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Original Research Article

Histopathological spectrum in patients of gastro-esophageal reflux disease: A prospective study in a Tertiary Care Institute

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Abstract

Introduction: Gastroesophageal reflux disease (GERD) presents with classical symptoms of heart burn and acid regurgitation. Endoscopically, it is divided into erosive reflux disese (ERD) and non-erosive reflux disease (NERD) depending on the presence or absence of visible mucosal breaks in esophagus.
Aims: This prospective study was undertaken with the aim to assess the histopathological spectrum in esophageal biopsies taken from patients with GERD.
Method: Esophageal biopsies were taken from 150 patients of GERD and 50 controls. GERD cases were defined by the presence of classical symptoms of heart burn and/or acid regurgitation. Endoscopically, GERD cases were subdivided into ERD (n=70) and NERD (n=80). Six histological parameters studied in esophageal biopsy were basal cell hyperplasia, papillary elongation, dilatation of intercellular spaces, intra-epithelial eosinophil, intra-epithelial neutrophil and erosion/necrosis. Each parameter was given a score (range: 0-2) and total score of microscopic lesions ranged from 0-12 and a cut-off value (score ≥2) was used to distinguish GERD patients from controls.
Results: Histological alterations were observed in 144 (96%) of GERD patients. All controls had esophageal biopsy score of <2, while 74(92.5%) NERD and 70(100%) ERD patients had a score≥2. Esophageal biopsy had a sensitivity of 96% and specificity of 92%.
Conclusion: The high sensitivity and specificity of histopathology findings leads us to strongly support the role of esophageal biopsy in diagnosing GERD especially NERD.
Keywords: GERD, esophageal biopsy, cut-off score>=2.

1. Introduction

Gastroesophageal reflux disease (GERD) is a chronic relapsing disorder diagnosed by the presence of classical symptoms (heartburn and/or reflux), endoscopic findings (erosive or non-erosive reflux disease) along with histological changes in esophageal biopsy. Prevalence in western countries is 10-29%.[1] It is now becoming an emerging digestive disease in India also.[2] This prospective study was undertaken with the aim to determine the histological spectrum in esophageal biopsies in patients of GERD and comparing them with controls.

2. Methods

This prospective study was conducted for a period of one year. 150 patients coming to the Gastroenterology OPD with complaints of heartburn and/or regurgitation were included as cases of GERD. 50 patients attending the

OPD with complaints other than heartburn or reflux and having normal endoscopic findings were included as controls. Patients with evidence of cancer or mass lesion in the esophagus, gastric lesions (ulcer, polyp, cancer), severe gastroparesis, oesophageal varices, inability to discontinue non-steroidal anti-inflammatory drugs or aspirin prior to the study, severe uncontrolled coagulopathy and prior history of oesophageal or gastric surgery were excluded from the study. On the basis of presence/absence of erosion/ulcer on endoscopy, GERD cases were further subdivided into non erosive reflux disease (NERD) and erosive reflux disease (ERD) group depending upon the absence or presence of visible erosions on endoscopy, respectively. Multiple esophageal biopsies were obtained from 2cm proximal to the gastroesophageal junction. Histopathological findings were assessed for a maximum histological score of 12.

1. Erosion/necrosis	Score 0:- absent; 2:- present
2. Intra-epithelial eosinophil	Score 0:- absent; 1:1/hpf; 2:>1/hpf
3. Intraepithelial neutrophils	Score 0:- absent; score 2: present
4. Basal cell hyperplasia	Score 0:- <15% of epithelial thickness
	Score 1:- 15%-30% of epithelial thickness
	Score 2:- >30% of epithelial thickness
5. Papillary elongation	Score 0:- <50% of epithelial thickness
	Score 1:- 50%-75% of epithelial thickness
	Score 2:- >75% of epithelial thickness
6. Dilated intercellular spaces	Score 0: absent; 1: small; 2: large [3]

2.1 Statistical analysis

Epi info software system (version 7.1) was used for statistical assessment of these histological parameters. The sensitivity and specificity were calculated for endoscopic biopsy by considering classical symptoms of heartburn and/or regurgitation as gold standard.

3. Results

Among the GERD (NERD+ ERD) patients (n=150), there were 89 males and 61 females with a male: female ratio of 1.45:1.

A statistically significant difference ($p<0.05$) was found between cases of GERD and controls in papillary elongation, basal cell hyperplasia, intra-epithelial neutrophils and dilated intercellular spaces (Table 1).

Table 1: Comparison of esophageal biopsy findings of GERD cases with controls

Histopathology findings	GERD (n=150)		Controls (n=50)		P value (Fisher's exact test)
	No	%	No	%	
Papillary elongation (PE)	106	70.66	1	2	<0.05
Basal cell hyperplasia (BCH)	98	65.33	0	0	<0.05
Dilated intercellular spaces (DIS)	99	66	3	6	<0.05
Intra-epithelial neutrophils (IEN)	18	12	0	0	0.0123
Intra-epithelial eosinophils (IEE)	1	0.66	0	0	0.562
Erosion/Necrosis (Er/N))	9	6	0	0	0.0763
Normal	6	4	46	92	<0.05

These parameters also revealed a statistically significant difference ($p<0.05$) when NERD and ERD were separately compared with the controls (Table 2 & 3).

Table 2: Comparison of esophageal biopsy findings of NERD cases with controls

Histopathology findings	NERD (n=80)		Controls (n=50)		P value (Fisher's exact test)
	No.	%	No.	%	
Papillary elongation (PE)	59	73.75	1	2	<0.05
Basal cell hyperplasia (BCH)	51	63.75	0	0	<0.05
Dilated intercellular spaces (DIS)	50	62.5	3	6	<0.05
Intra-epithelial neutrophils (IEN)	7	8.75	0	0	0.031
Intra-epithelial eosinophils (IEE)	0	0	0	0	0
Erosion/ Necrosis (Er/N))	0	0	0	0	0
Normal	6	7.5	46	92	<0.05

Table 3: Comparison of esophageal biopsy findings of ERD cases with controls

Histopathology findings	ERD (n=70)		Controls (n=50)		P value (Fisher's exact test)
	No	%	No.	%	
Papillary elongation (PE)	47	67.14	1	2	<0.05
Basal cell hyperplasia (BCH)	47	67.14	0	0	<0.05
Dilated intercellular spaces (DIS)	49	70	3	6	<0.05
Intra-epithelial neutrophils (IEN)	11	15.71	0	0	<0.05
Intra-epithelial eosinophils (IEE)	1	1.42	0	0	0.396
Erosion/Necrosis (Er/N)	9	12.85	0	0	0.076
Normal	0	0	46	92	<0.05

Histological alterations were observed in 144 GERD patients. 74(92.5%) out of 80 NERD and 70(100%) out of 70 ERD subjects had a score≥2. All controls had esophageal biopsy score of <2. 4 controls also showed some histological changes (but total histological score remained <2) (Figure 1).

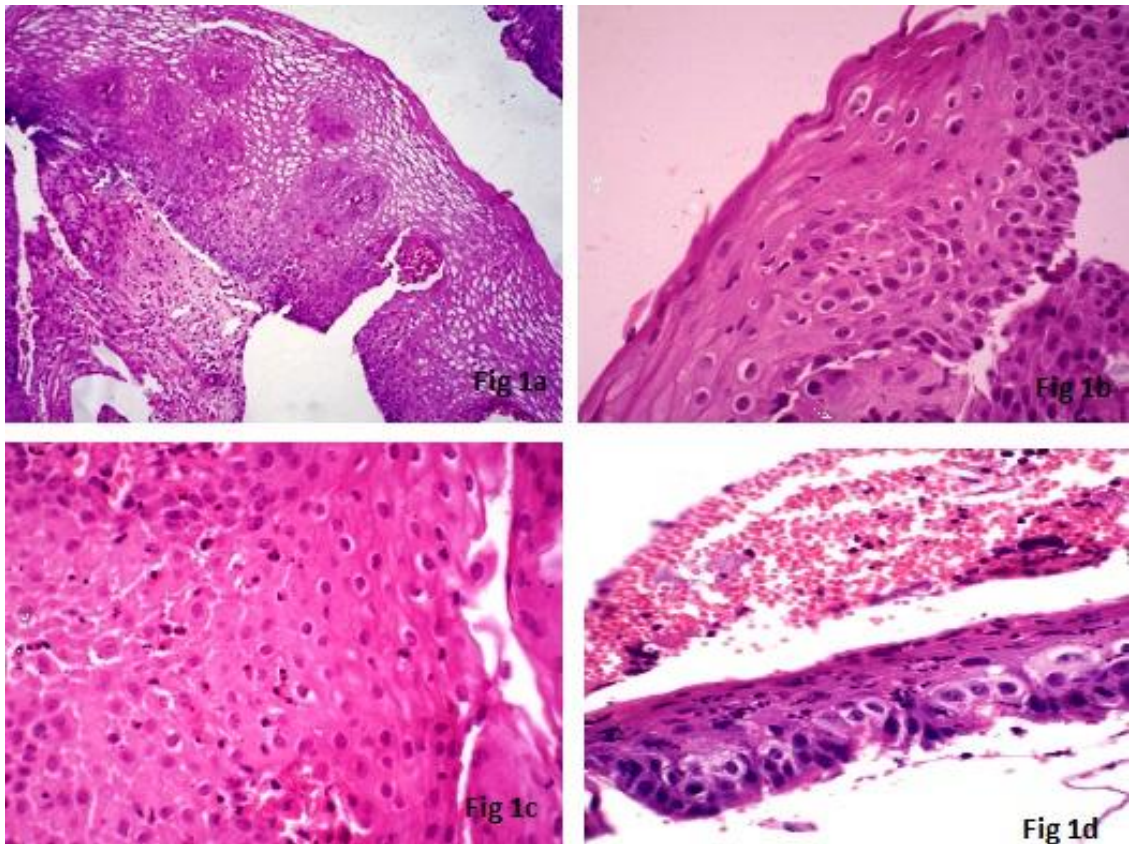

Figure 1: 1a) Papillary elongation >90% (score=2). 1b) Basal cell hyperplasia> 30% (score=2). 1c) Dilated intercellular spaces (score=2) & Intraepithelial eosinophils. 1d) Erosion and intra-epithelial neutrophils (H&E 40X)

A significant correlation (p<0.05) was seen between cases and controls. Thus, a cut-off score of ≥2 efficiently distinguished GERD cases from controls. In our study, the sensitivity of esophageal biopsy in diagnosis of GERD was 96% and specificity was 92%.

4. Discussion

Gastroesophageal reflux disease is an important health problem and its prevalence is rapidly increasing globally. The present one year study was undertaken to assess the histological features of gastroesophageal reflux disease.

In the present study, among the GERD patients, male: female ratio was 1.45:1. This is consistent with the study conducted by Zentilin P *et al* and Tadiparthi *et al* (2011).[3,4] However, in the study conducted by Nwokediuko *et al* (2011), male: female ratio was 0.75:1.[1]

In the study conducted by Zentilin P *et al*, abnormal histology was found in 100 of 119 GERD patients (84%) and in 3 of 20 controls (15%) and the difference was

highly significant.[3] Our results are consistent with their results wherein abnormal histology was found in 144 of 150 GERD cases (96%) and 4(8%) controls and there was a statistically significant difference ($p<0.05$).

When NERD cases were compared with controls, Zentilin P *et al* found histological changes in 54 of 71 NERD patients (76.05%) and in 3 of 20 controls (15%) and the difference between them was significant ($p<0.05$).[3] In the study conducted by Kiesslich *et al*, abnormal histology was found in 27 of 39 NERD patients (69.23%) and in 8 of 39 controls (20.51%) and the difference between them was significant ($p<0.05$).[5] Our results are consistent with these studies. Even though 4 controls (8%) revealed histological changes, the total score was <2 (Table 2).

When ERD cases were compared with controls, Zentilin P *et al* found abnormal histology in 46 of 48 patients (96%) and in 3 of 20 controls (15%) and the difference between them was significant.[3] Our results are consistent with this study (Table 3).

In our study, papillary elongation was seen in 70.6% cases and 2% control and there was statistically significant difference between cases and controls ($p<0.05$). The finding is consistent with Zentilin P *et al* who found papillary elongation in 62% cases while Takubo *et al* observed it in 61% cases and there was a statistically significant correlation in both the studies.[3,6]

Dilated intercellular spaces (DIS) were seen in 66% cases of GERD and 6% controls with a significant difference between cases and controls (p value <0.05) in our study. These results are consistent with those of Zentilin P *et al* who observed this in 86% cases and Takubo *et al* in 48% cases and both studies revealed a statistically significant correlation ($p<0.05$).[3,6] Tobey *et al* (1996) and Caviglia *et al* (2005) used electron microscopy to assess dilated intercellular spaces and concluded that dilated intercellular spaces is an objective structural marker of GERD symptoms as was observed in our study as well.[7,8] According to Tadiparthi *et al* (2011)and Villanacci *et al* (2001), light microscopic evaluation of DIS correlates reasonably well with the electron microscopy measurements.[4,9]

In our study, basal cell hyperplasia (BCH) was seen in 65.33% cases of GERD and none of the controls with a significant difference (p value <0.05). In the study by Zentilin P *et al* and Takubo *et al* basal cell hyperplasia was seen in 93% and 57% of the GERD cases respectively and a statistically significant difference was observed between cases and controls ($p<0.05$).[3,6]

Intra-epithelial neutrophils (IEN) were seen in 12% cases and were found to be a statistically significant finding ($p<0.05$). Similar results were noted by Zentilin P *et al* who observed intra-epithelial neutrophils in 7% cases of GERD and a statistically significant difference was reported ($p<0.05$).[3]

Intra-epithelial eosinophils (IEE) were seen in 0.66% of GERD and in none of the controls. The difference between cases and controls was not found to be statistically significant ($p>0.05$). Zentilin P *et al* noted intraepithelial eosinophils in 10% controls and 49% cases of GERD, but despite their high prevalence, they concluded that diagnostic impact of eosinophils was poor.[3] Takubo *et al* and Frierson *et al* also did not find statistically significant difference [6,10]

Erosion/necrosis (Er/N) was seen in 6% cases of GERD in our study but none of the controls and the difference was not significant statistically ($p=0.076$) while Zentilin P *et al* noted erosions in 49% cases and 10 % controls and the difference was statistically significant ($p=0.012$).[3]

The difference between NERD cases and controls when assessed for papillary elongation, basal cell hyperplasia, dilated intercellular spaces and intra-epithelial neutrophils was found to be statistically significant ($p<0.05$). Intra-epithelial eosinophils and erosion were not seen in any of NERD cases or controls. Our results are consistent with the results of Zentilin P *et al* and Kiesslich *et al*.[3,4]

In the present study, esophageal biopsies from subjects with ERD revealed statistically significant difference ($p<0.05$) between cases and controls in papillary elongation, dilated intercellular spaces, basal cell hyperplasia and intra-epithelial neutrophils. However, intra-epithelial eosinophils and erosion did not reveal statistically significant difference ($p>0.05$). These findings are consistent with the results of Zentilin P *et al*. However, in their study, intra-epithelial eosinophils were seen in 69% cases and had a significant correlation between cases and controls ($p<0.05$), which was not found in our study.

In our study, the sensitivity of esophageal biopsy in diagnosis of GERD was 96% and specificity was 92%. Vieth *et al* (2004) suggest a high specificity but very low sensitivity.[11] Our sensitivity is comparable to that found by Behar *et al* (1967), Ismail Begi *et al* (1970), Vieth *et al* (2004), Zentilin P *et al* (2005) and Mastracci *et al* (2009).[3,11-14] However, Seefeld *et al* (1977) and Kiesselich *et al* (2004) have reported a lower sensitivity of histology.[5,15] Our high sensitivity and specificity can be justified by the fact that cases and controls were selected very carefully. Also, six histological parameters were used to assess esophageal biopsies and each parameter was scored individually. The ultimate diagnosis of GERD was confirmed only if the total score of these individual parameters was ≥2.

5. Conclusion

The assessed histological parameters showed a good statistical correlation with GERD especially if a cut-off score>=2 is used and hence are important diagnostic indicators. The high sensitivity and specificity of histopathology findings lead us to strongly support the role of histology in diagnosis of GERD especially NERD.

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