Protocol (a) I8Z-MC-APCA

A Randomized, 9-Way, Single-Dose, Crossover Study to Evaluate the Pharmacokinetics, Pharmacodynamics, Safety, and Tolerability of LY3185643 and rGlucagon in Healthy Subjects

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LY3185643

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Clinical Pharmacology Protocol Electronically Signed and Approved by Lilly:
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1. Protocol Synopsis

Title of Study: A Randomized, 9-Way, Single-Dose, Crossover Study to Evaluate the Pharmacokinetics, Pharmacodynamics, Safety, and Tolerability of LY3185643 and rGlucagon in Healthy Subjects

Rationale: The availability of insulin pumps and continuous glucose monitors has transformed diabetes care by allowing continuous infusion of insulin with bolus doses administered based on food intake. Hypoglycemia may result from excessive insulin administration in relation to food intake, disease state, or physical activity. Hypoglycemia may be life-threatening; this is a risk with any insulin administration, and may be of concern with automated systems.

An artificial pancreas device system (APDS) may provide better control of blood sugar with less episodes of hypoglycemia. A bihormonal APDS combines automated delivery of both insulin and glucagon. Pilot studies using investigational, bihormonal APDSs have demonstrated that delivery of small doses of glucagon in response to declining glucose levels reduces the frequency of hypoglycemia. Glucagon opposes the effects of insulin in the liver and increases blood glucose levels.

The current commercial glucagon (recombinant glucagon [rGlucagon]) preparations are only available as lyophilized powder that must be reconstituted with a diluent before administration. The current Lilly formulation of rGlucagon also has poor solubility in aqueous buffers at or near physiological pH values and poor chemical and physical stability when formulated at low and high pH.

LY3185643 is a glucagon analog with improved solubility that is being developed for use in a bihormonal APDS. The current study is designed to assess the pharmacokinetic (PK) and pharmacodynamic (PD) profiles of LY3185643 and compare these to rGlucagon.

Objectives/Endpoints:

Primary Objective	**Endpoints**
The primary objective of this study is to evaluate the PK and PD profiles of LY3185643 and rGlucagon after single doses of study treatment	C_{max}, AUC, and terminal half-life of LY3185643 and rGlucagon C_{max} and AUC (change from predose) of blood glucose and C-peptide
Secondary Objective	**Endpoints**
To evaluate the safety and tolerability of LY3185643 after single doses of study treatment	Adverse events

Abbreviations: AUC = area under the concentration versus time curve; C_{max} = maximum concentration; PD = pharmacodynamic; PK = pharmacokinetic; rGlucagon = recombinant glucagon.

Summary of Study Design: This study is a Phase 1, single-center, subject- and investigator-blind, randomized, 9-way, 3-period crossover study in healthy subjects to evaluate the PK and PD of LY3185643 and rGlucagon after subcutaneous (SC) administration.

Up to 37 healthy men and women may be enrolled to target approximately 18 subjects to complete the study. Each subject will receive 3 doses on each of 3 dosing days (total of 9 doses: 5 doses of LY3185643 and 4 doses of rGlucagon) administered SC. Doses will be administered in a 9-way complete crossover design in 3 periods, and subjects will be randomized to predefined treatment sequences.

Each study period will consist of 3 days (Days -1 to 2). Subjects should return for the next period with a minimum of 3 days between treatment periods.

Blood samples will be collected for the determination of concentrations of LY3185643, glucagon, glucose, C-peptide, and for immunogenicity. Safety data will include clinical examinations, vital signs, electrocardiograms (ECGs; including telemetry), clinical laboratory tests, and a record of adverse events (AEs).

Treatment Arms and Duration: Each subject will be randomized to 1 of 9 treatment sequences comprising single SC doses of LY3185643 or rGlucagon. Subjects will receive 9 doses total during 3 treatment periods (3 doses per period). Each treatment period will consist of 3 days with a minimum of 3 days between each period. A follow-up visit will be required at least 30 days after completing the last dose of study treatment.

Number of Subjects: Up to 37 subjects may be enrolled so that approximately 18 subjects complete the study.

Statistical Analysis: PK and PD analyses will be conducted on the full analysis set. This set includes all data from all randomized subjects receiving at least 1 dose of the investigational product and with a baseline and at least 1 postbaseline measurement for each dose according to the treatment the subjects actually received. Safety analyses will be conducted for all enrolled subjects whether or not they completed all protocol requirements.

Sample Size: The sample size is customary for Phase 1 studies evaluating safety, PK, and/or PD parameters and is considered sufficient to evaluate the primary objective of this study.

Safety: All investigational product and protocol procedure AEs will be listed, and if the frequency of events allows, safety data will be summarized using descriptive methodology. Other safety parameters that will be assessed include safety laboratory parameters, vital signs, and ECG parameters. The safety parameters will be listed and summarized using standard descriptive statistics.

Pharmacokinetics: PK parameter estimates for LY3185643 and rGlucagon will be calculated by standard noncompartmental methods of analysis. The primary parameters for analysis will be maximum drug concentration (C_{max}) and area under the concentration versus time curve (AUC). Other noncompartmental parameters, such as half-life, apparent clearance, and apparent volume of distribution, may be reported. Population PK and exposure-response analyses may be performed using PK and glucose data.

PK parameters for LY3185643 and rGlucagon will be summarized for each dose group. PK parameter estimates will be evaluated to delineate effects of dose proportionality.

Pharmacodynamics: The PD time-action and response of LY3185643 and rGlucagon will be compared to establish the relative potency between LY3185643 and rGlucagon. The primary PD response variables of interest include time to C_{max} (T_{max}), C_{max}, and change from baseline to each time point, and incremental AUCs for glucose and C-peptide. A dose-proportionality parameter will also be estimated to establish the relative potency between LY3185643 and rGlucagon. Partial AUC for glucose over appropriate time intervals will be calculated using the trapezoidal rule.

Parameters will be analyzed using a nonlinear mixed-effects model with period, treatment (LY3185643 or rGlucagon), and dose (within treatment) as fixed effects, baseline glucose as a covariate, and subject as a random effect. A conversion factor will be defined in the model for each parameter to estimate the relative potency of LY3185643 to rGlucagon. In addition, a conversion factor will be estimated between LY3185643 and rGlucagon

for multiple parameters simultaneously using a similar mixed nonlinear model approach to account for the correlation between parameters for the same subject.

All PD parameters including the baseline-corrected parameters will be summarized by dose group and tabulated. Summary statistics will be provided. The individual observed and mean (by dose group) time profile of the PD measurements after dosing will be plotted.

Immunogenicity: The frequency of antibody formation to LY3185643 will be determined. If a neutralization assay is performed, the frequency of neutralizing antibodies will be determined. The relationship between the presence (or absence) of antibodies and AEs will be assessed. Likewise, the relationship between the presence of antibodies and the PK parameters and PD response to LY3185643 will be assessed.

Interim Analysis: No interim analyses are planned for this study.

The Lilly study team is unblinded, and the investigator will remain blinded until the study is completed. Data may be analyzed while the trial is ongoing, but no changes to the study design are planned. An assessment committee will not be formed.

2. Schedule of Activities

Study Schedule Protocol I8Z-MC-APCA

Procedure	Screen	Periods 1, 2, 3[a]			FU/ET[b]	Comments
	Up to Day -28	Day -1	Day 1[c]	Day 2		
Informed consent	X					
Admission to CRU		X				
Discharge from CRU				X		
Carbohydrate-rich meal		X				Subjects will be provided a meal approximately 10:00 PM and will remain fasted until completion of the study procedures on Day 1.
Treatment administration			X (3 doses)			Subjects will receive 3 doses of study treatment, each separated by at least 4 hours (for example, 8:00 AM, 12:00 PM, 4:00 PM).
Medical history	X					
Height	X					
Weight	X	X			X	
AEs/concomitant medications	X		X	X	X	Collected throughout the study.
Vital signs (minutes)	X		-20 (±10), 15 (±5), 30 (±5), 45 (±5), 60 (±5), 90 (±15), 120 (±15), 180 (±15), 210 (±15)	X	X	Supine blood pressure and/or pulse rate may be measured as clinically indicated. Sampling times are relative to the time of each study drug administration.
Clinical laboratory tests	X		Any time before the first dose		X	Period 1 only. See Appendix 2, Clinical Laboratory Tests, for details.
Pregnancy test	X	X			X	Urine pregnancy tests will be performed. See Appendix 2, Clinical Laboratory Tests, for details.
Physical examination/ medical assessment	X			X	X	Physical examination at screening. Thereafter, medical assessment and targeted examination, as appropriate.
12-lead ECG (minutes)	X		-20 (±10)		X	Single ECGs will be collected for safety. Predose ECGs must be recorded before collecting any blood for safety or PK tests.
ECG telemetry		X	X			The ECG waveform and cardiac rhythm will be monitored continuously using an ECG telemetry monitor during the inpatient stay at the CRU, from the night of Day -1 to 4 hours after the last dose on Day 1 for all 3 periods.

Study Schedule Protocol I8Z-MC-APCA

Procedure	Screen	Periods 1, 2, 3[a]			FU/ET[b]	Comments
	Up to Day -28	Day -1	Day 1[c]	Day 2		
Continuous ECG monitoring			X			15 ECGs will be extracted from the continuous ECG recording at -5, 30, 60, 120, and 180 minutes postdose for each Day 1 dose administration.
PK samples: LY3185643 or glucagon (minutes)			0 (predose), 15 (±2.5), 30 (±2.5), 60 (±5), 120 (±10), 180 (±10)			Sampling times are relative to the time of each study drug administration.
Glucose samples (minutes)			-5 (±2.5), 0 (predose), 5 (±1), 10 (±1), 15 (±2.5), 22 (±2.5), 30 (±2.5), 45 (±5), 60 (±5), 75 (±5), 90 (±5), 105 (±5), 120 (±5), 150 (±5), 180 (±5)			Blood concentrations using YSI glucose analyzer. Sampling times are relative to the time of each study drug administration.
PD samples: C-peptide, glucose (minutes)			-5 (±2.5), 0 (predose), 5 (±1), 15 (±2.5), 30 (±2.5), 60 (±5), 120 (±5)			Sampling times are relative to the time of each study drug administration.
Immunogenicity (LY3195643 antibodies) samples			Any time before the first dose		X	Period 1 and Period 3 only. Prior to the first dose only.

Abbreviations: AE = adverse event; CRU = clinical research unit; ECG = electrocardiogram; ET = early termination; FU = follow-up; PD = pharmacodynamics; PK = pharmacokinetics.

Note: The site should schedule activities as appropriate. In cases where several study procedures are scheduled at the same time, the order of priority for these procedures will be as follows: PK samples, PD samples, vital signs, ECG, clinical laboratory samples.

[a] Each treatment period will consist of 3 days (Days -1 to 2). Subjects should return for the next period with a minimum of 3 days between treatment periods.

[b] FU/ET visit will occur at least 30 days after completing the last dose of study treatment.

[c] Study drug will be administered 3 times on Day 1 in each period. Day 1 activities will be performed 3 times on Day 1 with the sampling times corresponding to each study drug administration.

3. Introduction

3.1. Study Rationale

The Diabetes Control and Complications Trial (DCCT) and its observational Epidemiology of Diabetes Interventions and Complications (EDIC) follow-up study demonstrated that intensive therapy (that lowers glycemia) reduced renal and cardiovascular disease, the predominant causes of early mortality in patients with type 1 diabetes mellitus (T1DM) (Nathan et al. 2005; DCCT/EDIC Research Group et al. 2011).

The availability of insulin pumps and continuous glucose monitors has transformed diabetes care by allowing continuous infusion of insulin with bolus doses administered based on food intake. Hypoglycemia may result from excessive insulin administration in relation to food intake, disease state, or physical activity. Hypoglycemia may be life-threatening; this is a risk with any insulin administration, and may be of concern with automated systems.

An artificial pancreas device system (APDS) may provide better control of blood sugar with less episodes of hypoglycemia. A bihormonal APDS combines automated delivery of both insulin and glucagon. Pilot studies using investigational, bihormonal APDSs have demonstrated that delivery of small doses of glucagon in response to declining glucose levels reduces the frequency of hypoglycemia (Russell et al. 2012). Glucagon is a 29-aminoacid peptide hormone produced in the pancreas that stimulates glucagon receptors in the liver, resulting in the breakdown of stored glycogen into glucose. Glucagon opposes the effects of insulin in the liver and increases blood glucose levels.

The current commercial glucagon (recombinant glucagon [rGlucagon]) preparations are only available as lyophilized powder that must be reconstituted with a diluent before administration. The current Lilly formulation of rGlucagon also has poor solubility in CCI [redacted]

LY3185643 is a glucagon analog with improved solubility that is being developed for use in a bihormonal APDS. The current study is designed to assess the pharmacokinetic (PK) and pharmacodynamic (PD) profiles of LY3185643 and compare these to rGlucagon. Understanding the relative potency of these molecules, as well as any differences in time-action profiles, will be used to direct modifications of APDS control algorithms that have been developed based on rGlucagon pharmacology. These modified control algorithms will be tested in future studies using LY3185643 in an investigational, bihormonal APDS.

3.2. Background

LY3185643 is a selective glucagon analog with potency similar to rGlucagon based on nonclinical studies (in vitro and in vivo). LY3185643 administration resulted in dose-dependent increases in blood glucose levels in the CCI [redacted] in vivo following subcutaneous (SC) dosing in a similar time pattern as rGlucagon. Peak glucose level was attained at 30 minutes and the glucose level returned to baseline at approximately 60 minutes postadministration as observed with rGlucagon.

The PK of LY3185643 was evaluated in rats and dogs. Exposure increased with increase in dose. LY3185643 exposure in dogs was generally dose proportional. No consistent sex differences in systemic exposure were observed in rats or dogs. The time to maximum drug concentration (T_{max}) ranged from approximately CCI in dogs. The half-life ($t_{1/2}$) of LY3185643 following SC administration in dogs ranged from approximately CCI.

Development of LY3185643 was supported by repeat-dose nonclinical studies in both rats and dogs receiving daily SC injections for approximately 1 month. No adverse findings were reported following the 1-month nonclinical toxicology studies in both rats and dogs. Administration of LY3185643 CCI decreased blood pressure and increased heart rate (refer to the LY3185643 Investigator's brochure [IB]), consistent with the acute effects of native and/or recombinant glucagon peptides previously described in dogs (CDER 1998; Eistrup et al. 1993). No other LY3185643-related cardiovascular changes were noted. The reported changes in animal physiology were consistent with the pharmacologically-related changes associated with repeated administration of native and/or recombinant glucagon peptide described previously (Root 1954; CDER 1998; Eistrup et al. 1993).

Further, signs of injection-site inflammation were observed in dogs at all doses which were attributed of the frequency of repeat SC administration; however, there were no local tolerance issues observed in the nonclinical repeat-dose studies.

3.2.1. Clinical Experience

LY3185643 has been administered SC to 27 healthy subjects as single doses of 0.01 mg, 0.03 mg, 0.05 mg, 0.06 mg, 0.1 mg, 0.2 mg, 0.3 mg, 0.48 mg, and 0.72 mg (n = 3 subjects for each dose level). Preliminary PK data available for doses up to 0.2 mg show dose-dependent increases in LY3185643 exposures. Over the dose range of 0.01 to 0.2 mg, the median time to peak concentrations ranged between 0.5 to 1.25 hours, mean peak concentrations ranged between 0.13 to 4.25 ng/mL, and mean terminal $t_{1/2}$ ranged between 0.5 to 0.75 hours. LY3185643 concentrations decreased below the limit of quantification by 4 hours postdose. Dose-dependent increases in glycemic response were observed up to 0.2-mg dose level. Maximum mean blood glucose elevations of up to 180 mg/dL were observed at doses above 0.2 mg of LY3185643. Peak glucose concentrations were reached between 0.5 to 2 hours postdose, and glycemic effects lasted for up to 3 hours postdose. This transient glycemic effect at high LY3185643 concentrations is consistent with rGlucagon findings and likely reflects a feedback inhibition of hepatic glucose output in response to exogenous glucagon administration (Mighiu et al. 2013).

Adverse events (AEs) included nausea, vomiting, light headed, headache, dizziness, vasovagal episode, erythema at injection site, loose stools, feeling hot and cold, abrasion of right leg, increased appetite, right thigh ache, drowsiness, feels sleepy, feels faint, decreased alertness, and dehydration. The most common (67% of 36 reported events) AEs were nausea (n=10), vomiting (n=6), headache (n=3), dizziness (n=3), and light headedness (n=2). No serious AEs (SAEs) were noted.

3.3. Benefit/Risk Assessment

AEs seen after LY3185643 administration in a single-dose study, Study I7U-MC-GAHA (Study GAHA), were consistent with AEs reported with currently marketed glucagon such as nausea and vomiting. Tolerability issues in Study GAHA were mild-to-moderate and dose dependent. No clinically significant safety or tolerability concerns were identified in 27 subjects who received single doses of LY3185643 up to 0.72 mg in Study GAHA.

CCI [redacted] However, no arrhythmias were observed with LY3185643 administration, CCI [redacted]

Nevertheless, continuous telemetry will be performed during dosing periods in the current study.

Potential risks are considered to be monitorable and manageable at the planned dose range of 10 to 200 µg (0.01 to 0.2 mg) for LY3185643 in healthy subjects for this proposed study, which will be conducted in a Phase 1 clinical research unit (CRU). Because of the short half-life of LY3185643, any pharmacologic effects are anticipated to be short-lived.

No benefit to study subjects is anticipated. More information about the known and expected benefits, risks, SAEs and reasonably anticipated AEs of LY3185643 are to be found in the IB; known and expected benefits, and risks of rGlucagon may be found in the package insert for rGlucagon (Glucagon for injection prescribing information, 2012).

4. Objectives and Endpoints

Table APCA.4.1 shows the objectives and endpoints of the study.

Table APCA.4.1. Objectives and Endpoints

Primary Objective	**Endpoints**
The primary objective of this study is to evaluate the PK and PD profiles of LY3185643 and rGlucagon after single doses of study treatment	C_{max}, AUC, and terminal half-life of LY3185643 and rGlucagon C_{max} and AUC (change from predose) of blood glucose and C-peptide
Secondary Objective	**Endpoints**
To evaluate the safety and tolerability of LY3185643 after single doses of study treatment	Adverse events
Exploratory Objective	**Endpoints**
To evaluate immunogenicity of LY3185643 after multiple single doses of study treatment	Anti-LY3185643 antibodies

Abbreviations: AUC = area under the concentration versus time curve; C_{max} = maximum concentration; PD = pharmacodynamic; PK = pharmacokinetic; rGlucagon = recombinant glucagon.

5. Study Design

5.1. Overall Design

This study is a Phase 1, single-center, subject- and investigator-blind, randomized, 9-way, 3-period crossover study in healthy subjects to evaluate the PK and PD of LY3185643 and rGlucagon after SC administration.

Up to 37 healthy men and women may be enrolled to target approximately 18 subjects to complete the study. Each subject will receive 3 doses on each of 3 dosing days (total of 9 doses: 5 doses of LY3185643 and 4 doses of rGlucagon) administered SC. Doses will be administered in a 9-way complete crossover design in 3 periods, and subjects will be randomized to predefined treatment sequences. Completers are defined as subjects who have received all 9 doses.

Subjects will be required to attend the CRU on at least 5 occasions:

- 1 screening visit (may occur up to 28 days before randomization)
- 3 treatment periods
- 1 follow-up visit (at least 30 days after completing the last dose of study treatment)

Each study period will consist of 3 days (Days -1 to 2). Subjects should return for the next period with a minimum of 3 days between treatment periods.

On Day -1, subjects will be admitted to the CRU. Subjects will be provided a carbohydrate-rich meal in the evening at approximately 10:00 PM and will remain fasted until completion of the study procedures on Day 1.

On Day 1, study drug will be administered SC as follows:

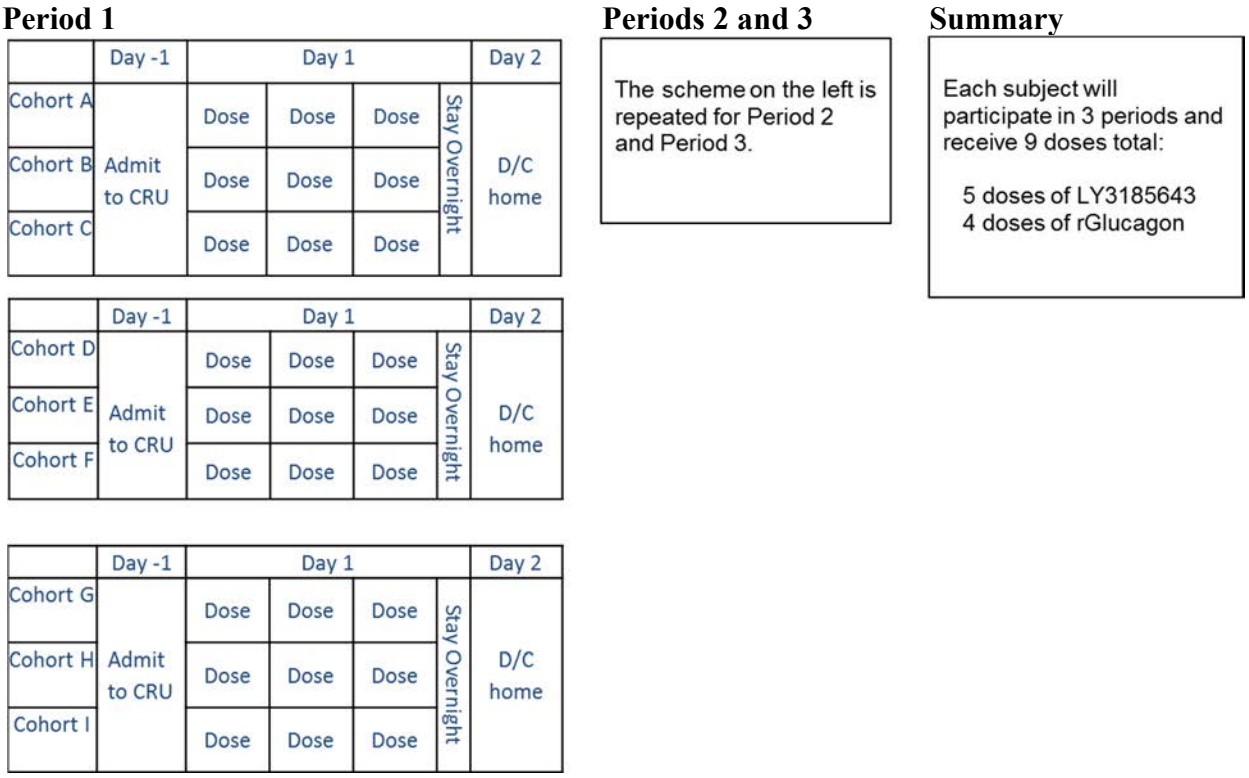
- Dose 1: morning of Day 1
- Dose 2: approximately 4 hours after administration of the first dose
- Dose 3: approximately 4 hours after administration of the second dose

Blood samples will be collected for the determination of concentrations of LY3185643, glucagon, glucose, C-peptide, and for immunogenicity as shown in the Schedule of Activities (Section 2).

Safety data will include clinical examinations, vital signs, electrocardiograms (ECGs; including telemetry), clinical laboratory tests, and a record of AEs.

All study procedures will end approximately 4 hours after the third dose. Subjects will be provided dinner and stay overnight in the CRU for observation. Subjects will be discharged the following day (Day 2) after completion of all assessments as shown in the Schedule of Activities (Section 2).

Figure APCA.5.1 illustrates the study design.



Abbreviations: CRU = clinical research unit; D/C = discharge.

Figure APCA.5.1. Study design.

Approximately 18 subjects are expected to complete the study. Each subject will be randomized to 1 of 9 treatment sequences comprising single SC doses of LY3185643 or rGlucagon (Table APCA.5.1).

Subjects who discontinue may be replaced to target at least 18 completers; the replacement subject will be assigned the treatment sequence of the discontinued subject and complete all 3 treatment periods (total of 9 doses).

Table APCA.5.1. Treatment Sequences

Cohort[a]	Treatment Period 1			Treatment Period 2			Treatment Period 3		
A	Dose 1	Dose 9	Dose 7	Dose 6	Dose 4	Dose 8	Dose 5	Dose 3	Dose 2
B	Dose 2	Dose 7	Dose 8	Dose 4	Dose 5	Dose 9	Dose 6	Dose 1	Dose 3
C	Dose 3	Dose 8	Dose 9	Dose 5	Dose 6	Dose 7	Dose 4	Dose 2	Dose 1
D	Dose 4	Dose 6	Dose 2	Dose 3	Dose 9	Dose 1	Dose 8	Dose 7	Dose 5
E	Dose 5	Dose 4	Dose 3	Dose 1	Dose 7	Dose 2	Dose 9	Dose 8	Dose 6
F	Dose 6	Dose 5	Dose 1	Dose 2	Dose 8	Dose 3	Dose 7	Dose 9	Dose 4
G	Dose 7	Dose 1	Dose 4	Dose 9	Dose 2	Dose 6	Dose 3	Dose 5	Dose 8
H	Dose 8	Dose 2	Dose 5	Dose 7	Dose 3	Dose 4	Dose 1	Dose 6	Dose 9
I	Dose 9	Dose 3	Dose 6	Dose 8	Dose 1	Dose 5	Dose 2	Dose 4	Dose 7

Note: This is an example table; subjects will be assigned a treatment sequence according to the actual treatment schedule provided to the site.

Dose assignment: Dose 1 = 10 µg LY3185643; Dose 2 = 25 µg LY3185643; Dose 3 = 50 µg LY3185643; Dose 4 = 100 µg LY3185643; Dose 5 = 200 µg LY3185643; Dose 6 = 10 µg rGlucagon; Dose 7 = 25 µg rGlucagon; Dose 8 = 50 µg rGlucagon; Dose 9 = 200 µg rGlucagon.

[a] In each cohort, LY3185643 and rGlucagon will be administered.

The Schedule of Activities (Section 2) details the procedures and tests occurring at specific times during the study.

5.2. Number of Participants

Up to 37 subjects may be enrolled so that approximately 18 subjects complete the study.

Subjects who receive all 9 doses of study treatment will be considered as completing the study.

5.3. End of Study Definition

End of the study is the date of the last visit or last scheduled procedure shown in the Schedule of Activities (Section 2) for the last subject.

5.4. Scientific Rationale for Study Design

A population of healthy subjects is selected based upon the likelihood of less physiologic variability in the absence of disease states that may affect multiple organ systems and the absence of other confounding factors such as concomitant medications. Furthermore, studies with native glucagon suggest that the PD responses being assessed in this study are similar between healthy subjects and subjects with diabetes who form the eventual patient population for this treatment (Nielsen et al. 1997; Matsuda et al. 2002).

Three doses of LY3185643 and/or rGlucagon will be administered on Day 1 of each study period; each dose will be separated by at least 4 hours. A 4-hour dosing cycle is considered appropriate because previous clinical data support that a 4-hour observation period is a sufficient amount of time to obtain both the PK and PD profiles. The study drug concentrations will be below the quantification limits by 4 hours and the glycemic levels will return back to baseline within 4 hours postdose. A previous study used a similar design with up to 4 doses given over the course of a single day and showed that this dosing schedule was well tolerated and that the PD response was not attenuated or refractory to repeat dosing (Blauw et al. 2016).

The use of a crossover design allows each subject to serve as his or her own control, thereby reducing variability.

The currently marketed glucagon product, rGlucagon, will be administered to provide a comparison of its PK/PD profile to the PK/PD profile of LY3185643.

5.5. Justification for Dose

The approved rGlucagon doses for hypoglycemia recovery are 0.5 and 1 mg administered parenterally. Small bolus doses of rGlucagon ranging between 5 to 50 µg are administered in the bihormonal APDS (Russell et al. 2014). In vitro binding affinity data and clinical glucose response indicate that the potency of rGlucagon and LY3185643 are similar. Thus, a dose range of 10 to 200 µg (0.01 mg to 0.2 mg) for both LY3185643 and rGlucagon was selected for evaluation of PK/PD to enable the dosing of LY3185643 in APDS. This dose range is supported by preclinical and clinical safety/tolerability data.

In a single dose-escalation safety/tolerability study (Study I7U-MC-GAHA [Study GAHA]), LY3185643 was safely administered to healthy subjects within a dose range of 0.01 to 0.72 mg. The proposed doses of LY3185643 in the current study are within that range and the proposed top dose of 0.2 mg is approximately 4-fold lower than the 0.72-mg dose administered in Study GAHA.

6. Study Population

Eligibility of subjects for study enrollment will be based on the results of screening medical history, physical examination, vital signs, clinical laboratory tests, and ECG.

The nature of any conditions present at the time of the physical examination and any preexisting conditions will be documented.

Screening may occur up to 28 days before enrollment. Subjects who are not enrolled within 28 days of screening may be subjected to an additional medical assessment and/or clinical measurements to confirm their eligibility.

Prospective approval of protocol deviations to recruitment and enrollment criteria, also known as protocol waivers or exemptions, are not permitted.

6.1. Inclusion Criteria

Subjects are eligible for inclusion in the study only if they meet all of the following criteria at screening and/or enrollment:

[1] are overtly healthy males or females, as determined by medical history and physical examination

[1a] male subjects:

Male subjects with female partners of childbearing potential will be required to use a condom in conjunction with a spermicidal gel, foam, cream or suppository. In addition, the female partner will be requested to use an additional effective form of contraception, which can be any of the following:

- female condom with spermicide
- diaphragm with spermicide
- cervical sponge
- cervical cap with spermicide
- combined oral contraceptive pill and mini-pill
- NuvaRing
- implantable contraceptives
- injectable contraceptives (such as Depo-Provera®)
- intrauterine device (such as Mirena® and ParaGard®)
- total abstinence

Men who have had a vasectomy with appropriate post-vasectomy documentation of the absence of sperm in the ejaculate are not required to use contraception. Male subjects

with a female partner meeting the definition of a woman not of childbearing potential will not be required to use contraception.

[1b] female subjects:

women not of childbearing potential may participate without using contraception, and include those who are:

a) infertile due to surgical sterilization (hysterectomy, bilateral oophorectomy, or tubal ligation), congenital anomaly such as mullerian agenesis; or
b) postmenopausal – postmenopausal is defined as women with an intact uterus who have not taken hormones or oral contraceptives within 1 year, who have had either cessation of menses for at least 1 year, or 6 to 12 months of spontaneous amenorrhea with follicle-stimulating hormone consistent with menopause (>40 mIU/mL)

[2] are between 21 and 60 years of age at the time of screening, inclusive

[3] have a body mass index of 18.0 to 30.0 kg/m^2, inclusive, at screening

[4] have clinical laboratory test results within normal reference range for the population or investigative site, or results with acceptable deviations that are judged to be not clinically significant by the investigator

[5] have venous access sufficient to allow for blood sampling as per the protocol

[6] are reliable and willing to make themselves available for the duration of the study and are willing to follow study procedures

[7] are able and willing to give signed informed consent

6.2. Exclusion Criteria

Subjects will be excluded from study enrollment if they meet any of the following criteria at screening and/or enrollment:

[8] are investigative site personnel directly affiliated with this study and their immediate families. Immediate family is defined as a spouse, biological or legal guardian, child, or sibling

[9] are Lilly employees

[10] are currently enrolled in a clinical trial involving an investigational product or any other type of medical research judged not to be scientifically or medically compatible with this study

[11] have participated, within the last 30 days, in a clinical trial involving an investigational product. If the previous investigational product has a long half-life, 3 months or 5 half-lives (whichever is longer) should have passed

[12] have previously completed or withdrawn from this study or any other study investigating LY3185643, and have previously received the investigational product

[13] have known allergies to LY3185643 or rGlucagon, related compounds, or any components of the formulation, or history of significant atopy

[14] have a history or ECG evidence of heart block, or any abnormality in the 12-lead ECG that, in the opinion of the investigator, increases the risks associated with participating in the study

[15] have an abnormal blood pressure as determined by the investigator

[16] have a history of recurring symptomatic postural hypotension irrespective of the decrease in blood pressure, or asymptomatic postural hypotension at screening as defined as a decrease in systolic blood pressure ≥20 mm Hg within 3 minutes when changing from supine to standing position

[17] have a history of vasovagal response such as fainting

[18] have a significant history of or current cardiovascular, respiratory, hepatic, renal, gastrointestinal, endocrine, hematological, or neurological disorders capable of significantly altering the absorption, metabolism, or elimination of drugs; of constituting a risk when taking the investigational product; or of interfering with the interpretation of data

[19] have a history of/current insulinoma and/or pheochromocytoma

[20] have known or ongoing psychiatric disorders

[21] have a history of drug abuse

[22] show evidence of human immunodeficiency virus (HIV) infection and/or positive human HIV antibodies

[23] show evidence of hepatitis C and/or positive hepatitis C antibody

[24] show evidence of hepatitis B and/or positive hepatitis B surface antigen

[25] intend to use over-the-counter or prescription medication within 7 and 14 days, respectively, before dosing (apart from vitamin/mineral supplements, occasional paracetamol, hormone replacement therapy, or thyroid-replacement therapy). If this situation arises, inclusion of an otherwise suitable subject may be at the discretion of the investigator and sponsor

[26] have had systemic exposure to glucocorticoids within 3 months before entry into the study (including oral or intra-articular glucocorticoids, or potent topical steroids applied to >20% of body surface area)

[27] have any chronic diseases or illness that interfere with glucose metabolism

[28] have known presence of hereditary problems of glycogen storage disease, and/or galactose

[29] have had lymphoma, leukemia, or any malignancy within the past 5 years except for basal cell or squamous epithelial carcinomas of the skin that have been resected with no evidence of metastatic disease for 3 years

[30] consume more than 10 cigarettes per day or the equivalent, or are unable or unwilling to refrain from nicotine use while in residence at the CRU

[31] have donated blood of 450 mL or more in the last 3 months or provided any blood donation within the last month before screening

[32] have an average weekly alcohol intake that exceeds 14 units per week or are unwilling to stop alcohol consumption from 24 hours before dosing until the completion of each trial period (1 unit = 12 oz or 360 mL of beer; 5 oz or 150 mL of wine; 1.5 oz or 45 mL of distilled spirits)

[33] in the opinion of the investigator or sponsor, are unsuitable for inclusion in the study

6.3. Lifestyle and/or Dietary Requirements

Throughout the study, subjects may undergo medical assessments and review of compliance with requirements before continuing in the study.

6.3.1. Meals and Dietary Restrictions

At approximately 10:00 PM on Day-1 of each study period, subjects will be provided with a mixed meal containing at least 100 g of carbohydrate and at least 600 calories total. Subjects should be encouraged to consume the entire meal.

For all treatment periods, subjects are expected to fast for approximately 8 hours before the first dose until 4 hours after the last dose. Water can be consumed freely during this period.

When at home, subjects will be encouraged to follow normal, unrestricted diet that contains at least 150 g of carbohydrate daily (in the form of simple sugars, starch, or complex carbohydrates) and refrain from strenuous exercise for at least 3 days before admission to the CRU.

6.3.2. Caffeine, Alcohol, and Tobacco

No alcohol will be allowed at least 24 hours before each dose and throughout the duration of each CRU visit.

No tobacco smoking or use of smokeless tobacco will be permitted while subjects are resident in the CRU.

Subjects should refrain from caffeine-containing food/beverages (for example, cola, chocolate, Milo, tea, and coffee) for at least 12 hours before each dose and throughout the duration of each CRU visit.

6.3.3. Activity

Subjects will be encouraged to maintain their regular exercise; however, they should not undertake vigorous or prolonged exercise at least 3 days before admission to the CRU. After dosing, subjects should remain recumbent or sitting in the CRU.

6.4. Screen Failures

Individuals who do not meet the criteria for participation in this study (screen failure) may not be rescreened.

7. Treatment

7.1. Treatment Administered

LY3185643 and rGlucagon will be supplied by Lilly or its representative in accordance with current good manufacturing practices and will be supplied with lot numbers.

Each vial of investigational product will contain 1 mg/mL of LY3185643 or rGlucagon (Lilly USA, LLC). LY3185643 or rGlucagon may be diluted for SC administration. Additional information can be found in the pharmacy instructions including detailed instructions for the preparation of the investigational products. rGlucagon will be reconstituted from the lyophilized commercial product on the day of use and will be administered within the time limits defined in the pharmacy instructions.

Table APCA.7.1 shows the treatment regimens.

Table APCA.7.1. Investigational Product Regimens

Product:	**LY3185643**	**rGlucagon (recombinant glucagon)**
Dose range:	10 to 200 µg	10 to 200 µg
Route of administration:	subcutaneous injection	subcutaneous injection

The investigator or designee is responsible for:

- explaining the correct use of the investigational product to the site personnel
- verifying that instructions are followed properly
- maintaining accurate records of investigational product dispensing and collection
- returning all unused medication to Lilly or its designee at the end of the study

Note: In some cases, sites may destroy the material if, during the investigative site selection, the evaluator has verified and documented that the site has appropriate facilities and written procedures to dispose of clinical trial materials.

Subjects will be instructed to contact the investigator as soon as possible if he or she has a complaint or problem with the investigational product so that the situation can be assessed.

Study treatment will be drawn up into a 0.3-mL insulin syringe with a 6-mm/31 G needle to ensure all injections are delivered to a consistent depth target into the SC space. Each syringe will be labelled appropriately and dispensed to qualified site personnel for administration.

The doses will be administered SC in the CRU by qualified site personnel (as designated by the investigator). Injection sites selected should be approximately 5 cm from the umbilicus and the treatment administered SC with the needle applied at approximately 90 degrees without pinching the skinfold. Each injection in a given study period will be administered into a different quadrant of the anterior abdominal wall (that is, left upper quadrant, left lower quadrant, and right upper quadrant) and the site of injection for each dose will be recorded. As far as it is

practical, the same staff will administer the injections. Please refer to the pharmacy binder for detailed instructions.

7.1.1. Packaging and Labeling

LY3185643 will be packaged by Lilly or its representative in a 2-mL vial.

rGlucagon will be packaged by Lilly or its representative in a 1-mL vial.

These clinical trial materials will be labeled in accordance with local regulatory requirements.

7.2. Method of Treatment Assignment

Randomization tables with treatment codes for allocation of LY3185643 or rGlucagon will be prepared by the statistician for the study and provided to the site pharmacists involved in dose preparation. The pharmacist will provide treatment to the blinded site staff who will administer the study drugs.

7.2.1. Selection and Timing of Doses

The doses will be administered in the CRU during the treatment period on Day 1 only. The actual time of all dose administrations will be recorded in the subject's case report form (CRF).

7.3. Blinding

This study is subject- and investigator-blind. Blinding will be maintained throughout the conduct of the trial until all data are cleaned to an acceptable level of quality and locked.

Emergency codes will be available to the investigator. A code, which reveals the treatment group for a specific study subject, may be opened during the study only if the subject's well-being requires knowledge of the subject's treatment assignment.

If a subject's study treatment assignment is unblinded, the subject must be discontinued from the study unless the investigator obtains specific approval from a Lilly clinical pharmacologist or clinical research physician (CRP) for the study participant to continue in the study. During the study, emergency unblinding should occur only by accessing the study subject's emergency code.

In case of an emergency, the investigator has the sole responsibility for determining if unblinding of a subject's treatment assignment is warranted for medical management of the event. Subject's safety must always be the first consideration in making such a determination. If the investigator decides that unblinding is warranted, the investigator should make every effort to contact the Lilly clinical pharmacologist or CRP before unblinding a study subject's treatment assignment unless this could delay emergency treatment of the subject. If a study subject's treatment assignment is unblinded, Lilly must be notified immediately.

Upon completion of the study, all codes must be returned to Lilly or its designee.

7.4. Dose Modification

There will be no dose modifications of LY3185643 and rGlucagon.

7.5. Preparation/Handling/Storage/Accountability

All clinical trial material provided to the investigator will be stored in a secure place and allocated and dispensed by appropriately trained persons. The allocation and dispensing of the investigational products will be fully documented and verified by a second person. Detailed records of the amounts of the investigational product received, dispensed, and remaining at the end of the study will be maintained.

Only participants enrolled in the study may receive investigational product and only authorized site staff may supply or administer study treatment. All study treatments should be stored in an environmentally controlled and monitored (manual or automated) area in accordance with the labeled storage conditions with access limited to the investigator and authorized site staff.

The investigator is responsible for study treatment accountability, reconciliation, and record maintenance (such as receipt, reconciliation and final disposition records). The investigator site will be permitted to destroy the investigational material after written approval is obtained from the sponsor.

7.6. Treatment Compliance

Every attempt will be made to select subjects who have the ability to understand and comply with instructions. Noncompliant subjects may be discontinued from the study.

The investigational product will be administered at the CRU and documentation of treatment administration will occur at the site.

The specifications in this protocol for the timings of safety, PK, and PD sampling are given as targets to be achieved within reasonable limits. Modifications may be made to the time points based upon the safety and PK information obtained. The scheduled time points may be subject to minor alterations; however, the actual time must be correctly recorded in the CRF. Failure to obtain samples due to clinical issues, such as problems with venous access, technical difficulty with obtaining samples, or subject no show for planned procedural visits will not be considered a protocol deviation but the site will still be required to notify the sponsor in writing.

Any major modifications that might affect the conduct of the study, subject safety, and/or data integrity will be detailed in a protocol amendment.

7.7. Concomitant Therapy

Subjects should not use over-the-counter or prescription medication within 7 and 14 days, respectively, before each dosing (apart from vitamin/mineral supplements, occasional paracetamol, hormone replacement therapy, or thyroid-replacement therapy).

If the need for concomitant medication arises, inclusion or continuation of the subject may be at the discretion of the investigator and, if possible, after consultation with a Lilly clinical pharmacologist or CRP. Any additional medication used during the course of the study must be documented.

7.8. Treatment after the End of the Study

LY3185643 and rGlucagon will not be made available to subjects after conclusion of the study.

8. Discontinuation Criteria

8.1. Discontinuation from Study Treatment

Discontinuation of the investigational product for abnormal liver tests **should be considered** by the investigator when a subject meets one of the following conditions after consultation with the Lilly designated medical monitor:

- alanine aminotransferase (ALT) or aspartate aminotransferase (AST) >5X upper limit of normal (ULN)
- ALT or AST >3X ULN along with one of the following criteria
    - sustained for more than 2 weeks or
    - total bilirubin level >2X ULN or
    - prothrombin time >1.5X ULN or
    - appearance of fatigue, nausea, vomiting, right upper-quadrant pain or tenderness, fever, rash, and/or eosinophilia (>5%)
- alkaline phosphatase (ALP) >3X ULN
- ALP >2.5X ULN and total bilirubin level >2X ULN
- ALP >2.5X ULN with the appearance of fatigue, nausea, vomiting, right quadrant pain or tenderness, fever, rash, and/or eosinophilia (>5%)

Subjects who discontinue the investigational product early will have procedures performed as shown in the Schedule of Activities (Section 2).

8.1.1. Discontinuation of Inadvertently Enrolled Subjects

If the sponsor or investigator identifies a subject who did not meet enrollment criteria and was inadvertently enrolled, a discussion must occur between the Lilly clinical pharmacologist/CRP and the investigator to determine if the subject may continue in the study. If both agree it is medically appropriate to continue, the investigator must obtain documented approval from the Lilly clinical pharmacologist/CRP to allow the inadvertently enrolled subject to continue in the study with or without continued treatment with investigational product.

8.2. Discontinuation from the Study

Subjects will be discontinued in the following circumstances:

- enrollment in any other clinical trial involving an investigational product or enrollment in any other type of medical research judged not to be scientifically or medically compatible with this study
- participation in the study needs to be stopped for medical, safety, regulatory, or other reasons consistent with applicable laws, regulations, and good clinical practice (GCP

- investigator decision
    - the investigator decides that the subject should be discontinued from the study
- subject decision
    - the subject, or designee (for example, parents or legal guardian) requests to be withdrawn from the study

Subjects who discontinue the study early will have end-of-study procedures performed as shown in the Schedule of Activities (Section 2).

8.3. Subjects Lost to Follow-Up

A subject will be considered lost to follow-up if he or she repeatedly fails to return for scheduled visits and is unable to be contacted by the study site. Site personnel are expected to make diligent attempts to contact subjects who fail to return for a scheduled visit or were otherwise unable to be followed up by the site.

9. Study Assessments and Procedures

Section 2 lists the Schedule of Activities, detailing the study procedures and their timing (including tolerance limits for timing).

Appendix 2 lists the laboratory tests that will be performed for this study.

Appendix 5 provides a summary of the maximum number and volume of invasive samples for all sampling during the study.

Unless otherwise stated in subsections below, all samples collected for specified laboratory tests will be destroyed within 60 days of receipt of confirmed test results. Certain samples may be retained for a longer period, if necessary, to comply with applicable laws, regulations, or laboratory certification standards.

Investigators must document their review of each laboratory safety report.

9.1. Efficacy Assessments

This section is not applicable for this study.

9.2. Adverse Events

Investigators are responsible for monitoring the safety of subjects who have entered this study and for alerting Lilly or its designee to any event that seems unusual, even if this event may be considered an unanticipated benefit to the subject.

The investigator is responsible for the appropriate medical care of subjects during the study.

The investigator remains responsible for following, through an appropriate health care option, AEs that are serious or otherwise medically important, considered related to the investigational product or the study, or that caused the subject to discontinue the investigational product before completing the study. The subject should be followed until the event resolves, stabilizes with appropriate diagnostic evaluation, or is reasonably explained. The frequency of follow-up evaluations of the AE is left to the discretion of the investigator.

After the informed consent form (ICF) is signed, study site personnel will record, via CRF, the occurrence and nature of each subject's preexisting conditions, including clinically significant signs and symptoms of the disease under treatment in the study. Additionally, site personnel will record any change in the condition(s) and the occurrence and nature of any AEs.

The investigator will interpret and document whether or not an AE has a reasonable possibility of being related to study treatment or a study procedure, taking into account the disease, concomitant treatment, or pathologies.

A "reasonable possibility" means that there is a cause and effect relationship between the investigational product and/or study procedure and the AE.

Planned surgeries should not be reported as AEs unless the underlying medical condition has worsened during the course of the study.

If a subject's investigational product is discontinued as a result of an AE, study site personnel must report this to Lilly or its designee via CRF.

9.2.1. Serious Adverse Events

An SAE is any AE from this study that results in one of the following:

- death
- initial or prolonged inpatient hospitalization
- a life-threatening experience (that is, immediate risk of dying)
- persistent or significant disability/incapacity
- congenital anomaly/birth defect
- events considered significant by the investigator based upon appropriate medical judgment

Study site personnel must alert Lilly, or its designee, of any SAE within 24 hours of investigator awareness of the event via a sponsor-approved method. If alerts are issued via telephone, they are to be immediately followed with official notification on study-specific SAE forms. This 24-hour notification requirement refers to the initial SAE information and all follow-up SAE information.

Although all AEs are recorded in the CRF after signing informed consent, SAE reporting begins after the subject has signed informed consent and has received investigational product. However, if an SAE occurs after signing informed consent, but before receiving investigational product, AND is considered reasonably possibly related to a study procedure then it MUST be reported.

Investigators are not obligated to actively seek AEs or SAEs in subjects once they have discontinued from and/or completed the study (the subject summary CRF has been completed). However, if the investigator learns of any SAE, including a death, at any time after a subject has been discharged from the study and he/she considers the event reasonably possibly related to the study treatment or study participation, the investigator must promptly notify Lilly.

9.2.1.1. Suspected Unexpected Serious Adverse Reactions

Suspected unexpected serious adverse reactions (SUSARs) are serious events that are not listed in the IB and that the investigator identifies as related to investigational product or procedure. United States 21 CFR 312.32 and European Union Clinical Trial Directive 2001/20/EC and the associated detailed guidances or national regulatory requirements in participating countries require the reporting of SUSARs. Lilly has procedures that will be followed for the recording and expedited reporting of SUSARs that are consistent with global regulations and the associated detailed guidances.

9.2.2. Complaint Handling

Lilly collects product complaints on investigational products and drug delivery systems used in clinical trials in order to ensure the safety of study participants, monitor quality, and to facilitate process and product improvements.

Subjects should be instructed to contact the investigator as soon as possible if he or she has a complaint or problem with the investigational product so that the situation can be assessed.

9.3. Treatment of Overdose

For the purposes of this study, an overdose of LY3185643 is considered any doses higher than the doses assigned through randomization. An overdose of rGlucagon is considered as any dose higher than 1 mg (the upper limit of the approved dosage in the prescribing information). Refer to the IB for LY3185643 and the prescribing information for rGlucagon (2012) for management of inadvertent treatment overdose.

9.4. Safety

9.4.1. Laboratory Tests

For each subject, laboratory tests detailed in Appendix 2 should be conducted according to the Schedule of Activities (Section 2).

9.4.2. Physical Examination

Physical examinations and routine medical assessments will be conducted as specified in the Schedule of Activities (Section 2).

9.4.3. Vital Signs

For each subject, vital signs measurements should be conducted according to the Schedule of Activities (Section 2).

Blood pressure and pulse rate should be measured after at least 5 minutes supine.

If orthostatic measurements are required, subjects should be supine for at least 5 minutes and stand for at least 3 minutes.

If the subject feels unable to stand, supine vital signs only will be recorded.

Additional vital signs may be measured during each study period if warranted at the discretion of the investigator.

9.4.4. Electrocardiograms

For each subject, ECGs should be collected according to the Schedule of Activities (Section 2).

Any clinically significant findings from ECGs that result in a diagnosis and that occur after the subject receives the first dose of the investigational product should be reported to Lilly, or its designee, as an AE via CRF.

9.4.4.1. Continuous 12-Lead ECG Monitoring

Continuous 12-lead ECG monitoring does not allow bedside printing or viewing of ECGs extracted from the signal recorded. Thus, for subject safety, ECGs must be recorded by a local, static 12-lead ECG device at the times specified in the Schedule of Activities (Section 2). These safety ECGs will not be transmitted to a central ECG laboratory. Safety ECGs must be interpreted by the investigator or qualified designee soon after the time of collection. This review will be documented and provides information for immediate subject management. If the ECG shows a clinically significant quantitative or qualitative change from baseline, the investigator or qualified designee will assess the subject for symptoms (for example, palpitations, near syncope, syncope) and determine if the subject can continue in the study. The investigator or qualified designee is responsible for subject management. If the investigator determines that the changes identified in the ECG are clinically relevant, an applicable AE should be recorded on the subject CRF.

9.4.4.1.1. Recordings for Data Analysis

Continuous 12-lead recordings will be performed according to the Schedule of Activities (Section 2). Patients must be in a quiet atmosphere without significant external stimulation (for example, TV, internet, etc.), remain in a supine position for at least 5 to 10 minutes before the specified ECG collection times, and remain supine but awake during ECG collection and for at least 10 minutes afterward. Subjects should be encouraged to remain still, if possible, during this time.

Digital recordings of all periods of continuous ECG recordings in individual subjects will be transferred to a central ECG laboratory designated by Lilly. The ECG laboratory will perform quality control checks for the time points of interest (for example, acquisition quality for ability to measure/interpret, demographics, and study details) and extract 15 unique 10-second ECGs at the times listed in the Schedule of Activities (Section 2).

The ECG laboratory will then store the recording as well as the extracted 10-second ECGs, and a cardiologist at the central ECG laboratory will conduct a full overread (including the measurement of all intervals) on one of the replicates that was extracted. For each set of replicates, the cardiologist will determine the RR and QT intervals and heart rate on the ECGs that were not fully overread. No reports will be issued from the central ECG laboratory back to the sites for any ECGs.

All data from the overreads will be placed in a Lilly or clinical research organization (CRO) database for analytical and study report purposes.

It is recognized that the ECG interpretations by the investigator (or qualified designee) and the cardiologist at the central ECG laboratory may be different. Interpretation of the ECG by the investigator (or qualified designee) will be used for study entry and immediate subject management. Interpretations of the ECG at the central laboratory will be used for data analysis and report-writing purposes.

9.4.4.2. Telemetry

The ECG waveform and cardiac rhythm will be monitored continuously using an ECG telemetry monitor during the inpatient stay at the CRU, from the night of Day -1 to 4 hours after the last dose on Day 1, for all 3 periods. Telemetry data from Day -1 up to approximately 1 hour before dosing will be reviewed by the investigator before administration of the first dose on Day 1. Subjects will not be dosed if any exclusion criteria are observed (for example, Exclusion Criterion [14]). Any other observations, such as changes in the ECG waveform and rhythm, should be evaluated by the investigator and the decision to dose will be at the investigator's discretion. Any confirmed abnormalities/events assessed to be clinically significant by the investigator during monitoring should be recorded in the medical record and reported as a preexisting condition or AE on the AE CRF page.

9.4.5. Safety Monitoring

The Lilly clinical pharmacologist or CRP will monitor safety data throughout the course of the study.

Lilly will review SAEs within time frames mandated by company procedures. The Lilly clinical pharmacologist or CRP will consult with the functionally independent Global Patient Safety therapeutic area physician or clinical research scientist when appropriate and periodically review:

- trends in safety data
- laboratory analytes
- AEs

If a study subject experiences elevated ALT ≥3X ULN, ALP ≥2X ULN, or elevated total bilirubin ≥2X ULN, clinical and laboratory monitoring should be initiated by the investigator. Details for hepatic monitoring depend upon the severity and persistence of observed laboratory test abnormalities. To ensure subject safety and compliance with regulatory guidance, the investigator is to consult with the Lilly designated CRP regarding collection of specific recommended clinical information and follow-up laboratory tests (Appendix 4).

9.4.6. Immunogenicity Assessments

At the visits and times specified in the Schedule of Activities (Section 2), blood samples for immunogenicity testing will be collected to determine antibody production against the investigational product. Justification for the lack of paired PK samples is based on the short $t_{1/2}$ of LY3185643 (less than 1 hour) and the long delay between the dose administration and sampling times.

Additional samples may be collected if there is a possibility that an AE is immunologically mediated. Immunogenicity will be assessed by a validated assay designed to detect anti-drug antibodies in the presence of the investigational product. Antibodies may be further characterized and/or evaluated for cross-reactive binding to native glucagon and/or their ability to neutralize the activity of the investigational product.

Samples will be retained for a maximum of 15 years after the last subject visit, or for a shorter period if local regulations and Ethical Review Boards (ERBs) allow, at a facility selected by the sponsor. The duration allows the sponsor to respond to future regulatory requests related to the investigational product. Any samples remaining after 15 years will be destroyed.

9.5. Pharmacokinetics

At the visits and times specified in the Schedule of Activities (Section 2), venous blood samples of approximately 4 mL each will be collected to determine the plasma concentrations of LY3185643 or glucagon. Instructions for the collection and handling of blood samples will be provided by the sponsor. The actual date and time (24-hour clock time) of each sampling will be recorded.

9.5.1. Bioanalysis

Samples will be analyzed at a laboratory approved by the sponsor and stored at a facility designated by the sponsor.

Concentrations of LY3185643 will be assayed CCI [redacted]

Concentrations of glucagon will be assayed using a validated LC/MS method.

Bioanalytical samples collected to measure investigational product concentrations will be retained for a maximum of 1 year following last subject visit for the study.

9.6. Pharmacodynamics

At the times specified in the Schedule of Activities (Section 2), venous blood samples will be collected and used to determine the PD effects of LY3185643 or rGlucagon. Blood will be collected to evaluate blood glucose and C-peptide (both analyzed at a central laboratory designated by the sponsor).

The sample(s) will be stored for up to a maximum of 1 year after the last subject visit for the study at a facility selected by the sponsor.

9.6.1. Glucodynamic Samples

Blood samples (approximately 0.2 mL each) will be obtained for the measurement of glucose at the times specified in the Schedule of Activities (Section 2) using a validated method (YSI glucose analyzer) that will be readily available at the investigative site during the inpatient periods in order to provide real-time glucose measurement. Repeat samples for counter-checking of apparent spurious results may be taken where indicated. Samples will be disposed of upon confirmation of results.

9.7. Genetics

This section is not applicable for this study.

9.8. Biomarkers

This section is not applicable for this study.

9.9. Health Economics

This section is not applicable for this study.

10. Statistical Considerations and Data Analysis

10.1. Sample Size Determination

The sample size is customary for Phase 1 studies evaluating safety, PK, and/or PD parameters and is considered sufficient to evaluate the primary objective of this study.

Subjects who discontinue the study may be replaced so that at least 18 subjects complete the study. Replacement subjects will be assigned the treatment sequence of the subject that discontinued the study.

10.2. Populations for Analyses

10.2.1. Study Participant Disposition

A detailed description of subject disposition will be provided at the end of the study.

10.2.2. Study Participant Characteristics

The subject's age, sex, weight, body mass index, height, race/subrace, or other demographic characteristics will be recorded and summarized.

10.3. Statistical Analyses

Statistical analysis of this study will be the responsibility of Eli Lilly and Company.

PK and PD analyses will be conducted on the full analysis set. This set includes all data from all randomized subjects receiving at least 1 dose of the investigational product and with a baseline and at least 1 postbaseline measurement for each dose according to the treatment the subjects actually received. Safety analyses will be conducted for all enrolled subjects whether or not they completed all protocol requirements.

Additional exploratory analyses of the data will be conducted as deemed appropriate. Study results may be pooled with the results of other studies for population analysis purposes.

10.3.1. Safety Analyses

10.3.1.1. Clinical Evaluation of Safety

All investigational product and protocol procedure AEs will be listed, and if the frequency of events allows, safety data will be summarized using descriptive methodology.

The incidence of symptoms for each treatment will be presented by severity and by association with investigational product as perceived by the investigator. Symptoms reported to occur before first dose will be distinguished from those reported as new or increased in severity during the study. Each symptom will be classified by the most suitable term from the medical regulatory dictionary.

The number of investigational product-related SAEs will be reported.

10.3.1.2. Statistical Evaluation of Safety

Safety parameters that will be assessed include safety laboratory parameters, vital signs, and ECG parameters. The ECG parameters that will be assessed include the ECG heart rate and the following intervals: QT, RR, PR, and QT corrected for heart rate (QTc) using Fridericia's formula (QTcF). The safety parameters will be listed and summarized using standard descriptive statistics. Any additional analyses will be detailed in the statistical analysis plan.

10.3.2. Pharmacokinetic Analyses

10.3.2.1. Pharmacokinetic Parameter Estimation

PK parameter estimates for LY3185643 and rGlucagon will be calculated by standard noncompartmental methods of analysis.

The primary parameters for analysis will be maximum drug concentration (C_{max}) and area under the concentration versus time curve (AUC). Other noncompartmental parameters, such as $t_{1/2}$, apparent clearance, and apparent volume of distribution, may be reported. Population PK and exposure-response analyses may be performed using PK and glucose data.

10.3.2.2. Pharmacokinetic Statistical Inference

PK parameters for LY3185643 and rGlucagon will be summarized for each dose group. PK parameter estimates will be evaluated to delineate effects of dose proportionality.

10.3.3. Pharmacodynamic Analyses

The PD time-action and response of LY3185643 and rGlucagon will be compared to establish the relative potency between LY3185643 and rGlucagon.

10.3.3.1. Pharmacodynamic Parameter Estimation

The primary PD response variables of interest include T_{max}, C_{max}, and change from baseline to each time point, and incremental AUCs for glucose and C-peptide. A dose-proportionality parameter will also be estimated to establish the relative potency between LY3185643 and rGlucagon. Partial AUC for glucose over appropriate time intervals will be calculated using the trapezoidal rule. The AUC for each subject after each dose will also be baseline-adjusted. Baseline for each dose will be defined as the average of predose values for that dose.

10.3.3.2. Pharmacodynamic Statistical Inference

PD parameters of interest may be transformed before statistical analysis if deemed necessary. Unless specified otherwise, parameters will be analyzed using a nonlinear mixed-effects model with period, treatment (LY3185643 or rGlucagon), and dose (within treatment) as fixed effects, baseline glucose as a covariate, and subject as a random effect. The use of other influencing variables as covariates will be explored as detailed in the statistical analysis plan.

A conversion factor will be defined in the model for each parameter to estimate the relative potency of LY3185643 to rGlucagon. In addition, a conversion factor will be estimated between LY3185643 and rGlucagon for multiple parameters simultaneously using a similar mixed nonlinear model approach to account for the correlation between parameters for the same subject. Model details will be discussed in the statistical analysis plan.

All PD parameters including the baseline-corrected parameters will be summarized by dose group and tabulated. Summary statistics will be provided. The individual observed and mean (by dose group) time profile of the PD measurements after dosing will be plotted.

10.3.4. Evaluation of Immunogenicity

The frequency of antibody formation to LY3185643 will be determined. If a neutralization assay is performed, the frequency of neutralizing antibodies will be determined. The relationship between the presence (or absence) of antibodies and AEs will be assessed. Likewise, the relationship between the presence of antibodies and the PK parameters and PD response to LY3185643 will be assessed.

10.3.5. Interim Analyses

No interim analyses are planned for this study. If an unplanned interim analysis is deemed necessary, the Lilly clinical pharmacologist, CRP/investigator, or designee will consult with the appropriate medical director or designee to determine if it is necessary to amend the protocol.

The Lilly study team is unblinded, and the investigator will remain blinded until all data collection has been completed. Data may be analyzed while the trial is ongoing, but no changes to the study design are planned. An assessment committee will not be formed.

11. References

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Appendix 1. Abbreviations and Definitions

Term	Definition
AE	adverse event: Any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product that does not necessarily have a causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product.
ALP	alkaline phosphatase
ALT	alanine aminotransferase
APDS	artificial pancreas device system
AST	aspartate aminotransferase
AUC	area under the concentration versus time curve
blinding	A procedure in which one or more parties to the trial are kept unaware of the treatment assignment(s). Unless otherwise specified, blinding will remain in effect until final database lock. A single-blind study is one in which the investigator and/or his staff are aware of the treatment but the subject is not, or vice versa, or when the sponsor is aware of the treatment but the investigator and/his staff and the subject are not. A double-blind study is one in which neither the subject nor any of the investigator or sponsor staff who are involved in the treatment or clinical evaluation of the subjects are aware of the treatment received.
CIOMS	Council for International Organizations of Medical Sciences
C_{max}	maximum drug concentration
complaint	A complaint is any written, electronic, or oral communication that alleges deficiencies related to the identity, quality, purity, durability, reliability, safety or effectiveness, or performance of a drug or drug delivery system.
compliance	Adherence to all the trial-related requirements, good clinical practice requirements, and the applicable regulatory requirements.
confirmation	A process used to confirm that laboratory test results meet the quality requirements defined by the laboratory generating the data and that Lilly is confident that results are accurate. Confirmation will either occur immediately after initial testing or will require that samples be held to be retested at some defined time point, depending on the steps required to obtain confirmed results.
CRF	case report form

CRP	Clinical Research Physician: Individual responsible for the medical conduct of the study. Responsibilities of the CRP may be performed by a physician, clinical research scientist, global safety physician or other medical officer.
CRU	clinical research unit
DCCT	Diabetes Control and Complications Trial
ECG	electrocardiogram
EDIC	Epidemiology of Diabetes Interventions and Complications
enroll	The act of assigning a subject to a treatment. Subjects who are enrolled in the trial are those who have been assigned to a treatment.
enter	Subjects entered into a trial are those who sign the informed consent form directly or through their legally acceptable representatives.
ERB	Ethical Review Board
GCP	good clinical practice
HIV	human immunodeficiency virus
IB	Investigator's Brochure
ICF	informed consent form
ICH	International Conference on Harmonization
informed consent	A process by which a subject voluntarily confirms his or her willingness to participate in a particular trial, after having been informed of all aspects of the trial that are relevant to the subject's decision to participate. Informed consent is documented by means of a written, signed and dated informed consent form.
interim analysis	An interim analysis is an analysis of clinical trial data, separated into treatment groups, that is conducted before the final reporting database is created/locked.
Investigational product	A pharmaceutical form of an active ingredient or placebo being tested or used as a reference in a clinical trial, including products already on the market when used or assembled (formulated or packaged) in a way different from the authorized form, or marketed products used for an unauthorized indication, or marketed products used to gain further information about the authorized form.
investigator	A person responsible for the conduct of the clinical trial at a trial site. If a trial is conducted by a team of individuals at a trial site, the investigator is the responsible leader of the team and may be called the principal investigator.
LC/MS	liquid chromatography with tandem mass spectrometry
Legal representative	An individual or judicial or other body authorized under applicable law to consent, on behalf of a prospective subject, to the subject's participation in the clinical trial.

Non-investigational product	A product that is not being tested or used as a reference in the clinical trial, but is provided to subjects and used in accordance with the protocol, such as: concomitant or rescue/escape medication for preventative, diagnostic, or therapeutic reasons, medication to ensure adequate medical care, and/or products used to induce a physiological response.
PD	pharmacodynamic(s)
PK	pharmacokinetic(s)
QTc	QT corrected for heart rate
QTcF	QTc using Fridericia's formula
rGlucagon	recombinant glucagon
SAE	serious adverse event
SC	subcutaneous(ly)
screen	The act of determining if an individual meets minimum requirements to become part of a pool of potential candidates for participation in a clinical trial.
SUSARs	suspected unexpected serious adverse reactions
$t_{1/2}$	half-life
T1DM	type 1 diabetes mellitus
T_{max}	time to maximum drug concentration
ULN	upper limit of normal

Appendix 2. Clinical Laboratory Tests

Laboratory Tests

Hematology[a]	Clinical Chemistry[a]
Hematocrit	Sodium
Hemoglobin	Potassium
Erythrocyte count (RBC)	Bicarbonate
Mean cell volume	Chloride
Mean cell hemoglobin	Calcium
Mean cell hemoglobin concentration	Phosphorus
Leukocytes (WBC)	Magnesium
Absolute counts of:	Glucose (fasting)
Neutrophils	Blood urea nitrogen
Lymphocytes	Total cholesterol
Monocytes	Total protein
Eosinophils	Albumin
Basophils	Total bilirubin
Platelets	Alkaline phosphatase
	Aspartate aminotransferase
Urinalysis[a]	Alanine aminotransferase
Specific gravity	Creatinine
pH	Gammaglutamyl transferase
Protein	
Glucose	Hepatitis B surface antigen[c]
Ketones	Hepatitis C antibody[c]
Bilirubin	HIV[c]
Urobilinogen	Pregnancy test[d]
Blood	FSH[e]
Nitrite	Thyroid-stimulating hormone[f]
Leukocytes	
Microscopy[b]	

Abbreviations: FSH = follicle-stimulating hormone; HIV = human immunodeficiency virus; RBC = red blood cells; WBC = white blood cells.

a Results will be validated by the local laboratory at the time of initial testing.

b If clinically indicated, per investigator's discretion.

c Performed at screening only. Tests may be waived if they have been performed within 6 months before screening and the corresponding reports are available for review.

d Only for females. Test will be performed at screening, at every admission to the clinical research unit, and at the follow-up or early termination visit.

e For women only when needed to confirm postmenopausal status.

f Only for subjects who are being treated with thyroxine or have a history of thyroid disease.

Appendix 3. Study Governance, Regulatory, and Ethical Considerations

Informed Consent

The investigator is responsible for:

- ensuring that the subject understands the potential risks and benefits of participating in the study
- ensuring that informed consent is given by each subject or legal representative. This includes obtaining the appropriate signatures and dates on the informed consent form (ICF) before the performance of any protocol procedures and before the administration of investigational product
- answering any questions the subject may have throughout the study and sharing in a timely manner any new information that may be relevant to the subject's willingness to continue his or her participation in the trial

Ethical Review

The investigator or appropriate local representative must give assurance that the ethical review board (ERB) was properly constituted and convened as required by International Conference on Harmonization (ICH) guidelines and other applicable laws and regulations.

Documentation of ERB approval of the protocol and the ICF must be provided to Lilly before the study may begin at the investigative site(s). Lilly or its representatives must approve the ICF before it is used at the investigative site(s). All ICFs must be compliant with the ICH guideline on good clinical practice (GCP).

The study site's ERB(s) should be provided with the following:

- the current IB for LY3185643 and the prescribing information for rGlucagon and updates during the course of the study
- ICF
- relevant curricula vitae

Regulatory Considerations

This study will be conducted in accordance with:

1) consensus ethics principles derived from international ethics guidelines, including the Declaration of Helsinki and Council for International Organizations of Medical Sciences (CIOMS) International Ethical guidelines
2) applicable ICH GCP guideline
3) applicable laws and regulations

Some of the obligations of the sponsor will be assigned to a third-party organization.

Protocol Signatures

The sponsor's responsible medical officer will approve the protocol, confirming that, to the best of his or her knowledge, the protocol accurately describes the planned design and conduct of the study.

After reading the protocol, each principal investigator will sign the protocol signature page and send a copy of the signed page to a Lilly representative.

Final Report Signature

The investigator or designee will sign the clinical study report for this study, indicating agreement with the analyses, results, and conclusions of the report.

The sponsor's responsible medical officer and statistician will sign/approve the final clinical study report for this study, confirming that, to the best of his or her knowledge, the report accurately describes the conduct and results of the study.

Data Quality Assurance

To ensure accurate, complete, and reliable data, Lilly or its representatives will do the following:

- provide instructional material to the study sites, as appropriate
- provide training to instruct the investigators and study coordinators. This training will give instruction on the protocol, the completion of the case report forms (CRFs), and study procedures
- make periodic visits to the study site
- be available for consultation and stay in contact with the study site personnel by mail, telephone, and/or fax
- review and evaluate CRF data and/or use standard computer edits to detect errors in data collection
- conduct a quality review of the database

In addition, Lilly or its representatives will periodically check a sample of the subject data recorded against source documents at the study site. The study may be audited by Lilly and/or regulatory agencies at any time. Investigators will be given notice before an audit occurs.

The investigator will keep records of all original source data. This might include laboratory tests, medical records, and clinical notes. If requested, the investigator will provide the sponsor, applicable regulatory agencies, and applicable ERBs with direct access to the original source documents.

Data Collection Tools/Source Data

An electronic data capture system will be used in this study. The site must define and retain all source records and must maintain a record of any data where source data are directly entered into the data capture system.

Study and Site Closure

Discontinuation of Study Sites

Study site participation may be discontinued if Lilly or its designee, the investigator, or the ERB of the study site judges it necessary for medical, safety, regulatory, or other reasons consistent with applicable laws, regulations, and GCP.

Discontinuation of the Study

The study will be discontinued if Lilly or its designee judges it necessary for medical, safety, regulatory, or other reasons consistent with applicable laws, regulations, and GCP.

Appendix 4. Hepatic Monitoring Tests for Treatment-Emergent Abnormality

Selected tests may be obtained in the event of a treatment-emergent hepatic abnormality and may be required in follow-up with subjects in consultation with Lilly or its designee clinical research physician.

Hepatic Monitoring Tests

Hepatic Hematology[a]	**Haptoglobin[a]**
Hemoglobin	
Hematocrit	**Hepatic Coagulation[a]**
RBC	Prothrombin time
WBC	Prothrombin time, INR
Neutrophils, segmented	
Lymphocytes	**Hepatic Serologies[a,b]**
Monocytes	Hepatitis A antibody, total
Eosinophils	Hepatitis A antibody, IgM
Basophils	Hepatitis B surface antigen
Platelets	Hepatitis B surface antibody
	Hepatitis B core antibody
Hepatic Chemistry[a]	Hepatitis C antibody
Total bilirubin	Hepatitis E antibody, IgG
Conjugated bilirubin	Hepatitis E antibody, IgM
Alkaline phosphatase	
Alanine aminotransferase	**Anti-nuclear antibody[a]**
Aspartate aminotransferase	
Gammaglutamyl transferase	**Anti-smooth muscle antibody (or anti-actin antibody)[a]**
Creatinine phosphokinase	

Abbreviations: Ig = immunoglobulin; INR = international normalized ratio; RBC = red blood cells; WBC = white blood cells.

a Assayed by Lilly-designated or local laboratory.

b Reflex/confirmation dependent on regulatory requirements and/or testing availability.

Appendix 5. Blood Sampling Summary

This table summarizes the approximate number of venipunctures and blood volumes for all blood sampling (screening, safety laboratories, and bioanalytical assays) during the study. Fewer venipunctures and blood draws may actually occur, but this will not require a protocol amendment.

Protocol I8Z-MC-APCA Sampling Summary

Purpose	Maximum Blood Volume per Sample (mL)	Maximum Number of Blood Samples	Maximum Total Volume (mL)
Screening tests[a]	17	1	17
Clinical laboratory tests[a]	11	2	22
Pharmacokinetics	4	54	216
Blood discard for cannula patency	0.25	135	33.75
Pharmacodynamics (glucose)	0.2	135	27
Pharmacodynamics (glucose and C-peptide)	2.5	63	157.5
Immunogenicity	7	3	21
Total			494.25
Total for clinical purposes [rounded up to nearest 10 mL]			500

[a] Additional samples may be drawn if needed for safety purposes.

Appendix 6. Protocol Amendment I8Z-MC-APCA(a) A Randomized, 9-Way, Single-Dose, Crossover Study to Evaluate the Pharmacokinetics, Pharmacodynamics, Safety, and Tolerability of LY3185643 and rGlucagon in Healthy Subjects

Overview

Protocol I8Z-MC-APCA(a) [A Randomized, 9-Way, Single-Dose, Crossover Study to Evaluate the Pharmacokinetics, Pharmacodynamics, Safety, and Tolerability of LY3185643 and rGlucagon in Healthy Subjects] has been amended. The new protocol is indicated by Amendment (a) and will be used to conduct the study in place of any preceding version of the protocol.

The overall changes and rationale for the changes made to this protocol are as follows:

- Minor typographical errors addressed.
- Inclusion of descriptions of effective birth control methods, as suggested by the FDA.
- The Exclusion Criteria for systemic exposure to glucocorticoids within 3 months to include the use of oral or intra-articular glucocorticoids, or extensive use topical steroids, as suggested by the FDA.
- Increase in the sample size as a result of higher than expected subject drop-out rate among early participants.
- Deletion of word *Holter,* since the site uses Mortara surveyor system.
- Clarification of the duration of telemetry (text is now consistent with the information in the Schedule of Activities).
- Deletion of the phrase *LY3185643 antibodies* listed in the Clinical Laboratory Tests table. These blood samples remain listed in the Schedule of Activities.

Revised Protocol Sections

Note:	All deletions have been identified by ~~strikethroughs~~. All additions have been identified by the use of underscore.

Section 1. Synopsis

Summary of Study Design: This study is a Phase 1, single-center, subject- and investigator-blind, randomized, 9-way, 3-period crossover study in healthy subjects to evaluate the PK and PD of LY3185643 and rGlucagon after subcutaneous (SC) administration.

Up to ~~27~~37 healthy men and women may be enrolled to target approximately 18 subjects to complete the study. Each subject will receive 3 doses on each of 3 dosing days (total of 9 doses: 5 doses of LY3185643 and 4 doses of rGlucagon) administered SC. Doses will be administered in a 9-way complete crossover design in 3 periods, and subjects will be randomized to predefined treatment sequences.

Number of Subjects: Up to ~~27~~ 37 subjects may be enrolled so that approximately 18 subjects complete the study.

Section 2. Schedule of Activities

Study Schedule Protocol I8Z-MC-APCA

Procedure	Screen	Periods 1, 2, 3[a]			FU/ET[b]	Comments
	Up to Day -28	Day -1	Day 1[c]	Day 2		
Continuous ECG ~~Holter~~ monitoring ~~(24 hour)~~			X			15 ECGs will be extracted from the continuous ECG ~~Holter~~ recording at -5, 30, 60, 120, and 180 minutes postdose for each Day 1 dose administration.
Immunogenicity (LY3195643 antibodies) samples			Any time before the first dose		X	Period 1 and Period 3 only. Prior to the first dose only.

5.1. Overall Design

Up to ~~27~~37 healthy men and women may be enrolled to target approximately 18 subjects to complete the study. Each subject will receive 3 doses on each of 3 dosing days (total of 9 doses: 5 doses of LY3185643 and 4 doses of rGlucagon) administered SC. Doses will be administered in a 9-way complete crossover design in 3 periods, and subjects will be randomized to predefined treatment sequences. Completers are defined as subjects who have received all 9 doses.

5.2. Number of Participants

Up to ~~27~~37 subjects may be enrolled so that approximately 18 subjects complete the study.

Section 6.1. Inclusion Criteria

[1a] male subjects:

~~agree to use an effective method of contraception for the duration of the study~~

Male subjects with female partners of childbearing potential will be required to use a condom in conjunction with a spermicidal gel, foam, cream or suppository. In addition, the female partner will be requested to use an additional effective form of contraception, which can be any of the following:

- female condom with spermicide
- diaphragm with spermicide
- cervical sponge
- cervical cap with spermicide
- combined oral contraceptive pill and mini-pill
- NuvaRing
- implantable contraceptives
- injectable contraceptives (such as Depo-Provera®)
- intrauterine device (such as Mirena® and ParaGard®)
- total abstinence

Men who have had a vasectomy with appropriate post-vasectomy documentation of the absence of sperm in the ejaculate are not required to use contraception. Male subjects with a female partner meeting the definition of a woman not of childbearing potential will not be required to use contraception.

[1b] female subjects:

women not of childbearing potential may participate without using contraception, and include those who are:

Section 6.2. Exclusion Criteria

[26] have ~~used~~ had systemic exposure to glucocorticoids within 3 months before entry into the study, ~~or~~ (including oral or intra-articular glucocorticoids, or potent topical steroids applied to >20% of body surface area)

Section 9.4.4.1. Continuous 12-Lead ~~Holter~~ ECG Monitoring

Continuous 12-lead ~~Holter~~ECG monitoring does not allow bedside printing or viewing of ECGs extracted from the signal recorded. Thus, for subject safety, ECGs must be recorded by a local, static 12-lead ECG device at the times specified in the Schedule of Activities (Section 2). These safety ECGs will not be transmitted to a central ECG laboratory. Safety ECGs must be interpreted by the investigator or qualified designee soon after the time of collection. This review will be documented and provides information for immediate subject management. If the ECG shows a clinically significant quantitative or qualitative change from baseline, the investigator or qualified designee will assess the subject for symptoms (for example, palpitations, near syncope, syncope) and determine if the subject can continue in the study. The investigator or qualified designee is responsible for subject management. If the investigator determines that the changes identified in the ECG are clinically relevant, an applicable AE should be recorded on the subject CRF.

Section 9.4.4.1.1. ~~Holter~~ Recordings for Data Analysis

Continuous 12-lead ~~Holter~~ recordings ~~(H12+)~~ will be performed according to the Schedule of Activities (Section 2). Patients must be in a quiet atmosphere without significant external stimulation (for example, TV, internet, etc.), remain in a supine position for at least 5 to 10 minutes before the specified ECG collection times, and remain supine but awake during ECG collection and for at least 10 minutes afterward. Subjects should be encouraged to remain still, if possible, during this time.

Digital recordings of all periods of continuous ECG recordings in individual subjects will be transferred to a central ECG laboratory designated by Lilly. The ECG laboratory will perform quality control checks for the time points of interest (for example, acquisition quality for ability to measure/interpret, demographics, and study details) and extract 15 unique 10-second ECGs at the times listed in the Schedule of Activities (Section 2).

The ECG laboratory will then store the ~~24-hour~~ recording as well as the extracted 10-second ECGs, and a cardiologist at the central ECG laboratory will conduct a full overread (including the measurement of all intervals) on one of the replicates that was extracted. For each set of replicates, the cardiologist will determine the RR and QT intervals and heart rate on the ECGs that were not fully overread. No reports will be issued from the central ECG laboratory back to the sites for any ECGs.

Section 9.4.4.2. Telemetry

The ECG waveform and cardiac rhythm will be monitored continuously using an ECG telemetry monitor during the inpatient stay at the CRU, from the night of Day -1 to 4 hours after the last dose on Day 1, ~~morning of Day 2~~ for all 3 periods. Telemetry data from Day -1 up to approximately 1 hour before dosing will be reviewed by the investigator before administration of the first dose on Day 1. Subjects will not be dosed if any exclusion criteria are observed (for example, Exclusion Criterion [14]). Any other observations, such as changes in the ECG waveform and rhythm, should be evaluated by the investigator and the decision to dose will be at the investigator's discretion. Any confirmed abnormalities/events assessed to be clinically significant by the investigator during monitoring should be recorded in the medical record and reported as a preexisting condition or AE on the AE CRF page.

Appendix 2. Clinical Laboratory Tests

Laboratory Tests

Hematology[a]	Clinical Chemistry[a]
Hematocrit	Sodium
Hemoglobin	Potassium
Erythrocyte count (RBC)	Bicarbonate
Mean cell volume	Chloride
Mean cell hemoglobin	Calcium
Mean cell hemoglobin concentration	Phosphorus
Leukocytes (WBC)	Magnesium
Absolute counts of:	Glucose (fasting)
Neutrophils	Blood urea nitrogen
Lymphocytes	Total cholesterol
Monocytes	Total protein
Eosinophils	Albumin
Basophils	Total bilirubin
Platelets	Alkaline phosphatase
	Aspartate aminotransferase
Urinalysis[a]	Alanine aminotransferase
Specific gravity	Creatinine
pH	Gammaglutamyl transferase
Protein	
Glucose	Hepatitis B surface antigen[c]
Ketones	Hepatitis C antibody[c]
Bilirubin	HIV[c]
Urobilinogen	Pregnancy test[d]
Blood	FSH[e]
Nitrite	Thyroid-stimulating hormone[f]
Leukocytes	~~LY3185643 antibodies as indicated in Section 2~~
Microscopy[b]	

Abbreviations: FSH = follicle-stimulating hormone; HIV = human immunodeficiency virus; RBC = red blood cells; WBC = white blood cells.

a Results will be validated by the local laboratory at the time of initial testing.

b If clinically indicated, per investigator's discretion.

c Performed at screening only. Tests may be waived if they have been performed within 6 months before screening and the corresponding reports are available for review.

d Only for females. Test will be performed at screening, at every admission to the clinical research unit, and at the follow-up or early termination visit.

e For women only when needed to confirm postmenopausal status.

f Only for subjects who are being treated with thyroxine or have a history of thyroid disease.

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