Non-endoscopic diagnosis of multifocal atrophic gastritis ; efficacy of serum gastrin-17, pepsinogens and *Helicobacter pylori* antibodies

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Abstract

Background/Aims : **Infection with *H.pylori* is an important risk factor for the development of gastric cancer and glandular atrophy is an intermediate stage in gastric carcinogenesis. While screening the patients with atrophic gastritis by endoscopy is unrealistic, a concept of "serological gastric biopsy" based on measurement of gastric secretory proteins and peptides should be further validated. We sought to determine if the laboratory panel composed of serum PGI and protein stimulated gastrin-17 might select patients with MAG, and what is diagnostic significance of *H.pylori* serology in population of high prevalence of *H.pylori* infection.**

Material and methods : **55 consecutive patients of both sexes (M/F 25/30 ; range of age 55 -81 years) were referred for gastroscopy with antrum and corpus mucosal biopsies. Patients with histological signs of glandular atrophy at any site of the stomach were considered to have multifocal atrophic gastritis. A first blood sample was collected for measurement of basal gastrin-17, pepsinogens and *H.pylori* IgG-antibodies, and second was taken 20 minutes after use of protein-rich drink to measure stimulated gastrin-17.**

Results : **Signs of mucosal atrophy were found in 19 patients, while 29 patients showed non-atrophic gastritis and seven *H.pylori*-negative patients had no histological pathology. Low serum level of stimulated gastrin-17 (< 5 pmol/l) and/or pepsinogen I (< 50 µg/l), were found in 16 of 19 patients (84,2%) with and in 7 of 36 patients (19,4%) without atrophy in the histological study. Combining of *H.pylori* serology with serum levels of secretory peptides had no significant effect on diagnostic sensitivity of the test panel.**

Conclusion : **The test panel composed of pepsinogen I and protein stimulated gastrin-17 may be used as the "serological gastric biopsy" detecting multifocal atrophic gastritis. The diagnostic sensitivity of this test panel is not increased by knowledge of *H.pylori* status.** (Acta gastroenterol. belg., **2004**, 67, **320-326**).

Key words : atrophic gastritis ; Gastrin-17 ; Pepsinogen I.

Introduction

Gastric carcinoma remains a major cause of morbidity and mortality worldwide despite its significant decline in recent years. It has been shown that infection with *H.pylori* is an important risk factor for the development of gastric cancer. Glandular atrophy and intestinal metaplasia, which are sequelae of long-term *H.pylori* infection are intermediate stages in gastric carcinogenesis (1,2). As prospective studies have shown that atrophic gastritis increases a risk for subsequent development of gastric cancer by at least three- to six-fold (3-6), an appropriate screening and preventive measures should be considered in patients with this condition.

H.pylori infection begins with non-atrophic gastritis, leading after several decades to loss of the glandular structures and fibrosis of the mucosa in more than half of the affected subjects (3,7). This process affects both the antrum and corpus of the stomach and has a multifocal aspect. It has been demonstrated that multifocal atrophic gastritis (MAG) and intestinal metaplasia improves and may even heal after eradication of *H.pylori.* This is especially true in early stages of mucosal atrophy (6,8-11).

According to the Maastricht 2000 - consensus the patients having mucosal atrophy with or without intestinal metaplasia should be treated with anti-*H.pylori* drugs if the presence of the bacterium can be evidenced. It seems, however, that early stages of mucosal atrophy cannot be delineated, as this condition is not associated with specific laboratory or clinical symptoms. On the other hand, gastroscopy with tissue sampling cannot be proposed to all patients infected with *H.pylori*, especially in countries where the infection rate is high. Non-invasive screening methods based on measurement of pepsinogens and gastrin in the serum have been tested for a long time. It is well known that a major loss of chief cells in the gastric corpus is associated with a decreases in both the serum pepsinogen I (PGI) and the ratio of serum PGI to PGII (12-16). The interest in non-invasive diagnosis of MAG has renewed, when it had been shown that moderate and severe mucosal atrophy in the antrum is accompanied by decreased serum levels of gastrin-17 (G17). Several studies have demonstrated that the histological condition of gastric mucosa and topography of atrophic gastritis may be evaluated by the laboratory panel (*Biohit* panel), based on combined measurement of G17, PGI and *H.pylori* antibodies (17-19). The aim of our study was to investigate the usefulness of the *Biohit* panel in detection of early stages of MAG in a country with a considerably higher incidence rate of both the *H.pylori* infection and gastric cancer, in comparison with populations on which the Biohit panel has been validated (20,21).

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Material and Methods

Subjects

All outpatients aged 55 years or older who required diagnostic upper GI endoscopy because of dyspeptic or refluxive symptoms were eligible for the study. By these criteria 55 consecutive patients of both sexes (25 men and 30 women ; age range 55-81 years, mean age 65 years) were included. This prospective study was performed over a 5-month period (February 2002 and June 2002). Exclusion criteria were treatment with antibiotics or bismuth containing drugs in the preceding 2 months, active gastrointestinal bleeding, previous gastric surgery, confirmed immune-mediated gastric atrophy of the corpus in the context of pernicious anemia, alcoholism or chronic administration of nonsteroidal anti-inflammatory drugs (excluding low-dose aspirin). The patients taking proton pump inhibitors were asked to stop the treatment one week before the study, and if they could not do without inhibition of gastric acid secretion they were switched to H2-blocker (they were told to stop the drug one day before the study). Relevant demographic and clinical data were obtained in each patient.

The study was approved by Ethics Committee of Silesian Medical University and all subjects gave informed consent.

Serological assays

The basal blood samples for assays of pepsinogens, PGI and PGII, gastrin-17 (G17) and IgG antibodies to *H.pylori* (IgG-*H.pylori*) were collected after an overnight fast. The samples for stimulated gastrin-17 (s-G17) were taken 20 minutes after drinking a protein meal (10 grams of chocolate flavored soy protein powder, Cat. No. 610 099, Biohit Plc, Finland). The blood was placed in chilled tubes and centrifuged at speed of 1000-1500xg. The serum was stored at -70°C until use.

The PGI, PGII and G17 serum levels were determined with EIA methods using monoclonal antibodies to PGI (Cat. No. 601 010, Biohit Plc, Finland), to PGII (Cat. No. 601 020, Biohit Plc, Finland) and amidated G17 (Cat. No 601 030, Biohit Plc, Finland) according to the instructions of the manufacturer adapted for automated ELISA systems. The G17 antibody detected amidated gastrin-17 only, but no other gastrin molecules or fragments (*e.g.* glycine extended gastrin-17, gastrin-34, gastrin-13 and CCK-(26-33). The sensitivity of the PGI test was 0,7 µg/l, of the PGII assay 1,0 µg/l and of the G17 test 2,2 pmol/l, enabling baseline measurements to be taken. IgG class antibodies to *H.pylori* were determined using specific EIA tests *Helicobacter pylori* IgG EIA Test Kit (Cat. No. 601 040, Biohit Plc, Helsinki, Finland). Assays were performed in batches of 40 samples on a microwell plate according to the instructions of the manufacturer.

All EIA techniques were based on measuring of the absorbance after a peroxidation reaction at 450 nm. Between the reaction steps the plates were washed using a BW50 Microplate Strip Washer (Biohit Plc, Finland). The absorbances were measured using a microwell plate reader (BP800 Microplate Reader, Biohit Plc, Finland). For determination of PGI and G-17 values 2nd order fit on standard concentrations was used to interpolate/extrapolate unknown sample concentrations (automatically, with the help of e-Lisa XL software, Biohit Plc, Finland). The *H.pylori* antibodies were expressed as enzyme immuno units (EIU) according to the formula included in the test kit. EIU levels above 38 were considered *H.pylori* positive.

Endoscopy and histological analysis

Endoscopes and biopsy equipment were thoroughly cleaned with a detergent, disinfected with 2% glutaraldehyde and rinsed with sterile water between all procedures.

All subjects underwent an endoscopy of upper digestive tract (Olympus GIF-Q30) with no use of image zooming or adjunctive mucosal staining techniques. Biopsies for histological examination were taken in each patient from the lesser and the greater curvatures in the mid-antrum and the middle body, one specimen from each location. Specimens were fixed in 10% formalin and embedded in paraffin. The sections (4 µm thickness) were deparaffinized, rehydrated, and stained separately with haematoxylin and eosin and alcian blue (pH 2.5) periodic acid Schiff to detect intestinal metaplasia. Histological examination was made in accordance with the updated Sydney system (22), assessing the degree of infiltration with neutrophils (activity) and mononuclear cells (chronic inflammation) scored 0 for none, 1 for mild, 2 for moderate and 3 for severe, respectively. Modified Giemsa stain was performed for assessment of *H.pylori* colonization. Diagnosis of MAG was made when a mild to severe loss of gastric glands with/without intestinal metaplasia was present at any site of the stomach. Histological examination was performed by two independent pathologists having no prior knowledge of endoscopic diagnosis or experimental results.

Statistical analysis

Difference in prevalence was evaluated by chi square test, and differences between means were evaluated by two-tailed *t* test. Sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) of the serological tests were estimated using histology as the "gold standard". Fisher's exact test for paired observations was used for the statistical analysis ; 95% confidence intervals were calculated for the differences as appropriate.

Results

In histological examination 29 patients presented non-atrophic pangastritis and in 7 patients the mucosa

Table 1. — **Histological characteristics of 55 subjects**

HISTOLOGY	Number of subjects	Gender M	Gender F	Metaplasia	Dysplasia
Normal	7	2	5	0	0
Non-atrophic pangastritis	29	11	18	8 (28%)	0
Atrophic gastritis (*total*)	19	12[a,b]	7	13 (68%)	1 (5%)
antrum-limited	11	10[a,c]	1	7 (64%)	1 (9%)
corpus-limited	4	1	3	3 (75%)	0
antrum & corpus	4	1	3	3 (75%)	0

[a] $p < 0.05$ as compared with normal ; [b] $p < 0,05$ as compared with non-atrophic gastritis ; [c] $p < 0,005$ as compared with non-atrophic gastritis.

Table 2. — ***Prevalence of H.pylori* infection evidenced by serological and/or histological methods**

HISTOLOGY	Number of patients	IgG-*H.pylori* positive	*Giemsa stain* positive
Normal	7	0	0
Non-atrophic gastritis	29	21[a]	18[a]
Atrophic gastritis	19	17[b]	9[c]
Total	55	38	23

Significantly more frequently than in patients with normal histology ; [a] $p < 0,0001$; [b] $p < 0,001$; [c] $p < 0,005$.

was normal (Table 1). Histological signs of atrophy were found in 19 patients (34,5%). Topography of atrophic gastritis and occurrence of intestinal metaplasia or dysplasia are shown in Table 1. Smoking cigarettes, consumption of alcohol in an amount of more than 3 drinks weekly, use of aspirin or regular consumption of vegetables were not significantly different in patients with atrophic gastritis than in subjects with no mucosal atrophy. Atrophy was significantly more frequent in men than in women (Table 1). There was no relationship between age of patients and histological diagnosis, including severity of mucosal atrophy.

In 13 patients the glandular atrophy in antrum and/or corpus was mild and in 6 patients of moderate or severe degree. In one patient the mucosal atrophy was present in all four tissue samples, in 2 patients in three samples, in 11 patients in two samples and in 5 patients only in one sample. Gastroscopic diagnosis of MAG was characterized by low sensitivity (21%) and this method was associated with 9 false positive diagnoses of mucosal atrophy.

Serological test detected infection with *H.pylori* in 69,1% of patients included in the study. However, in patients with MAG the *H.pylori* infection rate was of 89,5% and in patients with antral-dominant atrophic gastritis of 93,3%. The patients who had been positively tested for *H.pylori* and subsequently eradicated during the past 2 years have been included to *H.pylori* infected group, irrespective of the serological test result (among 10 patients eradicated 7 had IgG-antibodies to *H.pylori* < 38 EIU). The results of serological and histological evaluation of *H.pylori* status are shown in Table 2.

The serum levels of basal and protein stimulated G17, and pepsinogens ; PGI and PGII in relation with histological classification and severity of mucosal atrophy are presented in Tables 3 and 4, respectively. In 6 patients who stopped the use of proton pump inhibitors one week prior to the study serum levels of b-G17 and s-G17 were not significantly different to respective values in remaining subjects. The mean s-G17 serum level in patients with corpus atrophic gastritis was not significantly higher than in patients with no histological abnormalities ($59,8 \pm 84,2$ vs. $18,22 \pm 18,4$; $p > 0,05$). Histological presence of intestinal metaplasia had not significant influence on b-G17, s-G17 and PGI measurements. The scattergrams of s-G17 and PGI serum levels with regard to histological diagnosis are demonstrated in Figures 1 and 2.

Serum PGI in patients with corpus atrophic gastritis was significantly lower than in patients with antral atrophic gastritis ($36,6 \pm 26,5$ mg/l vs $91,6 \pm 51,4$ mg/l ; $p < 0,01$). Serum b-G17 in patients with antral atrophic gastritis was significantly lower than in patients with corpus atrophic gastritis ($2,16 \pm 2,11$ pmol/l vs $27,5 \pm 55,6$ pmol/l ; $p < 0,05$). Similarly, s-G17 in patients with antral atrophic gastritis was significantly lower than in patients with corpus atrophic gastritis ($5,84 \pm 4,52$ pmol/l vs $59,8 \pm 84,2$ pmol/l ; $p < 0,01$).

The diagnostic efficacy of all laboratory parameters depending on dominant topography of gastric mucosal atrophy and its severity is shown in Table 5. The highest sensitivity in detection of MAG by means of s-G17 and PGI serum levels were obtained for cut-off values of 5 pmol/l and 50 µg/l, respectively. The combination of

Table 3. — **G17 serum levels in basal conditions (b-G17) and after stimulation with protein drink (s-G17) in subjects with no histological signs of mucosal atrophy and in 15 patients with antral atrophic gastritis**

HISTOLOGY	b-G17 pmol/l	s-G17 pmol/l	Ratio s-G17 to b-G17
Normal	2,46 ± 1,68	18,22 ± 18,4	7,33 ± 4,55
Non-atrophic gastritis	12,74 ± 30,4	31,98 ± 47,8	4,82 ± 4,35
Antral atrophic gastritis (*total*)	2,16 ± 2,11	5,84 ± 4,52[a,b]	6,06 ± 12,5
Mild	2,52 ± 2,21	6,65 ± 4,73[a,b]	6,67 ± 13,6
Moderate or severe	0,72 ± 0,63	2,61 ± 0,55[b]	2,69 ± 0,72

[a] significantly lower than in patients with normal histology ; ($p < 0,05$)
[b] significantly lower than in patients with non-atrophic gastritis ; ($p < 0,05$).

Table 4. — **Pepsinogen I (PGI) and pepsinogen II (PGII) serum levels in subjects with no histological signs of mucosal atrophy and in 8 patients with corpus atrophic gastritis**

HISTOLOGY	PGI µg/l	PGII µg/l	Ratio PG I to PG II
Normal	124,5 ± 124,4	9,89 ± 7,76	11,64 ± 2,34
Non-atrophic gastritis	122,1 ± 92,5	13,0 ± 8,59	10,3 ± 4,0
Corpus atrophic gastritis (*total*)	36,6 ± 26,5[a,b]	7,69 ± 5,18	5,60 ± 3,42[a,b]
Mild	45,3 ± 31,1[b]	9,13 ± 5,97	5,95 ± 4,14[a,b]
Moderate or severe	22,1 ± 4,44[b]	5,29 ± 2,93	5,02 ± 2,43[a,b]

[a] significantly lower than in patients with normal histology ; ($p < 0,05$)
[b] significantly lower than in patients with non-atrophic gastritis ; ($p < 0,05$).

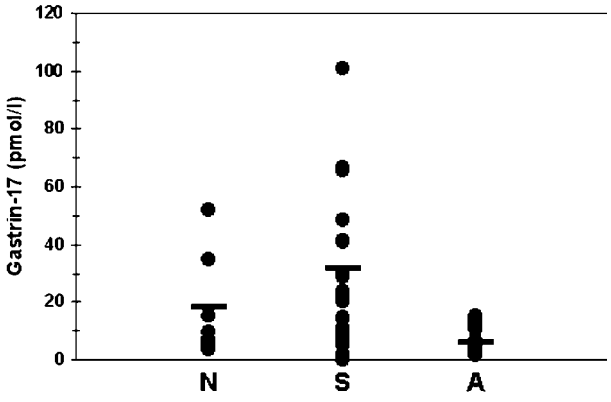

Fig. 1. — Individual and mean s-G17 serum levels in patients with normal mucosal histology (N), non-atrophic superficial gastritis (S) and atrophic gastritis (A). Serum level of s-G17 in patients with atrophic gastritis is significantly lower than in patients classified in groups N and S ($p < 0,05$).

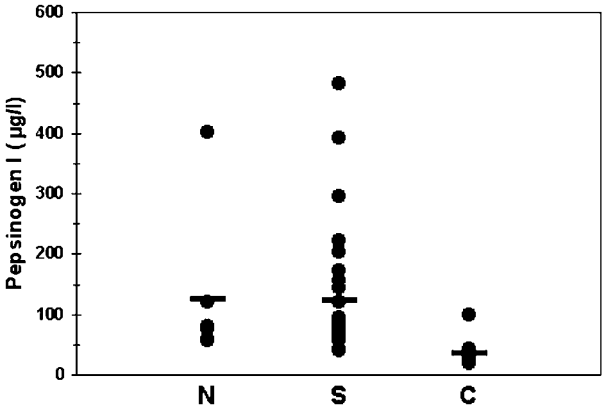

Fig. 2. — Individual and mean PGI serum levels in patients with normal histology (N), non-atrophic superficial gastritis (S) and atrophic gastritis (C). Serum level of PGI in patients with atrophic gastritis is significantly lower than in patients classified in groups N and S ($p < 0,05$).

these two peptides detected MAG with sensitivity of 84% (CI 61-96%). Correct diagnosis of MAG was provided by the test panel in 16 of 19 patients. Equal diagnostic accuracy was achieved by combination of s-G17 and PGI/PGI < 6. In subgroup of patients with moderate and severe atrophy there was no false negative diagnoses of MAG (sensitivity of 100%), however, false positive rate was of 31% (17/55). The test panel with PGI cut-off value < 30 µg/l better discriminated atrophic from non-atrophic gastritis with none false negative result and false positive rate of 21,8% (12/55). The sensitivity of *H. pylori* serological test in detection of MAG was 89,5% (specificity 27,6%). Inclusion of *H.pylori* antibodies > 38 EIU to the test panel resulted in lower number of false positive recognitions of MAG, but had no favorable effect on its diagnostic sensitivity (Tables 5 and 6).

Discussion

An early diagnosis of MAG and eradication of *H.pylori* form a basis for the treatment of atrophic gastritis and the prevention of related diseases such like

Table 5. — **Efficacy of different laboratory parameters in diagnosis of atrophic gastritis**

Parameter	Diagnosis	Sensitivity	Specificity	Accuracy	PPV	NPV
b-G17 < *2 pmol/l*[a]	A	53%	70%	36%	40%	80%
s-G17 < *5 pmol/l*	A	67%	90%	84%	71%	88%
s-G17 < *5 pmol/l*	A2	100%	79%	80%	21%	100%
s-G17 / b-G17 < *2*	A	38%	87%	74%	50%	80%
s-G17 < *5 pmol/l* & IgG-*H.pylori* +	A	67%	97%	89%	91%	89%
PGI < *30 µg/l*	C	62%	100%	94%	100%	94%
PGI < *50 µg/l*	C	88%	91%	91%	64%	98%
PGI < *30 µg/l*	C2	100%	96%	96%	60%	100%
PGI < *50 µg/l*	C2	100%	85%	85%	27%	100%
PGI / PGII < *6*	C	75%	94%	91%	67%	96%
PGI < *50 µg/l* & IgG-*H.pylori* +	C	75%	96%	93%	75%	96%
s-G17 < *5 pmol/l* and/or PGI < *50 µg/l*	MAG	84%	81%	82%	70%	91%
s-G17 < *5 pmol/l* and/or PGI/PGII < 6	MAG	84%	81%	82%	70%	91%
s-G17 < *5 pmol/l* and/or PGI < *50 µg/l* & IgG-*H.pylori* +	MAG	74%	94%	87%	87%	87%

Legend : PPV – positive predictive value ; NPV – negative predictive value

A- antral-dominant atrophic gastritis ; A2- antral-dominant moderate or severe atrophic gastritis ; C- corpus atrophic gastritis ; C2- corpus moderate or severe atrophic gastritis ; MAG- multifocal atrophic gastritis.

Table 6. — **Accuracy of three test panels in diagnosis of gastritis with and without atrophy. Test Panel I = s-G17 < 5 pmol/l and/or PGI < 50 µg/l ; Test Panel II = s-G17 < 5 pmol/l and/or PGI/PGII < 6 ; Test Panel III = s-G17 < 5 pmol/l and/or PGI < 50 µg/l combined with *H.pylori* infection approved by serological test**

Histology	Test Panel I		Test Panel II		Test Panel III	
	No atrophy	Atrophy	No atrophy	Atrophy	No atrophy	Atrophy
Normal	6	1	6	1	7	0
Non-atrophic gastritis	23	6	23	6	27	2
Atrophic gastritis	3	16	3	16	6	13
Total	32	23	32	23	40	15

Diagnostic efficacies of the test panels not significantly different in chi2 square test.

deficiency of vitamin B_{12}, gastric peptic ulcer and gastric cancer. The prevalence of MAG is dependent on a country-specific incidence rate of *H.pylori* infection. Since it generally takes 20 years before one-third of a population with *H.pylori*-related gastritis develops mucosal atrophy, the increasing age is associated with increasing proportion of individuals having histological signs of atrophic gastritis (23-25). In our study the prevalence of atrophic gastritis in dyspeptic patients aged over 55 years was 34,5%. In other regions of high incidence rate of *H.pylori* infection such like Columbia or Japan the prevalence of MAG in patients aged 60 years is estimated about 60% (2,26-28). However, it should be noted that an assessment of MAG prevalence in cross-sectional studies may be largely influenced by the selection of subjects and choice of diagnostic methods. Apart from age we did not use any preselection criteria of dyspeptic patients in whom four mucosal samples were obtained, as recommended by Sydney System (22). To the best of our knowledge there is no study evaluating the number of mucosal biopsies needed for an accurate histological diagnosis of MAG. Although for the purpose of our study the presence of atrophy even in a single mucosal sample was sufficient to diagnose MAG, we cannot rule out the possibility that small areas of atrophic mucosa escaped our detection.

The severity of atrophic gastritis, which should be a target-point for widespread screening is still unknown. The study by Lahner *et al.* (29) suggests that potential neoplastic lesions do not occur earlier than 4 years after the primary diagnosis of atrophic gastritis. Our subjects with MAG presented early stages of mucosal atrophy. This statement is based on histological observation that in 13 of 19 patients neither severe nor moderate atrophy was found in available specimens. Besides, in 16 of 19 patients the histological signs of glandular atrophy were present in no more than two of four collected samples.

Atrophic gastritis is a premalignant condition that is not systematically discovered, as it is not usually associated with dyspepsia. On the other hand, a tissue sampling in all *H.pylori* infected patients is not justified either on ethical or cost-benefit basis. In our study the endoscopic assessment of gastric mucosa had a negligible impact on recognition of MAG and was biased by high rate of false positive diagnoses. This finding is compatible with other endoscopic studies including ours (30-32), confirming that MAG is purely histological diagnosis as long as gastroscopy is not extended to adjunctive staining techniques (32-34). In this study we sought to determine, if the laboratory panel composed of serum PGI and protein stimulated G17 might select

patients with MAG, and what is diagnostic contribution of *H.pylori* serology in population highly-infected with this bacterium.

In a majority of infected patients, *H.pylori* causes chronic antral-dominant non-atrophic gastritis, and an increase or no change in acid secretion, which is the most likely scenario, will limit *H.pylori* colonization to the antrum. Indeed, 11 of 19 patients in whom MAG was diagnosed presented antrum-limited type of atrophic gastritis. The risk of mucosal atrophy depends upon the severity of gastritis, which is determined by various host- and bacteria-related factors. It is noteworthy that atrophic gastritis was significantly more frequent in males which implies that genetic or hormonal factors may play a role in the passage from chronic active to atrophic gastritis.

G17 is a gastrin isoform almost exclusively produced by antral G-cells. Basal serum levels of G17 are < 2,5 pmol/l and in an acidic stomach may be undetectable. The protein-rich meal stimulates G17 over twice that of baseline values and it has been shown that measurement of postprandial G17 is superior in recognition of antral mucosal atrophy to non-stimulated both total gastrin and G17 molecular form (16). In accordance, our study provides evidence that antral atrophic gastritis may be better distinguished from non-atrophic antritis by low protein stimulated than basal G17 serum level (cut-off value < 5 pmol/l). PGI is a precursor to pepsin enzyme and is produced by gastric body glands (35). In many previous studies low PGI serum levels and decreased ratio of PGI to PGII were used as indices of immune- or *H.pylori*-mediated corpus mucosal atrophy, however, the diagnostic sensitivity of these biomarkers was assessed in broad ranges from 6% to 84% (36-38). In our study detection of MAG by the combined protein-stimulated G17 and PGI measurements was characterized by 84% sensitivity. Replacing PGI by the ratio of serum PGI to PGII resulted in equal diagnostic efficacy of the test panel, however, an additional cost associated with measurement of PGII favors the use of PGI only. More advanced atrophy was diagnosed by the test panel with excellent 100% sensitivity, however, it yielded a false positive rate of 31% for the diagnosis of MAG.

In our study 89.5% of patients with histological evidence of MAG were infected with *H.pylori*, including those with recently eradicated bacterium and negative IgG-antibodies at the time of the study. These data are consistent with a general concept of strong association of *H.pylori* infection with atrophic gastritis, however, in two patients with MAG the bacterial infection was not confirmed either by serological test or histological study. An explanation for low titer of *H.pylori* antibodies in patients with MAG could be spontaneous loss of bacterium due to chronic hypochlorhydria or its involuntary pharmacological eradication (39,40). Finally, MAG might be caused by environmental non-bacterial factors. Lack of universal *H.pylori* positivity among patients with MAG was responsible for the fact, that *H.pylori* serology did not improve the diagnostic sensitivity of the test panel composed of secretory gastric peptides. Keeping in mind an occurrence of subjects with *H.pylori*-negative antral atrophic gastritis we would rather suggest not to incorporate *H.pylori* test into diagnostic algorithms as a key separator of MAG. Moreover, high prevalence of *H.pylori* infection may change interpretation standards of the test panel, as the bacterium deranges the homeostasis of secretory products of the gastric mucosa (35,41,42). In agreement with this opinion, we noted an elevated G17 serum level in subgroup of patients with antral non-atrophic gastritis, that may be explained by *H.pylori* direct and indirect effects on gastrin secretion (41). This finding should be perceived as a limitation to G17 efficacy in detection of mucosal atrophy, as this process is always accompanied by inflammation having a counteracting effect on G17 release. It is also well known that hypochlorhydria associated with mucosal atrophy of the gastric body may be another factor augmenting gastrin release (35). In accordance, the serum G17 levels were increased in our patients with corpus atrophic gastritis.

In conclusion, low level of serum PGI or protein stimulated G17 matches with histological diagnosis of MAG, therefore this laboratory panel may be used as the "serological gastric biopsy". *H.pylori* serology did not increase the number of patients correctly classified to atrophic gastritis group. The usefulness of measurement of gastric secretory peptides in screening programs to identify patients at risk for gastric cancer warrants further studies.

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