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Review Article

Carrier systems for the treatment of inflammatory bowel disease

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Abstract

This article reviews a number of drug carrier systems in use clinically or under development for the treatment of inflammatory bowel disease (IBD). The advantages and disadvantages of the various methods for delivering drugs to the inflamed colon are discussed, including physiological and pathophysiological problems. Local delivery of drugs to the colon was developed to improve efficacy and minimize side effect. Particularly, efforts have focused on developments for oral administration in order to adapt the carriers to the physiological requirements which would further increase the therapeutic efficacy and improve patient compliance. These conditions include variations in local pH, transit throughout the gastrointestinal tract, the potential role of gut microflora and drug dissolution in both the healthy and diseased large intestine. Effective and selective delivery of an otherwise nonspecifically acting drug could provide new therapeutic options in the treatment of IBD. New strategies for such treatments are also presented, including liposomal formulations, nanoparticles, bacterial cytokine expression and viral gene therapy.

Introduction

The 2 major types of inflammatory bowel disease (IBD) are ulcerative colitis (UC) and Crohn's disease (CD), and are usually characterized by the areas of the intestine that are affected. Both diseases are of unknown etiology. Transmural chronic inflammation may involve all intestinal segments in CD, with a focus usually in the distal parts of the small intestine. Mucosal inflammation is limited to the colon in UC. The natural course of both UC and CD consists of quiescent phases that are interrupted by relapses (1). The typical locations of UC and CD are shown in Figure 1. As can be seen, the site of inflammation in UC (Fig. 1a) extends to the more distal regions of the colon, whereas in CD (Fig. 1b) up to 40% of the colonic tissue is affected by the inflammation (2). In these cases, colonic drug delivery systems may be applicable in treating CD patients in addition to many UC patients.

Whereas the incidence of UC has remained stable at around 8-9/100,000 people, the incidence of CD has increased from below 1 to more than 5/100,000 per year during the last 3 decades. Although little is known about the inflammatory pathomechanisms specific to either CD or UC, a distinction between both diseases can often be made by clinical manifestations, endoscopic appearance or histologic characteristics. The immunology of mucosal inflammation in both diseases is characterized by an overshooting immune response to unknown antigens, probably luminal antigens. Several studies reported an increase in proinflammatory cytokines in patients apparently unable to adequately downregulate immune activation. More importantly, it appears that CD does not describe a single clinical entity. The development of certain disease characteristics (*e.g.*, fistula or stricture formation) varies between individuals and may be a constant characteristic that is not altered in the natural course of disease. From clinical observations, therefore, it can be concluded that disease subgroups may exist which could be distinct in disease pathophysiology and treatment response.

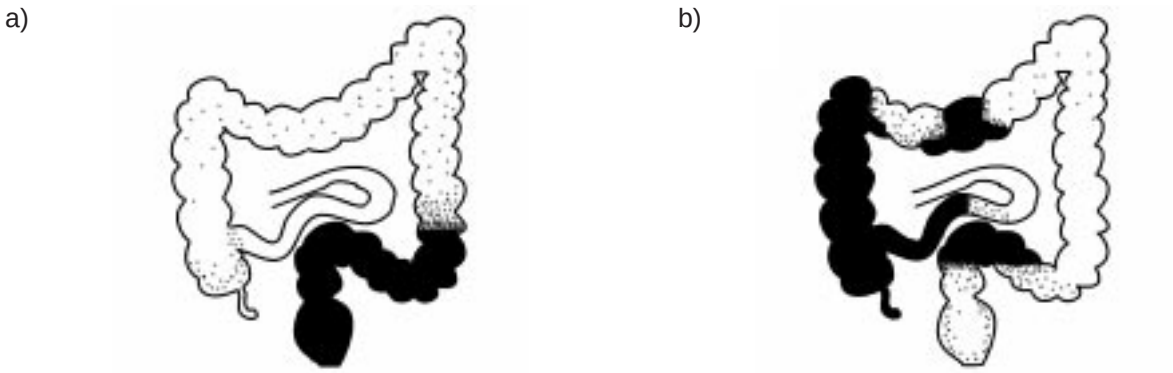

Fig. 1. Schematic representation of the large bowel. The most affected areas in ulcerative colitis (a) and Crohn's disease (b) are shaded in black.

The general principles of treatment of these diseases are to induce remission of outbreaks and to prevent outbreaks during remission. Available pharmaceutical products are 5-aminosalicylic acid preparations with different delivery profiles in the gastrointestinal tract, glucocorticoids and other immunosuppressants, especially azathioprine (3).

Topical glucocorticosteroids have become an alternative to standard glucocorticosteroids in ileocolonic CD since they cannot be used as maintenance therapy because of their well-known side effects such as Cushing's syndrome (4). The aim of glucocorticoid therapy of IBD, as for all other drugs, would be a potent local antiinflammatory effect in the mucosa with negligible amounts of the active agent absorbed into the systemic circulation to avoid systemic side effects. The inability to dissociate the antiinflammatory effects from the adverse effects of oral glucocorticoids has led to the development of topically active compounds. A good candidate is budenoside which is a relatively potent glucocorticoid with low oral bioavailability due to extensive first-pass hepatic metabolism (5, 6). The major interest for local delivery of glucocorticoids is their use in maintenance therapy, which is not yet possible.

Recent advances in the treatment of IBD have been characterized mainly by the more widespread use of immunosuppression. Azathioprine is currently used in CD with methotrexate as a second-line immunosuppressive drug (3, 7). Ciclosporin may become a drug of choice to treat severe UC but its long-term effects are probably insufficient (8). Beneficial effects in treating complicated, refractory CD have also been reported for thalidomide and tacrolimus (9-11).

Traditionally, 5-aminosalicylic acid (5-ASA) was considered effective for inducing a response in some patients with mild to moderate CD but has demonstrated little or no long-term benefit in maintenance therapy in controlled clinical trials. Glucocorticosteroid therapy is associated with higher rates of response in patients with active CD; however, clinical benefits are frequently offset by the common occurrence of corticosteroid-related toxicity. Oral controlled-release budesonide has demonstrated efficacy comparable to prednisolone with less risk for adverse effects, although many questions remain regarding the long-term use of this agent.

Sulfasalazine is a double molecule where the 5-ASA and sulfapyridine are linked together by an azo bond and it has been a standard in the treatment of UC for 60 years. Bacterial splitting of sulfasalazine within the colon allows delivery of 5-ASA for topical action (prodrug system). In UC, 5-ASA is effective in the treatment of mild to moderate acute attacks as well as for maintenance therapy. A meta-analysis showed a trend for 5-ASA to be superior to sulfasalzine for inducing remission, while the latter was more effective in maintaining remission. There are only a few studies comparing the efficacy of different new prodrugs.

In the past few years, new concepts have been formulated for the therapeutic management of difficult-to-treat IBD disease, based on better insights into the pathophysiology of the inflamed colonic mucosa. New immunomodulating agents with specific effects on intracellular processes involved in inflammation are now being developed, and infliximab, a TNF-α antibody, is now an accepted agent for use in severe, treatment-resistant cases of fistulizing CD.

The most significant development in recent years is the introduction of immunomodulatory therapy with cytokines and anticytokines. Clinical data show that the anti-TNF monoclonal antibody infliximab not only may result in rapid control of active CD but also may promote rapid tissue healing (12). Immunomodulatory therapy is very promising since early reset of the immunostat might be able to control inflammation in the long term (13).

Moreover, in the past few years several compounds have been developed which neutralize or impair the production of TNF-α such as the TNF receptor p75-Fc fusion protein, etanercept or p65 antisense oligonucleotide metalloproteinase inhibitors, as well as other compounds including CDP-571, rhIL-10 (14, 15) and opreleukin (rhIL-11) (16).

At present, successful treatment of active refractory CD has been reported with anti-TNF-α antibodies; more clinical studies are in progress or will be performed with

compounds that intervene in the activation, production and processing of TNF-α. However, important aspects of this type of immune intervention therapy still need to be elucidated (*e.g.*, long-term effects, identification of responders and nonresponders, *etc.*) (17).

After the colon-specific action of sulfasalazine was determined, attention was directed towards the colonic delivery of drugs and the design of colonic dosage forms. Because the delivery of peptide and protein drugs into the colon may enhance their oral bioavailability, drug delivery to the colon has become attractive to researchers interested in the delivery of peptide drugs to the large intestine. The degradation of those drugs has been reported to be reduced in this part of the intestine due to the estimated lower enzymatic activity in the mucosa (18). Thus, sustained-release dosage forms have been formulated delivering substantial drug amounts into the large intestine from where it is absorbed. Several reviews are available on the methods used in oral colonic drug delivery (19-22).

Progress has also been made in the development of drug carriers for the local treatment of IBD. Even if the aims are different, since one approach attempts to protect the drug during gastrointestinal transit to guarantee the most efficient absorption in the large bowel while the other wants to achieve a locally restricted effect, the general technologies and strategies are similar.

In order for a targeted drug delivery system to be efficient, the parameters of the disease that could influence the behavior of the carrier system must be taken into account. For example, the failure or success of a treatment could depend on different pH, motility and residence time at the inflamed tissues. Moreover, the different sites of inflammation in the gut are a challenge for the development of an appropriate delivery system and can also have an effect on the efficiency of drug delivery. Thus, a completely satisfactory therapeutic approach to the treatment of IBD is not yet available (23).

A wide range of antiinflammatory drugs are available for local administration. A major problem in IBD is the fact that the inflamed area is not easily accessible, so that any route of administration of these drugs would have the risk of severe systemic adverse effects due to the absorption of the free drug. Therefore, one strategy has been to increase the specificity of the drug itself, which has thus far not been accomplished. However, in most cases, local delivery systems can be used which further reduce the risk of adverse effects.

Prodrugs

The principle of prodrug development for the treatment of IBD is, in general, similar to other prodrug systems. A nonactive compound is cleaved by bacterial enzymes in the colon and the active part is subsequently delivered locally (24). Standard medical therapy for IBD is mainly restricted to aminosalicylates, corticosteroids and immunosuppressants. The choice of carrier depends on the functional group available on the drug molecule for conjugation with the carrier. For example, the hydroxyl group on corticosteroids can be used for a glycosidic linkage with various sugars (25); the carboxyl group of biphenylyl acetic acid can form an ester or amide conjugate with cyclodextrines (26). The most common example in this context may be sulfasalazine. In the large bowel, about 75% of this compound is split by bacteria into its 2 components. The sulfonamide was previously thought to have an antiinflammatory effect, but later experiments showed that this component is actually inactive and that mesalazine alone is the active component (27). The only function of sulfapyridine is to be the carrier for mesalazine. It prevents early absorption of this active part from the small intestine since splitting of the azo bond only occurs in the large bowel. Later it was shown that the inactive sulfonamide component is largely responsible for the high rate of adverse effects associated with sulfasalazine, especially the dose-dependent adverse reactions (28). A successful prodrug strategy has been followed for glucocorticoids as already mentioned above. When administered orally, dexamethasone-glucuronide was found to lead to higher intraluminal and mucosal tissue concentrations as compared to oral administration of pure dexamethasone which can be administered at a lower dose and subsequently causes less adverse effects (29). In general, similar strategies in the development of prodrugs can be followed by using amino-acid conjugates, glycoside conjugates, glucuronide and sulphate conjugates and azo conjugates (30).

Rectal administration

Rectally administered dosage forms were developed because of the well-known side effects of antiinflammatory drugs and because UC often affects distal parts of the colon. Enema formulations have been used extensively to treat distal UC or proctitis and proctosigmoiditis. In particular, glucocorticoids have been used effectively in these dosage forms to obtain a local therapeutic effect (31-34). One important issue associated with rectal preparations (enemas, foams, suppositories) is the ability of these dosage forms to reach only the lower part of the distal colon. Some studies have pointed out the limitations in the access range of 2 enema preparations of budesonide (35). Similar studies conducted with rectal foams and enemas containing mesalazine have shown that only the rectosigmoid region can consistently be reached in patients with active UC (36). Efficacy has been reported in UC patients for mesalazine-containing suppositories, especially when the rectum is involved. Satisfactory results were obtained in distal UC and proctosigmoiditis where symptoms improved within a few days (37). In contrast to foams and suppositories which are better accepted by UC patients, enemas and rectal preparations are generally unpopular (38, 39).

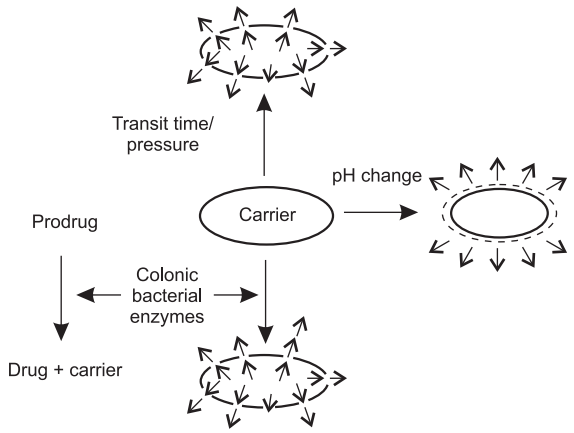

Fig. 2. Various standard strategies used in oral administration of antiinflammatory drugs.

Macroscopic carrier systems for oral colonic drug delivery

There are several approaches for delivering drugs to the large intestine after oral administration (Fig. 2). These include pH-dependent coatings designed to release drugs in the lower gastrointestinal tract; bioerodible coatings and matrices; enzyme-controlled drug delivery; and timed- and sustained-release dosage forms that release drugs as they pass through the small and large intestine.

Many colon-specific dosage forms have been developed, including cross-linked hydrogels, matrices and coated dosage forms. However, the synthesis of prodrugs is possible only if the drug has suitable moieties that can be bound to a carrier molecule, and with biodegradable hydrogels and matrices, slow degradation rates resulting in slow drug release often represent a problem especially for their use in IBD. A major disadvantage of these types of carriers appears to be the lack of consideration for the specific physiological changes occurring in IBD since they were mainly developed from a healthy human gut model. Therefore, use of prodrugs was recommended for maintenance therapy since they seem to be marginally better than slow-release preparations (40).

Detailed mechanisms of action of these delivery systems are addressed below in the context of factors that can affect their performance.

pH-Controlled drug release

Several commercial drug formulations designed for colon-specific drug delivery rely on the physiological difference between the luminal pH of the acidic stomach and that of the distal small intestine. If the intended site of drug release of the coated dosage form is the colon, one must take into consideration that, between the terminal ileum and the transverse or distal colon, there is a slightly acidic region in the proximal colon that may affect drug release profiles and the reproducibility of drug release.

If the terminal ileum or the colon is the target organ for the drug, the start of the drug release must be further controlled by increasing the thickness of the coating, allowing a pH- and time-controlled polymer dissolution and drug release process. Thus, drug release from many of the coated dosage forms designed for pH-controlled drug delivery to the terminal ileum or colon will also depend on the transit time through the small intestine. These dosage forms therefore usually represent combined pH- and time-controlled drug delivery systems for which time-controlled release can be achieved by selecting a suitable coating (41).

Both commercial and experimental delivery systems rely on differences in pH along the alimentary canal to control the site of drug release. Therefore, it is important to understand these differences and what role the disease state may play in the performance of these systems in patients. Local pH levels can affect intestinal enzyme activities and hence the performance of delivery systems that rely on these enzymes to trigger drug release from biodegradable dosage forms and from prodrugs.

Many commercial drug formulations for the oral treatment of IBD are coated with pH-sensitive enteric coating polymers such as Eudragit® L or S (Table I). These polymers have a dissolution pH of between 6 and 7, and are intended to release the drug as soon as the intestinal pH exceeds 6 or 7, respectively. Disintegration sites vary from the ileum to the splenic flexure, indicating a lack of site specificity. When dosage forms coated with Eudragit® S were used, which is designed to delay release of a drug until it reaches the terminal ileum or ascending colon, and given orally in tablet form to humans, drug release in the colon was not sufficiently reproducible. In accordance with the measured pH values along the length of the gastrointestinal tract, the tablets rapidly disintegrated at sites ranging from the ileum to the splenic flexure (Table II). The authors concluded that such data demonstrate a lack of site specificity (42, 43), although results from other studies support the feasibility of this targeting method.

The pH along the gastrointestinal tract is reasonably well characterized in normal subjects (44). There is a drop in pH to a mean of 6.4 ± 0.4 in the caecum. The pH remains between 6.4 and 7.0 from the ascending colon to the left colon. Only in the distal part of the colon is a pH of approximately 7 reached, and this differs only slightly from the average pH in the small intestine which is 6.5-6.8 (44, 45) (Fig. 3).

The luminal pH in patients with IBD can be lower than that measured in normal volunteers. The colonic pH ranged from 5.0 to neutral in some patients, while in 3 patients, the pH values were considerably lower (2.3, 2.9 and 3.4). Similar data have been collected in 4 CD patients (46, 47). Thus, certain developments in pH-dependent carriers seem to be applicable only in healthy subjects, since the normal pH of 6.4-7 may drop in the disease state to values between 2 and 5 and consequently no adequate drug release can be provided.

Table I: Coated dosage forms for the treatment of ulcerative colitis available on the German market.

Drug	Coating polymers	Dissol. pH	Trade name	Manufacturer
Mesalazine	Eudragit® L	6.0	Claversal®	SmithKline Beecham Pharm., Munich
Mesalazine	Eudragit® S	7.0	Asacolitin®	Henning Berlin GmbH & Co., Berlin
Mesalazine	Eudragit® L	6.0	Salofalk®	Dr. Falk Pharma GmbH, Freiburg
Mesalazine	Ethylcellulose	–	Pentasa®	Ferring Arzneimittel GmbH, Wittland
Sulfasalazine	Cellulose phthalate	6.2-6.5	Azulfidine®	Pharmacia & Upjohn GmbH, Erlangen
Sulfasalazine	Eudragit® L100-55	5.5	Colo-Pleon®	Henning Berlin GmbH & Co., Berlin
Budesonide	Eudragit® L100-55 ethylcellulose	5.5	Entocort®	ASTRA GmbH, Wedel
Budesonide	Eudragit®	6.0	Budenofalk®	Dr. Falk Pharma GmbH, Freiburg

From Leopold, C.S. *Coated dosage forms for colon-specific drug delivery.* Pharm Sci Technol Today 1999, 2: 197-204.

Table II: Gastrointestinal transit times (h) for Eudragit(R) S coated tablets.

Vol.	Gastric emptying	Time in lower small intestine	Time through ileocecal junction	Small intestine transit time	Time in ascending colon	Main position at 12 h
1	3.0	1.8	6.1	3.1	2.1	Hepatic flexure
2	1.6	>15	>15	>13	–	Ileocecal junction
3	1.4	5.5	8.7	7.3	0.5	Splenic flexure
4	6.1	3.6	10.2-12.0	5.1	–	Ascending colon
5	2.0	1.1	4.9	2.9	0.2	Hepatic flexure
6	1.3	>12	>12	>10	–	Ileocecal junction
7	1.3	0.0	5.0-5.5	4.0	0	Hepatic flexure

From Ashford, A. et al. *An in vivo investigation into the suitability of pH-dependent polymers for colonic targeting*. Int J Pharm 1993, 95: 193-9.

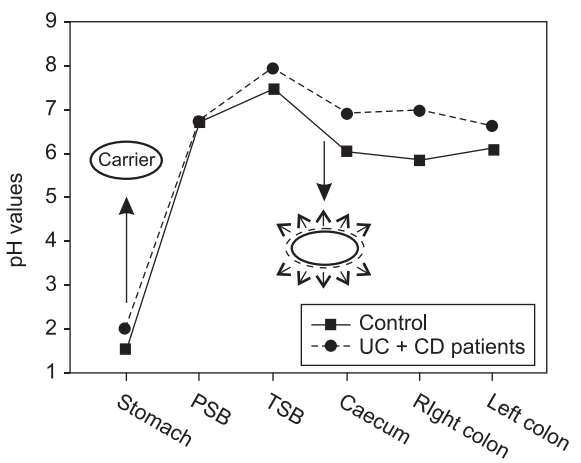

Fig. 3. pH profiles in ulcerative colitis (UC) and Crohn's disease (CD) patients compared with healthy volunteers. The pH-dependent drug release occurs when passing a certain zone in the gut as shown in the figure. PSB: proximal small bowel; TSB: terminal small bowel.

To address this problem, a formulation that releases the drug at an acidic pH was proposed (48), consisting of a drug core, an acid-soluble basic polymer layer such as aminoalkyl methacrylate copolymer (Eudragit® E) or polyvinyl-acetal diethylamino acetate and an enteric coating as a gastroresistant outer layer.

Time-controlled drug release

Drug release from a time-controlled colonic delivery system typically occurs after a predetermined lag time, which corresponds to or exceeds the small intestinal transit time, to ensure drug delivery to the large intestine. In the case of coated dosage forms designed for time-controlled drug release, the lag time usually depends on the coating thickness, and drug release can be triggered either by a change in pH, a change in the osmotic pressure or by disruption of the coating by swelling of the core.

Time-controlled drug release with pH-induced drug delivery is triggered by changes of pH within the dosage form itself, which is independent of the luminal pH of the gastrointestinal tract. Colon-specific drug formulations relying on the time-dependent dissolution of basic polymer layers under acidic conditions is one such system. Enteric Eudragit® E-coated gelatin capsules are filled with a solid organic acid that dissolves as soon as it comes into contact with the penetrating intestinal fluid initiating the dissolution of the coating material followed by the drug release (49, 50).

An oral swelling-induced drug delivery system consisting of a hydroxypropylmethylcellulose-coated drug core, covered by an Eudragit® L coating dissolves in the intestinal fluid (51, 52). The high viscosity inner layer begins to swell and slowly erodes over time after dissolution of the enteric coating.

Table III: Transit measurements and stool output during active and quiescent disease.

	Total colitis			Distal colitis		
	Active	Quiescent	P	Active	Quiescent	P
$T_{1/2}$ for gastric emptying (min)	46 ± 22	41 ± 6	n.s.	51 ± 23	78 ± 33	<0.05
Mouth-to-caecum transit (min)	298 ± 62	310 ± 60	n.s.	313 ± 87	293 ± 82	n.s.
Whole gut transit (h)	64 ± 22	56 ± 29	n.s.	68 ± 49	78 ± 55	n.s.
Mean daily stool weight (g)	253 ± 72	159 ± 62	<0.02	192 ± 99	144 ± 79	<0.05
Mean daily stool frequency	4.1 ± 1.0	1.9 ± 1.0	<0.002	3.0 ± 0.8	1.2 ± 0.7	<0.001

From Rao, S.S.C. et al. *Studies on the mechanism of bowel disturbance in ulcerative colitis.* Gastroenterology 1987, 93: 934-40.

A time-controlled explosion system consists of sucrose beads covered with a drug layer and coated with an inner layer of low substituted hydroxypropylcellulose and an outer ethylcellulose film (53). The intestinal fluid penetrates gradually through the outer ethylcellulose membrane and the swelling force of the hydroxypropylcellulose layer causes membrane disruption after a predetermined lag time. Drug release from the dosage form is pH-independent. Another time-controlled ethylcellulose capsule for colon-specific drug delivery is equipped with micropores on the bottom and basically consists of an ethylcellulose drug container (54).

Understanding the movement of a dosage form through the gastrointestinal tract in IBD patients is a prerequisite for the design of oral preparations targeted at the colon. If transit is very rapid through the gut, drug release may be insufficient and the therapy may not be effective. The average transit time of both pellets and tablets has been measured at about 3 ± 1 h (55). In contrast, the transit of tablets or nondisintegrating capsules through the colon tends to be more rapid than that of small pellets of about 1.0 mm in diameter (56, 57). Similar findings have been reported for different sizes of tablets, *e.g.*, smaller tablets (6 mm) were retained in the ascending colon longer than larger tablets (58). Results supporting this phenomenon of size dependency were reported from another study where large, single-unit dosage forms moved significantly faster through the proximal colon than did small pellets (59). The rapid transit rates of larger size dosage forms through the colon has been called streaming. Where distinct liquid and solid phases are present in the colon, the 2 phases can move at different rates so that there is streaming of contents into the colon (60).

Furthermore, gastrointestinal transit in IBD can vary from that in healthy state due to mucosal inflammation and the changes in the normal mechanism of absorption and transit. An additional common pathophysiological manifestation of UC patients is diarrhea. The inflamed colon presents abnormalities in fluid and electrolyte absorption and secretion (61). The physical consistency of stools is also important clinically and can potentially affect drug delivery to the colon based on an excess amount of fluid influencing certain swelling steps of the formulation. Transit data following oral administration of a radiolabeled liquid preparation in UC patients during active and quiescent disease are summarized in Table III. An evaluation of gastrointestinal transit in UC patients using actual pharmaceutical dosage forms showed that transit through the upper intestine was not distinctly different from that of the healthy subjects (62). The authors concluded that overall transit times were similar to those found in normal volunteers, although transit in the proximal colon of UC patients was on average slower than that in normal subjects. Accelerated transit through the rectosigmoid region of the colon results from distal irritability (63).

The transit of delivery systems can affect the site of drug release and exposure of the inflamed mucosa. Systems which are based on time-controlled release may be particularly susceptible to problems of variable transit times. The transit patterns of UC patients is theoretically less problematic for a delivery system based on enzymatic triggering of drug release. Due to proximal stasis, there should be adequate time for the degradation of the tablet coating or matrix. As the fecal material passes through the transverse and descending colon, maximal amounts of drug will be available for delivery into the inflamed mucosa. Greatly accelerated transit through the rectosigmoid region may, however, limit the amount of drug delivered to this region.

Enzyme-controlled drug release

Enzyme-controlled drug release relies on the presence of enzymes produced by microorganisms in the colon. The colonic microflora produce a variety of enzymes, including azoreductase, various glycosidases and, to lesser extent, esterases and amidases, that all can be exploited for colon-specific drug delivery. By taking advantage of these enzymes, prodrugs, hydrogels, matrices and biodegradable coating materials have been developed for enzyme-controlled drug release.

The regulatory requirements are a major drawback to the enzyme-based approach since each newly synthesized polymer must acquire federal agency approval. Biodegradable materials mainly comprise azo polymers, glycosidic polymers and matrices consisting of conventional polymers for sustained drug delivery, based on acrylate or cellulose polymers combined with biodegradable pore formers.

Azo polymers were the first coating materials to be investigated with regard to biodegradability in the colon.

The microbial degradation of these polymers is dependent on their hydrophilicity; however, sufficient resistance to gastric and intestinal fluids is observed only if the polymer is more lipophilic in nature (64, 65). The enzymatic degradation of the polymer and thus drug release depends on the degree of swelling.

There are several problems that need to be addressed when developing azo polymers for colon-specific drug delivery. Toxicity resulting from the microbial or chemical reduction of azo compounds to primary aromatic amines may be critical. The reduction of polymeric azo compounds often proceeds too slowly. Furthermore, reduction does not necessarily depend on the existence of the azoreductase, but can occur as a result of the low oxidation potential in the colon.

Various polysaccharides have been investigated for their potential suitability as colon-specific coating polymers. If galactomannans are used coating materials, the microbial degradation will be rapid if the mannose:galactose ratio is high. Therefore, galactomannans from locust bean gum with a mannose:galactose ratio of 4:1 appear to be suitable. However, the high water solubility of this polysaccharide has to be reduced by chemical modification. Another candidate is chitosan, which is a cationic polysaccharide susceptible to degradation by microbial enzymes in the colon. If used as a coating material for colon-specific drug delivery, it is important to protect the acid-soluble polymer with an enteric coating (66).

Polymers are structurally weakened when they arrive in the colonic region, allowing for swelling and subsequent fermentation of the amylose to occur which ultimately leads to drug release.

Galactomannans, such as guar gum, have been used as satisfactory coating films in combination with Eudragit® RL, RS and NE (67, 68). However, high amounts of the acrylic polymers and high coating levels are required to avoid an early drug release.

Although the commonly exploited enzymes such as azoreductase and various glycosidases are present only in the terminal ileum and colon and premature drug release does not occur, the activity of the microbial enzymes is even more susceptible to diet, drug intake (particularly antibiotics and certain laxatives) and environmental factors. Thus, reproducibility of enzymatic polymer degradation can be a problem, especially in the case of IBD.

Colonic enzymes vary in their activity during phases of disease activity leading to inter- and intrasubject variability. The proven therapeutic efficacy of several prodrugs suggests that variable enzymatic activity is probably of minor significance, especially when used in quiescent, slightly active disease. There are a number of studies evaluating the effect of the IBD state on gut microflora (69-71). In general, the types and concentration of gut microflora in UC are similar to those of healthy controls. Likewise, few differences in glycosidase activity are noted in UC patients when compared with healthy volunteers. In CD, however, differences have been noted both in concentration of microbes present and their enzyme activity (in general, glycosidase activity is reduced compared to healthy controls). Based on these observations, using an enzyme-based colonic delivery system is probably more feasible for treatment of UC rather than CD.

Regardless of the technological approach, any drug delivered into the colon must also be bioavailable even at the inflamed tissues of IBD patients. However, mechanical factors as well as hydrodynamic flow around tablet dosage forms are heterogeneous and subject to conditions of the colonic contents (72, 73). As a result, friction is progressively more important than other processes (such as intermolecular diffusion) in controlling flow as the dosage form and drug move down the intestine. Studies involving hydrodynamic flow reveal that when less viscous fluid moves towards fluid with a higher viscosity, interfacial ripples form, leading to meandering or fractal flow (74). These observations suggest that dissolution of poorly water soluble antiinflammatory drugs should be considered to occur in topologically constrained media. Thus, traditional criteria used to describe absorption of a drug from the colon may be unrealistic. The implications of such an approach in describing the highly variable and heterogeneous environment of the colon are as of yet unclear.

Microsize delivery forms

Due to the above mentioned streaming phenomenon, investigations were undertaken to reduce the size of the drug carrier systems by maintaining the conventional drug delivery strategies. In particular, microparticles were developed to slow down transit and prolong the drug release period.

Due to their bioadhesive properties, the use of chitosan microspheres was proposed for local release of prednisolone (75). The use of such an adhesive polymer would result in retention of the drug at the inflamed mucosal site and consequently a reduction in dose. Other strategies targeted simultaneous controlled drug release of a drug combination (76). In this case, biocompatible polyesters were used in order to avoid toxicological problems when carriers are taken up into the ruptures at the inflammation site.

Another system consists of budenoside-loaded cellulose acetate butyrate microspheres coated by an enteric polymer such as Eudragit® S. The *in vitro* drug release studies of pH-sensitive microcapsules containing the hydrophobic cores showed that no drug was released below pH 7 (77, 78). The use of the new budenoside-containing microparticulate delivery system significantly reduced the colon/body weight ratio compared with control formulations. Similarly, myeloperoxidase activity and macroscopic and histological damage to the inflamed colonic segments significantly decreased when the budenoside formulation was used as compared to oral administration of the drug suspension. However, there were no significant differences between the new system

and the control formulation consisting of simple enteric microparticles (79).

The combination of intravenous and soft gelatin-capsule ciclosporin is used to treat severe steroid-refractory UC. However, since it is associated with major toxicity, an oral microemulsion formulation was developed. The new formulation proved to be effective in achieving short-term remission in patients with steroid-refractory UC. Adverse effects were also reduced and there were no withdrawals from treatment (80-82).

New therapeutic strategies

Liposomes are used as a contrast agent in IBD (83) and even more interesting, in a liposomal drug delivery system for IL-4 and IL-10. Studies have shown that increased concentrations of IL-4 and IL-10 following rectal administration of these liposomal preparations were able to reduce TNF-α tissue concentrations (84). Recently, preliminary results were obtained from transfection studies using adenovirus carriers. The IL-10 gene was injected i.p. and was found to be efficient in preventing, but not in reversing, experimental colitis in rats and mice (85, 86). Further carrier systems, *e.g.*, cationic liposomes or *in vitro* transduced cells, have been proposed in gene therapeutic drug targeting for other autoimmune diseases and could be potential new approaches in the treatment of IBD as well (87). Another new strategy recently proposed for the treatment of IBD is IL-10-secreting bacteria. Intragastric administration of IL-10-secreting *Lactococcus lactis* caused a 50% reduction in colitis in mice treated with dextran sulfate sodium and prevented the onset of colitis in IL-10$^{-/-}$ mice (88).

Several studies have implicated macrophages and dendritic cells in active IBD. Manipulation of these cells is considered a very important therapeutic strategy for patients with IBD since it was reported that biodegradable microspheres can be sufficiently taken up by macrophages (89). Based on these findings, the direct uptake of antiinflammatory agents by macrophages (via microspheres) appears to have superior immunosuppressive effects and to be more effective for the treatment of patients with IBD (90, 91). Other studies have described the use of agents targeted at macrophages that rupture inside the ulcerated tissue in IBD for specific local drug delivery (92, 93). Such ruptures and the involved macrophage extravasation appear to be a very promising strategy (Fig. 4). The aim of this strategy is to selectively accumulate the drug delivery system at the site of action. Polymeric nanoparticulate carrier systems were expected to target the inflamed tissue in IBDs. Since no sedimentation occurs with colloidal drug carriers, they might be affected to a lesser extent or not at all by the streaming due to their diffusing properties. With the antiinflammatory drug rolipram, all drug-loaded nanoparticle formulations proved to be as effective as the drug in solution in mitigating experimental colitis. Inflammation activity scores and myeloperoxidase activity decreased significantly after oral administration of rolipram nanoparticles or solution. When drug administration was discontinued, those animals treated with the drug solution relapsed, whereas animals treated with the nanoparticles maintained reduced levels of inflammation. Moreover, when the free drug was administered, the rolipram solution group exhibited a high adverse event index, while the rolipram nanoparticles group had a significantly reduced index, demonstrating their potential to retain the drug from systemic absorption. An essential advantage of this strategy is the direct contact of the carriers with the inflammation site which allows for a much higher local drug concentration. Moreover, nanoparticles were found not only to accumulate in the ulcer but also to adhere to the mucus, allowing for an increased specificity to nonulcerated inflamed tissue since mucus production is significantly increased in inflamed tissue.

Fig. 4. Microscopic image of colon tissue from rats with experimental colitis showing the increased mucus layer on the mucosa and typical ulceration in the colon. The ulcerated area exhibits a distinct macrophage extravasation while mucosal cells show abundant mucus production (black areas stained by Astra blue).

These new delivery systems permit the desired drug to accumulate in the inflamed tissue with relatively high efficiency, which has 2 major advantages. The drug is concentrated at its site of action, which reduces possible adverse effects and enhances the effect of the administered dose. Moreover, the sustained drug release prolongs pharmacological effects due to the extended time the carrier system is present at the targeted inflamed area. This deposition of polymeric carrier systems in the inflamed tissue may be promising for the design of new carrier systems for the treatment of IBD.

Conclusions and perspectives

The ability to deliver an antiinflammatory drug into the large intestine during active ulcerative colitis or Crohn's disease depends on a number of factors, including interactions between the mechanism of drug delivery and the variables of pH, transit and gut microflora. All colonic drug

delivery systems rely to some extent on one or more of these variables to trigger drug release locally. In particular, the transit characteristics in the colon during active disease represent a challenge for adequate amounts of drug to reach the inflamed tissues after oral administration. In general, coated dosage forms with a simple design are suitable systems because of their ease of manufacture. More complex systems can cause problems with regard to production and reliability. The fact that colonic pH can drop dramatically in IBD has not been taken into consideration with the formulations that are currently available on the market for this indication. Finally, the traditional concepts of drug dissolution and local bioavailability may not be applicable, based on knowledge of the physical milieu of the colonic lumen both in the healthy and diseased state. Therefore, new therapeutic strategies must be promoted in order to solve problems based on physiological influences of the disease state, which are only just now being taken into account.

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