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Original Article

Herbal Mucoadhesive Buccal Films of Tinospora Cardifolia: Formulation and Characterization

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ABSTRACT

Background: Buccal drug delivery has gained significant attention due to its ease of administration, rich vascular supply, avoidance of hepatic first-pass metabolism, and improved patient compliance. The mucoadhesive buccal films are thin, flexible strips that adhere to the buccal mucosa and provide extended residence time for controlled drug release and improved therapeutic action. Tinospora cordifolia, a well-known ayurvedic medicinal plant, has been reported to possess antimicrobial, anti-inflammatory, antioxidant, and wound healing properties, which suggests its potential for the treatment of oral infections and ulcers.
Objective: The objective of the present investigation was to develop and evaluate mucoadhesive buccal films comprising Tinospora cordifolia extract for oral infections and ulcers.
Methods: Tinospora cordifolia extract was prepared using microwave-assisted extraction. Mucoadhesive buccal films were prepared by the solvent casting technique with hydroxypropyl methyl cellulose (HPMC E-15). The developed films were studied for physicochemical parameters, mechanical properties, functional parameters, and antimicrobial activity by agar well diffusion method.
Results: All the formulations were of uniform thickness and with good mechanical strength, near neutral surface pH, and good flexibility. Among the nine formulations, F3 exhibited characteristics such as the best physicochemical and mechanical properties, and controlled swelling behavior. The optimized formula showed a pronounced antimicrobial effect against the Staphylococcus aureus (mean zone of inhibition 19.0 ± 0.8 mm), indicating that the pharmaceutical activity of the herbal extract was preserved.
Conclusion: The prepared mucoadhesive buccal films of Tinospora cordifolia exhibited good film characteristics and significant antimicrobial activity, suggesting them to be a successful and safe herbal buccal drug delivery system for the treatment of oral infections.

Keywords: *Mucoadhesive buccal film, Tinospora cordifolia, Herbal drug delivery, Solvent casting, antimicrobial activity.*

INTRODUCTION

The oral cavity is a preferred route for drug delivery based on its accessibility, rich vasculature, and high patient compliance. In the numerous paths in the oral cavity, the buccal route has been found to be of extreme interest for local as well as systemic drug administration[1]. Buccal mucosa is the area between the upper gingiva and the cheek, which contains a perfused, relatively permeable lining that provides for effective drug absorption by passing directly into systemic circulation without being subjected to hepatic first-pass metabolism[2]. This property also renders buccal drug delivery particularly advantageous in enhancing the bioavailability of drugs that are excessively metabolised by first-pass metabolism following oral administration. Mucoadhesive buccal films are an innovative and patient-compliant dosage form formulated to attach to the buccal mucosa for prolong time period, achieving controlled and sustained release of the

drug[3]. These films have several advantages over traditional drug delivery systems, such as accurate dosing, rapid onset of action, improved drug bioavailability, reduced dosing frequency, and improved patient compliance. In addition, buccal films are thin, flexible, and non-invasive and cause minimal disturbance during speech or swallowing; they therefore can be used to treat chronic or acute conditions in the mouth. Oral ulcers are frequent lesions that can result from traumatic, nutritional deficit stress, or microbial infections, above all those produced by bacteria of the genus *Streptococcus*[4].

Traditional treatments normally involve topical gels, ointments, or mouth rinses, which are limited by short dwelling time at the target site, need for multiple dosing, and lack of patient compliance[5]. Thus, there is an increasing demand for a good local delivery system that can remain in close contact with the buccal mucosa to exert a continuous antimicrobial effect. In the recent past, medicinal plants have been explored by many researchers across the world due to their safety, cost-effectiveness, and low side effects when compared to synthetic drugs. *Tinospora cordifolia* (family: Menispermaceae), which is also called Guduchi, is a well-known, traditionally used medicinal plant in Ayurveda[6]. It has been pharmacologically shown to have antimicrobial, anti-inflammatory, immunomodulatory, antioxidant, and wound healing activities. The broad-spectrum antimicrobial activity of *Tinospora cordifolia* against several pathogens, including Streptococcus species, is suggestive of the potential to treat oral infections and ulcers[7].

Herbal extracts loaded mucoadhesive buccal film is an emerging area for site-specific drug delivery in the oral cavity. Appropriate selection of the polymers is necessary to dictate mechanical strength, mucoadhesive property, drug release kinetics, and overall performance of buccal film products[8]. Hydrophilic polymers such as hydroxypropyl methylcellulose (HPMC E15) and sodium carboxymethyl cellulose (SCMC) are extensively employed because of their good film-forming properties, biocompatibility, and mucoadhesion[9].

In the present study, an effort has been made to develop and evaluate mucoadhesive buccal films of *Tinospora cordifolia* extract using the solvent casting technique. The film-forming properties were optimized at various polymer conditions containing varying concentrations and combinations of polymers. The prepared films were assessed for their physicochemical, mechanical, and performance characteristics to get a suitable formula effective in treating oral ulcers induced by species of Streptococcus.

MATERIALS AND METHODS

Tinospora cordifolia (Willd.) Miers was freshly collected from Koregaon Park, Pune, Maharashtra, India, and authenticated before use. Hydroxypropyl methylcellulose (HPMC E15) and sodium starch glycolate were kindly supplied as gift samples by JRS Pharma, Mumbai, India. All other chemicals and reagents employed in the study were of analytical grade and were obtained from standard commercial suppliers.

METHODS

Preparation of *Tinospora cordifolia* Plant Extract

Fresh *Tinospora cordifolia* (stem and leaves) was obtained from Koregaon Park, Pune, Maharashtra, India. The plant material was washed several times with distilled water to remove adhering mud and extraneous matter. The extracted plant material was rinsed thrice with chloroform and shade dried for 7 days at room temperature. The dried product was milled in a mechanical grinder to obtain coarse powder. The powdered materials were then sifted through the no. 44 and no. 80 meshes in consecutive order to form a uniform particle size distribution[10].

Extraction Process

The microwave-assisted extraction (MAE) was used to extract the active compounds from *Tinospora cordifolia*. 20 g of the powdered plant materials was soaked in 300 mL of distilled water and left to stand at room temperature overnight. The next day, the dispersed solution was filtered, and the solid remaining on the filter paper was collected. The re-dissolved sediment was dissolved in 150 mL of deionized water and kept under aluminium foil to avoid evaporation. The mixture was reconstituted and extracted by microwave-assisted extraction at 240 W (corresponding to 35% of headspace power) during 20 min. After filtration, roughly 100 mL of filtrate containing the extracted phytoconstituents was obtained. The filtered extract was concentrated to give 1.36 g of dried material for subsequent formulation work[11].

Preparation of Various Concentrations of Drugs

A stock solution of *Tinospora cordifolia* extract was prepared by dissolving 1 g of powdered drug in 10 mL of ethanol to prepare 10% w/v. 1 mL of the stock solution was pipetted into a sterile test tube, which was designated as the 10% concentration of an extract. The resulting stock solution was diluted with ethanol into 7 different concentrations of the extract, that is; 2%, 3%, 4%,5%,6%,7% and 8 % w/v, respectively. All the solutions prepared were used for subsequent formulation studies.

Formulation of Mucoadhesive Buccal Films by Solvent-Casting Method

Mucoadhesive buccal films were prepared using the solvent casting technique, which is one of the simplest and most widely accepted methods for the preparation of buccal films. This method provides clearer films and a more constant film

thickness than extrusion-based methods[12]. In this method, the required quantity of polymer was dissolved in distilled water by stirring until a clear polymeric solution was obtained. Similarly, the active pharmaceutical ingredient (*Tinospora cordifolia* extract) and other additives were dissolved in a suitable solvent system, which remained part of their work. The two solutions were mixed, followed by stirring and heating in order to produce a homogeneous solution, designated as the "casting solution." A casting solution was cast in a suitable casting mould and dried under controlled conditions to release the solvent[13]. After completely drying, the resulting films were carefully peeled off and kept in a desiccator for further analysis (**Error! Reference source not found.**).

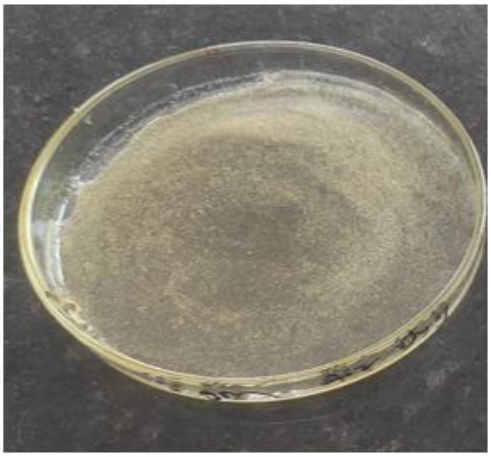

Figure 1.Prepared solution containing Tinospora cordifolia casted into a petri plate

Formulation of *Tinospora cordifolia* Extract-Loaded Mucoadhesive Buccal Films

The solvent casting method was used to prepare buccal mucoadhesive strips containing *Tinospora cordifolia* extract owing to its simplicity, cost, and ease of preparation. The schematic of the preparation process is displayed in Table 1. Various formulations were prepared from water-soluble polymers alone or in combination, as indicated in Table 1. Weighed amounts of HPMC E15 and SCMC-H (either alone or combined according to formulation design) were dissolved in 10 mL of distilled water initially. The polymer dispersion was left to stand for 15 minutes to cause the polymers to swell, and then it was continuously stirred at 300 rpm on a magnetic stirrer for 1 hour until a clear and homogeneous polymeric solution was obtained. Thereafter, glycerol was incorporated into the polymeric solution and mixed well. The specified amounts of *T. cordifolia* extract, sweetener (sucrose), citric acid, colorant, and flavoring agents (menthol oil) were dissolved in 10 mL of distilled water separately in another beaker. This solution was slowly dropwise added into the polymeric blend with constant stirring to ensure a homogeneous casting solution. The casted solution was poured on a pre-coated Petri plate and dried at room temperature for 24 h to ensure full evaporation of the solvent, with thin and even films. The films were then carefully peeled off the Petri plate after drying and tailored into specific sizes and shapes for further use[14].

Table 1.Composition of Various Batches of Tinospora cordifolia Mucoadhesive Buccal Films

Formulation ingredients	**F1**	**F2**	**F3**	**F4**	**F5**	**F6**	**F7**	**F8**	**F9**
T. cordifolia extract	635.85	635.85	635.85	635.85	635.85	635.85	635.85	635.85	635.85
HPMC E15 (mg)	100	250	500	-	-	-	400	350	250
SCMC-H (mg)	-	-	-	100	250	500	100	150	250
Glycerol (ml)	2	2	2	2	2	2	2	2	2
Sodium starch glycolate	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Citric acid (mg)	50	50	50	50	50	50	50	50	50
Sucrose (mg)	60	60	60	60	60	60	60	60	60
Peppermint oil	q.s	q.s	q.s	q.s	q.s	q.s	q.s	q.s	q.s
Water (ml)	20	20	20	20	20	20	20	20	20

Calculation of Drug Content per Batch

The amount of *Tinospora cordifolia* extract incorporated per batch was calculated based on the dose requirement per film using the following parameters:

- Dose of drug per film = 40 µL of 2% extract solution
- Area of one film = 4 cm^2
- Area of Petri plate = 63.585 cm^2

The drug content per batch was calculated using the formula:

$$\text{Drug per batch} = \frac{\text{Dose per film} \times \text{Area of Petri plate}}{\text{Area of one film}} = \frac{40 \times 63.585}{4} = 635.85\ \mu L$$

Thus, 635.85 µL of *Tinospora cordifolia* extract was incorporated per batch.

Evaluation of Mucoadhesive Buccal Films

Assessment is an important process in the manufacturing of oral thin films for comparing intra-batch and inter-batch uniformity, quality, and performance of the dosage form. The prepared mucoadhesive buccal films were evaluated for different physical and physicochemical parameters.

Weight Variation

Weight uniformity was tested and calculated in order to examine the consistency of the oral thin film formulations. Ten equal-sized pieces of each batch of formulated films were cut. The mass of each film was weighed using an analytical scale, and the average weight was determined[15]. The individual weight of each film from the mean weight was calculated using the equation:

$$\text{Weight variation (\%)} = \text{Average weight} - \text{Individual Weight} / \text{Average weight} \times 100$$

Thickness

The thickness of the oral thin films was determined for uniformity and ease of application. The thickness of each film was measured at various spots using a Vernier caliper and then averaged. Film thickness is a critical factor and affects mechanical strength, uniformity of drug content, and patient acceptance[16].

Determination of Surface pH

The surface pH of the oral thin films was measured for evaluating risk of irritation on the oral mucosa. A small film sample was cut and dissolved in distilled water under gentle shaking for 20 s. The pH value of the resultant dispersion was tested on a digital pH meter with calibration. The surface pH of the films was 7.30 ± 0.04, suggesting their potential for buccal application without causing mucosal irritation[17].

Moisture Uptake (Moisture Content)

Moisture is an important factor that affects the stability, flexibility, and friability of oral thin films. The moisture content of the films was determined using Karl Fischer titration. Firstly, the film was accurately weighed and then heated at 100~200°C for a predetermined time[18]. After heating, the final weight of the film was then determined, and the moisture content can be calculated according to:

$$\text{Moisture content (\%)} = \text{Initial Weight} - \text{Initial weight} / \text{Final weight} \times 100$$

The ideal moisture content of oral thin films should be less than 5% to ensure adequate mechanical strength and stability.

Tensile Strength

The breaking stress of a film is the maximum stress applied to it before rupture takes place. Tensile strength of the oral thin films was measured by a tensile testing apparatus with a 5-kg load. A strip of film was held clamped between two fixtures and elongated at an even velocity (mm/s) until rupture[19]. The tensile strength value was determined as the mass of the total weight attached to the string that was needed to tear the film.

Flexibility (Folding Endurance)

Flexibility of the oral thin films was characterized using the folding endurance parameter, which is an index of high importance regarding practical aspects during dispensing and handling. The film was folded repeatedly till it broke or upto 280 to 300 double folds without breaking at the same place by folding angle of 180°. The number of folds to which the film can be folded until break was measured as the folding endurance value[20].

Percentage Elongation

Percentage elongation represents the extent to which a film can stretch beyond its original length before breaking and indicates the elastic nature of the film[21]. The deformation-enabled elongation of the film under tensile stress was measured. The extension of the film was measured following tensile, along with the calculation of percentage elongation, which can be expressed with the following equation:

$$\text{Percent Elongation} = (L - L_o) \times 100 \div L_0$$

where

L_0= original length of the film

L= length of the film after elongation

Swelling Degree (Swelling Index)

The swelling index of the mucoadhesive buccal films was calculated by determining the percentage weight gain between initial and final weights[22]. The films were first dried and then weighed correctly (W 1). Thereafter, each film was submerged in de-ionized water for 2 minutes at room temperature. Excess surface water was gently blotted after removal, and the final weight (W2) was measured. The swelling ratio was determined by the following equation:

Swelling Index = $100\ (W_2\text{-}W_1) \div W_1$

where

W1=Initial weight and W2= final weight

Disintegration Test

The disintegration test was carried out with a conventional disintegration tester. A portion of the oral thin film was put into the basket of this apparatus, which performed at 30 up and down movements per minute. The test was continued until the film had completely dissolved, and no visible image of the film on top of the gauze could be seen[23]. This influx test aimed to monitor the amount and speed of swallow solution being formed from the unit.

Transparency

Opacity of films was evaluated spectrophotometrically using a UV-Vis spectrophotometer. The film was shaped of appropriate form and introduced into the cell of the spectrophotometer. Film transmittance was determined at a wavelength of 600 nm. Transparency was quantified with the equation:

$$\text{Transparency} = (\log T600)/B = -EC$$

where

T600= transmittance at 600nm and B = film thickness (mm)

Young's Modulus

Young's modulus, also known as the elastic modulus, was determined to assess the stiffness of the oral thin films. It was estimated from the tensile strength tests[24]. Young's modulus is the stress/strain ratio in the elastic deformation range and was calculated as follows:

$$\text{Young's Modulus} = \text{Slope} \times 100 \div \text{Film thickness} \times \text{cross head speed}$$

Hard and brittle film has high tensile strength and Young's modulus with small elongation.

Antimicrobial Study

Antimicrobial property of *Tinospora cordifolia* extract-loaded mucoadhesive buccal film was assessed for its efficacy at a particular concentration against the growth of microbes. Sterile nutrient agar medium was autoclaved at 120°C for 20 min. All the glassware used in the study were sterilized by a hot air oven at 160°C for 1 hr[25].

Sterile nutrient agar was aseptically transferred into sterile petri plates and allowed to solidify. After the media was solidified, 0.1 mL of the bacterial culture was spread on the agar using a spread plate method under aseptic conditions. Holes were aseptically bored into the agar wells using a sterile cork borer and filled with the developed buccal film samples. After 24 h, the plates were incubated at 37°C in a CO2 incubator[25]. The antimicrobial activity of the formulations was determined by measuring the width of inhibition zones around each well after incubation. All manipulations were performed in the aseptic chamber to avoid contamination.

Statistical Analysis

All experimental data were recorded and processed using Microsoft Excel. The results were expressed as mean ± standard deviation (SD). Each experiment was performed in triplicate (n = 3), and descriptive statistical analysis was used to evaluate the formulation parameters.

RESULTS

Evaluation of Mucoadhesive Buccal Film

The formulated mucoadhesive buccal films were investigated for various physicochemical, mechanical, and functional attributes. All experiments were conducted in triplicate (n = 3), and the data are shown as mean ± SD. The evaluation parameters of the optimized formulations F3 and F7 are tabulated in (Table 2). Both types rendered films with uniform consistency and with acceptable peelability and flexibility. The formulation F3 appeared smooth and white, whereas the formulation F7 presented a little bit bubbly with an off-white colour. The films from formulation F3 were clear, and those of formulation F7 were slightly translucent. The average thickness of F3 and F7 was 0.221 ± 0.006 mm and 0.268 ± 0.009 mm, respectively. Weight: The average weights of F3 and F7 were found to be 59.0 ± 1.0 mg and 65.0 ± 1.0 mg, respectively. The pH of the films was around neutral, and formulation F3 (pH = 7.20 ± 0.04) performed a lower value than that obtained for formulation F7 (pH = 7.80 ± 0.06). The folding endurance for F3 and F7 was 225 ± 5 and 198 ± 5, respectively. The swelling index of the F3 formula was found to be 41.4 ± 1.1% and of the F7 formula 39.6 ± 0.9%. The disintegration time of the films was 35 ± 2 s for F3 and 19 ± 1 s for F7.

Table 2Evaluation of Optimized Mucoadhesive Buccal Films (Triplicates and Mean ± SD, n = 3)

Parameter	Formulation	Replicate 1	Replicate 2	Replicate 3	Mean ± SD
Thickness (mm)	F3	0.215	0.222	0.226	0.221 ± 0.006
	F7	0.259	0.270	0.275	0.268 ± 0.009
Weight (mg)	F3	58	60	59	59.0 ± 1.0
	F7	64	66	65	65.0 ± 1.0
Surface pH	F3	7.15	7.22	7.23	7.20 ± 0.04
	F7	7.74	7.80	7.86	7.80 ± 0.06
Folding endurance (no. of folds)	F3	219	227	229	225 ± 5
	F7	193	198	203	198 ± 5
Swelling index (%)	F3	40.2	41.7	42.3	41.4 ± 1.1
	F7	38.8	39.4	40.6	39.6 ± 0.9
Disintegration time (s)	F3	33	36	37	35 ± 2
	F7	18	19	20	19 ± 1
Zone of inhibition (mm)	F3	18.2	19.1	19.7	19.0 ± 0.8

Antimicrobial Activity

The antimicrobial activity of the optimized formulation was evaluated against *Staphylococcus aureus* using the agar well diffusion method. The study was performed in triplicate, and the zone of inhibition was measured after incubation at 37°C (**Error! Reference source not found.**). Formulation F3 showed a mean zone of inhibition of 19.0 ± 0.8 mm.

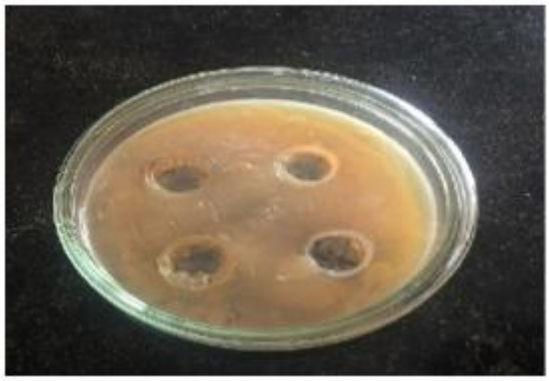

Figure 2.Microbial testing of drug containing film

DISCUSSION

In the current work, mucoadhesive buccal films were prepared and evaluated for *Tinospora cordifolia* extract by the solvent casting method. From the nine formulations formulated, F3 and F7 were chosen considering the initial aspects of peelability, homogeneity, and consistency shown by M films that suggested an adequate potential of film formation from the selected polymeric systems. Both the formulations yielded uniform films, with differences in physical appearance owing to polymer composition. Very smooth and transparent films were obtained in the case of formulation F3; on the contrary filmy formed using formulation F7 were slightly bubbling and less transparent, which can be due to high viscosity and air entrapped during solvent evaporation, affected by viscous blending of polymer. Polymer content and composition affected the thickness and weight of the films. Formulation F3 had a reduced thickness while being lighter in weight than formulation F7, which was consistent with even evaporation of solvent and effective formation of polymer matrix. Both formulations showed close to neutral surface pH (~ 6.0), which indicated that they were suitable for the buccal mucosa and had low irritation upon administration. The folding endurance values suggested that films possessed good mechanical strength and flexibility, with formulation F3 exhibiting a higher resistance to multiple folds, reflecting increased film integrity in addition to elasticity. These characteristics are important for manageability and patient compliance. The swelling of the mucoadhesive buccal films indicated that formulations based on F1 and F2 were properly hydrated. Higher swelling index of formulation F3 (compared to F2) may favor the mucoadhesive interaction due to better contact with the buccal mucosa. Disintegration studies revealed rapid disintegration of formulation F7 as compared to F3 due to the presence of blends of polymers and disintegrating agents, leading to the formation of porous characteristics for faster water uptake and matrix erosion. These variations in disintegration behavior indicate the effect of polymer composition on film functionality. Mechanical testing also confirmed the favorability of the fabricated films, with formulation F3 displaying a good balance between tensile strength and flexibility, while on the other hand, formulation F7 presented higher stiffness. The antimicrobial assay demonstrated retention of biological activity of the *Tinospora cordifolia* extract following incorporation into the buccal film matrix. The presence of a zone of inhibition against *S. aureus* for formulation F3 clearly demonstrates the antibacterial activity of the prepared film. This antimicrobial activity may be due to the bioactive

phytoconstituents like alkaloids, diterpenoids, and glycosides present in T. cordifolia.
In general, the results suggested that the concentration and composition of the polymer were a crucial factor in determining the physicochemical, mechanical, and functional properties of mucoadhesive buccal films. According to the overall evaluation parameters and antimicrobial activity, F3 was selected as the optimized formulation with good film characteristics, buccal compatibility, and biological activity.

CONCLUSION

The study formulated the mucoadhesive buccal films with *Tinospora cordifolia* extract by a simple and cost-effective method of solvent casting done at ambient temperature. Out of the nine formulations, F3 showed better physicochemical properties such as optimum thickness and controlled swelling, indicating good compatibility with buccal mucosa. Antimicrobial studies showed that *T. cordifolia* preserved its antibacterial activity in the films even against S. aureus. These are promising buccal films for delivery, which offer advantages such as convenient administration and enhanced patient compliance. Additional in vivo investigations and stability tests would be warranted to verify the formulation's applicability for clinical purposes.

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