Clinical Investigation of Efficacy of Tauroursodeoxycholic Acid (TUDCA) to Enhance Pancreatic Beta Cell Survival In Type 1 Diabetes by Reducing Endoplasmic Reticulum Stress

National Clinical Trial Identifier: NCT02218619

Document Date: March 9, 2021

1. Study Purpose and Rationale

Hypothesis: An agent (TUDCA) that reduces ER stress will protect against the deterioration of beta-cell function in new-onset T1D.

Rationale: Abnormalities of ER stress responses contribute to the beta cell failure in early-onset T1D. Treatment of pre-diabetic NOD mice with TUDCA preserves beta cell mass and insulin secretion (see Background). In mice, TUDCA treatment also decreases lymphocytic infiltration of the islets and improves survival and morphology of beta- cells. Patients with newly- diagnosed T1D have a residual beta cell mass of 30-50% (c-peptide often >0.2 pmol/mL); beta cell destruction continues for at least 12-24 months after diagnosis until C-peptide is generally low or undetectable.

In this pilot study in human subjects with T1D, we will enroll 20 new-onset (within 100 days of diagnosis) subjects with measurable stimulated C-peptide (>0.2 pmol/mL).

2. Study Design and Statistical Procedures Also see Trial Flow Diagram

Visit -1 -Screening visit (+/- 3 days) Each participant must sign a consent form prior to screening. The screening visit consists of a medical history, physical exam, assessment of other medications that they may be taking and a review of their insulin usage and blood glucose values over the previous three days, blood tests including liver function tests, a urine test and a combination 2 hour Mixed Meal Tolerance Test (MMTT).

For females only: A sample of their urine will be utilized for a urine pregnancy test during the screening. Only the participant will be told the results of this test. If the test is positive, then they will not be able to participate. Females who are lactating are also ineligible for this study.

Visit 2- Screening visit Week 0 (+/- 14 days) This visit and all subsequent visits will only occur if participants have been found eligible to continue on the trial. It will include a 3 mm punch of skin; discussion about any medications they may be taking, a review of home blood glucose monitoring results, a review of adverse events and a review of insulin usage over 3 days prior to this visit. A urine sample will also be tested for pregnancy if the participant is female and of childbearing potential. The participants will be provided with a 3-month supply of the study drug/placebo. You will start TUDCA/placebo at a dose of 1750 mg orally daily.

Visit 3- Interim Visit (Month 1) (+/- 14 days) This in-person visit will include discussion about any medications the particpant may be taking, review of home blood glucose monitoring results, a review of adverse events, and a review of insulin usage over 3 days prior to this visit. Blood tests will be done including liver function tests and a urine sample will also be tested for pregnancy if the participant is female and of childbearing potential. Participants will continue the study medication.

Visit 4- Interim Visit (Month 2) (+/- 14 days) This phone visit will include discussion about any medications participants may be taking, review of home blood glucose monitoring results, a review of adverse events, and a review of insulin usage over 3 days prior to this visit. Participants will continue the study medication.

Visit 5 - Interim Visit (Month 3) (+/- 14 days) This in-person visit will include a physical exam, a discussion about any medications the participant may be taking, review of home blood glucose monitoring results, a review of adverse events, and a review of insulin usage and blood glucose values over 3 days prior to this visit. The participant's blood will be drawn for liver function tests and HbA1c. A urine sample will also be tested for pregnancy if the participant is female and of childbearing potential. Participants will be provided with a 3-month supply of the study drug/placebo. Participants will continue the study medication.

Visit 6- Interim Visit (Month 6) (+/- 14 days) This in-person visit will include a physical exam, a discussion about any medications the participant may be taking, review of home blood glucose monitoring results, a review of adverse events, and a review of insulin usage, and blood glucose values over 3 days prior to this visit. The participant will complete a 2 hr Mixed Meal Tolerance Test and have blood drawn for a HbA1c and liver function tests. A urine sample will also be tested for pregnancy if the participant is female and of childbearing potential. Participants will be provided with a 3-month supply of the study medication. Participants will continue the study medication.

Visit 7- Interim Visit (Month 9) (+/- 14 days) This in-person visit will include a discussion about any medications participants may be taking, review of home blood glucose monitoring results, a review of adverse events, and a review of insulin usage over 3 days prior to this visit. Participants will have blood drawn for a HbA1c and liver function tests. Participants will be provided with a 3-month supply of the study drug/ placebo. Participants will continue the study medication.

Visit 8- Interim Visit (Month 12) (+/- 14 days) This visit will include a physical exam, a discussion about any medications participants may be taking, review of home blood glucose monitoring results, a review of adverse events, and a review of insulin usage and blood glucose values over 3 days prior to this visit. Participants will complete a 2 hr Mixed Meal Tolerance Test and have blood drawn for a HbA1c and liver function tests. Medication will be stopped at the end of 12 months.

Visit 9- Interim Visit (Month 15) (+/- 1 month) This phone visit will include discussion about any medications participants may be taking, review of home blood glucose monitoring results, a review of adverse events, and a review of insulin usage over 3 days prior to this visit.

This in-person visit will include a physical exam, a discussion about any medications participants may be taking, review of home blood glucose monitoring results, a

review of adverse events, and a review of insulin usage and blood glucose values over 3 days prior to this visit. Participants will complete a 2 hr Mixed Meal Tolerance Test and have blood drawn for a HbA1c and liver function tests.

i. Endpoints: The primary endpoints will be the area under the stimulated C-peptide curve of a 2- hour mixed meal tolerance test (MMTT) conducted at screening, and during the 12 months of drug treatment at 6 and 12 months and at 6 months after drug or placebo is stopped.

The study will also examine as secondary endpoints the effect of the experimental treatment on diabetes-specific autoantibodies and metabolic parameters. Interim metabolic assessments will consist of periodic review of glucose records and reports of hypoglycemia, insulin dose and HbA1c.

ii. Trial Design: This single-center double-masked, Phase II placebo-controlled study will examine the efficacy of TUDCA to arrest beta cell hypofunction/destruction in 20 subjects with new- onset T1D (ages 18-45 years), randomized within 100 days of diagnosis). All subjects will receive standard intensive diabetes treatment with insulin and dietary management, consistent with American Diabetes Association Standards of Care. Diabetes management will be done by the primary diabetes care team at the Berrie Center. Subjects will be randomly assigned 1:1 to receive either daily TUDCA 1750 mg/day or placebo for 12 months. All subjects will be followed for until study end and will be offered option for re-contact in the informed consent so that longer-term study follow-up is a possibility. Enrollment is expected to occur over 18 months. The mean c-peptide responses to MMTT in the TUDCA treatment group will be compared to the placebo-treated group at 0, 6, 12 and 18 months. The study will also examine the effects of TUDCA treatment on other T1D- related metabolic and immunologic outcome measures.

iii. Exploratory outcome: If a significant change in c-peptide responses are detected in both groups, we will compare the *in vivo* responses to ER stress relief to in-vitro responses to ER stress relief. In these subjects, we will compare the clinical response to an agent that reduce ER stress to the response of the iPS-derived beta cells, derived from the same subject, when treated with the same experimental agent to reduce ER stress.

iv. Treatment and Dose: Subjects will be treated with either TUDCA at a dose of 1750 mg/day or placebo for 12 months with an additional 6 months of follow-up. 1750 mg is the most commonly used dose used to treat biliary disease and was used in a recent study of human obesity and insulin resistance (39). In this 4 week study, an effects on ER stress was not observed on adipose cells; however we plan a longer treatment time period and hypothesize that since TUDCA undergoes extensive

hepatic metabolism, the exposure of pancreatic beta cells to drug at this dose may be sufficient to cause an effect on ER stress. In addition, in vitro studies will enable us to manipulate exposure of beta cells to a range of TUDCA concentrations, thus revealing extent of individual differences in sensitivity to the agent.

v. Duration: All subjects will be followed for 1 year of treatment and at least 6 months of follow-up.

vi DSMB: The progress of the study will be monitored by the DSMB which will review safety data and make recommendations regarding continuation, termination, or modification of the study. Based on an 18 month enrollment period and an additional study period of at least 6 months, the DSMB will formally review the safety data, enrollment, and clinical outcome data at least twice yearly.

vii The number of subjects who discontinue study treatment will also be included in the reports prepared for the DSMB.

Statistical analysis: These studies are pilots in which we anticipate substantial inter-individual differences in responses to the proposed *in vivo* intervention (TUDCA). We will compare the clinical endpoints between treated and control subjects using repeated measures ANOVA, treating (if indicated) age, gender and BMI as covariates. The endpoints of the in vitro studies will be analyzed for differences between individuals regardless of the experimental group to which they have been randomized. These analyses will inform us of the range of autonomous beta cell differences in response to ER stress, and to the mitigation conveyed by TUDCA. These data will then be related to clinical endpoints, probing for the ability of the *in vitro* studies to predict (correlate with) clinical course and response to TUDCA. We are experienced in all of the relevant analytic techniques and have the software on hand. If necessary, we have access to expert statistical consultations through the Columbia NIH CTSA.

3. Study Procedures MMTT and skin biopsy

Aim: To assess the performance of canonical ER stress response pathways, including the ability of TUDCA to meliorate such stress *in vitro* in beta cells of the subjects of this study.

Rationale: As noted, there is growing recognition that ER stress plays an important role in beta cell responses to the immunologic and metabolic stresses that are the hallmarks of T1D. Pharmacological “relief” of ER stress is beneficial in rodent models of diabetes. Molecules that have these effects (e.g. TUDCA) are very well tolerated in children and adults to whom they have been administered for other indications.

Hypothesis: In patients with very recent onset of clinical T1D, TUDCA

administration will slow or prevent the advance of the disease.

Anticipated outcome: Individuals treated with TUDCA will show a trend toward preservation of beta cell mass and function as reflected in stimulated C-peptide in MMTT at 6 and 12 months of treatment compared to placebo; it is possible that if observed, this effect will still be present 6 months after cessation of treatment or later.

4. Name of investigational product: TUDCA

i. Risks and Benefits: The known risks of drug treatment: In a summary of post-marketing data of 37 trials involving 1283 subjects treated with TUDCA up to 1750 mg/day for up to 1 year for severe liver disease, the most common side effect was diarrhea that occurred in .01% subjects. Less frequent adverse events were nausea, abdominal pain, pruritis, and headache.

ii. Study drug and placebo are manufactured and supplied by Bruschettini (Genoa, Italy) and provided at no charge to study participants. The study drug is administered orally twice daily.

iii. Population to be studied: New-onset T1D. We will recruit individuals between the ages of 18 and 45 with new- onset T1D (within 100 days of diagnosis). All patients will be in otherwise good health. Inclusion criteria will include: at least one diabetes- related autoantibody present (insulin autoantibody alone will not be sufficient in subjects treated with insulin for > 14 days) and a stimulated C-peptide levels > 0.2 pmol/ml measured during a mixed meal tolerance test (MMTT) conducted at least 21 days from diagnosis of diabetes and within one month (37 days) of randomization to either TUDCA or placebo. Pregnant women will be excluded. Participants of childbearing potential and/or their partners must agree to use acceptable methods of birth control beginning at the pre-study visit and throughout the study.
Examples of acceptable methods of birth control include abstinence, or 2 of the following: intrauterine device (IUD-with or without local hormone release), diaphragm, spermicides, cervical cap, contraceptive sponge, and /or condoms.

iv. Drug handling/distribution/administration is conducted in accordance with the policies of the Research Pharmacy and Columbia University Medical Center

5. Study Instruments (e.g., Questionnaires, Interview Outlines, Focus Group Guides): N/A

6. Study Subjects

I. Subject Inclusion Criteria

1. Be between the ages of 18 and 45 years.
2. Be within 3 months (100 days) of diagnosis of T1D based on ADA criteria.
3. Must have at least one diabetes-related autoantibody present.
4. Must have stimulated C-peptide levels > 0.2 pmol/ml measured during an MMTT conducted at least 21 days from diagnosis of diabetes and within one month (37 days) of randomization.
5. If participant is female with reproductive potential, she must be willing to avoid pregnancy and have a negative pregnancy test during the 12 months of treatment and for an additional 3 months.
6. Must be willing to comply with intensive diabetes management.
7. Must be willing to undergo 3 mm skin biopsy at randomization.

II. Subject Exclusion Criteria

1. Be currently pregnant, lactating or anticipating getting pregnant.
2. Ongoing use of medications known to influence glucose tolerance.
3. Have serologic evidence of current or past HIV, hepatitis B, or C infection.
4. Have any complicating medical issues or abnormal clinical laboratory results including abnormal liver function test (more than 1.5 x ULN) that interfere with study conduct or cause increased risk.
5. Have a history of malignancy.
6. Currently using non-insulin drugs that affect glucose control.
7. Currently using drugs that may interfere with TUDCA absorption or effect such as bile acid sequestering agents (cholestyramine and colestipol), aluminum based antacids, estrogens, oral contraceptives and clofibrate.
8. Currently participating in another T1D treatment trial or Sensitivity to TUDCA or other similar agent.

iii. Subject withdrawal criteria: An intent–to-treat approach will be used. Subjects will not be replaced. All data acquired prior to termination for the reasons outlined below will be included in the primary analysis unless a participant withdraws consent. Every effort will be made to conduct a final study visit with the participant and participants will be followed clinically until, if applicable, all adverse events resolve. All subjects will be offered clinical follow-up at the Berrie Center for T1D.

Reasons for withdrawal and stopping rules: The drug will be stopped by the investigator because of serious and unexpected adverse events, safety laboratory parameters and/or vital signs. Subject may be withdrawn by the investigator; the subject my withdraw from the study; there may be an Intercurrent illness or event that precludes further to the study site or ability to evaluate disease.

Treatment of Subjects: Participants will be treated with either TUDCA at a dose of 1750 mg/day or placebo for 12 months, with study visits every 3 months during treatment, and undergo at least 6 months of follow-up, depending upon time of randomization. The study will be stopped based on occurrence of serious and unexpected adverse events, safety laboratory parameters and/or abnormal vital signs. Subjects will agree to avoid the following drugs during the study: non-insulin drugs that affect glucose control, bile acid sequestering agents (cholestyramine and colestipol), aluminum based antacids, estrogens, oral contraceptives and clofibrate. Subjects will be offered option of ongoing clinical care at Berrie Center for T1D as well as option for re-contact in informed consent, providing possibility for longer- term follow-up studies to be performed in the future on these subjects.

Recruitment: Participants will be recruited from patients seen at the Berrie Center. Researchers will not directly approach a patient for recruitment until the patient has been informed by the study by their physician who has ascertained that the patient is willing to discuss the study with the investigators.

7. Informed consent process: Consent will be obtained by study investigator, Dr. Robin Goland or by the study coordinator Ellen Greenberg. Consent will be obtained verbally in-person.

v. Duration: All subjects will be followed for 1 year of treatment and at least 6 months of follow-up.

vi DSMB: The progress of the study will be monitored by the DSMB which will review safety data and make recommendations regarding continuation, termination, or modification of the study. Based on an 18 month enrollment period and an additional study period of at least 6 months, the DSMB will formally review the safety data, enrollment, and clinical outcome data at least twice yearly.

vii The number of subjects who discontinue study treatment will also be included in the reports prepared for the DSMB.

8. Confidentiality of Study Data: Subjects' medical and research records will be confidential to the extent permitted by law. Efforts will be made to keep subjects' personal information private. However, there is no guarantee for complete confidentiality. Subjects will be identified by a code, and personal information from your records will not be released without the subject's written permission. When information from this study is published, subjects will not be identified by name. All end-user devices will be encrypted and all data will be password protected.

9. Privacy Protections: Study will be carried out in accordance with all HIPPA regulations.

10. IND Holder Responsibilities

a) Ensure proper monitoring: The Sponsor-Investigator will ensure that the study protocol is followed, the data is accurate and the research subjects are safe. Safety data will be reviewed by the Sponsor-Investigator or designee as soon as it is received. As per the "AE Reporting" document that was submitted as part of the protocol, events that are considered to be serious and unexpected "Suspected Adverse Reactions" or SARs, will be submitted to the FDA as soon as possible, but no later than 15 calendar days from notification, and unexpected fatal or life-threatening SARs will be submitted no later than 7 calendar days from initial notification. Events that are considered to be "Unanticipated Problems" or UPs, will be submitted promptly, but no later than 7 calendar days following the occurrence of the UP or knowledge of the UP. The S-I will also notify the IRB and FDA of any serious deviations from the protocol and any new information indicating added risks to subjects.

The S-I will review 100% of all source documentation of data collected including all safety data. The regulatory binder, along with other essential study documents, will also be reviewed to ensure that it is complete and up to date. The first review will occur after the first patient is enrolled and will continue periodically with new enrollments. Any discrepancies will be discussed with relevant study personnel for clarification and/or correction.

b) Ensuring the study is conducted in accordance with the protocol: The Sponsor-Investigator will continually monitor the study to ensure that the study is conducted in compliance with the IRB approved study protocol. Research personnel will be trained on the protocol, and the S-I will provide oversight to staff to ensure compliance. Potential subjects, research subjects, and study progress will be discussed at the weekly Type 1 Diabetes Departmental meeting. The performance of all aspects of the study, including the methods used to obtain informed consent, will be conducted in accordance with all applicable state and federal guidelines as well as the principles for protection of research subjects as outlined in the declaration of Helsinki, ICH Guidelines for Good clinical practice, applicable local health authority regulations and US Title 21 of the Code of Federal Regulations.

c) Review of ongoing investigations; evaluate safety and efficacy and report to the FDA and IRB: The progress of study will be monitored by the Data and Safety Monitoring Board (DSMB) which will review safety data and make recommendations regarding continuation, termination, or modification of the study. The DSMB will formally review the safety data, enrollment, and clinical outcome data at least twice yearly. In addition, safety data will be reviewed by the Sponsor-Investigator or designee as soon as it is received, and will report findings to the FDA and IRB as appropriate.

d) Keep and retain records/documentation: Adequate and accurate case histories on

all subjects' participation in the trial will be kept. This includes source data, progress notes, concomitant medications, subject eligibility and documentation of consent as well as signature and date of staff obtaining data. A record will be maintained of all monitoring activities, findings, conclusions and actions taken to correct deficiencies. Submissions to the FDA and other FDA correspondence will also be filed. All data collected from the study will be maintained in charts and password protected computers in a locked facility with limited access. Only the S-I and designee(s) will have access to the records. Confidentiality of data and protection of privacy of subjects will be ensured following the IRB policy and federal regulations 45 CRF 46.111(a)(7) and 21 CRF 56.111(a)(7).

e) Submit amendments, IND Safety Reports, and Annual reports to the FDA: Protocol and information amendments will be submitted to the FDA in accordance with 21 CFR 312.30 and 21 CFR 312.31, respectively.

Safety reporting will be submitted to the FDA in accordance with 21 CFR 312.32 and as outlined in the "AE Reporting" document. Sponsor-Investigator will submit IND Safety Reports for any adverse experiences that are both serious and unexpected and associated with the use of the drug. Serious and unexpected suspected adverse reactions will be reported to FDA as soon as possible, but no later than 15 calendar days following the S-I's initial receipt of the information. Unexpected fatal or life-threatening suspected adverse reactions will be reported to FDA as soon as possible, but no later than 7 calendar days following the S- I's initial receipt of the information. Any relevant information not reported in the initial IND safety report will be submitted as a follow-up report no later than 15 calendar days after the S-I receives the information.

Annual reports will be submitted to the FDA within 60 days of the anniversary date that the IND went into effect. Annual reports include information on the progress of the investigation, as well as a summary of all safety reports submitted.