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Vivek S. Dave

St. John Fisher University, vdave@sjf.edu

Deepak Gupta

Lake Erie College of Osteopathic Medicine

Monica Yu

Lake Erie College of Osteopathic Medicine

Phong Nguyen

Lake Erie College of Osteopathic Medicine

Sheeba Varghese Gupta

University of South Florida

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Abstract

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Keywords

Permeability, BCS Class III, Prodrugs, Ion-pairing, Counterion, Nanotechnology, Liposome, Permeation enhancer

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Current and evolving approaches for improving the oral permeability of BCS Class III or analogous molecules

Vivek S. Dave^{1*}, Deepak Gupta^{2*}, Monica Yu², Phuong Nguyen², Sheeba Varghese Gupta³

¹St. John Fisher College, Wegmans School of Pharmacy, Rochester, NY, USA

²Lake Erie College of Osteopathic Medicine, School of Pharmacy, Pharmaceutical Sciences
Bradenton, FL, USA

³USF College of Pharmacy, Department of Pharmaceutical Sciences, Tampa, FL, USA

* Both authors contributed equally to this article.

Abstract

The Biopharmaceutics Classification System (BCS) classifies pharmaceutical compounds based on their aqueous solubility and intestinal permeability. The BCS Class III compounds are hydrophilic molecules (high aqueous solubility) with low permeability across the biological membranes. While these compounds are pharmacologically effective, poor absorption due to low permeability becomes the rate-limiting step in achieving adequate bioavailability.

Several approaches have been explored and utilized for improving the permeability profiles of these compounds. The approaches include traditional methods such as prodrugs, permeation enhancers, ion-pairing, etc., as well as relatively modern approaches such as nanoencapsulation and nanosizing. The most recent approaches include a combination/hybridization of one or more traditional approaches to improve drug permeability. While some of these approaches have been extremely successful, i.e. drug products utilizing the approach have progressed through the USFDA approval for marketing; others require further investigation to be applicable. This article discusses the commonly studied approaches for improving the permeability of BCS Class III compounds.

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Introduction

The Biopharmaceutics Classification System (BCS) of drugs has been developed primarily to provide a scientific approach for classifying oral immediate release drug formulations based on aqueous solubility and intestinal permeability, in combination with the dissolution properties [1]. Pharmaceutical compounds that belong to the BCS Class III are predominantly hydrophilic and have a poor permeability profile. This classification can be further extended to include new chemical entities (NCEs) with similar properties. Molecules in this class are known to demonstrate excellent pharmacological efficacy *in-vitro*, but present a challenge when formulated into orally administered dosage forms due to permeability related issues[2]. Over the years, our laboratories have worked with BCS Class III and similar molecules to address permeability issues; mainly by prodrug and permeation enhancer based approaches. Besides that, researchers have explored several other approaches with an aim to tackle the issue of low drug permeability without compromising critical solubility [3, 4, 5, 6, 7]. Among the more commonly studied approaches include prodrugs, permeation enhancers, ion-pairing, and other nanotechnology-based approaches. With this review article, we are bringing the foremost approaches under one umbrella to help facilitate further research on BCS Class III or analogous molecules.

This review highlights the different approaches utilized in recent years to increase the permeability of BCS Class III compounds. The article is organized such that the more established approaches are discussed earlier, followed by the relatively newer approaches. Each approach is discussed with respect to its concept, mechanism, advantages/drawbacks, and applicability to the oral and non-oral dosage forms. Specific examples of the compounds currently being researched, or currently being marketed, are provided as applicable.

Approaches for improving permeability

Prodrugs

Prodrugs have been described earlier as the derivatives of drug molecules that undergo an enzymatic or other biochemical transformation *in-vivo* in order to release its pharmacologically active parent drug [8]. The prodrug approaches have been historically utilized for improving the physicochemical and pharmacokinetic properties of pharmacologically active agents. Prodrugs provide opportunities to overcome several formulation and drug delivery barriers such as poor aqueous solubility, physical and chemical instability, insufficient oral absorption, rapid pre-

systemic metabolism, and inadequate tissue permeability [8]. An ideal prodrug is one that is readily transported to the desired site, selectively cleaved to release the active drug, and is retained at the site of action.

Improving passive diffusion

For a majority of drugs, the permeability across cell membranes is known to be significantly influenced by their partition coefficient, i.e. their log P value besides other factors like molecular size and concentration gradient. Previous reports have hypothesized a sigmoidal relationship between the log P value of drugs, and their cell membrane permeability[9]. Therefore, among of the more commonly employed prodrug strategies for improving the absorption of poorly permeable drugs is to increase the lipophilicity (log P) of drug [10]. Although, the use of log P to understand permeability is an over-simplification of the complex process, some correlation exists as majority of oral drugs fall between 1-5 log P values. A few outliers with atypical permeability behaviors are generally attributed to either active transport, paracellular transport, and/or the involvement of other influx-efflux mechanisms. The log P value of drugs can be increased to a certain extent by masking the hydrophilic, non-ionizable, or ionizable functional groups such as carboxylic, hydroxyl, amine, phosphate, and carbonyl groups. Typically, these functional groups are modified into carbonate esters, carbamates, amides, phosphate esters and oximes, which are frequently represented in marketed prodrugs. Ester prodrugs are among the most commonly designed prodrugs for increasing the lipophilicity, and thus enhancing the passive membrane permeability of water-soluble drugs by masking the charged functional groups. Once within the physiology, the ester bond is readily hydrolyzed by the esterases found in the blood, liver, tissues, and other organs. Figure 1 shows a few of the prominent examples of ester prodrugs including, enalapril (ethyl ester), pivampicillin (pivaloyloxymethyl ester), adefovir dipivoxil (a diester), and famciclovir (prodrug of penciclovir) [8].

The use of enzymatically labile spacer group moieties is one of the approaches researchers have utilized in order to help the drugs resist non-enzymatic hydrolysis during passive diffusion. The acyloxyalkyl compounds formed as a result serve as neutral lipophilic prodrugs capable of traversing the cell membranes by passive diffusion, followed by intracellular reversion to the parent di-acid after the cleavage of acyl group [11]. This is rapidly followed by the release of an aldehyde, thereby eliminating the spacer group. Methylene is a standard spacer

group, owing its popularity to the lack of chirality issues [12]. Researchers have applied this approach to synthesize prodrugs for a variety of antiviral agents, including adefovir.

Adefovir exists as a di-anionic compound at physiological pH, rendering it extremely polar. This highly polar nature of adefovir prevents it from undergoing passive diffusion across cellular membranes and intestinal mucosa, thus resulting in poor oral bioavailability. An approach for improving membrane permeability of adefovir involved esterification of the polar phosphonic acid group, resulting in a prodrug, adefovir dipivoxil (Figure 1), a compound with two pivaloyloxymethyl moieties. The masking of the polar phosphonic acid group resulted in an increase in the lipophilicity of adefovir, thereby allowing the prodrug to traverse the cell membranes by passive diffusion [11]. The oral bioavailability of adefovir dipivoxil in humans was found to be significantly higher (59 %), compared to that of adefovir (12 %) [13]. Besides, this prodrug of adefovir demonstrated significantly increased antiviral activity *in-vitro*, indicating increased penetration into infected cells. The clinical (human) safety and efficacy of adefovir dipivoxil was later established, and in 2002, an oral tablet formulation (Hepsera[®], Gilead Sciences, Inc., USA) was approved by the USFDA for the treatment of hepatitis B infections. **Perioli et al synthesized and evaluated prodrugs of several NSAIDs according to the chemical delivery approach developed by Bodor, with a goal to increase their permeability across the blood brain barrier (BBB) [14]. The prodrugs were prepared by attaching the carboxylic group of NSAIDs to the 1,4-dihydro-1-methylpyridine-3-carboxylate moiety, which acts as a carrier, via an amino alcohol bridge. The prepared prodrugs were found to be promising candidates for improved BBB permeability.**

Improving active transport

Prodrugs targeted towards specific membrane transporters are designed to have structural features that would allow them to be taken up by one of the endogenous transporters present at the intestinal epithelium [8]. Targeting specific endogenous transporter is critical for highly polar, charged compounds that may not demonstrate passive diffusion as a major absorption mechanism. Unlike passive diffusion that is influenced mainly by log P value, active transport of drug molecules depends on a wide variety of specific endogenous transporters. In fact, different classes of transporters such as, solute carrier family (SLC) and ATP-binding cassette (ABC) transporters have been explored with an aim to increase oral absorption [15]. Transporters such as organic cation transporter (OCT), organic anion transporter (OAT), bile acid transporters,

cholesterol transporter (ABCG5/8), monocarboxylate transporter 1 (MCT-1), etc. have been reported in literature to improve oral bioavailability of drugs [16, 17].

These transporters are known to have a high transport capacity and a broad substrate specificity. A detailed discussion of all transporters is beyond the scope of this article, however readers are encouraged to follow the references for detailed information. A few examples are provided to illustrate the importance of transporters for increasing the permeability of drug molecules. Well-known examples of prodrugs that are designed to exploit such transporter-mediated absorption include valacyclovir, valganciclovir, and gabapentin.

Peptide transporters are among the most widely distributed transporters throughout the small intestine. Valacyclovir is a *l*-valyl ester prodrug of acyclovir that is used for the treatment of herpes, varicella zoster, and cytomegalovirus (Figure 1). This orally administered prodrug was developed to improve the bioavailability of acyclovir, and is a classic example of utilizing the carrier-mediated transport. The absolute oral bioavailability of acyclovir (the parent drug) from this prodrug was found to be 54.4%, a 3- to 10-fold increase compared to that observed with the administration of its parent molecule [18]. The observed increase in the oral bioavailability of acyclovir is reported to be the result of favorable transport of valacyclovir into the gut lumen by an endogenous oligopeptide transporter (PEPT-1) [19, 20, 21].

A prodrug approach exploiting the carrier mediated transport was recently explored by our laboratory[4]. 4-guanidine oseltamivir carboxylate (GOCarb), Figure 1, is a potential influenza treatment that is reported to have a higher potency compared to oseltamivir carboxylate (OC). OC was earlier modified to produce an ethyl ester prodrug, which showed significantly higher membrane permeability, and was later approved as Tamiflu®. However, emergence of resistant strains and the need for better therapeutic outcomes lead us to target GOCarb. Simple esterification approach was not successful with GOCarb - a highly polar BCS Class III type molecule. A carrier-mediated approach was then used with GOCarb, which involved linking *l*-isoleucyl, *l*-leucyl, or *l*-valyl with the parent molecule to obtain amino acid prodrugs of GOCarb (Figure 1). These prodrugs of GOCarb demonstrated a significant advantage with respect to PEPT-1 mediated uptake, increasing its membrane permeability and bioavailability (Figure 2)[4].

Another example involving MCT-1 and sodium dependent multivitamin transporter (SMVT) is XP13512, a novel prodrug of gabapentin. The parent drug, gabapentin, is a structural analog of γ -aminobutyric acid (GABA) that is currently approved by the USFDA for the

treatment of epilepsy and post-herpetic neuralgia. This drug is thought to be absorbed from the intestines of mammals by a low-capacity solute transporter mainly located in the upper small intestine. Due to the saturation of this transporter, gabapentin demonstrates dose-dependent pharmacokinetics and high inter-patient variability, i.e. its bioavailability ranges from 30-60% of the dose (300 mg) used to treat neuropathic pain [22].

$(\pm) - 1 - \{[(\alpha - isobutanoyloxyethoxy) carbonyl] aminoethyl\} - 1 -$
cyclohexane acetic acid (XP13512, or gabapentin enacarbil), was later designed to increase the absorption of gabapentin throughout the intestine by high-capacity nutrient transporters MCT-1 and SMVT. Gabapentin shows no significant interactions with either MCT-1 or SMVT, probably due to its charged amine group; however, its prodrug XP13512, combines the properties required for recognition by both high-capacity transporters, allowing for its direct uptake across human embryonic kidney (HEK) cells [22]. The transcellular flux of XP13512 across CACO-2 cell monolayers was observed to be five times greater in the apical to basolateral direction, further suggesting that the prodrug interacts with both MCT-1 and SMVT. XP13512 is rapidly converted to gabapentin in the intestinal and liver tissues of animals and humans. The artificial membrane permeability studies at pH 6.5 and 7.4 with XP13512 showed that this prodrug may also have some ability to passively diffuse across cell membranes depending on the local pH. The pK_a of XP13512 is 5, suggesting that passive diffusion plays only a minor role, if at all, in its intestinal absorption. This prodrug was approved by the USFDA for the treatment of Restless Legs Syndrome (RLS) in 2011, and for the treatment of post-herpetic neuralgia in adults in 2012.

As discussed above, utilizing the active and passive mechanisms of transport have proven to be highly successful approaches for improving the permeability, and the overall bioavailability of BCS Class III or similar compounds. This can further be extended to address permeability issues of BCS Class IV molecules. Among the more popular formulation approaches for improving the membrane permeability of these compounds is the use of permeation enhancers. The applications of permeation enhancers as a formulation strategy form to increase drug permeability are discussed below.

Permeation enhancers

Permeation enhancers (PEs), also known as absorption enhancers are chemical moieties included in the formulations to facilitate the absorption of drugs from pharmaceutical products. **PEs can**

positively influence drug absorption by a variety of mechanisms such as modifying mucus rheological properties, altering fluidity of gastric membrane, inhibiting enzymes (e.g. proteases) and inhibiting efflux pumps [23]. PEs have been studied for several years in efforts to develop non-parenteral formulations for peptides, proteins, and other pharmacologically active compounds that have less than optimal membrane permeability [24]. PEs can be useful in optimizing drug delivery for oral as well as transdermal/transmucosal (e.g. nasal, buccal, sublingual, rectal etc.) absorption sites. However, safety in humans with these agents is always a concern and these agents need to be carefully evaluated before they can be approved by FDA. This review is limited to the discussions regarding the enhancement of oral absorption of drugs with poor permeability. Several PEs currently under investigation are illustrated in Table 1. This table also addresses the regulatory status of some of the permeation enhancers with USFDA. Most of the research on permeation enhancers is focused on the preclinical evaluation of permeability enhancement via *in-vitro* studies and possible toxicity.

Table 1: Common permeation enhancers

Permeation enhancer	Proposed mechanism	Remarks
Sodium dodecyl sulphate[25]	Decreases Trans Epithelial Electrical Resistance (TEER), and facilitates opening of the tight junctions	<ul style="list-style-type: none"> • Toxicity to the intestinal mucosa • Included in the FDA inactive ingredients guide (dental preparations; oral capsules, suspensions, and tablets; topical and vaginal preparations)
N ^α -deoxycholy-L-lysyl methylester[26]	Increases the lipophilicity of drug; drug-complex interacts with the bile acid transporter and increases absorption	<ul style="list-style-type: none"> • Toxicity to the gastrointestinal mucosa • Insulin carrier to enhance oral delivery
Rhamnolipids[27]	P-gp inhibition	<ul style="list-style-type: none"> • Demonstrated to be safe in cell viability hemolysis assays • Low toxicity, surface active properties and antimicrobial activities against several microbes (<i>Bacillus cereus</i>, <i>Micrococcus luteus</i>, <i>Staphylococcus aureus</i>, <i>Listeria monocytogenes</i>) thereby showing promising applications in pharmaceuticals and therapeutics[28, 29]
Phospholipid hexadecanol tyloxapol (PHT)[9]	Direct interaction with the tight junctions between cells resulting in intercellular permeation of peptide drugs	<ul style="list-style-type: none"> • No significant acute toxicity observed in the Lactose dehydrogenase (LDH) release assay
CriticalSorb TM (absorption enhancer based on Solutol [®] HS15)[30]	Decrease in TEER; P-gp inhibition (combination of increased paracellular and transcellular flux)	<ul style="list-style-type: none"> • No evidence of cytotoxicity in tissue histology, and CACO-2 assay

		<ul style="list-style-type: none"> Pharmaceutically acceptable excipient approved by the FDA as generally regarded as safe (GRAS)
Chitosan[31]	Opening of the epithelial tight junctions (TJ), and paracellular transport via translocation of JAM-1 (a trans-membrane TJ protein) resulting in the disruption of TJs	<ul style="list-style-type: none"> No apparent toxicity FDA approved
D-octaarginine-linked polymers[32]	Polymer induced macropinocytosis	<ul style="list-style-type: none"> Polymer-induced uptake is significantly suppressed by 5-(N-ethyl-N-isopropyl) amiloride, which is an inhibitor of micropinocytosis Currently being investigated as a novel penetration enhancer[33]
Thiolated polyacrylates[2]	P-gp inhibition	FDA approved for topical route (i.e. patch, film CR), also shown to improve the uptake of hydrophilic macromolecules from the GI tract.[34, 35]
Thiolated polycarbophils/ glutathione[36]	Protection against degrading enzymes and facilitation of transport across intestinal membranes	<ul style="list-style-type: none"> Included in the FDA Inactive Database (buccal (tablet) and ophthalmic (solution) preparations; topical patches; vaginal gel) PCP-Cys/GSH system might be a promising tool for the oral administration of oligonucleotides as it allows a significant protection toward degrading enzymes and facilitates their transport across intestinal membranes[37]
Partially quarternized poly(methacrylate) terpolymers[38]	pH-dependent decrease in TEER values	<ul style="list-style-type: none"> Reversible decrease in TEER values; recovery of cells after 6 hours Partially quarternized poly (methacrylate) terpolymers (Q-BBMCs) have been synthesized, based on the basic butylated methacrylate copolymer (BBMC/EUDRAGIT E), which is an excipient approved by the FDA[26]
Spermine (SPM) - polyacrylic acid (PAA) polymer ion-pair[39]	Decreased TEER; paracellular permeation via opening of tight junctions	<ul style="list-style-type: none"> Polyacrylic acid, sodium salt is listed as a food additive by the FDA Cytotoxicity studies in Caco-2 monolayers revealed the safety of the delivery system in the concentration range used for permeation enhancement.[40] Reversible opening of tight junctions; reversible decrease in TEER values

Sodium Caprate[41]	Opening of tight junctions; perturbation of membrane lipid or protein	<ul style="list-style-type: none"> • Increased intracellular calcium levels to induce contraction of calmodulin-dependent actin filaments • FDA approved
Decanoyl Carnitine[42]	Opening of tight junctions (TJ); perturbation of membrane lipid or protein	Decrease in the intracellular pH and the ATP levels. Intracellular acidosis. Increases in the calcium levels

Surfactants are the most frequently utilized wetting agents in pharmaceutical formulations for improving the dissolution and absorption of poorly soluble drugs. While surfactants are also considered as efficient enhancers, their use in formulations is limited due to potential adverse effects on the integrity of the intestinal mucosa. Additionally, they can interact with native surfactants (biles acids and phopspholipids) and can have add-on effect. Low molecular weight ionic surfactants like sodium dodecyl sulfate (SDS) are among the most commonly used classes of compounds for this purpose [43]. SDS has been reported to have significant effects on the paracellular permeability in CACO-2 cells by decreasing the TEER (Transepithelial electrical resistance) values, increasing the intracellular calcium levels, and opening of the tight junctions [25].

Gundogdu et al. assessed the effects of SDS in improving the permeability of fexofenadine hydrochloride (FEX) using CACO-2 cells.[44] FEX contains a basic amine, and an acidic carboxylic acid group, which results in FEX having a pH-dependent aqueous solubility. FEX also demonstrates a low, and a concentration-dependent intestinal permeability in CACO-2 cells *in-vitro*. Using TEER value as a standard indicator of permeability, the study results showed that exposing the cell monolayers to SDS for two hours resulted in a significantly increased permeability of FEX. The study also emphasized the importance of TEER value in predicting the permeation-enhancing effects of surfactants like SDS. The observed improvement in the drug solubilization was attributed to two mechanisms: improvement in the wetting characteristics of the drug, and possibly a micellar solubilization of the drug. The CACO-2 permeability of FEX from the apical to basolateral, and basolateral to apical direction was also found to significantly increase with increasing SDS concentration (10 μ M - 50 μ M). At the concentrations assayed, SDS was thought to increase the FEX permeability by relaxing the tight junctions while maintaining the integrity of the CACO-2 cell monolayer.

N ^{α} – deoxycholyl – L – lysyl – methyl ester (DCK) is a permeation enhancer of synthetic origin. DCK is composed of a hydrophobic domain from deoxycholic acid, and a

positively charged lysine that facilitates the ion-pairing interactions with anionic drugs. Such ion-pairing interactions could result in an improved absorption of drugs with poor permeability profile. DCK is also known to interact with a bile acid transporter in the intestinal membrane that could play a role in the enhancement of oral absorption of several drugs. Park et al. investigated the influence of DCK as a permeation enhancer on the permeability of ibandronate, a drug used in the treatment of osteoporosis [45]. Ibandronate is a highly potent nitrogen-containing bisphosphonate with a good safety and a tolerability profile. However, due to its poor absorption from the GI tract after oral administration, a relatively high dose (50mg/day, up to 96 weeks) is required to be administered to achieve therapeutic effectiveness. Park et al. synthesized ibandronate-DCK complex and evaluated the *in-vitro* permeability, and *in-vivo* bioavailability of the prepared complex in comparison to pure ibandronate in laboratory animals. The *in-vitro* permeability assessment was carried out using a vertical Parallel Artificial Membrane Permeability Assay (PAMPA) system. As compared to the parent drug, the prepared ibandronate-DCK complex demonstrated a significantly higher permeability of ibandronate, suggesting that ionic complexation with DCK can enhance the partition of ibandronate into the lipophilic membranes. The amphiphilic nature of DCK was thought to impart lipophilicity to ibandronate molecule without any chemical alteration, resulting in the observed increase in its absorption. The prepared ibandronate-DCK complex (1:2) was additionally analyzed for bioavailability after intra-jejunal administration in rats. The results showed that the prepared ibandronate-DCK complex exhibited 2.8- to 4.3-fold increases in the C_{max} and AUC values compared to those of pure ibandronate. Thus a significant increase in the relative bioavailability of ibandronate due to its complexation with DCK was observed [45].

Chitosan is a linear polysaccharide composed of randomly distributed β -(1-4)-linked D-glucosamine (de-acetylated unit) and N-acetyl-D-glucosamine (acetylated unit). It is a non-toxic, biocompatible, and biodegradable polymer, commercially prepared by deacetylation of chitin. Chitosan has been shown to be a useful excipient in a variety of drug delivery systems. The amine groups present in the polymer have been explored for its role in imparting permeation enhancement, controlled release, mucoadhesion, transfection, and inhibition of efflux pumps, etc. properties in pharmaceutical dosage forms. In its role as a permeation enhancer, chitosan is reported to effectively open the epithelial tight junctions, allowing paracellular transport of the compounds via translocation of the junction adhesional molecules [46]. Chitosan is also reported

to decrease the TEER values of the cell monolayers, and increase the paracellular permeability of compounds across them [47, 48]. Additionally, the disruption of the tight junctions by chitosan is shown to be a reversible process, ensuring the safety of its use as a permeation enhancer.

There are some studies in literature documenting the use of inorganic materials to alter physiochemical property of a drug. Literature has shown the use of inorganic materials such as layered double hydroxides; Perioli et al prepared hydrotalcite like compounds (HTlc) viz. MgAl-HTlc-FURO to improve furosemide dissolution. The study showed that furosemide dissolution was enhanced with these complexes by homogenously dispersing the drug and forming a molecular film that could be defined “shell-liquid state”, in the nanospaces of the interlayer region [49]. While this study was focused on enhancement of drug dissolution, in a later study the authors evaluated the influence of these complexes in enhancing the permeability of furosemide [50]. The authors’ main goal was to assess gastric mucus rheology and drug flux across gastric mucus layer *in vitro*. They also carried out permeation assays using the artificial membrane (*in vitro*) and porcine gastric mucosa (*ex vivo*). While the results obtained from artificial membrane did not correlate well, the study found a significant increase in furosemide permeability with MgAl-HTlc-FURO, when compared to furosemide alone. While this was an interesting study demonstrating the use of inorganic materials to enhance bot solubility and permeability of a compound, such studies are few and far between, and more inorganic compounds such as these need to be explored to understand their influence on the mechanistics of drug permeability.

Current Status of permeation enhancers

As shown in Table 2, significant advances have been made in the utilization of permeation enhancers to improve drug absorption in transdermal formulations, and several transdermal drug products using this approach have been approved by the USFDA. While the scientific merit regarding the use of this approach in oral formulations has been established in the literature, currently no oral drug product is approved for marketing. One of the barriers to the use of permeation enhancers in oral formulations is the lack of their established safety profile in advanced clinical trials. In recent years, a few drug products utilizing permeation enhancers for increasing oral drug delivery are either on the threshold of being approved, or have advanced through various stages of clinical trials (Table 2) [24].

Table 2: Permeation enhancers in development/market

Enhancer/technology	Drug/drug product	Company
Cyclopentadecalactone (CPE-215)[51]	Nasulin TM (nasal insulin)	CPEX Pharmaceuticals, Inc.
Sodium N-[8-(2-hydroxybenzoyl)amino] caprylate (SNAC)[52]	Eligen B12 TM (oral vitamin B ₁₂)	Emisphere Technologies, Inc.
Gipet [®] (medium chain fatty acids, salts, and derivatives)[53]	Almerol TM (MER-103) (alendronate)	Merrion Pharmaceuticals
Sodium caprate[54, 55]	Antisense oligonucleotide	Ionis Pharmaceuticals, Inc.
Sodium caprylate[56, 57]	Octeotride	Chisama, Inc.
Axcess TM (GRAS excipients)[58]	Capsulin TM (oral insulin)	Diabetology, Inc.
(ChiSys [®]) Chitosan[59]	Intranasal apomorphine	Archimedes Pharma
Nexact [®] - Dodecyl-2-N,N-dimethylamino propionate (DDAIP)[60]	Femprox [®] (Topical Alprostadil Cream)	Apricus biosciences, Inc. Nexmed (U.S.A.), Inc.

Ion-pairing

The use of counterions, or ion-pairing method to improve the passive permeability have been researched extensively in the past three decades [3, 19, 20, 61, 62]. The ion-pairing approach works primarily by complexing the molecule of interest with oppositely charged ionic species, resulting in an overall neutral ion-pair that increases lipophilicity, the membrane permeability, and the absorption of the molecule [63, 64]. Unlike the prodrug approach where the permeability enhancement is achieved via formation of a new chemical entity, the complex formed by ion-pairing is held together as a single unit by coulombic attractive forces rather than covalent bonding [42]. As stated above, the ion-pairing approach works by neutralizing the charge on the drug molecules to allow passive diffusion of the ion-pair complex through biological membranes. Assuming quasi-equilibrium conditions, this approach would follow the simple rules of the association constant as shown in the Figure 3, and equation 1 below, where $[BH^+]$ represents the drug and $[A^-]$ represents the counterion.

$$K = \frac{[BH^+A^-]}{[BH^+][A^-]} \quad (1)$$

The common advantages and drawbacks of this approach are summarized in Table 3.

As shown in this table, a major concern with utilizing the ion-pairing approach for permeability enhancement is the possibility of having insufficient and/or weak ionic interaction, which may result in the complex dissociating during the absorption phase through the biological membranes. This emphasizes the requirement of a high association constant (K) for these interactions.

Table 3: Advantages and drawbacks of ion-pairing approach

Advantages	Drawbacks
<ul style="list-style-type: none"> • Non-prodrug formation of complex; thus do not requiring enzymes for activation. • Eliminates the necessity of active transport for absorption. • Complex formed dissociates easily after absorption. • Equilibrium may be modified to enable complex absorption followed by facile dissociation of available drug. • Additives which may compromise membrane integrity, and lead to toxicity are not required 	<ul style="list-style-type: none"> • Possibility of premature complex dissociation during membrane permeation due to weak non-covalent interactions. • Many ion-pairing agents utilized may not be suitable for human intake due to known membrane irritation and/or toxicity challenges. • Overutilization of counterions may result in the formation of micelles, which may exacerbate the poor permeability issues. • Concentration-dependent permeability enhancement may lead to toxicity at high levels • Limited range of target partition coefficient: the Log P value of the ion-pair complex required to be between 2 and 5 to facilitate passive membrane permeability.

Among the more commonly utilized counterion is 1-hydroxy-2-napthoic acid (HNAP).

This counterion, due to its high lipophilicity, relatively strong binding constant, and a history of prior use with drugs such as salmeterol, zanamivir, and oseltamivir makes it an ideal agent to counterbalance the high polarity of several BCS Class III drugs [5, 6]. Miller et al. utilized HNAP as a counterion to prepare ion-paired complexes of the oral drugs with poor intestinal permeability such as zanamivir heptyl ester (ZHE) and guanidine oseltamivir (GO) [6]. The study results showed a significant (up to 3.7 log units) increase in the lipophilicity of the drugs due to ion-pairing. Furthermore, the study demonstrated the enhancement of permeability by plotting the apparent partition coefficient (P_{app}) of CACO-2 against the concentration of HNAP, and developing a quasi-equilibrium transport model with a linear relationship of the

slope close to 1. As shown in the Table 4, each formulation may utilize different types of counterion approaches to improve permeability.

Table 4: Common ion-pairing agents

Counterion	Counterion type	Remarks
Trifluoroacetic acid (TFA)[65]	Anionic ion-pairing	Reduces the pH of aqueous mobile phase
Heptafluorobutyric acid (HFBA), Pentafluoropropionic acid (PFPA), phosphoric acid[66]	Anionic ion-pairing	Increases the lipophilicity of the compound
1-hydroxy-2-napthoic acid (HNAP)[6]	Anionic ion-pairing	Increases the partition coefficient ($\text{Log}P$) of the compound
Tetraheptylammonium bromide (THAB), Tetrabutylammonium iodide (TBAI)[67]	Phase-transfer catalyst	Increases zeta potential
Hexadecyldimethylbenzyl ammonium chloride, Hexylsalicylic acid[38]	Absorption enhancer	Increased the lipophilicity and bioavailability of a beta-lactam antibiotic, cefpirom

Although this manuscript is mainly focused on approaches to improve oral permeability, these principles can be extended to topical, as well as parenteral formulations. Understanding the mechanistic principles of enhanced permeability of a given approach is important, and can help extrapolate this information to different routes of administration. A significant amount of ion pairing approach is applied to transdermal formulations. Counterions can serve as neutralizing agents by binding with the drug molecules via Coulomb forces, reducing their polarity, and promoting their transmembrane permeation. The ion-paired complex can then dissociate post-absorption, permitting drug molecules to diffuse into the epidermal and dermal tissue layers [40, 68]. Megwa et al. studied the influence of the extent of ion-pairing with alkylamines and quaternary ammonium ions on the *in-vitro* transmembrane permeability of salicylates by measuring the conductivity of the penetrant solution [69]. The study showed that an increase in the extent of ion-pairing neutralized the overall charge on the molecule, resulting in an increase in the flux of the drug across the membrane.

There are several other examples where counterion approach has increased transdermal permeability [70, 71]. There are also reports in literature where the ion-pairing approach have been explored for ocular formulations [72]. As mentioned above, exploring approaches utilized in non-oral route of administration can assist in identifying counter ions suitable for oral use. While studies indicating conceptual success of ion-pairing in improving the permeability of drugs are frequently found in scientific literature, additional investigations are required to further this concept to clinical trials, resulting in commercially successful products.

Nanotechnology-based approaches

In the recent years, nanotechnology based approaches are among the most aggressively researched, for enhancing the delivery of pharmaceutical drug formulations. Due to the perceived advantage of small particle size ($\leq 100\text{nm}$) in increasing transmembrane permeability, nanotechnology based products are presumed to provide a significant improvement in the bioavailability of several drugs [73]. **Several studies have been reported that link the bio-toxicity of nanoparticles to their small particle size. However, more comprehensive studies have shown that nanoparticle-induced bio-toxicities are a result of a complex interplay of the size, shape, surface chemistry, and the charge on the particles [74]. This review is focused purely on the merits of nanotechnology in aiding the drug permeability and bioavailability.** The section below discusses current advances in the use of nanotechnology-based approaches for the improvement of drug permeability, and their overall bioavailability.

Nanoparticles

The use of nanoparticles for increasing the permeability of drugs has been a subject of current scientific and biomedical research. Originally stemmed from the focus of delivering anti-cancer agents, this approach has gained popularity due to its reported ability to allow delivery of nucleic acids, peptides, etc. for the treatment of several diseases [75]. The emergence of this approach also stems from the drawbacks associated with the therapeutic interventions using macromolecules or micromolecules. Most macromolecular drugs are reported to have a high risk of proteolytic and/or hydrolytic degradation, resulting in low bioavailability and a shorter half-life. Micromolecular anticancer drugs are associated with toxic side effects or unsuitable bio-distribution [75]. Additionally, small-molecule anticancer agents may quite often be hydrophilic in nature, and suffer from poor permeability similar to that observed with BCS Class III drugs.

Thus, the primary aim of the nanoparticle-based approaches is to improve the permeability profiles of drugs, in addition to preventing the undesired side effects.

The nanoparticle based approach works by encapsulating the drug molecule in polymeric nanoparticles with a goal to overcome several challenges such as hydrophilicity, poor cellular penetration due to size, biological degradation, and toxicity of the drug molecules [75]. In addition, the significant reduction of the particle size to ≤ 100 nm is known to result in dramatic changes in the pharmacokinetics of the drug molecules, particularly the drugs' permeability and delivery efficiency [31]. Since low permeability is the greatest challenge for drugs in BCS Class III, the nanoparticle based approach provides a promising new direction for formulating these drugs. Nanoparticle based formulation of drugs can be achieved in several ways, i.e. entrapment of drug molecules in polymeric nanoparticles, adsorption of drug molecules on the surface of nanospheres, or formulation of aqueous-core nanocapsules. Table 5 summarizes some of the advantages and drawbacks of each of these approaches[7].

Nanoparticle-based formulations offer several advantages. First, drug encapsulation prevents biological macromolecules from degradation in the GI tract, and allows them to reach the target site in relatively higher concentration. Second, encapsulation may increase the oral bioavailability of these drugs while minimizing the plasma concentration fluctuations and the systemic side effects. This is due to the utilization by the nanoparticles, of different uptake mechanisms such as 'active transport system' or 'endocytosis', and the ability of the drug to remain in circulation for a prolonged period of time, mimicking a controlled-release profile. Third, nano-encapsulating drug molecules significantly decreases the size, allowing more passive diffusion, and penetration through the biological membranes. The uptake of nanostructures has been reported to be about 15-250 times greater than that of microparticles in the 1-10 μm range [31].

Table 5: Nanoparticle-based formulation strategies

Strategy	Advantages	Drawbacks
Entrapment in polymer nanoparticles	<ul style="list-style-type: none"> • Drug protection • Sustained drug release • Loading efficiency dependent on the formulation process 	<ul style="list-style-type: none"> • Lack of drug solubility in polymer • Reaction between drug and PACA/monomer • Use of organic solvent • Medium acidification during polyester degradation • Rapid degradation of alginate nanoparticles by cation exchange

Adsorption onto polymer nanoparticles	<ul style="list-style-type: none"> • Drug not submitted to the formulation process • Nature of polymer (charge, hydrophilicity) influences drug loading • No reaction between drug and polymer → possibility of modifying the nanoparticle surface 	<ul style="list-style-type: none"> • Drug leakage and rapid release • Nanoparticle destabilization by charge mask • Stealth properties modified • Lower biological efficacy • Use of organic solvent
Entrapment in Aqueous Core Nanoparticles (ACN) -drug protection	<ul style="list-style-type: none"> • Drug solubilized in the core • Reaction between drug and monomer is limited • Low polymer content • Sustained drug release 	<ul style="list-style-type: none"> • High energy devices for generating emulsions and nano-emulsions • Organic solvent

Several agents belonging to different chemical classes have been explored for their use as nano-encapsulating agents. These chemical classes with their unique attributes, and **current status of some** specific examples are summarized in Table 6; **enlisting all the examples and their current statuses is beyond the scope of this review**. Nanoparticulating or nanosizing of drugs involves a reduction of drug particle sizes to less than 100 nm. Nanoparticulation of drug particles have been reported to provide several advantages including enhanced drug delivery, reduction in the amount of expensive Active Pharmaceutical Ingredient (API) required, protection from biological degradation, and possible achievement of higher specificity with minimized side effects. Among the popular nanoparticulating techniques are formulation of liposomes, and the use of nanometric carriers, each with their unique contribution in enhancing drug delivery via an increase in permeability [27].

Table 6: Nanoparticle encapsulation agents

Category	Examples	Toxicity	Current status	Comments
Polymer based agents				
Polymer-drug conjugates	<i>Natural polymers:</i> <ul style="list-style-type: none"> • albumin, chitosan, and heparin <i>Synthetic polymers:</i> <ul style="list-style-type: none"> • N-(2-hydroxypropyl)-methacrylamide copolymer (HPMA) 	<ul style="list-style-type: none"> • Nontoxic and biodegradable 	<ul style="list-style-type: none"> • Paclitaxel-albumin (Abraxane®) Approved by the USFDA in 2005 for the treatment of breast, lung, pancreatic, and small cell lung 	<ul style="list-style-type: none"> • Synthetic or natural • Water-soluble • Selective drug accumulation in tumor tissue (EPR effect) • Receptor-mediated targeting to increase specificity

	<ul style="list-style-type: none"> • Polystyrene-maleic anhydride copolymer • Polyethylene glycol (PEG) • Poly-L-glutamic acid (PGA) 		<p>cancers [76, 77]</p> <ul style="list-style-type: none"> • Doxorubicin-HPMA copolymer (Phase II trials completed) [78] • Paclitaxel-Poly (glutamic acid) (Phase II trials completed) [79] • PEG-Pglu (SN38) (Phase II trials for breast cancer) [80, 81] 	toward cancer cells
<p>Polymeric micelles</p>	<ul style="list-style-type: none"> • Paclitaxel • Genexol-PM (PEG-poly(D,L-lactide)-paclitaxel) 	<ul style="list-style-type: none"> • Biodegradable and non-toxic 	<ul style="list-style-type: none"> • Paclitaxel (Paclitaxel polymeric micelle)(Phase III completed for ovarian cancer) [82] 	<ul style="list-style-type: none"> • Amphiphilic block copolymers • Suitable carrier for water-insoluble drugs • Biocompatible, self-assembling • Ease of modification • Targeting potential
<p>Dendrimers</p>	<ul style="list-style-type: none"> • Poly(amidoamine) 	<ul style="list-style-type: none"> • Cationic dendrimers exhibit hemolytic toxicity, cytotoxicity and hematological toxicity[83] • Neutral and negatively charged 	<ul style="list-style-type: none"> • Docetaxel-dendrimer conjugate (DEPTM- Docetaxel in Phase I trials for advanced metastatic cancer)[84] 	<ul style="list-style-type: none"> • Synthetic polymeric macromolecule with nanometer dimensions • Monodisperse size, modifiable surface functionality → high

		dendrimers are non-toxic[83]		structural and chemical homogeneity <ul style="list-style-type: none"> • Multivalency • Water solubility and biodistribution can be modified • Internal cavity • Controlled degradation
Lipid based agents				
Liposomes	<ul style="list-style-type: none"> • Pegylated liposomes • Non-pegylated liposomes 	<ul style="list-style-type: none"> • Cationic liposomes (non-pegylated liposomes) has shown to cause dose dependent toxicity and pulmonary inflammation in mice[85] 	<ul style="list-style-type: none"> • Doxil (Doxorubicin liposomes approved for the treatment of Kaposi's sarcoma, ovarian cancer and breast cancer)[86, 87] 	<ul style="list-style-type: none"> • Amphiphilic, biocompatible • Ease of modification • Targeting potential
Viral nanoparticles	<ul style="list-style-type: none"> • Cowpea mosaic virus • Cowpea chlorotic mottle virus • Canine parvovirus • Bacteriophages 	<ul style="list-style-type: none"> • Could elicit immune response in host[88] • Cowpea mosaic virus is reported to be safe [89] • Viral nanoparticles containing iron oxide are reported to be toxic[90] 	<ul style="list-style-type: none"> • Emerging field with applications in disease prevention, diagnosis, monitoring and therapy[91] 	<ul style="list-style-type: none"> • Multifunctional with specific tumor targeting • Multivalency – ability to allow various surface modifications • Defined geometry and uniformity • Biological compatibility and inert nature
Carbon nanotubes	-	<ul style="list-style-type: none"> • Reported to have multiple toxicities depending on the surface charge, shape, length, diameter, 	<ul style="list-style-type: none"> • Emerging area of research 	<ul style="list-style-type: none"> • Use of benzene ring • Multifunctional

		agglomeration and purity[91, 92]		
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Liposomes and other nanometric carriers

Liposomes are generally described as lipid-based microscopic vesicles with sizes ranging from 50 nm to over 100µm. Liposomes may possess a negative, positive, or a neutral surface-charge depending on their composition. In addition, depending on their size, liposomes may be categorized as unilamellar vesicles, or in the presence of more than one bilayers, multilamellar vesicles [93]. The lipid bilayers of the liposomes possess both hydrophilic and lipophilic properties, similar to that observed in the phospholipid bilayer in human cells. These properties provides liposomes with a specific advantage with respect to increasing permeability by facilitating enhanced interactions between the drug and the cell membrane. Enhanced permeability with liposomes is achieved via one or more of the four mechanisms: adsorption, endocytosis, fusion, and lipid transfer. Additionally, derivatized liposomes such as pegylated liposomes have been reported to display a prolonged drug circulation, and a slower release of drug molecules similar to controlled-release formulations [94, 95]. Liposomes are also reported to possess the ability to provide target-site specificity, as demonstrated by pegylated horse-radish-peroxidase loaded liposomes targeting the brain capillary endothelial cells [96].

Another nanotechnology-based approach for permeability enhancement of drugs is the use of nanometric carriers. This approach involves adsorbing, entrapping, or covalently attaching the drug molecules to sub-micron sized particulate carriers in an attempt to improve the drug permeability. Among the reported advantages of this approach are, ease of passage across even the smallest of the capillary vessels, achievement of higher penetration into the cells and organs, prolonged plasma circulation by avoiding phagocytic clearance, improved therapeutic effects of drugs, and the ease of functional modifications for improving stability [31]. Current research with regards to utilizing this approach for enhanced drug delivery is primarily focused on achieving target specificity, improving the drug delivery capacity, and the use of a combination of carrier materials to improve drug delivery efficiency. Among the commonly employed nanometric carrier materials include, poly-lactic acid (PLA), poly-glycolic acid (PGA), and poly-lactide-coglycolide (PLGA).

Smart drug delivery systems, polymer-drug conjugates, and multifunctional carrier systems

Achieving acceptable drug permeation and accumulation in the target tissue or organ calls for designing a drug delivery system that is multifunctional, and more specifically possessing a property of switching ‘on’ or ‘off’ certain functions when required [31]. While several attempts have been made to formulate such a drug delivery system utilizing nanotechnology based approaches such as liposomes or nanoparticles, only few successes have been reported [97].

The Smart Drug Delivery Systems (SDDS), also known as the stimuli-sensitive delivery systems function on a principle of ‘rapid transition of the physical-chemical properties of a polymer system upon contact with a stimulus’, the stimulus being either physical, chemical, or biological, to trigger a desired drug release [31]. A commonly employed approach for the development of SDDS is through the use of polymer micelles. The polymeric micelles contain both hydrophilic, and lipophilic domains, and when used to encapsulate hydrophilic drugs, are known to increase the permeability via passive diffusion. The release of drug occurs upon the degradation of the micellar domains *in-vivo*. A specific example of micelle forming polymeric agent is Poly (ethylene glycol)-b-polyhistidine (PEG-b-PHis). A unique property of PEG-b-PHis is the ability to adjust its *pKa* by changing its molecular weight. Several studies report successful utilization of PEG-b-PHis to prepare SDDS, and achieve targeted and controlled drug delivery [98, 99]. Higher localized drug accumulation have been achieved at the desired target site even with the parenteral administration of SDDS [100]. Chu et al. prepared a SDDS which consisted of a glucose-sensitive microcapsule with a porous membrane, with linear-grafted polyacrylic acid (PAAC) chains, and covalently bound glucose oxidase enzymes in the membrane pores acting as functional gates [101]. The study results showed that the prepared SDDS provided a self-regulated drug delivery systems capable of releasing insulin in response to changes in the plasma glucose concentration.

The polymer-drug conjugates has been a successfully utilized approach for improving the delivery of several drugs. However, this approach is mainly employed to improve the solubilization of drugs, rather than for improving permeability. Polymer-drug conjugates typically consists of water-soluble polymers that attach to the drug molecules, forming a conjugate with increased aqueous solubility and distribution. An advantage of utilizing polymer-drug conjugates is the formation of colloidal particles which may prolong the drug circulation time [31]. Multifunctional drug delivery systems (MDDS) are the systems that possess more than

one of the functional properties observed with the nanotechnology based approaches. These delivery systems exhibit properties such as stimuli-sensitivity, passive/active drug localization at targeted site, ability to achieve higher bioavailability, and/or increased blood circulation time. An example of a MDDS would be a mixture of PLA-b-PEG-b-PHis-biotin and PEG-b-PHis block copolymers [68].

Nanotechnology-based approaches provides a plethora of options for enhancing drug delivery via different routes, i.e. oral, parenteral, pulmonary, topical etc. Although parenteral formulations are devoid of challenges related to drug permeability (since they are directly injected into blood stream), it is still important to evaluate the benefits of using nanoparticle based formulations to ensure the distribution/bioavailability of the drug at the site of action. Zara et al. compared the pharmacokinetics of doxorubicin incorporated as ion-pair into Solid-Lipid Nanospheres (SLN) with that of the commercial solution of the drug in laboratory animals [102]. They observed that SLN showed a markedly higher concentration of doxorubicin in the blood, lung, spleen, and brain, at each time point than observed with the commercial solution. The study also found that the SLN-treated rats showed a lower doxorubicin concentration in liver, heart and kidney. Additionally, the results showed that SLN increased the area under the curve (AUC) of doxorubicin compared to a conventional doxorubicin solution. Since oral administration is the most preferred route of administration, it is also the focus of several nanotechnology-based studies for improving drug bioavailability. Kalaria et al. prepared and pharmacokinetically evaluated doxorubicin-loaded poly(lactic-co-glycolic acid) (PLGA) nanoparticles [103]. They found that the doxorubicin-loaded nanoparticles demonstrated a significant (363%) increase in the bioavailability of doxorubicin, and a six fold increase in its T_{max} .

Pulmonary drug delivery is another area that has benefitted from nanotechnology based approaches. Here, however, a nanoparticulate strategy via micronization of drug particles rather than drug encapsulation is utilized. Smaller drug sizes enable better penetration in the lung either via i.v. administration (through microspheres), nebulized solutions, or dry powder inhalations. An example of nanoparticle utilization for pulmonary administration is the microemulsion system based on water/lecithin/propanol/iso-octane [104]. The small size of nanoparticles allows them to be the ideal candidates for pulmonary administration. However, the effects of microsizing/nanosizing on the manufacturability, stability, and the safety of the drug products need further research and analysis. Due to the small size and higher penetration observed with

nanotechnology based products, nanoparticles have also found applications in topically administered products. In comparison to the conventional drug molecules, nanoparticles result in a more uniform drug particle and larger number of particles, leading to a higher occlusion factor and skin penetration (Figure 4). Moreover, compared to conventional formulations, a smaller dose is required with nanoparticles. This results in a significant reduction in the occurrence of irritation, dryness, itching, and/or burning sensation that can occur with conventional dosage forms.

A promising new application of nanoparticles is the possibility of achieving a target specific drug delivery. Gao et al. investigated the multivalent effect for up-regulating the intracerebral delivery of nanoparticles via receptor-mediated transcytosis [105]. They prepared nanoparticles labeled with near-infrared (NIR) fluorophore and different numbers of angiopep-2 peptides that specifically target low-density lipoprotein receptor-related protein (LRP) on the blood brain barrier (BBB). The *in-vivo* NIR image analysis showed that the multimetric association between the angiopep-2 peptides labeled on the nanoparticle, and the LRP receptors on the BBB significantly increased the intracerebral uptake of the nanoparticles.

Hybrid or combination approaches

With the advancement of technology, several studies employing a combination of one or more approaches above to improve drug permeability have been explored. Fattal et al. studies the effect of hybridization of polyalkylcyanoacrylate nanoparticle and an ion-pairing agent, cation hydrophobic detergent (CTAB) on the delivery of antisense oligonucleotides (ON) in modulating gene expression [106]. ON was loaded into the polyalkylcyanoacrylate nanoparticle that was formed via an emulsion polymerization process. The obtained complex was then coated with CTAB to create a lipophilic nanoparticle-ion paired complex. The functional characterization of this complex was then carried out via *in-vitro* and *in-vivo* analysis. The *in vitro* analysis showed an increase in the cellular uptake of the drug complex, as it survives the nuclease attack in the cell culture media and is actively taken up via the endocytotic/phagocytotic pathway. The *in-vivo* analysis demonstrated an increased stability of the drug complex in plasma and liver. While this method of hybridization between nanoparticle and the ion-pairing approach did not demonstrate an increased drug permeability, and instead increased the active uptake of the drug, it is still a feasible method to consider.

Another example of hybridization/combination of nanoparticle and ion-pairing approach is the formation of an insulin-sodium deoxycholate complex with an aim to promote the oral delivery of insulin [41]. In this study, an insulin-deoxycholate complex (Ins-SD-Comp) was first formed by the hydrophobic ion-pairing method to increase the lipophilicity of insulin. This complex was also hypothesized to enhance the bioavailability of insulin, as sodium deoxycholate is naturally produced by the liver and undergoes enterohepatic circulation up to 20 times a day. This Ins-SD-Comp was then encapsulated in poly (lactide-co-glycolide) (PLGA) nanoparticle via emulsion solvent diffusion method to form Ins-SD-Comp-PLGA-NP, to prevent physiological degradation and enable oral administration. The hypoglycemic effects of this hybrid complex were analyzed by the *in vivo* bioassay. The results showed that this hybrid complex demonstrated a statistically significant plasma glucose reduction when compared to oral administration of saline-free insulin solution.

Conclusions

GI absorption of the low permeability compounds such as those belonging to the BCS Class III or molecules with similar properties is an area of extensive research. For these drugs, the intestinal membrane permeability is expected to be the rate-limiting step in the overall drug absorption process. Amongst the various approaches used for improving the permeability include, prodrugs, permeation enhancers, ion-pairing, and nanotechnology based approaches.

The use of prodrugs or targeting carrier-mediated transport has proven to be a relatively successful in improving the permeability. Ion-pairing approach has yielded positive results in transdermal and parenteral drug delivery; however, further research for the application of this approach for oral delivery is still underway.

Nanotechnology-based approach have shown to provide a promising technology to enhance the permeability of the drugs. Both the nanoencapsulation and nanosizing strategies result in significant reduction in the particle size, resulting in improved drug permeability. While the majority of research utilizing nanotechnology is still under clinical trials, the current results from different routes of administration have demonstrated positive outcomes. Similar to ion-pairing approach, further research is required to assess the safety profile of nanotechnology-based products and the ease of their manufacturing. Once thought impossible, the hybridization of several approaches for improving drug permeability is now feasible. While the initial studies have demonstrated a success in the formation of nanoparticle-ion-paired hybrid complexes and

stable activity, it is usually either the ion-pairing effect or the nanoparticle effect that is found to be the key permeability enhancer. Nonetheless, further advancement in the technology and refinement of approaches will certainly make the hybrid-complexes a successful strategy in improving drug permeability.

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