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Advances of 3D printing in gastroenterology and where it might be going

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Abstract

Gastrointestinal (GI) system comprises a great number of organs and tissues of various functions, both hollow and solid. However, it is still a less well-developed area for three-dimensional (3D) printing (3DP) applications compared to orthopedics. Clinical applications of 3DP in the GI system are presently restricted to preoperative planning, surgical guidance, and education for students, residents, and patients, either for laparoscopy or endoscopy. Several surgery-related accessories have been designed to facilitate surgical procedures. The results are promising but not adequately proven due to a lack of reasonable study design and proper comparisons. Other important requirements for GI systems in clinical scenarios are structural reconstruction, replacement, defect repair, drug screening, and delivery. Many 3D-printed decellularized, cell-seeded, or even bioprinted scaffolds have been studied; however, most studies were conducted on small animal or *in vitro* models. Although encouraging results have been obtained, there is still a long way to go before products compatible with humans in size, histology, and functions can be printed. The key points to achieving this goal are the printing material, cell type and source, and printing technology. The ultimate goal is to print tissue and organ substitutes with physiological functions for clinical purposes in both time- and cost-effective ways.

Keywords: 3D printing; Bioprinting; Gastroenterology; Surgery; Endoscopy

1. Introduction

Three-dimensional (3D) printing (3DP) has evolved since Bill Master introduced the concept in 1984[1], and it has been popular in various fields, such as industry, architecture, education, and medicine. In medical areas, 3DP is predominantly used in orthopedics, maxillofacial surgery, stomatology, and cranial and spinal surgery, where tissues and organs are solid and non-deformable on radiological images. Most articles recognized the positive role of 3DP in reducing operational time, decreasing radiation, and improving outcomes[2]. 3DP is also particularly useful in producing patient-specific implantable objects[3], and it is particularly true in bone grafts[4]. In comparison, the gastrointestinal system is less well-explored, perhaps due to there being multiple organs and tissues involved and their complex anatomical relationships. They are different from solid organs as they are soft and deformable, with peristaltic ability and complex interactions with host microbiomes.

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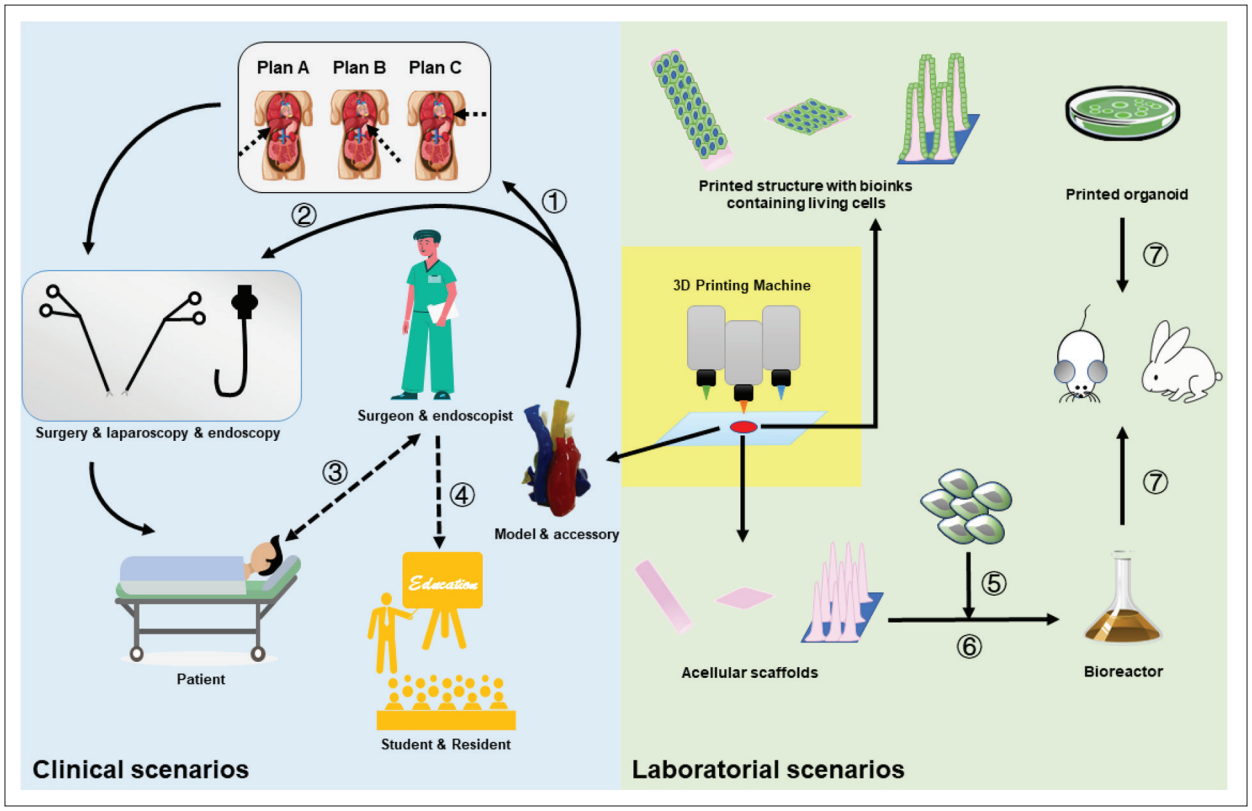

Figure 1. A schematic diagram of the applications of 3D printing and bioprinting in gastroenterology, both clinical and laboratorial. (1) Preoperative planning; (2) Accessory production for laparoscopy or endoscopy; (3) Communication and education for the patients; (4) Clinical education and skill training; (5) Cell seeding; (6) Maturation (cell expansion and differentiation); (7) Transplantation.

As gastrointestinal (GI) system is embodied in thoracic and abdominal cavities, different types of surgeries, such as laparotomy, laparoscopy, and gastrointestinal endoscopy, are performed. One unique characteristic of the alimentary tract is its abundant blood supply and variations of blood vessels[5]. It often requires the precise anatomy knowledge and spatial imagination of doctors to imaginatively reconstruct the 3D structures from two-dimensional (2D) images produced by computed tomography (CT) or magnetic resonance imaging (MRI) scans, which is not "real" enough. 3D printing can be seen as an extension of those medical images[6].

Patient-specific prototyping of anatomical structures accords with the growing popularity of precision medicine. 3DP models help clinicians, especially those with less experience, perceive a better understanding of anatomical structures before applying surgical or laparoscopic procedures[7,8]. Many researchers have tried 3DP gastrointestinal organs and tissues for various purposes, including preoperative planning, education, and implantable object production. However, their printing technologies vary greatly. There are approximately seven kinds of 3D printers, including vat photopolymerization, material jetting, binder jetting, material extrusion, powder bed fusion, sheet lamination, and direct energy deposition, which have been extensively summarized previously[9].

Unlike orthopedics, where 3DP and its productions are extensively studied both in laboratories and in patients, studies concerning gastroenterology are sparsely documented, and most of the state-of-the-art experiments are on rodents. In this review, we summarize what is going on with 3DP in gastrointestinal surgery, laparoscopy, and endoscopy from the perspectives of preoperative planning, education, and object production. Then, we discuss some progress of 3D bioprinting in this area and leave a short outlook at the end (Figure 1). 3D printing technologies and inks, which have been extensively discussed elsewhere[10-13], are not illustrated here.

2. 3D printing in gastrointestinal surgery

Gastrointestinal surgery is one of the most complex operations because it involves a wide range of organs and adjacent anatomic structures, such as the heart, lung, blood, and lymphatic vessels. At present, surgeons view and evaluate the operational area mainly through

tomographic images acquired by enhanced CT or MRI; or, better, through virtual 3D reconstruction by software. Compared to these imaging modalities, printed 3D models provide more detailed visual and tactile information and experience. Furthermore, surgeons could rehearse surgical procedures on the real model to select surgical devices, choose an optimized operative approach, and define other intraoperative matters that need attention.

2.1. Operative planning and guidance

Preoperative planning and intraoperative guidance are some of the most researched topics regarding the esophageal aspect. Most studies conclude that 3D models, especially of blood vessels, help identify anatomical variants, reduce operative time and blood loss, and optimize the operative approach. This is especially true when encountering complicated cases. In 2015, Dickinson *et al.* of Mayo Clinic reported two complex cases of aortoesophageal fistula complicated by previous cervical esophagostomy and thoracoplasty and multiple esophageal diverticula secondary to esophageal dysmotility[14]. The printed life-size models helped doctors examine anatomical structures closely and directly, providing valuable chances for operational rehearsal and multidisciplinary consultation. Hamada *et al.* also reported a case where a surgery for cT3N4M0 stage III esophageal cancer was complicated by major vascular malformation (double aortic arch)[15]. The authors firmly acknowledged the efficacy of the 3DP model for preoperative simulation. Much of the difficulties they faced during surgery were what they had expected when simulating on the model. A similar function was reported in a robotic surgery for guiding anti-reflux surgery complicated by tortuous thoracic aorta compression[16].

Regarding the colic aspect, Garcia-Granero *et al.* reported using 3DP models to facilitate the planning of laparoscopic right hemicolectomy and D3 lymphadenectomy[17]. The model contained only blood vessels because the lymph nodes are usually located around the gastrocolic trunk of Henle (GCTH), where its origin from the mesentery is often variable. No obvious intraoperative bleeding was recorded, and all D3 nodes were found negative. Hojo *et al.* also printed a 3D model to rehearse laparoscopic surgery of descending colon cancer complicated by a 69 mm abdominal aortic aneurysm[18]. The authors acknowledged the benefit of planning port sites and avoiding vascular injury and postoperative adhesion. For rectal surgery, Hamabe *et al.* printed 3D pelvic models for rectal cancer that could be sagittally cleaved for clearer inspection. However, the printing material was hard and not elastic, leading to less operational simulation value[19].

The 3DP model is particularly helpful for simulating solid organs, such as the liver, spleen, and pancreas, in addition to hollow organs, because of their non-deformable nature. Igami *et al.* reported that the 3DP liver model helped to plan a resection strategy for a small metastatic tumor that could be neither touched nor seen during laparoscopic surgery and increased the success rate of R0 resection[20]. The model could distinctly locate and describe tumors, bile ducts, and blood vessels to predict the functional status of the liver postresection. However, the printed model was whitish, so the vessels had to be dyed manually. Later, in 2017, Witowski *et al.* also reported a similar but much more cost-effective 3DP colorectal liver metastatic model[21]. The researchers used desktop-fused deposition modeling (FDM) printing technology, which is cheaper than jetting. However, this FDM printer could only print a small part of the whole model at a time, and assembling multiple parts was needed. Therefore, this printing strategy is semiautomated, time-consuming, and usually requires several printers, despite costing less than others. Similar successful outcomes were reported for retroperitoneal tumors[22] and for avoiding large-for-size syndrome in pediatric liver transplantation[23]. Interestingly, Villarreal *et al.* reported two cases in which 3DP models were used in the planning and practice of complex hepatic separation of conjoined twins[24]. In this case, the authors referred to similar 3DP models documented in previously published literature.

Fistulas are not uncommon in general surgery and abdominal diseases. They are sometimes complicated because the fistula may involve multiple organs or tissues, and the fistula canals may vary. Therefore, researchers used 3D printing as an isolation technique to illustrate the fistula's complete picture. Huang *et al.* employed 3D printing to construct a stent that could match well with the angled intestinal tract (105°) that connected the enterocutaneous fistula[25]. The stent significantly reduced enteric effluent loss, allowing enteric nutrition and future fistula resection; however, a small amount of effluent leakage still occurred after the postural shift, indicating a need for further improvement in printing precision or material. One of the most common non-traumatic causes for enteric fistula might be attributed to Crohn's disease. Guz *et al.* reported a case in which an MRI image-based 3DP model provided a direct rotatable view and tactility of the location and degree of Crohn's disease-related perianal fistula[26]. The authors argued that this technique could do more than merely surgical planning by possibly lowering interobserver bias in interpreting radiological information.

2.2. Education and research

Another major function of the 3DP model is facilitating activities in education for medical students, residents,

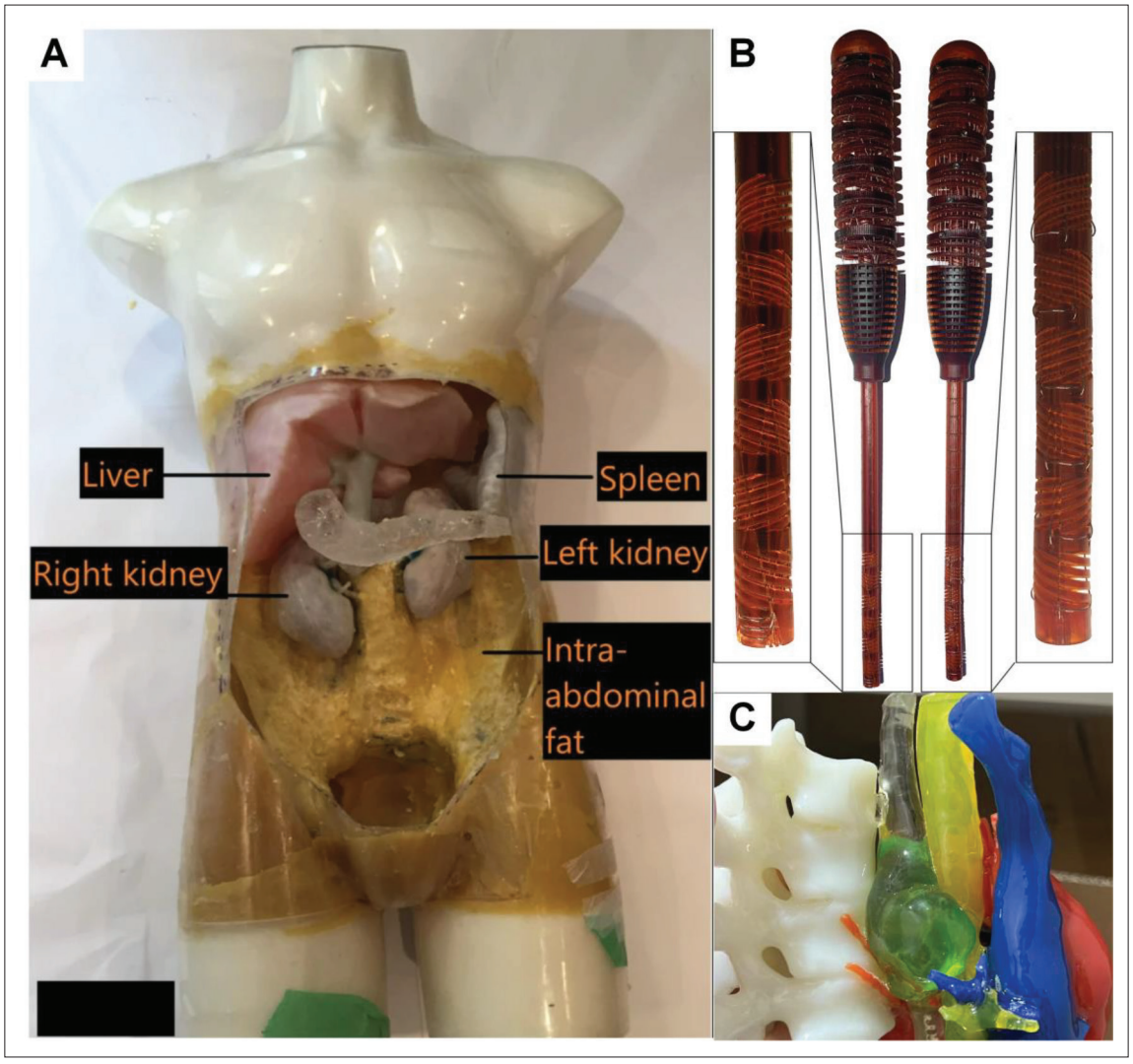

Figure 2. Illustrations of several applications of 3D-printed objects. (A) An abdominal cavity filled with 3D-printed organs that match the radiological density in real CT scan[35]; (B) 3D-printed steerable instruments that improve flexibility in laparoscopy[60]; (C) A 3D-printed model of esophageal submucosal tumor (green) to be resected using endoscopic submucosal dissection and its adjacent anatomy. Figure 2A–B are reprints of original images with permission (The images are licensed under Creative Commons Attribution 4.0 International License).

and patients, such as teaching, training, and counseling. For example, an anorectal fistula is intractable to assess because of its anatomical winding routes and complex connections. Most importantly, they are difficult to illustrate. Sahnan *et al.* used MRI images to construct a 3DP model of fistula canals and adjacent structures[27]. The authors believed that real 3D models improved surgeons' understanding of the complex anatomical relationship between the fistula and sphincter and provided better clinician–patient relationships and medical education. Hojo *et al.* of Tokyo University retrospectively created 3DP models of the superior mesenteric artery and superior mesenteric vein based on the surgeries carried out for laparoscopic right hemicolectomy with D3 lymphadenectomy in five patients[28]. Young surgeons could refer to the spatial relationships when reviewing operative videos of laparoscopic right hemicolectomy, which the author believed could shorten learning curves. Similar recognition of its usefulness in education and simulation has also been achieved among both experts and residents in bowel anastomosis[29], tracheoesophageal prosthesis placement[30], laparoscopic pyloromyotomy for neonates[31], laparoscopic preperitoneal inguinal hernia repair[32], laparoscopic bariatric surgery[33], and cystic duct variations in laparoscopic cholecystectomy[34].

For a purer educative and research purpose, Anwari *et al.* developed a 3DP anthropomorphic phantom based on CT images (Figure 2A)[35]. The phantom was created

organ-specifically and modularly. The highlight of the model is that different organs were printed and filled with different materials that mimicked radiological density in real CT scans, like water, liquid urethane rubber, agar-based solution, etc. The model could be scanned using CT, and the image was comparable to that of real human bodies. This model is particularly useful in anatomical and radiological courses for medical students for better correlation between anatomy and radiology. Similar demonstrations using 3D models for situs ambiguus of gastrointestinal organs[36], superior mesenteric artery plexus[37], and liver segments[38] were also documented.

The 3DP model is patient-friendly tool for communicating the medical-related information as well as facilitating the consent acquisition process. Almost all the cases reported above acknowledged that the printed model had positive effect on doctor–patient communication, patient's understanding of the operation's necessity, procedure, and expected difficulties or complications, and helped family members have realistic expectations. For postoperative care education, the model shows some benefits. For example, patient-specific postcolectomy stoma care training using a life-size printed model is helpful for patients to understand and reduce skin problems after self-practicing on the model[39].

The above studies are mainly presented as case reports, the conclusions of which were not validated[40]. A pilot study focusing on laparoscopic splenectomy enrolled 12 patients and 10 surgical residents, and both patients and residents highly confirmed the better illustrative and educative effects of 3DP models[41]. Luzon *et al.* included 23 patients and confirmed that the patient-specific 3DP model was useful in right colectomy and D3 mesenterectomy[42]. The accuracy of interanatomical structural distance measurement was validated by comparison with virtual 3D and intraoperative measurements. However, no clinical outcomes pertaining to surgery, e.g., time, blood loss, complications, hospital stay, cost, and prognosis, were included in these studies. In a prospective study, researchers from China found that 3DP models could help reduce the operation time (average 39.2 min), bleeding volume (average 45.1 ml), and medical expense (6.74%) while increasing rates of lymph node dissection (average 3.3) and patient satisfaction in right hemicolon cancer surgery[43]. Hojo *et al.* also reported a greater number of pelvic lymph nodes dissected for rectal cancer in the 3DP group, using a retrospective propensity score matching method[44].

In addition to education, there is also research concerning preoperative skill evaluation. Nishihara *et al.* previously constructed a 3DP model for the simulation of transabdominal preperitoneal (TAPP) inguinal hernia repair (IHR)[32]. Four years later, they used this TAPP model to evaluate surgeons' skills preoperatively and found that the model could distinguish surgeons with different levels of laparoscopic IHR experience[45]. A similar outcome of the laparoscopic choledochojejunostomy model was reported by Xia *et al.* based on the Objective Structured Assessment of Technical Skills (OSATS) scoring system[46]. Therefore, patient-specific 3DP models might be added to medical curricula for both education and evaluation. The 3DP model is more beneficial to less experienced residents and students rather than experts. The educative effect is particularly good among medical interns in understanding complex GI structures like Henle's trunk[47], practicing suturing[48], and perceiving oncopathological anatomy in different scenarios[49]. Quantitative analysis revealed that both surgeons and patients recognized 3DP models as useful, authentic, and favorable and scored median to high in preoperative planning, promoting learning, helping patients, and making effective and less conflicting decisions[7,50]. Surgical plans made upon 3DP models usually have a higher consistency with intraoperative findings[51].

The 3DP model is most useful in low-volume medical centers where rare and complex cases are hardly seen. While we have seen its many benefits, there remain problems to be solved: (i) Printing cost is high and time is long, and sometimes reprint is needed because of low quality[42]; (ii) Few comparisons are designed (traditional surgery vs. 3DP, 3DP vs. virtual reality); (iii) Evaluation criteria are mostly subjective; (iv) Sample size is too small to yield convincing result. A summary of the above-mentioned applications is listed in Table 1.

2.3. Accessory production

Apart from printing models for planning and education, printing-customized 3D tools might also be promising. Steinemann *et al.* tried to produce a space holder using 3DP to facilitate an intra-esophageal mucosal purse-string suture in Barrett's esophagus[52]. They found that the space holder helped resect more mucosa on sacrificed pigs but increased operational time and stitching variance. Another animal experiment of minipigs conducted by Yang *et al.* tested the usefulness of a 3DP biopolymer device in duct-to-mucosa pancreaticojejunostomy[53]. After 24 weeks of observation, they concluded that the device was promising for pancreatoenteric reconstruction with feasible procedural time and no adverse events. However, further controlled studies are warranted for validation. One special design might be the PLAFOKON operating platform, a flexible 3DP single-port overtube that was intervention-specific for individuals[54]. Other 3DP objects include spiral polymer stent for malignant esophageal stenosis[55], enteroatmospheric fistula tent[56,57], gastric

Table 1. 3D printing for surgical operation, education, and simulation

Author	Year	Image source	Data format	Image processing software	Output format	3D printing software	Printing machine
Igami *et al.*[20]	2014	CT/MRI	N/A	PLUTO	STL	PLUTO	AGILISTA3100
Dickinson *et al.*[14]	2015	CT	DICOM	Proprietary software	STL	Mimics software	Objet350 Connex
Pietrabissa *et al.* [41]	2016	CT	DICOM	ITK-SNAP	STL	—	Object 30 Pro 3D
Tominaga *et al.*[39]	2016	Artec 3D scanner	N/A	Geomagic FreeForm	STL	—	Objet260 Connex
Hamabe *et al.*[19]	2017	CT	DICOM	Zedview	N/A	Geomagic Freeform	Objet500 Connex3
Witowski *et al.*[21]	2017	CT	DICOM	Horos	STL	Meshmixer & Blender	Desktop Ultimaker 2+
Huang *et al.*[25]	2017	CT	N/A	N/A	STL	N/A	N/A
Nishihara *et al.*[32]	2017	CT	DICOM	N/A	N/A	N/A	Objet500 Connex3
Barber *et al.*[30]	2018	—	—	Fusion 360	N/A	—	Ultimaker 2+
Garcia-Granero *et al.*[17]	2018	CT	DICOM	Cella-supplied	STL	N/A	N/A
Sahnan *et al.*[27]	2018	MRI	DICOM	Open-source software	STL	Cura 3.0.4 & Ultimaker B.V.	Ultimaker 3 Extended
Luzon *et al.*[42]	2019	CT	DICOM	Osirix MD v. 8.5.2, Mimics Medical, 3-matic Medical	STL, MXP	PreForm	The Form1+
Hamada *et al.*[15]	2019	CT	N/A	N/A	N/A	N/A	Rais 3D N2 Plus
Marano *et al.*[16]	2019	CT	N/A	N/A	STL	N/A	N/A
Anwari *et al.*[35]	2020	CT	DICOM	Vitrea, v.6.9 & Slicer v4.7.0	STL	Blender v.2.78 & Cura v.15.04.5	Rostock Max V2
Chen *et al.*[47]	2020	CT	DICOM	—	N/A	Geomagic Studio 2014	N/A
Etherton *et al.*[36]	2020	CT/MRI	N/A	Analyze 12.0	STL	Autodesk Mesh mixer v.3.5.474	Raise3D N2 Plus & Ultimaker 2 Extended+
Sun *et al.*[22]	2020	CT	N/A	Mimics 16.0	N/A	Mimics 16.0	Stratasys C 350
Casas-Murillo *et al.*[34]	2021	MRI	DICOM	3D Slicer v.4.8.0	STL	Meshmixer v. 3.5.474	3D Zortrax
Guler *et al.*[49]	2021	CT/MRI	DICOM	3D Slicer v.4.10.1	STL	—	Mass Portal Pharaoh xd 20 & Form Labs2
Guz *et al.*[26]	2021	MRI	DICOM	3D Slicer v.4.8.0	STL	Blender 2.77a	3D ProJet 460Plus
Hojo *et al.*[28]	2021	CT	N/A	OsiriX MD	STL	Meshmixer v.3.5	Axiom Dual Extruder
Oxford *et al.*[29]	2021	—	—	Fusion 360	N/A	Cura	Ultimaker S3/S5
Luzon *et al.*[37]	2022	Nano-CT	DICOM	ITK-snap	STL	Ultimaker Cura v. 4.9.1	Ultimaker S3
Park *et al.*[23]	2022	CT	N/A	Mimics 21.0	N/A	Cinema 4D	Cubicreator & Cubicon Single Plus
Xia *et al.*[46]	2023	CT	DICOM	Mimics 23.0	STL	Magic24	N/A

Abbreviations: ABS, acrylonitrile butadiene styrene; CT, computed tomography; DICOM, digital imaging and communications in medicine; FDM, fused deposition modeling; GCTH, gastrocolic trunk of Henle; IHR, inguinal hernia repair; MRI, magnetic resonance imaging, PLA, polylactic acid; PU, polyurethane; PVA, polyvinyl alcohol; SLA, stereo lithography appearance; SMA, superior mesenteric artery; SMV, superior mesenteric vessel; STL, stereolithography; TAPP, transabdominal peritoneal; TEPP, tracheoesophageal puncture and prosthesis; TPU, thermoplastic polyurethane; —, not required; * refers to the percentage of lifesize organs.

Printing material	Printing technique	Model name	Model size*	Printing time	Printing cost
Rigid acrylic resin	N/A	Liver	70%	18 h–36 h	50,000–110,000 yen
Liquid photopolymer	Polyjet	Thorax	100%	N/A	N/A
Rigid photopolymer	Polyjet	Spleen	N/A	20 h	N/A
N/A	N/A	Stoma	N/A	Several days	100 USD
Ultraviolet-cured resin	N/A	Pelvic structure	N/A	34 h 20 min–37 h 30 min	250,000 Yen
PLA	FDM	Liver	N/A	72 h	150 USD
TPU	FDM	Fistula patch	100%	N/A	N/A
N/A	N/A	Laparoscopy simulator in TAPP IHR	N/A	N/A	N/A
PLA	N/A	TEPP placement simulator	N/A	N/A	N/A
ABS, PU rubber	N/A	GCTH	N/A	N/A	N/A
N/A	N/A	Perianal fistula	N/A	1 h	N/A
Resin-based polymers	SLA	SMV	100%	4 h–6 h	21–34 USD
N/A	N/A	Esophageal cancer, Double aortic arch	N/A	N/A	N/A
Photopolymer resin	SLA	Esophagus, proximal stomach, thoracic aorta, diaphragmatic crus	100%	48 h	23,000 euros
ABS, flexible urethane rubber, beeswax	FDM	Abdominal structure mannequin	100%	N/A	900 CAD
TPU, resin, silica gel	N/A	Gastrocolic trunk	N/A	N/A	N/A
PLA, TPU	FDM	Situs ambiguus	N/A	497 h	130 USD
N/A	N/A	Retroperitoneal tumor	N/A	18 h	N/A
ABS, P53 silicone rubber, and Elmer's slime	FDM	Laparoscopic simulator for cystic duct and its variants	N/A	N/A	0.9–11.7 USD
N/A	N/A	Cancer model	100%	15 h	N/A
VisiJet PXL Core	N/A	Fistula of Crohn's disease	N/A	6 h	N/A
PLA, TPU	FDM	SMV	100%	8 h–20 h	10 USD
N/A	N/A	Intestinal anastomosis simulator	N/A	N/A	2.67–131 USD
PLA, PVA	N/A	SMA plexus	50%	24 h–32 h	N/A
N/A	FDM	Abdominal cavity	N/A	9 h 36 min	1.6 USD
Silica gel	FDM	Choledochojejunostomy model	N/A	N/A	N/A

Table 2. 3D printing for endoscopic operation, education, and simulation

Author	Year	Application	Image source	Data format	Image processing software	Output format	3D printing software	Printing machine
Lee *et al.*[65]	2018	Endoscopic biopsy	CT	DICOM	3D Slicer v.4.5.0	STL	Netfabb professional v.5	Clone S270 & Clone K300
Yang *et al.*[61]	2018	ERCP	CT/MRI	DICOM	Mimics Innovation Suite v17.0	STL	N/A	ProJet 4500
Lee *et al.*[64]	2019	Endoscopic hemostasis	CT	DICOM	3D Slicer v.4.5.0	STL	Netfabb professional v.5	Form 2
Kwon *et al.*[67]	2020	ERCP	CT	N/A	In-house software	STL	MeshLab and MeshMixer	3DM Tough-3.6
Dhir *et al.*[109]	2015	EUS-guided biliary drainage	MRI	N/A	N/A	N/A	N/A	Viper SI2
Holt *et al.*[68]	2018	Endoscopic ampullectomy	N/A	N/A	N/A	N/A	Solid Works 2014	Connex 260v

Abbreviations: CT, computed tomography; DICOM, digital imaging and communications in medicine; ERCP, endoscopic retrograde cholangiopancreatography; FDM, fused deposition modeling; MRI, magnetic resonance imaging; PLC, polycarbonate; SLA, stereo lithography appearance; STL, stereolithography; *refers to the percentage of lifesize organs.

phantoms of different phases[58], device for sutureless end enterostomy[59], and multisteerable configurations for laparoscopy (Figure 2B)[60]. These are listed in Table 3.

3. 3D printing in gastrointestinal endoscopy

Similar to open surgery or laparoscopic surgery, 3D printing also facilitates GI endoscopy in preoperative planning and guidance (Table 2), education (Table 2), and accessory production (Table 3).

3.1. Preoperative planning and guidance

Yang *et al.* reported that the 3DP model could accurately display hilar cholangiocarcinoma (HCC) and its relationship with the surrounding bile duct, which could be used to guide endoscopic retrograde cholangiopancreatography (ERCP) in HCC patients and improve the success rate[61]. Recently, our team updated the concept of endoscopic submucosal dissection (ESD) based on 3DP (Figure 2C). A large esophageal submucosal tumor was successfully removed by endoscopy under the guidance of the 3DP model, which can distinctly display the tumor anatomy and details of important adjacent organs such as the bronchus, aorta, and spine[62].

3.2. Education

Endoscopy is an important tool for the diagnosis and treatment of digestive diseases, but it is a challenging technique that requires extensive training. Traditionally, novices learn basic endoscopic skills under the guidance of experienced endoscopists. Although this provides direct supervision and real-time evaluation by instructors, it may inadvertently lead to patient discomfort, prolonged operation time, and increased training costs. Experienced endoscopists usually have higher overall resection rates, shorter operative times, and fewer adverse events[63]. Adequate training models of different GI disease scenarios, which are often difficult in real clinical settings, are needed to train less-experienced learners. Therefore, simulator-based endoscopy training has been widely used and validated over the past few decades.

Lee *et al.* created a new 3DP stomach hemostatic simulator with two hemostasis modules for hemoclipping and injection, which can effectively train beginners in GI hemostatic skills[64]. They also created a 3DP gastric biopsy simulator that could improve biopsy skills[65]. For advanced endoscopic techniques such as endoscopic retrograde cholangiopancreatography (ERCP), anatomical differences, realistic sense, and durability are major concerns for simulation[66]. Kwon *et al.* optimized the ERCP model using the 3DP technique, which helped learners successfully and repeatedly complete basic biliary intubation, difficult intubation, stone extraction, mechanical lithotomy, stent implantation, and balloon dilation[67]. Should ERCP fail, a printed biliary duct prototype could also be used for training and practicing endoscopic ultrasound (EUS)-guided biliary drainage. Endoscopic ampullectomy is another technically challenging procedure. The effect of a 3DP endoscopic ampullectomy training model has been preliminarily confirmed, which may solve the problem of limited training opportunities[68].

3.3. 3D-printed endoscopic accessories

Similar to surgical applications, 3DP is equally useful in producing endoscopic accessories. Zizer *et al.* developed a 3DP overtube system and confirmed its efficacy in

Printing material	Printing technique	Model name	Model size*	Printing time	Printing cost
Platinum silicone rubber	FDM	Stomach	N/A	N/A	N/A
Visijet C4 Spectrum Core	N/A	Hilar cholangiocarcinoma and bile duct	N/A	N/A	N/A
Silicone	SLA	Stomach	N/A	N/A	N/A
Silicone	N/A	Stomach and duodenum	N/A	N/A	N/A
PLC	SLA	Bile duct	N/A	N/A	N/A
Silicone rubber, polymer resin	Polyjet	Stomach, duodenal ampulla	N/A	N/A	1482 USD

accelerating endoscopic submucosal dissection (ESD) progress in a porcine model[69]. To improve diagnostic and therapeutic effects, Ko *et al.* fabricated four types of tailored endoscopic caps for ESD, endoscopic mucosal resection (EMR), peroral endoscopic myotomy (POEM), and Trucut biopsy, and applied them in 39 patients[70]. To improve the adenoma detection rate, a sideoptic-enhanced cap was printed[71]. There are also 3DP versatile pedal fixators to improve ergonomics during endoscopic procedures[72] and devices to ease endoscopic cell sheet transplantation[73].

4. Scaffold production

A few attempts have been made on animals for GI tract reconstruction. In 2015, researchers covered artificial esophageal defects in rabbits with poly-ε-caprolactone (PCL) mesh[74]. Although the growth of smooth muscle and epithelial cells was observed, a relatively high proportion (9/15) of rabbits developed diverticula due to the fast degradation of the material. Later in 2016, Park *et al.* from South Korea refined the technique by coating printed PCL scaffolds with fibrin, thrombin, and rabbit mesenchymal stem/stromal cells (rMSCs). They found that the scaffold endured mechanical strength after implantation into rabbit esophageal defects without leakage and that the MSC-seeded scaffold had a complete covering with epithelial cells, while the nude scaffold did not[75]. For better structural simulation, a circumferentially printed acellular PCL model with improved strength was cultured in rat omentum and transplanted to repair this rat's esophageal transection after cellularization and vascularization[76]. However, the planted graft lacked peristalsis and was easily obstructed due to its small diameter. Kim *et al.* later compared this omentum bioreactor with a mesenchymal stem cell-based bioreactor, where a two-layered printed artificial esophageal scaffold was incubated[77]. They found that both bioreactors enabled over 80% mucosal regeneration in rat esophageal defects. It also seemed that different printing materials have different cellular activities. Park *et al.* revealed that in a rat esophageal defect model, both adipose-derived mesenchymal stem cell (ADSC)-seeded 3DP PCL and ADSC-seeded 3DP polyurethane-nanofiber (PU-Nf) had greater tissue regeneration than nude scaffold groups[78]. Interestingly, smooth muscle regeneration was greater in the PCL scaffold, while epithelium regeneration was greater in the PU-Nf scaffold. Perhaps cocktail formulations of inks might be considered for the expansion of different cells.

3DP scaffolds can also be used as loading systems for extracellular matrices and drugs. Ha *et al.* used 3DP to load esophagus-derived decellularized extracellular matrix (EdECM) hydrogel onto printed rod-shaped PCL stent[79]. They tested the stent in a radiation esophagitis rat model and observed fast remission of inflammation. Later in 2021, Kim *et al.* loaded tetracycline onto a 3DP PCL patch and implanted the patch into an artificial esophageal fistula in rats[80]. The patch could continuously release drugs for over 30 days and had good sealing, antibacterial, antimacrophage, and proregenerative effects. A similar printed stent loaded with 5-fluorouracil was applied in malignant esophageal stenosis[81]. In spite of these applications, 3DP scaffolds of intestinal microvilli and crypts with either hydrogels or silk fibroin protein can serve as models that better mimic physiological and barrier functions, which might be exploited to explore

Table 3. 3D printing of accessories

Author	Year	Application	Object name	Purpose	3D printing software	Printing machine	Printing technique	Printing material	Printing time	Printing cost
Maeda *et al.*[73]	2015	Endoscopy	Cell sheet carrier	Endoscopic cell sheet transplantation	N/A	Objet350 Connex	N/A	MED610	N/A	N/A
Ko *et al.*[70]	2016	Endoscopy	Tailored endoscopic caps	Facilitating EMRC, ESD, POEM, and TCB	N/A	Objet260 Connex	FDM & Polyjet	Silicon	N/A	N/A
Walter *et al.*[71]	2017	Endoscopy	Sideoptic caps	Colonoscopic examination	N/A	N/A	N/A	PLA	N/A	1 euro
Steinemann *et al.*[52]	2018	Laparoscopy	Space holder	Intraesophageal suturing	Autodesk Inventor 2016 & Autodesk Meshmixer v.10.10.170	Ultimaker B.V.	FFF	PLA	5 h	5 euros
Lin *et al.*[55]	2019	Endoscopy	Spiral polymer stent	Inoperative esophageal malignancy	CAD software	Ultimaker 2	N/A	TPU/PLA	N/A	N/A
Xu *et al.*[56]	2019	Surgery	Fistula stent	Plugging enteroatmospheric fistula	Solid work software	N/A	FDM	TPU	N/A	N/A
Yang *et al.*[53]	2019	Surgery	Biopolymer device	Simplify pancreaticojejunostomy	N/A	Fused Deposition Modeling	FDM	PLA	N/A	N/A
Durán Muñoz-Cruzado *et al.*[57]	2020	Surgery	Custom device	NPWT	Regemat 3D designer	Regemat 3D	N/A	PCL	4 h	N/A
Kwon *et al.*[58]	2020	Research	Anthropomorphic gastric phantom	Facilitate intragastric balloon investigation	3-matic 9.0	Objet500 Connex3	N/A	Agilus Transparent	N/A	N/A
Sejor *et al.*[59]	2020	Surgery	Custom device	Simplify enterostomy	Rhinoceros 6	Factory 2.0 & Raise 3D N2 Dual Plus	N/A	N/A	N/A	N/A
Culmone *et al.*[60]	2022	Laparoscopy	Multisteerable instruments	Reduce rigidity of surgical instruments	N/A	Perfactory Mini XL	Vat photopolymerization	Epoxy photopolymer resin	N/A	N/A
Bernhard *et al.*[54]	2022	Surgery	PLAFOKON	Specific robotic surgical platform	PLAFOKON planning tool	N/A	SLS	Polyamide 12	A few hours	N/A

Abbreviations: CAD, computer-aided; EMRC, endoscopic mucosal resection; ESD, endoscopic submucosal dissection; FDM, fused deposition modeling; FFF, fused filament fabrication; NPWT, negative pressure wound therapy; PCL, polycaprolactone; PLA, polylactic acid; POEM, peroral endoscopic myotomy; SLS, selective laser sintering; TCB, Trucut biopsy; TPU, thermoplastic polyurethane.

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disease pathogenesis, drug screening, and microbiome interaction[82,83]. The related literature is summarized in Table 4.

While the advantages of printed scaffolds are unanimously recognized (e.g., highly customized and controllable, compatible with many types of materials, and producing delicate micro structures)[84], there are still disadvantages (e.g., expensive equipment, time-consuming, low printing resolution, and requirement of bioreactor). The ideal printed scaffolds should have tubular structures with contiguous epithelial linings of different functions (e.g., acid and mucus secretion, absorption) and properties of nutrient provision, peristaltic pumping, and the microbiome. PCL is one of the most commonly used printing materials. However, it is not very friendly to cell adhesion, although it is reported to have fair biocompatibility, durability, processability, and relatively slow degradability[79,85]. Further refinement of both inks and experimental steps is required to meet experimental and clinical requirements.

5. What can 3D bioprinting do?

The above-mentioned printed scaffolds are primarily biomimetic structures without cells. Thus, bioprinting, where bioinks containing living cells, is later introduced (Table 5). There are generally three kinds of bioprinting technologies at the micrometric scale[10]: extrusion-based[86], jetting-based (inkjet and laser-assisted)[87], and vat photopolymerization (stereolithography and digital light processing)[88]. It is faster and more efficient than traditional methods as it excludes cell seeding and repopulation processes. Most importantly, 3D bioprinting is able to create grafts with spatial relocations of bioinks with living cells and with microenvironments for cell expansion. Therefore, 3D bioprinting is most suitable for stratified organs with different layers of cells like GI tract. The treatment of GI diseases requiring surgical interventions usually involves organ reconstruction, defect repair, and stenosis treatment. The success rate of intestinal allografts is still relatively low due to their immunogenicity. Therefore, implantable organs and materials that do not lead to immunological rejection, coagulopathy, pathogen transmission, and hazardous decomposition byproduct are needed. Functional tubular organs need a special design to mimic histological layers and physiological functions. 3D bioprinting has been attempted in creating hollow organs such as the esophagus, small intestine, and bile duct, but not yet in the stomach.

In 2019, Takeoka *et al.* bioprinted scaffold-free 3D tubular structures to repair rat esophageal defects. They found that a greater proportion of mesenchymal stem cells tended to induce greater strength and cellular activity[89]. Madden *et al.* used bioinks of human intestinal myofibroblasts (hIMFs) and human intestinal epithelial cells (hIECs) to print layer by layer onto the transwell membrane to form a bilayer structure. Differentiation into polarized and tightly joined epithelial subpopulations such as chromaffin cells and goblet cells was observed[90]. However, the researchers used this model to test drugs only rather than mechanical properties, peristaltic characteristics, and biocompatibilities. Further efforts can be made to develop implantable sheets and even hollow intestinal sections. Maina *et al.* 3D bioprinted a biopatch consisting of hydrogel, rat venous smooth muscle cells, and aortic fibroblast cells[91]. They implanted this patch into a rat enterostomy and found that the sealed intestine maintained integrity with the intraluminal pulsatile flow and exhibited robust histological formation of villi and crypts. To better mimic the histological architecture of villi, Kim *et al.* fabricated a collagen bioink-based intestinal model in which a single villus was 183 μm wide and 770 μm tall[92]. The model also contained vascular structure, making it perform better in cell growth, mucus secretion, barrier formation, and even absorption function than the 2D model and 3D model without vasculature. Kim further improved the bioink by adding decellularized small intestinal submucosa[93]. They demonstrated that the updated version had a better performance in cell activities than the previously reported version[92,93]. Very few studies have explored its application in the biliary system. Yan *et al.* printed models with ink containing cholangiocytes and laminin-like amphiphiles that comprise the base membrane. They found that the cells could organize and develop tubular structures with branches[94]. Boyer *et al.* also invented a 3D bioprinted biliary stent infused with collagen, human placental MSCs, and cholangiocytes, aiming to improve biliary stent patency and patient care[95].

Instead of tubular structures, 3D bioprinting of liver organoids has also been attempted in recent years. Yang *et al.* introduced a printed hepatorganoid that consisted of HepaRG cells and bioinks of sodium alginate and gelatin[96]. The organoid obtained functions of drug metabolism, synthesis of protein, and glycogen storage after proper culture both *in vitro* and *in vivo*. The planted organoid significantly prolonged the survival of liver failure mice.

The challenges for 3D bioprinting are finding the optimal formulation of biomaterials with cell components that meet the requirements of bioprinting, especially for hollow organs. Caution should be taken regarding questions about personalized ink formulation (i.e., biological composition, viscosity, mechanical properties, postprocessing gelatin, and clinical grade), nozzle clotting, cell damage, and prototype sterilization[97]. Hydrogels, whether natural or

Table 4. 3D printing of decellularized scaffolds

Author	Year	Animal model	Printed object	Application	Printing machine	Printing material
Park *et al.*[75]	2016	Rabbit	Artificial esophageal patch	Repairment of partial esophageal defect	3D Bioplotter	PCL
Chung *et al.*[76]	2018	Rat	Tubular scaffold	Repairment of transectional esophageal defect	BT-3000	PCL
Kim *et al.*[77]	2019	Rat	Esophageal graft	Repairment of transectional esophageal defect	3D Bioplotter	PCL/PU
Boyer *et al.*[95]	2019	*In vitro*	Biliary stent	Biliary procedures	MakerBot Replicator	PVA
Fouladian *et al.*[81]	2020	*In vitro*	Esophageal stent	Malignant esophageal stenosis	Ultimaker S5	PU+5-FU
Ha *et al.*[79]	2021	Rat	Esophageal stent	Treating radiation esophagitis	2RPS	PCL
Kim *et al.*[80]	2021	Rat	Artificial esophageal patch	Repairment of partial esophageal defect	Simplify 3D v. 4.0	PCL+TCN
Park *et al.*[78]	2021	Rat	Artificial esophageal patch	Repairment of partial esophageal defect	3D Bioplotter	PCL/PU

Abbreviations: ADSC, adipose-derived mesenchymal stem cell; EdECM, esophagus-derived decellularized extracellular matrix; FDM, fused deposition modeling; 5-FU, 5-fluorouracil; MSCs, mesenchymal stem cells; PMSCs, placental mesenchymal stem cells; PCL, polycaprolactone; PU, polyurethane; PVA, polyvinyl alcohol; TCN, tetracycline.

synthetic, are promising materials due to their biochemical ability to promote cellular activity. Therefore, hydrogels are often the first option in 3D bioprinting. However, it is weak in terms of mechanical properties, so it is still not the ideal option[10]. An ideal bioink should be one that has slow degradation and could be replaced by regenerative tissues. Yeleswarapu *et al.* solved this problem by innovatively using a stereolithography-based 3D printer[98]. They used esophageal muscle dECM to fabricate tubular structures, which sustained good biocompatibility and mechanical strength. From reported cases, we can see that bioinks derived from decellularized ECM seem to have better cellular activity. Furthermore, light-activated bioprinting materials might be a good choice to avoid thermal or cryogenic injury to cells in bioinks[99]. To refine printing techniques, Nam *et al.* developed an extrusion-based printing technique named "dragging technique" to fabricate a multilayered tubular scaffold with delicate pore characteristics, which previous techniques could not[100]. Pi *et al.* presented, in another way, a microfluidic bioprinting technology called a multichannel coaxial extrusion system (MCCES)[101]. The system could print circumferentially multilayered tubular structures, which were perfusable, with adequate cellular functionality in a single step.

The previously mentioned models were printed *in vitro*. Zhao *et al.* innovatively proposed a concept of *in situ* bioprinting *in vivo*, and brought this into reality by installing a micro bioprinter to the endoscope[102]. A printed circuit microelectromechanical system (PC-MEMS) technique was used to build the printing platform. They tested it by bioprinting a gelatin–alginate scaffold with human gastric epithelial and smooth muscle cells to repair a wound on a stomach model. Recently, Thai *et al.* also reported an *in situ* 3DP technique compatible with robotic surgery and tested it on colon phantoms and fresh porcine tissues[103]. However, it should be emphasized that these are not truly *in vivo*. In the future, live animal models are needed to simulate a real endoscopic procedure.

6. Outlook for 3D printing in gastroenterology

While we have seen encouraging reports of 3DP applications in gastroenterology, much is left unclarified and unsolved. Using traditional 3D printing, creating a 1:1 duplicate of anatomical structures in surgical areas instead of virtual ones that cannot be touched does lead to a seemingly better clinical outcome. However, most of the studies are presented as cases or case series of small samples. Few comparative studies have provided low-grade evidence about the effect of the 3DP model in preoperative planning and education. Many of the endpoints cannot be objectively evaluated, leaving suspicion about their credibility, even though comparisons have been made. Therefore, in future clinical studies assessing the applicability of 3DP models in helping surgery and education, several factors need to be addressed: (i) the studies need to be designed in a prospective manner with proper controls, either as randomized controlled trials (RCTs) or as cohort studies; (ii) validated endpoint events

Printing technique	Seeded cells	Extracellular matrix	Bioreactor	Results
Extrusion	Rabbit MSCs	Fibrin, thrombin	None	Better cell regeneration in MSC group
3D printing & electrospinning	None	None	Omentum	Better cell regeneration in MSC group
3D printing & electrospinning	Human MSCs	None	Custom-made & omentum	Satisfactory tissue regeneration with both bioreactors
N/A	Human PMSCs, human primary cholangiocytes	Collagen	Growth medium	Satisfactory cholangiocytes coating
FDM	None	None	None	Sustained release of 5-FU over 110 days
Extrusion	None	EdECM-based hydrogel	None	Rapid resolution of inflammatory response
Extrusion	None	None	None	Better tissue regeneration and antibacterial activity
3D printing & electrospinning	ADSC	Matrigel & fibronectin	Growth medium	Better cell regeneration in ADSC group

that make a subjective evaluation of improvement of skills and patient satisfaction more credible (e.g., structured scoring systems); and (iii) an adequate sample size that meets statistical principles.

When it comes to 3D printing, several limitations, such as high expenses, long printing time, change in size, and low printing resolution, hinder its widespread patient-specific application. Printing technology and materials need further refinement to achieve time-effective and cost-effective results while producing high-resolution[104], durable, and biocompatible models and objects. Implantable objects also have to endure sterile procedures and challenging physical or chemical environments *in vivo*. Researchers may also consider printing GI models with lifelike textures and histological layers (e.g., mucosa and submucosa) to provide better simulative effects. Another question for models concerning surgery and patient education is that who should cover the printing cost.

To better meet the clinical demands of organ replacement, reconstruction, and repair, either cell-seeded scaffolds or bioprinted scaffolds have to acquire physiological properties such as secretion, absorption, and peristalsis that resemble native tissues. While researchers have realized some of those properties, such as in the regenerations of multilayer epithelium and smooth muscle, they were mostly performed on mice, rats, or rabbits. Such experiments have not been conducted in larger mammals. Whether the scaffolds can be immediately transplanted or they should be left in a bioreactor after bioprinting remains to be explored. We expect the cells to self-assemble to form a native histological structure. Should 3D bioprinted grafts be applied to the human gastroenterological system, several questions must be answered first: (i) What kind of cells are needed and where do we get them? (ii) What kind of bioink best stimulates cell growth and differentiation? (iii) Is the bioink formulation a panacea or tissue-specific? (iv) Does the printing technology and material support a 1:1 duplicate of native human organ or tissue with mechanical, microbiological, immunological, and neurological functions as well as microenvironments of blood and lymphatic vessels, and how fast can it be? (v) Is 3D-printed organoid transplantation an alternative to organ or tissue transplantation, and for what kind of scenarios might it be suitable?

In the end, machine learning (ML) has been popular in the last decades, and several attempts have been made in process optimization, defect detection, dimensional accuracy analysis, bioink design, and cellular viability prediction[105-108]. While many challenges remain, how artificial intelligence might be integrated into tissue design, bioink formulation, cell sorting and culture, printing, and monitoring in gastroenterology is still an interesting task in the future.

7. Conclusion

Although much seems to have been tried, gastroenterology is still a less developed area for 3D printing and bioprinting. However, it is promising for vast clinical requirements. Preoperative planning, realistic simulation, evaluation

Table 5. 3D bioprinting of gastrointestinal grafts

Author	Year	Animal model	Printed object	Application	Printing machine	Bioink formulation	Printing technique	Bioreactor	Results
Kim *et al.*[92]	2018	*In vitro*	Small intestinal villi model with vessels	Mimicking human intestine	DTR3-2210 T-SG	HUVECs, Caco-2 cells + collagen	Extrusion	Growth medium	Successful mimicking of 3D geometry and physiology of human intestine
Madden *et al.*[90]	2018	*In vitro*	Human intestinal tissue model	Drug development	Novogen Bioprinter	IMF, Caco-2 cells + Novogel	N/A	Custom-made	Mimic injury response to drug-induced toxicity and inflammation
Yan *et al.*[94]	2018	*In vitro*	Nanostructural scaffold	Promoting bile duct formation	Envision TEC (GMBH) 3D Bioplotter	Cholangiocytes + thiolated gelatin, PA	Extrusion	Growth medium	Formation of functional tubular structure
Takeoka *et al.*[89]	2019	Rat	Tubular structure	Regeneration of esophagus	Regenova bio-3D printer	Cellular spheroids of NHDFs, HUVECs, human bone marrow-derived MSCs, HESMCs	Kenzan method	Custom-made	Greater proportion of MSCs with greater mechanical strength and tissue regeneration
Kim *et al.*[93]	2020	*In vitro*	Small intestinal villi model	Mimicking human intestine	DTR3-2210 T-SG	Caco-2 cells + SIS, collagen	Extrusion	Growth medium	Better epithelial mimicking than pure collagen
Maina *et al.*[91]	2021	Rat	Biopatch	Repairment of small intestinal injury	Novogen mmx-07	Venous SMC, aortic FC + hydrogel	Inkjet	Growth medium	Villi and crypt formation, and complete restoration of epithelium
Yang *et al.*[96]	2019	Mice	Hepatorganoid	Treating liver failure	Spp1603	HepaRG cells + alginate, gelatin	Extrusion	Growth medium	Display synthetic functions and prolong liver injury mice survival

Abbreviations: FC, fibroblast; HESMCs, human esophagus smooth muscle cells; HUVECs, human umbilical vein endothelial cells; MSCs, mesenchymal stem cells; NHDFs, normal human dermal fibroblasts; PA, peptides amphiphile; SIS, small intestinal mucosa; SMC, smooth muscle cell.

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of therapy response, and organ/tissue replacement for precision medicine are what we are now endeavoring to explore. More importantly, we need to discover areas that we have not tried with 3D printing, where it might boost medical progress and bring real benefits to patients.

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Conflict of interest

The authors declare no conflict of interest.

Author contributions

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Writing – original draft: Yuhang Zhang, Liuxiang Chen
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