

RESPONSE LETTER

Name of journal: *World Journal of Gastroenterology*

ESPS Manuscript NO: 28819

Title: Lymphovascular invasion in more than one-quarter of small rectal neuroendocrine tumors

Reviewer's code: 00699087

Dear Reviewers and editor

Thank you for the very kind and insightful comments on the manuscript. We have carefully reviewed the comments and have revised the manuscript accordingly. Our responses are given in a point-by-point manner below. Changes to the manuscript are shown in yellow highlighted text. A "Comments" section was also added according to the retrospective study guideline. We would be very honored if our manuscript is now considered acceptable for publication in the journal.

Sincerely,
Ho Suk Kang

~~~~~

**Comment 1)** I think it will be better if they contain more discussion of the importance of this study for future clinical problems (last paragraph of discussion section. I think there should be several cases of recurrence with more long time observation. Please emphasize these points more.).

**Response 1)** Thank you for your comment. I have inserted additional discussion in the last paragraph of the text.

"Small rectal neuroendocrine tumors have a favorable prognosis and successful outcomes following endoscopic resection. However, a low but real risk of metastasis remains, as in our results, and there are several cases of recurrence during long-time observation. Therefore, careful histologic examination for LVI and prospective studies with long-term follow up are needed to determine the natural course of small, endoscopically resected rectal NETs."

~~~~~

Comment 2) Last paragraph of Introduction "determined which clinicopathological risk factors are

associated with LVI” LVI itself might be the risk factor. So please reconsider this sentence, like determined the clinical impact of LVI or etc.

Response 2) Thank you for your comment. I have changed the sentence.

“In the present study, we used 2 methods, H&E and ancillary immunohistochemical staining (D2-40 and EVG), to compare the detected rate of LVI in 104 endoscopically resected small rectal NET specimens and to **determine the clinical impact of LVI.**”

.....

Comment 3) What is the indication of endoscopic resection of NETs? What kind of patients underwent surgical resection? Please describe the indication.

Response 3) Thank you for your comment. The first submission lacked an adequate description about the indications, so we have revised the methods section to add these details.

“Between June 2005 and December 2015, 138 patients with 139 tumors were diagnosed with rectal NET at Hallym university sacred heart hospital in Anyang, Korea. **Endoscopic gross tumor size ≤10 mm, and absence of lymph node involvement or distant metastasis on the abdominal CT were the indications for endoscopic resection.** The study inclusion criteria for small rectal NETs were as follows: (i) a tumor ≤10 mm, in diameter histologically, (ii) a tumor within 15 cm of the anus, (iii) no metastasis to lymph nodes or distal organs detected on abdominal computed tomography, and **(iv) a tumor resected in our institution for the first time.** Therefore, the following cases were excluded from this analysis: **2 patients who underwent radical surgical excision with lymph node dissection owing to large tumor (3 cm and 5 cm), 4 who underwent transanal resection based on the decision of the outpatient clinic surgeon regardless of size, 7 who underwent additional transanal resection after incomplete endoscopic resection at other clinics, 12 who did not undergo additional treatment after diagnosis, 4 who were treated at other clinics, 4 who could not be evaluated for LVI owing to an insufficient specimen, and 2 with endoscopically resected tumors exceeding 1 cm (1.2 cm and 1.7 cm).** As a result, 103 patients with 104 rectal NETs were included in this study; the related medical records were reviewed retrospectively.”

.....

Comment 4) Results What were the reasons for additional surgery in three patients? Also please provide more detail of the patients with lymph node metastasis (tumor size, LVI rate etc.)

Response 4) Thank you for your comment. The first submission lacked an adequate description about these patients, so we have changed the results section to add these details.

“Three patients underwent additional surgery **owing to the presence of LVI in our primary histologic reports before this study;** among them, 1 patient had lymph node metastasis. **This 21 year old man’s histologic evaluation showed a 5 mm tumor size, a Ki 67 index < 3%, and < 2 mitosis/10 HPFs; however, the vertical margin and lymphatic invasion were positive on the ESD specimen.”**

~~~~~

**Comment 5)** Several spelling error 1. Put spaces ① P6 line 17: 13 patients→13 patients ② P7 line 7: sub mucosal → submucosal ③ P8 line 3 from the bottom: werethen→were then ④ P10 line 3: 0.24 mm, 1.0 mm→0.24 cm, 1.0 cm (Please confirm) ⑤ P10 line4 from the bottom: 26(25.0%)→26 (25.0%) ⑥ P11 line 2: significant (P=0.648).LVI as...→). LVI (put space) ⑦ P15 line 3: ... of 4 a mm→of 4 mm

**Response 5)** We have made these corrections. Thank you very much.

~~~~~

COMMENTS

Background

Rectal neuroendocrine tumors (NETs) arise from enterochromaffin endocrine cells and are found incidentally during sigmoidoscopy or colonoscopy. On endoscopy, they typically appear as sessile, subepithelial tumors covered with yellow, discolored epithelium. Rectal NETs ≤ 10 mm in diameter, within the mucosal or submucosal layer, can be treated with endoscopic resection and have a good prognosis. However, lymphovascular invasion (LVI), a well-established risk factor for lymph node metastasis, is often found in endoscopically resected specimens, and there are no definite guidelines about these cases. Therefore, we investigated the frequency and prognostic significance of LVI in small, endoscopically resected rectal NETs

Research frontiers

Immunohistochemical analysis is not currently recommended for routine use to identify LVI in NETs. However, for accurate and reliable diagnosis of LVI, we undertook additional immunohistochemical staining using D2-40 and Elastica van Gieson staining to confirm the presence of LVI.

Innovations and breakthroughs

We observed LVI rates of 25% and 27.9%, higher than previously reported, through hematoxylin and eosin and additional immunohistochemical staining. On the other hand, LVI was not associated with lymph node metastasis or recurrence in small rectal NETs (<1 cm) during a 3 year-follow up period.

Applications

After endoscopic resection of rectal NETs, even in small tumors (≤ 10 mm), careful histologic examination for LVI is needed. Furthermore, long-term prospective studies are required to determine the natural course of endoscopically resected rectal NETs.

Terminology

NET: Neuroendocrine tumor, LVI: Lymphovascular invasion

Peer-review

In this article, the authors analyzed the frequency of lymphovascular invasion in endoscopically resected small rectal neuroendocrine tumors by precise methods and compared these results with conventional hematoxylin and eosin staining. By these methods, they found that the frequency of lymphovascular invasion was higher than the previously reported ratio. Although they could not determine the relationship between lymphovascular invasion and clinical outcome, such as survival and recurrence, this study provides very important insights for future study.