Preface

New materials and technologies in addressing complex medical challenges continue to remain one of the driving forces behind the landscapes of biomedical engineering and healthcare. Among some of the most exciting trends relating to bringing these systems together, particularly in drug delivery, polymers, nanoparticles, composites, and nanofibers, have highly varied applications in cancer treatment, tissue engineering, implants, imaging, medical devices, and other biomedical applications. Drug delivery systems (DDSs) have been investigated as vital strategies in cancer therapy research in recent years due to the rapid development of polymeric biomaterials. One of the most significant types of polymers, polyurethane (PU), has changed the field of regenerative medicine and the potential for medical treatments due to its exceptional biocompatibility and capacity to work in harmony with biological systems. Its easy production and versatile properties, which depend on its structure, make it a highly sought-after polymer. Moreover, the chain extenders, which are amino-acid and peptides, are hydrophilic in nature, which facilitates the drug release. In addition, the amino-acid and peptide grafting on PU increases the selectivity towards the cancer cells, and the targeted killing efficacy increases when conjugated with the drug. Here, the drug-PU conjugate follows the "Blitzkrieg Warfare" strategy to kill cancer cells. Being composed of amino-acid (Cystine), it also shows antimicrobial properties. As chemotherapy suppresses the immune system, cancer patients are more susceptible to bacterial infections, which can lead to the failure of treatment and impede cell proliferation as well. The risk of infections at the site of drug administration or within the body during treatment may be decreased if the drug delivery system includes built-in antibacterial qualities. The development of polymeric materials, such as Tryptophan combined with aromatic PU for a "Theranostics" approach (therapy as well as imaging), or grafting of GLY-GLY dipeptide with PU for giving an ECM mimicking surface for better integrin binding (like RGD) applicable for targeted killing, Cystine combined with aliphatic PU for drug delivery in cancer cells as well as bacterial infection prohibition, further broadens their utility in tumor treatment. Previously reported works of literature showed that cell adhesion and motility dynamics in three-dimensional (3D) matrices may greatly influence liposome-cell interaction in physiological environments. Fraley S.I.[1] characterized that focal adhesions,

extensive multi-protein complexes at the basal cell surface, are central to mediating cell signaling, force transduction, and adhesion. Interestingly, in 3D matrices, focal adhesion proteins such as vinculin, paxillin, talin, α-actinin do not form traditional aggregates but are diffusely distributed throughout the cytoplasm. This study shows that membrane protrusions constitute a critical motility/matrix-traction module that drives cell motility in a 3D matrix. Brian J Lestini et al[2] showed the development of liposomal surface modifications that address both targeting specificity and vesicle clearance. This study utilized an RGD-containing peptide to target liposomes to integrin GPIIb–IIIa on activated platelets and explored the role of oligodextran surfactants in modulating vesicle stability and clearance. These insights indeed form the foundation of our thesis work. Such pioneering discoveries find enormous scope within the ambit of modern healthcare providing a flexible solution that has transcended conventional methods to make an effective counter to varied clinical problems associated with cancer treatment, therapies, and diagnostics. This effort places a new age for medical innovation and improved patient care.

In **Chapter 1**, we offer a concise overview of cancer and the conventional methods used for tumor treatment. We offer an idea about polyurethanes, elucidating their modifications, and properties with a brief literature survey on biomaterials and various chain extenders. Our exploration extends to their potential applications in biomedical fields. This chapter summarizes the major contributions in this field and presents the research's important findings. The goal of the thesis is described based on the literature review, and as a consequence, the findings, discussions, and significant findings are provided in four distinct chapters, which are followed by conclusions.

In **Chapter 2**, we present a summary of the experiments utilized for sample preparation. The chapter also addresses characterization techniques that are used for data collection and calibration. The structural and microstructural analysis is carried out using NMR, FTIR, UV-VIS, and XRD. UTM, DSC, TGA, and DMA to compare thermal and mechanical responses with morphological investigation by SEM, BT-SEM, AFM, and TEM. Detailed discussions are provided for the biological characterizations, such as *in-vitro* drug release, the MTT (3-[4,5-dimethylthiazol-2-yl]-2,5 diphenyl tetrazolium bromide) assay, antibacterial assay, and ROS assay with *in-vivo* release, along with gelation kinetics and

H&E staining for localized treatment efficacy. UV-VIS spectrophotometer is used to check the drug-polymer interaction, along with mathematical modeling, and Docking is done to confirm the drug-polymer interaction.

Chapter 3 For the regulated release of the model drug PTX, four distinct polyurethane-based carrier systems have been developed. FTIR, NMR, and UV-VIS investigations are used to describe the polyurethane (PU) carriers, and their release behavior is examined in PBS. Interestingly, PU with aliphatic diisocyanates shows a release of about 80%, whereas those with aromatic diisocyanates show less than 20% drug release in PBS (pH 7.4). PU-I (IPDI-based PU) is determined to be more hydrophilic based on contact angle measurements. Furthermore, the well-spread morphologies of the PU samples with increased adherence to the cell surfaces are confirmed by cellular adhesion studies using SiHA cells. The presence of the −NH– bond is confirmed by FTIR analysis, which agrees well with the NMR spectra. These results highlight how important polyurethane's segmental structure is as a flexible instrument for regulating drug release.

After being confirmed that PU can act as a DDS, in **Chapter 4**, we synthesize Tryptophan (TRP) modified PU by varying the weight percentage from 30-70% using grafting process for its potential application in therapy as well as imaging. NMR confirmed the formation of PU-TRP, whereas FTIR and UV-Vis supported the fact of tagging. XRD shows the amorphous nature of modified PU post-polymerization. Further, the MTT assay confirms the higher cell viability of PU-TRP compared to PU in both SiHa and 3T3 cell lines. The drug-loaded material selectively kills up to 14% of cancer cells, leaving 95% of 3T3 cells alive at 20 μg/ml concentration. We have further confirmed that the material shows fluorescence inside the cells through a three-day study on cancer cells. Also, we did not observe any significant toxicity in the skin section after TNFα staining when the section of skin was collected after gelation kinetics study. *In-vivo* drug release confirmed that our material shows sustained release on both IV and IP modes in comparison to pure PTX. So, this material can be used in localized treatment for tumor killing as well as diagnostic purposes.

Chapter 5 L-cystine (dimer) based polyurethane has been synthesized using HMDI and PTMG as the hard and soft segments, to generate biodegradable polymer (PUs) with

advantageous biocompatibility and bioavailability qualities. NMR, FTIR, and UV studies confirmed the addition of cystine to PU chains. In comparison to pure polyurethane, CYS-modified PU releases up to 89%, whereas PP/pure polyurethane has only 58% in a similar time frame (48 hours). The data from the 24-hour cellular assessment confirms improved adherence of material (PU) to cancer cells, and the OD values for 3T3 and SiHA cells are 101% and 149%, respectively. These kinds of systems help treat cancer in a targeted manner. Modified PU (20 µg/ml) selectively kills 59% of SiHA cells following three-day research, while the same drug-loaded system retains 85% of 3T3 cells viable at the same dosage. In addition, the CYS-modified PU shows antibacterial properties against *S. aureus* with an MIC of 20 mg/ml. Cystine's antibacterial property adds to PU and reduces ROS of the system where oxidative stress is concerned, and effectively inhibits the growth of *S. aureus*. CYS-modified PU shows protracted drug release, high biocompatibility along with antibacterial properties, which makes it a suitable candidate to be used as an anti-cancer targeted drug delivery system.

In **Chapter 6**, we present a groundbreaking strategy for mimicking the ECM matrix with the simplest amino acid, Glycine, and its homopeptides (which act as an integrin-binding sequence) that uniquely provide an RGD-like sequence and help in killing cancer cells in a targeted manner. These are confirmed using NMR, UV-VIS, XRD, FTIR, and SEM analysis. MTT assay and cell adhesion tests on both SiHA and 3T3 cells demonstrate that peptide-grafted PU adheres to SiHA cells more readily than 3T3 cells because the peptides can mimic the ECM microenvironment of cancer cells. Further, the biomaterials are loaded with PTX and the molecular docking studies reveal the interaction between the drug and the polymer matrix, indicating that tripeptide-grafted PU (PU-G3) drug interaction is stronger than dipeptide-grafted PU (PU-G2) drug, although the latter shows 31% drug delivery compared to 12% release of PU-G3 in 24 hours in contrast to PU-G1 having 10% and pure PU with 9% release. Notably, introduction of PTX into PU-G2 matrix leads to low death rates (98% viable cells) in healthy cells (3T3), as opposed to a 54% mortality rate with the pure drug. PU-G2 (20 µg/ml) selectively kills 54% of SiHA cells in a three-day trial. For this reason, the "next generation" of DDS-based systems with greater efficacy should include peptides that can imitate ECM components like (RGD surface) or integrin-binding sequences and characteristics to achieve better treatment for cancer therapy.

In **Chapter 7**, a discussion of the current research's potential and a thorough description of the work completed are given. In this work, we have concentrated on investigating the *in-vitro* properties of modified polyurethane with different chain extenders, primarily amino acids and peptides, for biomedical applications like drug delivery, diagnosis, and antibacterial assay; further research through *in-vivo* studies are necessary.

A list of journals and books used to bind up the thesis has been given at the end as references.