Analysis of Pulpal Reactions to Restorative Procedures, Materials, Pulp Capping, and Future Therapies
Peter E. Murray, L. Jack Windsor, Thomas W. Smyth, Abeer A. Hafez and Charles F. Cox
CROBM 2002 13: 509
DOI: 10.1177/154411130201300607

The online version of this article can be found at:
http://cro.sagepub.com/content/13/6/509
ANALYSIS OF PULPAL REACTIONS TO RESTORATIVE PROCEDURES, MATERIALS, PULP CAPPING, AND FUTURE THERAPIES

Peter E. Murray*
L. Jack Windsor
Department of Oral Biology, Indiana University School of Dentistry, 1121 West Michigan Street, Indianapolis, IN 46202-5186; *corresponding author, petmurra@iupui.edu

Thomas W. Smyth
Department of Dentistry, St. Francis Hospital and Medical Center, Hartford, CT 06105

Abeer A. Hafez
School of Dentistry, University of Southern California, Los Angeles, CA 90089-0641

Charles F. Cox
Department of Restorative Dentistry, School of Dentistry, University of California at Los Angeles, Los Angeles, CA 90095-1668

ABSTRACT: Every year, despite the effectiveness of preventive dentistry and dental health care, 290 million fillings are placed each year in the United States; two-thirds of these involve the replacement of failed restorations. Improvements in the success of restorative treatments may be possible if caries management strategies, selection of restorative materials, and their proper use to avoid post-operative complications were investigated from a biological perspective. Consequently, this review will examine pulp injury and healing reactions to different restorative variables. The application of tissue engineering approaches to restorative dentistry will require the transplantation, replacement, or regeneration of cells, and/or stimulation of mineralized tissue formation. This might solve major dental problems, by remineralizing caries lesions, vaccinating against caries and oral diseases, and restoring injured or replacing lost teeth. However, until these therapies can be introduced clinically, the avoidance of post-operative complications with conventional therapies requires attention to numerous aspects of treatment highlighted in this review.

I) Introduction

The regeneration or replacement of oral tissues affected by inherited disorders, trauma, and neoplastic or infectious diseases is expected to solve many dental problems. Within the next 25 years, unparalleled advances in restorative dentistry are set to take place with the availability of artificial teeth, bone, organs, and oral tissues (Baum and Mooney, 2000; Baum et al. 2002), as well as the ability of growth factors to stimulate dental repair (Murray and Smith, 2002), regenerate lost tissues (Becker, 1994; Smith et al., 2002), produce vaccinations against viruses (Baum and O’Connell, 1995), and genetically alter disease pathogens to help eradicate caries and periodontitis (Hillman et al., 2000). Patient demand for tissue engineering therapy is staggering in both scope and cost. Each year, $400 billion is spent treating Americans suffering some type of tissue loss or end-stage organ failure. This includes 20,000 organ transplants, 500,000 joint replacements, and millions of dental and oral craniofacial procedures, ranging from tooth restorations to major reconstruction of facial soft and mineralized tissues (National Institute of Dental and Craniofacial Research [NIDCR], 2002).

Two-thirds of all restorative dentistry involves the replacement of failed restorations (Maupome and Sheiham, 1998; Burke et al., 1999). In the United States, it is estimated that, of 290 million restorations, 200 million are replacements for failed restorations (Arnst and Carey, 1998). Currently, a high proportion of these teeth develop symptoms requiring endodontic treatment (Zöllner and Gaengler, 2000), and millions of teeth are extracted, with 1.5 million US restorations requiring root canal therapy (NIDCR, 2002). This rate of treatment failure suggests that current restorative regimes are not yet optimized and have a potential for improvement. Often, clinical experience or empirical evidence is used to diagnose dental problems requiring treatment, because scientific evidence is lacking (Mjör, 2001a). Therefore, it is necessary to evaluate the biological changes taking place in teeth to help optimize current treatment regimens, and to use, stimulate, or augment natural regenerative processes to accomplish therapy by tissue engineering. Avoidance of tooth-healing complications requires examination of surgical trauma and injuries caused by a failure to use materials and procedures congruent with the natural regenerative activity of teeth (Murray et al., 2000a). There is limited information regarding sources of pulp injury, healing defects, bacterial leakage, and pulp inflammation, although these factors often create healing problems. Consequently, this review will investigate those aspects of restorative dentistry.
which may be optimized. It will also discuss how the natural regeneration of teeth could serve as the basis for tissue engineering approaches to solve common restorative problems. Tissue engineering is in the early stages of development, and it is uncertain exactly how this technology might be applied to solve many common dental problems. Nevertheless, it is important to formulate tissue-engineering protocols without delay, because the development phase of these new therapies is measured in decades.

(II) Treatment of Caries
A caries lesion is initiated when micro-organisms in dental plaque, commonly mutans streptococci and lactobacilli, ferment carbohydrates (mainly sugars) to produce acid (Featherstone, 1996). Dental caries is a process that brings about a progressive acid-demineralization of the inorganic component of the tooth, accompanied by an enzymatic disintegration of the organic portion. It is primarily a bacterial disease, but has multifactorial etiology (Glossary of Operative Dentistry Terms, 1983). The goals of treatment are to reduce the etiologic microbiota and contributing risk factors to halt the caries decay process and stimulate remineralization. Caries activity can be subdivided according to the rate of tooth demineralization. If demineralization stops, a caries lesion is regarded as “arrested”. Conversely, if the caries lesion is “active”, it can be either slowly or rapidly progressing (Bjørndal and Darvann, 1999). The defense and healing of tooth pulp tissue are more effective in response to slowly progressing or arrested lesions, and the pulp injury can be minimal. In contrast, rapidly progressing lesions can overwhelm pulp-defensive responses and cause severe pulp injuries (Bjørndal and Mjör, 2001).

(A) Optimization of Caries Lesion Treatment
Caries is cited as the most common reason for the need to restore teeth (Deligeorgi et al., 2000). The goals of restorative therapy are to restore teeth to a state of health, function, and aesthetic appearance and to prevent the recurrence of caries (Lutz et al., 1997). However, for the management of caries, it is difficult to achieve the correct balance between an eagerness to remove the lesion and the continued monitoring of lesion progression. The selection of either treatment strategy is relevant to the risk of creating pulp complications, because the selection of approach can mediate the quantity of caries excavation, risk of pulp injury and exposure, size of cavity preparation, and selection of capping materials. Although pediatric dentists continue to apply pit-and-fissure sealants to erupting permanent teeth, this is not done in many general practices. Minimally invasive procedures to treat small caries lesions are proving popular, because this approach may be advantageous to prevent lesion progression (White and Eakle, 2000). For larger penetrating lesions, a step-wise excavation approach may be used (Bjørndal and Thylstrup, 1998). This can prevent extensive pulp injury by leaving firm but stained carious dentin at the cavity floor during excavation. This dentin is often remineralizable (Cate, 2001), and the stepwise excavation of carious tissues can provide pulp protection by stimulating the remineralization of hard carious dentin and reactionary dentin (Leksell et al., 1996). It is recommended to use caries detector dyes to distinguish the unstained remineralizable dentin that should not be excavated (Fusayama and Terachima, 1972), but the use of these dyes may lead to excessive removal of tooth tissues (I.A. Mjör, personal communication). Furthermore, fluoride-containing pulp-capping materials, such as resin-modified glass ionomers, have been observed to remineralize adjacent caries (Donley and Grandgenett, 1998). However, simple sealing of caries lesions with composite resin or resin/amalgam techniques has been shown to arrest lesion progression for over 10 years (Mertz-Fairhurst et al., 1998). These developments in caries treatment planning demonstrate how the natural remineralizing activity of teeth can be used to optimize restorative treatment.

(B) Growth Factor Therapy to Stimulate Tissue Regeneration Beneath Caries
If tissue engineering is to have a significant impact on restorative dentistry, it must primarily focus on providing effective treatments for remineralizing caries lesions and arresting or reversing tooth decay. Toward this aim, increased understanding of the biological processes mediating tissue repair has allowed some investigators to mimic or supplement tooth-reparative responses. Dentin contains many proteins capable of stimulating tissue responses. Growth factors, especially those of the transforming growth factor-beta (TGF-β) family, are important in cellular signaling for odontoblast differentiation and stimulation of dentin matrix secretion. These growth factors are secreted by odontoblasts and deposited within the dentin matrix (Roberts-Clark and Smith, 2000), where they remain protected in an active form through interaction with other components of the dentin matrix (Smith et al., 1998). Carious demineralization of the dental tissues can lead to their release, as can application of cavity-etching agents and restorative materials (Smith et al., 2000). Indeed, it is likely that much of the therapeutic effect of calcium hydroxide may be due to its extraction of growth factors from the dentin matrix (Smith et al., 1995a). Once released, these growth factors may play key roles in signaling many of the events of tertiary dentinogenesis, a response of pulp-dentin repair. The addition of purified dentin protein fractions (Smith et al., 2001), bone morphogenic proteins (Rutherford, 2001), and TGF-β1 (Tziafas et al., 1998) following cavity preparation—all have stimulated an increase in tertiary dentin matrix secretion. This indicates the potential for the addition of growth factors prior to pulp capping, or incorporating them into restorative materials to stimulate dentin and pulp regeneration.

(C) Ameloblast Stem Cell Therapy and Transplantation of Enamel
The outer enamel layer of teeth is more than 95% mineral. During formative stages, the enamel consists of a protein matrix that forms the framework for mineral deposition (Diekwisch et al., 1995). Amelogenin secreted by ameloblasts accounts for 90% of the enamel matrix mineral (Zeichner-David et al., 1995). Other matrix proteins, including tuftelin and ameloblastin, have recently been identified and cloned (Takahashi et al., 2001). Isolating ameloblast progenitor cells and replicating the natural enamel-forming process in three-dimensional culture by the use of growth factors to stimulate enamel secretion offer the potential for transplantation. Transplants of ameloblasts and artificial enamel to the outer surfaces of teeth could replace the porcelain veneers and dental materials currently applied to restore tooth structure following caries, disease, erosion, and accidental trauma, or for esthetic reasons (Kihn and Barnes, 1998). The advantage of
applying artificial enamel and ameloblasts to teeth is that this procedure provides a natural regenerative ability to respond to subsequent erosion, attrition, abrasion, or traumatic losses of tooth mineral.

(D) GENE THERAPY TO VACCINATE AGAINST CARIES

The eradication of dental caries or periodontal disease may be successful if gene transfer therapy can mediate humoral and cellular immune responses to the pathogenic bacteria involved in these disease processes (Slavkin, 1996). This type of gene transfer therapy is called DNA vaccination, because DNA-containing antigens which can mediate an immune response are delivered in a plasmid to the target bacteria (Gurunathan et al., 2000a). Caries vaccine strategies may use the mucosal immune system in newborn infants, which is functional prior to the appearance of their first teeth, as an effective way to induce immunity against the colonization of teeth by mutans streptococci and protection against subsequent dental caries (Michalek et al., 2001). Currently, this approach is years away from clinical trials, because of the difficulty of vaccination against intracellular organisms requiring cell-mediated immunity (Gurunathan et al., 2000b), and the possibility of inducing tissue cross-reactivity (Bleiweis et al., 1992). A promising alternative approach to the eradication of caries may be to replace mutans streptococci with genetically altered strains, which have little or no caries-causing potential (Hillman et al., 2000). The eradication of caries and bacterial disease could reduce the demand for restorative treatments by two-thirds, which is approximately the annual proportion of restorative treatments devoted to the treatment of caries (American Dental Association; in Arnt and Carey, 1998).

(III) Guidelines for Optimizing the Success of Cavity Preparations

Non-invasive approaches to the sealing of early caries lesions with penetrating adhesive resins to arrest tooth demineralization and decay are under development (Robinson et al., 2001). If these techniques become introduced clinically, the avoidance of operative intervention should greatly prevent pulp injury. Furthermore, bacterial leakage and secondary caries may also be avoided. In the meantime, it is important to investigate pulp reactions to existing treatments. A review of surveys found that secondary caries and/or discoloration was the most frequent reason for replacement of restorations, followed by mechanical failures and various reasons such as bacterial leakage and inflammation associated with healing complications such as hypersensitivity (Deliegoiri et al., 2001). Age, diet, and oral hygiene characteristics of patients play a considerable role in restoration longevity. Several surveys have also showed how the placement of one type of restorative material in preference can make the critical difference between success and failure within a few years (Mjör et al., 1990; Maupome and Sheiham, 1998; Burke et al., 1999).

This demonstrates the different effects of restorative materials on restoration longevity, and the importance of examining the effects of cavity preparation and capping materials on pulp injury and regeneration.

(A) IMPORTANCE OF PRESERVING PULP VITALITY

The preservation of pulp vitality following restorative intervention is dependent on the degree to which the pulpal cell populations can survive, as well as the ability of these cells to detect and respond to injury to initiate an appropriate repair response (Murray et al., 2001). The most visible repair response to pulp injury is the deposition of a tertiary dentin matrix. Unlike primary or secondary dentin that forms along the entire pulp-dentin border, tertiary dentin is focally secreted by odontoblasts in response to primary and secondary dentin injury. The process of tertiary dentin secretion can be classified as being reactionary or reparative in origin, depending on the severity of the initiating response and the conditions under which the newly deposited dentin matrix was formed. In general, reactionary dentin is secreted by pre-existing odontoblasts, and reparative dentin is secreted by newly differentiated odontoblastoid cells (Smith et al., 1995a). The secretion of reactionary dentin is the main post-operative odontoblast repair response to the presence of a cavity carefully cut into the dentin of a tooth (Fig. 1a). Reparative dentin, in contrast, is secreted by a second generation of odontoblastoid cells when irreversible odontoblast injury has destroyed these primary cells (Fig. 1b). Reparative dentinogenesis is a much more complex process than reactionary dentinogenesis, and is generally observed following injurious cavity preparation or a pulp exposure situation (Mjör, 1985). Minimizing pulp injury during cavity prepara-
TABLE 1
Sequence of Cavity Preparation and Restoration Variables* Correlated to Pulp Injury

<table>
<thead>
<tr>
<th>Variable Correlations with Odontoblast Numbers as a Measure of Pulp Injury</th>
<th>Analysis of Variance (P value)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cavity remaining dentin thickness</td>
<td>0.0001</td>
</tr>
<tr>
<td>Cavity wall depth</td>
<td>0.0001</td>
</tr>
<tr>
<td>Cavity wall area</td>
<td>0.0001</td>
</tr>
<tr>
<td>Pulpal inflammation related to RDT</td>
<td>0.0001</td>
</tr>
<tr>
<td>Type of restoration material</td>
<td>0.0001</td>
</tr>
<tr>
<td>Total cavity surface area</td>
<td>0.0013</td>
</tr>
<tr>
<td>Etching (cavity conditioning)</td>
<td>0.0044</td>
</tr>
<tr>
<td>Cavity volume</td>
<td>0.0069</td>
</tr>
<tr>
<td>Reactionary dentin area</td>
<td>0.0172</td>
</tr>
<tr>
<td>Sex of patient</td>
<td>0.2063</td>
</tr>
<tr>
<td>Cavity floor width</td>
<td>0.2211</td>
</tr>
<tr>
<td>Patient age</td>
<td>0.2226</td>
</tr>
<tr>
<td>Bacteria within restorations</td>
<td>0.3892</td>
</tr>
<tr>
<td>Cavity floor area</td>
<td>0.6794</td>
</tr>
<tr>
<td>Bacteria in cut dentinal tubules</td>
<td>0.5677</td>
</tr>
<tr>
<td>Time elapsed since operative procedures</td>
<td>0.7557</td>
</tr>
</tbody>
</table>

* Among the variables most important in the creation of pulp injury were the cavity remaining dentin thickness and type of restoration material (About et al., 2001). Reprinted with permission from the Journal of Dentistry.

Pulp repair can be negatively influenced by the absence of coolant during cavity cutting, and the bur speed (Swerdlow and Stanley, 1958; Langeland and Langeland, 1968) in addition to cavity RDT, cavity wall depth, area, volume, inflammation related to RDT, type of restorative material, total cavity surface area, cavity conditioning treatment (Table 1), and the presence of bacteria (Cox et al., 1992) are important considerations. Although other variables were found to be less important (Table 1), this finding is not to say that they lack effect in terms of pulp injury or healing. All aspects of restorative treatment require careful consideration, and the sequence of variables presented in Table 1 indicates aspects of treatment which are most deserving of attention in an attempt to minimize injury and optimize pulp dentin regeneration.

(C) Pulp reactions to cavity’s remaining dentin thickness

Erosion, trauma, caries, and diseases can severely injure teeth; however, it has been frequently observed that greater injuries are sustained following cavity preparation and restoration with dental materials (Cox et al., 1992; Stanley, 1992; Kim and Trowbridge, 1998). While the dimensions of cavity preparations are under the control of the clinician, these are commonly determined by the extent and progression of disease, such as caries, as well as by the cavity preparation form necessary to retain the filling. Consequently, it is important to understand pulp reactions to a range of cavity preparation dimensions, particularly the cavity’s remaining dentin thickness (RDT). Odontoblast survival and reactionary dentin secretion were the two responses most sensitive to cavity RDT (Murray et al., 2002b). The relationship between these variables is shown in Fig. 2. To a lesser degree, pulp inflammation and cavity wall depth were influenced by cavity RDT, whereas patient variables and restorative factors had little effect (Murray et al., 2002b). The effects of RDT on odontoblast survival and reactionary dentin repair activity can be attributed to an increasing degree of cellular injury, due to reductions in the protective properties of dentin. Dentin protects pulp cells from potential sources of injury (Stanley et al., 1975). Deep cavities with small RDTs leave the pulp tissue less protected from preparation trauma, and also from the chemical activity of dental materials (Lee et al., 1992). The ability to understand and predict pulpal reactions to cavity preparation allows treatment decisions to be made which exploit the natural repair responses of the tooth. Deviations from the expected pulp responses to restorative treatment can be used to identify potential post-operative complications. Practitioners need to have the option to re-attempt restorative treatment or develop a new treatment plan before post-operative complications can progress in severity and become irreversible.

(D) Repair of fillings rather than total replacement

An excellent method to conserve RDT is to repair faulty restorations, because this is less invasive than having to replace fillings completely. However, most dental schools are reluctant to teach students how to repair faulty restorations for
fear that they would be regarded as second-class restorations or that severe, extensive decay will be overlooked. This often leads to the removal and replacement of functional restorations simply to repair a marginal defect affecting perhaps only 10% of the restoration. Part of the problem involves the philosophy of seeking ideal treatment. Another part of the problem is the clinician's inability to visualize gingival or subgingival marginal areas. However, it is possible to illuminate and visualize these areas under magnification on a video monitor, using the same fiber-optic devices that are used in periodontics and endodontics. Together with staining with caries-detector solutions, this technology allows sufficient image quality for the removal of carious tooth structure, scaling of the caries-affected dentin, and repair of the faulty filling according to the principles of minimally invasive dentistry (D.H. Pashley, personal communication).

(E) Restorative materials for non-exposed cavity preparations
Patient confidence is promoted by the placement of long-lasting restorations not subject to recurrent caries, pulp inflammation, allergic sensitization, or symptoms requiring endodontic treatment. Recognition over the last few years that the largest proportion of restoration failures arises from secondary (recurrent) caries or complications (Deligeorgi et al., 2000, 2001) has led to technological advancements in dental materials, in an attempt to reduce their leakage characteristics and improve their longevity (Kugel and Ferrari, 2000). Operator handling (Ciucchi et al., 1997; Finger and Balkenhol, 1999) and surgical skill remain important for influencing treatment success, most likely due to the need for restorations to be finished with a very high technical quality (Murray et al., 2001). It is a pity that many student dentists do not have laboratory exercises where they can perfect their bonding technique by measuring bond strengths to teeth bonded in vitro, before they begin using them clinically (D.H. Pashley, personal communication). However, several surveys have showed how the selection and placement of one type of restorative material in preference over another can make the critical difference between the failure and the success of treatment within a few years (Maryniuk and Kaplan, 1986; Burke et al., 1999).

(F) Pulp inflammation and bacterial leakage associated with restorative materials
The immune system triggers inflammatory reactions to limit tissue damage from invading or foreign molecules (Jontell et al., 1998). These inflammatory reactions can injure the pulpal cell populations and lead to pulp complications in response to cavity restorations that may initially appear to be successful (About et al., 2001). Severe forms of inflammatory activity can develop into total pulpal necrosis and periapical lesion development with local bone destruction (Bergenholtz, 1982). In less severe cases, the inflamed pulp is associated with hypersensitivity, so that thermal, mechanical, or osmotic stimuli encountered in normal function can cause intense pain (Ngassapa, 1996). Inflammation has been shown to produce a down-regulation of normal sodium channels in nerves (Waxman et al., 1999). These observations explain why immunological inflammatory activity is associated with the high rates of primarily vital teeth exhibiting pulpal complications following cavity restoration (Zöllner and Gaengler, 2000). Cavity preparation trauma as well as the chemical activity of restorative materials can stimulate the release of inflammatory mediators from sensory nerve fibers (Langeland et al., 1966; Okiji et al., 1997). However, most restorative materials appear to mediate low levels of pulp inflammation in teeth with highly permeable dentin, such as those that are newly erupted, except in the presence of bacterial leakage (Cox et al., 1987; Bergenholtz, 2000; Murray et al., 2002c). The selection of restorative materials has an important influence on bacterial leakage. Zinc oxide eugenol and resin-modified glass ionomer can prevent bacterial growth in 100% of cavity restorations for up to one year following treatment (Table 2). These observations can be attributed to the antibacterial activity of these agents and the direct sealing of cavity walls (Bergenholtz et al., 1982; Tarim et al., 1998). The placement of enamel-bonded resin composite and adhesive-bonded resin composite does not seem to result in a perfect seal with cavity walls, because bacteria were detected in 24 and 11%, respectively, of these restorations (Table 2). The favorable sealing characteristics of resin-modified glass ionomers explain why these materials are recommended for...
TABLE 2
Pulpal Inflammatory Activity and the Presence of Bacteria at the Tooth-Restoration Interface

<table>
<thead>
<tr>
<th>Restorative Material</th>
<th>Pulpal Inflammation (Percentage of Teeth)</th>
<th>Bacterial Leakage (Percentage of Teeth)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Absent/Slight</td>
<td>Moderate</td>
</tr>
<tr>
<td>Composite resin bonded to dentin</td>
<td>32</td>
<td>59</td>
</tr>
<tr>
<td>Composite resin bonded to enamel</td>
<td>38</td>
<td>56</td>
</tr>
<tr>
<td>Resin-modified glass ionomer</td>
<td>51</td>
<td>49</td>
</tr>
<tr>
<td>Zinc oxide eugenol</td>
<td>91</td>
<td>9</td>
</tr>
<tr>
<td>Calcium hydroxide</td>
<td>83</td>
<td>17</td>
</tr>
</tbody>
</table>

* These restorations were placed in the caries-free teeth of young adult patients. Severe inflammation was avoided by the use of materials such as resin-modified glass ionomer and zinc oxide eugenol that prevented bacterial growth (Murray et al., 2001). © 2001 American Dental Association. Reprinted by permission of ADA Publishing, a Division of ADA Business Enterprises, Inc.

Class V cavities in caries-prone patients (Bergenholtz et al., 1982). The detection of bacteria beneath composite resin restorations demonstrates the continued need for improvement in the adherence and marginal sealing ability of these materials, by the use of sandwich placement techniques to reduce composite resin shrinkage during polymerization (Gallo et al., 2000). The greater prevalence of bacteria in cavities following use of enamel-bonded resin composite suggests that inadequate or incomplete bonding to dentin may result in increased leakage of bacteria at the tooth/restoration interface. It would seem that restorative materials that form the most perfect sealing with tooth structure are most able to prevent bacterial microleakage. The prevention of bacterial microleakage will limit the severity of pulp inflammation and help maintain pulp tissue vitality.

(IV) Guidelines for Optimizing the Success of Pulp-exposed Cavity Preparations

The difference between the longevity and frequency of complications associated with exposed and non-exposed pulp restorations means that it is worthwhile to make every effort to avoid creating pulp exposure. The success of pulp capping depends on many factors (Mjör, 2002a,b), but it is reported to be 37% after 5 years and 13% after 10 years (Barthel et al., 2000), while the success of non-exposed cavity preparation/restorations is generally much higher and varies with different types of materials and conditions (Maupome and Sheiham, 1998; Burke et al., 1999). Nevertheless, there is an 86% success rate for equivalent teeth over 10 years for teeth that did not have a pulp exposure prior to restoration (Mertz-Fairhurst et al., 1998).

Pulp exposures are more problematic to restore than non-exposed pulp cavity preparations for several reasons. First, during surgery, it is necessary to control hemorrhage that contaminates the dentin surface and prevent blood clot formation, while identifying and removing diseased or infected tissue (Haefez et al., 2000). Second, mechanical or traumatic injury compromises the pulp-healing response (Mjör, 2001b). Third, the presence of dentin restricts the diffusion of potentially injurious agents in deep dentin; only 50% of the dentin surface is composed of tubules, while the other half is made up of solid buffer. The loss of dentin beneath the restorative material causes the pulp tissue to be more sensitive to the possible cytotoxic and irritational action of the capping agent (Murray et al., 2000e). Fourth, operative debris including dentin fragments, sometimes called “chipitis” (Stanley, 1998), and particles of capping materials may infiltrate the pulp, causing injury and inflammatory reactions (Walton and Langeland, 1978; Harsted et al., 1981). The failure to form tertiary dentin may leave pulp tissues vulnerable to subsequent injury, or the opposite effect: The excessive formation of tertiary dentin can be associated with a closure of the pulp chamber and root canals (Stanley, 1989), to make future endodontic treatment difficult or impossible.

(A) Pulp-capping therapy

Ultimately, after the pulp is exposed, the clinician must decide whether to place a pulp-capping material or to undertake pulpotomy. Most dentists will immediately opt for a pulpotomy (Ranly and Garcia-Godoy, 2000). The most important factor in this decision-making process is the likelihood of success. Factors which contribute to the likelihood of pulp-capping success include a young patient age, open tooth apex, good patient health, lack of pre-existing symptoms, good pulp response to stimuli, small size of exposure, and minor pulpal hemorrhage (Christensen, 1998; Görecka et al., 2000; Haefez et al., 2000). Tissue engineering may be able to revitalize pulp tissues by adding new stem cells or by transplanting new vital tissue. Until such approaches are available, however, it is important to use direct pulp-capping materials and placement techniques which will benefit pulp healing and preserve pulp vitality.

(B) Odontoblast Replacement by Odontoblastoid Cells

Following pulp exposure, all primary odontoblasts are irreversibly injured at the exposure site (Murray et al., 2001). These post-mitotic, terminally differentiated cells cannot proliferate to replace subjacent irreversibly injured odontoblasts. Consequently, these primary cells must be replaced by a new generation of odontoblastoid cells (Tziafas, 1994, 1995). The source of these cells has proved to be a source of much speculation. Autoradiographic studies have indicated that new odontoblastoids proliferate from within other pulp cell populations by a process of differentiation and migrate toward the site of pulp exposure, where reparative dentin is secreted (Fitzgerald et al., 1990). Possible progenitor cell populations for the new odontoblastoid cells were assumed to be the subjacent cells of the sub-odontoblast layer or pulp fibroblasts (Fitzgerald, 1979; Fitzgerald et al., 1990). However, cultures of pulp fibroblasts did not appear to differentiate into cells with odontoblast-like phenotypes or secretory characteristics (Hanks et al., 1998; MacDougall et al., 1998). However, recent histological investigations have observed the presence of periocyte and myofibroblast transitional cells in pulp tissue (Carlile et al., 2000; Alliot-Licht et al., 2001). These cells appear to migrate to the site of pulp exposure and secrete reparative
dentin (Murray et al., 2002e). Isolation of these cells in culture has produced human dentin secretion (About et al., 2000; Alliot-Licht et al., 2001). These specialized fibroblast-phenotype pulp cells have the ability to migrate in response to cytokine signaling molecules (Ellis et al., 1997). The similar phenotype between normal fibroblasts and odontoblastoid progenitor cells probably explains why the identification and activity of these cells have proved difficult to isolate. (A schematic representation of these cells is shown in Fig. 1b, step 3.) While these cells appear to lack some characteristics of true stem cells, they initially appear to be pluripotent mesenchymal cells with the potential to express both soft- and hard-tissue phenotypes, initially appear to be pluripotent mesenchymal cells with the potential to express both soft- and hard-tissue phenotypes, potentially giving rise to a variety of cell types, including fibroblasts, chondroblasts, and osteoblasts. Consequently, these cells offer exciting opportunities for regenerating decayed dentin for which there are few conservative treatment possibilities because of the limitations of current treatment options.

(C) DENTIN BRIDGE FORMATION

Dentin bridges are a type of tertiary dentin secreted by odontoblastoid cells at the site of pulp exposure (Fig. 1b, step 5). The secretion of dentin bridges can be influenced by pulp-capping materials, degree of mechanical injury, and the creation of dentin debris during operative procedures (Murray et al., 2002e). Inflammation and bacterial leakage also negatively influence dentin bridge formation (Cox et al., 1987). A practitioner’s attention to minimizing operative debris and carefully placing pulp-capping materials is postulated to be most beneficial for dentin bridge formation (Cox and Suzuki, 1994). Growth factor therapy is already used to promote new bone formation to correct periodontal defects (Heijl et al., 1997). This same approach should also be applied to stimulate healing of pulp tissues if pulp-reparative activity is compromised—for example, by injury or the age of the patient.

(D) PULP-CAPPING MATERIALS

The selection of direct pulp-capping materials and treatments has changed little in the past 30 years while, over the same time, the introduction of new composite resin materials has provided various options for the restoration of indirect cavity preparations (Cox and Suzuki, 1994). Calcium hydroxide is the most commonly used pulp-capping agent (Yoshida et al., 1994). Its use is largely based on histological evidence, such as: inducing healing of periapical lesions, promoting apical closure in incompletely developed teeth, and preventing or arresting root resorption (Mjör and Ferrari, 2002). The high pH of calcium hydroxide provides bactericidal activity and encourages tissue repair by promoting tertiary dentin secretion (Yoshida et al., 1994; Foreman and Barnes, 1990). However, calcium hydroxide suffers from unstable physical properties that allow material particles to migrate into pulp tissue, thus causing inflammation, which may develop into necrosis (Walton and Langeland, 1978). The first generations of calcium-hydroxide-containing materials had improved physical properties but were still not insoluble, and they eventually permitted bacterial leakage to occur (Cox and Suzuki, 1994). Calcium hydroxide may be considered to be a comparatively primitive material by current standards. The adhesive systems used to place composite resin materials appear to have more technological development potential. Future areas in which these adhesive systems could be developed include using bioactive molecules to mediate pulp repair (Tzifas et al., 2000), adding antibacterial activity (Imazato et al., 2001), including desensitizer, improving sealing and bonds strengths (Costa et al., 2000), as well as active caries-prevention activity (Christensen, 2000). Moreover, it must be recognized that the placement protocols for adhesive systems are not fully optimized, and improvements are needed to increase the ease and speed of placement, and to avoid problems. This is because globules of resin can migrate into pulp tissue and stimulate inflammation (Kitasako et al., 1999). In addition, polymerization-shrinkage during the placement of these materials can create marginal gaps to permit bacterial leakage to occur (Fashley, 1996). Knowing the benefits and problems associated with these restorative materials is helpful in assessment of the risks and types of post-operative complications.

(E) PULP INFLAMMATION FOLLOWING DIRECT PULP CAPPING

A comparison between pulp capping with resin composite and that with calcium hydroxide showed that composite resin was associated with a lower frequency of bacterial leakage (19.7% vs. 47.0%) (Murray et al., 2002e). These findings agree with reports suggesting that new composite resin products are superior to their predecessors in having improved sealing properties. The leakage of bacteria was highly correlated with pulp inflammation in the teeth of young adults (Murray et al., 2002e). Both composite resin and calcium hydroxide pulp capping appeared to have similar effects on pulp inflammation in the presence and absence of bacteria. Consequently, the selection of materials with the ability to seal the exposed pulp and prevent bacterial leakage is perceived as the most important factor in avoiding and minimizing pulp inflammation.

(F) TUNNEL DEFECTS

The pulp tissue can be protected following exposure by the deposition of tertiary dentin, referred to as dentin bridge formation (Cox et al., 1987, 1996). To act as an effective barrier against bacterial leakage and migration of particles from capping materials, the reparative dentin should not contain any tunnel defects (Cox et al., 1996). A tunnel defect is a discontinuity in the structure of reparative dentin, which allows for passage between the exposure site and pulp tissue (Cox et al., 1996). Recently, it was observed that most tunnel defects are associated with operative debris or particles of capping materials (Murray et al., 2002e). The presence of operative debris appears to interfere with the continuity of dentin bridging. Consequently, for more complete bridging of pulp exposures, slow bur speeds should be used together with the cleansing of operative debris from the site of exposure.

(V) Impact of Tissue Engineering on Restorative Dentistry

(A) THE PROBLEM OF NON-RESTORABLE TEETH

The treatment of caries and periodontal diseases has improved greatly over the last 50 years and has led to reductions in the numbers of teeth extracted (Greenberg, 1992). If teeth require extraction, patients have the option of having artificial tooth implants fitted or wearing dentures. Currently, 45 million Americans wear dentures, and 25% of them are dissatisfied (Lechner and Roessler, 2001). Even
with the implementation of tissue-engineering procedures in restorative dentistry, there will always be some teeth that are too severely damaged to be restorable, and these will still require extraction and replacement. Recent developments in tissue-culturing technology and improvements in controlling oral cell activity have facilitated the growth of teeth in organ culture (Murray et al., 2000c, 2002d) and human dentin to be synthesized and secreted in vitro (About et al., 2000). The next stage in this process will be to grow and harvest artificially grown teeth as a substitute for implants and prosthetics. Once teeth have been extracted. But injury to original tooth-supporting structures from disease and tooth extraction will likely compromise the success of tooth transplantation. However, studies are under way with proteins to improve gingival adhesion to implants (Tamura et al., 1997). It is hoped that this research will also benefit tooth implantation.

### (B) POTENTIAL APPLICATIONS OF TISSUE ENGINEERING TO REPLACE EXISTING THERAPIES

There appears to be no certainty regarding the introduction of any particular aspect of tissue-engineering therapy into restorative dentistry, and there is no way of knowing how successful these therapies will eventually prove to be. This explains a general reluctance of authors to be specific and speculate on how tissue engineering will transform individual aspects of restorative dentistry (Ranly and Garcia-Godoy, 2000; Mjör, 2002a, b). However, in any review of this type, the issue of using therapies in situations where they are likely to prove most beneficial should be addressed. Although restorative dentistry is too highly dependent on clinical opinion to be an exact scientific discipline (Mjör, 2001a), if we examine restorative dentistry by the degree of tooth injury requiring restoration, then some obvious applications of the tissue engineering discussed previously in this review become apparent. Examples of tooth injury with common restorative treatments vs. likely tissue-engineering therapies are shown in Table 3.

### (VI) Summary

Restorative dentistry is an intricate form of microsurgery. When removing caries, it is often difficult for clinicians to know how, when, and where to start, and vice versa for when to stop. Yet there are biological indicators as well as records of longevity for restorations, radiographs, stimuli testing, and patient opinions to be used for predicting the outcomes of different caries-treatment strategies, cavity-cutting methods, and restorative materials. From the biological perspective, each restorative variable has some effect on pulp vitality, injury, and regeneration. Deviations from normal pulp regeneration may be used to diagnose the onset of complications, such as bacterial leakage and pulp inflammation. Minimizing pulp injury during cavity preparation and placing materials which prevent bacterial microleakage will preserve pulp vitality. Furthermore, pulp regeneration will be used as the basis for tissue engineering to radically alter restorative dentistry and the prognosis of restored teeth. To avoid the need for operative dentistry, DNA vaccines may be used to arrest or prevent the development of caries lesions. Restorative materials may contain a “cocktail” of growth factors, delivered in a slow-release vehicle to regenerate replacement dentin from intra-coronal pulp matrix. In cases of partially decayed or fractured teeth, pluripotent cells may be implanted to regenerate tooth structure. Eventually, non-restorable or lost and missing teeth might be replaced by artificial implants of tooth tissues grown synthetically in an in vitro culture.

### Conclusion

Tissue-engineering therapies offer exciting treatment possibilities, but health hazards, technical problems, and high costs will delay their routine introduction for years to come. In the meantime, restorative dentistry will rely on the optimization of conventional therapies. The following guidelines are intended to avoid the need for restorative treatment, avoid the creation of pulp exposures, and optimize restorative outcomes.

### TABLE 3

<table>
<thead>
<tr>
<th>Degree of Tooth Injury</th>
<th>Minimal</th>
<th>Severe</th>
<th>Some Loss of Vital Tissue</th>
<th>Large Loss of Vital Tissue</th>
<th>Complete Loss of Vital Tissue</th>
</tr>
</thead>
<tbody>
<tr>
<td>Example of dental problem</td>
<td>Early non-arrested caries lesion</td>
<td>Arrested deep-penetrating caries lesion</td>
<td>Arrested or slowly progressing caries lesion extending to pulp tissue</td>
<td>Partially decayed or fractured tooth</td>
<td>Decayed tooth, or accidentally evulsed tooth.</td>
</tr>
<tr>
<td>Common current restorative therapy</td>
<td>Seal fissure or excavate caries and apply restorative materials.</td>
<td>Excavate caries and apply restorative materials.</td>
<td>Stepwise excavation of caries lesion, or pulp capping</td>
<td>Pulp capping, endodontic treatment, or tooth extraction</td>
<td>Remove injured tissue and place implant or prosthetic teeth.</td>
</tr>
<tr>
<td>Likely objective of tissue engineering in restorative therapy</td>
<td>Alter oral bacterial DNA to arrest and prevent subsequent enamel and dentin caries demineralization.</td>
<td>Stimulate pulp-dentin healing with growth factors.</td>
<td>Regenerate lost tissues and dentin repair with growth factors.</td>
<td>Implant progenitor cells to regenerate lost tissue and tooth mineralized structure.</td>
<td>Use progenitor cells and growth factors in 3-dimensional tissue culture to harvest artificial teeth for implantation.</td>
</tr>
</tbody>
</table>

(1) Monitor tooth lesions to distinguish between active and arrested caries. Only active caries should require treatment except for esthetic reasons.

(2) Soft caries should be excavated, but sometimes there appears to be no need to remove caries-affected dentin from the cavity floor; although this dentin is often discolored and stains with caries-detector dyes, it is usually remineralizable.

(3) Deep-penetrating caries lesions should be treated by slow stepwise excavation techniques to allow for dentin remineralization, regeneration, and pulp healing.

(4) Prior to replacing fillings, clinicians should consider repair possibilities rather than complete removal.

(5) Dental materials should be selected which reduce bacterial leakage. Prime examples include the use of long-lasting amalgam restorations for filling posterior teeth, resin-modified glass ionomers in non-load-bearing premolar teeth, and adhesive systems which often require less removal of tooth structure for filling retention.

(6) Clinicians not experiencing problems with existing restorative materials should continue with tried and tested approaches. If problems are encountered, aspects of operator handling should be evaluated, because this can influence the effects of materials.

(7) Direct pulp capping must be restricted to teeth with a good prognosis, young patients in good health, lack of pre-existing symptoms, fresh non-carious exposure, good pulp response to stimuli, small size of exposure, and minor pulp hemorrhage; otherwise, pulpotomy or tooth extraction is indicated.

(8) Minimizing the creation of operative debris with low-speed cutting, and cleansing away of all visible debris prior to pulp capping, will reduce pulp injury and permit the increased continuity of tertiary dentin formation.

Acknowledgments

The authors thank Dr. David H. Pasley and Dr. Ivar A. Mjör for their review and personal communications.

References


Diaz-Flores L, Gutierrez R, Gonzalez R, Verela H (1981). Inducible perivascular cells contribute to the neochondrogenesis in graft-


