EFFICACY OF MAGNIFYING COLONOSCOPY FOR THE DIAGNOSIS OF COLORECTAL NEOPLASIA: COMPARISON WITH HISTOPATHOLOGICAL FINDINGS

AKIHIKO OHTA, KENJI TOMINAGA AND YOSHIHIRO SAKAI

Division of Gastroenterology, Department of Internal Medicine, Toho University, Ohashi Hospital, Tokyo, Japan

Background: Some authors have reported a good correlation between the shape and arrangement in the orifice of mucosal crypt (pit pattern) in the diagnosis of colorectal lesions and histopathological findings. However, there remains no good consensus on the definition of irregularity in configurations.

Methods: We studied 110 colorectal neoplasia of 110 cases that were examined using a magnifying colonoscope and resected endoscopically or surgically between 2001 and 2003. We divided the lesions into two groups according to Fujii’s classification as follows: type Non-V showed type III or IV by Kudo’s classification and irregular pits not occurring in a demarcated area; and type V showed irregular pits, which occurred in a demarcated area in addition to type V by Kudo. The diagnosis by this classification correlated with the histopathological findings. We also addressed the causes of misdiagnosis in comparison with histopathological findings.

Results: The diagnostic accuracy for Non-V and V was 95.1% and 82.1%, respectively. Lesions classified as Non-V showed a significant correlation. Histologically, carcinoma consisted of high-grade atypia (P < 0.0001), an erosive change of surface epithelium (P < 0.0001) and the appearance of desmoplastic reactions (P = 0.002) in comparison with Non-V. Misdiagnosis was likely due to differences in the grade of atypia between superficial and deeper glands and explainable erosive changes on the surface, as well as simple misreading of the pit pattern.

Conclusion: Diagnosis of colorectal neoplasia by magnifying colonoscopy using our classification was useful for evaluating the depth of invasion and correlated well with histopathological findings.

Key words: colorectal neoplasia, magnifying colonoscopy, pit pattern, submucosal invasive carcinoma, type V pit.

INTRODUCTION

Magnifying colonoscopy permits the in vivo examination of shapes and arrangements of crypt orifice (pit pattern) in the colorectal mucosa. Kosaka was the first to report the stereomicroscopic observation of pit patterns of minute polyps accompanied by colorectal carcinoma in resected specimens. Since then, investigators in Japan have enthusiastically compared stereomicroscopic findings of colorectal mucosa with in vivo observation by magnifying colonoscope and currently recognize the latter as a useful procedure. Magnifying colonoscopy was regularly applied to discriminate between non-neoplastic and neoplastic lesions, and to estimate the depth of invasion in colorectal malignancies. During that time, some investigators divided type V pit into two subgroups to correlate with invasive depth of carcinoma. However, their definition differed between investigators, and the classifications did not always correlate with decisions for treatment. Therefore, the classification of pit patterns was recently unified into two subgroups: V (irregular pits considered to be a marker of intramucosal or slightly invasive submucosal carcinomas) and V (a decrease or absence of pit on the mucosal surface, which is considered to be a marker of deep invasive submucosal carcinomas).

Fujii et al. have reported that the diagnosis of deep invasive submucosal carcinomas by magnifying colonoscopy is important to appreciate the distribution of irregular pits in a demarcated area. Our initial studies supported this, but to now there are scarce data in the literature on the relationship between classification of carcinomas by magnifying colonoscopy and histopathological findings.

We therefore undertook the present study to evaluate the correlation between the new classification criteria of pit pattern and histopathological findings in colorectal neoplasia. In addition, because some cases were difficult to diagnose by magnifying colonoscope, we attempted to address the possible causes of misdiagnosis based on evaluation of the criteria in comparison with histopathological findings.

MATERIALS AND METHODS

We studied 110 lesions of colorectal neoplasia in 110 cases that were examined using a magnifying colonoscope and resected endoscopically or surgically between 2001 and
The diagnosis was 37 with adenoma, 39 with intramus-
cosal carcinoma (m), and 34 with submucosal invasive carci-
noma (sm). Adenoma was defined as lesions of larger than
10 mm in diameter or accompanied by a depressed area on
the surface in ordinary observation. The lesions were subdi-
vided into macroscopic types according to the Japanese
classification of colorectal carcinoma following ordinary
observation with 0.3% indigo carmine solution (IC) sprayed
over the lesions: protruding type, pedunculated (Ip), subpe-
dunculated (Isp) and sessile (Is); superficial elevated type,
IIa and IIa + IIc; or superficial depressed type, IIc and
IIc + IIa. Magnifying colonoscopy was performed using type
CF-240ZI and CF-H260ZAI endoscopes (OLYMPUS,
Tokyo, Japan). If the pit pattern could not be assessed,
0.05% Crystal Violet (CV) was applied to the lesion for sur-
face staining.

Pit pattern was evaluated according to Kudo’s classifica-
tion. Type I were round pits found on normal mucosa;
type II were star-shaped or onion-like pits found on hyper-
plastic change; type III, pits were large tubular structures
found on polypoid adenomas, type IIIIs were small, round
pits found on superficial depressed lesions; type IV were
large, branched or gyrus-like pits found on polypoid ade-
nomas; type V were irregular (type V) or almost destruc-
tive (so-called non-structure, type VN) pits found on
carcinomas. In this study, irregular pits (type V) were fur-
ther subdivided into two subgroups, based on whether they
were found in a demarcated area (Fig. 1). Irregular pits
without demarcated area were defined as Non-V in addition
to types III and IV pits (Fig. 2a,b). Irregular pits traced
clearly with demarcated area from surrounding area were
defined as V in addition to type VN (Fig. 2c,d). Magnifying
diagnosis was evaluated by a single operator at the time of
examination.

All resected specimens were examined histopathologi-
ically after hematoxylin and eosin (H&E) staining. The vertical
length of submucosal invasion was measured from the mus-
cularis mucosa to the deepest invasive portion. When the
muscularis mucosa was not apparent due to submucosal inva-
sion, vertical length was then measured from the surface of
the lesion to the deepest invasive portion. Lesions with a
vertical length of less than 1000 μm in the submucosal layer

Fig. 1. Relationship between Kudo’s classification and our
clinical classification.

<table>
<thead>
<tr>
<th>Kudo’s classification</th>
<th>Our clinical classification</th>
</tr>
</thead>
<tbody>
<tr>
<td>III·IV</td>
<td>Non-V</td>
</tr>
</tbody>
</table>

Fig. 2. (a) Highly magnified pictures showing irregular pits that
vary in size. (b) In low magnification colonoscopic images, the pits
do not occupy the entire surface; this pattern defined the Non-V le-
sion in the present study. (c,d) Magnifying pictures showing
irregular pits demarcated in a specific limited area (above the
dotted line), and showing type I or III, pit in the surrounding mu-
cosa (below the dotted line); this pattern defined the type V lesion
in the present study.
were classified as submucosal slightly invasive carcinoma (sm-s), and those more than 1000 μm as submucosal deep and massive invasive carcinoma (sm-d).

Association of pit pattern analysis and histology
Diagnostic accuracy was defined according to the following criteria: whether the Non-V lesions were limited to submucosal slightly invasive (sm-s); and whether the V lesions had invaded 1000 μm or more into the submucosal layer (sm-d). According to the classification of Watanabe et al., superficial glands in a lesion are classed as either carcinoma with low or high grade atypia.19,20 The condition of the surface epithelium on the lesion was classified as either preserved (preservation group) or degenerated, eroded (erosion group) (Fig. 3a–c). Desmoplastic changes superior to the lesion in submucosal carcinomas were classified into three groups: (−), no desmoplastic reaction (DR); (+), DR without a decrease in gland density; (++) DR with a decrease in gland density (Fig. 3d–f).

Relationship between macroscopic types and diagnosis after surface dyeing
The lesions were classified into two groups as follows: IC group, lesion that could be diagnosed on pit pattern only by
IC spraying; CV group, lesion that required CV staining to be diagnosed.

**Statistical analysis**

The significance of differences in variables was assessed using Fisher’s exact test and non-parametric test (Mann–Whitney U-test). Statistical significance was defined as $P < 0.05$.

**RESULTS**

Of the 110 lesions studied, 82 were classified as type Non-V and 28 as type V. The mean size of the type Non-V lesions was $15.5 \pm 7.1$ mm (mean ± SD); type V was $18.5 \pm 6.1$ mm; this represented a significant difference in size ($P = 0.009$). Macroscopically, the lesions were classified as 46 protruding types (Ip, 19; Isp, 17; Is, 10), 28 superficial elevated types, and eight superficial depressed types for the type Non-V lesions. For the 28 cases of type V lesions, there were 16 protruding types (Ip, 2; Isp, 12; Is, 2), 10 superficial elevated types, and two superficial depressed types.

**Magnifying diagnosis and depth of invasion**

The diagnostic accuracy was $95.1\%$ (78/82) for type Non-V lesions, $82.1\%$ (23/28) for type V lesions and $91.8\%$ (101/110) overall.

Submucosal carcinomas were diagnosed in $89.3\%$ (25/28) of type V lesions. Of 82 lesions with type Non-V, $11.0\%$ ($9/82$) were diagnosed as submucosal carcinomas histologically. And $4.9\%$ ($4/82$) were diagnosed as sm-d (Table 1).

**Relationship between pit pattern and grade of atypia**

With respect to the relationship between magnifying observation and grade of atypia in the superficial glands, carcinoma classed as high-grade atypia was only $23.9\%$ ($11/46$) in type Non-V, compared with $88.9\%$ ($24/27$) in type V. The incidence of carcinoma with high-grade atypia in type V was significantly higher than in type Non-V ($P < 0.0001$) (Table 1).

**Relationship between pit pattern and surface epithelium**

With respect to the condition of the surface epithelium, $67.1\%$ (55/82) of type Non-V were classified as preservation type. In contrast, only $7.1\%$ (2/28) of type V fell into the preservation group, with $92.9\%$ (26/28) classed as erosion group. The incidence of erosion group in type V cases was significantly higher than in type Non-V ($P < 0.0001$) (Table 1).

**Relationship between pit pattern and desmoplastic reaction**

Of the submucosal carcinomas in type Non-V lesions, $66.7\%$ (6/9) were DR (–) and $33.3\%$ (3/9) were DR (+), with no DR (++) observed. In type V lesion, $12.0\%$ (3/25) were DR (–), $60.0\%$ (15/25) were DR (+) and $28.0\%$ (7/25) were classed as DR (++), and appearance of DR was observed in $88.0\%$ of type V (22/25). The incidence of appearance of DR in type V was significantly higher than in type Non-V ($P = 0.002$) (Table 1).

| Table 1: Relationship between magnifying diagnosis and histopathological findings |
|-----------------------------|---------------------|-------------------|
|                            | Non-V              | V                 | $P$ value |
| $n$                        | 82                 | 28                |
| Depth of invasion          |                     |                   |
| adenoma                    | 36 (43.9%)         | 1 (3.6%)          |
| m                          | 37 (45.1%)         | 2 (7.1%)          |
| sm-s                       | 5 (6.1%)           | 2 (7.1%)          |
| sm-d                       | 4 (4.9%)           | 23 (82.1%)        |
| Accuracy rate, %           | 95.1%              | 82.1%             |
| Total %                    | 91.8               |
| Histological findings      |                     |                   |
| Grade of atypia            |                     |                   |
| Low-grade                  | 35 (76.1%)         | 3 (11.1%)         | $< 0.0001^*$ |
| High-grade                 | 11 (23.9%)         | 24 (88.9%)        |
| Surface epithelium         |                     |                   |
| Preservation               | 55 (67.1%)         | 2 (7.1%)          | $< 0.0001^*$ |
| Degeneration               | 17 (20.7%)         | 1 (3.6%)          |
| Erosion                    | 10 (12.2%)         | 25 (89.3%)        |
| Desmoplastic reaction      |                     |                   |
| (–)                        | 6 (66.7%)          | 3 (12.0%)         | 0.002*     |
| (+)                        | 3 (33.3%)          | 15 (60.0%)        |
| (++)                       | 0                  | 7 (28.0%)         |

* Fisher’s exact test.

SD, standard deviation; protruding, Ip (pedunculated polyp), Isp (subpedunculated polyp) and Is (sessile polyp); superficial elevated, Ha and Ha + Ic; superficial depressed, Hc and Hc + Ha; m, intramucosal carcinoma; sm-s, invasion of submucosal layer less than 1000 μm; sm-d, invasion of submucosal layer more than 1000 μm; (–), no desmoplastic reaction (DR); (+), DR without decrease in gland density; (++) quantity DR with a decrease in gland density.

**Relationship between observation after surface dyeing and macroscopic configuration**

On chromoscopic study, all type V lesions required CV staining to recognize the pit pattern. In contrast, $58.5\%$ (48/82) of type Non-V lesions were diagnosed by IC spraying alone. The ease of diagnosis by imaging following IC spraying was, in sequence, as follows: protruding type > superficial elevated type > superficial depressed type (Fig. 4).

**Possible causes of misdiagnosis by magnifying colonoscopy**

The present study indicated that the factors involved in misdiagnosis include estrangement in the grade of atypia between superficial and deeper glands, an erosive change and attached mucin on the mucosal surface or simple misreading of the pit pattern (Table 2).

**DISCUSSION**

The observation of colorectal lesions by magnifying colonoscopy developed from the comparison between stereomicroscopic observation and histopathological findings. Recent improvements on colonoscopic instrumentation now permit in vivo examination without changing scopes. However, differences remain between endoscopists in their definition of ‘irregular’ or ‘non-structure’ in the classification by Kudo and in the accuracy reported for the depth of invasion. This variability has led to confusion in diagnosis using
pit pattern, resulting in a lack of consistency in correlating diagnosis and treatment regimens. Some investigators have suggested the risk factors for lymph node metastasis in submucosal invasive carcinomas. Of those, only the depth of invasion in the submucosal layer can be determined before imaging treatment. Therefore, it is important to make this distinction with respect to whether the lesion is treated endoscopically or surgically, and the treatment decision relies on an accurate estimation of this parameter.

Fujii et al. reported that because submucosal massive invasive carcinomas comprise carcinomas with high-grade atypia and desmoplastic changes, it is important to determine by magnifying colonoscopy whether the irregular pits occur in a demarcated area. Based on histological findings, they divided the magnifying configuration into three categories: non-neoplastic pattern, types I and II pit, which do not require treatment; non-invasive pattern, types III, IV and irregular (V) pit without a demarcated area, which are treated endoscopically; and invasive pattern, where the irregular pits can be traced clearly in a demarcated area and should be removed surgically. In a study of early colorectal carcinomas, they reported a diagnostic accuracy of 95% for non-invasive patterns and 83% for invasive patterns. This classification was suitable to suggest as a basis for therapeutic decision.

Here, we sought to evaluate whether our criteria according to Fujii’s classification was able to estimate invasion of the submucosal layer to a depth of 1000 μm, and to clarify the correlation between our criteria based on magnifying colonoscopy and the histopathological findings. The diagnostic accuracy obtained for type Non-V and type V was 95.1% and 82.1%, respectively, and 91.8% overall. In addition, our criteria proved useful in estimating the depth of invasion as well as Fujii’s.

With respect to the grade of atypia in adenocarcinomas, Watanabe et al. reported that because many invasive lesions are carcinoma with high-grade atypia, it is important to distinguish grades of atypia into low and high. In practice, it was difficult to distinguish a cellular atypia that satisfies Watanabe’s criteria by magnifying colonoscopy. However, our criteria showed that type V lesions reflected carcinoma with high-grade atypia.

The condition of the surface epithelium can be affected by a few factors, including invasion of carcinomas and appearance of DR, in addition to artifact. However, according to our results, the incidence of erosive change in the mucosal surface of type V was higher than in type Non-V, suggesting that the changes were due to the invasive process.

Desmoplastic reaction is observed in stromal areas when the carcinoma invades into the deep submucosal layer. By magnifying colonoscopy, type VN correlates with DR according to the finding of a decrease or disappearance of pits and, in such cases, massively invasive carcinomas are revealed histologically. In this study, the degree of DR was classified as (−) (+) or (+ +). Type V was observed in 88.0% of patients.

![Fig. 4. Relationship between staining medium and each macroscopic configuration to assess diagnosis of pit pattern. □, Indigo carmine (IC); ■, IC + crystal violet (CV).](image)

**Table 2. Assessment of misdiagnosis for magnifying diagnosis**

<table>
<thead>
<tr>
<th>Difference of atypia between surface and deeper area</th>
<th>Size (mm)</th>
<th>Macroscopic type</th>
<th>Pit</th>
<th>Depth of invasion</th>
<th>Grade of atypia in gland</th>
<th>Surface epithelium</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>Isp</td>
<td>Non-V (IV)</td>
<td>sm-d</td>
<td>Surface: LG, sm deepest area: HG</td>
<td>Preservation</td>
<td></td>
</tr>
<tr>
<td>20</td>
<td>Ip</td>
<td>Non-V (IV)</td>
<td>sm-d</td>
<td>Surface: LG, sm deepest area: mucinous component</td>
<td>Preservation</td>
<td></td>
</tr>
<tr>
<td>20</td>
<td>Is</td>
<td>Non-V (IV)</td>
<td>sm-d</td>
<td>Surface: LG, partly: HG</td>
<td>Preservation</td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>Isp</td>
<td>Non-V (IV)</td>
<td>m</td>
<td>Surface: HG</td>
<td>Erosion</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>IIa + IIc</td>
<td>V</td>
<td>adenoma</td>
<td>Erosion</td>
<td></td>
<td></td>
</tr>
<tr>
<td>25</td>
<td>IIc + Ila</td>
<td>V</td>
<td>sm-s</td>
<td>LG</td>
<td>Erosion, mucin</td>
<td></td>
</tr>
<tr>
<td>Simple misreading of pit pattern</td>
<td>30</td>
<td>IIa</td>
<td>V</td>
<td>m</td>
<td>LG</td>
<td>Preservation</td>
</tr>
<tr>
<td>15</td>
<td>IIa + IIc</td>
<td>V</td>
<td>sm-s</td>
<td>LG</td>
<td>Erosion</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>Is</td>
<td>Non-V (IIIa)</td>
<td>sm-d</td>
<td>HG</td>
<td>Erosion</td>
<td></td>
</tr>
</tbody>
</table>

Ip, pedunculated polyp; Isp, subpedunculated polyp; Is, sessile polyp; IIa, superficial elevated lesion; IIa + IIc, superficial elevated lesion with depression; sm-s, invasion of submucosal layer less than 1000 μm; sm-d, invasion of submucosal layer more than 1000 μm; LG, carcinoma with low-grade atypia; HG, carcinoma with high-grade atypia.
and the rate of DR (++) observed was 28%, and type V was reflected with the appearance of DR due to submucosal massive invasion. This result is diagnostically important to assess the depth of invasion, and to determine whether the lesion should be treated endoscopically or surgically.

Indigo carmine is used initially as a contrast medium to determine the contour and morphology of a lesion. CV staining is used as a secondary dye medium if IC is not good enough, but CV is an absorptive dye and may confer long-term toxicity according to some in vitro and animal studies. Therefore, CV should only be used when small pits and disrupted pits are observed by IC. The present study highlighted differences in the macroscopic configuration and the value of both mediums. Many protruding lesions were diagnosed by IC. It showed III type, IV type as well as IV type, because they were larger than other pit types. In addition, many of the type Non-V showing carcinoma with low-grade atypia and preserved surface epithelium were considered easier to diagnose by IC. In contrast, type IIIIs found on superficial depressed lesions, and type V with accompanying structural atypia and erosive change in the surface epithelium required CV staining to assess configuration and distribution of pits. Our results therefore show that it is important to select a contrast agent by macroscopic configuration, and that the staining might actually provide a marker that correlates with the histopathological findings.

There were a few cases of misdiagnosis in the present study so they could not be classified thoroughly, although some causes were possible (Table 2). In the cases that were misdiagnosed because of the different grades of atypia in the same glands, three of the four showed type IV pits that retained mucosal structure, and were classified histologically as carcinoma with mainly low-grade atypia. In addition, the entire misdiagnosed lesions were classified as protruding type macroscopically. Tamura et al. reported that because the mucosal glands in protruding type are thicker than those in superficial type, protruding types are more difficult to diagnose than superficial types with respect to depth of invasion. All lesions misdiagnosed due to a secondary erosive change in the mucosal surface were found in lesions accompanying a depressed area on the surface.

In conclusion, magnifying colonoscopy is a valuable procedure in the diagnosis of colorectal lesions to assess the depth of invasion, and correlates well with histopathological findings. It is particularly useful in evaluating configuration and the distribution of pits in a demarcated area. This procedure using our classification may provide the gastroenterologist with an objective aid in the treatment decision when lesions are detected by colonoscopy.

ACKNOWLEDGMENT

We thank Dr Takahiro Fujii for invaluable comments.

REFERENCES

21. Tanaka S, Haruma K, Teixeira CR et al. Endoscopic treat-


