NANO – DRUG DELIVERY OF ANTI-CANCER DRUG DOXORUBICIN USING TAPIOCA STARCH NANOPARTICLES

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Introduction:

Cancer remains one of the leading causes of death globally, accounting for approximately 10 million deaths in 2020, according to the World Health Organization (WHO). With the global burden of cancer projected to rise to 28.4 million new cases by 2040, the need for advanced, efficient, and safer therapeutic approaches has become more necessary. Conventional chemotherapy, though a cornerstone of cancer treatment, is fraught with limitations such as non-specific cytotoxicity, low bioavailability, rapid systemic clearance, and multi-drug resistance.

To overcome these challenges, nanotechnology-based drug delivery systems have emerged as a transformative strategy in oncology. These systems enable targeted delivery, controlled release, and enhanced solubility of chemotherapeutic agents, thereby minimizing systemic toxicity and reducing side effects. The global nanomedicine market reflects this rapid advancement, valued at USD 138.8 billion in 2023, and is expected to reach USD 381.8 billion by 2032, growing at a CAGR of 11.7%.

This project explores the nanoencapsulation of doxorubicin, a widely used anthracycline chemotherapeutic agent, using tapioca starch-based nanoparticles as

the carrier system. Tapioca starch, a natural, biodegradable, biocompatible, and non-immunogenic polysaccharide, presents an attractive alternative to synthetic polymers in drug delivery. Its high amylopectin content offers superior swelling and gelling properties, contributing to high drug loading capacity, excellent nanoencapsulation efficiency, and sustained drug release profiles. Moreover, its minimal interaction with the encapsulated drug ensures better retention of pharmacological activity.

Objectives:

1. Nanoencapsulation of Doxorubicin using Tapioca starch
2. Nanoencapsulation Efficiency and Sustained Release Kinetic Studies of nano-encapsulated Doxorubicin
3. Characterization of nano-encapsulated Doxorubicin using FTIR and SEM
4. Validation of sustained release of Doxorubicin from Tapioca starch nanoparticles on MCF and HeLa cancer cell lines

Methodology:

1. Extraction of Tapioca starch
   Fresh cassava roots were washed, peeled, chopped, and ground into pulp. The pulp was mixed with water (1:10 w/v), filtered, and sedimented for 2 hours. The starch was dried at 55°C for 24 hours.

2. Preparation of Tapioca starch nanoparticles and its characterization:
   To prepare nanoparticles, the dried starch was ultrasonicated at 20 kHz for 30 minutes and stored aseptically.

3. Drug binding capacity
   Fixed doxorubicin (50 µg/mL) was added to varying starch amounts (50–1000 mg) in distilled water and incubated for 12 h. The supernatant and sediment were analyzed to determine unbound drug and sustained release using UV-Vis at 480 nm.

4. Drug loading capacity

Fixed starch amount was mixed with increasing doxorubicin concentrations (100–800 µg), incubated, and analyzed for release similarly.

5. Nanoencapsulation efficiency
   Encapsulation efficiency was evaluated by the formula:

   EE%= (Amount of bound drug /total amount of drug added) ×100

6. Calculation of yield
   Yield % = (Weight of Petri plate + nano-encapsulated drug) – (Weight of petri plate) = yield of nano-encapsulated drug in grams * 100

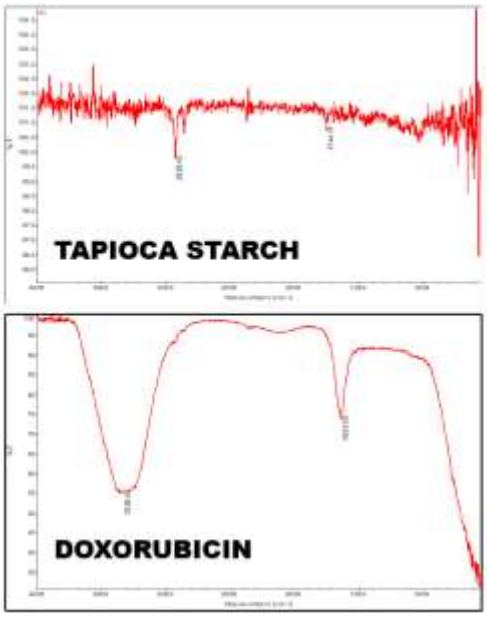
7. Sustained drug release kinetics study:
   Sustained release was assessed in saline at pH 7.2 and 37°C over 8 h, with OD readings taken hourly at 480 nm.

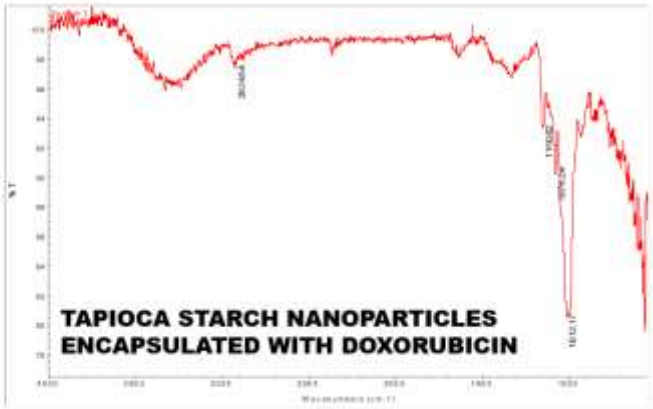
8. Validation of sustained drug release on cell lines.
   Cytotoxicity was validated using the MTT assay on cancer cell lines, measuring absorbance at 570 nm to assess cell viability post-treatment.

Result and Conclusion:

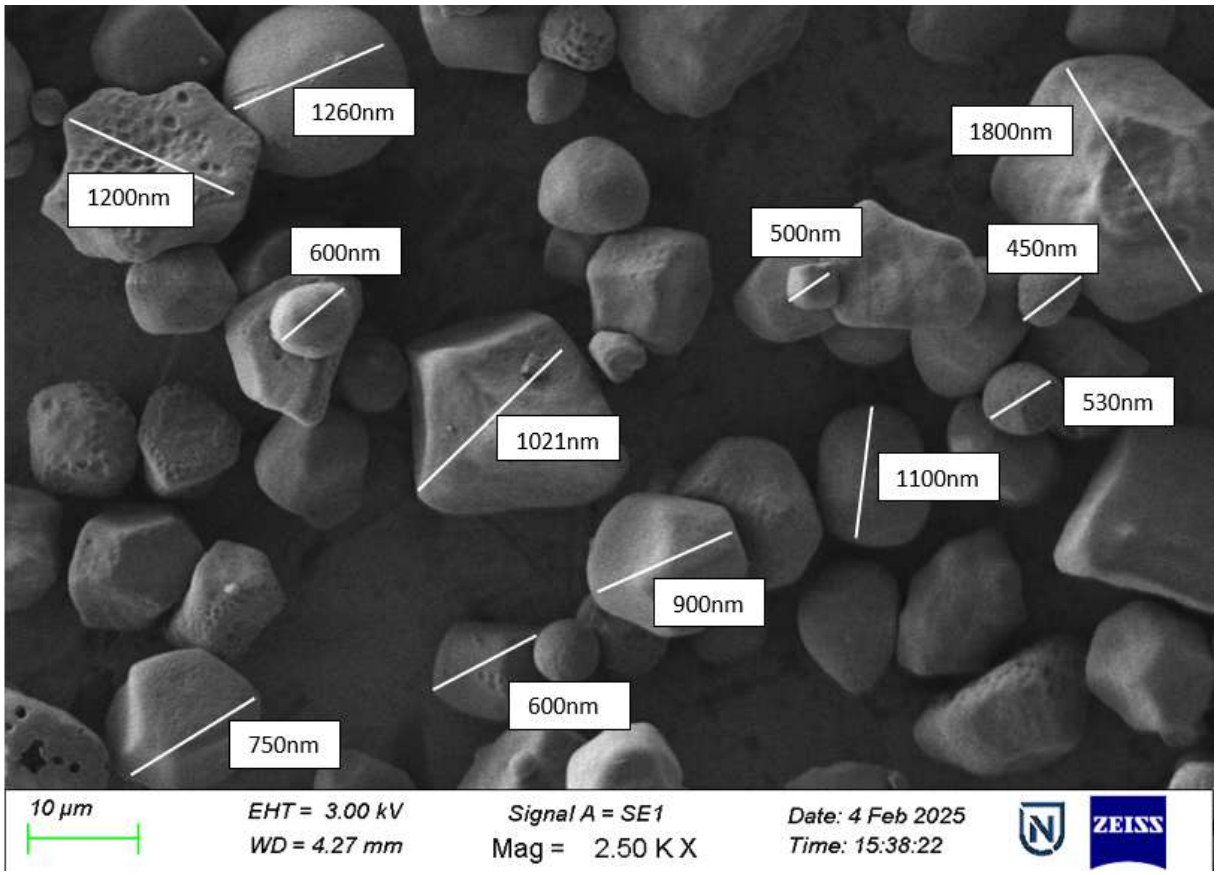
The optimal nanoparticle concentration (200 mg) exhibited the highest binding efficiency and sustained drug release, with 400 µg of doxorubicin showing maximum encapsulation efficiency (93.4%).

FTIR analysis confirmed successful drug loading with minimal interaction.

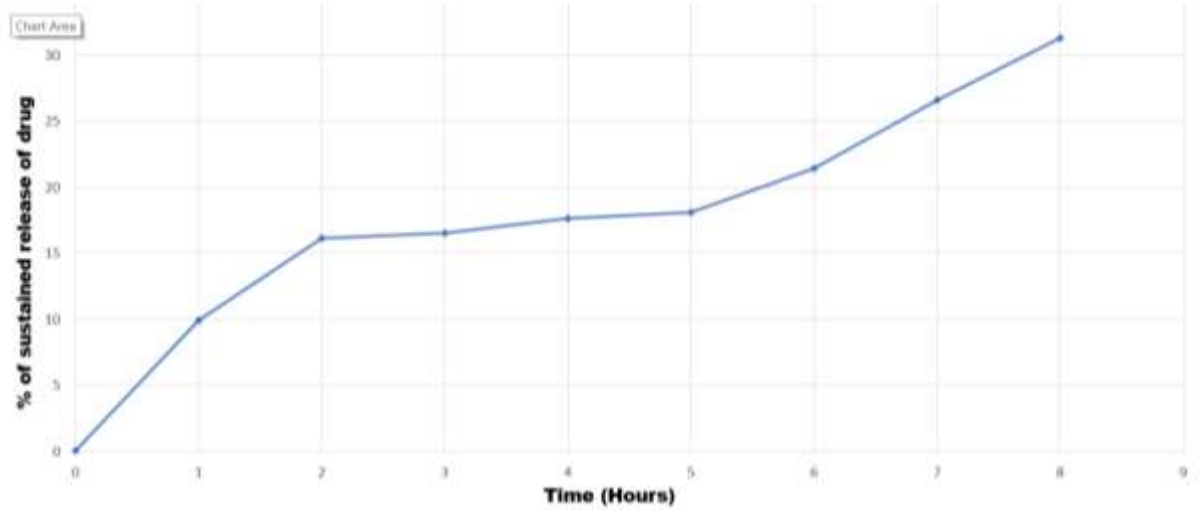


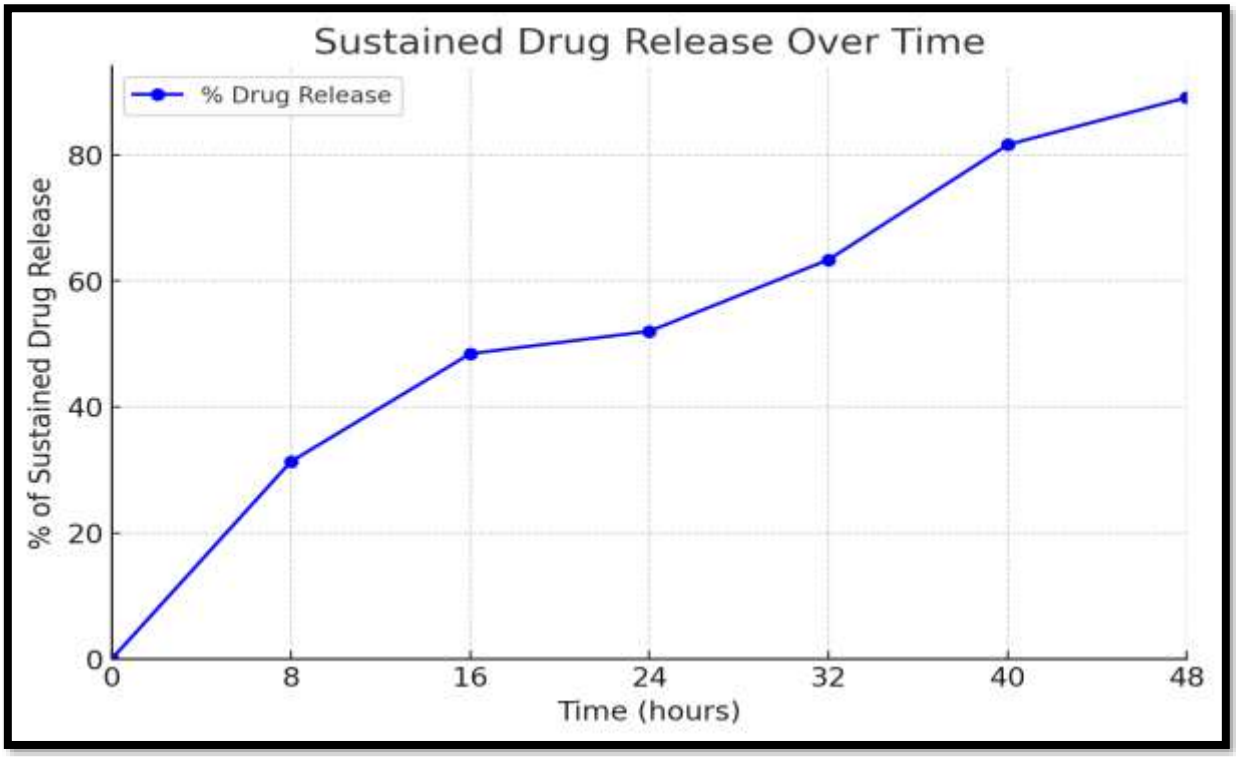


The nanoparticles, ranging from 500–1000 nm in size, exhibited spherical, polygonal, and irregular morphologies, facilitating efficient cellular uptake. It exhibited polydisperse nanoaparticles which is ideal for nano- drug delivery.



Sustained release kinetics demonstrated 31% drug release in 8 hours and 89% in 48 hours, without degradation. The validation studies to evaluate cytotoxicity studies on HeLa and MCF7 cell lines confirmed a dose-dependent decrease in cell viability.





In conclusion, given its biocompatibility, eco-friendliness, cost effectiveness, and controlled drug release capabilities, tapioca starch nanoparticles present a promising platform for drug delivery. The sustainable nature of tapioca starch makes it a promising candidate for future advancements in nanomedicine and drug delivery.

Future Scope:

Building on the promising results of using tapioca starch nanoparticles as a nano-carrier for doxorubicin, future research could expand the potential applications of this delivery system. Investigating other therapeutic agents for encapsulation could pave the way for broader clinical applications, particularly in targeted cancer therapies. Furthermore, exploring in-vivo models will be essential to validate these findings and understand the long-term biocompatibility and effectiveness of the nanoparticles. Additionally, mechanistic studies, including apoptosis, cell cycle analysis, ROS production, and protein expression profiling, could offer insights into the molecular pathways influenced by these nanoparticles, especially in breast and cervical cancer cells. Such studies would deepen our understanding of the therapeutic potential and optimize this delivery system for enhanced anti-cancer efficacy and enhanced therapeutic efficiency.

11. Project Outcome & Industry Relevance (10-15 lines):

Practical implications :

- **Controlled Release and Lower Cumulative Dose -** The nano-encapsulated form of doxorubicin allows for a controlled and sustained release of the drug, maintaining therapeutic levels over a prolonged period and potentially requiring a lower cumulative dose. Limiting systemic toxicity, including bone marrow suppression, and decreasing the cumulative exposure associated with secondary malignancies.

- **Minimized Extravasation Risks -** Nanoencapsulation in tapioca starch can stabilize doxorubicin within nanoparticles, reducing the likelihood of the drug leaking or extravasating from blood vessels during intravenous administration. Thereby, preventing necrosis and severe tissue ulceration.

- **Reduced Cardiac Toxicity** - With a controlled release, there's less risk of high peak concentrations that can lead to toxicity in non-target tissues, like the heart,

which reduces the risk of side effects like cardiac toxicity. This targeted delivery can help prevent acute and chronic cardiac toxicity, potentially reducing the risk of reversible arrhythmias, irreversible cardiomyopathy, and congestive heart failure.

This project is relevant in the pharmaceutical industry in its role in ensuring that drugs are delivered efficiently and effectively to the right part of the body at the right time. Companies like Johnson & Johnson ,Sun Pharmaceutical Industries Ltd. are involved in manufacture of nano-medicine for drug-delivery.

Working Model vs. Simulation/Study:

Starch as a nano-carrier was explored in this study. Tapoica starch nanoparticles were characterized using Scanning electron microscopy and FTIR. The validation was conducted on **HeLa and MCF7** cell lines indicated that there was **decrease in cell viability** with **increasing concentration of doxorubicin.**

Project Outcomes and Learnings:

Tapioca starch nanoparticles demonstrated significant potential as an efficient nano-carrier for doxorubicin delivery. The high amylopectin content of tapioca starch facilitated excellent drug encapsulation, with an encapsulation efficiency of 93.4%. The optimized nanoparticle concentration (200 mg) and doxorubicin loading (400 µg) achieved the highest binding efficiency and sustained drug release, with 31% released in 8 hours and 89% in 48 hours. The FTIR analysis confirmed successful encapsulation and sustained release without drug degradation. Furthermore, cytotoxicity studies on HeLa and MCF7 cell lines validated the nanoparticles' efficacy in reducing cancer cell viability. Given its biocompatibility, cost-effectiveness, and eco-friendly nature, tapioca starch nanoparticles offer a promising platform for controlled drug delivery and can be further explored for the delivery of various therapeutic agents.