University of Pennsylvania

CLINICAL STUDY PROTOCOL

A Multi-Center, Open Label, Collaborative Research Study to Treat HRS-AKI Patients with Continuous Terlipressin Infusion

Protocol Number:	TER001
Study Drug:	Terlipressin
Development Phase:	Phase 3
IND Number:	146962
Protocol Release Version 1 Date:	January 13, 2020
Protocol Release Version 2 Date:	September 15, 2020
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Protocol Release Version 4 Date:	June 29, 2021
Sponsor:	K. Rajender Reddy, M.D. University of Pennsylvania 2 Dulles, Liver Transplant Office 3400 Spruce Street, Philadelphia, PA 19104

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INVESTIGATOR'S AGREEMENT

I have read the attached protocol entitled "A Multi-Center, Open Label, Collaborative Research-Study to treat HRS-AKI patients with Continuous Terlipressin Infusion"and agree to abide by all provisions set forth therein.

I agree to comply with the International Conference on Harmonization (ICH) Tripartite Guideline on Good Clinical Practice (GCP), the ethical principles stated in the latest version of the Declaration of Helsinki, and the applicable local and international regulations, whichever provide the greater protection of the individual.

I agree to ensure that the confidential information contained in this document will not be used for any purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of the sponsor, University of Pennsylvania.

Investigator's Signature	Date

Institution Conducting Research

Sponsor Statement

This study protocol was subject to critical review and has been approved by the following sponsor representative.

K. Rajender Reddy — June 29, 2021

K. Rajender Reddy M.D — Date

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INFUSE: Summary of VERSION 4 Dated: June 29, 2021 Changes

Section: SYNOPSIS

- Expansion of inclusion criteria to include patients that are not liver transplant eligible or listed for liver transplantation
- Modification of inclusion criteria
- Modification of exclusion criteria to exclude patients 1) MELD ≥ 35 2) Advanced HCC with expected survival of < 6 months

Section: SAFETY AND EFFICACY ASSESSMENTS

- Clarification of secondary efficacy endpoint for Serum creatinine and incidence of dialysis at Day 30, Day 90, Day 180, and Day 365 after liver transplantation and those alive without transplantation (transplant eligible and ineligible patients)

Section: STUDY POPULATION

- Clarification that patients who are not liver transplant eligible or listed for liver transplantation will also be eligible for the study
- Modification of inclusion and exclusion criteria

Section: SECONDARY ANALYSES

- Clarification of the timepoint for those patients who were transplant ineligible or did not undergo a transplant.

General formatting and administrative clarifications were made throughout the protocol.

INFUSE: Summary of VERSION 3 Dated: March 30, 2021 Changes

Section: SYNOPSIS

- Clarification that patients who are liver transplant eligible with anticipation of being placed on the liver transplant wait list are eligible for the study
- Clarification of inclusion criterion to remove the restriction that eligible subjects are anticipated to be placed in the liver transplant wait list within 7 days of screening
- Clarification of total volume of 0.9% sodium chloride used for drug infusion

- Clarification that some lab assessments and procedures will only be collected if they are performed as per site's standard of care

Section: STUDY POPULATION

- Clarification that patients who are liver transplant eligible with anticipation of being placed on the liver transplant wait list are eligible for the study

Section: TERLIPRESSIN

- Update of stability and microbiology testing, including stability at refrigerated temperatures of 48 hours and room temperature for 24 hours
- Clarification of total volume of 0.9% sodium chloride used for drug infusion

Section: ASSESSMENTS AND PROCEDURES

- Clarification that assessments are completed per sites' Standard of Care.
- Clarification of the timepoints for echocardiogram
- Clarification of the timepoints for optional cystatin c and biomarker samples
- Clarification that optional biomarker samples may be drawn +/- 1 day of the target day
- Clarification that some lab assessments and procedures will only be collected if they are performed as per site's standard of care
- Clarification of acceptable window for follow up assessments

Section: STUDY ASSESSMENTS

- Clarification that some lab assessments and procedures will only be collected if they are performed as per site's standard of care

Section: DETAIL OF LABORATORY ASSESSMENTS AND PROCEDURES

- Clarification of the timepoints for optional cystatin c and biomarker samples
- Clarification that some lab assessments and procedures will only be collected if they are performed as per site's standard of care

Section: REFERENCES

- Addition of reference Bui 2020

Section: APPENDIX C

- Addition of Appendix C: Terlipressin Microbiology

General formatting and administrative clarifications were made throughout the protocol.

INFUSE: Summary of VERSION 2 Dated: September 15, 2020 Changes

Section: SYNOPSIS

- Added clarification for retrospective case collection time-period parameters.
- Exclusion criteria addition to exclude ACLF grade 3 subjects.
- Exclusion criteria addition to exclude subjects with SCr greater than 5.0 mg/dL unless approved by Sponsor Medical Monitor.
- Exclusion criteria clarification on participation in other clinical trials.
- Exclusion criteria clarification on prior use of vasopressors for prospective subjects.
- Dosing clarification that only 0.9% sodium chloride solution should be used.
- Lab clarification on collection of BUN and eGFR.

Section: INVESTIGATIONAL PLAN

- Added clarification for retrospective case collection time-period parameters.
- Exclusion criteria addition to exclude ACLF grade 3 subjects.
- Exclusion criteria addition to exclude subjects with SCr greater than 5.0 mg/dL unless approved by Sponsor Medical Monitor.
- Exclusion criteria clarification on participation in other clinical trials.
- Exclusion criteria clarification on prior use of vasopressors for prospective subjects.

Section: STUDY DRUG STORAGE AND PREP

- Corrected stability of study drug once reconstituted is 48 hours for this study.

Section: ADMINISTRATION

- Clarification on dosing procedures.

Section: SCHEDULE OF ASSESSMENTS

- Clarifications on assessments to be collected

Section: PHYSICAL EXAMINATION

- Added collection of Estimated Dry Body Weight.

Section: ECHOCARDIOGRAM

- Clarification on collection requirements.

Section: VITAL SIGNS

- Clarification of vital sign collection parameters.

Section: ADVERSE EVENTS

- Clarification that SAEs are collected through 30 days post treatment. Deaths are reported until study completion but will not be SAEs if occurred after SAE reporting timeframe.

Section: SERUM ELECTROLYTES & BIOCHEMISTRY

- Clarification of assessments to be collected.

Section: HEMATOLOGY

- Clarification of assessments to be collected.

Section: SERIOUS ADVERSE EVENTS

- Clarification on reporting requirements
- Deletion of Adverse Event information due to not applicable for protocol.
- Clarification on SUSAR reporting.

Section: COLLECTION OF ADVERSE EVENTS/SERIOUS OR UNEXPECTED

- Clarification of reporting procedures.

Section: SITE REPORTING TO SPONSOR

- Correction to reporting contact information.

Section: SAFETY VARABLES

- Clarification of death collection.

Section: STUDY AND STUDY SITE DISCONTINUATION CRITERIA

- Removal of incorrect Sponsor reference.

Section: REFERENCES

- Added a protocol reference for Electrocardiogram/Echocardiogram assessments.

General formatting and administrative clarifications were made to overall Protocol

1 SYNOPSIS

(synopsis to follow)

SYNOPSIS
Name of Sponsor/Company: University of Pennsylvania
Name of Investigational Product: Terlipressin
Name of Active Ingredient: Terlipressin
Title of Study: A Multi-Center, Open Label, Collaborative Research Study to Treat HRS-AKI Patients with Continuous Terlipressin Infusion
Study Centers: Approximately 5-10 sites in the United States of America
Phase of Development: Phase 3
Objectives: **Primary:** To assess safety/efficacy for continuous terlipressin infusion in adult subjects with Hepatorenal syndrome- Acute Kidney Injury (HRS-AKI) who are in one of two groups: 1. On the liver transplant wait list or liver transplant eligible with anticipation of being placed on the liver transplant wait list 2. Not eligible for liver transplant (limited to 25 patients)
Methodology: Prospective Terlipressin Group - Patients to be treated with open label terlipressin by infusion following an initial bolus dose of 0.5mg. Retrospective Control Group – Historical patients who would have met inclusion/exclusion criteria detailed below and received at least 48 hours of treatment for HRS-AKI according to center's standard-of-care (Midodrine/Octreotide (M&O), albumin, or other vasopressor)
Number of Subjects (Planned): Prospective Terlipressin Group - approximately 50 terlipressin treated subjects Retrospective Control Group - approximately 15-25 subjects per clinical site that were treated for HRS-AKI, that would have met the inclusion/exclusion criteria below as well as having critical clinical data points to enable a comparison of the Prospective and Retrospective groups. Retrospective cases collected from each site should include eligible patients starting from prior to site IRB approval for this protocol and going back

to January 2015. Subjects enrolled in a blinded clinical trial in this time period are excluded.

Diagnosis and Main Inclusion/Exclusion Criteria:

Adult subjects that are listed for liver transplant or liver transplant eligible with anticipation of being placed on the liver transplant wait list and have a diagnosis of HRS-AKI based on 2015 revised International Ascites Club (IAC) diagnostic criteria.

Inclusion Criteria:

1. Written informed consent by subject or legally authorized representative.
2. At least 18 years of age.
3. Cirrhosis and ascites.
4. No sustained improvement in renal function (less than 20% decrease in SCr) at least 48 hours after diuretic withdrawal and after plasma volume expansion with albumin (given daily for two days - 48 hours minimum from 1st dose).If SCr improves by ≥ 20 % but plateaus (≤ 10 % fluctuation in sCr) and remains above 1.5 mg/dl for ≥another 48 hrs and there are no features of acute tubular necrosis.
5. Increase in SCr by at least ≥ 0.3 mg/dl OR 1.5-2 fold above baseline (AKI stage 1 and above), to a SCr of ≥ 1.5 mg/dl at the time of initiating treatment. Baseline SCr is defined as the most recent, lowest SCr within last 6 months before date of current admission.
6. A.) On liver transplant wait list or liver transplant eligible with anticipation of being placed on the liver transplant wait list. B.) Patients not on the transplant waitlist or transplant eligible are also eligible for the trial (maximum 25 subjects)

Exclusion Criteria:

1. Serum creatinine level greater than 5.0 mg/dL. Subjects with value greater than 5.0 mg/dL may be enrolled with Sponsor prior approval.
2. MELD score of ≥ 35
3. Acute on Chronic Liver Failure (ACLF) grade 3 (according to the CLIF Consortium grading system)
4. Uncontrolled sepsis and/or uncontrolled bacterial infection (e.g., persisting bacteremia, persisting ascitic fluid leukocytosis, fever, increasing leukocytosis with vasomotor instability).
5. Shock.
6. Current or recent (within 4 weeks) treatment with or exposure to nephrotoxic agents: e.g., aminoglycosides, amphotericin, cyclosporine A, cisplatin, nonsteroidal anti-inflammatory drugs (NSAIDs: e.g., ibuprofen, naproxen, diclofenac), significant exposure to radiographic contrast agents (large doses or multiple injections of iodinated contrast media; e.g., during coronary or abdominal angiogram).

7. Estimated life expectancy of less than 7 days.
8. Advanced hepatocellular Carcinoma with expected life expectancy of < 6 months
9. Superimposed acute liver injury due to drugs (e.g., acetaminophen), dietary supplements, herbal preparations, viral hepatitis, or toxins (e.g., Amanita toxin with mushroom poisoning carbon tetrachloride), with the exception of acute alcoholic hepatitis.
10. Evidence of obstructive uropathy or parenchymal renal disease. Renal ultrasound or other imaging not required but should be taken if suspicious.
11. Tubular epithelial casts, heme granular casts (range of 1-3 granular casts acceptable), hematuria or microhematuria on urinalysis that is indicative of acute tubular necrosis and/or intrinsic renal disease.
12. Subjects known to be pregnant; all women of child-bearing age and potential must have a negative pregnancy test.
13. Severe cardiovascular disease, including, but not limited to, unstable angina, pulmonary edema, congestive heart failure, or persisting symptomatic peripheral vascular disease, myocardial infarction or stable chronic angina within the past 12 months, or any other cardiovascular disease judged by the investigator to be severe and creates a risk to subject with concurrent terlipressin use.
14. Current or recent (within 4 weeks) renal replacement therapy (RRT) or anticipation of RRT within 3 days on enrollment.
15. Participation in other clinical research involving investigational medicinal products within 30 days of starting study drug that would adversely affect participation in this or the current trial.
16. Transjugular intrahepatic portosystemic shunt (TIPS) within 30 days of starting study drug.
17. For the Prospective Group: All vasopressors must be stopped prior to treatment with terlipressin. Use of vasopressors (e.g., norepinephrine, epinephrine or vasopressin dopamine or other vasopressors) of ≥ 3 consecutive days within the prior 14-day screening period are excluded. Patients receiving a vasopressor other than midodrine within 24 hours of qualifying SCr are excluded, i.e, a 24-h washout is required prior to enrollment.

    Note: Patients receiving midodrine and octreotide may be enrolled. Midodrine and octreotide treatment must be stopped prior to enrollment.
18. Known allergy or sensitivity to terlipressin.

Investigational Product, Dosage and Mode of Administration:

Prospective Terlipressin Group:

Terlipressin will be administered intravenously as a bolus injection for the first dose, then by continuous IV infusion.

Retrospective Control Group:

Dose and method of administration for M&O, albumin, or other vasopressor

Dosage and Mode of Administration:

For Retrospective Control Group:

Dosing for M&O , albumin, or other vasopressor will be reported as it was recorded on the subject's chart.

For Prospective Terlipressin Group:

Terlipressin will be supplied in single-use, sterile 6-mL vials containing 1 mg of lyophilized Terlipressin acetate (equivalent to 0.85 mg Terlipressin free base) with 10 mg mannitol as a bulking agent/stabilizer.

Initial Dosing:

Terlipressin will be administered intravenously as a bolus injection (the vial will be reconstituted with 5 mL of sterile 0.9% sodium chloride solution and given over 1 minute at a dose of 0.5 mg.

- Continued dosing will be by 2 mg infusion per day. For continuous infusion, the dose of terlipressin is to be dissolved in 0.9% sodium chloride solution and infused with a pump.

Dose Modifications:

- If on Day 3 the SCr is not 30% lower from the baseline value, increase infusion dose to 4 mg per day.

- If on Day 5 the SCr is not 50% lower from the baseline value, increase infusion dose to 6-8 mg per day, per PI discretion. Dosing can be increased by more than 1 mg increment at Day 5 or beyond, up to a maximum of 8 mg/day.

- If the subject has infusion stopped for ≥ 4 hours, a 0.5 mg bolus should be given prior to restarting infusion.
- The drug should not be used in subjects with severe coronary artery disease or in the setting of circulatory overload, pulmonary edema, or bronchospasm as determined by the Investigator. If drug therapy has been initiated, Investigator can also decide not to increase the dose, based on the patient's status with regard to the above, based on individual subject condition.

- If dosing is interrupted due to an adverse event, terlipressin may be re-started, at the discretion of the investigator, at the same or lower dose as per protocol. Terlipressin will not be restarted if dosing was interrupted due to cardiac ischemia or mesenteric ischemia.

Treatment Discontinuation:

- Terlipressin may be continued until 24 hours after serum creatinine has reached the lowest value possible as determined by Principal Investigator, up to a maximum of 14 days.
- After a minimum of 72 hours post final dose escalation and tolerability, if the SCr is at or above enrollment value, terlipressin should be discontinued.
- If 72 hours after final dose escalation, there is no further improvement in SCr, continuation of terlipressin is at the discretion of the Principal Investigator at the clinical site.
- If a subject is initiated on RRT, terlipressin can be continued for a maximum of 72 hours after initiation of RRT.
- Terlipressin must be discontinued when the subject is to undergo liver transplantation, TIPS, or any other vasopressor therapy (single administration of vasopressor for cardiac stress test is acceptable)
- Dosing must be permanently discontinued if an event of cardiac ischemia or mesenteric ischemia occurs.

Retreatment:

If judged by the investigator to be potentially beneficial, subjects who demonstrate at least a partial response during the initial treatment course (at least 50% reduction in SCr) and develop recurrence of HRS-AKI may be retreated for a maximum of 14 days. To qualify for retreatment the subject must again meet the study inclusion/exclusion criteria. Retreatment may occur within 90 days of the subject's first dose of terlipressin.

Duration of Treatment:

For the Prospective Terlipressin Group, the active treatment period will be up to 14 days, allowing 1 retreatment cycle.

For both Prospective and Retrospective Subjects:

<u>All subjects will be followed:</u>

Post treatment clinical data collection will be through phone call and chart review. Creatinine, BUN and eGFR will be recorded per standard of care, recording mortality and the date of last dialysis (CVVH or HD) session for the following time points:

Day 7 +/- 2 days;

Day 14 +/- 3 days;

Day 30 +/- 1 week and

Day 90 +/- 1 week

Day 180 +/- 2 weeks and

Day 365 +/- 2 weeks.

For subjects who have a liver transplant Post Treatment

If the subject has a liver transplant recorded prior to the 90-Day visit (phone call or chart review), additional follow up will occur through one year after liver transplant.

Creatinine, BUN and eGFR will be recorded per standard of care, recording mortality and the date of last dialysis (CVVH or HD) session for the following time points:

Day 0 (date of transplant) – record allocation MELD and last calculated MELD prior to transplant;

Day 7 +/- 2 days;

Day 14 +/- 3 days;

Day 30 +/- 1 week,

Day 90 +/- 1 week,

Day 180 +/- 2 weeks and

Day 365 +/- 2 weeks.

Key Efficacy and Safety Data Assessment:

Primary Efficacy Assessment:

Improvement of renal function (SCr) from Day 1 through end of treatment, repeated measure analysis. SCr will be collected at baseline and daily from Day 1 through end of treatment.

Secondary Efficacy Endpoints:

- Improvement of renal function (SCr) from Day 1 through end of treatment, repeated measure analysis as compared to the Retrospective Control Group.

- For the Prospective Terlipressin Group alone and also in comparison to the Retrospective Control Group:
    - Incidence of dialysis by Day 30 and Day 90 of follow up.
    - Survival by Day 30 and Day 90 follow up.
    - Transplant by Day 30 and Day 90 follow up.
    - Serum creatinine at Day 30 and Day 90 follow up
    - Serum creatinine and incidence of dialysis at Day 30, Day 90, Day 180, and Day 365 after liver transplantation
    - Incidence of kidney transplant within one year after liver transplant
    - Descriptive analysis of number of simultaneous liver kidney transplants and episodes of graft rejection by Day 90 and Day 365 after liver transplant

Safety:

Safety review and assessments will be made only for the Prospective Terlipressin Group and will include the following:

- Serious adverse events up to 30 days after end of treatment.

Statistical Methods:

A formal sample size calculation is not applicable for this study. It is expected that at least 50 subjects across more than 5-10 institutions may receive terlipressin treatment during the course of the research study.

Additionally, up to 150 subjects across all clinical sites will have their Retrospective data collected. Descriptive analysis will be planned for the efficacy and safety data collected. Sponsor may perform additional analysis not specified in study protocol.

Contents

2 List of Abbreviations

Abbreviation	Definition
AASLD	American Association for the Study of Liver Diseases
ACLF	acute on chronic liver failure
AE	adverse event
AKI	acute kidney injury
ALP	alkaline phosphatase
ALT	alanine amino transferase, also known as SGPT
ANCOVA	analysis of covariance
AR	adverse reaction
AST	aspartate amino transferase, also known as SGOT
BUN	blood urea nitrogen
CBC	complete blood count
CFR	Code of Federal Regulations
CMH	Cochran-Mantel-Haenszel
CrCl	creatinine clearance
CRF	Case report form
EASL	European Association for the Study of the Liver
ECG	Electrocardiogram
eGFR	estimated glomerular filtration rate
EVH	esophageal variceal hemorrhage
FDA	Food and Drug Administration
GCP	Good Clinical Practice
HRS	hepatorenal syndrome
IAC	International Ascites Club
IAC-AKI	IAC-acute kidney injury
IB	Investigator's Brochure
ICF	informed consent form
ICH	International Conference on Harmonization
IME	important medical events
IND	Investigational New Drug
INR	international normalized ratio
IRB	Institutional Review Board
ITT	intent to treat
IV	Intravenous
LAR	Legally Authorized Representative
LVP	large volume paracentesis
MAP	mean arterial pressure

Abbreviation	Definition
MedDRA	Medical Dictionary for Regulatory Activities
MELD	model for end-stage liver disease
M&O	Midodrine/Octreotide
NDA	New Drug Application
NSAID	nonsteroidal anti-inflammatory drug
RBC	red blood cell
RRT	renal replacement therapy
SAE	serious adverse event
SUSAR	suspected unexpected adverse reaction
SCr	serum creatinine
SIRS	systemic inflammatory response syndrome
SOC	standard of care
SpO_2	pulse oximetric saturation
SRO	Safety Review Officer
TIPS	transjugular intrahepatic portosystemic shunt
US	United States
WBC	white blood cell
WHO	World Health Organization

3 Introduction

3.1 Hepatorenal Syndrome

Hepatorenal syndrome (HRS) Type 1, a potentially reversible renal failure, is a serious, rapidly progressing, fatal orphan disease complicating decompensated chronic liver disease associated with cirrhosis (Arroyo, 1996; Salerno, 2007; Angeli, 2015). The estimated United States (US) annual incidence for HRS Type 1 ranges between 9,000 and 20,000 patients (Marrero, 2003; Muir, 2002; Murkerjee, 2002; National Hospital Discharge Survey 2005), establishing it as an orphan disease. The death rate from chronic liver disease and cirrhosis has been rising, while the death rates for other major diseases such as stroke, cancer, heart disease, and diabetes have gone down in the past 15 years (Centers for Disease Control and Prevention 2013; Ma, 2015).

As indicated in the American Association for the Study of Liver Diseases (AASLD) Guidelines, at present, there is no available pharmacological therapy (i.e., approved or proven) in the US or Canada for HRS Type 1 (Runyon, 2013) and there remains a significant unmet need.

An increasing body of knowledge of the pathophysiology of HRS Type 1 has demonstrated that vasoconstrictive drug therapy may improve renal function in HRS Type 1 (Salerno, 2007; European Association for the Study of the Liver 2010). Terlipressin has been extensively studied as a splanchnic vasoconstrictor for the treatment of HRS Type 1 and is the standard of care for this condition wherever the drug is available (European Association for the Study of the Liver 2010). Although there has been some off-label use of other vasoconstrictors, a recent study has shown that the most commonly used agents (i.e., combination of midodrine and octreotide) are not effective (Cavallin, 2015).

There is a shift in HRS terminology to move away from HRS Type 1 and HRS Type 2 to the new designations of HRS-Acute Kidney Injury (HRS-AKI) and HRS-Chronic Kidney Disease (HRS-CKD), respectively, in order to better harmonize with the existing nomenclature in nephrology, the clinical consequences of kidney injury, and initiation of treatment at earlier time points when it is more effective (Angeli, 2015). HRS-AKI removes the condition of creatinine doubling over two weeks, as often patients with ascites, a precipitating event, and HRS physiology may not have had a creatinine checked in the preceding two weeks but would still benefit from HRS treatment. Additionally, HRS-AKI divides the kidney injury into stages (1-3) to mirror the AKI criteria set forth by the Acute Kidney Injury Network (AKIN) (Angeli, 2015).

3.2 Terlipressin

Terlipressin is a synthetic vasopressin analogue that acts as a systemic vasoconstrictor via the vascular vasopressin V_1 receptors. In HRS patients the strong V_1 receptor-mediated vasoconstrictor activity of terlipressin, particularly in the splanchnic area, increases effective intravascular volume and mean arterial pressure (MAP), ameliorates renin-angiotensin-aldosterone system and sympathetic nervous system hyperactivity, and improves renal blood flow. These corrective hemodynamic effects culminate in improved renal function, thereby providing the pharmacologic rationale for treatment of HRS with terlipressin (Arroyo, 2000; Ginès, 2003; Kiszka-Kanowitz, 2004).

After intravenous (IV) administration of terlipressin, the glycyl residues of terlipressin are cleaved by endogenous tissue proteases. Thus, terlipressin levels in the blood decrease rather rapidly and the pharmacologically active metabolite lysine-vasopressin is released gradually from tissues into the circulation. Intravenous terlipressin can be delivered quickly via bolus injection or slowly through continuous infusion. There is evidence that continuous infusion leads to fewer adverse events and lower total terlipressin doses to achieve HRS reversal (Cavallin, 2016). A bolus injection prior to continuous infusion will achieve therapeutic levels earlier than continuous infusion alone.

Terlipressin is approved in many countries and regions outside the US where it has been a standard of care for decades in the treatment of subjects with liver cirrhosis and esophageal variceal hemorrhage (EVH) (de Franchis, 2005; Ioannou, 2003), and has more recently become a standard of care for the treatment of subjects with HRS Type 1 where it is available (EASL, 2010). Terlipressin has been approved for the treatment of HRS Type 1 in Australia, France, Ireland, South Korea, Mexico, Taiwan, Spain, India, Brazil, Turkey, and Portugal.

3.3 Efficacy

Improvement in renal function is the goal of therapy for HRS-AKI; improvement in renal function is associated with improvement in clinical outcomes and improved prognosis, with or without liver transplantation. HRS reversal is a widely accepted, clinically meaningful response to treatment for HRS Type 1, as apparent inexorable, rapid progression of renal failure is arrested and normal renal function in the setting of decompensated liver disease is restored. The primary endpoint of HRS reversal (1 serum creatinine [SCr] value of $\leq$ 1.5 mg/dL) is the standard endpoint utilized in the literature and HRS treatment guidelines (Arroyo, 1996; Salerno, 2007; Angeli, 2015). Both terlipressin bolus and continuous infusion have been shown to be effective

in the treatment of HRS Type 1, with fewer adverse events occurring in the cohort receiving continuous infusion. (Cavallin, 2016).

3.4 Safety

The safety characteristics of terlipressin is captured in the literature and post-marketing data from the World Health Organization (WHO) database. In general, terlipressin has a well-established safety profile in the treatment of patients with HRS Type 1. The safety risks are predictable and recognizable. The adverse drug reactions can be readily managed and reversed by terlipressin dose reduction or interruption in hospital-care settings. Continuous infusion of terlipressin may be more tolerable than bolus injection (Cavallin, 2016).

3.5 Retrospective Control Group: M&O Treated Subjects

3.5.1 Midodrine and Octreotide

Midodrine is an oral alpha-receptor agonist that binds to peripheral alpha-receptors on both arterioles and veins, leading to an increase in blood pressure (Cruz, 2000). Midodrine is a prodrug; it is transformed into the active metabolite de-glymidodrine, a compound similar in structure to norepinephrine (Cruz, 2000). Midodrine was approved for use in the United States for treatment of orthostatic hypotension in 1996. Octreotide is somatostain analogue that binds to somatostain receptors and mimics the physiologic effects of somatostatin including inhibition of various hormones, some of which lead to vasodilation, thus octreotide can cause arteriolar vasoconstriction leading to increases in blood pressure (Gold Standard). Octreotide can be administered via intravenous or subcutaneous routes and was approved for use in the United States in 1988 for treatment of certain neuroendocrine tumors.

3.5.2 M&O Off-Label Use

The combination of midodrine/octreotide (M&O), in conjunction with albumin, has been used "off-label" in the United States for treatment of HRS Type 1 for years. Small studies, mostly single center, have demonstrated favorable outcomes for M&O in treatment of HRS Type 1 (Angeli, 1999; Wong, 2004; Esrailian, 2007). The use of M&O plus albumin is recommended in the 2012 AASLD guidelines for the treatment of HRS Type 1 (Runyon, 2013) , particularly in light of the fact that terlipressin was not available in the United States at the time of publication. However, the largest multi-centered study comparing terlipressin to M&O undertaken by the Italian Association for the Study of the Liver Study Group on Hepatorenal Syndrome, was stopped early due to the considerable differences in HRS reversal between terlipressin and M&O (55.5 vs 4.8%, $p<0.001$) (Cavallin, 2015).

Midodrine/Octreotide use in patients on the liver transplant wait list with HRS Type 1

Increasing the duration of time in which a patient with decompensated cirrhosis experiences renal dysfunction decreases the likelihood of renal recovery after liver transplant (Campbell, 2005). Wong et al retrospectively evaluated patients that had experienced HRS Type 1 without reversal prior to liver transplant at University of Toronto between 2001 to 2010 (n=62); M&O was used as treatment of HRS Type 1 in the majority of these patients (Wong, 2015). While none of the patients in the cohort had HRS reversal, 76% were free from dialysis 30 days after liver transplant and had a serum creatinine ≤ 1.5 mg/dL (Wong, 2015). The duration of HRS type 1 and the number of dialysis days prior to liver transplant were highly predictive of post-transplant renal function (Wong, 2015). Additionally, those patients that were not dialysis-free 30 days post-transplant with serum creatinine ≤ 1.5 mg/dL had higher mortality rates (Wong, 2015). This data suggests that even if HRS reversal is not possible, that limiting the dialysis days and length of HRS type 1 can lead improve renal outcomes post-transplant.

4 Study Rationale

The purpose of this open labeled study is to assess safety/efficacy for continuous terlipressin infusion in liver transplant-eligible adult subjects with Hepatorenal syndrome- Acute Kidney Injury (HRS-AKI).

5 Safety and Efficacy Assessments

5.1 Efficacy Variables

The efficacy variables are summarized below, and further details are provided in Section 13.

Primary Efficacy Assessment:

Improvement of renal function (SCr) from Day 1 through end of treatment, repeated measure analysis. SCr will be collected daily, from Day 1 through end of treatment. Baseline SCr will also be entered.

Secondary Efficacy Endpoints:

- Improvement of renal function (SCr) from Day 1 through end of treatment, repeated measure analysis as compared to the Retrospective Control Group.
- For the Prospective Terlipressin Group alone and also in comparison to the Retrospective Control Group:
    - Incidence of dialysis by Day 30 and Day 90 of follow up.

- Survival by Day 30 and Day 90 follow up.
- Transplant by Day 30 and Day 90 follow up.
- Serum creatinine at Day 30 and Day 90 follow up
- Serum creatinine and incidence of dialysis at Day 30, Day 90, Day 180, and Day 365 after liver transplantation and those alive without transplantation (transplant eligible and ineligible patients)
- Incidence of kidney transplant within one year after liver transplant.
- Descriptive analysis of number simultaneous liver kidney transplants and episodes of graft rejection at Day 90 and Day 365 after liver transplant.

Safety:

Safety review and assessments will be made only for the Prospective Terlipressin Group and will include the following:

- Serious adverse events up to 30 days after end of treatment.

6 Investigational Plan

6.1 Overall Study Design and Plan: Description

Prospective Terlipressin Group

This is a collaborative open-label, multicenter study of terlipressin infusion following an initial bolus dose of 0.5mg. Written informed consent will be obtained by the investigator, sub-investigator, or qualified designee from the subject or legally authorized representative (LAR) prior to the subject enrollment into the study. Approximately 50 subjects are planned to be enrolled at 5-10 sites in the US. Subjects will receive up to 14 days of terlipressin treatment.

Retrospective Control Group

This study will also seek clinical data from a Retrospective Control Group of patients who would have met inclusion/exclusion criteria detailed below and received at least 48 hours of treatment with Midodrine and Octreotide (M&O), albumin, or other vasopressor used for the treatment of HRS-AKI. Retrospective cases collected from each site should include eligible patients starting from prior to site IRB approval for this protocol and going back to January 2015. Subjects enrolled in a blinded clinical trial in this time period are excluded.

7 Study Population

7.1 Population Rationale

The study population consists of adult subjects with cirrhosis, ascites, and a diagnosis of HRS AKI based on based on the 2015 revised International Ascites Club (IAC) diagnostic criteria.

7.2 Number of Subjects to be Studied

Prospective Terlipressin Group - approximately 50 Terlipressin treated subjects. Retrospective Control Group - approximately 15-25 subjects per clinical site that were treated for HRS-AKI according to center's standard-of-care (Midodrine/Octreotide (M&O), albumin, or other vasopressor) that would have met the inclusion/exclusion criteria below as well as having critical clinical data points to enable a comparison of the Prospective and Retrospective groups. Retrospective cases collected from each site should include eligible patients starting from prior to site IRB approval for this protocol and going back to January 2015. Subjects enrolled in a blinded clinical trial in this time period are excluded.

7.3 Inclusion/Exclusion Criteria

Adult subjects with a diagnosis of HRS-AKI based on 2015 revised International Ascites Club (IAC) diagnostic criteria will be considered for enrollment. There are two groups: 1. listed for liver transplant or liver transplant eligible with anticipation of being placed on the liver transplant wait list; 2. Ineligible for liver transplant

Inclusion Criteria: Subjects must meet all of the following inclusion criteria to be eligible for enrollment:

1. Written informed consent by subject or legally authorized representative.
2. At least 18 years of age.
3. Cirrhosis and ascites.
4. No sustained improvement in renal function (less than 20% decrease in SCr) at least 48 hours after diuretic withdrawal and after plasma volume expansion with albumin (given daily for two days - 48 hours minimum from 1st dose). If SCr improves by ≥ 20 % but plateaus (≤ 10 % fluctuation in sCr) and remains above 1.5 mg/dl for ≥another 48 hrs and there are no features of acute tubular necrosis.

5. Increase in SCr by at least ≥ 0.3 mg/dl OR 1.5-2 fold above baseline (AKI stage 1 and above), to a SCr of ≥ 1.5 mg/dl at the time of initiating treatment. Baseline SCr is defined as the most recent, lowest SCr within last 6 months before date of current admission.
6. A.) On liver transplant wait list or liver transplant eligible with anticipation of being placed on the liver transplant wait list. B.) Patients not on the transplant waitlist or transplant eligible are also eligible for the trial (maximum 25 subjects)

Exclusion Criteria: If any of the following exclusion criteria are met, the subject will **not** be enrolled:

1. Serum creatinine level greater than 5.0 mg/dL. Subjects with value greater than 5.0 mg/dL may be enrolled with Sponsor prior approval.
2. MELD score ≥ 35
3. Acute on Chronic Liver Failure (ACLF) grade 3 (according to the CLIF Consortium grading system).
4. Uncontrolled sepsis and/or uncontrolled bacterial infection (e.g., persisting bacteremia, persisting ascitic fluid leukocytosis, fever, increasing leukocytosis with vasomotor instability).
5. Shock.
6. Current or recent (within 4 weeks) treatment with or exposure to nephrotoxic agents: e.g., aminoglycosides, amphotericin, cyclosporine A, cisplatin, nonsteroidal anti-inflammatory drugs (NSAIDs: e.g., ibuprofen, naproxen, diclofenac), significant exposure to radiographic contrast agents (large doses or multiple injections of iodinated contrast media; e.g., during coronary or abdominal angiogram).
7. Estimated life expectancy of less than 7 days.
8. Advanced Hepatocellular Carcinoma (HCC) with expected survival of < 6 months.
9. Superimposed acute liver injury due to drugs (e.g., acetaminophen), dietary supplements, herbal preparations, viral hepatitis, or toxins (e.g., *Amanita* toxin with mushroom poisoning carbon tetrachloride), with the exception of acute alcoholic hepatitis.
10. Evidence of obstructive uropathy or parenchymal renal disease. Renal ultrasound or other imaging not required but should be taken if suspicious.

11. Tubular epithelial casts, heme granular casts (range of 1-3 granular casts acceptable), hematuria or microhematuria on urinalysis that is indicative of acute tubular necrosis and/or intrinsic renal disease.
12. Subjects known to be pregnant; all women of child-bearing age and potential must have a negative pregnancy test.
13. Severe cardiovascular disease, including, but not limited to, unstable angina, pulmonary edema, congestive heart failure, or persisting symptomatic peripheral vascular disease, myocardial infarction or stable chronic angina within the past 12 months, or any other cardiovascular disease judged by the investigator to be severe and creates a risk to subject with concurrent terlipressin use.
14. Current or recent (within 4 weeks) renal replacement therapy (RRT) or anticipation of RRT within 3 days on enrollment.
15. Participation in other clinical research involving investigational medicinal products within 30 days of starting study drug that would adversely affect participation in this or the current trial.
16. Transjugular intrahepatic portosystemic shunt (TIPS) within 30 days of starting study drug.
17. For the Prospective Group: All vasopressors must be stopped prior to treatment with terlipressin. Use of vasopressors (eg, norepinephrine, epinephrine or vasopressin dopamine or other vasopressors) of ≥ 3 consecutive days within the prior 14-day screening period are excluded. Patients receiving a vasopressor other than midodrine within 24 hours of qualifying SCr are excluded, i.e, a 24-h washout is required prior to enrollment.

    Note: Patients receiving midodrine and octreotide may be enrolled. Midodrine and octreotide treatment must be stopped prior to enrollment.
18. Known allergy or sensitivity to terlipressin.

7.4 Subject Withdrawal Criteria

Subjects have the right to discontinue treatment and/or withdraw from the study at any time without prejudice. The investigator may discontinue any subject at any time for any reason.

7.5 Protocol Deviations

This study will be conducted as described in this protocol, except for an emergency situation in which the protection, safety, and wellbeing of the subject requires immediate intervention that

deviates from the protocol, based on the judgment of the investigator (or a responsible, appropriately trained professional designated by the investigator). In the event of a significant deviation from the protocol due to an emergency, accident, or error, the investigator or designee must contact the sponsor, or their agent, at the earliest possible time by telephone. This will allow an early joint decision regarding the subject's continuation in the study. The investigator and the sponsor will document this decision. The Institutional Review Board (IRB) will be informed of all protocol deviations by the investigator in accordance with the IRB established procedure. No deviations from the protocol of any type will be made without complying with all the IRB established procedures. Significant deviations from the protocol may not allow that patient's clinical data to be included in the Intent-To-Treat Analysis.

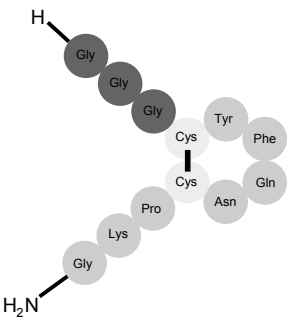
8 Terlipressin

Terlipressin will be supplied in single-use, sterile 6-mL vials containing 1 mg of lyophilized terlipressin acetate (equivalent to 0.85 mg terlipressin free base) with 10 mg mannitol as a bulking agent/stabilizer.

The lot numbers will be documented in the trial master file.

Figure 1: Terlipressin for Injection

Structure:



Molecular Formula: $C_{52}\ H_{74}\ N_{16}\ O_{15}\ S_2$

Molecular Weight: 1227.4 Daltons

Appearance: Homogenous lyophilized white to off-white solid

Solubility: Clear, colorless solution in saline

Vials:	Colorless glass vials containing 11 mg of a white to off-white solid, 1 mg active ingredient and 10 mg mannitol.

The active ingredient, N-[N-(N-glycylglycyl) glycyl]-8-L-lysinevasopressin, is a synthetically manufactured hormonogen of 8-lysine vasopressin, composed of 12 amino acids and having the characteristic ring structure of a cyclic nonapeptide with a disulfide bridge between the 4th and the 9th amino acid. Three glycyl-amino acids are substituted at position 1 (cysteine) of 8-lysine-vasopressin. By this N-terminal extension of 8-lysine-vasopressin the metabolic degradation rate of the active ingredient is significantly reduced, because the glycyl molecules inhibit rapid N-terminal enzymatic degradation.

8.1 Packaging and Labeling

Single-use, sterile 6-mL vials of a lyophilized solid containing 1 mg of terlipressin acetate (equivalent to 0.85 mg terlipressin free base) will be provided to the sites. Study drug labels will comply with local regulatory requirements, including sponsor name and address, protocol number, storage conditions and US Investigational New Drug (IND) caution statement.

8.2 Study Drug Storage and Preparation

Study drug should be stored in a secure location at 2ºC to 8ºC until reconstituted. Reconstituted study drug bolus and study drug infusion bags may be stored up to 48 hours at refrigerated storage conditions (2ºC to 8ºC) and at room temperature (25 ºC) for 24 hours. (Appendices A and C).

Terlipressin is stable at 2ºC to 8ºC up to 48 hours in 0.9% sodium chloride solution at a concentration of 1mg Terlipressin in 5mL solution (Appendix A).

Microbiology studies at a concentration of 2 mg Terlipressin in 50 mL of 0.9% sodium chloride solution are included in Appendix C. Stability at a concentration of 0.034 mg/mL has been previously demonstrated (Bui 2020). Study drug assignment, preparation, and dosing will be strictly limited to the investigators and their designees. Study doses will be labelled to ensure drug is used only for authorized investigational use only. The subject's initials will be placed on the label by the Pharmacists at the time of dispensing.

8.3 Reconstitution

For the first dose of terlipressin, each vial will be reconstituted with 5 mL of sterile 0.9% sodium chloride solution and administered intravenously as a bolus injection and given over 1 minute at a dose of 0.5 mg.

For continuous infusion, the dose of terlipressin is to be dissolved in 0.9% sodium chloride solution and infused with a pump (Appendix B).

8.4 Administration

Subjects in the Prospective Terlipressin Group:

Single-use, sterile 6-mL vials containing 1 mg of lyophilized terlipressin acetate (equivalent to 0.85 mg terlipressin free base) with 10 mg mannitol as a bulking agent/stabilizer.

- For initial bolus dose of 0.5mg, the vial will be reconstituted with 5 mL of sterile 0.9% sodium chloride solution and given over 1-minute push.
- Continued dosing will be by 2mg infusion per day for Days 1, and 2. For continuous infusion, the dose of terlipressin is to be dissolved in 0.9% sodium chloride solution and infused with a pump.
- If on Day 3 the SCr is not 30% lower from the baseline value, increase infusion dose to 4 mg per day.
- If on Day 5 the SCr is not 50% lower from the baseline value, increase infusion dose to 6-8 mg per day, per PI discretion. Dosing can be increased by more than 1 mg increment at Day 5 or beyond, up to a maximum of 8 mg/day.
- If the subject has infusion stopped for ≥ 4 hours, a 0.5 mg bolus should be given prior to restarting infusion.
- The drug should not be used in subjects with severe coronary artery disease or in the setting of circulatory overload, pulmonary edema, or bronchospasm as determined by the Investigator. If drug therapy has been initiated, Investigator can also decide not to increase the dose, based on the patient's status with regard to the above, based on individual subject condition.

- If dosing is interrupted due to an adverse event, terlipressin may be re-started, at the discretion of the investigator, at the same or lower dose as per protocol. Terlipressin will not be restarted if dosing was interrupted due to cardiac ischemia or mesenteric ischemia.

Subjects in the Retrospective Control Group:

The dose and method of administration for Midodrine/Octreotide (M&O), albumin, or other vasopressor will be recorded as noted in the patient's medical record. To be included as a completed subject in this study, patients must have been treated for at least 48 hours with albumin infusions beyond volume resuscitation and diuretic withdrawal. Vasopressors also need to be used for a minimum of 48 hours.

8.5 Study Drug Accountability

The investigator or designee must maintain an inventory record of study drug administered to assure the regulatory authorities and the sponsor that the investigational new drug will not be dispensed to any person who is not a subject under the terms and conditions set forth in this protocol. Neither the investigators nor any designees may provide study drug to any subject not formally enrolled in this protocol.

The study drug supplied for use in this study is to be prescribed only by the investigator or properly assigned designee and may not be used for any purpose other than that outlined in this protocol.

All unused investigational product will be handled per study drug supplier instructions at conclusion of the study. Periodic review of drug accountability records will be conducted by investigational site monitors. Final review and reconciliation of the accountability records will be performed by the sponsor or designee.

9 Assessments and Procedures

9.1 Schedule of Assessments and Procedures

Table 1 presents the schedule of assessments and procedures to be performed during the study. Assessments are completed per sites' Standard of Care.

Table 1 Schedule of Assessments

Study Assessment[k]	Screening Period	Pretreatment Period	Active Study Period								Follow-up Period (Days from Last Dose)					
		Baseline Assessment	Days 1 to 14[a] (as applicable)							End of Treatment	7 ±2 days	14 ±3 days	30 ±7 days	90 ±7 days	180 ±14 days	365 ±14 days
			1	2	3	4	5	6	7	8-14						
Diagnosis of HRS - AKI	X															
Informed consent	X															
Inclusion exclusion criteria	X	X														
Demographics	X															
Height, body weight, estimated dry weight[p]	X															
Pregnancy test[b]	X															
Prior medications	X	X														
Medical history	X	X														
Physical examination[d]		X														
12-lead ECG[e]	X	X														
Echocardiogram[n]		X														
Child-Pugh score		X														
Study drug dosing			X	X	X	X	X	X	X	X						

Study Assessment[k]	Screening Period	Pretreatment Period	Active Study Period									Follow-up Period (Days from Last Dose)					
		Baseline Assessment	Days 1 to 14[a] (as applicable)								End of Treatment	7 ±2 days	14 ±3 days	30 ±7 days	90 ±7 days	180 ±14 days	365 ±14 days
			1	2	3	4	5	6	7	8-14							
Biomarker Blood and Urine Sample[l]		X				X					X						
Cystatin c[m]		X				X					X						
Assessments																	
albumin dose if administered	X	X	X	X	X	X	X	X	X	X	X						
SCr[c] (BUN & eGFR if taken [f)]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
24 hr Urine Volume	X	X	X	X	X	X	X	X	X	X		X					
Vital signs[h]		X	X	X	X	X	X	X	X	X	X						
Encephalopathy score		X	X	X	X	X	X	X	X	X							
Serum electrolytes		X	X	X	X	X	X	X	X	X	X						
ALT, AST, ALP, protein, albumin, bilirubin[o]		X	X		X				X		X	X	X	X	X	X	X
Serum glucose, calcium, magnesium[o]		X	X		X				X		X						
INR		X	X		X				X		X	X	X	X	X	X	X
CBC[o]		X	X		X				X		X						
Concomitant medications[i]			X	X	X	X	X	X	X	X							
Assessments Throughout Study																	
Adverse events[g]			Monitor and record Serious Adverse Events throughout active study period to 30 days post-treatment.														
Record RRT, TIPS, transplant, and mortality[j]			Monitor and record throughout active study period and follow-up.														
Record all paracentesis events			Record all paracentesis performed until drug treatment discontinued														

All assessments are completed per Standard of Care.

[a]Maximum treatment of 14 Days.

[b]Female subjects of child-bearing potential must have a negative urine or serum pregnancy test within 14 days prior to the first dose of study treatment.

[c]The qualifying SCr value (SCr value at least 48 hours after both diuretic withdrawal and the beginning of albumin fluid challenge) is considered the Enrollment SCr and must be drawn prior to start of study drug. The qualifying SCr value must be ≥ 1.5 mg/dL AND Baseline SCr is defined as the most recent, lowest SCr within last 6 months before date of current admission.

[d]A general standard of care physical assessment of the major body systems evaluating any new clinically significant abnormalities within the following: general appearance, skin, head/eyes/ears/nose/throat, neck, heart, lungs, abdomen, extremities, and neurological systems. The investigator may perform additional unscheduled examinations to manage or evaluate a suspected AE as clinically necessary. The physical examination will also include weight and height.

[e]Collection of the data from the last electrocardiogram done prior to admission and the electrocardiogram performed during admission, if performed as per standard of care

[f]Must be performed at least once daily during active treatment AND until Day 14 (regardless of treatment status) or discharge, whichever occurs first. BUN & eGFR are recorded if taken per site standard of care. If assessments are performed more than once daily as part of the subject's medical care, all values are to be recorded on the CRF. If subject has SCr, BUN and/or eGFR, including Vital signs taken at any of the Follow-up Periods, they are to be recorded on the CRF.

[g]Serious Adverse Events will be required to be collected only for the Prospective Terlipressin Group.

[h]Vital sign measurements include body temperature, respiratory rate, systolic and diastolic blood pressure, SpO_2 (pulse oximetric saturation), and heart rate. Vitals will be taken at baseline (within 24 hours of start of study drug) and daily during study drug treatment days. The measurements should be assessed and recorded at a consistent, specific time of day. Preferable morning assessment/measurement.

[i]Concomitant medications include IV solutions and blood products.

[j]All follow-up visits will include assessments of survival, RRT, TIPS and liver transplant status. Study days will be counted from the last day of study drug administration. If the subject is in the hospital during any of the Follow-up Time Periods, the study assessment data will be recorded from the subject's medical record.

[k]All study assessments are to follow SOC

[l] Optional - Collection of Blood and Urine for future biomarker evaluation is optional and may be collected ± 1 day of target collection day

[m]Optional - Cystatin C – assessment of cystatin c levels is optional – but if measured we want it assessed 3 times (at Baseline/pretreatment, on day 4 of treatment and last day of treatment)

[n]Collection of last echocardiogram done prior to admission and any echocardiogram performed during admission, if performed as per standard of care

[o]Neutrophils, lymphocytes, total protein, and magnesium if collected Standard of Care

[p] Estimated Dry Body Weight is collected if calculated by site

Table 2 presents the schedule of assessments and procedures to be performed Post Transplantation.

Table 2 Schedule of Assessments Post Transplantation

Study Assessment[c]	Day of Liver Transplant	Follow-up Period (Days from Day of Liver Transplant						
		7 ±2 days[a]	14 ±3 days[a]	30 ±7 days[a]	90 ±7 days[a]	180 ±14 days[a]	365 ±14 days[a]	Last Known follow-up date[a]
Record events of RRT, TIPS and mortality	X	X	X	X	X	X	X	X
Record all paracentesis events	X	X	X	X	X	X	X	X
SCr (BUN & eGFR if taken[b])	X	X	X	X	X	X	X	X
MELD Score[d]	X							
Dialysis				X	X	X	X	X
Incidence of Kidney Transplant[e]	X	X	X	X	X	X	X	X

[a]All subjects will be contacted for all follow-up periods to assess survival, RRT, TIPS and liver transplant status. Follow-up days will be counted from days after transplantation. If the subject is in the hospital during any of the Follow-up Time Periods, the study assessment data will be recorded from the subject's medical record.

[b]BUN & eGFR are recorded if taken per site standard of care. If assessments are performed more than once daily as part of the subject's medical care, all values are to be recorded on the CRF. If subject has SCr, BUN and/or eGFR, including Vital signs taken at any of the Follow-up Periods, they are to be recorded on the CRF.

[c]All study assessments are to follow SOC.

[d] In addition to the Allocation MELD score (MELD score that qualifies subject for transplant), also record the date and the last calculated MELD score prior to transplant.

10 Study Assessments

10.1 Screening Period

The screening period occurs prior to enrollment and consists of establishing the diagnosis of HRS-AKI as per guidelines and standard medical practice and confirming eligibility for study participation. The investigator or qualified designee will not be required to obtain sponsor approval to enroll subjects. Enrollment of subjects determined not to qualify for the study during sponsor monitoring oversight may result in study site closure. Written informed consent will be obtained by the investigator, sub-investigator, or qualified designee from the subject or legally authorized representative prior to the subject qualification form being completed.

10.2 Pre-Treatment Period

The pre-treatment period occurs prior to administration of study drug and includes performing baseline assessments and collection of prior medication information.

The qualifying SCr value (SCr value at least 48 hours after both diuretic withdrawal and after the beginning of albumin fluid challenge) is considered the enrollment SCr and must be drawn prior to study drug administration.

10.3 Active Study Period (14 Days)

Dosing (Up to 14 Days)

The active study period extends from the initiation of study treatment through Day 14 or discharge from the hospital for any reason, whichever occurs first. Study drug will be administered as described in Section 7.3.2. Concomitant medications will be collected during this active study period. See Table 8.1 for the schedule of assessments during the active study period.

End of Treatment, Discharge or Day 14

Terlipressin may be continued until 24 hours after serum creatinine as reached the lowest value possible as determined by Principal Investigator, up to a maximum of 14 days.

10.4 Follow-up Period (Up to 365 Days after transplantation)

The follow-up period begins after the end of the study treatment and concludes Day 365 after liver transplantation.

10.5 Physical Examination

A physical examination, including weight, estimated dry weight (if calculated as per standard of care), and height, will be conducted at baseline and will follow the standard of care for these subjects.

The investigator may perform additional unscheduled examinations to manage or evaluate a suspected SAE as clinically necessary. The timing and scope of additional unscheduled examinations should be determined by the nature and severity of the SAE being evaluated.

10.6 Electrocardiogram

A 12-lead electrocardiogram (ECG) will be performed at baseline. Clinically relevant abnormalities found will be reported on the medical history page. The investigator may perform additional unscheduled examinations to manage or evaluate a suspected SAE as clinically necessary. The data of all subsequent ECGs will be reported on the ECG/Echocardiogram CRF.

10.7 Echocardiogram

The last echocardiogram done prior to admission will be collected as well as any echocardiogram performed during admission. Clinically relevant abnormalities found will be reported on the medical history page. The data of all subsequent Echocardiograms will be reported on the ECG/Echocardiogram CRF.

10.8 Child-Pugh Scores

Child-Pugh score will be calculated at baseline and entered in the CRF.

11 Detail of Safety Assessments and Procedures

11.1 Vital Signs

Vital sign measurements will be recorded at baseline and daily during study drug administration. The measurements should be assessed and recorded at a consistent, specific time of day, preferable morning assessment/measurement.

Systolic and diastolic blood pressure, heart rate, body temperature, respiratory rate, and pulse oximetric saturation (SpO_2) will be recorded at baseline and during study drug administration.

11.2 Encephalopathy Score

Clinically detectable encephalopathy is to be assessed prior to study drug administration and daily during study drug administration using the clinical criteria described in Table 3.

Table 3 West Haven Criteria for Semiquantitative Grading of Mental State

Grade 1	Trivial lack of awareness. Euphoria or anxiety. Shortened attention span. Impaired performance of addition.
Grade 2	Lethargy or apathy. Minimal disorientation for time or place. Subtle personality change. Inappropriate behavior. Impaired performance of subtraction.
Grade 3	Somnolence to semi-stupor, but responsive to verbal stimuli. Confusion. Gross disorientation.
Grade 4	Coma (unresponsive to verbal or noxious stimuli).

Source: Ferenci et al, 2002.

11.3 Model for End-Stage Liver Disease Score

Model for end-stage liver disease (MELD) scores will be calculated by the sponsor based on SCr, bilirubin and INR values at baseline and treatment termination.

11.4 Adverse Events

Only Serious Adverse Events will be recorded until 30 days after discontinuation of study drug.

Those events that are unanticipated serious adverse events possibly related to the study drug (SUSAR) must be reported to the sponsor or its designee for safety reporting in an expedited. See section 14.2 for SUSAR definition.

12 Detail of Laboratory Assessments and Procedures

12.1 Local Laboratory at Investigational Site

The laboratory tests specified in this section will be performed in the laboratory at the investigational site. All efforts should be made to take the blood sample from a clean new venous site, or the second option could be a central venous line to collect the blood specimen. Site

personnel should not draw the blood sample from an existing peripheral venous access site, or a line used for IV fluid administration.

12.2 Serum Electrolytes & Biochemistry

The following assessments are required at baseline, then once daily during treatment or hospital discharge (whichever occurs first):

- Creatinine level (SCr)
- Blood urea nitrogen (BUN, if taken as per standard of care)
- Estimated Glomerular Filtration Rate (eGFR, if taken as per standard of care)

If SCr, BUN and/or eGFR assessments are performed more than once daily as part of the subject's medical care, all values are to be recorded on the CRF.

SCr (BUN & eGFR if also taken standard of care) are to be collected and recorded at all Follow-up Visits including: Day 7, 14, 30, 90, 180 and 365 as per standard of care.

The following assessments are required at baseline and daily during treatment with study drug:

- Sodium
- Potassium
- Chloride
- HCO_3
- 24 Hour Urine Output (if available)

The following assessments are required at baseline and on treatment days 1, 3, 7, and, whenever possible, at treatment termination:

- Total bilirubin*
- Alkaline phosphatase (ALP)*
- Alanine aminotransferase (ALT)*
- Aspartate aminotransferase (AST)*
- Total protein*
- Albumin*
- Glucose

*Total Bilirubin, ALP, ALT, AST, Total Protein, and Albumin are to be collected and recorded at all Follow-up Visits including: Day 7, 14, 30, 90, 180 and 365 if taken as per standard of care.

The following assessments are optional:

- Collection of blood & urine for future biomarker evaluation - optional at baseline (pretreatment), on treatment day 4, and at treatment termination
- Cystatin C - optional at baseline (pretreatment), treatment day 4, and at treatment termination

12.3 Hematology

The following assessments are required at baseline and on treatment days 1, 3, 7, and, whenever possible, at treatment termination:

- International normalized ratio (INR)
- Complete blood count (CBC) (no differential required, but neutrophils and lymphocytes if available)

INR is to be collected and recorded at all Follow-up Visits including: Day 7, 14, 30, 90, 180 and 365 as per standard of care.

12.4 Total Volume of Blood Collected

The total volume of blood collected from each subject over the course of 14 days and end of study should not exceed approximately 310 mL, and is standard of care for the subject being treated. All blood draws and assessments requested are to follow standard of care procedures.

13 Detail of Efficacy Assessments and Procedures

Serum creatinine must be collected at enrollment (prior to study dosing), and at least once daily during treatment until Day 14 or hospital discharge, whichever occurs first. If SCr assessments are performed more than once daily as part of the subject's medical care, all values obtained each day will be recorded on the CRF.

13.1 Primary Efficacy Variable

Improvement of renal function (SCr) from Day 1 through end of treatment, repeated measure analysis. SCr will be collected at baseline and daily from Day 1 through end of treatment.

13.2 Secondary Efficacy Variables

- Improvement of renal function (SCr) from Day 1 through end of treatment, repeated measure analysis as compared to the Retrospective Control Group.

- For the Prospective Terlipressin Group alone and also in comparison to the Retrospective Control Group:
    - Incidence of dialysis by Day 30 and Day 90 of follow up.
    - Survival by Day 30 and Day 90 follow up.
    - Transplant by Day 30 and Day 90 follow up.
    - Serum creatinine at Day 30 and Day 90 follow up
    - Serum creatinine and incidence of dialysis at Day 30, Day 90, Day 180, and Day 365 after liver transplantation
    - Incidence of kidney transplant within one year after liver transplant
    - Descriptive analysis of number simultaneous liver kidney transplants and episodes of graft rejection at Day 90 and Day 365 after liver transplant

14 Serious Adverse Events

Only Serious Adverse Events will be collected for this study for the prospective group only. Safety review and assessments will be made only for the Prospective Terlipressin Group from time of first dose through 30 days after end of treatment. SAEs are collected from the start of study drug until 30 days after study drug was discontinued. Non-serious Adverse Events are not required to be collected for this study.

The investigator will record the nature, causality, severity, treatment, and outcome of the SAE, and will determine whether there is a "reasonable probability" that the study drug treatment caused the event.

14.1 SAE Definitions

The following definitions are based on:

- International Conference on Harmonization (ICH) E2A: International Conference on Harmonisation Tripartite Guideline: Clinical Safety Data Management: Definitions and Standards for Expedited Reporting (1994).
- ICH E6: International Conference on Harmonisation Tripartite Guideline: Guideline for Good Clinical Practice (1996) and Integrated Addendum [E6] (R2) June 2015.

- Office of Human Subjects Protection, Division of Department of Health and Human Services Policy; "Reviewing and Reporting Unanticipated Problems Involving Risks to Subjects or Others and Adverse Events" (2007).
- Code of Federal Regulations Title 21, Part 312.32 (2011).

Also included are events that are common to this subject population.

14.2 Suspected Unexpected Serious Adverse Reaction (SUSAR)

This is an unexpected SAE or suspected unexpected adverse reaction (SUSAR) which is not listed in the Investigator's Brochure (IB), nor listed at the specificity or severity observed. If an SUSAR is listed in the IB as occurring with a specific class of drugs, or occurring as part of a disease process, or from the pharmacological properties of the drug, but not specifically mentioned as occurring with the particular drug under investigation, it should be considered unexpected.

14.3 Serious Adverse Event Definition

Serious adverse events (SAE) are defined (in the view of the investigator or the sponsor) as any untoward medical occurrence that meets any of the following criteria:

- Resulted in death.
- Is life-threatening (if an AE or SUSAR places a subject at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it were more severe).
- Requires inpatient hospitalization or prolongation of existing hospitalization.
- Results in disability (persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions).
- Is a congenital anomaly/birth defect.
- Is an important medical event that may not result in death, be life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. Examples of an important medical event include allergic bronchospasm, blood dyscrasias, convulsions that do not result in inpatient hospitalization, or drug dependence/abuse.

14.4 Severity of an Adverse Event

Intensity of an AE, assessed by the investigator and graded as mild, moderate, or severe, irrespective of the relationship to the drug or seriousness of the event, and evaluated according to the following scales:

- Mild - awareness of the symptom but easily tolerated.
- Moderate - discomfort enough to interfere with normal activities.
- Severe - Incapacitating with the inability to perform normal activities.

14.5 Study Drug Causality

Study drug causality will be assessed as follows:

Unrelated: An AE that is clearly and incontrovertibly due to extraneous causes (disease, environment, etc.) and does not meet the criteria for 'possible' or 'probable.'

Possible: The connection between the AE and study treatment appears unlikely but cannot be ruled out with certainty. An AE may be considered 'possibly related' if it has at least 2 of the following:

- It follows a reasonable temporal sequence from administration of study drug.
- It may readily have been produced by the subject's clinical state or by environmental or toxic factors.
- It follows a known response pattern to study drug.

Probable: An AE that is considered to be related to study drug treatment with a high degree of certainty. An AE may be considered probably related if:

- It follows a reasonable temporal sequence from administration of study drug.
- It cannot be reasonably explained by the known characteristics of the subject's clinical state.
- It follows a known pattern of response to study drug treatment.
- It reappears upon rechallenge.

Table 4 lists adverse events that are common in this subject population (i.e., with HRS), even in the absence of exposure to the study drug (Rozov-Ung and Panesar, 2010; Arroyo et al, 2008).The investigator may use this as a guidance while assessing the causality.

Table 4: Adverse Events Common to the Hepatorenal Syndrome (HRS) Population

MedDRA System Organ Class	Preferred Term
Blood and Lymphatic System Disorders	Anemia Coagulopathy
Cardiac Disorders	Atrial fibrillation Bradycardia
Gastrointestinal Disorders	Abdominal pain Diarrhea Nausea Vomiting
General Disorders and Administration Site Conditions	Multi-organ failure
Hepatobiliary Disorders	Hepatic failure
Infections and Infestations	Pneumonia Urinary tract infection
Metabolism and Nutrition Disorders	Hyperglycemia
Nervous System Disorders	Headache
Respiratory, Thoracic and Mediastinal Disorders	Dyspnea Epistaxis Pulmonary edema Respiratory failure
Vascular Disorders	Hypotension

Source: Rozov-Ung and Panesar, 2010; Arroyo et al, 2008.

14.6 Overdose

Overdose is defined as an accidental or intentional/infusion or excessive dose of a product.

14.7 Collection of Adverse Events/Serious or Unexpected

Any adverse event occurring prior to signing the ICF and prior to 1st dose of study drug should be considered medical history or a pre-existing condition and will be collected on the Medical History CRF.

SAEs will be assessed and recorded beginning with study drug administration until 30 days after discontinuation of study drug.

14.8 Laboratory Abnormalities

The investigator will review clinical laboratory test results in a timely fashion. Laboratory abnormalities deemed clinically significant and serious should be reported to the sponsor as appropriate.

14.9 Hospitalizations

Planned hospital admissions and/or planned surgical operations for an illness or disease, which existed before the subject was randomized are not to be considered SAEs (e.g., re-hospitalization for liver transplantation). However, a baseline condition which deteriorates during the clinical study may be considered an SAE.

Following the initial hospital discharge, all re-hospitalizations within 30 days post end of treatment (except for planned hospital admissions or procedures as described above) are to be recorded as a SAE.

14.10 Complications of the Disease

Complications frequently associated with HRS - AKI are expected to occur, but should be recorded as a SAE, as applicable, e.g., EVH, hepatic encephalopathy, sepsis, pneumonia, urinary tract infection.

Since HRS - AKI is the indication under investigation, worsening or aggravation of HRS - AKI during the active treatment period (including non-response) will NOT be recorded as a SAE unless HRS – AKI is the cause of death or results in a subsequent re-hospitalization due to suspected recurrence.

14.11 Pregnancy

Pregnancy occurring in a subject is NOT considered an SAE. However, the investigator must collect pregnancy information for subject and/or subject's partner if the fetus was exposed to the study drug.

14.12 Reporting Serious or Unexpected Adverse Reactions or Adverse Events

Site Reporting to Sponsor

Investigators must report all SAEs to the sponsor (and applicable IRB) whether or not the events are study drug-related or expected.

All SAEs must be reported on a SAE form and faxed/scanned to International HealthCare (IHC) within **24 hours** of an investigator becoming aware of the event:

IHC Safety Review Officer
Email: infusealerts@ihcresearch.com

The initial SAE form does not need to be signed by the investigator; however, a signed SAE form is required for final SAE reporting. The initial scan by the investigator will include (whenever possible) a detailed description of the event including start and stop dates, causality assessment by the investigator, and supported as needed with written copies of medical records, autopsy reports, and other appropriate documents. Follow-up information (including information requested by the sponsor) should be reported by fax/scan within 24 hours of availability. The investigator is expected to make every effort to collect follow up information for each SAE in a timely fashion after the initial reporting of the SAE.

If there are any specific questions regarding a SAE classification, the investigator should contact the lead sponsor Principal Investigator for this study.

If there are any specific questions regarding the completion of the SAE form, please contact your site monitor.

Site Reporting to Institutional Review Board/Independent Ethics Committee

The investigator must notify the local IRB which approved the trial of the event in accordance with applicable guidelines.

Sponsor Reporting to Regulatory Authorities and Investigators

All reportable SAEs, including narrative safety reports of overall findings or data from aggregate analyses, will be reported to the applicable regulatory authorities by the sponsor or designee in accordance with the local regulations as described in the safety review plan for this study.
The reporting of SAEs will be conducted in accordance with ICH E2A (Clinical Safety Data Management: Definitions and Standards for expedited reporting) and regulatory guidelines.

In addition, study endpoints described in Section 5.1 of the protocol that are serious adverse events will only be reported in the final clinical study report unless the event was unusual,

unexpected, and there is evidence suggesting a causal relationship between the study drug and the event (e.g., death from anaphylaxis).

The sponsor will notify all participating Investigators of safety reports, in a blinded fashion, of potential serious risks as described in the safety review plan for this study.

15 Statistical Plan

Baseline characteristics, such as patient demographics, disease etiology, disease severity and factors present at time of listing for liver transplantation, will be compared between the terlipressin treatment group and the retrospective control group. Categorical variables will be presented as proportions and percentages and compared using chi-squared or Fisher's exact testing. Continuous variables will be presented as means and standard deviations or medians and interquartile ranges. Depending on the distribution of the variable, the Student's t-test or Wilcoxon rank sum will be used for comparison.

15.1 Primary Analysis

The primary analysis will utilize mixed effects regression modeling to estimate changes in serum creatinine (SCr) over the study period, accounting for baseline SCr and the correlation structure between the repeated measures. All potential confounders will be added to the model and sequentially removed if they do not confound the association between receipt of terlipressin and improvement in Scr. Those confounders resulting in a $\geq$ 10% change in the coefficient for the association between terlipressin and improvement in SCr will be retained in the final model. For all generated models, Aikaike Information Criteria (AIC) scores will be calculated and compared. The model with the smallest absolute AIC value will be accepted as the most parsimonious and will be used as the primary model for the explanation of study results. As has been undertaken in other terlipressin studies in HRS, a least-squares means analysis will be conducted using a repeated-measures analysis of variance. The results from this approach to the statistical analysis can then be directly compared to prior study results.

15.2 Secondary Analyses

A number of secondary outcomes will be examined for the study cohort. These outcomes include the incidence of dialysis, mortality, and liver transplantation at day 30 and 90. The

incidence of kidney transplant within one year of liver transplant will also be determined. Additionally, descriptive comparisons of SCr will be undertaken at day 30 and 90 of follow up and at 4 time points after receipt of liver transplantation (day 30, 90, 180 and 365) and also at similar time points for those patients who were transplant ineligible or did not undergo a transplant. Incidence will be calculated as the number of new events during the observation period divided by the total persons at risk. All statistical analyses will be conducted using Stata version 15.1 statistical software (StataCorp, College Park, TX). Statistical significance will be declared for a two-sided hypothesis test if the *P* value was <0.05.

15.3 Sample Size Considerations

A formal sample size calculation is not applicable for the proposed study. However, at least 50 subjects across more than 5-10 institutions may receive terlipressin treatment during the course of the intervention portion of the study. For the comparison of the terlipressin treatment group and retrospective controls, we expect to have 50 subjects in the intervention group and 150 subjects, assembled from the study sites, in the control group. Assuming a type 1 error of 0.05 and power of 80%, we expect to be able to detect a range of differences in serum creatinine (SCr) depending on the standard deviation of the change (primary endpoint) expected for the groups (see below). Based on results from the REVERSE trial, a change in SCr of as small as 0.05 mg/dL would be able to be detected if the standard deviation for the change approximated 0.10 mg/dL and a pre-specified sample size of 200 subjects, 50 of whom are in the intervention arm, is used for the calculation.

	Δ SCr mg/dL	Δ SCr mg/dL	Δ SCr mg/dL	Δ SCr mg/dL	Δ SCr mg/dL
Detectable Difference	0.46	0.34	0.23	0.11	0.05
Standard Deviation	1.00	0.75	0.50	0.20	0.10

Given the relatively small number of subjects proposed for inclusion in this terlipressin study, we will likely by underpowered to detect a difference in the other secondary outcomes being explored.

15.4 Safety Variables

Safety variables will include the following:

- Serious adverse events and all deaths. Safety details may include:
    - Vital signs (body temperature, blood pressure, heart rate, respiratory rate, and SpO_2).
    - MELD score.
    - Encephalopathy score.
    - Laboratory parameters.

Serious Adverse events will be not classified by Medical Dictionary for Drug Regulatory Activities (MedDRA) system organ class, preferred term, severity, and seriousness. SAEs will be summarized using site reported terminology, as well as by the investigator's assessment of the relationship of the SAE to the study drug.

15.5 Subject Accounting and Baseline Characteristics

A summary of the study completion status and reasons for discontinuation will be provided for both the Prospective Terlipressin Group and the Retrospective Control Group. Baseline characteristics (age, gender, race, etc.) will be summarized. The number of subjects with medical conditions within each body system will be reported. Summaries of the extent of exposure and protocol deviations will also be provided.

15.6 Safety Population

The safety population is defined as all subjects who received at least 1 dose of study drug.

16 Direct Access, Data Handling, and Record Keeping

16.1 Investigator

The investigator will permit study-related monitoring, audits, IRB review, and regulatory inspections by providing direct access to original source data and documents.

All subject information will be recorded on source documents. The CRFs must be fully completed and include all required data for all subjects enrolled. All CRF data must be submitted to the sponsor throughout and at the end of the study.

If an investigator retires, relocates, or otherwise withdraws from conducting the study, the investigator must notify the sponsor to agree upon an acceptable storage solution. Regulatory agencies will be notified with the appropriate documentation.

Any significant changes in study personnel will require an updated Statement of Investigator (e.g., FDA Form 1572) to be filed with the sponsor.

The investigator must notify their IRB of protocol violations in accordance with local regulatory and IRB requirements.

16.2 Sponsor

The CRF data are stored in a database and processed electronically. The data are reviewed for completeness, and logical consistency. Requests for data clarification are forwarded to the study site for resolution.

16.3 Records Retention

The investigator shall retain and preserve 1 copy of all data collected or databases generated in the course of the study, specifically including but not limited to those defined by Good Clinical Practice (GCP) as essential. Essential documents should be retained until at least 2 years after the last approval of a marketing application in an ICH region and until there are no pending or contemplated marketing applications in an ICH region or at least 2 years have elapsed since the formal discontinuation of clinical development of the investigational medicinal product.
These documents should be retained for a longer period, however, if required by the applicable regulatory requirements or by an agreement with the sponsor. It is the responsibility of the sponsor to inform the investigator/institution as to when these documents no longer need to be retained. Prior to destruction of any study essential documents, the investigator must first obtain written approval from the sponsor.

17 Quality Control and Quality Assurance

17.1 Study Monitoring

The sponsor has ethical, legal, and scientific obligations to carefully follow this study in a detailed and orderly manner in accordance with established research principles and applicable regulations. The investigator, as part of his responsibilities, is expected to cooperate with the sponsor, or their designee International HealthCare, in ensuring that the study adheres to the protocol and GCP requirements.

As part of a concerted effort to fulfill these obligations (maintain current personal knowledge of the progress of the study), the sponsor's monitor will visit the center(s) during the study in accordance with the monitoring plan set forth for this study as well as maintain frequent telephone and written communication. The investigator will permit the sponsor to monitor the study as frequently as is deemed necessary and provide access to medical records to ensure that data are being recorded adequately, that data are verifiable, and that protocol adherence is satisfactory.

17.2 Auditing

The sponsor may conduct audits at the study center(s). Audits will include, but not be limited to, drug supply, presence of required documents, the informed consent process, and comparison of case report forms with source documents. The investigator agrees to participate with audits conducted at a reasonable time in a reasonable manner.

Regulatory authorities worldwide may also audit the investigator during or after the study. The investigator should contact the sponsor immediately if this occurs and must fully cooperate with regulatory authority audits conducted at a reasonable time in a reasonable manner.

18 Ethics and Responsibility

This study will be conducted in compliance with the protocol, with the sponsor’s standard operating procedures and/or guidelines, with the FDA or local regulatory regulations, with the ICH GCP guidelines, and with the Declaration of Helsinki.

18.1 Institutional Review Board/Independent Ethics Committee

This protocol and the written informed consent form shall be submitted to the IRB identified with this responsibility at the research facility. Notification in writing of approval must come from the IRB chairman or secretary, to the investigator, either as a letter or as a copy of the appropriate section of the IRB meeting minutes where this protocol and associated informed consent form were discussed. The investigator will not participate in the decision. If the investigator is an IRB member, the written approval must indicate such non-participation.
The investigator will submit status reports to the IRB at least annually (when applicable).
The IRB must be notified by the investigator in writing of the interruption and/or completion of the study; the investigator must promptly report to the IRB all changes in research (protocol amendments) and will not make such changes without IRB approval except where necessary to eliminate apparent immediate hazards to human subjects. In these cases, the IRB must be

notified within 5 days of the change. The investigator will promptly report to the IRB all unanticipated problems involving risk to subjects or others. The investigator is required to maintain an accurate and complete record of all written correspondence to and received from the IRB and must agree to share all such documents and reports with the sponsor.

18.2 Protocol Amendments

Any change in the study plan requires a protocol amendment. An investigator must not make any changes to the study without IRB and sponsor approval except when necessary to eliminate apparent immediate hazards to the subjects. A protocol change intended to eliminate an apparent immediate hazard to subjects may be implemented immediately, but the change must then be documented in an amendment, reported to the IRB within 5 working days, and submitted to the appropriate regulatory agency in the required time frame.

18.3 Informed Consent

All subjects will have a signed document allowing the use of their medical data. For the Prospective Terlipressin Group, written informed consent will be obtained from all subjects (or their guardian or legal representative) before any study-related procedures (including any pre-treatment study-specific procedures) are performed. The investigator(s) has both ethical and legal responsibility to ensure that each subject being considered for inclusion in this study is given a full explanation of the protocol. This shall be documented on a written informed consent form, which shall be approved by the same IRB responsible for approval of this protocol. Each informed consent form shall include the elements required by regulatory regulations in 21 Code of Federal Regulations (CFR) Part 50 and ICH Guidance E6, Section 4.8. The investigator agrees to obtain approval from the sponsor of any written informed consent form used in the study, preferably prior to submission to the IRB.

Once the appropriate essential information has been provided to the subject and fully explained by the investigators (or a qualified designee) and it is felt that the subject understands the implications of participating, the subject and the investigator (or designee) shall sign the IRB-approved written informed consent form. The subject shall be given a copy of the signed informed consent form, and the original shall be kept in the site's regulatory file. A second copy may be filed in the subject's medical record, if allowed by the institution.

18.4 Confidentiality

All information generated in this study will be considered confidential and will not be disclosed to any persons not directly concerned with the study without written prior permission from the

sponsor. However, authorized regulatory officials and sponsor personnel will be allowed full access to the records. All medications provided and subject bodily fluids and/or other materials collected shall be used solely in accordance with this protocol, unless otherwise agreed to in writing by the sponsor.

Only initials and/or unique subject numbers in case report forms will identify subjects. Their full names may, however, be made known to a product regulatory agency or other authorized official if necessary.

18.5 Subject Injury

Subject injury provisions will be specified in the site's Clinical Trial Agreement.

18.6 Study Suspension, Termination, and Completion

The sponsor or designee may suspend or terminate the study or part of the study at any time for any reason.

If the investigator suspends or terminates the study, the investigator will promptly inform the sponsor or designee and the IRB and provide them with a detailed written explanation. The investigator will destroy all unused (after final drug accountability has been performed) and partially used investigational product per site standard operating procedure. Upon study completion, the investigator will provide the sponsor or designee, IRB, and regulatory agency with final reports and summaries as required by regulations. For IND application studies, or when the data will be used in support of an IND, the investigator must submit a written report to the sponsor or designee and the IRB within 3 months after the completion or termination of the study. Study termination and follow-up will be performed in compliance with applicable standard operating procedures.

18.7 Study and Study Site Discontinuation Criteria

The sponsor, investigator, medical monitor, or regulatory officials may discover conditions during the study that indicate that the study or study site should be terminated. This action may be taken after appropriate consultation between the sponsor and investigator. Conditions that may warrant termination of the study/study site include, but are not limited to:

- The discovery of an unexpected, serious, or unacceptable risk to the subjects enrolled in the study.

- The decision on the part of the sponsor to suspend or discontinue testing, evaluation, or development of the study drug.
- Failure of the investigator to enroll subjects into the study at an acceptable rate.
- Failure of the investigator to comply with pertinent regulatory regulations.
- Submission of knowingly false information from the study site to the sponsor, study monitor, or regulatory authority.
- Insufficient adherence to protocol requirements
- Study/study site termination and follow-up will be performed in compliance with the sponsors' standard operating procedures.

19 Registration of Study and Publication of Data

This study will be registered at the publicly accessible Web site www.clinicaltrials.gov. Registration at other publicly accessible registries will be performed as required.

The results of this study will be published and/or presented at scientific meetings in accordance with usual and customary academic, editorial, and ethical practices and requirements.

19.1 Investigator's Ability to Publish

Terms and provisions of publication rights are governed by the Publication Section in the clinical study agreement.

20 References

Al-Khafaji AH, Sharma S, Eschun G. Multiple organ dysfunction syndrome in sepsis. Medscape. March 2015 (http://emedicine.medscape.com/article/169640-overview).

Angeli P, Ginès P, Wong F, et al. Diagnosis and management of acute kidney injury in patients with cirrhosis: Revised consensus recommendations of the International Club of Ascites. J Hepatol. 2015 Apr;62(4):968-74. Epub 2015 Jan 28.

Angeli P, Volpin R, Gerunda G, et al. Reversal of type 1 hepatorenal syndrome with the administration of midodrine and octreotide. Hepatology. 1999;29(6):1690-1697.

Arroyo V, Ginès P, Gerbes AL, et al. Definition and diagnostic criteria of refractory ascites and hepatorenal syndrome in cirrhosis. International Ascites Club. Hepatology. 1996 Jan;23(1):164 76.

Arroyo V, Jiménez W. Complications of cirrhosis. II. Renal and circulatory dysfunction. Lights and shadows in an important clinical problem. J Hepatol. 2000;32(1 Suppl):157-70.

Arroyo V, Fernandez J, Gines P. Pathogenesis and Treatment of Hepatorenal Syndrome. Abstract. Seminars in Liver Disease. Vol. 29, 2008: 81-95.

Arroyo V, Moreau R, Jalan R, et al, on behalf of the investigators of the EASL-CLIF Consortium CANONIC Study. Acute-on-chronic liver failure: A new syndrome that will re-classify cirrhosis. J Hepatol. 2015;62:S131–S143.

Bui TNN, Sandar S, Luna G, et al. An investigation of reconstituted terlipressin infusion stability for use in hepatorenal syndrome. Sci Rep. 2020; 10, 21037.

Campbell MS, Kotlyar DS, Brensinger CM, et al. Renal function after orthotopic liver transplantation is predicted by duration of pretransplantation creatinine elevation. Liver Transplantation. 2005;11(9):1048-1055.

Cavallin M, Kamath PS, Merli M, et al. Terlipressin plus albumin versus midodrine and octreotide plus albumin in the treatment of hepatorenal syndrome: a randomized trial. Hepatology. 2015 Aug;62(2):567-74.

Cavallin M, Piano S, Romano A, et al. Terlipressin given by continuous intravenous infusion versus intravenous boluses in the treatment of hepatorenal syndrome: A randomized controlled study. Hepatology. 2016;63(3):983-992.

Centers for Disease Control and Prevention, National Center for Health Statistics, Death: Final data for 2013, http://www.cdc.gov/nchs/fastats/liver-disease.htm, last updated 6 February 2015.

Chalasani NP, Hayashi PH, Bonkovsky HL, et al, on behalf of the Practice Parameters Committee of the American College of Gastroenterology. ACG Clinical Guideline: The diagnosis and management of idiosyncratic drug-induced liver injury. Am J Gastroenterol. 2014;109:950-966.

Choi CH, Ahn SH, Kim DY, Lee SK, Park JY, Chon CY, Moon YM, Han KH. Long-term clinical outcome of large volume paracentesis with intravenous albumin in patients with spontaneous bacterial peritonitis: a randomized prospective study. J Gastroenterol Hepatol. 2005 Aug;20(8):1215-22.

Cruz DN. Midodrine: a selective α-adrenergic agonist for orthostatic hypotension and dialysis hypotension. Expert Opinion on Pharmacotherapy. 2000;1(4):835-840.

de Franchis R. Evolving consensus in portal hypertension. Report of the Baveno IV consensus workshop on methodology of diagnosis and therapy in portal hypertension. J Hepatol. 2005 Jul;43(1):167-76. Corrigendum in J Hepatol. 2005 Sep;43(3):547.

Esrailian E, Pantangco ER, Kyulo NL, Hu K-Q, Runyon BA. Octreotide/Midodrine Therapy Significantly Improves Renal Function and 30-Day Survival in Patients with Type 1 Hepatorenal Syndrome. Digestive Diseases and Sciences. 2007;52(3):742-748.

European Association for the Study of the Liver. EASL clinical practice guidelines on the management of ascites, spontaneous bacterial peritonitis, and hepatorenal syndrome in cirrhosis. J Hepatol. 2010 Sep;53(3):397-417.

Ferenci P, Lockwood A,Mullen K, Weissenborn K, Blei A. Hepatic encephalopathy-definition, nomenclature, diagnosis, and quantification: final report of the working party at the 11th World Congresses of Gastroenterology, Vienna, 1998. Hepatology. 2002 Mar;35(3):716-21.

Ginés P, Arroyo V, Quintero E, Planas R, Bory F, Cabrera J, Rimola A, Viver J, Camps J, Jiménez W, et al. Comparison of paracentesis and diuretics in the treatment of cirrhotics with tense ascites. Results of a randomized study. Gastroenterology. 1987 Aug;93(2):234-41.

Ginès P, Guevara M, Arroyo V, Rodés J. Hepatorenal syndrome. Lancet. 2003 Nov 29;362(9398):1819-27.

Gold Standard, Inc. Octreotide. Drug Monograph in ClinicalKey. https://www.clinicalkey.com/#!/content/drug_monograph/6-s2.0-449. Accessed October 7, 2019.

Hochberg Y. A sharper Bonferroni procedure for multiple tests of significance. Biometrika. 1988;75:800-802.

Ioannou GN, Doust J, Rockey DC. Systematic review: terlipressin in acute oesophageal variceal haemorrhage. Aliment Pharmacol Ther. 2003 Jan;17(1):53-64.

Izzy M, VanWagner LB, Lin G, et al. Redefining Cirrhotic Cardiomyopathy for the Modern Era. Hepatology. 2020;71(1):334-345. doi:10.1002/hep.30875Jalan R, Saliba F, Pavesi M, et al, on behalf of the CANONIC study investigators of the EASL-CLIF Consortium. Development and validation of prognostic score to predict mortality in patients with acute-on-chronic liver failure. J Hepatol. 2014;61:1038-1047.

Kao HW, Rakov NE, Savage E, Reynolds TB. The effect of large volume paracentesis on plasma volume - a cause of hypovolemia? Hepatology. 1985 May-Jun;5(3):403-7.

Kiszka-Kanowitz M, Henriksen JH, Hansen EF, Møller S, Bendtsen F. Effect of terlipressin on blood volume distribution in patients with cirrhosis. Scand J Gastroenterol. 2004 May;39(5):486-92.

Lang RM, Badano LP, Mor-Avi V, Afilalo J, Armstrong A, Ernande L, et al. Recommendations for cardiac chamber quantification by echocardiography in adults: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. J Am Soc Echocardiogr 2015;28:233-270

Ma J, Ward E, Siegel R, Jemal A. Temporal trends in mortality in the United States, 1969-2013, JAMA 2015; 314:1731-9.

Marrero JA. Hepatorenal Syndrome. NORD Guide to Rare Disorders. Philadelphia, PA: Lippincott Williams & Wilkins; 2003:344-345.

Muir AJ. Medical Encyclopedia: Hepatorenal syndrome. Medline Plus. Reprinted from: www.nlm.nih.gov/medlineplus/print/ency/article/000489.htm. April 25, 2002.

Murkerjee S, Roy H, Zetterman RK. Hepatorenal Syndrome. eMedicine. Reprinted from: www.emedicine.com/med/topic/1001.htm. July 12, 2002.

National Hospital Discharge Survey 2005. Annual Summary with Detailed Diagnosis and Procedure Data. Vital and Health Statistics; Series 13, Number 165. Table 45, Number of first-listed diagnoses from short-stay hospitals, page 69 (ICD-9 code 571, cirrhosis); Table 47, Number of all-listed discharges from short-stay hospitals, page 138 (ICD-9 code 572.4, HRS). December 2007.

Nagueh SF, Smiseth OA, Appleton CP, Byrd BF 3rd, Dokainish H, Edvardsen T, et al. Recommendations for the evaluation of left ventricular diastolic function by echocardiography: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. Eur Heart J Cardiovasc Imaging 2016;17:1321-1360Pinto PC, Amerian J, Reynolds TB. Large-volume paracentesis in nonedematous patients with tense ascites: its effect on intravascular volume. Hepatology. 1988 Mar-Apr;8(2):207-10.

Rodríguez E, Elia C, Solà E, et al. Terlipressin and albumin for type-1 hepatorenal syndrome associated with sepsis. J Hepatol. 2014;60: 955-61.

Rozov-Ung I, Panesar M. The Use of Terlipressin in Hepatorenal Syndrome. Dialysis and Transplantation. 2010 Oct;421-25.

Runyon BA; AASLD. Introduction to the revised American Association for the Study of Liver Diseases Practice Guideline management of adult patients with ascites due to cirrhosis 2012. Hepatology. 2013 Apr;57(4):1651-3.

Salerno F, Gerbes A, Ginès P, Wong F, Arroyo V. Diagnosis, prevention and treatment of hepatorenal syndrome in cirrhosis. A consensus workshop of The International Ascites Club. Gut. 2007 Sep;56(9):1310-18.

Titó L, Ginès P, Arroyo V, Planas R, Panés J, Rimola A, et al. Total paracentesis associated with intravenous albumin management of patients with cirrhosis and ascites. Gastroenterology. 1990 Jan;98(1):146-51.

Wong F, Leung W, Al Beshir M, Marquez M, Renner EL. Outcomes of patients with cirrhosis and hepatorenal syndrome type 1 treated with liver transplantation. Liver Transplantation. 2015;21(3):300-307.

Wong F, Pantea L, Sniderman K. Midodrine, octreotide, albumin, and TIPS in selected patients with cirrhosis and type 1 hepatorenal syndrome. Hepatology. 2004;40(1):55-64.

21 Appendix A: Terlipressin Stability Data:

Table 1: In-Use Stability Study Design of Terlipressin for Injection

Batch Tested	Lot No. 300953F	
Manufacturing Date	16 June 2013	
Study Start Date	January 2016	
Storage Conditions	2-8°C	
	25°C – Light Exposed (light level held at 1000 – 1200 Lux)	
	25°C – Dark (Control, foil wrapped in the light chamber)	
Study Intervals	Initial, 12 hours, 24 hours, 48 hours, 72 hours	
Test Parameters	Completeness and Clarity of Solution	The constituted solution is not significantly less clear than an equal amount of the diluent
	Assay	90.0 – 110.0%
	Related Substances	Lypressin: NMT 0.5% Unknown A: NMT 1.0% Acetyl-terlipressin: NMT 0.7% Individual unspecified: NMT 0.5% Total Impurities: NMT 5.0%
	pH	4.3 – 7.5
	Reconstitution Time	Dissolves completely within 30 sec, leaving no visible residue

Table 2: In-Use Chemical Stability Data after reconstitution with 0.9% sodium chloride at 25°C, Light Exposed

Diluent	**0.9% Sodium Chloride**					
Storage Conditions	**25°C, Light Exposed**					
Test Parameter	**Acceptance Criteria**					
		Initial	**12 Hours**	**24 Hours**	**48 Hours**	**72 Hours**
Completeness and Clarity of Solution	The constituted solution is not significantly less clear than an equal amount of the diluent	Complies	Complies	Complies	Complies	Complies
pH	4.3 – 7.5	4.9	4.9	4.9	4.9	4.9
Assay	90.0 – 110.0%	99.9%	100.1%	101.6%	99.4%	100.4%
Related Substances						
Lypressin	NMT 0.5%	0.15%	0.21%	0.21%	0.21%	0.19%
Unknown A[1]	NMT 1.0%	0.25%	0.26%	0.26%	0.22%	0.21%
Acetyl-Terlipressin	NMT 0.7%	0.33%	0.46%	0.39%	0.51%	0.42%
Individual Unspecified (largest reported)	NMT 0.5%	0.28% (RRT 1.08)	0.29% (RRT 1.08)	0.32% (RRT 1.08)	0.26% (RRT 1.08)	0.24% (RRT 1.08)
Total Impurities	NMT 5.0%	1.72%	2.04%	1.90%	1.92%	1.75%

1 Unknown A impurity was later identified as Impurity A

Table 3: In-Use Chemical Stability Data after reconstitution with 0.9% sodium chloride at 25°C, Dark Control (Wrapped in Foil)

Diluent	**0.9% Sodium Chloride**					
Storage Conditions	**25°C, Dark Control (Wrapped in Foil)**					
Test Parameter	**Acceptance Criteria**					
		Initial	**12 Hours**	**24 Hours**	**48 Hours**	**72 Hours**
Completeness and Clarity of Solution	The constituted solution is not significantly less clear than an equal amount of the diluent	Complies	Complies	Complies	Complies	Complies
pH	4.3 – 7.5	4.9	4.9	4.9	4.9	4.9
Assay	90.0 – 110.0%	99.9%	100.8%	102.5%	98.9%	98.8%
Related Substances						
Lypressin	NMT 0.5%	0.15%	0.23%	0.23%	0.22%	0.21%
Unknown A	NMT 1.0%	0.25%	0.32%	0.25%	0.24%	0.32%
Acetyl-Terlipressin	NMT 0.7%	0.33%	0.44%	0.43%	0.34%	0.47%
Individual Unspecified (largest reported)	NMT 0.5%	0.28% (RRT 1.08)	0.30% (RRT 1.08)	0.30% (RRT 1.08)	0.34% (RRT 1.08)	0.28% (RRT 1.08)
Total Impurities	NMT 5.0%	1.72%	2.01%	1.82%	1.83%	2.04%

Table 4: In-Use Chemical Stability Data after reconstitution with 0.9% sodium chloride at 2-8°C, Dark

Diluent	**0.9% Sodium Chloride**					
Storage Conditions	**2-8°C, Dark**					
Test Parameter	**Acceptance Criteria**					
		Initial	**12 Hours**	**24 Hours**	**48 Hours**	**72 Hours**
Completeness and Clarity of Solution	The constituted solution is not significantly less clear than an equal amount of the diluent	Complies	Complies	Complies	Complies	Complies
pH	4.3 – 7.5	4.9	4.9	4.9	4.9	4.8
Assay	90.0 – 110.0%	99.9%	100.0%	100.8%	99.5%	98.9%
Related Substances						
Lypressin	NMT 0.5%	0.15%	0.21%	0.21%	0.23%	0.21%
Unknown A	NMT 1.0%	0.25%	0.30%	0.30%	0.32%	0.30%
Acetyl-Terlipressin	NMT 0.7%	0.33%	0.43%	0.43%	0.33%	0.47%
Individual Unspecified (largest reported)	NMT 0.5%	0.28% (RRT 1.08)	0.28% (RRT 1.08)	0.26% (RRT 1.08)	0.30% (RRT 1.08)	0.26% (RRT 1.08)
Total Impurities	NMT 5.0%	1.72%	1.96%	1.94%	1.98%	1.93%

22 Appendix B: Sodium Chloride Solution:

NDC	00338-0049-31
Full FDB Description	SODIUM CHLORIDE 0.9% USP SOLUTION, SINGLE USE
Generic Name	0.9 % sodium chloride
Mfr/Supplier Part #	2B1308
FDA Application #	NDA 016677
Supplier	BAXTER
Strength	0.9 %

23 Appendix C: Terlipressin Microbiology

Table 1: Microbiological Stability Testing Protocol

Batch Tested:	Lot No. 870403F	
Study Start Date:	December 2020	
Product:	Terlipressin 2mg in 50mL NSS IV bag	
Storage Conditions:	2-8 °C – DEHP containing IV bag	
	2-8 °C – DEHP-free IV bag	
	25 °C – DEHP containing IV bag	
	25 °C – DEHP-free IV bag	
Study Intervals:	Initial, 12 hours, 24 hours, 48 hours, 72 hours (for Aerobic and Anaerobic testing) Initial, 72 hours (for Pyrogen testing)	
Test Parameters:	Aerobic bacteria	Negative for growth
	Anaerobic bacteria	Negative for growth
	Pyrogen	Less than 0.500 EU/mL

Table 2: Microbiological Stability Testing Results

Storage Conditions	**2-8 °C – DEHP containing IV bag**					
Test Parameter	**Acceptance Criteria**	**Initial**	**12 Hours**	**24 Hours**	**48 hours**	**72 hours**
Aerobic Bacteria	Negative for growth	Pass	Pass	Pass	Pass	Pass
Anaerobic Bacteria	Negative for growth	Pass	Pass	Pass	Pass	Pass
Pyrogen	Less than 0.500 EU/mL	Pass	n/a	n/a	n/a	Pass
Storage Conditions	**2-8 °C – DEHP-free IV bag**					
Test Parameter	**Acceptance Criteria**	**Initial**	**12 Hours**	**24 Hours**	**48 hours**	**72 hours**
Aerobic Bacteria	Negative for growth	Pass	Pass	Pass	Pass	Pass
Anaerobic Bacteria	Negative for growth	Pass	Pass	Pass	Pass	Pass
Pyrogen	Less than 0.500 EU/mL	Pass	n/a	n/a	n/a	Pass
Storage Conditions	**25 °C – DEHP containing IV bag**					
Test Parameter	**Acceptance Criteria**	**Initial**	**12 Hours**	**24 Hours**	**48 hours**	**72 hours**

Aerobic Bacteria	Negative for growth	Pass	Pass	Pass	Pass	Pass
Anaerobic Bacteria	Negative for growth	Pass	Pass	Pass	Pass	Pass
Pyrogen	Less than 0.500 EU/mL	Pass	n/a	n/a	n/a	Pass
Storage Conditions	**25 °C – DEHP-free IV bag**					
Test Parameter	**Acceptance Criteria**	**Initial**	**12 Hours**	**24 Hours**	**48 hours**	**72 hours**
Aerobic Bacteria	Negative for growth	Pass	Pass	Pass	Pass	Pass
Anaerobic Bacteria	Negative for growth	Pass	Pass	Pass	Pass	Pass
Pyrogen	Less than 0.500 EU/mL	Pass	n/a	n/a	n/a	Pass