Some Features of Intestinal Metaplasia in Children

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Abstract

Background and aims: Intestinal metaplasia(IM) of stomach mucosa in children is rare problem. Aim of the study is to apply the criteria used in adults for diagnostic and control of IM in pediatric cohort.

Patients and methods: From 2011 to 2015 endoscopy with NBI was performed on 1860 children. Endoscopic studies were performed using high-definition (HD) resolution equipment with electronic magnification and/or standard-definition (SD) resolution with optical magnification using Exera II and III by Olympus (Tokyo, Japan) systems. Narrow band imaging (NBI) was used for illumination. Two groups were founded. The main group included 24 children with morphologically verify intestinal metaplasia (IM) and 65 children were included in the control group. Patients in the main group have precision biopsy from IM area. The main and the control group also underwent multiple non-targeted biopsies in accordance with OLGA criteria. H. pylori was defined in two ways - morphologically and C13 breath test. Evaluation of pepsinogens I and II, gastrin-17 levels in blood was performed in group with IM also.

Results: The sensitivity and specificity of high-resolution endoscopy with NBI for the detection of intestinal metaplasia (IM) foci were 100% (85.18-100.00) and 98.59% (92.40-99.96), respectively. "Light blue crests" – was leading endoscopic symptom of IM. In 23 of 24 cases, the zone of IM was localised in the pre-pyloric zone, with trend to stomach angle. In the group with IM, *H.pylori* were detected in 2 cases (8.3%), and in the control group, in 35 cases (53.8%). The relative risk of detection of *H. pylori* in the presence of IM was 0.0774 (0.0112-0.5341), and the Odds ratio was 0.0373 (0.0047-0.2926). The pepsinogen I and pepsinogen II levels were significantly higher in the control group (P<0.01). The ratio of the pepsinogen I and pepsinogen II levels was reliably different in these groups. There were no reliable differences in the gastrin 17 levels.

Conclusions: Endoscopy with NBI for detecting of intestinal metaplasia is a universal diagnosis tool for this type of pathology. The incidence of intestinal metaplasia in the main group was 1.29%. The visualised foci of intestinal metaplasia in children are mainly associated with the pylorus and have a vector of propagation towards the lesser gastric curvature. The probability of identifying the association of *Helicobacter pylori* in children with intestinal metaplasia is very low.

Keywords: Intestinal Metaplasia; High-Resolution Endoscopy with NBI Illumination; Contamination of Gastric Mucosa With *H. Pylori*; Diagnostics; Children;

Introduction

Metaplasia of the stomach is a pathological process of replacing the cells of the gastric epithelium with intestinal type cells under various undesirable factors. The dynamics of this process can lead to dysplasia with further regeneration into cancer, and therefore intestinal metaplasia of the gastric mucosa (IMGM) is considered a precancerous mucosal change in adults [1]. Thus, it is important to note that intestinal metaplasia of the stomach is a benign stage of such successive changes. The process can be reversible with a timely response, the elimination of harmful factors, and treatment.

The early detection and diagnoses of IMGM is of particular importance for regions with a high prevalence of stomach cancer. In the Republic of Belarus, stomach cancer ranks No. 4 among men and No. 6 among women in the structure of adult oncological morbidity. In 2012, the Stomach Cancer Incidence Rates was more than 23.8 per 100,000 men of the population, in our country [2]. Stomach cancer was the third leading cause of mortality from cancer. There is a high incidence of stomach cancer in people under 30 years of age. [3].

The detection of metaplasia in the earliest age and in the initial stage a priori ensure high effectiveness of medical care and minimises further irreversible changes. Concomitantly, there are only a few publications on the prevalence of IMGM in children,

which affects 1.1-1.9 per 100,000 children according to different data. These data are either a morphological finding or combined data with structural macroscopic changes, such as the probable presence of an aberrant pancreas [4].

According to the Kyoto agreement, the most informative tool for detecting IMGM is high-resolution endoscopy with a maximum magnification and an illumination in narrow band imaging (NBI)*; the combination of these parameters provides the high sensitivity and specificity of this investigation method [5]. Japanese authors are pioneers in the use of such endoscopy

to identify IMGM foci [6]. This diagnostic method was also recommended by the MAPS agreement in 2011 [7].

*Narrow band imaging (NBI) is used in endoscopy and includes two components: 415 nm wavelengths reflected from mucosal surface structures and 540 nm wavelengths absorbed by haemoglobin-containing structures [8] (Figure 1 A and B).

Thus, the presence of IM in the gastric mucosa of children means a long exposure to pathological changes over the course of life and requires constant dynamic control considering the risk of oncogenesis in the context of such pathology in adults.
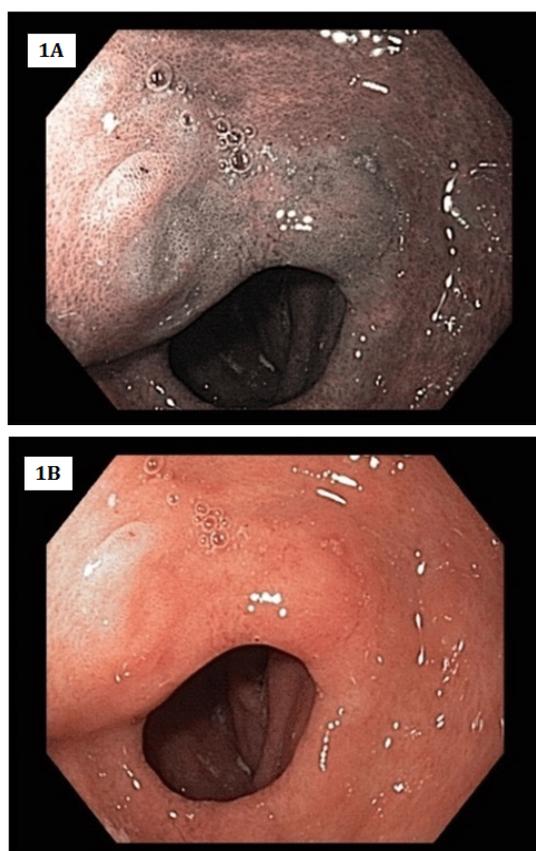

Figure 1: A: Visualisation of the intestinal metaplasia site conjugated to the pylorus using NBI.
B: Visualisation of the intestinal metaplasia site conjugated to the pylorus using white light illumination.

Objectives and Purposes

The objectives of this study were:

1. To reveal the sites of intestinal metaplasia in the gastric mucosa in children using HD endoscopy

2. To determine the topography of metaplasia sites in the gastric mucosa and detail the macro- and microscopic characteristics of these sites

3. To determine the features of the contamination of the gastric mucous membrane with *H. pylori* in children with and without

intestinal metaplasia in combination with an evaluation of pepsinogens I and II and gastrin-17 levels in the blood

Materials and Method

From 2011 to 2015, at Republican scientific and practical center «Mother and child», video esophagogastroduodenoscopy was performed on 1860 children using HD endoscopy with electronic magnification and an SD resolution with an optical magnification. In all cases, NBI was used for illumination.

This study had a case-controlled design.

Two groups were identified: the main group and the control group. The main group included 24 children and the control group included 65 children.

Children with endoscopically confirmed and morphologically verified IM in the gastric mucosa were included in the main group. The control group included random children with clinic signs of dyspepsia, but without endoscopic and morphological signs of intestinal metaplasia in gastric mucosa.

Two methods were used to include children in the study group:

The first method: if there were signs of the possible presence of IM in a primary endoscopy using an HD endoscope and NBI for illumination, a detailed study of the detected areas was performed using electronic magnification. Then the targeted biopsy of sites with microcharacteristics of IM for the morphological verification and the non-targeted biopsy using the Operative Link on Gastritis Assessment system (OLGA) was performed [9]. If there was a positive morphological outcome, the child was included in the study group.

The second method: if the child had a history of endoscopy with white light as an illumination, a non-targeted biopsy was performed and morphological signs of intestinal metaplasia were obtained; in this case, repeated endoscopy with an optical magnification and NBI was performed to clarify the topography of the focus and its microarchitectonics, and multiple targeted and non-targeted biopsies were performed. If there was IM morphological verification, the child was included in the study group.

The criteria for inclusion in the control group were:

1. The presence of clinical signs of dyspepsia (the evaluation of the manifestation degree was carried out in accordance with the Rome Criteria III Revision, classified as group H [10]).

2. The absence of intestinal metaplasia foci in a high-resolution or magnification endoscopy with NBI for illumination.

3. The absence of IM in the morphological description of the biopsy material Patients in the control group also underwent multiple non-targeted biopsies in accordance with OLGA criteria.

Statistical processing of the results was carried out using computer programmes. The analysis of consistency of signs distribution type to the normal distribution law was carried out using the Shapiro-Wilk test; the sign distribution was considered a departure from normality at $P<0.05$. Depending on the consistency/inconsistency of the distribution of the analysed signs to the normal distribution law, the parametric Student's t-test and the nonparametric Mann-Whitney U-test were used to evaluate differences between the groups.

Measures of central tendency and data dispersion were calculated using the methods of descriptive statistics; depending on the type of distribution, the quantitative parameters are presented as mean (M) and 95% confidence interval(95% CI) or as median (Me) and interquartile range (25% and 75% quartiles). $P<0.01$ was accepted as statistically significant in the testing of statistical hypotheses.

To evaluate the reliability of the endoscopic signs of mucosal changes and to select patients and separate them into groups, the χ2 (chi square) test, Fisher's test (φ), and Fisher's z-test were used.

The presence/absence of correlation between the degree of intestinal metaplasia and the focus area was evaluated using the correlation-regression analysis.

Endoscopy

An endoscopic study was performed using equipment by Olympus (Tokyo, Japan) using the Exera II and III systems and the video endoscopes GIF-HQ190, GIF-H180, and GIF-Q160Z. NBI was used for illumination.

In the search for focus mode, the mucosa of the digestive tract was examined at a distance of 2-3 cm with NBI illumination. When background light blue or blue staining was detected, the site was studied in search for focus mode according to the following signs:

1. Microarchitectonics

2. The presence of light blue crests (LBC) [11]

3. The presence of microfication (the dimensions of the microstructure elements of the mucosa in the site under study were two times less than that of the surrounding elements)

4. Heterogeneity (the microstructure elements are heterogeneous)

5. Enlargement (enlargement of the microstructure elements of the mucosa)

The search for the focus level was referred to as category A, and signs of focus characteristics were referred to as category B. If an endoscopic sign of category A and/or A and B was detected at the study site, a conclusion was made about the possible focus relationship to the intestinal metaplasia.

Morphological evaluation

The description of the biopsy material and the classification of chronic gastritis (the degree of activity, inflammation, and atrophy [metaplasia]) were performed in accordance with the Houston-Sydney system. Additionally, the severity and prevalence of metaplasia (atrophy) was evaluated using the OLGA gradation system [9].

The diagnosis of IM in a histological material stained with haematoxylin and eosin was carried out on the basis of generally accepted signs; (complete) small intestinal metaplasia was characterised by the presence of phenotypically small intestinal epithelium enterocytes with a well-defined brush border of goblet cells and Paneth cells. In incomplete intestinal metaplasia, there was a similarity to cells in the epithelium of the large intestine containing numerous mucin drops of various sizes and shapes in the cytoplasm, with no brush border [11] (Figure 2).
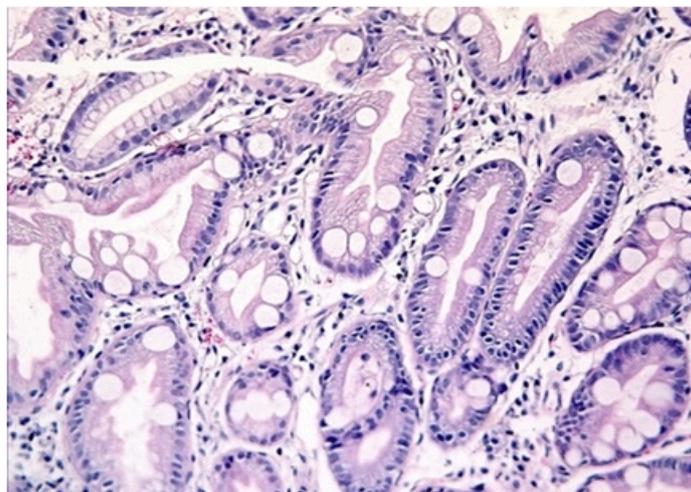
Figure 2: Small intestinal metaplasia of the surface epithelium and gland necks of the stomach antral part of moderate degree. The sample was stained with haematoxylin and eosin. Magnification: 250 times

Breath test

C13 breath test was performed in both groups of children with carbon 13-labeled urea to determine the presence of H. pylori. The infrared analyser FANci2 (Fischer Analysen Instrumente GmbH, Leipzig, Germany) was used to measure the pepsinogen I and II and gastrin-17 levels, and *H. pylori* antibodies were assessed using an assay kit (DRG international inc., NJ, USA).

Results

The study group (24 children) had an equal number of boys and girls (12), and the average age was 14.5 ± 2.06 years old. The mean age in the control group (65 children) was 12.75 ± 3.39, and girls (35) also predominated in this group (53.8%). No reliable difference in age and gender was found.

Localisation and area of intestinal metaplasia foci

In 23 of 24 cases, the zone of intestinal metaplasia was localised in the pre-pyloric zone and had a connection with the pylorus. In two cases, additional foci of intestinal metaplasia were found localised in the antral part. In one case, visually typical intestinal metaplasia was detected on the site of the aberrant pancreas in the antral part.

The zones of detected intestinal metaplasia belonged to the pyloric, but differed in the direction of distribution and area. To evaluate the area and the direction of the pylorus associated with the focus distribution, the following system was used: a virtual coordinate grid with the origin of the coordinates in the centre of the pylorus (point 0) was superimposed onto the image of the pre-pyloric zone made using NBI illumination, and the width of the open biopsy forceps was used as a unit measure. An 8 mm biopsy forceps was used in all of the studies. Eight vectors originating from the zero point were used. Four main vectors were arranged at right angles to each other as follows: vector A, from the starting point to the middle of the lesser gastric curvature; vector B, the back wall; vector C, the major curvature; vector D, the front wall; and the four additional vectors, AB, BC, CD, and DA. Each vector was graded.

Comparing the results obtained by the coordinate grid vectors, we detected reliable differences in the value of vector A compared to the other vectors AB, B, BC, C, CD, and D ($P<0.01$), and in the value of vector AD compared to B, BC, CD, and D ($P<0.01$). The data are presented in Table 1. No reliable differences were found in the other ratios of the vector values (Table 1).

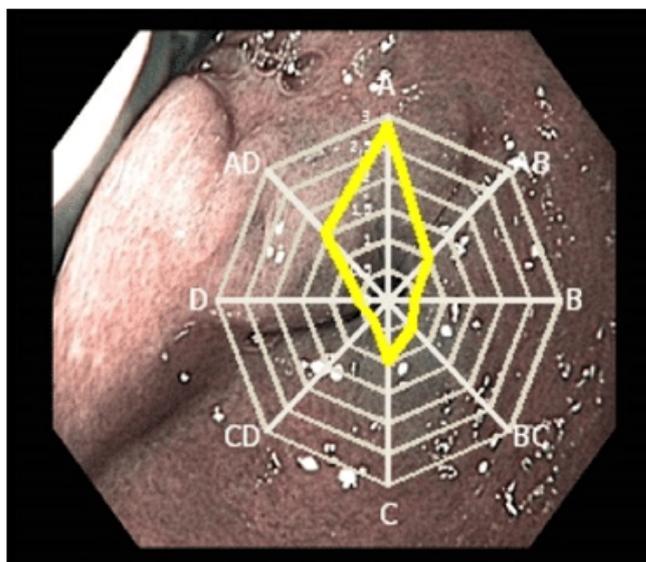| Table 1: Determination of reliable difference using Mann-Whitney U-test by coordinate grid vectors							
	AB	**B**	**BC**	**C**	**CD**	**D**	**AD**
A	P<0.001	P<0.0001	P<0.0001	P<0.001	P<0.0001	P<0.0001	P=0.0566
	AB	P<0.01	P=0.041	P=0.692	P=0.0157	P=0.0297	P=0.0270
	B		P=0.445	P=0.062	P=0.5575	P=0.4831	P<0.001
	BC			P=0.188	P=0.8162	P=0.9276	P<0.001
	C				P=0.1076	P=0.1608	P=0.0259
	CD					P=0.8874	P<0.0001
	D						P=0.0001

Figure 3: The intestinal metaplasia zone with the superimposition of the virtual system for the determination of its area and the diagram of distribution of intestinal metaplasia in the main group

Note: Considering the number of patients in the groups (more than 20) and a degree of freedom of 58, a z-test with the determination of critical values by Student t-test and Mann-Whitney U-test was used (Figure 3).

The correlation-regression analysis showed an absence of correlation between the IM degree and the focus area. However, the regression component of the analysis demonstrated the effect of the focus area, as an independent variable, on the intestinal metaplasia degree in this site, expressed as a percentage of metaplastic glands, as a dependent:

$\log(y)=1.3703+0.1994 \log(x)$, 1.3703 $(1.1097-1.6308)$, $t=11.3632$, $P<0.01$.

Endoscopic signs of intestinal metaplasia

The following endoscopic signs were used as the detection criteria (characteristics) of the intestinal metaplasia focus (Table 2) [12]:

Using the approach described above, foci with changes in colour characteristics of the mucosa were detected in 24 cases and morphologically verified for intestinal metaplasia in 23 cases.

The sensitivity of the high-resolution (magnification) endoscopy with NBI illumination in the detection of IM foci was 100% (85.18-100.00%) and the specificity was 98.59% (92.40-99.96%).

The analysis of the endoscopic signs for changes in the mucosa in the patients in the study group showed the adequacy of the applied method of IM diagnostics ($\chi2=68.963$ and $\varphi=0.972$, $P<0.01$). The Odds ratio (OR) for the detection of the intestinal metaplasia focus using these criteria was 1711.0 ($P<0.01$).

Table 2: Endoscopic signs of intestinal metaplasia sites

A: Search level signs B: Search level signs	Signs	Detected cases-patients ratio	Incidence of signs in patients (%)
A	Specific staining (presence of background lightblue or blue staining)	24/24	100
B	Presence of LBC	23/24	95.8
B	Microficationof elements	3/24	12.5
B	Heterogeneityof elements	4/24	16.67
B	Enlargementof elements (B)	14/24	58.3
B	Lack of structure(B)	0/30	0
B	Presence of pathological microvessels	0/30	0

Evaluation of pepsinogens I and II, gastrin-17 levels in blood, and contamination of gastric mucosa with H. pylori

The obtained results to determine the pepsinogen I and II levels, their ratio, as well as the gastrin 17 level in the study group and in the control group are presented in Table 3.

The pepsinogen I and pepsinogen II levels were significantly higher in the control group (P<0.01). The ratio of the pepsinogen I and pepsinogen II levels was reliably different in these groups. There were no reliable differences in the gastrin 17 levels.

H. pylori bacteria were detected using two methods, by the morphology and the urease breath test with C13 carbon radioisotope. To identify and evaluate the correlation ratio between the two above diagnostic methods, the Spearman rank correlation coefficient was used, which in the intestinal metaplasia group was 0.692 (0.400-0.856) and in the control group was 0.85 (0.765-0.906), P<0.01, which revealed the presence of a direct link between the two methods of diagnosis of H. pylori. In the main group, H. pylori bacteria were detected in 2 cases (8.3%), and in the control group, in 35 cases (53.8%). The relative risk of detection of H. pylori in the presence of IM was 0.0774 (0.0112-0.5341), and the Odds ratio was 0.0373 (0.0047-0.2926).

Table 3: Mean values of gastrin 17, pepsinogen I and II levels and their ratios in the study and control groups

Group	Pepsinogen I (mg/L) CI 95%	Pepsinogen II (mg/L) CI 95%	Pep I/Pep IICI 95%	Gastrin 17 (mmol/L) CI 95%
Main (IM)	38.21 (31.34-45.07)	4.68 (3.7-5.66)	9.23 (7.16-11.29)	408.5 (330.25-486.74)
Control (no IM)	51.2 (41.58-60.91)	8.97 (7.0-10.94)	6.30 (5.63-6.97)	476.48 (393.29-559.68)

Discussion

Considering intestinal metaplasia in the context of the Correa cascade, we assumed a reliable link between the presence of H. pylori in the gastric mucosa and the detection of intestinal metaplasia foci. The literature analysis on this topic showed that there are publications confirming this assumption, and those in which it is not established [13-17].

Our results also do not confirm the link of contamination of the gastric mucosa with H. pylori and the presence of metaplastic changes in children. In other words, in the children of the examined cohort living in the Republic of Belarus, H. pylori does not affect the development of intestinal metaplasia in the gastric mucosa. The same result was published from Bosnia and Herzegovina [20].

The incidence of intestinal metaplasia in children is open for discussion. Some publications report different numbers depending on the patients screened, ranging from 1.0% to 3.07% [4,18]. In our work, the screened group involved 1860 children. All had signs of dyspepsia. The incidence of intestinal metaplasia with respect to this group was 1.29%.

The high sensitivity of the method to detect intestinal metaplasia foci during high-resolution endoscopy and using NBI for the illumination of the mucosa is a proven fact [18]. This methodology makes it possible to perform a targeted biopsy with an error of only 0.5-1.0 mm.

The topography of the intestinal metaplasia foci is the subject of active study as an assumed factor determining the potential of the intestinal metaplasia focus [19]. The use of high-resolution endoscopy enables the clarification of the focus location within the stomach surface and provides more accurate information about its topography compared to morphologically oriented systems.

The presence of areas of intestinal metaplasia "conjugated" to the pylorus is noted also in adults. It is believed that these sites do not carry the maximum oncogenic potential. It can be assumed that there is some non-standard factor triggering the development of the intestinal metaplasia in relation to the zone topography. Concomitantly, the hypothesis about the effect of the lesion area on the percentage of the metaplastic glands in the targeted biopsy is confirmed.

The endoscopic characteristics of the detected intestinal metaplasia foci proposed for the characteristics of mucosal lesions in adults [7] are reproducible in children. In the search for focus mode, the most informative characteristic is the change in the colour characteristics of the mucosa towards light blue or blue segments of the spectrum. In the focus analysis mode, the most informative factor is the presence of LBC (which possibly determines the colour change in the remote examination of focus), the enlargement of focus structural elements, and their heterogeneity.

Conclusions

1. The detected foci of intestinal metaplasia in children are mainly associated with the pylorus and have a vector of propagation towards lesser gastric curvature.

2. The probability of detecting Helicobacter pylori in children with intestinal metaplasia is very low.

3. It is not possible to use criteria for detecting atrophic gastritis by pepsinogen I and pepsinogen II levels and their ratio in the children of the studied group.

4. The endoscopic criteria of the intestinal metaplasia foci detected in children using high-resolution (magnification) endoscopy with NBI illumination are highly sensitive and specific.

Summarised conclusion. The diagnosis of intestinal metaplasia in gastric mucosa may have adverse clinical outcomes, but in children, the conclusion about the risk of developing possible stomach cancer should be made with caution.

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