Objective & Plan of work

3.1 Aim of work

The aim of this research was to develop and optimize novel PHBV based polymeric nanoparticles containing docetaxel (BCS class IV) antineoplastic drug as an active pharmaceutical ingredient for the effective and improved treatment of breast cancer. To achieve the aim, all the developed formulations were characterized and evaluated for *in-vitro* and *in-vivo* performance. In addition, their anticancer potential against associated cell lines (MCF7) and anticancer efficacy against C3H/J strain mice were also evaluated.

3.2 Objective of work

1. To develop, optimize and characterize breast cancer targeted docetaxel loaded polymeric nanoparticulate based drug delivery system.
2. To improve its bioavailability, biological half-life, blood circulation time and therapeutic efficacy.

3.3 Rationale of work

❖ Selection of Drug - Docetaxel

- Docetaxel (DTX) is a BCS class IV antineoplastic drug. It is available as intravenous formulation. Here, solubility, as well as permeation both are rate-limiting step in drug absorption, which may affect its pharmacokinetics.
- During chemotherapy, drugs destroy cancer cells by stopping them from growing, multiplying and spreading, but healthy cells are also harmed

along with these cells, especially those which divide quickly. The selected drug docetaxel has a potent anti-tumor activity and generally, large dose is recommended for treatment, which may induce toxic side effects to normal cells and non-targeted organs.

- Due to lack of tissue specificity docetaxel shows systemic toxic side effects like bone marrow suppression, peripheral neurotoxicity, mucositis, neutropenia, mild asthenia, mild paresthesia, neurosensory disturbances, anaemia, hepatotoxicity, hypersensitive reactions and fluid retention.
- DTX also gets rapidly metabolized, offer tumor drug resistance and suffer from other shortcomings. So, a unique carrier system is needed for specific, sustained and targeted drug delivery.
- An increase in bioavailability of DTX may help in reducing inter-subject variability as well as prevention of development of drug resistance on prolonged usage.

❖ Development of Polymeric nanoparticles

- Polymeric nanoparticles offer advantages as drug carriers in terms of stability, drug targeting specificity, feasibility of incorporation of both hydrophilic and hydrophobic substances, ease of preparation and predefined rate or extent of drug release.
- Recently, the trend has been shifted towards natural polymers for developing drug delivery systems. Main advantages of these polymers are their low cost and good compatibility with the encapsulation of a wide range of drugs, and also minimal use of organic solvents.

- PHBV is biodegradable and biocompatible natural polymer. It is also included in FDA inactive ingredient database. It is rigid in nature and having lower glass transition temperature. This allows longer encapsulation time for the entrapped drug.

3.4 Plan of work

A. Preformulation studies

- FTIR interpretation
- Analytical method development (UV/HPLC)
- Standard calibration curve

B. Formulation of polymeric nanoparticles

- Selection of manufacturing method
- Polymeric nanoparticles (NPs) preparation
  - PHBV PVA polymeric NPs (PHBV-PVA-DTX)
  - PHBV PF127 polymeric NPs (PHBV-PF127-DTX)
  - PHBV TPGS polymeric NPs (PHBV-TPGS-DTX)
  - PHBV PLH PE-PEG polymeric NPs (PHBV-PLH-PEG-DTX)
- Formulation optimization
  - Identification of critical quality attributes (CQA)

- Selection of critical quality attributes (CQA)
- Placket-Burman design (PBD)
- Box-Behnken design (BBD)

C. Characterization

- Particle size (Zeta sizer)
- Surface charge (Zeta potential)
- Morphology (SEM, TEM, and AFM)
- Entrapment efficiency (%EE)
- Drug-excipient interaction analysis (FTIR/DSC)
- Analysis of crystallinity (XRD)

D. Drug release studies (*In vitro*)

E. Cytotoxicity assessment (MCF7 by SRB assay)

F. Cellular uptake study (confocal microscopy)

G. Evaluation of hemolysis

H. Platelets aggregation study

I. Drug-plasma interaction (SDS-PAGE)

J. Pharmacokinetic studies

K. *In silico* and *ivivc* studies

L. *In vivo* anticancer activity (C3H/J strain mice)

M. Stability studies

N. Statistical analysis