CHAPTER-1

Introduction

Colon canceris recognized as one of the common prevalent cancers and the fourth leading cause of cancer related death worldwide[Kraus et al., 2014].Itgradually progresses from benign polyp or cyst and if left untreated can metastases to the major organs of the gastrointestinal tract (GIT) such as liver, spleen, pancreas etc.[Kuppusamy et al., 2017]. This is very common with the patients suffering from inflammatory bowel disease. These people are considered at the greatest risk of the colon cancer[Mustafa et al., 2016]. The other contributory factors that promote the occurrence of colon cancer includes smoking, excessive alcohol consumption, diet, old age, lack of physical activity or exercise, and living lifestyle[Fedirko et al., 2011, Lee et al., 2012].Genetic risk is also observed as one of the factors, however, through a survey it has been identified that in the population of greater than 75-95% of people are affected with colon cancer with little or no genetic disorder[Watson and Collins, 2011]. Various strategies are available to screen colon cancer such as optical colonoscopy[Regula et al., 2006], flexible sigmoidoscopy[Atkin et al., 2010], CT colonography[Horvat et al., 2018], barium enema[Brady et al., 1994]. Colon cancer is curative at the early stage, if detected can be prevented by the removal of premalignant adenomas of the colon[Schulmann et al., 2002].Surgery is also preferred in the initial stage or stage I of the colon cancer with the curing rate of 95% and 75% at stage II. But this fails at stage III as this stage showsthe presence of nodal involvement that increases the chance of reoccurrence of the cancer[Markowitz et al., 2002]. Thus, when treatment with the available approaches (screening and surgery) is not possible,patients are provided with a combination of cytotoxic therapy along with targeted therapy [Hammond et al., 2016]. Though this conjugated treatment is an advancement towards systemic therapy, but this does not assure

the survival rate[Siegel et al., 2014]and also develops approximately 90% resistance to therapy[Longley and Johnston, 2005]. Thus, there isa requirement of change in the current treatment.Recently, several scientific advancements has been done in the field of polymeric drug delivery system[Allen and Cullis, 2004]. Utilization of polymers as an additive is done nearly in all dosage forms, but their application in conjugation with drugs and other therapeutics has led it towards a unique drug delivery system known as controlled drug delivery [Jagur-Grodzinski, 2009]. This form of delivery systemcontrols the drug release at the therapeutic site while encapsulating the drug within the polymericshell fora prolonged time without any toxic effect [Malekzad et al., 2018]. While designing a polymeric drug delivery system, the biodegradation of the polymers must be taken into account[Padmakumar et al., 2018]. Often, the degradation product of the polymer may exhibit the toxic effect. Several factors are there that supports biodegradation such as enzymatic or hydrolytic degradation, physical factors including change in the size, shape and porosity of the polymer or any change in chemical structure occurred by ionic exchange etc. These factors should be considered carefully for the proper biodegradation[De Souza et al., 2010]. Today, polymers have become the most popular material because of their versatile application, reasonable cost, excellent chemical resistance, easy processibility and tailoring as per the desired applications[Tiwari et al., 2014]. There exist several multiphase polymeric systems, such as graft copolymer, block polymers and polymers blends, chemical blends etc.Among all, the physical blended polymers are easy to prepare but during mixing or blending of thepolymers,they show coarse multiphase morphology due to thermodynamic incompatibility thus, they undergo phase separation. Graft and block copolymers are better than the physical blended but they are limited only to thermoplastic polymers[Molau, 1970].

Interpenetrating Polymeric Network (IPNs)are different from the above mentioned polymeric system. They are the novel type of polymeric blend (blend of either natural polymers or

synthetic polymers alone or in combination) that are either cross-linked or synthesized in the immediate vicinity of each other in such a way that network cannot be dispersed unless the chemical bonds are broken[Aminabhavi et al., 2015, Frisch, 1985]. IPNs are the only effective polymeric drug delivery system which intimately combines two cross-linked polymers without or low phase separation [Andrews et al., 2009].

Preparation of IPN involves generally two or more polymeric constituents and based on the polymeric structure they are categorized into two types

- Full IPN/IPN
- Semi IPN

When both the components are cross-linked then the IPN is said to be a full IPN. However, in both the components one polymer is linear and the other one is a network then the IPN is said to be semi IPN [Mandal et al., 2012]. A general schematic presentation of IPN is shown in Figure 1.1.

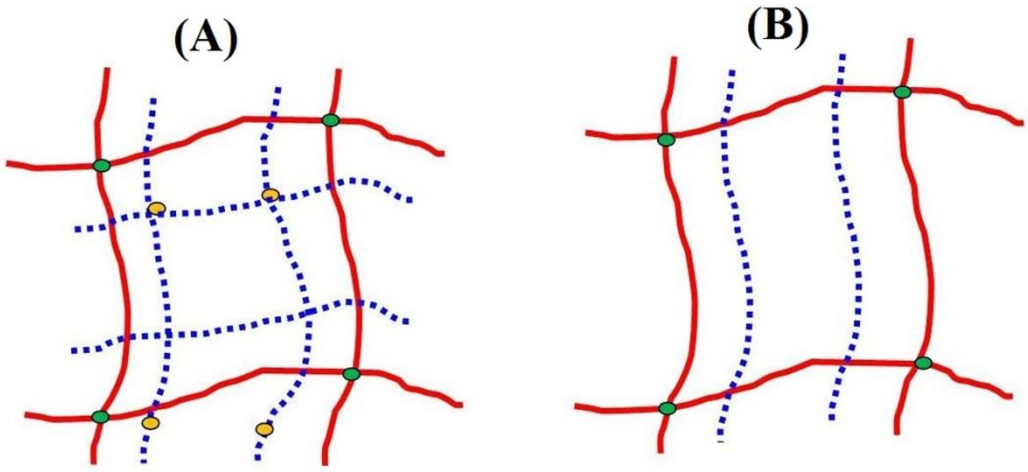

Figure 1.1 Types of IPN(A) Full IPN; (B) Semi IPN

Oral controlled multiparticulateunit dosage forms for example pellets, macromolecules (beads) and micro particles etc.are more popular than the conventional single unit dosage forms such as tablets and capsules etc. because they easily enter and distributes well over

large surface area of the absorbing mucosa of GIT, thus, avoids gastric emptying, and prevents exposure to the high drug concentration[Bechgaard and Nielsen, 1978].

Oral multi-particulate drug delivery using IPN is a novel idea in the field of medicine [Siepmann et al., 2008]. It is one such delivery vehicle that releases the bioactive molecule in a controlled way. IPNs generally involves the use of natural polymers that are cross-linked by means of divalent or trivalent metallic cation etc. thus, masks the demerits usually associated with the synthetic polymers such as bacterial contamination, loss of viscosity upon storage and uncontrolled hydration[Burugapalli et al., 2001, Changez et al., 2003].

Locust bean gum (LBG) is a natural plant polysaccharide composed of galactomannans unit[Mirhosseini and Amid, 2012].It is soluble in water with slight heating and non-ionic in nature[Chen et al., 2001]. LBG is remarkably utilized in oral drug delivery due to high drug loading capacity, excellent mucoadhesive property, and for the most desired property i.e. release of drug in a controlled way [Asane et al., 2008].

Sodium alginate (NaAlg) is another natural polysaccharide used widely in the pharmaceutical industry for controlled release[Gombotz and Wee, 2012]. It is obtained from brown seaweeds and made up of D-mannuronic acid and D-guluronic acid units.NaAlg imparts controlled release to the bioactives or drugs by formation of gels by cross-linking with di or trivalent cations that occur at the G-G sequence junction within the polymer, called as 'egg box junction' [Tripathi and Mishra, 2012].

The model drug selected for the research work is Capecitabine (CAP).CAP is an oral anticancer prodrug of 5- Fluorouracil (5-FU) andmarketed under brand name Xeloda®, approved by USFDA for the treatment of metastatic colon cancer[Schoener and Peppas, 2012]. It is a fluoropyrimidine carbamate derivative, developed to imitate the continuous 5-FU infusion.After its absorption into blood circulation it converts into 5-FU following three steps(Figure 1.2) (1) CAP converts into 5'DFCR (5-deoxy-5-fluorocytidine) by

carboxylesterase enzyme in liver; (2) 5'-DFCR then converts into 5'DFUR (5'-deoxy-5-fluorouridine) by cytidine deaminase present in liver and finally (3) 5'DFUR converts into 5-FU by enzyme thymidine phosphorylase (TP) present in the tumor site[Kaklamani and Gradishar, 2003].Since this orally administered prodrug was developed to mimic the continuous effect of 5-FU, however, after approximately 6 h,5-FU is eliminated completely and its recommended twice daily dosing of 1250 mg/m^2 creates a dosing exposure gap of 6 h in between two subsequent doses.Therefore, the present study is an effort towards the preparation of sustained/ controlledrelease dosage form for once / twice day medication with continuous maintenance of drug concentration in therapeutic range during the therapy.

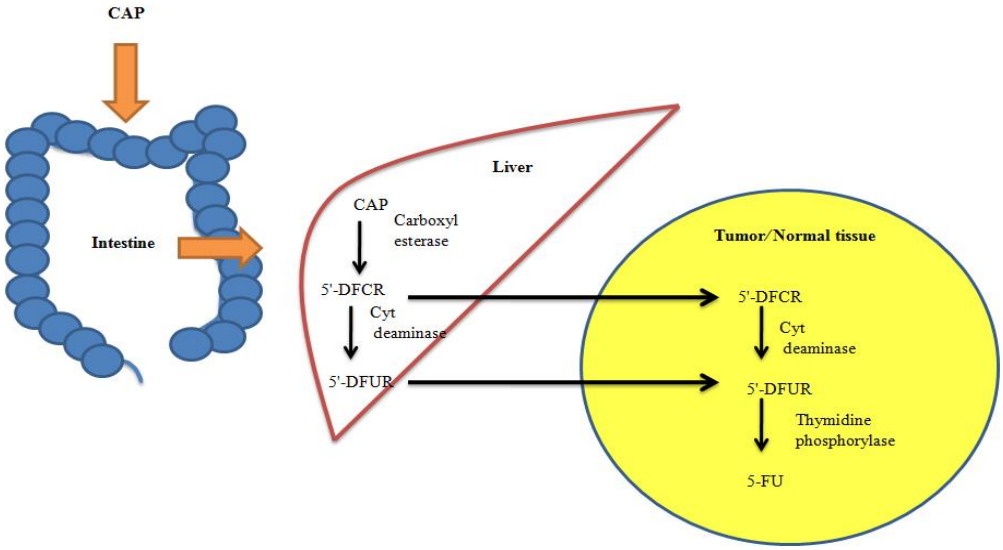

Figure 1.2 Bioconversion pathway of Capecitabine.

Designing an efficient pharmaceutical dosage forminvolvesrigorous and complex stepsas it requires a deep understanding between the process parameter and quality attributes to get a best characteristic pharmaceutical product [Crcarevska et al., 2015].Using one-factor at-a-time optimization is not an efficient tool to obtain an optimized formulation thus; there is a need to establish a design space (range of variability for each selected factors) in order to identify the target quality [Cavazzuti, 2013]. There are numerous experimental designs for the development of a formulation with less number of experimental runs[Chang et al., 2007,

Gohel and Amin, 1998]. Recently, application of statistical experimental design or Design of experiment (DOE) to a pharmaceutical formulation has shown a better understanding between dependent and independent variables with less number of experimental runs by its three key elements i.e. replication (involves, repetition of the experiments to obtain more precise result and also to estimate the experimental error); randomization (random orders in which theexperimental runs to be performed) and blocking (isolates the biased effect and prevent it from concealing the main effect)[Cavazzuti, 2013]. After screening out the main factors and removing the complicated variables, the screened factors are furtheremployed to the response surface methodology (RSM). RSM helps in identifying the possible interrelationship between independent and dependent variables through polynomial equations and two dimensional and three-dimensional plots.

The present work is an attempt to optimize, prepare and evaluatenatural polymers based oral extended delivery of Capecitabine through IPN drug delivery vehicle. Oral drug delivery is still regarded as an ideal and acceptable route of administration because it ensures patient compliance and comfort [Ensign et al., 2012]. Oral chemotherapy in the real sense can translate the concept of traditional chemotherapy as it provides long exposure of the drugs to the cancerous cell at relatively low concentration which leads to the fewer side effects and also a less chance for the cancerous blood vessels to grow[Bottomley, 2002, Feng and Chien, 2003].IPNs composed of natural polymers, LBG and NaAlg are sufficient in extending the release of drug from any delivery vehicle. However, to further sustain the release of a drug with such high dose, a concept of dual mechanism i.e. combination of floating and mucoadhesion would be an ideal approach for prolonging the release of CAP having short elimination half-life of 1-2 h[Bera et al., 2015]. Buoyant dosage forms remains in a floating condition and are sufficient to overcome gastric emptying time for an extended periodhowever, with the passage of time as the stomach empties the floating capacity of the

dosage form reduces as well as formulation also passes towards other parts of GI tract. At that time the property of mucoadhesion can provide theformulation to remain in intimate contact with the mucosal wall of the GI tract, thereby enhancing the bioavailibilty and prolong the release of the drug from delivery vehicle[Adebisi et al., 2015, Patil and Talele, 2015]. Swelling and mucoadhesion are the inherent feature of the polymers LBG and NaAlg, thus, the feature of our third formulation clearly depicts that after adding pore former the system will become porous and ensures that the formed dosage formulation will stay in the stomach region for extended time so that distribution and movement of CAP occurs slowly within gastric fluidand through mucoadhesion get adhere and absorbed at the sites to which it is suitable.

The present thesis thusincludes optimization, formulation, characterization and evaluations of three different preparations of IPN microbeads loaded with CAP

- Preparation of LBG and NaAlg based divalent calcium ion (Ca^{2+}) cross-linked IPN microbeads loaded with CAP
- Preparation of LBG and NaAlg based trivalent aluminium ion (Al^{3+}) cross-linked IPN microbeads loaded with CAP
- Preparation of LBG and NaAlg based trivalent aluminium ion (Al^{3+}) cross-linked IPN microbeads loaded with CAP exhibiting the dual mechanism of floating and mucoadhesiveness.

All three types of formulations of IPN microbeads prepared by ionotropic gelation method and their optimized batch were subjected to various characterization techniques such as Fouriertransforminfra red spectroscopy (FTIR), x-ray diffraction (XRD), differential scanning calorimetry (DSC) for drug-polymer interaction and change in the physical form.Energy dispersive x-ray (EDX) for thedetermination of elemental composition. For the size measurements of IPNmicrobeads, optical microscopy and

scanning electron microscopy (SEM) were utilized.Other experiments such as *in vitro* drug release, swelling, *in vitro*buoyancy,*ex vivo* mucoadhesion (in case of buoyant IPN microbeads) were also performed. Other studies such as *in vitro* cytotoxicity,*in vivo* pharmacokinetic and oral toxic, proved IPN to be a safe and effective drug delivery vehicle for oral administration. Further, in support to its treatment towards colon cancer, the results of*in vivo*anti tumor activity exhibiteddecrease in the tumor size andwith reference tothe buoyancy of the microbeads, gamma (γ) scintigraphy studywas alsoconducted.Thus, all these studies proved IPN to be an effective vehicle for the prolonged release of CAP from the microbeads.