refinement of ESD-related instruments, devices, equipment, and injection solutions will help facilitate increased use of colorectal ESD throughout the world.

REFERENCES

1. Ahmad NA, Kochman ML, Long WB, et al. Efficacy, safety, and clinical outcomes of endoscopic mucosal resection: a study of 101 cases. Gastrointest Endosc 2002;55:390–6.
2. Yokota T, Sugihara K, Yoshida S. Endoscopic mucosal resection for colorectal neoplastic lesions. Dis Colon Rectum 1994;37:1108–11.
3. Soetikno RM, Gotoda T, Nakanishi Y, et al. Endoscopic mucosal resection. Gastrointest Endosc 2003;57:567–79.
4. Saito Y, Fujii T, Kondo H, et al. Endoscopic treatment for laterally spreading tumors in the colon. Endoscopy 2001;33:682–6.
5. Kudo S, Kashida H, Tamura T, et al. Colonoscopic diagnosis and management of nonpolypoid early colorectal cancer. World J Surg 2000;24:1081–90.
6. Kitajima K, Fujimori T, Fujii S, et al. Correlations between lymph node metastasis and depth of submucosal invasion in submucosal invasive colorectal carcinoma: a Japanese collaborative study. J Gastroenterol 2004;39:534–43.
7. Hosokawa K, Yoshida S. Recent advances in endoscopic mucosal resection for early gastric cancer [in Japanese with English abstract]. Jpn J Canc Chemother 1998;25:476–83.
8. Ono H, Kondo H, Gotoda T, et al. Endoscopic mucosal resection for treatment of early gastric cancer. Gut 2001;48:225–9.
9. Yamamoto H, Kawata H, Sunada K, et al. Successful en-bloc resection of large superficial tumors in the stomach and colon using sodium hyaluronate and small-caliber-tip transparent hood. Endoscopy 2003;35:690–4.
10. Fujishiro M, Yahagi N, Kakushima N, et al. Outcomes of endoscopic submucosal dissection for colorectal epithelial neoplasms in 200 consecutive cases. Clin Gastroenterol Hepatol 2007;5:674–7.
11. Saito Y, Emura F, Matsuda T, et al. A new sinker-assisted endoscopic submucosal dissection for colorectal tumors. Gastrointest Endosc 2005;62:297–301.
12. Saito Y, Uraoka T, Matsuda T, et al. A pilot study to assess safety and efficacy of carbon dioxide insufflation during colorectal endoscopic submucosal dissection under conscious sedation. Gastrointest Endosc 2007;65:537–42.
13. Saito Y, Uraoka T, Matsuda T, et al. Endoscopic treatment of large superficial colorectal tumors: a cases series of 200 endoscopic submucosal dissections (with video). Gastrointest Endosc 2007;66:966–73.
14. Yamazaki K, Saito Y, Fukuzawa M. Endoscopic submucosal dissection of a large laterally spreading tumor in the rectum is a minimally invasive treatment. Clin Gastroenterol Hepatol 2008;6:e5–6.
15. Saito Y, Kawano H, Takeuchi Y, et al. Current status of colorectal endoscopic submucosal dissection in Japan and other Asian countries: progressing towards technical standardization. Dig Endosc 2012;24(Suppl 1):67–72.
16. Uraoka T, Saito Y, Matsuda T, et al. Endoscopic indications for endoscopic mucosal resection of laterally spreading tumours in the colorectum. Gut 2006;55:1592–7.
17. Sakamoto T, Saito Y, Matsuda T, et al. Treatment strategy for recurrent or residual colorectal tumors after endoscopic resection. Surg Endosc 2011;25:255–60.
18. Ono A, Fujii T, Saito Y, et al. Endoscopic submucosal resection of rectal carcinoid tumors with a ligation device. Gastrointest Endosc 2003;57:583–7.

19. Mashimo Y, Matsuda T, Uraoka T, et al. Endoscopic submucosal resection with a ligation device is an effective and safe treatment for carcinoid tumors in the lower rectum. J Gastroenterol Hepatol 2008;23:218–21.

20. Fujii T, Hasegawa RT, Saitoh Y, et al. Chromoscopy during colonoscopy. Endoscopy 2001;33:1036–41.

21. Matsuda T, Fujii T, Saito Y, et al. Efficacy of the invasive/non-invasive pattern by magnifying estimate the depth of invasion of early colorectal neoplasms. Am J Gastroenterol 2008;103:2700–6.

22. Sano Y, Ikematsu H, Fu KI, et al. Meshed capillary vessels by use of narrow-band imaging for differential diagnosis of small colorectal polyps. Gastrointest Endosc 2009;69:278–83.

23. Ikematsu H, Matsuda T, Emura F, et al. Efficacy of capillary pattern type IIIA/IIIB by magnifying narrow band imaging for estimating depth of invasion of early colorectal neoplasms. BMC Gastroenterol 2010;10:33.

24. Kikuchi T, Fu KI, Saito Y, et al. Transcutaneous monitoring of partial pressure of carbon dioxide during endoscopic submucosal dissection of early colorectal neoplasia with carbon dioxide insufflation: a prospective study. Surg Endosc 2010;24:2231–5.

25. Uraoka T, Fujii T, Saito Y, et al. Effectiveness of glycerol as a submucosal injection for EMR. Gastrointest Endosc 2005;61:736–40.

26. Saito Y, Uraoka T, Yamaguchi Y, et al. A prospective, multicenter study of 1111 colorectal endoscopic submucosal dissections (with video). Gastrointest Endosc 2010;72:1217–25.

27. Hotta K, Yamaguchi Y, Saito Y, et al. Current opinions for endoscopic submucosal dissection for colorectal tumors from our experiences: indications, technical aspects and complications. Dig Endosc 2012;24(Suppl 1):110–6.

28. Tamegai Y, Saito Y, Masaki N, et al. Endoscopic submucosal dissection: a safe technique for colorectal tumors. Endoscopy 2007;39:418–22.

29. Tanaka S, Oka S, Kaneko I, et al. Endoscopic submucosal dissection for colorectal neoplasia: possibility of standardization. Gastrointest Endosc 2007;66:100–7.

30. Friedland S, Sedehi D, Soetikno R. Colonoscopic polypectomy in anticoagulated patients. World J Gastroenterol 2009;15:1973–6.

31. Lee EJ, Lee JB, Lee SH, et al. Endoscopic submucosal dissection for colorectal tumors-1,000 colorectal ESD cases: one specialized institute's experiences. Surg Endosc 2013;27(1):31–9.

32. Kang KJ, Kim DU, Kim BJ, et al. Endoscopy-based decision is sufficient for predicting completeness in lateral resection margin in colon endoscopic submucosal dissection. Digestion 2012;85:33–9.

33. Hurlstone DP, Atkinson R, Sanders DS, et al. Achieving R0 resection in the colorectum using endoscopic submucosal dissection. Br J Surg 2007;94:1536–42.

34. Probst A, Golger D, Anthuber M, et al. Endoscopic submucosal dissection in large sessile lesions of the rectosigmoid: learning curve in a European center. Endoscopy 2012;44:660–7.

35. Antillon MR, Bartalos CR, Miller ML, et al. En bloc endoscopic submucosal dissection of a 14-cm laterally spreading adenoma of the rectum with involvement to the anal canal: expanding the frontiers of endoscopic surgery (with video). Gastrointest Endosc 2008;67:332–7.

36. Fukami N, Ryu CB, Said S, et al. Prospective, randomized study of conventional versus HybridKnife endoscopic submucosal dissection methods for the esophagus: an animal study. Gastrointest Endosc 2011;73:1246–53.

37. Saito Y, Fukuzawa M, Matsuda T, et al. Clinical outcome of endoscopic submucosal dissection versus endoscopic mucosal resection of large colorectal tumors as determined by curative resection. Surg Endosc 2010;24:343–52.

38. Hotta K, Fujii T, Saito Y, et al. Local recurrence after endoscopic resection of colorectal tumors. Int J Colorectal Dis 2009;24(2):225–30.

39. Moss A, Bourke MJ, Williams SJ, et al. Endoscopic mucosal resection outcomes and prediction of submucosal cancer from advanced colonic mucosal neoplasia. Gastroenterology 2011;140:1909–18.

40. Kiriyama S, Saito Y, Yamamoto S, et al. Comparison of endoscopic submucosal dissection with laparoscopic-assisted colorectal surgery for early-stage colorectal cancer: a retrospective analysis. Endoscopy 2012;44:1024–30.

41. Kobayashi N, Saito Y, Uraoka T, et al. Treatment strategy for laterally spreading tumors in Japan: before and after the introduction of endoscopic submucosal dissection. J Gastroenterol Hepatol 2009;24:1387–92.

42. Kiriyama S, Saito Y, Matsuda T, et al. Comparing endoscopic submucosal dissection with transanal resection for non-invasive rectal tumor: a retrospective study. J Gastroenterol Hepatol 2011;26:1028–33.

43. Park SU, Min YW, Shin JU, et al. Endoscopic submucosal dissection or transanal endoscopic microsurgery for nonpolypoid rectal high grade dysplasia and submucosa-invading rectal cancer. Endoscopy 2012;44:1031–6.

44. Parra-Blanco A, Gonzalez N, Arnau MR. Ex vivo and in vivo models for endoscopic submucosal dissection training. Clin Endosc 2012;45:350–7.

Endoscopic Submucosal Dissection

Editor

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complications, delayed bleeding, and perforation. A small-caliber-tip transparent hood is useful. Mechanical stretching of the submucosal tissue allows safe dissection and effective prevention of bleeding with minimum muscle injury under direct visualization of the submucosal tissue and blood vessels. A short double-balloon endoscope is useful to stabilize control of the endoscope tip in distal duodenal ESD. Selection of ESD in the duodenum should be made cautiously considering both benefits and risks of the procedure.

Yutaka Saito, Taku Sakamoto, Takeshi Nakajima, and Takahisa Matsuda

The number of medical facilities that perform colorectal endoscopic submucosal dissection (ESD) has been growing, and its effectiveness has been increasingly reported in recent years. Indications approved by the Japanese government's medical insurance system are early colorectal cancers with a maximum tumor size of 2–5 cm. ESD was an effective procedure for treating noninvasive colorectal tumors difficult to resect en bloc by conventional EMR, resulting in a higher en bloc resection rate that is less invasive than surgery. Based on the excellent clinical results of colorectal ESDs, the Japanese health care insurance system has approved colorectal ESD for coverage.

Haruhiro Inoue, Esperanza Grace Santi, Manabu Onimaru, and Shin-ei Kudo

Peroral endoscopic myotomy (POEM) is an evolving minimally invasive endoscopic surgical procedure, with no skin incision, intended for long-term recovery from symptoms of esophageal achalasia. POEM was developed based on both the already established surgical principles of esophageal myotomy and the advanced techniques of endoscopic submucosal dissection. This article relates how POEM was developed, and its use in practice is reported and discussed. As an extension of the POEM technique, submucosal endoscopic tumor resection is introduced.

Kazuki Sumiyama, Christopher J. Gostout, and Hisao Tajiri

Submucosal endoscopy with a mucosal flap (SEMF) safety valve technique is a global concept in which the submucosa is a free working space for endoscopic interventions. A purposefully created intramural space provides an endoscopic access route to the deeper layers and into the extraluminal cavities. The mucosa overlying the intramural space is protective, reducing contamination during natural orifice transluminal endoscopic surgery (NOTES) procedures and providing a sealant flap to repair the entry point and the submucosal space. In addition to NOTES, SEMF enables endoscopic achalasia myotomy, histologic analysis of the muscularis propria, and submucosal tumor removal.

CASE REPORT

Hereditary diffuse gastric cancer in a Japanese family with a large deletion involving *CDH1*

Masayoshi Yamada · Takeo Fukagawa · Takeshi Nakajima · Kiyoshi Asada · Shigeki Sekine · Satoshi Yamashita · Eriko Okochi-Takada · Hirokazu Taniguchi · Ryoji Kushima · Ichiro Oda · Yutaka Saito · Toshikazu Ushijima · Hitoshi Katai

Abstract Hereditary diffuse gastric cancer (HDGC), characterized by susceptibility to gastric signet ring cell carcinomas (SRCCs) and caused by *CDH1* germline mutations, is rare in the Japanese. We present here a Japanese family with HDGC identified by comparative genomic hybridization (CGH) analysis. A 55-year-old woman was treated with completion gastrectomy for multiple SRCCs, and pathological examination revealed approximately 200 foci of SRCC with loss of E-cadherin expression. Her 30-year-old son had surveillance endoscopy and was found to have multiple SRCCs. He underwent total gastrectomy, and 32 foci of SRCC with loss of E-cadherin expression were histologically found. Although no point mutations were detected in *CDH1* by sequencing, CGH revealed a 275-kb deletion involving exons 7–16 of *CDH1* in both patients. While only a few HDGCs have been reported in East Asia, patients with multiple SRCC may need to be offered appropriate genetic counseling and testing in this area.

Keywords Hereditary diffuse gastric cancer · Gastric cancer · E-cadherin · Comparative genomic hybridization

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Introduction

Hereditary diffuse gastric cancer (HDGC) is an autosomal dominant disease associated with multiple signet ring cell carcinomas (SRCCs) and is caused by a germline mutation in the E-cadherin gene (*CDH1*). Guilford et al. [1] first reported that HDGC is caused by truncating mutations of *CDH1* in New Zealand Maori families with early-onset, multiple SRCCs. In Western countries, inherited gastric cancers (GCs) are thought to account for 1–3 % of all GCs [2]. Clinically, approximately 25 % of families fulfilling the criteria for the diagnosis of HDGC have inactivating *CDH1* germline mutations [2, 3]. In contrast, in East Asian countries, including Japan, Korea, and China, with high incidences of GCs [4], HDGC has rarely been reported [5, 6]. Recently, Yamada et al. [7] reported two germline alterations in the *CDH1* gene in two Japanese familial GCs. However, HDGC is still rarely diagnosed in East Asian countries, and it is still unknown whether HDGC is really rare or overlooked because of the high incidence of coincidental familial GC. We report here clinical characteristics and genomic analysis of a Japanese HDGC family with a *CDH1* germline mutation.

Case reports

A 55-year-old female patient was referred to our hospital for treatment of multiple SRCCs detected by endoscopic examination during an annual health check. She had a past history of intramucosal SRCC at the age of 34 and had undergone distal gastrectomy with *Billroth I* reconstruction in another hospital. She had no past history of other malignancies, including lobular breast cancer. Family history of GC was noted, affecting the patient's father and paternal grandfather (Fig. 1). Esophagogastroduodeno-scopy (EGD) in our hospital detected 10–12 small pale mucosal patches, mainly in the greater curvature of the remnant stomach (Fig. 2a), but no atrophic gastritis indicative of *Helicobacter pylori* infection. The size of each focus was endoscopically estimated as less than 10 mm. In one of the largest lesions, narrow band imaging with magnification showed a wavy-shaped irregular microvessel pattern suggesting undifferentiated adenocarcinoma (Fig. 2b). All the biopsy specimens from six lesions demonstrated SRCC. Completion gastrectomy was performed, and the entire gastric mucosa was histologically examined.

Histopathological examination revealed approximately 200 SRCC foci, and their maximum size was 10 mm in diameter (Fig. 2f). None of the lesions showed submucosal invasion, and tumor cells were mostly confined to the upper mucosal layer (Fig. 2c). Periodic acid-Schiff (PAS) staining highlighted intracytoplasmic mucin in SRCC (Fig. 2d).
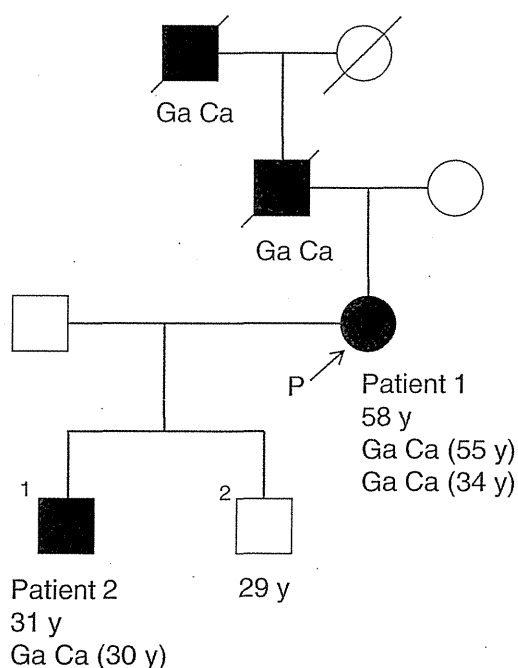

Fig. 1 Pedigree chart. The presence of the deletion mutation (Fig. 4) was confirmed in patients 1 and 2 in two generations

Immunohistochemistry was performed using anti-E-cad-herin antibody (NCH-38; 1:100 dilution; DAKO, Glostrup, Denmark) as previously described [8]. The results showed loss of E-cadherin expression in SRCCs. (Fig. 2e). These histopathological findings were exactly identical to the findings in the HDGC cases reported by Guilford et al. [1]. No lymph node metastasis was observed.

Based on the pathological findings of the proband (patient 1), we suspected that she might have HDGC. Therefore, we performed surveillance endoscopy of two sons of the proband. Neither son had a history of malignancy. EGD for the 30-year-old elder son (Patient 2) detected three tiny pale areas at the body-antrum junction of the stomach (Fig. 3a, b). Apart from these tiny pale patches, no other endoscopic findings were suggestive of SRCC, but biopsy specimens obtained from each of the three pale areas revealed SRCC. Based on these findings, we clinically diagnosed HDGC in both patients, and total gastrectomy was performed on patient 2. Histopathological assessment of the entire gastric mucosa revealed a total of 32 SRCCs (Fig. 3f) along with loss of E-cadherin expression in SRCCs (Fig. 3c, d, e). No apparent findings of recurrence or distant metastasis have been noted on the follow-up thus far. EGD in the younger son identified no significant endoscopic findings.

The presence of a 275-kb deletion involving *CDH1*

In order to perform genetic analysis, genomic DNA was extracted from peripheral leukocytes of patient 1, patient 2, and a healthy volunteer. Genomic DNA was also extracted from biopsy specimens obtained from the cancer site(s) in the stomachs of both patients. All 16 exons of *CDH1* were sequenced by PCR-direct sequencing from both directions using previously reported primers [9]. Promoter methylation was analyzed by bisulfite modification and quantitative real-time methylation specific PCR (MSP) as previously described [10]. Primer sequences for the methylated DNA were 5′-TCG TTT TGG GGA GGG GTT C-3′ (forward) and 5′-CAA ATA AAC CCC GAA AAC ACC G-3′ (reverse), and those for the unmethylated DNA were 5′-GGA GGT ATT GTT TTT TGT ATT-3′ (forward) and 5′-AAC AAA CCA TCA ACT CCA-3′ (reverse). However, no *CDH1* mutation or aberrant methylation was detected in peripheral lymphocytes or biopsy specimens of patient 1.

Array-comparative genomic hybridization (CGH) analysis was performed according to the manufacturer's protocol using genomic DNA of peripheral leukocytes and human reference DNA (Caucasian, male #5190-4370, female #5190-4371, Agilent Technologies, Santa Clara, CA). DNA was digested with *Alu*I and *Rsa*I, labeled with Cy5 and Cy3, respectively, using a SureTag DNA
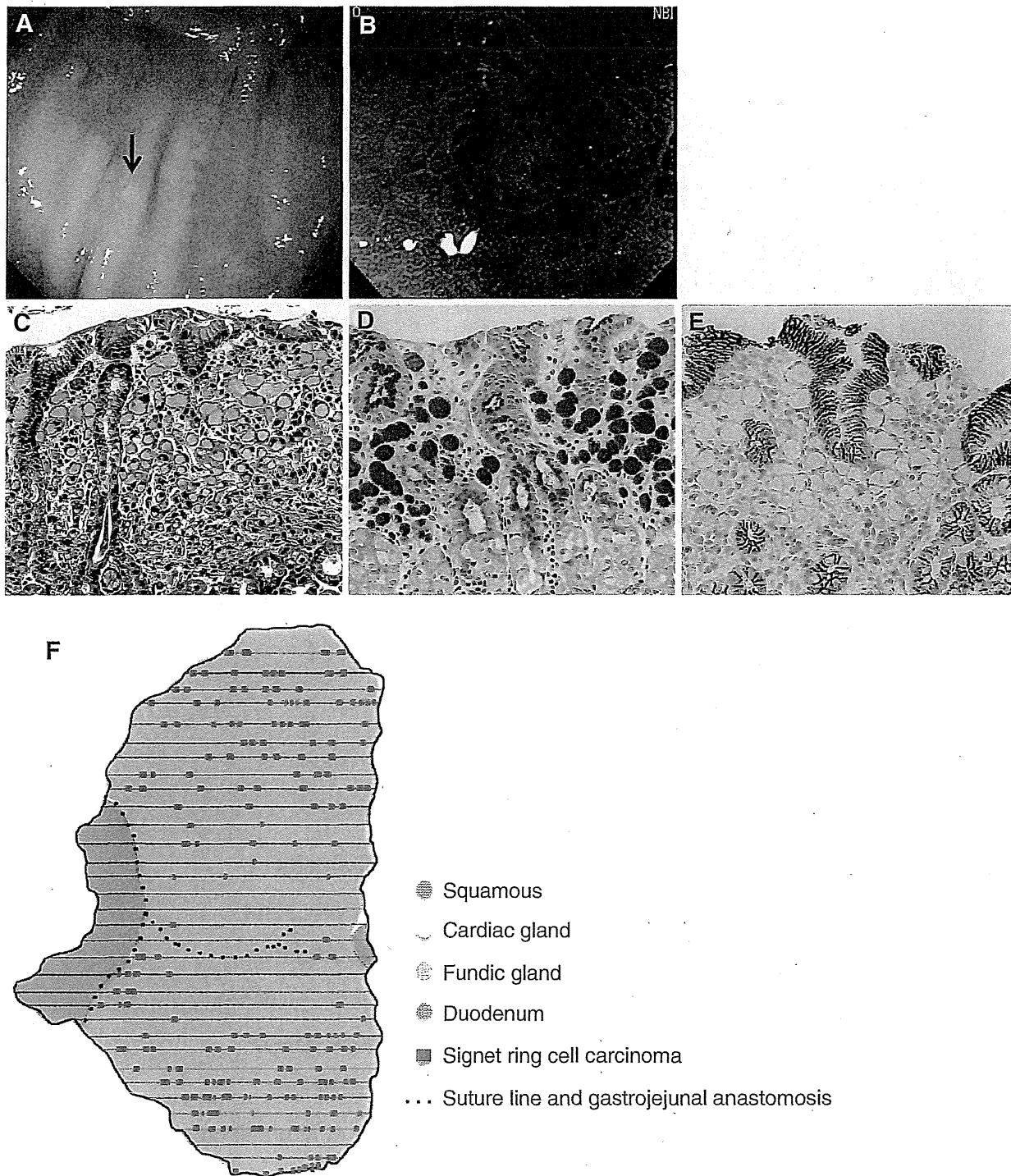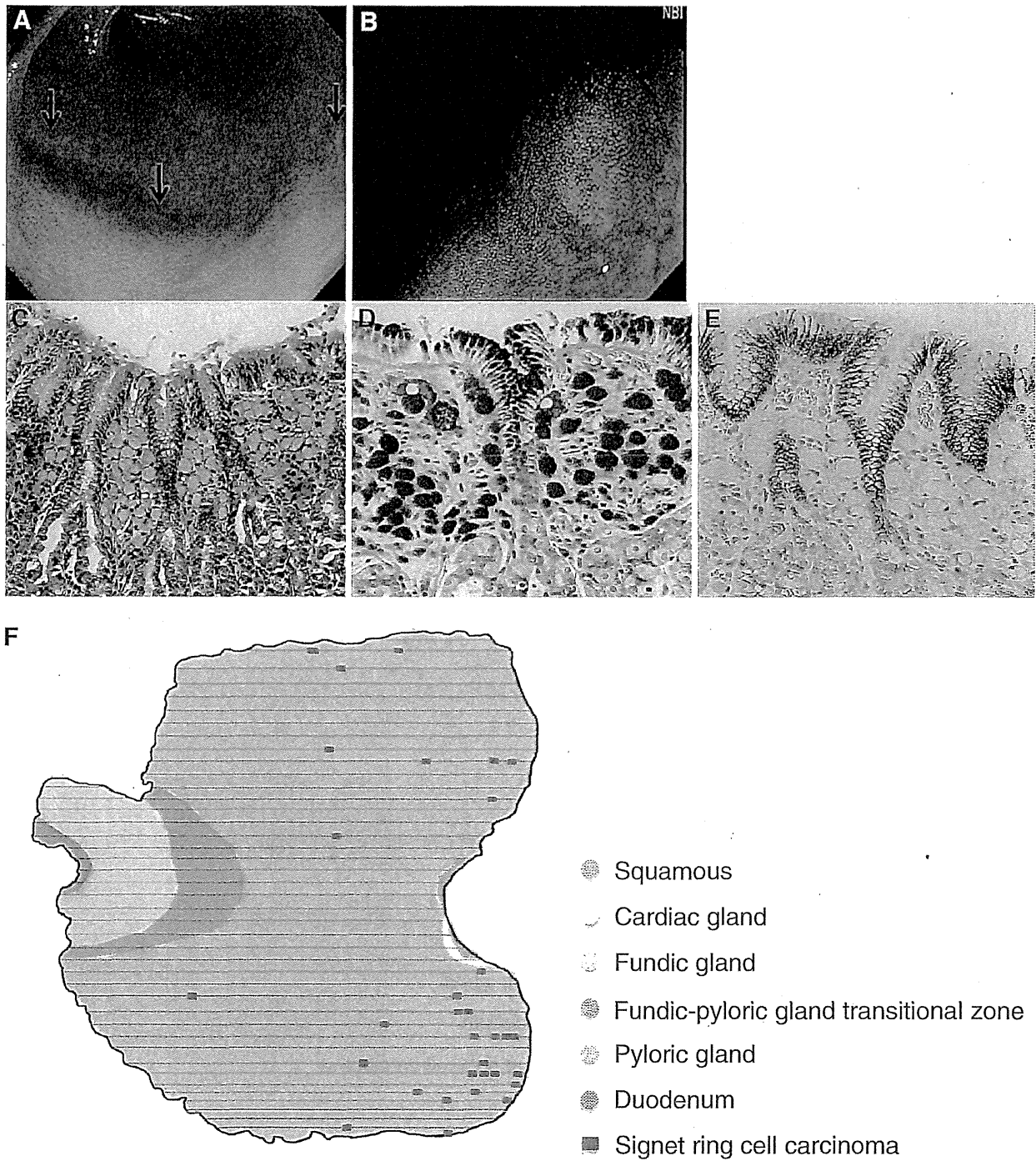
Fig. 2 Clinicopathological findings in patient 1. **a** Conventional endoscopy revealed pale lesions in the greater curvature of the remnant stomach (*arrow*). **b** Magnifying endoscopy with narrow-band imaging showed irregular microvessels. **c** Histology of the tumor showing SRCC proliferating in the upper layer of the fundic gland mucosa. **d** Periodic acid-Schiff (PAS) staining highlighted intracytoplasmic mucin in SRCC. **e** Immunohistochemistry for E-cadherin. Although normal gastric epithelial cells showed clear membrane staining, SRCCs showed loss of immunoreactivity. **f** Gastrectomy mapping study. Approximately 200 SRCC foci were observed in the resected specimen, predominantly near the greater curvature

Squamous

Cardiac gland

Fundic gland

Fundic-pyloric gland transitional zone

Pyloric gland

Duodenum

Signet ring cell carcinoma

Fig. 3 Clinicopathological findings in patient 2. a Conventional endoscopy revealed pale lesions at the body-antrum junction of the stomach (arrows). b Magnifying endoscopy with narrow-band imaging. c Histological analysis showed SRCC in the fundic gland mucosa. d PAS staining highlighted intracytoplasmic mucin in SRCC. e Immunohistochemistry revealed lack of E-cadherin expression in SRCC. f Gastrectomy mapping study. A total of 32 signet ring cell carcinoma foci were detected exclusively in the fundic gland area. The finding was similar to that in patient 1

Complete Labeling Kit (Agilent Technologies), and hybridized to a SurePrint G3 Cancer CGH + SNP 4 × 180K microarray (Agilent Technologies). The microarray was scanned with an Agilent G2565BA microarray scanner (Agilent Technologies), and the scanned data were processed using the Agilent CytoGenomics

Fig. 4 A 275-kb deletion involving exons 7–16 of *CDH1*. **a** Array CGH profiles obtained from peripheral leukocyte DNA of patient 1, patient 2, and a healthy control (33-year-old Japanese male without history of gastric cancer or family history of gastric cancer). The profiles of patients 1 and 2 showed a 275-kb deletion. Deleted regions and the *CDH1* locus are shown by a *green square* and a *red bar*, respectively. A deletion of an exactly identical region was observed in both patients 1 and 2. **b** The location of the breakpoint within the *CDH1* gene. The 5′-most probe of the deleted region (probe y, A_16_P03169109) was located on exon 7, and the 3′-most probe of the intact region (probe x, A_16_P03169108) was located on exon 6. **c** Copy numbers of *CDH1* exons 4, 6, 7, 8, 10, 13, and 16 in patient 1, Pptient 2, and a healthy control analyzed by TaqMan copy number assays. Deletion of *CDH1* exons 7–16 was confirmed in patients 1 and 2. The TaqMan probes for exons 4, 6, 7, 8, 10, 13, and 16 were Hs00296979_cn, Hs01313772_cn, Hs01453316_cn, Hs00466712_cn, Hs01438786_cn, Hs01952025_cn, and Hs02507752_cn, respectively. For the seven exons above, copy numbers of patients 1 and 2 relative to a healthy control were calculated by ΔΔCt method
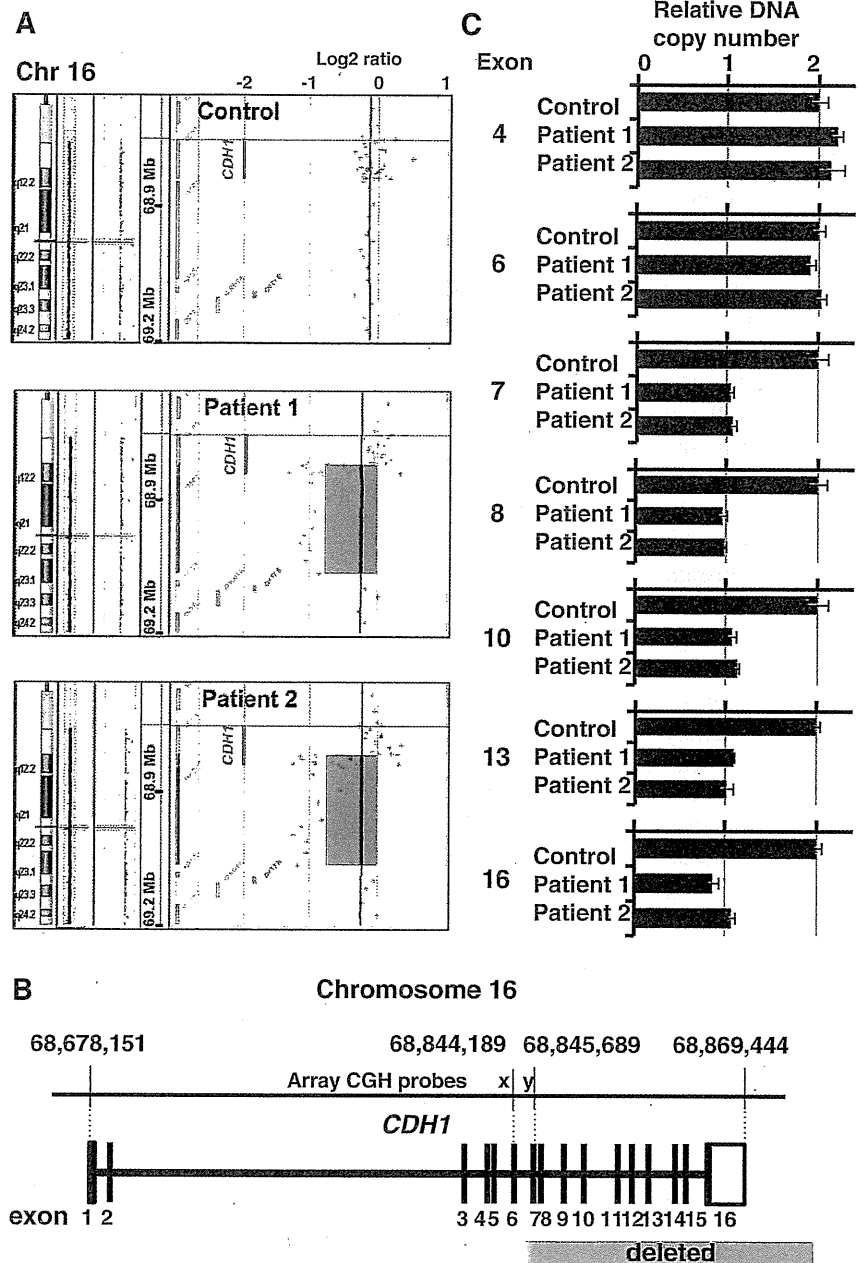
Software (Agilent Technologies). Regions that were covered by five probes or more and showed a mean log2 ratio >0.5 or <−0.5 were considered to have a gain or loss of the copy numbers, respectively. The CGH analysis revealed a 275-kb deletion in both patients 1 and 2 that involved exons 7–16 of *CDH1* (Fig. 4a, b). There were no other genomic regions that had altered copy numbers commonly in patients 1 and 2 but not in a Japanese healthy control (Supplementary Table 1). The deletion of exons 7–16 of *CDH1* was further confirmed by copy number analysis of *CDH1* exon 4, 6, 7, 8, 10, 13, and 16 using TaqMan copy

number assays (Life Technologies, Grand island, NY) (Fig. 4c).

Discussion

We presented a Japanese HDGC family with a 275-kb deletion involving exons 7–16 of *CDH1*. From the initial EGD, we strongly suspected patient 1 to suffer from HDGC according to the clinical course and histopathological findings, which, although they did not fulfill the first

diagnostic criteria for HDGC formulated by the International Gastric Cancer Linkage Consortium (IGCLC) in 1999 [11], fulfilled the criteria updated in 2009 [2], individuals with diffuse gastric cancer before the age of 40 with or without family history. We therefore recommended the proband's offspring to undergo EGD, and one son had three tiny pale areas at the body-antrum junction of the stomach that were confirmed to be SRCCs. Other than the tiny pale patches on endoscopy, there were no other endoscopic findings suggestive of SRCC. In the IGCLC guidelines, it is recommended that any endoscopically visible lesions should be targeted and that random sampling of six biopsies should additionally be taken for each of the following anatomical zones: antrum, transitional zone, body, fundus, and cardia. In total, a minimum of 30 biopsies is recommended. However, we were able to perform successful targeted biopsies of a minimal number of biopsy sites in patient 1 ($n = 6$) and 2 ($n = 3$) because good preparation was obtained as previously described [12]. Based on the presence of multiple SRCCs, patient 2 was also diagnosed with HDGC and was successfully treated with total gastrectomy.

The existence of numerous SRCCs strongly indicated that this family has HDGC. Although the expression of E-cadherin was absent or reduced in the tumor cells, we were unable to detect any small mutations in any exons of CDH1 or to detect aberrant methylation of CDH1 in DNA collected from the peripheral leukocytes of the two patients. Generally, 25–30 % of HDGC families are known to have germline mutations of the CDH1 gene [2]; 77.9 % of all reported CDH1 germline mutations are truncating mutations, and their locations span the whole length of the CDH1 gene without any hot spots [13]. At the same time, 4 % of reported CDH1 germline mutations exhibit large deletions that are not detectable by conventional DNA sequencing [14]. Therefore, we continued to search for a large deletion of the CDH1 gene in patients 1 and 2, and, by CGH analysis, we detected a pathogenic 275-kb deletion in both patients 1 and 2. To our knowledge, this is the first case of HDGC harboring a large germline deletion in the range of hundreds of kilobases involving CDH1.

GCs have marked geographic variation in their incidence, with high incidences in East Asian countries and low incidences in Western countries. On the other hand, the majority of HDGC with CDH1 mutations have been reported in the Western countries. To our knowledge, there have been only two Japanese HDCG families with germline CDH1 mutations reported so far [7]. However, it still remains unclear whether HDCG truly occurs less in Japan than in Western countries. The rarity of HDGC in Japan might be due to insufficient surveillance rather than reflecting the actual incidence. The high incidence of sporadic GCs is likely to increase "false-positive" familial

GCs and make it difficult to identify hereditary GCs. The IGCLC determined diagnostic criteria for HDGC, but these criteria are likely to be too broad to be used in areas with high GC incidence, including Japan [2, 11]. In addition, the prevalence of large deletions, as suggested by the study of Yamada et al. [7] and here, might have made detection of the CDH1 mutation difficult.

Although HDGC has been regarded as rare in East Asia, these two recent reports show that there are HDGC families in East Asia and suggest the importance of the analysis of large deletions for cases with multiple SRCCs. Genetic counseling and testing may be advised for patients with multiple SRCCs and their family members even in East Asia.

References

1. Guilford P, Hopkins J, Harraway J, McLeod M, McLeod N, Harawira P, et al. E-cadherin germline mutations in familial gastric cancer. Nature. 1998;392:402–5.
2. Fitzgerald RC, Hardwick R, Huntsman D, Carneiro F, Guilford P, Blair V, et al. Hereditary diffuse gastric cancer: updated consensus guidelines for clinical management and directions for future research. J Med Genet. 2010;47:436–44.
3. Park JG, Yang HK, Kim WH, Caldas C, Yokota J, Guilford PJ. Report on the first meeting of the International Collaborative Group on Hereditary Gastric Cancer. J Natl Cancer Inst. 2000;92: 1781–2.
4. Lynch HT, Grady W, Suriano G, Huntsman D. Gastric cancer: new genetic developments. J Surg Oncol 2005;90:114-33; discussion 33.
5. Wang Y, Song JP, Ikeda M, Shinmura K, Yokota J, Sugimura H. Ile-Leu substitution (I415L) in germline E-cadherin gene (CDH1) in Japanese familial gastric cancer. Jpn J Clin Oncol. 2003;33: 17–20.
6. Kim HC, Wheeler JM, Kim JC, Ilyas M, Beck NE, Kim BS, et al. The E-cadherin gene (CDH1) variants T340A and L599 V in gastric and colorectal cancer patients in Korea. Gut. 2000;47: 262–7.
7. Yamada H, Shinmura K, Ito H, Kasami M, Sasaki N, Shima H, et al. Germline alterations in the CDH1 gene in familial gastric cancer in the Japanese population. Cancer Sci. 2011;102:1782–8.
8. Yamada M, Sekine S, Ogawa R, Taniguchi H, Kushima R, Tsuda H, et al. Frequent activating GNAS mutations in villous adenoma of the colorectum. J Pathol. 2012;228:113–8.
9. Berx G, Cleton-Jansen AM, Nollet F, de Leeuw WJ, van de Vijver M, Cornelisse C, et al. E-cadherin is a tumour/invasion suppressor gene mutated in human lobular breast cancers. EMBO J. 1995;14:6107–15.
10. Asada K, Ando T, Niwa T, Nanjo S, Watanabe N, Okochi-Takada E, et al. FHL1 on chromosome X is a single-hit gastrointestinal tumor-suppressor gene and contributes to the formation of an epigenetic field defect. Oncogene. 2013;32:2140–9.
11. Caldas C, Carneiro F, Lynch HT, Yokota J, Wiesner GL, Powell SM, et al. Familial gastric cancer: overview and guidelines for management. J Med Genet. 1999;36:873–80.
12. Bhandari P, Green S, Hamanaka H, Nakajima T, Matsuda T, Saito Y, et al. Use of Gascon and Pronase either as a pre-endoscopic drink or as targeted endoscopic flushes to improve

visibility during gastroscopy: a prospective, randomized, controlled, blinded trial. Scand J Gastroenterol. 2010;45:357–61.

13. Carneiro F, Oliveira C, Suriano G, Seruca R. Molecular pathology of familial gastric cancer, with an emphasis on hereditary diffuse gastric cancer. J Clin Pathol. 2008;61:25–30.

14. Oliveira C, Senz J, Kaurah P, Pinheiro H, Sanges R, Haegert A, et al. Germline CDH1 deletions in hereditary diffuse gastric cancer families. Hum Mol Genet. 2009;18:1545–55.

Digestion

Endoscopic Mucosal Resection for Middle and Large Colorectal Polyps with a Double-Loop Snare

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Abstract
Background/Aims: This study aimed to analyze the endoscopic mucosal resection (EMR) with a novel uniquely shaped, double-loop snare (Dualoop, Medico's Hirata Inc., Tokyo, Japan) for colorectal polyps. *Methods:* This was a clinical trial conducted in two referral centers, Kyoto Prefectural University of Medicine and National Cancer Center Hospital in Japan. First, the firmness of various snares including 'Dualoop' was experimentally analyzed with a pressure gauge. Five hundred and eighty nine consecutive polyps that underwent EMR with 'Dualoop' were compared to 339 polyps with the standard round snare. Lesion characteristics, en bloc resection, and complications were analyzed. *Results:* 'Dualoop' had the most firmness among the various snares. The average tumor size was 9.3 mm (5–30), and en bloc resection was achieved in 95.4%. The rate of en bloc resection for middle polyps 15–19 mm in diameter was significantly higher with the 'Dualoop' than that with the round snare (97.9 vs. 80.0%, p < 0.05). The rate of en bloc resection was 64.7% for large polyps ≥20 mm in diameter using 'Dualoop'. Higher age, larger tumor size, and superficial polyps were associated with the failure of en bloc resection. *Conclusion:* EMR with 'Dualoop' was effective for resecting both middle and large polyps en-bloc.

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Introduction

Endoscopic mucosal resection (EMR) of adenomas and early colorectal cancer is performed worldwide [1–3]. However, tumors ≥20 mm in diameter are considered difficult candidates for en bloc resection because of the size of snares and the unstable state of mucosal elevation by injection. The rates of en bloc resection with EMR for such lesions are reported to be 25.0–48.1% [3–5]. In contrast, endoscopic submucosal dissection (ESD) enables en bloc resection of large early colorectal cancer lesions [6–9]. However, it can be time consuming and carries a higher risk of perforation than does EMR. For improving en bloc resections of large colorectal polyps in EMR, various injection solutions have been used to achieve sustained and higher mucosal elevation. Glycerol and dextrose provide better complete resection rates and longer-lasting mucosal elevation than does normal saline (NS), and these liquids are used for clinical EMR in Japan [10, 11].
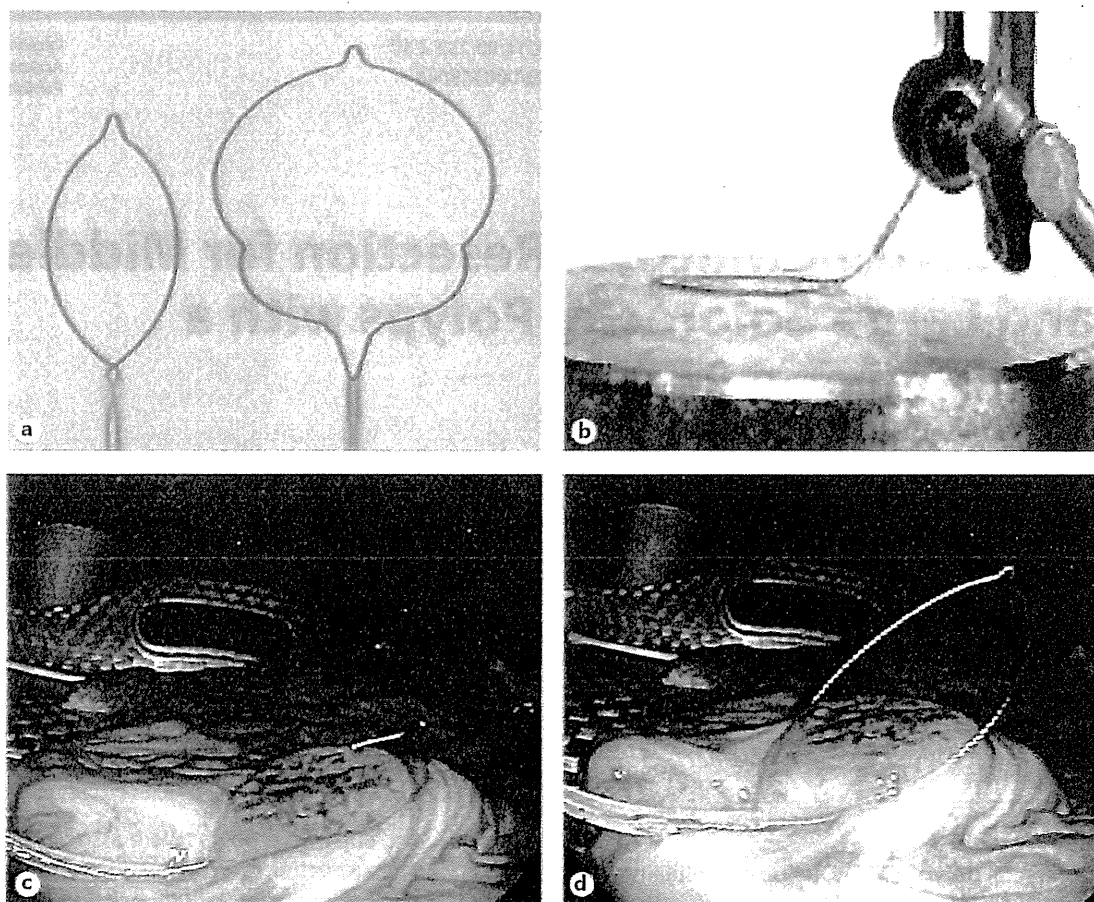
Fig. 1. Firmness of snare with respect to mucosal resection in the experimental model. **a** The shape of 'Dualoop' with double loops. **b** A snare is pushed into a pressure gauge at a 45-degree angle to calculate its pressure. **c** Pressure applied by the snare is more than the hardness of the mucosal elevation caused by HA. **d** Snaring is difficult when the hardness of the mucosal elevation caused by HA is more than pressure applied by the snare.

On the other hand, Yamamoto et al. first reported the efficacy of hyaluronic acid (HA) during a novel endoscopic resection technique for large colorectal polyps [12]. HA has been shown to create higher and more sustainable mucosal elevation than does NS [13–15]. A prospective randomized controlled trial about the efficacy of 0.13% HA in colorectal EMR proved that using 0.13% HA rather than NS during EMR was more effective for complete resection [16]. On the other hand, the viscosity of HA can make snaring difficult, especially in large polyps. Using a firm snare can overcome this problem in EMR using HA [16]. Recently, a novel uniquely shaped snare (Dualoop; Medico's Hirata Inc., Tokyo, Japan) was introduced. 'Dualoop' as its name suggests has a 'double loop' shape in one snare and is firm due to this unique shape. This novel feature may allow effective resection of large colorectal polyps. In the current study, we assessed the firmness of various snares used in EMR and analyzed the efficacy of EMR for middle and large colorectal polyps using 0.13% HA and 'Dualoop'.

Methods

Assess the Firmness of Various Snares

Various snares such as 'Dualoop', 'Captivator II' 33 mm in size (Boston Scientific Co., Tokyo, Japan), 'Captivator' 13 and 27 mm in size (Boston Scientific Co.), 'Captivator Small Hex' 13 mm in size (Boston Scientific Co.), 'SnareMaster' 15 and 25 mm in size (Olympus Medical Co., Tokyo, Japan), 'Spiral Snare' 20 mm in size (Olympus Medical Co.), 'Rotatable Snare' 13 and 20 mm in size (Olympus Medical Co.) were used. 'Dualoop' has two loops in one snare (fig. 1a). The diameter of the small loop is 16 mm and that of the large loop is 33 mm. A snare was pushed into a pressure

gauge at an angle of 45 degrees to determine its pressure (fig. 1b). The same experiment was performed twice for each snare. In general, pressure applied by the snare is more than the hardness of the mucosal elevation caused by HA (fig. 1c). On the contrary, snaring is difficult when the hardness of the mucosal elevation is more than the firmness of snare (fig. 1d). HA injection makes a hard mucosal elevation and, therefore a firmer snare is needed for definitive resection.

Effectiveness of 'Dualoop' for EMR of Colorectal Polyps

A multicenter prospective open label trial was performed on EMR with 0.13% HA and 'Dualoop' at the Kyoto Prefectural University of Medicine and National Cancer Center Hospital in Japan. Lesion characteristics and various other measurements were analyzed in 589 consecutive polyps in 345 patients who underwent EMR for colorectal polyps from January 2013 to July 2013 (Dualoop group). As a comparison, 339 consecutive polyps in 174 patients who underwent EMR for colorectal polyps using 0.13% HA and standard round snare from July 2013 to December 2013 were analyzed (Round snare group). Only 'SnareMaster' was used in the Round snare group. All patients provided written informed consent to undergo EMR, and the study was approved by each institution's review board, including the ethics committees of Kyoto Prefectural University of Medicine and the National Cancer Center Hospital. This study was carried out in accordance with the World Medical Association Helsinki Declaration and was registered in the University Hospital Medical Information Network Clinical Trials Registry (UMIN-CTR) as number UMIN000010152. The inclusion criteria were the presence of a neoplastic colorectal polyp ≥5 mm in diameter that had been diagnosed using a double-contrast barium enema or colonoscopy. Recurrent lesions after previous EMR, lesions diagnosed as deeply invasive submucosal cancer by colonoscopy, and non-neoplastic polyps were excluded. Similarly, colorectal polyps ≥31 mm were also excluded because they were not appropriate for EMR, and ESD was necessary for effective resection of these lesions. Some colorectal polyps 20–30 mm in diameter and lesions for which en bloc resection by EMR was considered difficult because of location, morphology, and fibrosis were decided for ESD, and those lesions were excluded from this study [17]. In terms of morphology, polyps were divided into the protruding and superficial type; protruding-type polyps were divided into the sessile and pedunculated type in accordance with the Japanese Classification of Colorectal Carcinoma [18]. The size of a polyp was defined as its maximum diameter and was calculated relative to the size of the snare and injection needle. If the histopathological diagnosis after EMR indicated a non-neoplastic polyp, the polyp was excluded from the analysis.

EMR with 'Dualoop' (fig. 2)

'Dualoop' made of a special wire has the above-mentioned unique shape, which makes it firm. The two-loop configuration is useful as the small loop can be used for polyps <15 mm in diameter and the large loop for polyps ≥15 mm in diameter. In general, it is impossible to resect large polyps en bloc with a small standard round snare. Similarly, a standard large snare is not appropriate for resecting small polyps because of the difficulty in maneuvering, especially for less-experienced endoscopists. Thus, both a small and a large snare are essential for the resection of small and large polyps. The two loops of 'Dualoop' have this combined facility to resect small and large polyps without having to exchange snares frequently.

A lower gastrointestinal endoscope with a single channel ((EC-590MP, Fujifilm Medical Co., Tokyo, Japan), and PCF-Q260AZI or CF-H260AZI (Olympus Optical Co., Ltd., Tokyo, Japan)) were used. The procedures were performed by 35 endoscopists at the two institutions, whose experience with colonoscopy ranged from 500 to 15,000 cases. Patients took sodium picosulfate the day before the examination and 2.0 l of polyethylene glycol solution in the morning before the examination to prepare their bowel. The injection solution was prepared from a 0.4% HA solution (Mucoup; Johnson & Johnson, Tokyo, Japan or Seikagaku Corporation, Tokyo, Japan), which was approved by the Japanese national health insurance system as an injection solution for EMR. The 0.13% HA solution was prepared by diluting a 0.4% HA solution with NS. The 0.13% HA was injected using a 25-gauge needle. A small amount of indigo carmine was added to the solution to facilitate the visualization of the mucosal elevation. No epinephrine was added to the injection solutions. 'Dualoop' was used in conjunction with an automatically controlled high-frequency generator (Swift 40W effect 3, VIO300D; Erbe Elektromedizin, Ltd., Tübingen, Germany). Endoscopists decided which loops to use according to the situation in the Dualoop group. Mostly, the large loop was used for polyps ≥15 mm in diameter and the small loop for polyps <15 mm in diameter. On the other hand, two sizes of snare such as 15 and 25 mm in 'SnareMaster' were used in the Round snare group and endoscopists chose the size of snare appropriately according to the size of polyps.

Histopathological Evaluation

Histopathological diagnosis was based on the World Health Organization classification scheme [19]. Adenoma was diagnosed according to mild-to-severe cytological and architectural features. The term 'intramucosal adenocarcinoma' was applied to lesions that showed histopathological evidence of invasion into the lamina propria or muscularis mucosa but not into the submucosa. Complete resection was defined histopathologically, as the absence of tumor in the lateral and vertical margins of the resected specimens.

Clinical Outcomes

We analyzed the characteristics of the lesions and several measurements of clinical outcomes, including en bloc resection, histopathologically defined complete resection, postoperative hemorrhage and perforation rate in the Dualoop group and the Round snare group. Postoperative hemorrhage was defined as hematochezia that required endoscopic treatment for hemostasis. Perforation was detected by endoscopy during the EMR procedure or by abdominal computed tomography after EMR. Moreover, the number of patients with both 5–14 mm and ≥15 mm polyps were calculated to examine the effects of 'Dualoop' as follows. With 'Dualoop', a single snare can be used to resect both small and large polyps, whereas two separate sizes of standard round snares are required for a similar EMR. As a subgroup analyses, the clinical outcomes of polyp size were examined in small polyps 5–14 mm, middle polyps 15–19 mm, and large polyps ≥20 mm in diameter in the Dualoop group and the Round snare group. Similar clinical outcomes were analyzed for colorectal polyps ≥20 mm in diameter in which pedunculated polyps were excluded. We also analyzed which factors are related to the failure of en bloc resection in the colorectal polyps ≥20 mm in diameter in the Dualoop group. Furthermore, the relationship between the experience of endoscopists and the clinical characteristics were analyzed in the Dualoop
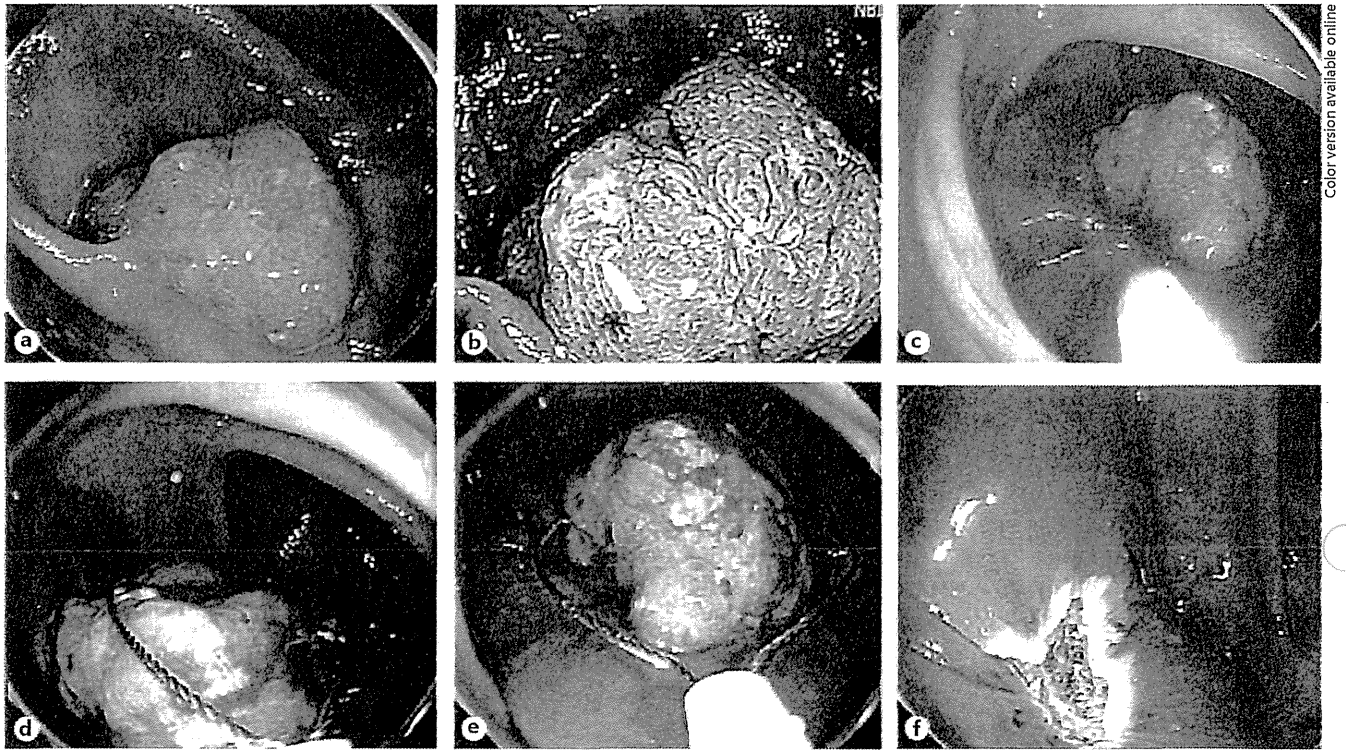
Fig. 2. Endoscopic mucosal resection with 'Dualoop'. **a** Colorectal polyp 0-Isp 25 mm on the ascending colon. **b** Narrow band imaging showing an irregular surface pattern and a vascular pattern. **c** Eight milliliters of 0.13% HA was injected and high mucosa elevation was achieved. **d** The small loop of 'Dualoop' was not appropriate for this lesion. **e** The size of the large loop of 'Dualoop' was bigger than the size of this lesion. **f** En bloc resection was performed. Histopathological diagnosis showed submucosally invasive cancer. Both the horizontal and vertical margins were negative.

group. The endoscopists were divided into the less experienced group and the experienced group according to the number of colonoscopies they had previously performed. Of the 35 endoscopists who participated in the study, 26 were classified as less experienced (having performed <5,000 colonoscopies) and 9 as experienced (having performed ≥5,000 colonoscopies).

Statistical Analysis
Statistical analyses were performed using the Mann-Whitney U test and the chi-square test (StatView 5.0, HULINKS). Continuous variables such as patient age and tumor size were analyzed using the Mann-Whitney U test. Categorical variables such as the rate of complete resection and other endpoints were analyzed using the chi-square test. A p value of <0.05 was considered statistically significant.

Results

In view of the firmness of snares, the large loop (33 mm) of 'Dualoop' (average pressure: 111 mN) was firmer than other 20–33 mm snares (average pressure: 75 mN,

range 50–100 mN) and the small loop (16 mm) of 'Dualoop' (average pressure: 187 mN) was firmer than other 13–15 mm snares (average pressure: 97 mN, range 55–135 mN) (fig. 3).

In this study, the average tumor size was 9.3 mm (5–30), and en bloc resection was achieved in 562 of the total 589 lesions (95.4%) in the Dualoop group (table 1). On the other hand, the rate of en bloc resection was 97.0% in the Round snare group. There were no significant differences in rates of en bloc resection and complete resection in the two groups. The rates of perforation and postoperative hemorrhage in the Dualoop group were 0 and 0.5% respectively. Thirty-eight of 345 patients (11.0%) had both polyps of 5–14 and ≥15 mm in diameter.

The results of subgroup analysis are shown in table 2. In the Dualoop group, the en bloc resection was achieved for 484 of 497 lesions (97.3%) in polyps 5–14 mm in diameter, 47 of 48 lesions (97.9%) in polyps 15–19 mm in diameter, 31 of 44 lesions (84.8%) in polyps ≥20 mm in diameter, and 22 of 34 lesions (64.7%) in polyps ≥20 mm

Colorectal EMR with a Double-Loop
Snare

Table 1. Clinical characteristics of endoscopic mucosal resection with 'Dualoop' for 598 colorectal polyps

|	Dualoop	Round snare	p
Number of tumors	589	339	
Median age (range)	67.3 (24–95)	68.3 (24–88)	n.s.
Patient number (male/female)	345 (230/115)	174 (117/57)	
Tumor size, mm	9.3 (5–30)	8.8 (5–30)	n.s.
Location (right-sided/left-sided/rectum)	311/214/64	185/124/30	n.s.
Morphology (protruding (peduncluated) or superficial)	433 (50)/156	252 (35)/88	n.s.
En bloc resection	95.4%, 562/589	97.0%, 329/340	n.s.
Histopathological diagnosis (Ad/M/SM)	520/52/17	307/26/6	n.s.
Complete resection	87.5%, 515/589	89.4%, 311/339	n.s.
Perforation	0	0	n.s.
Postoperative hemorrhage	0.5%, 3/589	1.1%, 4/339	n.s.
Patient with small and large polyps	11.0%, 38/345	9.8%, 17/173	n.s.

Ad = Adenoma; M = intramucosal cancer; SM = submucosally invaded cancer; n.s. = not significant.

Table 2. Clinical characteristics of colorectal endoscopic mucosal resection according to polyp size

D	5–14		15–19		≥20		≥20 without pedunculation	
	dualoop	round snare	dualoop	round snare	dualoop	round snare	dualoop	round snare
Number	497	298	48	25	44	16	34	7
Average tumor size, mm	7.5	7.5	15.4	15.8	22.2	23.2	22.0	21.1
Location (right-sided/left-sided/rectum)	270/172/55	170/102/26	22/23/3	11/12/2	19/19/6	4/10/2	18/12/4	3/3/1
Morphology (protruding (pedunculated) or superficial)	376 (31)/121	221 (18)/77	27 (9)/21	18 (8)/7	30 (10)/14	12 (9)/4	20/14	3/4
En bloc resection	97.3% (484/497)	99.6% (297/298)	97.9% (47/48)*	80.0% (20/25)**	84.8% (31/44)	82.3% (13/16)	64.7% (22/34)	57.1% (4/7)
Complete resection	89.4% (444/497)	91.3% (272/298)	91.7% (44/48)	79.2% (19/25)	61.4% (27/44)	75.0% (12/16)	52.9% (18/34)	42.9% (3/7)
Histopathological diagnosis (Ad/M/SM) (rates of cancer)	458/32/7 (7.8%)	275/19/4 (7.6%)	36/10/2 (25.0%)	21/3/1 (16.0%)	26/10/8 (40.9%)	11/4/1 (31.2%)	22/6/6 (35.3%)	6/1/0 (14.2%)

* vs. ** p = 0.02. Ad = Adenoma; M = intramucosal cancer; SM = submucosally invaded cancer.

Fig. 3. Firmness of 'Dualoop' and other snares. The large loop of 'Dualoop' 33 mm in diameter (average pressure: 111 mN) is firmer than other 20–33 mm snares (average pressure: 75 mN, range 50–100 mN). The small loop of 'Dualoop' 16 mm in diameter (average pressure: 187 mN) is firmer than other 13–16 mm snares (average pressure: 97 mN, range 55–135 mN).

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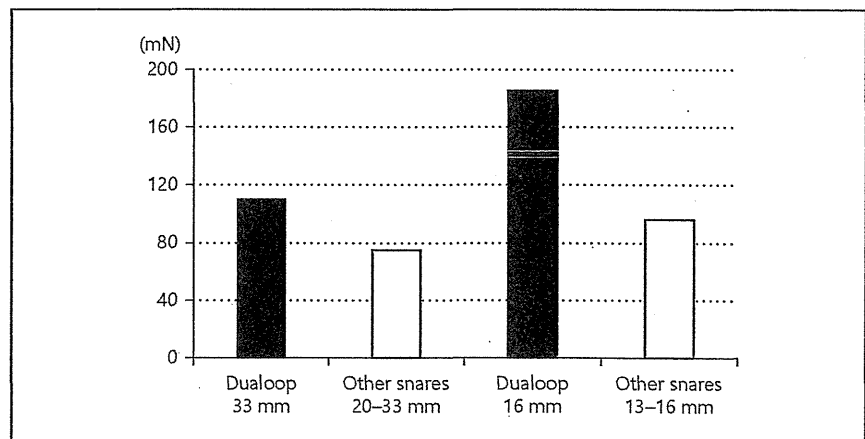
Table 3. Factors of failure of en bloc resection for endoscopic mucosal resection with 'Dualoop' for colorectal polyps 20 mm ≤ in diameter

	Success of en bloc resection	Failure of en bloc resection	p
Number of tumors	31	13	
Median age (range)	65.1 (50–83)	69.4 (58–91)	<0.05
Tumor size, mm	21.9 (20–30)	22.7 (20–30)	<0.05
Location (right-sided/left-sided/rectum)	13/15/3	6/4/3	n.s.
Morphology (protruding (pedunculated) or superficial)	24 (9)/7	6 (1)/7	<0.05
Histopathological diagnosis (Ad/M/SM) (rate of cancer)	18/8/5 (41.9%)	8/2/3 (23.1%)	n.s.

Ad = Adenoma; M = intramucosal cancer; SM = submucosally invaded cancer; n.s. = not significant.

Table 4. Clinical characteristics of colorectal endoscopic mucosal resection according to the experience of the endoscopist in the Dualoop group

	Less-experienced endoscopist	Experienced endoscopist	p
Number of tumors	287	302	
Median age	68.0	66.6	n.s.
Tumor size, mm	8.5±4.2	10.0±5.2	<0.05
Location (right-sided/left-sided/rectum)	136/111/40	175/103/24	n.s.
Morphology (protruded (pedunculated) or superficial)	239 (35)/48	194 (15)/108	n.s.
En bloc resection	94.7% (272/287)	96.0% (290/302)	n.s.
Histopathological diagnosis (Ad/M/SM) (rate of cancer)	241/35/11 (15.9%)	279/17/6 (7.6%)	<0.05
Complete resection	83.0% (238/287)	92.2% (277/302)	<0.05
Perforation	0%	0%	n.s.
Postoperative hemorrhage	0.7%	0.3%	n.s.

Ad = Adenoma; M = intramucosal cancer; SM = submucosally invaded cancer; n.s. = not significant.

in diameter without pedunculation (table 2). There was a significant difference about the rate of en bloc resection for middle polyps 15–19 mm in diameter between the Dualoop group and the round snare group (p = 0.02). There were no significant differences in the other rates in the two groups.

In the Dualoop group, colorectal polyps ≥20 mm in diameter that were successfully removed by en bloc resection were compared with lesions that could not be removed by en bloc resection (table 3). There were significant differences in median age, tumor size, and morphology between the two groups.

The rate of en bloc resection was similar for less-experienced and experienced endoscopists (94.7 and 96.0%) (table 4). The rate of complete resection was higher for experienced endoscopists than for less-experienced endoscopists (92.2% vs. 83.0%; p < 0.05).

Discussion

According to our results, larger snares are softer than smaller snares. Snaring is difficult when pressure applied by the snare is not more than the hardness of the mucosal elevation caused by the injection liquid. The hardness of the mucosal elevation is dependent on the kind of injection liquid, and HA makes the hard mucosal elevations. According to our estimation, snares should be chosen according to the hardness of mucosal elevation and the size of the polyps. 'Dualoop' has two different sizes of firm loops in the same snare and is the most appropriate for EMR using HA.

In our previous multicenter study of 624 colorectal polyps, EMR with various conventional round and oval snares and 0.13% HA showed rates of en bloc resection of 93.3% for colorectal polyps 5–20 mm in diameter (aver-

age tumor size, 8.3 mm) [22]. This rate was almost similar to the rate of en bloc resection (95.4%) for colorectal polyps 5–30 mm in diameter (average tumor size, 9.3 mm) using 'Dualoop' in the current study. On the other hand, the rate of en bloc resection for middle colorectal polyps 15–19 mm in diameter (average tumor size, 15.4 mm) using 'Dualoop' was 97.9% and it was higher than that using round snare (80.0%). In the previous multicenter study described above, the rate of en bloc resection for colorectal polyps 11–20 mm in diameter (average tumor size, 14.1 mm) was 85.1%. Thus, our data suggests that 'Dualoop' may be more successful at removing middle size polyps in en bloc resection compared to round snares. This was probably due to its firmness and unique shape. Additionally, the two loop configuration of 'Dualoop' is useful, such that endoscopists can choose each loop according to the polyp size at the timing of resection. On the other hand, with standard round snares, endoscopists have to choose separate small or big snares according to the size of polyps during the procedure. Due to the time constraints and economical reasons endoscopists would try to perform EMR using a single snare as much as possible, making resection of some polyps difficult. Middle polyps 15–19 mm in diameter require small-sized snares and these lesions are sometimes ineffectively resected by inappropriate sized snare. With respect to another effectiveness of 'Dualoop' with two configurations, 11.0% of patients had both a small polyp 5–14 in diameter and a large polyp more than 15 mm in diameter. In these patients, 'Dualoop' can be effectively used to resect all the polyps using one snare.

Previous reports revealed the rates of en bloc resection for polyps ≥20 mm to be 25.0–48.1% [2–4]. In our study, the rate of en bloc resection for colorectal polyps ≥20 mm in diameter without pedunculation (average tumor size, 22.2 mm) was as high as 64.7% although the average tumor size in this study was smaller than in previous studies. EMR with 'Dualoop' and 0.13% HA is expected to be useful for resecting large colorectal polyps. However, there were no significant differences between the Dualoop group and the round snare group. This may possibly be due to the lack of comparison of cases. Further examination should be performed to analyze the effectiveness of EMR with 'Dualoop' for larger lesions.

In view of the experience of endoscopists, our previous study showed that the EMR procedure time (using 0.13% HA) of less-experienced endoscopists for colorectal polyps 5–20 mm in diameter was longer than that of experienced endoscopists (2.4 ± 1.4 vs. 1.9 ± 1.4 min; $p < 0.01$) [20]. However, less-experienced endoscopists achieved a high rate of en bloc resection and complete resection similar to that of veteran endoscopists (en bloc resection: 92.2% vs. 94.8%, complete resection: 75.0% vs. 81.3%). Less-experienced endoscopists achieved a high quality of EMR similar to what experienced endoscopists achieved because HA can maintain mucosal elevation longer than do NS. In our present study, the rate of en bloc resection by less-experienced endoscopists was similar to that achieved by experienced endoscopists (94.7% vs. 96.0%; NS). With respect to complete resection, less-experienced endoscopists could not resect colorectal polyps with definite margin though experienced endoscopists could do that (83.0% vs. 92.2%; $p < 0.05$). According to the increase of experiences of EMR, the rate will be expected to improve. When considering complications, no perforation occurred in both groups and the rates of postoperative hemorrhage for less-experienced endoscopists were similar to those achieved by experienced endoscopists. We suggest that 'Dualoop' is useful for less-experienced endoscopists to get high quality of EMR without complication.

Unfortunately, EMR with 0.13% HA and 'Dualoop' did not achieve en bloc resection, especially in large lesions. For those lesions, the main reason for the incomplete en bloc resection was considered to be inappropriate and low mucosal elevation due to severe fibrosis, difficult locations, excessive amount of injection liquid, and long procedure times. Additionally, our previous study about round snares showed risk factors for failure of enbloc resection to colorectal polyps. Multivariate analysis showed that polyp 11–20 mm in size and location not in the left-side colon was significantly independent risk factors for failure of enbloc resection. ESD enables performing en bloc resection for such difficult and large lesions, although this procedure requires many devices, including special knives, and a longer procedure time. Another alternative treatment for those lesions is EMR with circumferential incision [21, 22]. This method is useful for resecting large colorectal polyps, and does not need special knives other than the standard snare used for EMR. Sakamoto et al. reported that the rate of en bloc resection for colorectal polyps 20–40 mm in diameter by using this method was 67% and the average procedure time was 40 min [21]. Toyonaga et al. called this method 'simplified ESD', and the rate of en bloc resection was 90.9% (average tumor size, 17 mm) [22]. The average procedure time was 27 min, and it was shorter than that for ESD. 'Dualoop' is a firm snare and it may be useful for this EMR with circumferential incision.

In conclusion, 'Dualoop' was the firmest snare and it was appropriate for mucosal elevation produced by HA.

EMR with 'Dualoop' was effective for resecting smaller polyps as well as larger polyps. A higher rate of en bloc resection is achieved in EMR for middle colorectal polyps 15–19 mm in diameter. Additionally, it contributes to high quality of EMR for less-experienced endoscopists.

Limitations

It remains possible that the inclusion of 35 endoscopists performing EMR in this study introduced some bias. There were a limited number of colorectal polyps ≥20 mm in diameter in the Dualoop group and the round snare group. Additionally, in both groups, some colorectal polyps 20–30 mm in diameter and lesions for which en bloc resection by EMR was considered difficult because of location, morphology, and fibrosis were decided for ESD, and those lesions were excluded from this study. No follow-up colonoscopy was performed to assess the recurrence.

References

1 Kudo S: Endoscopic mucosal resection of flat and depressed types of early colorectal cancer. Endoscopy 1993;25:455–461.
2 Saito Y, Fujii T, Kondo H, et al: Endoscopic treatment for laterally spreading tumors in the colon. Endoscopy 2001;33:682–686.
3 Tada M, Shimada M, Murakami F, et al: Development of the strip-off biopsy (in Japanese with English abstract). Gastroenterol Endosc 1984;26:833–839.
4 Saito Y, Fukuzawa M, Matsuda T, et al: Clinical outcome of endoscopic submucosal dissection versus endoscopic mucosal resection of large colorectal tumors as determined by curative resection. Surg Endosc 2010;24:343–352.
5 Tanaka S, Haruma K, Oka S, et al: Clinicopathologic features and endoscopic treatment of superficially spreading colorectal neoplasms larger than 20 mm. Gastrointest Endosc 2001;54:62–66.
6 Saito Y, Uraoka T, Yamaguchi Y, et al: A prospective, multicenter study of 1111 colorectal endoscopic submucosal dissections (with video). Gastrointest Endosc 2010;72:1217–1225.
7 Saito Y, Uraoka T, Matsuda T, et al: Endoscopic treatment of large superficial colorectal tumors: a case series of 200 endoscopic submucosal dissections (with video). Gastrointest Endosc 2007;66:966–973.
8 Yoshida N, Yagi N, Naito Y, et al: Safe procedure in endoscopic submucosal dissection for colorectal tumors focused on preventing complications. World J Gastroenterol 2010; 16:1688–1695.

9 Yoshida N, Wakabayashi N, Kanemasa K, et al: Endoscopic submucosal dissection for colorectal tumors: technical difficulties and rate of perforation. Endoscopy 2009;41:758–761.
10 Uraoka T, Fujii T, Saito Y, et al: Effectiveness of glycerol as a submucosal injection for EMR. Gastrointest Endosc 2005;61:736–740.
11 Varadarajulu S, Tamhane A, Slaughter RL: Evaluation of dextrose 50% as a medium for injection-assisted polypectomy. Endoscopy 2006;38:907–912.
12 Yamamoto H, Yube T, Isoda N, et al: A novel method of endoscopic mucosal resection using sodium hyaluronate. Gastrointest Endosc 1999;50:251–256.
13 Hyun JJ, Chun HR, Chun HJ, et al: Comparison of the characteristics of submucosal injection solutions used in endoscopic mucosal resection. Scand J Gastroenterol 2006;41:488–492.
14 Fujishiro M, Yahagi N, Kashimura K, et al: Comparison of various submucosal injection solutions for maintaining mucosal elevation during endoscopic mucosal resection. Endoscopy 2004;36:579–583.
15 Hirasaki S, Kozu T, Yamamoto H, et al: Usefulness and safety of 0.4% sodium hyaluronate solution as a submucosal fluid 'cushion' for endoscopic resection of colorectal mucosal neoplasms: a prospective multi-center open-label trial. BMC Gastroenterol 2009;9:1.

16 Yoshida N, Naito Y, Inada Y, et al: Endoscopic mucosal resection with 0.13% hyaluronic acid solution for colorectal polyps less than 20 mm: a randomized controlled trial. J Gastroenterol Hepatol 2012;27:1377–1383.
17 Tanaka S, Oka S, Kaneko I, et al: Endoscopic submucosal dissection for colorectal neoplasia: possibility of standardization. Gastrointest Endosc 2007;66:100–107.
18 Japanese Society for Cancer of the Colon and Rectum: Japanese Classification of Colorectal Carcinoma, English ed 2. Kanehara & Co., Ltd., Tokyo, 2009.
19 Hurlstone DP, Fu KI, Brown SR, et al: EMR using dextrose solution versus sodium hyaluronate for colorectal Paris type I and 0–II lesions: a randomized endoscopist-blinded study. Endoscopy 2008;40:110–114.
20 Yoshida N, Naito Y, Inada Y, et al: Multicenter study of endoscopic mucosal resection using 0.13% hyaluronic acid solution of colorectal polyps less than 20 mm in size. Int J Colorectal Dis 2013;28:985–991.
21 Sakamoto T, Matsuda T, Nakajima T, et al: Efficacy of endoscopic mucosal resection with circumferential incision for patients with large colorectal tumors. Clin Gastroenterol Hepatol 2012;10:22–26.
22 Toyonaga T, Man-I M, Morita Y, et al: The new resources of treatment for early stage colorectal tumors: EMR with small incision and simplified endoscopic submucosal dissection. Dig Endosc 2009;21(suppl 1):S31–S37.