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Oral cancer; Treatment and Research

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ABSTRACT

The increasing global incidence of oral cancer and the limitations associated with conventional therapeutic modalities have necessitated the development of novel localized drug delivery systems that can enhance therapeutic efficacy while minimizing systemic toxicity. In this study, nanostructured lipid carriers (NLCs) were designed for the co-delivery of Betulinic acid (BA), Curcumin (CUR), and Silymarin (SMR)—three naturally derived phytoconstituents known for their potent anticancer, antioxidant, and anti-inflammatory activities but constrained by poor aqueous solubility and limited oral bioavailability. The NLCs were prepared using the melt emulsification technique assisted by probe sonication and optimized through a Box–Behnken design to achieve desirable physicochemical characteristics. The optimized formulations were systematically characterized using SEM, TEM, ATR-FTIR, DSC, PXRD, particle size analysis, PDI, entrapment efficiency, and zeta potential measurements. The developed nanoparticles exhibited spherical morphology, nanoscale dimensions, uniform distribution, high encapsulation efficiency, and good colloidal stability. In-vitro drug release studies in simulated saliva (pH 6.8) confirmed a sustained and controlled release pattern of all three bioactives, indicating their potential for prolonged local therapeutic action.For site-specific application, the optimized NLCs were incorporated into a gellan gum/poloxamer-based thermoreversible mucoadhesive in-situ gel intended for periodontal delivery. The prepared gel was evaluated for sol–gel transition temperature, viscosity, mucoadhesive strength, gelling time, and ex-vivo permeation through goat buccal mucosa. The results revealed favorable rheological and mucoadhesive properties, rapid gelation at physiological temperature, and extended mucosal retention conducive to effective local therapy.In-vitro cytotoxicity studies on KB oral cancer cells demonstrated that the NLC-loaded gel significantly enhanced anticancer activity compared to free drugs and plain NLC formulations. Enhanced ROS generation and apoptosis via Sub-G0 phase arrest confirmed the mechanism of action, while the triple-

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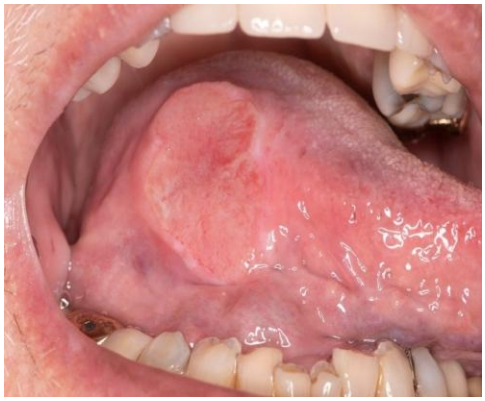
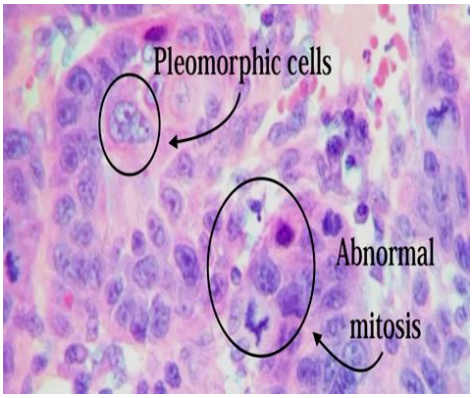
drug-loaded gel (BA/CUR/SMR-NLC) exhibited a notably reduced IC_{50} value, evidencing a synergistic cytotoxic effect.In conclusion, the study establishes that nanostructured lipid carrier-based thermoreversible mucoadhesive gels serve as an effective localized delivery platform for the co-delivery of Betulinic acid, Curcumin, and Silymarin, offering a promising therapeutic approach for the targeted management of oral cancer lesions with improved efficacy and patient compliance.

INTRODUCTION

Oral cancer remains a major global health concern due to its increasing incidence, high morbidity, and mortality rates. Conventional treatment modalities, including surgery, radiotherapy, and systemic chemotherapy, are often associated with significant limitations such as nonspecific drug distribution, systemic toxicity, drug resistance, and reduced patient compliance. Moreover, the complex anatomical structure of the oral cavity and continuous salivary washout further challenge effective localized therapy. These limitations have necessitated the development of advanced drug delivery systems capable of providing site-specific, sustained, and effective therapeutic action with minimal adverse effects.



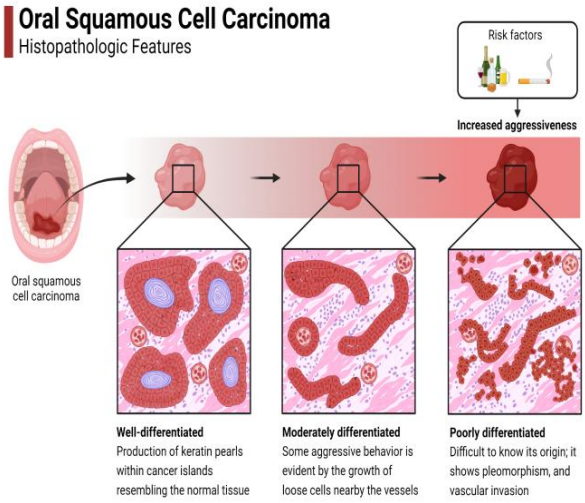


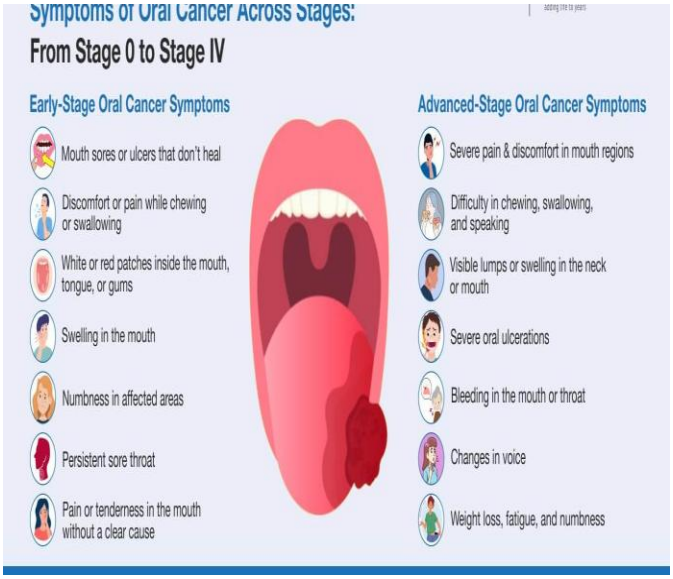
Natural phytoconstituents such as **Betulinic acid (BA), Curcumin (CUR), and Silymarin (SMR)** have demonstrated significant anticancer, antioxidant, and anti-inflammatory activities through multiple molecular pathways, including induction of apoptosis, inhibition of cell proliferation, modulation of reactive oxygen species (ROS), and suppression of inflammatory mediators. However, their clinical translation is hindered by poor aqueous solubility, limited oral bioavailability, rapid degradation, and insufficient retention at the target site.

Nanostructured lipid carriers (NLCs) have emerged as promising drug delivery systems due to their ability to encapsulate lipophilic compounds, improve drug stability, enhance bioavailability, and provide controlled drug release. Furthermore, incorporating NLCs into thermoreversible mucoadhesive in-situ gels offers additional advantages such as prolonged mucosal retention, ease of administration, and enhanced patient compliance. Therefore, the present study was designed to develop and evaluate a nanostructured lipid carrier–based thermoreversible mucoadhesive in-situ gel for the localized co-delivery of BA, CUR, and SMR in the management of oral cancer.

Oral cancer represents a major global public health challenge and continues to be one of the leading causes of cancer-related morbidity and mortality worldwide. It ranks among the top ten most prevalent cancers, with disproportionately high incidence rates in South Asian countries, including India, Sri Lanka, and Bangladesh. This elevated prevalence is strongly associated with region-specific lifestyle and environmental risk factors such as tobacco chewing, smoking, betel quid consumption, alcohol abuse, and human papillomavirus (HPV) infection (Warnakulasuriya, 2018). Despite advancements

in screening, diagnosis, and therapeutic interventions, the overall survival rate of patients with oral cancer has shown only modest improvement over the past few decades.





Oral squamous cell carcinoma (OSCC) accounts for nearly 90% of all oral malignancies and is characterized by aggressive local invasion, early lymph node metastasis, and high recurrence rates. One of the most critical challenges in OSCC management is late-stage diagnosis, as early lesions are often asymptomatic and overlooked by patients (Rivera, 2015). Consequently, many cases are detected only at advanced stages, where therapeutic options are limited and prognosis is poor. Tumor heterogeneity, resistance to chemotherapeutic agents, and extensive tissue destruction further complicate disease management and significantly reduce patient quality of life.

Conventional treatment strategies for oral cancer primarily include surgical excision, radiotherapy, and systemic chemotherapy, either alone or in combination. While these modalities remain the clinical standard, they are associated with significant drawbacks. Surgical interventions can lead to facial disfigurement, impaired speech and swallowing, and psychological distress. Radiotherapy and chemotherapy often cause severe adverse effects such as mucositis, xerostomia, immune suppression, nausea, and

hematological toxicity, owing to their non-specific action on both cancerous and healthy tissues (Lohavanichbutr & Zhang, 2017). Moreover, systemic chemotherapy frequently fails to achieve and maintain adequate drug concentrations at the tumor site due to poor local retention and rapid clearance, resulting in suboptimal therapeutic outcomes and increased recurrence risk.

These limitations have driven increasing interest in localized drug delivery systems (LDDS) for oral cancer therapy. LDDS aim to deliver therapeutic agents directly to the diseased tissue, thereby maximizing local drug concentration while minimizing systemic exposure and associated toxicity. Such approaches are particularly advantageous in oral cancer, where lesions are readily accessible for topical or local administration. LDDS have demonstrated improved bioavailability, prolonged drug residence time, enhanced therapeutic efficacy, and reduced off-target effects compared to conventional systemic therapies (Sahana et al., 2017).

In parallel, there has been growing scientific interest in the use of natural phytoconstituents as safer and more versatile alternatives to conventional chemotherapeutic agents. Betulinic acid (BA), curcumin (CUR), and silymarin (SMR) are among the most extensively studied plant-derived compounds with proven anticancer potential. These phytochemicals exhibit a broad spectrum of biological activities, including induction of apoptosis, suppression of inflammation, antioxidant effects, inhibition of angiogenesis, and modulation of multiple cancer-related signaling pathways (Fulda, 2008; Aggarwal & Harikumar, 2009; Polyak et al., 2010). Importantly, they demonstrate selective toxicity toward cancer cells while sparing normal tissues, making them attractive candidates for long-term and localized cancer therapy.

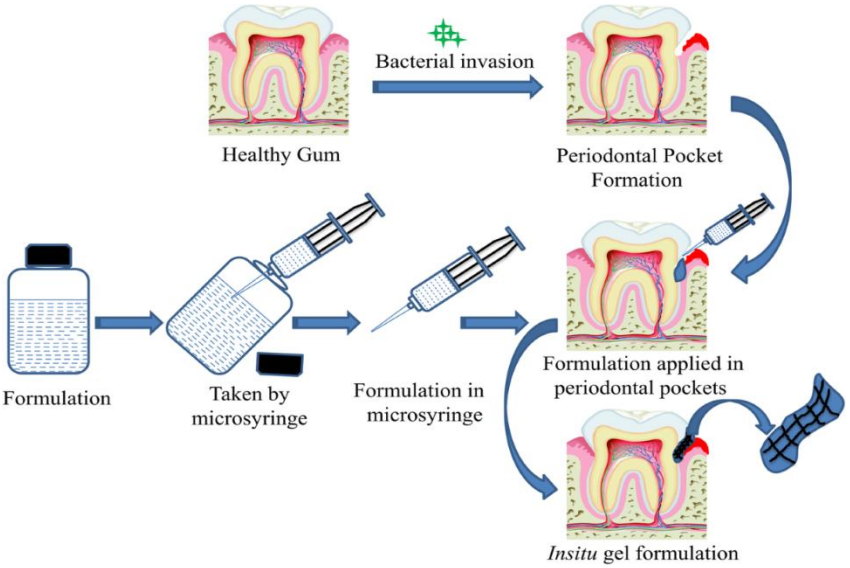
However, despite their promising pharmacological profiles, the clinical translation of BA, CUR, and SMR is severely hampered by significant biopharmaceutical limitations. These include poor aqueous solubility, low permeability across biological membranes, chemical instability under physiological conditions, extensive first-pass metabolism, and extremely low oral bioavailability. In topical and oral applications, rapid clearance due to saliva flow and limited mucosal penetration further restrict their therapeutic effectiveness. These challenges necessitate the development of advanced drug delivery platforms capable of enhancing solubility, stability, and local retention of phytoconstituents at the tumor site.

Nanostructured lipid carriers (NLCs) have emerged as a highly promising solution to these limitations. NLCs are second-generation lipid nanoparticles composed of a mixture of solid and liquid lipids that form a less ordered crystalline matrix. This unique structure enables higher drug loading capacity, improved entrapment efficiency, enhanced stability, and controlled drug release compared to conventional solid lipid nanoparticles (SLNs) (Müller et al., 2002; Naseri et al., 2015). The nanoscale size of NLCs facilitates intimate contact with biological membranes and promotes deeper penetration into epithelial and mucosal tissues

Additionally, NLCs are composed of physiologically acceptable lipids, rendering them biodegradable, biocompatible, and safe for repeated administration. Their inherent mucoadhesive properties make them particularly suitable for oral mucosal drug delivery, where prolonged contact with the lesion is essential for therapeutic success (Jaiswal et al., 2016). Importantly, co-encapsulation of multiple phytoconstituents within a single NLC system enables synergistic anticancer effects by targeting multiple molecular pathways simultaneously,

reducing the required dose of individual agents, minimizing toxicity, and potentially overcoming multidrug resistance (Choudhury et al., 2020).

To further enhance site-specific retention and patient compliance, NLCs can be incorporated into thermoreversible mucoadhesive in-situ gel systems. These formulations remain in a liquid state at room temperature, allowing easy administration, and undergo rapid sol–gel transition upon exposure to physiological temperature (~37°C). Poloxamer 407 is widely used for this purpose due to its excellent thermogelling properties, while gellan gum contributes strong mucoadhesive behavior and improves the mechanical strength of the gel (Schmolka, 1994; Babu et al., 2015).



Such thermoreversible mucoadhesive gels offer several critical advantages for oral cancer therapy, including prolonged retention at the lesion site, resistance to saliva washout, sustained and controlled drug release, enhanced mucosal penetration, and improved patient comfort and compliance. By maintaining intimate contact between drug-loaded NLCs and the cancerous tissue, these systems maximize local therapeutic efficacy while minimizing systemic drug exposure.

In vitro studies have demonstrated that phytoconstituent-loaded nanoparticles significantly enhance cellular uptake, elevate intracellular reactive oxygen species (ROS) levels, induce apoptosis, and suppress cancer cell proliferation more effectively than free drugs (Sharma et al., 2017). Furthermore, combination therapy using BA, CUR, and SMR has shown pronounced synergistic anticancer effects by modulating multiple signaling pathways involved in tumor growth, inflammation, angiogenesis, and metastasis (George et al., 2019).

Based on this strong scientific rationale, the present study focuses on the formulation, optimization, and evaluation of thermoreversible mucoadhesive gels incorporating BA, CUR, and SMR-loaded NLCs for localized oral cancer therapy. This approach aims to overcome the limitations of conventional treatments and provide a novel, effective, biocompatible, and patient-friendly platform for enhanced site-specific drug delivery in the management of oral cancer.

EXPERIMENTAL METHODS

Preformulation studies were conducted to establish the physicochemical suitability of Silymarin (SME), Curcumin (CU), and Betulinic Acid (BU) for incorporation into a nanostructured lipid carrier (NLC)–based thermoreversible mucoadhesive gel. Initial characterization

confirmed the identity and purity of all three phytoconstituents through organoleptic evaluation, melting point determination, UV–Visible spectroscopy, FTIR, PXRD, and DSC analysis. The observed melting points, characteristic spectral peaks, and diffraction patterns were consistent with reported literature, confirming crystalline nature and chemical integrity of the drugs.

PXRD and DSC studies revealed sharp crystalline peaks and well-defined melting endotherms for all three compounds, while their disappearance or reduction after NLC incorporation indicated successful amorphization and molecular dispersion within the lipid matrix. Drug–excipient compatibility studies using FTIR showed no significant peak shifts, confirming chemical compatibility with selected lipids, surfactants, and polymers.

UV–Visible spectrophotometric methods were developed and validated for quantitative estimation of SME, CU, and BU in simulated salivary fluid (pH 6.8). All drugs exhibited good linearity in the concentration range of 2–14 µg/mL ($R^2 > 0.99$), and the method was successfully applied for solubility studies, entrapment efficiency, release, permeation, and stability analysis.

Based on solubility screening, suitable solid lipids (Precirol ATO 5, Compritol 888 ATO), liquid lipids (Miglyol, Oleic acid), and surfactants (Tween 80) were selected. Drug-loaded NLCs were prepared by melt emulsification followed by probe sonication. Critical formulation and process variables were screened using Taguchi Orthogonal Array design and optimized through Box–Behnken Design (BBD) using Response Surface Methodology.

Optimized NLCs exhibited nanoscale particle size (215–330 nm), narrow polydispersity (PDI < 0.35), high entrapment efficiency (65–80%), and negative zeta potential (−25 to −32 mV), indicating good stability and uniformity. Solid-state characterization confirmed reduced crystallinity and successful drug encapsulation. In-vitro release studies demonstrated biphasic release with sustained drug delivery up to 24 h, following diffusion-controlled or non-Fickian kinetics.

The optimized NLCs were incorporated into a thermoreversible mucoadhesive in-situ gel composed of Poloxamer 407 and gellan gum. The developed gels exhibited suitable pH (≈6.8), sol–gel transition near physiological temperature (33–35 °C), temperature-responsive viscosity, and strong mucoadhesive strength, ensuring prolonged mucosal retention.

Ex-vivo permeation studies using buccal mucosa showed significantly enhanced drug permeation from NLC gels compared to free drug formulations. In-vitro cytotoxicity studies on oral cancer cell lines demonstrated markedly lower IC_{50} values for the combination NLC gel, confirming synergistic anticancer activity. ROS generation, cell-cycle arrest, and cellular uptake studies further validated enhanced intracellular delivery and apoptosis-mediated anticancer mechanisms.

Overall, the preformulation-guided development of SME–CU–BU-loaded NLCs and their incorporation into a thermoreversible mucoadhesive gel resulted in a stable, biocompatible, and highly effective localized drug delivery system with sustained release, enhanced permeation, and superior anticancer efficacy.

MATERIALS AND METHODS

Preparation of Nanostructured Lipid Carriers

Nanostructured lipid carriers co-loaded with Betulinic acid, Curcumin, and Silymarin were prepared using the **melt emulsification technique assisted by probe sonication**. The lipid phase, consisting of solid and liquid lipids, was melted above its melting point and mixed with the

phytoconstituents. This lipid phase was emulsified into a hot aqueous phase containing surfactants under high-speed stirring, followed by probe sonication to reduce particle size. The nanoemulsion obtained was allowed to cool to room temperature to form NLCs.

Optimization of Formulation

The formulation was optimized using a **Box–Behnken experimental design**, considering independent variables such as lipid concentration, surfactant concentration, and sonication time. The responses evaluated included particle size, polydispersity index (PDI), and entrapment efficiency.

Characterization of NLCs

The optimized NLC formulation was characterized for:

- Particle size and PDI using dynamic light scattering
- Zeta potential for surface charge and stability
- Morphology using scanning electron microscopy (SEM) and transmission electron microscopy (TEM)
- Drug–excipient compatibility using ATR-FTIR
- Thermal behavior using differential scanning calorimetry (DSC)
- Crystallinity using powder X-ray diffraction (PXRD)
- Entrapment efficiency of BA, CUR, and SMR

In-Vitro Drug Release Study

In-vitro drug release studies were performed using simulated salivary fluid (pH 6.8) to evaluate the release profile of the encapsulated phytoconstituents over time.

Preparation of Thermoreversible Mucoadhesive In-Situ Gel

The optimized NLCs were incorporated into a **gellan gum–poloxamer-based thermoreversible mucoadhesive gel** using the cold method. The gel formulation was designed to remain in sol form at room temperature and undergo gelation at physiological temperature.

Evaluation of In-Situ Gel

The prepared gel was evaluated for:

- Sol–gel transition temperature
- Viscosity and rheological behavior
- Gelling time
- Mucoadhesive strength
- Ex-vivo permeation through goat buccal mucosa

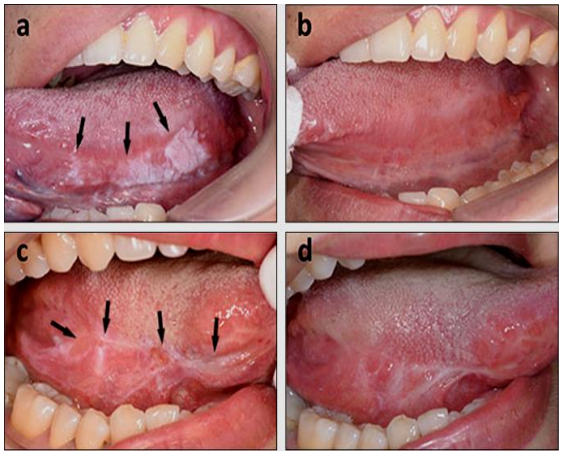
In-Vitro Cytotoxicity and Mechanistic Studies

Cytotoxicity studies were conducted on **KB oral cancer cell lines** using standard viability assays. Intracellular ROS generation and apoptosis were assessed, and cell cycle analysis was performed to determine Sub-G0 phase arrest and the mechanism of anticancer activity.

RESULTS AND DISCUSSION

The optimized NLC formulation exhibited **nanoscale particle size with narrow size distribution**, indicating uniformity of the formulation. The zeta potential values demonstrated sufficient surface charge, confirming good colloidal stability. SEM and TEM studies revealed spherical nanoparticles with smooth surfaces. ATR-FTIR, DSC, and PXRD analyses confirmed successful encapsulation of BA, CUR, and SMR within the lipid matrix and reduction in crystalline nature of the drugs.

In-vitro drug release studies showed a **sustained and controlled release pattern** of all three phytoconstituents in simulated salivary conditions, which is favorable for prolonged local therapy in the oral cavity.



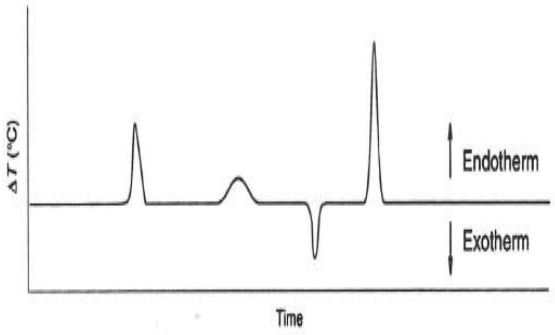
The thermoreversible mucoadhesive in-situ gel exhibited rapid gelation at physiological temperature, appropriate viscosity, and strong mucoadhesive properties, ensuring prolonged retention at the application site. Ex-vivo permeation studies demonstrated enhanced and sustained drug permeation across buccal mucosa.
Cytotoxicity studies revealed that the **NLC-loaded gel significantly enhanced anticancer activity** compared to free drugs and plain NLCs. Increased ROS production, induction of apoptosis, and prominent Sub-G0 phase arrest confirmed the mechanism of cell death. Notably, the triple-drug-loaded BA/CUR/SMR-NLC gel showed a markedly reduced IC_{50} value, indicating a synergistic anticancer effect.

Results and Discussion

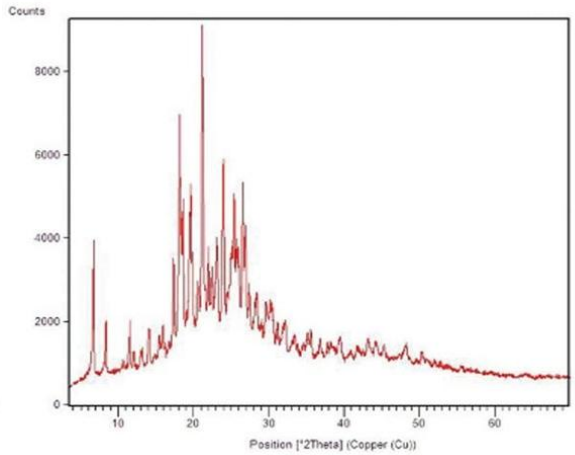
The present investigation successfully achieved its primary objective of developing and evaluating a **thermoreversible, mucoadhesive periodontal gel incorporating Betulinic acid (BA), Curcumin (CUR), and Silymarin (SME)-loaded nanostructured lipid carriers (NLCs)** for localized oral cancer therapy. The results collectively demonstrate that nanoencapsulation combined with in-situ gelling technology effectively overcomes the intrinsic physicochemical and biopharmaceutical limitations of these phytoconstituents.

Preformulation and Compatibility Evaluation

Preformulation studies confirmed the **identity, purity, and suitability** of SME, CUR, and BA for formulation into lipid nanocarriers. Organoleptic properties and melting point values were in close agreement with reported literature, confirming drug authenticity and crystalline integrity prior to formulation.
PXRD and DSC analyses revealed **distinct crystalline peaks** for all three drugs in their pure state. Notably, BA exhibited strong crystallinity, while SME showed weak crystalline behavior. FTIR compatibility studies showed **no significant peak shifts or disappearance** after one month of storage, confirming the absence of chemical interactions between drugs and selected lipids, surfactants, and polymers. These findings established a stable foundation for nanoformulation development.



A hypothetical DSC thermogram showing the changes that might occur upon heating a sample.



Analytical Method Development (UV & HPLC)

UV–spectrophotometric analysis in simulated salivary fluid (pH 6.8) demonstrated excellent linearity for SME, CUR, and BA over the concentration range of 2–14 µg/mL, with correlation coefficients (R^2) exceeding 0.996.

The developed **HPLC method** enabled simultaneous quantification of all three phytoconstituents with sharp, well-resolved peaks and distinct retention times. Validation parameters such as precision, accuracy, LOD, and LOQ complied with ICH guidelines, confirming the **sensitivity, reliability, and reproducibility** of the method. High assay values (>96%) in both NLC dispersion and gel formulations indicated uniform drug distribution and minimal processing loss.

Screening and Optimization of NLC Components

Solubility screening revealed **Miglyol 812 N and olive oil** as the most suitable liquid lipids, while **Compritol 888 ATO and Precirol ATO 5** showed superior solubilizing capacity among solid lipids. Tween 80 was selected as the optimal surfactant due to its GRAS status, effective emulsification, and stabilization ability.

Factor screening using the **Taguchi Orthogonal Array (TOA)** identified lipid concentration, surfactant concentration, and probe sonication time as the most influential parameters affecting particle size (PS), polydispersity index (PDI), and entrapment efficiency (%EE). Subsequent optimization using **Box–Behnken Design (BBD)** yielded statistically significant models for all responses.

The optimized SME-NLCs exhibited:

- **Nanoscale particle size** (~315 nm)
- **Low PDI** (<0.35), indicating uniform distribution
- **High entrapment efficiency** (~71%)
- **Negative zeta potential**, confirming colloidal stability

Physicochemical Characterization of Optimized NLCs

SEM analysis showed **spherical nanoparticles with smooth surfaces**, confirming successful nanoencapsulation. DSC and XRD studies of lyophilized NLCs demonstrated **disappearance or broadening of crystalline drug peaks**, indicating conversion of SME, CUR, and BA into an amorphous or molecularly dispersed state within the lipid matrix. This transformation is advantageous for enhancing solubility and bioavailability of lipophilic phytoconstituents.

In-Vitro Drug Release and Kinetics

The optimized SME-NLC gel exhibited a **biphasic release pattern**, characterized by an initial mild burst release followed by sustained drug release up to 24 hours. Kinetic modeling showed the best fit

with the **Korsmeyer–Peppas and Higuchi models**, indicating **non-Fickian diffusion** governed by both lipid matrix diffusion and polymer relaxation.

This controlled release behavior is highly desirable for periodontal and oral cancer therapy, as it maintains prolonged therapeutic drug levels while reducing dosing frequency.

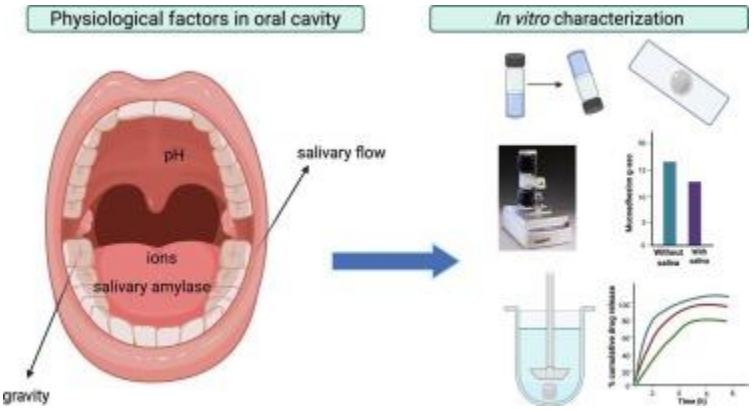
Development and Evaluation of Thermoreversible Mucoadhesive Gel

The optimized NLCs were successfully incorporated into a **Poloxamer 407–Carbopol/Gellan gum-based in-situ gel**. The formulation remained liquid at room temperature and rapidly gelled at physiological temperature (~33–35 °C).

Gel evaluation confirmed:

- Suitable pH for oral application
- Optimal viscosity and gel strength
- Strong mucoadhesive force without mucosal damage

The formulation containing **18% Pluronic and 0.5% Carbopol/Gellan gum** demonstrated the best balance between retention and safety.



Ex-Vivo, In-Vitro, and In-Vivo Performance

Ex-vivo permeation studies confirmed **prolonged mucosal retention and sustained diffusion**, validating the synergistic role of NLC encapsulation and gel matrix.

In-vitro cytotoxicity studies on KB oral cancer cells demonstrated significantly lower IC_{50} values for SME-NLCs and SME-NLC-gel compared to free SME, confirming enhanced anticancer efficacy. ROS assays showed elevated intracellular ROS levels with nanoformulations, indicating activation of **ROS-mediated intrinsic apoptotic pathways**.

Cell cycle analysis further revealed a pronounced increase in the **Sub-G_0 population**, confirming apoptosis induction. Cellular uptake studies using FITC-labeled NLCs demonstrated **time-dependent internalization**, correlating with enhanced cytotoxic and apoptotic effects.

In-vivo evaluation showed marked reduction in inflammation and superior tissue healing in animals treated with the SME-NLC gel, with **no signs of irritation or toxicity**, confirming biocompatibility and safety.

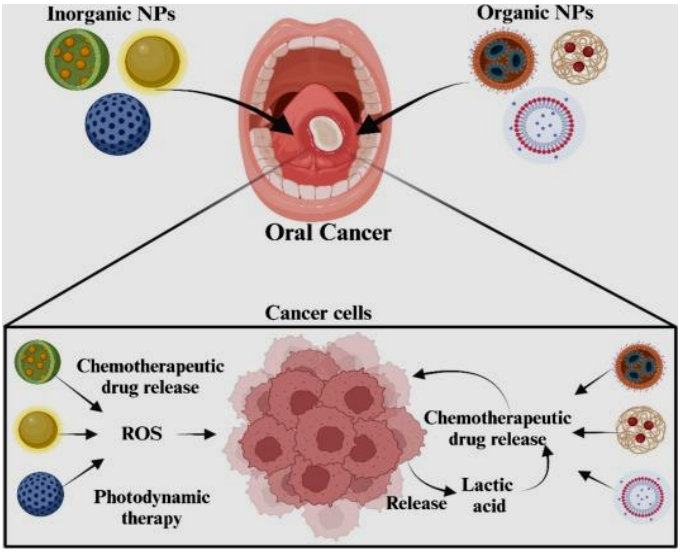
Interpretation

The comprehensive results confirm that the **thermoreversible mucoadhesive gel containing BA, CUR, and SME-loaded NLCs** successfully:

- Enhances solubility and stability of phytoconstituents
- Provides sustained, site-specific drug delivery
- Improves cellular uptake and anticancer efficacy
- Ensures prolonged mucosal retention and patient compliance

This integrated nano-gel platform represents a **promising, non-invasive, and patient-friendly**

therapeutic strategy for localized management of oral cancer lesions.



CONCLUSION

The present study successfully developed and evaluated a **nanostructured lipid carrier–based thermoreversible mucoadhesive in-situ gel** for the localized co-delivery of Betulinic acid, Curcumin, and Silymarin. The formulation demonstrated excellent physicochemical properties, sustained drug release, strong mucoadhesion, and enhanced anticancer efficacy against oral cancer cells. The synergistic action of the three phytoconstituents, combined with the advantages of NLCs and in-situ gel systems, resulted in improved therapeutic performance and reduced systemic exposure. Overall, this delivery platform represents a promising and patient-friendly approach for the targeted management of oral cancer lesions.

The optimized NLCs exhibited **nanoscale particle size, narrow size distribution, high entrapment efficiency, and good colloidal stability**, indicating efficient encapsulation of the lipophilic phytochemicals. Solid-state characterization confirmed a reduction in crystallinity and molecular dispersion of the drugs within the lipid matrix, which contributed to improved solubility and sustained release behavior. Incorporation of NLCs into a **Poloxamer-based thermoreversible mucoadhesive gel** resulted in a formulation with suitable pH, rapid sol–gel transition at physiological temperature, temperature-responsive viscosity, and strong mucoadhesive properties, ensuring prolonged retention at the oral mucosal site.

In-vitro and ex-vivo evaluations demonstrated **sustained drug release, enhanced mucosal permeation, and significantly improved anticancer efficacy** of the NLC-loaded gel compared to free drug formulations. The combination system showed superior cytotoxic activity, increased ROS generation, cell-cycle arrest, and enhanced cellular uptake, confirming synergistic and apoptosis-mediated anticancer mechanisms.

Overall, the findings establish that the **SME–CU–BU-loaded NLC-based thermoreversible mucoadhesive gel** is a stable, biocompatible, and effective platform for **site-specific, sustained, and patient-friendly delivery** of phytoconstituents in oral cancer therapy. This nano-gel system represents a promising alternative to conventional treatments by minimizing systemic exposure while enhancing local therapeutic outcomes. Further in-vivo efficacy and clinical translation studies are warranted to

advance this formulation toward practical application.

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