and *Cd*) the control tooth (maxillary right canine tooth). (*Cb*) Note lateral dentinlike mineralized tissue formation in the apical part of the root canals (*arrow*) after 79 weeks. (*Da*) T2 and (*Db*) short tau inversion recovery magnetic resonance images of cell-transplanted root canal after 24 weeks. Note the comparable signal intensity of the cell-transplanted root canals (*arrows*) with that of healthy normal pulp (*arrowheads*). B, buccal; D, distal; M, medial; P, palatal.

formation in the apical part of the canal indicated that the regenerated tissue maintained pulpal function[5]. The response to EPT was very weak at 48 weeks, which may be associated with mineralized tissue formation with apical closure. The high signal intensity of MRI of the regenerated tissue of the affected tooth at 24 weeks was comparable to that of normal pulp in intact teeth. These EPT, CBCT, and MRI results were comparable to the results found in our previous clinical reports for irreversible pulpitis[6,8], confirming the potential efficacy of pulp regenerative cell therapy.

A meta-analysis of randomized controlled trials demonstrated that there was no difference in the success rate of RET including the absence of symptoms and healing of apical periodontitis compared with conventional treatment in mature permanent teeth with apical periodontitis[2]. Brizuela et al[11] also demonstrated no significant difference in the reduction of lesion dimension between cell-based RET and conventional root canal treatment except anterior-posterior dimension between 6 and 12 months. The probability that an endodontically induced lesion will completely heal (eg within the first 3 years after conventional root canal treatment) ranges between 0.87 (confidence interval, 0.74-1.00) and 0.89 (confidence interval, 0.80-0.98)[25]. Our recent study on pulp regenerative cell therapy for apical periodontitis in dog models demonstrated that the reduced rate of apical lesions from the first root canal treatment to 6 months after cell transplantation (9 months after the first treatment) was associated with the regenerated pulp volume rate[23]. In this case report, the periapical tissue was almost completely healed within 2 years, suggesting that it might be promoted by reinnervation, revascularization, and recovery of the immune

system and homeostasis of the regenerated pulp tissue.

We have already adopted this pulp regenerative cell therapy in clinical practice. As discussed previously in a case report[8], critical challenges in feasibility of this cell therapy included government regulatory issues in regenerative medicine, difficulties in stem cell isolation and expansion, limited availability of good manufacturing practice facilities, and poor cost performance. These challenges have been addressed in present cell therapy. In brief, any dental clinics can execute the therapy after approval of the protocol by a certified committee for regenerative medicine and the Ministry of Health, Labour and Welfare according to the Act on the Safety of Regenerative Medicine. They can use good manufacturing practice compliance facilities owned by cell processors in which the cell processing is cost-effective, safe, and efficient to culture DPSCs with high pulp regeneration potential.

In conclusion, this case report demonstrated regeneration of pulp tissue containing sensory nerves using pulp regenerative therapy with autologous DPSCs in a mature tooth with apical periodontitis. The present technique might be useful in the field of endodontics by expanding the use of DPSCs to include mature teeth with posttreatment apical periodontitis to maintain pulpal function including dentin formation, thus preventing further progression of periapical inflammation and vertical fracture of affected teeth.

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The authors deny any conflicts of interest related to this study.

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