MEDICINAL PRODRUGS APPROACHES FOR COLON TARGETED RELEASE: A REVIEW

*Singh Deepak, Kumar Anoop and Shrivastava A. K.

*Department of Pharmaceutical Chemistry, Nandini Nagar Mahavidyalaya College of Pharmacy, Nawabganj, Gonda- 271303, U.P; India.

ABSTRACT

In the conventional therapeutic system the oral delivery of drugs are most easily accepted route of administration but gastrointestinal tract (GIT) may create several hindrance to delivery of therapeutic drugs at specific site like colon hence colon targeted drugs delivery approaches now a days widely accepted for treatment if disorder associated with colon like IBDs, Chron’s disease, ulcerative colitis, colorectal cancer and amoebiasis. Numerous prodrugs have been designed and developed since then to overcome pharmaceutical and pharmacokinetic barriers (degradation, release and absorption in the upper portion of the GI tract) and then to be ensured controlled release in the proximal colon in clinical drug application. This strategy increases the selectivity of drugs for intended target and there is a lower chance for attack on healthy cells with reduce side effect.

KEYWORDS: Prodrugs, IBDs, CTDDS, Medicinal approach.

INTRODUCTION

Prodrugs are inactive or less active bioreversible derivatives of active molecules which under gone enzymatic or chemical biotransformation and release the active drug moiety in the specific target tissue. Also for colon targeted release, can not only reduce the dose to be administered, but also decrease the adverse effects.[1,2] The colon is the highly efficient part of lower GI tract. The colon is being investigated as a site for administration of protein and peptides, which are degraded by digestive enzymes in the upper GIT. To achieve successful colonic delivery, a drug needs to be protected from absorption and the environment of the upper gastrointestinal tract (GIT) and then be abruptly released into the proximal colon,
which is considered the optimum site for colon-targeted delivery of drugs.\[^{1}\]\ One of the approaches used for colon specific drug delivery is the formation of prodrugs which optimizes drug delivery and improves drug efficacy. Many prodrugs have been evaluated for colon drug delivery. These prodrugs are designed to pass intact and unabsorbed from the upper GIT and undergo biotransformation in the colon releasing the active drug molecule. This biotransformation is carried out by a variety of enzymes, mainly of bacterial origin present in the colon e.g. azoreductase, β-glucuronidase, glycosidase, dextranase, esterase, nitroreductase, cyclodextranase.\[^{3,4}\]\ The classical example of colon specific prodrug is azo-prodrug of anti-inflammatory agent 5-aminosalicylic acid (5-ASA) i.e salicylazosulphapyridine (SASP) is bioactivated to 5-ASA and sulphapyridine by azoreductase, however sulphapyridine shows toxic adverse effect so similar azo-prodrugs are prepared such as Sulfasalazine, ipsalazide and olsalazine have been developed as colon-specific delivery systems for the treatment of inflammatory bowel diseases (IBDs).\[^{5,6}\]\ A number of colonic disease such as Inflammatory bowel diseases (IBDs) Crohn's diseases, ulcerative colitis, colorectal cancer and amoebiasis are still lacking appropriate medical treatment. The drugs which are given generally have poor bioavailability at the colonic site hence colon targeted prodrugs approach is designed.\[^{7}\]\ The prodrug approach, a chemical approach using reversible derivatives, can be useful in the optimization of the clinical application of a drug. The prodrug approach gained attention as a technique for improving drug therapy in the early 1970s.\[^{8}\]\ The colon specific drug delivery System (CDDS) is beneficial not only for the oral delivery of proteins and peptide drugs (degraded by digestive enzymes of stomach and small intestine) but also for the delivery of low molecular weight compounds used to treat diseases associated with the colon or large intestine such as ulcerative colitis, diarrhoea and colon cancer.\[^{9}\]\ Clinically relevant bioavailability may be achieved if the peptide can be protected from acid and enzymes in the stomach and upper intestine.\[^{9}\]\ The colon is having high water absorption capacity, the colonic contents are considerably viscous and their mixing is not efficient, thus availability of most drugs to the absorptive membrane is low. The human colon has over 400 distinct species of bacteria as resident flora, a possible population of up to 1010 bacteria per gram of colonic contents. Among the reactions carried out by these gut flora are azo reduction and enzymatic cleavage i.e. glycosides. These metabolic processes may be responsible for the metabolism of many drugs and may also be applied to colon-targeted
delivery of peptide based macromolecules such as insulin by oral administration. This strategy increases the selectivity of drugs for intended target and there is a lower chance for attack on healthy cells with reduce side effect.

**What is the concept of Prodrug?**
The term "prodrug" or "proagent" was first introduced by Albert to signify pharmacologically inactive chemical derivatives that could be used to alter the physicochemical properties of drugs in a temporary manner, to increase their usefulness and/or to decrease associated toxicity. Since Albert discussed the concept of prodrugs in the late 1950s, such compounds have also been called “bioreversible derivatives,” ”congeners,” and ”latentiated drugs” but now ”prodrug” is the most commonly accepted term. The prodrug approach can be applied to a variety of drug molecules, administration routes and formulations and is one way to improve the physicochemical, pharmaceutical and biopharmaceutical properties of pharmacologically potent structures and to overcome barriers to a drug's usefulness.

Prodrugs are well established as a strategy to:

- Improve solubility.
- Improve permeability.
- Improve drug targeting (eg. targeted drug delivery to organ such as colon, liver and brain etc.).
- Improve stability.
- Lower presystemic metabolism.

The characteristics of prodrugs must be improved for site specific drug delivery:

- The prodrug must be readily transported to site of action.
- The prodrug must be selectively cleaved to active drug utilizing specific enzymes.

Once prodrug is selectively generated at site of action, the tissue must retain the active drug without further degradation.

**Types of prodrug**
Silverman classified the ‘pro-drug’ into three categories.
1. Carrier - linked prodrugs

These include drug that attached via a metabolically labile linkage to another molecule, pro-moiety, which is not so necessary for activity but may impart certain desirable characteristic feature to the ‘drug’ viz., (a) site-directed delivery (b) enhanced water/lipid solubility.

These invariably comprise:

- Ideally the group removed is pharmacologically inactive and nontoxic while the connecting bond must be labile for efficient activation in vivo.
- Contain a group that can be easily removed enzymatically (such as ester) to reveal the true drugs.
- Bipartate - Composed of one carrier attached to the drug.
- Tripartate – Carrier group is attached via linker to drug.

2. Mutual Pro-drugs

The term was first use by wise in 1981. A mutual prodrug consists of two pharmacological active agents coupled together so that each acts as a pro-moiety for the other agent and vice versa. Here, the carrier used is another biological active drug instead of some inert molecule and may have the same biological action as that of the parent drug and thus might give synergistic action, or the carrier may have some additional biological action that is lacking in the parent drug. The carrier may also be a drug that might help to target the parent drug to a specific site or organ or cells or may improve site specificity of a drug.

3. Polymeric prodrugs

A conjugation of a drug with a polymer forms so-called “Polymeric prodrug”. Drug is incorporated into the polymeric system without formation of covalent bond between drug and polymer. The system also referred to as macromolecules prodrug system. Most commonly used polymers are proteins such as antibodies and lipoproteins, polypeptides as polylysine and polyglutamic acid, synthetic polymers as copolymers of N-vinyl pyrrolidine and polysaccharides as gum, dextran, starch, etc. are used. Polymeric conjugates of conventional drugs (polymeric prodrugs) have several advantages over their low molecular weight precursors. The main advantages of these areas:

a) An increase in H₂O solubility of low soluble or insoluble drugs and thereby, enhance bioavailability of drug.

b) An improvement in pharmacokinetics.
c) The ability to provide passive or active targeting of the drug specifically to the site of its action.

d) Protection of drug from deactivation and preservation of its activity during circulation, transport to targeted organ or tissue.

e) The possibility to form an advanced complex drug delivery system, with, in addition to drug and polymer carrier, may include several other active components that enhance the specific activity of the main drug.

Due to above advantages over free form of a drug, the polymeric prodrug conjugates has lead into a new era of a polymeric drug delivery system (PDDS). Three major types of polymeric prodrugs are currently being used.

- Prodrugs of the first type are broken down inside cells to form active substance.
- The second type of prodrug is usually the combination of two or more substances. Under specific intracellular conditions, these substances react forming an active drug.
- The third type of prodrug, targeted drug delivery system, usually includes three components: a targeting moiety, a carrier and one or more active components.

**What is colon targeted drug delivery system?**

The colon is a site where both local or systemic drug delivery could be achieved. In addition to restricted therapy, the colon can also be utilized as a portal for the entry of drugs into the systemic circulation. For example, molecules that are degraded/poorly absorbed in the upper gut, such as peptides and proteins, may be better absorbed from the more benign environment of the colon. Overall, there is less free fluid in the colon than in the small intestine and hence, dissolution could be problematic for poorly water-soluble drugs. In such instances, the drug may need to be delivered in a pre-solubilized form, or delivery should be directed to the proximal colon, as a fluid gradient exists in the colon with more free water present in the proximal colon than in the distal colon. Aside from drug solubility, the stability of the drug in the colonic environment is a further factor that warrants attention. The drug could bind in a nonspecific manner to dietary residues, intestinal secretions, mucus or general faecal matter, thereby the concentration of free drug. Moreover, the resident micro-flora could also affect reducing colonic performance via degradation of the drug.
Colon-specific formulation could also be used to prolong the drug delivery. It should be considered as beneficial in the treatment of colon diseases eg. Ulcerative colitis, crohn’s disease etc. A number of others serious diseases of the colon, e.g. colorectal cancer, might also be capable of being treated more effectively if drugs were targeted to the colon. Colonic drug delivery has gained increased importance not just for the delivery of the drugs for the treatment of local diseases associated with the colon but also for its potential for the delivery of proteins and therapeutic peptides.

Why CDDS is needed?
- Site-specific or targeted drug delivery system would allow oral administration of peptide and protein drugs, colon-specific formulation could also be used to prolonged the drug delivery.
- Colon-specific drug delivery system is considered to be beneficial in the treatment of colon diseases.
- Colon targeted drug delivery would ensure direct treatment at the disease site, lower dosing and fewer systemic side effect.

Table 1. Colon targeting disorders, Drugs and Sites\[12\]

<table>
<thead>
<tr>
<th>Target sites</th>
<th>Disease conditions</th>
<th>Drug and active agents</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Topical action</strong></td>
<td>Inflammatory Bowel Diseases, Irritable bowel diseaseCrohn’s disease</td>
<td>Hydrocortisone, Budenoside, Prednisolone, Sulfasalazine, Olsalazine, Mesalazine, Balsalazide</td>
</tr>
<tr>
<td><strong>Local action</strong></td>
<td>Chronic pancreatitis Cystic fibrosis Colorectal cancer</td>
<td>Digestive enzyme supplements 5-Flourouracil</td>
</tr>
<tr>
<td><strong>Systemic action</strong></td>
<td>To prevent gastric irritation To prevent first pass metabolism of orally ingested drugs Oral delivery of peptides Oral delivery of vaccines</td>
<td>NSAIDS Steroids Insulin Typhoid</td>
</tr>
</tbody>
</table>

Factors to be considered in the design of CDDS

Anatomy of colon

The large intestine extends from the distal end of the ileum to the anus. Human large intestine is about 1.5 m long.\[13\] The colon is upper five feet of the large intestine and mainly situated in the abdomen. The colon is a cylindrical tube that is lined by moist, soft pink lining called mucosa; the pathway is called the lumen and is approximately 2-3 inches in diameter.\[14\] The cecum forms the first part of the colon and leads to the right colon or the ascending colon
(just under the liver) followed by the transverse colon, the descending colon, sigmoid colon, rectum and the anal canal (Figure 1). The physiology of the proximal and distal colon differs in several respects that have an effect on drug absorption at each site.

![Figure 1: Anatomical features of Small and Large Intestine.](image)

**Table-2: Measured Surface area of GIT region**

<table>
<thead>
<tr>
<th>SN</th>
<th>Region of GIT Surface area/length</th>
<th>Region of GIT Surface area/length</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td><strong>Total GIT</strong></td>
<td>2-106 cm²</td>
</tr>
<tr>
<td>2.</td>
<td><strong>Small intestine</strong></td>
<td></td>
</tr>
<tr>
<td>2.1</td>
<td>Duodenum</td>
<td>20-30 cm</td>
</tr>
<tr>
<td>2.2</td>
<td>Jejunum</td>
<td>150-250 cm</td>
</tr>
<tr>
<td>2.3</td>
<td>Ileum</td>
<td>200-350 cm</td>
</tr>
<tr>
<td>3.</td>
<td><strong>Large intestine</strong></td>
<td></td>
</tr>
<tr>
<td>3.1</td>
<td>Cecum</td>
<td>6-7 cm</td>
</tr>
<tr>
<td>3.2</td>
<td>Ascending colon</td>
<td>20 cm</td>
</tr>
<tr>
<td>3.3</td>
<td>Descending colon</td>
<td>45 cm</td>
</tr>
<tr>
<td>3.4</td>
<td>Transverse colon</td>
<td>30 cm</td>
</tr>
<tr>
<td>3.5</td>
<td>Sigmoid colon</td>
<td>12 cm</td>
</tr>
<tr>
<td>4.</td>
<td><strong>Rectum</strong></td>
<td>3 cm</td>
</tr>
</tbody>
</table>

**pH level in stomach and intestine**

Generally, the release and absorption of orally administered drugs are influenced by the GI pH. In stomach, the pH is 1.5 - 2 in fasted and 3-6 in fed condition. The changes in the pH along the gastrointestinal tract have been used as a means for targeted colon drug delivery. Radio telemetry shows the highest pH (7.5 ±0.5) in the terminal ileum. On entry into the colon, the pH drops to 6.4 ± 0.6. The pH in the mid colon is 6.6 ± 0.8 and in the left colon 7.0 ± 0.7. There is a fall in pH on entry into the colon due to the presence of short chain fatty acids arising from bacterial fermentation of polysaccharides. For example lactose is
fermented by the colonic bacteria to produce large amounts of lactic acid resulting in pH drop to about 5.[17,18,19]

Table 3: pH at different sites of GI tract.

<table>
<thead>
<tr>
<th>Position</th>
<th>Site</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oral cavity</td>
<td></td>
<td>6.2-7.4</td>
</tr>
<tr>
<td>Oesophagus</td>
<td></td>
<td>5.0-6.0</td>
</tr>
<tr>
<td>Stomach</td>
<td>Fasted condition</td>
<td>1.5-2.0</td>
</tr>
<tr>
<td></td>
<td>Fed condition</td>
<td>3.0-5.0</td>
</tr>
<tr>
<td>Small intestine</td>
<td>Jejunum</td>
<td>5.0-6.5</td>
</tr>
<tr>
<td></td>
<td>Ileum</td>
<td>6.0-7.5</td>
</tr>
<tr>
<td>Large intestine</td>
<td>Right colon</td>
<td>6.4</td>
</tr>
<tr>
<td></td>
<td>Mild &amp; left colon</td>
<td>6.0-7.6</td>
</tr>
</tbody>
</table>

Microflora in Colon

The human alimentary canal is highly populated with aerobic and anaerobic bacteria and other microflora throughout the entire length. The upper region of the GIT has a very small number of bacteria. The concentration of bacteria in the stomach is usually less than $10^3$ colony forming units/ml (CFU/ml). The microflora of the proximal small bowel is similar to those of the stomach. In the distal part of the small intestine, a higher concentration of anaerobic bacteria is found. The concentration of bacteria in the human colon is $10^{11} - 10^{12}$ CFU/ml. The bacterial flora of the colon is predominantly anaerobic and composed of more than 400 strains. The important anaerobic bacteria are *Bacteroides*, *Eubacterium*, *Peptococcus*, *Ruminococcus* and *Clostridium*. The metabolic actions of the microflora are influenced by various factors such as age, intake of drugs, gastrointestinal disease and fermentation of dietary residues and can lead to inactivation of drugs or the enhancement of the action and side effects of the drugs.[20]

Table 4: Different microflora their enzymes released and action.

<table>
<thead>
<tr>
<th>Microorganism Enzyme</th>
<th>Microorganism Enzyme</th>
<th>Microorganism Enzyme</th>
</tr>
</thead>
<tbody>
<tr>
<td>Metabolic reaction</td>
<td>Metabolic reaction</td>
<td>Metabolic reaction</td>
</tr>
<tr>
<td>Clostridia, Lactobacilli</td>
<td>Hydrogenase</td>
<td>Reduces carbonyl groups &amp; aliphatic double bonds</td>
</tr>
<tr>
<td>Clostridia, Eubacteria</td>
<td>Glucosidase</td>
<td>Cleavage of β-glycosidase of alcohols &amp; phenols</td>
</tr>
<tr>
<td>E.coli, Bacteroids</td>
<td>Nitroreductase</td>
<td>Reduces aromatic &amp; heterocyclic nitro compounds</td>
</tr>
<tr>
<td>Eubacteria, Clostridia, Streptococci</td>
<td>Sulfatase</td>
<td>Cleavage of O-Sulphates &amp; Sulfamates</td>
</tr>
</tbody>
</table>
Gastrointestinal transit
Drug delivery to the colon upon oral administration depends mainly on gastric emptying and bowel transit time. Compared to other regions of the gastrointestinal tract, movement of materials through the colon is slow. The total time for transit tends to be highly variable and influenced by a number of factors such as diet, in particular dietary fiber content, mobility, stress, disease and drugs. Colonic transit times ranged from 20 to 35 hours. Stool weights increased significantly with the presence of active disease presumably due to exudates from inflamed epithelium, increased mucus secretion and reduction in reabsorption of fluid and electrolytes.\(^{[21,22]}\)

Table 5: Transit time of different parts of GIT.

<table>
<thead>
<tr>
<th>Part of GIT</th>
<th>Transit time</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stomach</td>
<td>Fasted state 10min – 2hr,</td>
</tr>
<tr>
<td></td>
<td>Fed state &gt;2hr</td>
</tr>
<tr>
<td>Small intestine</td>
<td>3-4hr</td>
</tr>
<tr>
<td>Colon</td>
<td>20-35hr</td>
</tr>
</tbody>
</table>

Advantages of CDDS over Conventional Drug Delivery
Chronic colitis, namely ulcerative colitis and Crohn’s disease are currently treated with glucocorticoids and other anti-inflammatory agents.\(^{[23]}\) Administration of glucocorticoids namely dexamethasone and methyl prednisolone by oral and intravenous routes produce systemic side effects including adenosuppression, immunosuppression, cushinoid symptoms, and bone resorption.\(^{[24]}\) Thus selective delivery of drugs to the colon could not only lower the required dose but also reduce the systemic side effects caused by high doses.\(^{[25]}\)

Criteria for Selection of Drug for CDDS
CTDDS are drugs which show poor absorption from the stomach or intestine including peptides.

The drugs used in the treatment of irritable bowel diseases (IBDs), ulcerative colitis, diarrhea, and colon cancers are prominent for local colon delivery.\(^{[26]}\)

- Drugs used for local effects in colon against GIT diseases.
- Drugs poorly absorbed from upper GIT.
- Drugs for colon cancer Drugs that degrade in stomach and small intestine.
- Drugs that undergo extensive first pass metabolism.
- Drugs poorly absorbed from upper GIT.
- Drugs for targeting.

Factors affecting colon targeted drug delivery
- Physiological factors.
- Gastric emptying.
- pH of colon.
- Colonic microflora and enzymes.
- Pharmaceutical factors.
- Drug candidates.
- Drug carriers.

Drug Carrier is another factor which influences CDDS. The selection of carrier for particular drugs depends on the physiochemical nature of the drug as well as the disease for which the system is to be used. Factors such as chemical nature, stability and partition coefficient of the drug and type of absorption enhancer chosen influence the carrier selection. Moreover, the choice of drug carrier depends on the functional groups of the drug molecule.\textsuperscript{[27]} For example, aniline or nitro groups on a drug may be used to link it to another benzene group through an azo bond. The carriers, which contain additives like polymers (may be used as matrices and hydro gels or coating agents) may influence the release properties and efficacy of the systems.\textsuperscript{[28]}

Drugs used to treat irritable bowel disease (IBD) require local delivery at drug to colon e.g. sulfasalazine, olsalazine, mesalazine, steroids like fludrocortisone, budesonide, prednisolone and Dexamethasone.

- Drugs to treat colonic cancer require local delivery e.g. 5-fluourouracil, doxorubicin and methotrexate.
- To treat infectious diseases (amoebiasis & helminthiasis) - requires site specific delivery e.g. metronidazole, mebendazole and albendazole.
- To treat rheumatoid arthritis (NSAIDS), nocturnal asthma, angina require delay in absorption due to circadian rhythms.
- Drugs showing more selective absorption in colon than small intestine due to small extent of paracellular transport e.g. glibenclamide, diclofenac, theophylline, ibuprofen, metoprolol and oxyprenolol.
**Prodrug approach to colon targeted drug delivery system**

Colonic release has gained increased importance, over last two decades, not just for the drug delivery to the colon for the treatment of local diseases associated with the colon but also for its potentials for absorption of therapeutic peptides and proteins and other drugs. To achieve successful colonic delivery, a drug needs to be protected from its absorption or metabolism in the environment before the entry to ascending colon where it should release the drug and exert its local action or otherwise absorbed. For CDDS, one approach includes covalent linkage of drug with carrier and other include deliver of intact molecule to colon. Various approaches that can be exploited for the development of colon targeted drug delivery systems are mentioned below.

**Approaches used for site specific Colon drug delivery (CDDS)**

[A] - Medicinal approaches for CDDS$^{[29,30]}$

- Microbially triggered Prodrug approach for drug delivery to colon.
  a) Azo conjugates.
  b) Amide conjugates.
  c) Cyclodextrin conjugates.
  d) Dextran conjugates.
  e) Ester conjugates.
  f) Glycosidic conjugates.
  g) Glucuronide conjugates.
  h) Polypeptide conjugates.
  i) Polymeric conjugates.

[A] Medicinal approaches of colon prodrugs delivery

Microbially triggered prodrugs for drug delivery to colon

The microflora of colon is in the range of $10^{11}$-$10^{12}$ CFU/mL$^{[31]}$, consisting mainly of anaerobic bacteria, e.g. Bacteroides, Bifidobacteria, Eubacteria, Clostridia, Enterococci, Enterobacteria and Ruminococcus etc. This vast microflora fulfills its energy needs by fermenting various types of substrates that have been left undigested in the small intestine, e.g. di- and tri-saccharides, polysaccharides etc.$^{[29]}$ For this fermentation the microflora produces a vast number of enzymes like glucoronidase, xylosidase, arabinosidase, galactosidase, nitroreductase, azoreducatase deaminase and urea dehydroxylase because of the presence of the biodegradable enzymes only in the colon, the use of biodegradable...
polymers for colon-specific drug delivery seems to be a more site-specific approach as compared to other approaches.\textsuperscript{[32]} These polymers shield the drug from the environments of stomach and small intestine and are able to deliver the drug to the colon. On reaching the colon, they undergo assimilation by micro-organism or degradation by enzyme or break down of the polymer back bone leading to a subsequent reduction in their molecular weight and thereby loss of mechanical strength. They are then unable to hold the drug entity any longer.\textsuperscript{[33]}

**(a) Azo conjugates prodrugs conjugates approach for drug delivery to colon**

The indigenous microfloras are responsible for a wide variety of metabolic processes, including the reduction of nitro and azo groups in environmental and therapeutic compounds. The use of these azo compounds for colon-targeting has been in the form of hydrogels as a coating material for coating the drug cores and as prodrug. In this the drug is attached via an azo bond to a carrier. This azo bond is stable in the upper GIT and is cleaved in the colon by the azo-reductases produced by the microflora.

In 1942, Svartz discovered sulfasalazine; the sulfanilamide prodrug of 5-amino salicylic acid (5-ASA) which is effective in the treatment of rheumatoid arthritis and anti-inflammatory disease. It is a salicylazosulphapyridine (SASP) where sulphapyridine is linked to a salicylate by an azo bond after administration, only a small proportion of the dose is absorbed from the small intestine and the sulphasalazine reaches the colon intact in bulk. In the colon, the azoreductases cleave the azo bond releasing the drug, 5-ASA and the carrier sulphapyridine\textsuperscript{[34]} in figure. 2.

![Diagram](https://via.placeholder.com/150)

**Figure 2: Cleavage of sulfasalazine (1) into 5-aminosalicylic acid (2) and sulfapyridine (3).**
Figure 3: The chemical structure of SASP, Balsalazide, Ipsalazide and Olsalazine showing the azo bond cleavage by bacteria leading to formation of the active drug 5-ASA.

Azo prodrugs of 5-aminosalicylic acid with histidine, 4-aminosalicylic acid with L-tryptophan and D-phenylalanine and 5-aminosalicylic acid with L-tryptophan were synthesized by coupling L-histidine with salicylic acid for targeted drug delivery to the colon in inflammatory bowel disease. Therapeutic efficacy of the carrier system and the mitigating effect of the azo conjugate were evaluated in trinitrobenzenesulphonic acid induced experimental colitis model. It was found to be equally effective in mitigating the colitis in rats, as that of sulfasalazine, without the ulcerogenicity of 5-ASA and adverse effects of sulfasalazine.

Table 6: Azo Prodrugs conjugates approach for drug delivery to colon.

<table>
<thead>
<tr>
<th>Carrier</th>
<th>Drug investigated</th>
<th>Linkage hydrolysed</th>
<th>In vitro/in vivo model used</th>
<th>Performance of the Prodrug/conjugates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Azo conjugates Suphapyridine (SP)</td>
<td>5-ASA[^34]</td>
<td>Azo linkage</td>
<td>Human</td>
<td>Site specific with a lot of side effects associated with SP</td>
</tr>
<tr>
<td>5-ASA</td>
<td>5-ASA[^35]</td>
<td>Azo linkage</td>
<td>Human</td>
<td>Delivers 2 molecules of 5-ASA as compared to sulphasalazine</td>
</tr>
<tr>
<td>Sulfapyridine</td>
<td>4-ASA and 5-ASA[^36]</td>
<td>Azo linkage</td>
<td>Human</td>
<td>Potential candidate for IBD patients intolerant to pancreatitis induced by oral administration of 5-ASA</td>
</tr>
<tr>
<td>Phenols</td>
<td>4-ASA and 5-ASA[^37]</td>
<td>Azo linkage</td>
<td>Human</td>
<td>Colon- targeting carriers for management of inflammatory bowel disease</td>
</tr>
</tbody>
</table>

(b) Amide conjugates prodrugs conjugates approach for drug delivery to colon

<table>
<thead>
<tr>
<th>Carrier</th>
<th>Drug investigated</th>
<th>Linkage hydrolysed</th>
<th>In vitro/in vivo model used</th>
<th>Performance of the Prodrug/conjugates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sulphamethoxazole, Sulphanilamide</td>
<td>Ibuprofen,[41] Diclofenac, Flurbiprofen</td>
<td>Amide linkage</td>
<td>Rat</td>
<td>Showed better activity profile in terms of analgesic and anti-inflammatory activity as compared to their respective parent drugs.</td>
</tr>
<tr>
<td>Amino acids</td>
<td>Dexibuprofen,[42]</td>
<td>Amide linkage</td>
<td>Rat</td>
<td>The histo-pathological studies revealed less ulceration in the gastric region when treated with prodrugs.</td>
</tr>
<tr>
<td>Amino acids</td>
<td>Ketorolac,[43]</td>
<td>Amide linkage</td>
<td>Human</td>
<td>Such prodrugs can be considered for sustained release purpose.</td>
</tr>
<tr>
<td>Tyrosine, Glycine</td>
<td>Mefenamic acid,[44]</td>
<td>Amide linkage</td>
<td>Human</td>
<td>Prodrugs are better in action as compared to MA, and are advantageous in having fewer gastrointestinal side effects</td>
</tr>
<tr>
<td>Amino acid conjugates glycine</td>
<td>Salicylic acid,[45]</td>
<td>Amide linkage</td>
<td>Rabbit</td>
<td>Absorbed from upper GIT, though metabolized by microflora of large intestine.</td>
</tr>
<tr>
<td>Tyrosine/methionine</td>
<td>Salicylic acid,[46]</td>
<td>Amide linkage</td>
<td>Rabbit</td>
<td>Absorbed from upper GIT, though metabolized by microflora of large intestine.</td>
</tr>
<tr>
<td>L – Alanin/D-Alanine</td>
<td>Salicylic acid,[47]</td>
<td>Amide linkage</td>
<td>In vitro</td>
<td>Salicylic acid-l-alanine was hydrolysed to salicylic acid by intestinal microorganism but salicylic acid-D-alanine showed negligible hydrolysis.</td>
</tr>
<tr>
<td>Glycine</td>
<td>5-ASA,[48]</td>
<td>Amide linkage</td>
<td>In vitro</td>
<td>Prodrug was stable in upper GIT and was hydrolysed by cecal content to release 5-ASA.</td>
</tr>
</tbody>
</table>

(c) Cyclodextrin conjugates prodrugs approach for drug delivery to colon

Cyclodextrin are cyclic oligosaccharides consisted of 6 to 8 glucose units linked through $\alpha-1,4$ glucosidic bonds. It has been executed for improving pharmacokinetic profile of certain drugs for instance stability, bioavailability etc.
Table 8: Cyclodextrin Prodrugs conjugates approach for drug delivery to colon.

<table>
<thead>
<tr>
<th>Carrier</th>
<th>Drug investigated</th>
<th>Linkage/conjugate hydrolysed</th>
<th>In vitro/in vivo model used</th>
<th>Performance of the Prodrug/conjugates</th>
</tr>
</thead>
<tbody>
<tr>
<td>β-cyclodextrin</td>
<td>5-ASA[50]</td>
<td>β-cyclodextrin-drug conjugate</td>
<td>Rat</td>
<td>CyD-5-ASA conjugates may be used as prodrugs for colon-specific drug delivery system</td>
</tr>
<tr>
<td>β-cyclodextrin</td>
<td>Biphenyl acetic acid[51]</td>
<td>β-cyclodextrin-drug conjugate</td>
<td>Rat</td>
<td>Ester-type drug conjugate of beta-cyclodextrin may serve as a colon-targeting prodrug.</td>
</tr>
<tr>
<td>α- or β - cyclodextrin</td>
<td>Naproxen, Sulindac, Flurbiprofen[52]</td>
<td>Amide linkage, β-cyclodextrin-drug conjugate</td>
<td>Rat</td>
<td>Reduced glutathione (GSH) levels in colon tissues</td>
</tr>
</tbody>
</table>

(d) Dextran conjugates prodrugs approach for drug delivery to colon

Dextran act as a successful carrier molecule for the delivery of NSAIDs with reduced gastrointestinal effect. The dextran backbone was not degraded and no release of 5-ASA was detected in buffer solutions of 1.2 and 6.8 pH, which indicate higher chemical stability.

Table 9: Dextran Prodrugs conjugates approach for drug delivery to colon.

<table>
<thead>
<tr>
<th>Carrier</th>
<th>Drug investigated</th>
<th>Linkage/conjugate hydrolysed</th>
<th>In vitro/in vivo/ex vivo model used</th>
<th>Performance of the Prodrug/conjugates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dextran with succinate spacer</td>
<td>Budesonide[53]</td>
<td>Dextran-drug conjugate</td>
<td>In vitro</td>
<td>Colon specific delivery system for budesonide to increase the efficacy in the treatment of ulcerative colitis.</td>
</tr>
<tr>
<td>Dextran with succinate and glutarate spacer</td>
<td>Dexamethasone[54] Methylprednisolone</td>
<td>Dextran-drug conjugate</td>
<td>Rat</td>
<td>Dextran conjugates may be useful in selectively delivering glucocorticoids to the large intestine for the treatment of colitis</td>
</tr>
<tr>
<td>Dextran with peptide spacer</td>
<td>Methylprednisolone[55]</td>
<td>Dextran-drug conjugate</td>
<td>Rat –ex vivo</td>
<td>Hepatic immunosuppressive activities</td>
</tr>
<tr>
<td>Dextran</td>
<td>Ketoprofen[56]</td>
<td>Dextran-drug ester conjugate</td>
<td>Pig</td>
<td>Selective colon delivery of drugs possessing a carboxylic acid derivatives</td>
</tr>
</tbody>
</table>
Dextran with succinate and glutarate spacer | Dexamethasone[57] | Dextran-drug conjugate | Rat | Glucocorticoid-dextran conjugates as potential prodrugs for colon-specific delivery

(e) Ester conjugates prodrugs approach for drug delivery to colon

Table 10: Ester Prodrugs conjugates approach for drug delivery to colon.

<table>
<thead>
<tr>
<th>Carrier</th>
<th>Drug investigated</th>
<th>Linkage/conjugate hydrolysed</th>
<th>In vitro/in vivo/ex vivo model used</th>
<th>Performance of the Prodrug/conjugates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acrylic type</td>
<td>5-ASA[58]</td>
<td>Ester linkage</td>
<td>Rat</td>
<td>Controlled release preparations of 5-aminosalicylic acid to target colon.</td>
</tr>
<tr>
<td>N-hydroxymethyl succinimide acid[60]</td>
<td>Ester linkage</td>
<td>In vitro</td>
<td>Analgesic, anti-inflammatory activities and with less GIT toxicity than the parent drug.</td>
<td></td>
</tr>
<tr>
<td>alpha-CyD</td>
<td>Prednisolone[61]</td>
<td>Ester linkage</td>
<td>Rat</td>
<td>Rational design of steroid prodrugs for the colon-specific drug delivery system</td>
</tr>
</tbody>
</table>

(f) Glycosidic conjugates prodrugs approach for drug delivery to colon

Glycoside conjugates also form the foundation of a new CTDDS and the activity of glycosidase enzyme secreted by the colonic microflora plays an important role in this context. Due to hydrophilic nature of drug glycosides, they are absorbed from the upper GIT and once such a glycoside reaches the colon it can be hydrolyzed or acted upon by glycosidase and subsequent release of free drug takes place which gets absorbed by the mucosa of colon. Naturally found glycosides, e.g. the sennosides have been utilized for laxative action since ages. These sennosides are activated by colonic microflora to generate rhein, anthones which shows the desired laxative effect.[62] Dexamethason-21-β-glucoside and Prednisolone-21-β-glucoside were synthesized for colonic delivery of these steroids.[63]
Table 1: Glycosidic Prodrugs conjugates approach for drug delivery to colon.

<table>
<thead>
<tr>
<th>Carrier</th>
<th>Drug investigated</th>
<th>Linkage/conjugate hydrolysed</th>
<th>In vitro/in vivo/ex vivo model used</th>
<th>Performance of the Prodrug/conjugates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saccharine</td>
<td>Dexamethasone/Prednisolone[27]</td>
<td>Glycosidic linkage</td>
<td>Rat</td>
<td>Dexamethasone prodrug was site specific and 60% of oral dose reached the cecum.</td>
</tr>
<tr>
<td>Glucose/Galactose/Celllobioside</td>
<td>Dexamethasone[64] Prednisolone Hydrocortisone, Fludrocortisone</td>
<td>Glycosidic linkage</td>
<td>In vitro</td>
<td>Less hydrolysis of the prodrug was seen in contents of stomach and proximal small intestine</td>
</tr>
</tbody>
</table>

(g) Glucuronide conjugates prodrugs approach for drug delivery to colon

Glucuronide conjugation is one of the important mechanisms for the inactivation and clearance of a variety of drugs. Bacteria of the lower GIT secrete β-glucuronidase and can deglucuronidate a variety of drugs in the intestine.[65] Thus the deglucuronidation process results in the release of the active drug again and enables its reabsorption.

Table 12: Glucuronide Prodrugs conjugates approach for drug delivery to colon.

<table>
<thead>
<tr>
<th>Carrier</th>
<th>Drug investigated</th>
<th>Linkage/conjugate hydrolysed</th>
<th>In vitro/in vivo/ex vivo model used</th>
<th>Performance of the Prodrug/conjugates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glucuronide conjugates glucuronic acid</td>
<td>Naloxone/Nalmefene[66]</td>
<td>Glucuronide linkage</td>
<td>Rat</td>
<td>When given to morphine dependent rats, these reversed the GIT side effects caused by morphine without causing CNS withdrawal symptom because of activation in large intestine followed by a resultant diarrheas which excreted the prodrug</td>
</tr>
<tr>
<td>Glucuronide conjugates glucuronic acid</td>
<td>Budesonide[67]</td>
<td>Glucuronide linkage</td>
<td>Rat</td>
<td>Was found to be superior than budesonide itself for treatment of colitis</td>
</tr>
</tbody>
</table>

(h) Polypeptide conjugates prodrugs approach for drug delivery to colon

Due to the hydrophilic nature of polar groups like -NH2 and -COOH, that is present in the proteins and their basic units (i.e. the amino acids), they reduce the membrane permeability of amino acids and proteins. Various prodrugs have been prepared by the conjugation of drug molecules to these polar amino acids.[68] Non-essential amino acids such as tyrosine, glycine, methionine and glutamic acid were conjugated to salicylic acid (SA) successfully, which
show higher hydrophilicity with decrease membrane permeability property. By having these desirable features, these amino acid prodrugs were found to show majestic results with least absorption from the upper gastrointestinal tract and found appropriate for targeted salicylic acid delivery to colon.

Table 13: Polypeptide Prodrugs conjugates approach for drug delivery to colon.

<table>
<thead>
<tr>
<th>Carrier</th>
<th>Drug investigated</th>
<th>Linkage/conjugate hydrolysed</th>
<th>In vitro/in vivo/ex vivo model used</th>
<th>Performance of the Prodrug/conjugates</th>
</tr>
</thead>
<tbody>
<tr>
<td>L-lysine amino acid trans-ferulic acid</td>
<td>5-amino salicylic acid[^69]</td>
<td>Amino acid-drug conjugate</td>
<td>In vitro</td>
<td>Useful in Crohn disease treatment and the evaluation of its antioxidant activity</td>
</tr>
<tr>
<td>amino acid</td>
<td>Celecoxib[^70]</td>
<td>Amino acid-drug conjugate</td>
<td>In vitro</td>
<td>Improved therapeutic potency and cardiovascular toxicity for chemoprevention of colorectal adenomas</td>
</tr>
</tbody>
</table>

(h) Polymeric conjugates prodrugs approach for drug delivery to colon

The polymeric prodrug systems under investigation may have potential use in the management of IBD in future because the drug dose will be sufficiently low.[^71] Recent advance of polymeric prodrugs including the effect of polymeric carrier material on the drug loading, biocompatibility and renal excretion.[^72] The polymeric prodrugs, their in vivo degradation by enzymatic or chemical action to generate nontoxic and biocompatible byproducts for further metabolism or excretion by normal physiological pathways.

Table 14: Polymeric Prodrugs conjugates approach for drug delivery to colon[^75]

<table>
<thead>
<tr>
<th>Carrier</th>
<th>Drug investigated</th>
<th>Linkage/conjugate hydrolysed</th>
<th>In vitro/in vivo/ex vivo model used</th>
<th>Performance of the Prodrug/conjugates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dextran</td>
<td>Dexibuprofen[^73]</td>
<td>Dexibuprofen-dextran polymeric</td>
<td>In vitro</td>
<td>Improvement in aqueous solubility, increased therapeutic efficiency and reduction of gastrointestinal side effects</td>
</tr>
<tr>
<td>Chitosan</td>
<td>Metronidazole[^74]</td>
<td>Chitosan-metronidazole conjugates</td>
<td>In vitro</td>
<td>Targeting delivery to colon.</td>
</tr>
</tbody>
</table>

CONCLUSIONS

This has been concluded from our review that the colon is an interesting site for the delivery of acid labile drugs as well as proteins and peptides. There are several approaches for colon targeting, such as pH dependent, pressure dependent, enzyme dependent etc, but each of
these approaches has certain limitation in terms of site specificity, toxicity, uncertainty pattern of drug release due to change in gastro-luminal pH or due to change in enzyme population. These limitations can be overcome by the use of natural polymers or a combination of polysaccharide with synthetic polymers. These types of combinations have the greatest potential for colon specific delivery in terms of site specificity and safety. This article has described the application of various chemical and medicinal approaches for colon targeting that are non-toxic and that are selectively degraded in the colon. So, challenges in the future will be to find the suitable prodrugs or its combinations to obtain colon specific molecules that exhibit high microbial and pH degradability in the colon.

REFERENCES
20. Krishnaiah YSR., Satyanarayana S. In: Jain NK (Ed.) Advances in Controlled and Novel Drug Delivery New Delhi, India., 2001; 89-119.


