Orlistat for the Treatment of Type I Hyperlipoproteinemia

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Funding Sponsor: Local funds, Southwestern Medical Foundation
Version: 2
Date: June18, 2018
IRB number – STU 012013-042

Introduction and Purpose:
Patients with Type I Hyperlipoproteinemia (T1HLP) have a rare form of hypertriglyceridemia marked by significant chylomicronemia and recurrent episodes of acute pancreatitis. T1HLP is caused by a deficiency of lipoprotein lipase or one of its cofactors. Many patients are a challenge to treat, as the only effective therapy available is an extremely low fat diet. This diet is exceedingly difficult to follow, and despite adherence, many patients still have chylomicronemia and develop acute pancreatitis.

Specific Aim: To determine the efficacy of a gastric and pancreatic lipase inhibitor, Orlistat, in reducing serum triglyceride levels in patients with T1HLP.

Hypothesis: Inhibiting dietary fat absorption using a lipase inhibitor will lower serum triglyceride concentrations in patients with T1HLP.

We expect that Orlistat will markedly reduce serum triglycerides in patients with T1HLP by inducing partial fat malabsorption, and we expect that this can be done safely with minimal side effects.

Rationale: As there are no therapeutic options in patients with T1HLP, the results of this study may help in designing a therapeutic approach to extreme hypertriglyceridemia in these patients.

Primary Endpoint: Fasting serum triglyceride levels

Secondary Endpoints:
1. Serum chylomicron-TG levels (Fasting and postprandial)
2. Postprandial serum TG levels during meal tolerance test
3. 72 hour fecal fat
4. Fat soluble vitamin levels

Primary Safety Endpoints:
1. Fat soluble vitamin deficiencies due to fat malabsorption
2. Gastrointestinal side effects (oily stools, anal leakage, diarrhea, flatus with discharge)

The side effects experienced with Orlistat use are mainly gastrointestinal due to fat malabsorption. We intend to reduce these side effects by limiting fat intake in the diet. Additionally, we will provide patients with a multivitamin to take daily in order to reduce risk of vitamin deficiencies.

The benefit of taking Orlistat is a reduction in serum triglyceride levels, and thus, a reduction in the risk of acute pancreatitis. There is significant morbidity and mortality associated with acute pancreatitis, and so we believe a reduction in this risk offers a greater benefit than mild gastrointestinal side effects.

Background:
Type I hyperlipoproteinemia (T1HLP) is a rare, autosomal recessive condition characterized by recurrent episodes of acute pancreatitis due to extreme hypertriglyceridemia as a result of accumulation of chylomicrons (1-2). In most of the patients, T1HLP is due to lipoprotein lipase (LPL) or apolipoprotein CII (APOC2) deficiency, however, recently mutations in lipase maturation factor 1 (LMF1), apolipoprotein A-5 (APOA5), and glycosyl-phosphatidylinositol anchored high density lipoprotein binding protein 1 (GPIHBP1) have been reported (3-4). In some patients, the

genetic basis of T1HLP remains unknown. Patients present with eruptive or tuberous xanthomas, pancreatitis, lipemia retinalis, and hepatosplenomegaly. Most importantly, acute pancreatitis is often a cause for significant morbidity and even mortality in these patients. Treatment of these patients poses a significant challenge, as the current medications for hypertriglyceridemia such as fibrates, niacin, and omega-3 fatty acids are ineffective (1-2). Some investigators have tried invasive procedures such as plasmapheresis to lower triglycerides (TG), however this is a temporary measure (5). Others are exploring gene therapy using intramuscular injection of AAV1-Lipoprotein lipase^{S447X} in patients with LPL deficiency. Although this initially lowers triglycerides, the effect is transient as patients develop antibodies to the capsid proteins (6).

The only effective therapy is a very low fat diet (<20% of total energy). Since the basic defect in T1HLP is the reduced clearance of chylomicrons due to impaired lipolysis of triglycerides, reduction in dietary fat by reducing chylomicron formation can lower serum triglycerides. However, some patients continue to have severe triglyceridemia and acute pancreatitis despite following a low fat diet (1).

There have been no formal clinical trials conducted in patients with T1HLP so far and most of the evidence about efficacy or lack of efficacy of medications or dietary intervention is anecdotal. We wish to investigate whether an inhibitor of intestinal lipase (Orlistat) will reduce serum triglyceride concentrations in patients with T1HLP. Orlistat is an inhibitor of gastric and pancreatic lipases and can reduce dietary fat absorption by 30% (7). Thus, Orlistat may reduce serum TG levels in patients with T1HLP by decreasing the substrate available for chylomicron formation.

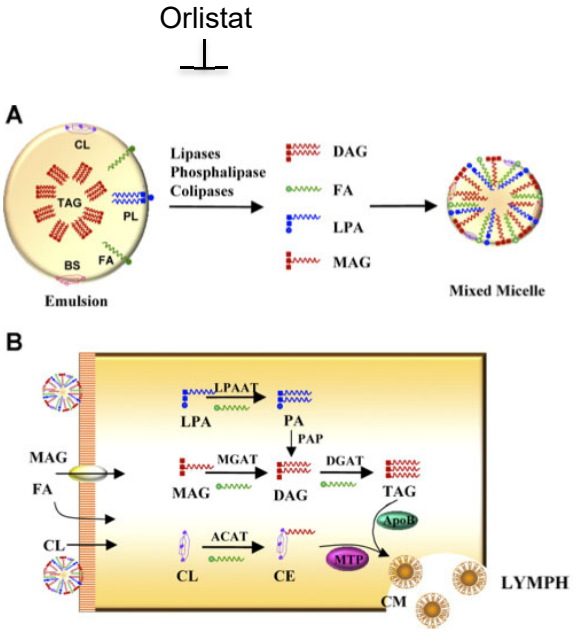


Fig. 1. The process of dietary lipid digestion and absorption. *A,* The digestion of dietary lipids begins with partial digestion by gastric lipase, forming large fat globules with a triacylglycerol (TAG) core surrounded by phospholipids (PL), free cholesterol (CL), fatty acids (FA), and ionizing proteins. In intestine, fat globules are mixed with bile salts (BS) and pancreatic lipases. Monoacylglycerol (MAG), lysophosphatidic acid (LPA), diacylglycerol (DAG), and FA that are released from lipid and PL hydrolysis join bile salts, CL, and fat-soluble vitamins to form mixed micelles for dietary fat absorption at the brush border of the enterocytes. *B,* After entering the enterocyte, MAG, LPA, and CL are reacylated. MAG is sequentially acylated by MGAT and DGAT enzymes to form TAG. LPA is acylated by LPA acyltransferase (LPAAT) to phosphatidic acid (PA) followed by dephosphorylation by PA phosphorylase (PAP) to yield DAG. Dietary cholesterol (CL) is acylated by acyl-CoA:cholesterol acyltransferase (ACAT) to cholesteryl esters (CE). Facilitated by microsomal triglyceride transfer protein (MTP), TAG joins CE and apolipoprotein B-48 (ApoB48) to form chylomicrons that are secreted in the lymph for delivery to circulation. Modified from Shi and Cheng (8). The step that is inhibited by Orlistat (an intestinal lipase inhibitor) is shown

Concise Summary of Project:

A two-treatment condition, four-period, two-sequence cross-over study design will be employed. Since T1HLP is a very rare disease, the study design will allow for patients to act as their own controls, and thus increase the power of the study. This will be an open-label trial.

Baseline period:
The baseline period will include a 4-week run-in period during which time dietary counseling will be performed. To be included in the study, subjects must have a fasting TG > 1000 mg/dL at the end of the baseline period (complete inclusion/exclusion criteria are listed below). All patients will be evaluated at the Clinical Research Unit (CRU) at UTSW for three days at the end of the baseline period. During this time, we will instruct them to consume a diet containing 15% of total energy from fat, 70% from carbohydrate and 15% from protein. The same diet will be used during subsequent evaluations at CRU. This is to ensure that serum lipid evaluation is made when patients are taking a low fat diet to avoid confounding effects of variability in dietary fat content. The subjects will also be asked to complete a 3-day food recall before admission to the CTRC to assess energy intake as well as dietary composition. All patients will then be asked to follow a low fat diet (< 20% energy from fat) for the remainder of the study period. The diet counseling will be done by a trained research fellow or CTRC dietician immediately after the baseline period and at the beginning each new treatment period and at 2 wk duration for the remainder of the study. The counseling will be done in person at baseline and at subsequent follow up visits and by phone in the intervening intervals.

Fasting blood samples will be obtained for serum lipid profile, chylomicron-TG, serum chemistry, and aminotransaminases for the course of three days. Blood will also be drawn once for determination of fat soluble vitamin levels (A, D, E, and K). Patients will collect 72 hour stool for determination of fecal fat. On the last day of the baseline period, a meal tolerance test will be performed.

Study phase:
The patients will then be randomized to receive 3 months of Orlistat or control condition with no drug, then will crossover to the opposite condition as shown in Figure 2. This will be repeated for a total of 12 months or four periods of study.. Orlistat is approved by the FDA at the higher dose of 120 mg three times a day for children over the age of 12 years. There are limited data regarding dosing of Orlistat for children, especially normal-weight children. One study used Orlistat for the treatment of Crigler-Najjar, which included children over the age of 7. The smallest child weighed 29 kg. In this study, the authors used a dose of 66 mg/m^2 body surface area three times daily, which is roughly equal to the amount for an adult per square meter body surface area (Hafkamp et al. Pediatric Research 2007;62(6):725-730). We are unable to precisely dose Orlistat since we will be using 60 mg capsules (Alli). Therefore, for our study, we will use a graded system for dosing based on the weight of the child (Table 1).

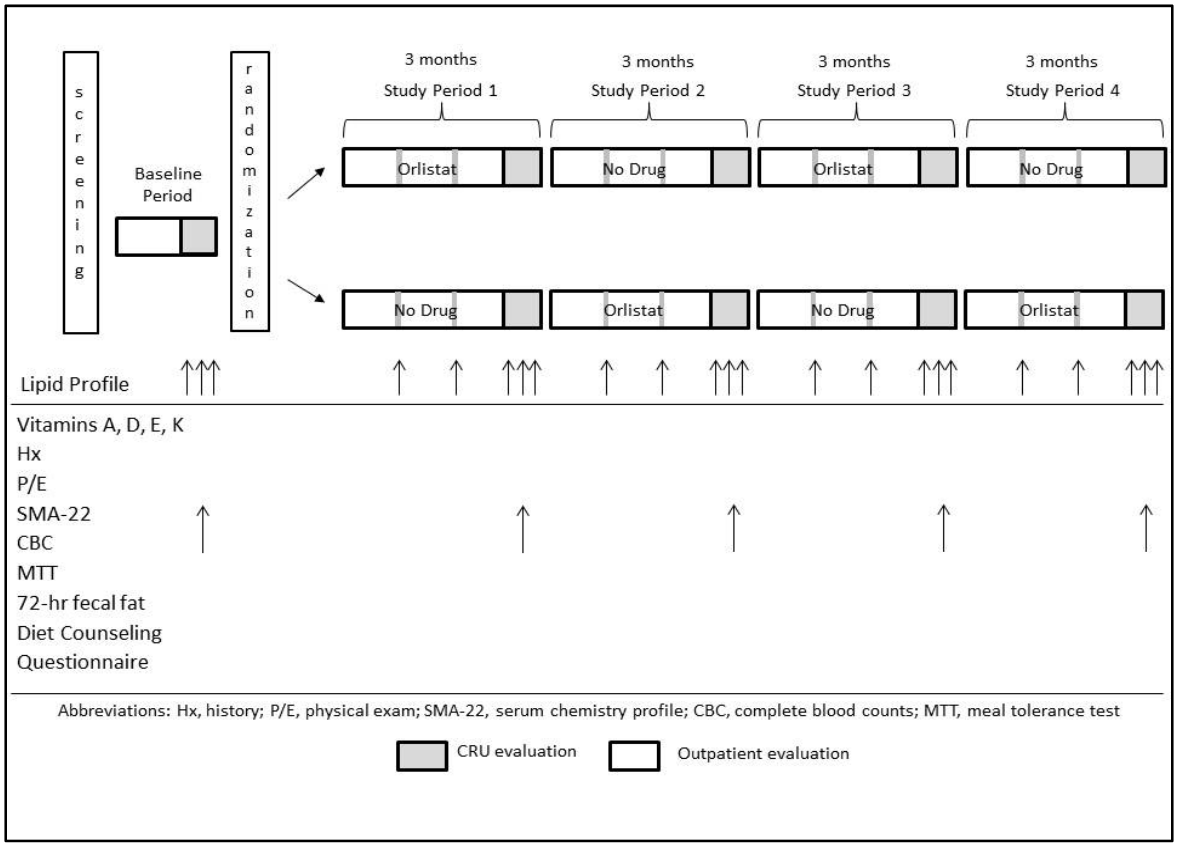
Table 1: Dosing of Orlistat by weight in pediatric subjects:

	Breakfast	Lunch	Dinner
≥50 kg or adult ≥18 yrs	120 mg	120 mg	120 mg
40-49.9 kg	60 mg	120 mg	120 mg
30-39.9 kg	60 mg	60 mg	120 mg
20-29.9 kg	60 mg	60 mg	60 mg

Orlistat has a short half-life of 1 to 2 hours, so no carry-over effects should be observed. Subjects will have their triglycerides measured monthly. All laboratory studies enlisted during the baseline

period (vitamin levels, liver function tests, complete blood count, and 72-hour fecal fat, etc.) will be repeated during the four study periods at CRU (see figure 2). The patients will also take a daily multivitamin supplement containing vitamin A 3,500 IU (71% as retinol and 29% as β-carotene), vitamin D 400 IU (as cholecalciferol), vitamin E 30 IU (as dl-α-tocopheryl acetate) and vitamin K 25 µg (as phytonadione). Patients will be instructed to take vitamins at least 2 hours apart from Orlistat dosing.

Figure 2: Study Flow Diagram



Orlistat is a gastric and pancreatic lipase inhibitor that is approved by the FDA for weight loss. It is available over-the-counter as 60 mg tablets under the trade name Alli, and available by prescription as 120 mg capsules under the trade name Xenical. At a dose of 120 mg three times daily, Orlistat reduces dietary fat absorption by 30% (7). Several studies in the past ten years have shown a beneficial effect of Orlistat on postprandial lipids in healthy volunteers and in patients with diabetes (9-12). In a 52-week study involving obese type 2 diabetic men, Orlistat

120 mg three times daily reduced triglycerides and apolipoprotein B levels significantly (9). The FDA approved dose for obese children 12-18 years is the same as that for adults, i.e., 120 mg three times a day.

Most of the patients with T1HLP present with recurrent abdominal pain and even acute pancreatitis (a potentially life threatening complication) along with extreme hypertriglyceridemia during childhood. Since there is no established therapy for T1HLP, these children are at high risk of acute pancreatitis. Therefore, we have included children in this study. We will carefully monitor each child during participation in the study for any possible side effects. Since variability in glycemic control could be a confounding factor and patients with poorly controlled diabetes mellitus can develop secondary hypertriglyceridemia, we have excluded patients with diabetes from the study.

Orlistat is currently approved for the treatment of obese children age 12 and older. Our study may incorporate younger children since there is currently no therapy available for these patients. A Swedish study was done to evaluate the safety of Orlistat in obese children ages 7-12. This study found no serious adverse effects (including liver toxicity and vitamin deficiencies). Most subjects did experience oily stools and diarrhea on at least one occasion, but this occurred after ingestion of a high fat meal, and was not severe enough to cause discontinuation of the study. No negative effect on psychological or physical well-being was detected, and no effect on linear growth was identified. The adult dose of Orlistat (120 mg three times a day) was used in this study (13). For this reason, we will use the adult dose, except for small children weighing less than 30 kg. In these children, a dose of 60 mg three times a day will be used. If the adult dose is not tolerated in children weighing $\geq$ 30 kg, it will be reduced to 60 mg three times a day.

Fasting serum TG is chosen as the primary end point variable as this determination is made during clinical follow up of patients with T1HLP. Since Orlistat is expected to affect chylomicron formation, fasting and postprandial levels of chylomicron TG and serum TG are included as secondary endpoint variables. We will also capture any episodes of acute pancreatitis and include them as a secondary endpoint variable. Tertiary end point variables include fat soluble vitamin levels to assess the potential effects of Orlistat on fat soluble vitamin absorption and fecal fat excretion to assess the potency of these drugs in blocking dietary fat absorption.

The majority of Orlistat's mechanism of action is believed to be exerted in the gastrointestinal tract. Pharmacokinetic studies have shown that Orlistat is infrequently detected in plasma in subjects taking Orlistat, and when it is detected, very low levels are found (14-15).

For this study, a maximum of 12 patients will be enrolled. The subjects will serve as their own controls during their period off of the medication. Conditions which would result in the subject exiting the study prior to the expected completion date include safety concerns, intolerable side effects, and subject withdrawal of consent.

Study Procedures:

Serum lipoproteins: Serum cholesterol, TG and high density lipoprotein cholesterol will be measured by Quest Diagnostics. Serum chylomicron TG will be measured according to the Lipid Research Clinics method after ultracentrifugation (16) using enzymatic kits.

Postprandial TG and chylomicron remnant clearance during meal tolerance test: On the last day of evaluation, after obtaining a fasting blood sample, a breakfast meal with a fixed dietary composition (600 kcal with 15% fat, 70% carbohydrate and 15% protein) will be given with 50,000

IU of vitamin A (Aquasol). A low fat meal will be prepared using natural food items and is chosen to avoid exacerbation of hypertriglyceridemia and chylomicronemia in these susceptible patients with T1HLP. Blood samples will be drawn at hourly intervals for 4 hours after the meal for determination of plasma triglyceride and retinyl palmitate concentrations. .

Plasma retinyl palmitate concentration will be determined as described previously (17). Briefly, neutral lipids will be extracted from plasma using methanol and hexane. All tubes used in the extraction will be covered with aluminum foil to protect retinyl esters from light. Plasma (500 µL) and retinyl undecanoate as internal standard (300 ng in 100 µL of ethanol) will be added dropwise to 5 mL methanol. After vortexing, 5 mL hexane will be added and the mixture will be vortexed again for 30 s. The samples will be allowed to stand for phase separation, and after removal of the upper phase, the lower phase will be washed with 5 mL hexane. The hexane phase will be then dried under nitrogen and the residue will be resuspended in 50 µL benzene.

Retinyl palmitate concentrations in the lipid extracts will be measured using reverse phase high performance liquid chromatography (HPLC) using a Waters Model 6000A chromatograph (Waters Associates, Milford, MA) and a 3 µM Ultrasphere ODS column 4.6 X 75 mm (Alltech Associates, Deerfield, IL). Column effluent will be monitored at 326 nm and chromatograms will be recorded on a Beckman 3390A integrator (Beckman Instruments, Palo Alto, CA) using an attenuation setting of three. Peaks will be quantitated by the area ratio method. Plasma triglyceride and retinyl palmitate data obtained during the oral fat tolerance test will be analyzed by calculating incremental area under the curves (normalized to the baseline value) using the trapezoidal rule.

Serum chemistry, complete blood counts and urinalysis: Complete blood counts will include hemoglobin, hematocrit, MCV, MCH, MCHC, WBC count, RBC count, Platelet count and differential WBC count. Serum chemistry will include SMA-22 panel with electrolytes (Na, K, Cl, carbon dioxide, urea nitrogen, creatinine, Ca, Mg, Phosphate), liver function tests (alanine aminotransferase, aspartate aminotransferase, Bilirubin, alkaline phosphatase, gamma glutamyl transferase), glucose, hemoglobin A1c, creatine phosphokinase, total protein, albumin, globulin, uric acid and lactate dehydrogenase. Urinalysis will include color, appearance, specific gravity, pH, glucose, bilirubin, ketones, occult blood, protein, nitrite, leukocyte esterase, WBC, RBC, squamous epithelial cells, bacteria and hyaline cast. These tests will be done as indicated in the flow diagram. Serum TSH and free T4 will be measured during each CRU visit for those patients who are taking thyroid hormone.

72 Hour Fecal Fat: Stool will be collected for 72 hours during each study period. The measurement will be performed by Quest Diagnostics. Oral carmine will be used as a marker to ensure 72 hour collection of the stools.

Fat soluble vitamin levels: Serum retinol, α-tocopherol, 25-OH Vitamin D and vitamin K will be measured once at baseline and during each study period. The measurements will be performed by Quest Diagnostics.

Three-Day Food Record: Dietary intake will be assessed by 3-day food record (two weekdays and one weekend day), a valid and reliable method. The records will be taken at baseline and at two week intervals. The participants will be instructed on how to record in detail all the food and drink consumed in the 3-day food record book provided. The booklet will be available in English and Spanish. Any questionable input or incomplete food entries recorded will be promptly addressed when the subjects return their food records. The food records will be analyzed for nutrient content using the University of Minnesota Nutrient Data System (NDS) for research.

Gastrointestinal Symptoms Questionnaire: All patients will complete a gastrointestinal symptoms questionnaire (see appendix 1) during visit to the CRU (18). This questionnaire includes questions about diarrhea and steatorrhea, the expected side effects of Orlistat.

Subjects will be evaluated every month. They will be evaluated at CRU for evaluation at the beginning of the study, and every 3 months thereafter until the end of the study. Each evaluation will be 3 days long. In addition, subjects will be seen in the CRU at the 1- and 2-month marks of each study period. This evaluation will last 3-4 hours and will not require an overnight stay. If patient is unable to come to CRU, they will send blood sample for lipid analysis.The study diagram above describes the timing of the procedures in more detail.

Subjects will not be responsible for any research-related costs during this study.

Sub-Study Procedures:
No sub-studies will be involved in this study.

Criteria for Inclusion of Subjects:

1. Type I hyperlipoproteinemia
2. Fasting serum triglyceride levels of greater than 1000 mg/dL
3. Age ≥ 8 years

Criteria for Exclusion of Subjects:

1. Secondary hypertriglyceridemias due to diabetes, renal disease, hypothyroidism, alcoholism and drug therapy such as estrogens and estrogen analogues, steroids, HIV-protease inhibitors, retinoic acid derivatives and interferons
2. Pregnant or lactating women
3. Significant liver disease (elevated transaminases > 2 times upper limit of normal)
   Alcohol abuse (> 7 drinks or 84 g per week for women and > 14 drinks or 168 g per week for men)
4. Severe anemia (hematocrit < 24%)
5. Drug use (cocaine, marijuana, LSD, etc.)
6. Major surgery in the past three months
7. Congestive heart failure
8. Serum creatinine greater than 2.5 mg/dL
9. Cancer within the past five years
10. Gastrointestinal surgery in the past
11. Current therapy with anti-coagulants, digoxin, and anti-arrhythmics
12. Current therapy with cyclosporine
13. Chronic malabsorption syndromes
14. Cholestasis
15. Acute illnesses such as acute pancreatitis in the last 8 weeks

Sources of Research Material:
Research material obtained in this study will consist of questionnaires, blood and stool specimens, physical examination, and 3-day food records obtained from the subjects according to the protocol. These will be obtained directly from subjects. Subjects will be asked to provide urine

and stool, and blood will be collected via venipuncture. New clinical data will be generated from these specimens. The specimens will be collected and analyzed solely for research purposes.

Recruitment Methods and Consenting Process:
Investigators will conduct chart review on Children's EPIC and Parkland EPIC to search for patients with above mentioned inclusion and exclusion criteria. The study subjects will be recruited from Children's Medical Center (now called Children's health), the lipid clinics at the Parkland Memorial Hospital, Veterans Affairs North Texas Health Care System, and UT Southwestern Aston Ambulatory Center. In the past, patients have been referred to the P.I. for diagnosis, clinical evaluation and management of T1HLP. These patients may be recruited for the study as well. Patients will be recruited and consented by the investigators. Investigators will obtain informed consent after explaining the study and the subjects having read or been read the consent form. For Spanish-speaking subjects, a consent form will be provided in Spanish. For other Non-English speaking subjects, an interpreter will be used via phone or in person. All information collected will be confidential and kept in a locked room. The subjects will be informed that participation is voluntary and will not affect their medical care or their relationship with the physician and institution. Minors may be included in this study, and consent for participation will be obtained from the subject's parent or guardian. Consent will be obtained prior to any study procedures being performed. A copy of the consent form will be given to the subjects and the original consent will be maintained in the subject's research records, which will be maintained in the CRU.

Potential Risks:
All subjects will undergo a history and physical examination. Subjects will be asked to collect stool and urine samples, for which there is minimal risk of psychological or physical discomfort. They will be asked to undergo blood drawing, for which there is an unlikely risk for bruising and infection. Phlebotomy volumes for pediatric subjects will be adjusted based on the child's weight using the National Institutes of Health Guidelines for Blood Drawn for Research Purposes in the Clinical Center 2003.

The potential risks of Orlistat include gastrointestinal side effects such as: diarrhea, oily stools, flatulence with discharge, and anal leakage (9, 19-20). These risks occur commonly but are significantly reduced by adherence to a low fat diet. The major risks for Orlistat are: drug-drug interactions and the reduction of cyclosporine and thyroid hormone levels in subjects taking thyroid supplementation (21), and fat soluble vitamin deficiencies due to malabsorption (9, 19-22). We will provide vitamin supplementation daily during the study and monitor the fat soluble vitamin levels periodically. Vitamins will be taken 2 hours apart from Orlistat dosing. For patients taking thyroid supplementation, thyroid hormone levels will be checked during each CRU visit and medication adjusted as needed. Patients will be instructed to take levothyroxine 4 hours apart from Orlistat dosing. Patients taking cyclosporine are excluded from the study.

Currently the standard care for patients with T1HLP is a very low fat diet. This therapy is ineffective and difficult to follow. The risks of Orlistat are mild when compared to the reduction in risk of pancreatitis that may be obtained from this medication. No placebo will be used in this study.

Subject Safety and Data Monitoring:
Potential risks will be minimized by utilizing study codes to identify subjects. The physical risks will be minimized by the inclusion and exclusion criteria and a complete physical examination

prior to initiating study procedures with careful monitoring throughout. Additionally, pregnant or lactating women are excluded from the study. Subjects will have close monitoring of their liver enzymes. In the event of adverse effects or events, the study subjects will be referred to an appropriate physician for treatment. A pregnancy test will be performed on all female patients of reproductive age at each CRU visit (every 3 months). Study subjects who become pregnant during the course of the study will be discontinued from the trial. Blood tests including chemistry, lipids and liver enzymes will be obtained periodically to ensure safety of the subjects. All subjects with a BMI <24 who lose >5% body weight from baseline will be required to undergo dietary counseling from a dietician or trained research fellow. If weight continues to decrease over the next month, subjects will be discontinued from the study.

We propose the creation of an internal Data and Safety Monitoring Committee composed of experts in statistics and lipid metabolism. Identified for the necessary expertise are individuals who are not associated with this trial and not collaborators of the principal investigator. 1. Song Zhang, PhD, Assistant Professor, Department of Clinical Sciences. 2. Scott Grundy, M.D., Ph.D. Professor of Internal Medicine 3. Perrin White, M.D., Professor of Pediatrics 4. Fredrick Dunn, M.D., Associate Professor of Internal Medicine.

The Committee will be asked to: a) Review the research protocol, informed consent documents and the plans for data and safety monitoring. b) Evaluate the progress of the study including periodic assessments of data quality and timeliness, recruitment, retention, and risk versus benefit. c) Report on the safety and scientific progress of the trial. d) Make recommendations to the PI, and if needed to the IRB, CTRC and FDA concerning continuation, termination or modifications of the trial based on the observed beneficial or adverse effects of the interventions under study. e) Conduct or review the interim analysis with regards to safety in accordance with stopping rules, which should be defined in advance of the data analysis. f) Ensure data integrity. g) Ensure the confidentiality of the data and results of monitoring. h) Maintain study integrity by commenting on any problems with study conduct, enrollment, statistics or data collection.

The PI will hold the primary oversight responsibility for this trial and as such he will be responsible for surveying the medical literature for scientific or therapeutic developments that may impact the safety of participants or the ethics of the study. He will insure that: subjects are fully informed of the study requirements throughout the trial, insure that study subjects receive a study calendar upon enrollment, are updated on any new information relevant to their continued participation or change in the risk versus benefit ratio of the interventions. The PI will be responsible for reporting adverse events to the IRB, CTRC and FDA in accordance with IRB policies and time frames. The PI is responsible for submitting continuing review reports to the IRB, and FDA.

Procedures to Maintain Confidentiality:
Study codes will be used to identify subjects to maintain confidentiality. All files will be kept locked, and all information on computers will be password protected. Access to research data is restricted to key personnel directly involved with the study who have been trained in the protection of human subjects and signed statements assuring their compliance with University policies protecting the privacy of research subjects. We do not intend to apply for a Certificate of Confidentiality from the NIH.

Potential Benefits:

The potential benefit of Orlistat therapy is an improvement in serum triglycerides, serum chylomicron-TG levels, and postprandial serum triglyceride levels.

If this therapy is effective, it will result in a reduction of the severe hypertriglyceridemia that is associated with T1HLP and ultimately decrease the incidence of acute pancreatitis. The knowledge to be gained by this study will have important implications in our understanding of how to effectively treat this rare condition.

Biostatistics:

Design. Due to the rare disease status of Type I Hyperlipoproteinemia, which will limit enrollment, we will implement a dual sequence, four-period crossover design to study the Orlistat and control treatment phases. For a small sample size, this design allows us to better estimate treatment and carryover effects that can be aliased in a two period design.

The sample size will be determined by the number of subjects with Type I Hyperlipoproteinemia meeting the inclusion criteria. Approximately 6-12 patients are expected to be enrolled. Therefore no formal power calculations are performed.

Statistical analysis. Each treatment condition will be summarized with descriptive statistics, graphics for each subject, and adverse events will be carefully tabulated. The treatment effect (Orlistat versus control) will be estimated from repeated measures mixed model and summarized with the model's least squares means estimate and corresponding 95% confidence interval.

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Appendix 1: GI Symptoms Questionnaire.

Gastrointestinal Symptoms Questionnaire

Name:_____ **Date:**_____ **Visit:**_____

	No Symptoms (0) → Severe Symptoms (5)					
Symptom	**0**	**1**	**2**	**3**	**4**	**5**
1. Abdominal pains	☐	☐	☐	☐	☐	☐
2. Heartburn	☐	☐	☐	☐	☐	☐
3. Acid regurgitation	☐	☐	☐	☐	☐	☐
4. Sucking sensations in the diaphragm area	☐	☐	☐	☐	☐	☐
5. Nausea and vomiting	☐	☐	☐	☐	☐	☐
6. Rumbling of stomach	☐	☐	☐	☐	☐	☐
7. Abdominal distension	☐	☐	☐	☐	☐	☐
8. Belching	☐	☐	☐	☐	☐	☐
9. Increased gas	☐	☐	☐	☐	☐	☐
10. Decreased passage of stools	☐	☐	☐	☐	☐	☐
11. Increased passage of stools	☐	☐	☐	☐	☐	☐
12. Loose stools	☐	☐	☐	☐	☐	☐
13. Hard stools	☐	☐	☐	☐	☐	☐
14. Urgent need for bowel movement	☐	☐	☐	☐	☐	☐
15. Feeling of incomplete evacuation	☐	☐	☐	☐	☐	☐
16. Oily stools	☐	☐	☐	☐	☐	☐
17. Stool leakage	☐	☐	☐	☐	☐	☐