

# Diagnosis of neoplastic and non-neoplastic lesions and prediction of submucosal invasion of early cancer during colonoscopy

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## Abstract

Magnifying chromoendoscopy is an exciting new tool that allows detailed analysis of the morphological architecture of mucosal crypt orifices. In this review, we principally describe the efficacy of magnifying chromoendoscopy and magnifying colonoscopy with narrow band imaging (NBI) for differential diagnosis of colorectal lesions, including distinction between non-neoplastic and neoplastic lesions, and also between endoscopically treatable early invasive cancers and untreatable cancers, based on a review of the literature. We have conducted a prospective study showing that a combination of magnifying colonoscopy and chromoendoscopy is currently a more reliable method than conventional endoscopy and chromoendoscopy for separating non-neoplastic from neoplastic lesions of the colon and rectum. Magnifying colonoscopy with NBI is convenient and as accurate as chromoendoscopy with magnification. We principally use only magnifying colonoscopy with NBI, rather than chromoendoscopy, to routinely distinguish neoplastic from non-neoplastic polyps. Colonoscopists can predict the depth of invasion of early colorectal cancer by magnifying chromoendoscopy, magnifying colonoscopy with NBI and the non-lifting sign. Among these approaches, magnifying chromoendoscopy is diagnostically the most reliable, with an accuracy, sensitivity, and specificity of 98.8%, 85.6%, and 99.4%, respectively. Although its reliability depends on the skill of magnifying observation, widespread applications of the magnification technique could influence the indications for biopsy sampling during colonoscopy and the indications for mucosectomy.

## Key words

Colorectal neoplasm, Differential diagnosis, Learning curve, Magnifying chromoendoscopy, Narrow band imaging (NBI).

## INTRODUCTION

The prognosis of patients with colorectal malignancies is strictly dependent on efficient detection of lesions at the pre-malignant or early malignant stage. Colonoscopy is the only technique currently available that has the potential to both find and remove not only premalignant lesions but also early cancers throughout the colon and rectum. At present, new powerful high-resolution endoscopy combined with image enhancement is an exciting new tool that facilitates detailed

analysis of the morphological architecture of mucosal crypt orifices (1, 2). Comparable to the rapid development of chip technology, new developments in optical techniques such as narrow band imaging (NBI), endocytoscopy (3), and laser-scanning confocal microscopy (LCM) (4), now allow unique observation of glandular and cellular structures.

This review highlights the efficacy of magnifying chromoendoscopy, and magnifying colonoscopy with narrow band imaging (NBI), for the diagnosis of colorectal lesions based on a review of the literature.

## MAGNIFYING CHROMOENDOSCOPY

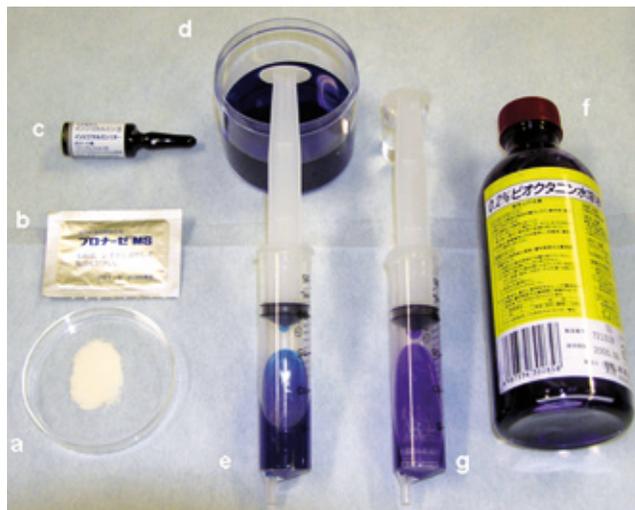
### When and how should magnifying chromoendoscopy be used?

In Japan, colorectal lesions are initially diagnosed by conventional-view colonoscopy, and then, if possible, by magnifying view and/or chromoendoscopy using indigo carmine. We routinely use a magnifying colonoscope because the insertion technique and manipulation are similar to those for an ordinary colonoscope (5). A magnifying endoscope with high resolution can provide low- to high-magnification (x80-100 maximum) images utilizing a one-touch operation electrical power system (2). In a prospective study, Konishi and colleagues reported that insertion of a magnifying colonoscope into the cecum was achieved successfully in 97% of cases, and that there were no significant differences in the average time taken to reach the cecum or the average total procedure time (6).

When a colonoscopist intends to perform chromoendoscopy, dye is sprayed as an aqueous solution via the biopsy channel in a volume of 3-5 ml, along with 15 ml of air, using a 20-cc syringe. Common dyes for the characterization of the colorectum are indigo carmine as a contrast stain (0.1–0.4%) and crystal violet (0.05%) and methylene blue (0.1%) as absorptive stains. Although indigo carmine and methylene blue are often used to screen for sporadic adenoma, crystal violet, as an absorptive stain, offers advantages for patients with early invasive cancer or for detailed observation using a non-traumatic catheter after washing the lesion with lukewarm water containing pronase (Pronase MS<sup>®</sup>) (Figure 1, 2) (7).

Is it advisable to spray dye over the whole of the colon and rectum to identify significant lesions? When should magnification be employed? Certainly, pan-mucosal chromoendoscopy significantly increases the rate of detection of small neoplastic and flat lesions, but this technique requires an excessive volume of dye and a significantly prolonged procedure (8-12). Therefore, colonoscopists use “selective” chromoendoscopy only for further examination of any subtle mucosal irregularity detected during standard colonoscopy. After the detection of mucosal abnormality, target chromoendoscopy with magnification is indispensable for confirming the surface structure and perimeter shape of the lesion in detail (Figure 3). When the examiner performs more detailed magnifying observation, a change in the patient’s posture to position the lesion opposite to the direction of gravity is very effective, because this prevents soaking the lesion in a dye pool. Needless to say, thorough preparation of the colon prior to colonoscopy is also important. Harewood *et al.* reported that there was a close association between adequate preparation and the suc-

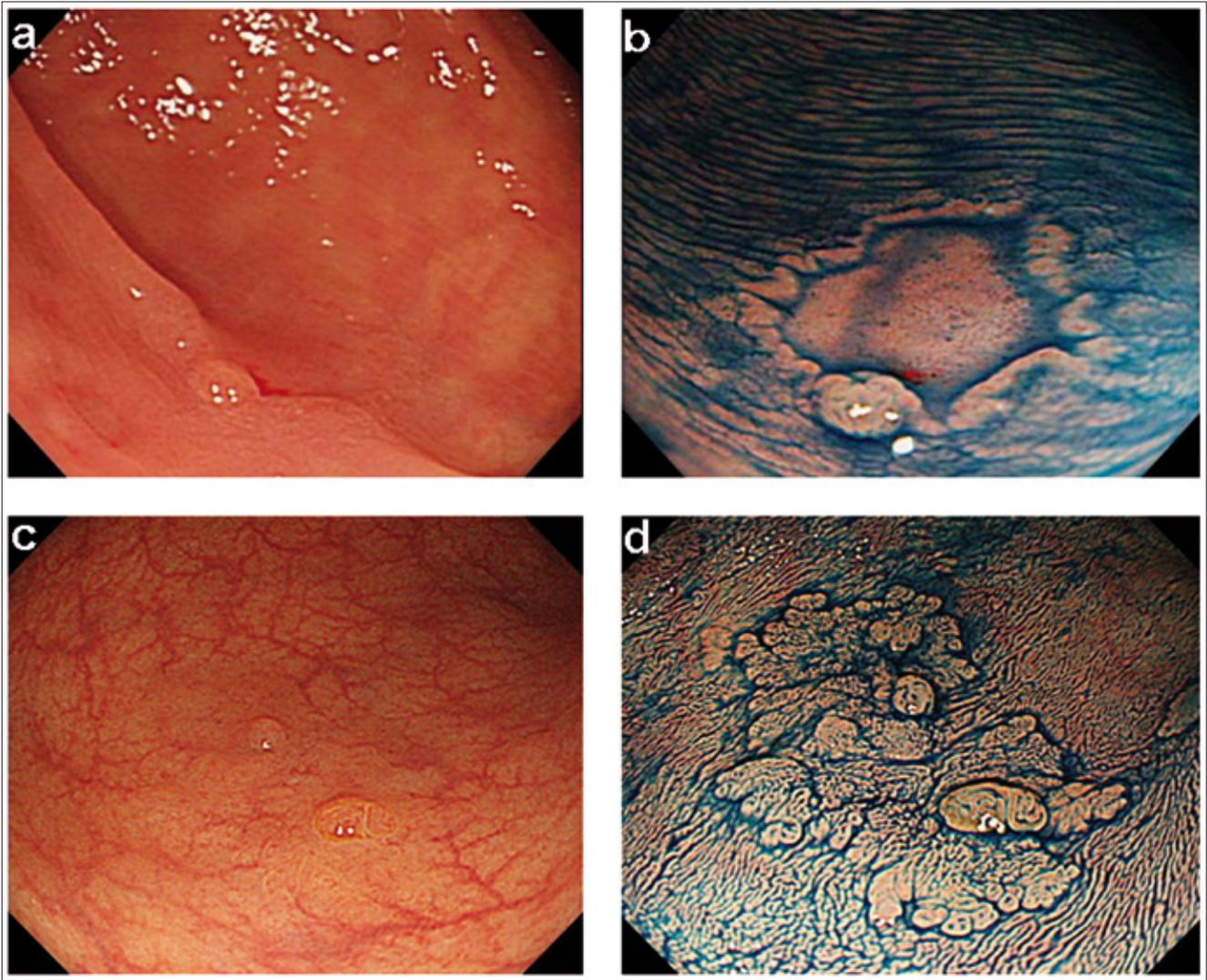
cessful identification of smaller lesions (odds ratio 1.23), compared with that of larger lesions (13). Inadequate preparation resulting in a residue of food or fecal material could obscure not only small polypoid lesions but also superficial depressed lesions (7, 14). However, this conclusion needs to be confirmed in further prospective studies.



**Figure 1.** Preparations for magnifying observation. a, b: Pronase MS<sup>®</sup>. Washing of the target lesion surface can be done with 500 cc of lukewarm water containing a packet of Pronase MS<sup>®</sup> (20000U). c: Indigo carmine (Daiichi Pharmaceutical Corp., Tokyo, Japan). d: The dye is a blue stain that accentuates the contours of a lesion, providing a detailed view of its border and shape. The dye is used as a 0.1-0.4% aqueous solution. e: This solution is flushed through the biopsy channel of the scope using a 20-cc syringe. Generally, 3-5 cc 5s used along with 15 cc of air. f: Crystal violet (Honzo Pharmaceutical Corp., Nagoya, Japan). The dye is a vital stain and is preferentially taken up by the Lieberkuhn gland openings (crypts), which appear as dots or pits. g: A few small drops of crystal violet in 0.05% solution are applied using a non-traumatic catheter (Olympus 6233064; Olympus Optical Co., Ltd., Tokyo, Japan).



**Figure 2.** Non-traumatic, globular-tip catheter. This catheter is used to remove mucus and to drop crystal violet solution onto the lesion. Better positioning for magnifying observation can be obtained by pushing and holding the surrounding mucosa.



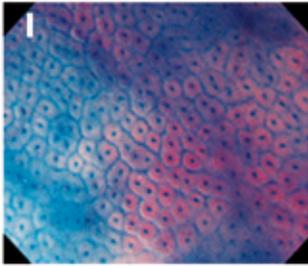
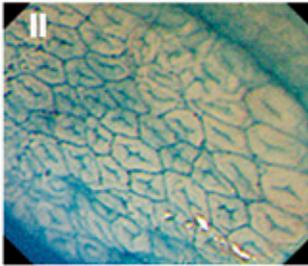
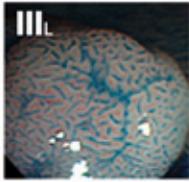
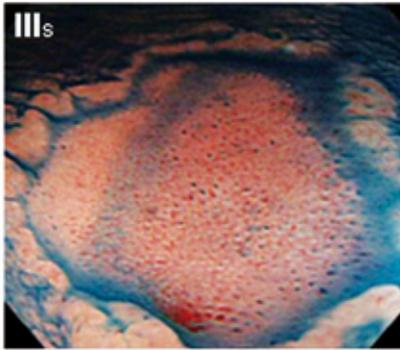
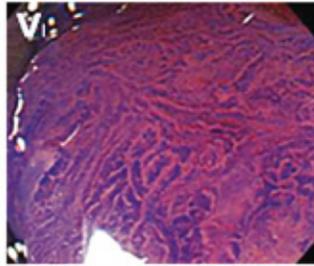
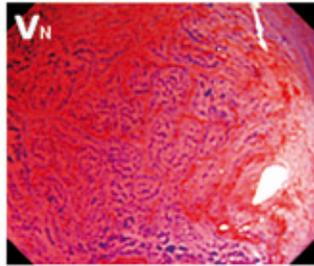
**Figure 3.** Usefulness of indigo carmine. a: Disruption of the mucosal fold and a slightly reddish area are observed, but the whole lesion is unclear. b: After spraying with indigo carmine dye, a 7-mm depressed lesion (0-IIc) is identified clearly. c: A slightly elevated lesion with an obscure superficial vascular component is evident, but the whole lesion is not recognized. d: A slightly elevated lesion measuring 18 mm is obviously detected using indigo carmine.

### Mucosal crypt (pit) patterns on the surface of colorectal lesions

Pit patterns have essentially been identified from stereomicroscopic observations of resected colonic specimens.<sup>1</sup> The introduction of magnifying colonoscopy has permitted observation of pit patterns *in vivo* (2). A magnifying colonoscope has functions essentially identical to a conventional colonoscope, but with the addition of 'zoom' magnification. Pit patterns are grossly classified into 7 types (Figure 4). In Kudo's classification, it has been suggested that type I and II pit patterns are characteristic of non-neoplastic lesions such as normal mucosa or hyperplastic polyps.

However, most lesions showing pattern types IIIS, IIIL, IV, and a subset of VI are intramucosal neoplastic lesions such as adenoma or intramucosal carcinoma. Lesions with a type VN pattern and a subset of type VI suggest deep invasive carcinoma (2).

In neoplastic lesions of the colorectum, the risk of lymph node metastases is nil when the cancer is intramucosal or submucosal limited to <1000  $\mu\text{m}$  of invasion, and the risk of lymph node metastasis reaches about 10% when a cancer invades the deep submucosa (15). In this regard, Fujii and colleagues proposed a clinical classification of crypt patterns in relation to treatment (7, 16), and categorized lesions into three basic patterns: "non-neoplastic", "non-

	Clinical classification		
	Non-neoplastic pattern	Non-invasive pattern	Invasive pattern
Kudo's classification	I · II	III <sub>L</sub> · III <sub>S</sub> · IV · (part of VI)	VI · VN
Endoscopic findings	 	 	 
Histology	Normal Hyperplastic polip	Adenoma *m **sm-slight	‡sm profundo
Treatment	No treatment	Endoscopic treatment (Polypectomy or EMR)	Surgical treatment

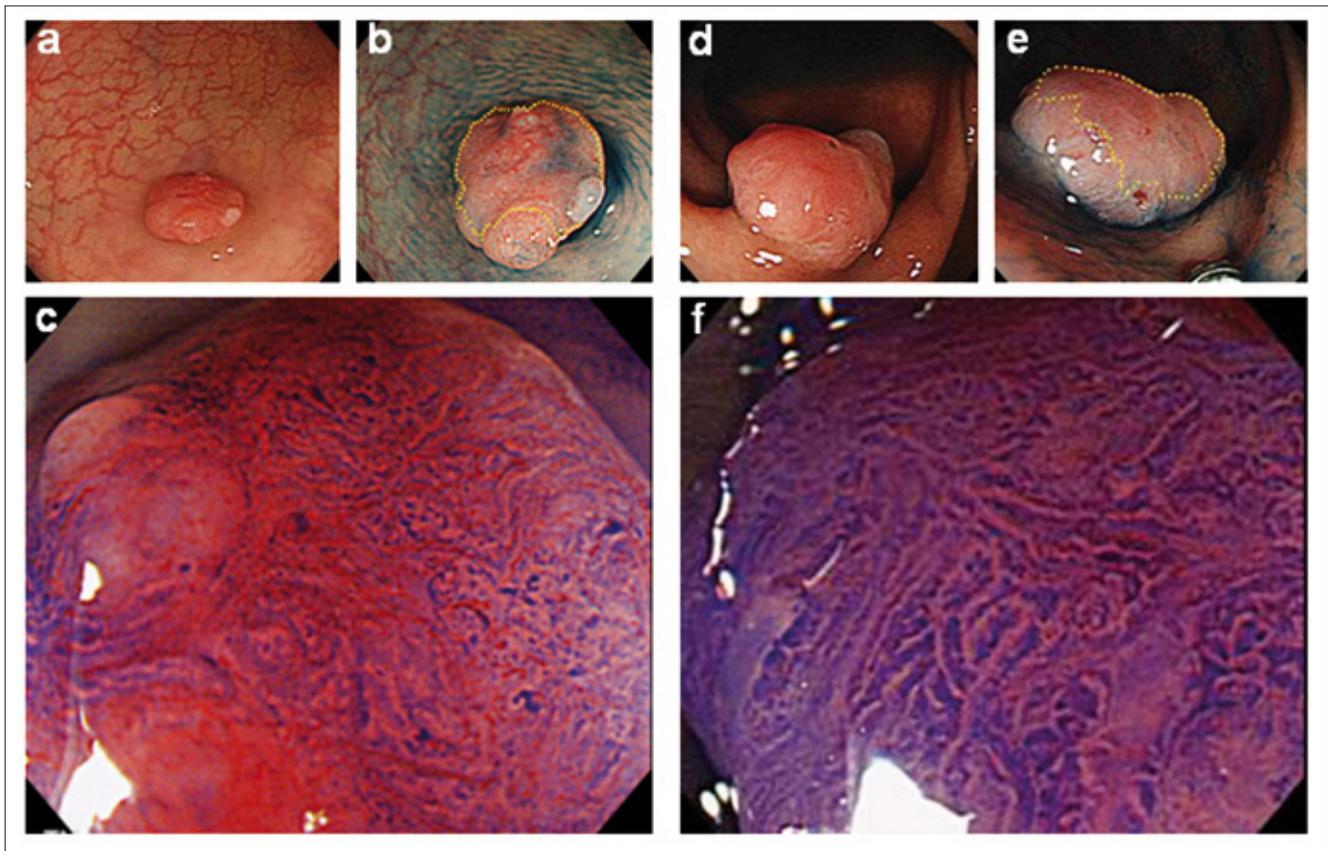
**Figure 4.** Pit pattern classification and clinicopathological findings.

invasive” and “invasive”. Basically, non-neoplastic patterns are equivalent to the type I and II pit patterns in Kudo's classification. The invasive pattern is defined as an irregular and distorted crypt observed in a demarcated area such as a depression, large nodule, or reddened area (Figure 5) due to the fact that histopathologically, lesions deeply invading the submucosa usually show a demarcated area on the surface. Therefore, types III<sub>S</sub>, III<sub>L</sub>, IV, and VI that do not show a demarcated area are classified as non-invasive patterns. Overall, Kudo's type VI pit pattern is classified into two groups – VI-non invasive and VI-invasive – based on the absence or presence of a demarcated area.

### Differential diagnosis of non-neoplastic and neoplastic lesions using magnifying chromoendoscopy

The lesions most frequently encountered using colonoscopy are colorectal polyps, of which 10-30% are reportedly non-neoplastic (17-19). Since removal of non-neoplastic

lesions not only wastes time and resources but may also increase procedure-associated complications, it is important to differentiate non-neoplastic from neoplastic lesions on colonoscopy (20-23). In addition, based on the adenoma-carcinoma sequence and evidence from randomized control trials, removal of all neoplastic lesions is considered to reduce the incidence of, and mortality from colorectal cancers (24). The differential diagnostic abilities of conventional view, chromoendoscopy and magnifying observation with chromoendoscopy have been reported to be 68-83%, 82-92%, and 80-96%, respectively (Table 1) (6, 25-31). According to previous reports, magnifying colonoscopy with chromoendoscopy seems to be the most effective method for differential diagnosis, although most of these results have been based on single-arm studies and are therefore not comparable. Konishi *et al.* conducted an excellent prospective study showing that the diagnostic ability to distinguish non-neoplastic from neoplastic lesions by magnifying colonoscopy was superior to that of



**Figure 5.** Invasive pattern. a) Endoscopic examination demonstrates a small (7-mm) flat elevated lesion in the sigmoid colon. b) Chromoscopy with indigo carmine shows a definite central depression. c) Magnification with crystal violet staining demonstrates an invasive pattern in a demarcated area. Based on these findings, the tumor was diagnosed as an early colon cancer with deep submucosal invasion, and surgical resection was recommended. Histopathological examination of the resected specimen demonstrated well differentiated adenocarcinoma, invasive to the submucosa (sm deep; 4000  $\mu\text{m}$ ). d) A sessile lesion Is (+IIc), 15 mm in diameter, identified in the upper rectum. e) Chromoscopy with indigo carmine: Reddish change and slight depression are observed on the surface of the tumor. f) Magnification with crystal violet staining demonstrates an invasive pattern. Histopathological examination of the resected specimen demonstrated well differentiated adenocarcinoma (sm deep; 4500  $\mu\text{m}$ ).

non-magnifying colonoscopy (6). Furthermore, we have conducted a prospective study to examine whether magnification and/or indigo carmine dye-spraying is more reliable than the conventional view for such differential diagnosis (32). The overall diagnostic accuracy of magnification in addition to chromoendoscopy using indigo carmine was 95.6%, being 10% and 5% more reliable than conventional endoscopy and chromoendoscopy, respectively. In addition, this method was significantly superior to conventional endoscopy and chromoendoscopy ( $p < 0.0001$  and  $p = 0.0152$ ). Therefore, based on these results, we are able to conclude that at present, a combination of magnifying colonoscopy and chromoendoscopy is the most reliable method for distinguishing non-neoplastic from neoplastic lesions of the colon and rectum. The reported results related to differential diagnostic abilities, including those of our study, are summarized in Table 1.

### Distinction between m, sm1 vs. sm2 or beyond using magnifying chromoendoscopy

As submucosal invasion of early colorectal cancer has a 6-13% risk of lymph node metastasis (33-38), surgery is indicated. In Japan, there is growing evidence supporting the theory that lesions with submucosal invasion limited to  $< 1000 \mu\text{m}$  without lymphovascular involvement and a poorly differentiated component lack LN metastases (39-42), and can be cured by endoscopic resection alone. The Paris endoscopic classification of superficial neoplastic lesions has also determined  $1000 \mu\text{m}$  to be the cut-off limit between sm1 and sm2 (15). It is important to determine the vertical depth of invasion of submucosal colorectal cancers prior to endoscopic resection, because endoscopic resection of early colorectal cancer with massive submucosal invasion carries a high risk of bleeding and perforation.

**Table 1.** Previous studies of overall diagnostic accuracy, sensitivity, specificity and predictive value for differentiating non-neoplastic lesions from neoplastic lesions.

Author	Colonoscopy apparatus	Number of lesions	Overall accuracy (%)	Sensitivity (%)	Specificity (%)	PPV* (%)	NPV** (%)
Chapius et al	Ordinary	120	82,5	84,5	77,7	89,8	68,3
Neale et al	Ordinary	81	80,2	69,2	85,4	69,2	85,5
Our results	Ordinary	206	84	88,8	67,4	93,4	63,3
Eisen et al	Chromoendoscopy	480	82,1	82	82	75	88
Kieslich et al	Chromoendoscopy	283	92,6	92,4	93,2	97,5	81
Our results	Chromoendoscopy	206	89,3	93,1	76,1	93,1	76,1
Axelrad et al	Magnifying	55	94,5	92,9	95,1	86,7	97,5
Togashi et al	Magnifying	923	88,4	92	73,3	94,2	85,2
Tung et al	Magnifying	175	80,6	93,8	64,6	76,3	89,5
Liu et al	Magnifying	954	86,1	90,8	72,7	90,4	73,6
Our results	Magnifying	206	95,6	96,3	93,5	98,1	87,8

With regard to conventional colonoscopy, Saitoh and colleagues have proposed characteristic colonoscopic features of depressed-type colon cancers that indicate the need for surgical treatment: 1) an expansive appearance, 2) a deeply depressed surface, 3) an irregular bottom in the depressed surface, and 4) folds converging toward the tumor, as revealed by combined use of videoendoscopy and chromoendoscopy (43). They have also reported that the invasion depth of depressed-type early colorectal cancers was correctly determined in 58 of 64 lesions (91%) on the basis of these findings. Matsuda and colleagues reviewed all conventional colonoscopic images of 123 non-polypoid submucosal colorectal cancers treated endoscopically or surgically between 1999 and 2003. They found that white spots (a chicken skin appearance),

redness, firm consistency and a deeply depressed area were significantly associated with an increased risk of submucosal deep invasion, on the basis of univariate analysis (Table 2) (44).

In previous studies, some authors have reported the clinical usefulness of detailed determination of the V pit pattern using magnifying chromoendoscopy to predict the depth of invasion prior to EMR for submucosal colorectal cancers. Kudo *et al.* reported that a non-structural pit pattern V (VN) was recorded essentially in intramucosal and submucosal cancers (93.3% of the type); 65% (128/195) of lesions classified as displaying this type corresponded to invasive cancer infiltrating the submucosa (45). Kato *et al.* reported that the diagnostic accuracy of magnifying colonoscopy for invasive (sm) cancers was 85% (81/95) (25).

**Table 2.** Relationship between findings of conventional endoscopy and submucosal depth of invasion.

	SM-Superficial (n=35)	SM-Deep (n=88)	Univariate Analysis (P value)	Diagnostic Sensitivity and specificity
Size (≥ 20mm)	16/35 (45,7%)	30/88 (34,1%)	0,23	Sens. 34,1% Spec. 54,3%
White spots (chicken skin) (+)	2/35 (5,7%)	29/88 (32,9%)	0,002	Sens. 32,9% Spec. 94,3%
Redness (+)	14/35 (40,0%)	62/88 (70,4%)	0,002	Sens. 70,4% Spec. 60,0%
Firm consistency (+)	11/35 (31,4%)	69/88 (78,4%)	<0,0001	Sens. 78,4% Spec. 68,6%
Expansion (+)	2/35 (5,7%)	18/88 (20,4%)	0,007	Sens. 20,4% Spec. 94,3%
Fold convergence (+)	4/35 (11,4%)	20/88 (22,7%)	0,24	Sens. 22,7% Spec. 88,6%
Deep depression (+)	15/35 (42,9%)	77/88 (79,5%)	<0,0001	Sens. 79,5% Spec. 57,1%

The clinical classification of pit patterns (invasive or non-invasive) was originally proposed by Fujii to discriminate between m-sm1 and sm2 or beyond (7). The diagnostic definition of an invasive pattern is identification of irregular or distorted crypts, in a demarcated area, where the orifice of each crypt cannot be traced clearly (Figure 5). This finding suggests that the lesion has already invaded deeply into the submucosa. The invasive pit pattern has been used to ascertain the depth of sm invasion in order to determine the ideal treatment, i.e. endoscopic resection or surgery. Indeed, in our recent large prospective series, the clinical classification of pit patterns (invasive or non-invasive) was proven to be effective for differentiating intramucosal or sm superficial invasion (<1000 µm) from sm deep invasion (≥1000 µm). In this study, histopathology confirmed epithelial neoplasia in 99.4% of 4037 lesions, with a non-invasive pattern and confirmed deep sm invasion in 86.5% of 178 lesions with an invasive pattern (46). Furthermore, the calculated sensitivity, specificity, positive predictive value, negative predictive value, and accuracy were 85.6%, 99.4%, 86.5%, 99.4%, and 98.8%, respectively, for differentiating intramucosal or sm superficial invasion (<1000 µm) from sm deep invasion (≥1000 µm) (Table 3).

**Table 3.** Relationship between clinical classification and histological findings of magnifying chromoendoscopy.

	Total no.	Adenoma no. (%)	Intramucosal (m) cancer, no. (%)	Submucosal (sm) cancer	
				sm-slight no. (%)	sm-deep* no. (%)
Invasive pattern	178	0 (0)	12 (7)	12 (7)	154 (86)
Non-invasive pattern	4037	3371 (83)	600 (15)	40 (1)	26 (0.6)

Sensitivity: 85,6% (15/180), Specificity: 99,4 (4011/4035), Accuracy: 98,8% (4165/4215).

NPV (negative predictive value): 99,4% (4011/4037) PPV (positive predictive value): 86,5% (154/178)

\*sm deep invasion (≥1000 µm)

These results suggest that magnifying colonoscopy is also effective for differentiating between m-sm1 and sm2 or beyond. Among lesions diagnosed endoscopically as having an invasive pattern, a high percentage showed invasive cancer, especially sm depth invasive cancer, for which surgical resection is undoubtedly the appropriate treatment. However, the lesions diagnosed endoscopically as having a non-invasive pattern were mostly intramucosal lesions, for which endoscopic resection is feasible.

## Learning curve for magnifying chromoendoscopy

The effort necessary for learning to identify mucosal crypt patterns is important, but has been little studied. Based on our limited experience, it took three months of training in our hospital for a foreign clinician without knowledge of pit patterns to achieve a differential diagnostic ability exceeding 90%, similar to that of well trained endoscopists in our hospital (unpublished data). Togashi *et al.* investigated the efficacy of magnifying colonoscopy for differential diagnosis of colorectal polyps and also described the associated learning curve (31). They reported that observational experience of at least 200 lesions at high magnification was needed for an adequate diagnostic knowledge of pit patterns.

Kobayashi *et al.* investigated the ease of acquisition of pit pattern diagnostic ability for inexperienced examiners (47). They gave short lectures about pit pattern diagnosis to five nurses without any prior knowledge of endoscopic diagnosis, and the nurses were then tested using pictures taken by magnifying colonoscopy. The accuracy rate of differential diagnosis of non-neoplastic from neoplastic polyps was 85.4%, while the accuracy rate for differential diagnosis of intramucosal from invasive cancers was 72.4%. In conclusion, differentiation between non-neoplastic and neoplastic polyps using magnifying colonoscopy is feasible even for observers with no prior knowledge of the procedure, if a short lecture is given before the observation. However, differentiation between intramucosal and invasive cancers using magnifying colonoscopy is not easy for an inexperienced examiner.

We also investigated whether experience with colonoscopy influences pit pattern diagnosis in a study using 119 cases of early colorectal cancer, comprising 71 intramucosal cancers and submucosal (sm) cancers with slight invasion (m-sm1), and 48 sm cases with deep invasion (sm2-3). Endoscopic pictures were assessed by 20 endoscopists without prior knowledge of any lesion. Each colonoscopist independently diagnosed whether the lesion was m-sm1 or sm2-3, first using pictures of the conventional view, followed by pictures of the magnifying view. The results indicated a difference in diagnostic accuracy between the experienced and inexperienced groups following the addition of magnification. The diagnostic accuracy in the experienced group without access to magnification was 84.4%, and this improved to 88.0% with magnification. In the inexperienced group, however, there was no such difference: the corresponding figures were 74.4% and 75.2%.

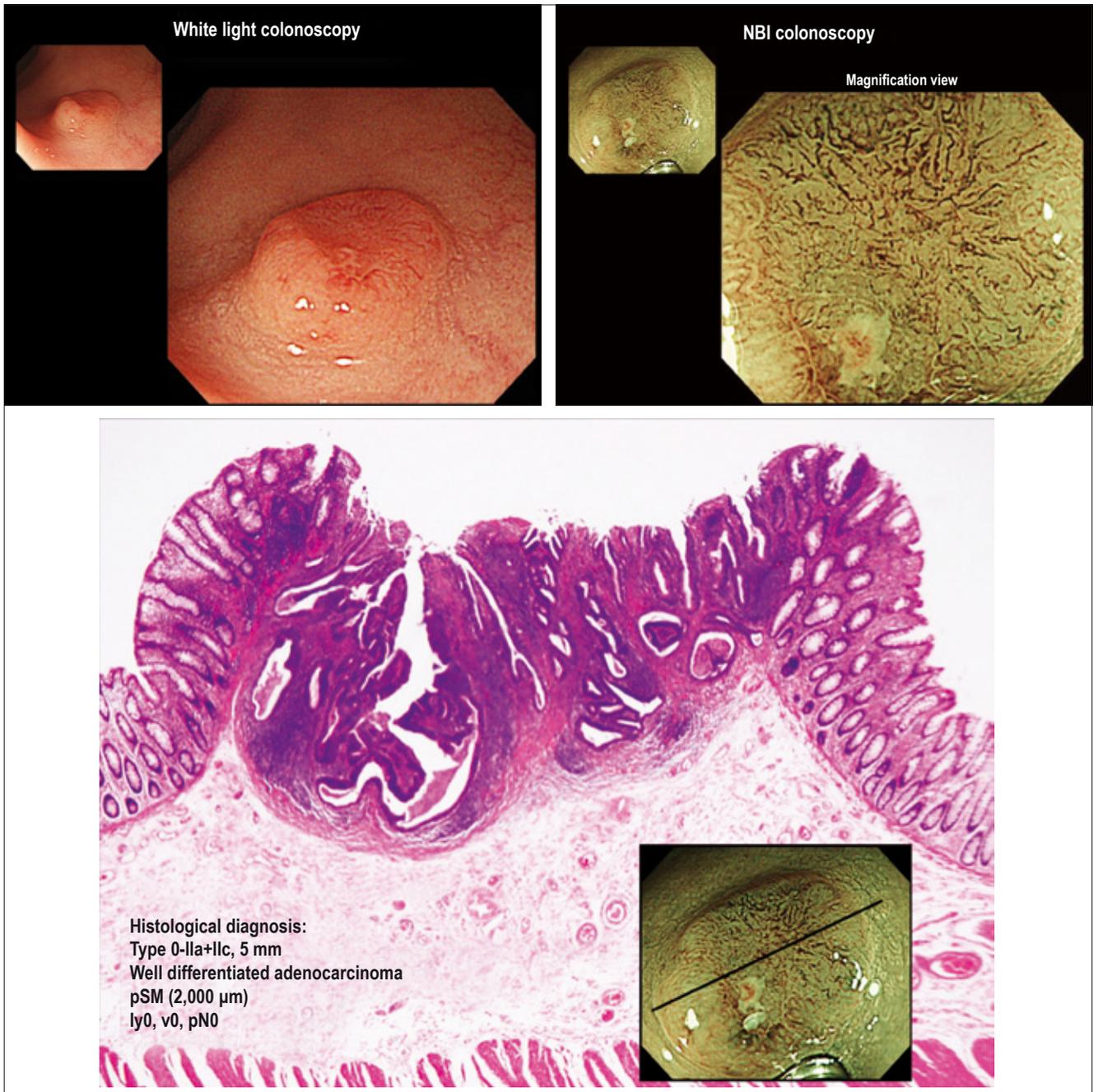




**Figure 7.** Early colorectal cancer showing CP type IIIA on NBI with magnification. Conventional endoscopic view, showing a depressed lesion (type 0-IIc, 4 mm). CP type IIIA on magnifying view with NBI. Histological findings show well differentiated intramucosal adenocarcinoma.

Ikematsu and colleagues recently conducted a prospective study to determine whether CP type IIIA/IIIB identified by magnifying NBI was effective for estimating the depth of invasion in 130 early colorectal neoplasms (59). There were 15 adenomas, 66 intramucosal cancers (pM) and 49 submucosal cancers (pSM): 16 pSM superficial (pSM1) and 33 pSM deep cancers (pSM2-3). The sensitivity, specificity and diagnostic accuracy of CP type III for

differentiating pM-ca or pSM1 ( $<1000 \mu\text{m}$ ) from pSM2-3 ( $\geq 1000 \mu\text{m}$ ) were 84.8%, 88.7% and 87.7%, respectively. The accuracy of CP type IIIA (NPV) was 94.5% (86/91), and that for lesions of CP type IIIB (PPV) was 71.8% (29/39) (Table 4). In their study, the rate of diagnostic agreement among the three observers was good, without variability (interobserver variability:  $\kappa = 0.68, 0.67, 0.72$ . intraobserver agreement:  $\kappa = 0.79, 0.76, 0.75$ ). Magnifying



**Figure 8.** Early colorectal cancer showing CP type IIIB on NBI with magnification. Conventional endoscopic view, showing a depressed lesion (type 0-IIaIIc, 5 mm). CP typeIIIB on magnifying view with NBI. Histological findings show well differentiated adenocarcinoma with deep submucosal invasion (2000μm).

colonoscopy with NBI for diagnosis on the basis of pit pattern (invasive/non-invasive) had similar accuracy (87.7% vs 87%) and sensitivity (84% vs 85%), but markedly lower specificity (88% vs 99%). In considering the reasons for these differences, the authors suggested that the inclusion of more than 3000 adenomatous lesions in their series and

the learning curve for estimating the depth of early colorectal neoplasms using NBI may have been contributory. Thus, from the viewpoint of diagnostic estimation of the depth of early colorectal cancer, magnifying colonoscopy with NBI is not superior to magnifying chromoendoscopy. With regard to the learning curve for endoscopy with NBI,

Higashi and colleagues reported the diagnostic skills of less-experienced endoscopists for differentiation of diminutive colorectal polyps using NBI and pit pattern analysis, with and without magnification, after an expanded training program. The use of high-magnification NBI increased the differential diagnostic skill of the less-experienced group after expanded training, making it equivalent to that of the experienced group (60).

**Table 4.** Diagnostic accuracy, NPV and PPV of CP type IIIA and type IIIB.

	Histological diagnosis	
	Adenoma, m*, sm-superficial (sm1)**	Sm-deep (sm2-3)***
CP type IIIA	86	5
CP type IIIB	11	28

Sensitivity: 84,8%. Specificity: 88,7. Accuracy: 87,7%.  
 NPV (negative predictive value): 94,5%. PPV (positive predictive value): 71,8%  
 \*intramucosal cancer  
 \*\* sm superficial invasion (<1000 µm)  
 \*\*\*sm deep invasion (≥1000 µm)

## NON-LIFTING SIGN

Submucosal saline injection is also useful, not only as a method for endoscopic mucosal resection (EMR) but also as a simple diagnostic tool for deeply invasive cancers. Although adenoma and intramucosal cancer are easily lifted by submucosal saline injection, deeply invasive cancer is not lifted because of the presence of a desmoplastic reaction and the invasive nature of the lesion.

Uno and colleagues originally proposed the non-lifting sign in 1994, and considered it to be positive in cases where the surrounding mucosa, but not the lesion, was elevated (61). Kobayashi and colleagues conducted a prospective multicenter trial to assess the accuracy of the non-lifting sign, in comparison to endoscopic diagnosis, as a diagnostic tool for determining whether the depth of invasion from the muscularis mucosae was less, or greater, than 1 mm (62). The overall sensitivity, specificity and accuracy of the non-lifting sign were 61.5% (16/26), 98.4% (241/245) and 94.8% (257/271), respectively (Table 5), whereas those of endoscopic diagnosis were 84.6% (22/26), 98.8% (242/245) and 84.6% (22/26) and 98.8% (242/245), respectively. Although the non-lifting sign showed high specificity, the sensitivity was insufficient in comparison with endoscopic diagnosis. On the basis of this result, they concluded that the non-lifting sign was unable to replace endoscopic diagnosis for experienced colonoscopists.

**Table 5.** Overall accuracy of the non-lifting sign for diagnosis of invasion depth.

	Histological diagnosis	
	Adenoma, m*, sm-superficial (sm1)**	Sm-deep (sm2-3)***
Non lifting sign (-)	241	10
Non lifting sign (+)	4	16

Sensitivity: 61,5%. Specificity: 98,4. Accuracy: 94,8%.  
 NPV (negative predictive value): 96,0%. PPV (positive predictive value): 80,0%  
 \*intramucosal cancer  
 \*\* sm superficial invasion (<1000 µm)  
 \*\*\*sm deep invasion (≥1000 µm)

## DISCUSSION

In the last 10 years, endoscope systems have been dramatically improved, being comparable to the rapid development of CCD chip technology, allowing video-endoscopy to be performed with new powerful high-resolution or high-vision endoscopes as a standard and routine procedure. By combining chromoendoscopy with magnification, allowing accurate in vivo prediction of colorectal lesion histopathology, it may be possible to reduce the risk through selective removal of clinically significant lesions endoscopically.

It is important to differentiate non-neoplastic from neoplastic lesions by colonoscopy, as this helps to minimize procedure-associated complications and wasted time and effort. Fu and colleagues conducted a valuable prospective study showing that the diagnostic accuracy of magnification in addition to chromoendoscopy using indigo carmine was 95.6%, thus making it 10% and 5% more reliable than conventional endoscopy and chromoendoscopy, respectively, for determining whether a colorectal lesion is non-neoplastic or neoplastic (32). Sano and colleagues found that magnifying colonoscopy with NBI had high accuracy, sensitivity and specificity (95.3%, 96.4%, and 92.3% respectively) for differentiating neoplastic from non-neoplastic lesions (57). It is likely that magnifying colonoscopy with NBI will eventually replace magnifying chromoendoscopy because it is convenient and requires no spraying of dye.

How is possible to predict submucosal invasion of early colorectal cancers prior to endoscopic resection? Endoscopic ultrasonography (EUS) is also a useful predictive modality, but requires additional training and equipment, and is time-consuming. Some authors have reported that EUS is superior to magnifying chromoendoscopy (63, 64), and Fu and colleagues have recently reported that

magnifying chromoendoscopy is as accurate as EUS for predicting the invasion depth of early colon cancer (87% vs 75%,  $P=.0985$ ) (65).

Magnifying chromoendoscopy and NBI with magnifying colonoscopy allow colonoscopists to make real-time diagnosis of the depth of early colon cancer when they find it, and the non-lifting sign allows diagnosis in real time once treatment has been initiated. What, then, is the gold standard method for prediction of invasion depth? In comparison to endoscopic diagnosis by magnifying chromoendoscopy, NBI with magnifying colonoscopy and the non-lifting sign, magnifying chromoendoscopy is the most reliable method currently available for predicting the depth of early colon cancer (Table 6).

Although the reliability of any method depends on the skill of magnifying observation, as indicated above, widespread application of the magnification technique could influence the indications for biopsy sampling during colonoscopy and those for mucosectomy. If so, how does a beginner acquire sufficient expertise? This probably requires close collaboration between the endoscopist and the

pathologist. In practical terms, the endoscopist takes the endoscopic mucosal resection material, suitably mounted, to the laboratory. There it is re-stained using half-diluted Carazzi's hematoxylin and examined jointly with the pathologist using a stereomicroscope. Histological sections would also be examined jointly, and the microscopic features compared with the endoscopic and stereomicroscopic images. Through these processes, both the endoscopist and the pathologist would become familiar with the prediction of histology from the mucosal crypt pattern. Moreover, to avoid misinterpretation, we recommend that endoscopists who are not familiar with the pit pattern or with magnifying colonoscopy should acquire knowledge not only from published papers but also from colonoscopists experienced in magnification techniques. As the proverb says, "Seeing is believing".

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**Table 6.** Comparison of endoscopic diagnosis for the depth of submucosal deeply invasive colon cancer.

Diagnostic method	Number of adenoma, m-ca* sm-slight-ca**	Number of sm deep-ca***	Overall accuracy (%)	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
Magnifying Chromoendoscopy (invasive pattern)	4035	180	98,8	85,6	99,4	86,5	99,4
NBI with magnifying colonoscopy (capillary pattern classification)	97	33	87,7	84,8	88,7	71,8	94,5
Non-lifting sign	245	26	94,8	61,5	98,4	80,0	96,0

\*intramucosal cancer

\*\* sm superficial invasion (<1000  $\mu$ m) cancer

\*\*\*sm deep invasion ( $\geq$ 1000  $\mu$ m) cancer

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