BARRETT´S ESOPHAGUS INCREASES THE RISK FOR PRENEOPLASTIC LESIONS IN CARDIA TYPE MUCOSA

Gamboa-Domínguez A, **Rubio-Tapia A**, Kershenobich-Stalnikowitz D, Angeles-Angeles A.

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Instituto Nacional de Ciencias Médicas y Nutrición
"Salvador Zubirán"

Dr. Alfonso Gulias Herrero
Profesor Titular del Curso de Especialización en Medicina Interna
Instituto Nacional de Ciencias Médicas y Nutrición
"Salvador Zubirán"

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Editorial Office Magdeburg:
Albert Roessner, M.D.
Professor and Chairman
Department of Pathology
University of Magdeburg
Leipziger Straße 44
D-39120 Magdeburg
Germany
Tel.: ++49(0)391/671 58 17
Fax: ++49(0)391/671 58 18
e-mail: Albert.Roessner@medizin.uni-magdeburg.de

Armando Gamboa-Domínguez, M.D., M.Sci
Institut für Pathologie
Technische Universität München
Klinikum rechts der Isar
Ismaningerstr. 22

81675 München

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Barrett's Esophagus Increases the Risk for Preneoplastic/Inflammatory Lesions in Cardia Type Mucosa

Dear Doctor Gamboa-Domíguez:

Thank you very much for the submission of the manuscript mentioned above, which Prof. Schaefer passed on to our Editorial Office for further handling. I have read it with interest and think that it is basically suitable for publication in our journal.

Yours sincerely,

Albert Roessner, M.D.
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ASUNTO: Autorización del trabajo de investigación del Dr. Alberto Rubio Tapia.

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Me permito informar a usted que el Dr. Alberto Rubio Tapia, alumno del curso de especialización en Medicina Interna en el Instituto Nacional de Ciencias Medicas y de la Nutrición Salvador Zubirán, presenta el trabajo de investigación intitulado ***"Barrett's esophagus increases the risk for preneoplastic lesions in cardia type mucosa".***

De conformidad con el artículo 21 capítulo 5º. de las Normas Operativas del Plan Unico de Especializaciones Médicas (PUEM) se considera que cumple con los requisitos para validarlo como el trabajo formal de Investigación que le otorga el derecho de la diplomación como especialista.

Sin otro particular de momento, reciba un cordial saludo.

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LRP*drf

Barrett´s esophagus increases the risk for preneoplastic lesions in cardia type mucosa

Armando Gamboa-Domínguez, Alberto Rubio-Tapia*, David Kershenobich-Stalnikowitz**, Arturo Angeles-Angeles.

Departments of Pathology, Internal Medicine* and Gastroenterology.** Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán. Mexico City. Mexico.

Address request:

Department of Pathology.
Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán.
Vasco de Quiroga No. 15. Tlalpan.
México 14000, D. F.
Tel: (5)573-1200.
Fax: (5)513-5395
e-mail: agd5@prodigy.net.mx

ABSTRACT

Aim: To compare the prenoplastic/inflammatory changes of cardia in patients with classical Barrett´s esophagus and of patients with gastritis and intestinal metaplasia in the distal stomach.

Materials and Methods: Biopsies were prospectively taken from esophagus, z-line, cardia, fundus, corpus and antrum. Cases were included when all sites were sampled and had diagnoses of either Barrett´s esophagus (group A) or gastritis of the lower stomach associated to intestinal metaplasia (group B). A format evaluating clinical manifestations and symptom duration was filled out in the endoscopy room. Biopsies were processed for HE and PAS/AA pH 2.5. Immunohistochemical detection of Ki-67, bcl-2 and p53, and morphological evaluation by two blinded pathologists, using the Sydney system were performed. Results were compared with t, X^2, Mann Withney U and associated risk with logistic regression.

Results: 94 patients were included in the study. Hiatal hernia ($p<0.0006$) and regurgitation ($p=0.02$) were frequently identified in group A. A painful feeling of hunger ($p=0.000$) as well as abdominal pain ($p=0.0005$) were seen in higher frequency in group B. Intestinal metaplasia in cardia type mucosa ($p=0.01$), and more extensive and incomplete forms of metaplasia in cardia were appreciated in group A ($p=0.04$). An odds ratio for cardia lesion was found to be 3.5 times higher in group A than group B. No differences in expression of Ki-67, bcl-2 or p53 were detected.

Conclusions: Preneoplastic changes in cardia mucosa were 3.5 times more frequent in patients with Barrett´s esophagus than in patients with gastritis associated to intestinal metaplasia.

INTRODUCTION

Barrett's esophagus is the main preneoplastic change associated to distal esophageal carcinoma.[1] Chronic gastritis associated to intestinal metaplasia is a common finding in the most frequent adenocarcinoma of the distal stomach.[2] The first condition is etiologically related to gastroesophageal reflux disease while the second is commonly associated to chronic *Helicobacter pylori* infection.[1,2] Both metaplastic changes are morphologically similar, observing substitution of stratified esophageal epithelium and cilindric antral epithelium by columnar cells and goblet acid mucous producing cells.[3] It has been considered that incomplete intestinal metaplasia of the gastric mucosa is histochemically identical to Barrett's epithelium. Some studies have even shown identical sucrase isomaltase gene expression in both Barrett's mucosa and gastric intestinal metaplasia.[4] These changes, which are the milestone in carcinogenic sequences, are proposed for esophageal adenocarcinoma as well as for intestinal type adenocarcinoma of the distal stomach. [5, 6]

Information obtained from epidemiological studies has shown a rising incidence of adenocarcinomas of both the esophagus and gastric cardia. [7-9] However, most of the documented lesions are large and it is therefore difficult to identify the primary origin site. [10, 11] This situation has been resolved by some authors describing the overall lesion as esophagogastric adenocarcinoma with presumed typical clinical and morphological characteristics. [12] Retrospective designs of most studies have also contributed to this dilemma, since lesions from the distal esophagus and gastric cardia were always analyzed together. Hence, it has been considered that distal esophageal and proximal

gastric adenocarcinomas are identical entities, although there is lack of evidence clearly supporting this idea. [10-12]

The significance of incomplete intestinal metaplasia in gastric cardia is unknown[13] and the evolution of metaplastic changes in the esophagus and distal stomach is poorly understood. [14-18] According to the sequential schema, these changes should evolve to dysplasia, carcinoma *in situ* and finally to invasive adenocarcinoma. [6] However, the morphological transformation in the gastric cardia is not well defined. Some authors favor the hypothesis that the changes in the gastric cardia resemble those observed in the distal stomach commonly associated to *H pylori* infection. [14, 16, 17] A number of studies support that cardia changes are related to esophagogastric reflux disease and therefore to Barrett's esophagus.[11, 15] However, some other studies suggest that the inflammatory and preneoplastic changes in cardia mucosa are actually independent of both. [19]

Identification of acid and sulfatated mucous production by histochemistry with alcian blue/PAS diastase is recommended and commonly used for diagnoses of intestinal metaplasia. However, few studies describe the cell kinetic of the epithelial cells in gastric cardia and their expression of bcl-2 and p53. [20, 21]

The aim of this paper is to describe the morphological spectrum of changes of the gastric cardia in patients providing the two natural "models", of chronic inflammatory damage of the upper gastrointestinal tract, i.e. Barrett's esophagus, and the other of chronic gastritis associated to intestinal metaplasia in the distal stomach.

MATERIALS AND METHODS

Patients referred to the Endoscopy Unit for gastroesophageal symptoms were invited to participate in the study. Signed consent and symptom evaluation questioners were obtained before endoscopic exploration. Patients with systemic disease complicating the endoscopic procedure or with intrinsic factor deficiency were excluded.

Tissue source and preparation: two biopsies were obtained from distal esophagus, esophagogastric junction, cardia mucosa, fundus, corpus and antrum. When hiatal hernia was present the biopsies were obtained one centimeter proximal to the terminal gastric folds. Biopsies were fixed with 10% fomalyne and paraffin embedded. HE and PAS-D/AA pH2.5 were obtained from each paraffin block and immunostains for Ki-67, bcl-2, and p53 (Dako, Carpinteria, CA), using an automated stainer (Ventana Nexes. Tucson, AZ) were performed. Diagnoses and a semi cuantitative value was blindly obtained from each section, using the updated Sydney system[22] for inflammation, atrophy, metaplasia and H. pylori.

Cases were included if patients had classical Barrett´s esophagus or chronic gastritis with intestinal metaplasia of the antrum, an adequate sampling of esophagus and stomach and well defined cardia type mucosa. Cardia mucosa was defined histologically as mucous glands lacking oxhyntic cells surrounded by edematous stroma and covered with columnar PAS positive secretory cells, resembling antral mucosa.

Differences were contrasted using two sided t, X^2, and Mann Whitney U. The risk of association to gastric cardia preneoplastic lesion was obtained using log Rank between groups.

RESULTS

One hundred and twenty patients accepted the extended sampling of the esophagogastric mucosa. Twenty six cases were excluded for absence of cardia type mucosa (16), absence of intestinal metaplasia in antrum (5), simultaneous presence of intestinal metaplasia in antrum and Barrett´s mucosa (3), or absence of lesions in both antrum and esophagus (2). The remainder 94 patients were distributed into two equal groups comparative in all aspects except for the presence of hiatal hernia ($p=0.0006$), table 1.

Clinical symptoms suggesting gatroesophageal reflux disease were mainly observed in the group with Barrett´s esophagus ($p=0.02$), whereas abdominal pain ($p<0.000$), dolorous hunger ($p=0.0005$) and nausea ($p=0.05$), classical signs and symptoms of gastritis, where found in the group with antral intestinal metaplasia, table 2.

Morphologic analysis showed a higher association of atrophy of the gastric cardia mucosa with antral intestinal metaplasia ($p=0.05$), and of incomplete intestinal metaplasia of cardia mucosa (figure 1) with Barrett´s esophagus ($p=0.01$). No differences were observed in frequency of complete intestinal metaplasia of the cardia type mucosa, in acute or chronic inflammation, between groups, table 3. When incomplete intestinal metaplasia in the cardia type mucosa was graded as mild, moderate or severe, according to the updated Sydney system,[22] lesions with the higher grade were mostly observed in the group with Barrett´s esophagus ($p=0.04$). Graphic.

A 3.5 higher risk of preneoplastic lesion in cardia type mucosa of patients with Barrrett´s esophagus was determined.

No bcl-2 or p53 expression was observed in epithelial cells of the cardia mucosa. Ki-67 was observed in less than 5% of epithelial cells of the glandular neck, thus reflecting normal proliferation and the incipient lesions of this study. In 10 cases of Barrett´s esophagus, expression of Ki-67 and p53 in the metaplastic epithelium was observed. The latter was strongly expressed in two cases, one with low grade and the other with high grade dysplasia.

DISCUSSION

There is small literature supporting early preneoplastic changes in gastric cardia; most of the reports have patients with Barrett´s esophagus or distal gastritis, but have never considered either condition as a natural "model" of cardia preneoplastic changes. In this study we found a risk 3.5 times higher of preneoplastic changes in gastric cardia in the presence of Barrett´s esophagus than in patients with antral gastritis with intestinal metaplasia. These data support the view that cardia and distal esophageal mucosa are pathogenetically related, and therefore that neoplastic proliferations in both sites are identical.[23, 24]

Incipient lesions in gastric mucosa anteceding dysplasia and adenocarcinoma include hyperplasia, atrophy and intestinal metaplasia. The latter is the most consistent finding in the vicinity of gastric intestinal type adenocarcinomas and supposes a higher risk for developing dysplasia,[5] besides the fact that it is relatively easy to identify morphologically. Incomplete intestinal metaplasia is a second step in the sequence and suggests a higher instability of the mucosa. Both changes were more prevalent and widespread in gastric cardia mucosa of patients with Barrett´s esophagus than in those with gastritis and antral intestinal metaplasia. This relatively early but definitive mucosal change allows us to suggest that at least in this phase of gastric carcinogenesis, the cardia mucosa is more similar to esophageal rather than antral metaplastic mucosa. These results are consistent with the view of considering gastroesophageal reflux as a condition for the development of cardia preneoplastic changes.[23] However, cell proliferation (Ki-67), cell growth control (bcl-2) and p53 overexpression were not

different, probably because of the incipient changes analyzed in this study or due to insensitivity of immunohistochemistry during the early phases of subcellular lesions.[6, 18, 20, 21] In this study not one case of dysplasia on the gastric cardia was found even though two cases of Barrett´s mucosa did show it, thus reflecting low progression to higher preneoplastic lesions in this type of mucosa.[25]

The differences found between the two groups are strong because of the prospective design of the study, the blind evaluation and inclusion of the cases on morphological basis. This precaution is paramount and must be considered when regions such as the gastric cardia are evaluated, i.e., a zone without anatomical landmarks, endoscopically poorly defined and with a position dependent on the dynamic situation of the patient.[17] Here we defined histologically cardia mucosa always ensuring that the biopsy be obtained proximal to the terminal gastric folds. When this histological definition was not achieved, the patient was not included, thus explaining 16 of the 26 excluded cases of this study. Also, excluding patients with intrinsic factor deficiency was a mean to obtain homogeneous groups only different in hiatal hernia and gastritis manifestations. Another interesting finding was the high frequency of chronic inflammation of the cardia mucosa which was present in nearly 100% of cases. This is coincident with the results previously reported for inflammatory damage in cardia mucosa in patients with esophagogastric reflux disease and chronic multifocal atrophic gastritis.[16, 17]

Our conclusions are that (i) in the early stages of gastric and esophageal carcinogenesis the cardia mucosa is more similar to Barrett´s mucosa and that (ii) cardia mucosa has 3.5 times higher risk of having preneoplastic change in the presence of classical Barrett´s esophagus than in the presence of antral gastritis with intestinal metaplasia.

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Table 1. Com arison of atients included in two grou s (n=94).

		Barrett´s esophagus (n=47)	Gastritis + intestinal metaplasia (n=47)	*p*
Gender	Female	20	28	0.09
	Male	27	19	
Age	Median + SD	56.6 (16.8)	56.9 (15.0)	0.90
Alcoholism		14	16	0.65
Tabaquism		25	21	0.41
Hiatal hernia		38	22	0.0006

Table 2. Clinical data evaluating gastroesophageal reflux and gastritis

	Barrett´s esophagus (n=47)	Gastritis + intestinal metaplasia (n=47)	*p*
Regurgitation	32	21	0.02
Dysphagia	11	4	0.08
Odinophagia	7	1	0.06
Pyrosis	22	26	0.41
Halitosis	3	6	0.48
Abdominal pain	9	33	0.000
Painful hunger	5	21	0.0005
Diarrhea	2	3	1.0
Plenitude	6	14	0.07
Nausea	2	9	0.054

Corrected X^2 Yates.

Table 3. Gastric cardia lesions

	Barrett´s esophagus (n=47)	Gastritis + intestinal metaplasia (n=47)	*p*
Atrophy	4	12	0.05
Complete IM	6	4	0.73
Incomplete IM	17	6	0.01
H. pylori	7	10	0.5
Acute inflammation	16	12	0.4
Chronic inflammation	46	46	0.5

IM intestinal metaplasia

Corrected X^2 Yates.