A Pilot Combined Endoscopic Ultrasound Guided Core Liver Biopsy and Intragastric Balloon Placement for Management of Nonalcoholic Steatohepatitis and Obesity

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Principal Investigator	Barham Abu Dayyeh M.D. MPH Division of Gastroenterology and Hepatology Mayo Clinic 200 First Street SW, Rochester MN 55905 Phone: 507-266-6931 Email: abudayyeh.barham@mayo.edu

Study Product:	Orbera Intragastric Balloon (Apollo Endosurgery, Austin, TX)
Protocol Number: (IRBe)	IRB 15-009262
IDE Number:	G160077

Initial version: **05/03/16** Version **(1.0)**
Revised: **6/23/2017 (3.0)**

Table of Contents

LIST OF ABBREVIATIONS

AE	Adverse Event/Adverse Experience
BMI	Body Mass Index
CFR	Code of Federal Regulations
CRF	Case Report Form
DSMB	Data and Safety Monitoring Board
EUS	Endoscopic Ultrasound
FDA	Food and Drug Administration
GCP	Good Clinical Practice
HIPAA	Health Insurance Portability and Accountability Act
IDE	Investigational Device Exemption
IGB	Intragastric Balloon
IRB	Institutional Review Board
MRE	Magnetic Resonance Elastography
MRS	Magnetic Resonance Spectroscopy
NAFLD	Non-alcoholic Fatty Liver Disease
NAS	Non-alcoholic Steatohepatitis Score
NASH	Non-alcoholic Steatohepatitis
PHI	Protected Health Information
PI	Principal Investigator
SAE	Serious Adverse Event/Serious Adverse Experience
SOP	Standard Operating Procedure
TBWL	Total Body Weight Loss
UADE	Unanticipated Adverse Device Effect

Study Summary

Title	A Pilot Combined Endoscopic Ultrasound Guided Core Liver Biopsy and Intragastric Balloon Placement for Management of Nonalcoholic Steatohepatitis and Obesity
Running Title	EUS and IGB for Management of NASH and Obesity
IRB Protocol Number	15-009262
Phase	Pilot
Methodology	Open Label

Overall Study Duration	60-64 Weeks
Subject Participation Duration	12 months
Objectives	Significant weight loss achieved with the IGB in NASH patients with BMI between 30 – 55 kg/m^2 and early fibrosis based on MRE is associated with ≥ 2 points improvement in NAS and regression of fibrosis on paired EUS guided liver core biopsies.
Number of Subjects	25
Diagnosis and Main Inclusion Criteria	Inclusion criteria to our study include NASH patients with evidence of early fibrosis (excluding advanced fibrosis or cirrhosis) by baseline liver biopsy when available, and/or MRE (magnetic resonance elastography), who are referred for an intragastric balloon placement for weight loss.
Study Device	Orbera Intragastric Balloon (Apollo Endosurgery Austin, TX)
Duration of Exposure	6 months
Statistical Methodology	We have the resources to enroll at total of 20 patients (additional 5 in case of early withdrawal) to this study. With a sample size of 20 we will have 80% power to detect a medium to large (effect size 0.66) improvement in NASH activity scores on paired liver biopsies using a paired t-test with a two sides p-value < 0.05. The table below shows that sample size needed to detect small, medium, or large improvement in NAS scores on paired liver biopsies at 80% power.

1 Introduction

This document is a protocol for a human research study. This study will be carried out in accordance with the procedures described in this protocol, applicable United States government regulations and Mayo Clinic policies and procedures.

1.1 Background

Obesity has reached pandemic proportions.[1] Mirroring this rise in obesity prevalence is a rise in its associated co-morbid conditions including metabolic syndrome and non-alcoholic fatty liver disease (NAFLD). [2] Indeed, NAFLD is thought to afflict about 70% and 50% of obese adults and children, respectively.[3] Of those, about 5% will progress to cirrhosis and end stage liver disease. Non-alcoholic steatohepatitis (NASH) is projected to be the leading cause of liver transplantation in the United States by 2020. [4]

Hepatic fibrosis regardless of etiology progresses to cirrhosis if the etiological agent is not removed or treated. [5] There is growing evidence for reversibility of steatohepatitis and hepatic fibrosis especially in early stages and with significant weight loss. [5] In a seminal study of 293 patients with histologically proven NASH who had life-style modification to reduce their weight over 52 weeks, 72 (25%) achieved resolution of steatohepatitis, 138 (47%) had reductions in nonalcoholic fatty liver disease activity score (NAS), and 56 (19%) had regression of fibrosis. [5] The degree of weight loss was independently associated with improvements in all NASH-related histologic parameters with all patients who lost ≥ 10% of their total body weight (%TBWL) had reductions in NAS, 90% had resolution of NASH, and 45% had regression of fibrosis, compared to 32% (reduction in NAS), 10% (resolution of NASH), and 16% (regression of fibrosis) in those who lost ≤5%. Life-style interventions for obesity; however, are associated with poorly sustained effects and relatively weak efficacy. Indeed, in this study less than 10% of the cohort achieved ≥ 10% TBWL with life-style modification alone and the majority (70%) achieved less than 5% TBWL.[5]

The Orbera™ Intragastric Balloon (Apollo Endosurgery, Austin, TX), previously known as the BioEnterics Intragastric Balloon (BIB) (Allergan, Irvine, CA) is an elastic spherical balloon made from silicone, filled with about 600 ml of saline and is placed in the stomach via upper endoscopy. This IGB is designed to stay in place for 6 months prior to endoscopic removal and has been widely used in Europe, South America, and the Middle East with excellent safety profile. In an ASGE Bariatric Endoscopy Task Force systematic review and meta-analysis the pooled %TBWL after the Orbera IGB implantation was 12.3% (95% CI, 7.91–16.73), 13.16% (95% CI, 12.37–13.95), and 11.27% (95% CI, 8.17–14.36) at 3, 6, and 12 months after implantation, respectively.[6] This was based on a review of 55 studies. This balloon is currently FDA approved for use in the United States after a US randomized multicenter pivotal trial demonstrated similar efficacy.

The rates of adverse events after implantation of the Orbera balloon are pooled from a manual review of 68 studies (8500 implantations) and are summarized in Figure 1. Pain and nausea are frequent side-effects after Orbera balloon implantation, occurring in up to 33.7% of subjects. Medications such as proton pump inhibitors, antispasmodic drugs, and antiemetics are usually prescribed prophylactically before, during, and after balloon

placement to prevent or minimize these expected common side effects. Serious side-effects with Orbera balloon are rare with an incidence of migration and gastric perforation of 1.4% and 0.1%, respectively. Most of the reported perforations with the Orbera were in patients who had undergone previous gastric surgeries, which is a contraindication for use in the US.[6]

More particularly, the safety and efficacy of the Orbera intragastric balloon has been previously demonstrated in patients with suspected NASH. A small study randomized 18 obese or overweight patients with histologically proven nonalcoholic steatohepatitis (NASH) to lifestyle modification plus Orbera balloon placement or to lifestyle modification plus a sham procedure. Weight and liver histology were assessed before and 6 months after balloon insertion or the sham procedure. The Orbera balloon placement group had a significantly higher reduction in mean BMI (1.52 vs 0.8; P = .0008) and a superior improvement in nonalcoholic fatty liver disease activity scores at the end of treatment (2 [SD 0.75] vs 4 [SD 2.25]; P = .03).[7] Three other single arm studies demonstrated improvement in liver steatosis by cross sectional imaging and liver biochemical testing with the Orbera balloon in patient with suspected NASH. [8-10]

The detection of early fibrosis with steatohepatitis is critical given the more aggressive nature of disease, which requires a more aggressive treatment plan. However, early fibrosis is difficult to diagnose with non-invasive imaging modalities without the need for a liver biopsy. Although modalities such as transient elastography (Fibroscan) have good diagnostic accuracy for cirrhosis, its performance is suboptimal for earlier stages of fibrosis. [11] Several studies have established that liver magnetic resonance elastography (MRE) is an accurate technique for the detection of earlier stages of liver fibrosis and steatosis. MRE has therefore emerged as a potential alternative to liver biopsy in establishing the diagnosis of NASH. [12, 13] **However, despite MRE accuracy in establishing the diagnosis, there are no studies to demonstrate its sensitivity to change in response to an intervention such as weight loss. Furthermore, there are no standardized validated criteria to objectively assess improvement in NASH and fibrosis with MRE as exit for liver biopsy, which remains the gold standard in clinic trial evaluating the impact of intervention on NASH**.[14]

Endoscopic ultrasound guided liver biopsy has been shown to be very safe and capable of adequately and reproducibly sampling widely separated liver regions under direct guidance; thus yielding more representative and suitable liver tissue for the staging and histopathological follow-up of NASH, especially in the confounds of clinical trial.[15-17] EUS-guided core liver biopsy can produce equal or higher number of complete portal tracts compared to traditional percutaneous approach, while avoiding sampling error in a paired liver biopsy design (pre and post intervention), which is currently a pitfall of the precautious liver biopsy approach given the patchy nature of non-alcoholic fatty liver disease. [18]

Given the limited data of the impact of weight loss produced by the intragastric balloon on NASH with early fibrosis, we propose a prospective study to evaluate the impact of weight loss produced the Orbera intragastric balloon on objective NASH histopathological parameters.

1.2 Investigational Device

The Orbera (Apollo Endosurgery, Austin Tx) is an elastic spherical balloon made of silicone, filled with 450 to 700 mL of saline solution, and is FDA approved for weight loss in adult US patients with body mass index between 30 to 40 kg/m^2. The deflated balloon comes preloaded on a catheter, which is blindly advanced transorally into the stomach. An endoscope is then advanced alongside it to ensure accurate placement of the balloon in the fundus. Under direct visualization, the balloon is then inflated by injecting saline solution through the external portion of the catheter. The Orbera balloon is currently used in many countries outside the United States and is typically implanted for 6 months and then retrieved endoscopically.

1.3 Preclinical Data

See PMA study of the Orbera intragastric balloon

1.4 Clinical Data to Date

The Orbera™ Intragastric Balloon (Apollo Endosurgery, Austin, TX), previously known as the BioEnterics Intragastric Balloon (BIB) (Allergan, Irvine, CA) is an elastic spherical balloon made from silicone, filled with about 600 ml of saline and is placed in the stomach via upper endoscopy. This IGB is designed to stay in place for 6 months prior to endoscopic removal and has been widely used in Europe, South America, and the Middle East with excellent safety profile. In an ASGE Bariatric Endoscopy Task Force systematic review and meta-analysis the pooled %TBWL after the Orbera IGB implantation was 12.3% (95% CI, 7.91–16.73), 13.16% (95% CI, 12.37–13.95), and 11.27% (95% CI, 8.17–14.36) at 3, 6, and 12 months after implantation, respectively.[6] This was based on a review of 55 studies. This balloon is currently FDA approved for use in the United States after a US randomized multicenter pivotal trial demonstrated similar efficacy.

The rates of adverse events after implantation of the Orbera balloon are pooled from a manual review of 67 studies (8500 implantations) and are summarized in Figure 1. Pain and nausea are frequent side-effects after Orbera balloon implantation, occurring in up to 33.7% of subjects. Medications such as proton pump inhibitors, antispasmodic drugs, and antiemetics are usually prescribed prophylactically before, during, and after balloon placement to prevent or minimize these expected common side effects. Serious side-effects with Orbera balloon are rare with an incidence of migration and gastric perforation of 1.4% and 0.1%, respectively. Most of the reported perforations with the Orbera were in patients who had undergone previous gastric surgeries, which is a contraindication for use in the US.[6]

More particularly, the safety and efficacy of the Orbera intragastric balloon has been previously demonstrated in patients with suspected NASH. A small study randomized 18 obese or overweight patients with histologically proven nonalcoholic steatohepatitis (NASH) to lifestyle modification plus Orbera balloon placement or to lifestyle modification plus a sham procedure. Weight and liver histology were assessed before and 6 months after balloon insertion or the sham procedure. The Orbera balloon placement group had a significantly

higher reduction in mean BMI (1.52 vs 0.8; P = .0008) and a superior improvement in nonalcoholic fatty liver disease activity scores at the end of treatment (2 [SD 0.75] vs 4 [SD 2.25]; P = .03).[7] Three other single arm studies demonstrated improvement in liver steatosis by cross sectional imaging and liver biochemical testing with the Orbera balloon in patient with suspected NASH. [8-10]

1.5 Study Rationale and Risk/Benefits

1.5.1 Study Rationale

NASH with early fibrosis is serious condition that afflicts a significant portion of patients with obesity with the potential to progress to cirrhosis. Weight loss of ≥ 10 % TBWL has been definitively shown to be an effective treatment of NASH with regression of early fibrosis, however, this magnitude of weight loss is only achieved in the minority (≤ 10%) of patients with life style modification. The IGB achieves a mean %TBWL weight loss of about 13% at 6 months, and has previously shown efficacy in management of NASH in a small cohort outside the US. In 8 patients with biopsy proven NASH and obesity, who underwent the Orbera IGB placement and paired percutaneous liver biopsies before and 6 months after Orbera placement, the NAS significantly improved with IGB placement group compared to the life-style intervention alone (4 ±2.25 vs. 2 ± 0.75 vs. P = .03). However, this study demonstrated no improvement in fibrosis scores as only the minority of the cohort had true early fibrosis. From a recent seminal paper we know that achieving ≥ 10% TBWL is clearly associated with regression in fibrosis, although this magnitude of weight loss is difficult to achieve with life-style modification only. Thus, in this protocol we will be selecting patients with NASH and early fibrosis diagnosed by MRE for placement of intragastric balloon and concomitant EUS guide liver biopsies at the time of balloon placement and removal (6 months after) to evaluate the impact of weight loss achieved by the Orbera IGB on NAS and fibrosis regression.

The use of paired EUS guided liver biopsies is crucial for our study design as despite MRE accuracy in establishing the diagnosis of NASH with early fibrosis for patient inclusion to our study, there are no studies to demonstrate its sensitivity to change in response to an intervention such as weight loss. Furthermore, there are no standardized validated criteria to objectively assess improvement in NASH and fibrosis with MRE as exists for liver biopsy, which remains the gold standard in clinic trial evaluating the impact of intervention on NASH.

Spectroscopic and biochemical hepatic fat quantitative assessment pre- and post-intervention – Rationale:

The degree of lipid saturation and polyunsaturation can be measured noninvasively by proton magnetic resonance spectroscopy [19]. This technique adds no cost, as the series can be simultaneously captured during MRE acquisition session, adding few minutes to the procedure. Evidence suggests that depletion of hepatic polyunsaturated fatty acids (PUFAs) reduces hepatic fatty acid oxidation and triglyceride export. Furthermore, it is associated with augmentation in in hepatic fatty acid and triglyceride synthesis, leading to increased triglyceride aggregation. Moreover, this process has been thought to increase inflammation and facilitates the development of steatohepatitis. Indeed, studies have shown that alterations

in tissue lipid saturation may portend sinister clinical outcomes more reliably than simple means of fatness measurements [20].

To the best of our knowledge, no studies have corroborated the sensitivity of H-MRS detection of SI and PUI changes, as well as other indices of hepatic lipid composition, with weight loss, and its correlation to decrease in total steatosis, changes in liver stiffness, as well as changes in fibrosis scores on histology.

Our current study offers a rare window to accurately examine and capture these changes due to the availability of liver tissue histology pre- and post-intervention. This will allow the determination H-MRS reliability in predicting lipidomic analysis changes in a NASH afflicted liver, obviating the need of invasive testing.

Furthermore, an unmet need in the understanding of nonalcoholic fatty liver disease is the determination of the interactions between the changes in hepatic lipid polyunsaturated fatty acids (PUFA) depletion and liver injury; considering the expected improvement in liver histology parameters with our proposed intervention, our study would offer a unique opportunity to correlate these improvements with PUFA depletion[19, 21, 22].

1.5.2 Anticipated Risks

The rates of adverse events after implantation of the Orbera balloon are pooled from a manual review of 68 studies (8500 implantations) and are summarized in Figure 1. Pain and nausea are frequent side-effects after Orbera balloon implantation, occurring in up to 33.7% of subjects. Medications such as proton pump inhibitors, antispasmodic drugs, and antiemetics are usually prescribed prophylactically before, during, and after balloon placement to prevent or minimize these expected common side effects. Serious side-effects with Orbera balloon are rare with an incidence of migration and gastric perforation of 1.4% and 0.1%, respectively. Most of the reported perforations with the Orbera were in patients who had undergone previous gastric surgeries, which is a contraindication for use in the

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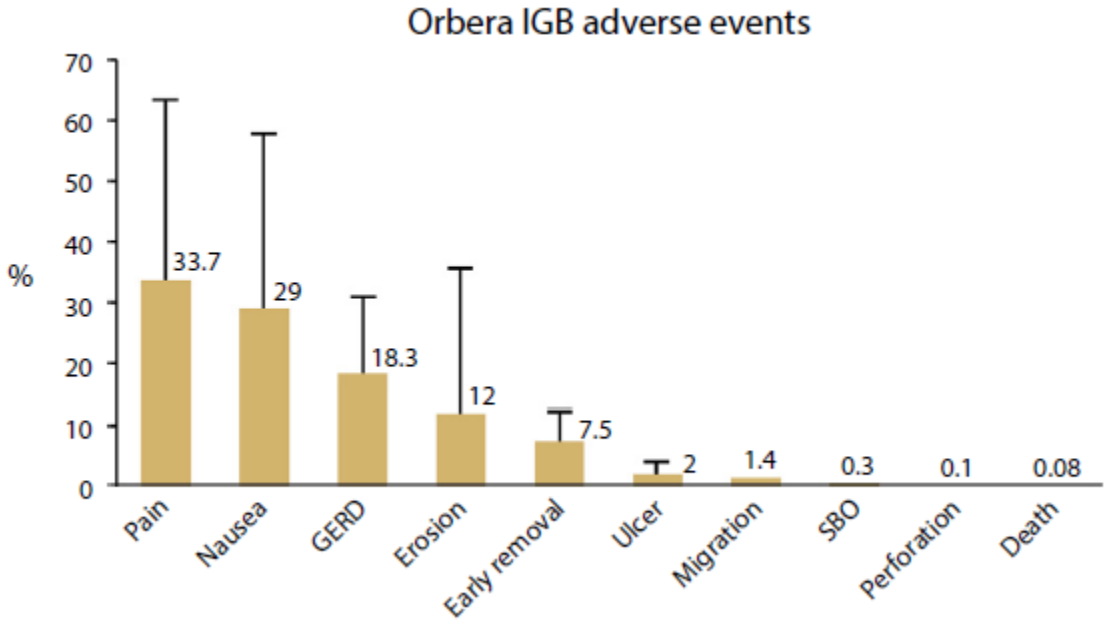


Figure 1. Pooled Orbera Complications from 68 studies (8500 patients) [6]

More particularly, the safety of the Orbera intragastric balloon has been previously demonstrated in patients with suspected NASH. A small study randomized 18 obese or overweight patients with histologically proven nonalcoholic steatohepatitis (NASH) to lifestyle modification plus Orbera balloon placement or to lifestyle modification plus a sham procedure. Weight and liver histology were assessed before and 6 months after balloon insertion or the sham procedure. The Orbera balloon placement group had a significantly higher reduction in mean BMI (1.52 vs 0.8; P = .0008) and a superior improvement in nonalcoholic fatty liver disease activity scores at the end of treatment (4 [SD 2.5] vs 2 [SD 20.75]; P = .03), with no increased incidence of adverse events.[7] Three other single arm studies demonstrated improvement in liver steatosis by cross sectional imaging and liver biochemical testing with the Orbera balloon in patient with suspected NASH with excellent safety profile in these cohorts with suspected NASH. [8-10]

EUS guided core liver biopsy has been established as a safe, adequate, and cost effective mean to obtain diagnostic liver tissue to diagnose and stage chronic liver disease. In more than 200 patients reported in the literature, EUS guided liver biopsy was capable in providing similar or more adequate liver sample than percutaneous or trans-jugular approach with no reported incidence of any adverse events. [15-17, 23] Our EUS group has conducted over 800 EUS guided liver biopsies, with no reported incidence of immediate or delayed adverse events. In this protocol using paired EUS guided liver biopsies at the time of balloon insertion and removal will be cost effective, safe, only add few extra minutes to the procedure, and will allow us to minimize sampling error and isolate the effects of weight loss achieved by the intragastric balloon on gold standard histological endpoints.

1.5.3 Potential Benefits

Hepatic fibrosis regardless of etiology progresses to cirrhosis if the etiological agent is not removed or treated. [5] There is growing evidence for reversibility of steatohepatitis and hepatic fibrosis especially in early stages and with significant weight loss. [5] In a seminal study of 293 patients with histologically proven NASH who had life-style modification to reduce their weight over 52 weeks, 72 (25%) achieved resolution of steatohepatitis, 138 (47%) had reductions in nonalcoholic fatty liver disease activity score (NAS), and 56 (19%) had regression of fibrosis. [5] The degree of weight loss was independently associated with improvements in all NASH-related histologic parameters with all patients who lost ≥ 10% of their total body weight (%TBWL) had reductions in NAS, 90% had resolution of NASH, and 45% had regression of fibrosis, compared to 32% (reduction in NAS), 10% (resolution of NASH), and 16% (regression of fibrosis) in those who lost ≤5%. Life-style interventions for obesity; however, are associated with poorly sustained effects and relatively weak efficacy. Indeed, in this study less than 10% of the cohort achieved ≥ 10% TBWL with life-style modification alone and the majority (70%) achieved less than 5% TBWL.[5]

The Orbera intragastric balloon is expected to results in 12.3% (95% CI, 7.91–16.73), 13.16% (95% CI, 12.37–13.95), and 11.27% (95% CI, 8.17–14.36) TBWL at 3, 6, and 12 months after implantation, respectively.[6] This was based on a review of 55 studies. This balloon is currently FDA approved for use in the United States after a US randomized multicenter pivotal trial demonstrated similar efficacy. Thus, with the anticipated weight loss seen with the Orbera Intragastric balloon, we expect improvement in histological NASH parameters and regression of fibrosis with the weight loss achieved with the intragastric balloon.

The ability to quantify and capture hepatic fat change, and correlate it to spectroscopic changes could offer important non-invasive means of NASH progression/regression in afflicted individuals. This study would offer a unique opportunity to corroborate the utility of MRS in accurate and reliable evaluation of this parameter.

2 Study Objectives

2.1 Primary Objective

NASH with early fibrosis is serious condition that afflicts a significant portion of patients with obesity with the potential to progress to cirrhosis. Weight loss of ≥ 10 % TBWL has been definitively shown to be an effective treatment of NASH with regression of early fibrosis, however, this magnitude of weight loss is only achieved in the minority (≤ 10%) of patients with life style modification. The IGB achieves as mean %TBWL of 13%. Therefore, we aim to study the impact of weight loss achieved by the Orbera intragastric balloon on NAS and liver fibrosis in a cohort selected by MRE to have NASH with early fibrosis but not cirrhosis. This will be assessed with a paired EUS guided biopsies at the time of balloon placement and removal after six months.

Hypothesis 1: Significant weight loss achieved with the IGB in NASH patients with BMI between 30 – 55 kg/m^2 and fibrosis based on MRE is associated with ≥ 2 points improvement in NAS and regression of fibrosis on paired EUS guided liver core biopsies.

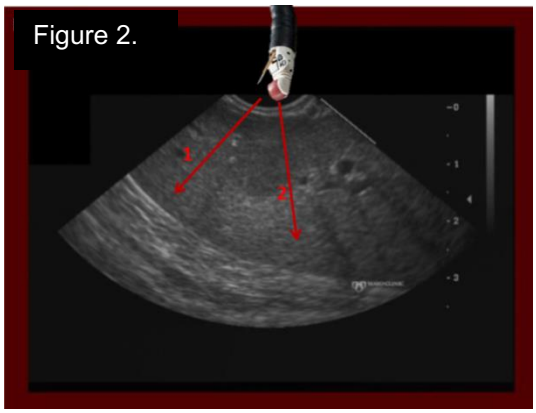
2.2 Secondary Objective

- Evaluate safety and tolerance of the Orbera intragastric balloon and concomitant paired EUS guided liver biopsy at time of balloon placement and removal.
- Obtaining lipid saturation (SI) and polyunsaturation (PUI) indices via H-MRS technology pre- and post-intervention
- Obtaining hepatic triglycerides content (HTGC) via H-MRS technology pre- and post-intervention.
- Obtaining percent unsaturated fatty acyl chains present in this cohort of NASH patients, as well as changes that occur in the setting of weight loss.
- Obtaining percent polyunsaturated fatty acyl chains present in this cohort of NASH patients, as well as changes that occur in the setting of weight loss.
- Measurement of hepatic lipid composition by needle biopsy and lipidomic analysis techniques before and after weight loss.
- Correlation of MRS obtained lipid composition to lipidomic analysis composition.

3 Study Design

3.1 General Design

Patients with suspicion for NASH at our center undergo a combined MRE/MRS (magnetic resonance elastography/magnetic resonance spectroscopy) sequence to classify their disease according to four stages. These stages are numbered from 1-4 with stage 4 signifying more advanced fibrosis. The MRE/MRS sequence also quantifies hepatic fat content. Those with stage 1-3 fibrosis will be referred for IGB placement for weight loss as a management strategy for NASH. Once referred to the endoscopic bariatric clinic a multidisciplinary team that includes endocrinology, gastroenterology, psychology, and nutrition will evaluate them. If found to be appropriate candidates, they will be enrolled in this study by a member of the study team who will discuss the study protocol, risks, benefits, and alternatives with patients and have them sign an informed consent. Patient will undergo intragastric balloon placement and an EUS guided liver biopsy during the same endoscopy session will be performed under monitored anesthesia sedation at our outpatient endoscopy unit. The EUS guided liver biopsy will be performed before balloon insertion from the left liver lobe under EUS guidance using a 22 gauge core needle (Sharkcore, Covidien, Minneapolis, MN). Three passes will be performed from two separate liver segments (Figure 2). An additional pass may be performed at the discretion of the PI if the three performed passes do not yield enough tissue on visual inspection of the core to ensure adequate histology analysis. The maximum number of passes will not exceed four. Doppler ultrasound will be used to exclude intervening vessels. Two samples will be submitted for histopathology review in formalin, and one or two will be submitted for total fat



quantification analysis. After the EUS guided liver biopsy (usually take 10 minutes to perform), the Orbera intragastric balloon will be advanced to the stomach under endoscopy guidance and inflated with 600 ml of normal saline. Patient transient nausea and pain symptoms will be managed with few days course of antiemetic and antispasmodics as per our clinical protocol, and patients will be required to take a daily proton pump inhibitor if not already taking. Patient will then undergo a moderate intensity life-style intervention consisting of daily phone call for the first week (nurse and or study coordinator) then monthly group visits with a health-care coach / nutrition to administer the life-style intervention program over 6 months period as per our clinical protocol for the intragastric balloon. At 6 months patients will return for balloon removal at our complex endoscopy unit under monitored anesthesia care. The balloon will be removed using the standard balloon retrieval kit approved by the FDA. After balloon removal a second EUS guided liver biopsy from two separate liver segments similar to the first biopsies site will be obtained with three or four passes with the 22 gauge core liver biopsy needle (Sharckcore, Covidien, Minneapolis, MN). The anticipated procedural length for both the EUS guided liver biopsy and balloon insertion and removal is less than 6 minutes. After balloon removal, all patients will be offered additional six months of moderate life-style intervention program administered as monthly group or individual visits with a health care coach to emphasize healthy lifestyle intervention and behaviors. Weight, height, waist circumference, and demographic information will be collected a baseline, time of balloon removal (6 months), and at 12 months (6months after balloon removal). Of note the cost of balloon placement, removal, medications to manage symptoms after balloon placement, EUS guided liver biopsy and pathologic interpretation, and the 12 months life-style intervention program will be covered by the study. The initial evaluation at the obesity clinic prior to enrollment in the study will be part regular clinical care.

Of note in this protocol will include patients with diagnosis of NASH with fibrosis (stage 1-3) on MRE/MRS (magnetic resonance elastography/magnetic resonance spectroscoy), which has demonstrated high accuracy in making the diagnosis, but has unknown sensitivity and specificity to detect change over time and has no standardized validated criteria to objectively assess improvement in NASH and fibrosis as exists for liver biopsy, which remains the gold standard in clinic trial evaluating the impact of intervention on NASH. **In the rare instance that the MRE result was falsely positive and the patient was not found to have NASH on liver biopsy. The patient will continue with balloon therapy for weight loss, but we will not repeat liver biopsy at 6 months, since the balloon is indicated for obesity with or without NASH with clear health benefits to weight loss in this cohort.** The balloon cost will remain covered by the study under these rare circumstances.

3.2 Primary Study Endpoint

Number of subjects with ≥ 2 points improvement in NASH activity scores on paired liver biopsies before and 6 months after intragastric balloon placement.

3.3 Secondary Study Endpoints

- Regression of fibrosis on paired EUS guided liver core biopsies.

- Percent total body weight loss at 6 months after Orbera balloon placement.
- Obtaining lipid saturation (SI) and polyunsaturation (PUI) indices via H-MRS technology pre- and post-intervention
- Obtaining hepatic triglycerides content (HTGC) via H-MRS technology pre- and post-intervention.
- Obtaining percent unsaturated fatty acyl chains present in this cohort of NASH patients, as well as changes that occur in the setting of weight loss.
- Obtaining percent polyunsaturated fatty acyl chains present in this cohort of NASH patients, as well as changes that occur in the setting of weight loss.
- Measurement of hepatic lipid composition by needle biopsy and lipidomic analysis techniques before and after weight loss.
- Correlation of MRS obtained lipid composition to lipidomic analysis composition

3.4 Primary Safety Endpoints

- Incidence of all serious adverse events including unanticipated adverse effects (balloon migration, small bowel obstruction, luminal GI perforation, ulcer formation, liver abscess formation, bleeding, biliary obstruction or injury, aspiration, infection, abdominal pain requiring hospitalization, nausea and dehydration requiring intravenous fluids, and death). All adverse events will be captured and graded for severity using standardized scales. [24]

- Early balloon intolerance requiring removal.

- Balloon deflation rates (see Orbera PMA)

4 Subject Selection, Enrollment and Withdrawal

We plan to enroll patients with obesity and NASH with fibrosis but no advanced fibrosis or cirrhosis as diagnosed by MRE/MRS . We are expanding the cohort of patient with obesity to include patients with BMI between 30 to 55 kg/m^2 given recent meta-analysis demonstrating similar weight loss with the Orbera intragastric balloon in obese patients with higher BMIs (Figure 3).[6] Given that more than 10% TBWL is associated with improvement in NASH [5], we feel that including patients with higher BMI will be important in this pilot study.

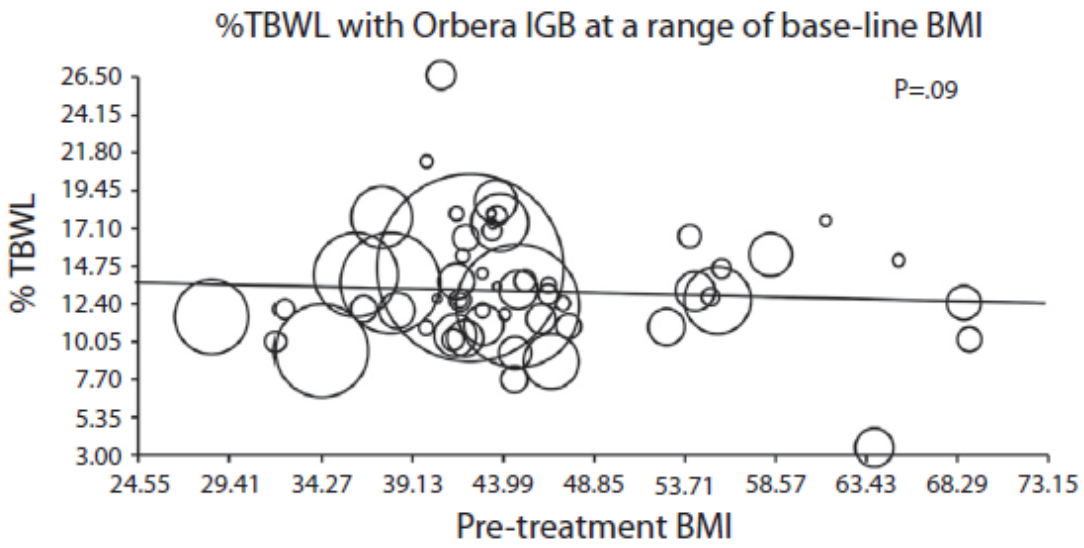


Figure 3. Meta-regression linear plot depicting the best-fit regression line of the association between baseline body mass indexes (BMIs) and percentage of excess weight loss (%EWL) at 6 months after Orbera intragastric balloon (IGB) implantation. The sample size of individual studies is proportional to the diameter of the circle by which it is represented on the graph. [6]

4.1 Inclusion Criteria

- Age 21 – 70 years
- Male and Female
- BMI between 30 to 55 kg/m^2
- NASH with fibrosis (excluding advanced fibrosis or cirrhosis). This is determined by a baseline liver biopsy when available showing stage 1-3 fibrosis, but *not* stage 4 (advance fibrosis or cirrhosis), and / or magnetic resonance elastography (MRE) showing a liver stiffness measurement range that spans a score of ≥ 3.0kPa and with a mean liver stiffness value of < 5 kPa. Both of these tests are valid for study inclusion within 6 months of enrollment. When both a biopsy and MRE are available prior to study enrollment the fibrosis designation will be based on the gold standard histology.
- Subjects will have attempted and failed more conservative weight reduction alternatives, such as supervised diet, exercise and behavior modification programs (per Orbera's approved PMA: P140008)
- Able to provide a written informed consent

4.2 Exclusion Criteria

- Age <21 or >70 years
- Pregnancy or plan for pregnancy during study period
- Lactation
- Previous history of gastric surgery

- Hiatus hernia > 5cm
- Current or recent (within 6 months) gastric or duodenal ulcers
- Gastroparesis
- Liver cirrhosis
- Coagulopathy or active use of anticoagulation
- Inflammatory bowel disease involving the upper GI tract.
- Active substance abuse
- Active psychiatric condition deemed as a contraindication by our psychology team evaluation
- Concomitant chronic liver disease other than NASH
- Potential upper gastrointestinal bleeding conditions such as esophageal or gastric varices, congenital or acquired intestinal telangiectasis, or other congenital anomalies of the gastrointestinal tract such as atresias or stenosis
- Hiatal hernia with associated severe or intractable gastro-esophageal reflux symptoms
- Structural abnormality in the esophagus or pharynx such as a stricture or diverticulum that could impede passage of the delivery catheter and/or an endoscope
- Achalasia
- Gastric mass
- Patients receiving aspirin, anti-inflammatory agents, anticoagulants or other gastric irritants, not under medical supervision
- Patients who are unable or unwilling to take prescribed proton pump inhibitor medication for the duration of the device implant.

4.3 Subject Recruitment, Enrollment and Screening

Patients with suspicion for NASH at our center undergo MRE (magnetic resonance elastography) sequence to classify their disease as grade 0 (simple steatosis), grade 1 (steatohepatitis with no fibrosis), grade 2 (steatohepatitis with early fibrosis), grade 3 (steatohepatitis with advance fibrosis/cirrhosis). Those with stage 1-3 fibrosis and interested in weight loss with the intragastric balloon will be referred for the intragastric balloon clinic evaluation, which consist of one day evaluation by nutrition, psychology, and gastroenterology in the obesity clinic. Those who meet the inclusion / exclusion criteria will be consented to participate in the study after discussion of risk, benefits, and alternatives by one of our study team members and a written consent is provided. Those who do not qualify for the study will be offered other alternatives for weight loss as per out clinical protocol. Of note the cost of balloon placement, removal, medications to manage symptoms after balloon placement, EUS guided liver biopsy and pathologic interpretation, and the 12 months life-style intervention program will be covered by the study. The initial evaluation at the obesity clinic prior to enrollment in the study will be part regular clinical care.

4.4 Early Withdrawal of Subjects

4.4.1 When and How to Withdraw Subjects

Study will be stopped:

- 1 death

- 2 or more severe adverse events or unanticipated adverse device effects deemed of enough severity to stop the study by an independent data safety and monitoring board consisting of three Mayo consultants independent from the study

Patient will be withdrawn from the study:

- Subject safety issues, adverse device effects
- Failure of subject to adhere to protocol requirements
- Disease progression to cirrhosis within the time frame of the study
- Subject decision to withdraw from the study (withdrawal of consent)

Replacement of withdrawn subjects:

If a patient is withdrawn from the study for reasons other than severe adverse events, the intragastric balloon will be removed from the patient at a time that works for the patient and within six months of insertion. The patient will be followed as per the standard clinical protocol for the intragastric balloon to ensure safety. Weight and safety data will be collected until the intragastric balloon is removed. After balloon removal subjected will be followed clinically as per our protocol for additional three months to ensure safety. We will replace up to 5 subjects withdrawn for reasons other than severe adverse events.

4.4.2 Data Collection and Follow-up for Withdrawn Subjects

If a patient is withdrawn from the study for reasons other than severe adverse events, the intragastric balloon will be removed from the patient at a time that works for the patient and within six months of insertion. The patient will be followed as per the standard clinical protocol for the intragastric balloon to ensure safety. Weight and safety data will be collected until the intragastric balloon is removed. After balloon removal subjected will be followed clinically as per our protocol for additional three months to ensure safety. We will replace up to 5 subjects withdrawn for reason other than severe adverse events.

5 Study Device

5.1 Description

The Orbera (Apollo Endosurgery, Austin Tx) is an elastic spherical balloon made of silicone, filled with 450 to 700 mL of saline solution, and is FDA approved for weight loss in adult US patients with body mass index between 30 to 40 kg/m^2. The deflated balloon comes preloaded on a catheter, which is blindly advanced transorally into the stomach. An endoscope is then advanced alongside it to ensure accurate placement of the balloon in the fundus. Under direct visualization, the balloon is then inflated by injecting saline solution through the external portion of the catheter. The Orbera balloon is currently used in many countries outside the United States and is typically implanted for 6 months and then retrieved endoscopically.

5.2 Method for Assigning Subjects to Treatment Groups

All enrolled subjects will get the Orbera intragastric balloon and paired EUS guided liver biopsies.

5.3 Preparation and Administration/Implantation of Investigational Device

The intragastric balloon will be used as described by the Orbera PMA application approved by the FDA.

5.4 Subject Compliance Monitoring

Patient will undergo intragastric balloon placement and an EUS guided liver biopsy during the same endoscopy session will be performed under monitored anesthesia sedation at our outpatient endoscopy unit. The EUS guided liver biopsy will be performed before balloon insertion from the left liver lobe under EUS guidance using a 22 gauge core needle (Sharkcore, Covidien, Minneapolis, MN). Three passes will be performed from two separate liver segments (Figure 2). An additional pass may be performed at the discretion of the PI if the three performed passes do not yield enough tissue on visual inspection of the core to ensure adequate histology analysis. The maximum number of passes will not exceed four. Doppler ultrasound will be used to exclude intervening vessels. Two samples will be submitted for histopathology review in formalin, and one or two will be submitted for total fat quantification analysis. After the EUS guided liver biopsy (usually take 10 minutes to perform), the Orbera intragastric balloon will be advanced to the stomach under endoscopy guidance and inflated with 600 ml of normal saline. Patient transient nausea and pain symptoms will be managed with few days course of antiemetic and antispasmodics as per our clinical protocol. Patient will also be required to take a proton pump inhibitor -if not already taking- throughout the 6 months of balloon therapy. Patient will then undergo a moderate intensity life-style intervention consisting of daily phone call for the first week (nurse and or study coordinator) then monthly group visits with a health-care coach / nutrition to administer the life-style intervention program over 6 months period as per our clinical protocol for the intragastric balloon. At 6 months patients will return for balloon removal at our complex endoscopy unit under monitored anesthesia care. The balloon will be removed using the standard balloon retrieval kit approved by the FDA. After balloon removal a second EUS guided liver biopsy from two separate liver segments similar to the first biopsies site will be obtained with three or four passes with the 22 gauge core liver biopsy needle (Sharckcore, Covidien, Minneapolis, MN). The anticipated procedural length for both the EUS guided liver biopsy and balloon insertion and removal is less than 6 minutes. After balloon removal, all patients will be offered additional six months of moderate life-style intervention program administered as monthly group or individual visits with a health care coach to emphasize healthy lifestyle intervention and behaviors. Weight, height, waist circumference, and demographic information will be collected a baseline, time of balloon removal (6 months), and at 12 months (6 months after balloon removal). Of note the cost of balloon placement, removal, medications to manage symptoms after balloon placement, EUS guided liver biopsy and pathologic interpretation, and the 12 months life-style intervention program will be covered by the study. The initial evaluation at the obesity clinic prior to enrollment in the study will be part of regular clinical care. The PI will evaluate all patients within 1 week of balloon placement and removal.

5.5 Prior and Concomitant Therapy

Not applicable

5.6 Packaging and Labeling

Upon receipt of the IGB devices from Apollo Endosurgery, assurance of the investigational device label will be affixed to the packaging of each device. See below:

"CAUTION – Investigational Device. Limited by Federal (or United States) law to investigational use"

5.7 Masking/Blinding of Study

Not applicable

5.8 Receiving, Storage, Distribution and Return

5.8.1 Receipt of Investigational Devices

The Orbera intragastric balloon devices will be shipped to the study coordinator from Apollo Endosurgery in Austin, Texas. Upon receipt of the devices, the study coordinator will perform an inventory and fill out a device accountability log to ensure that all devices are accounted for. Any discrepancies, damaged, or unusable devices in a given shipment will be documented in the study files and the company will be notified. The devices will then be taken by the study coordinator to where they will be stored in the Operating Room Core.

5.8.2 Storage

The Orbera intragastric balloons will be stored with the rest of the supplies that will be stored in the Operating Room Core which is a secure location with limited access to prevent unintended or unauthorized use of the device.

5.8.3 Distribution of Study Device

The Orbera intragstric balloons will be distributed according to the clinical protocol of how clinical intragastric balloons are distributed. The serial number on the Orbera intragastric balloon will be matched up to a subject's medical record number. This de-identified data will then be recorded on the device accountability log using subject number instead of medical record number.

5.8.4 Return or Destruction of Study Device

At routine intervals and at the completion of the study, there will be a reconciliation of devices shipped, devices utilized, and devices remaining. This reconciliation will be logged on the Device Accountability form, signed and dated. Any discrepancies noted will be documented, the sponsor-investigator will be notified and an investigation will be conducted to determine the cause of the discrepancy. Devices destroyed on site will be documented in the study files.

6 Study Procedures

Patients with suspicion for NASH at our center undergo a combined MRE/MRS (magnetic resonance elastography/magnetic resonance spectroscopy) sequence to classify their disease according to four stages. These stages are numbered from 1-4 with stage 4 signifying more advanced fibrosis. The MRE/MRS sequence also quantifies hepatic fat content. Those with stage 1-3 fibrosis and interested in weight loss with the intragastric

balloon will be referred for the intragastric balloon clinic evaluation, which consist of one day evaluation by nutrition, psychology, and gastroenterology in the obesity clinic. Those who meet the inclusion / exclusion criteria will be consented to participate in the study after discussion of risk, benefits, and alternatives by one of our study team members and a written consent is provided. Following this, patients will be scheduled for their intragastric balloon placement and EUS guided liver biopsy.

Patients will undergo intragastric balloon placement and an EUS guided liver biopsy during the same endoscopy session will be performed under monitored anesthesia sedation at our outpatient endoscopy unit. The EUS guided liver biopsy will be performed before balloon insertion from the left liver lobe under EUS guidance using a 22 gauge core needle (Sharkcore, Covidien, Minneapolis, MN). Three passes will be performed from two separate liver segments (Figure 2). An additional pass may be performed at the discretion of the PI if the three performed passes do not yield enough tissue on visual inspection of the core to ensure adequate histology analysis. The maximum number of passes will not exceed four. Doppler ultrasound will be used to exclude intervening vessels. Two samples will be submitted for histopathology review in formalin, and one or two will be submitted for total fat quantification analysis. After the EUS guided liver biopsy (usually take 10 minutes to perform), the Orbera intragastric balloon will be advanced to the stomach under endoscopy guidance and inflated with 600 ml of normal saline. Patient transient nausea and pain symptoms will be managed with few days course of antiemetic and antispasmodics as per our clinical protocol. Patient will also be required to take a proton pump inhibitor -if not already taking- throughout the 6 months of balloon therapy. Patient will then undergo a moderate intensity life-style intervention consisting of daily phone call for the first week (nurse and or study coordinator) then monthly group visits with a health-care coach / nutrition to administer the life-style intervention program over 6 months period as per our clinical protocol for the intragastric balloon. At 6 months patients will return for balloon removal at our complex endoscopy unit under monitored anesthesia care. The balloon will be removed using the standard balloon retrieval kit approved by the FDA. After balloon removal a second EUS guided liver biopsy from two separate liver segments similar to the first biopsies site will be obtained with three or four passes with the 22 gauge core liver biopsy needle (Sharckcore, Covidien, Minneapolis, MN). The anticipated procedural length for both the EUS guided liver biopsy and balloon insertion and removal is less than 6 minutes. After balloon removal, all patients will be offered additional six months of moderate life-style intervention program administered as monthly group or individual visits with a health care coach to emphasize healthy lifestyle intervention and behaviors. Weight, height, waist circumference, and demographic information will be collected a baseline, time of balloon removal (6 months), and at 12 months (6 months after balloon removal). Of note the cost of balloon placement, removal, medications to manage symptoms after balloon placement, EUS guided liver biopsy and pathologic interpretation, and the 12 months life-style intervention program will be covered by the study. The initial evaluation at the obesity clinic prior to enrollment in the study will be part of regular clinical care. The PI will evaluate all patients within 1 week of balloon placement and removal.

6.1 Follow up for Patients with Device-Related Adverse Events at the Time of Device Removal

Patients with any device related adverse events (AE) or gastric ulcerations at the time of device removal will continue on medical proton pump inhibitors therapy with dose and duration of therapy determined by the study PI. These patients will be followed clinically until complete resolution. Repeat endoscopy to assess for resolution will be done at the discretion of the study PI depending on the severity of endoscopic finding at the time of removal and the clinical need based on best clinical guidelines and practices.

6.2 Schedule of Events

Study Activity	Screening Visit	Procedure	Week 1	Month 1	Month 2	Month 3	Month 4	Month 5	Month 6	Months 7-11	Month 12
Informed consent	X										
History and demographic information	X										
Physical exam[a]	X										
MRE/MRS[b] (magnetic resonance elastography/spectroscopy)	X								X		
One day evaluation in obesity clinic[c]	X										
B-HCG[d]	X										
Endoscopic ultrasound (EUS)		X							X		
EUS guided liver biopsy		X							X		
Placement of intragastric balloon		X									
Daily phone calls life Style Intervention			X								
Monthly group visit health-care coach/nutrition				X	X	X	X	X	X	X	X
Follow-up phone calls by bariatric clinic				X	X	X	X	X	X	X	X
Removal of intragastric balloon									X		

a Weight, height, waist circumference, demographic information
b MRE=Magnetic Resonance Elastography, MRS=Magnetic Resonance Spectroscopy
c Nutrition, psychology, gastroenterology
d Serum pregnancy test (women of childbearing potential)

6.3 Tissue lipidomics workup

A total of 50 mg of liver tissue will be collected via EUS-guided biopsy. Sharkcore needle is 22 gauge. Diameter is ~0.7 mm. ~270 mm of tissue length will be acquired and will deliver a volume of ~50 mg. The sample will be frozen in liquid nitrogen immediately upon harvesting in the endoscopy suite and placed in a 1.5 conical Eppendorf tube. Samples shall not sit more than few minutes before flash freezing to ensure adequate analysis. Also, inspection before freezing will occur to ensure that the tissue is relatively clean specimen on inspection.

7 Statistical Plan

Baseline subject characteristics will be described by their means, medians and and/or frequencies. For our primary study endpoint of improvement in NASH activity scores on paired liver biopsies at baseline and after 6 months, we will be using a paired t-test with two sided alpha <0.05. With a sample size of 20 subjects, we will have 80% power to detect a medium to large difference in activity scores (0.66). Please see table below for minimum sample size needed for each desired effect size.

	Small effect size 0.5	Medium effect size 0.66	Large effect size 0.8
Sample (N)	34	20	15

Additionally, we will run a linear regression analysis with the change in NASH activity scores as the outcome with baseline BMI and diabetes as the covariates on a univariable and multivariable fashion to investigate their effects on the primary outcome.

For our secondary endpoint of number of subjects with regression of fibrosis in paired liver biopsies, we will be reporting the percentage of subjects with regression. As far as the secondary endpoint of percent total body weight 6 months after Orbera balloon placement, we will be reporting the mean or median percent total body weight loss at 6 months.

For our secondary endpoint of quantifying total fat liver and triglycerides content, we will be reporting the mean and median percent of change at 6 months.

8 Safety and Adverse Events

All adverse events occurring during the study, including those not meeting the criteria of an Unanticipated Adverse Device Effect (UADE) will be recorded on the appropriate case report form. Records of these events will be maintained and reports submitted to the FDA and IRB according to the regulatory requirements. Expected clinical adverse events and nonsignificant (not serious) clinical adverse events will not be reported. Expected clinical adverse events and anticipated adverse device effects are those listed in Section 1.5.2.

8.1 Definitions

Unanticipated Adverse Device Effect (UADE)

A UADE is any serious adverse effect on health or safety or any life-threatening problem or death caused by, or associated with, a device if that effect, problem or death was not previously identified in nature, severity, or degree of incidence in the investigational plan or IDE application (including a supplementary plan or application), or any other unanticipated serious problem associated with a device that relates to the rights, safety, or welfare of subjects.

Adverse Effect (Event)

Any untoward medical occurrence in a subject involved in clinical study of an investigational device; regardless of the causal relationship of the problem with the device or, if applicable, other study related treatment(s).

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.

Hospitalization, Prolonged Hospitalization or Surgery

Any adverse event that results in hospitalization or prolonged hospitalization should be documented and reported as an unanticipated adverse device effect 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 an adverse event.

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

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 local investigator should instruct each subject to report, to the local investigator, any subsequent event(s) that the subject, or the subject's personal physician, believes might reasonably be related to participation in this study. The local investigator should notify the study regulatory 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 local investigator should become aware of the development of problems, cancer or of a congenital anomaly in a subsequently conceived offspring of a subject that has participated in this study.

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.

Unanticipated Problems Involving Risk to Subjects or Others (UPIRTSO)

Any unanticipated problem or adverse event that meets all of the following three criteria:

- Serious: Serious problems or events that results in significant harm, (which may be physical, psychological, financial, social, economic, or legal) or increased risk for the subject or others (including individuals who are not research subjects). These include: (1) death; (2) life threatening adverse experience; (3) hospitalization - inpatient, new, or prolonged; (4) disability/incapacity - persistent or significant; (5) birth defect/anomaly; (6) breach of confidentiality and (7) other problems, events, or new information (i.e. publications, DSMB reports, interim findings, product labeling change) that in the opinion of the local investigator may adversely affect the rights, safety, or welfare of the subjects or others, or substantially compromise the research data, **AND**
- Unanticipated: (i.e. unexpected) problems or events are those that are not already described as potential risks in the protocol, consent document, not listed in the Investigator's Brochure, or not part of an underlying disease. A problem or event is "unanticipated" when it was unforeseeable at the time of its occurrence. A problem or event is "unanticipated" when it occurs at an increased frequency or at an increased severity than expected, **AND**
- Related: A problem or event is "related" if it is possibly related to the research procedures.

8.2 Adverse Event Reporting Period

For this study, the study treatment follow-up period is defined from enrollment in the study to 6 months after balloon removal (12 months from balloon insertion). Adverse events will be captured and reported from enrollment to the end of the study treatment follow-up.

8.3 Recording of Adverse Events

Adverse events will be solicited at each study visit or contact with patient. All adverse events will be recorder on the below case report form:

- Subject Study Number/Identifier
- Device information (model and serial number)
- Date of event onset
- Description of the event
- Indication if study treatment was discontinued, or if investigational device was removed
- Subject current status, or if the event was resolved
- Principal Investigator assessment of if the event was serious and justification for determination

- Principal Investigator assessment of causality and relationship to study treatment.

8.4 Sponsor-Investigator Reporting of Unanticipated Adverse Device Effects and Unanticipated Problems

When an adverse event has been identified, the study team will take appropriated action necessary to protect the study participant and then complete the Study Adverse Event Worksheet and log. The sponsor-investigator will evaluate the event and determine the necessary follow-up and reporting required.

The sponsor-investigator will promptly review documented Unanticipated Adverse Device Effects and as necessary shall report the results of such evaluation to FDA within 10 working days and Mayo IRB within 5 working days of initial notice of the effect. Thereafter the sponsor-investigator will submit such additional reports concerning the effect as requested.

8.4.1 Sponsor-Investigator Reporting, Notifying Mayo IRB

The sponsor-investigator will report to the Mayo IRB any UPIRTSOs and Non-UPIRTSOs according to the Mayo IRB Policy and Procedures.

8.4.2 Sponsor-Investigator Reporting: Notifying the FDA

The sponsor-investigator will report to the FDA all unanticipated adverse device effects according to the required reporting timelines, formats and regulations.

The sponsor-investigator will submit a completed FDA Form 3500A to the FDA's Center for Devices and Radiological Health for any observed or reported adverse effect that is determined to be an unanticipated adverse device effect. A copy of this completed form will be provided to all participating sub-investigators. (Note: DSMP)

The completed FDA Form 3500A will be submitted to the FDA as soon as possible and, in no event, later than 10 working days after the sponsor-investigator first receives notice of the adverse effect.

If the results of the sponsor-investigator's follow-up evaluation shows that an adverse effect that was initially determined to not constitute an unanticipated adverse device effect does, in fact, meet the requirements for reporting; the sponsor-investigator will submit a completed FDA Form 3500A as soon as possible, but in no event later than 10 working days, after the determination was made.

For each submitted FDA Form 3500A, the sponsor-investigator will identify all previously submitted reports that that addressed a similar adverse effect experience and will provide an analysis of the significance of newly reported adverse effect in light of any previous, similar report(s).

Subsequent to the initial submission of a completed FDA Form 3500A, the sponsor-investigator will submit additional information concerning the reported adverse effect as requested by the FDA.

Reporting Process

Unanticipated Adverse Device Effect reports will be submitted on FDA Form 3500A. The contact information for submitting reports is:

Food and Drug Administration
Center for Devices and Radiological Health
Document Mail Center - WO66-G609
10903 New Hampshire Avenue
Silver Spring, Maryland 20993-0002

Deviations from the investigational plan

The sponsor-investigator shall notify Mayo IRB (see 21 CFR 56.108(a) (3) and (4)) of any deviation from the investigational plan to protect the life or physical well-being of a subject in an emergency. Such notice shall be given as soon as possible, but in no event later than 5 working days after the emergency occurred. Except in such an emergency, prior approval by the sponsor-investigator is required for changes in or deviations from a plan, and if these changes or deviations may affect the scientific soundness of the plan or the rights, safety, or welfare of human subjects, FDA and IRB notification in accordance with 21 CFR 812.35(a) also is required.

8.5 Unblinding Procedures

Not applicable

8.6 Stopping Rules

- 1 death
- 2 or more severe adverse events or severe unanticipated adverse device effect deemed of enough severity by the primary investigator and co-investigator to stop the study due to safety concerns that can't be mitigated by the study team. In that case the study will not resume until approval is obtained from the FDA and IRB.

8.7 Medical Monitoring

It is the responsibility of the sponsor-investigator to oversee the safety of the study. 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 10 Auditing, Monitoring and Inspecting). Medical monitoring will include a regular assessment of the number and type of serious adverse events.

8.7.1 Internal Data and Safety Monitoring Board

The data safety will be monitored directly by the primary investigator.

8.8 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 (long term survival status that the subject is alive) at the end of their scheduled study period.

8.9 Source Documents

Source data comprise 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. When applicable, information recorded on the CRF shall match the Source Data recorded on the Source Documents.

8.10 Case Report Forms

A Case Report Form (CRF) will be completed for each subject enrolled into the clinical study. The investigator-sponsor will review, approve and sign/date each completed CRF; the investigator-sponsor's signature serving as attestation of the investigator-sponsor's responsibility for ensuring that all clinical and laboratory data entered on the CRF are complete, accurate and authentic.

The study case report form (CRF) 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, write "N/D". If the item is not applicable to the individual case, write "N/A". All entries should be printed legibly in black ink. If any entry error has been made, to correct such an error, draw a single straight line through the incorrect entry and enter the correct data above it. All such changes must be initialed and dated. Do not obliterate, erase, or use "white-out" for errors. For clarification of illegible or uncertain entries, print the clarification above the item, then initial and date it. If the reason for the correction is not clear or needs

additional explanation, neatly include the details to justify the correction. Any missed, unused, corrected and/or spurious data will be discarded in HIPPA compliant manner.

Data Management

A database will be created and maintained by the study coordinator in order to ensure all data on case report forms are completed and accurate. This database will be stored on a secure Mayo Clinic computer and not be accessible to anyone outside the institution.

Data Security and Confidentiality

Any study data with identifiers will be kept in a locked file cabinet and stored electronically on a password protected server.

Data Quality Assurance

Source documents will be verified to ensure they align with case report form data.

Data Clarification Process

N/A

8.11 Records Retention

The sponsor-investigator will maintain records and essential documents related to the conduct of the study. These will include subject case histories and regulatory documents.

The sponsor-investigator will retain the specified records and reports for:

1. Up to 2 years after the marketing application is approved for the drug; or, if a marketing application is not submitted or approved for the drug, until 2 years after shipment and delivery of the drug for investigational use is discontinued and the FDA has been so notified. OR
2. As outlined in the Mayo Clinic Research Policy Manual –"Retention of and Access to Research Data Policy" http://mayocontent.mayo.edu/research-policy/MSS_669717, whichever is longer.

9 Study Monitoring, Auditing, and Inspecting

9.1 Study Monitoring Plan

The investigator will allocate adequate time for such monitoring activities. The Investigator will also ensure that the monitor or other compliance or quality assurance reviewer is given access to all the study-related documents and study related facilities (e.g. pharmacy, diagnostic laboratory, etc.), and has adequate space to conduct the monitoring visit.

This study will be monitored during the conduct of the trial by staff from the Mayo Clinic Office of Research Regulatory Support. Clinical trial monitoring may include review of the study documents and data generated throughout the duration of the study to help ensure the validity and integrity of the data along with the protection of human research subjects. This will assist sponsor-investigators in complying with Food and Drug Administration regulations.

9.2 Auditing and Inspecting

The sponsor-investigator will permit study-related monitoring, audits, and inspections by the IRB, the monitor, and government regulatory agencies, of all study related documents (e.g., source documents, regulatory documents, data collection instruments, study data etc.). The sponsor-investigator will ensure the capability for inspections of applicable study-related facilities (e.g., pharmacy, diagnostic laboratory, etc.).

Participation as a sponsor-investigator in this study implies acceptance of potential inspection by government regulatory authorities and applicable compliance offices.

10 Ethical Considerations

This study is to be conducted according to United States government regulations and Institutional research policies and procedures.

This protocol and any amendments will be submitted to a properly constituted local Institutional Review Board (IRB), in agreement with local legal prescriptions, for formal approval of the study. The decision of the IRB concerning the conduct of the study will be made in writing to the sponsor-investigator before commencement of this study.

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. This consent form will be submitted with the protocol for review and approval by the IRB for the study. The formal consent of a subject, using the Approved IRB consent form, must be obtained before that subject undergoes any study procedure. The consent form must be signed and dated by the subject or the subject's legally authorized representative, and the individual obtaining the informed consent.

11 Study Finances

11.1 Funding Source

This study will be sponsored by an internal Mayo Clinic development grant and an endoscopic research grant from the American Society for Gastrointestinal Endoscopy.

11.2 Subject Stipends or Payments

None

12 Publication Plan

This is an investigator initiated and funded study. The investigator holds the primary responsibility for publication. The trial will be registered on ClinicalTrials.gov prior to subject recruitment and enrollment. The results will also be posted to ClinicalTrials.gov within 12 months of final data collection for the primary outcome.

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