Is it time for Cold-Endoscopic Submucosal Dissection? A feasibility study in an esophageal and colorectal live porcine model

Authors

Adolfo Parra-Blanco[1], Miguel Fraile-López[1,2,3]

Institutions

1 NIHR Nottingham Biomedical Research Centre, Department of Gastroenterology, Nottingham University Hospitals NHS Trust and University of Nottingham, Nottingham, UK
2 Department of Gastroenterology, Hospital Universitario Central de Asturias, Oviedo, Spain
3 Instituto de Investigación Sanitaria del Principado de Asturias (ISPA), Oviedo, Spain

submitted 24.4.2020
accepted after revision 12.6.2002

Bibliography
Endoscopy International Open 2020; 08: E1595–E1602
DOI 10.1055/a-1223-2127
ISSN 2364-3722

Corresponding author
Adolfo Parra-Blanco MD, PhD, FJGES, Head of Endoscopy Unit, Nottingham City Hospital Campus, Nottingham University Hospitals NHS Trust, Hucknall Rd, Nottingham NG5 1PB, UK
Fax: +0115 8405821
Adolfo.Parra-Blanco@nottingham.ac.uk

ABSTRACT

Background and study aims Use of cold endoscopic resection has increased due to excellent results with it and the ability to avoid electrosurgery related complications. The aim of this study was to evaluate the feasibility and safety of cold-endoscopic submucosal dissection (C-ESD) in an in vivo porcine model.

Patients and methods C-ESD with circumferential incision and submucosal dissection with a predominantly cold technique was tested in the esophagus and colorectum. Incision and dissection were attempted with a cold technique with a biopsy forceps and an endoscopic Maryland dissector. Large vessels were pre-coagulated with the latter device. Different traction methods were applied.

Results Twelve dissections were performed: four esophageal, four colonic, and four rectal. Tunnel and pocket methods were applied successfully. Full C-ESD was possible in the colorectum. In the esophagus, an initial incision had to be done with electrocautery. No major bleeding occurred. Two perforations occurred in the colon, one was endoscopically treated.

Conclusions Full C-ESD is feasible in the colorectum, whereas a small hot incision is needed in the esophagus. However, in 50 % of the colonic cases, there were perforations caused by the biopsy forceps making the circumferential incision. Therefore, potential benefits of endoscopic resection without cautery would warrant futures studies in humans initially in esophagus and rectal locations.

Introduction

Endoscopic submucosal dissection (ESD) is a widely-used technique for removal of premalignant and malignant lesions in the gastrointestinal tract. Use of cold endoscopic resection techniques has recently increased due to their safety. Piecemeal cold snare has proven safe and effective for large flat lesions [1 – 3]. The advantages of avoiding cautery are well known, but lesion size represents a major limitation for cold "en bloc" resection.

The hypothesis of this study was that cold technique can be applied to ESD. The aim of this study was to evaluate the feasibility and safety of predominantly Cold-ESD (C-ESD) in a live porcine model in the esophagus and the colorectum.

Patients and methods

This was a prospective, preclinical, proof-of-concept, non-survival animal study. It was conducted between June and November 2019.

Animals and preoperative preparation

This animal study was developed at the Hospital Virtual de Valdecilla (Santander, Spain) and at Centro de Investigación Experimental Biomédica (Lleida, Spain), following study protocol approval by the Animal Ethical and Welfare Committee of Hospital Virtual de Valdecilla.

Three domestic pigs were used with a mean weight of 41.36 kg (range 39.7 – 43.2). Bowel preparation was performed with a low-fiber diet and a purgative split preparation (Moviprep/Plenvue; Norgine, Harefied, UK). General anesthesia was administered using propofol and oxygen after endotracheal intubation with continuous monitoring of oximetry and electrocardiogram.

Cold-ESD technique

Two endoscopists participated in this study; one of them with extensive experience in ESD, and the second with experience in therapeutic endoscopy but not in ESD. Esophageal ESD was performed with a gastroscope (EG29 *i*10), and colorectal ESD with a colonoscope (EC3490TFI) with a high-definition video processor (EPK-*i*7010) (Pentax Medical, Spain). CO_2 insufflation, irrigation pump and an electrosurgical unit (VIO300D; Erbe, Türbingen, Germany) were utilized. Hypothetical lesions approximately 20 mm in size (except a colonic case, 50 mm) were marked with Hybrid-Knife T-Type (HK-T) (Erbe Elektromedizin Gmbh, Tübingen, Germany).

The C-ESD technique consists of submucosal injection with a mixed solution of saline and indigo carmine, followed by mucosal incision with multiple stepwise bites with a biopsy forceps (Radial Jaw 4/Needle; Boston Scientific, France), previously described by our group [4]. Once inside the submucosa, dissection was performed using an endoscopic Maryland dissector (Coag Dissector, Ovesco, Tübingen, Germany). This rotatable ESD forceps allows blunt dissection of the submucosa when opening and closing the clamp, and also coagulation of large vessels. In principle, this would be the only non-cold intervention with this technique, although HK-T would be allowed if needed in case of unsuccessful circumferential incision or dissection. Distal attachments were used (DH-28GR, 29CR; Fujifilm; D-201 – 11304; Olympus, Tokyo, Japan).

We wanted to test the feasibility of C-ESD with different ESD techniques: Conventional, pocket-creation method (PCM) [5], and tunneling. Usual traction methods were applied. To reduce the number of animals to a minimum, and because this was a proof-of-concept study, four resections in each location (esophagus, colon, and rectum) were planned.

Depending on the technique applied, the location and the organ, either a circumferential incision was attempted from the beginning, or a partial incision was undertaken.

Esophagus

1. For standard esophageal C-ESD, a C-shaped incision was performed, followed by trimming. Then, after the circumferential incision was concluded, the dissection was completed after applying traction by clip line.
2. For tunnel C-ESD, the anal and oral aspects were incised with HK-T, then the tunnel was completed. The clip line traction was applied, and then both lateral pillars were cut.

Colon and rectum

1. Stepwise circumferential incision and dissection, and traction with clip-band, or clip line (low rectum) was performed.
2. PCM was performed with or without clip-band [6, 7] depending on endoscopist preference.

Coagulation with the dissector was only applied when bleeding occurred, or prophylactically for vessels larger than 1 mm.

Post-resection evaluation

After study completion, the animals were sacrificed. Post-mortem examination was performed in cases with a suspected perforation. All specimens were examined for size and resection margins and each area was calculated using the minor and major axis (π *x major axis x minor axis/ 4*), for calculation of dissection speed.

Statistical analysis

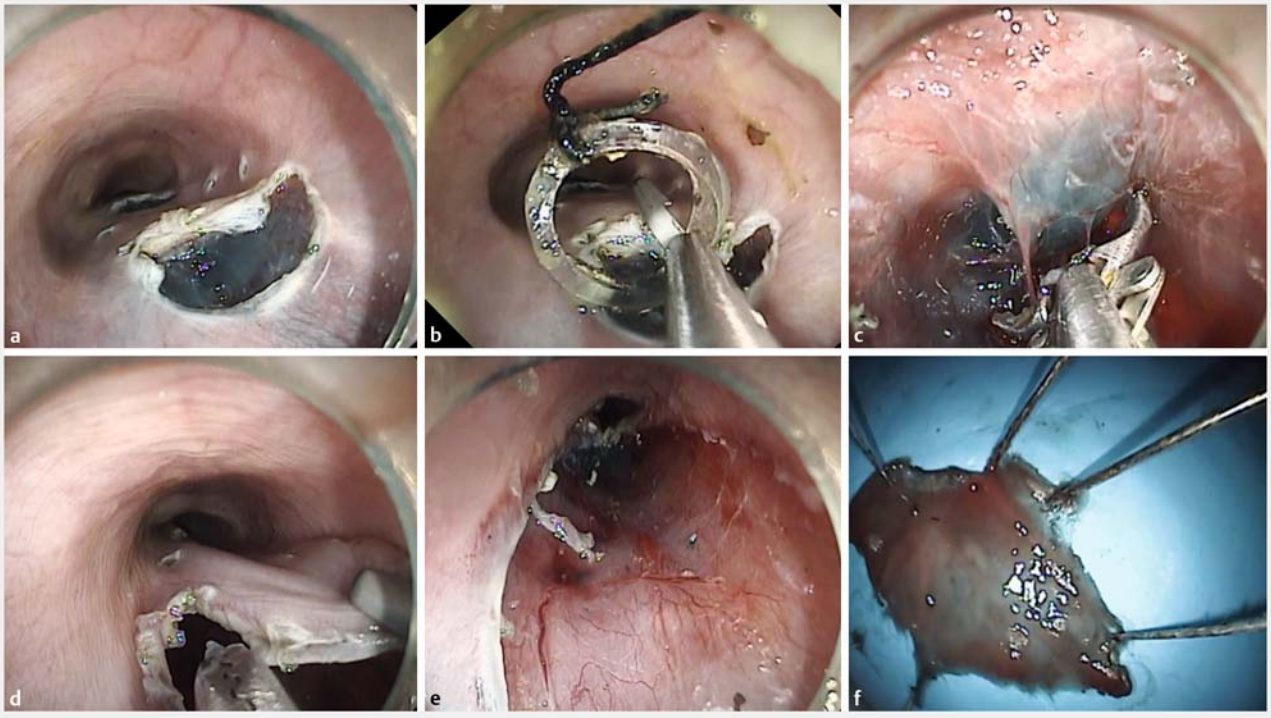
Data analysis was performed using SPSS 20 (Chicago, Illinois, United States). Continuous variables were expressed as medians with interquartile ranges and were analyzed by using a non-parametric 2-tailed Mann-Whitney U test. $P<0.05$ was considered significant.

Results

A total of 12 dissections were performed: four esophageal, four colonic and four in the rectum. Median dissection times, including circumferential incision and dissection, for each location were 62, 64.5, and 88 minutes, respectively. Median number of biopsies for circumferential incision was 55.5 for colon resections and 61.5 for the rectal ones. For colorectal dissections, the median number of biopsies for circumferential incision using the PCM was 63 for a median resected area of 647.9 mm^2 vs. 44 bites for 141.4 mm^2 with the conventional one. With PCM, a median of one biopsy was performed per 10.62 mm^2 vs. one biopsy per 3.21 mm^2 with the circumferential cut (n.s.). Five of 8 (62 %) colorectal dissections were performed completely free of cautery. All esophageal cases needed hot mucosal incision, and the dissection phase was completely cold in two of four cases (50 %).

Esophagus

Full C-ESD was attempted, but it was not possible to reach the submucosa with the bite of the biopsy forceps, and at least an initial incision with HK-T was necessary to gain submucosal access (▶ **Fig. 1**). The standard technique was applied in two cases, and the tunnel technique in the remaining two. The clip-band line traction method was applied in all cases to improve the submucosal view for dissection [8]. The subsequent dissection was completed with a cold technique, exposing a neat muscular layer. No bleeding or perforation occurred, and prophylactic coag-

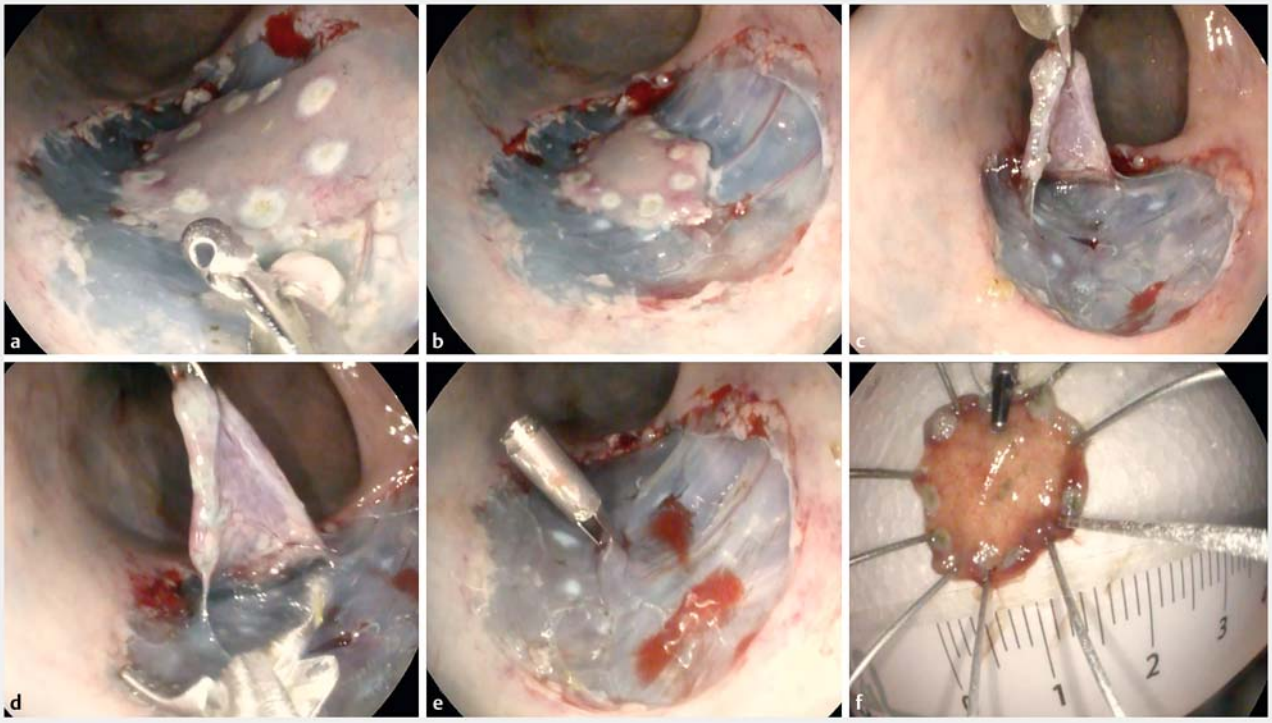


▶ **Fig. 1** Esophagus C-ESD (Tunnel technique). **a** Oral/anal incision with Hybrid Knife Type T. **b** Early application of clip-band line traction method. **c** Submucosal dissection with Maryland dissector under direct vision. **d** Lateral cut with Maryland dissector. **e** Clear scar with muscular layer exposure. **f** Specimen 'en bloc' resected.

▶ **Table 1** Esophagus Cold-ESD procedures.

	Case 1 **ESD Expert**	**Case 2** **Non-Expert**	**Case 3** **Non-Expert**	**Case 4** **ESD Expert**
Dissection technique	Circumferential incision	Circumferential incision	Tunnel	Tunnel
Traction, yes/no (method)	Yes (Clip-band line)	Yes (Clip-band line)	Yes (Clip-band line)	Yes (Clip-band line)
Specimen size, mm (mm^2)	19×11 (164.2)	23×20 (361.3)	22×18 (311)	22×11 (190.1)
Dissection time, min	77	111	47	47
Dissection speed, mm^2/min	2.13	3.25	6.62	4.04
Biopsy forceps bites to complete circumferential incision (after hot incision), n	0	0	47	36
Coagulation times, n	0	1	1	0
Injection volume, mL	28	40	30	22
Hybrid Knife Type-T use, yes/no	Yes (Circumferential incision)	Yes (Circumferential incision)	Yes (Oral/anal incision)	Yes (Oral/anal/anti-gravity side)
"En bloc" resection, yes/no	Yes	Yes	Yes	Yes
Bleeding episodes, n	0	0	0	0
Perforation, yes/no (treatment)	No	No	No	No

ESD, endoscopic submucosal dissection.



▶ **Fig. 2** Colonic C-ESD (Hybrid Biopsy EMR technique). **a** Circumferential incision with biopsy forceps (Hybrid Biopsy EMR). **b** Full circumferential incision. **c** Clip-band traction method exposing the submucosal dissection plane. **d** Cold dissection with Maryland dissector. **e** Scar with small vessel clipped. **d** Specimen 'en bloc' resected.

Video 1 Esophagus C-ESD (tunnel technique).

ulation was applied once in two cases. Information about esophageal C-ESD cases is presented in ▶ **Table 1** and ▶ **Video 1**.

Colon

Complete C-ESD was performed in two cases using sequential circumferential incision and dissection, and in the remaining two cases the PCM was applied (▶ **Fig. 2**). Clip-band traction method was used in one case, clip-band line in one, and the remaining two required no traction. No major bleeding was encountered. Coagulation was used twice for minor bleeding, and twice for prophylaxis both in the same case. Two perforations occurred, both during circumferential incision with the biopsy forceps. The first was in a case undertaken by the expert and the defect was immediately noticed and clipped. The second one occurred in a case by the non-expert; initially there was a small perforation, which was not recognized immediately and as such, resulted in a full-thickness resection, which was then impossible to close endoscopically. Post-mortem evaluation confirmed a very thin colonic wall with large perforation contained by the serous layer. Details of colonic C-ESD are summarized in ▶ **Table 2** and can be seen in ▶ **Video 2**.

Rectum

All four cases were completed using the C-ESD technique; the PCM was used in three cases (two with clip-band, one no traction), and conventional step-by step circumferential ESD with clip-band line traction in one. No major bleeding occurred; prophylactic coagulation was used on four occasions (two cases) and there were no perforations. Rectal C-ESD details are shown in ▶ **Table 3**. Technical details can be seen in ▶ **Video 3**.

Procedure times according to Cold-ESD location and comparison of procedure times between both endoscopists can be seen in ▶ **Table 4** and ▶ **Table 5**.

▶ **Table 2** Colon Cold-ESD procedures.

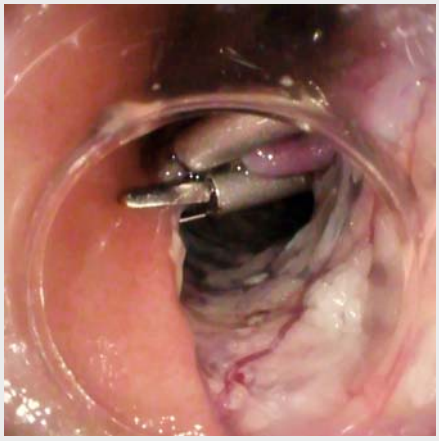
	Case 1 ESD Expert	Case 2 ESD Non-expert	Case 3 ESD Expert	Case 4 ESD Non-expert
Dissection technique	Circumferential incision	Circumferential incision	PCM	PCM
Traction, yes/no (method)	Yes (Clip-band)	No	No	Yes (Clip-band line)
Specimen size, mm (mm^2)	17 × 12 (131.9)	18 × 10 (141.4)	52 × 33 (1347.7)	26 × 23 (469.7)
Dissection time, min	42	30	141	87
Dissection speed, mm^2/min	3.14	4.71	9.56	5.39
Incision time, min	10	22	4	11
Biopsy forceps bites to access to Sm with PCM, n	–	–	10	28
Biopsy forceps bites for circumferential incision, n	48	44	82	63
Coagulation times, n	0	0	4	0
Injection volume, mL	20	14	30	50
Hybrid Knife Type-T use, yes/no	No	No	No	No
"En bloc" resection, yes/no	Yes	Yes	Yes	Yes
Bleeding episodes, n	0	0	2	0
Perforation, yes/no (treatment)	No	Yes (Not possible)	Yes (Clipped)	No

ESD, endoscopic submucosal dissection; Sm, submucosal layer; PCM, pocket creation method.

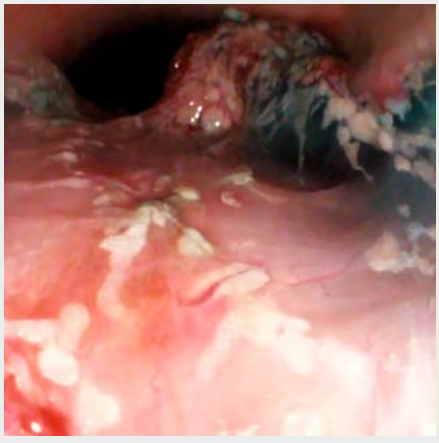
▶ **Table 3** Rectum Cold-ESD procedures.

	Case 1 ESD Expert	Case 2 Non-Expert	Case 3 ESD Expert	Case 4 Non-Expert
Dissection technique	Circumferential incision	PCM	PCM	PCM
Traction, yes/no (method)	Yes (Clip-band line)	Yes (Clip-band)	No	Yes (Clip-band)
Specimen size, mm (mm^2)	33 × 30 (777.5)	28 × 26 (571.8)	33 × 25 (647.9)	36 × 31 (876.5)
Dissection time, min	69	90	100	86
Dissection speed, mm^2/min	11.27	6.35	6.48	10.19
Incision time, min	16	11	3	5
Biopsy forceps bites to access to Sm with PCM, n	–	28	7	9
Biopsy forceps bites for circumferential incision, n	33	69	61	62
Coagulation times, n	0	2	2	0
Injection volume, ml	29	50	42	36
Hybrid Knife-T, yes/no	No	No	No	No
"En bloc" resection, yes/no	Yes	Yes	Yes	Yes
Bleeding episodes, n	0	0	0	0
Perforation, yes/no (treatment)	No	No	No	No

ESD, endoscopic submucosal dissection; Sm, submucosal layer; PCM, pocket creation method.



Video 2 Colon C-ESD (pocket technique).



Video 3 Rectum C-ESD (pocket technique).

▶ **Table 4** Procedure times according to Cold-ESD location

	Esophagus ESD	Colon ESD	Rectum ESD
Dissection time (min), median (IQR)	62 (55.5)	64.5 (94.5)	88 (24.25)
Dissection speed (mm^2/min), median (IQR)	3.64 (3.56)	5.05 (4.98)	8.33 (4.62)

ESD, endoscopic submucosal dissection; Min, minutes; IQR, interquartile range.

▶ **Table 5** Comparison of procedure times between both endoscopists.

	Non-Expert	Expert	*P* Value
Dissection time (min), median (IQR)	86.5 (52.5)	73 (64.5)	0.567
Dissection speed (mm^2/min), median (IQR)	5.87 (3.17)	5.26 (7.1)	1.000
Area (mm^2), median (IQR)	415.5 (379.37)	419 (763.92)	1.000

ESD, endoscopic submucosal dissection; Min, minutes; IQR, interquartile range; Min, minutes. *P* values obtained from a 2-tailed Mann-Whitney U test.

Discussion

In the era of "the cold resection revolution", we evaluated for the first time in this porcine model study the feasibility of C-ESD in the colorectum and esophagus. Full C-ESD has been proven feasible in the colorectum, and an initial hot incision has been shown to be needed for esophageal ESD in this porcine model. Importantly, 50 % of colonic cases resulted in a perforation.

Von Renteln et al previously described the advantages of gastric C-ESD in a porcine model comparing hot circumferential incision followed by blunt dissection with standard ESD [9]. They also reported a successful case in a pig colon with this technique [10].

The advantages of not using electrocautery in endoscopic resections are well established [11]. The risk of deep mural injury is reduced, which results in a lower risk of post-polypectomy syndrome, delayed bleeding, and perforation; specimen resection margins will be free of electrocautery artifacts, which should improve the quality of a specimen for histological analysis. Moreover, size should not be a limitation for "en bloc" resection with C-ESD. It could be hypothesized that C-ESD would be technically less challenging compared with standard ESD in the esophagus and the colorectum, both being thin-walled organs, in which perforations can happen even with minimal applications of cautery.

The risk of intraprocedural bleeding needs to be considered, and minor bleeding can be expected to be common, although in our in vivo porcine model study, this was not proven to be the case. In series reporting piecemeal cold resection of large flat lesions (>20 mm), risk of significant bleeding has been shown to be very low (0.7 – 1.3 % intraprocedural, 0 – 0.5 % delayed) [1]. Therefore, we would not anticipate an increase in significant bleeding with C-ESD.

While perforations did occur in this study, it is thought that in humans, the risk of perforation would be lower, considering the extreme thinness of the colonic wall in the 40-kg pigs in our study [12]. It is of note that both perforations were caused by the biopsy forceps during circumferential incision. However, reports of colorectal perforations in humans with a biopsy forceps are very rare in the literature. In addition, the authors have accumulated experience in Hybrid Biopsy Endoscopic Mucosal Resection (EMR) in humans using a needleless forceps, with no perforations during the circumferential incision. Therefore, a

needle-free forceps would be recommended, at least for colorectal C-ESD, because the amount of tissue plucked with each bite is less, and the perforation risk may be lower. Although saline solution was used in this study, we would advocate use of longer-lasting solutions to facilitate C-ESD incision in different locations of the gastrointestinal tract.

Compared to other cold resection methods recently reported, such as monopolar scissors, we believe that the method we propose may be safer for intraprocedural bleeding and immediate perforation, because tissue and vessels are cold dissected applying traction instead of transversal cut as scissors-type knives do, but this should be proven in comparative studies [13]. This technique can be considered relatively inexpensive, because the same clamp can be used to dissect and coagulate vessels without needing a dedicated coagulation forceps, and the price of the biopsy forceps is negligible compared with additional knives that are frequently used for circumferential incision.

We found a clip-band line traction method during esophageal C-ESD to be particularly useful, enabling observation confidently of the dissection plane, as previously reported in a randomized study in a porcine model [14], and in a recent multicenter, randomized controlled trial in humans [15]. In the colorectal cases undertaken with a traction method, it was felt by the endoscopists that identification of the dissection plane was easier. We believe that C-ESD and a traction method could be an advantageous combination to facilitate the procedure.

This feasibility study has some limitations. First, it was not a survival study, so delayed complications could not be assessed. Second, the size of the resections may appear small; a measurement tool was not used to mark the width of the hypothetical lesions, and due to the extreme thinness of the pieces obtained, they were probably not stretched to the maximum level to avoid fragmentation. Third, given the absence of a control group, no comparisons can be made with the standard technique in terms of procedure time or costs.

Fourth, no histopathological evaluation was made of the resected specimens. Therefore, it is not possible to confirm that this technique allows precise histopathological examination. Although the specimens were not fragmented, and no mucosal defects were observed in them, this issue should be investigated in future studies.

Conclusion

In conclusion, C-ESD is feasible in the colorectum in a porcine model, and an initial hot incision is needed for esophageal resections. Five of eight (62 %) colorectal cases were completed without application of any electrocautery. Perforations occurred in 50 % of colonic cases, caused by the biopsy forceps, but there were no perforations with the dissector in the study. Although colonic perforation with a cold biopsy is extraordinary in humans, precautions should be taken, and some modifications in the design of the biopsy forceps would be desirable to facilitate the application of C-ESD. Further studies will be needed to assess the feasibility of C-ESD in humans, initially in the esophagus and the rectum, and to compare its safety and efficacy with standard ESD techniques.

Acknowledgments

Joaquín de la Peña, MD. Gastroenterology Department. Hospital de Valdecilla. Santander. Spain.
Sabina Beg, MD, PhD. NIHR Nottingham Biomedical Research Centre, Nottingham University Hospitals, United Kingdom.
Pentax Medical Iberia, Spain.
Ovesco AG, Germany, Izasa Hospital S.L.U, Spain.
Hospital Virtual de Valdecilla (HVV). Santander. Spain.
Centro de Investigación Biomédica Aplicada (CREBA). Lleida. Spain.

Competing interests

The authors declare that they have no conflict of interest.

References

[1] Chandrasekar VT, Spadaccini M, Aziz M et al. Cold snare endoscopic resection of nonpedunculated colorectal polyps larger than 10 mm: a systematic review and pooled-analysis. Gastrointest Endosc 2019; 89: 929–36.e3

[2] Tate DJ, Awadie H, Bahin FF et al. Wide-field piecemeal cold snare polypectomy of large sessile serrated polyps without a submucosal injection is safe. Endoscopy 2018; 50: 248–252

[3] Chandrasekar VT, Aziz M, Patel HK et al. Efficacy and safety of endoscopic resection of sessile serrated polyps 10 mm or larger: a systematic review and meta-analysis: resection of sessile serrated polyps ³ 10mm. Endoscopy 2019; 51: 941–949

[4] Fraile-López M, Jawhari A, Asgari S et al. Hybrid-biopsy endoscopic mucosal resection: an effective and simple technique for flat colorectal lesions. Endoscopy 2019; 51: E201–E203

[5] Harada H, Nakahara R, Murakami D et al. Saline-pocket endoscopic submucosal dissection for superficial colorectal neoplasms: a randomized controlled trial. Gastrointest Endosc 2019; 90: 278–287

[6] Parra-Blanco A, Nicolas D, Arnau MR et al. Gastric endoscopic submucosal dissection assisted by a new traction method: the clip-band technique. A feasibility study in a porcine model. Gastrointest Endosc 2011; 74: 1137–1141

[7] Jacques J, Charissoux A, Legros R et al. Double-clip counter-traction using a rubber band is a useful and adaptive tool for colonic endoscopic submucosal dissection. Endoscopy 2018; 50: 179–181

[8] Fraile-López M, Parra-Blanco A. Double tunnel and double Clip-band-line traction circumferential ESD for squamous esophageal neoplasia. Endoscopy 21.02 2020: doi:10.1055/a-1109-2365

[9] Von Renteln D, Pohl H, Vassiliou MC et al. Endoscopic submucosal dissection by using a flexible Maryland dissector: a randomized, controlled, porcine study (with videos). Gastrointest Endosc 2010; 71: 1056–1062

[10] Von Renteln D, Phol H, Vassiliou MC et al. Colonic endoscopic submucosal dissection using a flexible Maryland dissector. Endoscopy 2009; Suppl2: E263

[11] Tutticci NJ, Kheir AO, Hewett DG. The cold revolution: how far can it go? Gastrointest Endosc Clin N Am 2019; 29: 721–736

[12] Yahagi N, Neuhaus H, Schumacher B et al. Comparison of standard endoscopic submucosal dissection (ESD) versus an optimized ESD technique for the colon: an animal study. Endoscopy 2009; 41: 340–345

[13] Kantsevoy SV, Levihim S, Pennington D et al. A novel approach for endoscopic submucosal dissection: scissors versus electricity. VideoGIE 2019; 23: 472–475

[14] Jin P, Fu KI, Yu Y et al. Traction using a clip-with-line is a preferred method for trainees in performing esophageal endoscopic submucosal dissection: an animal model study. Therap Adv Gastroenterol 2017; 10: 343–351

[15] Yoshida M, Takizawa K, Nonaka S et al. Conventional versus traction-assisted endoscopic submucosal dissection for large esophageal cancers: a multicenter, randomized controlled trial (with video). Gastrointest Endosc 2020; 91: 55–65.e2