Introduction
The goal of endodontics is to preserve the natural dentition in a state of physiologic health and function.\(^1\) In mature permanent teeth where endodontic therapy is indicated, this is typically accomplished through chemo-mechanical cleaning and shaping, obturation, and restoration.\(^1\) However, these techniques are challenging in the treatment of immature adult teeth with necrotic pulps because of incompletely formed root apices, thin dentin walls, and short crown to root ratios.\(^1\) Implants are also typically contraindicated in this young patient population due to their continued skeletal growth.\(^2\) Therefore, the treatment plan of choice has traditionally been tooth preservation through apexification, which induces a calcified barrier in a necrotic root with an open apex.\(^3\)

In 2004, Banchs and Trope published a treatment alternative which demonstrated promise in addressing the limitations of apexification: regenerative endodontics.\(^4\) Jeeruphan et al. reported regenerative endodontic procedures increase root length and thickness significantly more than either calcium hydroxide [Ca(OH)\(_2\)] or MTA apexification.\(^3\) Regenerative endodontics has a growing body of case reports documenting bony lesion resolution, continued root development, and in limited cases, re-establishment of an immunocompetent and functional pulp tissue.\(^1\) Murray, et al. defined regenerative endodontics as “biologically based procedures designed to replace damaged structures, including dentin and root structures, as well as cells of the pulp-dentin complex.”\(^5\)

This Clinical Update provides clinicians with the current AAE protocol for regenerative endodontic procedures (REPs) and presents the rationale behind recent protocol changes.

Indications/Case Selection for REPs
1. Young patient (6-18 years)\(^1\)
2. Permanent tooth with necrotic pulp & immature apex\(^1\)
3. No/minimal previous canal instrumentation\(^1\)
4. Not previously avulsed\(^6\)
5. No post space needed for restoration\(^6\)

Also, REPs should be referred to an endodontist or an experienced endodontic provider.

2016 AAE Protocol
The treatment regimen for the pulpless immature tooth is based on stem cell research and successful clinical cases. The body of knowledge in this field is steadily growing, and as it does, the treatment protocol is modified. The AAE has the most current recommended protocol: [http://www.aae.org/uploadedfiles/publications_and_research/research/currentregenerativeendodonticconsiderations.pdf](http://www.aae.org/uploadedfiles/publications_and_research/research/currentregenerativeendodonticconsiderations.pdf)

At the time of publication of this Clinical Update, the recommended protocol for REPs is summarized in Figure 1.

### Current Recommended Protocol

<table>
<thead>
<tr>
<th><strong>1st Visit-Disinfection (Phase 1)</strong></th>
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</thead>
<tbody>
<tr>
<td>Anesthesia, access, no instrumentation ↓</td>
</tr>
<tr>
<td>Canal irrigation (20ml 1.5% NaOCl 5 min., then 20ml 17% EDTA 5 min.) ↓</td>
</tr>
<tr>
<td>Paper point dry canal ↓</td>
</tr>
<tr>
<td>Ca(OH)(_2) paste in canal ↓</td>
</tr>
<tr>
<td>Provisional restoration ↓ (1 to 4 weeks)</td>
</tr>
</tbody>
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<table>
<thead>
<tr>
<th><strong>2nd Visit-Regeneration (Phase 2)</strong></th>
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<tbody>
<tr>
<td>Confirm absence of exudate (repeat Phase 1 if necessary) ↓</td>
</tr>
<tr>
<td>Local anesthesia without epinephrine (3% mepivacaine) ↓</td>
</tr>
<tr>
<td>Canal irrigation 20ml 17% EDTA ↓</td>
</tr>
<tr>
<td>Paper point dry canal ↓</td>
</tr>
<tr>
<td>Gently induce bleeding (#25 K-file with an apical bend instrumented 2mm beyond apex) ↓</td>
</tr>
<tr>
<td>Allow clot to form apical to CEJ (optional barrier/Gelfoam®) ↓</td>
</tr>
<tr>
<td>MTA/Biodentine™ over clot ↓</td>
</tr>
<tr>
<td><strong>Definitive restoration (GI, composite)</strong></td>
</tr>
</tbody>
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Figure 1. Regenerative endodontic procedure protocol.\(^7\)

Protocol Changes and Rationale
Research and clinical outcomes have increased understanding and led to changes in the clinical protocol recommendations. **Irrigants.** Previously, sodium hypochlorite (NaOCl) and chlorhexidine were the recommended irrigants for REPs. However, that combination was found to inhibit stem cell adhesion to dentin.\(^8\) Fortunately, the use of sodium hypochlorite and EDTA does not have the inhibitory effect on stem cell adhesion.\(^9\) Regarding stem cell viability, the concentration of sodium hypochlorite has been reduced from 6% to 1.5% to increase stem cell survival.\(^10\) The combination of 1.5% sodium hypochlorite and 17% EDTA improves stem cell viability over other irrigants tested, including sodium hypochlorite and saline as well as the previously advocated chlorhexidine.\(^10,11\) Additional benefits of EDTA include its ability to release growth factors already present within dentinal tubules and to induce stem cell differentiation.\(^12\)

**Intra-canal medicament.** Ca(OH)\(_2\) is the recommended intracanal disinfectant.\(^7\) Treatment with Ca(OH)\(_2\) (Ultracal...
XS™) significantly increases stem cell survival and proliferation compared to controls. An application time of greater than 1 week is required for antibacterial efficacy and less than 4 weeks minimizes weakening of dentinal walls compared to long-term application. Triple antibiotic paste (TAP) and double antibiotic paste (DAP) are still acceptable intracanal medications. However, the concentration of 1000 mg/mL for each was found to be cytotoxic to stem cells. Fortunately, a lower concentration of 1 mg/mL has been reported to be non-toxic to stem cells while retaining their antibacterial efficacy. Regarding tooth staining, TAP at 1 mg/ml was still found to stain teeth, but 1 mg/mL DAP and Ca(OH)₂ had no visible staining. 

The second visit begins with an assessment of pathosis and inflammation. If exudate is observed, the initial visit protocol is repeated, or based on clinical judgement, an alternative treatment performed. If exudate is absent and the tooth is asymptomatic, the REP can begin. The use of a local anesthetic without epinephrine, 3% mepivacaine, is recommended to allow bleeding induction. Bleeding from the apical tissues carries mesenchymal stem cells into the root canal space and the subsequent clot forms a scaffold, an essential element of tissue regeneration.

After blood clot formation, the tooth should be permanently restored with MTA and a glass ionomer, followed by a composite or alloy restoration. Since MTA has the potential to stain teeth, an alternative bioceramic material, such as Biodentine®, is acceptable for teeth in the esthetic zone.

ADA CDT Procedural Codes used for REPs:

**D3355-Pulpal Regeneration-Initial Visit**
- Includes opening tooth, preparation of canal spaces, and placement of medication

**D3356-Pulpal Regeneration-Interim Medication Replacement**

**D3357-Pulpal Regeneration-Treatment Completion**
- Does not include final restoration

**Follow-Up and Outcome Assessment**

The success of REPs is based on the extent to which it achieves its goals. The primary goal is resolution of infection, which is essential for success. A tooth may become asymptomatic after the initial visit and resolution of the apical radiolucency often occurs by six months after treatment. The secondary goal is increased root thickness and length. This can often be observed between six months to one year. This result is highly desirable but not necessarily essential. The tertiary goal is a positive response to pulp vitality testing, which represents the highest degree of success in regenerative endodontics. Follow-up intervals are not mandated, but have been suggested at 3, 6, 12, 18, and 24 months, and annually for five years. All providers and patients with REPs should be aware these teeth can have a radiographic appearance mimicking a tooth with incomplete endodontic therapy even though treatment is complete. Figure 2 represents the radiographic appearance of a successful REP.

**Conclusions**

Regenerative endodontics for the necrotic permanent immature tooth has made significant progress in achieving the goals of resolving apical periodontitis, increasing dentinal wall thickness and root length, and regaining immunocompetency through a revitalization of pulp tissue. Protocols for regenerative endodontic procedures continue to evolve as best treatment practices are being formulated by combining results of stem cell research and documented clinical outcomes. The AAE website, under the Regenerative Endodontics tab, should be referred to for future protocol updates.

**References**


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The views expressed in this article are those of the authors and do not necessarily reflect the official policy or position of the Department of the Navy, Department of Defense, or the U.S. Government.

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