NO: 89039

Dear Dr. Kurtcehajic,

We are pleased to inform you that, after preview by the Editorial Office and peer review, we believe that the academic quality, language quality, and ethics of your manuscript (**Manuscript NO.: 89039, Observational Study**) basically meet the publishing requirements of the World Journal of Gastrointestinal Endoscopy. As such, we have made the preliminary decision that it is acceptable for publication after your appropriate revision.

Please resolve all issues in the manuscript based on the peer review report and make a point-by-point response to each of the issues raised in the peer review report, and highlighted the revised/added contents with yellow color in the revised manuscript. Note, authors must resolve all issues in the manuscript that are raised in the peer-review report(s) and provide point-by-point responses to each of the issues raised in the peer-review report(s); these are listed below for your convenience:

Dear Editor,

Thank you for reviewing our manuscript and for giving us the opportunity to revise it according to the reviewrs' comments. We hope that we have made satisfactory adjustments according to the reviewer's comments and so improved the manuscript. The comments have been answered point by point.

Reviewer #1:

Specific Comments to Authors:

Conclusion: Accept (Minor revision) In this study, by comparing the microscopic details of gastric mucosa with WLE, NF, NF- NBI, AA-NF and AA-NF- NBI, NF- NBI is the most effective endoscopic model for evaluating RAC, SECN and GP, and AA-NF-NBI is the most effective endoscopic model for analyzing CO and IP. It provides a higher resolution observation tool for evaluating the relationship between the progress of gastric diseases and the existence of gastric venules, the regularity/irregularity of capillary network, the shape and size of gastric fossa and recess.

We thank the reviewer for the suggestion for improving conclusion of the study. Regards suggestion we have changed Conclusion section of the main text.

This article is concise and easy to read, which leads us to understand the optimization process of endoscope. However, the advantages of NF-NBI or AA-NF-NBI in evaluating the microscopic details of gastric mucosa and the practical application of clinical diagnosis have not been further explained. I reserve my opinion that the application of NF-NBI reduces the need for biopsy (pathological specimen, RUT) and other laboratory and serological tests related to gastric mucosal diseases. To achieve scientific publishing standards, several suggestions need to be considered and the quality of manuscripts should be seriously improved. Details are as follows:

- This study puts forward the advantages of NF-NBI in observing the gastric microstructure, and wants to understand the gastric microstructure and disease progress through NF-NBI. However, the background of gastric microstructure RAC, SECN, GP, CO and IP related to disease progression is rarely introduced in this paper. It is suggested that the introduction of the relationship between gastric microstructure RAC, SECN, GP, CO and IP and disease progression should be supplemented in the introduction and discussion to explain the importance of NF-NBI in observing gastric microstructure. This part is very important to attract interests of the readers.

We thank the reviewer for the suggestion. We have made changes to the main text (Introduction section, 1st paragraph and Discussion section, 3th paragraph) in accordance with the recommendation. Also in the Article highlights section (Research perspectives) we highlighted relation between the microscopic features of stomach mucosa and progression of gastric diseases.

- Many studies have reported that NF-NBI has successfully replaced ME-NBI in the diagnosis of pathological changes and diseases of pharynx, esophagus and stomach. So what is the innovation of NF-NBI in high-resolution observation of gastric microstructure?

NF-NBI endoscopic mode contrast to the conventional magnification-NBI has provided the visualisation of gastric crypts.We have higlighted it in Discussion section, 2nd paragraph.
"*In the NF-NBI mode, from the point of view all three observers, besides the RAC, SECN, GP and crypts were clearly seen at levels of 95.5%, 97% and 92.6%, respectively*"
Additionally with applying the acetic acid gastric crypts would be analysed in more detail (shape and size).We have higlighted it in Discussion section, 4th paragraph.
"*By applying the acetic acid in the area of observation, we highlighted a unique pattern containing regular round brown cryprs*"

- In the material method, I have doubts. The author mentioned that "340 images were classified in the same order into five groups regarding the above endoscopic modalities." This grouping method related to the inspection method of speculum will not affect the observer's subjective score.

We thank the reviewer for the suggestion.
We have made changes in the Abstract section (Results).
"*Regarding the WLE, NF and NF-NBI endoscopic modes 204 images were classified in the same order into three groups*"
We have made changes in the Result section, 1st paragraph of the main text.
"*Regarding the AA-NF and AA-NF-NBI endoscopic modes, 136 images (couple by the patient) were classified in the same order*"
The same order per endoscopic mode is mandatory for the interobserver diagnostic agreement - Kappa value.
I hope with the additional clarifications as well as the changes we made in the text, we removed the ambiguities pointed out by the reviewer and in this way we made the text clear.

- The article should pay attention to typesetting problems, such as the indentation of the first line of a paragraph needs to be unified.

Thank to the suggestion. We accept reviewer's remark and we have made changes.

- Note that the picture annotation of the article should be consistent with the text part. The thumbnail in figure 1 includes A, B, C, D, but the caption in the part of Endoscopic patterns and scoring and the table 1 show 1a, 2a, B, C, without D. The same problem occurs in figure 3.

Thank to the suggestion.We have corrected it.

- The format of Tables are suggested to be modified, and an intuitive three-line table is used to mark the meaning of rows and columns.

Thank to the suggestion. We accept reviewer's remark and we have made changes.

- The title should be changed to a specific one, related to the conclusion.

We thank the reviewer for the suggestion. We have corrected it.

Near focus endoscopy, narrow-band imaging and acetic acid in the visualization of stomach mucosa (*was*)

Could near focus endoscopy, narrow-band imaging and acetic acid improve the visualization microscopic features of stomach mucosa? (*now*)

EDITORIAL OFFICE'S COMMENTS

Authors must revise the manuscript according to the Editorial Office's comments and suggestions, which are listed below:

(1) Science editor:

1 Conflict of interest statement: Academic Editor has no conflict of interest.

2 Academic misconduct: No academic misconduct was found.

3 Scientific quality: The author submitted a study of near focus endoscopy, narrow-band imaging and acetic acid in the visualization of stomach mucosa. The manuscript is overall qualified.

4 Language evaluation: The English-language grammatical presentation needs to be improved to a certain extent. There are many errors in grammar and format, throughout the entire manuscript. Before final acceptance, the authors must provide the English Language Certificate issued by a professional English language editing company. Please visit the following website for the professional English language editing companies we recommend: https://www.wjgnet.com/bpg/gerinfo/240.

5 Specific comments: (1) Please provide the Figures cited in the original manuscript in the form of PPT. All text can be edited, including A,B, arrows, etc. With respect to the reference to the Figure, please verify if it is an original image created for the manuscript, if not, please provide the source of the picture and the proof that the Figure has been authorized by the previous publisher or copyright owner to allow it to be redistributed. All legends are incorrectly formatted and require a general title and explanation for each figure. Such as Figure 1 title. A: ; B: ; C: .

6 Recommendation: Transfer to other BPG journals (World Journal of Gastrointestinal Endoscopy).

Language Quality: Grade C (A great deal of language polishing)

Scientific Quality: Transfer to another BPG Journal

(2) Company editor-in-chief:

I recommend the manuscript to be published in the World Journal of Gastrointestinal Endoscopy. The 2023 Edition of Journal Citation Reports® cites the 2022 impact factor (IF) for WJGE as 2.0.

When revising the manuscript, it is recommended that the author supplement and improve the highlights of the latest cutting-edge research results, thereby further improving the content of the manuscript. To this end, authors are advised to apply PubMed, or a new tool, the RCA, of which data source is PubMed. RCA is a unique artificial intelligence system for citation index evaluation of medical science and life science literature. In it, upon obtaining search results from the keywords entered by the author, "Impact Index Per Article" under "Ranked by" should be selected to find the latest highlight articles, which can then be used to further improve an article under preparation/peer-review/revision. Please visit our RCA database for more information at: https://www.referencecitationanalysis.com/, or visit PubMed at: https://pubmed.ncbi.nlm.nih.gov/.

I recommend the manuscript to be published in the World Journal of Clinical Cases. Before final acceptance, when revising the manuscript, the author must supplement and improve the highlights of the latest cutting-edge research results, thereby further improving the content of the manuscript. To this end, authors are advised to apply a new tool, the Reference Citation Analysis (RCA). RCA is an artificial intelligence technology-based open multidisciplinary citation analysis database. In it, upon obtaining search results from the keywords entered by the author, "Impact Index Per Article" under "Ranked by" should be selected to find the latest highlight articles, which can then be used to further improve an article under preparation/peer-review/revision. Please visit our RCA database for more information at: https://www.referencecitationanalysis.com/.

Corrected according to the suggestion.

At the end, we would like to thank for patience, time and useful suggestions that helped us to improve our Orginal article (Observational study).
We hope that in this revised form it is going to be acceptable for your respected journal.

With deepest respect, Dr Admir Kurtcehajic
On behalf of all authors.

Name of Journal: **World Journal of Gastrointestinal Endoscopy**

Manuscript NO: **89039.** Manuscript Type: **ORIGINAL ARTICLE**

Observational Study, Title:

~~Near focus endoscopy, narrow-band imaging and acetic acid in the visualization of stomach mucosa~~

Could (can) near focus endoscopy, narrow-band imaging and acetic acid improve the visualization microscopic features of stomach mucosa?

Short title: Endoscopic microanatomy of the gastric mucosa

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Word count ~~3155~~

Conflict of interest disclosure and funding declaration:

Admir Kurtcehajic, Enver Zerem, Tomislav Bokun, Ervin Alibegovic, Suad Kunosic, Ahmed Hujdurovic, Amir Tursunovic and Kenana Ljuca have no conflicts of interest or financial ties to disclose.

ABSTRACT

Background: Conventional magnifying endoscopy with narrow-band imaging (NBI) observation of the gastric body mucosa, reporting dominant patterns in relation to regular arrangement of collecting venules (RAC), subepithelial capillary network (SECN) and gastric pits (GP).

Aims: In this study, we evaluate the possibilities of a new one - dual (near) focus, NBI mode, in the assessment of the microscopic features of gastric body mucosa, which we use instead of conventional magnification.

Methods: During 2021 and 2022, 68 patients underwent proximal gastrointestinal endoscopy using magnification endoscopic modalities, subsequently applying acetic acid (AA). The scope GIF-190HQ series NBI system, with dual focus capability, was used for the investigation of gastric mucosa.At the time of the endoscopy, the gastric body mucosa of all enrolled patients was photographed using the white light endoscopy (WLE), near focus (NF), NF-NBI,AA-NF and AA-NF-NBI modes.

Results: From all patients,~~340~~regarding the WLE, NF and NF-NBI endoscopic modes 204 images were classified in the same order into ~~five~~ three groups.~~the above endoscopic modalities.~~Regarding the AA-NF and AA-NF-NBI endoscopic modes, two images by the same patient in the couple were classified in the same order. According to all three observers independently, NF magnification was significantly superior to WLE ($p<0.01$), and the NF-NBI mode was significantly superior to NF magnification ($p<0.01$). After applying the AA, according to all three observers independently, AA-NF-NBI was significantly superior to AA-NF ($p<0.01$). Interobserver kappa values for WLE were 0.609, 0.704 and 0.598, respectively, and in the case of NF magnification, they were 0.600, 0.721 and 0.637, respectively. For the NF-NBI mode, the values were 0.378, 0.471 and 0.553, respectively, for AA-NF, they were 0.453, 0.603 and 0.480, respectively and for AA-NF-NBI, they were 0.643, 0.506 and 0.354, respectively.

Conclusion: When investigating gastric mucosa at microscopic detail level, among the five endoscopic modalities which took part in this study, NF-NBI was the most powerful endoscopic mode for assessing RAC, SECN and GP. AA-NF-NBI was the most powerful endoscopic mode for analysing crypt opening and intervening part.

Keywords: gastric mucosa, endoscopic microanatomy, magnifying endoscopy, near focus,

narrow-band imaging, acetic acid

Core tip: Narrow-band imaging have enabled the analysis gastrointestinal mucosa at microscopic detail level. However, this technique gives a dark image and makes it impossible to identify the colour and structural microanatomy changes of the stomach mucosa, and it is necessary to combine it with the mechanical addict on the top of the scope (rubber) - conventional magnification. These additions improve the visualization of the gastric mucosa, but significantly complicate the procedure. We present a new endoscopic mode called “near focus” that achieves the same or better visualisation and doesn’t require any additional accessories (rubbers).

Abbrevations:

AA - acetic acid

CO - crypt opening

GP - gastric pits

HP - Helicobacter pylori

HD – high definition

IEE – image enhanced endoscopy

IP - intervening part

ME - magnifying endoscopy

NBI – narrowband imaging

NF – near focus

RAC - regular arrangement of collecting venules

RUT - rapid urease test

SECN - subepithelial capillary network

WLE - white light endoscopy

INTRODUCTION

Currently, endoscopic platforms offer high-resolution images, image-enhanced endoscopy (IEE) techniques and magnification, allowing for the inspection of gastric mucosa at a detailed level.

Endoscopic microscopic features (microanatomy) of the gastric body mucosa are classified into the microvascular architecture, such as the regular arrangement of collecting venules (RAC) and the regular honeycomb subepithelial capillary network (SECN). The micro-surface structure is characterized by regular round gastric pits (GP), regular oval crypt opening (CO) and the intervening part (IP), which constitutes the space between the crypts. In gastric related diseases such as Helicobacter pylori (HP) infection, intestinal metaplasia, and gastric atrophy the microanatomy has been structurally changed - absence of venules, irregularity of capillary network, enlarging pits and crypts [1, 2, 3, 4, 5, 6].

In previous reports, white light endoscopy (WLE) has failed to assess endoscopic microanatomy. The need and wish to improve the differentiation of normal, inflammatory, and malignant lesions by gastrointestinal endoscopy has fuelled research to accelerate the development of novel types of video endoscopy systems, based on new optical technologies. Electronic chromoendoscopy via the most useful tool in the many clinical trials, namely, narrow-band imaging (NBI), has highlighted the vascular patterns of gastric mucosa [1, 3, 6, 7, 8].Three studies focused on acetic acid (AA) which enhances and determines the pathology of the gastric lesions [9, 10, 11].

Aiming to assess the endoscopic microscopic features in more detail, there has been a need for magnification. Conventional magnifying endoscopy (ME)with NBI observation of the normal gastric mucosa has been described previously; studies reported dominant patterns with RAC, SECN and GP [2, 3, 4, 6, 7].

Recent, optical innovation relating to the dual focus function allows the endoscopist to select between a normal mode and a near focus (NF) mode. The NF mode is optimized for near-field observation with 45-fold magnification. Studies have reported that NF-NBI successfully replaced ME-NBI in the detection of pathological lesions in the oesophagus and pharynx, as well as the identification of celiac disease [12, 13, 14, 15]. Recent studies reported the ability of the NF-NBI mode in the detection of

early gastric cancer lesions [16, 17]. Two recently published studies considered NF endoscopy in the evaluation of gastric atrophy, intestinal metaplasia, and Helicobacter pylori infection [18, 19].

Conventional ME using a soft rubber at the top of the scope due to the demanding manipulation in stomach remaining to be finally replaced with the simple, more pragmatic, and novel endoscopic way of magnification. Therefore, in this study we evaluate the possibilities of a new one - dual (near) focus, NBI mode, in the assessment of RAC, SECN, GP and even more CO in the gastric body (microscopic features of the mucosa), which we use instead of conventional magnification.

The first aim of this study was to determine the clinical usefulness of the NF-NBI mode in the observation of gastric microvascular architecture and micro-surface structure.

Secondly, by applying the 1.5% AA, we compare the power of visualization of the AA-NF and AA-NF-NBI mode, when assessing micro-surface patterns containing CO and IP.

MATERIALS AND METHODS

Patients

Between *September 2021* and *May 2022*, 68 patients underwent proximal gastrointestinal endoscopy using conventional WLE, NF magnification and the NF-NBI mode, with the subsequent application of AA. The patients consisted of 30 men and 38 women: the mean age was 38.5 years and the range, 25-65 years.

The study excluded patients with HP positive, either one serology or rapid urease test (RUT), those who had received anticoagulant therapy or drugs for chronic metabolic diseases (diabetes mellitus, hypothyroidism) and systemic inflammation disease, as well as nonsteroidal anti-inflammatory drugs and anxiolytics, and patients with chronic decompensated liver and kidney diseases.

The study was approved by the ethical committee of the Blue Medical Group, and written, informed consent was obtained from all participants.

Endoscopic modalities

The endoscopic video information system, EVIS EXERA III CLV-190, was used with an Olympus high-resolution endoscope, GIF-190HQ series NBI system, with dual focus capability for the investigation of the gastric mucosa. This scope allows to switch between two focus settings: “normal mode” and “near focus mode”. The “normal mode” or WLE suits normal observation at a distance of 5–100 mm and a 170° field of view, while the NF magnification of up to 70X allows close observation of the finest mucosal surfaces, at a distance of 2–6 mm. When switching to the NF mode at the simple touch of a button, the field of view will remain almost the same (160°).

NBI is based on the principle that depth of light penetration into tissues is directly proportional to the wavelength, which implies that the shorter the wavelength, the more superficial the penetration. The NBI resembles chromoendoscopy without dye, focusing on the capillaries.

AA via magnification enables vivid observation of the CO of the glandular epithelium, which has a deep brown appearance; the IP has a whitish appearance, because of reversible alterations in the

molecular structure of the cellular proteins that are induced by the AA, and last from several seconds to minutes.

Endoscopic procedure

Two hours before the procedure all patients took 80 mg of simethicone with a small amount of water, aiming to remove gastric mucus; the procedures were performed under the intravenous application of the propofol. A single experienced gastroenterologist (AK) performed all the procedures. The whole oesophagus, stomach and duodenum were examined to exclude obvious lesions with the conventional WLE, followed additionally by the manufacturer incorporated NF-NBI mode in the scope, by applying the 3 ml AA via a single catheter. The focus area was the anterior wall or greater curvature of the upper gastric body. Biopsies were taken from the antrum and corpus mucosa and RUT was performed to evaluate HP infection.

Endoscopic patterns and scoring

At the time of the endoscopy, the gastric body mucosa of all enrolled patients was photographed using the WLE, NF, NF-NBI, AA-NF and AA-NF-NBI modes.

Regarding the WLE, NF and NF-NBI endoscopic modalities, the regular/clear appearance of each mucosa microscopic feature, RAC, SECN, GP and CO scored 1 point; the unclear appearance of each scored a half point and the absence of each scored 0.

In relation to the WLE mode, the WLE-1a and WLE-2a patterns clearly show the RAC and score 1 point; WLE-2a presents a faded appearance of the RAC; the WLE-b pattern shows the RAC less clearly and scores a half point and the WLE-c pattern does not show the RAC and scores 0 (*Figure 1*).

With reference to the NF magnification, the NF-a pattern clearly shows the RAC, SECN and GP and scores 3 points. The NF-b pattern shows the RAC less clearly but clearly shows the SECN and GP and scores 2.5 points. The NF-c pattern does not show the RAC but clearly shows the SECN and GP and scores 2 points (*Figure 2*).

Regarding the NF-NBI endoscopic visualization, the NF-NBI-1a and NF-NBI-2a patterns clearly show the RAC, SECN, GP and CO and score 4 points; the 2a pattern has less distribution of the RAC and a

slightly enlarged GP and CO than the 1a pattern. The NF-NBI-b pattern does not show the RAC, clearly shows the SECN and GP, shows the CO less clearly and scores 2.5 points (*Figure 3*).

According to the aforementioned scoring rules, one pattern could score the most points (4) (clear presence of all microscopic features) or the least number of points (0) (absence of all microscopic features).

After enhancing the area of observation with AA, a pattern was suddenly visible on the AA-NF and AA-NF-NBI, containing CO and IP. On the AA-NF mode, the pattern shows small oval brown CO and light white IP. On the AA-NF-NBI mode, the pattern shows small oval black CO and dense white IP (*Figure 4*). The strong contrast between the CO regarding the shape/size and the IP on these two patterns within the same patient grades with 1 point, medium contrast grades with a half point and low contrast grades with 0 points. Endoscopic patterns were observed and scored by three independent endoscopists.

Statistical analysis

The differences between the scoring of each endoscopic modality for all observers were compared using the Wilcoxon Matched Pairs Test. P values < 0.05 were considered significant.

The interobserver diagnostic agreement was analysed with a kappa value. In theory, perfect disagreement has a kappa value of −1, and perfect agreement has a kappa value of +1. A value of 0 means an agreement by chance alone. As per the Landis and Koch scale, kappa values were graded as follows: 0.01–0.2 slight, 0.21–0.4 fair, 0.41–0.6 moderate, 0.61–0.8 substantial and 0.81–1.0 almost perfect. Cohen's suggested interpretation may be too lenient for health-related studies because it implies that a score as low as 0.41 might be acceptable (9, 14, 20). Data were analysed using SPSS 23 (IBM, US).

RESULTS

Following the meeting criteria relating to long-term epigastric discomfort and non-specific abdominal pain, the study initially included 74 patients. During the endoscopy, four patients (three male, one female) did not undergo NF magnification, due to one patient having benign stenosis of the distal oesophagus, one having cancer of the cardia and two having pyloric stenosis. Two patients (one male, one female) were ruled out due to severe bile reflux.

Finally, 68 patients underwent proximal gastrointestinal endoscopy, using the WLE, NF and NF-NBI modes to analyse the microscopic features of gastric body mucosa. From all patients, 204 images were classified in the same order into three groups, in relation to the above endoscopic modality by endoscopist AK; they were observed separately and scored by two experienced endoscopists, JF (observer I) and PJ (observer II) and one inexperienced endoscopist, OZ (observer III).

Moreover, after applying AA in the area of observation, the CO and IP were suddenly visible. Regarding the AA-NF and AA-NF-NBI endoscopic modes, 136 images (couple by the patient) were classified in the same order. The contrast between the CO and IP in the same patient was observed and graded separately by the three observers.

All observers had previously passed a live course regarding NF magnification and NBI chromoendoscopy mode with AA enhancing. The course was based on the 12 endoscopic patterns which would form part of this study; each pattern presented with 10 images.

The frequency and scoring for the WLE, NF and NF-NBI endoscopic modalities from the point of view of all three observers are shown in *Table 1*. According to the experienced observers (observer I and observer II), NF magnification was significantly superior to WLE ($p<0.01$) and the NF-NBI mode was significantly superior to NF magnification ($p<0.01$). Regarding the third inexperienced observer, NF magnification was significantly superior to WLE ($p<0.01$) and the NF-NBI mode was significantly superior to the NF magnification ($p<0.01$).

The frequency and scoring for the AA-NF and AA-NF-NBI endoscopic modalities from the point of view of all three observers are shown in *Table 2*. According to the experienced observers (observer I

and observer II), AA-NF-NBI was significantly superior to AA-NF ($p<0.01$). For the third inexperienced observer, AA-NF-NBI was significantly superior to AA-NF ($p<0.01$).

The interobserver diagnostic agreement for all five endoscopic modalities was analysed with a kappa value. Interobserver kappa values for WLE were 0.609 for observer I and observer II, 0.704 for observer I and observer III and 0.598 for observer II and observer III. Interobserver kappa values for NF magnification were 0.600 for observer I and observer II, 0.721 for observer I and observer III and 0.637 for observer II and observer III. Interobserver kappa values for the NF-NBI mode were 0.378 for observer I and observer II, 0.471 for observer I and observer III and 0.553 for observer II and observer III. Interobserver kappa values for AA-NF were 0.453 for observer I and observer II, 0.603 for observer I and observer III and 0.480 for observer II and observer III. Interobserver kappa values for AA-NF-NBI were 0.643 for observer I and observer II, 0.506 for observer I and observer III and 0.354 for observer II and observer III.

DISCUSSION

According to the results of our research into the investigation of gastric mucosa at a detailed microscopic level, of the five endoscopic modalities used in this study, NF-NBI was the most powerful endoscopic mode for evaluating the RAC, SECN and GP, and AA-NF-NBI was the most powerful endoscopic mode for analysing the CO and IP. There have been many advances in endoscopic imaging technologies. Standard definition endoscopy produces image signals with a resolution of one to four hundred thousand pixels. High-resolution or high-definition (HD) endoscopy produces image signals with a resolution of up to a million pixels, which has the same effect as visualizing a surface at a 30- to 35-fold magnification. A novel IEE technique is electronic chromoendoscopy, which includes NBI, i-Scan and flexible spectral imaging colour enhancement. Over the last 15 years, NBI has been used most in clinical practice, however, it is too dark to identify the colour and structural mucosa changes in organs with a large lumen, such as the stomach, therefore, these should be combined with ME [1, 6, 8, 14].

Over the last two decades, conventional ME was carried out with a soft rubber; before the procedure, soft black rubber was attached at the top of the scope. In this way, the area of interest was magnified but the view for normal observation was reduced. Magnification with soft rubber requires skill and special training and for this reason, ME is not frequently used in Western countries [1, 2, 3, 4, 6].

Several studies using HD endoscopy, IEE, conventional magnifying, and histopathology have confirmed the normal appearance of gastric body mucosa with the RAC, SECN and GP[1-3, 6-8, 21-25].

In our study, we successfully replaced the conventional ME with the dual focus (NF magnification). Regarding our results, all three observers noted independently that the NF magnification showed more power of visualization than WLE ($p<0.01$). NF magnification from the point of view of all three observers assessed the microscopic features of the gastric body mucosa, such as the RAC at 94%, 95% and 92%, respectively and the SECN and GP at 100%, in the same way as it was reported in studies

powered by the conventional ME [4, 22, 23]. The diagnostic interobserver agreement for the NF magnification showed a “substantial” level.

A retrospective study from the United Kingdom demonstrated that NF magnification improved the diagnostic yield of upper gastrointestinal mucosal lesions, however, its usefulness for gastric lesions is questionable [15].

In our study, all three observers noted independently that the NF with the NBI showed significantly more power of visualization than the NF magnification ($p<0.01$). In the NF-NBI mode, from the point of view all three observers, besides the RAC, SECN, GP and CO were clearly seen at levels of 95.5%, 97% and 92.6%, respectively. The diagnostic interobserver agreement in relation to the NF-NBI mode from the perspective of the experienced observers (one side) and the third inexperienced observer was at the “moderate” level. The diagnostic interobserver agreement among the experienced observers was at the “fair” level. The clinical explanation could be that NF-NBI “1a” and “1b” patterns were scored the same and that the differences were qualitative (distribution of the RAC, size of GP and CO). In the progression of gastric related diseases microscopic features of the stomach mucosa such as venules, capillary network, the shape and size of gastric fossa and recess have been structurally changed.

For the first time, two studies used the NF-NBI mode in the stomach for the evaluation of a tumour lesion and its margin [16, 17].The role of the NF-NBI mode has been assessed recently for atrophic gastritis, according to the Kimura-Takemoto classification and intestinal metaplasia (tubular/granular GP pattern of the corpus) [18].This study considered the shape of the GP without analysing the RAC, SECN and CO. In the absence of the previously verified gastric NF-NBI magnification pattern, it couldn't be appropriate define the pathology pattern.

In a recently published study by Fiuza et al., NF magnification was evaluated for the assessment of the mucosal surface pattern in HP related gastritis [19]. This study used the NF mode without NBI chromoendoscopy and was unable to consider the appearance of the CO.

Conventional ME requires more time, skills, and special endoscopic training; the mechanical addict on the top of the scope (rubber) and the view of visualization becomes less. On the other hand, NF magnification may be easily manipulated, there is no need for the mechanical attachment, the view of

visualization remains normal and finally, in our study, NF magnification with NBI chromoendoscopy beside RAC, SECN and GP provided presence of CO.

The current era of NF magnification endoscopic technology may be contrasted with the research of Cho et al. [25] who recently used conventional ME for the evaluation of HP associated gastritis. The forthcoming studies via NF-NBI endoscopy aim to evaluate the presence/absence of RAC, the regularity/irregularity of SECN and the shape and size of the GP and CO in relation to the HP infection, intestinal metaplasia, and atrophic gastritis, etc.

In the previous, aforementioned studies, the focus on the CO was less; a study by Kawamura et al. [26] evaluated the role of conventional ME (without NBI) by analysing the CO as part of HP related gastritis. The whiteness of the CO was classified as the “white-edged dark spot” type, the “white” type and the “dense white pit” type.

Regarding our results, the NF-NBI mode assessed the presence of the CO as a black point, but to visualize the CO in more detail, enhancement was carried out using AA. By applying the AA in the area of observation, we highlighted a unique pattern containing regular round brown CO and whiteish IP in the normal gastric body mucosa. In the AA-NF-NBI mode, the CO suddenly became black, consequently there is a clear contrast between the CO and IP.

Our results, independently noted by all three observers, showed that AA-NF-NBI was superior to AA-NF, $p<0.01$. Our results clearly push up the AA-NF-NBI mode in terms of analysing the shape and size of the CO and IP. The diagnostic interobserver agreement for the AA-NF-NBI mode among the experienced observers was at a “substantial” level. The diagnostic interobserver agreement between the experienced observers (one side) and the third inexperienced observer was at the “moderate” level and “fair” level, respectively. An explanation for this could be that there was no question about the existence of the contrast, instead the question related to the grade of the contrast between the CO and IP.

One limitation of this study (as well as other studies related to this issue) is the relatively small number of patients included in the research. Additional research (preferably randomized trials or prospective collaborative studies) is required in order to improve the endoscopic investigation of gastric mucosa at

a detailed microscopic level and to create the conditions for a better diagnosis and treatment of these diseases.

CONCLUSION

~~When investigating gastric mucosa at microscopic detail level, among the five endoscopic modalities which took part in this study, NF-NBI was the most powerful endoscopic mode for assessing RAC, SECN and GP. AA-NF-NBI was the most powerful endoscopic mode for analysing CO and IP. Our results have brought clinical and endoscopic usefulness of the NF-NBI and AA-NF-NBI; in the case of presenting RAC, regular appearance of SECN, GP, CO, and IP there is reduced need for the biopsy (pathology specimen, RUT), and other laboratory, serology tests related to the gastric mucosa diseases.~~ In this study, by investigating the microscopic details of gastric body mucosa among the five endoscopic modalities which took part in this study, NF-NBI was the most effective endoscopic mode for evaluating RAC, SECN and GP. AA-NF-NBI was the most effective endoscopic mode for analysing CO and IP. It provides a higher resolution observation tool for evaluating the relationship between the progress of gastric diseases and the existence of gastric venules, the regularity/irregularity of capillary network, the shape and size of gastric pits and recess.

ARTICLE HIGHLIGHTS

Research background

Narrow-band imaging (NBI)is too dark to identify the colour and structural microanatomy of stomach mucosa due to large lumen, therefore, these should be combined with magnification.

Research motivation

Conventional magnification endoscopy using a soft rubber at the top of the scope due to the demanding manipulation in stomach remaining to be finally replaced with the simple, more pragmatic, and novel endoscopic way of magnification.

Research objectives

We evaluate the possibilities of a near focus (NF) magnification, NBI mode with acetic acid (AA), in the assessment of venules, capillary network, pits and crypts in the gastric body mucosa.

Research methods

The endoscopic video information system, EVIS EXERA III CLV-190, was used with an Olympus high-resolution endoscope, GIF-190HQ series NBI system, with dual focus capability for the investigation of the gastric mucosa. At the time of the endoscopy, the gastric body mucosa of all enrolled patients was photographed using the white light endoscopy (WLE), NF, NF-NBI, AA-NF and AA-NF-NBI modes.

Research results

From 68 patients, 204 images were classified in the same order into three groups (WLE, NF and NF-NBI). They were observed separately and scored by two experienced endoscopists and one inexperienced endoscopist. According to all three observers independently, NF magnification was significantly superior to WLE ($p<0.01$), and the NF-NBI mode was significantly superior to NF magnification ($p<0.01$). Interobserver kappa values for WLE were 0.609, 0.704 and 0.598, respectively, and in the case of NF magnification, they were 0.600, 0.721 and 0.637, respectively. For the NF-NBI mode, the values were 0.378, 0.471 and 0.553, respectively.

For the endoscopic modalities with the AA, according to all three observers independently AA-NF-NBI was significantly superior to AA-NF ($p<0.01$). Interobserver kappa values for the AA-NF were

0.453, 0.603 and 0.480, respectively and for AA-NF-NBI, they were 0.643, 0.506 and 0.354, respectively.

Research conclusions

Among the five endoscopic modalities which took part in this study, NF-NBI was the most powerful endoscopic mode for assessing venules, capillary network, and gastric pits. AA-NF-NBI was the most powerful endoscopic mode for analysing crypts and space between crypts.

Research perspectives

The forthcoming studies via NF-NBI and AA-NF-NBI endoscopic modalities aim to evaluate the presence/absence of venules, the regularity/irregularity of capillary network and the shape and size of the gastric pits and crypts in relation to the Helicobacter pylori infection, intestinal metaplasia, and atrophic gastritis, etc.

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AUTHORS' CONTRIBUTIONS

Admir Kurtcehajic,Tomislav Bokun and Ervin Alibegovic provided the basic idea, and designed, edited and wrote the core of the manuscript. Enver Zeremreviewed the manuscript and provided intellectual input and academic writing. Suad Kunosicanalysed data and performed the statistical analysis. Ahmed Hujdurovic, Amir Tursunovic and Kenana Ljucawroteextended version of the manuscript and reviewed the literature data; all authors read and approved the final version of the manuscript.

REFERENCES

1. **Kim JW**. Usefulness of Narrow-Band Imaging in Endoscopic Submucosal Dissection of the
Stomach. Clin Endosc 2018; 51:527-533 [PMID: 30453446DOI: 10.5946/ce.2018.186]

2. **Yao K**. Clinical Application of Magnifying Endoscopy with Narrow-Band Imaging in the Stomach. Clin Endosc 2015; 48:481-490 [PMID: 26668793 DOI: 10.5946/ce.2015.48.6.481]

3. **Li HY**, Ge ZZ, Fujishiro M, Li XB. Current clinical applications of magnifying endoscopy with narrow band imaging in the stomach. Diagn Ther Endosc 2012; 2012:271914[PMID: 23024577 DOI: 10.1155/2012/271914]

4. **Qi Q**, Guo C, Ji R, Li Z, Zuo X, Li Y. Diagnostic Performance of Magnifying Endoscopy for Helicobacter pylori Infection: A Meta-Analysis. PLoS One 2016; 11:e0168201[PMID: 27992489DOI: 10.1371/journal.pone.0168201]

5. **Glover B**, Teare J, Patel N. A systematic review of the role of non-magnified endoscopy for the assessment of H. pylori infection. Endosc Int Open 2020; 8:E105-E114 [PMID: 32010741 DOI: 10.1055/a-0999-5252]

6. **Ji R**, Li YQ. Diagnosing Helicobacter pylori infection in vivo by novel endoscopic techniques.World J Gastroenterol 2014;20:9314-9320[PMID:25071325 DOI: 10.3748/wjg.v20.i28.9314]

7. **Okubo M**, Tahara T, Shibata T, Nakamura M, Kamiya Y, Yoshioka D, Maeda Y, Yonemura J, Ishizuka T, Arisawa T, Hirata I. Usefulness of magnifying narrow-band imaging endoscopy in the Helicobacter pylori-related chronic gastritis. Digestion 2011; 83:161-166 [PMID: 21266810 DOI: 10.1159/000321799]

8. **Jang JY**. The Past, Present, and Future of Image-Enhanced Endoscopy. Clin Endosc 2015; 48:466-475 [PMID: 26668791 DOI: 10.5946/ce.2015.48.6.466]

9. **Shibagaki K**, Amano Y, Ishimura N, Taniguchi H, Fujita H, Adachi S, Kakehi E, Fujita R, Kobayashi K, Kinoshita Y. Diagnostic accuracy of magnification endoscopy with acetic acid enhancement and narrow-band imaging in gastric mucosal neoplasms.
Endoscopy 2016; 48:16-25 [PMID: 26158242 DOI: 10.1055/s-0034-1392542]

10. **Kadowaki S**, Tanaka K, Toyoda H, Kosaka R, Imoto I, Hamada Y, Katsurahara M, Inoue H, Aoki M, Noda T, Yamada T, Takei Y, Katayama N. Ease of early gastric cancer demarcation recognition: a comparison of four magnifying endoscopy methods. J Gastroenterol Hepatol 2009; 24:1625-1630 [PMID: 19788603 DOI: 10.1111/j.1440-1746.2009.05918.x]

11. **Sha J**, Wang P, Zhu B, Zhu M, Li X, Gao F. Acetic Acid Enhanced Narrow Band Imaging for the Diagnosis of Gastric Intestinal Metaplasia. PLoS One 2017; 12:e0170957[PMID: 28135297 DOI: 10.1371/journal.pone.0170957]

12. **Singh R**, Shahzad MA, Tam W, Goda K, Yu LH, Fujishiro M, Uedo N, Ruszkiewicz A. Preliminary feasibility study using a novel narrow-band imaging system with dual focus magnification capability in Barrett's esophagus: is the time ripe to abandon random biopsies? Dig Endosc 2013; 25 Suppl 2:151-156 [PMID: 23617668 DOI: 10.1111/den.12106]

13. **Goda K**, Dobashi A, Yoshimura N, Aihara H, Kato M, Sumiyama K, Toyoizumi H, Kato T, Saijo H, Ikegami M, Tajiri H. Dual-focus versus conventional magnification endoscopy for the diagnosis of superficial squamous neoplasms in the pharynx and esophagus: a randomized trial. Endoscopy 2016; 48:321-329 [PMID: 26878247 DOI: 10.1055/s-0035-1569644]

14. **Gulati S**, Emmanuel A, Ong M, Pavlidis P, Patel M, El-Menabawey T, Vackova Z, Dubois P, Murino A, Martinek J, Sethi A, Neumann H, Haji A, Hayee B. Near-focus narrow-band imaging classification of villous atrophy in suspected celiac disease: development and international validation. GastrointestEndosc 2021;94:1071-1081[PMID: 34228981 DOI: 10.1016/j.gie.2021.06.031]

15. **Bond A**, Burkitt MD, Cox T, Smart HL, Probert C, Haslam N, Sarkar S. Dual-focus Magnification, High-Definition Endoscopy Improves Pathology Detection in Direct-to-Test Diagnostic Upper Gastrointestinal Endoscopy. J Gastrointestin Liver Dis 2017; 26:19-24 [PMID: 28338109 DOI: 10.15403/jgld.2014.1121.261.gen]

16. **Kim JW**, Jung Y, Jang JY, Kim GH, Bang BW, Park JC, Choi HS, Cho JH; Research Group for Endoscopic Instruments and Stents of Korean Society of Gastrointestinal Endoscopy. Narrowband imaging with near-focus magnification for discriminating the gastric tumor margin before endoscopic resection: A prospective randomized multicenter trial. J Gastroenterol Hepatol 2020; 35:1930-1937 [PMID: 32433790 DOI: 10.1111/jgh.15109]

17. **Kakushima N**, Yoshida N, Doyama H, Yano T, Horimatsu T, Uedo N, Yamamoto Y, Kanzaki H, Hori S, Yao K, Oda I, Tanabe S, Yokoi C, Ohata K, Yoshimura K, Ishikawa H, Muto M. Near-focus magnification and second-generation narrow-band imaging for early gastric cancer in a randomized trial. J Gastroenterol 2020; 55:1127-1137 [PMID: 33021688 DOI: 10.1007/s00535-020-01734-3]

18. **Na HK**, Choi KD, Park YS, Kim HJ, Ahn JY, Lee JH, Jung KW, Kim DH, Song HJ, Lee GH, Jung HY. Endoscopic scoring system for gastric atrophy and intestinal metaplasia: correlation with OLGA and OLGIM staging: a single-center prospective pilot study in Korea.

Scand J Gastroenterol 2022; 57:1097-1104 [PMID: 35387540 DOI: 10.1080/00365521.2022.2055974]

19.**Fiuza F**, Maluf-Filho F, Ide E, Furuya CK Jr, Fylyk SN, Ruas JN, Stabach L, Araujo GA, Matuguma SE, Uemura RS, Sakai CM, Yamazaki K, Ueda SS, Sakai P, Martins BC. Association between mucosal surface pattern under near focus technology and Helicobacter pylori infection. World J GastrointestEndosc 2021; 13:518-528 [PMID: 34733412 DOI: 10.4253/wjge.v13.i10.518]

20. **McHugh ML**. Interrater reliability: the kappa statistic.
Biochem Med (Zagreb) 2012; 22:276-282 [PMID: 23092060]

21. **Garcés-Durán R**, García-Rodríguez A, Córdova H, Cuatrecasas M, Ginès À, González-Suárez B, Araujo I, Llach J, Fernández-Esparrach G. Association between a regular arrangement of collecting venules and absence of Helicobacter pylori infection in a European population. GastrointestEndosc 2019; 90:461-466 [PMID: 31108089 DOI: 10.1016/j.gie.2019.05.027]

22. **Yagi K**, Nakamura A, Sekine A. Comparison between magnifying endoscopy and histological, culture and urease test findings from the gastric mucosa of the corpus. Endoscopy 2002; 34:376-381 [PMID: 11972268 DOI: 10.1055/s-2002-25281]

23. **Anagnostopoulos GK**, Yao K, Kaye P, Fogden E, Fortun P, Shonde A, Foley S, Sunil S, Atherton JJ, Hawkey C, Ragunath K. High-resolution magnification endoscopy can reliably identify normal gastric mucosa, Helicobacter pylori-associated gastritis, and gastric atrophy. Endoscopy 2007; 39:202-207 [PMID: 17273960 DOI: 10.1055/s-2006-945056]

24. **Tahara T**, Shibata T, Nakamura M, Yoshioka D, Okubo M, Arisawa T, Hirata I. Gastric mucosal pattern by using magnifying narrow-band imaging endoscopy clearly distinguishes histological and serological severity of chronic gastritis. GastrointestEndosc 2009; 70:246-253 [PMID: 19386303 DOI: 10.1016/j.gie.2008.11.046]

25. **Cho JH**, Jeon SR, Jin SY, Park S. Standard vs magnifying narrow-band imaging endoscopy for diagnosis of Helicobacter pylori infection and gastric precancerous conditions. World J Gastroenterol 2021; 27:2238-2250 [PMID: 34025076 DOI: 10.3748/wjg.v27.i18.2238]

26. **Kawamura M**, Sekine H, Abe S, Shibuya D, Kato K, Masuda T. Clinical significance of white gastric crypt openings observed via magnifying endoscopy. World J Gastroenterol 2013; 19:9392-9398 [PMID: 24409067 DOI: 10.3748/wjg.v19.i48.9392]

Footnotes

Institutional review board statement: The study was reviewed and approved by the Ethical Committee of Blue Medical Group, 75000 Tuzla, Bosnia and Herzegovina.

Informed consent statement: All study participants, or their legal guardian, provided informed written consent prior to study enrolment.

Conflict-of-interest statement: There are no conflicts of interest to report.

Data sharing statement: No additional data are available.

STROBE statement: The authors have read the STROBE Statement-checklist of items, and the manuscript was prepared and revised according to the STROBE Statement-checklist of items.

Figures legends

Figure 1. White light endoscopy (WLE) from the point of view of the first observer. A image (WLE-1a pattern) and B image (WLE-2a pattern): clear appearance of the regular arrangement of collecting venules (RAC) - white arrow. C image (WLE-b pattern): less clear appearance of the RAC - white arrow. D image (WLE-c pattern): absence of the RAC.

Figure 2. Near focus (NF) mode from the point of view of the first observer. A image (NF-a pattern): clear appearance of the regular arrangement of collecting venules (RAC) - white arrow, subepithelial capillary network (SECN) - black arrow and gastric pits (GP) - blue square. B image (NF-b pattern): less clear appearance of the RAC - white arrow, clear appearance of the SECN - black arrow and GP - blue square. Cimage (NF-c pattern): clear appearance of the SECN - black arrow and GP - blue square.

Figure 3. Near focus (NF), narrow-band imaging (NBI) mode from the point of view of the first observer. A image (NF-NBI-1a pattern) and B image (NF-NBI-2a pattern): clear appearance of the regular arrangement of collecting venules (RAC) - white arrow, subepithelial capillary network (SECN) - black arrow, gastric pits (GP) - blue square and crypt opening (CO) - green arrow, right upper corner. C image (NF-NBI-b pattern): clear appearance of the SECN - black arrow and GP - blue square. Less clear appearance of the CO - green arrow, right upper corner.

Figure 4. Acetic acid (AA), near focus (NF) and AA-NF, narrow-band imaging (NBI) mode from the point of view of the first observer. A1 image (AA-NF pattern) was scored at 0.5, B1 image (AA-NF-NBI pattern) was scored at 1. A2 image (AA-NF pattern) was scored at 1, B2 image (AA-NF-NBI pattern) was scored at 1. A3 (AA-NF pattern) was scored at 0, B3 image (AA-NF-NBI pattern) was scored at 0.5.

Tables

Table 1*– Frequency and scoring of the endoscopic patterns on the white light endoscopy (WLE), near focus (NF) and NF, narrow-band imaging (NBI) mode.*

Table 2*– Frequency and scoring of the endoscopic patterns on the acetic acid (AA), near focus (NF) and AA-NF, narrow-band imaging mode (NBI) mode.*