NON-ENDOSCOPIC SURVEILLANCE FOR BARRETT'S ESOPHAGUS FOLLOWING ABLATIVE THERAPY

Principal Investigator

Nicholas J. Shaheen, MD, MPH
Professor of Medicine and Epidemiology
Director, Center for Esophageal Diseases and Swallowing
University of North Carolina School of Medicine
Chapel Hill, NC

Phone: (919) 966-7047
Fax: (919) 843-2508
Email: nicholas_shaheen @med.unc.edu

Co-Investigators

Rebecca C. Fitzgerald, MA, MD
Cambridge University
Cambridge, U.K.

Funding Sponsor: National Institutes of Health (NIH)
Covidien GI Solutions now part of Medtronic

UNC IRB Number: 13-2618

Protocol Version: 1.0

Protocol Version Date: 08 December 2015

National Clinical Trial Number: NCT02106910

SIGNATURE PAGE

The signature below constitutes the approval of this protocol and the attachments, and provides the necessary assurances that this trial will be conducted according to all stipulations of the protocol, including all statements regarding confidentiality, and according to local legal and regulatory requirements and applicable US federal regulations and ICH guidelines.

Principal Investigator or Clinical Site Investigator:

Signed: _____ Date: _____

Name: Nicholas Shaheen, MD, MPH

Title: Principal Investigator, Professor of Medicine and Epidemiology

Table of Contents

List of Abbreviations

Item	Definition
BE	Barrett's Esophagus
eCRF	Electronic Case Report Form
EAC	Esophageal Adenocarcinoma
EC	Ethics Committee
EGD	Esophagogastroduodenoscopy
EMR	Endoscopic Mucosal Resection
HGD	High grade dysplasia
IMC	Intramucosal Carcinoma
IRB	Institutional Review Board
LGD	Low grade dysplasia
PDT	Photodynamic Therapy
RFA	Radiofrequency Ablation

Study Summary

Title	Non-Endoscopic Surveillance for Barrett's Esophagus Following Ablative Therapy
Methodology	This is a cross-sectional study of subjects with dysplastic Barrett's Esophagus (BE) who have undergone successful endoscopic ablation with radiofrequency ablation (RFA), to assess the utility of the Cytosponge assay as a non-endoscopic method for monitoring the post-ablation patient. In addition, we will enroll a small cohort of up to 50 subjects with a current diagnosis of BE to collect pilot data for a larger scale study. Subjects presenting to UNC Hospitals for routine endoscopic examinations for current BE or after successful ablation will be offered enrollment in the study. After informed consent, and the same day as the endoscopic procedure, the subject will undergo administration of the Cytosponge assay and complete a questionnaire. The subject will then undergo routine endoscopic surveillance, using a standard Seattle biopsy surveillance protocol. The Cytosponge will be placed in fixative and shipped to the Fitzgerald laboratory for processing according to their established protocols. If the Cytosponge tissue specimen is inadequate, the patient will be recalled for a repeat sponge procedure (not endoscopy) 30 days later. Routine care tissue biopsies will undergo standard processing and H&E staining, with assessment by expert gastrointestinal pathologists. Subjects will be contacted via phone 7 days (+/- 2 days) after Cytosponge administration to complete additional questionnaires.
Study Duration	5 Years
Study Center(s)	University of North Carolina, Chapel Hill, NC
Objectives	*Primary objective:* To assess the acceptability of a novel, minimally invasive esophageal mucosal sampling technique, the Cytosponge, in subjects undergoing surveillance after radiofrequency ablation. Based on previous data, we hypothesize that the sponge-based sampling technique will be associated with low levels of patient distress, and will be preferred by patients, when compared to standard sedated upper endoscopy, for surveillance of their esophageal mucosa. *Secondary objective:* To assess the operating characteristics of this technique against a gold standard of upper endoscopy with biopsies for endoscopic surveillance in subjects with a history of successful radiofrequency ablation for dysplastic BE. We hypothesize that the assay will demonstrate both a sensitivity and specificity of >90% in the detection of recurrent BE following radiofrequency ablation. Further, we expect higher accuracy in those with a larger burden of recurrent disease.
Number of Subjects	374 (324 Post-ablation, 50 BE)

Diagnosis and Main Inclusion Criteria	Inclusion Criteria: 1. Male or female subjects, age 18-80 years, 2. Meets the following: 2.1. Previous diagnosis of BE with dysplastic LGD or HGD, as evidenced by both classical endoscopic appearance of salmon-colored mucosa in the tubular esophagus, as well as endoscopic biopsies from the involved areas demonstrating columnar metaplasia with goblet cells. The diagnosis of dysplasia must have been confirmed by a second expert pathologist. Previous EMR of focal nodular HGD or superficial intramucosal cancer (IMC) is allowable, as long as the EMR specimen shows complete resection of any IMC with clear margins, and biopsies following ablation confirm excision of the lesion, **AND** 2.1.1. A history of complete eradication of both dysplasia and intestinal metaplasia by radiofrequency ablation. Complete eradication is defined as a normal endoscopic appearance of the tubular esophagus, and histologic confirmation by biopsies in 4 quadrants every cm from throughout the length of the previous BE (post-RFA cohort).**OR** 2.2. Current diagnosis of BE, presenting for routine care endoscopy (BE cohort). 3. Good general health, with no severely debilitating diseases, active malignancy, or condition that would interfere with study participation. Exclusion Criteria: 1. Current use of blood thinners such as coumadin, warfarin, clopidogrel, heparin and/or low molecular weight heparin (requires discontinuation of medication 7 days prior to and 7 days after esophagogastroduodenoscopy [EGD] and Cytosponge administration, aspirin use is OK). 2. Known bleeding disorder 3. For the post-RFA cohort, prior ablative therapy of the esophagus other than radiofrequency ablation (RFA), including photodynamic therapy (PDT), more than one session of spray cryotherapy, and any other ablation therapies is exclusionary. However, prior endoscopic mucosal resection (EMR) is acceptable and up to two prior treatments of thermal/coagulation therapy (other than RFA) for focal residual disease following otherwise successful RFA therapy is acceptable. The BE cohort must be treatment naive and have no history of ablation, but prior EMR is acceptable. 4. History of esophageal stricture precluding passage of the endoscope or sponge, 5. Pregnancy, or planned pregnancy during the course of the study, 6. Any history of esophageal varices, liver impairment of moderate or worse severity (Child's- Pugh class B & C) or evidence of varices noted on any past endoscopy, 7. Any history of esophageal surgery, except for uncomplicated fundoplication, and, 8. History of coagulopathy, with INR>1.3 and/or platelet count of <75,000. 9. Planned ablation therapy within 3 days of Cytosponge administration (endoscopic mucosal resection and submucosal dissection is OK).

Statistical Methodology	For the primary objective, to assess the acceptability of the novel mucosal sampling technique in subjects after successful ablation, we will assess the distribution of Impact of Events Scale scores, and the intrusiveness and avoidance subscales. We will generate measures of central tendency and distribution of these data. Bivariate analysis will be performed to assess for predictors of low tolerance of Cytosponge surveillance, and a logistic regression model created to assess these factors while controlling for potential confounders. Data will be compared to population norms using parametric statistics. VAS scores will be calculated, and measures of central tendency and distribution reported. Subjects' preferences for Cytosponge vs. endoscopic surveillance, as well as willingness to undergo the procedure again, will be measured as proportions, with bivariate and multivariate analyses for predictors of preference performed. For the secondary objective, to assess the operating characteristics of Cytosponge against a gold standard of upper endoscopy, initially 2x2 tables will be constructed demonstrating Cytosponge and the gold standard findings (Y/N for BE). Sensitivity, specificity, positive predictive value, negative predictive value and accuracy will be calculated. Because Cytosponge positivity may vary based on the burden of BE, we will perform sensitivity analyses, defining "positive" cases as those with recurrent BE of ≥1 cm in length, and then ≥2 cm in length, to assess impact of disease burden on operating characteristics. Multivariate models controlling for age, sex, period of time from last ablation, burden of residual disease, and other potential confounders will be constructed, to assess the impact of these factors on test accuracy. Although we do not expect to see an association between the degree of dysplasia and Cytosponge positivity, exploratory analyses will be performed using degree of dysplasia as a predictor variable, and Cytosponge positivity as the outcome variable.

1 Introduction

This document is a protocol for a human research study. The study will be conducted in accordance with the International Conference on Harmonisation guidelines for Good Clinical Practice (ICH E6), the Code of Federal Regulations on the Protection of Human Subjects (45 CFR Part 46), and applicable federal regulations and institutional policies. All personnel involved in the conduct of this study have completed human subjects protection training..

1.1 Background

Barrett's esophagus (BE) is a premalignant condition associated with the development of esophageal adenocarcinoma (EAC). BE is an extremely common condition, and 1-2% of the general adult population harbor this lesion. In the majority of such subjects, the condition will be indolent, and EAC will not develop. However, in <5% of these subjects, BE will progress to EAC. If BE does progress, it is thought to do so through worsening degrees of dysplasia, from no dysplasia, to low-grade dysplasia (LGD), to high-grade dysplasia (HGD) and on to EAC. The prognosis of EAC is dismal, with a <15% five year survival. For that reason, endoscopic intervention is suggested in the setting of dysplastic BE to avert the development of cancer.

Radiofrequency ablation (RFA) is a safe and effective method for inducing reversion of BE to neosquamous epithelium. RFA is associated with a >90% risk reduction of EAC in the setting of dysplastic BE. However, BE recurs in as many as a quarter of subjects by 36 months following successful RFA. For this reason, surveillance endoscopy is recommended following RFA with complete eradication of intestinal metaplasia. Because this surveillance endoscopy is performed at frequent intervals (every 3-12 months), these procedures obligate the patient to recurrent risks associated with sedation and upper endoscopy, and the inconvenience of these exams. Importantly, the costs associated with this effort greatly impact the cost-effectiveness of RFA treatment.

A minimally-invasive, sponge-based technique for sampling the esophageal mucosa, the Cytosponge, has recently been described. This simple device is comprised of an abrasive sponge encapsulated in a gelatin coating, which is attached to a string. The subject ingests the capsule, which dissolves after a short exposure to gastric secretions. The sponge, now freed from the capsule, is drawn back through the hiatus and distal esophagus and out of the mouth by the string, sampling cardiac and esophageal epithelium. The resulting sample is immunostained for trefoil factor 3, a marker both sensitive and specific for the presence of BE. Preliminary work from collaborators in the U.K. demonstrates high sensitivity and specificity of this assay for detecting BE.

This is a comparative effectiveness study to assess the utility of this technique in endoscopic surveillance of patients with BE following successful RFA. The central hypothesis of this work is that this novel surveillance tool could supplant upper endoscopy in subjects having undergone RFA for BE, providing less invasive and more cost-effective surveillance of this large and growing patient population.

1.2 Rationale

Esophageal adenocarcinoma is a lethal cancer with a rapidly increasing incidence. In stark contrast to recent progress in other solid tumors, incidence and death rates from esophageal adenocarcinoma continue to rise rapidly in the U.S. There has been a 500% increase in the incidence of esophageal adenocarcinoma (EAC) from the 1970's to the 1990's,[1] and a near-parallel increase in mortality (*Figure 1*), underscoring the need for more effective prevention and treatment for this lethal cancer. Esophageal adenocarcinoma is thought to develop through a series of metaplastic, then dysplastic, changes of the mucosa.[2] Chronic gastroesophageal reflux (GERD) precipitates a metaplastic change from the normal squamous epithelium, to a more acid-resistant columnar histology.[3] When this columnar epithelium contains goblet cells, it is termed specialized or intestinalized metaplasia. When endoscopically evident, columnar metaplasia with goblet cells in the esophagus is termed Barrett's esophagus (BE).[4,5]

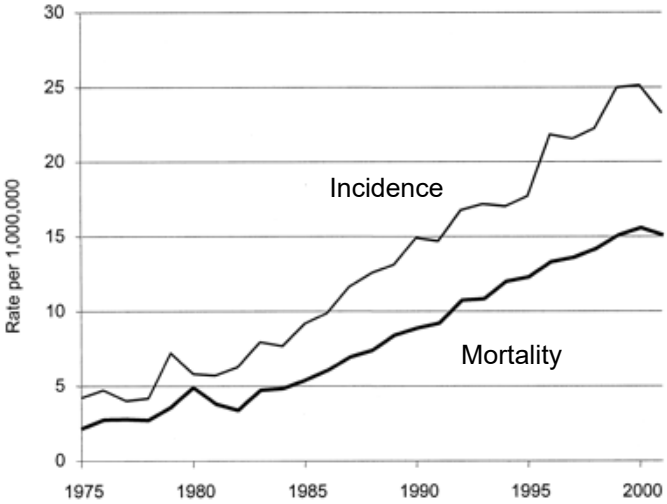


Figure 1 Incidence (top curve) and mortality (lower) from esophageal adenocarcinoma[33]

Barrett's esophagus is the strongest risk factor for esophageal adenocarcinoma. BE is associated with a risk of esophageal adenocarcinoma that is 40-120 times that of the general population.[6,7] Furthermore, BE is an extremely common condition, present in approximately 10% of subjects with chronic GERD,[8] and 1-2% of the general population.[9] Since 10- 20% of adult Americans have at least weekly symptoms of GERD,[10,11]the number of cases of BE in the U.S. is thought to be >2 million.22 BE does not generally spontaneously regress; barring an intervention, the patient will have BE for life. Most subjects harboring BE will not progress to EAC. However, in a proportion (0.2-0.5%/year,)2,[12] the metaplastic tissue will progress from low-grade dysplasia (LGD) to high-grade dysplasia (HGD), culminating in EAC.[13,14] Given the poor prognosis of cancer diagnosed symptomatically,[15,16] current effort is directed toward early detection and treatment. Current strategies for prevention of EAC focus on endoscopic screening and surveillance.[17] In the approach most commonly used in the U.S., subjects with chronic heartburn are offered a screening endoscopy to assess for BE. Patients found to have BE are then enrolled in endoscopic surveillance, consisting of periodic endoscopy at intervals governed by the presence of dysplasia.24 In current American College of Gastroenterology guidelines,[4] subjects with BE and no dysplasia undergo endoscopy every 3 yrs. Subjects with BE and LGD have endoscopy yearly. Subjects with BE and HGD are effectively managed with endoscopic therapy,[18] but may also opt for esophagectomy or continued surveillance. Endoscopic exams for GERD and BE are common and costly,

with over 330,000 exams/year in Medicare patients alone,[19] and an average total cost in ambulatory care centers of >$2,000/exam,[20] making for $660M in endoscopy costs in the Medicare population alone annually.

Endoscopic ablation induces reversion of Barrett's Esophagus and decreases progression of disease. Due to the increasing incidence of EAC and the poor prognosis once cancer has developed, endoscopic ablative therapies are commonly employed in subjects with dysplastic BE. In the most commonly used approach, radiofrequency ablation (RFA), high radiofrequency waves, are delivered by an endoscopic balloon covered with an electrode array, to cause epithelial destruction (*Figure 2*). Because of the regular spacing of the electrodes, and the delivery of a pre-set amount of energy, the depth of injury is well-controlled and reproducible. Data from our center and others demonstrates that RFA is highly effective in inducing reversion to squamous epithelium in subjects with dysplastic BE.[21–23] Additionally, data suggest that subjects with dysplastic BE who have undergone ablative therapy with RFA are less likely to progress to either more severe dysplasia or adenocarcinoma.[24] For these reasons, the newest societal guidelines recommend endoscopic ablative therapy for subjects with HGD.[18] Further, RFA is cited as a preferred endoscopic ablative therapy for these subjects. Not surprisingly, RFA has become the predominant method for ablation of BE in the U.S., and to date, over 100,000 of these procedures have been performed in the U.S. (personal communication, D. Utley, CMO, GI Solutions).

Figure 2 Circumferential Ablation Catheter

Recurrence of BE following ablative therapy. Early enthusiasm for endoscopic ablative therapy was stoked by the hope that, in addition to being an effective anti-neoplastic measure, successful ablation might allow elimination of surveillance upper endoscopy afterward. Because periodic upper endoscopy is used frequently in these subjects (every 3-12 months, depending on the baseline degree of dysplasia),[24] omission of these examinations would result in substantial healthcare savings, and would reduce risk and inconvenience to patients. Unfortunately, data demonstrate a risk of recurrence of BE following successful eradication. Recent data published by the candidate and colleagues from the AIM Dysplasia study demonstrate that approximately 25% of subjects who experience successful eradication of dysplastic BE will develop recurrent BE, almost all in the first year following successful therapy (*Figure 3*).[24] While most of these recurrences are non-dysplastic BE, dysplastic BE and even adenocarcinoma have been noted to occur after endoscopic ablative therapy.[25] Therefore, following successful endoscopic ablation, patients receive ongoing endoscopic surveillance. Current post ablation surveillance practices are governed by expert opinion, but reported intervals are frequent (every 3-12 months), and involve a copious number of biopsies. The cost and frequency of these examinations impacts the cost-effectiveness of ablative therapy for BE.[26] Additionally, each of these examinations imparts a small, but real risk to the patient.[27] There is substantial inconvenience and negative impact in quality of life associated with enrollment in endoscopic surveillance programs,[28] especially since the center performing the ablation may be far removed from the patient's home, and because currently most exams are done

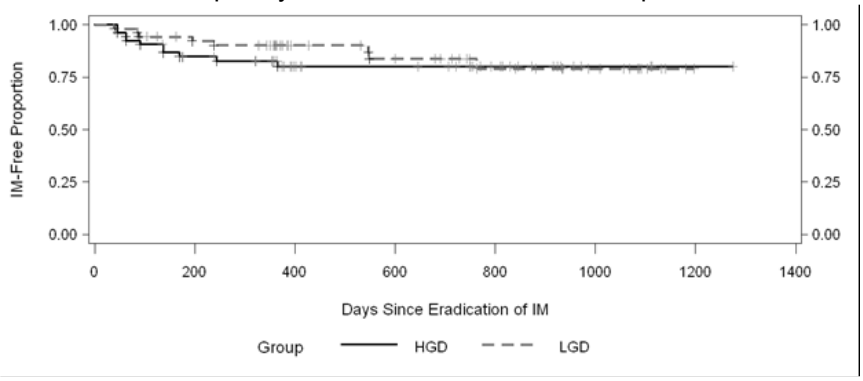


Figure 3 Durability of Eradication of IM[34]

under sedation, requiring both a day missed from work and an accompanying driver for transportation home. Because the number of subjects having undergone RFA in the U.S is now large, and continuing to rise, this is a substantial and growing healthcare cost.

A novel non-endoscopic technique as a potential surveillance tool after successful endoscopic ablation. Given the large number of patients with chronic GERD and BE, and the expense and inconvenience of endoscopic examinations for BE, investigators have sought less expensive, non-endoscopic modalities of assessing patients for BE. Early attempts using a non-endoscopic balloon demonstrated inadequate sensitivity, in part due to inadequate cytological samples.[29] More recently, a simple, non-endoscopic device, termed the Cytosponge, has been developed for endoscopic screening of subjects at risk for BE by investigators at the University of Cambridge in the U.K. The Cytosponge is an ingestible gelatin capsule enclosing a compressed spherical mesh of 3 cm diameter, the center of which is attached to a string (*Figure 4*). The capsule and string are swallowed with water. The string is held at the mouth without tension, allowing the capsule to move into the stomach. After 5 minutes (during which the gelatin capsule dissolves and the sponge is liberated), the sponge is withdrawn by gentle traction on the string. The sponge is placed in fixative for 48 hours, then the cells are pelleted, and processed into paraffin blocks. The pellets are immunostained with trefoil factor 3, which is interpreted simply as either positive or negative by the presence of any staining. In a recent study of 504 subjects with chronic GERD symptoms in the U.K. who underwent both Cytosponge analysis and upper endoscopy, the Cytosponge demonstrated a sensitivity of 90% and a specificity of 94% for the detection of BE (Prague classification C2 or more).[30] Follow-up work with an additional 334 subjects (186 controls, 148 BE) demonstrated similar results, with a sensitivity of 84% and a specificity of 92% (data courtesy of Rebecca Fitzgerald, MA(Cantab),MD). These results suggest that the Cytosponge may have suitable operating characteristics to serve as a surveillance tool in subjects with BE or dysplastic BE who have undergone ablative therapy.

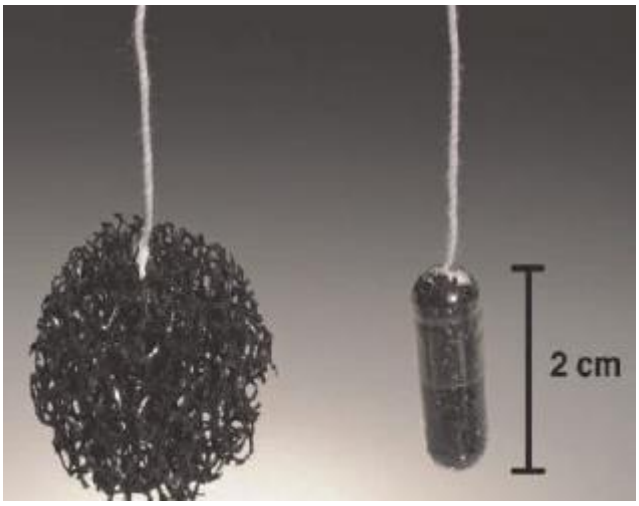


Figure 4 Cytosponge

Figure 5 CytospongeTM (left) and Cytosponge I (right) for comparisonCytospongeTM (left) and Cytosponge I (right) for comparison

Medtronic, formerly Covidien GI Solutions, has developed a more refined version of the Cytosponge I (referred hereafter as the Cytosponge™ or "Cytosponge"). The Cytosponge was developed from the Cytosponge I specification and design, with the additional priority of a more reproducible manufacturing process, standardization of dimensions, and other quality related features (Figure 5). Because we seek to assess a tool for widespread clinical usage, this study will use the Cytosponge provided by Covidien GI Solutions (now part of Medtronic).

1.3 Device Description

The Cytosponge™ Cell Collection Device (Cytosponge) is intended to collect surface cells from the esophagus. The device consists of a swallowable capsule, which dissolves in the body cavity, releasing a self-expandable sponge. The sponge is then retrieved from the esophagus using an attached cord. During the retrieval process, the sponge collects cells from the most superficial layer of the esophageal mucosa. Once removed from the body cavity, the sponge and cells are retained for investigation and/or testing.

Figure 6 Cytosponge with planned packaging and retrieval cord

The Cytosponge™ Cell Collection Device (Cytosponge) received 510(k) clearance from the FDA on November 26, 2014 (K142695). The Cytosponge ™ Cell Collection device is a Class II product under 21 CFR 874.4710 esophagoscope (flexible or rigid) and accessories. This study uses the Cytosponge in accordance with its labeling and is therefore exempt from an IDE per 21CFR812.2.

1.3.1 Prior Utilization

Dr. Fitzgerald and colleagues administered Cytosponge to 504 patients in a primary care setting and found it to be safe and well-tolerated. Of these patients, 501 (99%) were able to successfully swallow the capsule. Unsurprisingly, given pill-swallowing difficulty in the general population, 3 patients were unable to swallow the pill, feeling it was too large. No adverse events were noted, and patients demonstrated a low level of anxiety associated with the test. These 504 administrations are documented in a report in the British Medical Journal[30]. These investigators have, since this investigation, administered the Cytosponge to an additional 831 patients as part of the BEST2 study (a multicenter, prospective study to determine whether BE patients can be risk stratified using the Cytosponge) and an interim data analysis shows similarly excellent safety and tolerance profile, with no adverse events reported. [31] Overall, to date, there have been 1,335 documented administrations of the sponge, with no adverse events. Several hundred additional uses of the device have occurred in Cambridge, UK, without adverse event (personal communication, Dr. Fitzgerald), but have not yet been reported in the peer-reviewed literature.

Cytosponge is extremely similar to Cytosponge I. Although no complications have been reported with this device, there are several theoretical risks associated with the administration of this device. There is the possibility of aspiration whenever instrumentation of the esophagus or stomach is performed. Because the device does not render the upper esophageal sphincter incompetent, this risk is expected to be minimal. Additionally, although the sponge is soft and non-abrasive, subjects could bleed from any mucosal surfaces of the mouth, stomach or esophagus which come in contact with it, and/or experience a sore or irritated throat following administration. Finally, to date there have been >1,000 administrations and detachment of the sponge from the string has occurred in less than 1% of cases. Should the sponge detach from the string, it will be retrieved during the routine care upper endoscopy immediately following administration.

The current study has been designed to minimize occurrence of these theoretical risks. Specifically we plan to exclude those patients who:

- Are unable to discontinue clopidogrel, and/or warfarin for 7 days prior and 7 days after procedure,
- Have a history of esophageal stricture,
- Have any history of esophageal varices, liver impairment of moderate or worse severity (Child's-Pugh class B & C), or evidence of varices noted on any past endoscopy,
- Have any history of esophageal surgery (except uncomplicated fundoplication), and history of coagulopathy, with INR >1.3 and/or platelet count of <75,000.

Importantly, all patients enrolled in this study will have previously and recently received an upper endoscopy during which absence of findings putting the subject at higher risk for these theoretical concerns (conditions such as varices and stricture) will be documented.

Cytosponge administration will occur after an overnight fast to minimize the possibility of aspiration of any gastric contents. Every administered sponge will be assessed post-procedure for signs of fracture or incomplete retrieval of the sponge. In the unlikely case of incomplete retrieval, the sponge will be retrieved with a Roth net at the standard of care endoscopy which will routinely immediately follow the administration of the sponge per the study protocol. Any bleeding noted, either clinically following the sponge administration or due to blood on the sponge itself, will be similarly investigated, and, as necessary, treated during the subsequent endoscopy. Because study inclusion/exclusion criteria are designed to exclude those at highest risk for a bleeding complication, the risk of bleeding in this study should be extremely low.

2 Study Objectives

Primary objective:
To assess the acceptability of a novel, minimally invasive esophageal mucosal sampling technique, the Cytosponge, in subjects undergoing surveillance after radiofrequency ablation. Based on previous data, we hypothesize that the sponge-based sampling technique will be associated with low levels of patient distress, and will be preferred by patients, when compared to standard sedated upper endoscopy, for surveillance of their esophageal mucosa.

Secondary objective:
To assess the operating characteristics of this technique against a gold standard of upper endoscopy with biopsies for endoscopic surveillance in subjects with a history of successful radiofrequency ablation for dysplastic BE. We hypothesize that the assay will demonstrate both a sensitivity and specificity of >90% in the detection of recurrent BE following radiofrequency ablation. Further, we expect higher accuracy in those with a larger burden of recurrent disease.

3 Study Design

3.1 General Design

This is cross-sectional study of subjects with dysplastic BE who have undergone successful endoscopic ablation with RFA, to assess the utility of the Cytosponge assay as a non-endoscopic method for monitoring the post-ablation patient. In addition, we will enroll a small cohort of up to 50 subjects with a current diagnosis of BE to collect pilot data for a larger scale study. Subjects presenting to UNC Hospitals for routine endoscopic examinations for current BE or after successful ablation will be offered enrollment in the study. After informed consent, and the same day as the endoscopic procedure, the subject will undergo administration of the Cytosponge assay. The patient will then undergo routine endoscopic surveillance, using a standard Seattle biopsy surveillance protocol. The Cytosponge will be placed in fixative and shipped to the Fitzgerald laboratory for processing according to their established protocols. Tissue biopsies will undergo standard processing and H&E staining, with assessment by expert gastrointestinal pathologists at UNC. The primary outcome variables will be sensitivity and specificity of the novel assay, compared against the gold standard of the presence of recurrent BE as detected by upper endoscopy with biopsies. Secondary outcomes include acceptability of the nonendoscopic assay to the patient (assessed by a standardized tool, the Impact of Events Scale, as well as a visual analogue scale), and likelihood of assay positivity as a function of amount of residual disease (as measured by Prague criteria).

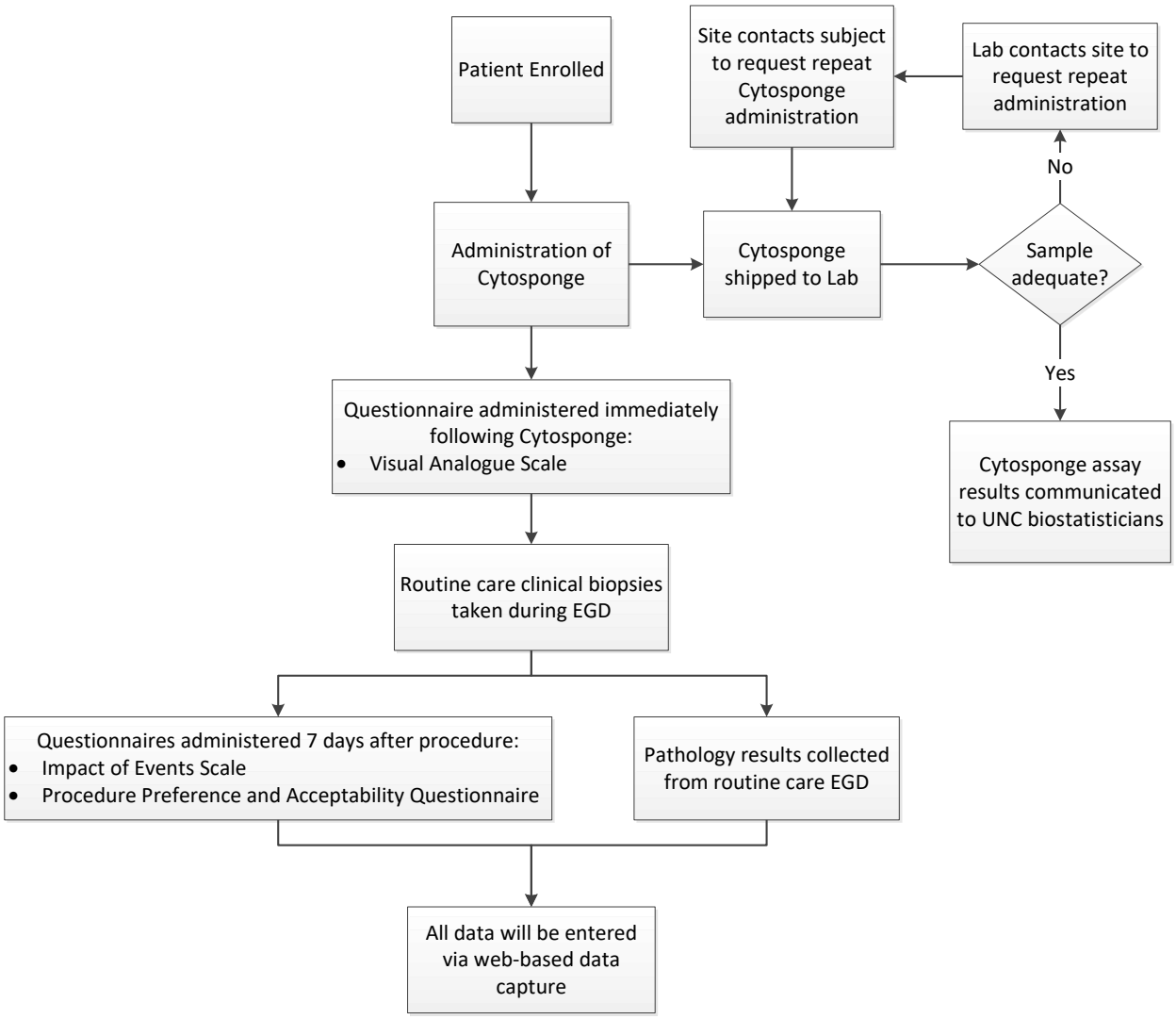
If the initial administration of the Cytosponge demonstrates an inadequate sample, defined as a sample that does not demonstrate at least one columnar cell on hematoxylin & eosin staining, repeat Cytosponge administration will be performed at 30 days (+/- 10 days) from the initial administration.

Subject recruitment will be from consecutive eligible patients presenting for routine care surveillance endoscopy of current BE or following endoscopic ablation of BE. Potentially eligible subjects will be contacted by telephone in advance of their procedure and their interest in study participation assessed. Subjects interested in participating will be asked to present to the endoscopy unit one hour prior to their scheduled procedure. At that time, inclusion and exclusion criteria will be reviewed, and eligible subjects will give informed consent. Subjects will then undergo the Cytosponge assay. Following Cytosponge assay, subjects will complete the procedure acceptability measures described below. Subjects will then attend their usual surveillance endoscopy session. Upper endoscopy will be performed and biopsies taken as part of routine care. Importantly, no record of the results of the standard endoscopy and biopsies will be provided to those assessing the Cytosponge results and these individuals will be masked to all clinical data. Similarly, the pathologists interpreting the histological specimens from the endoscopy will have no knowledge of the outcomes of the Cytosponge assay. Only the study biostatistician will have access to these data.

Primary assessment of acceptability will be via the Impact of Events Scale. This widely used scale was developed to assess the distress associated with a specific life event. It includes measures of both the intrusiveness of the event, and any avoidance responses by the subject in response to the event. Final scores are between 0-75, with low scores demonstrating a low impact of the event. The scale will be administrated 7 days after the sampling in order to allow the subjects to have time to reflect on the experience and to compare with the EGD. Secondary acceptability outcomes will include a visual analogue scale of acceptability of the Cytosponge, performed after the Cytosponge is administered. Also, the subject will be asked whether he/she would be willing to repeat the assay, and, assuming similar accuracy between Cytosponge and upper endoscopy, whether he/she would rather undergo surveillance by Cytosponge or standard EGD with biopsies.

There are no plans for repeat testing or duplicate enrollment. Enrolled subjects will be administered the cytosponge one time, on the date of an endoscopy scheduled for routine care, prior to the endoscopy. Subjects can only participate one time. The only exception is for subjects who have an inadequate sample on the initial Cytosponge. These subjects will be asked to return for repeat Cytosponge administration 30 days (+/- 10 days) after the initial administration.

3.2 Protocol Map



3.3 Primary Outcomes

The primary outcome variables will be sensitivity and specificity of the novel assay, compared against the gold standard of the presence of recurrent BE as detected by upper endoscopy with biopsies.

3.4 Secondary Outcomes

Secondary outcomes include acceptability of the non-endoscopic assay to the patient (assessed by a standardized tool, the Impact of Events Scale, as well as a visual analogue scale), and likelihood of assay positivity as a function of amount of residual disease (as measured by prague criteria).

4 Subject Selection and Withdrawal

4.1 Inclusion Criteria

1. Male or female subjects, age 18-80 years,
2. Meets the following:
   2.1. Previous diagnosis of BE with dysplastic LGD or HGD, as evidenced by both classical endoscopic appearance of salmon-colored mucosa in the tubular esophagus, as well as endoscopic biopsies from the involved areas demonstrating columnar metaplasia with goblet cells. The diagnosis of dysplasia must have been confirmed by a second expert pathologist. Previous EMR of focal nodular HGD or superficial intramucosal cancer (IMC) is allowable, as long as the EMR specimen shows complete resection of any IMC with clear margins, and biopsies following ablation confirm excision of the lesion, **AND**
      2.1.1. A history of complete eradication of both dysplasia and intestinal metaplasia by radiofrequency ablation. Complete eradication is defined as a normal endoscopic appearance of the tubular esophagus, and histologic confirmation by biopsies in 4 quadrants every cm from throughout the length of the previous BE (post-RFA cohort).**OR**
   2.2. Current diagnosis of BE, presenting for routine care endoscopy (BE cohort).
3. Good general health, with no severely debilitating diseases, active malignancy, or condition that would interfere with study participation.

Exclusion Criteria:

4.2 Exclusion Criteria

1. Current use of blood thinners such as coumadin, warfarin, clopidogrel, heparin and/or low molecular weight heparin (requires discontinuation of medication 7 days prior to and 7 days after esophagogastroduodenoscopy [EGD] and Cytosponge administration, aspirin use is OK).
2. Known bleeding disorder
3. For the post-RFA cohort, prior ablative therapy of the esophagus other than radiofrequency ablation (RFA), including photodynamic therapy (PDT), more than one session of spray cryotherapy, and any other ablation therapies is exclusionary. However, prior endoscopic mucosal resection (EMR) is acceptable and up to two prior treatments of thermal/coagulation therapy (other than RFA) for focal residual disease following otherwise successful RFA therapy is acceptable. The BE cohort must be treatment naive and have no history of ablation, but prior EMR is acceptable.
4. History of esophageal stricture precluding passage of the endoscope or sponge,
5. Pregnancy, or planned pregnancy during the course of the study,
6. Any history of esophageal varices, liver impairment of moderate or worse severity (Child's- Pugh class B & C) or evidence of varices noted on any past endoscopy,
7. Any history of esophageal surgery, except for uncomplicated fundoplication, and,
8. History of coagulopathy, with INR>1.3 and/or platelet count of <75,000.
9. Planned ablation therapy within 3 days of Cytosponge administration (endoscopic mucosal resection and submucosal dissection is OK).

4.3 Subject Recruitment and Screening

Potential subjects will be identified during their GI clinic or procedure visits at their treating institutions. All subjects will be screened and enrolled using EC/IRB-approved and HIPAA compliant methods.

An investigator, study coordinator, or other qualified personnel will obtain written informed consent prior to any study procedures. Potential subjects will have an opportunity to carefully review the consent form. The details of the study will be reviewed verbally, and all questions will be answered to the satisfaction of the patient. Only adults with the ability to consent will be eligible for enrollment in this study. After the subject signs the consent, a copy of the signed consent will be provided to the subject. Once written consent has been obtained, the coordinator will collect demographic and historical information from the patient pertaining to history of Barrett's Esophagus and ablation therapy.

The consent process will be documented by the coordinator in the patient's study file.

4.4 Early Withdrawal of Subjects

Subjects will be considered to have completed the study after completion of the last study visit (follow-up phone call). Subjects may be withdrawn prior to this for any of the following reasons:

- Death, or
- Lost to Follow-Up, or
- Withdrawal of consent, or
- Discontinuation by the investigator.

Documentation must be maintained at the site for any subject withdrawals. Subjects unable to complete Cytosponge administration will be withdrawn from the study (discontinued by investigator). Three attempts at contact using two different methods are required prior to determination that the subject is lost to follow-up. Attempts at contact must be with certified letters OR documented telephone contact. If a subject is withdrawn prior to completion of the study, the site should complete and submit a change of status case report form. See section 8.3 for additional information on case report forms. Subjects who withdraw after completion of the initial Cytosponge administration will not be replaced. Withdrawn subjects will not be followed, unless they have an active adverse event (AE) at the time of withdrawal. Subjects withdrawn while experiencing an adverse event will be followed until resolution of the AE.

5 Study Procedures

Assessment	Screening/Enrollment Visit	Follow-Up Phone Call (7 days +/- 2 days after Enrollment)	Repeat Cytosponge (30 days +/-10 days after Enrollment)
Informed Consent Form	X		
Demographics	X		
Medical History	X		
Inclusion/Exclusion Criteria	X		
Cytosponge Administration	X		X[1]
Visual Analogue Scale	X[2]		
Routine Care Endoscopy with Biopsy	X[3]		
Impact of Events Scale		X	
Procedure Preference and Acceptability Questionnaire		X	
Adverse Events[4]	X	X	X

[1]If the initial Cytosponge sample is inadequate, sites will be notified and subjects will be asked to return for repeat Cytosponge administration 30 days (+/- 10 days) after the initial Cytosponge administration. Only the Cytyosponge administration will be repeated. Questionnaires and the endoscopy procedure will not be repeated on these patients.
[2]VAS should be administered immediately following completion of the Cytosponge and prior to the upper endoscopy.
[3]Routine care biopsies should be taken during the endoscopy following the Cytosponge as this is considered standard of care for the target population. No research-specific biopsies will be obtained during the procedure.
[4]Only those events that are potentially related to participation in this research study must be reported. See section 7.2 for definition of a reportable adverse event.

5.1 Screening/Enrollment

During screening/enrollment, eligibility is assessed and those eligible and interested in participating are consented on the study. Once consent is obtained, subjects will undergo administration of the Cytosponge, complete a questionnaire and proceed with routine care upper endoscopy immediately following completion of the Cytosponge in which biopsies are taken for clinical purposes and sent to pathology.

5.1.1 Assessments

The following will be completed during the screening/enrollment visit:

- Eligibility review
- Informed consent
- Cytosponge administration
- Visual Analogue Scale
- Routine care upper endoscopy with biopsy
- Adverse event assessment
- Enrollment Case Report Form (eCRF): This captures demographics including race, ethnicity, gender, and year of birth, relevant BE medical history including documentation of endoscopic procedures received to date as well as pathology findings and endoscopic

history related to current diagnosis. This will also capture the date the Cytosponge was sent to the Fitzgerald Lab for analysis

5.1.2 Consenting Procedure

If a subject is screened eligible and interested in the study, the subject will be consented on the study prior to any study procedure. Written informed consent will be obtained by qualified study personnel. Documentation of the consent process will be maintained in the subject's research record.

Subjects will be given ample time to review the consent document and ask any questions they may have. A copy of the written consent form will be provided to the subject and the original maintained in the subject's research record.

If subjects meet all inclusion and none of the exclusion criteria and consent to the study, they will be enrolled in the study. Subjects will be assigned a unique subject code. Each institution will be provided a list of subject codes to use.

5.1.3 Cytosponge Administration

The Cytosponge™ device (referred to hereafter as the "Cytosponge") will be supplied by Covidien GI Solutions to the participating sites. Study sites will be responsible for storage and accountability of the device. The Cytosponge lifetime/use by date will be confirmed on the product packaging. The device received FDA 510(k) clearance on November 26, 2014 (K142695). The Cytosponge device consists of a spherical 3.0 cm diameter reticulated polyester foam compressed and encapsulated in a standard vegetarian capsule (size 00).

Subjects will undergo administration of the Cytosponge™ according to the IFU. Briefly, subjects will be placed in the seated position and will swallow the capsule with 150 – 250 mL of water. Additional water may be used if necessary. The sponge is attached to a length of suture material which passes out through the capsule. The suture is affixed to a retainer card which is held by the subject or administrator to prevent inadvertent swallowing of the suture. The string is to be held without tension as peristalsis and gravity advance the capsule into the stomach.

The capsule dissolves in the stomach, allowing the sponge to expand to its full size. Seven minutes and 30 seconds to ten minutes after ingestion, the sponge is then withdrawn by gentle traction on the suture, collecting cells from the lining of the esophagus in passing.

After retrieval, the string is cut and the retrieved foam sphere containing the cytological specimen is immersed in fixative and stored refrigerated (1° to 12°*C* [34° to 54°F]) until shipped. Samples will be shipped in batches to the Fitzgerald Lab in Cambridge. On arrival at the Fitzgerald lab, the fixative is spun in a centrifuge, and the pelleted cells are embedded in paraffin using standard techniques. The blocks are sectioned and stained for trefoil factor 3, where any staining is considered positive for the assay.

If a subject fails to swallow the Cytosponge, the subject will be asked to swallow again. Subjects who are willing to try again will be asked to wait 5 minutes before the Cytosponge is presented to them again. Subjects will be able to try up to three times before they are classified as "Cytosponge swallowing failure" and discontinued by the investigator.

5.1.4 Routine Care Endoscopy with Biopsy

After Cytosponge administration is complete, subjects will undergo routine care upper endoscopy, with assessment of BE (where applicable), and biopsy per accepted surveillance or screening recommendations. Routine care tissue biopsies will undergo standard processing and H&E staining at the home institution, with assessment by expert gastrointestinal pathologists.

5.1.5 Adverse Event Assessment

Subjects should be assessed for any adverse events that occur before, during, or after Cytosponge administration. Only those events that are potentially related to participation in this research study must be reported to the lead site. See section 7.2 for the definition of a reportable adverse event. Sites are responsible for following local IRB/EC guidelines for reporting adverse events to their local IRB/EC.

5.2 Follow-Up Phone Call

Subjects will be contacted 7 days (+/- 2 days) following the Cytosponge and upper endoscopy procedures. During this phone call, adverse events will be assessed and subjects will complete questionnaires. Participation in the study is complete when subjects have completed the follow-up phone call.

5.2.1 Assessments

The following data will be collected from subjects during the follow-up phone call:

- Impact of Events Scale
- Procedure Preference and Acceptability Questionnaire
- Adverse event assessment
- Follow-Up Case Report Form (eCRF): This captures relevant information for questionnaire completion and assessment of adverse events.

5.2.2 Impact of Events Scale

The impact of events scale will be completed with the subjected during the follow-up phone call and measures subjective distress related to administration of the Cytosponge.

5.2.3 Procedure Preference and Acceptability Questionnaire

The procedure preference and acceptability questionnaire will be completed with the subject during the follow-up phone call. This assessment collects subject preference for the Cytosponge or traditional upper endoscopy as well as willingness to undergo the procedure again.

5.2.4 Follow-up Phone Call Adverse Event Assessment

During the follow-up phone call, subjects should be assessed for any adverse events that have occurred since administration of the Cytosponge. Only those events that are potentially related to participation in this research study must be reported. See section 7.2 for the definition of a reportable adverse event. Sites are responsible for following local IRB/EC guidelines for reporting adverse events to their local IRB/EC.

5.3 Repeat Cytosponge Administration

If the initial Cytosponge sample is inadequate, sites will be notified and subjects will be asked to return for repeat Cytosponge administration 30 days (+/- 10 days) after the initial Cytosponge administration. Only the Cytyosponge administration will be repeated. Questionnaires and the endoscopy procedure will not be repeated on these patients. See section 5.1.3 for details on the Cytosponge administration procedure. The Repeat Administration eCRF should be completed.

6 Statistical Plan

6.1 Sample Size Determination

Previous data suggest a sensitivity of Cytosponge analysis for detecting BE ranging from 84-90%, and a specificity of 92-94%, using upper endoscopy as a gold standard. It is unlikely that this technique will be appropriate for surveillance if the true sensitivity is much lower than 80%, given the unacceptably high rate of false negatives. Previous data document a rate of recurrent BE of 13-38% in subjects assessed with surveillance endoscopy. If we wish to assess the accuracy of the Cytosponge compared to upper endoscopy, assuming a baseline sensitivity of 87%, we will need to enroll 81 subjects with recurrent BE to

yield a 95% confidence interval of 10% or less. If we further assume a rate of recurrent BE of 25% in the post-ablation population, a total of 324 post-ablation subjects will need to be enrolled in the trial.

6.2 Statistical Methods

For the primary objective, to assess the acceptability of the novel mucosal sampling technique in subjects after successful ablation, we will assess the distribution of Impact of Events Scale scores, and the intrusiveness and avoidance subscales. We will generate measures of central tendency and distribution of these data. Bivariate analysis will be performed to assess for predictors of low tolerance of Cytosponge surveillance, and a logistic regression model created to assess these factors while controlling for potential confounders. Data will be compared to population norms using parametric statistics. VAS scores will be calculated, and measures of central tendency and distribution reported.

Subjects' preferences for Cytosponge vs. endoscopic surveillance, as well as willingness to undergo the procedure again, will be measured as proportions, with bivariate and multivariate analyses for predictors of preference performed.

For the secondary objective, to assess the operating characteristics of Cytosponge against a gold standard of upper endoscopy, initially 2x2 tables will be constructed demonstrating Cytosponge and the gold standard findings (Y/N for BE). Sensitivity, specificity, positive predictive value, negative predictive value and accuracy will be calculated. Because Cytosponge positivity may vary based on the burden of BE, we will perform sensitivity analyses, defining "positive" cases as those with recurrent BE of ≥1 cm in length, and then ≥2 cm in length, to assess impact of disease burden on operating characteristics. Multivariate models controlling for age, sex, period of time from last ablation, burden of residual disease, and other potential confounders will be constructed, to assess the impact of these factors on test accuracy. Although we do not expect to see an association between the degree of dysplasia and Cytosponge positivity, exploratory analyses will be performed using degree of dysplasia as a predictor variable, and Cytosponge positivity as the outcome variable.

6.3 Subject Population for Analysis

The population whose data will be subjected to the study analysis will include all subjects enrolled in this study that completed the Cytosponge administration and have pathology results available from the subsequent routine care endoscopy with biopsy.

7 Safety and Adverse Events

7.1 Definitions

Unanticipated Problems Involving Risk to Subjects or Others
Any incident, experience, or outcome that meets all of the following criteria:

- Unexpected in nature, severity, or frequency (i.e. not described in study-related documents such as the IRB-approved protocol or consent form, the investigators brochure, etc.)
- Related or possibly related to participation in the research (i.e. possibly related means there is a reasonable possibility that the incident experience, or outcome may have been caused by the procedures involved in the research,
- Suggests that the research places subjects or others at a greater risk of harm (including physical, psychological, economic, or social harm) than was previously known or recognized.

Adverse Event
An ***adverse event*** (AE) is any symptom, sign, illness or experience that develops or worsens in severity during the course of the study. Intercurrent illnesses or injuries should be regarded as adverse events. Abnormal results of diagnostic procedures are considered to be adverse events if the abnormality:

- results in study withdrawal
- is associated with a serious adverse event
- is associated with clinical signs or symptoms

- leads to additional treatment or to further diagnostic tests
- is considered by the investigator to be of clinical significance

Serious Adverse Event
Adverse events are classified as serious or non-serious. A ***serious adverse event*** is any AE that is:

- fatal
- life-threatening
- requires or prolongs hospital stay
- results in persistent or significant disability or incapacity
- a congenital anomaly or birth defect
- an important medical event

Important medical events are those that may not be immediately life threatening, but are clearly of major clinical significance. They may jeopardize the subject, and may require intervention to prevent one of the other serious outcomes noted above. For example, drug overdose or abuse, a seizure that did not result in in-patient hospitalization, or intensive treatment of bronchospasm in an emergency department would typically be considered serious.

All adverse events that do not meet any of the criteria for serious should be regarded as ***non-serious adverse events***.

Adverse Event Reporting Period
The study period during which adverse events must be reported is normally defined as the period from the initiation of any study procedures to the end of the study treatment follow-up. For this study, the study treatment follow-up is defined as 30 days following completion of the biomarker panel.

Preexisting Condition
A preexisting condition is one that is present at the start of the study. A preexisting condition should be recorded as an adverse event if the frequency, intensity, or the character of the condition worsens during the study period.

General Physical Examination Findings
At screening, any clinically significant abnormality should be recorded as a preexisting condition. At the end of the study, any new clinically significant findings/abnormalities that meet the definition of an adverse event must also be recorded and documented as an adverse event.

Post-study Adverse Event
All unresolved adverse events should be followed by the investigator until the events are resolved, the subject is lost to follow-up, or the adverse event is otherwise explained. At the last scheduled visit, the investigator should instruct each subject to report any subsequent event(s) that the subject, or the subject's personal physician, believes might reasonably be related to participation in this study. The investigator should notify the study sponsor of any death or adverse event occurring at any time after a subject has discontinued or terminated study participation that may reasonably be related to this study. The sponsor should also be notified if the investigator should become aware of the development of cancer or of a congenital anomaly in a subsequently conceived offspring of a subject that has participated in this study.

Abnormal Laboratory Values
A clinical laboratory abnormality should be documented as an adverse event if <u>any one of the following</u> conditions is met:

- The laboratory abnormality is not otherwise refuted by a repeat test to confirm the abnormality
- The abnormality suggests a disease and/or organ toxicity
- The abnormality is of a degree that requires active management; e.g. change of dose, discontinuation of the drug, more frequent follow-up assessments, further diagnostic investigation, etc.

Hospitalization, Prolonged Hospitalization or Surgery
Any adverse event that results in hospitalization or prolonged hospitalization should be documented and reported as a serious adverse event unless specifically instructed otherwise in this protocol. Any condition responsible for surgery should be documented as an adverse event if the condition meets the criteria for and adverse event.

Neither the condition, hospitalization, prolonged hospitalization, nor surgery are reported as an adverse event in the following circumstances:

- Hospitalization or prolonged hospitalization for diagnostic or elective surgical procedures for a preexisting condition. Surgery should ***not*** be reported as an outcome of an adverse event if the purpose of the surgery was elective or diagnostic and the outcome was uneventful.
- Hospitalization or prolonged hospitalization required to allow efficacy measurement for the study.
- Hospitalization or prolonged hospitalization for therapy of the target disease of the study, unless it is a worsening or increase in frequency of hospital admissions as judged by the clinical investigator.

7.2 Reportable Adverse Events

For this study, only those events that are related to participation in the study must be reported. This includes events related to:

- Cytosponge administration
- Questionnaire administration
- Any adverse event that may be **related** to participation in this study or use of the Cytosponge device (possibly, probably, or definitely related)
- Any adverse event in which the Cytosponge may **have caused or contributed** to the event.
- Any event required to be reported to the FDA and/or manufacturer per 21CFR803 including:
    - Device-related deaths;
    - Device-related serious injuries.

****Deaths due to the expected progression of disease do not need to be reported as adverse events but should be reported as an outcome for the patient.*

7.2.1 Reporting Timeline

Serious adverse events (meeting the definition of a reportable AE) or unanticipated problems involving risk to subjects or others must be reported within **24 hours** of learning of the event. To report such events, sites must complete the Reportable Event electronic case report form. In addition to completion of the eCRF, sites must also fax or email the form to +1 919 843-2508 or cedas@med.unc.edu. If for any reason the form cannot be completed within 24 hours, a phone call should be made to the lead site +1 919 966-7655 to meet the reporting timeline. In the case of a telephone report, sites must still complete the electronic reportable event form at the earliest possible opportunity, and no later than 72 hours following learning of the event.

All other reportable events should be reported within **15 days** of learning of the event.

7.2.2 Recording of Adverse Events

At each contact with the subject, the investigator must seek information on adverse events by specific questioning and, as appropriate, by examination. Information on all adverse events should be recorded immediately in the source document, and also in the appropriate adverse event module of the electronic case report form (eCRF). All clearly related signs, symptoms, and abnormal diagnostic procedures results should recorded in the source document, though should be grouped under one diagnosis.

All adverse events occurring during the study period must be recorded. The clinical course of each event should be followed until resolution, stabilization, or until it has been determined that the study treatment or

participation is not the cause. Serious adverse events possibly related to the study that are still ongoing at the end of the study period must be followed up to determine the final outcome. Any serious adverse event that occurs after the study period and is considered to be possibly related to the study procedures or study participation should be recorded and reported immediately.

7.2.3 Reporting Adverse Events to Lead Site

All adverse events that meet the criteria of a "reportable" adverse event as described in section 7.2 above must be reported by completing the electronic reportable event form in the study database.

All events will be reported using the National Cancer Institute's Common Terminology Criteria for Adverse Events (CTCAE) version 4.0. A quick reference guide can be accessed from the following website: http://www.acrin.org/Portals/0/Administration/Regulatory/CTCAE_4.02_2009-09-15_QuickReference_5x7.pdf

7.2.3.1 Initial Report

If a patient experiences an event that should be reported as described in section 7.2 the site should complete and submit a reportable event form.

If a patient experiences more than one event, sites should report each event using a separate reportable event form.

7.2.3.2 Follow-Up Reports

All reported adverse events should be followed until resolved or a reason documented if resolution will not occur. Any new information or updates to a previously reported event should be reported as a follow-up to the event. To report a follow-up to an event, sites will update the previously completed electronic reportable event CRF.

7.2.4 Reporting Adverse Events to Local EC/IRBs

Investigators must conform to the adverse event reporting timelines, formats and requirements of the various entities to which they are responsible. All investigators are responsible for safety reporting to their local institutional review board (IRB) or ethics committee (EC). Investigators are responsible for complying with their local EC/IRB's reporting requirements, though must submit the required reports to their IRB no later than 10 working days. Copies of each report and documentation of IRB notification and receipt will be kept in the investigator's files. The definition of a reportable event for a local EC/IRB may not be the same as the definition used for this pilot study.

7.2.5 Notifying the FDA

The facility/institution and device manufacturer are required to report events to the FDA as defined per 21CFR803 (medical device reporting).

1. *Reports of death*. Facilities must submit a report to the FDA as soon as practicable but no more than 10 work days after the day that you become aware of information, from any source, that reasonably suggests that the device has or may have caused or contributed to the death of a patient of your facility. Facilities and institutions must report the following to the FDA via an FDA MEDWATCH Form 3500A (this form can be completed on paper or submitted electronically. You may obtain this form from http://www.fda.gov/medwatch/getforms.htm.
2. *Reports of serious injury*. Facilities must submit a report to the manufacturer of the device no later than 10 work days after the day that you become aware of information, from any source, that reasonably suggests that a device has or may have caused or contributed to a serious injury to a patient of your facility.

Facilities and manufacturers are responsible for all other FDA reporting requirement per 21CFR803 including semi-annual reporting. For additional guidance on reporting to the FDA including where to send reports please visit:

http://www.fda.gov/MedicalDevices/Safety/ReportaProblem/default.htm.

To review 21CFR803 please visit:
http://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/CFRSearch.cfm?CFRPart=803&showFR=1

7.2.6 Lead Site Reporting to Participating Investigators

It is the responsibility of the lead site (UNC) to notify all participating investigators of any adverse event associated with the study that is both serious and unexpected.

7.3 Medical Monitoring

It is the responsibility of the Principal Investigator to oversee the safety of the study at his/her site. This safety monitoring will include careful assessment and appropriate reporting of adverse events as noted above, as well as the construction and implementation of a site data and safety-monitoring plan (see section 9 Auditing, Monitoring and Inspecting). Medical monitoring will include a regular assessment of the number and type of serious adverse events.

8 Data Handling and Record Keeping

8.1 Confidentiality

Information about study subjects will be kept confidential and managed according to the requirements of the Health Insurance Portability and Accountability Act of 1996 (HIPAA). Those regulations require a signed subject authorization informing the subject of the following:

- What protected health information (PHI) will be collected from subjects in this study
- Who will have access to that information and why
- Who will use or disclose that information
- The rights of a research subject to revoke their authorization for use of their PHI.

In the event that a subject revokes authorization to collect or use PHI, the investigator, by regulation, retains the ability to use all information collected prior to the revocation of subject authorization. For subjects that have revoked authorization to collect or use PHI, attempts should be made to obtain permission to collect at least vital status (i.e. that the subject is alive) at the end of their scheduled study period.

8.2 Source Documents

Source data is all information, original records of clinical findings, observations, or other activities in a clinical trial necessary for the reconstruction and evaluation of the trial. Source data are contained in source documents. Examples of these original documents, and data records include: hospital records, clinical and office charts, laboratory notes, memoranda, subjects' diaries or evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, copies or transcriptions certified after verification as being accurate and complete, microfiches, photographic negatives, microfilm or magnetic media, x-rays, subject files, and records kept at the pharmacy, at the laboratories, and at medico-technical departments involved in the clinical trial.

8.3 Case Report Forms

This study will utilize electronic case report forms (eCRFs). The study case report form (eCRF) is the primary data collection instrument for the study. All data requested on the CRF must be recorded. All missing data must be explained. If a space on the CRF is left blank because the procedure was not done or the question was not asked, this should be documented in the comments field.

8.3.1 Case Report Form Completion Table

Form	Screening/Enrollment	Follow-Up	PRN (As Needed)
Enrollment CRF	X		
Follow-Up CRF		X	
Repeat Administration CRF			X
Reportable Event CRF			X
Change of Status CRF			X

8.4 Records Retention

The data compiled in this study will be stored for a period of at least 2 years following study termination.

9 Study Monitoring, Auditing, and Inspecting

9.1 Auditing and Inspecting

The investigator will permit study-related monitoring, audits, and inspections by the EC, IRB, the sponsor, the lead site, government regulatory bodies, and University compliance and quality assurance groups of all study related documents (e.g. source documents, regulatory documents, data collection instruments, study data etc.). The investigator will ensure the capability for inspections of applicable study-related facilities (e.g. pharmacy, diagnostic laboratory, etc.).

Participation as an investigator in this study implies acceptance of potential inspection by government regulatory authorities and applicable University compliance and quality assurance offices.

10 Ethical Considerations

This study is to be conducted according to the International Conference on Harmonisation Good Clinical Practice (ICH/GCP) as well as US federal regulations (21 CFR parts 11, 50, 56, and 312, and 45CFR46), as well as all applicable local and state government regulations and Institutional research policies and procedures.

This protocol and any amendments will be submitted to a properly constituted independent Ethics Committee (EC) or Institutional Review Board (IRB), in agreement with local legal prescriptions, for formal approval of the study conduct. The decision of the EC/IRB concerning the conduct of the study will be made in writing to the investigator and a copy of this decision will be provided to the sponsor before commencement of this study. The investigator should provide a list of EC/IRB members and their affiliate to the sponsor.

All subjects for this study will be provided a consent form describing this study and providing sufficient information for subjects to make an informed decision about their participation in this study. Sample consent forms will be provided by the lead site. These consent forms include a consent for the study as well as a consent for storage of samples for future use. All consent forms will be submitted with the protocol for review and approval by the EC/IRB for the study. The formal consent of a subject, using the EC/IRB-approved consent form, must be obtained before that subject undergoes any study procedure. The consent forms must be signed by the subject or legally acceptable surrogate, and the investigator-designated research professional obtaining the consent.

11 Study Finances

11.1 Funding Source

This pilot study is funded by the National Institutes of Health (NIH) and *Covidien* GI Solutions now part of Medtronic.

11.2 Conflict of Interest

Any investigator who has a conflict of interest with this study (patent ownership, royalties, or financial gain greater than the minimum allowable by their institution, etc.) must have the conflict reviewed by a properly constituted Conflict of Interest Committee with a Committee-sanctioned conflict management plan that has been reviewed and approved by the study sponsor prior to participation in this study. All University of North Carolina investigators will follow the University conflict of interest policy.

12 Publication Plan

Neither the complete nor any part of the results of the study carried out under this protocol, nor any of the information provided by the sponsor for the purposes of performing the study, will be published or passed on to any third party without the consent of the lead investigator, Dr. Nicholas Shaheen. Any investigator involved with this study is obligated to provide the lead investigator with complete test results and all data derived from the study.

13 References

1. Pohl H, Welch HG. The role of overdiagnosis and reclassification in the marked increase of esophageal adenocarcinoma incidence. *Journal of the National Cancer Institute*. 2005;97(2):142–6.

2. Shaheen N, Ransohoff DF. Gastroesophageal reflux, barrett esophagus, and esophageal cancer: Scientific review. *JAMA*. 2002;287(15):1972–1981.

3. Jovov B, Itallie C Van. Claudin-18: a dominant tight junction protein in Barrett's esophagus and likely contributor to its acid resistance. *Am J Physiol Gastrointest Liver Physiol*. 2007;27599:1106–1113.

4. Wang KK, Sampliner RE. Updated guidelines 2008 for the diagnosis, surveillance and therapy of Barrett's esophagus. *The American Journal of Gastroenterology*. 2008;103(3):788–97.

5. Spechler S, Sharma P, Souza R. American Gastroenterological Association technical review on the management of Barrett's esophagus. *Gastroenterology*. 2011;140(3):1–53.

6. Cameron A, Ott B, Payne W. The incidence of adenocarcinoma in columnar-lined (Barrett's) esophagus. *New England Journal of Medicine*. 1985;313(14):857–859.

7. Spechler SJ. Barrett's esophagus. *New England Journal of Medicine*. 2002;346(11):836–842.

8. Winters C, Spurling TJ, Chobanian SJ, et al. Barrett's esophagus: A prevalent, occult complication of gastroesophageal reflux disease. *Gastroenterology*. 1987;92(1):118–124.

9. Ronkainen J, Aro P, Storskrubb T, et al. Prevalence of Barrett's esophagus in the general population: an endoscopic study. *Gastroenterology*. 2005;129(6):1825–31.

10. Locke 3rd GR, Talley NJ, Fett SL, Zinsmeister AR, Melton 3rd LJ. Prevalence and clinical spectrum of gastroesophageal reflux: A population-based study in Olmsted County, Minnesota. *Gastroenterology*. 1997;112(5):1448–1456.

11. Nebel O, Fornes M, Castell D. Symptomatic gastroesophageal reflux: incidence and precipitating factors. *Am J Dig Dis*. 1972;21(11):953–956.

12. Hvid-Jensen F, Pedersen L, Drewes A, Sorensen H, Funch-Jensen P. Incidence of adenocarcinoma among patients with Barrett's esophagus. *New England Journal of Medicine*. 2011;365(15):1375–1383.

13. O'Connor J, Falk G, Richter J. The incidence of adenocarcinoma and dysplasia in Barrett's esophagus: report on the Cleveland Clinic Barrett's Esophagus Registry. *The American Journal of Gastroenterology.* 1999;94(8):2037–2042.

14. Drewitz DJ, Sampliner RE, Garewal HS. The incidence of adenocarcinoma in Barrett's esophagus: a prospective study of 170 patients followed 4.8 years. *The American Journal of Gastroenterology*. 1997;92(2):212–215.

15. Streitz M, Andrews C, Ellis FJ. Endoscopic surveillance of Barrett's esophagus. Does it help? *J Thorac Cardiovasc Surg*. 1993;105(3):383–387.

16. Eloubeidi M, Mason A, Desmon R, El-Serag H. Temporal trends (1973–1997) in survival of patients with esophageal adenocarcinoma in the United States: a glimmer of hope? *The American Journal of Gastroenterology*. 2003;98(7):1627–1633.

17. Shaheen NJ, Provenzale D, Sandler RS. Upper endoscopy as a screening and surveillance tool in esophageal adenocarcinoma: a review of the evidence. *The American Journal of Gastroenterology*. 2002;97(6):1319–27..

18. Spechler SJ, Sharma P, Souza RF, Inadomi JM, Shaheen NJ. American Gastroenterological Association medical position statement on the management of Barrett's esophagus. *Gastroenterology*. 2011;140(3):1084–91.

19. Sonnenberg A, Amorosi SL, Lacey MJ, Lieberman D a. Patterns of endoscopy in the United States: analysis of data from the Centers for Medicare and Medicaid Services and the National Endoscopic Database. *Gastrointestinal Endoscopy*. 2008;67(3):489–96.

20. Russo A, Elixhauser A, Steiner C, Wier L. Hospital-Based Ambulatory Surgery, 2007. *Statistical Brief #86*. 2010:1–16.

21. Bulsiewicz WJ, Kim HP, Dellon ES, et al. Safety and Efficacy of Endoscopic Mucosal Therapy With Radiofrequency Ablation for Patients With Neoplastic Barrett's Esophagus. *Clinical Gastroenterology and Hepatology : The Official Clinical Practice Journal of the American Gastroenterological Association*. 2012.

22. Pouw RE, Wirths K, Eisendrath P, et al. Efficacy of radiofrequency ablation combined with endoscopic resection for barrett's esophagus with early neoplasia. *Clinical Gastroenterology and Hepatology*. 2010;8(1):23–9.

23. Pouw RE, Gondrie JJ, Sondermeijer CM, et al. Eradication of Barrett esophagus with early neoplasia by radiofrequency ablation, with or without endoscopic resection. *Journal of Gastrointestinal Surgery*. 2008;12(10):1627–36.

24. Shaheen NJ, Sharma P, Overholt BF, et al. Radiofrequency ablation in Barrett's esophagus with dysplasia. *New England Journal of Medicine*. 2009;360(22):2277–2288.

25. Titi M, Overhiser A, Ulusarac O, et al. Development of subsquamous high-grade dysplasia and adenocarcinoma after successful radiofrequency ablation of Barrett's esophagus. *Gastroenterology*. 2012;143(3):564–6.e1.

26. Inadomi JM, Somsouk M, Madanick RD, Thomas JP, Shaheen NJ. A cost-utility analysis of ablative therapy for Barrett's esophagus. *Gastroenterology*. 2009;136(7):2101–2114.e1–6.

27. Eisen GM, Baron TH, Dominitz JA, et al. Complications of upper GI endoscopy. *Gastrointestinal Endoscopy*. 2002;55(7):784–793.

28. Lippmann QK, Crockett SD, Dellon ES, Shaheen NJ. Quality of life in GERD and Barrett's esophagus is related to gender and manifestation of disease. *The American Journal of Gastroenterology*. 2009;104(11):2695–703.

29. Falk G, Chittajallu R, Goldblum J, et al. Surveillance of patients with Barrett's esophagus for dysplasia and cancer with balloon cytology. *Gastroenterology*. 1997;112(6):1787–1797.

30. Kadri S, Lao-Sirieix P, O'Donovan M, et al. Acceptability and accuracy of a non-endoscopic screening test for Barrett's oesophagus in primary care: cohort study. *BMJ: British Medical Journal*. 2010.

31. Ross-Innes C, Varghese S, Lao-Sirieix P, Fitzgerald R. Cytosponge: A tool for screening and risk stratification of Barrett's Esophagus. In: *Ninth AACR-Japanese Cancer Association Joint Conference*. 2013:D44.

32. Food and Drug Administration. Information Sheet Guidance For IRBs, Clinical Investigators, and Sponsors: Significant Risk and Nonsignificant Risk Medical Device Studies. 2006.

33. Zhu Y, Terry NG, Woosley JT, Shaheen NJ, Wax A. Design and validation of an angle-resolved low-coherence interferometry fiber probe for in vivo clinical measurements of depth-resolved nuclear morphology. *Journal of Biomedical Optics*. 2011;16(1):011003.

34. Shaheen NJ, Overholt BF, Sampliner RE, et al. Durability of radiofrequency ablation in Barrett's esophagus with dysplasia. *Gastroenterology*. 2011;141(2):460–8.