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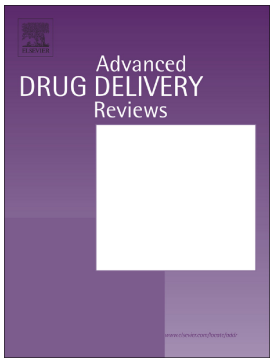

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Micro and Nanoscale Technologies in Oral Drug Delivery

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Abstract

Oral administration is a pillar of the pharmaceutical industry and yet it remains challenging to administer hydrophilic therapeutics by the oral route. Smart and controlled oral drug delivery could bypass the physiological barriers that limit the oral delivery of these therapeutics. Micro- and nanoscale technologies, with an unprecedented ability to create, control, and measure micro- or nanoenvironments, have found tremendous applications in biology and medicine. In particular, significant advances have been made in using these technologies for oral drug delivery. In this review, we briefly describe biological barriers to oral drug delivery and micro and nanoscale fabrication technologies. Micro and nanoscale drug carriers fabricated using these

technologies, including bioadhesives, microparticles, micropatches, and nanoparticles, are described. Other applications of micro and nanoscale technologies are discussed, including the fabrication of devices and tissue engineering models to precisely control or assess oral drug delivery *in vivo* and *in vitro*, respectively. Strategies to advance translation of micro and nanotechnologies into clinical trials for oral drug delivery are mentioned. Finally, challenges and future prospects on further integration of micro and nanoscale technologies with oral drug delivery systems are highlighted.

Keywords: Drug delivery devices; Micro and nanocarriers; Micro and nanoscale technologies; Oral drug delivery; Tissue models.

1. INTRODUCTION

Oral delivery has been one of the most commonly used approaches for drug administration in the body due to its high patient compliance, low cost, non-invasiveness, and ease of use (1, 2). A multitude of therapeutic compounds, including synthetic small molecules and biologics have been administered orally. However, oral drug delivery poses significant challenges to achieving efficient therapeutic outcomes. This is primarily due to the multitude of biological barriers that are present throughout the gastrointestinal (GI) tract that a drug carrier must navigate through. Some of these biological barriers to drug delivery include harsh acidic pH environments in the stomach, degrading enzymes that render drugs ineffective, the inefficient penetration of drugs across GI tissue barriers and into systemic circulation, and the eventual clearance of drugs through the GI tract, which may occur prior to drug release. To overcome these barriers, the delivered dosage of drugs is often higher than what is needed therapeutically, as the

bioavailability of the compound is often reduced due to factors like enzymatic degradation and poor permeation through the intestinal wall. However, it is important that the drug concentration should not exceed the level that can cause toxicity in the body, as has been observed with some DNA and protein based drugs above critical concentrations (3, 4). On the other hand, if a drug at a non-toxic concentration level passes the physiological barriers of the GI tract, its delivered dosage to target site may not be effective (5).

An ideal oral drug delivery system has yet to be realized, by which drugs can be delivered to a biological target at appropriate concentrations with tunable dosage windows. Several limitations of delivery of drugs through the oral cavity are due to physiochemical properties of drugs. In particular, Lipinski rules or the Rule of Five (ROF) are used as a rule of thumb for discovering new drugs and to improve the efficacy of developed drugs (6). The ROF details four properties of drug molecules including: that the molecular weight should not exceed 500 Da, logP values should be under 5, total number of hydrogen bond donors should be 5, and number of the hydrogen acceptors should not be more than 10. The total ROF score lies between 0 and 4. Molecules with an ROF score more than 4 are considered to be marginal drug molecules and need to have further development. However, some drug molecules do not follow the Lipinski rules, such as proteins and RNA molecules. One of the most well-studied protein therapeutics for oral administration is insulin. Currently, insulin is administered via daily injections and 45-60% of diabetic patients intentionally skip insulin doses out of dread of injections (7). It is therefore likely that oral administration of insulin would increase patient compliance and improve therapeutic outcomes in diabetic patients. However, the bioavailability of orally administered insulin is severely limited by the physiological barriers of the GI tract, such as acidic pH, presence of proteases, and the limited transport of insulin across GI epithelial barriers into the

bloodstream (8).

By incorporating drugs such as insulin into materials-based carriers, it may be possible to overcome or circumvent the physiological barriers that limit oral administration efficacies. In designing materials for oral drug delivery, the drug carrier should preserve therapeutic efficacy of the drug cargo for effective use in humans. There are two major goals for designing materials for oral drug delivery: (1) the effective targeting of drugs to a GI section of interest, and (2) the release of drugs from the GI into the bloodstream for systemic circulation. For both of these goals, the design and development of drug delivering materials needs to account for the mucosal microenvironment of the GI system, intestinal physiology, and target diseases. Moreover, chemistry, size, shape, metabolism, and bioavailability of drugs play a crucial role in the design of effective oral drug delivery systems.

Micro and nanotechnologies have seen widespread use in oral drug delivery systems with the goal of improving the efficiency of delivery systems. These technologies have been used for many applications, including drug discovery via development of high throughput screening assays (9-11), miniaturization of therapeutic and diagnostic tools, tissue engineering and oral drug delivery (12, 13). Some major problems in oral drug delivery have been solved by fabrication of micro and nanocarriers with precise control over their architecture and size. These efforts are a part of controlled drug delivery systems dating back to the 1950s (14, 15). In recent years, dynamic oral delivery systems have been fabricated using micro and nanofabrication technologies by which sensing, recording, and stimulating of biological systems can be achieved for optimized drug delivery (16, 17). In addition, microfabrication techniques have been used to

make biomimetic GI tract *in vitro* models in which the body's response to drugs can be recapitulated and used for better design of drugs.

In this work, a brief review of physiological barriers to oral drug delivery is given (**Figure 1**). We then discuss micro- and nanofabrication techniques and the subsequently fabricated drug carriers that have been used as oral drug delivery systems, with a focus on the material components, fabrication technologies, and drug loading efficiencies. We describe in detail the general chemical and physical strategies to functionalize diverse types of drug carriers for oral drug delivery applications with a focus on bioadhesion and tissue barrier remodeling. Specific examples of how engineered drug carriers have been used successfully to navigate the GI tract and improve oral drug delivery are then provided. We then discuss applications of fabrication technologies for modeling of the GI tract. Following that, clinical trials in oral drug delivery systems using fabrication technologies are mentioned. Finally, we highlight challenges and future directions in using micro- and nanofabrication technologies for oral drug delivery.

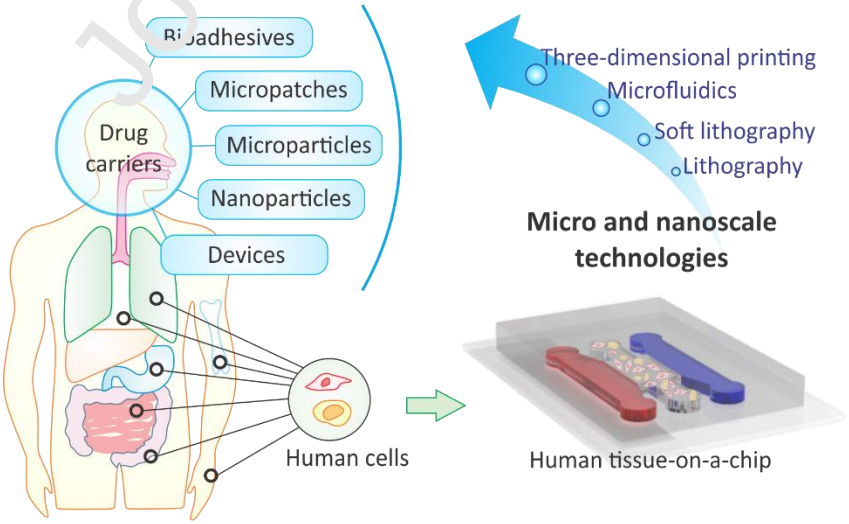

Figure 1. Micro- and nanoscale technologies enable fabrication of oral drug carriers as well as human tissue-on-a-chip models for precision medicine applications.

2. PHYSIOLOGICAL BARRIERS TO ORAL DRUG DELIVERY

Some limitations of oral drug delivery systems are governed by GI anatomy, physiology, and biochemistry. The skin is the largest interface between the human body and the external environment (18) (19). In a healthy adult, the human skin has a surface area of approximately 2 m^2. By comparison, the absorption mechanism of orally delivered drugs in the intestinal epithelium has more chemical and physical restrictions, as it has a much larger surface area (300 – 400 m^2) (20). In general, the drug is swallowed and enters the GI tract and it release at the intestine proceeds by diffusing inside the mucus layer as shown in **Figure 2**. The mucus in the small intestine is discontinuous, whereas in the stomach and large intestine (colon) there are two layers (21). The drug is delivered through the mucus layer and from there diffuses through pathways involving a long path through tight junctions (TJs) as well as epithelium cells. This process continues until the drug is carried all the way through the capillary layer covering the epithelium layer.

The GI tract consists of the oral cavity, esophagus, stomach, small intestine and colon, each with different properties that need to be considered when designing delivery systems and studying drug release mechanisms (**Table 1**) (22). In general, drug uptake in the GI tract is restricted by complex physiological barriers in the different GI tract regions. The GI tract has a naturally low permeability to the bloodstream and foreign molecules, such as orally delivered drugs (22). The

bottlebrush-like architecture of mucin in the lipid-rich matrix of mucus, embedded gastric glands in the stomach with the acidic environment, residence time, microbiome, and permeability across the intestinal epithelium should be considered for the design of carriers that facilitate oral delivery of small molecules, proteins, and peptides (23). The main obstacles that exist for oral drug delivery are the biochemical, mucus diffusional, and cellular permeability barriers of the GI tract. The site of drug absorption is determined by the type of drug, as well as local environmental conditions such as pH, enzymes, mucus barriers, drug residence time, and GI surface area (24).

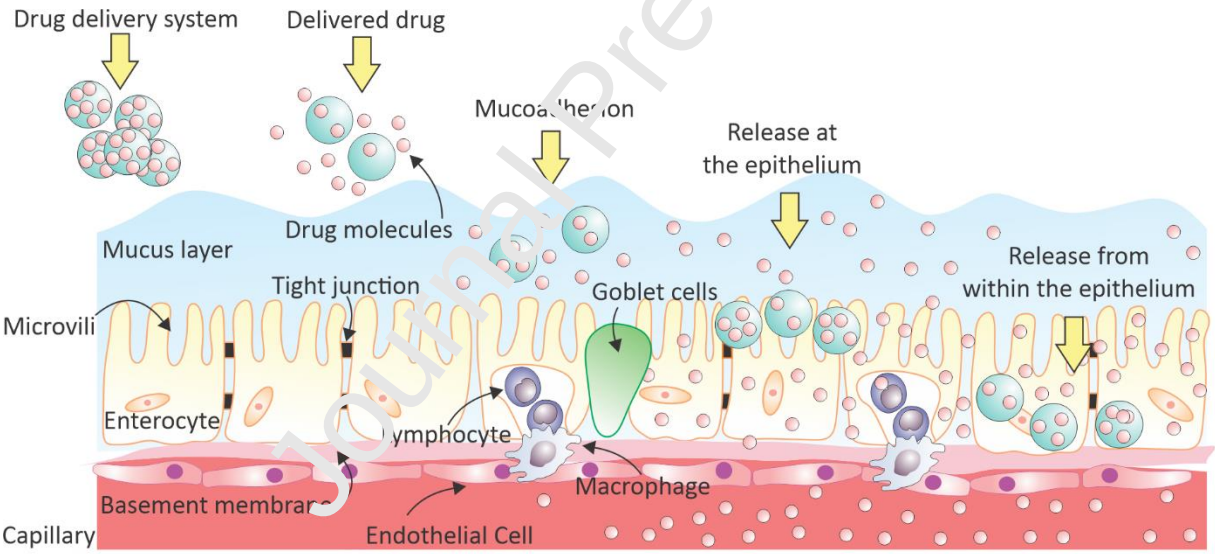

Figure 2. Schematic illustration of drug release and absorption mechanisms for orally delivered drugs in the large surface area of human intestinal epithelium.

Table 1. Characteristics of different segments of the human GI tract (22).

	pH	Length (cm)	Mean Diameter (cm)	Mucus Thickness (μm)	Mucus Turnover Rate (hours)
Stomach	0.8 – 5	20	N/A	245 ± 200	24 - 48
Duodenum	~ 7	17 - 56	4	15.5	24 - 48
Jejunum	≥ 7	280 - 1000	2 - 2.5	15.5	
Ileum	≥ 7		3	15.5	
Colon	7 - 8	80 - 313	4 - 4.8	135 ± 25	24 - 48

2.1. Biochemical Barriers

Enzymatic and pH degradation act together as the major biochemical barriers for the bioavailability of orally administered therapeutics (**Figure 3a**). The presence of drug-degrading enzymes and acidic pH results in an approximately 94-98% loss of ingested biologic drugs due to deamidation, oxidation, or hydrolysis (25). The stomach's digestive fluid is composed of hydrochloric acid, protein-digesting enzyme pepsin, and mucus secreted by gastric glands, which cause an acidic environment (pH=1.2-3). In addition to the harsh acidic environment of the stomach, digestive enzymes such as pepsin also pose challenges for oral drug delivery. Lipases in the stomach can also contribute to the hydrolysis of drugs with hydrophobic regions. The small intestine can also account for the digestion of drugs, as digestive enzymes, such as trypsins, chymotrypsins, carboxypeptidases, and elastases are present in high concentrations (26). Finally, the colon provides a longer residence time of up to 20 h, low concentrations of

digestive enzymes, and relatively neutral pH values of 6-6.7, as well as low fluid volumes to drug ratios (27).

2.2. Mucosal Diffusion Barrier

In addition to the above-mentioned physiological barriers (pH and enzymes), mucus with a viscoelastic and hydrogel-like structure creates a strong barrier for the penetration of therapeutics from the lumen to the underlying epithelium (**Figure 3b**). The direct interaction of therapeutics with epithelial cells is restricted by two mucus layers: the outer loosely adherent layer and the inner firmly adherent layer (26). Mucus is secreted by goblet cells, with turnover rates of every 24-48 h, to eliminate the attachment of potentially harmful compounds and bacteria. The majority of mucus is composed of mucin glycoproteins, which form a viscous gel to entrap foreign particles (26). Mucus is also composed of proteins, carbohydrates, nucleic acids, lipids, salts, antibodies, and other active proteins (23). Thus, it creates a safeguard and facilitates a nutrient-rich environment for bacterial colonization and antimicrobial molecules.

2.3. Cellular Permeability Barrier

The intestinal epithelium is the outermost layer of cells exposed to luminal contents. It is composed of TJs and three different kinds of cells: enterocytes, goblet cells, and Microfold cells (M-cells) (**Figure 3c**) (28). Enterocytes are the most abundant cells of the epithelium layer and enhance the transportation of nutrients and water from the gut lumen to the bloodstream. Mucus-secreting goblet cells comprise 10–20% of epithelial cells, while M-cells that cover Peyer's

patches represent <1%. M-cells are responsible for antigen sampling and are important drug targets since they are less shielded by mucus (26). TJs are paracellular barriers for the transportation of drugs between intestinal epithelial cells (29).

Methods for drug absorption into the bloodstream relies on interactions between the therapeutic and epithelial cells whether the drug is transported through the cell or between the cells through TJs. The absorption pathways are: a) transcellular pathways through epithelial cells; b) paracellular pathways through the TJs between adjacent epithelial cells; c) lymphatic absorption via M-cells of Peyer's patches; d) receptor and transcytosis-mediated endocytosis, commonly conducted by the vitamin B12 uptake pathway or by hydrogen-coupled peptide transporters, transferring receptors, and IgG neonatal receptors (26). In the next sections we will discuss how micro and nanoscale technologies allow for the bypass of biochemical and mucosal diffusion barriers to enable successful cellular uptake.

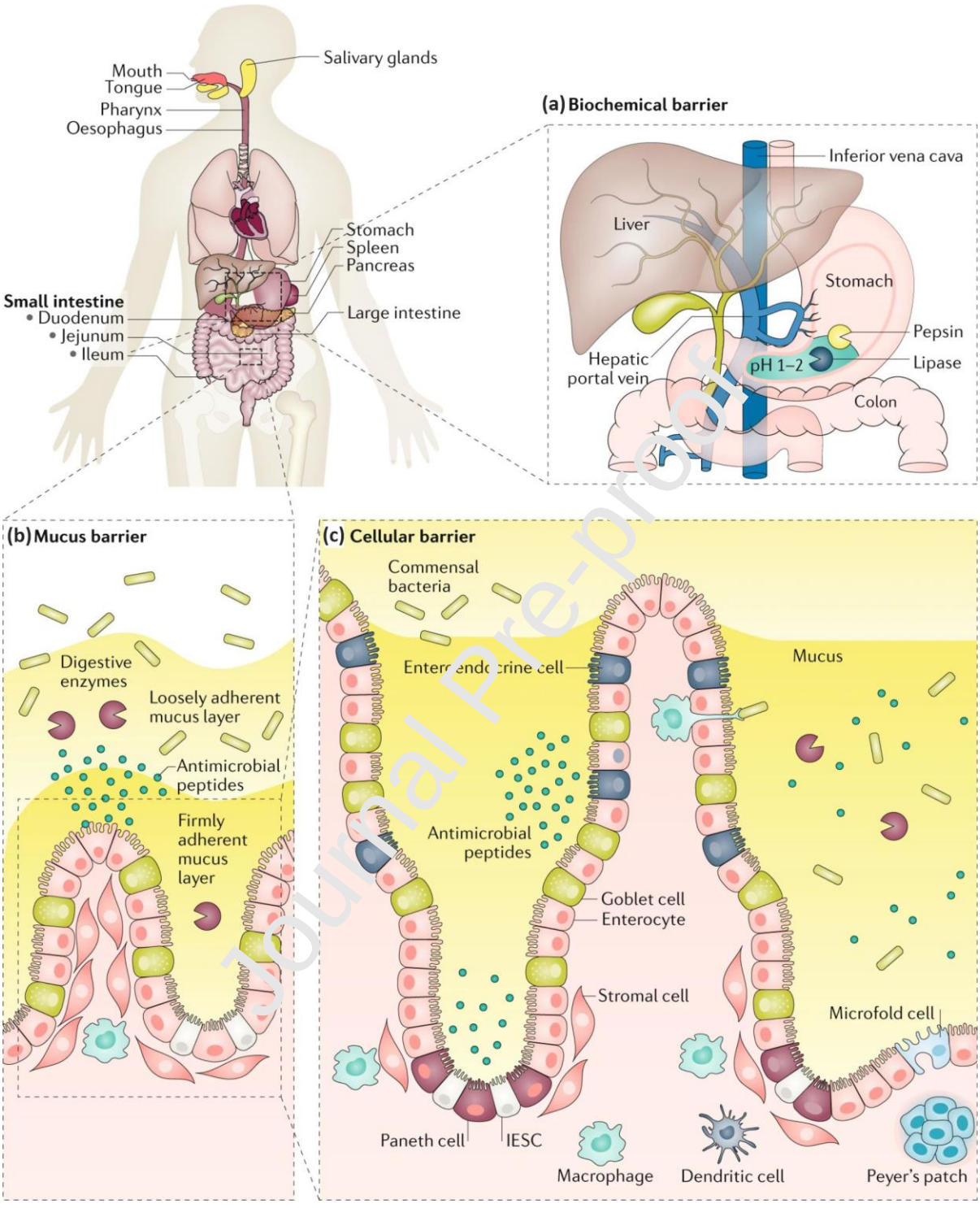

Figure 3. A schematic of physiological barriers in oral drug delivery including: (a) biochemical barriers, (b) mucus barrier, and (c) cellular barriers to oral drug delivery. Reprinted by permission from Springer Nature (26) Copyright (2019).

3. MICRO AND NANOSCALE FABRICATION TECHNIQUES

Micro-/nanoscale fabrication technologies, including lithographic techniques and microfluidics, have opened up important opportunities to further develop the fields of tissue engineering and drug delivery (1, 30). Microfabrication has been implemented for drug delivery because of its capability to combine different characteristics, including the ability to make precise shapes and sizes (e.g. needles, non-symmetrical features) that increase the contact area of the drug delivery system with the GI tract and precise sizes or reservoirs (multiple or single) to control drug release. These microfabricated devices can be further engineered to be stimuli-responsive and bioadhesive. In addition to drug delivery applications, microfabrication technologies provide great advantages in the generation of biomimetic GI tract prototypes, integrating physiological cues, flow, and biomimetic structures (1, 31). There are a variety of methods to fabricate micro- and nanoscale systems for controlled drug delivery applications, including emulsion, assembly, photolithography, mold replication, microchining, micromilling, deposition, etching, and laser ablation (32, 33).

3.1 Emulsion and self-assembling systems

Emulsion and self-assembly based systems of fabrication are some of the most widely used techniques for the development of nano and microparticles. Emulsion fabrication, as well as nano-precipitation techniques, rely on the phase separation of hydrophobic polymers in aqueous solution (34-36). Such oil-in-water emulsions are frequently used with biocompatible polymers, such as poly(lactic-co-glycolic acid) (PLGA) and polycaprolactone (PCL). These emulsion

techniques are compatible with drug encapsulation, either with hydrophobic drugs and single emulsion techniques, or with hydrophilic drugs and a double emulsion technique, which creates water-oil-water particles. Emulsion techniques allow for the fabrication of particles of varying size, ranging from tens of nanometers to hundreds of microns (37). The size of the particles is dictated by the sheer imparted during the emulsification process which is affected by the properties of solutions, as well as the concentration of polymer and surfactant in the aqueous phase. The surfactant controls the phase separation behavior during emulsification (38). PLGA-based reverse emulsions are not able to generate hydrophilic polymer particles in oil phase. Therefore, these emulsions are less common for clinical applications in oral drug delivery where the particle needs to stably traverse aqueous environments. However, in general, some PLGA-loaded particles have been used in clinic for other drug delivery systems.

Self-assembly fabrication approaches are directed by a variety of noncovalent forces such as hydrophobic-hydrophilic and electrostatic interactions, which can offer more precise control than standard emulsion or precipitation techniques and allow for the fabrication of diverse shapes including spherical, fibrillar, and ellipsoidal particles (39-41). Self-assembling systems are typically size-limited to the nanoscale, with the particles commonly reported in the tens of nanometers to low hundreds of nanometers in diameter. Similar to emulsion techniques, self-assembling systems have been used to encapsulate both hydrophobic and hydrophilic cargo for oral drug delivery applications.

3.2. Electrospinning

In electrospinning, a high voltage source is used to make micro- or nanofibers from a polymer solution or melt. An electrostatic interaction between a grounded collector and charged polymer solution is formed when the polymer expelled from a metal needle, forming a cone at the base of the needle called a Taylor cone. A fiber jet is ejected from the Taylor cone as the electric field strength exceeds the surface tension of the liquid. By travelling the fiber jet through the air, the solvent evaporates, consequently results in the deposition of solid polymer fibers on the collector. Fibers generated by this process usually have diameters on the order of some hundred nanometers. The capability to simply generate materials at different sizes in a rapid and simple manner has made a great interest in electrospinning for tissue engineering and drug delivery applications (42, 43). Researchers have widely used this technology in drug delivery because it is easy to modulate the release profile of drugs based on properties of polymeric materials and it is compatible with a variety of drugs and biopolymers (44).

3.3. Lithography

Photolithography involves the transferring of a photomask's pattern onto a photoresist by exposure to light (1, 45, 46). The photoresist layer is employed to transfer the pattern to a material after development. The wavelength of light exposure is the dominant limitation of photolithography. High resolution nanostructures can be made using more advanced techniques, such as ion beam lithography and electron beam lithography (33).

Soft lithography is a complementary version of photolithography. While photolithography has worked well to deal with photoresists (47), soft lithography expands the capabilities of

photolithography. Soft lithography can process a variety of elastomeric materials. Polydimethylsiloxane (PDMS) is commonly used for soft lithography applications due to its properties of biocompatibility, low cost, chemical inertness, low toxicity, mechanical flexibility and durability, and versatile surface chemistry. Additionally, fabricating PDMS devices requires minimal equipment (48, 49).

3.4. Microfluidics

Microfluidic systems are capable of handling and transporting small volumes of fluids through microchannels and they can be created with photo and soft lithography. Microfluidics have been used as a great tool for designing drug delivery systems (45, 50). Drug delivery to target sites can be done in an efficient and well-controlled manner with desired rates using microfluidic platforms via implantation, localization, precise control, automation, and integration of the platform (51). Microfluidics can fabricate materials with high precision and recapitulate *in vivo* conditions for drug screening (52-54) and drug discovery (54, 55) because of its capability to provide physiologically relevant fluid flow (1). This technology has become an essential part of cellular assays for the analysis of oral drug absorption.

Recently, different microfluidic-based platforms have been developed to produce and screen drug nanocarriers. Microfluidic drug development platforms provide high-throughput, reproducible, and low-cost methods for producing, screening, and optimizing nanocarriers. The properties of synthesized nanocarriers, such as morphology, drug loading capacity, and release kinetic parameters, can be easily and effectively modified and optimized by adjusting the

channel geometries and flow rate. Microfluidics facilitate the efficient and low cost production of various micro and nanoparticles, composed of different materials and therapeutic agents, with high loading capacity and controlled release at small scale, which minimizes the amount of required reagents, as compared to bulk mixing methods (56).

Microfluidic-based synthesizers are classified as diffusion and droplet-based methods (**Figure 4**) (57). Hasani-Sadrabadi *et al*. fabricated a microfluidic device for generating core-shell chitosan-based nanoparticles for oral delivery of hydrophobic anti-cancer drugs to treat colorectal cancer tumors (58). The core of the nanoparticles was composed of a hydrophobic modified *N*-palmitoyl chitosan for efficient loading of hydrophobic drugs. The core also allowed for the formation of nanoparticles through the self-assembly of chitosan chains, without using a cross-linking agent. The self-assembly of chitosan chains occurred in the first microreactor with hydrodynamically focused flow controlling the mixing time of flow streams. A Tesla micromixer was also designed for efficient mixing and coating of nanoparticles by pH-responsive layer of Eudragit ((pH-sensitive poly(methyl methacrylate)) in a controlled manner. The Coanda effect generated in this Tesla-designed micromixer enhances mixing efficiency. It results in deflection of a part of the flow toward the narrow side of the channel and flow of other part through the curved side for an efficient mixing of two flow streams. In the latter study, the thickness of the shell in synthesized particles was controlled by the ratio of sheath flow rate to the main flow rate.

Although the generation of particles can be controlled by tuning the flow rate in diffusion-based mixing methods, the continuous flow regime limits efficient diffusion and the reaction between materials boundaries between separate flow streams. In droplet-based techniques, each droplet serves as a microreactor for an independent reaction, resulting in higher production efficiency for

drug-loaded nanocarriers. This method allows for precise control of nanocarrier size, drug loading efficiency, and total amount of nanocarrier produced. Nano-in-micro platforms can be introduced based on droplet-based methods to prepare nanoparticles encapsulated inside microparticles. Araujo *et al*. produced a multifunctional composite for the oral delivery of a mixture of glucagon-like peptide-1 and an enzymatic inhibitor (dipeptidyl peptidase 4) as antidiabetic drugs for synergistic therapy using droplet-based microfluidic techniques (59). The glucagon-like peptide-1 was first loaded in different PLGA and mesoporous silicon biomaterials to limit its rapid degradation in the intestine and the resulting nanoparticles were further functionalized by mucoadhesive polymers, such as chitosan and cell penetrating peptides. Similar Nano-In-Micro platforms were utilized in several other investigations to encapsulate nanoparticles inside of micro structures, such as halloysite nanotubes-polymer, mesoporous silicon-solid lipid, and mesoporous silicon-polymer composites, for oral drug delivery applications (60),(61),(62).

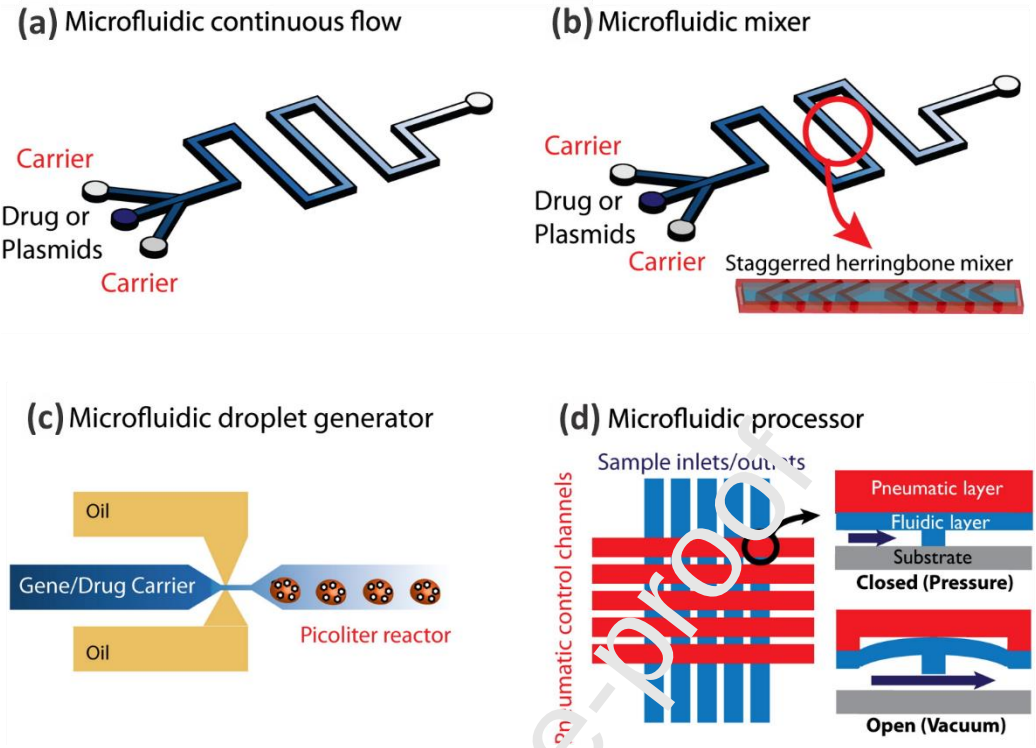

Figure 4. Microfluidic approaches to fabricate nanocarriers for oral drug delivery. Different diffusion- and droplet-based microfluidic platforms for preparation of nanoparticles including (a) microfluidic continuous flow, (b) microfluidic mixer, (c) microfluidic droplet generator, (d) microfluidic processor. Reprinted from (57) Copyright (2013), with permission from Elsevier.

3.5. Three-dimensional Printing

Three-dimensional (3D) printing technology is a promising fabrication technique that has received wide interest in biomedical engineering and drug delivery applications to provide complex drug release profiles, precise drug dosing, novel drug delivery devices, and 3D printed polypills (63). 3D printing technology offers low-cost applications compared to conventional systems since it does not need several unit operations and requires minimal human intervention

(64-67). Typically, 3D printing works through the digitally-controlled and layer-by-layer deposition of materials to make different 3D constructs and desired geometries without the need for molds or machining (68-70). This technology can offer precise and personalized dosing for treatment of different patients (64, 71). However, 3D printing technology may need multiple steps and sophisticated equipment to synthesize oral delivery platforms in a commercial setting.

Spritam® (72) was approved as the first 3D printed drug tablet by the Food and Drug Administration (FDA) and this led to great interest into the implementation of 3D printed drug delivery systems. The capability to handle low volumes of fluids with spatial control facilitates the preparation of devices with interesting compositions and geometries. The flexibility of 3D printing enables the preparation of systems with multiple drugs and specific release profiles (73). Various 3D printing technologies can be employed for the development of pharmaceutical formulations. Technologies such as digital light processing, selective laser sintering, continuous liquid interface production, stereolithography, fused deposition modelling (FDM), material jetting, inkjet deposition, and binder jetting are the most common 3D printing technologies that have been implemented in pharmaceutical research and customized drug formulation (68, 74). The most challenging part of using 3D printing for the fabrication of drug delivery systems is the development of functional inks that retain the features necessary for sustain release and bioadhesion. Some examples of such inks are commercially available (e.g., Resomer® filaments for 3D printing by Evonik). These could be potentially used for the development of oral delivery carriers.

Progress in 3D printing technology has led to innovative medical devices, as well as customized drug delivery systems. 3D printing technology offers multiple formulation options compared to

conventional drug delivery systems. Moreover, it provides an opportunity to load multiple drugs into a single device and make multifunctional drug delivery systems and dimension-specific drug formulations to attain tunable drug release profiles (63). More recently, two-photon lithography technologies were designed to offer 3D printing capabilities at nanoscale that could be interesting for oral delivery systems where a nanoparticle has to be of specific shape.

4. MICRO AND NANOSCALE CARRIER TYPES

The unique physiochemical properties and high surface area to volume ratio of nanoparticles facilitate high loading of drugs into them through encapsulation or formation of chemical-physical bindings with their functional groups. The stability of some nanoparticles in aqueous physiological environments allows for successful loading and delivery of poorly water-soluble drugs (75). Additionally, nanoparticles can be functionalized using different targeting or imaging agents in order to be utilized for imaging and targeted drug delivery applications (76). Specifically, drug delivery using biocompatible nanocarriers has been introduced as an effective solution to overcome some challenges involved in oral drug administration, particularly for drugs with low stability, bioavailability, and solubility. Nanoparticles can protect drugs from the acidic environment of GI and the secretion of mucus to enhance membrane permeability, which promotes drug absorption and bioavailability. Bioadhesive properties of nanoparticles enhance the permeation of drugs by increasing residence time in the GI tract (77).

Microparticles are common oral delivery systems in addition to nanoparticles, and offer the means to improve the bioavailability of pharmaceuticals through the control over shape, size,

geometry, and functional characteristics of the particle (78-81). During the past few decades, microparticle technology has been extensively applied for various applications in therapeutic and pharmaceutical fields, such as the delivery of anti-inflammatories (82, 83), antibiotics (84, 85), chemotherapeutics (86, 87), proteins (88), and vitamins (89, 90). Microparticle sizes range from 1 to 1000 μm and they exist in various structures (91). Microparticles may be characterized as either homogenous or heterogeneous structures depending on the formulation and processing.

Using the techniques described above, drug carriers can be fabricated in multiple size regimes from different materials, with tunable physical and chemical properties. We will briefly describe some of the different classes of carriers, before discussing functionalization strategies and methods of targeting the GI system or systemic bloodstream circulation via oral administration of these carriers.

4.1. Lipid-based Nanoparticles

Lipid-based nanoparticles, such as solid lipid nanoparticles (SLNs) and liposomes, are a group of nanoparticles used extensively for oral drug delivery due to their excellent biocompatibility, similarity with biological membranes, and drug loading capacity. Liposomes were the first nanocarriers approved by FDA for clinical use (92). They are composed of an aqueous core, which encapsulates hydrophilic drugs, and an amphiphilic lipid bilayer, which allows for the loading of hydrophobic drugs (93) (**Figure 5a**). Additionally, they can be utilized as the carrier of biomolecules like peptides, antigens or antibodies which are covalently attached to the polyethylene glycol (PEG)-coated (PEGylated) surface of liposomes (**Figure 5c**).

Functionalization of liposomes with PEG limits their recognition by phagocytic cells, resulting in longer circulation time and enhanced biodistribution (94). Furthermore, different targeting agents can be conjugated to the external surface of the liposomes for the enhanced targeted delivery of therapeutic agents to specific cells. The encapsulation of hydrophilic drugs and biologics inside of liposomes significantly improves their cellular absorption (95),(96). However, the efficiency of conventional phospholipid or cholesterol-based liposomes is seriously affected by instability of their lipid vesicles and phospholipid hydrolysis or oxidation in the GI track. Therefore, chemical and physical functionalization is employed to increase their residence time in the intestine and enhance their stability, as we will discuss in later sections.

SLNs contain a monolayer phospholipid shell and a solid lipid core (97). These nanoparticles can be utilized for encapsulation of lipophilic ingredients and insoluble drugs. SLNs are prepared from synthetic or natural biodegradable and biocompatible lipids, such as fatty acids, triglycerides, steroids, and phospholipids. They have shown high stability in the harsh conditions of the GI, as compared to other lipid nanoparticles, such as liposomes. Functionalizing and non-covalent coating of SLNs with carboxymethyl chitosan can further enhance their stability and drug bioavailability (98). Several investigations reported higher oral bioavailability of hydrophobic drugs, such as nitrendipine and nimodipine, loaded in SLNs (99),(100). However, the encapsulation of hydrophilic drugs in SLNs is limited due to the particle's hydrophobic nature. Incorporation of a SLN core with hydrophilic viscosity-enhancing polymers, such as PEG, through a water-oil-water double emulsion method is proposed for higher loading efficiency of hydrophilic drugs in the core of SLNs. In this strategy, the core of orally administered SLNs consists of a solid lipid core and a hydrogen-bonded rich aqueous phase encapsulating insulin, which is either dispersed in the lipid phase or is formed like a central core

in the lipid matrix (**Figure 5b**) (101). Although surface functionalization of SLNs with PEG enhances their hydrophilicity, a reduction in muco-adhesion of SLNs is also observed.

4.2. Polymeric Nano and Microparticles

Natural polymers, synthetic polymers or their combinations, cellulose derivatives, polysaccharides or proteins, and waxes of plant or animal origin can be used to prepare nano or microstructural oral drug delivery materials. Polymeric particles can interact with the mucus through electrostatic, van der Waals, hydrophobic, or hydrogen-bonding interactions, which may lead to long residence time of drugs in the absorption region (102). However, more research is required to reduce their undesired adhesion to non-target regions (103). Several studies have demonstrated more efficient absorbance of hydrophobic polymeric nanoparticles to the Peyer's patches when compared to less hydrophobic or hydrophilic particles (104), showing the important role of hydrophobicity in polymeric nanoparticles.

Polymeric nano and microparticles made of PLGA, poly(lactic acid) (PLA), PLA-PLGA copolymer, poly(acrylic acid) (Carbopol) and poly(N-isopropylacrylamide) have been extensively explored in the pharmaceutical field as carriers for oral drug delivery due to their biocompatibility, enzymatic degradation, bioadhesion, and rapid removal in the mucus. The biodegradability and biocompatibility of these polymers have been approved for various medical and pharmaceutical applications, including drug delivery by both the FDA and the European Medicine Agency (102).

Of the polymers available, PLGA is one of the most commonly researched polymer for oral drug delivery, owing to its FDA approval (105), biocompatibility, and biodegradability. In particular, PLGA is suitable for oral delivery of water insoluble anti-cancer drugs, such as paclitaxel and curcumin, due to its hydrophobic nature (106). The encapsulation of hydrophobic anti-cancer drugs in PLGA nanoparticles resulted in enhanced bioavailability of drugs, which is due to improved aqueous stability of loaded drugs in PLGA nanoparticles, compared to free drug, and sustained drug release by degradation of the PLGA nanoparticles (106).

In addition to nanoparticles, different microparticle, polymer-based systems have been introduced for oral drug delivery for *in vivo* and *in vitro* studies which are summarized in **Table 2**. A natural polymer-based microencapsulation system was proposed by Vasiliu *et al*. They prepared microparticles based on polyelectrolyte complexes between two polysaccharides (xanthan gum and gellan) and an acrylic ion exchange resin to obtain a novel antibiotic delivery system. The effects of contact time, temperature, and drug concentration on the patch efficacy were optimized using batch adsorption studies (107). Wang *et al*. reported a monodisperse and temperature-induced self-bursting microcapsules for encapsulating hydrophobic compounds. The proposed microcapsules had a hydrophobic core and a thermo-responsive shell comprised of poly(*N*-isopropyl acrylamide) and embedded superparamagnetic Fe_3O_4 nanoparticles (108). Koetting *et al*. designed hydrogel microparticles and used them for oral delivery of therapeutic proteins (109). More efficient surface engineering technologies, advanced bioadhesive functionalization, and combination with smart materials will result in the development of highly functional microparticle-based oral drug delivery systems.

Another widely used nanocarrier for oral drug delivery is the polymer micelle, which is formed by self-assembly of amphiphilic polymers above the critical micelle concentration. This fabrication process provides a core-shell structure, which allows loading of poorly water soluble drugs in hydrophobic cores with enhanced bioavailability and stability (110),(111). For example, an amphiphilic block copolymer consisted of a micellar shell-forming PEG block and a core-forming poly(2-(4-vinylbenzyloxy)-*N*,*N*-diethylnicotinamide) block, while *N*,*N*-Diethylnicotinamide in the micellar inner core resulted in effective paclitaxel solubilization and stabilization (110). Suitable copolymers for oral drug delivery should have self-assembling capabilities in water, water, biodegradability, biocompatibility, high stability, and residence time in the GI track. Different polymeric micelles containing polyethers or polyesters have been proposed for oral drug delivery. Pluronics composed of poly(ethylene oxide) (PEO)-poly(propylene oxide)-PEO copolymer, also known as of *Poloxamer* are commonly used polymers for micelle assembly. Some polymeric micelles exhibit pH-responsive disassembly with lower release rate in acidic environments, reducing initial burst release of drug (112). In addition to micelles, reverse micelles prepared in an oily solution with an interior hydrophilic core and exterior hydrophobic layer can be used for the encapsulation and sustained release of hydrophilic drugs (113).

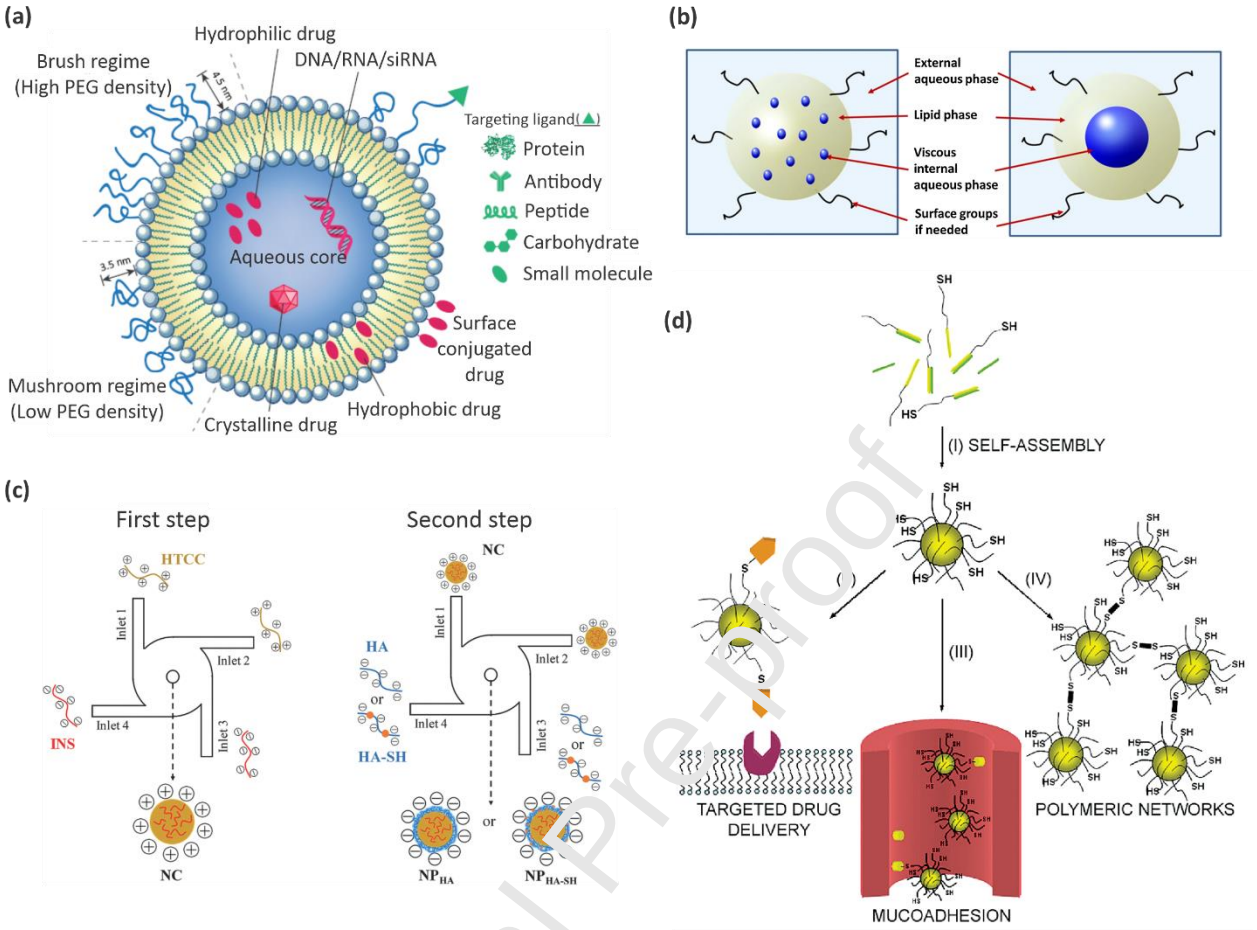

Figure 5. Fabrication and characterization of nanocarriers for oral drug delivery. (a) Schematic image of possibilities for drug loading and functionalization with different targeting and therapeutics ligands in liposomes. Reprinted from (92) with permission from Elsevier. (b) A strategy for loading hydrophilic drugs in the core of solid nanoparticles (blue color) by generation of a hydrophilic viscose phase in the core. Reprinted from (101), Copyright (2016), with permission from Elsevier. (c) A two-step preparation method for insulin-loaded core-shell nanoparticles composed of a modified chitosan core coated with thiolated hyaluronic acid through electrostatic (114). Copyright (2018) Wiley-VCH Verlag GmbH & Co. KGaA. Reproduced with permission. (d) Self-assembly of cationic copolymers (yellow color) with anion biomacromolecules (green color) to form polymer micelles with targeting agents can improve

mucoadhesion and can generate polymeric networks of micelles. Reprinted (adapted) with permission from (115) Copyright (2005) American Chemical Society.

Table 2. Microparticle systems for applications in oral drug delivery.

Material	**Model of Drug**	**Applications and Benefits**	**References**
PLA	Insulin	A solvent extraction method was used to prepare different sized microcapsules and the highest insulin release profile was obtained in 7-12 h.	(116)
	Lovastatin	PLA microspheres enhanced the bioavailability of drugs for gastroretentive drug delivery and prolonged the drug circulation time *in vivo*.	(117)
PLGA	Amifostine	A solvent evaporation technique was used for Amifostine encapsulation and oral controlled release. It was observed that 50% of the drug was released within the first 6 h and 92% within 12 h.	(118)
	Plasmid DNA (pDNA)	pDNA vaccine encapsulated PLGA microcapsules was synthesized via a solvent evaporation method. The pDNA was protected from degradation in the GI system.	(119)
	Insulin	Magnetic nanocrystals and insulin were encapsulated in PLGA microparticles to delay drug transition using a magnetic field.	(120)
	Curcumin	PLGA particles with different molecular weights were prepared by an emulsification-solvent evaporation method to encapsulate curcumin. The results showed that the bioavailability of high molecular weight PLGA particles was better than that of low molecular weight PLGA particles and curcumin.	(121)
		Physicochemical properties and *in vivo* therapeutic activities of porous and nonporous PLGA microparticles were studied. Ammonium bicarbonate was used to create the porosity and *in vivo* experiments showed that oral administration of porous microparticles exhibited therapeutic efficacy	(122)

		against Ulcerative colitis compared to nonporous microparticles.	
PCL	Bovine serum albumin	PCL microparticles for use in oral vaccine applications were produced in sizes (5-10 microns) that can be taken by M cells in Peyer's patches.	(123)
	Manidipine dihydrochloride	In order to treat high blood pressure, PCL microparticles containing Manidipine dihydrochloride with an antihypertensive effect for up to 24 h were developed.	(124)
Polyvinyl alcohol (PVA)	Ornidazole	Controlled release of the drug molecule in the GI tissue was provided with PVA microparticles prepared using different ratios of PVA to starch.	(125)
Methylcellulose	Thymol	Methylcellulose and hydroxypropyl methylcellulose phthalate were used to produce Thymol encapsulated microspheres. *In vivo* pharmacokinetic studies showed that the microparticles could be used for local treatment of intestinal infections.	(126)
Ethylcellulose	Propranolol	Ethylcellulose microparticles containing Propranolol hydrochloride were prepared using a modified solvent evaporation method, and its use for the treatment of hypertension was studied.	(127)
Carboxymethyl cellulose sodium	Flurbiprofen	Chitosan-coated and uncoated sodium carboxymethyl cellulose and polyvinyl alcohol microspheres were synthesized and crosslinked with Fe^{3+} ions. The chitosan-coating provided a slower release and a lower burst effect.	(128)
	Progesterone	Low methoxy amidated pectin-sodium carboxymethyl cellulose microspheres were prepared, and Zn^{2+} and Al^{3+} ions were used for crosslinking. The particles were tested in colon-targeted drug delivery.	(129)
Chitosan	Ovalbumin	Porous chitosan microparticles, which can be taken up by the epithelium of the Peyer's patches, were synthesized and used as a vaccine delivery system	(130)
	Curcumin	A sustained release of curcumin in the intestinal tract was reported for N-trimethyl chitosan modified SLNs	(131)

	Progesterone	Zn-pectinate/chitosan particles were made to increase the oral bioavailability of progesterone and to use the particles as the colon targeting system.	(132)
Sodium hyaluronate	Vancomycin	Drug loading capacity of vancomycin in porous and degradable hyaluronic acid (HA) microparticles were increased by the HA porosity, and the drug release degree could be modified by the degradability of the particles.	(133)
Sodium alginate	Curcumin	Alginate microparticles crosslinked by ion gelation were used for controlled release curcumin solubilized in the lipid phase.	(134)
	Insulin	The efficacy of microparticles prepared using different amounts of mucin and alginate on controlled insulin release was assessed.	(135)
Gelatin	Vascular endothelial growth factor (VEGF)	Gelatin microparticles were designed for the controlled release of VEGF, and a regular controlled release was achieved by modifying the degree of microparticle crosslinking.	(136)
	Bone morphogenetic protein-2	Gelatin microparticles were evaluated for controlled release of bone morphogenetic protein-2, and the release profiles were compared with PLGA microparticles.	(137)
	Ciprofloxacin	Ciprofloxacin, a water-insoluble antimicrobial drug, was encapsulated in gelatin as a result of a one-step process by spray drying an aqueous solution.	(138)
Polymethacrylic acid-polyethylene glycol-chitosan	Insulin	Surface thiolation was used to increase the drug release performance of hydrogel-based oral insulin delivery systems.	(139)
Chitosan-carboxymethyl starch	5-aminosalicylic acid	Chitosan-carboxymethyl starch particles were synthesized via a casting technique with high encapsulation performance as a drug delivery system for the colon.	(140)
Chitosan-graft-polyacrylamide	Ibuprofen	Chitosan-graft-polyacrylamide copolymer was produced by cerium (IV) ammonium nitrate-induced free radical graft polymerization, and the release profile as a function of crosslinker amount and drug to polymer ratio was investigated.	(141)
Poly(butylmethacrylate-co-(2-dimethylaminoethyl) methacrylate-co-	Micronutrients (iodine, zinc, iron, and vitamins (B2, B12, C, D, and A),	Poly(butylmethacrylate-co-(2-dimethylaminoethyl) methacrylate-co-methylmethacrylate) was used to encapsulate different micronutrients, and the	(142)

methylmethacrylate)	biotin, folic acid, and niacin)	encapsulation was shown to provide stability against a variety of factors.	

4.3. Inorganic Nano and Microparticles

In addition to organic nano and microparticles, inorganic particles, including mesoporous silica, gold, silver, iron oxide, quantum dots, carbon nanotubes, and graphene oxide nanoparticles, have found wide applications for oral drug delivery due to their exceptional physiochemical properties (143),(144),(145). High stability in aqueous conditions, along with acidic and enzymatic environments (1), feasibility of functionalization (2,3), large surface area with a high loading capacity (4,5), enhanced membrane permeability to the cells (6,7), and optical and magnetic properties (8,9) all suggest promising applications of inorganic nanoparticles not only in drug delivery, but also in bioimaging. However, their clinical applications are restricted by their poor biodegradability and biocompatibility, which requires functionalization with other biomaterials. Functionalization of inorganic nanoparticles with biocompatible ligands can be achieved during their synthesis procedure or after their preparation. Silica nanoparticles, with high porosity and surface area, possess silanol groups, which facilitate their functionalization. For example, silica nanoparticles coated with PEG slowed the release of insulin both in acidic and neutral pH (144). PEG, chitosan, and alginate coated silicate nanoparticles have also been utilized for oral delivery of insulin to enhance their mucoadhesion and biocompatibility (144),(146). Poly(amidoamine)-functionalized multiwalled carbon nanotubes loaded with hydrophobic therapeutics were modified with a carboxylate group to increase loading capacity and drug dissolution (137). Insulin-loaded silica nanoparticles ranging from 289 nm to 625 nm showed increased interaction with mucin when coated with chitosan (146).

4.4. Micropatches

Micropatches (also called wafers or films) can be designed as drug carriers with a typical size of 2-10 cm^2 and a thickness of 20-500 µm (147-149). They can be classified as melt away, rapid disintegrating, and sustained-release, referring to their different drug release rates and disintegration times as detailed by Kirsch *et al*. (150). The drug release rate can be defined by the polymer used but can also be multilayered with varying disintegration times. Oral patches are usually fabricated with laminated structure, a drug-containing bioadhesive layer, and an impermeable backing layer for increased retention (151). To improve the retention of micropatches within the intestinal lining, micropatches are designed to be thin and flat (**Figure 6a,b**). This design also minimizes exposure to the constant flow of fluids from the intestine (**Figure 6c**).

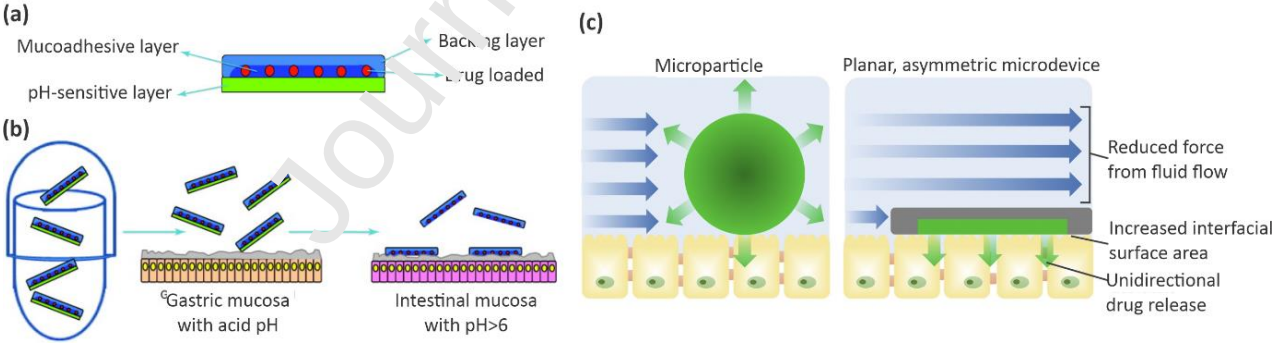

Figure 6. Micropatches in oral drug delivery. (a) Schematic representation showing a GI patch and (b) working mechanism of the hard capsule filled with mucoadhesive patches. Reprinted from (152), Copyright (2011), with permission from Elsevier. (c) In contrast to microspheres, asymmetric and planar microdevices facilitate proximal and unidirectional drug release, while

increasing residence time in the GI tract. Reprinted from (29), Copyright (2015), with permission from Elsevier.

4.5. 3D Fabricated Microdevices

Encapsulation of drugs in polymeric matrices can be achieved using several methods, such as injection molding, pressing, and 3D printing (153-156). First, a drug polymer blend can be fabricated via mixing the polymer with the desired drug and then either molded or shaped in the form of filaments for 3D printing. The release rate can be tuned by manipulating the polymer or drug concentration.

Khaled *et al.* utilized 3D extrusion-based printing to fabricate a polypill for patients under complex medication regiments. The polypill contained five distinct drugs with two independently well-defined and controlled release profiles. The drug formulations aimed to improve the drug usage for patients taking a variety of different drugs and to allow for the tailoring of a drug regimen with distinct release kinetics for each individual. The polypill demonstrated rapid and sustained release profiles based on the excipient/active ratio (157). Maroni *et al*. proposed printed 3D capsular and multi-compartment devices. The devices are separated by a 600 or 1200 μm thickness wall, for two-pulse oral drug delivery. The devices were manufactured by FDM 3D printing, which allows for larger scale production (158). Melocchi *et al*. investigated FDM 3D printing for the manufacturing of capsular devices with a shell-thickness of 600 μm using a swellable and erodible polymer (hydroxypropyl cellulose) for oral pulsatile release. This study focused on the fabrication of hollow structures via FDM and the

production of hydroxypropyl cellulose filaments by hot melt extrusion. After assembly of the capsular devices, the study reported an initial slow release followed by a quantitative and rapid liberation of the drug (159).

Li *et al*. showed that 3D printing can be utilized to fabricate oral drug delivery devices consisting of various materials with customized designs (**Figure 7a**) (160). FDM was employed for the 3D printing of glipizide-loaded filaments, which were fabricated by melt extrusion of PVA and glipizide (154). Then, the drug-loaded filament was 3D printed into a tablet shape. The tablet was composed of a core and shell comprised of various contents of glipizide. By this new design, both controlled and delayed release were possible as the composition of the outer layer controlled the release behavior of the core. In another study, Maroni *et al*. employed FDM and IM technologies to fabricate a capsular device for two pulse oral drug administration (**Figure 7b**) (158). Commercial PVA and other formulations of polymers were used for FDM and injection molding. The capsules were composed of two hollow halves with desired thicknesses with a middle partition. Varied thicknesses and compositions in each half led to faster or slower drug release in the respective halves of the capsule. FDM enabled customization of the drug delivery system and injection molding was suitable for its high throughput production. Kirtane *et al*. designed a novel oral dosage form for weekly and sustained drug release (**Figure 7c**) (161). The dosage form released long-acting antiretroviral for the prevention and treatment of human immunodeficiency virus (HIV). The device was composed of an elastomeric core connected to six separate polymeric arms that were flexible enough to be folded and placed into a capsule. The arms were embraced by a polymeric shell and filled with a drug-polymer composite, which was fabricated by melt mixing. Poly(adipic anhydride), poly(sebacic anhydride), and PEG polymers and three antiretrovirals, dolutegravir, cabotegravir, and rilpivirine, were selected for the drug-

polymer mixtures. Various polymers for drug-polymer composites enabled different drug release rates and a modular release system.

Reservoir-based microdevices are another type of microdevices designed to protect the drug against degradation and deactivation for an efficient drug release (162-166). Precise control over the amount of drug loaded into a device can be obtained by drop-on demand inkjet printing of the drugs. Marizza *et al*. developed reservoir-based microdevices for oral drug delivery of active ingredients (**Figure 7d**) (163). They employed lithography techniques to fabricate microcontainers in desired dimensions. Then, the microcontainers were filled with a precise amount of polyvinylpyrrolidone (PVP) solution via inkjet printing and ketoprofen was soaked into the PVP when supercritical carbon dioxide was used as the loading medium. The amounts of the printed polymer and loaded drug were modulated by varying printing and soaking parameters. Thus, a controlled and reproducible drug release was achieved.

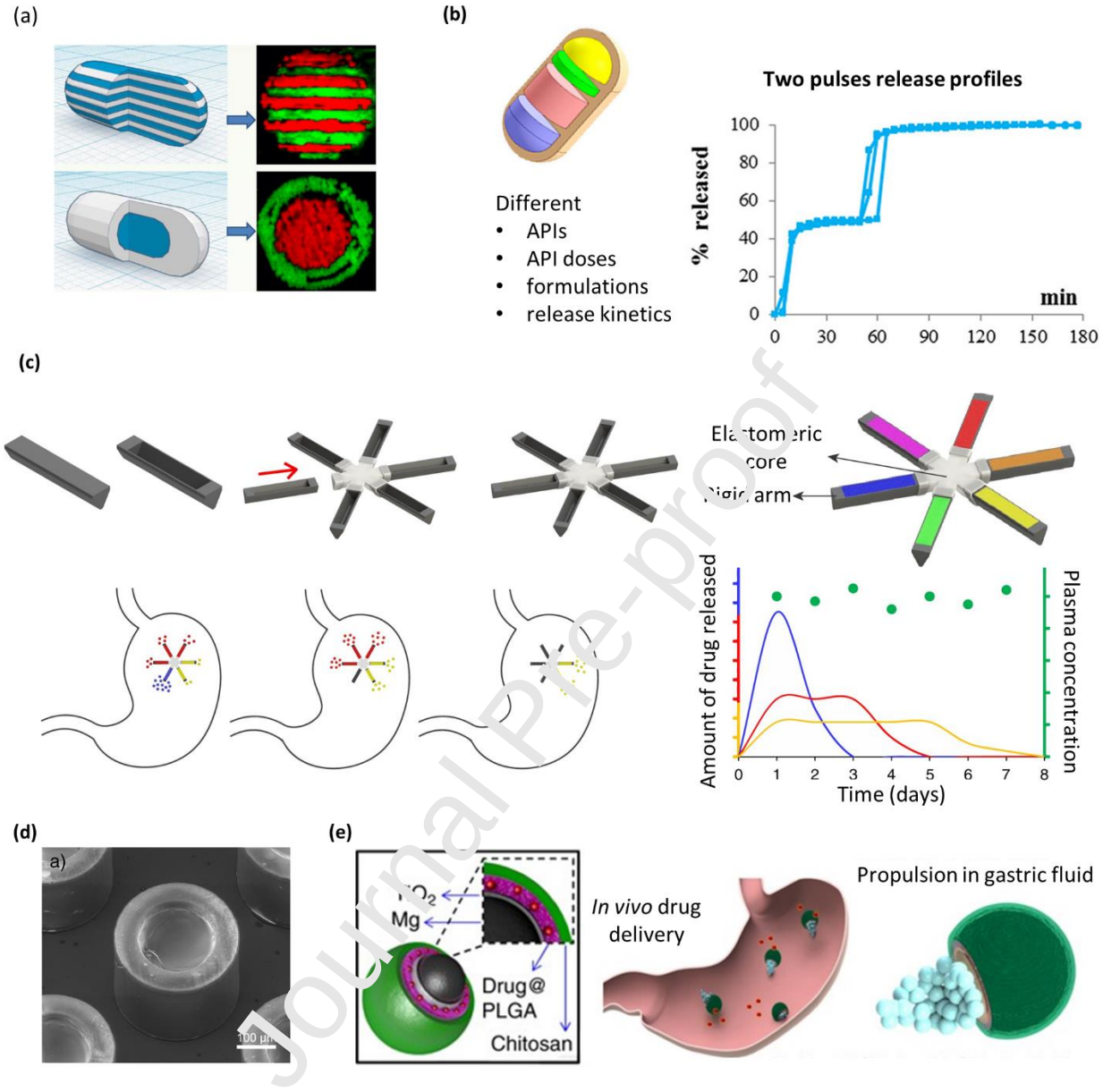

Figure 7. Fabricated devices for oral drug delivery. (a) Caplets fabricated by 3D printing with various designs of multiple materials showed by different colors. Reprinted with permission from (160). Copyright (2016) American Chemical Society. (b) 3D printed multi-compartment capsular devices with two phase release profiles. Reprinted from (158) Copyright (2017), with permission from Elsevier. (c) The device consisted of an elastomeric part (core) and six drug-loaded arms. Various polymers (blue, red and yellow) released the drug at different rates. Material from (161),

published 2018, Nature Springer. (d) Scanning electron microscopic (SEM) image of the microcontainer filled with polymer and impregnated with ketoprofen. Scale bar is 100µm. Reprinted from (163) Copyright (2014), with permission from Elsevier. (e) Schematic of the drug- loaded micromotor and drug delivery in stomach. Reprinted by permission from (163). Nature, Copyright (2017).

4.6. Dendrimers

Dendrimers have been extensively studied for oral drug delivery. Dendrimers have complex structure, divided into three parts: core, branch, and terminal groups. Dendrimers are monodispersed, usually symmetric and their molecular weight can be controlled. Additionally, physicochemical properties of dendrimers can be tuned based on their chemical structure, surface functionalization, and core structure. Most importantly, the terminal functional group in dendrimers has significant role as it can be conjugated to various biological active molecules, such as enzymes and antibodies. Polyamidoamine is the most commonly used dendrimer with core structure consists of alkyl diamine and tertiary amine branches. Several drugs, such as clotrimazole, sulfamethoxazole, propranolol, ketoconazole, triclosan have been conjugated to polyamidoamine dendrimer to test their efficacy through oral drug delivery (167-169). The latter studies proved that the activity of the drug molecules was significantly increased in conjugation with the dendrimer compared to their pure state.

5. STRATEGIES TO IMPROVE BIOADHESION OF DRUG CARRIERS

Over the years, many types of micro- and nanocarriers have been designed and fabricated for oral drug delivery. A combination of different materials, versatile fabrication technologies, and different sizes and shapes of carriers have been explored to develop oral drug delivery systems. The aim of fabricated micro- and nanocarriers is to obtain dynamic and accurate control over the drug delivery process. These drug carriers can be engineered to improve their interactions with biological systems, such as mucus barriers. This can be achieved either through the increased adhesion and retention of the carrier within a biological environment (termed bioadhesive), which has been shown to improve oral drug delivery to various regions of the GI tract. To improve bioadhesion, two main strategies are used to functionalize the drug carriers, i.e., chemical modification to the surface of the carriers and engineering the physical materials-biology interface.

The design of carriers with bioadhesive properties is considered an advance in oral, transmucosal, and transdermal delivery systems. The integration of bioadhesive properties improves the oral drug delivery of fabricated carriers. A key purpose of utilizing bioadhesive materials is to delay the transit of cargos for sustainable release of drugs at target sites, prolong the residence time in order to enhance the drug absorption process, and increase material-cellular contact, thus improving bioavailability. Bioadhesive materials have also demonstrated additional properties of inducing TJ rearrangement to enhance drug transport across epithelial barriers (29). Achieving bioadhesive properties can be classified into chemical approaches and physical approaches. In this section, we describe strategies that are used across multiple applications in oral drug delivery to further functionalize drug carriers to improve their overall bioadhesion and the interactions with tissue barriers (1, 29, 170).

5.1. Chemical Approaches to Improve Bioadhesion

Chemical bioadhesion is achieved through the combination of a material's chemical composition and structure on its surface. A common chemical approach to enhance adhesion of a material is the immobilization of target lectin and carbohydrate binding proteins onto the surface of microparticles, microdevices, or micropatches, to increase the specificity of binding receptors of intestinal epithelial cell lines. As binding to cell surface receptor results in receptor-based endocytosis of nanocarriers, this approach facilitates target delivery of carriers into cells (171).

The surface charge of carriers is another effective parameter for particle uptake in oral administration. Considering the negative charge of sugar moieties on the mucins, positively charged nanoparticles can enhance mucoadhesion through the formation of electrostatic interactions (23). Liu *et al.* showed the effect of surface chemical properties of a drug carrier in the GI tract. N3-O-toluyl-fluorouracil (TFu) was loaded into cationic SLNs (TFu-SLNs) and the particles were studied to improve the uptake of TFu. They observed that cation coated TFu-SLNs elevated the oral absorption of TFu about 2-fold in comparison with TFu suspension (**Figure 8c**). The plausible mechanism to enhance the ability of controlled drug release is increased bioadhesion of the carrier by electrostatic interaction between the negatively charged absorption mucosal surface and positively charged colloidal particles (172).

Another important method for modifying drug carriers is through the incorporation of functional biomaterials into the design of oral drug delivery systems. Chemically modified polysaccharides, such as chitosan, alginate, pectin, gelatin, and dextran, are considered the most important natural

polymers used in oral drug delivery due to their biocompatibility, bioadhesion, and enzymatic degradation. Chitosan with its positive charge enhances drug absorption and, consequently, coating nanoparticles with chitosan is an effective method to promote mucoadhesion (173). Chitosan variants with higher molecular weight have shown better mucoadhesion (174). Also, chemical modification of chitosan for enhanced mucoadhesion, physiological stability, permeability and bioavailability has been utilized for more efficient oral delivery of various drugs, including anti-cancer and peptide drugs (175),(176). For instance, thiol functionalization of chitosan exhibited enhanced mucoadhesion, which is due to covalent bonds between the cysteine and thiol groups on mucus glycoproteins (177). In a similar approach, Tian *et al*. introduced a two-step flash nanocomplexation process to fabricate core-shell nanoparticles coated with thiolated hyaluronic acid to be utilized for oral insulin delivery (114). First, a positively charged insulin-loaded nanoparticle core was fabricated through electrostatic interaction between insulin and N-(2-hydroxypropyl)-3-trimethyl ammonium chloride-modified chitosan under turbulent mixing conditions. Subsequently, the prepared positively charged nanoparticle core was coated with polyanionic thiolated hyaluronic acid to synthesize the final product (**Figure 5c**). A combination of chitosan with alginate and dextran have also been used for oral delivery of insulin (178).

Combining PLGA nanoparticles with chitosan has also been investigated to improve mucoadhesion of nanoparticles and facilitate their functionalization. In one example, Abd El Hady *et al*. studied a delivery system of diosmin in PLGA microparticles coated with chitosan and reported its effects on the gastric retention of diosmin (179). In another study, folic acid (FA)-functionalized nanoparticles were prepared through a double emulsion method by surface coating insulin-loaded PLGA nanoparticles with chitosan-FA conjugates through electrostatic

interactions to enhance their uptake and targeting abilities through FA receptors (180). Multi-layered nanocapsules of PLGA and chitosan were prepared through layer-by-layer self-assembly of PLGA and chitosan via electrostatic interactions

Functionalization of copolymers with mucoadhesive functional groups is an efficient way to enhance the mucoadhesion of micelles as well. Dufresne *et al*. prepared a copolymer micelle based on PEG-poly-(2-(N,N-dimethylamino)ethyl methacrylate) and functionalized the PEG chains with thiol groups to form disulfide bonds with the mucin, leading to improved mucoadhesion of micelles (115). In this method, thiolated copolymers with opposite charge were self-assembled in an aqueous solution to form thiolated polymer micelles with the potential for functionalization with targeting agents and improving mucoadhesion and the ability to form redox-sensitive polymer networks (**Figure 5d**).

These strategies have also been applied to liposomal particles. Coating liposomes with mucoadhesive polymers, such as chitosan through non-covalent interactions increased the residence time and bioavailability of highly hydrophilic drugs with low permeability (181),(182). Modifying liposomes with thiol groups through functionalization with thiolated polymers (thiomers) imbued liposomes with desirable properties, such as mucoadhesion and enzyme inhibition (183),(184). In addition to mucoadhesive polymers, conjugation of liposomes with ligands, such as biotin, whose receptors are expressed in the intestine, can enhance the efficacy of conventional liposomes in oral drug delivery (185). Glycans are binding sites for lectins in the membrane of cells in the GI tract; thus, functionalization of liposomes with lectin is another method to increase mucoadhesion (186).

In addition to mucoadhesive polymers, other hydrophilic polymers such as D-α-tocopheryl

poly(ethylene glycol) 1000 succinate (TPGS) (a PEG-conjugated vitamin E) can be used to enhance the residence time and bioavailability of particles for oral drug delivery. TPGS can prolong the circulation time and cellular uptake of the coated nanoparticles. TPGS increases oral bioavailability by enhancing cell membrane permeability through the inhibition of P glycoproteins (P-gp) (187, 188). This is particularly important for increasing the oral bioavailability of anticancer drugs. For example, low concentrations of TPGS increased the intestinal permeability of paclitaxel, attributed to the inhibition of P-gp (189). Therefore, TPGS stands as a potent adjuvant for orally administered chemotherapy.

5.2. Physical Approaches to Enhance Bioadhesion

The physical properties of materials, such as mechanical properties, surface area, and surface morphology affect their adhesion properties to surfaces (29). One of the physical methods to enhance bioadhesion is the utilization of different geometries/shapes of drug carriers. Tao and Desai made a direct comparison between conventional microspheres (multidirectional release) and their flat and thin device designed for unidirectional release, both coated with bioadhesive tomato lectin-poly(methyl methacrylate). They showed that despite the larger surface area of the microspheres compared to their device, the spheres remained less bounded to Caco-2 monolayers after consecutive washes. This was attributed to the smaller fraction of microsphere surface which is in direct contact to the cells (190). Similarly, in nano-adhesive conditions, increasing bioadhesion of drug carriers is associated with the multivalence of adhesive elements per surface area. As the number of adhesive elements increases, Van der Waals adhesion also increases, resulting in greater absorption of the drug. The microvilli on mucosal epithelia have protrusions

that increase surface area. Thus, fabricating nanostructured microdevices with multivalency could be a potential way to strengthen the bioadhesion of materials for targeting microvilli coated intestinal epithelium (191).

Microfabrication techniques are an advanced technology to design microdevice bodies with protruding microneedles and microposts. This permits the particle to interact with mucosa by strongly adhering to the mucosa surface and penetrating the mucus layer. As a result, an enhanced drug permeability occurs. Microneedle platforms were developed to increase the retention time in drug delivery systems (**Figure 8b**). For instance, Guan *et al*. fabricated a crosslinked bilayered system made of PEG with chitosan microparticles. These fabricated systems were structured with self-folding arms. As expected, the arms anchored to the cell surface by penetrating into mucus layer. Thus, retention time of the device, as well as resistance to surface erosion of the mucus layer, increased (192).

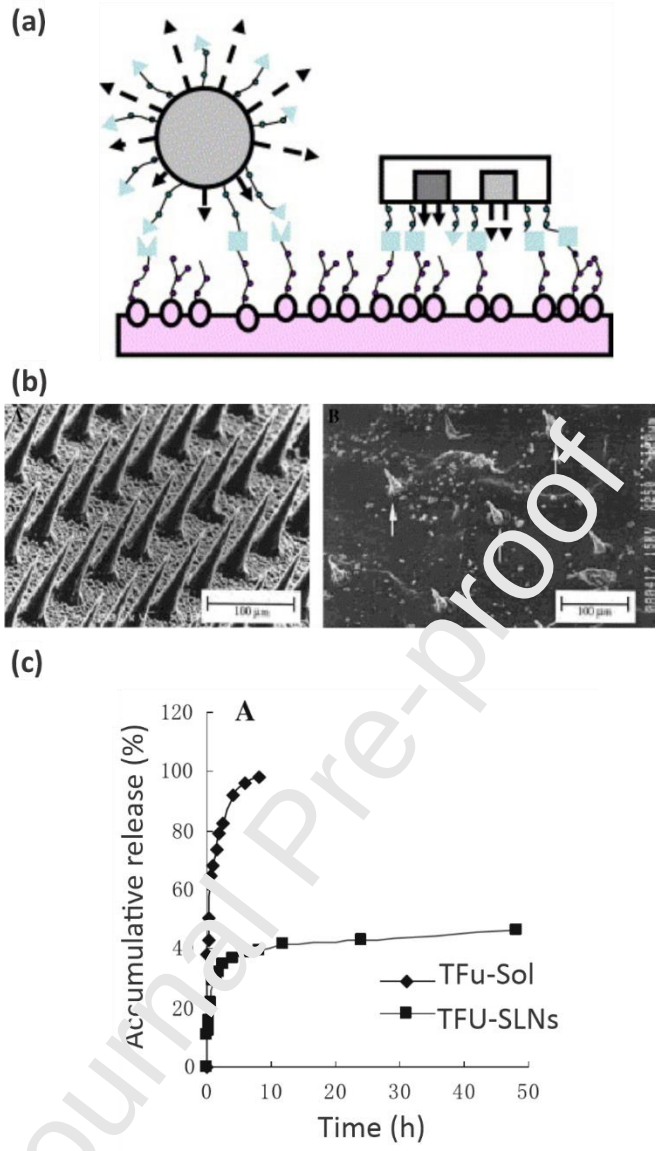

Figure 8. Physical and chemical approaches to oral drug delivery. (a) Performance of traditional drug delivery platforms (left) compared to the developed tomato lectin-modified poly(methyl methacrylate) drug delivery microdevices. (b) SEM images of the microneedles fabricated via reactive ion etching technique (left) and insertion of needle tips into the epidermis (right). (c) Release curves of TFu-SLNs and TFu-Sol in artificial intestinal juice and artificial gastric juice. Reprinted with permission from (193).

6. TARGETING THE GASTROINTESTINAL TRACT VIA ORAL ADMINISTRATION WITH MICRO AND NANOSCALE TECHNOLOGIES

For both the gastric and intestinal tracts, innovative micro and nanoscale technologies have been developed for enhanced tissue targeting and oral drug delivery. These technologies range from polymeric nano and microparticles to 3D-printed wearables. Although many different oral drug delivery techniques have been developed so far, there are many considerations for any specific active ingredient and one delivery method does not fit all drugs. Thus, it is essential to customize the delivery technique based on the type of drugs and the patient needs. Selected examples of micro and nanotechnologies used for improving the efficacy of oral administration and drug delivery to the esophagus, stomach, intestines, and colon are described in this section.

6.1. Esophageal and Stomach Delivery

Delivery of drugs to the esophagus for treatment of esophageal diseases including infections, gastric reflux, and cancers is limited by transient nature and low permeability of the esophagus. Therefore, designing an effective esophageal-targeted system with sufficient retention time during rapid transit through esophagus is of great importance. Drug-loaded nano and microparticles with prolonged contact and enhanced adhesion to the esophageal mucosa are introduced for targeting of therapeutic agents to the esophagus. Kockisch *et al*. developed an *in vitro* mucoadhesion tensile test and demonstrated efficient adhesion of polymeric microparticles, synthesized from carbopol, polycarbophil, and chitosan through water-in-oil emulsification to the

porcine esophageal mucosa (194). Additionally, magnetic systems are suggested for more efficient targeted therapy of esophageal cancer using an external magnetic force. Ito *et al*. prepared magnetic granules composed of ultrafine ferrite and a mixture of bioadhesive polymers containing hydroxypropyl cellulose and Carbopol 934. The granules were administered orally in rabbit models and were guided to the esophagus by applying an external magnet in less than 2 min, and results showed that almost all granules were retained in the target region for 2 h (195).

Rapid degradation, low stability, and poor absorption of drugs in the GI tract can be solved using stable drug delivery capsules to deliver drugs to the stomach, which is dominated by the gastric emptying time of 1-4 hours (196). In one approach, Jiang *et al*. studied oral delivery of mussel-inspired and protein-functionalized electrospun nanofibers for treating gastric cancer (197). Through the incorporation of a pH-responsive release mechanism, the authors demonstrated doxorubicin release from PCL nanofibers in acidic conditions of the stomach over neutral medium. In addition to the acidic stomach environment, there are a number of other challenges for the delivery of oral peptides, including low permeation through the intestinal epithelium, inactivation, and proteolytic degradation in the GI tract (198).

The incorporation of microneedles into pills that push into the GI lining (intra-enteral injection) to bypass these challenges has the capacity to improve bioavailability of a biologically active macromolecule (199). An illustration of the working mechanism of such microneedle pill is represented in **Figure 9**. Recent studies suggest that macromolecule drug delivery may be possible via ingestible self-orienting millimeter-scale applicator (SOMA) capsules, which contain a tiny needle to autonomously position the capsule to engage the GI lining of the stomach in a safe manner to increase retention and enable the escape of insulin cargo from the GI

into the bloodstream (196) (**Figure 10**). The use of these methods is an alternative to injectable delivery of biologics medications, such as insulin and peptides, and enhances patient convenience, as well as increases safety and efficacy.

Microneedle capsules can deliver not only small molecules but also proteins, peptides, vaccines, hormones, and other macromolecules. Traverso *et al*. successfully deployed a microneedle-containing device, modeled after current FDA-approved ingestible devices (200), and monitored the administration of insulin in the stomach (198).

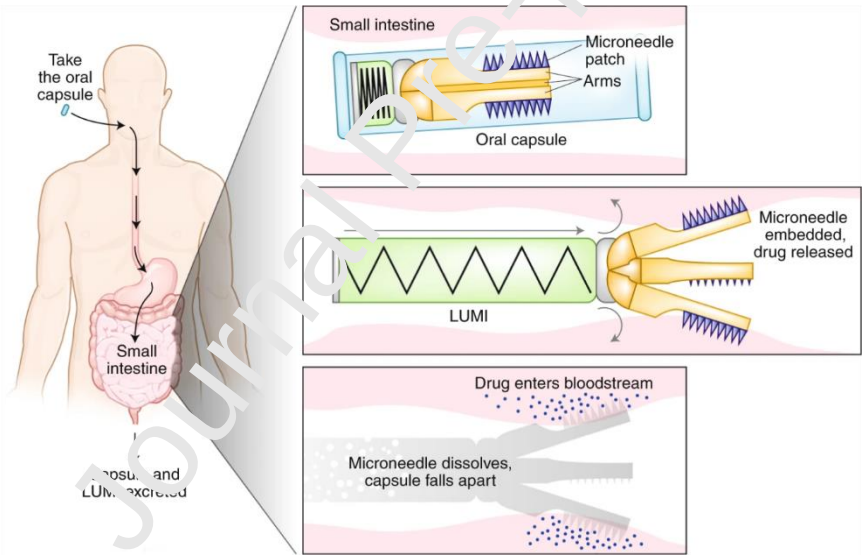

Figure 9. A microneedle approach for the delivery of biologics via oral administration. Delivery of biologics via the GI using a luminal unfolding microneedle injector (LUMI). Reprinted by permission from (199). Nature, Copyright (2019).

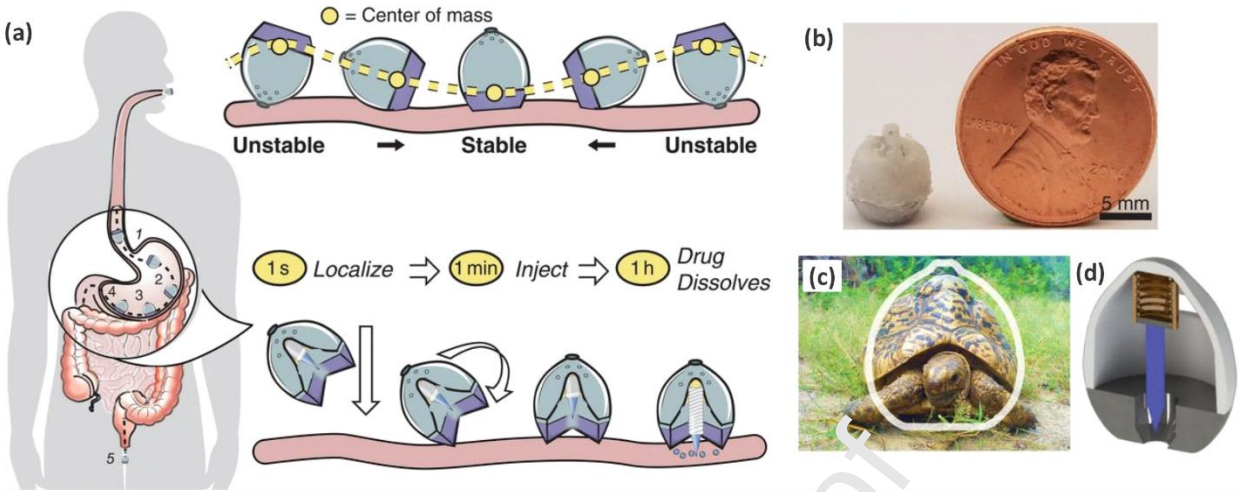

Figure 10. (a) A schematic illustrating SOMA capsules for oral drug delivery. SOMA capsules reach a stable point of orientation and deliver biologics through GI lining and into systemic circulation, (b) scale of fabricated SOMA, (c) the shape of SOMA capsules were inspired by the leopard tortoise shell, (d) mechanism of drug release after needle injection to the mucus through the spring ejection in caramelized sucrose. Reprinted from (201). Reprinted with permission from AAAS.

Recently, some novel self-propelled microdevices, known as microrockets or micromotors, have been developed for delivery of drugs to target locations of organs. Microrockets should be biocompatible and be degraded in the gastric acid. Zhou *et al*. developed a self-propelled microdevice for target-based delivery of doxorubicin (202). The microdevice consisted of a core of Zinc (Zn) surrounded by a thin layer of Fe and poly(aspartic acid) (PASP) microtube layer. Doxorubicin was adsorbed on the PASP surface by electrostatic interaction. The Zn particles electrodeposited into the PASP/Fe layers could propel the microdevice by creating hydrogen bubbles in an acidic environment and prevent the oxidation of Fe. The microdevice was

magnetically controlled due to presence of Fe layer and navigated to the target site of the stomach by locating a strong magnet near it. The microdevice was trapped into gastric layer and started releasing the drug in an acidic environment. In another work, Ávila *et al*. proposed a magnesium (Mg)-based micromotor for active delivery of antibiotics for treatment of gastric bacterial infections (203). The *in vivo* treatment of *H. pylori* infection using Mg-based micromotor loaded with clarithromycin was demonstrated in a mouse model. The micromotor consisted of a core of Mg microparticles coated with a thin layer of titanium oxide (TiO_2) and then with a film of PLGA loaded with clarithromycin, while a small opening for the contact with acid was left. Then, the micromotor was coated with a thin layer of chitosan to enable electrostatic adhesion of the micromotor to the mucus of the stomach. In the acidic environment of stomach, Mg core reacted with acid and Mg gradually dissolved, while created hydrogen gas that led to self-propulsion of the micromotor in the gastric fluid and delivery of the drug. The *in vivo* studies illustrated high efficiency in delivery of antibiotics when using Mg-based micromotors compared to the passive drug delivery approaches.

6.2. Intestines and Colon Delivery

Enhancing intestinal drug delivery is primarily driven through the increased adhesion of materials to the intestinal lining and mucosal barriers, both to facilitate local release of drugs to intestinal drug targets, and to increase the release of drugs into the bloodstream for systemic circulation. Thus, the majority of oral drug delivery research is focused on delivery to this region of the GI tract. We will discuss significant examples of successful intestinal delivery of

therapeutics, first discussing tissue-targeting strategies to achieve controlled drug release, and then discussing approaches focused at the cellular level of targeted intestinal drug delivery.

One of the most widely used drugs in oral drug delivery studies is insulin, with the goal of increasing insulin bioavailability via oral administration so that regular insulin injections are no longer required. One of the key goals with biologic delivery in the intestine is to protect the drug cargo from the harsh environments of the stomach prior to improving bioadhesion and controlled release of insulin within the intestines. Recently, Nemeth *et al*. reported on the loading of Eudragit® microwell devices via picoliter inkjet 3D printing for oral drug delivery applications (204). This method provides a high throughput and reproducible means of loading biologics such as insulin into flat devices to increase tissue retention and improve oral drug delivery. In another study, Fox *et al*. employed a multi-step lithography process to develop a microdevice including a drug reservoir sealed by a nanostraw membrane (205). The proposed device could facilitate drug loading and enable tunable release by manipulating the nanostraw inner diameter and density. The proposed microdevice could adhere to the GI tissue due to the presence of nanostructural topology (i.e., nanostraws) on the surface of the device. Thus, the drugs could be locally released over an adjustable time period, while the drugs were not exposed to drug-degrading biomolecules and digestive enzymes. The proposed microdevice can improve the oral absorption, which is of great importance for drugs with poor bioavailability such as insulin.

To increase the release of biomolecules from the intestine into the bloodstream, Abramson *et al*. developed a capsule named LUMI for the oral administration of various biomacromolecule drugs, using insulin as the potential target drug (206). The device consisted of 3 degradable arms with drug-loaded microneedles packed into a capsule that was 9 mm in diameter and 30 mm in

length. The microneedles were generated at the end of each arm and were composed of PVP with insulin powder at the tip. After taking the capsule, the poly(methacrylic acid-co-ethyl acrylate) coating was dissolved at pH levels above 5.5 and then the PEG coating embracing the compressed spring actuator was dissolved and the device was pushed out of capsule when the arms were in a random direction to the intestinal wall. Then, the drug was delivered, and the rest of device was dissolved. The actuation time of the capsule could be tuned by varying the molecular weight of the PEG in an environment with the proper pH condition. After actuation, the capsule broke apart and was transported through GI tract. The LUMI device showed consistent release of drugs into the small intestinal mucosa during the *in vivo* studies. However, one limitation of this devices is that the LUMI delivery method may cause discomfort for patients due to scratching the intestine after activation of the device.

In another work, oral administration of insulin-loaded liposomes, containing three kinds of bile salts including sodium glycocholate, sodium taurocholate, and sodium deoxycholate, resulted in enhanced bioavailability and an increase in blood insulin for 20 h (207),(208),(208). Liposomes containing ergosterol as the stabilizer, instead of cholesterol, have also shown a significant improvement in stability (209).

Recently, Lamson *et al*. reported on the development of anionic silica nanoparticles for the enhanced oral delivery of insulin (210). The researchers found that both the size and charge of the silica nanoparticles influenced not only their interactions with mucus barriers, but also the permeabilization of epithelial barriers for enhanced insulin release into the bloodstream. Interestingly, 50 nm silica nanoparticles in juxtaposition to 20 nm or 100 nm particles, provided the optimal balance between increased mucus penetration and integrin-mediated TJ relaxation.

Thus, 50 nm silica nanoparticles loaded with insulin displayed enhanced insulin bioavailability over what would normally be possible with oral drug delivery. This study lends support to other research that demonstrated that nanostructural cues can influence TJ remodeling (29, 205), and provides an example of the combined strengths of chemical and physical engineering of nanoscale drug carriers.

In addition to adjusting the physicochemical properties of drug carriers, the incorporation of stimuli-responsive materials in the reservoir-based drug delivery devices can advance the control over drug release. Using these materials, devices can be designed to be triggered and release the drug only under conditions simulating the GI tract. Nielsen *et al*. developed a platform of microwells for pH-triggered release of drugs (211). The microwells were fabricated by hot embossing PLA. The microwells were then filled with amorphous sodium salt of furosemide (ASSF) powder via an improved screen-printing process, as a proof of principle for oral drug delivery. The release of ASSF occurred at pH 6.5 (intestinal pH). The proposed microwells can be used for to protect the drug and prevent release before entering the intestine. Malachowski *et al*. developed a stimuli-responsive multi-fingered device that enabled site-specific delivery of various types of drugs by actively gripping the tissue at body temperature. It was proposed that gripping action increases the efficiency of drug delivery specially under flow conditions as this happens in the GI tract. The stimuli-responsive gripper was composed of rigid poly(propylene fumarate) and thermo-responsive poly(N-isopropylacrylamide-co-acrylic acid) hinges fabricated by photolithographic patterning (212). The grippers had a porous structure that allowed loading of the device with commercially available drugs. When the device entered the body (above 32°C), it gripped the tissue and delivered the loaded drug over time (up to seven days for mesalamine and doxorubicin). Both *in vitro* and *in vivo* studies confirmed the improved delivery

efficiency of doxorubicin.

In a different approach, scientists were inspired by transdermal patches to generate intestinal patches (152). Intestinal patches are typically millimeters in size and have a mucoadhesive drug reservoir layer, a pH-sensitive layer, and a backing layer. Also, intestinal patches can be composed of water-insoluble polymers, pH-sensitive polymers, colors, fragrances, absorption enhancers, buffer substances, and preservatives (150). While transdermal patches are designed for drug release of up to one week, intestinal patches are expected to release drugs over the course of hours (213). Intestinal patches were invented to increase drug bioavailability, reduce the drug disruption by the GI tract, and prevent painful drug injection, such as anticancer drugs for cancer chemotherapy or insulin for diabetes (214).

Illustrations of various intestinal micropatch structures are shown in **Figure 11**. The two-layer patches comprised of a waterproof backing layer and a drug-laden mucoadhesive layer. The mucoadhesive layer provides strong adhesion to the intestinal mucosa. Cui *et al*. developed a novel two-layered micropatch drug delivery system for oral delivery of proteins. The micropatch consisted of bilaminated films, a hydrophobic ethylcellulose layer, a mucoadhesive chitosan-ethylenediaminetetraacetic acid hydrogel layer, and also carboxylated chitosan-grafted nanoparticles (215). The adhesion of micropatches to intestinal mucosa is driven by the attachment of cationic polymers to negative residues on mucin, or through polymer chain entanglement and hydrogen bonding. Banerjee *et al*. developed mucoadhesive intestinal patches that combine intestinal devices with dimethyl palmitoyl ammonia propane sulfonate as a permeation enhancer for oral delivery of insulin. The patches were delivered from a capsule coated with a pH-responsive coating. The patches adhered to intestinal mucosa, released cargo

unidirectionally, and prevented enzymatic degradation in the gut (216). As shown in **Figure 11**, a typical 3-layer patch consists of a pH-responsive layer, a backing layer, and a drug-laden mucoadhesive layer. Four-layer patches typically have individual mucoadhesive and drug layers. Grabovac *et al*. made a three-layered oral delivery system for insulin delivery composed of thiolated polycarbophil as a polymeric matrix layer, a water-insoluble backing layer, and an enteric coating (217). Eaimtrakarn *et al*. developed a four-layer patch consists of a backing layer of ethyl cellulose to protect protein drug from enzymatic hydrolysis, a pH-sensitive surface layer, a drug-carrying middle layer, and a mucoadhesive layer. They tested three different pH-sensitive layers to achieve dissolution in different sections of the small intestine (218).

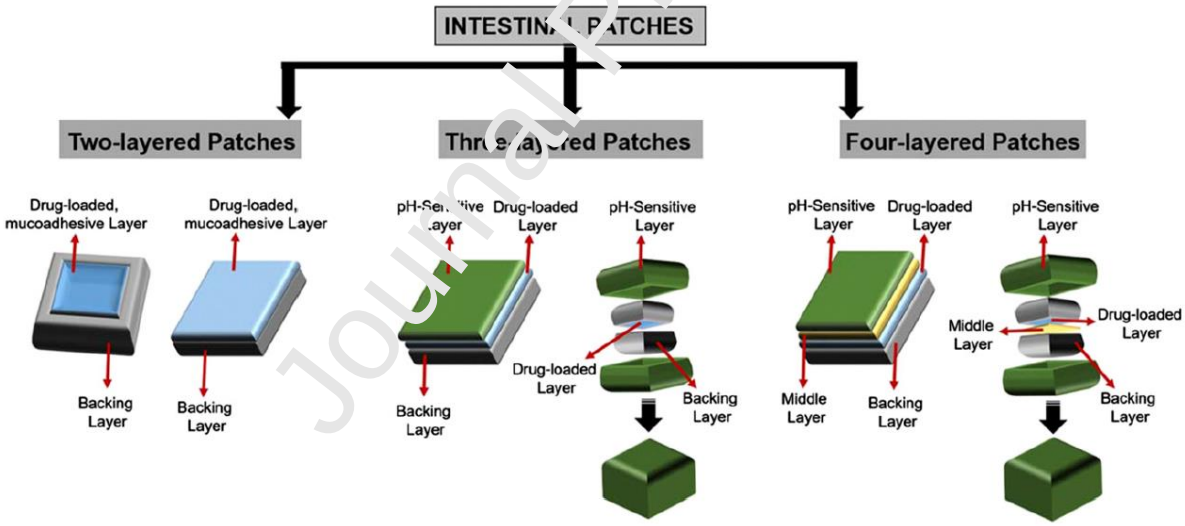

Figure 11. Examples of different intestinal patch structures including two-layered, three-layered, and four-layered patches. These patches deliver drugs with additional supportive layers. Reprinted from (219), Copyright (2015), with permission from Elsevier.

Colon-targeting approaches have received increased attention in the field of oral drug delivery, as the longer retention time within the large intestine and colon provide opportunities for sustained drug release and drug escape into the bloodstream. Varshosaz *et al*. used dextran to prepare a natural polymer-based microparticle system for targeted delivery to the colon of budesonide for ulcerative colitis treatment. Microcapsules were prepared with different drug-to-dextran ratios and three molecular weights of the polymer. Their results showed that budesonide encapsulated microcapsules could target the colon, increasing the specificity of drug delivery, resulting in higher reduction of macroscopic damage and efficacy than mesalamine suspension (220).

In addition to microparticle systems, electrospun medicated shellac nanofibers have received interest for the fabrication of a colon-targeting delivery capsule to improve the bioavailability of poorly water soluble drugs and provide sustained release in the colon (**Figure 12**) (221),(222). Wang *et al*. reported a simple method for the fabrication of capsule shells from a coaxial electrospinning method with a core fluid of shellac and ferulic acid (FA) and N,N-dimethylformamide as the shell (222). Due to the insolubility of shellac at low pH, a small percentage of drug was released into solution at pH 2, mimicking the stomach pH, but sustained release was observed after 2 h incubation and transitioning to a neutral dissolution medium (**Figure 12b**) Recently, there has been interest in the encapsulation of nanoparticles, such as micelles, liposomes, vesicles, and nanoparticles, in shellac nanofibers. In one study, Henning *et al*. developed liquid-filled shellac capsules to obtain colonic release of pectinate (223). They showed that an external shellac layer significantly protected the pectinate from enzymes in the GI tract and extended the material retention time to several hours. Colon-targeted drug delivery has recently gained significant interest for bioactive proteins (224),(197). Ravi *et al*. developed a colon-targeted delivery system using inulin as an inner coating, followed by shellac as outer

coating with diltiazem hydrochloride as a model drug. The release study showed that polymer-drug shellac tablets were insoluble in the stomach and intestinal environment and released the maximum amount of drug in the colonic environment to increase drug release into the bloodstream (221). Wen *et al*. developed a core/shell structured nanofilm, using alginate as the outer shell layer and chitosan nanoparticles as the inner core layer for delivery of a model protein, bovine serum albumin (224). In general, colon targeting fibrous materials could be useful for specific protein targeting to the colon, with the aim of minimizing side effects and improving the local efficacy of proteins.

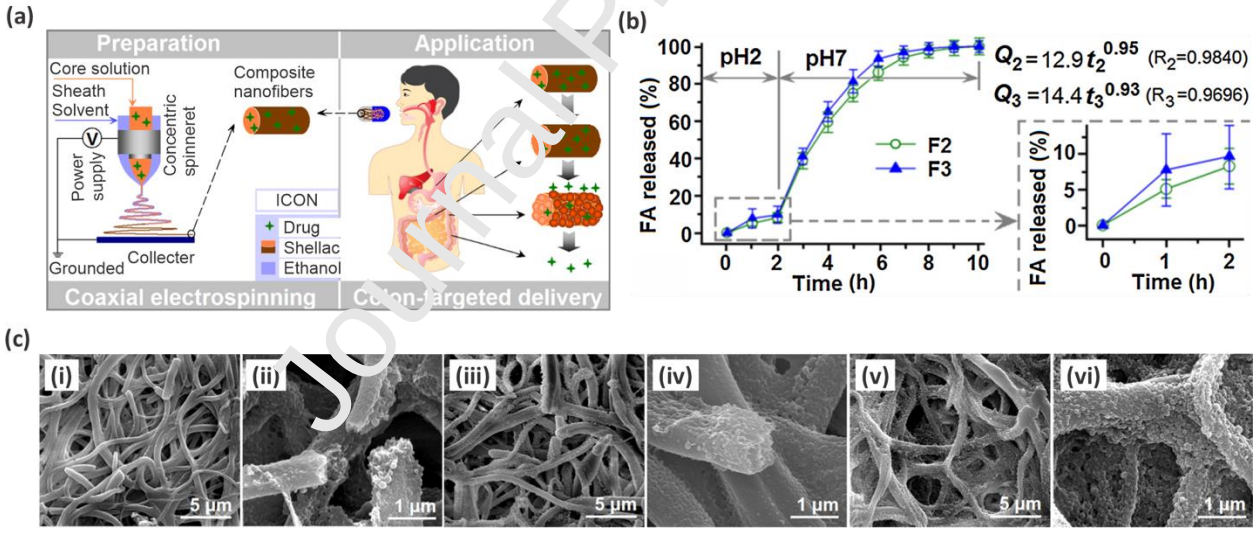

Figure 12. Preparation and characterization of shellac nanofibers and their applications in oral drug delivery. (a) A schematic illustrating the design strategy of medicated shellac nanofibers and the results of in vitro dissolution tests. (b) The FA release profiles and (c) SEM images (i, ii)

just after dissolution, (iii, iv) 3h after dissolution, (v, vi) 7h after dissolution. Reprinted from (222), Copyright (2015), with permission from Elsevier.

In addition to controlling the drug carrier tissue-targeting properties, cellular interactions must also be controlled to improve bioadhesion and drug delivery efficacies. To target intestinal cells and improve cellular bioadhesion, chemical functionalization of materials is often performed. Antibodies and peptides specific for intestinal cells have recently been utilized for functionalization of drug carriers not only to increase adhesion, but also for target based drug delivery to specific types of intestinal cells (23, 103). In one study, nanoparticles were grafted with trastuzumab (Herceptin, human epidermal growth factor receptor-2 antibody). The antibody-coated nanoparticles showed an increase in uptake by Caco-2 (225). Another strategy is to use adhesion promoters, such as tethered or linear polymer chains to stimulate bioadhesion in drug delivery (226, 227). In another example, Ainslie *et al.* designed a microdevice with di-methacrylate hydrogel as a backbone to which avidin was covalently conjugated. Subsequently, biotinylated tomato lectin was added to increase the bioadhesion of the microdevice, as biotin has a strong binding affinity to avidin. *In vitro* testing results demonstrated that the Caco-2 epithelial colorectal cells were more attached to microdevices functionalized with lectin than non-functionalized devices (228). Microdevices were conjugated with two lectins: (1) tomato, which is capable of attachment to Caco-2 cells, stable in low pH environments, and selective to small intestine epithelium; and (2) peanut, which has non-specific lectin sites. The lectin conjugated microdevices showed a 2-4 fold higher degree of binding when compared to control microdevices. The tomato lectin conjugate showed significant difference of 2 folds over peanut

lectin (229, 230). Similar strategies were applied for microspheres and micropatch systems to enhance the adhesion of the material.

To deliver drugs into systemic circulation from the intestine, epithelial barriers and TJs must be overcome. Drug carriers have been shown to rearrange of TJs and influence protein expression to enhance drug permeation across epithelial barriers. This is primarily achieved through the installation of nanotopography on microparticle carriers or on thin films. Uskokovic *et al*. demonstrated that nanowires protruding from the surface of silica microparticles facilitates the rearrangement of ZO-1 TJ proteins in Caco-2 monolayer models (231). Subsequent work improved on this design through the fabrication of planar nanowire-coated surfaces for enhanced tissue contact and TJ rearrangement (**Figure 13a**) (232). Nanostructured thin films have also been observed to reversibly loosen epithelial barriers, allowing for increased permeation of antibodies across TJs (**Figure 13b**) (233). In addition to influencing TJ behavior, recent work by Levy *et al*. demonstrated that PEG microdevices can inhibit P-gp efflux pump expression on Caco-2 monolayers, which may serve as a means of enhancing intestinal drug adsorption for oral drug delivery applications (234). Although this study did not use nanostructured materials, it provides further evidence that physical parameters of nano and microtechnologies can influence cellular behavior to improve oral drug delivery applications. Taken together, these studies demonstrate that the physical engineering of nano and microscale materials can serve as important methods for enhancing drug penetration across epithelial barriers to improve systemic drug delivery via oral administration.

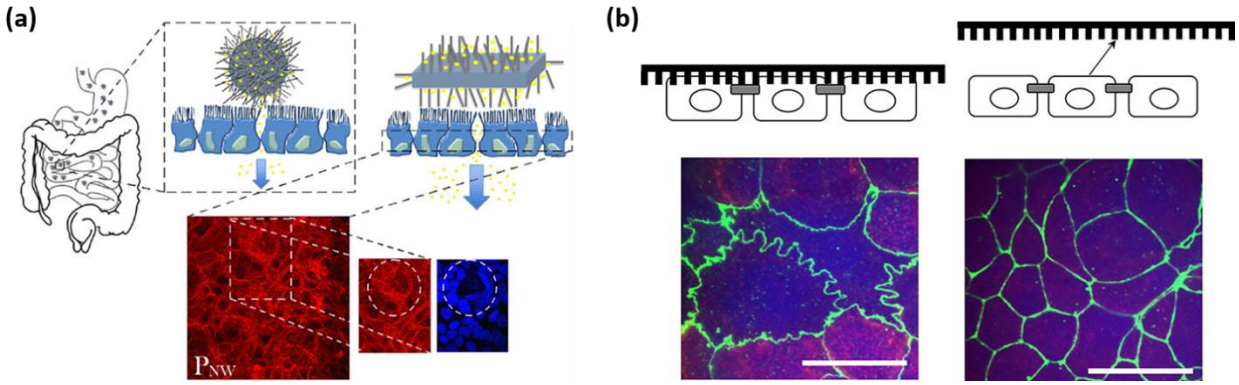

Figure 13. Physical approaches to modulate TJs for oral drug delivery. (a) Nanowire-coated silica microparticles and planar microdevices Reprinted with permission from (232). Copyright (2012) American Chemical Society (b) nanostructured thin films initiate ZO-1 TJ rearrangement to enhance drug penetration through epithelial barriers. Reprinted with permission from (233). Copyright (2013) American Chemical Society.

7. MICROFABRICATED *IN VITRO* MODELS

In vitro modelling of the GI system is significantly helpful to investigate the performance of various oral drug delivery systems in GI tissues. In particular, *in vitro* GI tract models allow for the studying of drug permeation across biological barriers, as well as studying complex host-microbe interactions, while decreasing the costs and ethical issues involved with preclinical animal studies (235). The physiological environment of the GI tract, including its functions and dynamic conditions, makes it a challenging organ to be modeled *in vitro*. Several *in vitro* models, such as organoids, trans-well co-culture, and macroscale bioreactors have been developed to study the GI tract and are reviewed in previous literature (236). Here, we review systems that use microscale or nanoscale approaches to recapitulate the topography, motility, and flow present

of the GI tract in microfabricated *in vitro* models and organ-on-a-chip platforms.

3D spheroid models better recapitulate *in vivo* biological conditions and have seen immense use as both basic science and drug discovery tools (237). Micro and nanoscale technologies offer opportunities to improve on standard spheroid models and better mimic *in vivo* physiology. Several examples have been reported on the use of microparticles to direct cellular assembly and architecture in spheroid models (238-241). In a recent example, Samy *et al*. demonstrated that the growth of Caco-2 cells around a Matrigel microparticle initiates faster TJ formation when compared to the standard transwell model system (242). This system also induces the proper polarization of apical and basal membranes, with actin and ZO-1 TJs displayed on the outer membrane of the spheroid, allowing easier drug access to the spheroid TJs in order to model intestinal drug transport *in vitro*.

Organ-on-a-chip platforms often provide a chamber to colonize cells in a specific arrangement that resembles the architecture of designed tissues or organs and mimics the physiological function of tissues or organs in a microfluidic system (243, 244). Kimura *et al*. developed a micro pumping system on a-chip to model the gut epithelial using Caco-2 cells (**Figure 14**). The device was used to investigate the perfusion and transportation of fluorescent compounds for applications in drug toxicity (245). In further advancement of GI modeling on a chip, Imura *et al*. made a microchip-based system using Caco-2 cells to test drug absorption (246). They also developed two organs on-a-chip system using Caco-2 cells for intestine and HepG2 (liver hepatocellular) cells for liver. The intestinal absorption and hepatic metabolism were tested to demonstrate the feasibility of their device for *in vitro* assays (247). In another work, Bricks *et al*. used a microfluidic platform to study the interaction between Caco-2 and HepG2. Their study

showed that such coculture is important for proper metabolism and transformation of a tested compound (248). A similar device was also used to study host-microbe molecular interactions in the gut. The intestinal epithelial barrier plays an important role in protecting our body from foreign organisms. To model this barrier, Tan *et.al*. used a commercial chopstick-style electrode to record the trans epithelial electrical resistance and quantify the integrity of cellular lining in their system. The trans epithelial electrical resistance was also used to measure the permeability across the intestinal barrier (249). In a more recent study, a modular GI tract-liver system was developed, using primary human intestinal epithelial cells and 3D liver micro-lobe like constructs (250). The authors showed that the primary intestinal cells formed a monolayer and exhibited comparable cellular integrity to native intestine.

The inclusion of dynamic conditions and fabrication of structures mimicking the intestinal architecture (e.g., crypts or villi) are important components in order to fully model the GI tract. To tackle this challenge, advanced models to recapitulate the microenvironment of the epithelium were recently developed. For instance, Costello *et al*. made *in vitro* small intestine tissue using polymeric scaffolds that mimic the 3D tissue architecture. The integrity data derived from trans epithelial electrical resistance showed that the inclusion of dynamic flow improved the integrity of the cell barrier as compared to static condition (251). Shim *et al*. fabricated a collagen scaffold that mimics the intestinal villi to study the epithelium's permeability (252). This study showed that the inclusion of flow in the microfluidic tract, in addition to the incorporation of 3D structure, further increased the relevance of their model to the intestinal villi.

To further improve the biomimicry of *in vitro* models of GI, inclusion of gut motility is another mechanical cue that needs to be considered in the device design. To tackle this, a variety of cell

stretchers were used to apply stain to a cell monolayer in different directions (uniaxial, biaxial, and radial stretching). Stretching is an important modulator of cell physiology in the GI tract. In particular, the epithelium in the intestine is stimulated by repetitive deformation caused by peristaltic movement and repetitive shortening of the villi. To investigate the effect of mechanical stimulation, Basson *et al*. cultured Caco-2 cells on an elastic membrane and subjected the cells to 10% strain. They showed that the cells proliferated faster and expressed brush border enzymes (253). In a recent work, Caco-2 cell junctions were disrupted to analyze the impact of cyclic stretch on the TJ in cells. The results showed the increased paracellular permeability and re-organization of the junction's proteins after the disruption (254).

In one example of highly sophisticated human intestine models, known as "human gut-on-a-chip", a microfluidic device was used flow to create both shear stress and cell stretching on a stretchable porous silicone membrane coated with extracellular matrix (ECM) proteins (**Figure 14a,b**) (255). The flow rate was adjusted to 30 µl/h to produce a shear stress of 0.02 dyne/cm^2 to mimic the flow rate of the intestine. They also applied cyclic uniaxial strain (10%; 0.15 Hz) that mimicked the physiological peristaltic motions and facilitated the fast polarization of the epithelium (**Figure 14c**). The cell monolayer developed with high integrity. This system was employed to co-culture intestinal epithelium cells with commensal microbes (256). After one-week, endoxins and immune cells stimulated the epithelial cells to generate proinflammatory cytokines. The inflammatory cascade was induced by the villi injury and compromised the intestinal barrier function. The latter study showed that gut-on-a-chip is suitable to study the interaction between the microbiome and intestine in a pathophysiological environment. In a recent work, Kasendra *et al*. made a small intestine-on-a-chip system for culturing human intestinal vascular endothelium and primary epithelial cells (257). The device also provided both

uniaxial cyclic deformation and flow to the cells and showed the formation of villi-like structures (**Figure 15a**) and functional gut epithelium and endothelium layers with TJs ((**Figure 15b**).). This system also provided increased transcriptional similarities to the human duodenum.

Taken together, the abovementioned studies are the most advanced micro-physiological models of the GI tract to date (258) Although the inclusion of a 3D and mature tissue composed of ECM, muscle cells, vasculature, and capillaries is missing from those models and would help to create a more biomimetic model of human GI tract. However, such simplified microphysiological models of GI tissues can be used to tackle challenges in oral drug delivery and improve our knowledge about the GI action in diverse microenvironments.

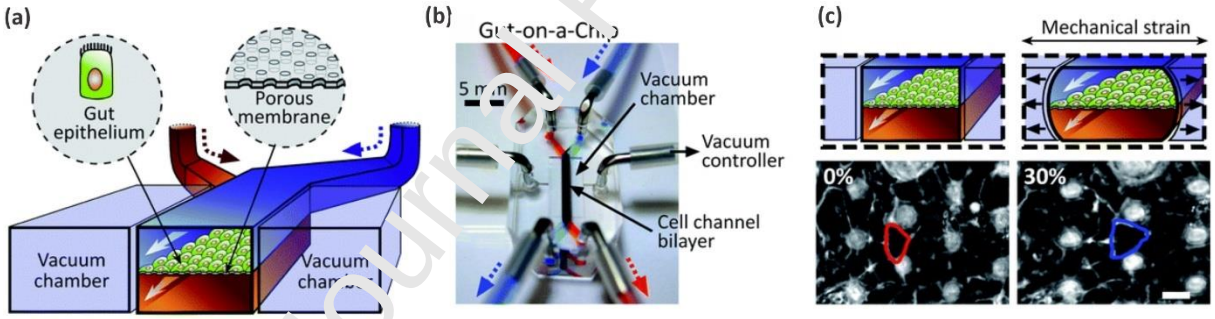

Figure 14. The human gut-on-a-chip. (a) Schematic of the gut-on-a-chip device showing the porous ECM-coated membrane covered with gut epithelial cells and side vacuum chambers to apply mechanical strain on membrane mimicking the role of peristaltic motion. Top channel (blue) represents the gut lumen and the bottom channel (red) represents the capillary bed underlaying the epithelial cells. (b) An actual image of the gut-on-a-chip device made of PDMS elastomer. Arrows show the flow direction and red and blue dyes in tubing correspond to the lower and upper microchannels, respectively, for channel visualization. (c) Schematics (of

intestinal monolayers cultured on the gut-on-a-chip porous membrane in the presence (right) or absence (left) of 30% mechanical strain applied by vacuum chambers and corresponding micrographs of epithelial cells on the porous membrane. Scale bar is 50 µm. Reproduced with permission. (255) Copyright 2012, Royal Society of Chemistry

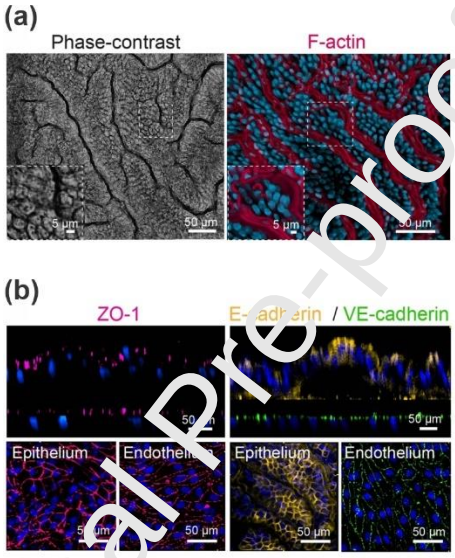

Figure 15. Morphological and microscopical characterization of the primary human intestine on-a-chip. (a) Microscopic images of the intestinal epithelium grown on-a-chip after 12 days under cyclic strain and fluid flow showing the formation of epithelial villi-like protrusions. The images are stained f F-actin (magenta, brush border) and for nuclei (DAPI, blue). (b) Immunofluorescence images showing the intact TJs in the intestinal epithelium and underlying endothelium immunostained with ZO-1 (magenta), E-cadherin for epithelial cells (yellow), VE-cadherin for endothelial cells (green), and nuclei (DAPI, blue). Scale bars are 50 µm (257).

8. BRINGING MICRO AND NANOSCALE TECHNOLOGIES INTO THE CLINIC

It is crucial to push highly advanced drug delivery systems and devices towards clinical applications. Nanocarriers were first used in clinical trials in the 1960s to deliver small molecules that have poor pharmacokinetic profile, low solubility, and high off-target toxicity (259). Among the developed particles for oral drug delivery, some polymer-based particles have been approved in clinical studies, and several liposomal formulations are under clinical investigations (260). Additionally, other nanoparticle carriers, such as silica nanoparticles and calcium phosphate nanoparticles have been used in oral drug delivery and have begun to see translation into clinical trials (261). These nanocarriers have been primarily used to deliver insulin, as well as small lipophilic peptides (~1-2 kDa molecular weight) with a cyclic structure, which are resistive to peptidase degradation

Oral administration is a pillar of pharmaceutical industry. Currently 62% of FDA-approved drugs are orally administered drugs (262), thanks to ease of administration and high patient acceptance. While small hydrophobic molecules have been successful in oral administration, hydrophilic small molecules or peptides and proteins are yet to find their way to the clinical translation. Indeed, there is an unmet need for the delivery of proteins and peptides for which oral bioavailability is as low as <2% with current technologies (22). Micro and nanotechnology have been showing to be promising to overcome these barriers. Although many microfabricated drug delivery systems are close to clinical trials, only a few formulations have seen translation into the clinic and numerous challenges remain for the future oral practice (263).

The oral delivery of biologicals could potentially improve the life of millions of patients and huge efforts are being put to push such solutions to the clinic (261). A self-emulsifying system

for oral delivery is marketed for delivering immunosuppressing agent cyclosporine A (Sansimmune, CH). Similarly, self-emulsifying oral testosterone received FDA approval in 2019 (Jatenzo) (264). Liposomal insulin formulation, which delivers drug to the liver, known as Hepatocyte-directed vesicular (HDV) insulin, showed promising results in phase 1 and 2 clinical trials (265). The oral nanoformuation of this liposomal HDV insulin, composed of HDV conjugated insulin and a biotin-phosphatidylanolamine hepatic tag, is available now and is under phase 2, and 3 of clinical trials (266). Liver-targeting liposomes for insulin delivery reached Phase III (NCT00814294, Silica nanoparticles also delivering insulin orally are currently in Phase II (NCT01973920, Oshadi Drug Administration, IL). Nanoparticles with a calcium phosphate core and pegylated salts of fatty acids, coated with carbomer and cellulose acetate phthalate for the delivery of insulin reached Phase I (ChiCTR-TRC-12 001 872, NOD/NodlinTM, CN). Nanomega Corp is encapsulating insulin in chitosan shelled gamma poly(g-glutamic acid)-based nanoparticles. These are key steppingstones that have to be leveraged for progressing micro and nanofabricated technologies for oral delivery of many therapeutic proteins. This knowledge can also be used for the delivery of hydrophilic small molecules. The challenges currently faced by micro and nanofabricated technologies are presented below.

Poor bioavailability has been the major bottleneck of translation of micro and nano technologies for oral delivery. Some practical and simple approaches have been proposed to tackle this problem. For example, nanoparticles can be coated with hydrophilic polymers, such as chitosan and PEG, and TPGS (a PEG-conjugated vitamin E) or can be synthesized with these polymers to further incorporate hydrophilic elements in the polymeric nanoparticles (267). As a result, the permeability, solubility, stability, and thereby oral bioavailability of nanoparticles are enhanced.

Another important challenge for orally administered formulations is that nano- and microfabrication come at a cost that must be justified to make a product commercially cost-effective. This is achievable with high-throughput emulsion or self-assembly fabrication methods. However, more advanced microfabricated systems (3D printed or lithography enabled casting), have typically a complex engineering design and low-throughput manufacturing processes that result in high cost of fabrication (29),(268),(269),(170). For example, the current workflow for 3D printed drug delivery systems is multi-step (3D printing, drug loading, sealing, substrate release) and involves expensive equipment (3D printer, inkjet printer, spray coater) (270). Recently, rapid and large-scale 3D printing was achieved using a mobile liquid interface that minimize heat buildup (271). The evolution of such technology to print micro-scale elements in high-throughput could enable the progress of application of oral deliveries.

Expensive biologicals are frequently encapsulated in micro and nano-fabricated systems. Interestingly, a daily capsule semaglutide requires a 100 times higher dose and more frequent administration, to achieve a similar efficacy compared to its weekly injection counterpart for the treatment of type 2 diabetes. A study was conducted to confirm that the additional costs incurred to produce extra semaglutide for the oral formulation would still generate a cost-effective oral formulation, given the greater quality of life experienced by the patients receiving the daily capsules (272). This study testifies the need to carefully assess the dosing frequency against the efficacy when a molecule is selected for encapsulation in micro and nano-fabricated systems. This work also teaches the importance of direct comparison of oral delivery to other routes of administration.

Of particular interest is the applications of micro- and nanoscale technologies for delivery of

therapies in preclinical and clinical trials for coronavirus disease 2019 (COVID-19). The outbreak of COVID-19 is considered as one of the deadliest diseases that has caused the death of approximately 550,000 people worldwide so far. Antiviral drugs, such as remdesivir, chloroquine, and hydroxychloroquine have been proposed as promising drug candidates for treating coronavirus disease (273-275). These antiviral drugs have been administered orally to inhibit virus infection with mild to moderate doses daily for a long time, which causes the adverse effects in patients. In order to overcome such issues, we suggest employing micro and nanotechnology-based drug delivery approaches to increase the efficacy of these drugs to prevent coronavirus infection.

Improving the standardization of preclinical parameters and procedures, including biodistribution, toxicity, protein adsorption, and device removal (27, 269) will enable faster translation of technologies into the clinic. Despite these challenges, the opportunities for improving oral drug delivery using micro and nanoscale technologies is immense, and therefore the field should continue the push for translation into large animal models and eventual clinical trials.

9. CONCLUSION

During the past decade, researchers have had great leverage to create nano and microdevices that enable oral administration of different biomolecules. Indeed, the clinical translation of nanotechnology for drug delivery through the intravenous route, gave us a deep understanding of such systems. In parallel, the great progress of microfabrication methods was leveraged to serve

the oral administration field. This progress was only recently reflected to marketed products. To unlock the full potential of such technologies, researchers should focus on inventing methods that would ensure robust manufacturing scale-up and solid proof of efficacy. It is crucial to push highly advanced drug delivery systems and devices towards clinical applications as they could be leading to an evolution of pharmaceutical industry towards more patient-friendly oral administration.

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References

1. Sant S, Tao SL, Fisher OZ, Xu Q, Peppas NA, Khademhosseini A. Microfabrication technologies for oral drug delivery. Adv Drug Deliv Rev. 2012;64(6):496-507. Epub 2011/12/15. doi: 10.1016/j.addr.2011.11.013. PubMed PMID: 22166590; PMCID: PMC3534972.
2. Sastry SV, Nyshadham JR, Fix JA. Recent technological advances in oral drug delivery–a review. Pharmaceutical science & technology today. 2000;3(4):138-45.
3. Jiang D, England CG, Cai W. DNA nanomaterials for preclinical imaging and drug delivery. J Control Release. 2016;239:27-38. Epub 2016/08/17. doi: 10.1016/j.jconrel.2016.08.013. PubMed PMID: 27527555; PMCID: PMC5037045.
4. Maham A, Tang Z, Wu H, Wang J, Lin Y. Protein-based nanomedicine platforms for drug

delivery. Small. 2009;5(15):1706-21. Epub 2009/07/03. doi: 10.1002/smll.200801602. PubMed PMID: 19572330.
5. Koziolek M, Grimm M, Schneider F, Jedamzik P, Sager M, Kuhn JP, Siegmund W, Weitschies W. Navigating the human gastrointestinal tract for oral drug delivery: Uncharted waters and new frontiers. Adv Drug Deliv Rev. 2016;101:75-88. Epub 2016/04/03. doi: 10.1016/j.addr.2016.03.009. PubMed PMID: 27037063.
6. Petit J, Meurice N, Kaiser C, Maggiora G. Softening the rule of five—where to draw the line? Bioorganic & medicinal chemistry. 2012;20(18):5343-51.
7. Peyrot M, Rubin RR, Kruger DF, Travis LB. Correlates of insulin injection omission. Diabetes care. 2010;33(2):240-5.
8. Gedawy A, Martinez J, Al-Salami H, Dass CR. Oral insulin delivery: existing barriers and current counter-strategies. Journal of pharmacy and pharmacology. 2018;70(2):197-213.
9. Ahadian S, Civitarese R, Bannerman D, Mohammadi MH, Lu R, Wang E, Davenport-Huyer L, Lai B, Zhang B, Zhao Y. Organ-on-a-chip platforms: a convergence of advanced materials, cells, and microscale technologies. Advanced healthcare materials. 2018;7(2):1700506.
10. Ahadian S, Ramón-Azcón J, Estili M, Obregón R, Shiku H, Matsue T. Facile and rapid generation of 3D chemical gradients within hydrogels for high-throughput drug screening applications. Biosensors and Bioelectronics. 2014;59:166-73.
11. Bae H, Chu H, Edalat F, Cha JM, Sant S, Kashyap A, Ahari AF, Kwon CH, Nichol JW, Manoucheri S, Zamanian B, Wang Y, Khademhosseini A. Development of functional biomaterials with micro- and nanoscale technologies for tissue engineering and drug delivery applications. J Tissue Eng Regen Med. 2014;8(1):1-14. Epub 2012/06/20. doi: 10.1002/term.1494. PubMed PMID: 22711442; PMCID: PMC4199309.
12. Caldorera-Moore M, Peppas NA. Micro- and nanotechnologies for intelligent and responsive biomaterial-based medical systems. Adv Drug Deliv Rev. 2009;61(15):1391-401. Epub 2009/09/18. doi: 10.1016/j.addr.2009.09.002. PubMed PMID: 19758574; PMCID: PMC2788786.
13. Pan J, Chan SY, Lee WG, Kang L. Microfabricated particulate drug-delivery systems. Biotechnol J. 2011;6(12):1471-87. Epub 2011/11/15. doi: 10.1002/biot.201100237. PubMed PMID: 22076813.
14. Khademhosseini A, Langer R. Drug delivery and tissue engineering. Chem Eng Prog. 2006;102(2):38-42.
15. Park K. Controlled drug delivery systems: past forward and future back. J Control Release. 2014;190:3-8. Epub 2014/05/06. doi: 10.1016/j.jconrel.2014.03.054. PubMed PMID: 24794901; PMCID: PMC4142099.
16. Dahan A, Hoffman A. Rationalizing the selection of oral lipid based drug delivery systems by an in vitro dynamic lipolysis model for improved oral bioavailability of poorly water soluble drugs. J Control Release. 2008;129(1):1-10. Epub 2008/05/24. doi: 10.1016/j.jconrel.2008.03.021. PubMed PMID: 18499294.
17. Son G-H, Lee B-J, Cho C-W. Mechanisms of drug release from advanced drug formulations such as polymeric-based drug-delivery systems and lipid nanoparticles. Journal of Pharmaceutical Investigation. 2017;47(4):287-96.
18. Prausnitz MR, Elias PM, Franz TJ, Schmuth M, Tsai J-C, Menon GK, Holleran WM, Feingold KR. Skin barrier and transdermal drug delivery. J Dermatology. 2012;3:2065-73.
19. Groeber F, Holeiter M, Hampel M, Hinderer S, Schenke-Layland K. Skin tissue

engineering—in vivo and in vitro applications. Advanced drug delivery reviews. 2011;63(4-5):352-66.
20. Prausnitz MR, Elias PM, Franz TJ, Schmuth M, Tsai J-C, Menon GK, Holleran WM, Feingold KR. Skin barrier and transdermal drug delivery.
21. Li H, Limenitakis JP, Fuhrer T, Geuking MB, Lawson MA, Wyss M, Brugiroux S, Keller I, Macpherson JA, Rupp S. The outer mucus layer hosts a distinct intestinal microbial niche. Nature communications. 2015;6(1):1-13.
22. Homayun B, Lin X, Choi HJ. Challenges and Recent Progress in Oral Drug Delivery Systems for Biopharmaceuticals. Pharmaceutics. 2019;11(3):129. Epub 2019/03/22. doi: 10.3390/pharmaceutics11030129. PubMed PMID: 30893852; PMCID: PMC6471246.
23. Ensign LM, Cone R, Hanes J. Oral drug delivery with polymeric nanoparticles: the gastrointestinal mucus barriers. Adv Drug Deliv Rev. 2012;64(6):557-70. Epub 2012/01/04. doi: 10.1016/j.addr.2011.12.009. PubMed PMID: 22212900; PMCID: PMC3322271.
24. Renukuntla J, Vadlapudi AD, Patel A, Boddu SH, Mitra AK. Approaches for enhancing oral bioavailability of peptides and proteins. Int J Pharm. 2013;447(1-2):75-93. Epub 2013/02/23. doi: 10.1016/j.ijpharm.2013.02.030. PubMed PMID: 23428883; PMCID: PMC3680128.
25. Moreno Raja M, Lim PQ, Wong YS, Xiong GM, Zhang Y, Venkatraman S, Huang Y. Polymeric Nanomaterials. Nanocarriers for Drug Delivery. 2019. p. 557-653.
26. Brown TD, Whitehead KA, Mitragotri S. Materials for oral delivery of proteins and peptides. Nature Reviews Materials. 2019:1-22.
27. Durán-Lobato M, Niu Z, Alonso MJ. Oral delivery of biologics for precision medicine. Advanced Materials. 2019:1901935.
28. Han Y, Gao Z, Chen L, Kang L, Huang W, Jin M, Wang Q, Bae YH. Multifunctional oral delivery systems for enhanced bioavailability of therapeutic peptides/proteins. Acta Pharm Sin B. 2019;9(5):902-22. Epub 2019/10/28. doi: 10.1016/j.apsb.2019.01.004. PubMed PMID: 31649842; PMCID: PMC6804447
29. Fox CB, Kim J, Le LV, Nemeth CL, Chirra HD, Desai TA. Micro/nanofabricated platforms for oral drug delivery. J Control Release. 2015;219:431-44. Epub 2015/08/06. doi: 10.1016/j.jconrel.2015.07.033. PubMed PMID: 26244713; PMCID: PMC4656086.
30. LaVan DA, Lynn DM, Langer R. Moving smaller in drug discovery and delivery. Nat Rev Drug Discov. 2002;1(1):77-84. Epub 2002/07/18. doi: 10.1038/nrd707. PubMed PMID: 12119612.
31. Belliveau NM, Huft J, Lin PJ, Chen S, Leung AK, Leaver TJ, Wild AW, Lee JB, Taylor RJ, Tam YK, Hansen CL, Cullis PR. Microfluidic Synthesis of Highly Potent Limit-size Lipid Nanoparticles for In Vivo Delivery of siRNA. Mol Ther Nucleic Acids. 2012;1:e37. Epub 2013/01/25. doi: 10.1038/mtna.2012.28. PubMed PMID: 23344179; PMCID: PMC3442367.
32. Li XJ, Zhou Y. Microfluidic devices for biomedical applications: Elsevier; 2013.
33. Nguyen NT, Shaegh SA, Kashaninejad N, Phan DT. Design, fabrication and characterization of drug delivery systems based on lab-on-a-chip technology. Adv Drug Deliv Rev. 2013;65(11-12):1403-19. Epub 2013/06/04. doi: 10.1016/j.addr.2013.05.008. PubMed PMID: 23726943.
34. Kawaguchi H. Functional polymer microspheres. Progress in polymer science. 2000;25(8):1171-210.
35. Makadia HK, Siegel SJ. Poly lactic-co-glycolic acid (PLGA) as biodegradable controlled drug delivery carrier. Polymers. 2011;3(3):1377-97.

36. Yang Y, Fang Z, Chen X, Zhang W, Xie Y, Chen Y, Liu Z, Yuan W. An Overview of Pickering Emulsions: Solid-Particle Materials, Classification, Morphology, and Applications. Front Pharmacol. 2017;8:287. Epub 2017/06/08. doi: 10.3389/fphar.2017.00287. PubMed PMID: 28588490; PMCID: PMC5440583.
37. Varde NK, Pack DW. Microspheres for controlled release drug delivery. Expert Opin Biol Ther. 2004;4(1):35-51. Epub 2003/12/19. doi: 10.1517/14712598.4.1.35. PubMed PMID: 14680467.
38. McClements DJ. Advances in fabrication of emulsions with enhanced functionality using structural design principles. Current Opinion in Colloid & Interface Science. 2012;17(5):235-45.
39. Branco MC, Schneider JP. Self-assembling materials for therapeutic delivery. Acta Biomater. 2009;5(3):817-31. Epub 2008/11/18. doi: 10.1016/j.actbio.2008.09.018. PubMed PMID: 19010748; PMCID: PMC2729065.
40. Rösler A, Vandermeulen GWM, Klok H-A. Advanced drug delivery devices via self-assembly of amphiphilic block copolymers. Advanced drug delivery reviews. 2012;64:270-9.
41. Torchilin VP. Micellar nanocarriers: pharmaceutical perspectives. Pharm Res. 2007;24(1):1-16. Epub 2006/11/17. doi: 10.1007/s11095-006-9132-0. PubMed PMID: 17109211.
42. Sill TJ, von Recum HA. Electrospinning: Applications in drug delivery and tissue engineering. Biomaterials. 2008;29(13):1989-2006. doi: https://doi.org/10.1016/j.biomaterials.2008.01.011.
43. Ignatious F, Sun L, Lee C-P, Baldoni J. Electrospun Nanofibers in Oral Drug Delivery. Pharmaceutical Research. 2010;27(4):576-88. doi: 10.1007/s11095-010-0061-6.
44. Thakkar S, Misra M. Electrospun polymeric nanofibers: New horizons in drug delivery. European Journal of Pharmaceutical Sciences. 2017;107:148-67. doi: https://doi.org/10.1016/j.ejps.2017.07.001.
45. Riahi R, Tamayol A, Shaegh SAM, Ghaemmaghami A, Dokmeci MR, Khademshosseini A. Microfluidics for Advanced Drug Delivery Systems. Curr Opin Chem Eng. 2015;7:101-12. Epub 2015/02/01. doi: 10.1016/j.coche.2014.12.001. PubMed PMID: 31692947; PMCID: PMC6830738.
46. Dou M, Sanjay ST, Dominguez DC, Liu P, Xu F, Li X. Multiplexed instrument-free meningitis diagnosis on a polymer/paper hybrid microfluidic biochip. Biosens Bioelectron. 2017;87:865-73. Epub 2016/09/23. doi: 10.1016/j.bios.2016.09.033. PubMed PMID: 27657849; PMCID: PMC5125860.
47. Weibel DB, DiLuzio WR, Whitesides GM. Microfabrication meets microbiology. Nat Rev Microbiol. 2007;5(3):209-18. Epub 2007/02/17. doi: 10.1038/nrmicro1616. PubMed PMID: 17304250.
48. Whitesides GM, Stroock AD. Flexible Methods for Microfluidics. Phys Today. 2001;54(6):42-8. doi: 10.1063/1.1387591. PubMed PMID: WOS:000168917400018.
49. Catarino SO, Rodrigues RO, Pinho D, Miranda JM, Minas G, Lima R. Blood Cells Separation and Sorting Techniques of Passive Microfluidic Devices: From Fabrication to Applications. Micromachines (Basel). 2019;10(9):593. Epub 2019/09/13. doi: 10.3390/mi10090593. PubMed PMID: 31510012; PMCID: PMC6780402.
50. Shamloo A, Abdorahimzadeh S, Nasiri R. Exploring contraction–expansion inertial microfluidic-based particle separation devices integrated with curved channels. AIChE Journal. 2019;65(11):e16741.
51. Sanjay ST, Zhou W, Dou M, Tavakoli H, Ma L, Xu F, Li X. Recent advances of controlled drug delivery using microfluidic platforms. Adv Drug Deliv Rev. 2018;128:3-28.

Epub 2017/09/19. doi: 10.1016/j.addr.2017.09.013. PubMed PMID: 28919029; PMCID: PMC5854505.
52. Wu MH, Huang SB, Lee GB. Microfluidic cell culture systems for drug research. Lab Chip. 2010;10(8):939-56. Epub 2010/04/02. doi: 10.1039/b921695b. PubMed PMID: 20358102.
53. Khademhosseini A, Langer R, Borenstein J, Vacanti JP. Microscale technologies for tissue engineering and biology. Proc Natl Acad Sci U S A. 2006;103(8):2480-7. Epub 2006/02/16. doi: 10.1073/pnas.0507681102. PubMed PMID: 16477028; PMCID: PMC1413775.
54. Kang L, Chung BG, Langer R, Khademhosseini A. Microfluidics for drug discovery and development: from target selection to product lifecycle management. Drug Discov Today. 2008;13(1-2):1-13. Epub 2008/01/15. doi: 10.1016/j.drudis.2007.10.003. PubMed PMID: 18190858; PMCID: PMC2700821.
55. Dittrich PS, Manz A. Lab-on-a-chip: microfluidics in drug discovery. Nat Rev Drug Discov. 2006;5(3):210-8. Epub 2006/03/07. doi: 10.1038/nrd1985. PubMed PMID: 16518374.
56. Jun Y, Kang E, Chae S, Lee SH. Microfluidic spinning of micro- and nano-scale fibers for tissue engineering. Lab Chip. 2014;14(13):2145-60. Epub 2014/03/22. doi: 10.1039/c3lc51414e. PubMed PMID: 24647678.
57. Tsui JH, Lee W, Pun SH, Kim J, Kim DH. Microfluidics-assisted in vitro drug screening and carrier production. Adv Drug Deliv Rev. 2013;65(11-12):1575-88. Epub 2013/07/17. doi: 10.1016/j.addr.2013.07.004. PubMed PMID: 23856409; PMCID: PMC3834230.
58. Hasani-Sadrabadi MM, Taranejoo S, Dashtimoghadam E, Bahlakeh G, Majedi FS, VanDersarl JJ, Janmaleki M, Sharifi F, Bertsch A, Hourigan K, Tayebi L, Renaud P, Jacob KI. Microfluidic Manipulation of Core/Shell Nanoparticles for Oral Delivery of Chemotherapeutics: A New Treatment Approach for Colorectal Cancer. Adv Mater. 2016;28(21):4134-41. Epub 2016/03/24. doi: 10.1002/adma.201502697. PubMed PMID: 27001745.
59. Araujo F, Shrestha N, Shahbazi MA, Liu D, Herranz-Blanco B, Makila EM, Salonen JJ, Hirvonen JT, Granja PL, Sarmento F, Santos HA. Microfluidic Assembly of a Multifunctional Tailorable Composite System Designed for Site Specific Combined Oral Delivery of Peptide Drugs. ACS Nano. 2015;9(8):8291-302. Epub 2015/08/04. doi: 10.1021/acsnano.5b02762. PubMed PMID: 26235314.
60. Zhang H, Liu D, Shahbazi MA, Makila E, Herranz-Blanco B, Salonen J, Hirvonen J, Santos HA. Fabrication of a multifunctional nano-in-micro drug delivery platform by microfluidic templated encapsulation of porous silicon in polymer matrix. Adv Mater. 2014;26(26):4497-503. Epub 2014/04/17. doi: 10.1002/adma.201400953. PubMed PMID: 24737409.
61. Liu D, Herranz-Blanco Br, Mäkilä E, Arriaga LR, Mirza S, Weitz DA, Sandler N, Salonen J, Hirvonen J, Santos HlA. Microfluidic templated mesoporous silicon–solid lipid microcomposites for sustained drug delivery. ACS applied materials & interfaces. 2013;5(22):12127-34.
62. Li W, Liu D, Zhang H, Correia A, Makila E, Salonen J, Hirvonen J, Santos HA. Microfluidic assembly of a nano-in-micro dual drug delivery platform composed of halloysite nanotubes and a pH-responsive polymer for colon cancer therapy. Acta Biomater. 2017;48:238-46. Epub 2016/11/07. doi: 10.1016/j.actbio.2016.10.042. PubMed PMID: 27815166.
63. Khan FA, Narasimhan K, Swathi CSV, Mustak S, Mustafa G, Ahmad MZ, Akhter S. 3D Printing Technology in Customized Drug Delivery System: Current State of the Art, Prospective and the Challenges. Curr Pharm Des. 2018;24(42):5049-61. Epub 2019/01/15. doi: 10.2174/1381612825666190110153742. PubMed PMID: 30636582.

64. Khatri P, Shah MK, Vora N. Formulation strategies for solid oral dosage form using 3D printing technology: A mini-review. Journal of Drug Delivery Science Technology. 2018;46:148-55.
65. Davoodi E, Montazerian H, Haghniaz R, Rashidi A, Ahadian S, Sheikhi A, Chen J, Khademhosseini A, Milani AS, Hoorfar M. 3D-Printed Ultra-Robust Surface-Doped Porous Silicone Sensors for Wearable Biomonitoring. ACS nano. 2020.
66. Montazerian H, Mohamed M, Montazeri MM, Kheiri S, Milani A, Kim K, Hoorfar M. Permeability and mechanical properties of gradient porous PDMS scaffolds fabricated by 3D-printed sacrificial templates designed with minimal surfaces. Acta biomaterialia. 2019;96:149-60.
67. Davoodi E, Fayazfar H, Liravi F, Jabari E, Toyserkani E. Drop-on-demand high-speed 3D printing of flexible milled carbon fiber/silicone composite sensors for wearable biomonitoring devices. Additive Manufacturing. 2020;32:101016.
68. Ashammakhi N, Ahadian S, Xu C, Montazerian H, Ko H, Nasiri R, Barros N, Khademhosseini A. Bioinks and bioprinting technologies to make heterogeneous and biomimetic tissue constructs. Materials Today Bio. 2019:100008.
69. Fetah K, Tebon P, Goudie MJ, Eichenbaum J, Ren L, Barros N, Nasiri R, Ahadian S, Ashammakhi N, Dokmeci MR. The emergence of 3D bioprinting in organ-on-chip systems. Progress in Biomedical Engineering. 2019;1(1):012001
70. Seyedmahmoud R, Çelebi-Saltik B, Barros N, Nasiri R, Banton E, Shamloo A, Ashammakhi N, Dokmeci MR, Ahadian S. Three-Dimensional Bioprinting of Functional Skeletal Muscle Tissue Using Gelatin Methacryloyl-Alginate Bioinks. Micromachines. 2019;10(10):679.
71. Alomari M, Mohamed FH, Basit AW, Gaisford S. Personalised dosing: printing a dose of one's own medicine. International journal of pharmaceutics. 2015;494(2):568-77.
72. https://www.accessdata.fda.gov/drugsatfda_docs/nda/2015/207958Orig1s000TOC.cfm.
73. Prasad LK, Smyth H. 3D Printing technologies for drug delivery: a review. Drug Dev Ind Pharm. 2016;42(7):1019-31. Epub 2015/12/03. doi: 10.3109/03639045.2015.1120743. PubMed PMID: 26625986.
74. Lim SH, Kathuria H, Tan JJY, Kang L. 3D printed drug delivery and testing systems—a passing fad or the future? Advanced drug delivery reviews. 2018;132:139-68.
75. Nasrollahi F, Varshosaz J, Khodadadi AA, Lim S, Jahanian-Najafabadi A. Targeted delivery of docetaxel by use of transferrin/poly (allylamine hydrochloride)-functionalized graphene oxide nanocarrier. ACS applied materials & interfaces. 2016;8(21):13282-93.
76. Nasrollahi F, Sana B, Paramelle D, Ahadian S, Khademhosseini A, Lim S. Incorporation of Graphene Quantum Dots, Iron, and Doxorubicin in/on Ferritin Nanocages for Bimodal Imaging and Drug Delivery. Advanced Therapeutics.1900183.
77. Zhang L, Wang S, Zhang M, Sun J. Nanocarriers for oral drug delivery. J Drug Target. 2013;21(6):515-27. Epub 2013/04/30. doi: 10.3109/1061186X.2013.789033. PubMed PMID: 23621127.
78. Augustine R, Ashkenazi DL, Arzi RS, Zlobin V, Shofti R, Sosnik A. Nanoparticle-in-microparticle oral drug delivery system of a clinically relevant darunavir/ritonavir antiretroviral combination. Acta Biomater. 2018;74:344-59. Epub 2018/05/04. doi: 10.1016/j.actbio.2018.04.045. PubMed PMID: 29723705.
79. Moreno JAS, Mendes AC, Stephansen K, Engwer C, Goycoolea FM, Boisen A, Nielsen LH, Chronakis IS. Development of electrosprayed mucoadhesive chitosan microparticles.

Carbohydr Polym. 2018;190:240-7. Epub 2018/04/10. doi: 10.1016/j.carbpol.2018.02.062. PubMed PMID: 29628244.
80. Park CG, Huh BK, Kim SN, Lee SH, Hong HR, Choy YB. Nanostructured mucoadhesive microparticles to enhance oral drug bioavailability. Journal of Industrial and Engineering Chemistry. 2017;54:262-9. doi: 10.1016/j.jiec.2017.06.001.
81. Swider E, Koshkina O, Tel J, Cruz LJ, de Vries IJM, Srinivas M. Customizing poly(lactic-co-glycolic acid) particles for biomedical applications. Acta Biomater. 2018;73:38-51. Epub 2018/04/14. doi: 10.1016/j.actbio.2018.04.006. PubMed PMID: 29653217.
82. Lamprecht A, Rodero Torres H, Schäfer U, Lehr C-M. Biodegradable microparticles as a two-drug controlled release formulation: a potential treatment of inflammatory bowel disease. Journal of Controlled Release. 2000;69(3):445-54. doi: 10.1016/s0168-3659(00)00331-x.
83. Valot P, Baba M, Nedelec JM, Sintes-Zydowicz N. Effects of process parameters on the properties of biocompatible ibuprofen-loaded microcapsules. Int J Pharm. 2009;369(1-2):53-63. Epub 2008/12/17. doi: 10.1016/j.ijpharm.2008.10.037. PubMed PMID: 19084583.
84. Petitti M, Barresi AA, Vanni M. Controlled release of vancomycin from PCL microcapsules for an ophthalmic application. Chemical Engineering Research and Design. 2009;87(6):859-66. doi: 10.1016/j.cherd.2008.12.008.
85. Zhang Z, Zhu Y, Yang X, Li C. Preparation of azithromycin microcapsules by a layer-by-layer self-assembly approach and release behaviors of azithromycin. Colloids and Surfaces A: Physicochemical and Engineering Aspects. 2010;362(1):135-9. doi: https://doi.org/10.1016/j.colsurfa.2010.04.006.
86. Moore C, Kosgodage U, Lange S, Inal JM. The emerging role of exosome and microvesicle- (EMV-) based cancer therapeutics and immunotherapy. Int J Cancer. 2017;141(3):428-36. Epub 2017/03/02. doi: 10.1002/ijc.30672. PubMed PMID: 28247946.
87. Tang K, Zhang Y, Zhang H, Xu P, Liu J, Ma J, Lv M, Li D, Katirai F, Shen GX, Zhang G, Feng ZH, Ye D, Huang B. Delivery of chemotherapeutic drugs in tumour cell-derived microparticles. Nat Commun. 2012;3:1282. Epub 2012/12/20. doi: 10.1038/ncomms2282. PubMed PMID: 23250412.
88. McQuilling JP, Arenas-Herrera J, Childers C, Pareta RA, Khanna O, Jiang B, Brey EM, Farney AC, Opara EC. New alginate microcapsule system for angiogenic protein delivery and immunoisolation of islets for transplantation in the rat omentum pouch. Transplant Proc. 2011;43(9):3262-4. Epub 2011/11/22. doi: 10.1016/j.transproceed.2011.10.030. PubMed PMID: 22099771; PMCID: PMC3241996.
89. Glowka E, Stasiak J, Lulek J. Drug Delivery Systems for Vitamin D Supplementation and Therapy. Pharmaceutics. 2019;11(7):347. Epub 2019/07/22. doi: 10.3390/pharmaceutics11070347. PubMed PMID: 31323777; PMCID: PMC6680748.
90. Han FY, Thurecht KJ, Whittaker AK, Smith MT. Bioerodable PLGA-Based Microparticles for Producing Sustained-Release Drug Formulations and Strategies for Improving Drug Loading. Front Pharmacol. 2016;7:185. Epub 2016/07/23. doi: 10.3389/fphar.2016.00185. PubMed PMID: 27445821; PMCID: PMC4923250.
91. Lengyel M, Kállai-Szabó N, Antal V, Laki AJ, Antal I. Microparticles, Microspheres, and Microcapsules for Advanced Drug Delivery. Scientia Pharmaceutica. 2019;87(3). doi: 10.3390/scipharm87030020.
92. Noble GT, Stefanick JF, Ashley JD, Kiziltepe T, Bilgicer B. Ligand-targeted liposome design: challenges and fundamental considerations. Trends in biotechnology. 2014;32(1):32-45.
93. Çağdaş M, Sezer AD, Bucak S. Liposomes as potential drug carrier systems for drug

delivery. Application of nanotechnology in drug delivery. 2014:1-100.
94. Immordino ML, Dosio F, Cattel L. Stealth liposomes: review of the basic science, rationale, and clinical applications, existing and potential. International journal of nanomedicine. 2006;1(3):297.
95. Law SL, Huang KJ, Chiang CH. Acyclovir-containing liposomes for potential ocular delivery. Corneal penetration and absorption. J Control Release. 2000;63(1-2):135-40. Epub 2000/01/21. doi: 10.1016/s0168-3659(99)00192-3. PubMed PMID: 10640587.
96. Li P, Nielsen HM, Müllertz A. Oral delivery of peptides and proteins using lipid-based drug delivery systems. Expert opinion on drug delivery. 2012;9(10):1289-304.
97. Mehnert W, Mäder K. Solid lipid nanoparticles: production, characterization and applications. Advanced drug delivery reviews. 2012;64:83-101.
98. Sanjula B, Shah FM, Javed A, Alka A. Effect of poloxamer 188 on lymphatic uptake of carvedilol-loaded solid lipid nanoparticles for bioavailability enhancement. J Drug Target. 2009;17(3):249-56. Epub 2009/03/04. doi: 10.1080/10611860902718672. PubMed PMID: 19255893.
99. Kumar VV, Chandrasekar D, Ramakrishna S, Kishan V, Rao YM, Diwan PV. Development and evaluation of nitrendipine loaded solid lipid nanoparticles: influence of wax and glyceride lipids on plasma pharmacokinetics. Int J Pharm. 2007;335(1-2):167-75. Epub 2006/12/13. doi: 10.1016/j.ijpharm.2006.11.004. PubMed PMID: 17161566.
100. Luo CF, Yuan M, Chen MS, Liu SM, Zhu L, Huang BY, Liu XW, Xiong W. Pharmacokinetics, tissue distribution and relative bioavailability of puerarin solid lipid nanoparticles following oral administration. Int J Pharm. 2011;410(1-2):138-44. Epub 2011/03/12. doi: 10.1016/j.ijpharm.2011.02.064. PubMed PMID: 21392565.
101. Boushra M, Tous S, Fetih G, Korzekwa K, Lebo DB, Xue HY, Wong HL. Development and evaluation of viscosity-enhanced nanocarrier (VEN) for oral insulin delivery. Int J Pharm. 2016;511(1):462-72. Epub 2016/07/16. doi: 10.1016/j.ijpharm.2016.07.016. PubMed PMID: 27418563.
102. Woodley J. Bioadhesion: new possibilities for drug administration? Clin Pharmacokinet. 2001;40(2):77-84. Epub 2001/03/05. doi: 10.2165/00003088-200140020-00001. PubMed PMID: 11286325.
103. des Rieux A, Fievez V, Garinot M, Schneider Y-J, Préat V. Nanoparticles as potential oral delivery systems of proteins and vaccines: a mechanistic approach. Journal of controlled release. 2006;116(1):1-27.
104. Eldridge JH, Hammond CJ, Meulbroek JA, Staas JK, Gilley RM, Tice TR. Controlled vaccine release in the gut-associated lymphoid tissues. I. Orally administered biodegradable microspheres target the Peyer's patches. Journal of Controlled Release. 1990;11(1-3):205-14.
105. https://www.accessdata.fda.gov/scripts/publications/search_result_record.cfm?id=63227.
106. Khalil NM, do Nascimento TCF, Casa DM, Dalmolin LF, de Mattos AC, Hoss I, Romano MA, Mainardes RM. Pharmacokinetics of curcumin-loaded PLGA and PLGA–PEG blend nanoparticles after oral administration in rats. Colloids and Surfaces B: Biointerfaces. 2013;101:353-60.
107. Vasiliu S, Bunia I, Racovita S, Neagu V. Adsorption of cefotaxime sodium salt on polymer coated ion exchange resin microparticles: Kinetics, equilibrium and thermodynamic studies. Carbohydrate Polymers. 2011;85(2):376-87. doi: 10.1016/j.carbpol.2011.02.039.
108. Wang W, Liu L, Ju XJ, Zerrouki D, Xie R, Yang L, Chu LY. A novel thermo-induced self-bursting microcapsule with magnetic-targeting property. Chemphyschem. 2009;10(14):2405-9.

Epub 2009/08/26. doi: 10.1002/cphc.200900450. PubMed PMID: 19705393.
109. Koetting MC, Guido JF, Gupta M, Zhang A, Peppas NA. pH-responsive and enzymatically-responsive hydrogel microparticles for the oral delivery of therapeutic proteins: Effects of protein size, crosslinking density, and hydrogel degradation on protein delivery. Journal of Controlled Release. 2016;221:18-25.
110. Lee SC, Huh KM, Lee J, Cho YW, Galinsky RE, Park K. Hydrotropic polymeric micelles for enhanced paclitaxel solubility: in vitro and in vivo characterization. Biomacromolecules. 2007;8(1):202-8. Epub 2007/01/09. doi: 10.1021/bm060307b. PubMed PMID: 17206808; PMCID: PMC2532792.
111. Coimbra M, Rijcken CJ, Stigter M, Hennink WE, Storm G, Schiffelers RM. Antitumor efficacy of dexamethasone-loaded core-crosslinked polymeric micelles. J Control Release. 2012;163(3):361-7. Epub 2012/10/09. doi: 10.1016/j.jconrel.2012.09.014. PubMed PMID: 23041274.
112. Satturwar P, Eddine MN, Ravenelle F, Leroux JC. pH-responsive polymeric micelles of poly(ethylene glycol)-b-poly(alkyl(meth)acrylate-co-methacrylic acid): influence of the copolymer composition on self-assembling properties and release of candesartan cilexetil. Eur J Pharm Biopharm. 2007;65(3):379-87. Epub 2006/11/25. doi: 10.1016/j.ejpb.2006.09.012. PubMed PMID: 17123802.
113. Vrignaud S, Anton N, Gayet P, Benoit JP, Saulnier P. Reverse micelle-loaded lipid nanocarriers: a novel drug delivery system for the sustained release of doxorubicin hydrochloride. Eur J Pharm Biopharm. 2011;79(1):197-204. Epub 2011/02/25. doi: 10.1016/j.ejpb.2011.02.015. PubMed PMID: 21345371.
114. Tian H, He Z, Sun C, Yang C, Zhao P, Liu L, Leong KW, Mao HQ, Liu Z, Chen Y. Uniform core–shell nanoparticles with thiolated hyaluronic acid coating to enhance oral delivery of insulin. Advanced healthcare materials. 2018;7(17):1800285.
115. Dufresne MH, Gauthier MA, Leroux JC. Thiol-functionalized polymeric micelles: from molecular recognition to improved mucoadhesion. Bioconjug Chem. 2005;16(4):1027-33. Epub 2005/07/21. doi: 10.1021/bc050067b. PubMed PMID: 16029046.
116. Ma XY, Pan GM, Lu Z, Hu JS, Bei JZ, Jia JH, Wang SG. Preliminary study of oral polylactide microcapsulated insulin in vitro and in vivo. Diabetes Obes Metab. 2000;2(4):243-50. Epub 2001/02/28. doi: 10.1046/j.1463-1326.2000.00080.x. PubMed PMID: 11225658.
117. Guan Q, Chen W, Hu X. Development of lovastatin-loaded poly(lactic acid) microspheres for sustained oral delivery: in vitro and ex vivo evaluation. Drug Des Devel Ther. 2015;9:791-8. Epub 2015/02/25. doi: 10.2147/DDDT.S76676. PubMed PMID: 25709403; PMCID: PMC4330030.
118. Mandal TK, Bostanian LA, Graves RA, Chapman SR, Womack I. Development of biodegradable microcapsules as carrier for oral controlled delivery of amifostine. Drug Dev Ind Pharm. 2002;28(3):339-44. Epub 2002/05/25. doi: 10.1081/ddc-120002849. PubMed PMID: 12026226.
119. Tian J, Sun X, Chen X, Yu J, Qu L, Wang L. The formulation and immunisation of oral poly(DL-lactide-co-glycolide) microcapsules containing a plasmid vaccine against lymphocystis disease virus in Japanese flounder (Paralichthys olivaceus). Int Immunopharmacol. 2008;8(6):900-8. Epub 2008/04/30. doi: 10.1016/j.intimp.2008.02.006. PubMed PMID: 18442796.
120. Cheng J, Teply BA, Jeong SY, Yim CH, Ho D, Sherifi I, Jon S, Farokhzad OC, Khademhosseini A, Langer RS. Magnetically responsive polymeric microparticles for oral

delivery of protein drugs. Pharm Res. 2006;23(3):557-64. Epub 2006/01/03. doi: 10.1007/s11095-005-9444-5. PubMed PMID: 16388405.
121. Prasad S, Tyagi AK, Aggarwal BB. Recent developments in delivery, bioavailability, absorption and metabolism of curcumin: the golden pigment from golden spice. Cancer Res Treat. 2014;46(1):2-18. Epub 2014/02/13. doi: 10.4143/crt.2014.46.1.2. PubMed PMID: 24520218; PMCID: PMC3918523.
122. Zhou X, Chen Q, Ma Y, Huang Y, Gou S, Xiao B. Porous Polymeric Microparticles as an Oral Drug Platform for Effective Ulcerative Colitis Treatment. J Pharm Sci. 2019;108(7):2238-42. Epub 2019/02/20. doi: 10.1016/j.xphs.2019.02.001. PubMed PMID: 30779888.
123. Benoit MA, Baras B, Gillard J. Preparation and characterization of protein-loaded poly(epsilon-caprolactone) microparticles for oral vaccine delivery. Int J Pharm. 1999;184(1):73-84. Epub 1999/07/30. doi: 10.1016/s0378-5173(99)00109-x. PubMed PMID: 10425353.
124. Barboza FM, Machado WM, Olchanheski Junior LR, Padilha de Paula J, Zawadzki SF, Fernandes D, Farago PV. PCL/PHBV microparticles as innovative carriers for oral controlled release of manidipine dihydrochloride. ScientificWorldJournal. 2014;2014:268107. Epub 2014/02/20. doi: 10.1155/2014/268107. PubMed PMID: 24510699; PMCID: PMC3914481.
125. Chattopadhyay H, De AK, Datta S. Novel Starch-PVA Polymer for Microparticle Preparation and Optimization Using Factorial Design Study. Int Sch Res Notices. 2015;2015:261476. Epub 2015/01/01. doi: 10.1155/2015/261476. PubMed PMID: 27347511; PMCID: PMC4897262.
126. Rassu G, Nieddu M, Bosi P, Trevisi P, Colombo M, Priori D, Manconi P, Giunchedi P, Gavini E, Boatto G. Encapsulation and modified-release of thymol from oral microparticles as adjuvant or substitute to current medications. Phytomedicine. 2014;21(12):1627-32. doi: https://doi.org/10.1016/j.phymed.2014.07.017
127. Pandav S, Naik J. Preparation and In Vitro Evaluation of Ethylcellulose and Polymethacrylate Resins Loaded Microparticles Containing Hydrophilic Drug. J Pharm (Cairo). 2014;2014:904036. Epub 2014/01/01. doi: 10.1155/2014/904036. PubMed PMID: 26556206; PMCID: PMC4590829.
128. Bulut E. Chitosan coated- and uncoated-microspheres of sodium carboxymethyl cellulose/polyvinyl alcohol crosslinked with ferric ion: flurbiprofen loading and in vitro drug release study. Journal of Macromolecular Science, Part A. 2019;57(1):72-82. doi: 10.1080/10601325.2019.1671770.
129. Gadalla HH, El Gibaly I, Soliman GM, Mohamed FA, El-Sayed AM. Amidated pectin/sodium carboxymethylcellulose microspheres as a new carrier for colonic drug targeting: Development and optimization by factorial design. Carbohydr Polym. 2016;153:526-34. Epub 2016/08/27. doi: 10.1016/j.carbpol.2016.08.018. PubMed PMID: 27561525.
130. van der Lubben IM, Verhoef JC, van Aelst AC, Borchard G, Junginger HE. Chitosan microparticles for oral vaccination: preparation, characterization and preliminary in vivo uptake studies in murine Peyer's patches. Biomaterials. 2001;22(7):687-94. Epub 2001/03/15. doi: 10.1016/s0142-9612(00)00231-3. PubMed PMID: 11246962.
131. Ramalingam P, Ko YT. Enhanced oral delivery of curcumin from N-trimethyl chitosan surface-modified solid lipid nanoparticles: pharmacokinetic and brain distribution evaluations. Pharm Res. 2015;32(2):389-402. Epub 2014/08/02. doi: 10.1007/s11095-014-1469-1. PubMed PMID: 25082210.
132. Gadalla HH, Soliman GM, Mohammed FA, El-Sayed AM. Development and in vitro/in vivo evaluation of Zn-pectinate microparticles reinforced with chitosan for the colonic delivery

of progesterone. Drug Deliv. 2016;23(7):2541-54. Epub 2016/10/18. doi: 10.3109/10717544.2015.1028602. PubMed PMID: 25853478.
133. Sahiner N, Suner SS, Ayyala RS. Mesoporous, degradable hyaluronic acid microparticles for sustainable drug delivery application. Colloids Surf B Biointerfaces. 2019;177:284-93. Epub 2019/02/16. doi: 10.1016/j.colsurfb.2019.02.015. PubMed PMID: 30769230.
134. Zhang Z, Zhang R, Zou L, Chen L, Ahmed Y, Al Bishri W, Balamash K, McClements DJ. Encapsulation of curcumin in polysaccharide-based hydrogel beads: Impact of bead type on lipid digestion and curcumin bioaccessibility. Food Hydrocolloids. 2016;58:160-70. doi: 10.1016/j.foodhyd.2016.02.036.
135. Builders PF, Kunle OO, Okpaku LC, Builders MI, Attama AA, Adikwu MU. Preparation and evaluation of mucinated sodium alginate microparticles for oral delivery of insulin. Eur J Pharm Biopharm. 2008;70(3):777-83. Epub 2008/07/23. doi: 10.1016/j.ejpb.2008.06.021. PubMed PMID: 18644444.
136. Patel ZS, Ueda H, Yamamoto M, Tabata Y, Mikos AG. In vitro and in vivo release of vascular endothelial growth factor from gelatin microparticles and biodegradable composite scaffolds. Pharm Res. 2008;25(10):2370-8. Epub 2008/07/30. doi: 10.1007/s11095-008-9685-1. PubMed PMID: 18663411.
137. Patel ZS, Yamamoto M, Ueda H, Tabata Y, Mikos AG. Biodegradable gelatin microparticles as delivery systems for the controlled release of bone morphogenetic protein-2. Acta Biomater. 2008;4(5):1126-38. Epub 2008/05/14. doi: 10.1016/j.actbio.2008.04.002. PubMed PMID: 18474452; PMCID: PMC3018807.
138. Silva DM, Vyas HKN, Sanderson-Smith ML, Sencadas V. Development and optimization of ciprofloxacin-loaded gelatin microparticles by single-step spray-drying technique. Powder Technology. 2018;330:201-9. doi: 10.1016/j.powtec.2018.02.029.
139. Sajeesh S, Vauthier C, Gueutin C, Ponchel G, Sharma CP. Thiol functionalized polymethacrylic acid-based hydrogel microparticles for oral insulin delivery. Acta Biomater. 2010;6(8):3072-80. Epub 2010/02/11. doi: 10.1016/j.actbio.2010.02.007. PubMed PMID: 20144748.
140. Saboktakin MR, Tabatabaie RM, Maharramov A, Ramazanov MA. Synthesis and in vitro evaluation of carboxymethyl starch-chitosan nanoparticles as drug delivery system to the colon. Int J Biol Macromol. 2011;48(3):381-5. Epub 2010/10/20. doi: 10.1016/j.ijbiomac.2010.10.005. PubMed PMID: 20955723.
141. Bulut E. Ibuprofen microencapsulation within acrylamide-grafted chitosan and methylcellulose interpenetrating polymer network microspheres: Synthesis, characterization, and release studies. Artif Cells Nanomed Biotechnol. 2016;44(4):1098-108. Epub 2015/03/10. doi: 10.3109/21691401.2015.1011802. PubMed PMID: 25749277.
142. Anselmo AC, Xu X, Buerkli S, Zeng Y, Tang W, McHugh KJ, Behrens AM, Rosenberg E, Duan AR, Sugarman JL, Zhuang J, Collins J, Lu X, Graf T, Tzeng SY, Rose S, Acolatse S, Nguyen TD, Le X, Guerra AS, Freed LE, Weinstock SB, Sears CB, Nikolic B, Wood L, Welkhoff PA, Oxley JD, Moretti D, Zimmermann MB, Langer R, Jaklenec A. A heat-stable microparticle platform for oral micronutrient delivery. Sci Transl Med. 2019;11(518):eaaw3680. Epub 2019/11/15. doi: 10.1126/scitranslmed.aaw3680. PubMed PMID: 31723037.
143. Mo R, Jiang T, Di J, Tai W, Gu Z. Emerging micro- and nanotechnology based synthetic approaches for insulin delivery. Chem Soc Rev. 2014;43(10):3595-629. Epub 2014/03/15. doi: 10.1039/c3cs60436e. PubMed PMID: 24626293.
144. Andreani T, de Souza AL, Kiill CP, Lorenzon EN, Fangueiro JF, Calpena AC, Chaud MV,

Garcia ML, Gremiao MP, Silva AM, Souto EB. Preparation and characterization of PEG-coated silica nanoparticles for oral insulin delivery. Int J Pharm. 2014;473(1-2):627-35. Epub 2014/08/05. doi: 10.1016/j.ijpharm.2014.07.049. PubMed PMID: 25089510.
145. Zheng X, Wang T, Jiang H, Li Y, Jiang T, Zhang J, Wang S. Incorporation of Carvedilol into PAMAM-functionalized MWNTs as a sustained drug delivery system for enhanced dissolution and drug-loading capacity. asian journal of pharmaceutical sciences. 2013;8(5):278-86.
146. Andreani T, Miziara L, Lorenzon EN, de Souza AL, Kiill CP, Fangueiro JF, Garcia ML, Gremiao PD, Silva AM, Souto EB. Effect of mucoadhesive polymers on the in vitro performance of insulin-loaded silica nanoparticles: Interactions with mucin and biomembrane models. Eur J Pharm Biopharm. 2015;93:118-26. Epub 2015/04/07. doi: 10.1016/j.ejpb.2015.03.027. PubMed PMID: 25843239.
147. Colley HE, Said Z, Santocildes-Romero ME, Baker SR, D'Apice K, Hansen J, Madsen LS, Thornhill MH, Hatton PV, Murdoch C. Pre-clinical evaluation of novel mucoadhesive bilayer patches for local delivery of clobetasol-17-propionate to the oral mucosa. Biomaterials. 2018;178:134-46. Epub 2018/06/22. doi: 10.1016/j.biomaterials.2018.06.009. PubMed PMID: 29929183.
148. Banerjee A, Chen R, Arafin S, Mitragotri S. Intestinal iontophoresis from mucoadhesive patches: a strategy for oral delivery. J Control Release. 2019;297:71-8. Epub 2019/02/02. doi: 10.1016/j.jconrel.2019.01.037. PubMed PMID: 30707901.
149. Whitehead K, Shen Z, Mitragotri S. Oral delivery of macromolecules using intestinal patches: applications for insulin delivery. J Control Release. 2004;98(1):37-45. Epub 2004/07/13. doi: 10.1016/j.jconrel.2004.04.013. PubMed PMID: 15245887.
150. Kirsch K, Hanke U, Weitschies W. An overview of intestinal wafers for oral drug delivery. Eur J Pharm Biopharm. 2017;114:135-44. Epub 2017/01/24. doi: 10.1016/j.ejpb.2017.01.003. PubMed PMID: 28111241.
151. Shinkar DM, Dhake AS, Setty CM. Drug delivery from the oral cavity: a focus on mucoadhesive buccal drug delivery systems. PDA J Pharm Sci Technol. 2012;66(5):466-500. Epub 2012/10/05. doi: 10.5731/pdajpst.2012.00877. PubMed PMID: 23035030.
152. Teutonico D, Ponchel G. Patches for improving gastrointestinal absorption: an overview. Drug Discov Today. 2011;16(21-22):991-7. Epub 2011/06/21. doi: 10.1016/j.drudis.2011.05.013. PubMed PMID: 21683153.
153. Gioumouxouzis CI, Katsamenis OL, Bouropoulos N, Fatouros DG. 3D printed oral solid dosage forms containing hydrochlorothiazide for controlled drug delivery. Journal of Drug Delivery Science and Technology. 2017;40:164-71.
154. Li Q, Wen H, Jia D, Guan X, Pan H, Yang Y, Yu S, Zhu Z, Xiang R, Pan W. Preparation and investigation of controlled-release glipizide novel oral device with three-dimensional printing. Int J Pharm. 2017;525(1):5-11. Epub 2017/04/06. doi: 10.1016/j.ijpharm.2017.03.066. PubMed PMID: 28377316.
155. Gioumouxouzis CI, Baklavaridis A, Katsamenis OL, Markopoulou CK, Bouropoulos N, Tzetzis D, Fatouros DG. A 3D printed bilayer oral solid dosage form combining metformin for prolonged and glimepiride for immediate drug delivery. Eur J Pharm Sci. 2018;120:40-52. Epub 2018/04/22. doi: 10.1016/j.ejps.2018.04.020. PubMed PMID: 29678613.
156. Buanz ABM, Belaunde CC, Soutari N, Tuleu C, Gul MO, Gaisford S. Ink-jet printing versus solvent casting to prepare oral films: Effect on mechanical properties and physical stability. Int J Pharm. 2015;494(2):611-8. Epub 2014/12/21. doi: 10.1016/j.ijpharm.2014.12.032.

PubMed PMID: 25526674.
157. Khaled SA, Burley JC, Alexander MR, Yang J, Roberts CJ. 3D printing of five-in-one dose combination polypill with defined immediate and sustained release profiles. J Control Release. 2015;217:308-14. Epub 2015/09/24. doi: 10.1016/j.jconrel.2015.09.028. PubMed PMID: 26390808.
158. Maroni A, Melocchi A, Parietti F, Foppoli A, Zema L, Gazzaniga A. 3D printed multi-compartment capsular devices for two-pulse oral drug delivery. J Control Release. 2017;268:10-8. Epub 2017/10/17. doi: 10.1016/j.jconrel.2017.10.008. PubMed PMID: 29030223.
159. Melocchi A, Parietti F, Loreti G, Maroni A, Gazzaniga A, Zema L. 3D printing by fused deposition modeling (FDM) of a swellable/erodible capsular device for oral pulsatile release of drugs. Journal of Drug Delivery Science and Technology. 2015;30:360-7. doi: 10.1016/j.jddst.2015.07.016.
160. Goyanes A, Wang J, Buanz A, Martinez-Pacheco R, Telford R, Gaisford S, Basit AW. 3D Printing of Medicines: Engineering Novel Oral Devices with Unique Design and Drug Release Characteristics. Mol Pharm. 2015;12(11):4077-84. Epub 2015/10/17. doi: 10.1021/acs.molpharmaceut.5b00510. PubMed PMID: 26473653
161. Kirtane AR, Abouzid O, Minahan D, Bensel T, Hill AL, Selinger C, Bershteyn A, Craig M, Mo SS, Mazdiyasni H, Cleveland C, Rogner J, Lee YA, Booth L, Javid F, Wu SJ, Grant T, Bellinger AM, Nikolic B, Hayward A, Wood L, Eckhoff PA, Nowak MA, Langer R, Traverso G. Development of an oral once-weekly drug delivery system for HIV antiretroviral therapy. Nat Commun. 2018;9(1):2. Epub 2018/01/11. doi: 10.1038/s41467-017-02294-6. PubMed PMID: 29317618; PMCID: PMC5760734.
162. Lim SH, Chia SM, Kang L, Yap KY. Three-Dimensional Printing of Carbamazepine Sustained-Release Scaffold. J Pharm Sci. 2016;105(7):2155-63. Epub 2016/06/13. doi: 10.1016/j.xphs.2016.04.031. PubMed PMID: 27290630.
163. Marizza P, Keller SS, Müllertz A, Boisen A. Polymer-filled microcontainers for oral delivery loaded using supercritical impregnation. Journal of Controlled Release. 2014;173:1-9.
164. Marizza P, Pontoni L, Rindzevicius T, Alopaeus J, Su K, Zeitler J, Keller SS, Kikic I, Moneghini M, De Zordi N. Supercritical impregnation of polymer matrices spatially confined in microcontainers for oral drug delivery: Effect of temperature, pressure and time. The Journal of Supercritical Fluids. 2016;107:45-52.
165. Guan J, He H, Lee LJ, Hansford DJ. Fabrication of particulate Reservoir-Containing, capsulelike, and Self-Folding polymer microstructures for drug delivery. Small. 2007;3(3):412-8.
166. Mazzoni C, Tentor F, Strindberg SA, Nielsen LH, Keller SS, Alstrom TS, Gundlach C, Mullertz A, Marizza P, Boisen A. From concept to in vivo testing: Microcontainers for oral drug delivery. J Control Release. 2017;268:343-51. Epub 2017/10/22. doi: 10.1016/j.jconrel.2017.10.013. PubMed PMID: 29054373.
167. D'Emanuele A, Jevprasesphant R, Penny J, Attwood D. The use of a dendrimer-propranolol prodrug to bypass efflux transporters and enhance oral bioavailability. Journal of controlled release. 2004;95(3):447-53.
168. Gardiner J, Freeman S, Leach M, Green A, Alcock J, D'Emanuele A. PAMAM dendrimers for the delivery of the antibacterial Triclosan. Journal of enzyme inhibition and medicinal chemistry. 2008;23(5):623-8.
169. Winnicka K, Wroblewska M, Wieczorek P, Sacha PT, Tryniszewska E. Hydrogel of ketoconazole and PAMAM dendrimers: formulation and antifungal activity. Molecules. 2012;17(4):4612-24.

170. Li H, Yu Y, Faraji Dana S, Li B, Lee CY, Kang L. Novel engineered systems for oral, mucosal and transdermal drug delivery. J Drug Target. 2013;21(7):611-29. Epub 2013/07/23. doi: 10.3109/1061186X.2013.805335. PubMed PMID: 23869879.
171. Tao SL, Desai TA. Microfabricated drug delivery systems: from particles to pores. Adv Drug Deliv Rev. 2003;55(3):315-28. Epub 2003/03/12. doi: 10.1016/s0169-409x(02)00227-2. PubMed PMID: 12628319.
172. Liu D, Liu C, Zou W, Zhang N. Enhanced gastrointestinal absorption of N3-O-toluyl-fluorouracil by cationic solid lipid nanoparticles. Journal of Nanoparticle Research. 2009;12(3):975-84. doi: 10.1007/s11051-009-9648-4.
173. Kawashima Y, Yamamoto H, Takeuchi H, Kuno Y. Mucoadhesive DL-lactide/glycolide copolymer nanospheres coated with chitosan to improve oral delivery of elcatonin. Pharm Dev Technol. 2000;5(1):77-85. Epub 2000/02/12. doi: 10.1081/pdt-100100522. PubMed PMID: 10669921.
174. Takeuchi H, Thongborisute J, Matsui Y, Sugihara H, Yamamoto H, Kawashima Y. Novel mucoadhesion tests for polymers and polymer-coated particles to design optimal mucoadhesive drug delivery systems. Adv Drug Deliv Rev. 2005;57(11):1583-94. Epub 2005/09/20. doi: 10.1016/j.addr.2005.07.008. PubMed PMID: 16169120.
175. Iqbal J, Shahnaz G, Perera G, Hintzen F, Sarti F, Bernkop-Schnurch A. Thiolated chitosan: development and in vivo evaluation of an oral delivery system for leuprolide. Eur J Pharm Biopharm. 2012;80(1):95-102. Epub 2011/10/04. doi: 10.1016/j.ejpb.2011.09.010. PubMed PMID: 21964316.
176. Hosseinzadeh H, Atyabi F, Dinarvand R, Ostad SN. Chitosan–Pluronic nanoparticles as oral delivery of anticancer gemcitabine: preparation and in vitro study. International journal of nanomedicine. 2012;7:1851.
177. Bravo-Osuna I, Vauthier C, Farabollini A, Palmieri GF, Ponchel G. Mucoadhesion mechanism of chitosan and thiolated chitosan-poly(isobutyl cyanoacrylate) core-shell nanoparticles. Biomaterials. 2007;28(13):2233-43. Epub 2007/01/31. doi: 10.1016/j.biomaterials.2007.01.005. PubMed PMID: 17261330.
178. Sarmento B, Ferreira D, Jorgensen L, Van De Weert M. Probing insulin's secondary structure after entrapment into alginate/chitosan nanoparticles. European Journal of Pharmaceutics and Biopharmaceutics. 2007;65(1):10-7.
179. Abd El Hady WE, Mohamed EA, Soliman OAE, El-Sabbagh HM. In vitro-in vivo evaluation of chitosan-PLGA nanoparticles for potentiated gastric retention and anti-ulcer activity of diosmin. Int J Nanomedicine. 2019;14:7191-213. Epub 2019/10/01. doi: 10.2147/IJN.S213836. PubMed PMID: 31564873; PMCID: PMC6732519.
180. Xu B, Jiang G, Yu W, Liu D, Liu Y, Kong X, Yao J. Preparation of poly(lactic-co-glycolic acid) and chitosan composite nanocarriers via electrostatic self assembly for oral delivery of insulin. Mater Sci Eng C Mater Biol Appl. 2017;78:420-8. Epub 2017/06/04. doi: 10.1016/j.msec.2017.04.113. PubMed PMID: 28576004.
181. Hejazi R, Amiji M. Chitosan-based gastrointestinal delivery systems. J Control Release. 2003;89(2):151-65. Epub 2003/04/25. doi: 10.1016/s0168-3659(03)00126-3. PubMed PMID: 12711440.
182. Kowapradit J, Apirakaramwong A, Ngawhirunpat T, Rojanarata T, Sajomsang W, Opanasopit P. Methylated N-(4-N,N-dimethylaminobenzyl) chitosan coated liposomes for oral protein drug delivery. Eur J Pharm Sci. 2012;47(2):359-66. Epub 2012/07/10. doi: 10.1016/j.ejps.2012.06.020. PubMed PMID: 22771544.

183. Leitner V, Marschütz M, Bernkop-Schnürch A. Mucoadhesive and cohesive properties of poly (acrylic acid)-cysteine conjugates with regard to their molecular mass. European journal of pharmaceutical sciences. 2003;18(1):89-96.
184. Werle M, Hoffer M. Glutathione and thiolated chitosan inhibit multidrug resistance P-glycoprotein activity in excised small intestine. J Control Release. 2006;111(1-2):41-6. Epub 2005/12/27. doi: 10.1016/j.jconrel.2005.11.011. PubMed PMID: 16377016.
185. Zhang X, Qi J, Lu Y, He W, Li X, Wu W. Biotinylated liposomes as potential carriers for the oral delivery of insulin. Nanomedicine: nanotechnology, biology and medicine. 2014;10(1):167-76.
186. Zhang N, Ping QN, Huang GH, Xu WF. Investigation of lectin-modified insulin liposomes as carriers for oral administration. International journal of pharmaceutics. 2005;294(1-2):247-59.
187. Collnot EM, Baldes C, Schaefer UF, Edgar KJ, Wempe MF, Lehr CM. Vitamin E TPGS P-glycoprotein inhibition mechanism: influence on conformational flexibility, intracellular ATP levels, and role of time and site of access. Mol Pharm. 2010;7(3):642-51. Epub 2010/03/09. doi: 10.1021/mp900191s. PubMed PMID: 20205474.
188. Zhang Z, Tan S, Feng S-S. Vitamin E TPGS as a molecular biomaterial for drug delivery. Biomaterials. 2012;33(19):4889-906.
189. Varma MVS, Panchagnula R. Enhanced oral paclitaxel absorption with vitamin E-TPGS: effect on solubility and permeability in vitro, in situ and in vivo. European journal of pharmaceutical sciences. 2005;25(4-5):445-53.
190. Tao SL, Desai TA. Micromachined devices: the impact of controlled geometry from cell-targeting to bioavailability. J Control Release. 2005;109(1-3):127-38. Epub 2005/11/09. doi: 10.1016/j.jconrel.2005.09.019. PubMed PMID: 16274828.
191. Arzt E, Gorb S, Spolenak R. From micro to nano contacts in biological attachment devices. Proc Natl Acad Sci U S A. 2003;100(19):10603-6. Epub 2003/09/10. doi: 10.1073/pnas.1534701100. PubMed PMID: 12960386; PMCID: PMC196850.
192. Guan J, He H, Lee LJ, Hansford DJ. Fabrication of particulate reservoir-containing, capsulelike, and self-folding polymer microstructures for drug delivery. Small. 2007;3(3):412-8. Epub 2007/02/08. doi: 10.1002/smll.200600240. PubMed PMID: 17285662.
193. Henry S, McAllister DV, Allen MG, Prausnitz MR. Microfabricated microneedles: a novel approach to transdermal drug delivery. J Pharm Sci. 1998;87(8):922-5. Epub 1998/08/04. doi: 10.1021/js980042+. PubMed PMID: 9687334.
194. Kockisch S, Rees GD, Young SA, Tsibouklis J, Smart JD. Polymeric microspheres for drug delivery to the oral cavity: an in vitro evaluation of mucoadhesive potential. Journal of pharmaceutical sciences. 2003;92(8):1614-23.
195. Reddy PC, Chaitanya KSC, Rao YM. A review on bioadhesive buccal drug delivery systems: current status of formulation and evaluation methods. DARU Journal of Pharmaceutical Sciences. 2011;19(6):385.
196. Abramson A, Caffarel-Salvador E, Khang M, Dellal D, Silverstein D, Gao Y, Frederiksen MR, Vegge A, Hubalek F, Water JJ, Friderichsen AV, Fels J, Kirk RK, Cleveland C, Collins J, Tamang S, Hayward A, Landh T, Buckley ST, Roxhed N, Rahbek U, Langer R, Traverso G. An ingestible self-orienting system for oral delivery of macromolecules. Science. 2019;363(6427):611-5. Epub 2019/02/09. doi: 10.1126/science.aau2277. PubMed PMID: 30733413; PMCID: PMC6430586.
197. Jiang J, Xie J, Ma B, Bartlett DE, Xu A, Wang CH. Mussel-inspired protein-mediated

surface functionalization of electrospun nanofibers for pH-responsive drug delivery. Acta Biomater. 2014;10(3):1324-32. Epub 2013/11/30. doi: 10.1016/j.actbio.2013.11.012. PubMed PMID: 24287161; PMCID: PMC3946753.
198. Traverso G, Schoellhammer CM, Schroeder A, Maa R, Lauwers GY, Polat BE, Anderson DG, Blankschtein D, Langer R. Microneedles for drug delivery via the gastrointestinal tract. J Pharm Sci. 2015;104(2):362-7. Epub 2014/09/25. doi: 10.1002/jps.24182. PubMed PMID: 25250829; PMCID: PMC4349565.
199. Prausnitz MR, Gomaa Y, Li W. Microneedle patch drug delivery in the gut. Nat Med. 2019;25(10):1471-2. Epub 2019/10/09. doi: 10.1038/s41591-019-0606-0. PubMed PMID: 31591600.
200. https://www.fda.gov/media/107708/download.
201. Abramson A, Caffarel-Salvador E, Khang M, Dellal D, Silverstein D, Gao Y, Frederiksen MR, Vegge A, Hubálek F, Water JJ. An ingestible self-orienting system for oral delivery of macromolecules. Science. 2019;363(6427):611-5.
202. Zhou M, Hou T, Li J, Yu S, Xu Z, Yin M, Wang J, Wang X. Self-propelled and targeted drug delivery of poly (aspartic acid)/iron–zinc microrocket in the stomach. ACS nano. 2019;13(2):1324-32.
203. de Ávila BE-F, Angsantikul P, Li J, Lopez-Ramirez MA, Ramírez-Herrera DE, Thamphiwatana S, Chen C, Delezuk J, Samakapiruk R, Ramez V. Micromotor-enabled active drug delivery for in vivo treatment of stomach infection. Nature communications. 2017;8(1):1-9.
204. Nemeth CL, Lykins WR, Tran H, ElSayed MEH, Desai TA. Bottom-Up Fabrication of Multilayer Enteric Devices for the Oral Delivery of Peptides. Pharm Res. 2019;36(6):89. Epub 2019/04/21. doi: 10.1007/s11095-019-2613-3. PubMed PMID: 31004235.
205. Fox CB, Cao Y, Nemeth CL, Chirra HD, Chevalier RW, Xu AM, Melosh NA, Desai TA. Fabrication of Sealed Nanostraw Microdevices for Oral Drug Delivery. ACS Nano. 2016;10(6):5873-81. Epub 2016/06/03. doi: 10.1021/acsnano.6b00809. PubMed PMID: 27268699; PMCID: PMC5435488.
206. Abramson A, Caffarel-Salvador E, Soares V, Minahan D, Tian RY, Lu X, Dellal D, Gao Y, Kim S, Wainer J, Collins J, Tamang S, Hayward A, Yoshitake T, Lee HC, Fujimoto J, Fels J, Frederiksen MR, Rahbek U, Roxhed N, Langer R, Traverso G. A luminal unfolding microneedle injector for oral delivery of macromolecules. Nat Med. 2019;25(10):1512-8. Epub 2019/10/09. doi: 10.1038/s41591-019-0593-9. PubMed PMID: 31591601.
207. Niu M, Lu Y, Hovgaard L, Guan P, Tan Y, Lian R, Qi J, Wu W. Hypoglycemic activity and oral bioavailability of insulin-loaded liposomes containing bile salts in rats: the effect of cholate type, particle size and administered dose. Eur J Pharm Biopharm. 2012;81(2):265-72. Epub 2012/03/01. doi: 10.1016/j.ejpb.2012.02.009. PubMed PMID: 22369880.
208. Niu M, Lu Y, Hovgaard L, Wu W. Liposomes containing glycocholate as potential oral insulin delivery systems: preparation, in vitro characterization, and improved protection against enzymatic degradation. International journal of nanomedicine. 2011;6:1155.
209. Cui M, Wu W, Hovgaard L, Lu Y, Chen D, Qi J. Liposomes containing cholesterol analogues of botanical origin as drug delivery systems to enhance the oral absorption of insulin. International journal of pharmaceutics. 2015;489(1-2):277-84.
210. Lamson NG, Berger A, Fein KC, Whitehead KA. Anionic nanoparticles enable the oral delivery of proteins by enhancing intestinal permeability. Nat Biomed Eng. 2020;4(1):84-96. Epub 2019/11/07. doi: 10.1038/s41551-019-0465-5. PubMed PMID: 31686002.
211. Nielsen LH, Nagstrup J, Gordon S, Keller SS, Ostergaard J, Rades T, Mullertz A, Boisen

A. pH-triggered drug release from biodegradable microwells for oral drug delivery. Biomed Microdevices. 2015;17(3):9958. Epub 2015/05/20. doi: 10.1007/s10544-015-9958-5. PubMed PMID: 25981751.
212. Malachowski K, Breger J, Kwag HR, Wang MO, Fisher JP, Selaru FM, Gracias DH. Stimuli-responsive theragrippers for chemomechanical controlled release. Angew Chem Int Ed Engl. 2014;53(31):8045-9. Epub 2014/03/19. doi: 10.1002/anie.201311047. PubMed PMID: 24634136; PMCID: PMC4315180.
213. Banerjee A, Mitragotri S. Intestinal patch systems for oral drug delivery. Curr Opin Pharmacol. 2017;36:58-65. Epub 2017/08/30. doi: 10.1016/j.coph.2017.08.005. PubMed PMID: 28850875.
214. Kaur G, Arora M, Ravi Kumar MNV. Oral Drug Delivery Technologies-A Decade of Developments. J Pharmacol Exp Ther. 2019;370(3):529-43. Epub 2019/04/24. doi: 10.1124/jpet.118.255828. PubMed PMID: 31010845; PMCID: PMC6806634.
215. Cui F, He C, Yin L, Qian F, He M, Tang C, Yin C. Nanoparticles incorporated in bilaminated films: a smart drug delivery system for oral formulations. Biomacromolecules. 2007;8(9):2845-50. Epub 2007/08/02. doi: 10.1021/bm070339e. PubMed PMID: 17665945.
216. Banerjee A, Lee J, Mitragotri S. Intestinal mucoadhesive devices for oral delivery of insulin. Bioeng Transl Med. 2016;1(3):338-46. Epub 2016/08/19. doi: 10.1002/btm2.10015. PubMed PMID: 29313019; PMCID: PMC5689539.
217. Grabovac V, Foger F, Bernkop-Schnurch A. Design and in vivo evaluation of a patch delivery system for insulin based on thiolated polymers. Int J Pharm. 2008;348(1-2):169-74. Epub 2007/08/21. doi: 10.1016/j.ijpharm.2007.06.052. PubMed PMID: 17706903.
218. Eaimtrakarn S, Itoh Y, Kishimoto H, Yoshikawa Y, Shibata N, Takada K. Retention and transit of intestinal mucoadhesive films in rat small intestine. Int J Pharm. 2001;224(1-2):61-7. Epub 2001/07/27. doi: 10.1016/s0378-5173(01)00738-4. PubMed PMID: 11472815.
219. Banerjee A, Lee J, Mitragotri S. Intestinal mucoadhesive devices for oral delivery of insulin. Bioengineering & translational medicine. 2016;1(3):338-46.
220. Varshosaz J, Ahmadi F, Emami J, Tavakoli N, Minaiyan M, Mahzouni P, Dorkoosh F. Microencapsulation of budesonide with dextran by spray drying technique for colon-targeted delivery: an in vitro/in vivo evaluation in induced colitis in rat. J Microencapsul. 2011;28(1):62-73. Epub 2010/12/22. doi: 10.3109/02652048.2010.529947. PubMed PMID: 21171817.
221. Ravi V, Siddaramaiah, Pramod Kumar TM. Influence of natural polymer coating on novel colon targeting drug delivery system. J Mater Sci Mater Med. 2008;19(5):2131-6. Epub 2007/11/14. doi: 10.1007/s10856-007-3155-x. PubMed PMID: 17999035.
222. Wang X, Yu DG, Li XY, Bligh SW, Williams GR. Electrospun medicated shellac nanofibers for colon-targeted drug delivery. Int J Pharm. 2015;490(1-2):384-90. Epub 2015/06/06. doi: 10.1016/j.ijpharm.2015.05.077. PubMed PMID: 26043827.
223. Henning S, Leick S, Kott M, Rehage H, Suter D. Sealing liquid-filled pectinate capsules with a shellac coating. J Microencapsul. 2012;29(2):147-55. Epub 2011/12/01. doi: 10.3109/02652048.2011.635220. PubMed PMID: 22126315.
224. Wen P, Feng K, Yang H, Huang X, Zong M-H, Lou W-Y, Li N, Wu H. Electrospun core-shell structured nanofilm as a novel colon-specific delivery system for protein. Carbohydrate polymers. 2017;169:157-66.
225. Sun B, Ranganathan B, Feng SS. Multifunctional poly(D,L-lactide-co-glycolide)/montmorillonite (PLGA/MMT) nanoparticles decorated by Trastuzumab for targeted chemotherapy of breast cancer. Biomaterials. 2008;29(4):475-86. Epub 2007/10/24. doi:

10.1016/j.biomaterials.2007.09.038. PubMed PMID: 17953985.
226. Morishita M, Goto T, Nakamura K, Lowman AM, Takayama K, Peppas NA. Novel oral insulin delivery systems based on complexation polymer hydrogels: single and multiple administration studies in type 1 and 2 diabetic rats. J Control Release. 2006;110(3):587-94. Epub 2005/12/06. doi: 10.1016/j.jconrel.2005.10.029. PubMed PMID: 16325951; PMCID: PMC4467730.
227. Morishita M, Goto T, Peppas NA, Joseph JI, Torjman MC, Munsick C, Nakamura K, Yamagata T, Takayama K, Lowman AM. Mucosal insulin delivery systems based on complexation polymer hydrogels: effect of particle size on insulin enteral absorption. J Control Release. 2004;97(1):115-24. Epub 2004/05/19. doi: 10.1016/j.jconrel.2004.03.008. PubMed PMID: 15147809.
228. Ainslie KM, Lowe RD, Beaudette TT, Petty L, Bachelder EM, Desai TA. Microfabricated devices for enhanced bioadhesive drug delivery: attachment to and small-molecule release through a cell monolayer under flow. Small. 2009;5(24):2857-63. Epub 2009/09/30. doi: 10.1002/smll.200901254. PubMed PMID: 19787677.
229. Tao SL, Lubeley MW, Desai TA. Bioadhesive poly(methyl methacrylate) microdevices for controlled drug delivery. J Control Release. 2003;88(2):215-28. Epub 2003/03/12. doi: 10.1016/s0168-3659(03)00005-1. PubMed PMID: 12628320.
230. Tao SL, Lubeley MW, Desai TA. Synthesis of cytoadhesive poly(methylmethacrylate) for applications in targeted drug delivery. J Biomed Mater Res A. 2003;67(2):369-75. Epub 2003/11/19. doi: 10.1002/jbm.a.10047. PubMed PMID: 14619915.
231. Uskoković V, Lee PP, Walsh LA, Fischer KE, Desai TA. PEGylated silicon nanowire coated silica microparticles for drug delivery across intestinal epithelium. Biomaterials. 2012;33(5):1663-72.
232. Uskokovic V, Lee K, Lee PP, Fischer KE, Desai TA. Shape effect in the design of nanowire-coated microparticles as transepithelial drug delivery devices. ACS nano. 2012;6(9):7832-41.
233. Kam KR, Walsh LA, Bock SM, Koval M, Fischer KE, Ross RF, Desai TA. Nanostructure-mediated transport of biologics across epithelial tissue: enhancing permeability via nanotopography. Nano Lett. 2013;13(1):164-71. Epub 2012/11/29. doi: 10.1021/nl3037799. PubMed PMID: 23186530; PMCID: PMC4418930.
234. Levy ES, Samy KE, Lamson NG, Whitehead KA, Kroetz DL, Desai TA. Reversible inhibition of efflux transporters by hydrogel microdevices. Eur J Pharm Biopharm. 2019;145:76-84. Epub 2019/10/23. doi: 10.1016/j.ejpb.2019.10.007. PubMed PMID: 31639417; PMCID: PMC6919324.
235. Cencič A, Langerholc T. Functional cell models of the gut and their applications in food microbiology—a review. International journal of food microbiology. 2010;141:S4-S14.
236. Costa J, Ahluwalia A. Advances and Current Challenges in Intestinal in vitro Model Engineering: A Digest. Front Bioeng Biotechnol. 2019;7(144):144. Epub 2019/07/06. doi: 10.3389/fbioe.2019.00144. PubMed PMID: 31275931; PMCID: PMC6591368.
237. Edmondson R, Broglie JJ, Adcock AF, Yang L. Three-dimensional cell culture systems and their applications in drug discovery and cell-based biosensors. Assay and drug development technologies. 2014;12(4):207-18.
238. Abbasi F, Ghanian MH, Baharvand H, Vahidi B, Eslaminejad MB. Engineering mesenchymal stem cell spheroids by incorporation of mechanoregulator microparticles. J Mech Behav Biomed Mater. 2018;84:74-87. Epub 2018/05/12. doi: 10.1016/j.jmbbm.2018.04.026.

PubMed PMID: 29751274.
239. Bratt-Leal AM, Kepple KL, Carpenedo RL, Cooke MT, McDevitt TC. Magnetic manipulation and spatial patterning of multi-cellular stem cell aggregates. Integr Biol (Camb). 2011;3(12):1224-32. Epub 2011/11/15. doi: 10.1039/c1ib00064k. PubMed PMID: 22076329; PMCID: PMC4633527.
240. Cerchiari AE, Samy KE, Todhunter ME, Schlesinger E, Henise J, Rieken C, Gartner ZJ, Desai TA. Probing the luminal microenvironment of reconstituted epithelial microtissues. Sci Rep. 2016;6(1):33148. Epub 2016/09/14. doi: 10.1038/srep33148. PubMed PMID: 27619235; PMCID: PMC5020616.
241. Yajima Y, Yamada M, Utoh R, Seki M. Collagen microparticle-mediated 3D cell organization: a facile route to bottom-up engineering of thick and porous tissues. ACS Biomaterials Science & Engineering. 2017;3(9):2144-54.
242. Samy KE, Levy ES, Phong K, Demaree B, Abate AR, Desai TA. Human intestinal spheroids cultured using Sacrificial Micromolding as a model system for studying drug transport. Scientific reports. 2019;9(1):1-12.
243. Bhatia SN, Ingber DE. Microfluidic organs-on-chips. Nat Biotechnol. 2014;32(8):760-72. Epub 2014/08/06. doi: 10.1038/nbt.2989. PubMed PMID: 25093883.
244. Christenson C, Baryeh K, Ahadian S, Nasiri R, Dokmeci MR, Goudie M, Khademhosseini A, Ye JY, editors. Enhancement of label-free biosensing of cardiac troponin I. Label-free Biomedical Imaging and Sensing (LBIS) 2020; 2020: International Society for Optics and Photonics.
245. Kimura H, Yamamoto T, Sakai H, Sakai Y, Fujii T. An integrated microfluidic system for long-term perfusion culture and on-line monitoring of intestinal tissue models. Lab Chip. 2008;8(5):741-6. Epub 2008/04/25. doi: 10.1039/b717091b. PubMed PMID: 18432344.
246. Imura Y, Asano Y, Sato K, Yoshimura E. A microfluidic system to evaluate intestinal absorption. Anal Sci. 2009;25(12):1403-7. Epub 2009/12/17. doi: 10.2116/analsci.25.1403. PubMed PMID: 20009325.
247. Imura Y, Sato K, Yoshimura E. Micro total bioassay system for ingested substances: assessment of intestinal absorption, hepatic metabolism, and bioactivity. Anal Chem. 2010;82(24):9983-8. Epub 2010/11/26. doi: 10.1021/ac100806x. PubMed PMID: 21090751.
248. Bricks T, Paullier P, Legendre A, Fleury MJ, Zeller P, Merlier F, Anton PM, Leclerc E. Development of a new microfluidic platform integrating co-cultures of intestinal and liver cell lines. Toxicol In Vitro. 2014;28(5):885-95. Epub 2014/03/26. doi: 10.1016/j.tiv.2014.02.005. PubMed PMID: 24662032.
249. Tan H-Y, Trier S, Rahbek UL, Dufva M, Kutter JP, Andresen TL. A multi-chamber microfluidic intestinal barrier model using Caco-2 cells for drug transport studies. PLoS One. 2018;13(5).
250. Chen HJ, Miller P, Shuler ML. A pumpless body-on-a-chip model using a primary culture of human intestinal cells and a 3D culture of liver cells. Lab on a Chip. 2018;18(14):2036-46.
251. Costello CM, Phillipsen MB, Hartmanis LM, Kwasnica MA, Chen V, Hackam D, Chang MW, Bentley WE, March JC. Microscale Bioreactors for in situ characterization of GI epithelial cell physiology. Sci Rep. 2017;7(1):12515. Epub 2017/10/04. doi: 10.1038/s41598-017-12984-2. PubMed PMID: 28970586; PMCID: PMC5624909.
252. Shim KY, Lee D, Han J, Nguyen NT, Park S, Sung JH. Microfluidic gut-on-a-chip with three-dimensional villi structure. Biomed Microdevices. 2017;19(2):37. Epub 2017/04/30. doi: 10.1007/s10544-017-0179-y. PubMed PMID: 28451924.

253. Basson MD, Li GD, Hong F, Han O, Sumpio BE. Amplitude-dependent modulation of brush border enzymes and proliferation by cyclic strain in human intestinal Caco-2 monolayers. J Cell Physiol. 1996;168(2):476-88. Epub 1996/08/01. doi: 10.1002/(SICI)1097-4652(199608)168:2<476::AID-JCP26>3.0.CO;2-#. PubMed PMID: 8707883.
254. Samak G, Gangwar R, Crosby LM, Desai LP, Wilhelm K, Waters CM, Rao R. Cyclic stretch disrupts apical junctional complexes in Caco-2 cell monolayers by a JNK-2-, c-Src-, and MLCK-dependent mechanism. Am J Physiol Gastrointest Liver Physiol. 2014;306(11):G947-58. Epub 2014/04/12. doi: 10.1152/ajpgi.00396.2013. PubMed PMID: 24722904; PMCID: PMC4042113.
255. Kim HJ, Huh D, Hamilton G, Ingber DE. Human gut-on-a-chip inhabited by microbial flora that experiences intestinal peristalsis-like motions and flow. Lab on a Chip. 2012;12(12):2165-74.
256. Kim HJ, Li H, Collins JJ, Ingber DE. Contributions of microbiome and mechanical deformation to intestinal bacterial overgrowth and inflammation in a human gut-on-a-chip. Proc Natl Acad Sci U S A. 2016;113(1):E7-15. Epub 2015/12/17. doi: 10.1073/pnas.1522193112. PubMed PMID: 26668389; PMCID: PMC4711860.
257. Kasendra M, Tovaglieri A, Sontheimer-Phelps A, Jalili-Firoozinezhad S, Bein A, Chalkiadaki A, Scholl W, Zhang C, Rickner H, Richmond CA. Development of a primary human Small Intestine-on-a-Chip using biopsy-derived organoids. Scientific reports. 2018;8(1):1-14.
258. Ronaldson-Bouchard K, Vunjak-Novakovic G. Organs-on-a-Chip: A Fast Track for Engineered Human Tissues in Drug Development. Cell Stem Cell. 2018;22(3):310-24. Epub 2018/03/03. doi: 10.1016/j.stem.2018.02.011. PubMed PMID: 29499151; PMCID: PMC5837068.
259. Ragelle H, Danhier F, Préat V, Langer R, Anderson DG. Nanoparticle-based drug delivery systems: a commercial and regulatory outlook as the field matures. Expert opinion on drug delivery. 2017;14(7):851-64.
260. Ventola CL. Progress in nanomedicine: approved and investigational nanodrugs. Pharmacy and Therapeutics. 2017;42(12):742.
261. Durán-Lobato M, Niu Z, Alonso MJ. Oral delivery of biologics for precision medicine. Advanced Materials.1901935.
262. Zhong H, Chan G, Hu Y, Hu H, Ouyang D. A comprehensive map of FDA-approved pharmaceutical products. Pharmaceutics. 2018;10(4):263.
263. Lu Y, Chen SC. Micro and nano-fabrication of biodegradable polymers for drug delivery. Adv Drug Deliv Rev. 2004;56(11):1621-33. Epub 2004/09/08. doi: 10.1016/j.addr.2004.05.002. PubMed PMID: 15350292.
264. https://www.accessdata.fda.gov/drugsatfda_docs/nda/2019/206089Orig1s000TOC.cfm.
265. Geho WB, Geho HC, Lau JR, Gana TJ. Hepatic-directed vesicle insulin: a review of formulation development and preclinical evaluation. Journal of diabetes science and technology. 2009;3(6):1451-9.
266. Caster JM, Patel AN, Zhang T, Wang A. Investigational nanomedicines in 2016: a review of nanotherapeutics currently undergoing clinical trials. Wiley Interdisciplinary Reviews: Nanomedicine and Nanobiotechnology. 2017;9(1):e1416.
267. Mei L, Zhang Z, Zhao L, Huang L, Yang XL, Tang J, Feng SS. Pharmaceutical nanotechnology for oral delivery of anticancer drugs. Adv Drug Deliv Rev. 2013;65(6):880-90. Epub 2012/12/12. doi: 10.1016/j.addr.2012.11.005. PubMed PMID: 23220325.
268. Li H, Kochhar JS, Pan J, Chan SY, Kang L. Nano/microscale technologies for drug

delivery. Journal of Mechanics in Medicine Biology. 2011;11(02):337-67.
269. Kam KR, Desai TA. Nano- and microfabrication for overcoming drug delivery challenges. J Mater Chem B. 2013;1(14):1878-84. Epub 2013/06/05. doi: 10.1039/C3TB00048F. PubMed PMID: 23730504; PMCID: PMC3666043.
270. Vaut L, Juszczyk JJ, Kamguyan K, Jensen KE, Tosello G, Boisen A. 3D Printing of Reservoir Devices for Oral Drug Delivery: From Concept to Functionality through Design Improvement for Enhanced Mucoadhesion. ACS Biomaterials Science & Engineering. 2020;6(4):2478-86.
271. Walker DA, Hedrick JL, Mirkin CA. Rapid, large-volume, thermally controlled 3D printing using a mobile liquid interface. Science. 2019;366(6463):360-4.
272. Abramson A, Halperin F, Kim J, Traverso G. Quantifying the value of orally delivered biologic therapies: a cost-effectiveness analysis of oral semaglutide. Journal of pharmaceutical sciences. 2019;108(9):3138-45.
273. Gao J, Tian Z, Yang X. Breakthrough: Chloroquine phosphate has shown apparent efficacy in treatment of COVID-19 associated pneumonia in clinical studies. Bioscience trends. 2020.
274. Gautret P, Lagier J-C, Parola P, Meddeb L, Mailhe M, Doudier B, Courjon J, Giordanengo V, Vieira VE, Dupont HT. Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. International journal of antimicrobial agents. 2020:105949.
275. Grein J, Ohmagari N, Shin D, Diaz G, Asperges E, Castagna A, Feldt T, Green G, Green ML, Lescure F-X. Compassionate use of remdesivir for patients with severe Covid-19. New England Journal of Medicine. 2020;382(24):2327-36.

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