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REVIEW

Recent advances and perspectives in topical oral anesthesia

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ABSTRACT

Introduction: Topical anesthesia is widely used in dentistry to reduce pain caused by needle insertion and injection of the anesthetic. However, successful anesthesia is not always achieved using the formulations that are currently commercially available. As a result, local anesthesia is still one of the procedures that is most feared by dental patients. Drug delivery systems (DDSs) provide ways of improving the efficacy of topical agents.

Areas covered: An overview of the structure and permeability of oral mucosa is given, followed by a review of DDSs designed for dental topical anesthesia and their related clinical trials. Chemical approaches to enhance permeation and anesthesia efficacy, or to promote superficial anesthesia, include nanostructured carriers (liposomes, cyclodextrins, polymeric nanoparticle systems, solid lipid nanoparticles, and nanostructured lipid carriers) and different pharmaceutical dosage forms (patches, bio- and mucoadhesive systems, and hydrogels). Physical methods include pre-cooling, vibration, iontophoresis, and microneedle arrays.

Expert opinion: The combination of different chemical and physical methods is an attractive option for effective topical anesthesia in oral mucosa. This comprehensive review should provide the readers with the most relevant options currently available to assist pain-free dental anesthesia. The findings should be considered for future clinical trials.

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1. Introduction

The isolation of cocaine and recognition of its local anesthetic (LA) properties in the second half of nineteenth century revolutionized dentistry, because patients could be treated painlessly while awake [1]. Since then, better anesthetic agents and armamentaria have been developed in order to achieve the best quality of local anesthesia.

However, painless treatment does not necessarily mean painless anesthesia. LA solutions must be injected through a needle, causing pain due to needle penetration/placement and solution deposition. Figure 1(b) and (d) illustrate LA injections in the maxillary buccal fold and palate, respectively. The aim of dental topical anesthesia (Figure 1(a) and (c)) is to eliminate the pain associated with dental anesthesia procedures [2].

Dental topical anesthesia formulations are commercially available for oral use as gels, ointments, solutions, and adhesive patches [1]. The most common anesthetic agents used in topical formulations are benzocaine, lidocaine, tetracaine hydrochloride, and the combination of benzocaine, butamben, and tetracaine. The eutectic mixture of lidocaine and prilocaine (EMLA® cream, AstraZeneca), which is designed for skin anesthesia, is also used on oral mucosa [3].

The efficacy of these formulations has been evaluated in many studies, with conflicting results. Most studies are difficult to compare, mainly due to the absence of a placebo or randomization, insufficient application time, and single-blind design [4]. However, double-blind randomized placebo-controlled studies have demonstrated that topical anesthetics reduce the pain induced by needle insertion but are unable to control the pain from anesthetic injection in the palate [5–8]. Even EMLA, which has been used as a positive control in many studies of topical anesthetic efficacy, is not effective in reducing pain during LA injection in the palate [8].

However, recent studies showed improvements in *in vitro* permeation and *in vivo* palatal efficacy of liposomal lidocaine formulations, offering new possibilities for effective oral topical anesthesia [9,10]. In addition, complexation of LAs with cyclodextrins (CDs) has also shown potential for improvement of topical anesthesia, by either providing rapid onset of anesthesia or prolonging its effect [11].

The present comprehensive review describes the state of art of a variety of drug delivery systems (DDSs) designed to enhance permeation and improve dental topical anesthetic efficacy or promote superficial anesthesia. Chemical methods include nanostructured carriers (liposomes, CDs, polymeric

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Article highlights

- Topical anesthesia offers the possibility of pain-free dental anesthesia.
- The greatest challenge in formulation development is to overcome the oral mucosa epithelium barrier, wrongly considered a highly permeable tissue.
- Although there are several studies reporting formulation development and promising *in vitro* performance, only a few formulations have been evaluated in clinical trials.
- All the clinical trials involving chemical methods (DDSs and pharmaceutical dosage forms) described throughout the review are summarized in Table 1. Nine studies are highlighted because they evaluated formulations still under development (not yet commercially available), in Phase I clinical trials. Seven of these tested liposomal formulations and two used polymeric films.
- Clinical trials indicate that liposomal encapsulation and the pre-cooling technique promote improved topical anesthetic efficacy in different oral mucosa sites, including the palatal mucosa, and seem to be auspicious strategies in dental topical anesthesia.
- Further studies are needed to evaluate the effectiveness of combinations of different chemical and physical methods, since this represents the most promising option for overcoming topical anesthesia challenges.

This box summarizes key points contained in the article.

nanoparticle (PN) systems, solid lipid nanoparticles [SLNs], and nanostructured lipid carriers [NLCs]), as well as different pharmaceutical dosage forms (patches, bio- and mucoadhesive systems, and hydrogels). Physical methods include the use of precooling, vibration, iontophoresis, and microneedle arrays. Emphasis is given to the clinical trials that have evaluated these methods in different sites of the oral mucosa. Chemical permeation enhancers will not be discussed, since this topic was previously reviewed by Hassan and colleagues [12].

2. Oral mucosa: structure, barrier properties, permeability, and permeation pathways

Knowledge of the structure, permeability, permeation pathways, and barrier properties of the oral mucosa is essential in the development of a successful oral topical anesthetic formulation. Permeability refers to the ability of the oral mucosa to be penetrated by drugs such as LA, while permeation pathways (or penetration routes) refer to the routes taken by the drugs to permeate the tissue. For detailed information on these topics, the reader is referred to the literature published by Squier and colleagues [13–15].

The oral cavity is covered by a protective lining tissue called the oral mucosa. This is composed of a stratified squamous epithelium and connective tissue (*lamina propria*) consisting of cells, blood vessels, fibers, and nerves (the site of action of LAs) dispersed in an amorphous tissue [15]. There are three different types of oral mucosa, classified according to anatomical position and epithelium structure. The masticatory mucosa has a keratinized epithelium and *lamina propria* attached to the periosteum of the underlying bone (maxillae or alveolar bone) and is found in the hard palate and gingiva (~25% of the oral cavity), where it is constantly subjected to mechanical forces. This tissue, called the mucoperiosteum, provides a firm and inelastic attachment (Figure 2(a)). To ensure resistance to shear forces and abrasion, its *lamina propria* is dense and filled with a compact complex of collagen fibers in the form of large, closely packed bundles. Due to these characteristics, LA injections in this tissue are more likely to be painful [14,15].

Approximately, 60% of the oral mucosa area is composed of the lining mucosa, which covers the soft palate, ventral side of the tongue, buccal region, inner side of the lips, and floor of the mouth. This tissue is composed of nonkeratinized epithelium and *lamina propria*, with the ability to deform during normal movements. Its softness is related to a submucosa region under the *lamina propria*, composed of loose fat cells, glandular tissue, blood vessels, and nerves (Figure 2(b)). This layer is absent in both masticatory and specialized mucosa, which are firmly connected to the periosteum and tongue muscle, respectively [14,15].

The dorsal side of the tongue is covered by a specialized mucosa. This has unique properties due to the presence of

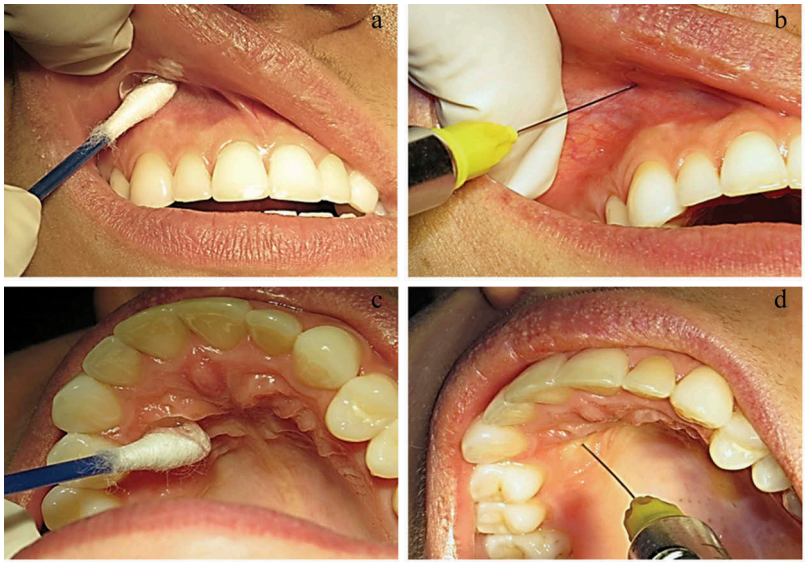

Figure 1. Topical and local anesthesia procedures commonly performed for dental treatment. Maxillary buccal fold topical anesthesia (1a) and injection (1b). Palate fold topical anesthesia (1c) and injection (1d).

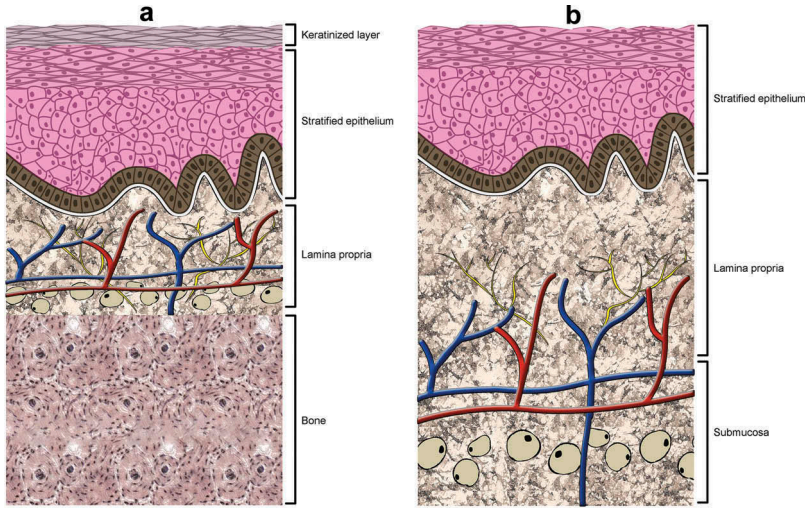

Figure 2. Schematic illustration of the composition of the keratinized (a) and non-keratinized (b) oral mucosa.

both keratinized and nonkeratinized epithelia, papillae, and taste buds responsible for the taste sensations of sweetness, saltiness, sourness, bitterness, and umami [14,15].

The barrier function of the oral mucosa is due to the upper region of the epithelium, comprising an intercellular lipid material released by the membrane-coating granules [16]. An additional barrier is provided by the strong adhesion among the keratinocytes, the main epithelium cell type, provided by desmosomes [15]. Permeation across the oral mucosa can occur according to two possible pathways, considering the epithelium structure: paracellular (or intercellular) and/or transcellular (or intracellular), with the paracellular route being used most frequently [17–19].

Structural variables and differences among the oral mucosa regions (considering thickness, keratinization, and lipid composition) can influence drug delivery [20,21]. Differences in epithelium permeability have been demonstrated, with permeability increasing in the order: hard palate < buccal mucosa < floor of the mouth mucosa [13,22]. As a result, topical anesthesia is less likely to be successful in the hard palate than in any other mucosa region [8,23].

In addition, permeation across the oral mucosa epithelium can be affected by biological factors (age and tissue condition), physicochemical characteristics of the drug (size, partition coefficient, diffusion coefficient, and solubility), and other characteristics such as the final dosage form and application method (considering occlusion ability, viscosity, drug concentration, frequency of application, and mucoadhesive properties) [19,20].

Due to the effectiveness of the oral mucosa barrier against drug penetration, successful transbuccal drug delivery, including topical anesthesia, remains a great challenge [18]. Therefore, efforts have been focused on increasing drug penetration and retention in order to achieve topical anesthesia success.

3. Chemical methods

Chemical methods are important tools for improving dental topical anesthesia. Table 1 summarizes the clinical trials conducted to evaluate topical anesthetic efficacy of LAs in humans.

3.1. Nanostructured carriers

Previous reviews have been published concerning DDSs based on technologies including liposomes, biopolymers, CDs, lipid nanoparticles, hydrogels, and patches, designed to prolong the anesthetic effect and to decrease the toxicity of LAs in infiltrative administration in medicine and dentistry [36], and for skin anesthesia [37]. Specific focus on DDSs for topical anesthesia on oral mucosa was outside the scope of these revisions, so this topic is therefore addressed in the present review.

3.1.1. Liposomes

Liposomes are lipid vesicles formed after agitation of phospholipids in water. They can have one or more lipid bilayers, where the hydrophobic lipid tails are directed toward the core and the polar heads face the bilayer surface, in contact with the aqueous phase. Liposomal drug delivery is a technological platform with recognized clinical acceptance, since a variety of drugs can be encapsulated into liposomes [38]. Because liposomes are typically made from natural lipid molecules, they are biocompatible [39], biodegradable, nontoxic [40], and show no or very little immunogenicity [39,41]. Although lipid bilayers present an intrinsic immune-adjuvant effect [42] that is potentiated in modified (pegylated and cationic) liposomes [41,43], there are no reports on adverse immune effects of liposomes topically administered in the mouth.

The first report regarding the use of LAs encapsulated in liposomes was in 1996. Zed and coworkers [24] demonstrated that the topical application of a liposomal 5% tetracaine formulation provided better pain relief than a 20% benzocaine gel during infiltrative injection of 4% prilocaine in 30 volunteers.

Liposome-encapsulated ropivacaine in a Carbopol® gel formulation applied to oral mucosa prior to LA injection improved the pain relief of needle insertion in a simulated local anesthesia procedure in healthy volunteers [32,35]. However, it was

Table 1. Summary of clinical trials investigating the efficacy of different formulations in promoting dental topical anesthesia at oral mucosa.

Formulations tested	Test method	Site and duration of application	Clinical study characteristics	Most effective formulation	Reference
Liposome encapsulated 5% tetracaine versus 20% benzocaine gel	**Pain during needle insertion and LA injection**	**Information not available**	**30 volunteers; double-blind, split-mouth design**	**Liposomal tetracaine**	[24]
(Part I) saliva-activated bioadhesive drug delivery system containing 20 mg lidocaine hydrochloride (SABDDS); (Part II) SABDDS versus 2% lidocaine gel (Xylocaine gel); (Part III) SABDDS	(Parts I and II) pinprick with a moon's probe; (Part III) exodontia	(Part I) Canine buccal fold of the mandible; (Part II) Canine buccal fold of the mandible/maxilla; (Part III) buccal and palatal/lingual region of the tooth to be extracted – time not defined	(Part I) 3 volunteers; (Part II) 5 volunteers; (Part III) 41 patients	(Part II) SABDDS not different to placebo; (Part III) only SABDDS was evaluated and promoted anesthesia	[25]
HPC film of tetracaine in association with analgesics, antibiotics, and antifungal agents versus Xylocaine™ and/or systemic analgesics	**Treatment of acute radiation-induced oral mucositis**	**Painful area – 1 h**	**52 patients who had received definitive radiation therapy for squamous cell carcinoma**	**HPC film of tetracaine in association with analgesics**	[26]
HPC-three layer mucosa-adhesive film containing dibucaine	**Alleviation of severe pain on chewing due to oral ulcers caused by chemotherapy and/or radiotherapy**	**Painful area – 30 min before taking a meal**	**Case report, 2 patients**	**Only dibucaine film was evaluated and was effective**	[27]
(Part I) Transoral lidocaine delivery system (DentiPatch) versus placebo; 20% benzocaine gel (Hurricane®, Beutlich Pharmaceuticals LLC) versus placebo; (Part II) DentiPatch versus 20% benzocaine gel	Pain during needle stick with a 25-G needle; pain during subgingival periodontal instrumentation	Buccal gingiva apical to the molar–bicuspid pair in the maxilla or mandible – 15 min DentiPatch and 30 s Hurricane	(Part I) 20 patients, randomized, placebo-controlled; (Part II) 20 patients, randomized	DentiPatch	[28]
(Part I) Transoral lidocaine delivery system (DentiPatch) versus placebo; 20% benzocaine gel (Hurricane) versus placebo; (Part II) DentiPatch versus 20% benzocaine gel; (Part III) DentiPatch versus placebo	Pain during needle stick with a 25-G or 27-G needle; pain during LA injection; pain during subgingival periodontal instrumentation	Buccal gingiva apical to the molar–bicuspid pair in the maxillary arch – 15 min DentiPatch and 30 s Hurricane	(Part I) 20 patients, randomized, placebo-controlled, double-blinded; (Part II) 20 patients, randomized; (Part III) 20 patients, randomized, placebo-controlled, double-blinded	DentiPatch	[29]
Lidocaine mucoadhesive patch (DentiPatch) versus 20% benzocaine gel (Hurricane)	Gingival anesthesia for rubber dam clamp placement	Buccal or lingual gingiva ~1 mm from the first and second premolars and permanent molars in the maxilla and mandible – 5 min DentiPatch and 1 min Hurricane	28 children, randomized, split-mouth design	All formulations tested were equally effective	[30]
Lidocaine transmucosal delivery system (DentiPatch)	Pain during scaling and root planing	Buccal gingiva apical to the treated area in maxilla or mandible – 5 min	40 patients, single-blind, randomized, four period, two treatment, placebo-controlled, cross-over design	Only DentiPatch was evaluated and was effective	[31]
1% liposomal ropivacaine gel versus 1% plain ropivacaine gel; 20% benzocaine gel; eutectic mixture of 2.5% lidocaine and 2.5% prilocaine (EMLA cream)	**Pain during needle insertion**	**Maxillary right canine buccal fold – 2 min**	**30 volunteers, randomized, blinded, cross-over study**	**Liposomal ropivacaine and EMLA**	[32]
Intraoral topical anesthetics lidocaine 20% patch (DentiPatch) and lidocaine 5% gel	Pain during repeated needle insertions and LA injection	Maxillary buccal fold of the first premolar and palatal mucosa – 15 min	31 children, randomized, unblinded, cross-over study	All formulations tested were equally effective	[33]
10% liposomal benzocaine gel versus 10% plain benzocaine gel; 20% benzocaine gel	**Pain during needle insertion**	**Maxillary right canine buccal fold – 2 min**	**30 volunteers, randomized, double-blinded, cross-over study**	**Liposomal benzocaine**	[34]
2% liposomal ropivacaine gel versus 20% benzocaine gel; liposomal placebo gel and placebo gel	**Pain during needle insertion**	**Maxillary lateral incisor buccal fold (bilaterally) – 30 min**	**40 volunteers, randomized, double-blinded, placebo controlled, cross-over study**	**Liposomal ropivacaine and benzocaine gel**	[35]
2% liposomal ropivacaine gel versus 1% liposomal ropivacaine gel; EMLA cream; liposomal placebo gel	**Pain during needle insertion and LA injection at palate**	**Palatal mucosa at the right canine region – 5 min**	**40 volunteers, randomized, single-blinded, placebo controlled, cross-over study**	**No difference among anesthetic formulations and placebo**	[8]
Liposome-encapsulated 2% lignocaine with epinephrine 1:100,000 versus 18% benzocaine/2% tetracaine hydrochloride gel (OneTouch®)	**Repeated pinprick tests with a periodontal probe (Part I) and LA injection using a 30-G needle (Part II)**	**At the junction between the vertical and horizontal parts of the mucosa near the roots of the upper first molars; (Part I) palatal mucosa – 4 min both formulations; (Part II) 4 min liposomal lidocaine and 1 min OneTouch**	**10 volunteers (Part I) and 22 volunteers (Part II)**	**Liposomal lignocaine with epinephrine**	[9]
Liposome-encapsulated 2.5 or 5% lidocaine versus 5% lidocaine gel (Xylocaine); EMLA cream; liposomal placebo gel and placebo gel	**Pain during needle insertion and LA injection at palate**	**Maxillary buccal fold at the canine region bilaterally – 2 min**	**40 volunteers, randomized, double-blinded, placebo controlled, cross-over study**	**Liposomal lidocaine and EMLA**	[10]

Phase I clinical trials are highlighted in bold type. HPC: Hydroxypropyl cellulose; LA: local anesthetic; EMLA: eutectic mixture of lidocaine and prilocaine.

demonstrated that the same liposomal formulation or other commercially available topical anesthetic formulations, including EMLA, were not able to reduce the pain during LA injection into the palatal mucosa [8].

In another study, a liposome-encapsulated 5% lidocaine formulation presented high *in vitro* permeation through porcine palatal epithelium. The same formulation showed topical anesthetic efficacy equivalent to that of EMLA, and both were able to reduce pain during needle insertion and LA injection in the palatal mucosa of healthy volunteers [10].

In addition, Paphangkorakit and coworkers [9] compared a liposomal 2% lidocaine formulation and a commercial gel (18% benzocaine/2% tetracaine), both used to reduce pain during anesthetic injection in the palatal mucosa of healthy volunteers. Despite the imprecise and unsophisticated methodologies employed for liposome preparation (vibration of an equimolar egg phosphatidylcholine:cholesterol mixture during 1 min, in an ultrasonic dental scaler) and anesthesia verification (repeated pin-pricks using a split-mouth, unblinded anesthesia protocol), the liposomal formulation showed better results than the commercial formulation.

The greater permeability of the oral mucosa (up to 4000 times more permeable, compared to intact skin [13]) would suggest that some of the liposome-encapsulated LA applied to this mucosa could penetrate the cortical bone, resulting in pulpal (tooth) anesthesia. This hypothesis was tested in human volunteers, comparing liposomal benzocaine and ropivacaine gels with commercial formulations [32,34,35]. Liposome-encapsulated 1% ropivacaine, 20% benzocaine, and EMLA were not able to induce pulpal anesthesia after 2-min application in the maxillary buccal fold [32]. Even after 30-min application in the same oral region, liposomal 2% ropivacaine was not able to induce pulpal anesthesia [35]. Furthermore, a liposomal 10% benzocaine formulation induced longer soft tissue anesthesia than a 20% benzocaine commercial gel, after a 2-min application in the maxillary buccal fold, but neither formulation induced pulpal anesthesia [34].

Hence, despite the conflicting results reported in the literature, in some cases, the encapsulation of LA into liposomes increased topical anesthesia efficacy, suggesting that this may be an important strategy that could be improved in order to achieve pain-free dental anesthesia. If combined with other nanotechnology tools (in a hybrid approach, see below), we can look forward to the emergence of many more clinical DDSs based on liposomal LAs in the near future.

3.1.2. CDs

CDs are cyclic oligosaccharides with a hydrophilic outer surface and a lipophilic central cavity. Their basic constitution is six or more α-1,4-linked D-glucopyranose units. CDs, especially β-cyclodextrin (β-CD) and derivatives, offer advantages for drug delivery including improved solubilization, protection against physicochemical and enzymatic degradation, and potential enhancement of absorption. CDs are useful for transporting drugs through aqueous phases, enabling their partitioning into membranes [36,44,45]. Most commercially available LAs have a benzene ring that docks inside β-CD or its derivatives. The association constants (K_a) resulting from this interaction show that esters (tetracaine, benzocaine, and proparacaine) present higher K_a values in comparison to amides (bupivacaine, lidocaine, prilocaine, and ropivacaine), hence exhibiting stronger interactions with CDs [46–52].

However, there are few studies reporting the use of LA-CD complexes in oral mucosa topical formulations. Arakawa and colleagues [53] investigated the ability of a low molecular weight β-CD polymer (4–5 β-CD units cross-linked with epichlorohydrin) for controlled release of drugs in a mucoadhesive buccal film. A film containing water-soluble low molecular weight β-CD polymers was prepared, using hydroxypropyl cellulose (HPC) or poly(vinyl alcohol) (5% w/v) as a film base. Lidocaine (0.5%) was complexed with this β-CD polymer and the *in vitro* release of lidocaine in artificial saliva was assessed. The results showed a 40% delay in release of lidocaine from the mucoadhesive β-CD polymer film, compared to the control base film. This was a clear demonstration that the addition of low molecular weight β-CD polymer to the film could control the *in vitro* release of the drug [53].

In 2010, Jug and coworkers [11] prepared and characterized binary systems for bupivacaine with β-CD or β-CD polymer (units of β-CD cross-linked with epichlorohydrin), with the aim of producing a new buccal mucoadhesive formulation to deliver bupivacaine through oral mucosa. Solid-state analysis revealed more intense interactions of bupivacaine with the β-CD polymer than with the native β-CD. *In vitro* dissolution studies using artificial saliva in Franz diffusion cells demonstrated that it was possible to change the dissolution characteristics of bupivacaine by complexation with CDs. The drug dissolution rate was reduced for bupivacaine complexed with native β-CD, and increased with the β-CD polymer. Therefore, the authors suggested that the bupivacaine release rate could be modulated according to the desired effect, with rapid onset of anesthesia for simple dental procedures, or prolonged effects in postsurgical procedures.

Animal studies of the toxicity of β-cyclodextrin have indicated that nephrotoxicity may occur following infiltrative administration [54]. However, β-CD derivatives such as hydroxypropyl-β-CD and sulfobutyl-β-CD are less toxic and have been approved by the FDA for injection [36].

The use of CD formulations directly on mucosa is considered safe for both natural CDs and their hydrophilic derivatives in a broad range of concentrations, while in the case of methylated CD derivatives, the concentration and duration of application should be monitored [55]. Boulmedarat and coworkers [44] evaluated the toxicity of randomly methylated β-CD in a buccal mucosa model (reconstituted human oral epithelium). Measurements were made of cell viability (MTT assay), membrane damage (lactate dehydrogenase release), and inflammatory effects (expression of interleukin-1α). The results indicated cytotoxic and inflammatory effects of 10% methylated β-CD, depending on the time of exposure. Lower concentrations (2% or 5%) did not induce any damage in the buccal tissue.

3.1.3. Lipid nanoparticles

The first and second generations of lipid nanoparticles, namely SLNs and NLCs, respectively, were developed as alternatives to

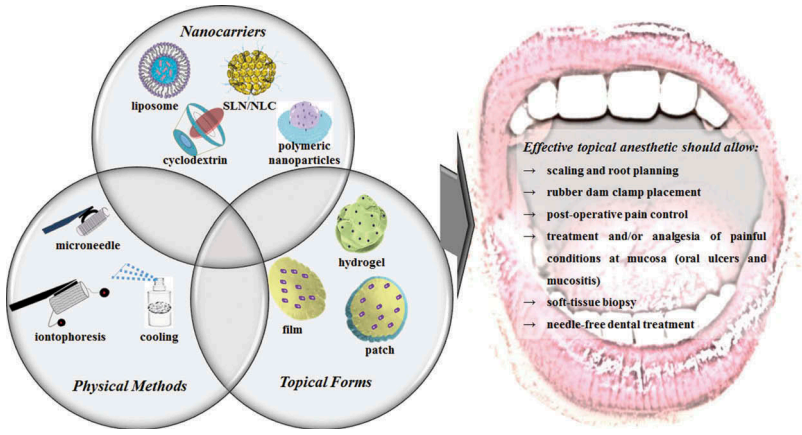

Figure 3. Schematic representation of drug delivery systems and physical methods used to increase topical anesthetic efficacy, as well as their possible combinations to achieve a better anesthetic effect and their possible clinical applications.

the existing nanosystems in an attempt to overcome limitations of liposomes such as poor physicochemical stability and low encapsulation efficiency for hydrophobic drugs. SLNs and NLCs are very similar nanocarriers, differing in their internal matrices. The SLN matrix consists of solid lipids, while NLCs are composed of a blend of solid and liquid lipids, with the resulting internal disorganization leading to better entrapment of a drug and prevention of its expulsion over time. However, the ability of these lipid nanosystems to encapsulate hydrophilic molecules is lower, compared to liposomes. They have been studied since the 1990s and are excellent options for use in DDSs for topical application, due to their nanometric size, large contact surface area, biocompatibility, biodegradability, and occlusive effect, which improves the permeation of active molecules [56]. SLNs and NLCs have been successfully used for targeting drugs such as cyclosporine A, curcumin, clotrimazol, and miconazole to the oral mucosa, proving their potential as excellent carriers for use in transbuccal administration [57–60].

Only a few studies have been published concerning the encapsulation of LA by SLNs and NLCs for topical applications, most of them involving transdermal delivery. NLCs loaded with lidocaine and benzocaine have exhibited desirable physicochemical properties, excellent *in vitro* permeation, sustained release profiles, and absence of cytotoxic effects [61–63].

In the case of the transbuccal route, no studies have yet been published concerning lipid nanoparticle systems for topical anesthesia. However, it is worth mentioning a study [64] in which optimized NLC systems for lidocaine–prilocaine presented physicochemical stability during up to 14 months of storage at 25°C, in terms of size, polydispersity index, and zeta potential. The release of the anesthetics was sustained for up to 20 h and the system protected both LAs against degradation. Rapid onset and long duration of anesthesia indicated the potential of the system for transbuccal topical anesthesia.

Nevertheless, the low viscosity of the colloidal systems mentioned above is not desirable for topical anesthesia of oral mucosa, because an efficient adhesion is required [65]. Recent advances in pharmaceutical nanotechnology include the exploration of hybrid approaches such as combinations of lipid and PN systems, nanoparticles and polymer-based topical pharmaceutical forms, nanoparticles and physical methods, and polymer-based topical pharmaceutical forms and physical methods (Figure 3). These multiple associations combine the best properties of each excipient in the final formulation [66] and they can provide versatile strategies for the development of new and improved mucoadhesive formulations that are engineered to present sustained release profiles (*smart DDSs*). Wang et al. [67] compared the performance of liposomes and lipid–polymer hybrid nanoparticles with encapsulated lidocaine for topical application. The lipid–polymer hybrid nanoparticles showed smaller particle size (<100 nm) and higher encapsulation efficiency (>85%) than liposomes, as well as the best *in vitro* sustained release and permeation profile of lidocaine. Studies have also described the incorporation of SLN dispersions into biopolymeric matrices to produce different mucoadhesive formulations, such as lipid–polymer gels and sponges, for the transbuccal release of curcumin [68,69].

3.1.4. PN systems

PNs are able to permeate the oral mucosa [70]. They can typically be prepared as polymeric micelles, nanospheres, and nanocapsules [71]. These nanosystems have been successfully used for transbuccal applications, due to their nanometric sizes, sustained release profiles, and good adhesion. Applications include cancer and periodontic treatments, as well as reduction of tooth hypersensitivity [72]. Despite their useful properties, to date, there are no reports regarding the use of these nanocarriers in topical transbuccal anesthesia.

However, LAs encapsulated in different PNs have been widely reported for a variety of applications. Optimized formulations of benzocaine loaded in poly-(D,L-lactide-*co*-glycolide) nanocapsules have been described, with desirable structure-dependent properties and sustained release profiles [73]. Nanocapsules based on poly(L-lactide) showed longer anesthetic action after sensory sciatic nerve blockade in mice, compared to other benzocaine-loaded PNs [74]. Alginate nanoparticles encapsulating bupivacaine were able to reduce the toxicity of the LA and prolong the anesthetic effect in mice [75]. The delivery of lidocaine using nanospheres based on poly(ε-caprolactone) resulted in increased intensity and duration of *in vivo* anesthesia in sciatic nerve blockade in mice [76].

De Melo et al. [77] evaluated different polymeric nanocarriers for articaine delivery. The best nanocapsule system was composed of poly(ethylene glycol)-poly(ε-caprolactone). This system minimized the toxicity of the drug and exhibited sustained *in vitro* release, probably due to strong interaction between articaine and the aqueous nucleus of the nanocapsule. These achievements support the potential use of PNs for topical anesthesia of oral mucosa.

3.2. Polymer-based topical pharmaceutical forms: bio- and mucoadhesive films, patches, and hydrogels

The main drawbacks of topical administration of drugs to the oral mucosa using traditional semisolid formulations is the short time of contact with the tissue, resulting in a low efficacy of the treatment, as well as the lack of unidirectional drug release, which can lead to systemic absorption.

The use of biopolymers as matrices for the incorporation of therapeutic agents is a versatile strategy that confers important properties on the systems, such as protection from photochemical degradation, a sustained release profile, and adequate swelling, permeation, and mucoadhesion, besides the possibility of unidirectional drug release. The resultant materials are extensively used as DDSs [78] and can be prepared as hydrogels, films, and patches, circumventing the limitations of the oral mucosa route. Lidocaine, the gold-standard LA in dentistry, has been extensively studied for sustained delivery systems targeting the oral mucosa [79–81]. However, reports about systems incorporating other LAs still remain scarce.

In 1996, the FDA approved DentiPatch™ (Noven Pharmaceuticals, Inc.), the first 'lidocaine transoral delivery system intended to provide topical anesthesia for the prevention of pain from injection and soft tissue dental procedure' [82]. DentiPatch contains 41.6 mg of lidocaine in 2 cm^2 of a karaya gum adhesive matrix, covered by a polyester film, and according to the manufacturer is able to induce anesthesia in 2.5 min. Its safety and efficacy have been proved in both adults [28,29,31,83,84] and children [30,33,85–87], although poor adhesion to oral mucosa has been reported [30].

Almost all films and patches can generally be prepared by the casting/solvent evaporation technique, but other methods have also been described, such as the lamination technique and hot-melt extrusion. The most widely studied cellulose derivative for the formulation of buccal films containing LA is HPC [26,27,81,88–90]. Films of HPC have been successfully used *in vivo* for relief of the pain of oral diseases. Yamamura et al. [27] used a three-layer HPC mucosa-adhesive film containing dibucaine for the treatment of oral ulcer. Oguchi et al. [26] employed a film of HPC containing tetracaine, in association with other drugs, for treating acute radiation-induced oral mucositis. The association of HPC with hydroxypropyl methyl cellulose (HPMC) or ethyl cellulose (EC) has proven to be useful in modulating drug release and for film mucoadhesion *in vitro*. In particular, the use of HPMC in combination with HPC for the preparation of hot-melt extruded films containing lidocaine promoted increased adhesion to intestinal rabbit mucosa (used as a biological substrate model) and a decrease of the lidocaine release rate, indicative of a prolonged analgesic effect [90]. The addition of EC to HPC solid dispersion films enabled modulation of lidocaine release, with an optimum obtained when the polymers were used in a 1:1 ratio. The optimized film showed significant mucoadhesion to the buccal mucosa *in vivo* in men [88].

An alternative to cellulose derivatives is provided by Carbopol, a water-insoluble acrylic polymer that shows good bioadhesion. Abu-Huwaij et al. [91,92] proposed mucoadhesive Carbopol films for the delivery of lidocaine hydrochloride. The mucoadhesive properties were influenced by the type and concentration of the plasticizer used, as well as by the concentration of lidocaine, which interacted with the polymer and reduced its bioadhesion [91]. The coupling of this film with an ethyl vinyl acetate backing, in order to ensure unidirectional drug release, together with the introduction of permeation enhancers such as oleic acid and Tween, amongst others, significantly prolonged the permeation of lidocaine across pig buccal mucosa, compared to a control solution. The system also showed good *in vivo* mucoadhesion in rabbits [92]. A Carbopol film containing HPMC was developed by Cavallari [79] for the buccal delivery of lidocaine. A hybrid approach, using the addition of Compritol microspheres containing lidocaine, resulted in a bimodal controlled release of the drug, with 40% release in 30 min and 100% release in 3 h.

Finally, poly(vinyl alcohol) was used in the development of a film for the buccal delivery of lidocaine, produced by the lamination technique [93]. The association with an acrylic polymer improved the transport of lidocaine across pig esophageal epithelium (used as a buccal mucosa model), possibly due to greater mucoadhesion of the film.

Hydrogels are tridimensional semisolid substances composed of networks of natural or synthetic polymers, with functional groups that interact synergistically with mucosal tissue. Their advantages as bases for topical anesthetic formulations include biodegradability, easy handling, patient compliance, softness, mucoadhesion, and rapid onset of anesthesia [94].

Abdel-Hamid et al. [95] proposed the application of mebeverine (an antispasmodic agent) as a LA contained in Poloxamer-407 hydrogels for the treatment of painful oral conditions such as lichen planus, recurrent aphthae, erythema multiforme, and Behçet's syndrome. Clinical evaluation of pain reduction efficiency showed a greater anesthetic effect of the proposed hydrogel, compared to a commercially available formulation, together with improved wound healing of the buccal mucosa.

Xu et al. [96] developed hydrogels based on genipin-cross-linked catechol–chitosan for the buccal mucosa delivery of lidocaine as a model drug. A hydrogel was obtained that showed good *in vitro* mucoadhesion to porcine buccal mucosa tissue, together with good mechanical and rheological properties. Evaluation of the *in vivo* drug release in rabbits confirmed the sustained release profile. Similarly, Pignatello et al. [97] suggested a mucoadhesive lidocaine-loaded hydrogel for buccal applications, based on chitosan glutamate, with clinical evaluation confirming the potential of the formulation to minimize pain associated with oral mucosa disorders.

Hirsh et al. [98] patented a gelled anesthetic preparation for oral and buccal mucosa application, which was especially indicated for the treatment of periodontal pockets. The formulation showed adequate viscosity and adhesion and provided long-term gingival anesthesia. The LAs that were most

compatible with the excipients of this patented formulation included benzocaine, tetracaine, and butamben, as well as blends of these drugs.

These interesting studies suggest the ability of biopolymers to act as excellent matrices for the incorporation of other LAs in the preparation of mucoadhesive transbuccal formulations.

4. Physical methods

Several physical methods are able to reduce pain during needle insertion and LA injection. They are also used to improve the efficacy of topical anesthetic agents applied to the oral mucosa. One of the most widely reported methods is the precooling technique.

Cryoanesthesia is based on the application of ice or cold to a surface area of the body, causing reduction of action potentials and resulting in sensorial nerve conduction blockade [99]. In dentistry, application of precooling to oral mucosa prior to LA infiltration can alter the perception of pain. Harbert et al. [100] observed reduced pain perception during LA infiltration when ice was applied topically on the palatal mucosa before and during the infiltration. Duncan et al. [101] reported relief of the discomfort caused by needle insertion after applying a cotton pellet saturated with dichlorodifluoromethane spray for 5 s on a small portion of palatal tissue.

Kosaraju and Vandewalle [102] used a split-mouth design in 16 volunteers to compare the topical anesthesia effectiveness of a refrigerant spray (1,1,1,3,3-pentafluoropropane/1,1,1,2-tetrafluoroethane) applied during 5 s and a 20% benzocaine gel applied during 2 min, both used prior to a palatal anesthetic injection. The volunteers rated their experience using a visual analog scale (VAS). The refrigerant spray was more effective than the benzocaine gel in reducing the pain during injection. Lathwal et al. [99] compared the effects of the same refrigerant spray (5 s), benzocaine (1 min), and ice (1 min) on pain perception during bilateral local anesthesia nerve block in 160 patients aged between 5 and 8 years. Significantly higher anesthesia efficacy was observed for ice, compared to benzocaine or the refrigerant spray.

In other work, 160 patients aged between 5 and 6 years were randomly allocated to receive pretreatment with topical anesthesia, alone or with ice cooling, before administration of inferior alveolar nerve blockage. A significant reduction of pain perceived during injection of the LA was observed when ice was used [103]. Ghaderi et al. [104] used a cross-over design in 50 healthy pediatric patients, evaluating pain perception during LA buccal infiltration after using topical anesthesia (20% benzocaine, 1 min) with or without application of an ice pack (1 min). It was observed that cooling the injection site before infiltration of the LA reduced the perceived pain.

Overall, the use of precooling with a 5-s application of a refrigerant spray or a 1-min application of ice seems to improve (or to itself promote) superficial anesthesia, leading to reduced pain perception during LA administration.

The mechanical stimulus (pressure or vibration) technique has been used to minimize the pain experienced during administration of LA agents. The stimulation of mechanical receptors by vibration or pressure excites inhibitory interneurons in the spinal cord, resulting in the elimination of transfer of nociceptive information by Aδ and C fibers to the second order neurons of the spinal cord [105]. Some devices are able to cause vibration while others simultaneously produce both vibration and pressure during injection of the anesthetic. The data published concerning these devices remains conflicting, with some studies showing that use of the devices leads to painless injection, while others have found no significant decrease in the perception of pain in the oral mucosa [106,107].

In contrast to these positive results obtained with the different techniques, in terms of reduced discomfort of anesthetic injection, Wiswall et al. [108] demonstrated that the application of topical anesthetics, precooling, and pressure did not reduce pain perception during anesthetic injection and deposition in bilateral greater palatine nerve blockage. A cross-over study was employed, with 42 volunteers who received the following pretreatments: (1) no stimulation; (2) pressure alone; (3) pressure + topical anesthetic (20% benzocaine); and (4) pressure + cold (1,1,1,2-tetrafluoroethane). The pain perception was rated using the Heft-Parker VAS, and no significant differences were found among the pretreatments. In addition, 1,1,1,2-tetrafluoroethane caused injuries in the oral mucosa when used together with pressure for 10 s [108].

Other different approaches, such as iontophoresis and microneedle arrays, have been studied in the last decade in order to achieve deeper and more effective topical anesthesia in the oral mucosa. Iontophoresis is a technique based on the application of a low-density electric current to facilitate the transfer of drugs through biological membranes. Iontophoresis is used in medicine for transdermal drug delivery but has been little explored in transbuccal delivery.

Hu et al. [109] studied the use of iontophoresis to enhance transdermal and transmucosal delivery of lidocaine and other drugs. The authors performed *in vitro* transdermal and transbuccal permeation experiments in Franz diffusion cells, using porcine skin and buccal tissues. The application of iontophoresis enhanced drug delivery, with a greater effect on transdermal than on transbuccal permeation.

Cubayachi et al. [110] recently evaluated the use of iontophoresis to enhance the permeation of prilocaine and lidocaine salts, combined in a mucoadhesive semi-solid formulation. *In vitro* studies were performed using pig esophageal mucosa as a barrier to simulate buccal mucosa. Iontophoresis resulted in increased retention of the LAs in the mucosal epithelium. Although only *in vitro* data were available, the authors hypothesized that this strategy might be used in needle-free applications in buccal anesthesia.

Microneedles have been extensively studied as a mechanical technology for transdermal delivery [111]. They are sufficiently long to cross the epithelium barrier and deliver the drug, but short enough to avoid the free nerve endings located in the dermal and *lamina propria* layers [112]. Kochhar et al. [113] observed high transdermal delivery and a faster onset of anesthesia for lidocaine in microneedles in a transdermal patch, compared to a traditional patch. In other work, porcine skin pretreated with microneedles showed enhanced permeation of a formulation of lidocaine in a sodium carboxymethyl cellulose/gelatin hydrogel [114].

Other studies have also investigated microneedle-based drug delivery for transdermal applications [115], and the use of coated microneedles has been reported to be a successful method for the delivery of lidocaine across pig skin [116]. The combination (in a hybrid approach) of drug encapsulation in nanoparticles and the use of coated microneedles has also been described for buccal topical use. Microneedles coated with doxorubicin encapsulated in PNs (poly(lactic-*co*-glycolic) acid) were found to effectively promote uniform distribution of the drug in a porcine cadaver buccal tissue [117].

Despite the lack of studies to demonstrate that the use of microneedles significantly increases the effectiveness of topical anesthetic in oral mucosa, this physical method (as well as its combination with other DDSs) seems to be a promising tool for use in future clinical studies.

5. Conclusions

Different DDSs and pharmaceutical dosage forms have been successfully developed with the aim of increasing the effectiveness of topical anesthesia in oral mucosa. Most of these promising formulations have shown interesting properties *in vitro*. Although there have been only a few *in vivo* clinical trials, these studies have confirmed the pharmacological improvements, especially in the case of liposomal formulations. In addition, the precooling of oral mucosa has been shown to be a valid tool for promoting pain-free local anesthesia. The continuing development of new systems and formulations for clinical use in the oral cavity will help to achieve significant improvements in topical oral anesthesia.

6. Expert opinion

Even today, it is utopic to conceive of a single nanosystem that might solve all the requirements for drug delivery in the oral mucosa, as reflected in the disappointing results for some of the clinical trials listed in Table 1. Despite substantial advances in pharmaceutical nanotechnology, including in dentistry, all the current systems have limitations in applications involving the oral mucosa. In order to select the most appropriate topical nanosystem, it is crucial to consider the physicochemical properties of the anesthetic agent and the specific characteristics of the site of administration (such as the gingival or palate mucosa). The use of a suitable excipient to maintain the drug at the site of application increases the potential success of the formulation, as shown for cyclodextrin, lipid-based, and polymeric nanosystems.

The association of two or more DDSs, in nanohybrid approaches (Figure 3), seems to be the most promising strategy to achieve successful topical anesthesia in the oral mucosa, combining the desirable properties of each material in the pharmaceutical formulation. The systems can be tailored to behave according to specific features of the oral cavity, such as temperature, humidity, salivary flow, free ions available in saliva, and the anionic mucosal surface. This can result in synergism between the formulation components and the biological interface, creating *smart DDSs*. The main objective, which remains a challenge, is the creation of a versatile material able to remain functional despite the large number of physiological variables that can hinder the success of formulations. Further development of nanohybrid approaches associated with physical methods seems to be the best way forward in terms of improved superficial anesthesia.

The use of biopolymers also deserves more attention, since these are derived from inexpensive, biocompatible, abundant, and multifunctional raw materials. In addition, multidisciplinary tools can be used to help in elucidating the performance of each component, as well as the interactions among them in the final formulation. This can minimize the likelihood of failure of the formulation and enable extrapolation to the delivery of other drugs by the transbuccal route.

An effective anesthetic formulation intended for topical oral applications should have additional clinical benefits besides the prevention of pain during dental anesthesia. An effective formulation could also provide postoperative pain control, treatment and/or analgesia of painful mucosal conditions such as oral ulcers and mucositis, gingival anesthesia prior to rubber dam clamp placement, scaling, and root planing, and anesthesia in soft tissue biopsy. These aspects have been considered in several of the clinical trials mentioned in Table 1 (as proposed in Figure 3). Among all the possible advantages, one should be highlighted, namely topical anesthesia to replace traditional dental anesthesia, hence enabling needle-free dental treatment. In terms of this goal, conflicting results have been reported. Although some studies have reported positive results [25,118–120], others have failed to reach such conclusions [32,34,35,121,122]. Nonetheless, this remains the greatest aspiration among both patients and clinicians in dentistry.

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Declaration of interest

The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.

References

Papers of special note have been highlighted as either of interest (•) or of considerable interest (••) to readers.

1. Meechan JG. Intraoral topical anesthesia. Periodontol 2000. 2008;46:56–79.
•• **A review on the pharmacology, clinical use, and toxicity of topical anesthetics and the factors influencing topical anesthetic efficacy.**
2. Hmud R, Walsh L. Dental anxiety: causes, complications and management approaches. J Minim Interv Dent. 2009;2:67–78.
3. Malamed S. Handbook of local anesthesia. 6th ed. St. Louis: Elsivier; 2013.
4. Meechan JG. Intra-oral topical anaesthetics: a review. J Dent. 2000;28:3–14.

5. Hutchins HS, Young FA, Lackland DT, et al.. The effectiveness of topical anesthesia and vibration in alleviating the pain of oral injections. Anesth Prog. 1997;44:87–89.
6. Bhalla J, Meechan JG, Lawrence HP, et al. Effect of time on clinical efficacy of topical anesthesia. Anesth Prog. 2009;56:36–41.
7. de Freiras GC, Pozzobon RT, Blaya DS, et al.. Efficacy of benzocaine 20% topical anesthetic compared to placebo prior to administration of local anesthesia in the oral cavity: a randomized controlled trial. Anesth Prog. 2015;62:46–50.
8. Franz-Montan M, de Paula E, Groppo FC, et al. Liposomal delivery system for topical anaesthesia of the palatal mucosa. Br J Oral Maxillofac Surg. 2012;50:60–64.
9. Paphangkorakit J, Sangsirinakagul C, Priprem A. Relief of palatal injection pain by liposome-encapsulated 2% lignocaine prepared by ultrasonic dental scaler. Br J Oral Maxillofac Surg. 2012;50:784–787.
10. Franz-Montan M, Baroni D, Brunetto G, et al. Liposomal lidocaine gel for topical use at the oral mucosa: characterization, in vitro assays and in vivo anesthetic efficacy in humans. J Liposome Res. 2015;25:11–19.
• **A very interesting paper which correlates palatal mucosa permeation and pulpal anesthesia.**
11. Jug M, Maestrelli F, Bragagni M, et al.. Preparation and solid-state characterization of bupivacaine hydrochloride cyclodextrin complexes aimed for buccal delivery. J Pharm Biomed Anal. 2010;52:9–18.
12. Hassan N, Ahad A, Ali M, et al.. Chemical permeation enhancers for transbuccal drug delivery. Expert Opin Drug Deliv. 2010;7:97–112.
• **An interesting review about chemical permeation enhancers for topical application at oral mucosa.**
13. Lesch CA, Squier CA, Cruchley A, et al. The permeability of human oral mucosa and skin to water. J Dent Res. 1989;68:1345–1349.
14. Squier CA. The permeability of oral mucosa. Crit Rev Oral Biol Med. 1991;2:13–32.
15. Squier C, Brogden K. Human oral mucosa: development, structure & function. Ames (IA): Wiley-Blackwell; 2011.
•• **A complete book that covers all aspects of the oral mucosa.**
16. Squier CA. Effect of enzyme digestion on the permeability barrier in keratinizing and non-keratinizing epithelia. Br J Dermatol. 1984;111:253–264.
17. Mashru R, Sutariya V, Sankalia M, et al.. Transbuccal delivery of lamotrigine across porcine buccal mucosa: in vitro determination of routes of buccal transport. J Pharm Pharm Sci. 2005;8:54–62.
18. Patel VF, Liu F, Brown MB. Advances in oral transmucosal drug delivery. J Control Release. 2011;153:106–116.
19. Sanz R, Calpena AC, Mallandrich M, et al.. Enhancing topical analgesic administration: review and prospect for transdermal and transbuccal drug delivery systems. Curr Pharm Des. 2015;21:2867–2882.
20. Paderni C, Compilato D, Giannola LI, et al.. Oral local drug delivery and new perspectives in oral drug formulation. Oral Surg Oral Med Oral Pathol Oral Radiol. 2012;114:e25–e34.
21. Morales JO, McConville JT. Novel strategies for the buccal delivery of macromolecules. Drug Dev Ind Pharm. 2014;40:579–590.
22. Harris D, Robinson JR. Drug delivery via the mucous membranes of the oral cavity. J Pharm Sci. 1992;81:1–10.
23. Meechan JG, Howlett PC, Smith BD. Factors influencing the discomfort of intraoral needle penetration. Anesth Prog. 2005;52:91–94.
24. Zed C, Epstein J, Donaldson D. Topical liposome encapsulated tetracaine versus benzocaine - A clinical investigation. J Dent Res. 1996;247. Abstract number: 1840.
25. Taware CP, Mazumdar S, Pendharkar M, et al. A bioadhesive delivery system as an alternative to infiltration anesthesia. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 1997;84:609–615.
26. Oguchi M, Shikama N, Sasaki S, et al. Mucosa-adhesive water-soluble polymer film for treatment of acute radiation-induced oral mucositis. Int J Radiat Oncol Biol Phys. 1998;40:1033–1037.
27. Yamamura K, Yotsuyanagi T, Okamoto T, et al.. Pain relief of oral ulcer by dibucaine-film. Pain. 1999;83:625–626.
28. Carr MP, Horton JE. Clinical evaluation and comparison of 2 topical anesthetics for pain caused by needle sticks and scaling and root planing. J Periodontol. 2001;72:479–484.
29. Carr MP, Horton JE. Evaluation of a transoral delivery system for topical anesthesia. J Am Dent Assoc. 2001;132:1714–1719.
30. Stecker SS, Swift JQ, Hodges JS, et al.. Should a mucoadhesive patch (DentiPatch) be used for gingival anesthesia in children? Anesth Prog. 2002;49:3–8.
31. Perry DA, Gansky SA, Loomer PM. Effectiveness of a transmucosal lidocaine delivery system for local anaesthesia during scaling and root planing. J Clin Periodontol. 2005;32:590–594.
32. Franz-Montan M, Silva ALR, Cogo K, et al. Liposome-encapsulated ropivacaine for topical anesthesia of human oral mucosa. Anesth Analg. 2007;104:1528–1531.
33. Bågesund M, Tabrizi P. Lidocaine 20% patch vs lidocaine 5% gel for topical anaesthesia of oral mucosa. Int J Paediatr Dent. 2008;18:452–460.
34. Franz-Montan M, Silva ALR, Fraceto LF, et al. Liposomal encapsulation improves the duration of soft tissue anesthesia but does not induce pulpal anesthesia. J Clin Anesth. 2010;22:313–317.
35. Franz-Montan M, de Paula E, Groppo FC, et al. Liposome-encapsulated ropivacaine for intraoral topical anesthesia. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2010;110:800–804.
36. de Paula E, Cereda CMS, Fraceto LF, et al. Micro and nanosystems for delivering local anesthetics. Expert Opin Drug Deliv. 2012;9:1505–1524.
•• **Review of DDSs for local anesthetics.**
37. de Araújo DR, da Silva DC, Barbosa RM, et al. Strategies for delivering local anesthetics to the skin: focus on liposomes, solid lipid nanoparticles, hydrogels and patches. Expert Opin Drug Deliv. 2013;10:1551–1563.
38. Allen TM, Cullis PR. Liposomal drug delivery systems: from concept to clinical applications. Adv Drug Deliv Rev. 2013;65:36–48.
39. Torchilin V. Liposomes in drug delivery. In: Siepmann J, et al., editors. Fundamentals and applications of controlled release drug delivery. New York (NY): Springer; 2012.
40. Gubernator J. Active methods of drug loading into liposomes: recent strategies for stable drug entrapment and increased in vivo activity. Expert Opin Drug Deliv. 2011;8:565–580.
41. Maheswaran A, Brindha P, Mullaincharam A, et al.. Liposomal drug delivery systems - a review. Int J Pharm Sci Rev Res. 2013;23:295–301.
42. Szebeni J, Barenholz Y. Complement activation, immunogenicity and immune suppression. In: Peer D, et al., editors. Harnessing biomaterials for nanomedicine: preparation, toxicity and applications. Boca Raton (FL): Pan Stanford Publishing Pte Ltd; 2009.
43. Peer D. Immunotoxicity derived from manipulating leukocytes with lipid-based nanoparticles. Adv Drug Deliv Rev. 2012;64:1738–1748.
44. Boulmedarat L, Bochot A, Lesieur S, et al.. Evaluation of buccal methyl-beta-cyclodextrin toxicity on human oral epithelial cell culture model. J Pharm Sci. 2005;94:1300–1309.
45. Loftsson T, Vogensen SB, Brewster ME, et al.. Effects of cyclodextrins on drug delivery through biological membranes. J Pharm Sci. 2007;96:2532–2546.
46. Moraes CM, Abrami P, de Paula E, et al. Study of the interaction between S(–) bupivacaine and 2-hydroxypropyl-beta-cyclodextrin. Int J Pharm. 2007;331:99–106.
47. de Araujo DR, Tsuneda SS, Cereda CMS, et al. Development and pharmacological evaluation of ropivacaine-2-hydroxypropyl-beta-cyclodextrin inclusion complex. Eur J Pharm Sci. 2008;33:60–71.
48. Moraes CM, Abrami P, de Araújo DR, et al. Characterization of lidocaine: hydroxypropyl-β-cyclodextrininclusion complex. J Incl Phenom Macrocycl Chem. 2007;57:313–316.
49. Cabeça LF, Figueiredo IM, de Paula E, et al.. Prilocaine–cyclodextrin–liposome: effect of pH variations on the encapsulation and topology of a ternary complex using 1H NMR. Magn Reson Chem. 2011;49:295–300.
50. Franco de Lima RA, de Jesus MB, Saia Cereda CM, et al. Improvement of tetracaine antinociceptive effect by inclusion in cyclodextrins. J Drug Target. 2012;20:85–96.
51. Pinto LMA, Fraceto LF, Santana MHA, et al. Physico-chemical characterization of benzocaine-beta-cyclodextrin inclusion complexes. J Pharm Biomed Anal. 2005;39:956–963.
52. Cabeça LF, Fernandes SA, de Paula E, et al.. Topology of a ternary complex (proparacaine-beta-cyclodextrin-liposome) by STD NMR. Magn Reson Chem. 2008;46:832–837.

53. Arakawa Y, Kawakami S, Yamashita F, et al.. Effect of low-molecular-weight beta-cyclodextrin polymer on release of drugs from mucoadhesive buccal film dosage forms. Biol Pharm Bull. 2005;28:1679–1683.
•• **A significant *in vitro* study showing that β-cyclodextrin polymer retarded the release of lidocaine from mucoadhesive buccal film.**
54. Rajewski RA, Traiger G, Bresnahan J, et al.. Preliminary safety evaluation of parenterally administered sulfoalkyl ether beta-cyclodextrin derivatives. J Pharm Sci. 1995;84:927–932.
55. Cal K, Centkowska K. Use of cyclodextrins in topical formulations: practical aspects. Eur J Pharm Biopharm. 2008;68:467–478.
56. Müller RH, Alexiev U, Sinambela P, et al. Nanostructured Lipid Carriers (NLC): the second generation of solid lipid nanoparticles. In: Dragicevic N, Maibach H, et al., editors. Percutaneous penetration enhancers chemical methods in penetration enhancement. Berlin: Springer; 2016. p. 161–185.
•• **Recent book chapter on nanostructured lipid carriers, describing the potential of these systems for topical application.**
57. Mendes AI, Silva AC, Catita JA, et al. Miconazole-loaded nanostructured lipid carriers (NLC) for local delivery to the oral mucosa: improving antifungal activity. Colloids Surf B Biointerfaces. 2013;111:755–763.
58. Esposito E, Ravani L, Contado C, et al. Clotrimazole nanoparticle gel for mucosal administration. Mater Sci Eng C Mater Biol Appl. 2013;33:411–418.
59. Karavana SY, Gökçe EH, Rençber S, et al. A new approach to the treatment of recurrent aphthous stomatitis with bioadhesive gels containing cyclosporine A solid lipid nanoparticles: in vivo/in vitro examinations. Int J Nanomedicine. 2012;7:5693–5704.
60. Hazzah HA, Farid RM, Nasra MMA, et al. Gelucire-based nanoparticles for curcumin targeting to oral mucosa: preparation, characterization, and antimicrobial activity assessment. J Pharm Sci. 2015;104:3913–3924.
61. Basha M, Abd El-Alim SH, Kassem AA, et al. Benzocaine loaded solid lipid nanoparticles: formulation design, in vitro and in vivo evaluation of local anesthetic effect. Curr Drug Deliv. 2015;12:680–692.
62. Pathak P, Nagarsenker M. Formulation and evaluation of lidocaine lipid nanosystems for dermal delivery. AAPS PharmSciTech. 2009;10:985–992.
63. Puglia C, Sarpietro MG, Bonina F, et al. Development, characterization, and in vitro and in vivo evaluation of benzocaine- and lidocaine-loaded nanostructrured lipid carriers. J Pharm Sci. 2011;100:1892–1899.
64. Ribeiro LN, Franz-Montan M, Breitkreitz MC, et al. Nanostructured lipid carriers as robust systems for topical lidocaine-prilocaine release in dentistry. Eur J Pharm Sci. 2016;93:192–202.
65. Souto EB, Wissing SA, Barbosa CM, et al.. Evaluation of the physical stability of SLN and NLC before and after incorporation into hydrogel formulations. Eur J Pharm Biopharm. 2004;58:83–90.
66. Hadinoto K, Sundaresan A, Cheow WS. Lipid-polymer hybrid nanoparticles as a new generation therapeutic delivery platform: a review. Eur J Pharm Biopharm. 2013;85:427–443.
67. Wang J, Zhang L, Chi H, et al.. An alternative choice of lidocaine-loaded liposomes: lidocaine-loaded lipid-polymer hybrid nanoparticles for local anesthetic therapy. Drug Deliv. 2016;23:1254–1260.
•• **This paper presents an innovative hybrid system for local anesthetic delivery, with improvement of the sustained release and permeation profile of lidocaine.**
68. Hazzah HA, Farid RM, Nasra MMA, et al. Lyophilized sponges loaded with curcumin solid lipid nanoparticles for buccal delivery: development and characterization. Int J Pharm. 2015;492:248–257.
69. Hazzah HA, Farid RM, Nasra MMA, et al. A new approach for treatment of precancerous lesions with curcumin solid-lipid nanoparticle-loaded gels: in vitro and clinical evaluation. Drug Deliv. 2016;23:1409–1419.
70. Sandri G, Poggi P, Bonferoni MC, et al. Histological evaluation of buccal penetration enhancement properties of chitosan and trimethyl chitosan. J Pharm Pharmacol. 2006;58:1327–1336.
71. Estanqueiro M, Amaral MH, Conceição J, et al.. Nanotechnological carriers for cancer chemotherapy: the state of the art. Colloids Surf B Biointerfaces. 2015;126:631–648.
72. Mazzarino L, Borsali R, Lemos-Senna E. Mucoadhesive films containing chitosan-coated nanoparticles: a new strategy for buccal curcumin release. J Pharm Sci. 2014;103:3764–3771.
73. Moraes CM, de Matos AP, Grillo R, et al. Screening of formulation variables for the preparation of poly(epsilon-caprolactone) nanocapsules containing the local anesthetic benzocaine. J Nanosci Nanotechnol. 2011;11:2450–2457.
74. De Melo NFS, De Araújo DR, Grillo R, et al. Benzocaine-loaded polymeric nanocapsules: study of the anesthetic activities. J Pharm Sci. 2012;101:1157–1165.
75. Grillo R, de Melo NFS, de Araújo DR, et al. Polymeric alginate nanoparticles containing the local anesthetic bupivacaine. J Drug Target. 2010;18:688–699.
76. Ramos Campos EV, Silva de Melo NF, Guilherme VA, et al. Preparation and characterization of poly(ε-caprolactone) nanospheres containing the local anesthetic lidocaine. J Pharm Sci. 2013;102:215–226.
77. De Melo NFS, Campos EVR, Gonçalves CM, et al. Development of hydrophilic nanocarriers for the charged form of the local anesthetic articaine. Colloids Surf B Biointerfaces. 2014;121:66–73.
78. Ribeiro LNM, Alcântara ACS, Darder M, et al. Bionanocomposites containing magnetic graphite as potential systems for drug delivery. Int J Pharm. 2014;477:553–563.
79. Cavallari C, Fini A, Ospitali F. Mucoadhesive multiparticulate patch for the intrabuccal controlled delivery of lidocaine. Eur J Pharm Biopharm. 2013;83:405–414.
80. Loughlin RG, Tunney MM, Donnelly RF, et al. Modulation of gel formation and drug-release characteristics of lidocaine-loaded poly (vinyl alcohol)-tetraborate hydrogel systems using scavenger polyol sugars. Eur J Pharm Biopharm. 2008;69:1135–1146.
81. Preis M, Woertz C, Schneider K, et al. Design and evaluation of bilayered buccal film preparations for local administration of lidocaine hydrochloride. Eur J Pharm Biopharm. 2014;86:552–561.
82. Noven Pharmaceuticals. DentiPatch (lidocaine hydrochloride) FDA prescribing information. May 1999. Available from: https://www.drugs.com/pro/dentipatch.html.
83. Hersh EV, Houpt MI, Cooper SA, et al. Analgesic efficacy and safety of an intraoral lidocaine patch. J Am Dent Assoc. 1996;127:1626–1626.
84. Houpt MI, Heins P, Lamster I, et al. An evaluation of intraoral lidocaine patches in reducing needle-insertion pain. Compend Contin Educ Dent. 1997;18:309-10,312-4, 316.
85. Kreider KA, Stratmann RG, Milano M, et al. Reducing children's injection pain: lidocaine patches versus topical benzocaine gel. Pediatr Dent. 2001;23:19–23.
86. Wu SJ, Julliard K. Children's preference of benzocaine gel versus the lidocaine patch. Pediatr Dent. 2003;25:401–405.
87. Leopold A, Wilson S, Weaver JS, et al.. Pharmacokinetics of lidocaine delivered from a transmucosal patch in children. Anesth Prog. 2002;49:82–87.
88. Kohda Y, Kobayashi H, Baba Y, et al. Controlled release of lidocaine hydrochloride from buccal mucosa-adhesive films with solid dispersion. Int J Pharm. 1997;158:147–155.
89. Okamoto H, Taguchi H, Iida K, et al.. Development of polymer film dosage forms of lidocaine for buccal administration. I. Penetration rate and release rate. J Control Release. 2001;77:253–260.
90. Repka MA, Gutta K, Prodduturi S, et al. Characterization of cellulosic hot-melt extruded films containing lidocaine. Eur J Pharm Biopharm. 2005;59:189–196.
91. Abu-Huwaij R, Assaf S, Salem M, et al.. Mucoadhesive dosage form of lidocaine hydrochloride: I. Mucoadhesive and physicochemical characterization. Drug Dev Ind Pharm. 2007;33:855–864.
92. Abu-Huwaij R, Assaf S, Salem M, et al.. Potential mucoadhesive dosage form of lidocaine hydrochloride: II. In vitro and in vivo evaluation. Drug Dev Ind Pharm. 2007;33(4):437–448.
93. Padula C, Pozzetti L, Traversone V, et al. In vitro evaluation of mucoadhesive films for gingival administration of lidocaine. AAPS PharmSciTech. 2013;14:1279–1283.

94. Hoare T, Kohane D. Hydrogels in drug delivery: progress and challenges. Polymer. 2008;49:1993–2007.
95. Abdel-Hamid SM, Abdel-Hady SE, El-Shamy AA, et al.. Formulation of an antispasmodic drug as a topical local anesthetic. Int J Pharm. 2006;326:107–118.
96. Xu J, Strandman S, Zhu JXX, et al. Genipin-crosslinked catechol-chitosan mucoadhesive hydrogels for buccal drug delivery. Biomaterials. 2015;37:395–404.
97. Pignatello R, Basile L, Puglisi G. Chitosan glutamate hydrogels with local anesthetic activity for buccal application. Drug Deliv. 2009;16:176–181.
98. Hirsh M, Hirsh J, Trumbore M. Gelled periodontal anesthetic preparation. US20070232695 A1. 2007.
99. Lathwal G, Pandit IK, Gugnani N, et al.. Efficacy of different precooling agents and topical anesthetics on the pain perception during intraoral injection: a comparative clinical study. Int J Clin Pediatr Dent. 2015;8:119–122.
100. Harbert H. Topical ice: a precursor to palatal injections. J Endod. 1989;15:27–28.
101. Duncan JD, Reeves GW, Fitchie JG. Technique to diminish discomfort from the palatal injection. J Prosthet Dent. 1992;67:901–902.
102. Kosaraju A, Vandewalle KS. A comparison of a refrigerant and a topical anesthetic gel as preinjection anesthetics: a clinical evaluation. J Am Dent Assoc. 2009;140:68–72.
103. Aminabadi NA, Farahani RMZ. The effect of pre-cooling the injection site on pediatric pain perception during the administration of local anesthesia. J Contemp Dent Pract. 2009;10:43–50.
104. Ghaderi F, Banakar S, Rostami S. Effect of pre-cooling injection site on pain perception in pediatric dentistry: "a randomized clinical trial". Dent Res J. 2013;10:790–794.
105. Saxena P, Gupta SK, Newaskar V, et al.. Advances in dental local anesthesia techniques and devices: an update. Natl J Maxillofac Surg. 2013;4:19–24.
106. Nanitsos E, Vartuli R, Forte A, et al. The effect of vibration on pain during local anaesthesia injections. Aust Dent J. 2009;54:94–100.
107. Elbay M, Şermet Elbay Ü, Yıldırım S, et al. Comparison of injection pain caused by the DentalVibe Injection System versus a traditional syringe for inferior alveolar nerve block anaesthesia in paediatric patients. Eur J Paediatr Dent. 2015;16:123–128.
108. Wiswall AT, Bowles WR, Lunos S, et al. Palatal anesthesia: comparison of four techniques for decreasing injection discomfort. Northwest Dent. 2014;93:25–29.
109. Hu L, Silva SMC, Damaj BB, et al.. Transdermal and transbuccal drug delivery systems: enhancement using iontophoretic and chemical approaches. Int J Pharm. 2011;421:53–62.
110. Cubayachi C, Couto ROD, de Gaitani CM, et al. Needle-free buccal anesthesia using iontophoresis and amino amide salts combined in a mucoadhesive formulation. Colloids Surf B Biointerfaces. 2015;136:1193–1201.
111. Pierre MBR, Rossetti FC. Microneedle-based drug delivery systems for transdermal route. Curr Drug Targets. 2014;15:281–291.
112. Gill HS, Denson DD, Burris BA, et al.. Effect of microneedle design on pain in human volunteers. Clin J Pain. 2008;24:585–594.
113. Kochhar JS, Lim WXS, Zou S, et al. Microneedle integrated transdermal patch for fast onset and sustained delivery of lidocaine. Mol Pharm. 2013;10:4272–4280.
114. Nayak A, Das DB, Vladisavljević GT. Microneedle-assisted permeation of lidocaine carboxymethylcellulose with gelatine co-polymer hydrogel. Pharm Res. 2014;31:1170–1184.
115. Gill HS, Prausnitz MR. Coated microneedles for transdermal delivery. J Control Release. 2007;117:227–237.
116. Ma Y, Gill HS. Coating solid dispersions on microneedles via a molten dip-coating method: development and in vitro evaluation for transdermal delivery of a water-insoluble drug. J Pharm Sci. 2014;103:3621–3630.
117. Ma Y, Boese SE, Luo Z, et al. Drug coated microneedles for minimally-invasive treatment of oral carcinomas: development and in vitro evaluation. Biomed Microdevices. 2015;17:44.
118. Vickers ER, Punnia-Moorthy A. Pulpal anesthesia from an application of a eutectic topical anesthetic. Quintessence Int. 1993;24:547–551.
119. Vickers ER, Marzbani N, Gerzina TM, et al. Pharmacokinetics of EMLA cream 5% application to oral mucosa. Anesth Prog. 1997;44:32–37.
120. Munshi AK, Hegde AM, Latha R. Use of EMLA: is it an injection free alternative? J Clin Pediatr Dent. 2001;25:215–219.
121. Meechan JG, Donaldson D. The intraoral use of EMLA cream in children: a clinical investigation. ASDC J Dent Child. 1994;61:260–262.
122. Franz-Montan M, Ranali J, Ramacciato JC, et al. Ulceration of gingival mucosa after topical application of EMLA: report of four cases. Br Dent J. 2008;204:133–134.