COMPARISON OF THE EFFECT OF AZITHROMYCIN VERSUS ERYTHROMYCIN ON ANTRODUODENAL PRESSURE PROFILES OF PATIENTS WITH GASTROINTESTINAL DYSMOTILITY

By

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A THESIS PRESENTED TO THE GRADUATE SCHOOL
OF THE UNIVERSITY OF FLORIDA IN PARTIAL FULFILLMENT
OF THE REQUIREMENTS FOR THE DEGREE OF
MASTER OF SCIENCE

UNIVERSITY OF FLORIDA

2009

To Mehri & Massoud Moshiree and Nanneh,
and to all patients suffering from gastroparesis

ACKNOWLEDGMENTS

I would like to express my appreciation for the protected research time I have received as an assistant professor, from the Division of Gastroenterology and the Department of Medicine at the University of Florida to complete the requirements of this Master of Science. I am also especially grateful to the Chief of Gastroenterology, Christopher E. Forsmark, M.D., for sheltering me from the clinical load so that I can further pursue my research goals. Additionally, I thank Philip P. Toskes, M.D. for his selfless guidance, mentorship and enthusiasm for my career advancement and for this project. I am grateful also to Thomas Baumgartner, Pharm.D. for his unique idea regarding Azithromycin's lack of P450 interactions and Jane Y. Douglas Ph.D. for her eager help with the manuscript writing portion. I appreciate Marian Limacher, M.D. greatly for starting the Advanced Postgraduate Program in Clinical Investigation (APPCI) in the first place and nurturing Junior Faculty and fellows' research careers. Finally, I thank James I. Slaff, M.D. who was such a passionate physician scientist, friend, colleague and a true inspiration to anyone interested in academic medicine.

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LIST OF ABBREVIATIONS

ADM	Antroduodenal manometry
AF	Activity front
ANOVA	Analysis of Variance
AZI	Azithromycin
BL	Baseline
CYP3A	Cytochrome P450 3A
EES	Erythromycin
GERD	Gastrointestinal reflux disease
GES	Gastric emptying scintigraphy
GID	Gastrointestinal dysmotility
GP	Gastroparesis
IRB-01	Institutional Review Board-01
IBS	Irritable bowel syndrome
IV	Intravenous
MI	Motility index
MMC	Migratory motor complex
PSC	Primary sclerosing cholangitis
SEM	Standard error of the mean
SSRIs	Selective reuptake inhibitors
TdP	Torsades de pointes
UC	Ulcerative colitis

Abstract of Thesis Presented to the Graduate School
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August 2009

Chair: Marian Limacher
Major: Medical Sciences—Clinical and Translational Science

Current treatments for gastroparesis (GP) with prokinetic agents have been disappointing due to the limited options available. Erythromycin ethylsuccinate (EES) is a potent prokinetic agent that stimulates gastric emptying. Recently, EES has been possibly linked to the occurrences of sudden cardiac death due to prolongation of QT intervals thought to be related to its interaction with inhibitors of cytochrome P-450 3A isoenzymes. Azithromycin (AZI) is a semisynthetic macrolide similar in structure to EES but unlike EES it does not interact with the cytochrome P-450 isoenzymes.

The purpose of our study was to determine if AZI stimulates gastric antral activity and small bowel activity in patients with chronic gastrointestinal pain and refractory GP. In the first phase, small bowel manometric data on 30 patients undergoing clinical evaluation for chronic digestive problems or documented refractory GP were reviewed. For the second phase, another consecutive series of 21 patients with gastrointestinal dysmotility were prospectively studied. Antral and duodenal activity was measured after infusion of EES 250 mg IV and AZI (500 mg IV or 250 mg IV) given at different intervals during the small bowel manometry. The parameters

measured included the total duration of effect, mean amplitude of antral contractions, duration of the highest antral contraction phase, number of cycles per minute, and the motility index (MI).

The data were analyzed using repeated measures ANOVA for comparison of each medication. Comparison of EES and AZI showed that the mean amplitude and duration of antral activity, and the motility index were significantly increased with AZI ($p = 0.006$, $p = 0.03$, and $p = 0.007$, respectively as compared to EES). Overall comparison of AZI and EES in the duodenum showed that AZI induced more migratory motor complexes in the duodenum with origin of activity fronts in the antrum than did EES (p value = 0.03). We conclude that AZI stimulates antral activity similar to EES and moreover has a longer duration of effect and may even promote small bowel motility. However, AZI unlike EES does not have significant drug-drug interactions and may be a potential new and safer treatment for gastrointestinal dysmotility.

CHAPTER 1
INTRODUCTION

Gastroparesis (GP) is a debilitating chronic gastrointestinal motility disorder resulting from impaired transit of intraluminal gastric contents from the stomach into the duodenum associated with delayed gastric emptying in the absence of any mechanical outlet obstruction. Symptoms of GP are variable but include early satiety, nausea, vomiting, epigastric abdominal pain, weight loss and bloating. The prevalence of GP is very difficult to estimate due to the incomplete correlation of symptoms with gastric emptying, the large number of misdiagnosed patients who actually have this disorder, and the lack of widely available and standardized diagnostic tests or treatments. Although the true prevalence of the disorder is unknown, an estimated one third of diabetic patients in tertiary care settings have abnormal gastric emptying studies (4). In addition, up to 4% of the adult population suffers with symptoms of this condition. Furthermore as many as 25-40% of adults and children initially diagnosed with dyspepsia are subsequently properly diagnosed with GP (4). Such misdiagnosis can result in significant healthcare costs due to work days lost and prolonged hospitalizations (5, 6). Our extensive clinical experience with abdominal pain in chronic pancreatitis indicates that a remarkably large number of patients initially diagnosed with chronic pancreatitis in fact have gastroparesis and/or small bowel dysmotility which responds to prokinetic therapy (7).

Diagnosis of Gastroparesis

After demonstrating no evidence of obstruction and a normal upper endoscopy, the diagnosis of GP is usually made by finding a delayed gastric emptying time using gastric emptying scintigraphy (GES). This method is regarded as the gold standard for the diagnosis of GP. Another sensitive indicator of gastrointestinal dysmotility (GID), however, is a less commonly used method, antroduodenal manometry (ADM). Currently in the U.S., ADM is

available in only a handful of tertiary referral centers with expertise in treating patients with GID. Antral hypomotility diagnosed using ADM correlates well with gastric stasis found using GES showing a prolongation of solid lag time and slower emptying rates of solids as compared with controls (8). However, ADM is more invasive and far less available a diagnostic tool than GES. Regardless of the method used for diagnosis of GP however, once the diagnosis is made, treatment is based on the underlying etiology. For example, in the diabetic patient, correction of the glycemic control is a pivotal part of management and prokinetics such as EES are not effective unless hyperglycemia is first corrected (9,10).

Pathophysiology

The etiology of gastroparesis in adults is multifactorial. The three most common etiologies are diabetes, idiopathic, and post-surgical, especially if surgery causes damage to the vagus nerve which is involved in inducing gastrointestinal motility. Other causes are certain medications, collagen vascular disorders, thyroid dysfunction, liver disease and cirrhosis, chronic renal insufficiency, Parkinson's disease, intestinal pseudo-obstruction and other miscellaneous causes (4).

The slow waves responsible for gastric motility originate in the region of the interstitial cells of Cajal (ICCs) and are responsible for initiating the electrical impulses across the stomach and toward the pylorus. These slow waves do not directly result in contraction of the gastric smooth muscle but instead they cause a simultaneous release of neurotransmitters from the enteric nerve endings, leading to smooth muscle contraction (11). Although the complete neuro-humoral control of gastric emptying is incompletely understood, both motilin and ghrelin are peptides secreted by the gastrointestinal endocrine cells that have been shown to increase gastric motor function (12, 13). This mechanism is thought to be responsible for EES's action on gastric motility (14-17).

In general, several factors affect gastric motility. These include motor dysfunction such as pyloric spasm, sensory dysfunction (such as impaired fundic relaxation, accommodation and abnormal sensation), electrical dysfunction (such as gastric arrhythmias and abnormal propagation), central nervous system effects resulting in nausea and vomiting, and others such as bacterial overgrowth in patients with GID (18). But, in most patients, including those with diabetes mellitus, treatment is still challenging due to the lack of available effective treatment options in the U.S. and abroad and the many side effects of the prokinetics currently available.

Treatments

Currently, the most potent treatment for GP is erythromycin (EES) which accelerates gastric emptying and promotes the dumping of food and nondigestible material from the stomach into the duodenum (14-16). The prokinetic effects of EES have been well documented in several studies done in both humans and dogs and are thought to occur through its effects as an agonist on smooth muscle motilin receptors (19-22). However, studies have failed to conclusively establish that the antral contractions EES triggers are solely mediated by release of motilin (17). Presently, treatment of GP has focused on the acceleration of gastric emptying to relieve symptoms of nausea, vomiting and abdominal pain. Furthermore, EES has been shown to be better than other commonly used prokinetics such as metoclopramide, cisapride and domperidone with EES resulting in as much as 30-60% improvement in gastric emptying (23).

Erythromycin and Cardiac Risk

For all of its benefits, EES still has its drawbacks. First, several reports of arrhythmias associated with use of either oral or intravenous (IV) EES have been published (1, 24, 25). Second, in 2004, a large cohort study of Tennessee Medicaid patients from 1988 to 1993 showed a possible association between EES with risk of sudden cardiac death through its extensive metabolism by the cytochrome P-450 3A isoenzymes (CYP3A) (1). In this study, the adjusted

rate of sudden death from cardiac causes was five times higher in patients on EES than in patients on other antibiotics such as amoxicillin. The incidence rate ratio was 5.35 (95% CI 1.72-16.64; p=0.004) among those patients on EES and concurrently on CYP3A inhibitors such as calcium channel blockers, statin drugs, and Selective Serotonin Reuptake Inhibitors (SSRIs). Furthermore, of all the macrolides, erythromycin has the greatest arrhythmogenic liability while azithromycin (AZI) has the least (24, 25).

Rationale for Using Azithromycin

Given these potentially problematic drug-drug interactions with use of EES, we sought to study another macrolide, AZI, for use in patients with GP given its lack of inhibition of the cytochrome isoenzymes as compared with other macrolides (26). Initially approved by the FDA for use as an antibiotic in November, 1991, AZI is now available even in generic form. In a single blinded, placebo-controlled manometric study of eleven healthy patients comparing oral AZI, midecamycin acetate to placebo, Sifrim et al. found that oral AZI significantly increased the postprandial antral motility index as compared with placebo ($p < .05$). (27). Furthermore, a recent case report using AZI in an elderly patient with diabetic GP showed symptom improvement after a three day treatment with IV AZI (28). Nevertheless, no data exist demonstrating the effectiveness of AZI in patients with GP confirmed by either ADM or GES in comparison to EES. Finally, although EES at high doses does not induce migratory motor complexes (MMC) in the small bowel, we do not yet know the effect of AZI on small bowel activity in patients with documented small bowel dysmotility (29, 30).

Study Aims

As a result, we sought out to study the effect of AZI in comparison to EES on ADM parameters of patients with GID as diagnosed by ADM. The aim of our present study, therefore, was to determine whether AZI stimulates antral activity similar to EES and if AZI's

effectiveness can be demonstrated using previously defined ADM parameters. We hypothesized that AZI will stimulate antral activity similar to EES given that both medications are motilin agonists and should then act similarly to promote antral activity. A secondary aim of this study was to investigate the effect of AZI as compared to EES on duodenal activity in patients with gastrointestinal dysmotility (GID).

CHAPTER 2
RESEARCH METHODS AND DESIGN

Study Design

This study is a case-series analysis of all patients who underwent ADM testing with provocative testing in the Clinical Motility Laboratory at the University of Florida from 2005-2007. In the first phase of the study, ADM data on 30 consecutive patients who were evaluated for either refractory GP or chronic functional abdominal pain were analyzed. In the second phase of the study, ADM data on 21 consecutive patients with GID were analyzed using similar parameters to compare the effect of AZI and EES on duodenal motility indices. The research protocol was approved by the University of Florida Health Science Center Institutional Review Board (IRB-01 Project #: 552-2004) on November 7, 2004.

Patient Selection

Thirty consecutive patients (mean age 50 ± 14 years, age range 24-75) with suspected functional or organic dysmotility had ADM performed routinely as part of their medical workup to clarify the physiology for their continued abdominal pain and to rule out an underlying gastrointestinal motility disorder. Symptoms on presentation included chronic abdominal pain in 23, abdominal distension in 3, nausea and vomiting in 13, weight loss in 4, constipation in 3, and diarrhea in 2 patients. After review of each patient's hospital records, we included 30 patients in the study. Prior surgeries in these patients included liver transplantation in one patient with ulcerative colitis (UC) and primary sclerosing cholangitis (PSC) with normal hepatic function, umbilical hernia surgery, and cholecystectomy. All patients had a normal physical examination with exclusion of any obstructive symptoms. Other patient diagnoses for those included in the study at the time of presentation for the ADM were as follows: Gastrointestinal Reflux Disease (GERD) in 1, Sphincter of Oddi Dysfunction in 1, diabetes mellitus in 1, Crohn's disease in 1,

UC in 1, chronic cholecystitis in one who had cholecystectomy, chronic idiopathic intestinal pseudo obstruction in 2, post-trauma in 2, hypothyroidism in 3, small bowel bacterial overgrowth in 3, chronic pancreatitis in 4, and irritable bowel syndrome (IBS) in 5. Twelve of the thirty patients who underwent GES testing, were diagnosed with GP based on GES studies showing a half-time (t ½) > 90 minutes. A normal gastric emptying study was defined as an emptying t ½ of 45-90 minutes. This study excluded patients with a prior history of obstruction, psychiatric illness, eating disorders, malignancy, and those with allergies to any macrolide antibiotics. Patients with systemic diseases other than diabetes and collagen vascular diseases were also excluded. None of the patients was on any chronic narcotic analgesia for at least three days prior to the study.

In the second phase of the study, we studied the effect of AZI and EES on duodenal motility in 21 consecutive patients referred to our Clinical Motility Lab. These patients were undergoing ADM for evaluation of chronic abdominal pain and found to have small bowel dysmotility by ADM criteria (31). Only those patients with GP *and* small bowel dysmotility were included. The same criterion for diagnosis of GP applied to these patients.

Manometric Technique

Thirty patients underwent ADM at the University of Florida for complaints of chronic digestive problems from 2005 to 2007. Since no standardized protocol for performing ADM exists, we describe here our protocol here at the University of Florida.

A solid state six-channel manometric microtransducer catheter (Unisensor™ USA Inc., Hampton, New Hampshire, Type: K10600-L5-0353-D) was placed intranasally under fluoroscopic guidance and advanced through the pylorus into the duodenum and positioned so that the distal tip was seen at the ligament of Treitz (30). Our recording started in the morning at 7:30 a.m. routinely after an overnight fast that started at 10:00 p.m. the night before the patients

arrived to the motility laboratory and was stopped after 23-24 hrs. Patients were required to stop all medications that can affect gastrointestinal motility 72 hours prior to the study. The manometric probe was calibrated just before the study with external application of atmospheric zero and 50 mm Hg by means of water column and was then passed under fluoroscopy. The calibration device was used to adjust and check all the sensors of the pressure transducer catheter. The pressure sensors were then adjusted to an accuracy of the sensitivity of ± 2% at 100 mmHg. The catheter used had a 7mm diameter with three distal transducers placed 5 cm apart in the duodenum and three proximal transducers 10 cm in the distal antrum for a total of six pressure transducers. A portable digital pressure recorder was used to record the antroduodenal pressures seen onto an MS-DOS server for later retrieval and as an online display. An event keyboard was also used to record patient symptoms, time for sleep, meal times, and medications administered (EES and AZI). Once the catheter was positioned, the interdigestive and digestive motor activity in the distal antrum was analyzed with the proximal three probes located in the stomach and the next three in the duodenum. The catheter was then secured in place with tape at patient's nose.

Data Analysis

These data were then analyzed retrospectively using the GastroTrac™ 2007 computer software and by visual inspection. For analytical purposes, the total recording time was divided into three periods: a fasting (interdigestive) period of 5-6 hours, followed by a therapeutic portion including a 2 hour fed (digestive) period during which patients were given a standard meal consisting of two cans of Two Cal-HN each 470 calories, and a postprandial (interdigestive) period during which an intravenous (IV) injection of EES then AZI was given. In this postprandial period after the two hour fed state, patients were first given an IV injection of Erythromycin® Lactobionate over 20 minutes. After 4 hours of observation, patients were then

given an IV injection of AZI infused over 30 minutes with another four hours for observation. The motility recording was continued for 4 hours after EES infusion to allow for enough time for washout of EES which has a hepatic excretion time of 1.5 hours. The two medications were therefore given with the same ordering for each patient and sequentially after 4 hours. Characterization of the different phases of the MMC was done according to accepted criteria (31). Although all 30 patients received EES, 15 patients were given 500 mg of IV AZI (Group AZI-A) and the other 15 patients received 250 mg IV of AZI (Group AZI-B). For the 21 patients with small bowel dysmotility and GP in the second portion of the study, all patient received AZI and EES at a dose of 250 mg IV.

In each patient, the pressure activity was analyzed for the amplitude, frequency of antral pressure activity and characteristics of the activity front (AF) of the MMC (32). The ADM reading for all 30 patients was consistent with postprandial antral hypomotility as per the simplified criteria by Thumshirn et al., defining an average of less than one contraction per minute postprandially as significant hypomotility (33). Nineteen of these patients had only decreased antral activity during the postprandial state, 10 had antral hypomotility and small bowel dysmotility, and 1 patient had low amplitude AF's of the MMC with antral and small bowel hypomotility consistent with myopathy. All 21 patients in the second portion of the study had GP and small bowel dysmotility. Antral and duodenal motility before and after each medication was analyzed with medication-induced AFs compared for each medication.

Measurements

The recording software used to analyze the test results was Gastrorac™2007 version: 4.3.0.47 from Alpine Biomedical APS Medical Devices. Measurements taken included the antral activity after postprandial infusion of EES 250 mg IV and AZI either 500 mg IV (AZI-A, n=15) or 250 mg IV (AZI-B, n=15) at different intervals during the ADM. The fasting state

(interdigestive phase) was used as a guide to identify minute by minute the most distal antral recording site that recorded up to 3 waves per minute (antral waves). The number of contractions for each recording site was also determined visually which allowed identification and characterization of the different phases of the MMC according to accepted criteria (32-34). Contractions in the antrum were defined by changes in amplitude from baseline of more than 10mmHg (1.3kPa) and durations of more than one second. Then the amplitude and duration of the contractions were analyzed using GastroTrac™ software and by visual inspection. Cluster activity was defined as a sequence containing a burst of contractions seen in at least two sensors followed by a period of motor quiescence lasting at least 15 seconds. Once these were identified, our data analysis focused on the most distal antral lead. The postprandial time was based on timing after meal and not the phases of MMC.

Phase One Measurements

Antral contractions with high amplitude pressure waves of frequencies of 0-4 cycles per min were identified (35, 36). The amplitude and duration of contractile waves were analyzed and mean values over a four hour period each after administration of EES or AZI were calculated. This analysis excluded any giant retrograde contractions. Finally, we compared AZI to EES with respect to the following parameters measured: the total duration of antral contractions (min) after administration of the drug (either EES or AZI), the mean amplitude of antral contraction (Kpa), duration of the highest AF in the antrum (minutes), number of cycles per min for each AF and the motility index (MI) defined as log {Σ (Ampl X number of contractions) + 1} (30). The total value for each parameter is the sum of all the contractions seen during the 4 hour postprandial period after administration of each medication.

Phase Two Measurements

For the 21 patients with both GP and small bowel dysmotility, pressure profiles were recorded similarly to that described above in three stages: baseline period (BL), fed state after the standardized meal, and postprandial after the administration of EES (250 mg IV) and AZI (250 mg IV). Only patients with known gastroparesis (diagnosed by gastric emptying Scintigraphy with t ½ > 90 minutes) and small bowel dysmotility (antroduodenal manometric criteria of < 3 MMC's in a 6 hour BL, MMC amplitudes < 5 mm Hg) were included in this portion of the study. The interdigestive phases (baseline phase) was defined as follows: 1. Phase I: motor quiescence beginning after phase III ends, 2. Phase II: pressure waves of > 2 κPa occurring at a rate higher than the two per 10 min and less than the maximum frequency of the duodenum (10-12 contractions/min), and 3. Phase III: rhythmic contractile activity which begins in the antrum for at least a min with three contractions/min and duodenum (10-12 contractions/min) for at least 2 min. Phase III activity must be seen in at least 2 of the recording sites and followed by Phase I activity. The measured parameters included the characteristics of the MMCs and AFs such as site of origin (antrum versus duodenum), duration of phase III activity, amplitude of phase III MMCs in duodenum and number of cycles per minute. A manometry reading depicting phase III activity after administration of AZI is shown in Figure 2-1.

Statistical Analysis

The manometric data obtained during the postprandial period were compared using repeated measures analysis of variance (ANOVA) with the SAS Software Version 9.1 (SAS institute, Cary, NC). The pressure tracings were reviewed on the GastroTrac™ software and analyzed visually with a print-out of the entire recording. All values are expressed as mean ± standard error of the mean (SEM). Comparisons of the two drugs, EES and AZI were completed using this method.

Antral Activity Front (AF)

MMCs in duodenum

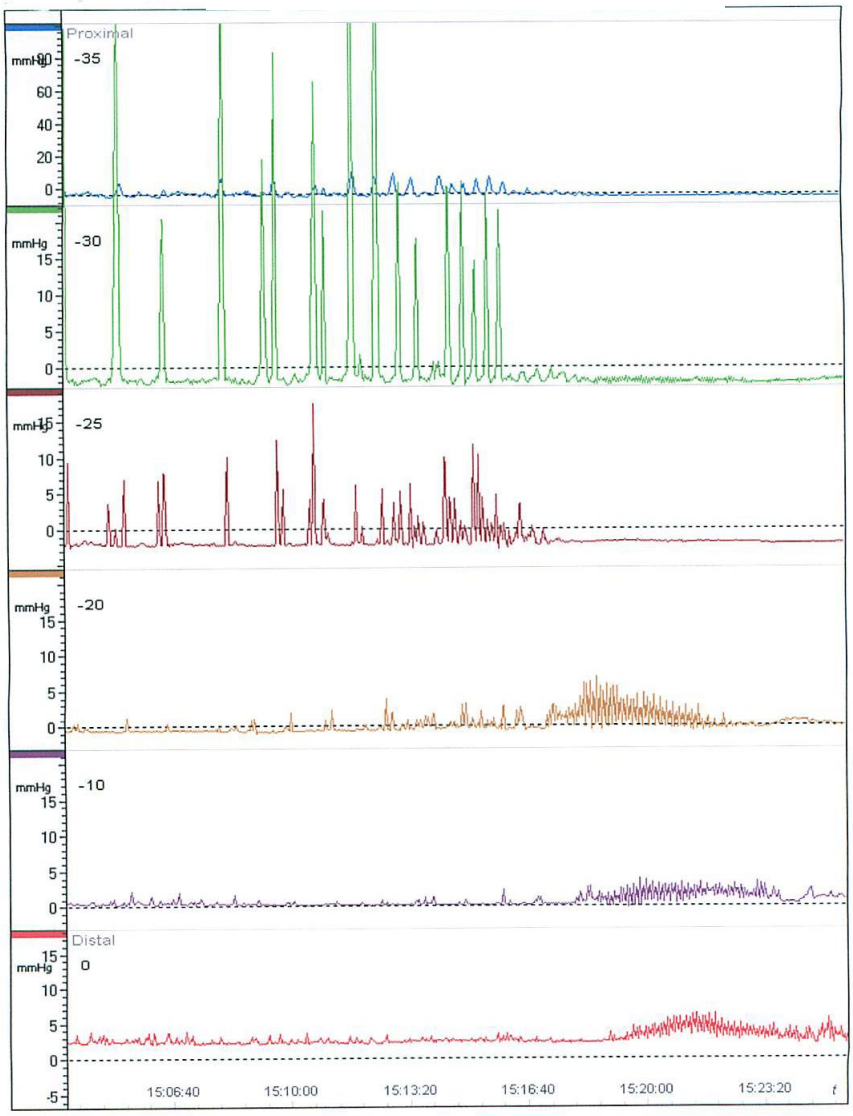

Figure 2-1. Antroduodenal manometry tracing after administration of Azithromycin (AZI). The tracing shows a phase III contraction with activity front (AF) starting in the antrum (first three channels) and migratory motor complex (MMC) in the duodenum (last three channels).

CHAPTER 3
RESULTS

The AZI-1 group included 15 patients given 500 mg of AZI and the AZI-2 group included patients given 250 mg IV of AZI. All patients were given EES. These two medications and the different doses of AZI given were then compared with respect to different manometric parameters (Table 3-1). The most prominent change in motility pattern observed after infusion of EES or AZI was the high-amplitude antral contractions seen in the first 30 min after the start of infusions. Antral contractions were seen for an average duration of 1 hour and 44 minutes after administration of EES and for 2 hours 22 minutes on average after administration of either dose of AZI. MMC-like AF's were induced by both AZI and EES.

Phase One Results

Repeated measures ANOVA showed no difference between EES and AZI-1 with respect to the total duration of antral contractions ($p = 0.11$) or the number of cycles per min of antral contractions seen ($p = 0.31$). Furthermore, when EES was compared to the lower dose of AZI (AZI-2), no statistically significant difference was seen in any of the parameters measured.

However, comparison of EES to AZI-1 showed an increased mean amplitude of antral contractions with AZI compared to EES during the postprandial period (112.4 ± 55.4 versus 87.4 ± 58.1, $p = 0.006$), duration of highest contractions (25 ± 24 versus 16.5 ± 13.4, $p = .03$), and MI (4.9 ± 0.9 versus 4.4 ± 1, $p = 0.007$) (Figures 3-1, 3-2, 3-3). When comparing the two doses of AZI (250 mg and 500 mg IV), only the duration of highest antral activity was statistically significant.

The most significant results were seen with the 500 mg AZI dose with no differences seen between the lower dose of AZI used. No statistically significant difference was seen between EES and AZI (any group) with respect to the number of cycles per minute visualized.

Overall for the two medications given, AZI induced a higher mean value of antral contractions (p = 0.0043) and higher total duration of antral contractions than EES (p = 0.05).

Phase Two Results

For the second phase of the study, comparison of AZI with EES was done in patients with both GP and small bowel dysmotility with duodenal motility indices compared with baseline (BL) interdigestive phase (Table 3-2). Overall comparison of AZI and EES showed that AZI induced more MMCs in the duodenum with origin of AF's in the antrum than did EES (18 patients with AZI versus 10 patients with EES, p value = 0.03). The average duration of MMCs and antral AFs were also longer in AZI group as compared with EES (AZI mean = 18.5 min, EES mean = 9.7 min, SEM 3.6 min, p value = 0.0172) (Figure 3-4). However, this result was only statistically significant with respect to antral AF's and not the individual duration of MMCs. Further comparison of pressure profiles seen at BL in the duodenum with AZI given in the postprandial state showed statistically significant improvement in the number of MMCs (BL mean = 1.2, AZI mean = 2.4, SEM = 0.42, p value = 0.0024) (Figure 3-5) and number of AFs in the antrum (p value = 0.0256). No significant difference between AZI and BL fasting state was seen with respect to the duration of MMCs or number of cycles per minute.

Table 3-1. Effect of Erythromycin (EES) and Azithromycin (AZI) on postprandial antral motility

Antroduodenal measures	EES (n= 30)	AZI-1 (n=15)	AZI-2 (n=15)
Mean amplitude of antral contractions (mm Hg)	87.4 ± 58.1	112.4 ± 55.4* p = .006	99.7 ± 50.1
Duration of highest antral contraction (min)	16.5 ± 13.4	25.0 ± 24.2* p = 0.03	15.6 ± 15.6
Total duration of antral contractions (min)	104.7 ± 58.0	136.2 ± 79.5* p = 0.05	148.1 ± 65.3* p = 0.05
Number of cycles/min (no/min)	2.1 ± 2.3	2.19 ± 2.0	1.7 ± .9
Motility index log (mm Hg·no)	4.4 ± 1.0	4.9 ± 0.9* p = 0.007	4.3 ± 1.0

The parameters assessed include mean amplitude of contractions, duration of highest contraction, total duration of antral contractions, number of cycles per minute and motility index. Data reported as means ± standard deviations. AZI-1 group was given 500 mg IV AZI and AZI-2 group received 250 mg IV AZI.
P values are given for each parameter analyzed.
*Denotes statistical significance with p values given for comparison of EES to AZI.

Table 3-2. Effect of Erythromycin (EES) and Azithromycin (AZI) on postprandial antroduodenal motility. Baseline (BL) is the pre-prandial fasting state prior to administration of any medications. Recorded are numbers of Migratory Motor Complexes (MMCs) and Activity Fronts (AFs) in the duodenum.

Antroduodenal measurements (mean ± SEM)	BL	EES	AZI
Avg # MMCs	1.2 ± .83	1.0 ± 1.18	2.4 ± 1.7
Avg # AFs	0.9 ± 0.77	0.9 ± 1.1	1.8 ± 1.5
MMC duration (min)	7.5 ± 4.6	5.5 ± 6.0	8.4 ± 6.0
AF duration (min)	16.9 ± 16.5	9.7 ± 11.7	18.5 ± 11.5
MMC mean amplitude (mmHg)	17.1 ± 10.6	12.9 ± 15.1	18.3 ± 13.0
MMC cycles per minute (no/min)	3.1 ± 1.9	1.5 ± 1.8	3.1 ± 1.7

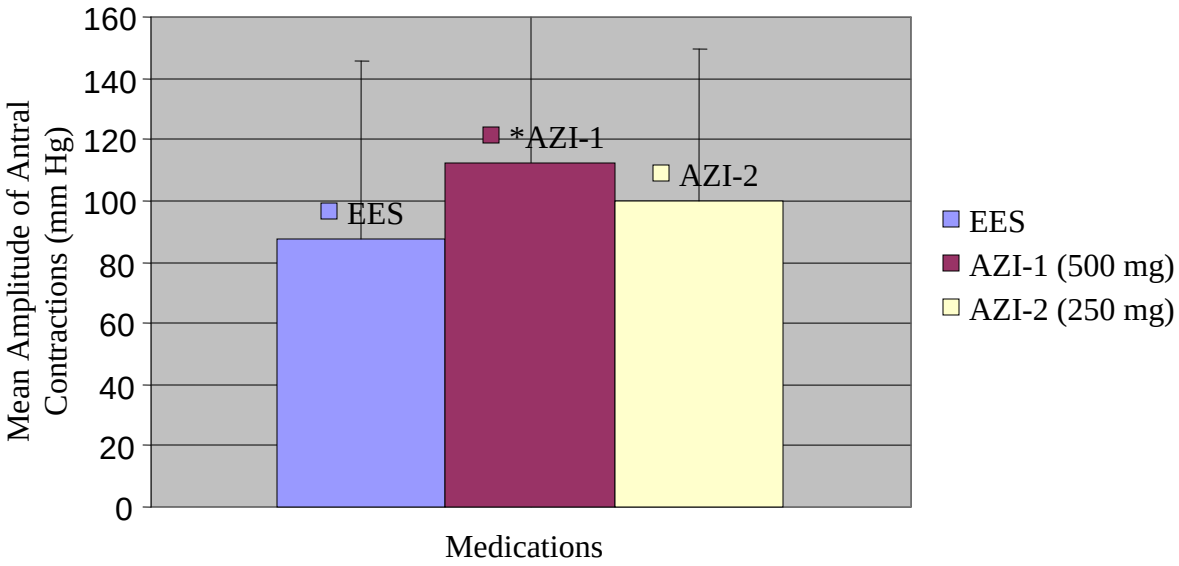

Figure 3-1. Comparison of erythromycin (EES) and azithromycin (AZI) with respect to mean amplitudes of antral contraction. * Denotes statistical significance with p values given for comparison of EES to AZI.

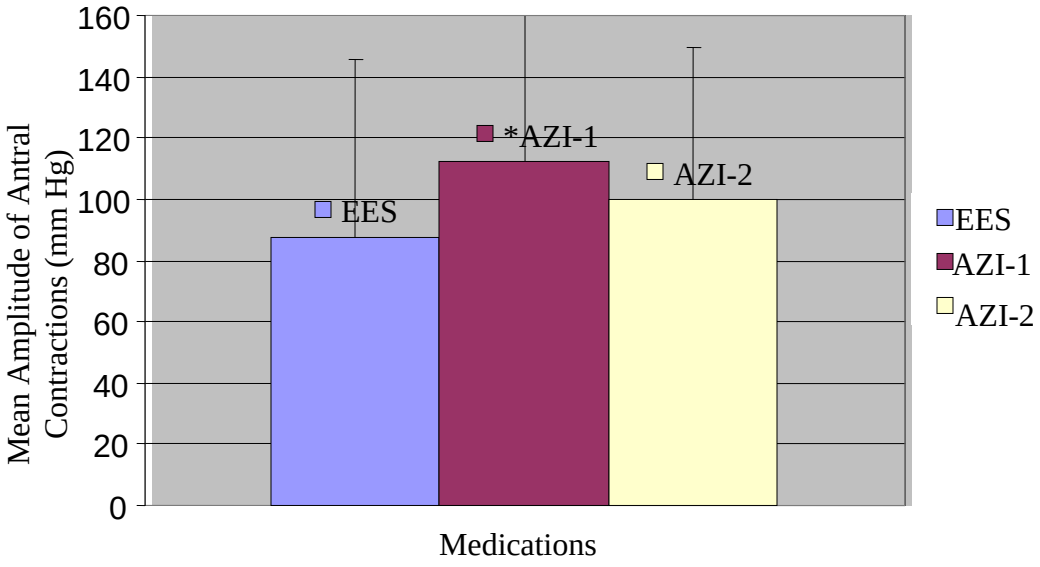

Figure 3-2. Comparison of duration of highest antral contractions in the postprandial period during antroduodenal manometry (ADM). * Denotes statistical significance with p values given for comparison of EES to AZI.

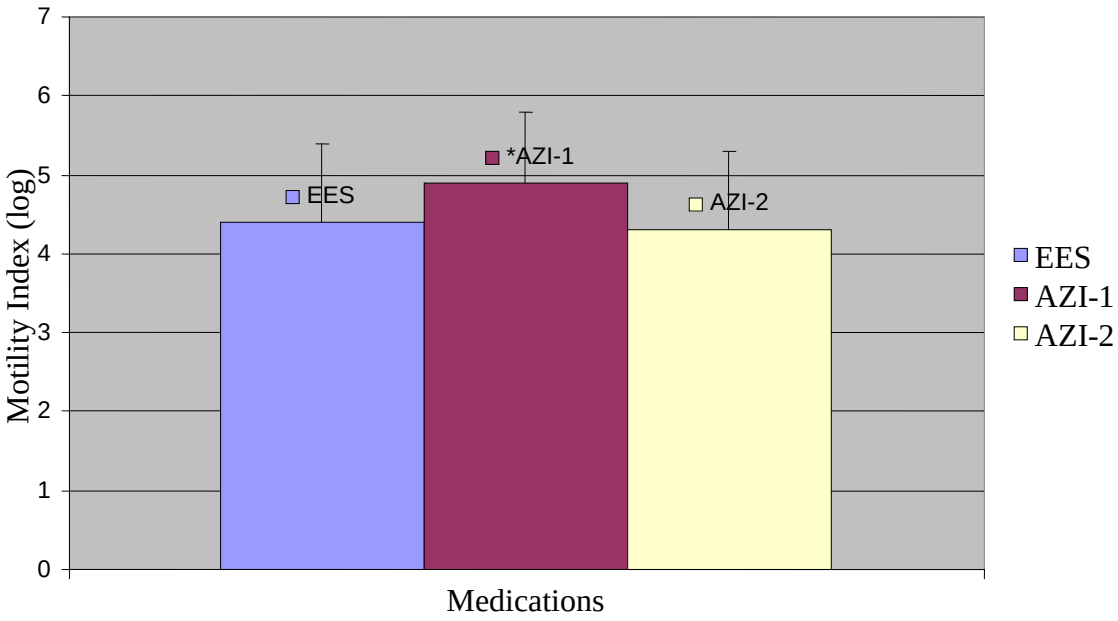

Figure 3-3. Comparison of the motility index for each prokinetic during Antroduodenal Manometry (ADM). * Denotes statistical significance with p values given for comparison of EES to AZI.

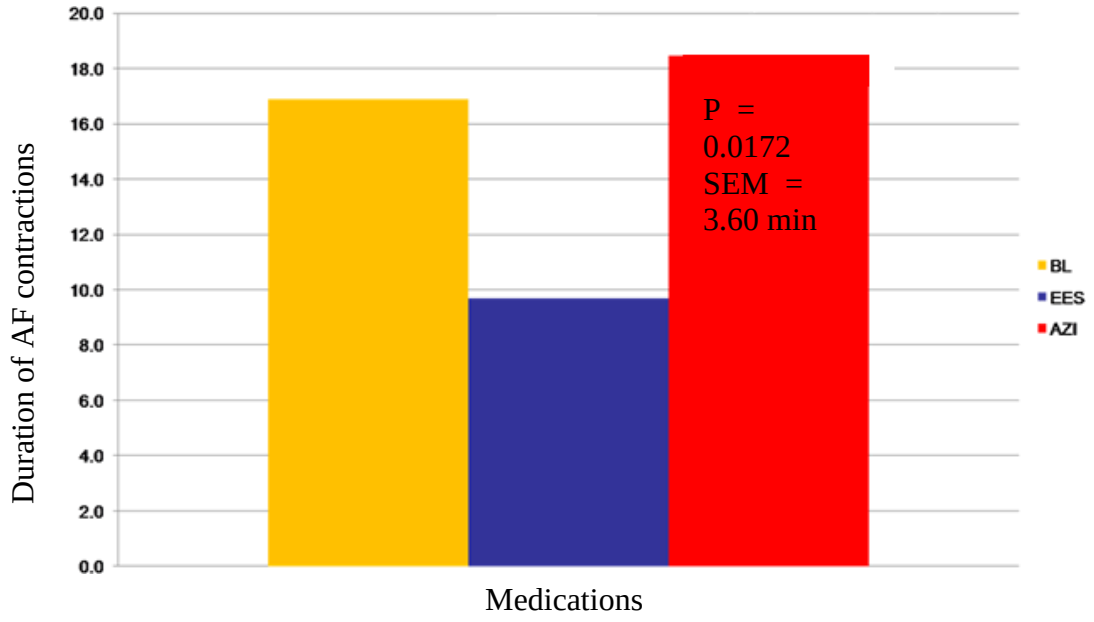

Figure 3-4. Duration of activity fronts (AFs) and migratory motor complexes (MMCs) in the duodenum at baseline and after administration of Erythromycin (EES) and Azithromycin (AZI) in minutes. P value listed is for comparison of AZI and EES with Standard Error of the Mean (SEM).

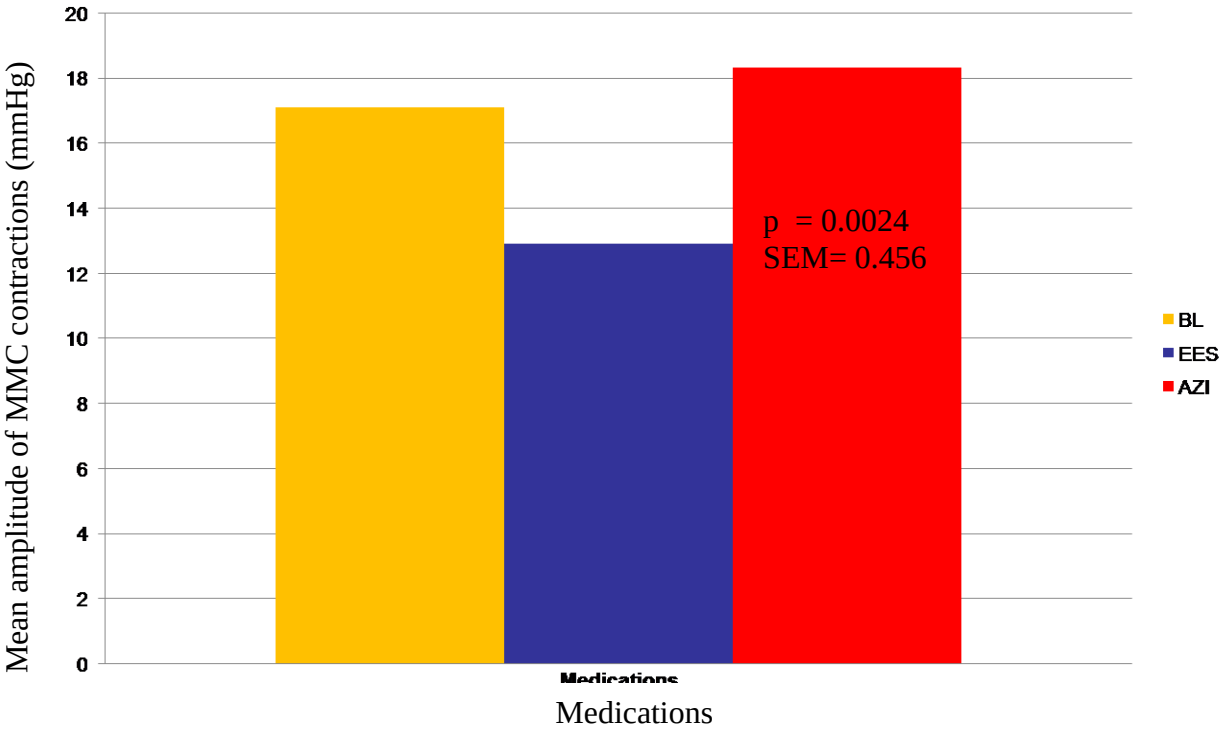

Figure 3-5. Mean amplitude of migratory motor complexes in the duodenum at baseline and after administration of Erythromycin (EES) and Azithromycin (AZI) in mm Hg. P values listed for comparison of EES to AZI, and Standard Error of the Mean (SEM).

CHAPTER 4
DISCUSSION

Our findings here suggest that AZI could be used as an alternative prokinetic for the treatment of GID and GP. The higher dose of AZI created a statistically significant difference in the MI, the duration of highest antral contractions, and the mean amplitude of antral contractions seen in the postprandial period on ADM. Moreover, even the lower dose of AZI was as effective as EES in stimulating antral contractions thus supporting the potential use of AZI instead of EES – the current most potent prokinetic – for treatment of GP. Ultimately, our results support human studies done previously by Sifrim et al. that show AZI may act on motilin similar to EES in promoting antral contractile activity (27).

In this study the effects of IV EES on postprandial ADM were compared to IV AZI- a 15-membered semi-synthetic macrolide known to have both less gastrointestinal side effects and less cardiac risk than EES. These data were obtained using prolonged ambulatory ADM in patients found to have gastric dysmotility defined by antral hypomotility. The comparison was done for the purpose of assessing whether AZI can be used as a substitute to EES in patients with known cardiac disease (prolonged QT) or in patients on concomitant medications which also interact with the cytochrome P450 enzymes like EES does. Patients with unexplained abdominal pain are often on antidepressants most of which interact with the CYP3A isoenzymes as does EES. In contrast, AZI does not inhibit the CYP3A isoenzymes.

Furthermore, AZI induces AFs followed by duodenal contractions more frequently than EES, even in patients with GID. Furthermore, analysis of duodenal MMC and AF characteristics indicates that the average duration of effect is also longer with AZI as compared to EES. As a result this study aimed to determine whether another macrolide with fewer drug interactions than EES, could treat small bowel dysmotility.

Of all the other macrolides available, AZI has the least effect on cytochrome P450 isoenzymes and is therefore not implicated in clinically significant drug-drug interactions. In fact, no dosage adjustments are necessary even when AZI is given in conjunction with warfarin (26). In one study, Milberg et al. found that although EES and Clarithromycin® cause early afterdepolarizations and torsades de pointes (TdP) by lowering K+ concentrations, AZI did not cause either depolarizations or TdP and had the lowest pro-arrhythmic potential out of all three macrolides (25). In some isolated case reports, QT prolongation and TdP have been reported in elderly patients taking AZI (38, 39). A one year, large randomized controlled study with patients prospectively analyzed with stable CAD to receive 600 mg AZI or placebo weekly for a year showed no alteration in the risk of cardiac events. This study concluded that AZI cannot be administered for secondary prevention of coronary events, however in the same respect, no increased risk of cardiac disease was seen with use of AZI (40).

Another benefit in using AZI instead of EES and other macrolides is the lower incidence of side effects seen with AZI (41). Common side effects seen with EES include nausea, vomiting and diarrhea which are seen to a lesser degree with AZI. Another advantage is that AZI reaches higher intracellular concentrations, thus increasing both its duration of action and its efficacy. This outcome was seen in our data when the total duration of antral contractions was compared between EES and AZI with the finding that the duration of antral contractions was longer with AZI than with EES. Furthermore, AZI is absorbed rapidly with food, thus increasing its absorption so that it can be taken with or without food. Moreover, in contrast to EES, AZI has a higher oral bioavailability. This bioavailability gives AZI an extensive distribution throughout the body with high tissue levels and at the same time it has poor central nervous system penetration. The second phase showed improvement of number of MMCs in the duodenum with

AZI from baseline and as compared to EES in patients with GID. This is the first study to show AZI as a promising prokinetic in patients with both gastric *and* small bowel dysmotility. Finally, the long half-life of AZI being up to 68 hours can potentially allow for easier administration due to long duration of effect. Perhaps then this medication could be dosed once daily as opposed to four times daily administration required with oral dosing with EES. This once daily dosing may also improve patient compliance with taking the medication.

This study has three significant limitations. First, like many studies done with prokinetic agents, this study lacked randomized control data and had an open-label design making it methodologically weak and subject to bias. This study had an uncontrolled design, small sample size, was of short duration, failed to randomize the order of drug administration and lacked symptom assessment, all of which limited the generalizability of the study. Nevertheless, this study's purpose was to illustrate whether EES and AZI can be used interchangeably with a similar prokinetic effect, but without AZI interacting with other drugs which are metabolized through the P450 metabolic pathway. Second, this study is limited by the lack of standardized criteria for reading of ADM with differences in between studies in regards to meal composition, analysis for research, and even the motility indices with differences in measurement. These different systems make comparison to other studies difficult given the marked variability in analysis and recordings with different durations for the postprandial, digestive and interdigestive phases. Still, we tried to minimize this effect by having two gastroenterologists trained in reading ADM studies interpret the ADM separately with agreement found in 90% of studies interpreted for clinical purposes. Third, 18 of the patients did not have GP by GES criteria but had antral hypomotility seen on ADM. Despite these limitations, we hope that in future studies, symptom improvement can be used as the endpoint using the cardinal GP symptom Index for assessment

of symptoms and for comparison of the two prokinetics in conjunction with GES or breath testing analysis.

Finally, AZI stimulates postprandial antral activity to a higher degree than EES. The duration, MI and mean amplitude of contractions in patients with documented GP by ADM are increased with AZI. This increase may be due to agonist properties at the motilin receptor initiating the gastric antral contractions, or perhaps another yet unknown mechanism. Depoortere et al. showed that only 12 and 14 membered macrolides displaced motilin while the others notably failed to act as motilin agonists at all (42). Therefore, as a 15-membered macrolide, AZI may exhibit its prokinetic properties through another mechanism altogether. Since as many as a third of patients with GP also have concurrent small bowel dysmotility, the extension of our investigation into the small bowel effect of AZI is very promising given EES has been shown to inhibit small bowel motility. The potential mechanism for this effect of AZI on induction of MMC-like AFs in the duodenum is unknown but warrants further investigation. Ultimately, further research is necessary to determine definitively whether the finding of increased antral contractions with use of IV AZI during ADM will translate into a prokinetic oral indication for use of AZI. A well designed, randomized controlled blinded study will need to be performed to show significant improvement of patient's symptoms, with a decreased side effect profile, using this very promising macrolide, AZI. This could provide the focus of future studies on treatment of gastroparesis.

CHAPTER 5
FUTURE GOALS

A recent submission of my proposal "Comparison of Two Macrolides, Azithromycin and Erythromycin, for Symptomatic Treatment of Gastroparesis" has now been accepted as of May, 2009 for funding by the Clinical and Translational Science Institute's Pilot and Collaborative Project Program for Junior Faculty in the University of Florida's K30 APPCI Program. This study aims to evaluate the effect of AZI in comparison to EES on gastrointestinal motility by using a non-invasive, inexpensive, 13C Octanoic Acid Breath Test for measurement of gastric emptying, and by monitoring of symptoms through the Gastroparesis Cardinal Symptom Index (GCSI) in a double-blind, cross-over study. This study hopes to demonstrate the effectiveness of AZI as compared to EES and later form the framework for larger randomized-controlled parallel studies to investigate use of AZI for treatment of GP. Quality of life measures will also be performed with this pilot cross-over study. This project will hopefully pave the way for more opportunities for funding from the National Institute of Health and for promoting my career in the field of motility disorders.

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BIOGRAPHICAL SKETCH

Baharak Moshiree received her undergraduate degree from the University of Florida in 1997, and she was accepted into the Junior Honors Program in 1996. Her medical degree is from the University of Florida where she graduated in the top 30% in her medical school class. She then completed training in internal medicine at the Medical College of Virginia in 2003. She successfully completed subspecialty training in gastroenterology at the University of Florida in 2006 with an advanced training in motility disorders. She joined the College of Medicine at the University of Florida as faculty in the Division of Gastroenterology, Hepatology, and Nutrition in September, 2007. The Master of Science with a concentration in Clinical Investigation was completed during her appointment as Assistant Professor in the Division of Gastroenterology and Hepatology. She is currently the Director of Motility at the University of Florida and Co-Director of the Pelvic Floor Program which was initiated at the University of Florida in collaboration with the colo-rectal surgery division.