

Mast Cells in Addition to Eosinophils Are Markedly Elevated in Patients with Eosinophilic Gastritis and/or Gastroenteritis

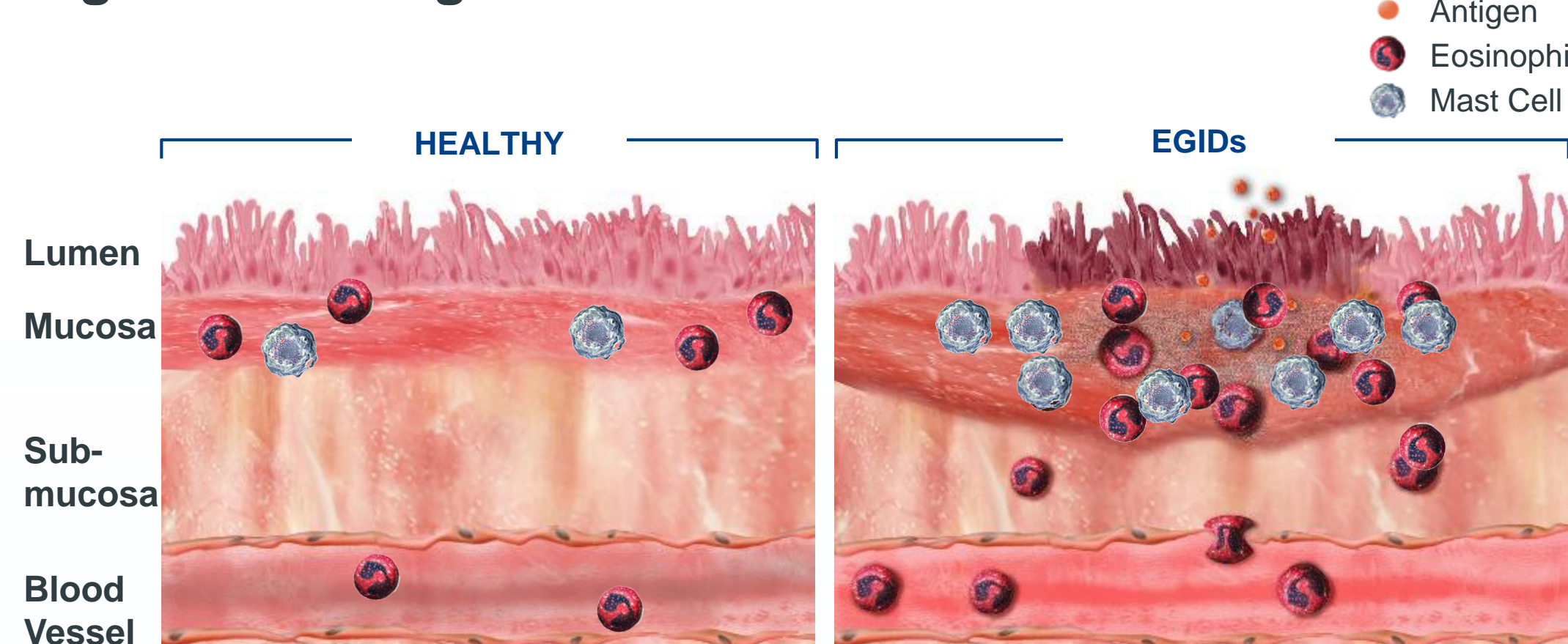
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BACKGROUND

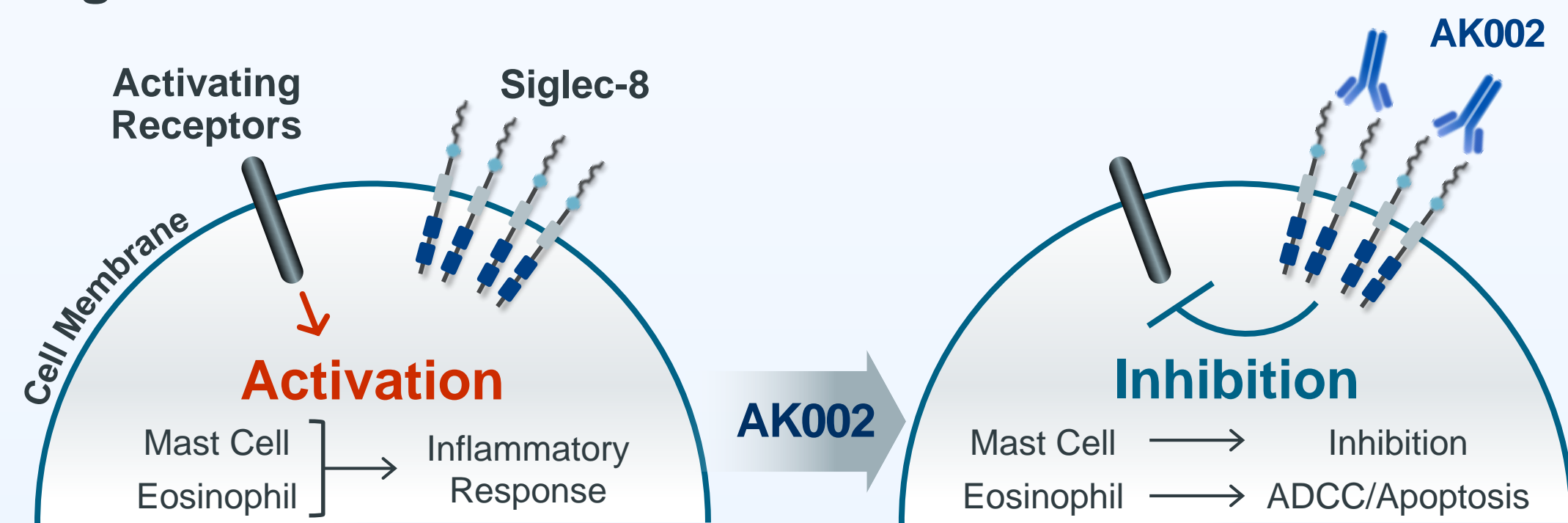
- Pathologic accumulation and over-activation of eosinophils are implicated in multiple chronic inflammatory diseases in the GI tract including eosinophilic esophagitis (EoE), gastritis (EG), gastroenteritis (EGE), and colitis - collectively termed eosinophilic gastrointestinal disorders (EGIDs)
- Patients with EGIDs have decreased quality of life due to debilitating symptoms such as dysphagia, abdominal pain, nausea, vomiting, and diarrhea
- While the pathogenesis of EGIDs has historically been thought to be driven by eosinophils, mast cells have also been shown to be elevated in EoE.^{1,2}
- The role of mast cells in other EGIDs, has yet to be established

Figure 1. Pathogenesis of EGIDs



- EG and EGE affect 45,000 - 50,000 patients in the US, though this number may be significantly underestimated³
- Current treatment options such as diet restriction and corticosteroids have limited efficacy and/or are inappropriate for chronic use
- There is a significant unmet need for novel therapies

Figure 2. AK002 Mechanism of Action



- Siglec-8 is an inhibitory receptor selectively expressed on human eosinophils and mast cells, and therefore represents a novel target for the treatment of EGIDs
- AK002 is a novel, humanized, non-fucosylated IgG1 monoclonal antibody to Siglec-8
- Engagement of Siglec-8 receptor by AK002 triggers:
 - Antibody dependent cell mediated cytotoxicity (ADCC) against eosinophils (blood)
 - Inhibition of mast cells and apoptosis of tissue eosinophils (tissue)
- ENIGMA, a Phase 2 multi-center, randomized, double-blind, placebo-controlled study of AK002, represents the largest clinical trial of patients with EG and EGE

OBJECTIVE

- To quantify the extent of eosinophil and mast cell infiltration in gastric and duodenal biopsies from symptomatic patients diagnosed with EG/EGE

METHODS

Figure 3. Screening Protocol

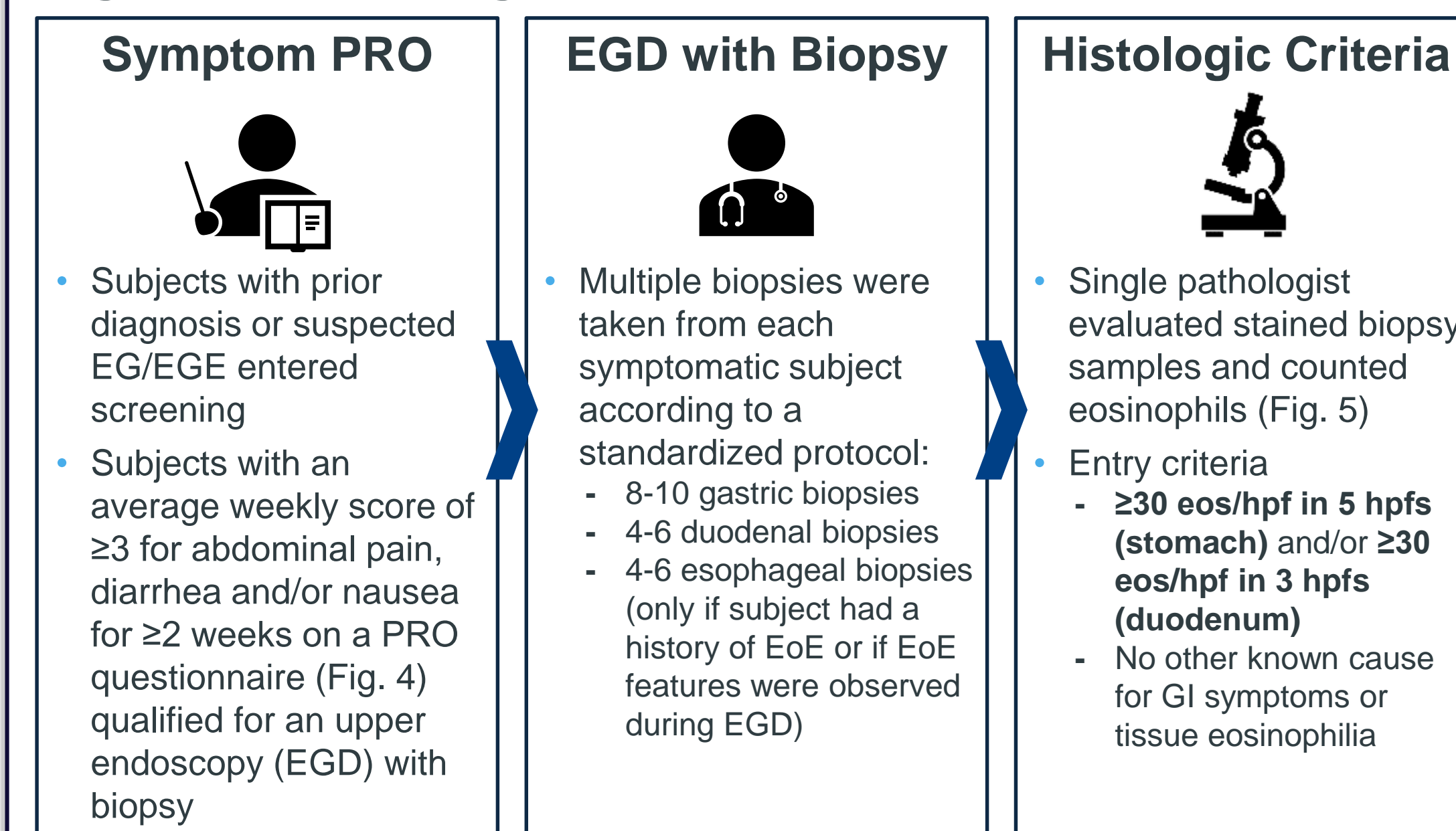


Figure 4. Daily Symptom Questionnaire

Instructions: Think of the last 24 hours and choose the number that best describes the worst experience of your EG/EGE symptoms during that time.

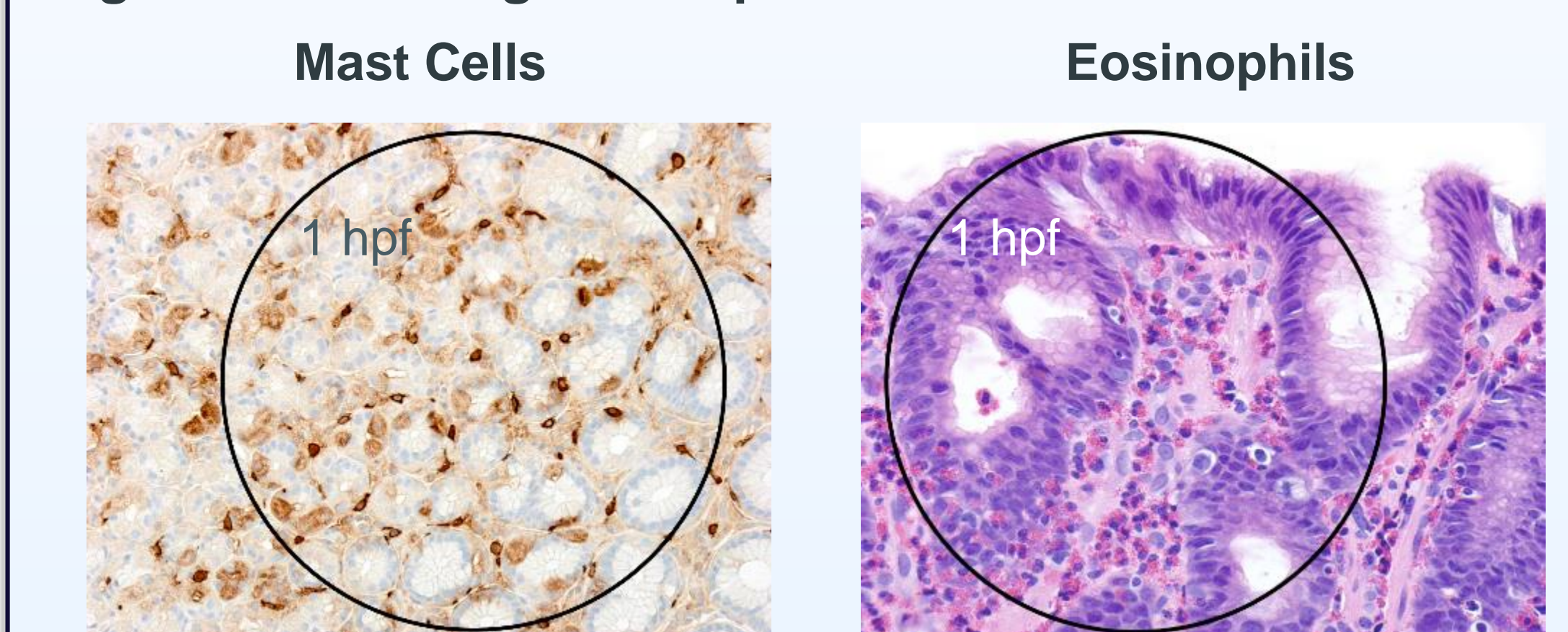
Over the past 24 hours, please rate the intensity of your

0	1	2	3	4	5	6	7	8	9	10
No Symptom										Worst Possible

- Abdominal pain
- Nausea
- Diarrhea
- Vomiting
- Fullness before finishing meal
- Loss of appetite
- Abdominal cramping
- Bloating
- Difficulty swallowing (EoE only)

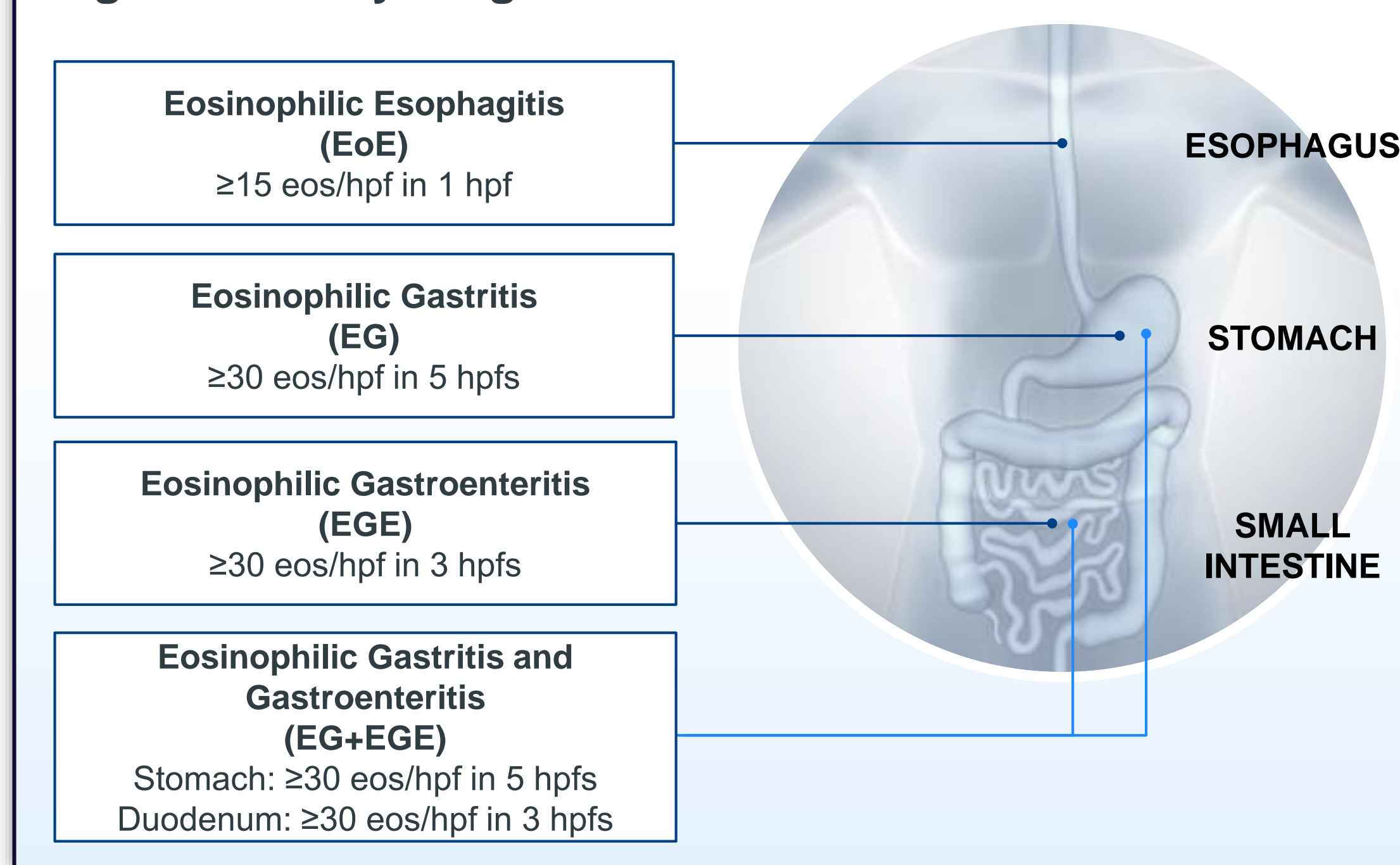
To qualify for a screening endoscopy and biopsy for this study, patients must have reported ≥3 average score for at least 2 weeks for abdominal pain and/or nausea and/or diarrhea

Figure 5. Counting Eosinophils and Mast Cells



- Eosinophils and mast cells were identified by H&E and immunohistochemistry (IHC), respectively. IHC was performed by staining with a mouse anti-human mast cell tryptase primary antibody (Clone AA1) followed by incubation with a peroxidase-labelled anti-mouse polymer secondary antibody and then counterstained with hematoxylin.
- Mast cells and eosinophils were counted in 5 separate non-overlapping high-power field (hpf; area of 0.237 mm²) for each biopsy sample
- The patients qualified for study if the average of the peak counts in 5 hpfs in the stomach (or 3 hpf in the duodenum) was ≥ 30 eos/hpf
- The patients were determined to have elevated mast cells if the average of the peak counts in 5 hpfs in the stomach (or 3 hpfs in the duodenum) was ≥30 mast cells/hpf

Figure 6. Study Diagnostic Criteria



RESULTS

Table 1. Baseline Characteristics

	EG (n=10)	EGE (n=25)	EG+E (n=30)	Enrolled (n=65)
Age, Median Years (Range)	39 (18-56)	40 (19-74)	41 (18-68)	40 (18-74)
Female, n (%)	7 (70%)	16 (64%)	17 (57%)	40 (62%)
Race - White, n (%)	10 (100%)	23 (92%)	27 (90%)	60 (92%)
History of Atopic/Allergic Disease ^a , n (%)	10 (100%)	17 (68%)	23 (77%)	50 (77%)
Peripheral Blood Eos (cells/μL), Mean	318	492	978	690

^a A Medical history at screening of asthma, rhinitis, food allergy, atopic dermatitis, seasonal allergy, environmental allergy, or pollen allergy

Figure 7. Distribution of Patients by Diagnosis

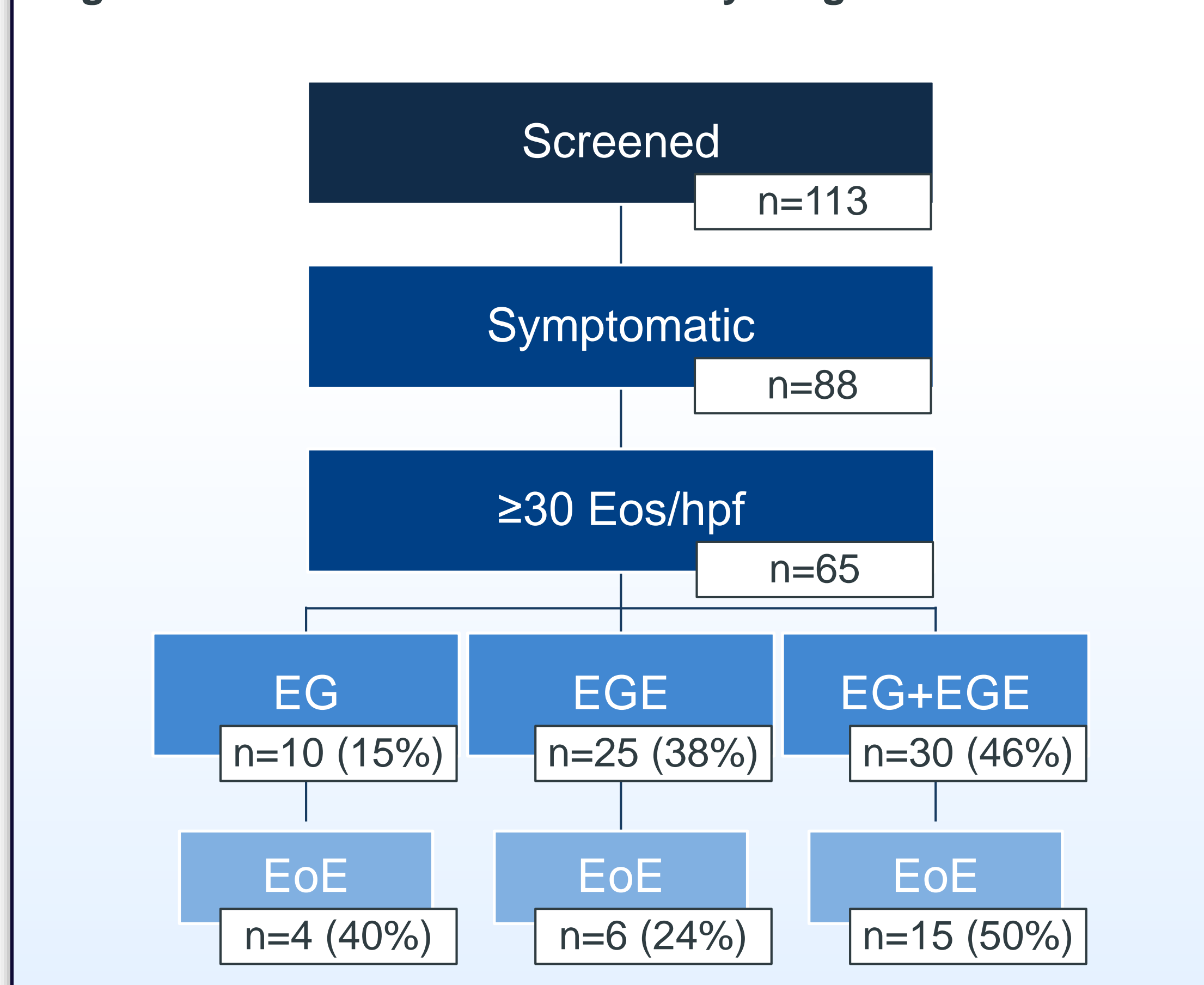


Figure 8. Eosinophils and Mast Cells Are Consistently Elevated in Gastric and Duodenal Biopsies

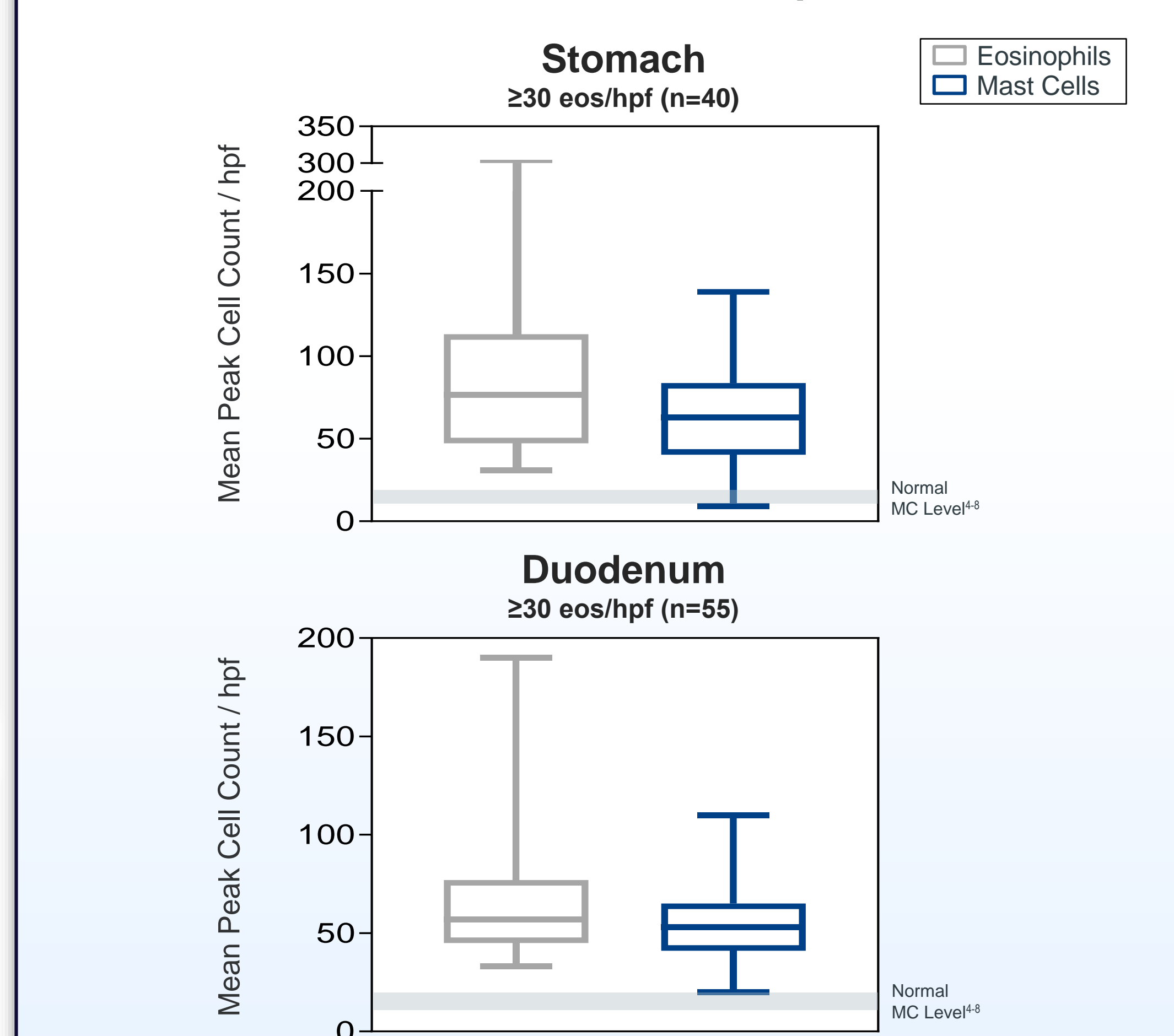


Figure 8. Data are plotted as box and whisker plots of peak mean cell count/HPF. The box plot represents the 25th and 75th percentiles, the internal horizontal line indicates the median and the T-bars indicate the minimum and maximum counts. The shaded gray bar indicates normal mast cell counts in the stomach & small intestine.⁴⁻⁸

Table 2. Tissue Eosinophil and Mast Cell Counts

	Stomach ^a (n=40)		Duodenum ^b (n=55)		Esophagus ^c (n=25)	
	Eos	MC	Eos	MC	Eos	MC
Mean	90	65	64	56	82	48
Median	77	63	57	53	87	45
SD	56	27	27	20	39	26

Eosinophils and mast cell counts quantified in (a) 5 peak hpfs (b) 3 peak hpfs (c) 1 peak hpf

CONCLUSIONS/DISCUSSION

- 65 patients with active EG, EGE, or EG+E were enrolled in this Phase 2 randomized controlled trial
- In addition to elevated tissue eosinophils, 64 of 65 (98%) EG/EGE patients also had elevated mast cell counts in gastric and/or duodenal tissue biopsies
- In patients with EoE, mast cells were also elevated in esophageal biopsies
- These data suggest the potential for a pathogenic role for both mast cells and eosinophils in EGIDs and that treatments for EGIDs may need to target both cell types for optimal effect

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References: 1) Caldwell et al. J Allergy Clin Immunol. 2014. 2) Youngblood et al. Gastroenterology. 2019. 3) Jensen ET, Martin CF, Kappelman MD, Dellon ES, J Pediatr Gastroenterol Nutr. 2016; Jan;62(1):36-42. 4) Hirano et al. Am J Surg Pathol. 2007; Nov;31(11):1668-76. 5) Tison et al. J Allergy Clin Immunol. 2010; Feb;125(2):481-87. 6) Walker et al. Aliment Pharmacol Ther. 2009; Apr 1;29(7):765-73. 7) Martinez et al. Gut. 2013; Aug;62(8):1160-8. 8) Doyle et al. Am J Surg Pathol. 2014; Jun;38(6):832-43.