Adhesive Tablet Effective for Treating Canker Sores in Humans

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ABSTRACT: A new mucoadhesive tablet, which releases natural active agents for pain reduction and rapid healing of canker sores, has been prepared and characterized. Adhesive tablets were prepared by compression molding of mixed powders of crosslinked polyacrylic acid and hydroxypropyl cellulose, absorbed with citrus oil and magnesium salt. The rate of tablet erosion and the rates of citrus oil and magnesium release were determined as well as the adhesiveness of the tablet using bovine gingival tissue and an Instron tensiometer. A clinical trial was conducted on 248 volunteers who had canker sores. Tablets adhere well to the mucosal tissue and gradually erode for 8 h releasing the citrus oil in a zero-order pattern whereas the magnesium is released during a period of 2 h. Both experimental and plain tablets were effective in reducing pain and decreasing healing time (p < 0.05) without adverse side effects. However, the tablets loaded with active agents were more effective. © 2004 Wiley-Liss, Inc. and the American Pharmacists Association J Pharm Sci 93:2927–2935, 2004

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INTRODUCTION

Canker sores are ulcers that affect mucous membranes in the mouth and usually develop on the inner cheeks, gums, lips, and occasionally, the tongue. They may occur singly as aphthous stomatitis lesions or in groups as recurrent aphthous stomatitis (RAS).1,2 The histopathology of the ulcerated lesions is similar to that which occurs under sites of acute inflammation of the skin.3 The canker sore is most painful during the first 3–4 days. The discomfort gradually diminishes and the sore heals in 10–14 days, usually without scarring.3–5 Before it becomes visible, the canker sore may produce a tingling or burning sensation. After 6–24 h, the ulcer appears as a small round depression 3–9 mm in diameter, surrounded by a red area of inflammation. It has been suggested that RAS is caused by viral or bacterial infections5 but this theory has not been supported by cultures, serology, cytology, or electron microscopy.6 Genetic factors seem to have a major role in patients with RAS because it is more frequently found in patients with a family history of the condition.6

Corticosteroids and other antiinflammatory agents7 are used to treat the inflammation associated with canker sores. These medications, which are supplied in a gel, cream, or paste formulation, are applied three to four times a day.8

The effect of essential oils on ulcers is known.9 Citrus reticulata (mandarin) peel, which possesses antibacterial activity, is used as a food biopreservative.10 Citrus oil is used because it has anesthetic and antimicrobial properties. Galati et al.11

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used hesperidin (a citrus flavonoid obtained from the solid residue of orange peel) as an antiinflammatory and analgesic drug. Citrus oil contains mainly monoterpenes including \( \text{d-limonene} \) (\( \sim 90\% \)), sabine (\( \sim 2.2\% \)), myrcene (2%), and gamma-terpinene (\( \sim 1\% \)).

Magnesium has mild anesthetic properties, and is often administrated alone or in addition to other drugs (usually intravenously). Adhesive dosage forms have been reported for buccal administration of drugs for many years. Suitable adhesive carriers include linear and partially crosslinked polymers. An ideal buccal device should be elastic, soft, and able to withstand breakage caused by stress from mouth activities. It must also possess bioadhesive properties to ensure that it is retained in the mouth for the required period of time.

Buccal delivery formulations containing hydroxypropyl cellulose (HPC) and carbopol have been in use for many years, with various ratios of the two polymers. A formulation with an HPC/carbopol 934 (CP) ratio of 1:2 was chosen for this study after conducting preliminary tests, which focused on the tablet’s adhesiveness, erosion time, and flexibility. This assay is not included in this article.

Whereas mucoadhesive delivery systems have been reported for several different drugs, there have been only a few reports about their use in the treatment of oral mucosal disorders such as canker sores. This article describes a simple method for preparing a mucoadhesive tablet, loaded with citrus essential oil and magnesium salts for treating canker sores in humans. The \textit{in vitro} release rate of the active ingredients and time of tablet erosion as well as a double-blind, placebo-controlled clinical study of these tablets are described.

**MATERIALS AND METHODS**

**Materials**

HPC, with average molecular weight of 1,150,000 Da and viscosity of 1500–3000 mPa/s (1% solution), was obtained from Hercules Co., Ltd. (Klucel HF, Wilmington, DE).

CP was obtained from Goodrich Co., Ltd. (Cleveland, OH). Citrus oil was obtained from Ginsol Co., Ltd. (Jerusalem, Israel). \( \text{d-limonene} \) was obtained from Sigma-Aldrich Co., Ltd. (Poole, Dorset, UK). Magnesium salt (\( \text{MgCl}_2 \)) was obtained from Dead Sea Works Ltd., Israel.

**Manufacture of Tablets**

HPC (23 mg) and CP (46.5 mg) were mixed with the magnesium salt (7 mg) and absorbed with citrus oil (3.5 mg), using a mortar and pestle. Tablets of 9-mm diameter, 2-mm thick, weighing 80 mg, were pressed by a laboratory Carver press (Carver Machine Works, Inc., Washington, NC), using a pressure of 5 ton/cm\(^2\) for 30 s. Tablets of the same size and weight, which did not contain citrus oil and the magnesium chloride, were prepared by compression molding of the polymer powder.

**In Vitro Tablet Water Absorption and Erosion**

Tablets were stuck to the bottom of 20-mL vials (one tablet per vial) containing 6 mL 0.1 M phosphate buffer, pH 6.5 (8.88 g \( \text{NaH}_2\text{PO}_4 \cdot \text{H}_2\text{O} \), 12.71 g \( \text{Na}_2\text{HPO}_4 \), and deionized water made up to 1000 mL). The vials were kept at 37\(^\circ\)C with constant shaking (75 rpm). At each time point, three experimental vials with the tablet were removed from the study. The tablets were weighed after being surface dried with filter paper. Tablets were then totally dried at 60\(^\circ\)C overnight to determine the net weight at each time point.

**In Vitro Release of Citrus Oil and Magnesium**

Tablets were attached to the bottom of 20-mL vials (one tablet per vial) and 4 mL of phosphate buffer (simulating gingival fluid) was added to each vial followed by 3 mL of hexane. The vials were incubated at 37\(^\circ\)C with constant shaking of 75 rpm. At each time point, the upper hexane layer and the buffer were replaced with fresh hexane and buffer. The optical density of the main ingredient of citrus oil, \( \text{d-limonene} \) (\( \sim 90\% \)) in hexane was determined by ultraviolet spectroscopy (Ultrospec 2100 Pro; Biochrom, Cambridge, UK) at a wavelength of 254 nm. Pure \( \text{d-limonene} \) showed good correlation with citrus oil absorbance levels. Assays for magnesium release were performed using standard clinical laboratory procedures for analysis of minerals in blood and urine. Tests were performed using a 950 Vitros analyzer (Ortho-Clinical Diagnostic; Johnson and Johnson, Rochester, NY). The analyzer uses dry chemistry slide technology and a reflection spectrophotometer. A known concentration of magnesium in phosphate buffer was used as a control. Each experiment was repeated three times and the mean result calculated. The result was the mean of three experiments.
In Vitro Determination of Bioadhesive Performance

The adhesiveness of the tablet when placed onto mucosal tissue was determined by using an Instron universal testing machine (model 4502) controlled by a model 4500 computer assist module (Canton, MA). Bovine gingival tissue was received fresh from a local abattoir. After careful removal of adherent fat, 2 x 2 cm tissue pieces were prepared. Each tablet was attached onto the flat surface of the moving crimp, by using butyl cyanoacrylate adhesive. Gingival tissue specimens were placed onto the flat surface of the lower, stationary crimp with the mucosal side facing the tablet and attached by a frame connected to the surface with four screws (see Fig. 1). The tablet was lowered at a rate of 0.1 mm s⁻¹ until contact with the buccal mucosa was obtained. A contact force of 0.5 N was maintained for 10, 30, 120, 300, or 600 s, after which the probe was withdrawn from the buccal membrane at a rate of 5 mm s⁻¹. The peak detachment force (Newton) was recorded as a function of extension diagram. The amount of energy (mJ) needed to break the bond between the tablet and mucosa was calculated from the detachment force and the displacement at maximal load. The results are an average of three independent measurements.

Clinical Study

The efficacy of the tablet was tested on 248 adult volunteers 10–75 years old, who had canker sores. Before the onset of the trails, a dentist assessed the patients on the basis of medical history, an oral examination, and general health. The volunteers were requested to maintain their usual diet and activities during the evaluation period. All participants were asked to fill in a questionnaire (Fig. 2) within 24 h after complete erosion of the tablet. Those having a one-time canker sore (with a frequency of less than once every month) were documented as cases of one-time aphthous ulcer. Over a period of 2 years, 94 patients with one-time aphthous and 154 patients with RAS were included in the study. A single tablet, with or without citrus oil and magnesium salt, was given to each patient in the double-blind trial. A dentist estimated the time of healing. Patients were also examined for possible damage to mucosal tissue during and after tablet dissolution.

Statistical Analysis

Statistical comparisons of the findings were made by one-way analysis of variance. Comparison of means was performed by the least significant difference test. Data analysis was performed using a statistical software package (Instat; GraphPad Software, San Diego, CA). The significance level was set at \( p < 0.05 \).

RESULTS AND DISCUSSION

In Vitro Water Absorption and Erosion of Tablets

The water absorption of the mucoadhesive tablets when immersed in phosphate buffer (pH 6.5) is shown in Figure 3A. Tablets (0.08 g) started swelling immediately after being immersed in the solution and continued swelling at a constant rate, reaching a maximum weight of approximately 1.65 g after 5 h. Then, a gradual decrease in the tablet weight was observed (about 0.54 g/h) followed by complete dissolution-erosion after 3 h. The net erosion rate of the tablet is shown in Figure 3B. This in vitro experiment shows that the tablet main erosion (75%) takes place only after maximal water absorption, when small fragments of the fully swollen gels erode from the tablet.

The HPC and CP hydrophilic components of the gel, together with the CP carboxylic acid groups, enable tablet hydration and mucoadhesion. The crosslinked polyacrylic acid gel, CP, prevents the tablet from dissolving but allows the gel to absorb water and gradually erode.²⁷
In Vitro Release of Citrus Oil and Magnesium

In vitro release of citrus oil (Fig. 4) was estimated by measuring the d-limonene content. Tablets remained attached to the bottom of the vial for the duration of the study although some pieces of gel detached from the eroding tablet during the last hour of the experiment. Thirty minutes from the onset of the experiment, the citrus oil was released from the tablet at a constant rate. From 5–6 h, the rate of release declined with tablet erosion until no citrus oil remained (8 h after the beginning of the experiment). The release rate of citrus oil from the tablet is controlled by a combination of two concurrent processes: oil diffusion and erosion of the tablet’s swollen polymer matrix. The diffusion is apparently the main factor influencing citrus oil release in the first 5 h and erosion is the main factor in the following 3 h.

Divalent cations such as Mg$^{+2}$ form salts with carboxylic acid groups of the CP, and form bridges between polymer chains, which might affect the release rate.$^{28}$ A similar pattern of release was found for both tablets with or without magnesium.
Magnesium was included in the tablet because of its anesthetic properties. The mean cumulative release of the magnesium ions is shown in Figure 4. After 1 h, 66% of the Mg$^{2+}$ ions was released into the medium, with complete release after 2 h. This pattern of release is due to the high solubility of magnesium ions in the aqueous solutions which diffuse from the hydrophilic gel into the surrounding medium. The release of hydrophobic and hydrophilic agents from HPC/CP tablets has been reported by Han et al. They examined the release rate of nalbuphine prodrugs from buccal tablets made of HPC and CP. When a hydrophilic prodrug was used, the release rate increased with the increase in drug loading. In contrast, the release rate of a hydrophobic prodrug was significantly slower and was inversely related to the drug loading.

### Adherence to Mucosal Tissue

The data demonstrating the adhesiveness of the tablets to mucosal tissue are presented in Table 1. Attachment of the tablets to the gingiva for 10 s is sufficient to produce a tensile force of 0.25 N (~25 g). The detachment force increased during the first 300 s to 2.5 N (~250 g), and reached a plateau. The energy needed for detachment of the tablet from the mucosal tissue was calculated

![Figure 3](image)

**Figure 3.** Water absorption and erosion of tablet. (A) Tablets were placed in phosphate buffer (pH 6.5) at 37°C and tablet weight gain was determined gravimetrically. (B) Tablets were placed in phosphate buffer (pH 6.5) at 37°C and at each time point the tablet was surface dried and weighed.

![Figure 4](image)

**Figure 4.** Release (%) of citrus oil and magnesium ions from the adhesive tablet. Tablets were placed in phosphate buffer (pH 6.5) at 37°C, n = 3. (○) Release of citrus oil from tablets containing magnesium salt. (□) Release of citrus oil from tablets without magnesium salt. (△) Release of magnesium ions from tablets loaded with citrus oil and magnesium salt.

### Table 1. Adhesiveness of the Tablet When Placed on Mucosal Tissue

<table>
<thead>
<tr>
<th>Time from Attachment (s)</th>
<th>Detachment Force (N)</th>
<th>Displacement at Maximal Load (mm)</th>
<th>Adhesion Energy (mJ)</th>
</tr>
</thead>
<tbody>
<tr>
<td>10</td>
<td>0.3 (0.0)</td>
<td>0.4 (0.1)</td>
<td>1.5 (0.6)</td>
</tr>
<tr>
<td>30</td>
<td>1.5 (0.1)</td>
<td>1.1 (0.3)</td>
<td>1.9 (0.8)</td>
</tr>
<tr>
<td>120</td>
<td>2.1 (0.4)</td>
<td>2.8 (0.5)</td>
<td>2.9 (1.4)</td>
</tr>
<tr>
<td>300</td>
<td>2.7 (0.1)</td>
<td>3.2 (0.7)</td>
<td>3.2 (1.8)</td>
</tr>
<tr>
<td>600</td>
<td>2.6 (0.1)</td>
<td>3.4 (0.7)</td>
<td>6.2 (1.2)</td>
</tr>
</tbody>
</table>

The adhesiveness of the tablet was determined using an Instron universal testing machine. Values shown are the mean of three independent experiments ± standard deviation.
from the area under the stress–strain curve of the output. This energy increases from $1.5 \pm 0.6 \text{ mJ}$ after $10 \text{ s}$ of adhesion, to $6.2 \pm 1.2 \text{ mJ}$ after $600 \text{ s}$. CP is a slightly crosslinked polyacrylic acid, capable of forming hydrogen bonds with mucosal components, resulting in good mucoadhesion. The mucoadhesive properties of CP have been extensively reported for mucoadhesive liposomes and interpolymer complex systems.\textsuperscript{30,31}

The patient response (see Clinical Study) indicated that in most cases, when a tablet adheres to the mucosa during the first minute of application, it will remain for the rest of the treatment. Intimate molecular contact is a prerequisite for the development of strong adhesive bonds, where wetting equilibrium and the dynamic behavior of the bioadhesive polymeric material with the mucus is critical. Interfacial forces are responsible for the adhesive strength.\textsuperscript{32}

**Clinical Study**

A total of 94 patients with a single, painful aphthous ulcer and 154 patients with RAS were treated. The efficacy of treatment with the tablet was documented, as well as patient satisfaction. Two types of tablets were used: tablets loaded with citrus oil and magnesium salt and plain tablets without active additives. The data for untreated sores were taken from each patient before treatment with the tablet. The questionnaire, which was given to all patients, is shown in Figure 2.

Aphthous stomatitis (one-time aphthous) untreated patients reported that there was a significant level of pain that lasted for an average of $70 \pm 15 \text{ h}$ from the canker sore appearance (Fig. 5A). The level of pain was significantly reduced among the patients who were treated with either the plain tablet or the tablet loaded with citrus oil and magnesium salts. The pain disappeared within $8 \text{ h}$ for patients treated with plain tablets, and within $2 \text{ h}$ for those treated with tablets loaded with the active ingredients. In both groups of patients who received tablets, some patients reported pain relief within minutes. This reduction in pain suggests that the pain is mostly related to the exposure of ulcers to mouth fluids and mechanical stress. Thus, protection of the canker sore by physical sealing alone reduced the pain. The average time for healing was approximately $1.5$, $5$, and $9 \text{ days}$ for patients receiving experimental tablets, plain tablets, and for untreated patients, respectively ($p < 0.05$).

A similar experiment was performed for RAS patients. The mean time for pain disappearance and healing in RAS patients is shown in Figure 5B. There was a significant reduction in the time for pain disappearance and healing in patients treated with tablets containing active ingredients compared with the patients treated with plain tablets, and untreated patients ($p < 0.05$). The average time for pain disappearance was reduced from $134 \text{ h}$ for the untreated patients, to $48 \text{ h}$ for the control group treated with plain tablets, and to $5 \text{ h}$ in patients treated with the active tablets. The time for healing is similar to RAS and aphthous stomatitis patients. The time of healing was approximately $1.5$, $6$, and $10 \text{ days}$ for patients who were treated with loaded tablets, plain tablets, and for untreated patients, respectively ($p < 0.05$).

Figure 5. Clinical results of treatments of aphthous and RAS patients with citrus oil/magnesium salt adhesive tablets or plain tablets (double blind study, $p < 0.05$). (A) Time for pain relief and healing of aphthous ulcers. Data are the mean of results obtained from 94 patients. (B) Time for pain relief and healing of RAS. Data are the mean of results obtained from 154 patients. Significance differences between loaded tablet, plain tablet, and untreated groups ($p < 0.05$) are indicated by an asterisk.

Differences between the loaded tablets and plain tablets are shown when comparing pain
reduction among aphthous stomatitis and RAS patients. The plain tablet was found to be as effective as the experimental tablet among aphthous stomatitis patients but less effective for RAS patients. These findings may be related to the differences in etiology of the two conditions. In RAS, these ulcerations may be indicative of underlying systemic diseases ranging from vitamin deficiency to autoimmunity and less of trauma or infection. This could explain why antibacterial and antiinflammatory agents are less effective among these patients.

The tablet exhibited satisfactory adhesion to the gum tissue for >8 h. The general tolerance was good (>90%), with only minor local reactions at the site of application (slight crumpling of the mucosa). During the first 12 h, about 25% of the patients reported a sensation of local dryness. This was probably the result of a superficial mucosal dehydration by the tablet which may have been subsequently balanced by occlusion. Nevertheless, after 24 h, absolutely normal mucosal tissue was observed in all patients.

The importance of a long-lasting barrier to the aphthous canker was clearly demonstrated in the clinical trial. Pain was significantly reduced among both aphthous and RAS groups when using tablets without active ingredients. Sealing and protecting the canker sore from irritating oral fluids and mouth activity are crucial for pain reduction, as demonstrated by Nagai in 1985.

Colgate Oral Pharmaceuticals recently introduced a patient-applied topical medication, 2-octyl cyanoacrylate, which, on contact with oral soft tissue, polymerizes to form a protective barrier that lasts up to 6 h. The purpose of this medication is to create a long-lasting barrier that will reduce or eliminate pain associated with oral ulcerative disease. Results from a recently concluded multicenter trial demonstrated that this barrier significantly reduced pain associated with aphthous ulcerations. The rationale behind participation in a patient satisfaction study was to determine patient acceptance of the new medication for symptomatic control of pain associated with oral wounds. Yamamura et al. reported that a water-soluble three-layered oral mucoadhesive film made from HPC containing dibucaine (0.25 mg of drug/cm) induced pain relief that lasted for 2–5 h after application of the film.

CONCLUSIONS

A mucoadhesive tablet made from the common pharmaceutical polymers, hydroxypropyl cellulose (HPC-HF) and crosslinked poly(acrylic acid) (carbopol 934), which releases citrus oil and magnesium chloride was found to be highly effective for pain reduction and healing time in both single-ulcer and RAS patients. The effect on pain reduction by blocking the canker sore from the mouth environment was also demonstrated.
REFERENCES


