Title: Effect of liraglutide on neural responses to high fructose corn syrup in individuals with obesity.

ClinicalTrials.gov ID: NCT03500484

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Document: Protocol and SAP

# STUDY PROTOCOL

**STUDY TITLE:** Effect of liraglutide on neural responses to high fructose corn syrup in individuals with obesity.

**ClinicalTrials.gov Identifier:** NCT03500484

**IND Number:** N/A

**Investigational Product:** Liraglutide

**Yale Protocol Number:** 2000022407

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APPROVED BY THE YALE UNIVERSITY IRB 1/6/2021

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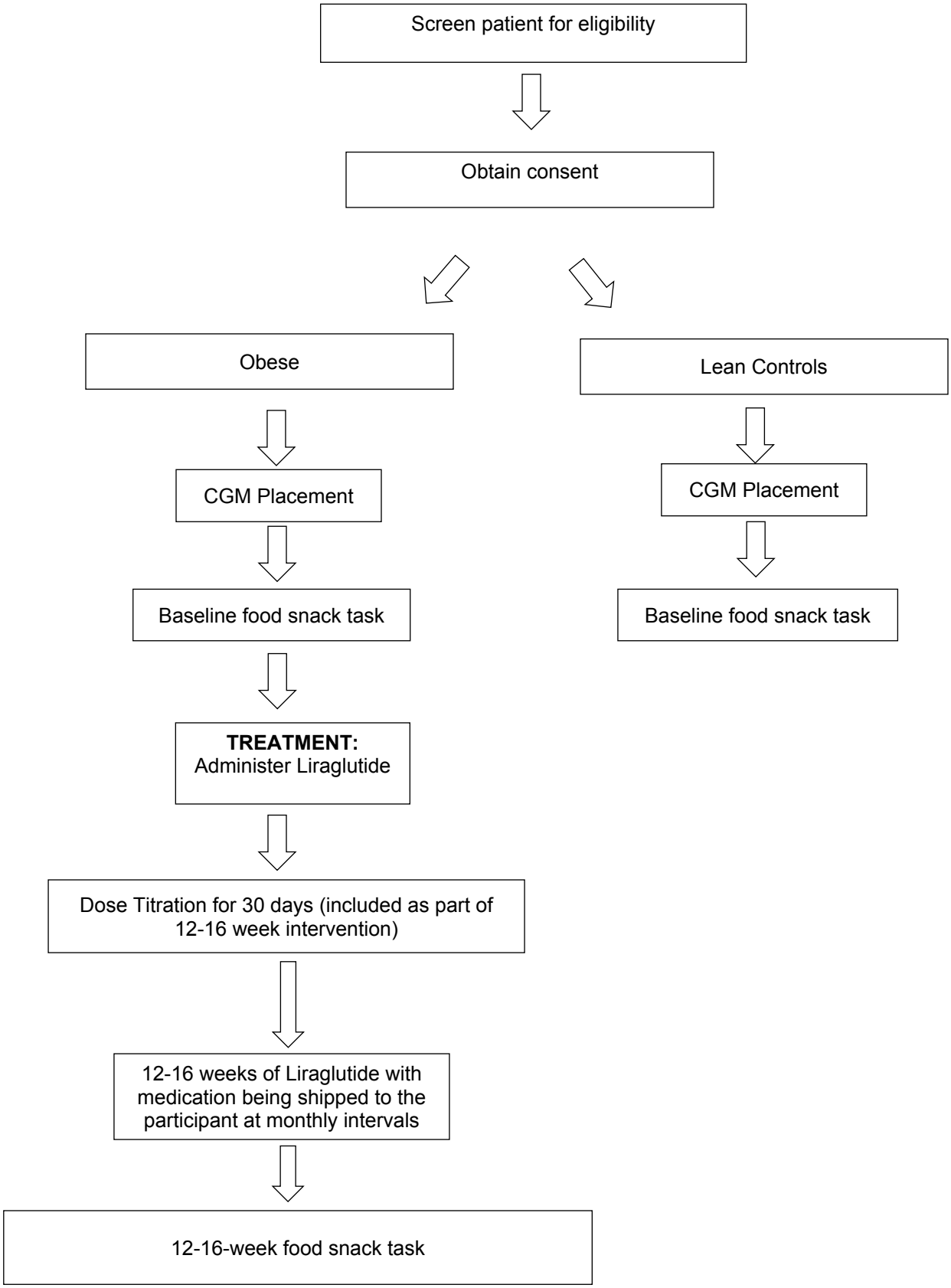
**TRIAL SUMMARY**

| | |
|---|---|
| Abbreviated Title | Effect of liraglutide on neural responses to HFCS in individuals with obesity |
| Trial Phase | 2A |
| Indication | Healthy lean (n=15) and obese individuals (n=30) |
| Control | N/A |
| Product | Liraglutide |
| Number of Subjects | 45 |
| Estimated Enrollment Period | 24 months |
| Aims | **Aim 1** – To investigate the effect of 12-16-week intervention with GLP-1 analogue (liraglutide 3.0mg) on eating behavior and food craving in individuals with obesity (age 15-39 yrs) who chronically consume SSBs (≥5 SSBs/week). |
| Inclusion Criteria | 1. 15-39 years old<br>2. Lean (BMI 18.5-24.9 kg/m$^2$) with normoglycemia with fasting plasma glucose (FPG) <100 mg/dL, 2-hour post oral glucose tolerance test (75gm glucose) glucose level <140 mg/dL and glycosylated hemoglobin (HbA1c) <5.7% **or**<br>3. Obese (BMI 30-45 kg/m$^2$) with or without prediabetes (insulin resistance) but without diabetes $<200$ mg/dL,<br>4. Ingest ≥5 sugar sweetened beverages per week,<br>5. Weight stable for at least 6 months,<br>6. Able to provide written and verbal informed consent,<br>7. Able to read and write in English Able to read and write in English, |
| Exclusion Criteria | 1. Current active participation in a weight loss program or weight loss of ≥10% of total body weight during the previous 6 months,<br>2. Prior bariatric surgery or current gastric balloon,<br>3. Following a vegetarian/vegan diet or dieting/restricting food due to significant food allergies, including but not limited to lactose intolerance and celiac disease,<br>4. Significant medical condition such as cardiac or pulmonary disease, coagulopathy, gastrointestinal disorder, known history of Type 1 or T2D, current psychiatric disorders (including suicidal ideation and depression), including eating disorders (DSM-IV criteria), neurological injury or illness<br>5. Current use of weight-loss medications or supplements, psychiatric medications or anti-hyperglycemic medications (stable dose of metformin for $\geq$ 6 months is acceptable),<br>6. History of or family history of multiple endocrine neoplasia type 2 (MEN-II) or medullary thyroid cancer, alcoholism, or previous history of pancreatitis,<br>7. Females who are pregnant or lactating, or who are unwilling to use proper contraception or remain abstinent, |
| Treatment Plan | Eligible obese subjects will self-administer Liraglutide once daily for 12-16weeks. |

**STUDY SCHEMA**



**LIST OF ABBREVIATIONS**

AE – adverse event
FDA – Food and Drug Administration
fMRI – functional magnetic resonance imaging
HbA1c – hemoglobin A1c
HFCS – high fructose corn syrup
GLP-1 – Glucagon-like peptide 1
PFC – pre frontal cortex
PI – principal investigator
RCT – randomized controlled trial
SAE – serious adverse event
SSB – Sugar sweetened beverage
T2D – Type 2 Diabetes
YCCI – Yale Center for Clinical Investigation

## RATIONALE AND BACKGROUND

Obesity and overweight has reached epidemic proportions in America with the current prevalence rate of 35% in adolescents[1]. This is particularly alarming since over 20% of obese adolescents develop prediabetes[2] and the majority of obese adolescents will be obese in adulthood[3] predisposing them to a myriad of diseases such as type 2 diabetes (T2D). In parallel with the obesity epidemic, added-sugar consumption has markedly increased in Western diets, and is postulated to be a contributor to this public health crisis[4]. Notably, adolescents are the highest consumers of added sugar, with ~20% of their caloric intake accounted for by sugar and nearly half of those calories from sugar-sweetened beverages (SSBs)[5] which contain high fructose corn syrup (HFCS). Evidence indicates that increased consumption of sugars such as HFCS found in SSBs is associated with future weight gain[6,7]. Therefore, understanding the physiologic mechanisms modulating brain reward responses that may promote excess sugar consumption may provide insight into novel interventions to quell SSB ingestion, thus attenuating obesity in adolescents.

Indeed, excessive sugar intake is highly rewarding in the brain[8]. In recent studies, we have found that simple sugars such as glucose and fructose affect brain pathways implicated in regulation of appetite and reward. Specifically, glucose stimulates homeostatic (hypothalamic) brain regions and fructose stimulates hedonic (striatal) regions and both require activation of the prefrontal cortex (PFC), which exerts regulatory control over impulses, choices, and decision-making, to regulate these brain regions to control food intake[9]. We previously reported that obese adolescents have decreased medial PFC (mPFC) control responses to consumption of glucose and fructose9, suggesting diminished capacity to down-regulate hedonic (striatal) and homeostatic (hypothalamic) brain responses that may affect eating behavior. Furthermore, we found hyperactivation of reward (striatal) responses to fructose ingestion in obese adolescents. Notably, these two highly rewarding simple sugars are rarely, if ever, consumed in isolation, but rather are ingested in combination forms such as HFCS[10]. Thus, the question arises: what interventions may attenuate excess sugar (SSB) consumption in obese adolescents by potentially decreasing the neural reward response to HFCS.

Increased ingestion of added sugars, such as HFCS in SSBs is associated with the obesity epidemic4, long- term weight gain[6,7], T2D, and cardiovascular disease (CVD)[22]. Furthermore, ingestion of SSB's may contribute to the development of T2D and CVD independent of obesity[22.] Notably, adolescents are the highest consumers of added-sugars[5], thus studying this vulnerable population to elucidate novel therapeutic strategies to prevent the development of T2D is vital. It is striking that adolescents with obesity and prediabetes (insulin resistance) progress to develop T2D much more rapidly than adults with prediabetes, within 21 months vs. 5 years, respectively[21]. Additionally, once adolescents develop obesity and T2D they often are not successful in maintaining glycemic control with lifestyle and metformin therapy or losing weight with lifestyle interventions, and subsequently their glycemic control deteriorates necessitating more aggressive pharmacological treatment such as insulin therapy[23]. It is clear that prevention of T2D in adolescents is imperative and interventions to mitigate SSB consumption, halt weight gain, and progression to T2D are critical. Yet, therapies to decrease added-sugar consumption, weight gain, and progression to T2D in adolescents are blatantly lacking. GLP-1 analogues improve glycemic control, contribute to weight loss, and may attenuate food preference for sweets[15,16]. Thus, it is important to elucidate how GLP-1 analogues effect neural responses in reward-motivation pathways of the brain to affect food preference and contribute to weight loss in obese prediabetic adolescents. The expected results may significantly contribute to our understanding of neural mechanisms involved in GLP-1 analogue action to affect food preference and curb excess sugar intake in the setting of adolescent obesity and prediabetes and have important implications for clinical practice in the

care of obese prediabetic adolescents to prevent progression to T2D.

**Rationale for Trial and Selected Patients**

Adolescents are the highest consumers of SSB potentially because their developing brains are most susceptible to the rewarding neural effects of sugar consumption. The prefrontal cortex (PFC) (implicated in control of impulses, choices, and decision-making) is not fully developed in adolescents[24], undergoing significant development into young adulthood[25,] and thus has not yet gained mature regulatory capacity over reward-motivation (striatal) and hunger-satiety (hypothalamic) brain regions, to control SSB intake and potentially mitigate excessive consumption in adolescents. Thus, we specifically target this vulnerable population (adolescents) to understand neural mechanisms involved in excess sugar consumption which predisposes to the development of obesity, prediabetes, and T2D.

As the original target study population is adolescents, we are now expanding the age group to include 15-17 year olds; we are basing this on several new studies which have been conducted recently and published, as well as the FDA approval of liraglutide for children age 10-17 who have type 2 diabetes. As of June 2019, liraglutide is now FDA approved for treatment of type 2 diabetes in children 10 years and older[68, 69]. Liraglutide has been FDA approved for treatment of obesity in adults >18 years old since 2014, and was approved in December, 2020 for the treatment of obesity in adolescents aged 12-17 years; additionally, several clinical trials have been conducted in children and adolescents (age 7-17 years old) with obesity alone without type 2 diabetes to assess safety, tolerability and dose. Liraglutide has been shown to have a similar safety, tolerability and pharmacokinetic profile in younger children (age 7-11 years) with obesity as in adults[71]. In a safety and tolerability trial, liraglutide was found to have a similar profile in terms of side effects and pharmacokinetics as in adults with obesity and adolescents with obesity (age 12-17 years) at the same dose used for weight management in adults[70]. Earlier in 2020, a clinical trial entitled “Effect of Liraglutide for Weight Management in Pubertal Adolescent Subjects With Obesity: 56-week Double-blind, Randomised, Parallel-group, Placebo-controlled, Multi-national Trial Followed by 26-week Period Off Study-drug” was completed, published, and liraglutide 3.0mg is now FDA-approved for treatment of adolescents (age 12-17 years old) with obesity, without diabetes[72, 73]. Given that liraglutide has been shown to have a similar safety profile in adolescents with obesity as adults with obesity, we believe it would be beneficial to lower the study’s age minimum to mid-late adolescents (age 15-17) to better understand the neural mechanisms in adolescents with excess sugar consumption. Indeed, adolescents are the highest consumers of added sugar and sugar sweetened beverages and the polultation we are most interested in both for understanding underlying mechanisms which lead to or quell SSB consumption as well as treatment for this high risk population.

The neural mechanisms involved in the reward response to “real-life” sugars, such as HFCS, may provide insight into novel interventions to quell SSB ingestion. Excessive sugar intake is highly rewarding in the brain[8]. Glucose stimulates homeostatic (hypothalamic) brain regions and fructose stimulates hedonic(striatal) regions and both require the prefrontal cortex (PFC) to regulate these brain region’s responses to control food intake[9]. In preliminary work, we found that obese adolescents have decreased medial PFC (mPFC) executive control response to ingestion of glucose and fructose[9], suggesting diminished capacity to down-regulate hedonic (striatal) and homeostatic (hypothalamic) brain regions that may affect eating behavior. Furthermore, we found hyperactivation of reward (striatal) response to fructose ingestion in obese adolescents[9]. This is important because fructose in combination with

glucose makes up “real-life” sugar in forms such as HFCS found in SSBs[10], but the impact of “real-life” sugar ingestion on neural responses in adolescents is not known. Thus, we chose to investigate brain response to ingestion of HFCS (60% fructose/40% glucose) as it is contained in the five most commonly consumed SSBs in the United States, including Pepsi, Coke, Dr. Pepper, Mountain Dew, and Sprite[10].

Therapies to mitigate added-sugar consumption, weight gain, and progression to T2D in adolescents are blatantly lacking. GLP-1 analogues decreased food intake and potentially affect food preference away from sugar-containing food. We will elucidate neural mechanisms by which the GLP-1 analogue liraglutide tempers food intake and potentially alters food preference. Importantly, GLP-1 analogues may prove to be a novel intervention to curb excess sugar consumption by shifting preference away from beverages containing sugar and, thus, contribute to prevention of T2D.

## STATEMENT OF PURPOSE/OBJECTIVES

**Aim 1** – To investigate the effect of GLP-1 analogue (liraglutide 3.0mg) on eating behavior and food craving in individuals with obesity (age 15-39 yrs) who chronically consume SSBs (≥5 SSBs/week).

**Exploratory Aim**: To investigate the role of specific metabolic/hormonal responses (serum leptin, insulin, glucose, GLP-1, acyl-ghrelin) induced by food consumption, and in the setting of liraglutide treatment

## PHARMACEUTICAL AND THERAPEUTIC BACKGROUND

### Product Description & Rationale for Use

Liraglutide was approved for use in adults by the FDA in 2010 for T2D treatment and in 2014 for obesity treatment. It is a glucagon-like peptide-1 (GLP-1) receptor agonist indicated as an adjunct to a reduced-calorie diet and increased physical activity for chronic weight management in adult patients with an initial body mass index (BMI) of 30 kg/m2 or greater (obese) or 27 kg/m2 or greater (overweight) in the presence of at least one weight-related comorbid condition (e.g. hypertension, type 2 diabetes mellitus, or dyslipidemia). Liraglutide, a GLP-1 analogue produced by Novo Nordisk used in the treatment of both obesity and T2D, contributes to weight loss[11,13] and improves glycosylated hemoglobin levels[11,12].

It is an intriguing therapeutic intervention to potentially affect food preference and decrease sugar consumption and attenuate weight gain. Liraglutide is an analogue of an incretin hormone, glucagon-like peptide-1 (GLP-1), which is secreted by L cells in of the small intestine in response to food ingestion[17]. GLP-1 dysregulation is noted in individuals with obesity and T2D with decreased GLP-1 levels post-parandially[18]. Of particular interest is how GLP-1 analogues, such as liraglutide, decrease food intake, and alter food preference.

Preclinical studies have demonstrated that liraglutide not only decreases food intake but also changes food preference[14.] Rats offered chow and candy, obtained 75% of their calories from candy and gained 15- 20% more weight that chow feed counterparts[14]; subsequently with 12-16 weeks of liraglutide treatment the rats decreased their food intake and reversed food preference with an increase in chow intake and decrease in candy ingestion. In another study, rats treated with a GLP-1 analogue no longer preferred the environment which contained chocolate pellets in a conditioned place preference test[16]. Furthermore, in a mouse model, GLP-1 was released from taste-bud cells in response to sweet compounds[37] and GLP-1 receptor knockout mice demonstrated decreased taste response to sweeteners[38] and reduced behavioral response to sweet compounds (short-term lick test) compared to wild-type mice[37.] These studies suggest that GLP-1 is important in sweet taste sensitivity and that GLP-1 analogues may shift food preference away from sugar-containing foods.

### Pharmacokinetics

**Absorption** - Following subcutaneous administration, maximum concentrations of liraglutide are achieved at 11 hours post dosing. The average liraglutide steady state concentration ($AUC_T$/24) reached approximately 116 ng/mL in obese (BMI 30-45 kg/m2) subjects following administration of Saxenda®. Liraglutide exposure increased proportionally in the dose range of 0.6 mg to 3 mg. The intra-subject coefficient of variation for liraglutide AUC was 11% following single dose administration. Liraglutide exposures were considered similar among three subcutaneous

injection sites (upper arm, abdomen, and thigh). Absolute bioavailability of liraglutide following subcutaneous administration is approximately 55%.

**Distribution** - The mean apparent volume of distribution after subcutaneous administration of liraglutide 3 mg is 20-25 L (for a person weighing approximately 100 kg). The mean volume of distribution after intravenous administration of liraglutide is 0.07 L/kg. Liraglutide is extensively bound to plasma protein (greater than 98%).

**Metabolism** - During the initial 24 hours following administration of a single [3H]-liraglutide dose to healthy subjects, the major component in plasma was intact liraglutide. Liraglutide is endogenously metabolized in a similar manner to large proteins without a specific organ as a major route of elimination.

**Elimination** - Following a [3H]-liraglutide dose, intact liraglutide was not detected in urine or feces. Only a minor part of the administered radioactivity was excreted as liraglutide-related metabolites in urine or feces (6% and 5%, respectively). The majority of urine and feces radioactivity was excreted during the first 6-8 days. The mean apparent clearance following subcutaneous administration of a single dose of liraglutide is approximately 0.9-1.4 L/h with an elimination half-life of approximately 13 hours, making liraglutide suitable for once daily administration.

**Drug Interactions**

Liraglutide has low potential for pharmacokinetic drug-drug interactions related to cytochrome P450 (CYP) and plasma protein binding.

**Drug Supply and Dose**
Liraglutide will be supplied in the following package sizes containing disposable, pre-filled, multi-dose pens. Each individual pen delivers doses of 0.6 mg, 1.2 mg, 1.8 mg, 2.4 mg or 3 mg (6 mg/mL, 3 mL). The study drug will be purchased from Novo Nordisk.

3 x Saxenda® pen NDC 0169-2800-13
5 x Saxenda® pen NDC 0169-2800-15

Each Saxenda® pen is for use by a single patient. A Saxenda® pen should never be shared between patients, even if the needle is changed.

To minimize side effects; liraglutide will be titrated over the first month: starting at a dose of 0.6mg daily for 7 days, increasing to 1.2mg on day 8 for the next 7 days, increasing to 1.8mg on day 15 for the next 7 days, increasing to 2.4mg on day 22 for the next 7 days, and then increasing to 3.0mg on day 39 and continuing this dose for the remainder of the study (12-16 weeks total). If a subject is not able to increase dose to 3.0mg, they will be maintained on the highest dose they are able to tolerate, at a minimum of 1.2mg. If a subject is not able to tolerate the medication at all or at a dose of at least 1.2mg, the medication will be discontinued and the subject will not continue in the study.

**Storage and Stability and Handling Requirements**

Prior to first use, Saxenda® should be stored in a refrigerator between 36ºF to 46ºF (2ºC to 8ºC)

| Prior to first use | After first use | |
|---|---|---|
| Refrigerated 36°F to 46°F (2°C to 8°C) | Room Temperature 59°F to 86°F (15°C to 30°C) | Refrigerated 36°F to 46°F (2°C to 8°C) |
| Until expiration date | 30 days | |

It should not be stored in the freezer or directly adjacent to the refrigerator cooling element. Do not freeze Saxenda® and do not use Saxenda® if it has been frozen.

After initial use of the Saxenda® pen, the pen can be stored for 30 days at controlled room temperature (59°F to 86°F; 15°C to 30°C) or in a refrigerator (36°F to 46°F; 2°C to 8°C). Keep the pen cap on when not in use. Saxenda® should be protected from excessive heat and sunlight. Always remove and safely discard the needle after each injection and store the Saxenda® pen without an injection needle attached. This will reduce the potential for contamination, infection, and leakage while also ensuring dosing accuracy.

**Contraindications**

Personal or family history of medullary thyroid carcinoma or Multiple Endocrine Neoplasia syndrome type 2, Hypersensitivity to liraglutide or any product components and Pregnancy.

**Warnings and Precautions**

Warnings and precautions include the following Thyroid C-cell Tumors, Acute Pancreatitis, Acute Gallbladder Disease, Serious Hypoglycemia, Heart Rate Increase, Renal Impairment, Hypersensitivity Reactions, and Suicidal Behavior and Ideation.

**Adverse Reactions**

Most common adverse reactions, reported in greater than or equal to 5% are: nausea, hypoglycemia, diarrhea, constipation, vomiting, headache, decreased appetite, dyspepsia, fatigue, dizziness, abdominal pain, and increased lipase. These will be reviewed with the participants.

**Availability**

Liraglutide will be ordered directly from Novo Nordisk by the investigational pharmacy and shipped directly to the investigational pharmacy.

**Ordering**

The study drug will be purchased from Novo Nordisk by the Investigational Pharmacy.

Product will be shipped to the investigational pharmacy and over labeled as an IP. The label will include address and telephone number, the protocol number, IP name, dosage form and strength (where applicable), amount of IP per container, lot number, expiration date (where applicable), medication identification/kit number, dosage instructions, storage conditions, and required caution statements and/or regulatory statements as applicable, as per local

regulations. Additional information may be included on the label as applicable per local regulations.

**Accountability**

Only the pharmacist or designee will dispose study drug. A record of drug dispensed and administered to each patient must be maintained. The pharmacist will document the dose dispensed in the appropriate study records which will be reviewed during monitoring visits. In addition, the pharmacist needs to maintain an inventory log of all IP delivered, stored, dispensed and destroyed (including expiry date and batch number).

**Destruction**

The sponsor (or designee) will review with the investigator and relevant site personnel the procedures for documenting receipt of IP, as well as the procedures for counting, reconciling IP, and documenting this process. Used (and/or empty) syringes of enoxaparin will be destroyed locally. Unused syringes will be destroyed locally after sponsor's approval for destruction. Documentation of all used and unused drug disposal must be maintained on site for review during monitoring visits.

# STUDY PLAN AND PROCEDURES

## Study Population

We will enroll 45 adolescents/young adults age 15-39 who present at any of the Principal Investigator's clinics where routine care is provided or may be recruited from the Yale New Haven Community via methods described below. 15 lean controls and 30 obese patients will be enrolled. We will enroll controls (lean) subjects (lean adults and lean adolescents) who are at risk for developing obesity (i.e. individuals with a family history of obesity or metabolic disease).

## Screening Procedures

Subjects will be identified and recruited from the local community via recruiting personnel in the Yale Center for Clinical Investigation (YCCI) and from the Yale Pediatric Obesity Clinic and the Yale Diabetes Center. We will utilize existing YCCI methods of recruitment including, fliers, web advertising, Yale research websites, and HelpUsDiscover. Potential participants will be initially screened for inclusion and exclusion criteria by phone by the research coordinator, and then, if initial study criteria are met, will be invited for an in-person visit at the Yale outpatient research unit (CSRU) or hospital research unit (HRU). Healthy lean and obese participants will be frequency matched on age, race/ethnicity and sex.

## Inclusion Criteria

1. 15-39 years old
2. Lean (BMI 18.5-24.9 kg/m$^2$) with normoglycemia with fasting plasma glucose (FPG) <100 mg/dL, 2-hour post oral glucose tolerance test (75gm glucose) glucose level <140 mg/dL and glycosylated hemoglobin (HbA1c) <5.7% **or**
3. Obese (BMI 30-45 kg/m$^2$) with or without prediabetes (insulin resistance), which is defined as two of the following:
    a. FPG <126 mg/dL
    b. 2-hour post oral glucose tolerance test (75gm glucose)
    c. glucose level <200 mg/dL
    d. HbA1c <6.5%
4. Ingest >=5 sugar sweetened beverages per week
5. Weight stable for at least 6 months
6. Able to read and write in English
7. Able to provide written and verbal informed consent

## Exclusion Criteria

1. Current active participation in a weight loss program or weight loss of >=10% of total body weight during the previous 6 months
2. Prior bariatric surgery or current gastric balloon
3. Following a vegetarian/vegan diet or dieting/restricting food due to significant food allergies, including but not limited to lactose intolerance and celiac disease
4. Significant medical condition such as cardiac or pulmonary disease, coagulopathy, gastrointestinal disorder, known history of Type 1 or Type 2 diabetes, current psychiatric disorders (including suicidal ideation and depression), current eating disorders (DSM-IV criteria), neurological injury or illness
5. Current use of weight-loss medications or supplements, psychiatric medications or anti-hyperglycemic medications (stable dose of metformin for $\geq$ 6 months is acceptable)
6. History of or family history of multiple endocrine neoplasia type 2 (MEN-II) or medullary

thyroid cancer, alcoholism, or previous history of pancreatitis

7. Females who are pregnant or lactating, or who are unwilling to use proper contraception or remain abstinent,

**Consent Process**

Research staff and investigators permitted to consent patients will describe the trial in detail including the data collection requirements, risks, procedures, and alternative treatments. Patients will be made aware that they are under no obligation to a participate, that failure to participate will not adversely affect their care, and that they may withdraw consent at any time. Data will continue to be collected until the patient withdraws consent to use it. They will be given an opportunity to ask questions and have these answered to their satisfaction. The consent form will then be signed and dated by both the patient and the study personnel obtaining consent. If there are any questions regarding competence, the trial will not be offered to the patient. The consent process will be documented in the medical record.

**Study Entry/Registration**

Eligibility will be determined by the PI and/or research team using an eligibility checklist. Once a patient is confirmed eligible and consent is obtained the patient will be assigned a unique study ID. The study ID will be used when the patient is registered into OnCore, Yale's Clinical Trials Management System and included on all related study documents. Subjects will be entered into OnCore by the study research team.

## TREATMENT REGIMEN WITH LIRAGLUTIDE

To minimize side effects; liraglutide will be titrated over the first month: starting at a dose of 0.6mg daily for 7 days, increasing to 1.2mg on day 8 for the next 7 days, increasing to 1.8mg on day 15 for the next 7 days, increasing to 2.4mg on day 22 for the next 7 days, and then increasing to 3.0mg on day 29 and continuing this dose for the remainder of the study (12-16weeks total). This titration schedule (Table 1) will serve as a guide for study clinicians for liraglutide titration. Liraglutide will not be titrated up any faster than the above table outlines. If a participant experiences side effects (such as nausea and vomiting) the study drug will be titrated up more slowly by the study clinicians with close oversight, guidance, and approval from Dr. Jastreboff (the study PI). If a subject is not able to increase dose to 3.0mg (due to side effects such as nausea or vomiting), they will be maintained on the highest dose they are able to tolerate, at a minimum of 1.2mg. If the subject is not able to tolerate a dose of at least 1.2mg, the medication will be discontinued and the subject will not continue in the study.

The participant will be provided with an information sheet detailing the study visits and the titration schedule (Appendix 3).

The study drug will be purchased from Novo Nordisk.

**Table 1: Dose Titration Over One Month**

| **Drug** | Day | Dose/ Potency | Dose Frequency | Route | Treatment Period |
|---|---|---|---|---|---|
| **Liraglutide** | 1-7 | 0.6mg | 1x daily | Subcutaneous | 1 week |
| **Liraglutide** | 8-14 | 1.2mg | 1x daily | Subcutaneous | 1 week |
| **Liraglutide** | 15-21 | 1.8mg | 1x daily | Subcutaneous | 1 week |
| **Liraglutide** | 22-28 | 2.4mg | 1x daily | Subcutaneous | 1 week |
| **Liraglutide** | 29-35 | 3.0mg | 1x daily | Subcutaneous | Until 12 - 16 week MRI |

### Administration

The medication is administered once daily subcutaneously via an injection pen.
Each study participant will be taught how to administer the medication by the PI who will follow-up with each subject at specified interval time points as outlined in Appendix 1 to ensure that participants are tolerating the liraglutide and adhering to taking the medication daily. The participant's first administration will be observed to ensure that it is being doing properly.

### Discontinuing Liraglutide

If a subject is not able to tolerate the medication at all due to persistent vomiting or intolerable nausea, the medication will be discontinued and the subject will not continue in the study. If the subject's mood changes while participating in the study, as assessed by PHQ-9, the PI will contact the subject and assess whether they need intervention and determine if they can continue in the study or if the drug should be discontinued.

**Holding the Dose**

Liraglutide will be stopped temporarily if ANY the following occur:

- Persistent vomiting or nausea
- Moderate-severe dehydration (as a result of nausea/vomiting)
- Significant persistent abdominal pain
- Other side effects which the participant attributes to starting study drug

**Restarting the Dose**

- If participant is amenable to trying the study drug again, the medication will be restarted within 4 days with phone call follow up to assess tolerability. If participant is not able to tolerate the study drug on re-initiation then they will not continue in the study. If the participant does not want to participate in the study at any time point, they may withdraw from the study.

- If the participant withdraws from the study at any point, they will be asked to complete the end of study questionnaires, dietary recalls and to return for a final weight, body composition measurement and end-of-study blood work. If the participants complete these visits, they will be compensated the same as if these vists had been completed at the end of the study.

**Safety Laboratory Procedures and Assessments**

Participants' screening/baseline laboratory assessment will include amylase/lipase, creatinine/ GFR, lipid profile, TSH, hemoglobin A1c, AST/ALT, calcitonin and hematocrit. If participants' screening laboratories are not within normal range, (they will be excluded from participation in the study.) they will be assessed by the study investigators and decision about participation in the study with be determined on an individual basis. For example, if the AST/ALT are greater than 3x the upper limit of normal the subject will not be included in the study.

- Participants' screening visit will also include a questionnaire to assess suicidality and depression, using the Patient Health Questionnaire (PHQ-9) (Appendix 2). Because the co-occurrence of depression is higher in individuals with obesity, we anticipate that many of our subjects may have minimal to mild depression. Subjects will be excluded from participation in the study if the result of the screen is greater than or equal to 10 (moderate to severe depression). They will also be referred to a psychologist if they score $\geq$ 10. Any concern for suicide or self harm will be immediately addressed by implementing emergency petition (EP) procedures and/or calling 911.
  Obese participants being treated with Liraglutide will have weekly phone calls with the study physicians or nurse practitioner to monitor for any adverse effects. In addition, during the first week, the study physicians or nurse practitioner will follow-up with the participant with two additional phone calls to answer any questions he/she may have about medication administration as well as to discuss any potential side effects. Participants will be provided with contact information should they need to contact the study physicians or nurse practitioner at any time throughout the study. The participants will be asked to report any side effects or worsening of depression, suicidal thoughts or behavior, and/or any unusual changes in mood or behavior, to the study physicians or nurse practitioner right away.
  Obese participants being treated with Liraglutide will have safety laboratory assessments performed at the following interval time points: week 0 (screening labs as above) and week 12-16 (end of study). These labs will include amylase, lipase, AST,

ALT, calcitonin and a urine pregnancy test. During phone sessions at weeks 4 and 8 to monitor for any adverse events, study personnel will also administer the PHQ-9 to again assess suicidality and any changes in mood. The PHQ-9 will also be administered at the at the second food snack task (week 12-16, end of study). Since the questionnaire relies on patient self-report, all responses will be verified by the PI and the PI will contact the subject to assess whether they need intervention and determine if they can continue in the study or if the drug should be discontinued. If at any time the subject scores $\geq$ 10 on the PHQ-9, they will be referred to a psychologist, and any concern for suicide or self harm will be immediately addressed by implementing emergency petition (EP) procedures and/or calling 911.

**Concomitant Medications**

- Other medication use will be assessed per exclusion criteria. If the participant is to take/start a new medication while in the study they will be asked to inform the study PI immediately to assess concomitant medication use.

**Diet and Other Considerations**

There are no dietary or food restrictions

**Duration of Therapy**

For Obese patients, Liraglutide will be administered daily for 12-16 weeks. If any of the following occur, the medication will be stopped:

- Pancreatitis (severe abdominal pain, increased amylase/lipase)
- Severe dehydration
- The participant chooses not to continue in the study

Lean controls will not be given study drug.

**OTHER STUDY PROCEDURES**

**Oral Glucose Tolerance Test (OGTT) and Anthropometric Measures**

After consent is obtained and at baseline prior to any study procedures, Anthropometric measures will be taken including weight, percent fat mass, and total fat mass (measured with a Tanita Body Fat Analyzer) and height (measured with a stadiometer). BMI will be calculated (weight [kg] /height [m2]). A urine pregnancy test will be performed on females.

Lean young adults will complete 1 OGTT (at baseline) and each obese young adult will complete 2 OGTTs (one at baseline and one at week 10 of the intervention with liraglutide).

Subjects will receive 75g glucose drink orally (Glucola). An intravenous (IV) catheter will be placed and blood samples will be taken for glucose and insulin assays at time points -20, 0, 10, 20, 30, 45, 60, 75, 90, 105, 120 minutes. In addition, fasting acyl-ghrelin, leptin, HgbA1c, GLP-1 will be drawn. Whole body insulin sensitivity index (WBISI) will be calculated (WBISI=10,000/[□(FPG x fasting plasma insulin) x (mean OGTT glucose concentration x mean OGTT insulin concentration)]49 as a measure of insulin resistance. No more than 50 ml of blood will be drawn on the OGTT day.

**Continuous Glucose Monitor (CGM)**

CGM enables measurement of glucose levels continuously throughout the day. To assess glucose variability, participants will be asked to use the CGM (FreeStyle Libre Pro) for up to 14 days. During their screening/baseline visit, a CGM sensor/transmitter will be placed on the participant's upper arm and attached with an adhesive patch by a trained study staff member. The sensor includes a wire-like tip which will be under the participant's skin in their fat tissue. The participants will be taught how to use the CGM. There are no restrictions on the participants while they wear the transmitter/sensor with regard to physical activity except for swimming. Participants will be blinded to the CGM monitor glucose levels as not to affect their activity or eating habits. The sensor will be removed by a trained study staff member by the end of day 14 of wear.

The CGM will be placed for a second time at the 8-week time interval during Liraglutide treatment and will be removed after 14 days of wear. Only obese participants will wear the CGM for a second time.

After the CGM is placed, participants will be provided with a BOOST® Original Nutritional Drink. They will be instructed to drink the BOOST on the follow day between 7:30 and 10:30am as a breakfast meal replacement (must drink on an empty stomach).

**fMRI**

The original study included use of functional MRI to assess brain perfusion in the response to drinking HFCS in healthy lean and obese adolescent/young adult males and females. However, in light of the recent pandemic we would like to shift the focus of the study towards the evaluation of the effect of GLP-1 analogue on food craving and intake as well as glycemic variailbity and metabolic hormonal changes in response to food.

**Plasma Collection**

Prior to the food snack task, a nurse will insert one IV catheter to sample blood for metabolite and hormone analysis (heated hand). Baseline measurements of leptin/ghrelin, insulin, GLP-1, cortisol and glucose will be collected. In addition, to assess how metabolic response to drinking HFCS relates to the neural responses observed, plasma samples will be obtained for leptin/ghrelin, insulin, GLP-1, glucose, and cortisol at specified interval time points. No more than 110mLs of blood will be drawn on each food snack task day.

**Eating Behavior Assessments**

To assess food preference and food intake before vs. after liraglutide intervention, eating behavior will be assessed in three ways;

**Lab-based Food Brunch Task**: At baseline and during the last week of treatment, each participant will undergo the food brunch task. Participants will be given the opportunity to eat ad libitum for 30 minutes with detailed measurement of food/drink consumed and video recording to assess eating topography. We will obtain signed permission prior to any video recording and the participant will know when they are being videotaped. The videotape recording will be used for research only. The recording will not be shown to the general public and will not be identified with the subject's name, although their face may be included in the recording. Declining to be videotaped will not affect the participant's study participation or

treatment in any way.

The food brunch task is adapted from Dr. Sinha on the basis of previous work on exposure to food cues and foods in the laboratory, which indicates that sufficient quantities, multiple food options and sufficient time available for eating are important variables modulating food intake. In the food brunch task, participants will be presented with a choice of the following foods, presented in 6 discrete bowls: skittles, M&Ms, chips, pretzels, cookies and popcorn. They will be offered the food and drinks and allowed to eat and drink as much as they like for 30 minutes. Video recordings will be assessed for frequency of times each bowl is touched/reached for, frequency of pick-ups and number of bites taken. The type and amount (weight and kilocalories) of food/drink consumed by each subject will be assessed before and after each session. All remaining items will be weighed by trained staff in the Yale Center for Clinical Investigation Metabolic Kitchen.

The instructions will state: “There are different foods and drinks on the tray. We will leave these here for you and you can eat and drink as much or as little as you like during the next half hour”. We will draw labs at the following time intervals during the food brunch task: 0, 15 and 30 minutes.

Participants will also be asked to complete the following questionnaires during each food snack test visit at 4 time points (baseline and then q 10 minute intervals): Hunger Scale, Anxiety Scale, DES-R and Food Craving Scale.

**Real-life dietary intake**: Participants will complete a detailed 1 week food record via a smartphone app, MetricWire. This will be administered during the 14 days that the participant is wearing the CGM. For obese participants, this will be completed a total of 2 times (obese participants wear the CGM 2 times). For lean participants, this will be completed once (lean participants wear the CGM once). During these time points, a 24-hour food recall, the National Cancer Institute’s (NCI) Automated Self-Administered 24-hour (ASA24®) dietary assessment tool, will also be administered and must be completed on 2 week days and 1 weekend day. Study personnel will call participants to complete the ASA24 over the phone.

Each participant will be taught how to accurately record their food and drink intake by the study registered dietician, doctors and/or nurse practitioner.

Participants will also be asked to complete the following questionnaires at the screening visit to assess food cravings and consumption and eating behaviors: Yale Food Addiction Scale (YFAS), Food Craving Inventory (FCI) and Dutch Eating Behavior Questionnaire (DEBQ). For obese participants only, these questionnaires will be repeated at the second food snack task visit.

> **ASA24:** The National Cancer Institute (NCI) created the Automated Self-Administered 24-hour (ASA24®) dietary assessment tool, a web-based tool that enables multiple, automatically coded, self-administered 24-hour recalls. The ASA24 system is freely available for use by researchers, clinicians, and teachers and can be used by researchers for epidemiologic, interventional, behavioral, or clinical research. Clinicians can utilize this system to collect 24-hour recalls or food records from patients and receive complete nutrient analysis of the foods and beverages consumed during the collection timeframe. Since ASA24 was released in 2009, greater than 3,900 studies have registered to use ASA24 and over 355,000 recall or record days were collected, as of December 2017. The ASA24 system consists of a Respondent Website used to collect dietary intake data and a Researcher Website

used to manage study logistics and obtain nutrient and food group data files. Researchers do not provide the National Cancer Institute (NCI) or the ASA24 system with any identifying data for study Respondents. Rather, Researchers specify a user ID for each Respondent and download system-generated usernames and encrypted passwords that Respondents use to access the application. The ASA24 system also does not collect any identifying data directly from Respondents. However, IP address information is accessed for the purpose of routing information between the server and the Respondent's computer -- often the IP address is that of the user's Internet Service Provider (ISP). IP addresses are not stored or tracked by the ASA24 system. However, logs of connections are kept for audit trail purposes. This information is not mined in any way but would be available if there were a legal obligation to release it.

**Other:** Subjects unable to attend in person visits due to COVID-19 will be sent a weight measurement scale.

## ASSESSING AND REPORTING ADVERSE EVENTS

### Definition

An adverse event is defined as any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product and which does not necessarily have to have a causal relationship with this treatment. An adverse event can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of a medicinal product or protocol-specified procedure, whether or not considered related to the medicinal product or protocol-specified procedure. Any worsening (*i.e.,* any clinically significant adverse change in frequency and/or intensity) of a preexisting condition that is temporally associated with the use of enoxaparin is also an adverse event.

Adverse events may occur during the course of the use of the IP in clinical trials or within the follow-up period specified by the protocol, or prescribed in clinical practice, from overdose (whether accidental or intentional), from abuse and from withdrawal.

**Serious adverse event.** A serious adverse event is an AE occurring during any study phase (i.e., treatment, follow-up), that fulfills one or more of the following criteria:

1. Results in death
2. Is immediately life-threatening
3. Requires in-patient hospitalization or prolongation of existing hospitalization
4. Results in persistent or significant disability or incapacity
5. Results in a congenital abnormality or birth defect
6. Is an important medical event that may jeopardize the subject or may require medical intervention to prevent one of the outcomes listed above

Each AE will be graded for severity per the NCI CTCAE version 4.0, and these criteria must be used in grading the severity of AEs. The criteria can be found at: http://ctep.cancer.gov/reporting/ctc.html

For those AEs, which are not listed as part of the NCI CTCAE version 4.0, the same grading system should be used, where:

- **Mild** corresponds to an event not resulting in disability or incapacity and which resolves without intervention

- **Moderate** corresponds to an event not resulting in disability or incapacity but which requires intervention
- **Severe** corresponds to an event resulting in temporary disability or incapacity and which requires intervention
- **Life-threatening** corresponds to an event in which the patient was at risk of death at the time of the event
- **Fatal** corresponds to an event that results in the death of the patient

**AE Expectedness**

AEs can be 'Unexpected' or 'Expected'. Expected AEs include Bleeding development of HIT, pain from subcutaneous administration of the drug, and anemia from blood draws.

Unexpected AEs are those AEs occurring in one or more patients participating in the research protocol, the nature, severity, or frequency of which is not consistent with either:

- The known or foreseeable risk of AEs associated with the procedures involved in the research that are described in (a) the protocol-related document, such as the IRB-approved research protocol, any applicable investigator brochure, and the current IRB-approved informed consent document, and other relevant sources of information, such as product labeling and package inserts; or
- The expected natural progression of any underlying disease, disorder, or condition of the patient(s) experiencing the AE and the patient's predisposing risk factor profile for the AE.

**AE Attribution**

The participating site investigator must attempt to determine if an AE is in some way related to the use of the study drug and define an attribution category. This relationship should be described as follows:

- Definite – The AE *is clearly related* to the study treatment.

  The event follows a reasonable temporal sequence from administration of the study drug, the event follows a known response pattern to the study drug and based on the known pharmacology of the study drug, the event is clearly related to the effect of the study drug. The AE improves upon discontinuation of the study drug and reappears upon repeat exposure.

- Probable – The AE *is likely related* to the study treatment.

  The event follows a reasonable temporal sequence from administration of the study drug, the event follows a known response pattern to the study drug AND the event cannot have been reasonably explained by an intercurrent medical condition OR the event cannot be the effect of a concomitant medication.

- Possible – The AE *may be related* to the study treatment.

  The event follows a reasonable temporal sequence from administration of the study drug and the event follows a known response pattern to the study drug BUT the event could have been produced by an intercurrent medical condition which, based on the pathophysiology of the condition, and the pharmacology of the study drug, would be unlikely related to the use of the study drug OR the event could be the effect of a concomitant medication.

- Unlikely – The AE *is doubtfully related* to the study treatment.

  AE does not have temporal relationship to intervention, could readily have been produced by the patient's clinical state, could have been due to environmental or other interventions, does not follow known pattern of response to intervention, does not reappear or worsen with reintroduction of intervention

- Unrelated – The AE *is clearly NOT related* to the study treatment.

The event is clearly due to causes distinct from the use of the study drug, such as a documented preexisting condition, the effect of a concomitant medication, or a new condition which, based on the pathophysiology of the condition, and the pharmacology of the study drug, would be unrelated to the use of the study. All SAEs will be reported to the project manager within 24 hours of the site becoming aware of the event.

**Procedure for Reporting Adverse Events**

All AEs will be recorded from the time the permission form is signed through the last dose of Liraglutide. AEs will be recorded on an Adverse Event log at and entered in OnCore within 72 hours of the PI becoming aware of the event.

All AEs will be reported to regulatory authorities, IRB and investigators in accordance with all local applicable laws and regulations.

All SAEs that are unexpected, which occur any time after the patient has consented up to the last dose of Liraglutide that are possibly, probably, or definitely related to the research notify the Yale Human Investigation Committee (within 5 days).

**Minimizing Risks**

**Intravenous (IV) catheter insertion and blood drawing:** The risks of bruising, clotting, and infection will be minimized by having venipuncture performed by trained and experienced personnel under sterile conditions. To avoid injury due to fainting, the IV will be inserted when the subjects are recumbent. The blood draws during the OGTT will be obtained from the already inserted catheter to minimize discomfort. Subjects will be asked to not participate in other studies which involve blood draws for 8 weeks after study completion. Additionally, nursing staff have been trained to be conscious of minimizing blood loss by re-infusing saline discards.

**Oral Glucose Tolerance Test (OGTT):** We use the Yale-New Haven Hospital (YNHH) standard of care guideline (blood sampling from a peripheral catheter) to re-infuse the blood saline discards. This method is used to prevent physiological anemia. Any symptoms which may occur during the IV catheter placement or OGTT will be addressed appropriately and if further interventions are indicated the proper physician referrals will be made.

**Continuous Glucose Monitor (CGM):** The CGM poses no major risks to the subjects. Participants may feel a mild discomfort (pin prick sensation) during the sensor insertion. Some individuals may experience black and blueness of the skin at the insertion site of the CGM sensor, which resolves by itself in a few days. Redness and discomfort (inflammation) can occur at the sensor insertion site. Rarely sensors may fracture and a small piece may remain under the skin which will need to be removed by one of the study doctors. This may cause mild discomfort, bruising, or temporary bleeding. The CGM catheter will be placed under sterile

conditions by experienced study doctors or nurse practitioner. In case of signs of inflammation or bruise at the sensor insertion site, the sensor will need to be removed immediately and standard care should be applied. The participants will be given our contact information if they have any questions, concerns or problems.

**Liraglutide (study drug):** Screening labs will be done prior to the start of the study to ensure that participants renal and pancreatic (exocrine) function is within normal limits. Phone calls will be done to ensure study drug is being tolerated by participants (as shown in detail in Appendix 1). Safety labs will be drawn at interval time points (weeks 4, 8 and 12-16) to ensure that renal, pancreatic, liver, and thyroid function remains within normal limits. In addition, a urine pregnancy test will be performed at these time points. The participant will be provided with a wallet card that includes the name of the drug and our contact information in the event that the participant receives outside care or experiences side effects and needs to contact us promptly (Appendix 4).

**Confidentiality**: Study participation is voluntary and subjects are informed that they are free to drop out at any time without penalty. All data will be kept confidential except in cases of imminent danger to the participants. Such limits to confidentiality will be clearly explained to participants verbally and in the written consent forms. Confidentiality in regard to collected materials will be maintained via a numbered reference system maintained by the investigators. Subjects' names will appear only on a consent form and a "key" form kept by the Project Director in locked filing cabinets. Only the PI, mentor, and main investigators will have access to any forms specifying both participant name and subject number. All number coded subjective and biological data will be kept in locked offices with access only to investigators and research staff. Furthermore, good clinical and research practice procedures and HIPAA regulations will be followed. No subjects are identified by name in any of the published literature and only by code in major data storage areas. The ASA24 have been designed to be compliant with HIPAA regulations. Both platforms encrypt data and do not collect any identifying data directly from participants.

## ANALYSES AND STATISTICAL CONSIDERATIONS

Analysis of subjective ratings, plasma hormonal measures, Brunch Food Task, and diet 3-day food measures will be assessed using a mixed-model repeated measures analysis (PROC MIXED SAS V9.4, Cary, NC) to assess main effects and interactions for the fixed factors of obesity status (lean vs. obese) and time (before vs. after intervention), and subjects will be the random effect factor to accommodate correlation between repeated measures. Covariate adjustment will be included for age and sex.

Using these random effects models applied to whole brain analysis for neural responses and mixed effects models for other analyses, the following specific hypotheses will be tested for each aim:

Sample size estimates are based on effect size determinations from our preliminary work and related previous research. Based on Jastreboff et al (2016)[9] findings from lean (n=14) vs. obese (n=24) adolescents, we found an overall large effect sizes between d=0.65 and 0.56 for the mPFC responses in the lean and obese adolescent groups, and d=0.68 and 0.53 for the striatal (caudate) response in the lean and obese adolescents as well as an effect size d=0.569 for lean vs. obese for the PFC connectivity with the striatum. The described fMRI liraglutide study by Farr et al.[20] (n=18) found effect size placebo=0.53 and liraglutide=-0.47 for decreased activation in the parietal cortex. There are no specific studies examining the effect of liraglutide on CBF response to HFCS ingestion. Based on these earlier studies, we will include 45 subjects in our study; specifically, 15 healthy lean and 30 obese adolescents to

test the proposed hypotheses.

For the adolescents, the goal is to have pilot data inclusive of adolescents with the food snack task alone – to obtain funding inclusive of adolescent age and add to literature. The effects of the medication on eating behavior are fairly robust which is why I believe we were able to see differences in initial pilot data. The main goal is to investigate the impact on eating behavior (assessed in the food snack task) before and after the medication (so baseline vs. post intervention). The lean participants serve as “controls” in that after the medication, the individuals with obesity have eating behavior more similar to that observed in lean. We also do within subject comparison (obesity before medication vs. after) - that I the main question.

## DATA MANAGEMENT AND RECORD KEEPING

The principal investigator is required to prepare and maintain adequate and accurate case histories designed to record all observations and other data pertinent to the investigation on each individual treated in the investigation. Data that are derived from source documents and reported on the CRF must be consistent with the source documents or the discrepancies must be explained. OnCore will be the designated electronic data capture tool. All data should be entered onto OnCore within 1 week of study entry.

## ADMINISTRATIVE AND REGULATORY

### Data safety and monitoring plan

Authority for patient safety will reside exclusively with the Principal Investigator. She will ensure data quality and completeness. The PI has the authority to recommend that the study protocol be terminated, temporarily suspended, or amended.

The data safety monitor for this study will be Jennifer Sherr, MD, PhD. Dr. Sherr is an Associate Professor in the department of Pediatrics (Pediatric Endocrinology) at Yale University School of Medicine. Dr. Sher is a licensed pediatric endocrinologist with over a decade of experience in conducting clinical trials in children and young adults with diabetes. Dr. Sherr is very familiar with the requirements for data safety monitoring. She also has several ongoing protocols with the Yale Human Investigation Committee (HIC). Dr. Sherr is not an investigator on the proposed study and would function as an independent monitor. Dr. Sherr will evaluate all Adverse and Serious Adverse Events and will assist the PI in preparing and sending the pertinent expedited reports to the appropriate persons as outlined above. She will monitor the study quarterly and review all adverse event sheets completed during that period. She will assist the PI in making critical decisions regarding subject's continuation in the study for safety reasons. She will review the summary of all Adverse Events for this study, which will be reported annually to the Yale Human Investigation Committee (HIC) and the ADA.

### Project Team Meetings

Scheduled meetings will be held weekly and will include the Principal investigators and research staff involved with the conduct of the protocol.

During these meetings, the investigators will discuss:

1. Safety of protocol participants (adverse events and reporting)
2. Validity and integrity of the data (data completeness on CRFs and complete source documentation)
3. Enrollment rate relative to expectation of target accrual (eligible and ineligible participants)
4. Retention of participants, adherence to the protocol and protocol violations
5. Protocol amendments

## CONFIDENTIALITY & SECURITY OF DATA

Data will be entered into OnCore. De-identified data will then be downloaded in Excel format for statistical analysis, which will be done on a HIPAA compatible, password protected encrypted laptop computer. All data entry will be performed by the study personnel. All source documentation will be retained at the research clinic in locked cabinets in locked offices.

## APPENDIX 1: TRIAL FLOW CHART

| | *Lean & Obese* | | *Obese Only* | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | **Visit 1 CSRU** | **Visit 2 CSRU** | | | | | | | **Visit 3 CSRU** | **Visit 4 CSRU** |
| | **Screen Visit/CGM placement** | **Food Snack Task** | **Week 1** | **Weeks 2-3** | **Week 4** | **Weeks 5-7** | **Week 8** | **Weeks 9-11** | **Week 10: CGM placement** | **Week 12-16** |
| **Date** | | | | | | | | | | |
| **Compensation** | $50 | $50 | | | | | | | $50 | $50 *+ $50 study completion bonus* |
| **Phone Call** | | | 2 calls | | | | | | | |
| **Side Effect Assessment** | | | | | | | | | | |
| **OGTT** | | | | | | | | | | |
| **Screen Labs** | | | | | | | | | | |
| **Labs** | | | | | | | | | | |
| **FCI & DEBQ** | | | | | | | | | | |
| **PHQ-9** | | | | | | | | | | |
| **CGM+BOOST, ASA 24 & App Food Record** | Remove 14 days later | | | | | | | | Remove 14 days later | |

## APPENDIX 2: PATIENT HEALTH QUESITONNAIRE (PHQ-9)

*Over the last 2 weeks, how often have you been bothered by any of the following problems?*

| | **Not at all** | **Several days** | **More than half the days** | **Nearly every day** |
|---|---|---|---|---|
| 1. Little interest or pleasure in doing things | 0 | 1 | 2 | 3 |
| 2. Feeling down, depressed, or hopeless | 0 | 1 | 2 | 3 |
| 3. Trouble falling or staying asleep, or sleeping too much | 0 | 1 | 2 | 3 |
| 4. Feeling tired or having little energy | 0 | 1 | 2 | 3 |
| 5. Poor appetite or overeating | 0 | 1 | 2 | 3 |
| 6. Feeling bad about yourself – or that you are a failure or have let yourself or your family down | 0 | 1 | 2 | 3 |
| 7. Trouble concentrating on things, such as reading the newspaper or watching television | 0 | 1 | 2 | 3 |
| 8. Moving or speaking so slowly that other people could have noticed. Or the opposite – being so fidgety or restless that you have been moving around a lot more than usual | 0 | 1 | 2 | 3 |
| 9. Thoughts that you would be better off dead, or of hurting yourself | 0 | 1 | 2 | 3 |

| **Total Score** | **Depression Severity** |
|---|---|
| 1-4 | Minimal depression |
| 5-9 | Mild depression |
| 10-14 | Moderate depression |
| 15-19 | Moderately severe depression |
| 20-27 | Severe depression |

## APPENDIX 3: PARTICIPANT INFORMATION SHEET

**Participant Information**

Subject ID: LIRA___

| **Appointments for this Study** | | | **Estimated Length:** |
|---|---|---|---|
| **Visit 1,** Screening/OGTT/CGM | **Date:** | **Time:** | **3 hours** |
| *This visit will involve blood work, a physical exam, and questionnaires. You will have an IV placed and get blood drawn. Study personnel will place a continuous glucose monitor. You will wear the CGM for 14 days. There are no restrictions with regard to physical activity except for swimming. The CGM will be removed by a physician or nurse practitioner at the end of 14 days.* | | | |
| **Visit 2,** Food Snack Task | **Date:** | **Time:** | **2 hour** |
| *This visit will involve blood work and a food snack task in the mid-morning. You will be taught about the medication, how to administer it and we will give you supplies. Once you start the medication, we will call you weekly to check in and make sure you are feeling well.* | | | |
| **Day 1,** Medication Starts | **Date:** | **Dose: 0.6mg** | |
| **Day 8,** Medication Dose Changes | **Date:** | **Dose: 1.2mg** | |
| **Day 15,** Medication Dose Changes | **Date:** | **Dose: 1.8mg** | |
| **Day 22,** Medication Dose Changes | **Date:** | **Dose: 2.4mg** | |
| **Day 29,** Medication Dose Changes | **Date:** | **Dose: 3.0mg** | |
| **Visit 3,** OGTT/CGM | **Date:** | **Time:** | **3 hour** |
| *This visit will involve questionnaires and blood work. We will also ask about any side effect you may be experiencing. We will place the CGM for a second time for you to wear for 14 days.* | | | |
| **Visit 4,** Food Snack Task | **Date:** | **Time:** | **2 hours** |
| *You will repeat the food snack task with blood work and questionnaires. After this visit the study medication will be discontinued.* | | | |

**Study Medications**

Take your medication as prescribed. Bring all study medication empty containers to each visit.

**Other Medications**

If during the course of the study you are prescribed any new medications, please call us before beginning the medication.

**Wallet Card**

If you require any medical care outside of the study while participating in this study, please show the other medical personnel this card. This card also has our contact information in the event that you experience any side effects and need to contact us promptly.

## APPENDIX 4: WALLET CARD

**RESEARCH STUDY PARTICIPANT AT YALE**

Participant name _____
is participating in a study of **Liraglutide.**
Protocol #2000022407
Please call Dr. Jastreboff at (203) 737- 4777
before taking any new medications.
Also call in case of a serious medical problem
or questions.

Thank you for your help.

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