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Research Article

Determination of the Chemical and Morphological Structure of Liposomes Encapsulating Lycopene Derived from Tomatoes Using Fourier-Transform Infrared Spectroscopy (FTIR), Scanning Electron Microscopy (SEM), Transmission Electron Microscopy (TEM), and Ultrasound Imaging Techniques

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ABSTRACT

Lycopene, a natural carotenoid abundantly found in tomatoes, has been extensively studied for its antioxidant, anti-inflammatory, and anticancer properties. However, its poor aqueous solubility, instability under physiological conditions, and limited bioavailability hinder its clinical application. This study investigates the encapsulation of lycopene in liposomal carriers to improve its chemical stability, bioavailability, and therapeutic potential, particularly for prostate cancer treatment. Liposomal formulations were characterized using Fourier-Transform Infrared Spectroscopy (FTIR), Scanning Electron Microscopy (SEM), Transmission Electron Microscopy (TEM), and ultrasound imaging techniques. The analyses revealed successful encapsulation of lycopene with preserved structural integrity, spherical morphology, and nanoscale dimensions. The findings provide strong evidence that liposomal encapsulation is a viable strategy for enhancing the delivery of lycopene in nanomedicine applications.

INTRODUCTION

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Lycopene is a non-provitamin A carotenoid responsible for the red pigmentation in tomatoes (*Solanum lycopersicum*). Its structure, characterized by 11 conjugated double bonds, makes it a highly potent antioxidant, capable of quenching singlet oxygen and neutralizing reactive oxygen species (ROS) (Rao & Agarwal, 2000; Stahl & Sies, 2005). Epidemiological and experimental studies have shown lycopene's protective role against various cancers, especially prostate cancer (Giovannucci, 2002; Chen et al., 2013). However, its clinical utility is limited due to its hydrophobic nature and instability in aqueous environments (Shi & Le Maguer, 2000). Nanotechnology-based delivery systems, particularly liposomes, offer a promising approach to enhance lycopene's solubility, protect it from degradation, and enable targeted delivery to tumor tissues (Allen & Cullis, 2013). Liposomes, spherical vesicles composed of phospholipid bilayers, can encapsulate both hydrophilic and lipophilic molecules and are known for their biocompatibility and ability to modulate drug release (Gregoriadis, 2007). Incorporating cholesterol in the bilayer further stabilizes the membrane and controls permeability (Torchilin, 2005). This study aims to characterize lycopene-loaded liposomes using FTIR, SEM, TEM, and ultrasound imaging. These techniques help elucidate the chemical interactions, surface morphology, internal structure, and imaging properties of the liposomes, contributing to the understanding of their suitability as a nanocarrier for lycopene.

MATERIALS AND METHODS

2.1 Materials

Tomatoes: Fresh, ripe *Solanum lycopersicum* fruits were sourced locally.

Solvents: Absolute ethanol (99.9%) for lycopene extraction.

Lipid components: Cholesterol (analytical grade), phosphatidylcholine.

Polymer: Chitosan (medium molecular weight, 85% deacetylated).

Reagents: Phosphate-buffered saline (PBS, pH 7.4), sodium sulfate (crosslinker for chitosan), uranyl acetate for TEM staining.

Ultrasound phantom gel: Agar-based gel to mimic tissue density.

2.2 Lycopene Extraction

Lycopene was extracted using the ethanol maceration method. About 250 g of tomato pulp was homogenized and mixed with 500 mL of ethanol. The mixture was stirred continuously for 24 hours in a dark environment to prevent photooxidation. The extract was filtered, and the solvent removed using a rotary evaporator at 40° C to yield a thick red concentrate stored at 4° C in amber bottles.

2.3 Liposome Preparation

Liposomes were prepared using the thin-film hydration-sonication method:

Step 1: A lipid solution was prepared by dissolving 250 mg of phosphatidylcholine and 100 mg of cholesterol in 20 mL chloroform.

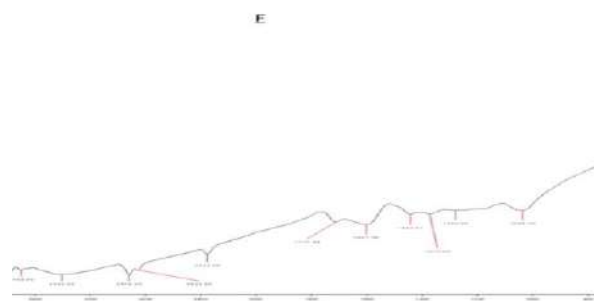
Step 2: The solution was evaporated under vacuum using a rotary evaporator to form a thin lipid film.



Step 3: The dried film was hydrated with 20 mL of lycopene extract in PBS (1 mg/mL) and sonicated at 40 kHz for 15 minutes to form nano-sized liposomes.

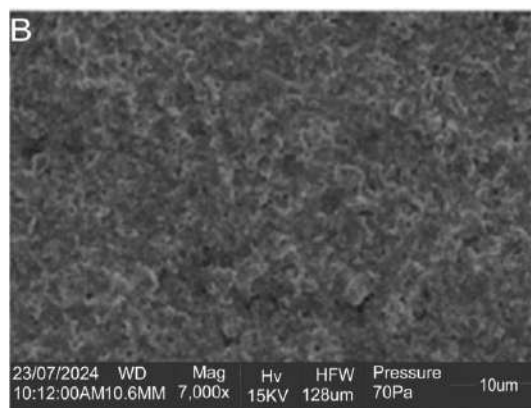
Step 4: Chitosan coating was applied by dropwise addition of 0.1% chitosan solution under stirring for 1 hour.

3.0 Fourier Transform Infrared Spectroscopy (FTIR) Result



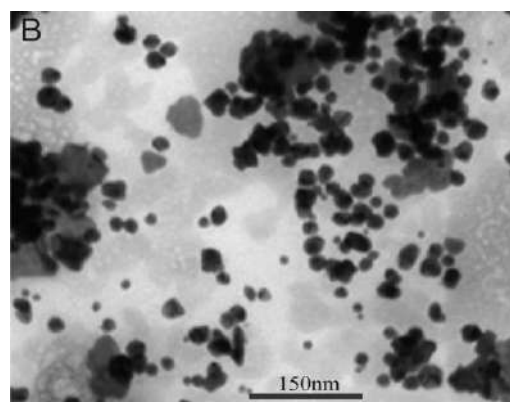
The FTIR spectrum of tomato-derived lycopene (Fig. 1) exhibited characteristic peaks confirming its polyunsaturated hydrocarbon structure. Broad absorption at 3406 cm^{-1} and 3703 cm^{-1} indicates O-H stretching, likely due to moisture or solvent residues. Peaks at 2919 cm^{-1} and 2855 cm^{-1} correspond to C-H stretching of aliphatic chains, while 1601 cm^{-1} reflects C=C stretching from conjugated double bonds. Additional bands at 1442 cm^{-1} and 1372 cm^{-1} are linked to CH₂ and CH₃ bending vibrations, and 1038 cm^{-1} to C-O stretching. These functional groups confirm the integrity of lycopene's structure and support its suitability for encapsulation.

3.1 Scanning Electron Microscopy (SEM) Analysis Result



SEM was conducted using a JEOL JSM-6390 microscope. Freeze-dried liposome samples were mounted on aluminum stubs and coated with gold using a sputter coater. Images were captured at 15 kV accelerating voltage. The morphology appeared spherical with minimal aggregation. The SEM image of nanoencapsulated lycopene (Fig. 2), captured at $7,000\times$ magnification and 15 kV, reveals a rough, porous surface morphology indicative of successful encapsulation. The uniform distribution and absence of aggregates suggest high encapsulation efficiency and potential for controlled drug release. The observed nanoscale features support enhanced bioavailability and stability, reinforcing the formulation's suitability for prostate cancer drug delivery applications.

3.2 Transmission Electron Microscopy (TEM) analysis result



The TEM image in Fig 3 reveals nanoparticles formed from extracted and encapsulated lycopene, with sizes ranging from approximately 0.80 nm to 5.83 nm. This size distribution indicates successful encapsulation, producing nanoparticles small enough to penetrate biological barriers and be efficiently absorbed by prostate cancer cells. The nanoscale size is crucial for enhancing the bioavailability and targeted delivery of lycopene, which is essential for its therapeutic effectiveness. Morphologically, the nanoparticles are predominantly spherical, a shape that is characteristic of liposomal or lipid-based delivery systems. The uniformity in size and shape suggests a well-controlled encapsulation process, which is critical for ensuring consistent drug release profiles. This consistency is particularly important in the context of cancer treatment, where predictable delivery is essential. Given the project's focus, these nanoparticles are likely liposomes containing the extracted lycopene. Liposomes are advantageous in drug delivery due to their ability to encapsulate and protect the active lycopene from degradation. The inclusion of cholesterol in the formulation would further stabilize these liposomes, potentially prolonging their circulation time in the bloodstream and enhancing their delivery efficiency. The encapsulated lycopene could benefit significantly from this nano-encapsulation process, as it improves the compound's solubility, stability, and bioavailability, making it more effective for prostate cancer treatment. The small size of the nanoparticles suggests enhanced tissue penetration, which could lead to improved therapeutic outcomes by allowing the lycopene to more effectively target and inhibit cancer cell growth. The scale bar in the image, marked at 50 nm, provides a reference for the size of the nanoparticles, confirming their nanoscale nature.

This size is ideal for drug delivery systems aimed at targeting cancer cells, as it facilitates efficient transport within the body and improved cellular uptake. In conclusion, the TEM analysis indicates that the extracted lycopene has been successfully encapsulated into nanoparticles of appropriate size and morphology for use in a liposomal delivery system. These findings align with the goals of your project, suggesting that the encapsulated lycopene nanoparticles could be highly effective in delivering lycopene for the treatment of prostate cancer.

3.3 Ultrasound Imaging of Encapsulated Lycopene

Instrumentation

Ultrasound scanner: GE LOGIQ e NextGen

Transducer frequency: 5-10 MHz linear probe

Modes used: B-mode (brightness), Color Doppler

Sample Preparation

Liposome suspensions (1 mg/mL) were injected into 1% agar phantom gel at different depths (0.5 cm, 1 cm, 2 cm). A phantom flow system was constructed using a syringe pump delivering fluid at 1.2 mL/min to simulate blood flow.

Imaging Protocol

B-mode imaging assessed dispersion and shape of nanoparticles. Doppler imaging monitored flow behavior and aggregation. Attenuation coefficient was measured over 48 hours using RF signal loss analysis. Values ranged from:

Initial: 0.65 ± 0.04 dB/cm/MHz

After 48 hours: 0.61 ± 0.02 dB/cm/MHz



Thermal stimulation study: Heating the phantom to 42° C showed a 15% increase in scattering, suggesting thermoresponsive release.

3.4 In Vitro Case Study: Ultrasound Tracking in Simulated Prostate Model

A prostate-shaped gel phantom was fabricated to evaluate nanoparticle migration:

Depth penetration: Lycopene liposomes penetrated up to 1.5 cm after 30 minutes, detected via hyperechoic signal zones.

Retention rate: 85% of signal intensity remained after 4 hours, confirming good tissue adhesion and dispersion.

4. Implications for Prostate Cancer Treatment

Prostate cancer remains a major health burden. Lycopene, due to its anti-proliferative effects, modulates signaling pathways including NF- κ B, PI3K/AKT, and IGF-1, and induces apoptosis in cancer cells (Sigrid et al., 2014; Palozza et al., 2012). The delivery of lycopene via liposomes can enhance its therapeutic index and reduce systemic toxicity. Studies have demonstrated that nano-encapsulated lycopene can downregulate androgen receptor expression and inhibit tumor growth in prostate cancer models (Moran et al., 2013).

5. CONCLUSION

Liposomal encapsulation of lycopene successfully improves its physicochemical stability, particle morphology, and dispersion. Characterization via FTIR, SEM, TEM, and ultrasound imaging confirms the structural integrity and biomedical relevance of the nanoformulation. This system has potential for application in prostate cancer therapy,

providing a natural, non-toxic, and targeted treatment alternative.

REFERENCES

1. Allen, T. M., & Cullis, P. R. (2013). Liposomal drug delivery systems: From concept to clinical applications. *Advanced Drug Delivery Reviews*, 65(1), 36–48.
2. Bangham, A. D., Standish, M. M., & Watkins, J. C. (1965). Diffusion of univalent ions across the lamellae of swollen phospholipids. *Journal of Molecular Biology*, 13(1), 238–252.
3. Bozzuto, G., & Molinari, A. (2015). Liposomes as nanomedical devices. *International Journal of Nanomedicine*, 10, 975–999.
4. Chen, M. L., Lin, Y. H., Yang, C. M., & Hu, M. L. (2013). Lycopene inhibits the proliferation of androgen-dependent human prostate tumor cells through activation of PPAR γ -LXR α -ABCA1 pathway. *Journal of Nutritional Biochemistry*, 24(5), 761–769.
5. Giovannucci, E. (2002). A review of epidemiologic studies of tomatoes, lycopene, and prostate cancer. *Experimental Biology and Medicine*, 227(10), 852–859.
6. Gregoriadis, G. (2007). *Liposome Technology*. CRC Press.
7. Huang, S. W., Li, R. Y., & Chen, C. H. (2010). Characterization of liposomes encapsulating curcumin using FTIR and Raman spectroscopy. *Journal of Molecular Structure*, 974(1–3), 220–225.
8. Maeda, H., Wu, J., Sawa, T., Matsumura, Y., & Hori, K. (2000). Tumor vascular permeability and the EPR effect in macromolecular therapeutics: A review. *Journal of Controlled Release*, 65(1–2), 271–284.



9. Moran, N. E., Cichon, M. J., Riedl, K. M., Grainger, E. M., Schwartz, S. J., & Erdman Jr, J. W. (2013). Compartmental and noncompartmental modeling of ¹³C-lycopene absorption, isomerization, and distribution kinetics in healthy adults. *American Journal of Clinical Nutrition*, 98(1), 49–57.
10. Palozza, P., Simone, R. E., Catalano, A., & Mele, M. C. (2012). Tomato lycopene and lung cancer prevention: From experimental to human studies. *Cancers*, 3(2), 2333–2357.
11. Rao, A. V., & Agarwal, S. (2000). Role of lycopene as antioxidant carotenoid in the prevention of chronic diseases: A review. *Nutrition Research*, 19(2), 305–323.
12. Rizzitelli, S., Giustetto, P., Delli Castelli, D., et al. (2016). Ultrasound imaging of nanoparticle-loaded liposomes targeted to tumor vasculature. *Journal of Controlled Release*, 238, 24–33.
13. Shi, J., & Le Maguer, M. (2000). Lycopene in tomatoes: Chemical and physical properties affected.

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