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Evaluation of Mast Cell and Eosinophil Count in Duodenal Mucosa- A Study on 69 Functional Dyspepsia Cases from Bangladesh

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Abstract
The exact cause of functional dyspepsia is not known. This study was designed to investigate the association between duodenal infiltration of immune-mediating cells, namely eosinophils and mast cells with functional dyspepsia. Total of 69 patients presenting with symptoms of functional dyspepsia and matched healthy 42 controls were included in this cross-sectional study. Tissue from duodenum was taken from both patients and controls, and the number of eosinophils and mast cells per five high power fields were counted. 't' test and chi square tests were done to find out the association of immune-mediating cells with functional dyspepsia. p value <0.05 was taken as significant. Duodenal eosinophil count ranges from 1-92 with mean 21.59 in cases and from 3-51 with mean 14.90 in controls. Eosinophil count was abnormally high among 29 (42.02%) and 08 (19.02%) among cases and controls, respectively, and the difference was significant (P=0.013). Mast cell density was seen among 56 cases and 23 controls from the same sample. Mast cell per 5 high power fields varied from 1-139 (mean 20.0536) and 3-57 (mean 20.86) were found in cases and controls groups, respectively (P=0.627). This study showed that functional dyspepsia is associated with a significant increase of eosinophils count in duodenum, but no significant association could be identified between of mast cell count in duodenum and functional dyspepsia.

Keywords: Functional Dyspepsia, Duodenum, Eosinophil, Mast Cell

Introduction
Functional dyspepsia (FD) is defined as unexplained pain or discomfort centered in the upper abdomen affecting 10% of the world’s population (El-Serag & Talley,2004; Zagari et al.,2010). Many who suffer with dyspepsia take medication, and up to 23% visit the general practitioner in a year (Penston & Pounder ,1996). Although FD
is generally non-life-threatening, this disorder places a substantial burden on affected individuals because of a decreased quality of life compared with healthy subject and incurs high direct and indirect health care costs (Nyrop et al., 2007). Current diagnostic criteria use a symptom-based classification, the Rome III criteria and patients are defined by symptoms (epigastric pain or burning, postprandial fullness, or early satiation) but no pathology (Tack et al., 2006).

Currently, various mechanisms, including visceral hypersensitivity, Helicobacter pylori (H. pylori) infection, altered gut microbiome, and psychosocial dysfunction, have been proposed (Futagami et al., 2011). However, the pathogenesis of FD still remains poorly understood. Recent evidence implicates the duodenum in altering gastric accommodation and emptying (Lee & Tack, 2010). Researchers recently noticed close relation of FD with duodenal immune activation (Friesen, Schurman, Colombo, & Abdel-Rahman, 2013). Studies from Sweden (Talley et al., 2007), and UK (Walker et al., 2010) found significant duodenal eosinophilia in subjects with FD compared with controls. Similarly, in post infectious FD in Japan (Futagami et al., 2010) and in children with FD in USA (Friesen, Sandridge, Andre, Roberts, & Abdel-Rahman, 2006; Friesen, Garola, Hodge, & Roberts, 2002), duodenal eosinophilia has been observed, indicating that these observations are highly likely to be clinically relevant.

Furthermore, mast cells (MCs) are known to be involved in and essential for eosinophilic inflammation. There is a crosstalk between MCs and eosinophils, apparently because the 2 types of cells interact with each other (Walker & Talley, 2008). Recent studies have demonstrated that MCs can initially induce eosinophils into the gastric mucosa and these eosinophils, in turn, promote MC survival, proliferations, maturation and degranulation by secretion of various growth factors (Friesen et al., 2013; Walker & Talley, 2008). Thus, eosinophils and MCs are co-dependent in the development of visceral hypersensitivity, and their interaction may be a leading cause of symptoms of functional gastrointestinal disease (Powell et al., 2010).

With this background, this study was designed to see the duodenal eosinophil count and mast cell count in patients with functional dyspepsia, especially patients with postprandial fullness, early satiation and to compare those with healthy individuals.

Material and methods

Selective patients presenting with postprandial fullness, early satiation, bloating, and distress syndrome having no lesion at endoscopy of upper GIT were included in the study. Patients having GIT surgery, pancreatico-biliary disease, pregnant lady, age below 18 years and unwilling to take part in the study were excluded from the study. Patients with overt or medical conditions known to increase peripheral and tissue eosinophils like inflammatory bowel disease, coeliac disease, vasculitis, connective tissue disease, active infection, and allergy were also excluded. Consecutive (age-matched) 42 patients undergoing upper GIT endoscopy with indications other than above-mentioned symptoms were taken as controls. Two grasps of tissue using biopsy forceps were taken from the second part of duodenum for histological examinations from cases and controls. Tissue samples were examined for eosinophil counts and mast cell using Hematoxylin and Eosin stain (H-E) (Figure 1) and Giemsa stains, respectively. Eosinophil counts < 22 / 5 HPF were taken as normal9.

Statistical analysis

Both eosinophil counts and mast cell counts per 5 high power fields were analyzed using SPSS version 20. "t" test, and chi square tests were done to find out the differences between cases and controls. P value <0.05 was taken as significant.

Result

Total 69 patients (male 50, female 19) of functional dyspepsia (symptoms – postprandial fullness, bloating, and early satiety with normal upper GIT at endoscopy) were enrolled as cases. Age of them varied from 18 to 85 (mean 34.73 and SD 13.61). Forty-two controls undergoing upper GI endoscopy for symptoms other than dyspepsia age varying from 18 to 51 (mean 29.18 and SD 9.16) were taken as controls (Table 1). Duodenal eosinophil counts per 5 high power fields varied from 1-92 with mean 21.59 and SD 18.812 were found in cases. Eosinophil counts in duodenal mucosa in controls varied from 3-51 with mean 14.9048 and SD 10.74, and the difference was statistically significant (P=0.009. Eosinophil count was abnormally high among 29 (42.02%) and 08 (19.02%) among cases and controls respectively, and the difference was also significant (P=0.013).
On the other hand, mast cell density was seen among 56 cases and 23 controls from the same sample. Mast cell per 5 high power fields varied from 1-139 (mean 20.0536 and SD 24.28) and 3-57 (mean 20.86 and SD 16.44) were found in cases and controls groups respectively (Table 2) which was almost similar (P=0.627). Neither the cases nor the controls had the feature of celiac disease or parasite infestation in duodenal tissue.

![Hematoxylin and eosin staining for eosinophil](image)

**Figure 1:** Hematoxylin and eosin staining for eosinophil (Eosinophils are indicated by red arrows, Magnification, x 40).

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<th>Table 1: Eosinophil count in duodenum</th>
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*Functional dyspepsia patients

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<th>Table 2: Mast cell count in duodenum</th>
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* Functional dyspepsia patients

**Discussion**

The etiology and pathophysiology of FD are not completely established, and no single physiologic abnormality can be implicated as the cause of symptoms in every patient. Inflammation of the upper gastrointestinal tract has been implicated in the development of functional gastrointestinal disorders (Friesen et al., 2013; Collins, 1996). Among the inflammatory cells, mast cells and eosinophils are especially important (Friesen et al., 2013; Walker et al., 2011; Theoharides & Cochrane, 2004).
In this study, counts of eosinophil in the second part of duodenum in 69 patients and 42 controls and eosinophil counts were found significantly higher among cases. Recruitment and activation of eosinophils are usually accompanied by an inflammatory response that is initiated by a number of internal and external triggers (Thumshirn, 2002). The internal triggers, such as anxiety and stress, act via brain-gut axis and external triggers, such as microbes, allergens, stimulate inflammation (Tack et al., 2004). During inflammatory response, degranulation of eosinophils leads to neural stimulation and smooth muscle contraction, which consequently elicits gastrointestinal symptoms, including flatulence, cramps, and abdominal discomfort (Powell, 2010; Rothenberg, 2001).

Previous clinical studies have reported that children and adults with FD had eosinophilia in duodenum (Friesen et al., 2002; Walker et al., 2008). In our study, we found significantly high eosinophil count in the second part of duodenum, which is also consistent with the findings by Walker et al. (2011).

Our study does not show an increase in mast cell infiltration in duodenum which is consistent with the report by Walker et al. (2013) But Schurman et al. (2010) found increased mast cell count in duodenum in pediatric patients with FD. It is to mention that the number of control in case of mast count in our study was very small.

**Conclusion**

Functional dyspepsia is associated with a significant increase of eosinophil infiltration in duodenum. But the association of mast cell infiltration in the duodenum in FD could not be established in our study.

**References**


